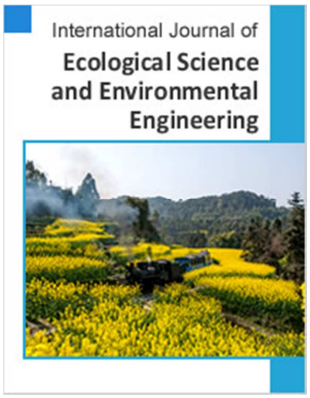
International Journal of Ecological Science and Environmental Engineering
2018; 5(2): 34-42
http://www.aascit.org/journal/ijesee
ISSN: 2375-3854

AASCIT American Association for Science and Technology



Keywords
Analysis,
Microbes,
Isolates,
Physiochemical Parameters,
Incubation and Water

Received: July 24, 2017
Accepted: December 11, 2017
Published: January 25, 2018

Microbiological and Physicochemical Analysis of Orogodo River, Agbor, Delta State, Nigeria

Omonigho Solomon Esharegoma[1], Nkem Chinedu Awujo[2], Inetianbor Jonathan[2, *], Igbeta Peace Nkonye-asua[1]

[1]Department of Microbiology, Faculty of Life Sciences, University of Benin, Benin City, Nigeria
[2]Department of Microbiology, Faculty of Pure and Applied Sciences, Federal University Wukari, Wukari, Nigeria

Email address
inetianbor@fuwukari.edu.ng (I. Jonathan)
[*]Corresponding author

Citation
Omonigho Solomon Esharegoma, Nkem Chinedu Awujo, Inetianbor Jonathan, Igbeta Peace Nkonye-asua. Microbiological and Physicochemical Analysis of Orogodo River, Agbor, Delta State, Nigeria. *International Journal of Ecological Science and Environmental Engineering*. Vol. 5, No. 2, 2018, pp. 34-42.

Abstract
The microbiological quality and physicochemical attributes of Orogodo river in Agbor, Delta state, Nigeria was investigated. Samples were taken from the river at two different points (upstream and downstream) f September, 2015 to February, 2016. Samples were collected bimonthly and were immediately transported to the laboratory for microbiological and physicochemical analysis using standard methods. In the rainy season, the mean bacterial counts ranged from $1.1\pm0.4x10^3$cfu/ml to $8.6\pm6.7x10^3$cfu/ml for the downstream and from $0.9\pm0.5x10^3$cfu/ml to $3.4\pm2.8x10^3$cfu/ml for the upstream. In the dry season, the mean bacterial counts ranged from $0.1\pm0.07x10^3$cfu/ml for the upstream to $1.1\pm0.4x10^3$ for downstream. In the rainy season, the mean fungal counts ranged from $0.5\pm0.3x10^3$cfu/ml for the downstream to $2.7\pm1.6x10^3$cfu/ml for the upstream. In the dry season, the mean fungal counts ranged from $1.3\pm0.8x10^3$cfu/ml for the downstream to $1.4\pm0.8x10^3$cfu/ml for the upstream. Bacterial isolates from the sampling points were *Bacillus subtilis, Klebsiella oxytoca, Alkaligenes facaelis, Escherichia coli, Staphylococcus aureus, Bacillus cereus, Pseudomonas fluorescens, Enterococcus facaelis, Citrobacter freundii, and Enterobacter aerogens* while fungal isolates were *Penicillium digitatum*, *Aspergillus flavus*, *Aspergillus niger*, *Fusarium oxysporium*, *Trichoderma* sp, *Mucor* sp. and *Saccharomyces* sp. The physicochemical parameter analysed showed a significant difference in the parameters of the downstream and upstream samples. This study has shown that surface water such as those from River are not microbiologically safe without treatment and as such, not fit for human consumption.

1. Introduction

A river is a natural flowing water course, usually fresh water, flowing towards an ocean, sea, lake or another river. In some cases, a river flows into the ground and becomes dry at the end of its course without reaching another water body. Small rivers can be referred to as stream, creek, brook, rivulet, and rill. There are no official definitions for the generic term river as applied to geographic features, although in some

countries or communities a stream is defined by its size. Many names for small rivers are specific to geographic location; examples are "run" in some parts of the United States, "burn" in Scotland and northeast England, and "beck" in northern England. Sometimes a river is defined as being larger than a creek, but not always: the language is vague [1]. A river can be termed upstream if the direction of flow is towards the source and against the current or downstream if the direction of flow is away from the source and towards the "mouth" of the river, in which the current flows [2]. Water is a basic natural resource for sustainable living on earth and though, this resource is very abundant on earth, it is not immediately available to the teeming human, animal, and agro-plant population all over the world. Besides, where water is available, it's quality is also a serous source of concern and this is more so, against the backdrop that the quality of water anywhere in the world is subject to natural, geographical, human- anthropological, and environmental factors and activities [3]. The quality of drinking water therefore comes with a premium to governments and individual all over the world and this is coordinated by the United Nation; as enshrined in the Millennium Development Goals of the world governing body in 2000. Hence according to the UN Secretary General in 2000, "No single measure would do to reduce disease and save lives in the developing world than bringing safe water and adequate sanitation to all". Adults require at least 1.6 litres of drinking water daily to replace lost fluid. However, contaminated water serve as a medium of transmitting dangerous pathogens and toxic chemicals into the body [4]. Water quality is defined in terms of the chemical, physical, and biological characteristics of water, usually in respect to its suitability for an intended purpose [5]. No single measure constitutes good water quality; this is because the quality of water appropriate for recreational purposes differs from that used for industrial processes. Worldwide, indicator organism such as coliform bacteria, *Escherichia coli* and faecal streptococci have been used to assess faecal pollution and deterioration in fresh water sources such as lakes, rivers, underground waters and streams [6]. Obiri-Danso and his colleagues [7] monitored coliform bacteria and enterococci in Subin, an urban river located in Kumasi Ghana, to determine the microbial pollution in the river. Faecal contamination of rivers is a major water quality issue in many fast growing cities where population growth far exceeds the rate of development of wastewater collection and treatment [7]. Despite global efforts during the United Nations water and sanitation decades of 1980s, improvement in water and sanitation infrastructure has not kept pace with population growth and urbanization in most developing countries. [7] Water quality is an important determinant to water availability because water which is not fit for use is in effect unavailable. The quality and quantity of available water have implication on the health status of a community. Polluted water is the major reason for the spread of many endemic diseases like skin and eye infections, cholera, tuberculosis, typhoid, diarrhoea, viral hepatitis A and even death [8]. Mortality in children less than five years due to water related diseases annually is estimated to be about 4 million in under developed countries and World Health Organization also estimated that 3.4 million people, mostly children, die every year from water-related disease [9]. Orogodo river serves as the drainage point for almost all run-off wastewater produced industrially and domestically within the Agbor metropolis and beyond as majority of the drainage channels constructed by the State and Local Government within the Agbor metropolis and surrounding villages, aimed at controlling flooding and checking erosion are channeled directly into the river at various points along the river length. The river also serves other domestic purpose such as cooking, washing, swimming and drinking and this necessitated study aimed at determining the microbiological qualities and physicochemical parameters of Orogodo River.

