The Impact of Accumulated Waste on the Soil and Underground Water in Bayelsa (Otuokpoti) Region of Nigeria

Abaraogu J. U., Uzoukwu C. S., Anyanwu V.K., Eziefula U. G., Ikpa P. N., Emejulu D. A., Abbah E. C., Nwokorobia G. C., Ogbonna S. N., Osukalu E. J,

ABSTRACT

The study deal on the negatives effects of wastes disposal on the groundwater pollution in different locations of a southern state in Nigeria. Water samples were collected from shallow wells located near the selected dumpsites in the selected areas. The analysis of physical, chemical and biological properties of the raw water samples from these locations shows that these wastes produce Leachates and gases when they are decomposed and percolated or/and infiltrated through rainwater into the groundwater. However, from the physio-chemical results, some of the sample water parameters tested fall within acceptable range when compared to the drinking water quality standard: NSDWQ (2015), these includes parameter like PH, Chromium, Nitrite, Lead, Ammonium, Iron and Chloride. It is therefore recommended that efforts should be made to improve the waste management system of these locations and Ogbia community in general; also, there should be some level of treatment on the waters around these locations and similar locations around the Ogbia Community to prevent any form of future outbreak/disease that may arise from the consumption of these contaminated waters. **Keywords:**Leachate, LesiccatorsVibriocount, Total heterotrophic bacteria count, Total coliform count Salmonella and Shigella count.

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I. BACKGROUND OF STUDY

Municipal Solid Wastes consists of refuse from households, hazardous refusefrom industrial, commercial and institutional establishments (including hospitals), market waste, yard waste, and street sweepings (Ogwueleka,2009). Refusedumps constitute integral parts of the soil hydrological system and pose a serious pollution threat to both groundwater and downstream surface water (Dahlin et al., 2010). In a refusedump, high concentrations of materials such as heavy metals, nutrients, and organic substances lead to a risk of pollution of the surrounding environment. The pollutant load to the environment depends on the quantity and quality of the water that percolates through the waste dump into the surroundings. According to Christensen et al. (1992), the major local environmental problem of refusedump is the discharge of leachates into surrounding ground and surface waters. Indeed, the leakage from municipal refusedeposits is usually associated with high ion concentrations and hence very low resistivities. This makes geoelectrical imaging techniques particularly interesting for mapping the three-dimensional extent of contamination around landfills (Bernstone and Dahlin, 1999).

In Nigeria and other developing countries, waste disposal management has become a problem (Agunwamba, 1998; Ogwueleka, 2009). Large refuse dumpsites that need remediation are seen surrounded by residential quarters in our urban cities. In the majority of these dumpsites the contaminants are heavy metals. However, the levels of contamination are often unknown (Ogwueleka, 2009). Pollution of groundwater under and near waste disposal sites happens when rain falls and water passes through the waste dump, producing a waste fluid called leachate which can infiltrate across the unsaturated zone into the water table. Every conceivable inorganic and organic material may be present in the leachate that can degrade the groundwater quality thereby putting the local community under serious health risk.

PROBLEM STATEMENT

The problem of solid wastes and its effect on the quality of underground water is now the focus of attention all over the world. The problem of waste generation in the country in the recent past had been quite tremendous, necessitating the need for this project. In fact, the presence of wastes scattered all over cities and towns and even in our institutions of higher learning is a threat to health and embarrassment to the pleasant

standard expected of these areas. The menace of refuse generation is most prominent in developed and industrialized countries; this is because complexity of generated waste is directly proportional to development in science and technology. Construction and operation of land filling material, recovery system, and incineration systems have become very costly. It is noteworthy that the urban populace generates more wastes than the rural areas. This is because of the various forms of commercial and industrial activities going on in cities and which have brought an increase in the volume and diversity of waste generated daily in the country. These have some great effects on underground water consumption (borehole and well). It is therefore believed that in consumption of underground water such as bore hole and well water, certain tests are needed to be carried out before consumption in regulation with the principles of World Health Organization. These tests are:

pH Test Total coliform test Test for nitrate [No₃⁻] Aerobic and sulphate reducing bacteria⁻

These tests are the most important tests carried out to know the physical chemical and biological effects of solid wastes on underground water before it could be suitable for consumption. In additional, wastes are such items which people are required to discard. It can be defined as unavoidable material resulting from an activity which has no immediate economic demand and must be disposed-off. Items such as household rubbish, sewage sludge, waste from manufacturing can give to a large variety of different wastes from different sources. Wastes are generated every day in all parts of the world. This mainly made up of waste coming from household, commercial activities (e.g. shops, restaurant, hospital), industry (e.g. pharmaceutical companies, clothes manufacturers etc.), agriculture (slurry) construction and demolition projects, mining and quarrying activities and from the generation of energy. With such vast quantities of wastes being produced, it is of vital importance that it is managed in such a way that does not cause any harm to either human health or to the environment.

