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Physicochemical and Microbial Properties of Dredged Oyibo River in Ehime Mbano, Imo State, Nigeria

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ABSTRACT

This study investigated the impact of dredging on the physicochemical, and microbial properties of Oyibo River in Ehime Mbano, Imo State, Nigeria. This study was carried out during the raining season (August) and dry season (January). Samples of water were collected using standard methods (collection from downstream to upstream). pH, temperature, flow rate, depth, width, conductivity, total dissolved solids (TDS), Dissolved Oxygen (DO) were analyzed in–situ using digital pH meter, thermometer, current meter, calibrated meter stick, measuring tape, Conductivity/ TDS meter and Dissolved Oxygen meter respectively. Other parameters (microbial, and some physicochemical parameters) were carried out ex-situ using existing standard methods. The river in the raining season had an average depth range of 1.60±0.00-2.00±0.00m; width range of 6.20±0.00-15.00±0.00m, pH, 5.60±0.00-7.15±0.05; temperature, 27.20±0.00 - 29.30±0.00^{0}C and flow rate range of 0.00±0.00 - 0.40±0.00 m/s. In the dry season the depth ranged from 0.08 - 0.50m; width, 6.20 - 11.40; pH, 2.8±0.00 - 5.50±0.00; temperature, 25.80±0.00 - 27.90±0.00^{0}C; and flow rate 6.20 - 11.40m. The water samples during the rainy season were found to be turbid (17.00±.0.00-48.44±0.00 NTU) while Chemical Oxygen Demand (COD), Biochemical Oxygen Demand (BOD), dissolved oxygen (DO), conductivity, total hardness and total alkalinity were within the Federal Ministry of Environment permissible limits. Statistically, there was no significant difference between the results in the raining season and dry season ($p \leq 0.05$). Microbial values exceeded their permissible limits with *Bacillus* spp, *Staphylococci* spp, *Enterococcus* spp, *Psendomonas* spp, *Micrococcus* spp, *Salmonella* spp and *Enterobacter* spp present in the water samples across the river points. *Asperigillus fumigatus, Asperigillus niger, Penicillum* spp, *Drechslera* spp, *Candida* spp, *Penicillum* spp, *Asperigillus niger, Asperigillus fumigatus and Paecilomyces* spp were among the fungi isolates from water and sediments samples. During the dry season, COD, BOD, DO, Total Hardness & Total chloride were observed to be within the permissible limits. No growth was observed for the Total Fecal Coliform Count while Total Bacteria count, Total Coliform count and Total Fungi Count were observed to be above permissible limits. Organisms observed in the surface water include, *Streptococcus* spp, *Klebsiella* spp, *Yersinia* spp, *Vibrio* spp, *Bacillus* spp, *Yersinia* spp, *Pseudomonas* spp, *Vibrio* spp and *Citrobacter* spp. Values of the sediment parameters were higher in the dry season. *Asperigillus niger, Candida* spp, *Asperigillus fumigatus, Penicillum* spp, *Asperigillus fumigatus* and *Paecilomyces* spp were the fungi that were observed in the surface water while *Candida* spp, *Penicillum* spp, *Dreschela* spp, *Penicillum* spp and *Asperigillus fumigatus* were observed in the sediment samples during the dry season. Some of the parameters are not within the federal Ministry of Environment (FMEnv.) acceptable limits, hence, the River in this study is not portable for drinking.

1. Introduction

The degradation of physical and chemical water quality due to human influences is often gradual and can only be easily detected through physicochemical and bacteriological analysis, otherwise, the degradation may not be readily detected until a dramatic shift in ecosystem condition occurs. This informed the need to assess the dredging impact of Oyibo River in Ehime Mbano, Imo State, Nigeria. Water quality is influenced by human activities, weathering of bedrock minerals, atmospheric processes of evapotranspiration and the deposition of dust and salt by wind, by the natural leaching of organic matter and nutrients from soil, by hydrological factors that lead to runoff, and by biological processes within the aquatic environment that can alter the physical and chemical composition of water (Akaishi *et al*., 2004). The human activities are not limiting to sand

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mining (River dredging), Stone/gravel mining, sink for wastes from agriculture and industry etc (Agbabiaka & Oyeyiola, 2012). Continuous assessment of water sources is important in maintaining the human health and health of the ecosystem since these waters are sourced for domestic water supply, irrigation, fishery development etc (Ayo & Arotupin, 2017).

Mbano is found in Southeastern part of Nigeria. Mbano shares boundaries with communities in Orlu, Okigwe and Owerri that constitute the three Geo-political zones of Imo State (Okoro, Uzochukwu & Chimezie, 2014). The area embraces Isiala and Ehime Mbano local government areas. Geographically, Mbano occupies an expanse of land of more than 205.30 square kilometers (Okoro, Uzochukwu & Chimezie, 2014). Mbano consists of Osu, Ehime, Mbama and Ugiri clans. The people are predominantly farmers. They are known for the production of palm wine (*mmanya-ngwo)*, from the raffia tree, and in much smaller quantity, (*mmanya-nkwu*), palm oil and kernels, cassava, yams, three-leaved yams (*una)*, coco-yams (*ede)*, native plantain (*unyere ojii)* etc (Okoro, Uzochukwu & Chimezie, 2014). They also keep live-stock such as goats, dogs, fowl, and pigs which provide them with a means of livelihood. The neighbours of Mbano are as follows: Ahiazu Mbaise on the West, Ikeduru/Mbaitolu on the South, Okwelle-Onuimu on the North and Agbaja-Isu in Nwangele on the eastern end, in Orlu zone. The contiguous communities that form neighbours of Mbano are, Eziama in Ekwereazu Mbaise, Inyishi, Amaimo, Atta and Umudim in Ikeduru, Etiti, Abajah Isu, Ogwa, and Okwelle in Onuimo. The boundaries which supposedly divide Mbano and its neighbours are no 'Berlin walls' that could prevent interaction (Okoro, Uzochukwu & Chimezie, 2014).

2. Materials and methods

Study area

This study was carried out in Ehime Mbano Local Government in Imo State, Nigeria. The study area is within $05^0$40.414'N $007^0$18.839'E and $05^0$40.468'N $007^0$16.866'E. It is in the tropical rainfall zone and has two distinct seasons, the wet and dry season. The wet season starts from April through October while dry season commences November through March annually.

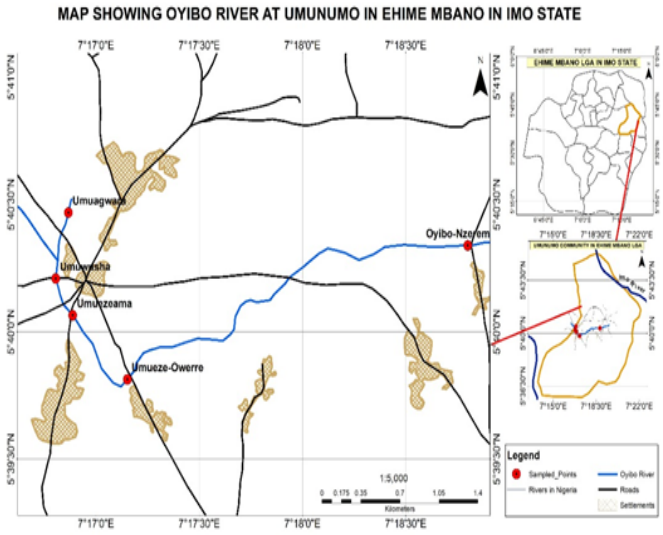

Fig 2.1. Map showing River Oyibo (Global Positioning System)

Water Sample Collection

This study was carried out between August, 2020 and January, 2021 at different locations: Ibeafor Umunumo, Umuogwara, Umuwosha, Umuezeama, Umueze-owere and Oyibo-Nzerem, all in Ehime Mbano, Imo State, Nigeria. Samples were collected using procedures of US EPA (2020). Water samples were collected from upstream to downstream in duplicates using $1dm^3$ sterile bottles while sediments were collected using a sterile stainless – steel scoop and placed in a sterile plastic bag. Both samples were collected from five (5) different sampling locations guided by where human, and other sundry activities were spotted across the communities through which the river transverses. The following parameters were analyzed in–situ; pH, temperature, flow rate, depth, width, conductivity, total dissolved solids (TDS), Dissolved Oxygen (DO) using digital pH meter/thermometer, Conductivity/ TDS meter and Dissolved Oxygen meter respectively. Other samples (water and sediments) for ex-situ investigations were collected and transported to the laboratory in iced coolers to maintain their integrity. However, water samples for the measurement of the 5days biological oxygen demand (BOD), were collected in 250ml brown sterile bottles while submerged in water.

Water analysis

Physicochemical analysis (who, 2011)

Determination of pH of Water Sample (ISO:3025 Electrometric Method)

pH meter was switched on for 30 minutes before the test. Buffer solutions: 4.0, 7.0 and 9.0 were prepared. The pH meter was calibrated to 9.2 and then 4.0 using the buffer and by adjusting the calibration knob. The pH meter was then calibrated to 7.0 using buffer solution. pH meter was inserted into the sample and read. The electrode was rinsed after removing from each buffer and sample.