2. Materials and Methods

2.1. Study Area

This study was carried out in Orogodo river in Agbor, Delta state. Orogodo river is located at longitude 6 1/4 N° 6 1/4 E° [10]. The river is surrounded on both sides by sloppy terrain of the Agbor metropolis.

2.2. Sample Collection

Samples were collected from two points, the upstream and the downstream of the river into a 250ml sterilized sample bottles. The samples were stored in a portable ice box after collection and were transported to the laboratory for analysis.

2.3. Serial Dilution and Culturation

A ten-fold serial of the samples was carried out as previously described [11]. Samples for the bacterial isolates were cultured using nutrient agar (NA) while those for the fungal isolates were cultured using potato dextrose agar (PDA) by pour plate technique. The NA plates were incubated at 37°C for 24H and the PDA plates were incubated at 22°C for 96H. Colonies were counted after the incubation and were recorded in colony forming unit per litre.

2.4. Enumeration of Microorganisms

The total bacterial and fungal counts of the isolates were enumerated by counting the discrete colonies on the NA and PDA were selected and counted. The mean colony counts on the media plates at each dilution was used to estimate the total viable count for the samples in colony forming unit per ml cfu/ml. Total coliform count was determined by obtaining the most probable Number (MPN) using MacConkey broth and positive result was indicated by acid and gas production after incubation at 37°C for 48H. Presumptive test of the sample was carried out by transferring one (1) ml of each sample into sterile test tubes containing Lactose broth and inverted Durham tubes and incubated for 24H at 37°C after

which the tubes were checked for gas production. The confirmatory test was achieved by streaking the inocula from the gas positive tubes of the presumptive onto Eosin Methylene Blue agar plates and was again incubated at 37°C for 24H. After incubation, colonies which showed bluish black colour with green metallic sheen and reddish/brown colonies were noted and isolated on agar slants. Complete test was achieved by sub-culturing colonies which had formed green metallic sheen on Eosin Methylene Blue agar from the confirmatory test onto into tubes containing lactose broth and they were incubated at 37°C for 24H after which they were observed for gas production.

2.5. Microbial Characterization and Identification

The bacterial isolates were characterized and identified based on standard microbiological methods such as cultural characteristics (shape, Colour, size, cell arrangement, surface, opacity, texture, elevation and pigmentation), Gram staining reaction, acid-fast staining reaction, spore staining test, motility testing, Sugar fermentation test (glucose, mannitol, lactose, sucrose and mannose) and appropriate biochemical assays (catalase activity, indole test, oxidase test, motility test, urease test and coagulase activity) as described [12]. The fungal isolates were characterized by both morphological and the Lactophenol assay. The assay was carried out by placing a drop of lactophenol blue stain on a clean grease free sterilized glass slide. A sterile inoculating wire loop was used to pick a mycelium unto the glass slide from the fungal culture and a smear was made by spreading the mycelium evenly on the slide and then covered gently with cover slips and this was allowed to dry for few seconds after which it was observed with the microscope with the X40 objective lens. The colonial and morphological characteristics of each isolate were determined and compared with standard fungal reference books [13-14].

2.6. Physicochemical Analysis

The physicochemical characteristics of the sample was determined according to standard method by the WHO water guidelines 2004 [9].

2.6.1. Temperature

Measurements were made with a Celsius thermometer at the sampling points by immersing the thermometer directly in the river for about a minute. The temperatures were recorded to the nearest 0.5°C.

2.6.2. pH Measurements

A pH meter was used and the pH values were reported to the nearest 0.1 pH unit.

2.6.3. Electrical Conductivity (EC)

The electrical conductivities of the river water samples were recorded at room temperature (25°C) on a WTW LBR conductivity bridge with a cell constant of 1.63.

2.6.4. Total Suspended Solids (TSS)

A glass fiber filter paper was dried to a constant weight in an oven at 103–105°C. The glass fiber filter paper was affixed to a Gooch funnel and suction flask connected to vacuum pump. 250 ml of the river water samples were withdrawn with pipette into the filtration setup and filtered using vacuum pump. The glass fiber filter paper was carefully removed and again dried to a constant weight. The TSS was measured as the difference in weight of the glass fiber filter paper before and after the filtration using the formula:

$$\text{TSS (mg/L)} = \text{mg of suspended solids} \times 1000 \text{ ml of sample water used}$$

2.6.5. Total Dissolved Solids (TDS)

The TDS was obtained by the difference between the total solids (TS) and the total suspended solids (TSS)

$$\text{TDS} = \text{TS} - \text{TSS}$$

Prior to TS determination, a clean evaporating dish was dried in the oven at 103 – 105°C to a constant weight. 100 ml of the river water sample was withdrawn after thorough mixing into the evaporating dish and subsequently evaporated to dryness on a steam bath at 103 – 105°C to a constant weight. The difference in weight of the dish is noted. The TS is calculated using the formula:

$$\text{TS (mg/L)} = \text{mg of total solids} \times 1000 \text{ ml of sample water used}$$

