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Bacteriological and Physicochemical Assessment of Akwaka River in Port Harcourt Nigeria Receiving Abattoir Effluent During Dry Season

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Abstract

A total of 50 samples were collected from five different locations (stations) along the River. The media used were nutrient agar, MacConkey agar, Eosine methylene blue agar, *Salmonella-Shigella* agar, proteolytic medium, Lipolytic medium, Amylolytic medium and cellulolytic medium. The method of isolation was pour plating technique. Colonial morphology, gram staining and biochemical tests were carried out using the standard microbiological methods. The results were subjected to statistical analysis- ANOVA. The mean count in log_{10}cfu/ml of TAPC ranged from 4.04±1.0 – 6.25 ± 5.2; *Escherichia coli count*, 1.84 ± 1.0 – 4.15 ± 1.62; coliform count, 2.0 ± 1.8 – 4.06 ± 1.2; *Salmonella- Shigella* count, 1.01 ± 0.2 – 1.35 ± 1.0; proteolytic count, 0.52 ± 0.3 – 1.98 ± 1.0; amylolytic count, 0.76 ± 0.1 – 1.15 ± 1.03; cellulolytic count, 0.44 ± 0.2 – 1.01 ± 0.4; lipolytic count, 0.43 ± 0.3 – 1.05 ± 0.23.The bacteria isolated and the percentage occurrence were *Bacillus species, 13%, Salmonella species,* 13%,*shigella species 8.5%, Pseudomonas specie 8.5%, Micrococcus species 12.3%, Staphlococcus auerus 16.2%, Klebsiella species 13.9%, and E.coli14.6%.* Temperature, P^H, conductivity, total dissolved solid. Biochemical oxygen demand, Chemical Oxygen demand and Sulphate were not significantly higher. Phosphates were significantly higher. This study revealed that the river is heavily contaminated and toxic.

Keywords: *Bacteriological, Physicochemical, Lipolytic, Cellulolytic, Amylolytic, Proteolytic*

Introduction

Water is the most precious natural resource that exists on our planet. Without the seemingly invaluable compound comprised of hydrogen and oxygen, life on earth would be non-existent. It is essential for everything on our planet to grow and prosper. Although we as humans recognize this fact, we disregard it by polluting our rivers, lakes and oceans. Subsequently, we are slowly but surely harming our planet to the point where organisms are dying at a very alarming rate. Not only organisms dying off but our drinking water has

become greatly affected (Reston, 2001). Water pollution is a major problem in the global context. It has been suggested that, it is the leading worldwide cause of deaths and diseases and accounts for the deaths of more than 14,000 people daily (West, 2006).

Water pollution is caused by anthropogenic contaminant and also natural phenomena such as volcanoes, algal blooms, storms and earthquakes. Water pollution due to pathogens cause many illnesses that range from typhoid and dysentery to minor respiratory and skin diseases. Pathogens include such organisms as bacteria, viruses and protozoa. These pollutants enter waterways through untreated sewage, storm drain, septic tanks, runoff from farms and particularly boats that dump sewage (Moe, 1997).

An abattoir has been defined as a premise approved and registered by the controlling authority for hygienic slaughtering and inspection of animals, processing and effective preservation and storage of meat products for human consumption (Alonge, 1991). Livestock waste spills can introduce enteric pathogens and excess nutrients into surface waters (Meadow, 1995). Also, if processors do not manage Abattoir wastes properly, it may cause other environmental problems like soil pollution with dung and atmosphere with methane from decomposing waste.

The aim and objective of this research is to ascertain the level of microbiological and physicochemical qualities of this river and advise the public on the health implications.

Materials and Methods

Study Area

The Akwaka River is situated at Rumuodomaya along Ikwerre Road in Port Harcourt, Rivers State. The river cuts across Aluu and Elelenwo in Obio/Akpor LGA of Rivers State. The river flows into Bonny River and beside the river is situated the Rumuodomaya Slaughter House.

Collection of Water Samples

Samples of water for this study were obtained from five different stations along the Akwaka River, where human activities take place. The sample was collected 500m apart. A total of 50 samples were collected. The samples were collected with sterile containers which were rinsed with the river water before collection and were transported immediately to the laboratory in a container containing ice packs and analyzed. The collection was between February to March 2010.

Enumeration of Heterotrophic Bacteria

After serial dilution of the samples, 1ml each of the appropriate dilution factor was pour plated into MacConkey agar, Eosin Methylene blue agar, *Salmonella-Shigella* agar, nutrient agar, lipolytic medium, proteolytic medium, Amylolytic medium and Cellulolytic medium. Colony Counter was used to estimate the total number of colony forming units.