Determination of Flow rate

The flow rate was determined using a current meter. The current meter was placed at the center of the stream and counting the number of revolutions of the rotor during a measured interval of time. The velocity/flow rate was then determined by dividing the number of revolutions by the time interval.

$$Flow\ rate = \frac{Nunber\ of\ revolutions}{Time} \qquad \text{Equ 2.1}$$

Determination of Depth

This was measured using a calibrated meter stick. Three points of the stream depths were measured and the average taken.

Determination of Width

This parameter was determined by using a measuring tape. The measuring tape was extended from the point where the bank meets the water on one side of the river to the same point on the other side. The measuring was held about 20cm above the water level.

Determination of Conductivity/Total Dissolved Solid (TDS) of Water Sample (ISO: 3025 Electrometric Method)

Conductivity meter was switched on (for at least 30 minutes before the test). A 0.1 M potassium chloride (KCL) was prepared. The conductivity meter was then calibrated to 14.12 mhos using the standard 0.1M KCl by adjusting the calibration knob. The conductivity meter was then inserted in the sample and reading recorded in µS/cm. This was later switched over to the TDS mode and recorded the reading in mg/l.

Total Solids (TS) (Iso:3025 Gravimetric Method)

The water sample was evaporated in a weighed dish on a steam bath and was dried to a constant mass in an oven at 105^0C. Clean evaporating dish 100 ml was dried at 105^0C in

an oven until constant weight was achieved. The dish was cooled to room temperature in a desiccator and weighed (w_1). Some of water sample (100ml) was pipetted into the dish after thorough mixture, and was evaporated to dryness on a steam bath. The outside of the dish was wiped, and the residue was dried in an oven at 105^0C for 1 hour. The dish was quickly transferred to the desiccator, cooled and weighed. The dish was returned to the oven to dry further for 10 -20 minutes, cooled in a desiccator and re-weighed. The drying was repeated until the weight of the dish plus residue was constant to within 0.05 mg (W_2). The weight of the dish (W_1) was subtracted from (W_2) to obtain the weight of the total solids.

$$Total\ Spolids = \frac{mg.\ total\ solids\ X\ 1000}{ml\ of\ sample} \quad \text{Equ 2.2}$$

Determination of Total Suspended Solid (TSS)

The total suspended solid was obtained by calculating from the difference between the total solid and total dissolved solids.

$$TS = TDS + TSS \quad \text{Equ 2.3}$$

Where;

TS = Total Solid; TDS = Total Dissolved Solids; TSS = Total Suspended Solids

Dissolved Oxygen (DO_2) Method: Electrometric

The DO meter was switched on and allowed to stabilize for about 15 minutes before calibrations were carried out following manufacturer's procedure. The DO_2 meter was calibrated prior the field trip for in-situ measurements by inserting the probe in 5% sodium sulphate solution for zeroing the equipment (0.0 mg/l O_2). The meter was put on and allowed to stabilize for about 10 minutes. The probe of the meter was then inserted into the water body, and a steady reading was recorded in mg/l O_2.

Biological Oxygen Demand (BOD_5). Electrometric & Incubation.

The water sample was carefully collected in-situ using Winkler's bottle by avoiding bubbling of water. The Winkler's bottle was covered inside the water body without air bubbles. The sample was placed in the cooler and transported to the laboratory. It was incubated at room temperature for 5 days. The DO_2 of the incubated sample was determined by inserting the DO Probe and the reading recorded in mg/l. The DO_2 of the 5 days was subtracted from the DO_2 of the day 1.

$BOD_5 = DO_2(1) - DO_2(5)$ Equ 2.4

Where;

DO_1= Average O_2 content of dilution water at the beginning of the assay (mg/l)

DO_5 = Average O_2 content of dilution water after 5days incubation (mg/l)

Temperature

Temperature analysis was carried out using Temperature Probe meter. The Temperature probe meter was switched on. The probe was inserted into the water body. A steady reading was recorded in ^{0}C.

Turbidity: Nephelometry

Spectrophotometer was switched on. The wavelength for turbidity (860 nm) was selected and the programmed number (750) was entered. Some 25 ml of distilled/de-ionized water(blank) was poured into one of the 25 ml cuvettes to the mark, and was inserted into the cell compartment or light shield then the zero button was pressed to zero the spectrophotometer. The water sample was vigorously shaken and 25 ml poured into the second cuvette. The blank cuvette was then replaced with the sample cuvette in the light shield. The READ button was pressed and the value of turbidity in the water sample was then digitally displayed in NUT (Nephelometric Turbidity Units)

Total Hardness (calcium + magnesium)

Some 25 mL of the sample was pipetted into a 250 mL conical flask with 50 ml of distilled water. Ammonia buffer solution (1 mL) was added followed by 2 drops of EBT (Eriochrome Black-T) indicator. The solution was titrated with EDTA (Ethylene Diamine Tetra Acetic Acid) solution as a distinct blue endpoint on addition of EDTA. Volume of EDTA added was noted.

$$CaCO3\ (mg/l) = \frac{v1\ x\ E\ (CaCO3)\ x\ 1000}{50} \quad \text{Equ 2.5}$$

Where;

S= mg $CaCO_3$ equivalent to 1 ml of EDTA titrant;

V = 1 mg $CaCO_3$

CHLORIDES(Cl^-):

(ARGENTOMETRIC TITRATION METHOD)

In 100ml of sample, 1ml of K_2CrO_4 indicator was added and titrated against 0.02N $AgNO_3$ till brick red precipitates were formed.

$$mg/L\ Cl = \frac{B.R\ x\ N\ x35.45\ x1000}{Vol\ of\ sample\ (ml)} \quad \text{Equ 2.6}$$

Where;

B.R= Burette reading (amount of titrant used);

N= normality of $AgNO_3$;

35.45= Equivalent weight of Chloride.

Sulphate (SO^{2-}_4) (Turbidimeter)

The photometer was switched on and the wavelength selected at 466 nm. Measure 10 ml of water sample was poured into 2 separate cuvettes. One of the samples (blank) was used to zero the photometer. One packet of barium chloride reagent was added to the second sample cuvette. The first cuvette was then replaced with the second reacted sample cell cuvette and timed for 5 minutes. The READ button was pressed at the end of the countdown time to display the reading in mg/l SO^{2-}_4

Colour (Colorimetric Platinum Cobalt)

The photometer was switched on, and the wavelength for colour was selected at 420 nm. A 10 ml cuvette was filled with distilled water to the mark and used as blank to zero the photometer. The cuvette was then replaced with another 10 ml cuvette filled with water sample. The READ button was pressed to display the reading in PCU (platinum Cobalt Unit).

Chemical Oxygen Demand (COD) (Titrimetry)

A 0.4g H_2SO_4 was weighed into a 250 ml Refluxing flask. Measured 20 ml of water sample or aliquot diluted to 20 ml was added into the flask. Sulphuric acids (2 ml) was added to the flask if NO_2-N is present in the sample. With the aid of pipette, 10 ml standard $K_2Cr_2O_7$ Solution was added in to the flask and then several glass beads previously dried at 600^oC for 1 hour, slowly added. $AgSO_4$ (30 ml) solution was added by gentle swirling. The flask was then connected to the condenser. A blank mixture was prepared and the mixture refluxed for 2.0 hours, cooled and the condenser was washed with distilled water into Erlenmeyer flask and diluted to 150 ml. The refluxed sample and the blank were cooled to room temperature and titrated with excess dichromate with standard FAS using 2-3 drops of ferroin as the indicator

$$Mg/L\ COD = \frac{(vb - vs)\ x\ M\ x\ 16{,}000}{ML\ sample} \quad \text{Equ 2.7}$$

Where;

vb = mL FAS used for blank;
Vs = mL FAS used for ample;
M = molarity of FAS

Phosphorus (Ortho Phosphate) (Ascorbic Acid Spectrophotometric)

Measured 50ml of water sample was mixed with 1 drop of phenol phltjalein indicator in a 125 ml conical flask. Red colouration was removed by adding $5NH_2SO_4$ with 8ml combined reagent and mixed. Absorbance of each sample was measured at 880nm after 15mins, Using distilled water blank with the combined reagent. A graph of absorbance versus phosphate concentration was plotted which gave a straight line.

$$Posphate\ (mg\ /L) = \frac{mg\ P\ from\ the\ calibration\ curve\ x\ 1000}{ml\ sample} \quad \text{Equ 2.8}$$

Nitrite (Colorimetric Method)

pH of 25ml sample was adjusted to between 5.0 and 9.0 and 75ml of NH4Cl- EDTA solution was added into the sample and mixed. Colour reagent (2 ml) was added to it. The colour was immediately read at 543nm using UV-Visible spectrophotometer (model spectrumlab 755s) indicating O.D. A standard graph was plotted to obtain the factor. Distilled water was used as Reagent blank to set the instrument.

$$Nitrate\ (mg\ /L) = \frac{O.D\ x\ Factor}{Vol\ of\ sample(ml)} \quad \text{Equ 2.9}$$