2.6.6. Metal ions

The metal ion such as Pb^{2+}, Cu^{2+}, Ni^{2+}, Fe^{3+}, Na^{+}, K^{+}, were determined using atomic absorption spectrophotometric method as previously described [15]. Titremetric methods was employed in the determination of Ca^{2+} and Mg^{2+} at controlled pH. Initially, the water samples were titrated with EDTA to determine the total content of Ca^{2+} and Mg^{2+} ions using Eriochrome black as indicator. For the determination of Mg^{2+} ions alone, the Ca^{2+} ions were first precipitated as its oxalate and removed by filtration. The solution changed from wine red to blue at the end point, indicating a positive result.

2.6.7. Chlorides

In the determination of nitrate, the mercuric nitrate method was used. Chloride ions reacted with mercuric ions to form a highly stable complex, which was titrated with a standard solution of mercuric salt. The end point was determined by a diphenylcarbazone indicator, which formed a blue violet complex with an excess of mercuric ions.

2.6.8. Chemical Oxygen Demand (COD)

The dichromate method which is the reference method for COD determination was used. The river water samples were boiled under reflux with potassium dichromate and silver sulphate in strong sulfuric acid. Parts of the dichromate was reduced by organic matter and the remainder was titrated with ferrous sulphate. The solution changed from its previous blue-green to reddish brown colour when the end point was reached.

2.6.9. Biological Oxygen Demand

Water samples were neutralized to PH 7.0 with 1N

H_2SO_4/NaOH. Dilution water was prepared by aerating the distilled water till it became saturated with oxygen. Then, the desired volumes of distilled water were placed in a suitable bottle and one ml each of phosphate buffer solution, magnesium sulphate solution, calcium chloride solution and ferric chloride solution were added per litre of water. Several dilutions of sample in the range of 0.1 to 1% were made to obtain required dilution. Three bottles A, B, C each of 300 ml capacity, were chosen and filled with dilution water and stoppered without formation of any air bubble in them. Dissolve oxygen content of bottle 'A' was determined immediately. Bottle 'B' was used as a blank and in bottle 'C', 2 ml effluent samples was added. These two bottles were incubated at 20C for five days. Calculations

BOD = (Initial D.O – Final D.O) / ml of water volume of BOD bottles

2.7. Statistical Analysis

Mean and standard error for each parameter were calculated for the various samples collected. One-way analysis of variance (ANOVA) was computed to know the level of significant difference among the stations. The physicochemical parameters were subjected to Pearson's correlation analysis to investigate the level of interaction between the variability of the parameters with the aid of SPSS statistical software and the level of significance was determined at P = 0.5.

3. Results

In order to determine the microbial load of Orogodo river in Agbor, Delta state, Nigeria, both coliform count and the heterotrophic counts were carried out for the bacterial isolates as well as the fungal count for the fungal isolates. Table 1 shows the total heterotrophic bacterial counts in nutrient agar at 10^3cfu/ml of the samples. Total bacterial counts in Orogodo River varied with respect to sampling sites and sampling period. In week 1 of the various sampling months, bacterial counts in downstream water was highest in September (rainy season) and lowest in January (dry season). There was no significant difference in bacterial counts in the months of September, October and February while bacterial counts in January was significantly different from other months. Bacterial counts in downstream at the fourth week were statistically similar in the months of September, October and January while significant difference was observed in the month of February. All values of bacterial counts in upstream water were not significantly different at week 4. However, in week 1, bacterial counts were significantly different in the month of February. Result showed that bacterial counts were significantly higher in the downstream water compared to upstream water. Table 2 shows the total coliform bacterial counts at 10^3cfu/ml dilution of the sample. There was a significant difference in the coliform counts between the samples collected in the upstream. There was also a significant difference in the counts of the samples collected in rainy seasons (September and October) and those collected in dry season (January and February). Table 3 shows the fungal count at 10^3cfu/ml dilution of the samples collected at the two points. The fungal counts revealed higher fungal density in downstream water compared to upstream water. counts were also observed to be more in rainy season compared to dry season. In week 1, highest fungal counts in downstream water were observed in in the month of September while lowest count was observed in the month of January. However, in the upstream water, highest fungal counts were observed in the month of January while lowest counts were in the month September. Table 4 shows the results of various biochemical test carried out which help in the characterization and identification of the bacterial isolates. Figure 1 shows the pie chart of the amount of bacteria isolated from the samples. *Escherichia coli* was the highest bacteria isolated while *Klebseilla oxytoca* was the least bacteria isolated. Figure 2 shows the pie chart of the amount of fungi isolated from the samples. *Saccharomyces* sp was the highest fungal isolate while *Trichoderma* sp was the least fungal isolate. Table 5 show the result of the physiochemical parameter analyzed. There was a significant difference in the parameters were between the downstream water and upstream water. This difference was also observed among the seasons and month sampled.

Table 1. *Total heterotrophic bacterial counts at 10^3cfu/ml dilution in Orogodo River.*

Months	Downstream Samples		Upstream samples		*p*-value
	wk1	wk4	wk1	wk4	
Sept.	8.6 ± 6.7^{d}	8.1 ± 6.4^{c}	3.4 ± 2.8^{b}	2.8 ± 2.0^{a}	0.000
Oct.	2.9 ± 2.0^{d}	3.3 ± 2.3^{c}	2.2 ± 1.4^{b}	1.5 ± 0.7^{a}	0.000
Jan.	1.1 ± 0.4^{c}	1.1 ± 0.5^{c}	0.9 ± 0.5^{b}	0.6 ± 0.2^{a}	0.001
Feb.	2.1 ± 1.4^{d}	1.2 ± 0.4^{b}	1.5 ± 0.8^{c}	1.0 ± 0.5^{a}	0.000

Values are means of triplicates ± standard errors (SE) of the mean. Mean differences are presented as a, b, c and d across column and values with significant difference carry different alphabets.

