WATER QUALITY ASSESSMENTAND ANTIBIOTIC SENSITIVITY PATTERN OF BACTERIAL ISOLATES IN OBAGIE-UHI RIVER, EDO SOUTH, NIGERIA

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Abstract: Water quality assessment and antibiotic sensitivity pattern of bacterial isolates in Obagie-Uhi River, Edo South was investigated. Physicochemical and bacteriological analyses were carried out using standard analytical methods. The parameters values range were; pH (6.48 – 6.98), electrical conductivity (51.8 – 142.4 µS/cm), turbidity (1.1 – 4.4 NTU), total suspended solid (1.1 – 9.2 mg/l), total dissolve solids (25.5 – 72.1 mg/l), dissolve oxygen (5.5 – 6.7 mg/l), biological oxygen demand (1.1 – 3.2 mg/l), chemical oxygen demand (6.7 – 22.4 mg/l), chloride (13.5 – 35.6 mg/l), nitrate (0.64 – 3.11 mg/l). Heavy metals were all less than the prescribed World Health Organisation permissible levels for drinking water. Iron was observed to have the highest concentration which ranged from 0.43 – 0.95 mg/l and nickel was not detected in all the analyzed water samples. Water quality index for the upstream and downstream water was 61.34 and 71.90, indicating poor quality. The heterotrophic bacterial counts, was in the order of 10^3 cfu/ml and total coliform counts ranged from 5 to 9 MPN/100ml (upstream) and 9 to 14 MPN/100ml (downstream). The highest *Escherichia coli* counts (7 MPN/100ml) were recorded for downstream point. The most predominant bacterial isolates are *Micrococcus* sp., *Pseudomonas aeruginosa, Staphylococcus epidermidis, Klebsiella aeruginosa, Enterobacter aerogenes*. The widest zones of inhibition were observed for ofloxacin, recephin, ciprofloxacin and gentamicin and septrin showed no zone of inhibition on the tested bacterial isolates. In conclusion, the results obtained indicate that the water had poor bacteriological and overall water quality index, as such was unsuitable for direct human consumption.

Keywords: Antibiogram, physicochemical, water quality, bacteria

Introduction

The significance of water to human and other biological systems cannot be over emphasized and there are numerous scientific and economic facts that, water shortage or its pollution can cause severe decrease in productivity and deaths of living species (Garba *et al*., 2008). Water is the second most important life sustainer after oxygen (Aluyi *et al*., 2006). It is the medium in which all biochemical reactions of the body take place and constitutes about 65-70% of the body weight (Aluyi *et al*., 2006). The availability of water through surface and groundwater resources has become more critical from day to day (Aluyi *et al*., 2006; Ranee and Vasantha, 2010). Water bodies are constantly used as receptacles for untreated waste-water or poorly treated effluents accrued from domestic and industrial activities (Agbaire and Obi, 2009). This consequently renders the water bodies unsuitable for both primary and/or secondary usage. One of the most critical challenges in developing countries is the lack of adequate potable water, with the usual sources of drinking water being streams, rivers, wells and boreholes, which are usually not treated (Agbaire and Obi, 2009; Bisi-Johnson *et al*., 2017). Water quality problems have intensified through the ages in response to the increased growth and concentration of population and increased industrial activities (Lehloesa and Muyima, 2000). Water-borne diseases are diseases caused by the ingestion of water contaminated by human or animal faeces or urine containing microbial pathogens (WHO, 2006). Major factors affecting the microbiological quality of surface water are discharges from sewage works and runoff from informal settlements (Zamxaka *et al*., 2004). High total and faecal coliform counts in water are usually manifested in the form of diarrhoea, fever and other secondary complications (Fatoki *et al*., 2001). Nigeria, the most populous country in Africa, is endowed with generous water resources and this water provides resources for fishery, transportation, irrigation, recreational and domestic uses. In majority of the rural areas, the populaces do not have access to potable water and therefore depend on water from wells, streams and rivers (Adekunle *et al*., 2007).

The aim of this study was to determine the level of quality and antibiogram profile of the bacterial isolates from Obagie-uhi River which is located in a densely populated remote community called Obagie-uhi, Uhunwonde Local Government Area, Edo State, Nigeria.