Where; O.D= Optical density

Nitrate (Cadmium Reduction Method)

pH of 25ml sample was adjusted to between 7.0. NH4Cl-EDTA solution (75ml) was added to the sample and mixed well. Nitrite is used to reduce Nitrate in the presence of Cadmium (Cd). This method uses commercially available cadmium granules coated with 2% copper sulphate ($CuSO_4$) packed in a glass column. The Nitrate (NO_3^-) produced was determined by diazotizing it with 2.0 ml colour reagent containing sulphanilamide coupled with N-(1-napthyl)-ethylenediamine diphydrochloride (NEDO) to form highly coloured azo dye. The colour developed was measured colorimetrically at 410nm. A correction was made for any NO_3^- present in the sample by analyzing the sample without the reduction of step.

$$Nitrate\ mg/l = \frac{O.D\ x\ Factor}{Vol\ of\ sample(ml)} \quad \text{Equ 2.10}$$

Where;

O.D= Optical density; Bi-Carbonate of Water Sample. (Titration Method)

Measured 100 mL of the water sample was pipetted into a clean flask and Barium Chloride solution (in excess) added to precipitate the carbonate which does not affect the bicarbonate. Two (2) drops of phenolphthalein indicator was then added to the solution. Then titrated with 0.02 M standard HCl (hydrochloric Acid). The volume of acid use was then recorded and calculated as:

$$Bi-carbonate = \frac{V \times M \times 100{,}000}{mL\ of\ sample\ used} \quad \text{Equ 2.11}$$

Carbonate: Titration Method

Carbonate was determined by titration method. To 50 mL of water sample in a clean conical flask was added 2 drops of methyl orange, shook well and titrated to the end point with 0.0 2 M standard HCl (Hydrochloric Acid), at pH 4.6. The colour changed from yellow to orange. The total conversion for this corresponds to the equation:

$OH^- + CO_3^{2-} + HCO_3^- + 4H^+ = 3H_2O + 2CO_2$

And calculate thus;

$$Cabonate = \frac{V \times M \times 100{,}000}{mL\ of\ sample\ used} \quad \text{Equ 2.12}$$

Microbial analysis

Inoculation

Each 1ml of each water samples were pipetted into a test tube containing 9mls of sterile distilled water. 10-fold serial dilution of the water sample were prepared using sterile distilled water as the diluents. Aliquots (0.1ml) of each (10^{-2}, 10^{-3}, 10^{-4} or 10^{-5}) of the test tube were inoculated in a Nutrient agar plate, Sabouraud dextrose agar and Mineral Salt Agar by spread plate technique and incubated at 37°C for 24hrs for bacteria and 25°C for 78hrs-96hrs for fungi on the incubator. After incubation, colonies were observed on different plates and counted and recorded.

$$No\ of\ Microorganisms = \frac{NO\ OF\ COLONY}{0.1}\ X\frac{1}{DF} \quad \text{Equ 2.13}$$

DF= Dilution factor (10^{-1} ,10^{-2} , 10^{-3} e.t.c)

Characterization and identification of microbial isolates

Microbial isolates were characterized based on cultural (colonial), microscopic and biochemical methods with reference to standard manuals (Nduka *et al*., 2020). The identities of the isolates were cross-matched with reference to standard manuals for the identification of bacteria.

Microscopic characterization (Nduka *et al*., 2020).

Gram staining test

A smear of each isolate was made on grease free glass slide with a drop of water and allowed to dry. The smear was fixed by mild heating, flooded with crystal violet and allowed to stand for 30 seconds. The crystal violet was rinsed off with water; Lugol's iodine was added and allowed to stand for 30 seconds. This was washed off with water and acid alcohol, till discoloration. It was counter stained with Safranin for 10 seconds and rinsed with water. The wet slide was allowed to air dry. A drop of oil immersion was added on the slide and viewed using x100 objective lens of the microscope.

Spore staining test

The spore stain was used to confirm the presence of spores when indicated in the Gram stain. Isolates were heat fixed on a slide and flooded with 5% malachite green. It was steamed for 3 minutes (without allowing it to boil), dried and cooled. It was then rinsed off and stained with Safranin for 30 seconds. This was rinsed, dried with filter paper and viewed under the microscope using oil immersion lens. The positive spores showed green while the vegetative cells were stained pink.

Motility test

This test was used to determine the motility of bacteria isolated. The test was carried out on a semi-solid agar medium in which motile bacteria swarm and gave a diffuse spreading growth. The medium was dispensed into test tubes, sterilized and allowed to set in an upright position. It was then inoculated using an inoculation needle by stabbing it into the medium in the test tube. This was incubated at 37°C for 24 hours. Diffuse growth from the straight line of inoculation was recorded as positive result.

Biochemical characterization of bacteria isolates

Microorganisms that were not identified by the colonial and microscopic characteristics were further subjected to few biochemical tests.

Catalase test

This test demonstrates the presence of catalase, an enzyme that catalyses the release of oxygen from hydrogen peroxide. One drop of 3% hydrogen peroxide solution was

placed on a clean slide. A loop full from 24 h culture was added. The release of bubbles (of oxygen) indicated the presence of catalase in the culture under test

Coagulase test

Coagulase is enzymes that clot blood plasma by a mechanism that is similar to normal clotting. The coagulase test identifies whether an organism produces this exoenzyme. This enzyme clots the plasma component of blood. The only significant disease-causing bacteria of humans that produce coagulase are *Staphylococcus aureus.* Thus, this enzyme is a good indicator of *S. aureus.* In the test, the sample was added to rabbit plasma and held at 37^0C for a specified period of time. Formation of clot within four hours indicated positive result and indicative of a virulent *S. aureus* strain. The absence of coagulation after 24 hours of incubation was a negative result, indicative of an avirulent strain.

Oxidase test

Oxidase test is used to check for the presence of terminal enzyme cytochrome c oxidase or cytochrome a. The presence of cytochrome c oxidase in the organisms is determined by the reagent tetramethyl-p-phenylene diamine dihydrochloride. It is used for the identification of *Pseudomonas* spp, *Aeromonas* spp, *Neisseria* spp, *Vibro* spp and *Pasteurella* spp, all of which produce oxidase. Members of entrobacteriaceae give negative oxidase test. A piece of paper was placed in a clean petri dish and 2-3 drops of freshly prepared oxidase reagent (1% tetramethyl-p- phenylene diamine dihydrochloride) was added. A small portion of culture was placed in the filter paper with the help of a sterile glass rod and a smear was made. Colour change was examined within 10 secounds. Blue-purple colour indicated oxidase positive.

Sugar fermentation/oxidation

This test is used to differentiate between bacteria groups that oxidize carbohydrate such as members of Enterobacteriaceae. One milliliter (1 ml) of 10% glucose, maltose, lactose, fructose, mannitol, and sucrose were separately under aseptic conditions transferred into duplicate tubes containing 9ml of sterile Hugh and Leifson's medium to obtain a final concentration of 1% of each of sugar. The tubes were stab-inoculated in duplicates while two uninoculated tubes served as control. Vaseline was used to cover one set of the duplicate tubes, one control to discourage oxidative utilization of sugar. All tubes were incubated at 37^oC for 48 h. After the incubation, they were observed for acid production in the culture. Yellow colouration indicated acid production in the open tubes, suggesting oxidative utilization of the sugar while acid production in the sealed tubes suggested a fermentative reaction.

Hydrogen sulphide production (H_2S) test

The test isolates were aseptically inoculated into a tube containing triple sugar iron agar started by stabbing the agar to the bottom and streaking the surface of the slant. The inoculated tube was incubated at 37^oC for 72 h and was examined daily. Black precipitation and yellow colouration was checked for. Black precipitate indicated H_2S production and yellow colouration for sucrose, lactose and glucose fermentation.

Urease test

Urease Agar slant in McCartney bottle was inoculated with the bacteria isolate at 30^oC for 4 hours and then overnight. A pink colour in the medium indicated a positive result.

IMViC TEST

This test consists of four different test; they are Indole production, Methyl-Red test, Voges Proskaeur test and Citrate utilization test. This test is specifically designed to determine the physiological properties of microorganism. They are especially useful in the differentiation of Gram-negative intestinal bacilli, particularly *Escherichia coli* and the *Enterobacter-Klebsiella* group.

Indole test

This test demonstrates the ability of certain bacteria to decompose the amino acid-Tryptophan to Indole. The bacteria isolates were inoculated into the medium and incubated at 37°C for 48 hours. At the end of incubation period, 3 drops of Kovac's reagents was added and then shaken. A red colour ring at the interface of the medium denotes a positive result.

Methyl red test

Methyl red test was preformed to demonstrate the capacity of different organisms to produce acid from the fermentation of sugar (dextrose). Methyl red reactions: Inoculated glucose phosphate medium was incubated at 37°C for 2 days. Two drops of methyl red solution were added shaken well and examined. Red colour indicated positive reaction and yellow indicated negative reaction.