Table 2. *Total coliform bacterial counts at 10^3cfu/ml dilution in Orogodo River.*

Months	Downstream samples		Upstream samples		*p*-value
	Dwk1	Dwk4	Uwk1	Uwk4	
Sept.	4.5 ± 2.9^{c}	4.5 ± 3.3^{c}	1.9 ± 0.9^{b}	1.7 ± 0.7^{a}	0.001
Oct.	2.9 ± 2.1^{c}	1.5 ± 0.8^{b}	1.4 ± 0.8^{b}	0.9 ± 0.5^{a}	0.001
Jan.	1.1 ± 0.4^{d}	0.9 ± 0.5^{c}	0.1 ± 0.07^{a}	0.7 ± 0.2^{b}	0.000
Feb.	1.7 ± 1.1^{d}	1.1 ± 0.4^{c}	0.08 ± 0.04^{a}	0.5 ± 0.2^{b}	0.000

Values are means of triplicates ± standard errors (SE) of the mean. Mean differences are presented as a, b, c and d across column and values with significant difference carry different alphabets.

Table 3. *Total fungal counts 10^3cfu/ml dilution in Orogodo River.*

Months	Downstream samples		Upstream samples		*p*-value
	Dwk1	Dwk4	Uwk1	Uwk4	
Sept.	2.7 ± 1.6^{c}	1.5 ± 1.2^{b}	0.5 ± 0.3^{a}	0.6 ± 0.2^{a}	0.000
Oct.	2.2 ± 1.4^{c}	1.6 ± 0.8^{c}	1.1 ± 0.5^{a}	1.3 ± 0.3^{b}	0.000
Jan.	1.4 ± 0.8^{c}	0.8 ± 0.2^{b}	1.3 ± 0.8^{c}	0.6 ± 0.2^{a}	0.000
Feb.	1.8 ± 1.1^{c}	1.0 ± 0.5^{b}	0.6 ± 0.2^{a}	0.9 ± 0.5^{b}	0.001

Values are means of triplicates ± standard errors (SE) of the mean. Mean differences are presented as a, b, c and d across column and values with significant difference carry different alphabets.

Table 4. *Biochemical assay of the Bacteria Isolates from the samples.*

Bacteria isolates	MOT	GS	SP	CAT	URE	OXI	COA	IND	CIT	H_2S	GLU	LAC
Bacillus subtilis	-	+	+	-	-	-	-	-	+	-	+	-
Klebsiella oxytoca	-	-	-	+	-	-	-	+	+	-	+	+
Alcaligenes faecalis	+	-	-	+	-	+	-	-	+	-	-	-
Escherichia coli	+	-	-	+	-	-	-	+	+	-	+	+
Staphylococcus aureus	+	+	-	+	-	-	+	-	+	-	+	+
Bacillus cereus	+	+	+	-	-	+	-	-	-	-	+	-
Pseudomonas fluorescens	+	-	+	+	-	+	-	-	-	-	+	-
Enterococcus facaelis	-	+	-	-	-	-	-	-	-	-	+	+
Citrobacter freudi	+	-	-	+	-	-	-	-	+	-	+	+
Enterobacter aerogene	-	-	-	+	-	-	-	-	+	-	+	+

Key: GM = Gram staining reaction, SP = Spore staining reaction, CAT = catalase, OXI = oxidase, COA = coagulase, URE = urease, MOT = Motility, IND = indole production, CIT = citrate utilization. = Glucose, LAC = Lactose, A = Acid production, + = Positive reaction/response and - = Negative/ no reaction.

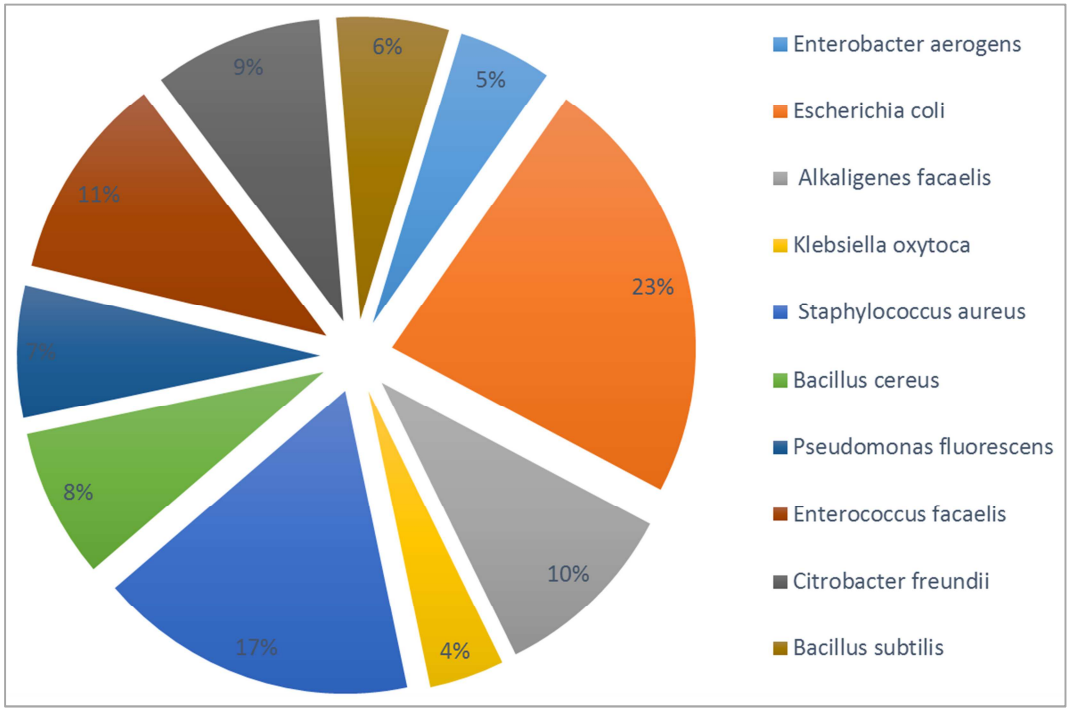


Figure 1. *Pie chart showing the quantity of bacterial isolates from Orogodo river. Escherichia coli and Klebsiella oxytoca were the highest and least bacteria isolated respectively.*