Study Area

In Bayelsa State, South-South of Nigeria with eight local governments, my focus area was limited to just one of the major local government areas in Bayelsa state which is Ogbia local government of Bayelsa state. Some locations around the communities in Ogbia local government area were considered.Ogbia is a Local Government Area of Bayelsa State in the Niger Delta region of Nigeria. Its headquarters is in the town of Ogbia in the south of the area at 4°39′00″N 6°16′00″E.It has an area of 695 km² and a population of 179,926. It is well known for its historic value to the today Nigerian state economy mainstay, i.e., its oil industry, being the local government area encompassing Oloibiri the first-place oil was discovered on Sunday 15 January 1956. In this town different wells were examined especially those that are very close to the

Region and Area

Longitudinally, the study area is located between longitudes 6" 28" East of the Greenwich meridian. It is bisected by Latitude 4°15" and 5°08 North of the equator. The study area is within the Niger Delta Region of Nigeria, specifically in Ogbia Local Government Area of Bayelsa State. The study area is bounded by Nembe Local Government Area at the southern Ijaw Local Government Area at the south-west and lastly, Yenagoa Local Government Area at the North.

Geomorphology/ Geology in the Area

The study area is part of the landscape of the Niger Delta Environment, South of Nigeria. According to Fubara (1987), the limit of Nigerian coast is within 30 to 40 km from the coastline inland. This delimitation appears to be realistic since the tidal influences are felt up to 45km inland (Oyegun, 1990). Based on that, the study area falls within the coastline and the inland alluvial plains of the coastal lowland of the Niger Delta. Akpokodje (1987, 1988) asserted that the area under study is about 8 meters above mid sea level (A.M.S.L)

Structurally, the study area is dominated by low-lying plain, belongs to the sedimentary environment of the modern Niger Delta. The land slopes gently from North- South direction to the boundary between Ogbia and Nembe Local Government Areas. The sedimentary sequence underlying the area constitutes the body of deltaic deposits laid down 50-70 million years ago. Since the past two million years, the Delta has changed in response to the constantly rising and falling of sea level as a result of climatic conditions.

The present topographic configuration has evolved from the sedimentary patterns of the last 75,000 years ago (Izeogu and Aisuebeogun, 1989). The geology of Ogbia is dated back to the Eocene era. This was accompanied by the built-up of fine sediments, which were ended by the River Niger and Ekole Creek.

The materials were transported and re-deposited to form the geomorphologic unit of Ogbia. The surface geology of the Niger Delta is made up of three tertiary lithostratigraphic units, Benin, Agbada and Akata formations (Akpokdje, 1987 and 1988).

Lithostratigraphically, the Benin formation consists of over 90% of sand and shale intercalations. It covers the whole coastline of Bayelsa state. The materials of the Benin Formation dates back to the Oligocene epoch, which the upper part of the formation generally ranges from Miocene to the recent age (Assez, 1970).

The Agada formation consists of a sequence of shale, which sand unit predominates the upper part and with minor shale interactions. The shale unit becomes thicker toward the boundary between it and the Akata formation. The Akata formation is made up of uniform shale deposits consisting of dark grey sandy, silt shales with plant remains at the top of the formation. Thin lenses are also known to occur near the top of this formation, particularly at the zone between Agada and Akata formations. Various types of quaternary to recent deposits including the Chicoco mud overlie the three units of formation (Akpokodji, 1988).

The sub-arial Niger Delta is made up of recent deposits. The quaternary deposits consist of either relatively uniform lithology or alternating sequence of sand, silt and clay- peat or sand-silt-clay mixture with clay and silt increasing seawards (Akpokodje, 1987). The oldest among these formations is the Akata formation.

Vegetation Type and Climatic Conditions

The Ogbia kingdom is dominated by two major soils - the fresh water/ salt water transition soil and the coastal plain terrace soils. Among these are other smaller soil units which also occur. These soils occurring in patches are likened to the soils of the high lying levees. Soils of the low- lying levees, meander belt soils, soils of the basins, silted river bed soils and soil of the transitional zone (Ogbia master plan ibid).

The study area lies in the rainy belt of the Niger Delta. It is hot and wet throughout the year. There are two types of air masses- the tropical maritime and the tropical continental air masses dominating the climatic events in the area. The former is associated with the south-west trade wind blowing from the Atlantic Ocean onshore. While the tropical continental air mass is usually associated with the cool dry and dusty Harmattan wind (Udo, 1981).

The study area is crisscrossed by two major vegetation zones. Broadly, the vegetation of the study is the fresh water swamp forest type with transitional zone of the Brackish and swamp forest (mangrove Swamp Forest) to the south and the fresh water swamp forest is characterized by raffia palms to the north (Udo, 1980; Oguntoyinbo et al. 1983 and Nyananya, 1999).

Drainage of System of the Area

The entire geographical space of Ogbia is characterized by a maze of rivers, creeks as well as swamps crisscrossing the low lying plain. The area is drained by large and medium to small channels, rills, rivulets and steams of high tides (Ogbia Master Plan, 1973). The major drainage artery is the Ekole River flowing from the River Nun through Yenagoa, entering the northern part of Ogbia from Otuokpoti down to the south where it links Nembe and Southern Ijaw. The river plain provides sites for all the settlements in Anyama District of Ogbia. The river is characterized by meanders, thereby resulting to the problems of river bank erosion and deposition. It is linked at the south with the tributary of Owubiyo anal to join the Otuaka (Otuoke) river leading to Ogbia town, Abobiri (Otuabo) off Oloibiri via Nembe.