Characterization and Identification of Bacterial Isolates

Characterization of the isolates was carried out by collecting from the mixed culture. This enables pure culture growth. Identification of the isolates was based on their microscopic examination, cultural morphology, carbohydrate fermentation and other biochemical tests.

Gram staining: Gram staining technique was used in identification/classification of bacteria into gram positive and Gram-negative organisms (Adeoye, 2007).

Statistical Analysis

Data obtained from this study was analyzed using the Statistical Package of Social Sciences (SPSS) version 17.0 for windows. Analyses of variance (ANOVA), independent Sample t-test were done at 0.05 level of significance. The result obtained thereof is represented as mean ± SEM (Agwung-Fobellah, 2007).

Results

The results of bacteriological and physiochemical characteristics of Akwaka River are shown in tables 1-3

Table 1 shows the mean counts of bacteria isolated in $\log_{10}$cfu/ml of TAPC ranged from 4.04±1.0 – 6.25 ± 5.2; *Escherichia coli count,* 1.84 ± 1.0 – 4.15 ± 1.62; coliform count, 2.0 ± 1.8 – 4.06 ± 1.2; *Salmonella- Shigella* count, 1.01 ± 0.2 – 1.35 ± 1.0; proteolytic count, 0.52 ± 0.3 – 1.98 ± 1.0; amylolytic count, 0.76 ± 0.1 – 1.15 ± 1.03; cellulolytic count, 0.44 ± 0.2 – 1.01 ± 0.4; lipolytic count, 0.43 ± 0.3 – 1.05 ± 0.23. Table 2 shows the bacteria isolated and the percentage occurrence were *Bacillus species, 13%, Salmonella species,* 13%,*shigella species 8.5%, Pseudomonas specie 8.5%, Micrococcus specie 12.3%, Staphlococcus auerus 16.2%, Klebsiella species 13.9%, and E.coli14.6%%.*Table 3 shows the result of the physicochemical parameters analyzed: Temperature 26.3-26.5^{0}C, pH ranged from 5.9-7.0, conductivity 80-1,240µs/cm. TSS ranged from 6-180mg/L, TDS 10.0-69mg/L, turbidity, 5.0-38NTU and BOD 1.8-16.1mg/L. Also, COD ranged from 6.4-59.7mg/L, sulphate 1.0-8.0mg/L,

Nitrate 0.25-5.26mg/L and phosphate 1.0-8.4mg/.

Temperature, pH, conductivity, total dissolved solid, biochemical oxygen demand, chemical oxygen demand and sulphate were not significantly higher. Phosphate were significantly higher ($p<0.05$) in the effluent entrance station compared to other station.

Table 1 also shows that the microbial mean count for total aerobic plate count was higher in bathing station more than the washing stations. The effluent station has the highest mean count compare with other stations. Total aerobic plate count was significantly higher ($p<0.05$) in all the water samples compared to those of *Escherichia* count, *coliform* count *Salmonella-Shigella* count, proteolytic count, cellulolytic count, amylolytic count and lipolytic count.

However, total aerobic plate count was not significantly different ($p<0.05$) between water samples from effluent station and bathing stations.

The effluent station had significantly higher ($p<0.05$) mean in *Escherichia* count, *coliform* count, proteolytic count, amylolytic count and lipolytic count compared to other stations.

However, there was no significant difference ($p<0.05$) in the mean of *Salmonella-Shigella* count and cellulolytic count in all the stations. *S. aureus* was significantly higher ($p<0.05$) in all the water samples compared to other bacteria.

Table 1: Mean Count of Microorganisms isolated

Log_{10}cfu/mL

Stations					
Stn 5	Stn 1	Stn 2	Stn 3	Stn 4	Stn 5
4.86 ± 1.2	5.55± 1.2	4.14 ± 1.23	6.25 ± 5.2	4.12 ± 1.3	4.04 ± 1.0
2.65 ± 0.3	4.15 ± 1.62	3.0 ± 2.4	3.12 ± 2.5	1.84 ± 1.0	3.32 ± 0.5
3.26 ± 2.0	2.15 ± 2.0	2.40 ± 1.2	4.06 ± 1.2	3.14 ± 2.2	2.0 ± 1.8
1.33 ± 0.3	1.35 ± 1.0	1.02 ± 0.5	1.52 ± 0.4	1.21 ± 1.3	1.01 ± 0.2
1.97 ± 0.5	1.29 ± 0.3	1.98 ± 1.0	2.4 ± 2.1	0.52 ± 0.3	1.43 ± 0.3
1.0 ± 0.1	0.97 ± 0.5	1.06 ± 0.1	1.15 ±1.03	0.88 ± 0.1	0.76 ± 0.1
0.98 ± 0.2	1.01 ± 0.4	0.44 ± 0.2	0.75 ± 0.3	0.72 ±0.32	0.45 ± 0.1
1.05 ± 0.2	0.43 ± 0.3	0.54 ± 0.1	1.05 ±0.23	1.03 ± 0.4	0.94 ± 0.5
3.94 ± 0.2	2.05 ± 1.6	2.98 ± 1.4	4.54 ± 1.56	4.00 ± 2.3	2.71 ± 0.1