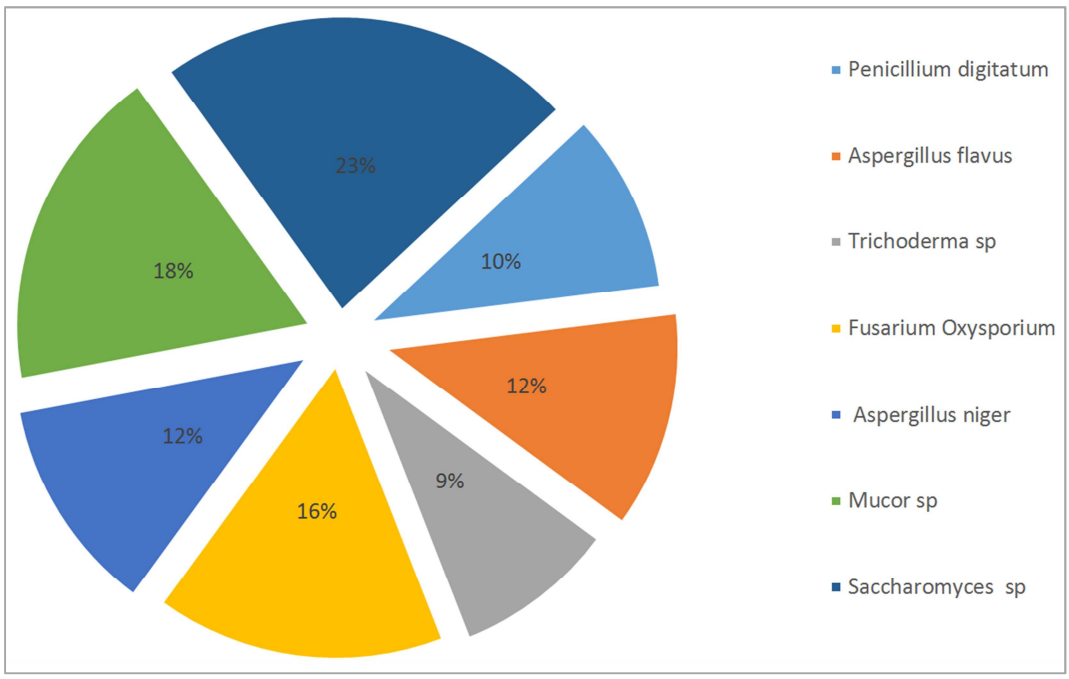


Figure 2. *Pie chart showing the quantity of the fungal isolates from Orogodo river. Saccharomyces sp and Trichoderma sp were the highest and least fungi isolated respectively.*

