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BIOELECTRICITY PRODUCTION FROM WASTEWATERS USING MICROBIAL FUEL CELL

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ABSTRACT

Electricity supply in Nigeria has been a recurring decimal as huge amount of capital is involved. The aim of the study is to produce bioelectricity in a microbial fuel cell (MFC) from wastewaters. Wastewater from abattoir, fish pond, and stream were used to construct a microbial fuel cell and their physiochemical and microbiological properties were determined and the voltage of electricity produced was measured. Wastewaters were separately inoculated using pour plate to determine bacterial counts and type of bacteria in the wastewater. MFC abattoir waste gave the highest voltage (0.77V), followed by fish pond MFC with 0.58V, and least was stream MFC with 0.30V. The pH of the three wastewaters ranged from 6.24 – 5.60. The MFC with Abattoir wastewater had the highest electrical conductivity (15,150 μ/cm^3), total dissolved solids (10,230), dissolved oxygen (8.20 mg/L), and BOD (6.00 mg/L), while MFC with stream waste water had the least electrical conductivity (600 μ/cm^3), total dissolved solids (390), dissolved oxygen (4.20 mg/L), and BOD (2.00 mg/L). Connection in series gave a voltage 1.89V from three microbial fuel cells constructed with wastewaters from the abattoir. Wastewater from Abattoir obtained the highest count both for aerobic (2.56×10^{23} cfu/ml) and anaerobic (1.35×10^{23} cfu/ml), while wastewater from the stream gave the least count 1.32×10^{23} cfu/ml (aerobic) and at 6.55×10^{22} cfu/ml (anaerobic). Isolated bacterial species includes; *Actinobacillus, Aeromonas, Bacillus, Citrobacter, Clostridium, Enterobacter, Enterococcus, Escherichia coli, Klebsiella, Micrococcus, Neisseria, Proteus, Psuedomonas aeruginosa, Samonella, Serrentia, Shigella, Staphylococcus and Streptococcus*.

1.0 INTRODUCTION

MFC is considered a sustainable technology for energy production and wastewater treatment. Biofuel is produced through complementary biological processes such as agriculture and anaerobic digestion. It refers to any fuel whose energy is obtained through a process of biological carbon fixation. It includes fuels derived from biomass, liquid fuels and various biogases. Biofuels are gaining increased public and scientific attention, due to the need for increased energy security and concern over greenhouse gas emission from bio-fossil fuels. Examples of biofuel include bioethanol from alcohol fermentation, biodiesel from vegetable oils and animal fats (Hall *et al.,* 2011).

Bioenergy is a renewable energy available from materials derived from biological sources. Renewable source of energy is a need of developing countries to fulfill their present and future energy deficiency. Electrical energy demand up to some extent can be fulfilled by microbial fuel cell (MFC). Microbial fuel cell technology represents a new form of renewable energy by generating electricity from waste. They are capable of converting chemical energy available in organic substrates into electrical energy using microorganisms as a biocatalyst to oxidize the biodegradable substrates. MFCs are capable of converting chemical energy available in organic substrates into electrical energy using bacteria as a biocatalyst which oxidizes the biodegradable substrates.

Electrical energy produced using microorganisms through microbial fuel cells is sustainable and renewable and is one of the most efficient sources of electricity (HaoYu *et al.,* 2007). The fact that bacteria can oxidize the substrates to produce electricity makes MFCs an ideal solution for wastewater treatment and domestic energy production (Schwartz, 2007). Logan (2010) reported that MFCs can generate power densities as much as 1kW/m^3 of reactor volume. MFCs as a source of bioenergy production has accelerated research-wise worldwide and the technical aspects of MFCs have been reviewed extensively (Pant, 2010).

The ability of certain bacteria to produce electrical current in the laboratory was for many decades a scientific curiosity, with little hope initially for practical applications. Many microorganisms possess the ability to transfer the electrons derived from the metabolism of organic matters to the anode. Wastewater, soil, marine sediment, sludge and animal wastes are all rich sources of substrate for these microorganisms.