The Otuoke River links the Kolo creek close to Ogbia town. The Kolo creek is the major drainage connecting all the communities in Emeyal-Kolo group and some of the settlements in Oloibiri group. The Oloibiri river, Emakalakala canal and the Otuoke rivers are influenced by the tidal movement of the sea water and for most of the dry season, these rivers experience brackish water (Ogbia Master Plan, 1975)

The major characteristic of these rivers and creeks is that they all flow in north- south direction emptying their water into the Atlantic Ocean. The exception to this rule is the Otuoke River, Oloibiri River and Emakalakala canal where the changes its flow direction in every six hours. The water is normally clear in the dry season but becomes muddy and cloudy during the rainy and the flood seasons. All these rivers have a mean depth often in excess of six meters (6m), sinvosity ratio exceeds 1.9, while velocity increases in downstream direction (Izeogu and Aisuobogun, 1989). They also affirmed that because of the dense vegetal cover of the area, chemical weathering predominates other forms of weathering. This is clearly shown by the concentration of minerals and the colour of the water.

Sources of Solid Wastes

Wastes are produced at the source. The quality and composition of refuse are determined by the features or the source, these sources are:

Domestic/Residential Solid Wastes: This category includes rejected solid materials that emanated from shop owners and house-hold unit.

Garbage: This result from marketing preparation and consumption in relation to ft residential units, it contains organic materials. It usually produces a very bad odor.

Rubbish / Trash: this category consists of paper and product plastics cans, bottles, glass, metals, ceramic dirt, dust and garden waste. They are mainly non-putrescible.

Ashes: this type of waste is the residue from any contribution process e.g. fire woodetc, resulting from house hold activities.

Physical and Chemical Hazard of Solid Wastes

A problem frequently overlooked is that of physical and chemical hazards that can result from certain components for example, Explosion hazard of solvent, gasoline etc. Toxic chemical used as pesticides solvent etc.Direct contact infected cuts, abrasions resulting from contact with broken glasses and sharp object, heavily contaminated with bacteria.

Sources of Water:

Water is a liquid material comprises of two elements, Hydrogen and Oxygen. It is the most important components in the universe and it is required in all chemical reactions.

Surface Water: This occurs as the result from the precipitation that is not absorbed by the ground or evaporated back into the atmosphere. Surface water becomes surface run-off as it flows into stream, lake and wet lands, reservoirs etc.

Ground Water: This results from the precipitations that infiltrate into the ground and seep downward through fractures, pores and other spaces in the soils and rocks Sources of water include rainwater, spring, river and lakes, surface wells, deep or artesian wells and tap water.

Rain Water: This takes up dusts and gases from the air and organic matters from the roofs over which it is collected. The long storing cistern gives bacteria opportunity to grow in large numbers, causing the water to be unsafe for drinking.

Spring Water: This water is a source of pure water supply, if is not contaminated by passing through soil which is polluted.

River and Lakes: These are common sources of water supply but they may be made unfit for drinking, if the surface water and sewage from towns and cities are allowed to drain into them.

Surface Wells: These are unsafe sources of drinking water supply, and the water should never be used when there are cesspools, drains, barnyards or any other sources of contamination with a radius of 60m of them.

Deep Artesian Well: These furnish pure water as a rule, unless the piping is not tightly jointed, when impure water from a subsoil stream near the surface may enters the pipes.

Tap Water: This is a drinking water which comes from an indoor tap or spigot. It is part of a larger indoor pumping system which requires a complex supportive infrastructure including stable water sources piping and filtration to keep the water safe. It is the savviest source of drinking. The germs present in the water are being killed through tests (e.g. biological, chemical and physical tests) before it is being consumed by each individual.

Materials

II. MATERIALS AND METHODS

The apparatus employed in the experiments of this project work include, burettes, pipettes, (1ml, 10ml, 25ml), mercury-in-glass centigrade thermometer, pH-meter (Hach model), water checker, conical flasks, beakers, white polyethylene bottles, steam bath, oven, desiccator, mettle balance, hot plate, flame emission photometer (FEP), Atomic absorption spectrophotometer (AAS) unicam 8625 UV\VIS spectrometer and so on.

Reagents: 0.02MAgNO3, 0.01M EDTA. CO2 free NaOH, 0.02MHCl, NH3\NH4Cl buffer, MnCl2 solution, alkaline iodideazide reagent, 0.05M potassium hydrogen phthalate, 0.0125MNa2S2O3 5HO2 starch solution, Erichrome Black T indicator, Brucine sulphate, stock nitrate solution, stock phosphate vanadate-molybdate reagent, gelatin-BaCl reagent, stock phosphate, carbon tetrachloride, concentrated (Conc) HCl, H2SO4, HNO3 Analar grades from British Drug House (BDH), etc.

III. Methods

In order to achieve the stated objectives, several procedures were taken in order to arrive at an economic but modern method of waste collection, waste disposal, waste processing and siting of wells at reasonable distances in the study areas by putting economics and engineering factors into consideration. Two methods includes (i) Physical and personal inspection and (ii) Water sampling and separation analysis

Step1:Collection of Data

From the primary sources of the information that was gathered, three wells from which the samples were gotten were located at very close proximity to dumpsites in the selected communities within Ogbia Local government area. The three communities were the samples are gotten from are Kolo 1 community, Kolo 3 community and Otuokpoti community all in located within Ogbia local government area of Bayelsa state. According to the people in the area the wells have been dug before the existence of the dumping sites. The constituents of the wastes on the dumpsites in the three locations comprises majorly of domestic and every day household items such as kitchen wastes, polythene papers and bags, plastics, scrap metals, etc. The samples were immediately taken to the laboratory where water quality tests were carried out on the three samples. The Physiochemical and Microbiological Tests were carried out for the water samples