Parameter	S	Stn 5
TAPC		4.04 ± 1.0

KEY:

TAPC - Total Aerobic Plate Count
EC - *Escherichia* Coli Count
CC - *Coliform* Count
SSC - *Salmonella-Shigella* Count
PC - Proteolytic Count
AC - Amylolytic Count
CL - Cellulolytic Count
LC - Lipolytic Count

Table 2: Bacteria isolated and their percentage occurrence
STATION

Bacteria	**Stn 1**	**Stn 2**	**Stn 3**	**Stn 4**	**Stn 5**	**TNI**	**%Occurrence**
Escherichia coli	4(80)	4(80)	5(100)	3(60)	3(60)	19	15.5
Klebsiella species	4(80)	3(60)	5(100)	3(60)	3(60)	18	14.6
Staphylococcus aureus	4(80)	4(80)	5(100)	3(60)	3(60)	19	15.5
Micrococcus species	0	4(80)	5(100)	3(60)	4(80)	16	13.0
Pseudomonas species	0	0	4(80)	3(60)	3(60)	10	8.1
Shigella species	3(60)	2(40)	4(80)	2(40)	3(60)	14	11.4
Salmonella species	3(60)	2(40)	4(80)	0	0	9	7.3
Bacillus species	4(80)	3(60)	5(100)	3(60)	3(60)	18	14.6
Total	25	22	37	17	22	123	100

KEY:

TNI – Total Number of Isolates

Table 3: The mean value of the parameters of the River Water Samples
Station

Parameter	**Stn 1**	**Stn 2**	**Stn 3**	**Stn 4**	**Stn 5**
Temperature	26.5	26.3	26.4	26.4	26.4
P^H	6.4	6.1	6.4	6.5	6.4
Odour	**Unobjectionable**				
Conductivity (µs/cm)	890	550	1,640	430	380
TSS(mg/L)	8.0	16	156	83	12
TDS (mg/L)	52.0	32.7	109	29.0	40
Turbidity (NTU)	5.0	6.0	28	15.0	8.0
BOD (mg/L)	3.9	2.6	31.2	13.6	3.8
COD (mg/L)	15.0	8.6	49.6	22.3	6.1
Sulphate (mg/L)	3.0	2.0	10.2	2.0	1.0
Nitrate (mg/L)	1.93	0.47	5.78	0.62	0.15
Phosphate (mg/L)	2.5	3.8	9.3	1.6	1.8

Key:

TSS– Total Suspended Solid
TDS – Total Dissolved Solid
BOD – Biochemical Oxygen Demand
COD – Chemical Oxygen Demand

Discussion

Abattoir operation is very beneficial to man in that it provides meat for human consumption and other useful by-products but it can be hazardous to public health in respect to the waste it generates (Adeyemi and Adeyemo, 2007). Bacteriological assessment of the samples indicated heavy contamination of the river. The bacteria isolates reported are in agreement with those of Adesemoye *et al*.,2006.

The bacteria isolated were *Bacillus* species, *Salmonella* species, *Shigella* species *Pseudomnas* species, *Micrococcus* species, *Staphylococcus aureus*, *Klebsiella* species *E. coli.* Their percentage occurrence indicates that *Staphylococcus aureus* was the most prevalent. This is because it is a normal flora of both human and animals and enters the river through humans bathing there *Staphylococus* is the leading cause of skin and soft tissue infections such as abscesses, furuncle and cellulitis. Although most Staphylococcus infections are not serious, it can cause serious infections such as bloodstream infection, pneumonia or bone and joint infections. Staph infections can spread by direct contact infected person, using contaminated objects or by inhaling infected droplets through sneezing and coughing. Humans using the water are most likely to be infected with *Staphylococcus* infection (Meadows, 1995).