Table 5. *Physicochemical analysis of water samples from Orogodo River.*

Parameters	Rainy Season				Dry Season				*p*-value
	Sept		Oct		Jan		Feb		
	D	U	D	U	D	U	D	U	
pH	6.52 ± 0.62^{B}	5.85 ± 0.32^{AB}	6.07 ± 0.03^{AB}	5.94 ± 0.02^{AB}	5.50 ± 0.03^{A}	5.57 ± 0.03^{A}	5.55 ± 0.02^{A}	5.45 ± 0.02^{A}	0.101
Temp (°C)	26.16 ± 0.16^{A}	26.43 ± 0.31^{AB}	26.76 ± 0.14^{ABC}	27.23 ± 0.43^{BCD}	28.06 ± 0.12^{D}	27.93 ± 0.08^{D}	28.03 ± 0.27^{D}	27.36 ± 0.37^{CD}	0.000
EC (µS/cm)	109.6 ± 0.30^{E}	128.39 ± 0.21^{F}	177.54 ± 0.18^{G}	194.45 ± 0.29^{H}	84.41 ± 0.53^{C}	90.82 ± 0.36^{D}	72.14 ± 0.18^{B}	61.66 ± 0.24^{A}	0.000
TDS (mg/l)	54.39 ± 0.29^{AB}	64.15 ± 0.10^{B}	88.21 ± 0.22^{C}	114.46 ± 7.18^{D}	41.73 ± 0.14^{A}	64.26 ± 6.36^{B}	45.53 ± 7.37^{A}	44.86 ± 3.14^{A}	0.000
Cl^- (mg/l)	31.74 ± 0.84^{C}	38.87 ± 0.14^{E}	53.50 ± 0.25^{F}	72.30 ± 0.81^{G}	27.77 ± 1.46^{B}	34.94 ± 0.13^{D}	22.02 ± 0.74^{A}	23.57 ± 0.27^{A}	0.000
Na^+ (mg/l)	4.17 ± 0.11^{C}	5.12 ± 0.06^{D}	6.75 ± 0.28^{E}	9.45 ± 0.24^{F}	3.35 ± 0.08^{B}	4.33 ± 0.14^{C}	2.73 ± 0.17^{A}	3.10 ± 0.06^{AB}	0.000
K^+ (mg/l)	3.33 ± 0.03^{B}	3.65 ± 0.22^{B}	5.29 ± 0.03^{C}	7.25 ± 0.12^{D}	2.39 ± 0.10^{A}	3.48 ± 0.28^{B}	2.12 ± 0.06^{A}	2.30 ± 0.06^{A}	0.000
Ca^{2+} (mg/l)	0.39 ± 0.02^{BC}	0.42 ± 0.01^{C}	0.56 ± 0.01^{D}	0.8 ± 0.01^{E}	0.25 ± 0.01^{A}	0.37 ± 0.01^{B}	0.24 ± 0.01^{A}	0.26 ± 0.01^{A}	0.000
Mg^{2+}(mg/l)	1.37 ± 0.01^{C}	1.57 ± 0.01^{D}	2.09 ± 0.04^{E}	3.09 ± 0.04^{F}	1.03 ± 0.01^{B}	1.41 ± 0.01^{C}	0.85 ± 0.03^{A}	0.93 ± 0.01^{A}	0.000
Fe^{3+} (mg/l)	2.07 ± 0.03^{B}	2.31 ± 0.07^{B}	3.29 ± 0.15^{C}	4.63 ± 0.02^{D}	2.02 ± 0.22^{B}	2.17 ± 0.09^{B}	1.38 ± 0.01^{A}	1.42 ± 0.02^{A}	0.000
Cu^{2+} (mg/l)	0.42 ± 0.01^{B}	0.48 ± 0.03^{B}	0.74 ± 0.02^{C}	0.33 ± 0.18^{B}	0.35 ± 0.01^{B}	0.08 ± 0.01^{A}	0.30 ± 0.01^{B}	0.07 ± 0.02^{A}	0.000
Ni^{2+} (mg/l)	0.07 ± 0.01^{D}	0.05 ± 0.01^{BCD}	0.05 ± 0.00^{CD}	0.04 ± 0.01^{ABC}	0.03 ± 0.00^{ABC}	0.03 ± 0.00^{ABC}	0.01 ± 0.00^{A}	0.02 ± 0.01^{AB}	0.015
Pb^{2+} (mg/l)	0.11 ± 0.00^{B}	0.11 ± 0.01^{B}	0.15 ± 0.01^{C}	0.21 ± 0.01^{D}	0.06 ± 0.01^{A}	0.13 ± 0.01^{BC}	0.05 ± 0.01^{A}	0.05 ± 0.01^{A}	0.000
COD (mg/l)	62.45 ± 0.17^{G}	34.89 ± 0.09^{C}	61.05 ± 0.03^{F}	65.46 ± 0.47^{H}	47.16 ± 0.17^{E}	31.06 ± 0.07^{B}	41.10 ± 0.11^{D}	20.94 ± 0.02^{A}	0.000
DO (mg/l)	7.26 ± 0.07^{B}	13.4 ± 0.17^{E}	11.9 ± 0.02^{D}	10.62 ± 0.01^{C}	5.59 ± 0.02^{A}	6.99 ± 0.03^{B}	5.86 ± 0.07^{A}	7.35 ± 0.02^{B}	0.000
BOD (mg/l)	3.54 ± 0.19^{D}	5.19 ± 0.03^{E}	6.05 ± 0.02^{F}	6.36 ± 0.12^{G}	2.88 ± 0.01^{C}	3.08 ± 0.02^{C}	2.5 ± 0.02^{B}	2.07 ± 0.01^{A}	0.000

Values are means of triplicates ± standard errors (SE) of the mean. Mean differences are presented as A, B, C, D, E, F G and H across column and values with significant difference carry different alphabet. Key: U = Upstream, D = downstream

4. Discussion

There were variations in the total heterotrophic bacterial counts in surface water from Orogodo River, with respect to different sampling months. At the first week of every sampling month, bacterial counts in downstream water ranged from $1.1\pm0.4\times10^{3C}$cfu/ml (January) to $8.6\pm6.7\times10^{3D}$cfu/ml (September) while bacterial counts from the upstream ranged from $0.9\pm0.5\times10^{3B}$cfu/ml (January) to $3.4\pm2.8\times10^{3B}$cfu/ml (September). In the fourth week of sampling months, bacterial counts from downstream ranged from $1.1\pm0.5\times10^{3C}$cfu/ml (January) to $8.1\pm6.4\times10^{3C}$cfu/ml (September) while values in upstream ranged from $0.6\pm0.2\times10^{3A}$cfu/ml (January) to $2.8\pm2.0\times10^{3A}$cfu/ml (September). This finding is similar to that obtained by Olatunji and his colleagues [16] who reported microbial counts from Asa River in Ilorin. It is shown from above that bacterial counts from Orogodo River were more during the raining season compared to the dry season. This could be attributed to high run off of nutrients and debris from land during rainy seasons. Bacterial counts obtained in