Materials and Methods

Collection of surface water samples

Water samples were collected at two sampling points (upstream and downstream) along the Obagie-Uhi River from December, 2018 to April, 2019 with the aid of sterile clean labelled plastic containers. Upon collection, samples were immediately transported to the laboratory for analysis and stored at 4°C in a refrigerator.

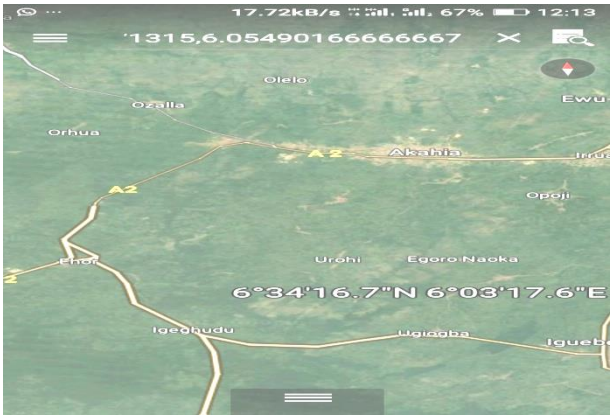

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Fig. 1: Google map of study area

Plate 1: Pictorial view of the upstream of Obagie-Uhi River

Plate 2: Pictorial view of the downstream of ObagieUhi River

Physicochemical parameters: The parameters analyzed are pH, Electrical conductivity, Total Suspended Solids, Total dissolved solids, colour, turbidity, Dissolved Oxygen (DO), Chloride, Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), nitrate, sulphate, phosphorus and Total Hydrocarbon Content (THC) (APHA, 1993).

Heavy metal analysis: Iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), chromium (Cr), cadmium (Cd), nickel (Ni), lead (Pb) and vanadium (V) were analysed using Atomic Absorbance Spectrophotometer (AAS) (Solar 969 Unicam series, UK) (APHA, 1993).

Enumeration of mean heterotrophic bacterial counts: Serial dilution of the respective surface water samples was done up to 10^{-6} with sterile Peptone water utilized as diluent. The mean viable heterotrophic bacterial counts were determined using pour plate technique with Nutrient agar (NA) utilized as general purpose media respectively (Harley and Prescott, 2002). The sterilized molten NA was dispensed into Petri dishes containing 1ml of the diluted aliquot appropriate dilution for the isolation of the heterotrophic culturable bacteria. The agar plates were swirled and allowed to solidify. The Nutrient agar (NA) plates were incubated at 35^{O}C and room temperature for 48 h and 7 days, respectively. The resultant bacterial colony counts on the agar plates was enumerated manually and recorded.

Determination of total coliform and *E. coli* counts: The method as described by Cheesebrough (2001) was used for the determination of the total coliform and fecal coliform (*Escherichia coli*) contents of the respective sample. Each count was conducted in three stages;

Determination of antibiotic sensitivity pattern of the water borne bacterial isolates

The antibiotic sensitivity pattern (antibiogram) of the isolates was determined using the disc diffusion method as described by Harley and Prescott (2002). The bacterial isolates were transferred to sterile Nutrient broth under aseptic conditions and incubated overnight. The turbidity of the broth cultures were adjusted to match an opacity standard ($BaSO_4$ turbidity standard). The resulting broth culture had a microbial cell density of about 10^8cfu. Nutrient agar plates were prepared and appropriately labeled. These plates were inoculated with the standardized bacterial broth cultures by spread plate technique (Harley and Prescott, 2002). The inoculated plates were left to dry for 15 min. Commercially available antibiotic discs containing varying concentrations of various types of antibiotics was placed at adequate distances on each of the seeded agar plates with the aid of sterile forceps under aseptic conditions. The antibiotic discs were; Ciprofloxacin (CPX) (10 µg), Chloramphenicol (CH) (30 µg), Sparfloxacin (SP) (10 µg), Amoxacillin (AM) (30 µg), Augmentin (AU) (30 µg), Gentamicin (CN) (30 µg), Pefloxacin (PEF) (10 µg), Streptomycin (S) (30 µg), Erythromycin (E) (10 µg), Ampiclox (APX) (30 µg), Zinnacef (Z) (20 µg), Ofloxacin (OFX) (5 µg) and Recephin (R) (25 µg). These plates were incubated for 12 hours and the resultant visible zones of inhibition were measured using a ruler (Harley and Prescott, 2002).