Voges-Proskaeur test

The Voges-Proskaeur test demonstrates the ability of organisms to produce acetoin from glucose metabolism. Some organisms metabolize glucose to produce pyruvic acid which is further broken down to yield Butane-diol and acetyl-methyl carbinol as an intermediate product. Into one milliliter of the culture was added one milliliter of six percent alcoholic solution of alpha-naphtol and one milliliter of 16% KOH and stood for 15-20 minutes. Development of red to pink colour was a positive test.

Citrate utilization test

This is one of the several techniques used to assist in the identification of Enterobacteria. Principle of the test is based on the ability of an organism to use citrate as its only source of carbon. The test was carried out using Simmon's citrate agar.

The slopes of the media were prepared in bijou bottles as recommended by the manufacturers. A sterile straight wire was used to the slope with a saline suspension of the test organisms before stabbing the butt. The bottles were incubated at 35^oC for 48 h. Bright blue colours in the medium means positive test while no change in colour of medium indicates negative citrate test.

Statistical Analysis

Data were subjected to analysis of variance and t-Test at 95% probability.

3. Results and Discussion

Physicochemical Result of water samples during raining season

Fig 3.1 and Table 3.1 shows some physical properties and some physicochemical properties of water samples during raining season. From Fig 3.1, the depth of the river ranged between 1.60±0.00 to 2.00±0.00 m while the width ranged between 6.20±0.00 to 15.00±0.00 m. The temperature of the river across sampling locations ranged from 27.20±0.00 in Umuezema (SWQ 3) to 29.30±0.00^{0}C in Umuogwara (SWQ 5). pH was slightly Akaline and Slightly acidic, ranging between 5.60±0.00 in Umuogwara (SWQ 5) to 7.15±0.05 in Oyibo-Nzerem (SWQ 1) and Umueze-Owere, (SWQ 2). Appearance was clear and odor, unobjectable.

Total hardness values were between 20.73±0.09 in Umueze-Owere, (SWQ 2) to 26.77±0.00 mg/l ($Ca/MgCO_3$) in SWQ1, SWQ 3 and SWQ 4. The total hardness values, conductivity, turbidity, dissolved solid, chloride, nitrate, nitrite, phosphate and total alkalinity were within the Federal Ministry of Environment (FMEnv) standards. Dissolved oxygen ranged between 1.25±0.05 in SWQ 5 and 6.05±0.05 mg/l O_2 in SWQ 2 which are below FMEnv standards. There is no significant difference (P≤0.05) across the locations.

Microbial properties of water samples from Oyibo River during raining season

Table 3.2 shows the Total Bacteria Count (TBC), Total Coliform Count (TCC), Total Fecal Coliform Count (TFCC) and Total Fungi Count (TFC) of Oyibo River water samples during raining season. From the result, there was no fungi in water samples at points SWQ1, SWQ2 and SQW3. The Total Bacteria Count (TBC) and Total Coliform Count (TCC) were within the FMEnv Standards. TBC ranged between $2.5x10^5$±0.00 cfu/ml in (SWQ2, SWQ3) and $8.0x10^5$±0.00 cfu/ml (SWQ1). TCC values ranged between $3.0x10^3$±0.00 cfu/ml (SWQ1) and $1.8x10^4$±0.00 cfu/ml (SWQ2). There is no significant difference in the sample points sampled (P≤0.05). *Bacillus* spp, *Staphylococci* spp, *Enterococcus* spp, *Psendomonas* spp, *Micrococcus* spp, *Salmonella* spp and *Enterobacter* spp were the most frequently isolated bacteria from Oyigbo River samples as shown in Table 4.3.

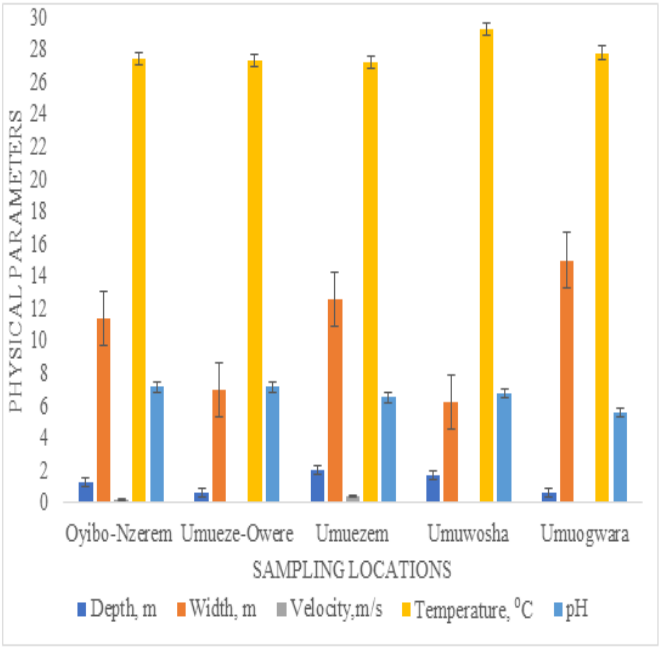

Fig 3.1. Some physical parameters of water samples during raining season.

All values were expressed as mean±SEM

Table 3.1. Physicochemical properties of water samples during raining season

PARAMETERS	FMEnv Standard	Mean± STDEV Oyibo-Nzerem, (SWQ 1) N05^{0}40.414' E007^{0}18.839' Elevation: 175.6ft	Mean± STDEV Umueze-Owere, (SWQ2) N05^{0}39.813' E007^{0}17.152' Elevation: 173.8ft	Mean± STDEV Umuezeama, (SWQ3) N05040.065' E007016.887' Elevation: 329.3ft	Mean± STDEV Umuwosha, (SWQ4) N05^{0}40.209' E007^{0}16.805' Elevation: 318.5ft	Mean± STDEV Umuogwara (SWQ5) N05^{0}40.468' E007^{0}16.866' Elevation: 358.1ft
Colour, PCU	15.00	176.5±1.50	129.00±1.00	129.00±1.0	209.00±0.00	6.00±0.00
Appearance	Clear	Slightly brownish	Slightly brownish	Slightly brownish	Slightly brownish	Clear
Odour	Unobjectionable	Objectionable	Objectionable	Objectionable	Objectionable	Unobjectionable
Conductivity, µS/cm	1000.00	26.00±0.00	23.00±0.00	22.00±0.00	20.00±0.00	29.50±0.50
Dissolved Oxygen, mg/l	>7.50	5.50±0.10	6.05±0.05	5.10±0.00	3.20±0.00	1.25±0.05
Biochemical Oxygen Demand, mg/l	NS	1.30±0.10	2.75±0.05	1.70±.0.00	1.20±.0.00	0.70±.0.00
Chemical Oxygen Demand, mg/l	NS	132.48±0.00	103.04±0.00	69.92±0.00	294.40±.0.00	125.12±0.00
Turbidity, NTU	10.00	42.66±0.14	47.02±0.02	43.84±0.00	48.44±0.00	17.00±.0.00
Total Solid, mg/l	500.00-1000.00	38.00±0.00	225.00±0.00	91.00±0.00	17.00±1.00	67.00±0.00
Total Dissolved Solid, mg/l	500.00	16.90±0 .00	14.95±0 .00	14.30±0 .00	13.00±0 .00	19.18±0.33
Total Suspended Solid, mg/l	<10.00	21.10±0.00	210.05±0.00	76.70±0.00	35.00±0.00	48.15±0.00
Total chloride, mg/l Cl-	250.00	27.99±0.00	35.99±1.00	29.99±0.00	24.99±0.00	35.49±0.50
Total Hardness, mg/l	200.00	26.77±0.00	20.73±0.09	26.77±0.00	26.77±0.00	23.38±0.42
Nitrate, mg/l	50.00	21.00±1.00	17.50±0.50	29.50±0.50	27.00±0.00	36.00±1.00
Nitrite, mg/l	0.30	0.28±0.01	0.26±0.00	0.18±0.00	0.23±0.00	0.21±0.01
Phosphate, mg/l	5.00	0.62±0.02	0.68±0.01	0.68±0.01	0.40±0.-00	1.28±0.02
Total Alkalinity, mg/l	150.00	14.00±0.00	14.00±0.00	16.00±0.00	20.00±0.00	16.00±0.00
Bicarbonate, mg/l	30.00	13.97±0.00	13.98±0.00	15.99±0.00	19.99±0.00	15.99±0.00
Sulphate, mg/l	200-400.00	10.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	10.00±0.00
Total Sulphide, mg/l	NS	6.80±0.00	9.20±0.00	8.20±0.00	4.60±0.00	9.00±0.00

mg/l- milligram per litre; **ND-** None Detected**; µS/cm-** microSiemens per centimeter, NS- Not Stated;

Table 3.2. Microbial Properties of water samples from Oyibo River during raining season

PARAMETERS	FMEnv Standard	Mean± STDEV Oyibo-Nzerem, SWQ 1 N05^{0}40.414' E007^{0}18.839' Elevation: 175.6ft	Mean± STDEV Umueze-Owere, SWQ2 N05^{0}39.813' E007^{0}17.152' Elevation: 173.8ft	Mean± STDEV Umuezeama, SWQ3 N05040.065' E007016.887' Elevation: 329.3ft	Mean± STDEV Umuwosha,SWQ4 N05^{0}40.209' E007^{0}16.805' Elevation: 318.5ft	Mean± STDEV Umuogwara SWQ5 N05^{0}40.468' E007^{0}16.866' Elevation: 358.1ft
Total Bacteria count, Cfu/ml	0-30	$8.0x10^5$±0.00	$2.5x10^5$±0.00	$2.5x10^5$±0.00	$2.6x10^5$±0.00	$3.0x10^5$±0.00
Total Coliform count, cfu/ml	0-10	$3.0x10^3$±0.00	$1.8x10^4$±0.00	$7.0x10^3$±0.00	$5.0x10^3$±0.00	$1.2x10^4$±0.00
Total Faecal Coliform Count, cfu/ml	0	NG	NG	NG	$4x10^2$±0.00	$4x10^2$±0.00
Total Fungi Count, cfu/ml	0	$2.0x10^3$±0.00	$4.0x10^3$±0.00	$4.0x10^3$±0.00	$7.0x10^3$±0.00	$3.0x10^3$±0.00

cfu/100ml- colony forming per 100 millilitre; NG- No Growth.