The idea of using microbial cells in an attempt to produce electricity was first conceived at the turn of the nineteenth century by *M.C. Potter* who first performed work on the subject in 1911. A professor of botany at the University of Durham, Potter managed to generate electricity from *Escherichia coli*, but the work was not to receive any major coverage. In 1931, however, Barnet Cohen drew more attention to the area when he created a number of microbial half fuel cells that, when connected in series, were capable of producing over 35 volts, though only with a current of 2 milliampere. It is now known that electricity can be produced directly from the degradation of organic matter in the microbial fuel cell. Sugar when utilized by microorganisms under aerobic condition release carbon dioxide and water, but when oxygen is not present the end product is carbon dioxide, protons and electrons as described below.

$$C_{12}H_{22}O_{11} + 13H_2O \text{ ---> } 12CO_2 + 48H^+ + 48e^-$$

The anodic electron transfer mechanism in MFC is a key issue in understanding the theory of how MFCs work. There are two terminals; one is positive and the other negative. Electricity flows from the negative terminal to the positive terminal (Singh and Songera, 2012).

2.0 MATERIALS AND METHODS

2.1.0 MATERIALS

2.1.1 Collection Area

Samples were collected at early hours of the day from Lafia modern abattoir, Shingeh, Alubo fish pond and Gandu stream in Lafia.

2.1.2 Samples Collection:

A total of 3,000mL of wastewater samples and sediments were collected from three different locations. One thousand milliliters each was collected from the different venue. Samples were collected by scooping the wastewater along with the sediment using a shovel into the bowl, and collection was in the morning to avoid influence of anthropogenic activities around the sample.

2.1.3 Processing of Wastewater

2.1.3.1 Wastewater from Abattoir

The samples collected from the Lafia modern abattoir located in Shingeh consists of wastes from large amount of slaughtered farm animals which include cows, sheeps, goats, rams and chicken. These include feacal wastes of the animals, blood, and normal flora from the skin of the animals, channeled into the abattoir dumping site where they are used as manure. Samples were collected from the dumping site.

2.1.3.2Wastewater from Alubo fish pond

The pond is made up of nine smaller ponds filled with hundreds of fish. The wastewater from each pond is channeled into a pit during the daily change of the pond water. Wastewaters together with sediments were collected from the collecting pit.

2.1.3.3 Wastewater from Gandu stream

The Gandu stream is popularly known for agricultural and recreational uses by villagers of Gandu. Samples were collected along the side of the stream.

2.1.4 Construction of Microbial Fuel Cell

MFC was constructed as described by Cheng *et al.,* (2006). MFC assembly consists of two containers connected with the external circuit and proton pump.

Apparatus used includes the following;

2.1.4.1 Plastic containers

One thousand milliliters plastic containers were used for each microbial fuel cell. One of the containers served as the anode jar which contains the wastewater which the second container served as the cathode jar which contains distilled water.

2.1.4.2 Copper wire

Copper wires with the diameter 1.40mm were used at both the anode and the cathode. The copper wires used were measured using micrometer screw jack with minimum measurement of 0.1mm.

2.1.4.3 Proton pump

The proton pumps used are molded hollow plastic tube filled with wick soaked in salt water solution for 2 hours.

2.1.4.4 Digital multimeter

Digital multimeter is a test tool used to measure two or more electrical values, principally voltage (volts), current (amps) and resistance (ohms). It is a standard diagnostic tool for technicians in the electrical/electronic industries. The digital multimeter model ALDA DT-830D was used. The digital multimeter is made up of four components; display; where measurement readout are viewed, buttons, for selecting various functions, dial or rotatory switch for selecting primary measurement values which includes volts, amps and ohms and input jacks where test leads are inserted.

The meter was turned on, and the probes were inserted into the correct connections as shown in Figure 2.1, Figurer 2.2 and Figure 2.3. The switch was set to the volts. The range was optimized for the best reading. Readings were recorded.

2.1.5 MFC set up:

Step 1: Two plastic containers are needed for the setup. One of the containers is constructed as anaerobic jar with tightly sealed cover and serves as the anode which is the negative terminal, while the other container is constructed as an aerobic jar with air supply to the cathode.