Evaluation of the water quality index of the sampled surface waters

The water quality index value for surface water sample sourced from the upstream and downstream sampling points on Obagie-Uhi river was determined using weighted arithmetic method as described by Boah *et al.* (2015). The water quality index (WQI) classification is 0-25 (excellent), 26-50 (good), 51-75 (poor), 76-100 (very poor) and above 100 (unsuitable for drink) (Akoteyon *et al*., 2011; Etim *et al*., 2013).

Results and Discussion

The results of the physicochemical quality of Obagie-Uhie River upstream and downstream samples are shown in Table 1. Generally, most parameters were within international and national recommended acceptable limits for drinking water. The parameters mean value range were; pH (6.48 – 6.98), electrical conductivity (51.8 -142.4 µS/cm), turbidity (1.1 -4.4 NTU), total suspended solid (1.1 – 9.2 mg/l), total dissolve solids (25.5 – 72.1 mg/l), dissolve oxygen (5.5 – 6.7 mg/l), biological oxygen demand (1.1 – 3.2 mg/l), chemical oxygen demand (6.7 – 22.4 mg/l), chloride (13.5 – 35.6 mg/l), nitrate (0.64 – 3.11 mg/l). Nitrate concentration in water sources are attributed to agricultural activities and sewage disposal, etc. (Akoteyon *et al*., 2011, Bisi-Johnson *et al*., 2017). The presence of heavy metals in water may be due to the chemistry of the geological composition of the river bed-rock, anthropogenic sources, leaching of corrosive materials and run-off due to erosion (Jaji *et al*., 2007). The high levels of magnesium in drinking water have been implicated with heart and kidney diseases in human (Etim *et al*., 2013). Findings from the heavy metal composition of Obagie-Uhie River are stated in Table 2. The results shows that investigated metals were all less than the prescribed World Health Organisation permissible levels for drinking water. Iron was observed to have the highest concentration which ranged from 0.43 – 0.95 mg/l and nickel was not detected in all the analyzed water samples. The low concentrations of trace metals in all the analyzed water samples was in agreement with an earlier report by Oguzie and Okhagbuzo (2010) which evaluated the heavy metal concentration of fresh water samples abstracted from several sampling points on the Ikpoba River, Benin City. The calculated water quality index (Table 3) shows that the status of the upstream and downstream water was 61.34 and 71.90. This finding revealed that the water from Obagie-Uhie River was of poor quality, therefore will require a routine monitoring and evaluation of the surface water, alongside with adequate treatment prior to consumption.

Table 1: Physico-chemical quality of water samples of Obagie-Uhi River

Parameters	Jan., 2019		Feb., 2019		March, 2019		April, 2019		May, 2019		WHO (2008)
	US	DS	US	DS	US	DS	US	DS	US	DS	
pH	6.56	6.59	6.77	6.54	6.48	6.58	6.51	6.52	6.52	6.98	6.5-8.5
EC (µS/cm)	97.2	99.6	135.9	142.4	91.6	142.3	59.1	51.8	84.3	120.7	400
Turbidity (NTU)	1.9	3.2	1.7	2.0	2.8	4.4	1.1	2.1	1.7	2.5	5
TSS (mg/l)	3.6	5.2	1.1	1.1	5.7	9.2	3.6	5.3	4.4	5.1	NS
TDS (mg/l)	47.6	48.8	68.6	68.6	45.8	72.1	29.8	25.5	42.9	61.1	500
DO (mg/l)	5.9	6.4	6.7	6.7	5.8	5.5	6.2	5.7	6.1	5.9	5
BOD_5 (mg/l)	1.8	2.5	1.1	1.1	1.6	2.4	1.3	3.2	1.1	3.2	NS
COD (mg/l)	8.9	11.3	7.4	7.4	7.2	16.5	8.6	22.4	6.7	12.5	NS
Cl (mg/l)	17.7	19.5	19.5	19.5	26.4	35.6	13.5	17.7	17.7	24.2	250
NO_3 (mg/l)	0.64	0.82	1.21	1.21	1.70	3.11	1.35	2.49	1.13	1.75	50