All the bacteria isolates have their highest frequency of occurrence in station 3 because that station is the point of discharge of the effluent. *Micrococcus* species were found in all stations except station 1, *Pseudomonas* species were not found in stations 1 and 2. Again *Salmonella* species were not found in station 4 and 5.

The presence of cellulolytic, amylolytic, proteolytic and lipolytic bacteria showed that they were responsible for the degradation of cellulose, starch, protein and lipid materials contained in the abattoir wastes entering into the river (Eze and Ikeri, 2010). Also, as rain falls, it carries nutrients into the river which support microbial growth, thereby increasing their population (Eze and Okpokwasili, 2010). The presence of *E. coli* and *S. aureus* could be as a result of contamination of fresh meat or meat products during slaughtering or beef processing or unhygienic handling of the meat right from the slaughtering, butchering plants or contamination from skin, mouth, nose of the handlers that might have been introduced directly into by processors with lesion caused by *S. aureus* on hands coming in contact with food or by coughing and sneezing

(Sobukola *et al*., 2009; Okonko *et al*.,2008).

E. coli reduces nitrate to nitrite and so can survive easily in water receiving abattoir effluent because the waste from the animals is rich in nitrate. It therefore implies that contamination with water can cause *E. coli* infection such as diarrhea, urinary tract infection and wound infection.

Also *E. coli* can be introduced into the body of the water through faecal contamination from humans. *Klebsiella* and *pseudomonas* are associated with wound and urinary tract and so must have been introduced to the water body through human activities. The presence of *Coliforms* indicates faecal contamination.

Shigella dysenteriae causes bacillary dysentery, it infects human and transmission is by the faecal oral route with poor sanitation (Cheesbrough, 2005).

The values of BOD are at variance with the WHO (2004) permissible limit of 0.0mg/l for drinking water. This implies that it is dangerous to discharge the effluent directly into water without treatment, as this would deplete the water of dissolved oxygen that is needed by aquatic lives.

The high BOD (Biochemical Oxygen Demand) leads to less dissolved oxygen which is detrimental to aquatic lives-indicates less oxygen availability to higher aquatic life (Akpor and Muchie, 2011). This high BOD could be attributed to only partial or non-treatment of the effluents before releasing into the Akwaka River (Chukwu *et al.,* 2007). The high level of COD (Chemical Oxygen Demand) indicates the presence of chemical oxidants in the soil resulting to reduced nutrient availability rate to plants. Organize waste such as biodegradation industrial wastes can be treated to reduce their oxygen demand by a set of processes that are often referred to as primary secondary and tertiary treatment (Chukwu *et al.,* 2007).

The total dissolved solids values in all the stations were within WHO tolerance limits of 500mg/L (WHO, 2004). Thus, the contamination is not high enough to be worrisome. The suspended solids may also be related to the presence of organic matter and mainly to the fact that the abattoir does not treat its waste at all.

Temperature is a very important environmental feature in waste water. It controls behavioural characteristics of organisms, solubility of gases and salts in water (Joanne *et al*., 2011).

The temperature values of the water samples in all the stations were almost the same which could be due to the fact that the effluent was not coming from thermal pollution or a power plant (Ram *et al*., 2011), this would not have any effect, on the environment but if allowed to increase; it would greatly reduce the level of dissolved oxygen.

Conductivity is a method of obtaining an estimate of dissolved solids in waste water sample. The high values of conductivity in station C may likely be due to increase in dissolved solids and/or due to the presence of metallic ions (Ogbonnaya, 2008). The pH values of the water samples in all the stations indicate that the water is slightly acidic. The nitrate, phosphate and sulphate value of the water samples were due to abundance of them in the animal waste.

The use or consumption of this water therefore can cause nitrate, phosphate and sulphate toxicity. If the values of those parameters increase, it will cause nitrification.

Conclusion

The introduction or presence of substances such as animal faeces, fat, urine, bile, blood and bone in the body of water, favours the growth of microbes capable of utilizing starch, lipid, protein, cellulose etc. Some bacteria degrade these substances into smaller substances which also are used by other microorganisms. Also, fungi are capable of degrading component from abattoir effluents. Physicochemical quality of water receiving abattoir effluent is poor, therefore unfit for human use.

It is recommended from this research that abattoir effluent should be treated before disposal or stopped. Animal handlers should not wash the meat inside the river and people should not take bath or wash fruits or food items inside the river.

Relevant governmental agencies such as NAFDAC should monitor abattoir in Nigeria to ensure proper disposal of waste and place sanction on washing of meat inside the river. Also, conducive working environment should be created for Abattoir, so that the safety of workers and consumers of meat from the Abattoir is guaranteed.

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