Step 2: The proton exchange bridge is constructed by soaking cotton wick in salt water. The soaked wick is placed into the plastic pipe. This serves as a passage for the movement of ions from the anode to the cathode.

Step 3: The proton exchange bridge in connected to the plastic containers by boring hole by the side of the container and fixing the proton exchange bridge tightly into the bored hole. The proton exchange bridge is tightly fixed to hold it in place and to prevent leaking.

Step 4: Copper wires are coiled up inside each container with a point at the top for external connection.

Step 5: The two containers were connected with an external circuit which is made up of copper wires connected to the point at the top of each container and connected to a digital multimeter.

Step 6: The sludge containing the microorganisms and glucose is added to the anaerobic chamber and the cover is sealed. While regular water is added to the aerobic chamber. Conductivity is increased by adding a pinch of salt.

Step 7: Voltage current is measured every two hours and results obtained were recorded.

The setup is as shown in Figure 2.4.

Step 8: The setup was repeated into three forms using wastewaters from the three different locations in the anode of each setup.

2.1.6 Measurement of electricity

The bioelectricity produced is determined by measuring the electricity produced per day and using the digital multimeter and the voltage is measured in millivolts.

Figure 2.1: Microbial fuel cell constructed with wastewater from Shingeh abattoir

Figure 2.2: Microbial fuel cell constructed with wastewater from fish pond

Figure 2.3: Microbial fuel cell containing wastewater from stream

Figure 2.4: Microbial fuel cell setup

2.2.0 METHODS

2.2.1 Analysis of various physicochemical parameters

Analysis of some physicochemical parameters was based on the method described by American Public Health Association (1995) with modifications. These include: Total Dissolved Solids (TDS), electrical conductivity (EC), dissolved oxygen (DO), biological oxygen demand (BOD), chlorine, pH, temperature and color, was carried out on the wastewaters.

2.2.2 Total dissolved solids (TDS):

Fifty centimeter cube ($50cm_3$) of the sample to be determined was filtered through a filter paper whose initial weight has been determined. The filtrate was collected into an evaporating dish, which has originally been weighed. The filtrate was then allowed to evaporate to dryness by heating on a water bath. The residue was allowed to cool to room temperature. It was then weighed and the value is recorded as the difference between the combined weight of the evaporating dish and residue and that of the empty evaporating dish is recorded as the weight of the dissolved solids.

2.2.3 Electrical conductivity (EC)

Electrical conductivity is the ability of a conductor to allow the passage of electric current. Electrical conductivity can serve as an indirect way of measuring dissolved minerals in the wastewater. The magnitude of electrical conductivity is useful in the indication of total concentration of ionic solutes. The EC was measured using the conductivity meter HQ14d – HACH company, employing APHA 2510-B(electrometric) method.

2.2.4 Dissolved oxygen

Dissolved oxygen in water is the amount of oxygen that is dissolved in water either through aeration diffusion or photosynthesis activity of algae measured in mg/L. it is traditionally measured with Winker's Azide modification method or Digital method. The digital method was used in this work with APHA 4500-OC/4500-OG as the method and VWR Oxygen meter as the instrument.

2.2.5 Biochemical oxygen demand (BOD)

This test is to calculate the amount of dissolved oxygen needed by aerobic organisms to break down organic material present in a given water sample at certain temperature over a specific period of time. Wastewater sample was collected into a BOD bottle with sample water without making air bubble. 2ml of manganese sulphate was added to the BOD bottle by inserting the pipette just below the surface of the water. 2mls of alkali-iodide-azide reagent was added to the BOD bottle in the same manner. The mixture was well homogenized until a brownish cloud appears as an indicator of the presence of oxygen. The brown precipitate is allowed to settle to the bottom and 2mls of concentrated H_2SO_4 was carefully added. The mixture was homogenized to dissolve the precipitate. The BOD bottle was kept in the incubator for 5 days.