Table 3.3. Biochemical Identification of Some Bacteria Isolated from Oyigbo River.

SAMPLING POINTS	COLONY MORPHOLOGY	MICROSCOPIC CHARACTERISTICS	GRAM STAIN	SLANT	BUTT	GLUCOSE	LACTOSE	GAS	H_2S	CITRATE	CATALASE	MOTILITY	OXIDASE	V.P	METYL RED	MOST PROBABLE ORGANISM
OYIBO-NZEREM	Creamy colony on Nutrient agar	Rod	+	B	A	+	-	-	-	-	+	+	+	+	-	*Bacillus spp Staphylococci spp*
		Cocci	+	B	A	-	-	-	-	-	+	+	-	+	-	
UMUEZE–OWERE	Creamy colony on nutrient agar	Cocci	+	B	A	+	-	-	-	-	-	-	+	+	-	*Enterococcus spp*
UMUEZAMA	Shiny creamy colony on Nutrient agar	Rod	-	B	B	-	-	-	-	-	+	+	+	+	-	*Psendomonas spp*
		Cocci	+	B	A	+	-	-	-	-	+	-	-	-	+	*Micrococcus spp*
UMUWOSHA	Mucoidal creamy colony on Nutrient agar	Rod	-	B	A	+	-	-	-	+	+	+	-	-	+	*Salmonella spp*
UMUOGWARA	Shiny creamy colony on Nutrient agar	Rod	-	B	A	+	-	+	-	-	+	+	-	-	+	*Enterobacter spp*

A = Acidic condition; **B** = Basic condition; **+** = positive; **-** = negative

Properties of Sediments samples from Oyibo River during raining season

Table 3.4 shows the properties of sediments samples from Oyibo River during raining season. pH of the sediments at the various points were observed to be acidic ranging from 3.20 to 4.80. which are within the FMEnv standard. Sulphate was not detected in samples from SWQ1, SWQ2, SWQ3 and SWQ4. Microbial Counts were observed to be above the standard limits. No significant difference ($P \leq 0.05$) was observed from the water samples. *Bacillus* spp, *Shigella* spp, *Salmonella* spp, *E.coli* spp, *Enterococcus* spp, *Moraxella* Spp, *Enterobacter* spp were isolated, and their distribution shown in Table 3.5.

The Fungi Species in water samples from Oyibo River during raining season

The distribution of the fungi species in water samples from Oyibo River during raining season as shown in Table 4.6. *Asperigillus fumigatus, Asperigillus niger, Penicillum spp, Drechslera spp, Candida spp, Penicillum spp, Asperigillus niger, Asperigillus fumigatus* and *Paecilomyces spp,* were the most frequently isolated fungi. While *Candida* spp, *Asperigillus niger, Drechslera* spp, *Drechslera* spp and *Penicillum* spp were observed to be present in the sediments.

Physicochemical properties of water samples from Oyibo River during dry season

Fig 3.3. representing the physical properties of Oyibo River at different points during dry season shows that the river depth ranged from 0.08±0.00 to 0.50±0.00m; width, 6.20±0.00 to 11.40±0.00 and velocity (flow rate) range from 0.00±0.00 to 0.09±0.00m/s. The temperature range was between 25.80±0.00 (SWQ5) to 27.90±0.00^{0}C (SWQ1).

From Table 3.7, Colour was slightly brownish with objectable odour at SWQ5. At SWQ 4,3,2, colour was observed to be slightly turbid with objectable odour while SWQ1 was clear with unobjectable odour. Temperature, conductivity, dissolved oxygen, total solids, total chloride, total hardness, nitrate, phosphate, total alkalinity were observed to be within the FMEnv standard limits. Dissolved oxygen ranged from 8.00±0.00 (SWQ4) between 14.10±0.00 mg/l (SWQ1). At SWQ1, nitrite was observed to have higher values together with total suspended solid, total solids, dissolved oxygen and colour. There is no significant difference ($P \leq 0.05$) observed from the physicochemical properties from the water sample points.

Table 3.4. Properties of Sediments samples from Oyibo River during raining season.

PARAMETER	*FMEnvStd 2006*	*SQ1 N05^{0}40.414' E007^{0}18.839' Elevation: 175.6ft Oyibo-Nzerem*	*SQ2 N05^{0}39.813' E007^{0}17.152' Elevation: 173.8ft Umueze-Owere*	*SQ3 N05^{0}40.065' E007^{0}16.887' Elevation: 329.3ft Umuezeama*	*SQ4 N05^{0}40.209' E007^{0}16.805' Elevation:318.5ft Umuwosha*	*SQ5 N05^{0}40.468' E007^{0}16.866' Elevation: 358.1ft Umuogwara*
Ph	6.50	4.70	3.90	3.20	4.80	3.60
Conductivity, µS/cm	1000.00	196.00	154.00	130.00	246.00	132.00
Total chloride, mg/kg Cl^-	250.00	159.95	107.99	53.98	63.98	75.98
Phosphate, mg/kg PO^{-3}_4	>100.00	0.49	0.64	2.74	1.59	0.57
Nitrate, mg/kg NO^-_3	20.00	23.00	10.00	40.00	30.00	27.00
Nitrite, mg/kg NO^-_2	NS	1.00	0.91	1.48	0.87	0.84
Sulphate, mg/kg SO^{-2}_4	100.00	0.00	0.00	0.00	15.00	0.00
Total Sulphide, mg/l S^{-2}	NS	17.92	20.48	15.68	18.88	21.76
Iron, mg/kg Fe	400.00	0.84	1.13	1.32	0.81	0.72
Total Bacteria count, Cfu/g	0-30	$1.7x10^8$	$7.0x10^6$	$6.0x10^6$	$4.5x10^6$	$6.0x10^4$
Total Coliform count, cfu/g	0-10	$4.0x10^4$	$9.0x10^3$	$7.05x10^5$	$6.0x10^3$	$8.0x10^3$
Total Faecal Coliform Count, cfu/g	0	$2.0x10^2$	$7.0x10^2$	$2.0x10^2$	$2.0x10^3$	$1.4x10^4$
Total Fungal Count, cfu/g	0	$1.6x10^4$	$1.4x10^4$	$3.0x10^3$	$9.0x10^3$	$1.4x10^3$

mg/l- milligram per litre; ND- None Detected; µS/cm- microSiemens per centimeter, NS- Not Stated; cfu/100ml- colony forming per 100 millilitre; NG- No Growth

Table 3.5. Biochemical Identification of Bacteria Isolates in Sediment Across Oyibo River during Raining Season.

SAMPLING LOCATIONS	COLONY MORPHOLOGY	MICROSCOPIC CHARACTERISTICS	GRAM STAIN	SLANT	BUTT	GLUCOSE	LACTOSE	GAS	H_2S	CITRATE	CATALASE	MOTILITY	OXIDASE	V.P	METYL RED	MOST PROBABLE ORGANISM
OYIBO-NZEREM	Creamy colony on Nutrient agar	Rod	+	B	A	+	-	-	-	-	+	+	+	+	-	*Bacillus Spp*
UMUEZE – OWERE	Creamy colony on nutrient agar	Rod	-	B	A	+	-	-	-	+	+	-	-	-	+	*Shigella spp*
UMUEZAMA	Shiny creamy colony on Nutrient agar	Rod	+	B	A	+	-	-	-	-	+	+	-	-	+	*Salmonella spp*
UMUWOSHA	Mucoidal creamy colony on Nutrient agar	Rod Cocci	- +	A B	A A	+ +	+ -	+ -	+ -	+ +	+ -	+ +	- -	- +	+ -	*E.coli spp* *Enterococcus spp*
UMUOGWARA	Shiny creamy colony on Nutrient agar	Cocci Rod	- -	A B	A A	+ +	+ -	+ -	+ -	- -	+ +	+ +	- -	- +	+ -	*Moraxella Spp* *Enterobacter spp*

A = Acidic condition; **B** = Basic condition; **+** = positive; **-** = negative

Table 3.6. The Distribution of The Fungi Species in Water Samples from Oyibo River During Raining Season.