MacConkey agar were observed to be higher in downstream water compared to upstream water and more in rainy season than dry season. For example, value of $4.5\pm2.9\times10^3$ and $1.9\pm0.9\times10^3$ cfu/ml for downstream and upstream respectively were obtained in the first week of sampling in the month of September. During dry season, counts of $1.1\pm0.4\times10^3$ and $0.1\pm0.07\times10^3$cfu/ml for downstream and upstream respectively were obtained in the first week of sampling during January. This suggests Presence of coliforms in River and usually is an indication that such water body has been contaminated with human or animal fecal materials and such water is usually not fit for human consumption. Fungal counts were observed to be lower than bacterial counts from sampled river water. again, there were more counts in downstream than upstream and in rainy season compared to dry season 2.7 ± 1.6 x10^3 and 0.5 ± 0.3 x10^3cfu/ml for downstream and upstream respectively were obtained during rainy season while 1.4 ± 0.8 x10^3 and 1.3 ± 0.8 x10^3cfu/ml fungal counts were obtained from downstream and upstream samples during dry season. Bacterial isolates from Orogodo River included *Bacillus subtilis, Klebsiella oxytoca, Alkaligenes facaelis, Escherichia coli, Staphylococcus aureus, Bacillus cereus, Pseudomonas fluorescens, Enterococcus facaelis, Citrobacter freundii, and Enterobacter aerogens* while fungal isolates were *Penicillium digitatum*, *Aspergillus flavus*, *Aspergillus niger*, *Fusarium oxysporium*, *Trichoderma* sp, *Mucor* sp. and *Saccharomyces* sp. Obire and his friends also reported the presence of these organisms in Elechi creek [17]. Microorganisms abound in the natural environment including water bodies. They function to bring about biodegradation of organic matters in the environment. Many of these organisms help to bring about bioremediation of contaminants in the environment. There were variations in the physicochemical parameters of Orogodo River. Hydrogen ion concentration (pH) was highest in downstream of river in the different months with values ranging from 5.50±0.03-6.52±0.62 and lowest upstream with range of values of 5.45±0.02-5.94±0.02. In the same vein, temperature ranged from 26.16±0.16-28.06±0.12 in downstream water and 26.43±0.31-27.93±0.08°C in upstream water. Electrical conductivity, 72.14±0.18-177.54±0.18 µS/cm and 61.66±0.24-194.45±0.29 µS/cm; total dissolve solid (41.73±0.14-88.21±0.22 mg/l and 44.86±3.14-114.46±7.18 mg/l); chemical oxygen demand (41.10±0.11-62.45±0.1mg/l and 20.94±0.02-65.46±0.4mg/l); dissolved oxygen (5.59±0.02-11.9±0.02 mg/l and 6.99±0.03-13.4±0.17 mg/l) and biochemical oxygen demand (2.5±0.02-6.05±0.02mg/l and 2.07±0.01-6.36±0.12mg/l). metal and non-metal ions were also assayed and range of values in downstream and upstream water are as follows: copper (0.30±0.01-0.74±0.02mg/l and 0.07±0.02-0.48±0.03mg/l) and lead (0.05±0.01-0.15±0.01 and 0.05±0.01-0.21±0.01 mg/l); sulphate (0.24±0.02-0.52±0.01 and 0.25±0.02-0.74±0.01 mg/l); nitrate (0.18±0.01-0.33±0.02 and 0.18±0.01-0.52±0.01 mg/l). The pH of water is extremely important and the pH values was found to be analyzed using pH meter was found to be more or less similar for each sample. Normal pH of water should be in the range of 6.5 to 8.5 for drinking and domestic purposes [9]. The mean temperature values of the water samples are not statistically different from each other ($p < 0.05$) and also fall within the normal temperature range supportive of good surface water quality which is 0°C to 30°C [16]. Hence, the temperature of the water from Orogodo River could not be implicated as influencing the observed variations in the bacterial population as well as in other physicochemical parameters. However, the observed range of the temperature allows for optimum proliferation of most of the bacteria isolated from the water samples. Enterobacteriaceae are mesophiles most of which grows optimally at temperature range of 20°C and 32°C [18]. Total dissolved solids (TDS) of the water from Orogodo River falls below the WHO standard limit for good water quality which is 1,000 mg/L [16]. Total dissolved solid, indicative of materials carried in solid [19]. Suspended solids whose source includes natural and anthropogenic (human) activities in the water shed, such as natural or excessive soil erosion from agriculture, forestry or construction, urban run-off, industrial effluents or phytoplankton growth [20]. A study carried out by other researchers to determine the average content of clay, silt, and sand in the bottom sediment of Orogodo River, as well as the concentrations of heavy metals (zinc, lead, copper, cadmium, nickel, manganese, iron, and chromium) by collecting and testing sediment at the river bed from five sampling sites along the length of the river, shows that the bottom sediment of Orogodo river comprises mainly sand and silt with higher silt content than sand, and small amount of clay [21]. The concentration of heavy metals in the sediments is low, but the iron content in some areas is close to its background value and may result in significant contamination of the environment in the future [21]. The high BOD and very high COD values are indicative of the presence of organic and inorganic pollutants, respectively. The mean BOD values of all exceeded the recommended maximum allowable concentration (RMC) set by the European Union for good quality water for fisheries and other aquatic life which is 3.0–6.0 mg/L [16]. Unpolluted waters typically have BOD values of 2 mg/L or less, whereas those receiving wastewaters may have values up to 10 mg/L or more [22, 16]. The COD usually includes all, or most of the BOD as well as some other chemical demands. The significantly high mean COD values also exceeded the acceptable concentrations for unpolluted surface water quality which is 20 mg/L or less. It however falls within the range of polluted waters (20–200 mg/L) [16]. The high BOD and COD values may be due to the discharge of untreated or incompletely treated industrial effluents into the river. The BOD values are high probably due to discharge of domestic wastes especially defecation activities and poorly executed agricultural activities near the river banks. Presence of magnesium and calcium is attributable to the geological formation of the source of the water samples and usually results in hardening of water. Hardness of water prevents lather formation with soap and increases the boiling point of water. Hardness of water mainly depends upon the amount of

calcium or magnesium salts or both. All the samples showed values lower than the acceptable standard values. Hardness has bad effects of developing corrosion and encrustation in pipes, consumes more soap, develops scales in boilers, influences drying processes and develops bad taste. The sulphate content of natural waters is an important consideration in determining their suitability for public and industrial supplies. Sulphate occurs naturally in water as a result of leaching from gypsum and other common minerals. Discharge of industrial wastes and domestic sewage tend to increase its concentration. The values of sulphate (SO_4) in all the samples were lower than the established standards. Excess amounts of nitrates in water cause an increase in algal growth and in drinking water can be toxic to humans. Sources of nitrates may include human and animal wastes, industrial pollutants and non-point source, runoff from heavily fertilized croplands and lawns. High levels of nitrates in drinking water have been linked to serious illness and even death in infants. Its presence here could be as a result of decomposition of natural vegetation and the use of chemical fertilizer in crop production in the area which may have entered the surface and groundwater. This can cause methanemoglobineamia in infants (blue babies). Phosphorus is an essential element for all life. It is part of the backbone of DNA. Phosphorous stimulates algal growth which can endanger other aquatic lives. None of the phosphate values were up to the NIS values of 10mg/l in all the water samples. Chloride is invariably present in small amounts in almost all natural waters and its contents go up appreciably with increasing salinity. High concentration of chlorides is considered to be indicator of pollution due to organic wastes of animal or industrial origin. Values of physicochemical parameters obtained in this work is not completely in agreement with earlier work of Issa and his colleagues who worked on the river sediment [23]. Difference may have arisen from the different sampling sites. Chlorides are troublesome in irrigation water and also harmful to aquatic life. The chlorine content of the water samples was all within the acceptable range. It has no adverse health impact, but excess of it impacts bad taste to the drinking water.