After incubation, 50ml of the sample water was titrated with 0.025N Sodium thiosulphate to a pale yellow colour. 2ml of starch solution was added till the sample turns blue. The titration continues till the sample gets clear and readings are noted. The concentration of the dissolved oxygen in the sample is equivalent to the number of milliliters of the titrant used.

2.2.6 Colour

Fifty milliliters of the wastewater sample was placed in a colourless test tube and compared with deionized water. The tubes were observed vertically downwards towards a white surface placed at an angle that light is reflected upwards through the columns of liquid. The colour were determined and shown in the table below.

2.2.7 Chlorine

The test method employed was APHA 4500-Cl using HACH 223101 (CN-66) Chlorine test kit and results were recorded.

2.2.8 Temperature

Direct measurement using mercury in glass centrigrade thermometer was employed. A clean thermometer was placed vertically in the containers containing the sample and allowed to stand for 3 – 5 minutes without touching the edges of the containers until the temperature became steady, which was recorded. This was done for three consecutive days.

2.2.9 pH

The pH meter was first activated using potassium chloride (KCl) solution so that the sensitive electrode would be active. The pH meter model HI 2214/ORP Meter was then calibrated and conditioned using standard buffers of 4, 7 and 12. The pH of 4 was used for acidic medium, 7 for neutral medium and 12 for alkaline medium. The pH for each wastewater sample was determined by measuring 25ml of the sample into a beaker and then the electric pH meter electrode was dipped into the solution. The pH value for each sample was read and recorded. This was done for three consecutive days.

2.3 Isolation of microorganisms

Isolation of microorganisms from wastewater was based on the method described by Zhang *et al.* (2009) with modifications. Aseptically, 1mL of the wastewater was measured into a beaker containing 9mL of sterile physiological saline which served as the stock and was allowed to stand for 1 minute. The stock was serially diluted to 15 dilutions. 1mL of 10^{-15} was prepared by pour plate method in nutrient agar for total coliform count. 1mL aliquot from 10^{-15} of each sample was inoculated by pour plate method into molten nutrient agar and was allowed to gel, it was inverted and incubated for 24 hours at $35^{o}C$ at aerobic and anaerobic conditions using the incubator and candle jar respectively.

2.4 Identification of isolates

Microorganisms were identified macroscopically based on their appearance on the plate and microscopically based on their Gram staining properties. Biochemical tests of isolates were carried out according to the method described by Zhang *et al*. (2009) with modifications.

2.4.1 Gram staining

Twenty four hours old presumptive colony of each isolate was emulsified on a clean grease-free glass slide containing a drop of distilled water and allowed to air dry. The smear was heat fixed. It was flooded with crystal violet as primary stain and allowed to stand for 60 seconds and rapidly washed off with clean distilled water. The smear was then covered with iodine for 60 seconds and rinsed with running water. The smear was decolorized with alcohol and immediately rinsed with clean water. Counter staining was done for 60 seconds using safranin and rinsed with clean water. The slide was allowed to air dry and viewed under the microscope using the oil immersion objective.

2.4.2 Catalase test

The slide test method of detecting the enzyme catalase was used. Catalase test was carried out on each isolates by placing a pure colony of each isolate on a clean grease free slide containing a drop of distilled water and smeared. A drop of 3% hydrogen peroxide (H_2O_2) was dispensed on the smear and it was observed for rapid and immediate bubbling which signified a positive result.

2.4.3 Coagulase test

The slide test method of detecting the enzyme coagulase was used. A drop of plasma was placed on a slide and a portion of the test organism was added to it. The slide was observed for the presence of agglutination of the cells on the slide within one to two minutes which indicates the presence of the enzyme coagulase.

2.4.4 Indole test

This test detects the production of indole from the amino acid tryptophan. The test was carried out by dispensing 3ml of sterile tryptone water into sterile testtube. The test organism was inoculated into the typtone water and incubated for 48 hours at 35^{o}C. After incubation, a drop of Kovac's reagent was added to the bijou bottle and agitated gently. The testtube was observed for the formation of red coloration on the surface of the mixture within 10 minutes which signifies a positive result.