NS – Not stated, US (upstream), DS (downstream), over all mean values

Table 2: Heavy metals concentrations of water samples of ObagieUhi River

Parameters	Jan., 2019		Feb., 2019		March, 2019		April, 2019		May, 2019		WHO (2008)
	US	DS	US	DS	US	DS	US	DS	US	DS	
Fe (mg/l)	0.68	0.95	0.23	0.51	0.43	0.84	0.56	0.71	0.38	0.51	0.3
Mn (mg/l)	0.073	0.112	0.045	0.067	0.035	0.061	0.029	0.042	0.028	0.092	0.2
Zn (mg/l)	0.26	0.48	0.08	0.19	0.26	0.39	0.18	0.22	0.09	0.16	3
Cu (mg/l)	0.015	0.019	0.005	0.012	0.011	0.018	0.008	0.027	0.008	0.013	1.0
Cr (mg/l)	0.020	0.034	ND	ND	0.007	0.013	0.011	0.019	ND	ND	0.05
Cd (mg/l)	0.018	0.026	ND	ND	0.003	0.027	0.005	0.016	ND	0.017	0.003
Ni (mg/l)	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.02
Pb (mg/l)	0.004	0.011	ND	ND	0.004	0.042	0.028	0.064	ND	0.028	0.01

NS – Not stated, US (upstream), DS (downstream), over all mean values

Table 3: Water quality index (WQI) of water samples of Obagie-Uhi River

Parameters	Observed values (V_i)		Standard values (S_i)		Unit Weight (W_i)		Quality rating (qi)		$\Sigma w_i q_i$		WQI ($\Sigma w_i q_i$/ ΣWi)	
	US	DS	US	DS	US	DS	US	DS	US	DS	US	DS
pH	6.53	6.52	6.5-8.5	6.5-8.5	0.2190	0.2190	98.10	118.5	21.48	25.95		
Electrical Conductivity(µS/cm)	83.3	111.36	400	400	0.3710	0.3710	20.83	27.84	7.73	10.33		
Total Dissolved Solids (Mg/l)	46.94	55.22	500	500	0.0037	0.0037	9.39	11.04	0.04	0.04		
Chlorides (Mg/l)	18.96	23.3	250	250	0.0074	0.0074	7.58	9.32	0.06	0.07		
Nitrates (Mg/l)	0.0168	1.876	50	50	0.0412	0.0412	0.034	3.75	0.001	0.16		
Dissolved Oxygen (Mg/l)	6.14	6.04	5	5	0.3723	0.3723	122.8	120.80	45.72	44.97		
BOD (Mg/l)	1.38	2.48	5	5	0.3723	0.3723	27.60	49.60	10.28	18.47		
Total Suspended Solids (Mg/l)	5.18	5.18	500	500	0.0037	0.0037	1.036	1.04	0.004	0.0038		
					ΣWi=1.39	**ΣWi=1.39**			**85.27**	**99.9**	**61.34**	**71.90**

US (Upstream), DS (Downstream)

Table 4 shows the results of the heterotrophic bacterial counts, which is in the order of 10^3 cfu/ml. The mean heterotrophic bacterial counts for the upstream sample ranged from 3.1×10^3 to 7.8×10^3 cfu/ml and downstream range from $5.3x10^3$ to $11.6x10^3$ cfu/ml. The total coliform counts recorded for water samples obtained from the upstream sampling point ranged from 5 to 9 MPN/100 ml and downstream ranges from 9 to 14 MPN/100ml. The highest *Escherichia coli* counts (7 MPN/100 ml) were recorded for downstream point. The most predominant bacterial isolates are *Micrococcus* sp., *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Klebsiella aeruginosa*, *Enterobacter aerogenes*. Finding could be the direct result of a plethora of anthropogenic activities such as bathing, disposal of domestic wastes and natural phenomena such as surface run offs arising from precipitation. However, maximal microbial counts observed were recorded for water samples sourced at the downstream sampling point (Table 4); a reflection of concomitant increase of human activities leading to pollution of the water body. The detection of bacterial isolates such as; *Klebsiella* sp., *Escherichia coli*, *Enterobacter aerogenes*, *Streptococcus* sp., and *Pseudomonas aeruginosa* are of public health concern (Anazoo and Ibe, 2005) and it renders the water unfit for human consumption as the level of these indicator organisms were higher than the permissible limits stipulated by the Standards Organization of Nigeria for portable water (SON, 2007) and World Health Organization (2008). Furthermore, the isolation of *E. coli* from the water samples source points is indication that the water body is a reservoir for fecal contamination.