Physiochemical test: Under the Physiochemical aspect of the water analysis, the following tests were carried out by the researcher; Color of the water samples, Odor of the water samples, Turbidity of the water samples, Temperature of the water samples, pH of the water samples, Dissolve Oxygen of the water Samples, Total Dissolve solid of the water Samples, Total hardness of the water Samples, Presence of Calcium (ca) in the water Samples, Presence of Nitrate (NO₃) in the water Samples, Presence of Nitrate (NO₂) in the water Samples and Presence of chloride (cl-) in the water Samples. Etc

Step 2: Sample Analysis

Other quality parameters determined in the laboratory include suspended solids (SS), total dissolved solids (TDS), total solids (TS), dissolved oxygen (DO), biochemical oxygen demand (BOD), alkalinity, acidity, total hardness (TH), chloride (Cl⁻), sulphates (SO⁻⁴) nitrates (NO⁻³) and chemical oxygen demand (COD). The analysis for each parameter was performed in triplicates.

Determination of DO by Winkler's method (Ademoroti, 1996a)

To each water sample contained in the BOD bottles, 2ml conc. HCI was added by allowing the acid to run down the neck of the bottle, stoppered, and mixed by gentle inversion until dissolution was complete making sure the iodine was uniformly distributed throughout the solution. 100ml of the solution was titrated against standard $0.0125MNa_2S_2O_3$, 5H2O to a pale straw yellow color, on the addition of 2-drops of starch indicator the color became blue. This titration was continued by adding the thiosulphate solution drop wise until the blue color disappeared.

Determination of acidity (A.O.A.C. 1984)

i Phenolphthalein acidity (PA): 50ml of each water sample was measured into 250ml conical flasks. Three (3) drops of phenolphthalein indicator were added in each case and the solution titrated with CO_2 free NaOH solution (0.02M) until the appearance of a faint pink color (pH 8.3) was observed. This indicated the end point of the titration.

ii Methyl orange acidity (MOA): To 50ml of waste water and effluent samples, three (3) drops of methyl orange indicator were added and the solution titrated with CO_2 free NaOH solution (0.02M) to the end point. Determination of alkalinity (A.O.A.C., 1984)

i Phenolphalein alkalinity (PAK): 50ml of each water sample was measured into different (250ml) conical flasks Three (3) drops of the phenolphthalein indicator were added. The solution remained colorless indicating a zero PAK.

ii Methyl orange alkalinity (MOAK): To the solution mixture obtained in 3.5.3 (i) above, three drops of methyl orange indicator were added forming yellow color. The solution was titrated to a second end point with 0.02MHCl until the yellow coloration changed to a faint yellow, and to pink.

iii Total alkalinity (TA): The addition of PAk and MOAk expressed as CaCO₃.



<u>Step 3:</u>Alkaline-Azide Modification of Winkler Method: Procedures:

Firstly, Stream water was filled into a 100ml brown bottle. After then, 2cm^3 of Mncl was added followed by 2cm^3 of alkali-iodide (AL-KI) reagent just below the surface of the liquid.In addition of these two solutions ppt was formed and was allowed to settle until a clear supernatant is observed. Fourthly, 2cm^3 of HCl was added to the liquid by allowing the acid to run down the neck of the bottle, this was rest opened, mixed by gentle inversion until dissolution is completed. Then a known volume needed was withdrawn for titrate on with 0.0125M Na²S²0³ to give a pale straw color (pale yellow).and finally 2cm3 of the starch solution was added to give a blue color, titration continued by adding the thiousulphate solution dropwise until the blue-black color disappears.

Solution,

Considering volume of the bottle with stopper in place, V1 Volume of aliquot taken for titration, V2 Volume of thiousulphate needed for titration, V and Molarity of the thiousulphate, M mg/1 DO = $\underline{M \times V \times 16,000}$ (V₁-2) *(V₂/V₁)

If the content of the bottle was titrated all. The formula below Mg/1 DO = (16,000 x M x V)(V₁-2)

Temperature (0°C)

A Celsius thermometer (mercury in glass centigrade thermometer O'C-110°C) was used in taking the temperature. The thermometer was vertically immersed the bulb containing the mercury in the water sample for about five (5) minutes. The reading was taken as the mercury rose to a steady state. The pH was taken at the site. No holding.

Determination of the pH value

A pH meter model 3320 JENWAY (electronic) was initially standardized using buffer solution 4 and 9 respectively to serve as check for proper Instrument response before sampling measurement. The electrode rinsed with distilled water and then inserted in the (water sample. The pH value recorded on the meter scale at a steady point. After each determination, the pH sensing bulb was rinsed with distilled water before next determination.

Turbidity

Step 4: Apparatus: (Water checker U-10 Horiba)

Procedure: Measurement of the turbidity was done using water checker Instrument. However, the water was filled to a standard level cup of the water checker to enhance the reading. The electrode was put into the water and the selector adjusted to turbidity position until a steady value appears indicating the turbidity of the water. 1 mg/l = 1 unit turbidity

Ref C. M. A. Ademoroti (1966) Standard method for water and effluent analysis. Foludex Press Ltd, Ibadan.