2.4.5 Citrate utilization test

This test detects if carbon is used as the main source of energy by the microorganism. Slants of Simmon citrate agar was prepared by preparing the agar according to manufacturer's instruction and poured into sterile test tubes. Using a sterile inoculating needle, a saline suspension of the organism was streaked on the slants. The inoculated slants were incubated for 24 to 48 hours at 35^{o}C. Color change from green to blue was observed which signifies a positive result.

2.4.6 Oxidase test

This test is to detect the presence of the enzyme oxidase. An oxidase strip was moistened with a drop of sterile water and emulsified with the test organism that was aseptically removed from the petri dish using sterile applicator stick. Color change from red to purple was observed within 20 seconds which signifies a positive result.

2.4.7 Haemolysis test

Isolated organisms were subcultured onto blood agar plates and incubated for 24 – 48 hours at 37^{o}C. Beta, alpha and gamma hemolysis was observed. Colour change from red to yellow or green around colonies signified a positive result.

3.0 RESULTS AND DISCUSSION

3.1 Statistical analysis

One way analysis of variance (ANOVA) and paired t-test was carried out using IBM SPSS STATISTICS version 22.

3.2 Physiochemical properties of the wastewater

As shown in Table 3.1, electrical conductivity was highest in wastewater from abattoir (15, 150 µ/cm3), while the lowest was in sand collected from stream (600 µ/cm3). Sand sample from the stream had lowest total dissolved solids, biological oxygen demand, dissolved oxygen and chlorine concentration of 390, 2.00 mg/L, 4.20 mg/L and 14.18 mg/L respectively.

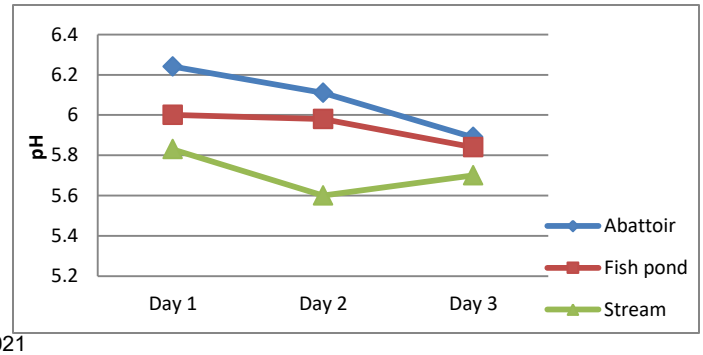
Table 3.1: Physiochemical properties of the wastewaters

Parameters	Abattoir	Fish pond	Stream
Electrical conductivity (µ/cm^3)	15,150	2,640	600
Total dissolved solids (TDS)	10,230	1,740	390
Dissolved oxygen (DO) (mg/L) 10th dilution	8.20	6.20	4.20
Biochemical oxygen demand (BOD) (mg/L)	6.00	4.00	2.00
Chlorine (mg/L) 5th dilution	50.00	35.45	14.18
Colour	**Black**	**Dark Brown**	**Brown**
Temperature	**31.4**	**31.2**	**30.9**
pH	**6.02**	**5.94**	**5.71**

3.3 pH fluctuation of the various wastewaters

Wastewater samples from the abattoir had the highest value of 6.24 on day 1 and the value decreased to 5.89 on day 3 (Figure 4.1). Stream sample had the lowest pH values ranging between 5.83 – 5.60.

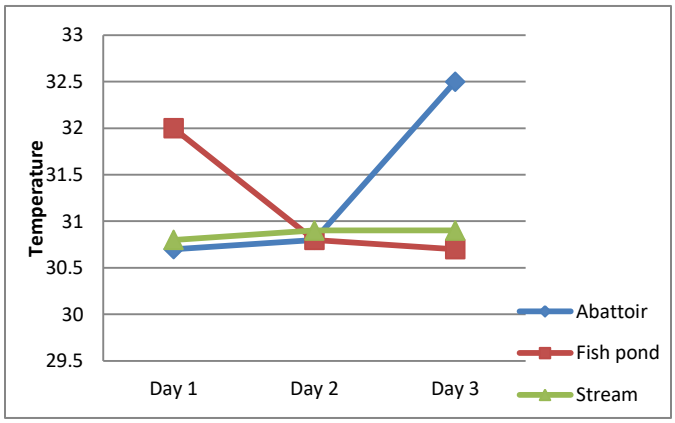
Figure 3.1: pH fluctuation of the various wastewaters



3.4 Temperature of the wastewaters

As shown in Figure 3.2, Abattoir on day 3 had the highest temperature of 32.5°C, while the lowest value was on day 3 and day 1 for abattoir and fish pond (30.7°C). Temperature values rose and fell as shown in the figure.