S/N	LOCATION	MOST PROBABLE FUNGI	OCCURRENCE	TCFU/ml
	SURFACE WATER			
1	OYIB – NZEREM	Asperigillus fumigatus	2	$2.0X10^3$
2	UMUEZE-OWERE	Asperigillus niger Penicillum spp Drechslera spp	1 2 1	$4.0X10^3$
3	UMEZEAMA	Candida spp Penicillum spp	1 3	$4.0X10^3$
4	UMUWOSHA VILLAGE	Asperigillus niger Asperigillus fumigatus	3 4	$7.0x10^3$
5	UMUOGWARA VILLAGE	Paecilomyces spp	3	$3X10^3$
	SENDIMENT			TCFU/g
1	OYIB – NZEREM	Candida spp Asperigillus niger	13 3	$1.6X10^4$
2	UMUEZE-OWERE	Candida spp Drechslera spp	12 2	$1.4X10^4$
3	UMEZEAMA	Candda spp Drechslera spp	2 1	$3.0X10^3$
4	UMUWOSHA VILLAGE	Candida spp Drechsler spp Penicillum Spp	6 1 2	$9.0X10^3$
5	UMUOGWARA VILLAGE	Candida spp	14	$1.4X10^3$

Microbial Properties of water samples from Oyibo River during dry season

From Table 3.8, the values of Total Bacteria Count (TBC), Total Coliform Count (TCC) and Total Fecal Coliform (TFC) across the five sampling points of Oyibo River were above the FMEnv limits. No Fungi was isolated at the five points. TBC ranged from $1.1x10^5$±0.00 (SWQ3) to $2.5x10^5$±0.00 cfu/ml (SWQ2), TCC ranged between $2.0x10^4$±0.00 (SWQ3) and $9.5x10^4$±0.00 cfu/ml **(**SWQ1). TFC ranged between $2.0x10^3$±0.0 SWQ1, 3 and $5.0x10^3$±0.00 cfu/ml (SWQ4). There was no significant difference (P≤0.05) in Oyibo water sampled points. Observed bacteria isolates were: *Streptococcus* spp, *Klebsiella* spp, *Yersinia* spp, *Vibrio* spp, *Bacillus* spp, *Yersinia* spp, *Pseudomonas* spp, *Vibrio* spp and *Citrobacter* spp with their distributions shown in Table 3.9.

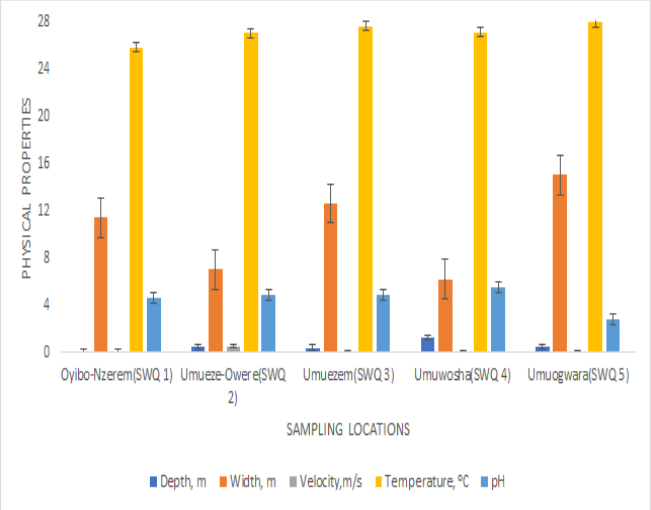

Fig 3.3. Physical Properties of Oyibo River at Different Points During Dry Season.

TABLE 3.7. Physicochemical Properties of water samples from Oyibo River during dry season

PAlRAMETERS	FMEnv Standard	Mean ± STDEV Oyibo-Nzerem, SWQ1 N05040.414' E007018.839' Elevation: 175.6ft	Mean ± STDEV Umueze-Owere, SWQ2 N05039.813' E007017.152' Elevation: 173.8ft	Mean ± STDEV Umuezeama, SWQ3 N05040.065' E007016.887' Elevation: 329.3ft	Mean ± STDEV Umuwosha, SWQ4 N05^{0}40.209' E007^{0}16.805' Elevation: 318.5ft	Mean ± STDEV Umuogwara SWQ5 N05040.468' E007016.866' Elevation: 358.1ft
Colour, PCU	15.00	129.50±9.19	63.00±49.00	68.00±0.00	86.50±0.71	10.5±3.54
Appearance	Clear	Slightly brownish	Slightly turbid	Slightly turbid	Slightly Turbid	Clear
Odour	Unobjectionable	Objectionable	Objectionable	Objectionable	Objectionable	Unobjectionable
Conductivity, µS/cm	1000.00	20.00±0.00	20.00±0.00	20.00±0.00	24.00±0.00	29±0.00
Dipsolved Oxygen, mg/l O_2	>7.50	14.10±0.00	12.00±0.00	9.00±0.00	8.00±0.00	10.8±0.00
Biochemical Oxygen Demand, mg/l BOD5	NS	13.0±0.00	11.00±0.00	8.30±0.00	7.90±0.00	9.4±0.00
Chemical Oxygen Demand, mg/l O_2	NS	100.8±6.79	244.50±6.36	168.00±6.79	153.60±27.15	86.4±13.57
Turbidity, NTU	10.00	59.05±0.00	47.60±0.00	31.50±0.00	59.20±0.00	28.6±0.00
Total Solid, mg/l	500.00-1000.00	72.0±7.07	50.00±0.00	38.00±2.83	54.00±1.41	49.5±4.95
Total Dissolved Solid, mg/l	500.00	13.0±0 .00	13.00±0.00	13.00±0.00	15.60±0.00	18.85±0.00
Total Suspended Solid, mg/l	<10.00	59.0±7.07	37±0.00	25.00±2.83	38.40±1.41	30.65±4.95
Total chloride, mg/l Cl-	250.00	1.395±0.43	1.31±0.00	1.75±0.30	1.75±0.30	2.40±0.00
Total Hardness, mg/l Ca/$MgCO_3$	200.00	3.81±0.00	6.19±0.00	10.95±0.00	3.81±0.00	5.00±0.00
Nitrate, mg/l $N0^-_3$	50.00	2.16±0.38	2.635±0.00	5.26±0.92	11.15±1.43	18.985±0.09
Nitrite, mg/l $N0_2$	0.30	0.56±0.02	0.145±0.00	0.27±0.04	0.15±0.00	0.165±0.01
Phosphate, mg/l $PO-^3_4$	5.00	0.26±0.01	0.325±0.00	0.29±0.28	0.49±0.01	0.27±0.00
Total Alkalinity, mg/l HCO_3 & $C0_3$	150.00	2.00±0.00	3.00±0.00	2.00±0.00	2.5±0.71	2.5±0.71
Bicarbonate, mg/l HCO_3	30.00	2.00±0.00	3.00±0.00	2.00±0.00	2.5±0.71	2.5±0.71
Sulphate, mg/l SO^{-2}_4	200-400.00	26.85±4.22	28.42±0.00	34.22±0.25	30.45±0.63	22.28±0.75
Total Sulphide, mg/l S^-_2	NS	31.36±0.91	30.4±0.45	24.32±0.91	32.32±1.36	18.74±21.47

mg/l- milligram per litre; ND- None Detected; µS/cm- microSiemens per centimeter, NS- Not Stated;

Table 3.8. Microbial Properties of water samples from Oyibo River during dry season

PARAMETERS	FMEnv Standard	Mean ± STDEV Oyibo-Nzerem , SWQ 1 N05^{0}40.414' E007^{0}18.839' Elevation: 175.6ft	Mean ± STDEV Umueze-Owere, SWQ2 N05^{0}39.813' E007^{0}17.152' Elevation: 173.8ft	Mean ± STDEV Umuezeama, SWQ3 N05040.065' E007016.887' Elevation: 329.3ft	Mean ± STDEV Umuwosha,SWQ4 N05^{0}40.209' E007^{0}16.805' Elevation: 318.5ft	Mean ± STDEV Umuogwara SWQ5 N05040.468' E007016.866' Elevation: 358.1ft
Total Bacteria count, Cfu/ml	0-30	$1.25x10^5$±0.00	$2.5x10^5$±0.00	$1.1x10^5$±0.00	$1.2x10^5$±0.00	$1.7x10^5$±0.00
Total Coliform count, cfu/ml	0-10	$9.5x10^4$±0.00	$1.8x10^5$±0.00	$2.0x10^4$±0.00	$5.0x10^4$±0.00	$1.0x10^5$±0.00
Total Faecal Coliform Count, cfu/ml	0	NG	NG	NG	NG	NG
Total Fungi Count, cfu/ml	0	$2.0x10^3$±0.0	$4x10^3$±0.00	$2.0x10^3$±0.00	$5.0x10^3$±0.00	$3.0x10^3$±0.00

cfu/100ml- colony forming per 100 millilitre

Table 3.9. Biochemical Identification of Bacteria Isolated from Oyibo River Samples.