Table 4: Bacteriological quality of water samples of Obagie-Uhi River

Parameters	Jan., 2019		Feb., 2019		March, 2019		April, 2019		May, 2019		WHO (2008)
	US	DS	US	DS	US	DS	US	DS	US	DS	
THBC (10^3 x cfu/ml)	3.1	5.3	6.2	8.3	6.3	7.5	6.5	9.7	7.8	11.6	2
TCC(MPN/100ml)	7	10	7	9	5	12	7	14	9	13	0.0
E. coli counts (MPN/100ml	0	3	1	2	0	1	2	5	5	7	0.0

NS – Not stated, US (upstream), DS (downstream), THBC, Total heterotrophic bacterial count, THFC, Total heterotrophic fungal count, TCC, Total coliform count, over all mean values

Table 5: Zone(s) of inhibition (mm) representing antibiotic susceptibility patterns of bacterial isolates

G+ve	No. of isolates	PEF (10µg)	CN (10µg)	APX (30µg)	Z (20µg)	AM (30µg)	R (25µg)	CPX (10µg)	S (30µg)	SXT (30µg)	E (10µg)
Bacillus subtilis	15	15.0	10.0	0.0	17.0	0.0	14.0	19.0	12.0	0.0	20.0
Staph. epidermidis	9	0.0	18.0	0.0	15.0	0.0	22.0	16.0	10.0	0.0	15.0
Micrococcus sp.	12	0.0	18.0	0.0	10.0	0.0	17.0	20.0	10.0	0.0	15.0
G-ve		**SXT (30µg)**	**CH (30µg)**	**SP (10µg)**	**CPX (10µg)**	**AM (30µg)**	**AU (30µg)**	**CN (10µg)**	**PEF (30µg)**	**OFX (10µg)**	**S (30µg)**
Escherichia coli	5	0.0	0.0	16.0	20.0	17.0	15.0	15.0	0.0	24.0	8.0
Klebsiella sp.	*4*	0.0	0.0	20.0	17.0	20.0	16.0	20.0	0.0	20.0	13.0
Enterobacter aerogenes	*8*	0.0	10.0	18.0	22.0	18.0	15.0	18.0	10.0	22.0	10.0
Alcaligenes sp.	*8*	10.0	10.0	19.0	24.0	10.0	10.0	22.0	15.0	20.0	15.0
Pseudomonas aeruginosa	*5*	0.0	0.0	0.0	20.0	0.0	0.0	20.0	0.0	20.0	0.0

Ciprofloxacin (CPX), Chloramphenicol (CH), Sparfloxacin (SP), Amoxacillin (AM), Augmentin (AU), Gentamicin (CN), Pefloxacin (PEF), Streptomycin (S), Erythromycin (E), Ampiclox (APX), Zinnacef (Z), Ofloxacin (OFX), Recephin (R), Septrin (SXT)

The results of the zones of inhibition (mm) of antibiotics test are presented in Table 5. The widest zones of inhibition were observed for ofloxacin, recephin, ciprofloxacin and gentamicin against Gram positive isolates (*Staphylococcus epidermidis* and *Micrococcus* sp.) and Gram negative isolates (*E. coli*, *Klebsiella* sp., *Enterobacter aerogenes* and *Pseudomonas aeruginosa*). It was also showed that septrin had no zone of inhibition on the tested bacterial isolates.There was no significant difference observed differences ($P>0.05$) between values recorded for the respective upstream and downstream water samples.

In conclusion, the results obtained indicate that Obagie-Uhie River had poor bacteriological and overall water qualities index, as such was unsuitable for direct human consumption. It is recommended that enlightenment programmes should be organized by relevant governmental and non- governmental agencies (NGOs) to educate the people residing within the catchment area of the Obagie-Uhi River on the need to stop disposing domestic waste and faeces into the river.

Conflict of Interest

The authors declare that there is no conflict of interest reported in this work.

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