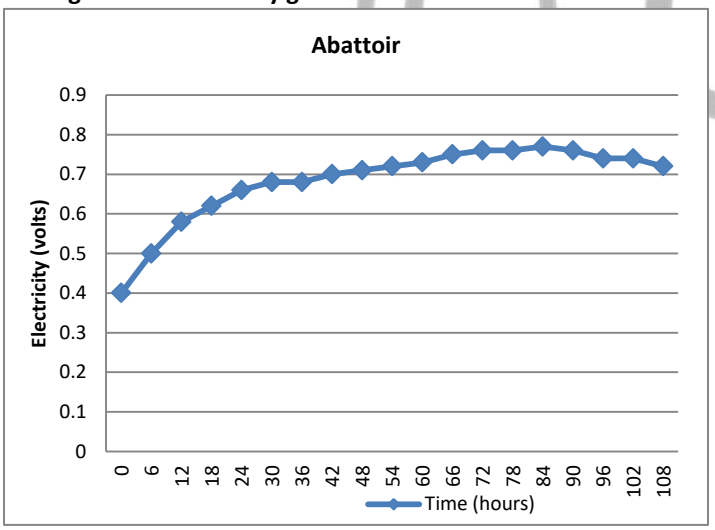
Figure 3.2: Temperature of the wastewaters



3.5 Electricity generated from abattoir wastewater

As shown in Figure 4.4, the highest electricity generated from abattoir wastewater was after eighty four (84) hours (0.77 volts). There was linear increase in the production of electricity followed by a gradual decrease after eighty four hours.

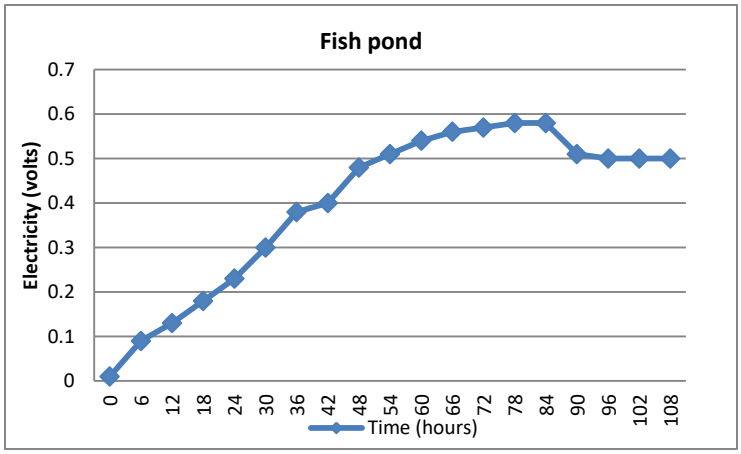
Figure 3.3: Electricity generated from Abatoir wastewater



3.6 Electricity generated in fish pond

As shown in Figure 4.4, there was a gradual increase in the amount of electricity produced. When the highest electricity, 0.58 volts, was produced, after seventy two (72) hours, there was drastically decrease in the amount of electricity produced.