Conductivity: Determination of conductivity was done using conductivity meter, model: Suntex sc-120 The electrode was properly rinsed with distilled water before taking the measurement. For every subsequent set of water determination, the electrode must be rinsed at each interval. Data were obtained from the reading. Conductivity in UScm⁻¹

Determination of COD (AO.A.C, 1984): 100ml of each water sample was measured into different conical flasks. 5ml of dilute H_2SO_4 (1:3) was added and the solution quickly transferred to a steam bath to boil. Then 15ml of $0.01MH_2C_2O_4$ was added followed by dropwise addition of KMnO₄ (0.01M) from the burette until the solution turned pink.

Determination of TDS: A clean glass dish was dried at 103°C to 105°C in an oven until constant weight was achieved cooled in desiccators and weighed. 100ml of filtrated water samples were evaporated on a water bath followed by a drying in an oven 103°C-105°C for about an hour. The dish was cooled in desiccators, weighed again and the increase in weight recorded.

Determination of SS: The Whatman filter paper No. 42 was dried to constant weight, cooled to room temperature in desiccators and the weight noted. 100ml of thoroughly mixed water samples were measured and filtered. The filtered residue was dried at a temperature of 103°C- 105°C in an oven for 30-40 minutes. The filter paper and the residue were cooled and weighed.

Determination of TS: The sum of TDS and SS gave TS expressed in mg \L

Determination of Cl (Ademoroti, 1996a): To 100ml of each water sample, three (3) drops of 5% K2CrO4 indicator were added and titrated with a standard $0.02MAgNO_3$ until the color changed from yellow to brick red.

Determination of BOD: The method for the determination of BOD was similar to that for DO, but in the BOD test, the water samples were kept in an incubator in the dark preset at 20°C for 5 days. After incubation, the DO test was repeated.

Determination of NO_3N^- (Ademoroti, 1996a): 10ml of each water sample was transferred into different 25ml standard flasks and 2ml of Brucine reagent was added; then 10ml of conc H_2SO_4 was also added rapidly. It was mixed for about 30 seconds and allowed to stand for 5 minutes. The flasks were set in cold water for about 5 minutes and then made up to volume with deionized water. The absorbance was read at 470nm with Unicam 8626 UV/VIS spectrometer.

<u>Step 5:</u>Microbiological Analysis of Water Samples

Procedure for the Estimation of Bacteria and Fungi in water Samples.

A total of 3 water samples were analyzed. These samples were Kolo 1, Otuokpoti, and kolo3. Precisely, 10ml of each water sample was accurately measured and mixed with 90mls of sterile water in a conical flask. Flasks were labelled accordingly. Prescott 2004.

Serial Dilution of Samples:

Serial dilutions of the samples were carried out according to the method of Cheesbrough (2004). Precisely, 10ml of water were measured and mixed in 90ml of sterile water (aliquot), dilutions were further diluted by transferring 1ml from the aliquot into a sterile 9ml of dilution blank in a test tube, dilutions were further carried out till 10-3 dilution factors were obtained.

Estimation of Microbial Loads in water Samples:

Standard microbiological techniques described by Horrgan and MacCance (1990), Prescott (2004) were employed for the microbiological analysis of these water samples. The total heterotrophic bacteria count (THBC) Coliform counts, Salmonella Shigella count, Vibrio count faecal coliform count staphylococcus count and fungal count were estimated by the Pour plates methods except fungal count that used the spread plates method. These procedures used Nutrient agar (NA), MacConkey agar (MCA), Salmonella Shigella agar (SSA),

Thiosulphate Citrate bile-salt Agar (TCBS), Eosine Methylene Blue Agar (EMB), Mannito Salt Agar (MSA) and Sabouraud Dextrose agar (SDA) as analytical media respectively.

Incubation of culture media and counting of microbial colonies:

The bacterial plates were incubated for 24 hrs at 28[°] C using a Gallenkamp incubator and fungi plates at room temperature for 5-7days. Fungi were incubated at room temperature. Microbial colonies that emerged on the incubated plates were counted with the aid of a Quebee Colony counter and recorded as colony forming unit (CFU/ml) of soil sample.

Purification and Maintenance of Microbial Isolates:

Representatives or discrete colonies from culture plates were picked for characterization. Bacterial colonies were repeatedly sub-cultured into freshly prepared Nutrient agar plates by streaking methods and incubated for growth at 28°C for 24hrs before transferring them to agar slants (Cheesbrough, 2004). The pure isolates of bacteria and fungi were maintained on agar slants as stock and preserved in the refrigerator for further used.

Characterization and Identification of Microbial isolates:

Bacterial isolates were characterized and identified presumptively based on their morphological, cultural and physiological characteristics, confirmatory identification was based on Biochemical reactions. The following Biochemical test were carried out. Gram staining, motility test, coagulase test, catalase test, spore staining test, oxidase test, urease test, citrate test, starch hydrolysis test, methyl red-Voges Proskaures (MR-VP) test and sugar fermentation test (lactose, glucose, mannitol, maltose, galactose, fructose and sucrose). The results derived from the test for various isolates were collected and the identification was carried out by comparing the characteristics with a known taxa using the scheme of Bergy's Manual of Determinative Bacteriology (1994). Fungi isolates were identified according to the method of Bernet and Hunter (1973). The procedure involved the visual examination of isolates in culture (pigmentation, texture, reverse side, colony surface) and by observation of stained preparation under microscope (nature of conidia, hyphae and spores). These were carried out by placing a drop of lactophenol in cotton blue on a clean slide, a small piece of mycelium free medium was picked with the help of a sterile inoculating needle. The mycelium was placed on the sterile and gently teased to avoid distorting of the mycelium using inoculating needle. A cover slide was placed with care to avoid contact of the objective with the stain, it was then viewed under the microscope at x10 and x40 objectives.