SAMPLING LOCATIONS	COLONY MORPHOLOGY	MICROSCOPIC CHARACTERISTICS	GRAM STAIN	SLANT	BUTT	INDOLE	GLUCOSE	LACTOSE	GAS	H2S	CITRATE	CATALASE	MOTILITY	OXIDASE	V.P	METYL RED	MOST PROBABLE ORGANISM
OYIBO- NZEREM	Creamy colony on Nutrient agar	Cocci	+	A	A	-	+	+	-	-	+	+	+	-	+	-	*Streptococcus* spp
		Rod	-	A	A	-	+	+	-	-	+	+	-	-	+	-	*Klebsiella* spp
UMUEZE – OWERE	Creamy colony on nutrient agar	Rod	-	B	A	-	+	-	-	-	-	+	+	-	+	+	*Yersinia spp*
UMUEZAMA	Shiny creamy colony on Nutrient agar	Rod	-	B	A	-	+	-	-	-	-	+	+	+	+	-	*Vibrio* spp
UMUWOSHA	Mucoidal creamy colony on Nutrient agar	Rod	+	B	A	-	+	-	-	-	-	+	+	-	+	-	*Bacillus* spp
		Rod	-	B	A B	-	+	-	-	-	-	+	+	-	+	+	*Yersinia* spp
		Rod	-	B		-	-	-	-	-	-	+		-		-	*Pseudomonas* spp
UMUOGWARA	Shiny creamy colony on Nutrient agar	Rod Rod	-	B A	A A	-	+	-	-	-	+	+	+	+	-	+	*Vibrio* spp
			-			-	+	+	-	+	+	+	+	-		-	*Citrobacter* spp

A = Acidic condition; **B** = Basic condition; **+** = positive; **-** = negative

Properties of sediments from Oyibo river samples during dry season

Table 3.10 shows properties of sediments from Oyibo river samples during dry season. pH of the sediment ranged between 4.64 (slightly acidic) at location SWQ5 and 9.10 (Alkaline) at location SWQ4. Conductivity, total chloride, phosphate and nickel were within the standard limits. For heavy metals, arsenic levels were observed to be within the standard limit while other heavy metals were slightly above standard limits too. TBC, TCC and TFC1 values were above standard limits. TFC was not detected at locations SWQ1 and SWQ3. There is no significant difference ($P \leq 0.05$) observed in the sampled points.

Fungi Species frequently isolated from water and sediment samples of Oyibo River during dry season

Asperigillus niger, *Candida* spp, *Asperigillus fumigatus*, *Penicillum* spp, *Asperigillus fumigatus* and *Paecilomyces* spp were frequently isolated from water samples while *Candida* spp, *Penicillum* Spp, *Dreschela* spp, *Penicillum* Spp and *Asperigillus fumigatus* were found in the sediments and their distibutions shown in Table 3.11

Discussion

Physicochemical Result of water samples

The pH values of Oyibo river at different five locations varied between slight acidly in the raining season and high acidity in the dry season (5.60±0.00 to 7.15±0.05; 2.8±0.00 - 5.50±0.00 respectively). Okereke *et al*. (2015) reported the pH of the river before dredging to range from 6.52 to near neutral during both seasons. This is at variance with the present findings especially during the dry seasons (2.80 to 5.50). Akaminwor & Egwim (2006) attributed the pH of water bodies to the presence of humic acids generated by some dead aquatic life forms affected by anthropogenic activities. The river had more depth in the rain season (1.60 to 2.00m) than in the dry season (0.08 to 0.50). The flow rate during raining season (0.00 to 0.40 m/s) and dry season (0.00 to 0.09 m/s); width (rain, 6.20 to 15.00m; dry, 6.20 to 11.40m) followed the same trend as the depth.

Table 3.10. Properties of Sediments from Oyibo River Samples During Dry Season

PARAMETER	FMEnvStd 2006	SQ1 N05^{0}40.414' E007^{0}18.839' Elevation: 175.6ft Oyigbo-Nzerem	SQ2 N05^{0}39.813' E007^{0}17.152' Elevation: 173.8ft Umueze-Owere	SQ3 N05^{0}40.065' E007^{0}16.887' Elevation: 329.3ft Umuezeama	SQ4 N05^{0}40.209' E007^{0}16.805' Elevation:318.5ft Umuwosha	SQ5 N05^{0}40.468' E007^{0}16.866' Elevation: 358.1ft Umuogwara
Ph	6.50	6.34	5.02	5.67	9.10	4.64
Conductivity, µS/cm	100.00	47.00	10.00	16.00	23..00	7.00
Total chloride, mg/kg Cl^-	250.00	43.60	26.16	104.65	139.53	69.11
Phosphate, mg/kg PO^{-3}	>100.00	0.27	0.47	0.25	0.21	0.124
Nitrate, mg/kg NO^-_3	20.00	13.53	22.70	11.96	7.84	13.38
NitrIte, mg/kg NO^-_2	NS	0.16	0.21	0.24	1.24	0.25
Sulphate, mg/kg SO^{-2}	100.00	614.04	196.49	3628.07	4596.49	63.16
Total Sulphide, mg/l S^{-2}	NS	204.16	210.56	251.52	274.56	51.84
Iron, mg/kg Fe	400.00	0.10	0.23	0.07	0.17	0.01
Total Bacteria count , Cfu/g	0-30	120,000	350,000	40,0000	100,000	200,000
Total Coliform count, cfu/g	0-10	30,000	100,000	190,000	50,000	195,000
Total Faecal Coliform Count, cfu/g	0	NG	75,000	NG	55,000	110,000
Total Fungal Count, cfu/g	0	70,000	70,000	4000	8000	1400

FMEnv. STD – Federal Ministry Environment Standard; **mg/kg**-milligram per kilogram, **NS**- Not Stated; **ND**- None Detected, **µS/cm**- milligram per centimeter; **NG**-No Growth; **% - percentage; BDL**- Below Detection Limit

Table 3.11. Biochemical Identification of Some Bacteria Isolated from Sediments samples from Oyibo River During Dry Season.

ISOLATE CODE	COLONY MORPHOLOGY	MICROSCOPIC CHARACTERISTICS	GRAM STAIN	SLANT	BUTT	GLUCOSE	LACTOSE	GAS	H2S	CITRATE	CATALASE	MOTILITY	OXIDASE	V.P	METYL RED	MOST PROBABLE ORGANISM
OYIBO- NZEREM																*Bacillus* Spp
	Creamy colony on	Rod	+	B	A	+	-	-	-	-	+	+	-	+	-	
	Nutrient agar	Rod		B	A	+	-	-	-		+	+				*Serratia* spp
		Rod	-	B	A	+	-	-	-	+	+	+	-	+	+	*Vibrio* spp
			-							-			+	-	-	
UMUEZE – OWERE	Creamy															
	colony on nutrient agar	Rod	-	B	A	+	-	+	-	+	+	+	-	+	+	*Enterobacter* spp
		Rod	-	A	A	+	+	+	-	+	+	+	-	+	+	*Enterobacter*
		Rod	+	B	A	+	-	-	-	-	+	+	-	+	-	*Bacillus* spp
		Rod	+	B	A	+	-	+	-	+	+	+	-	-	+	*Providencia* spp
UMUEZAMA	Shiny creamy colony on Nutrient agar															
		Rod	+	B	A	+	-	-	-	-	+	+	-	+	-	*Bacillus* spp
		Rod		B	A	+	-	-	-	-	+		-			*Bacillus* spp
		Rod	+	B	A	+	-	-	-	-	+	+	-	+	-	*Shigella* spp
			-									-		-	+	
UMUWOSHA	Mucoidal creamy															
	colony on	Rod	-	B	A	+	-	+	-	+	+	+	-	-	+	*Providencia* spp
	Nutrient agar	Rod	-	B	A	+	-		-		+	+		-		*Vibrio* spp
								-		-			+		-	
UMUOGWARA	Shiny creamy colony on Nutrient agar	Rod Rod	+	B	B A	A +	-	-	-	+	-	+	-	-	-	*Bacillus* spp
			-			+	-	+	+	+	-	+	-	+	+	*Enterobacter* spp

A = Acidic condition; **B** = Basic condition; **+** = positive; **-** = negative

Table 4.12. The Fungi Species isolated from river samples during dry season.