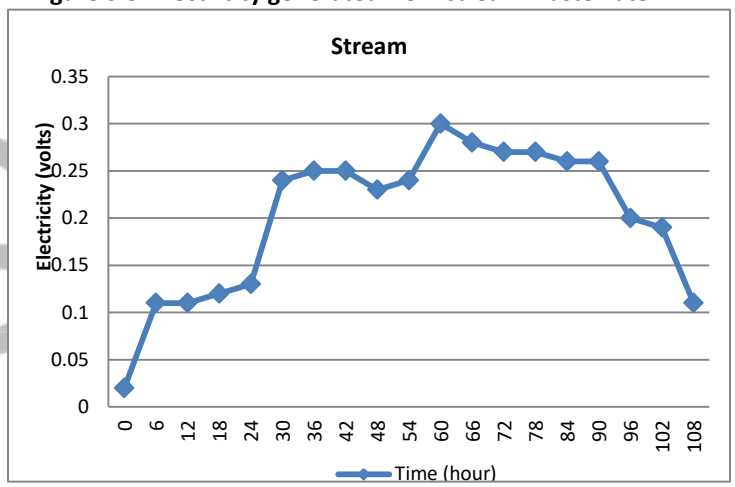
Figure 3.4: Electricity generated from fish pond wastewater



3.7 Electricity generated in Stream wastewater

As shown in Figure 4.4, the highest electricity generated (0.30), was after sixty (60) hours after which there was a gradual decrease of electricity generated.

Figure 3.5: Electricity generated from stream wastewater



3.9 Mean electricity generated

The mean electricity generated was calculated by dividing the total electricity produced by each wastewater by the total hours it was left to stand. The mean electricity increase in each wastewater in day one, two and three but decreases drastically in day four. The wastewater from abattoir has the highest mean which is 0.72 volts. This is shown in Table 3.2.

Table 3.2: Mean electricity generated

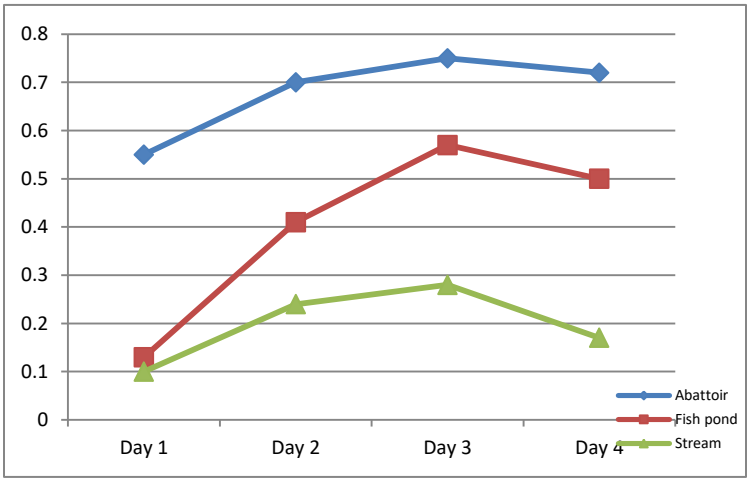
Day	Abattoir (volts)	Fish pond (volts)	Stream (volts)
1.	0.552±0.1030	0.128±0.0843	0.098±0.4444
2.	0.698±0.0180	0.414±0.0836	0.242±0.0084
3.	0.754±0.0150	0.567±0.0167	0.276±0.0152
4.	0.722±0.0430	0.500±0.0071	0.172±0.0569

3.10 Graphical representation of mean electricity generated

The electricity generated was represented graphically as shown in Figure 3.5. The graph shows a spontaneous increase and decrease of electricity generated by the various wastewaters. Wastewater from abattoir shows the highest amount of electricity generated while the wastewater from

the stream shows the wastewater with the least amount of electricity generated.

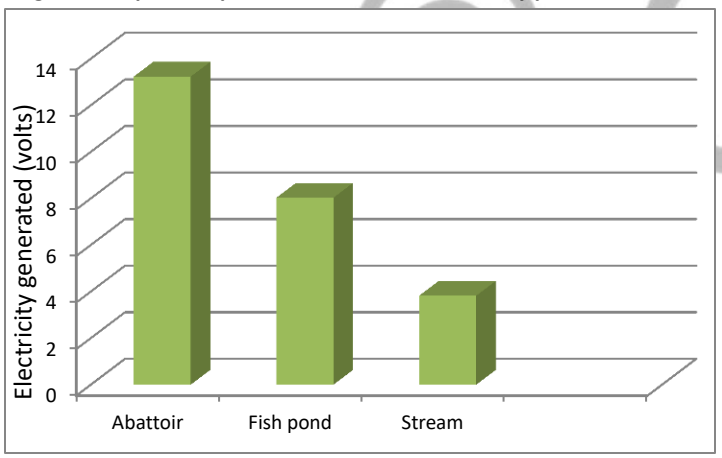
Fig. 3.6 Graphical representation of mean electricity generated