Step 6: Procedures for Biochemical Tests

Morphological and Biochemical Characterization of the Bacterial Isolates: Standard characterization tests (such as Gram staining, catalyses, coagulate, motility, starch hydrolysis, methyl-red Vogue Proskauer, indole, citrate utilization, urease, spore staining, hydrogen sulfide production and sugar fermentation) were performed. The pure culture was identified on the basis of its cultural, morphological and physiological features with those in Bergeys Manual of Determinative Bacteriology (Cowan, 1974; Buchanan and Gibbons, 1974). Details of the methods used in characterization of the isolates are as presented below;

Gram Staining: This differential staining was used to group bacteria in to two major groups; Gram positive and Gram-negative bacteria based on their cell wall composition. In this staining process, a heat-fixed smear was prepared and air-dried. The smear was covered with crystal violet for 60 seconds, rinsed gently with running water stained with iodine and allowed for 60 seconds, before being rinsed off again. The smear was decolorized with 70% alcohol for 10 seconds and rinsed off immediately. The smear was counterstained with safranin for 30 seconds and finally washed with clean water and blotted dry. The stained smear was observed under oil immersion objective lens (100x) microscope. Gram positive bacteria-stained purple (the colour of the primary stain) while the Gram-negative bacteria stained red (the colour of the counter stain).

Citrate Utilization Test: This test is used to determine the potential of the bacterial isolate to use citrate as a carbon source and ammonia as its only nitrogen source. The test organisms were inoculated on the surface of sterile molten Simon citrate agar and incubated for 24 hours at 37°C. Colour change from green to blue around the growth streak indicates a positive reaction for the organism.

Catalase Test: This test was carried out to demonstrate the presence of catalase in the organism. Catalase is an enzyme that catalysis the release of oxygen from hydrogen peroxide (H_2O_2) . Two to three drops of hydrogen peroxide were placed on a clean grease free slide. The test organism was transferred to the slide with a sterile loop. Positive result was indicated by an immediate gas bubbling or effervescence off the mixture.

Coagulate Test: This test depends on the ability of some bacteria to produce an extra- cellular enzyme coagulase. The coagulase has the ability to coagulate certain blood plasma especially the rabbit and human plasma. It is specifically employed to differentiate Staphylococcus aureus from Staphylococcus species. A loopful of normal saline was placed on grease free slide. An inoculum of the test organism was emulsified on

the saline. A drop of human plasma was added to the suspension and mixed for 5 seconds. Clumping of the plasma indicated a positive result.

Oxidase Test: Oxidase enzymes play a significant role in the processes carried out by the electron transport system during aerobic respiration. Cytochrome oxidase uses O_2 as an electron acceptor during the oxidation of reduced cytochrome to form water and oxidized cytochrome. A filter paper was placed on a clean petri dish. Two drops of oxidase reagent were added on the filter paper. With a sterile glass rod, the fresh test organism was smeared on the filter paper. A blue coloration after 10 minutes indicated a positive result.

Methyl Red and Voges Proskauer Test: This test is based on the ability of the test bacteria to ferment glucose with the production of acid. This process lowers the pH of the medium to pH 4.5. Glucose phosphate broth was prepared and autoclaved at 121° C for 15 minutes; it was allowed to cool and inoculated with the test organism. The tubes were incubated for 48 hours at 37° C.

Methyl Red test: Approximately 3 drops of methyl red was added into tube containing 2 ml of the broth culture, shaken gently and observed for the formation of bright red coloration.

Voges Proskauer: Approximately 1ml of the mixture of aqueous potassium hydroxide (KOH) and 30% alcohol alpha-naphthol was added into each test tube and shaken vigorously. Production of a pink coloration indicated a positive reaction.

Spore Formation: Spore forming is one of the modes of survival for organisms during unfavorable conditions. Spore staining is used to identify spore forming organisms. A heat fixed smear was prepared from 24 hours old culture of the test organism. The slide was placed over boiling beaker and the smear was flooded with Malachite green staining solution for about 5-6 minutes until steam rises. Saturation was avoided by continuous flooding with the solution. The slide was cool before rinsing with water for 30 seconds. The smear was then counter stain with safranin for 60 to 90 seconds and rinsed again water for 30 seconds. The slide was then blot dried from the edge with filter paper and examined under oil immersion. The spores stained green while the vegetative cells-stained red.

Motility: This test demonstrates the ability of an organism to move from one place to the other with the aid of a locomotive structure like the flagella. The test tube method was used where nutrient broth was stabbed inoculated with fresh test isolate in a straight line. The test tube was covered with a cover and incubated for 3-5 days. The test tube was examined for spread from the line of stab. An observable spread indicated a positive result.