S/N	LOCATION	MOST PROBABLE FUNGI	OCCURRENCE	TCFU/ml
	SURFACE WATER			
1	OYIBO- NZEREM	Asperigillus niger	2	$2.0X10^3$
2	UMUEZE – OWERE	Candida spp Asperigillus fumigatus	3 1	$4.0X10^3$
3	UMUEZAMA	Penicillum spp	2	$2.0X10^3$
4	UMUWOSHA	Asperigillus fumigatus	5	$5.0x10^3$
5	UMUOGWARA	Paecilomyces spp	1	$3X10^3$
	SENDIMENT			TCFU/g
1	OYIBO- NZEREM	Candida spp Penicillum Spp	2 5	$7.0X10^4$
2	UMUEZE – OWERE	Candida spp	7	$7.0X10^4$
3	UMUEZAMA	Dreschela spp Penicillum Spp	3 1	$4.0X10^3$
4	UMUWOSHA	Candida spp Asperigillus fumigatus	7 1	$8.0X10^3$
5	UMUOGWARA	Candida spp Dreschela spp	4 10	$1.4X10^3$

The observable difference in the depth, width and flow rate in the dry season can be attributed to the non-replenishment from inflow of other water sources (rain water, wash-offs). The colour of the Oyibo River was above the permissible limits except in Umuogwara (SWQ5) where the water sample was clear in both seasons, this can be attributed to the fact that SWQ5 is the water source (upstream). The source is often cleared of weeds and planktons by the villagers. This is in consonance with the work of Nayla (2019) who stated that the colour and appearance of river from its source is often within the acceptable range since there is no much contamination from both organic and inorganic matters. The total solids values are lower than the FMEn permissible limit. The effects of consuming water with high solids has been reported by Akubugwo *et al.* (2013). Total dissolved solids (TDS) and total suspended solids (TSS) is an indication of materials carried in suspension and solid respectively (Verla, Verla & Amaobi, 2018). TDS and TSS were above the limits during the raining season, this is an indication of pollution from runoff of soils around the area. This may be attributed to the release of nutrients as a result of re-suspension of sediments during dredging operations (Seiyaboh *et al*., 2013). During the dry season, TDS and TSS values revealed a reduction in pollution due runoffs. Conductivity was observed to be within the limits. Conductivity is a measure of total dissolved salts and greatly affect the taste of water (Verla, Verla & Amaobi, 2018). Dissolved Oxygen (DO) did not meet the permissible limit in the raining season but met the FMEn permissible limit during the dry season (1.25±0.05-6.05±0.05; 8.00±0.00-14.10±0.00 respectively). According to Garg *et al.* (2010), dissolved oxygen concentration of more than 5.00mg/l support aquatic life, this is evidenced by the presence of fish in some of the river points. The low levels of DO during the raining season may be attributed to pollution from flooding and other anthropogenic activities such as wash offs from farm lands, household wastes etc. This agrees with the findings of Seiyaboh *et al*., (2019) who observed low levels of DO in wet season and its higher levels in dry season while working on dredged Igbedi Creek, Upper Nun River in Niger Delta. Biochemical Oxygen Demand (BOD) levels were lower than their respective DO. This could be indication that the oxygen demand generally did not exceed the oxygen production and aeration rate for each sampled point in both seasons. According to Moore & Moore (1976) as reported by Joseph *et al*., (2019) category of water based on BOD levels, the dredged Oyibo River can be said to fairly clean since its BOD levels is lower than the DO levels. This is at variance with the work of Elijah *et al*., (2008) who observed higher BOD levels in dredged river in Niger Delta. In both seasons, turbidity and total hardness values showed that although the river samples from the five points were turbid, it was not a hard water. This is in tandem with the findings of Iwuoha (2011) who stated that dredging activity increases river turbidity but not necessarily hardness. The minerals found to be present in this water sample (nitrate, nitrite, phosphate, bicarbonate, sulphate, sulphide) may be linked to the weathering of bedrocks, deposition of dust and salt by winds, by the natural leaching of organic matter and nutrients from soil, by hydrological factors that lead to runoff, and by biological processes within the aquatic environment (Akaishi *et al*., 2004). These processes can alter the physical and chemical composition of water (Akaishi *et al*., 2004).

Microbial properties of water samples

In both seasons, high microbial values that were above FMEnv permissible limits indicated high organic matter in the river water. Microbial presence in a water body is an index of biological pollution. *Bacillus* spp, *Staphylococci* spp, *Enterococcus* spp, *Psendomonas* spp, *Micrococcus* spp, *Salmonella* spp and *Enterobacter* spp were the probable organisms observed at the different locations of the river during the raining season while *Streptococcus* spp, *Klebsiella* spp, *Yersinia* spp, *Vibrio* spp, *Bacillus* spp, *Yersinia* spp, *Pseudomonas* spp, *Vibrio* spp and *Citrobacter* spp were observed to be present at the different locations during the dry season . These isolated organisms have been implicated as agents of different diseases (Anudike, Duru & Uhegbu, 2019). In the rain season, Total Bacteria Count (TBC), Total Coliform Count (TCC), Total Fecal Coliform Count (TFCC) were observed to be higher in the upstream (SWQ5). TFC which only had growth for SQW 4 and SQW5 can be attributed to bird droppings (birds usually come to the water source to drink water) as no TFC growth was observed in the water samples during the dry season. Statistically there was no difference ($P \leq 0.05$) in the microbial load during raining and dry seasons. Before dredging, the average values of total heterotrophic bacteria were 5.8×10^3 cfu/ml and 1.51×10^3 cfu/ml for rainy and dry seasons respectively (Okereke *et al*., 2015); Bacterial isolates included *E. coli*, *Salmonella* spp, *Lactobacilli* spp, *Klebsiella* spp, *Staphylococcus* spp, *Proteus* spp, *and Pseudomonas* spp. Generally, the high microbial values from this present study can be attribute to surge in

various human, agricultural and industrial activities. Rivers near populated areas receive surface runoffs from agricultural and pasture land, animal wastes and effluent runoff discharges from the industrial, and plantation are faced with possible risk of bacterial contamination (Sui *et al*., 2018). Temperature is another factor that could influence the levels of pathogenic organisms in surface water. Bacteria grow faster at higher temperature than at lower temperature. High levels of nutrient also influence the growth rate of bacteria (Edokpayi, 2018). This is in line with the findings of Victor *et al*., (2019) who carried out microbial assay of dredged Otamiri River and its sediments in parts of Owerri, Imo State.

Sediments result of water samples

Sediments are important components of the aquatic ecosystem and serves as habitant and or spawning/ breeding grounds for a wide range of aquatic organisms, usually referred to as benthic organisms, the maintenance of its health and by extension that of the organism it supports is important (Wokoma & Friday, 2017). Many toxicants such as heavy metals and organic contaminants tend to settle on sediments which serves as a sink (Seiyaboh *et al*., 2013). In rain season, pH of the sediments at the various points were observed to be more acidic ranging from 3.20 to 4.80. Similar trend was also reported by Okechi & Chukwura (2020). The physicochemical parameters analyzed (pH, conductivity, total chloride, phosphate, nitrate, nitrite, sulphate and total sulphide) were within the FMEnv permissible limits. TBC, TCC, TFC and Total Fungi Count (TFC1) of the sediments which are above the permissible limits were also above that of the sampled surface water from the river. *Bacillus* spp, *Shigella* spp, *Salmonella* spp, *E.coli* spp, *Enterococcus* spp, *Moraxella* spp, *and Enterobacter* spp were observed to grow in the cultured samples in the raining season in exception to *Moraxella* which was only observed in the sediments. In the dry season, pH of the sediment ranged between 4.64 (slightly acidic) in SWQ5 and 9.10 (Alkaline) in SWQ4. This agrees with the work of Seiyaboh *et al*., (2013) who stated that the pH of dredged river sediments is mostly acidic which can adversely impact the fisheries distribution. Conductivity, total chloride, phosphate and nickel were within the standard limits. There is no significant difference in the sampled locations. *Bacillus* spp, *Serratia* spp, *Vibrio* spp, *Enterobacter* spp, *Bacillus* spp, *Providencia* spp, *Shigella* spp, were organisms observed to be present in the water sediment samples. This is in consonance with findings of Victor et al., (2019) who observed most of these microbes in dredged Otamiri sediments.

The Fungi Species of sampled river surface water and its sediments

During raining season, *Asperigillus fumigatus, Asperigillus niger, Penicillum spp, Drechslera spp, Candida* spp, *Penicillum* spp, *Asperigillus niger, Asperigillus fumigatus* and *Paecilomyces spp*, were the fungi that were observed in the surface water while *Candida spp, Asperigillus niger, Drechslera* spp, *Drechslera* spp and *Penicillum* spp were observed to be present in the sediments. In the dry season, *Asperigillus niger, Candida* spp, *Asperigillus fumigatus, Penicillum* spp, *Asperigillus fumigatus and Paecilomyces* spp were the fungi that were observed in the surface water while *Candida* spp, *Penicillum* spp, *Dreschela* spp, *Penicillum* spp *and Asperigillus fumigatus.* These fungi observed in the water surface tend to settle to the water beds (sediments) as water flows as most of the surface water fungi were also observed in the sediments. This agrees with the observations of Adieze *et al*., (2016) who noted that most fungi tend to settle at bottom of the water where there are relatively more nutrients.

4.0 Conclusion

This study shows that the dredging of Oyibo river in Ehime Mbano Local Government Area of Imo State triggered some physicochemical and microbial changes in both wet and dry seasons. The river was acidic and some of the parameters were not within the federal Ministry of Environment (FMEnv.) acceptable limits, hence, the River cannot serve as good source of water supply to the rural populace that depend on it.

Recommendations

- Simple Water treatment procedures such as boiling with filtration should be applied for drinking and domestic uses.
- Local population that depends this river should be properly enlightened on the need to purify water from the river before using it for drinking and other domestic purpose.

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