5. Conclusion

This work has shown that surface water from Orogodo River is highly contaminated with microorganisms including those of public health significance. Downstream water was shown to be more contaminated with upstream water. Significant variation in microbial load and physicochemical parameters was observed between rainy and dry season. It is recommended that surface water such as that of Rivers should be properly treated with appropriate method before consumption or any other domestic usage.

Conflict of Interest

The authors declare that there is no conflict of interest.

References

[1] Akoto, O. and Adiyah, J. (2007). Chemical analysis of drinking water from some communities in the Brong Ahafo Region. *International Journal of Environmental Science and Technology* 4 (2): 211-214.

[2] Aluyi, H. S. A., Ekhaise, F. O. and Adelusi, D. M. (2006). Effect of Human Activities and Oil Pollution on the Microbiological and Physicochemical Quality of Udu River, Warri, Nigeria. *Journal of Applied Sciences* 6 (5): 1214-1219.

[3] United Nation Department of Social and Economic Affairs (UNDSEA, 2001). Indicators of sustainable development: Guidelines and methodologies. 2nd edition. United Nations, New York. 99p.

[4] Hodgkinson, B., Evans, D., and Wood, J. (2003). Maintaining oral hydration in older adults: a systematic review. *International journal of nursing practice* 9: 19-28.

[5] Meybeck, M., Kimstach, V. and Helmer, R. (1996). Strategies for water quality assessment. In: Chapman, D. (Ed.) water quality assessment: A guide to the use of Biota, sediments and water in Environmental monitoring. 2nd edition. Chapman and Hall, London, pp 23-57.

[6] APHA, 1995. Standard Methods for the examination of water and wastewater, 19th edition, American Public Health Association, Washington DC, 52pp.

[7] Obiri-Danso, K., Weobong, C. A. A. and Jones, K. (2005). Aspects of health-related microbiology of the Subin, an urban river in Kumasi, Ghana. Journal of water and health 3 (1): 69-76.

[8] Herschy, R. W. (1999). Hydrometry principle and practices. 2nd ed. John Wiley and Sons, Chichester.

[9] World Health Organization, 2004. Guideline for drinking water quality. 2nd edition. Geneva, pp 231-233.

[10] Egun, N. K. (2010). Effect of Channeling Wastewater into Water Bodies: A Case Study of the Orogodo River in Agbor, Delta State. *Journal of Human Ecology* 31 (1): 47-52.

[11] Inetianbor, J. E., Ehiowemwenguan, G., Yakubu, J. M. and Ogodo, A. C. (2014). In-vitro antibacterial activity of commonly used toothpastes in Nigeria against dental pathogens. *Journal of Advanced Scientific Research* 5 (2): 40-45.

[12] Cheesbrough, M. (2000). District laboratory practice in Tropical countries part 2. Cambridge University Press PP 290-320.

[13] Mackie, J., McCartney, R., Andrew, G. F., Anthony, S., Barrie, P. M., Gerald, J. C. (1996). Fungal identification. *Practical Manual of Microbiology* 4: 715.

[14] Patrick, R. M., Ellen, J. B., Michael, A. P., Fred, L. T. and Robert, H. Y. (1995). Medical mycology. *Manual of clinical Microbiology* 6: 699-703.

[15] Ramchander, J., Rajitha, B., Sunitha, G., Praveen, E., Anjaneyulu, A., Sunitha, J. and Sayaji, R. (2015). Quantitative determination of heavy metals in the water sample of four areas of Hyderabad in Telangana state. *International Organization of Scientific Research-Journal of applied chemical* 8 (7): 18-19.

[16] Olatunji, M. K., Kolawole, T. A., Albert, B. O. and Anthony, I. O. (2011). Assessment of water quality in Asa River (Nigeria) and its indigenous *Clarias gariepinus* Fish. *International Journal of Environmental Research and Public Health* 8: 4332-4352.

[17] Obire, O., Tamuno, D. C. and Wemedo, S. A. (2005). The Bacteriological Water Quality of Elechi Creek in Port Harcourt, *Nigeria Journal of Applied Science-Environmental Management* 9 (1): 79–84.

[18] Fransol, G., Villers, G. andMasschein, W. J. (1985). Influence of temperature on bacterial development in water. *Ozone Science* 7: 205-227.

[19] Oladiji, A. T., Adeyemi, O. andAbiola, O. O. (2004). Toxicological evaluation of the surface water of Amilegbe River using rats. *Nigerian Society of Experimental Biology and Biochemistry* 16: 94-101.

[20] USEPA. (1997). Exposure factors Handbook-Final report. U.S. Environmental Protection Agency, Washington DC, USA. 447pp.

[21] Puyate, Y. T., Rim-Rukeh, A. and Awatefe, J. K. (2007). Metal Pollution Assessment and Particle Size Distribution of Bottom Sediment of Orogodo River, Agbor, Delta State, Nigeria. *Journal of Applied Sciences Research* 3 (12): 2056-2061.

[22] Okokoyo, P. A. and Rim-Rukey, A. (2003). Pollution pattern of Orogodo River, Agbo, Delta state, Nigeria. *African Journal of Applied Zoology and Environmental Biology* 5: 25-28.

[23] Issa, B. R., Arimoro, F. O., Ibrahim, M., Birma, G. H. and Fadairo E. A. (2011). Assessment of Sediment Contamination by Heavy Metals in River Orogodo (Agbor, Delta State, Nigeria). *Current World Environment* 6 (1): 29-38.