Urease Test: This test demonstrates the ability of an organism to elaborate urease enzyme. The enzyme is responsible for the breaking down urea to produce ammonia and carbon dioxide. 24.5g of Christiansen urea agar was diluted in 1,000 ml of water and autoclave for 15mins at 121oC. 10ml of 20% urea was dissolved into agar and shaken to mix; it was then poured into Petri dishes. The test organism was then inoculated via streaking on the media and incubated for 24 hours at 37oC. Development of pink coloration indicated a positive urease test while not pink color indicated negative urease test

Starch Hydrolysis: This test demonstrates the ability of an organism to hydrolyze starch. To determine this, the test isolates were streaked on starch supplemented agar plates and incubated for 24 hours. After the 24-hour incubation time, iodine is added to the plate and observed. Appearance of transparent clear zones around the colonies indicated positive result, i.e., the test organism is able to hydrolyze starch while dark blue coloration indicates negative result i.e., the unhydrolyzed starch forms the colored complex with starch.

Hydrogen Sulphide (H_2S) Test: This test demonstrates the ability of the isolate to reduce Sulphur containing compounds to hydrogen sulphide during metabolism. Here, the test isolates were inoculated into test tubes containing Klinger Iron Agar (KIA) and incubated for 24 hours. After 24 hours incubation the test tubes were observed. Development of blacken spots indicated positive result while absent of blacken spot indicated negative result.

Sugar Fermentation Test: This test demonstrates the ability of microorganism to ferment carbohydrate by utilization of sugar as source of carbon to produce acid and gas. Exactly 1 gram of sugar was added to 1% peptone water. Then 0.01% phenol red indicator was added into the medium and 10 ml were dispensed into the test tubes with inverted Durham's tubes. The test tubes and their contents were sterilized in the autoclave. Each test organism was inoculated into each sugar tube and incubated at 37°C for 24 hours. Tubes with yellow

coloration indicated acid production, while tubes with yellow coloration and gas bubbles in the inverted Durham's tubes indicated both acid and gas production.

IV. RESULTS

From thorough adherence of the analytical procedures in the preceding chapter, the results for the physiochemical analysis of the three water samples obtained from three locations in Ogbia local Government area of Bayelsa state Nigeria, are given in Table 1.1 and 1.2.

S/N	Parameters	Unit	Otuokpoti	Kolo 1	Kolo 3	NSDWQ 2015
1	Temperature	°C	28.9	29	29	Ambient
2	рН		9	8.7	8.5	6.5 - 8.5
3	Conductivity	µs/cm	724	286.6	287	1000
4	Turbidity	NTU	0.51	1.06	2.41	5
5	TDS	mg/l	361	143.5	142.5	500
6	Salinity	ppm	0.04	0.01	0.01	4
7	Ammonia	mg/l	0.34	0.22	2.28	1
8	Ammonium	mg/l	0.36	0.22	2.56	0.5
9	Nitrogen	mg/l	0.28	0.17	1.88	
10	Phosphate	mg/l	3.65	6.9	11.6	100
11	Phosphorus	mg/l	12	2.3	3.8	
12	Chromium	mg/l	0.02	0.01	0	0.05
13	Nickel	mg/l	0.55	0.6	0.6	0.02
14	Iron	mg/l	0.01	0.14	0.32	0.3
15	Lead	mg/l	0.125	0.06	0.011	0.01
16	Nitrate	mg/l	31.73	28.32	28.45	50
17	Nitrite	mg/l	0.32	0.15	0.35	0.2
18	Total Hardness	mg/l	62.11	48.92	54.6	100

Table 1.1 Physio-Chemical of Water Samples Results

Table 1.2 Physio-Chemical of Water Samples Results

19	Magnesium	mg/l	21.4	16.2	35	20
20	Zinc	mg/l	2.03	1.1	1.03	3
21	Copper	mg/l	0.77	0.63	0.86	1
22	Cadmium	mg/l	0.063	0.014	0.022	0.003
23	Chloride	mg/l	113.6	63.9	56.8	100
24	Sodium	mg/l	26.8	19.8	3.03	200
25	Alkalinity	mg/l	440	224	216.8	100
26	Potassium	mg/l	12.05	9.2	4.4	100
27	Fluoride	mg/l	0.53	0.42	0.4	1.5
28	Dissolved Oxygen(DO)	mg/l	9.35	10.4	9.6	
29	BOD	mg/l	5.6	6	5.75	
30	COD	mg/l	34.54	42.61	39.74	
31	TSS	mg/l	194.1	220.8	156	500

Colonycolour	Types	Natureofh	Vegetationstr	Asexualspore	Specialreproductive	Conic	Vesicleshape	Probableorga
	ofsoma	yphae	ucture		structure	al		nism
						head		
Densefeltyellowgre	Filament	Septate	Footcell	Globoseconidia	Phialides bornedirectly	Radiat	Sub globose	Aspergillusfl
encolony	ous	-			onthe vesiclesclerotia	e	-	avus
Smokyor	Filament	Septate	Footcell	Globoseconidia	Shortconidiophorees	Typic	Domeshapedbrod	Aspergillus
graygreencolony	ous	-			_	ally	lyclavate	fumigatus
						colum	-	_
						nar		
Green	Filament	septate	Broom-	Subgloboseconidia	Highly 3-stage	-	-	Penicilliumno
	ous	-	likeappearan	-	branchconidiosphores			tatums
			ce					
Creamy	Pseudohy	Septate	Anamorphs	Blastoconidiachlom	Buddingcells	Radiat	Dome shape	Candidaalbic
whitemoistcolony	phae	-	_	ydospores	-	e	_	ans
Whitebecominggre	Filament	Coenocy	Stolonsrhizoi	Ovoidsporangiospor	Tallsporangiophores	-	-	Rhizopus
yishbrown	ous	Tic	ds	es	ingroups,black-			stolonifer
-					brownsporangia			
Creamishyellowcol	Filament	Coenocy	-	Sporangiosp	Sympodiallybranchedspo	-	-	Mucorsp.
ony	ous	Tic		Ore	rangiophore			-

Table 1 Macroscopic and microscopic characteristics of fungal isolated from three (3) water samples

However, all the water samples allowed the growth of bacteria. The results of all the tests are given in the tables below:

Table 3: Chemical characterization and identification of bacterial isolated from three (3) water samples

Gram	Shape	Catal	Coagu	Motil	Starch	Citr	Ure	MV	Spore	H2	Oxid	Ind	Gluc	Malt	Xyl	Lact	Fruct	Sucr	Mann	Galact	Probable
Reacti ons		ase	lase	ity	hydrol ysis	ate	ase	P R	format ion	S	ase	ole	ose	ose	ose	ose	ose	ose	itol	ose	organisms
+	Rod	+	-	+	+	-	-	- +	+	-	-	-	AG	A	-	-	A	A	-	A	Bacillus cercus
+	Rod	+	-	+	+	+	-	- +	+	-	-	-	AG	A	A	-	A	-	-	A	Bacillus sublilis
+	Rod	-	-	+	+	+	-		+	+	-	-	A	A	-	-	A	A	A	AG	Chromatiumsp
-	Shortrod	+	-	+	-	-	-	+-	-	-	-	+	AG	AG	A	+	-	-	AG	-	Escherichiacol i
+	Coccinpai rs	+	-	-	+	+	+	+-	-	-	+	-	-	A	A	-	А	-	А	A	Micrococcus sp
-	Rod	+	-	+	-	+	-	+-	-	+	-	-	A	A	A	-	-	-	AG	-	Salmonella sp
-	Rod	+	-	+	-	-	-	+-	-	+	-	-	A	AG	A	-	-	AG	AG	A	Shigellasp
+	Cocciincl usters	+	-	-	-	+	-	- +	-	-	-	-	А	A	-	-	A	A	-	A	Staphylococus albus
+	Cocciincl usters	+	+	-	-	+	-	- +	-	-	-	-	A	A	-	A	AG	-	AG	А	Staphylococcu saureus
-	Comma	+	-	+	+	-	+	- +	-	-	+		A	A	-	-	A	A	-	A	Vibroheamolyt icus

Table 4: Microbial load of water Samples

Samples	ТНВС	TCC	FC	SSC	SC	VC	FC
Unit	(Cfu/ml)	(Cfu/ml)	(Cfu/ml)	(Cfu/ml)	(Cfu/ml)	(Cfu/ml)	(Cfu/ml)
1	5.6 x 10 ⁴	1.42 x 10 ⁵	2.8×10^4	1.02×10^4	4.0 x 10 ⁴	$2.4 \text{x} 10^4$	$2.0 \text{ x} 10^4$
2	2.10 x 10 ⁵	1.63 x 10 ⁴	6.3 x10 ⁴	5.6x10 ⁴	4.1 x10 ⁴	3.9 x10 ⁴	2.0 x10 ⁴
3	2.50x 10 ⁵	1.70 x 10 ⁴	9.1x10 ⁴	8.3 x 10 ⁴	7.6 x 10 ⁴	1.02×10^5	1.3 x 10 ⁴

V. DISCUSSIONS

From the results of the tests carried out the Faculty of Science of the University of Uyo, it was discovered that most of the values for the physiochemical parameters measured were within the specified limits set by the NSDWQ (2015). Few of the Parameters such as the alkalinity, nickel, pH, Lead etc. as was tested in the lab were a bit higher than the maximum permissible limit by NSDWQ (2015). I also noticed that sample 1, had the highest values for most of the tested parameters such as Conductivity, acidity, alkalinity dissolved solids etc.

It was discovered that the well water samples examination carried out allowed the growth of bacteria in them. The values of the tested parameters from the results obtained from the physiochemical analysis of the water samples doesn't give a clear indication of the negative impact of the proximity of the dump sites to the water underground, since most of the parameters for the three samples are within the maximum permissible limit by NSDWQ and only a few parameters are above the limit. In spite of this, people are liable to contact diseases such as cholera, typhoid fever, shigella (etc.) because the wells are open and the poor sanitary conditions of the locations in which they are situated which are very dangerous to human health. Appropriate measures should be taken by the community (Ogbia) to treat the wells at least three times in a year to kill the growth of the bacteria before consumption. The World Health Organization procedure for regulation of drinking water must be adhered to. Nevertheless, appropriate measures should be taken by relevant authority on the method of waste disposal in the areas affected.

VI. CONCLUSION

The presence of some quantity of bacteria in the water samples mean that leachate is altering the water quality of water in the vicinity of the dumpsite. Bacteriological concentration of water from all three samples exceeded the standards. Hence the water in the vicinity of the dumpsite presents significant threat to public health. From the above findings on the subject of effect of solid wastes on the quality of underground water, the following recommendations are suggested:

The wells should be sited at least 30m from solid wastes dump site. Secondly Appropriate place should be located at the outskirt of the town for refuse / disposal and finally the wells should be treated at least three times in a year.

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