3.11 Graphical representation of total electricity produced

The total amount of electricity produced by each wastewater was represented on the graph as shown in Figure 4.3. The wastewater from the abattoir has the highest production of electricity and the wastewater from the stream has the least production of electricity.

Fig. 3.7: Graphical representation of total electricity produced



3.12 Number of colonies in each wastewater

Sequel to growth on nutrient agar for day 1 and colonies were counted. Table 4.5 shows the number of growth on the agar. Abattoir had the highest number of colonies.

Table 3.3 Number of colonies in each wastewater

Wastewater	Colony Forming Units			
	Day 1		Day 2	
	Aerobic (cfu/ml)	Anaerobic (cfu/ml)	Aerobic (cfu/ml)	Anaerobic (cfu/ml)
Abattoir	2.84×10^{23}	1.02×10^{23}	2.27×10^{23}	1.68×10^{23}
Fish pond	1.64×10^{23}	9.1×10^{22}	1.53×10^{23}	1.32×10^{23}
Stream	1.23×10^{23}	6.2×10^{22}	1.41×10^{23}	6.9×10^{22}

3.13 Biochemical properties of isolated microorganisms

Samples of the wastewater were allowed to grow on nutrient agar and morphological characterization was taken, and colonies isolated were subjected to various biochemical tests which include their Gram's reaction, catalase, oxidase, citrate, coagulase, indole and hemolysis tests. Table 4.5 below shows the reactions of the isolated microbes to the different biochemical tests.

Table 3.4: Biochemical characteristics by isolated microorganisms

Suspected Organisms	Gram's stain	Aerobic/Anaerobic	Catalase	Oxidase	Citrate	Coagulase	Indole	Hemolysis
Actinobacillus sp.	-ve rods	A/AN	+ve	+ve	Weak	-ve	+ve	Gamma
Aeromonas sp.	-ve rods	A/AN	+ve	+ve	+ve	-ve	+ve	Beta
Bacillus sp.	+ve rods	A/AN	+ve	+ve	+ve	-ve	-ve	Gamma
Chromobacterium sp.	-ve rods	AN	+ve	+ve	-ve	-ve	-ve	Gamma
Citrobacter sp.	-ve rods	A/AN	+ve	-ve	+ve	-ve	-ve	Alpha
Clostridium sp.	+ve rods	AN	-ve	-ve	+ve	-ve	-ve	Gamma
Enterobacter sp.	-ve rods	A/AN	+ve	-ve	+ve	-ve	-ve	Gamma
Enterococcus sp.	+ve cocci	A/AN	-ve	-ve	-ve	-ve	-ve	Alpha
Escherichia coli	-ve rods	A/AN	+ve	-ve	-ve	-ve	+ve	Beta
Klebsiella sp.	-ve rods	A/AN	+ve	+ve	+ve	-ve	-ve	Gamma
Micrococcus sp.	+ve cocci	A	+ve	+ve	-ve	-ve	-ve	Beta
Neissera sp.	-ve diplococci	A	+ve	+ve	+ve	-ve	-ve	Gamma
Proteus mirabilis	-ve rods	A/AN	+ve	-ve	+ve	-ve	-ve	Beta
Pseudomonas aeruginosa	-ve rods	A	+ve	+ve	+ve	-ve	-ve	Gamma

Samonella sp.	-ve rods	A/AN	+ve	-ve	-ve	-ve	-ve	Gamma
Serrentia sp.	-ve rods	A/AN	+ve	-ve	+ve	-ve	-ve	Beta
Shewanella sp.	-ve rods	AN	+ve	+ve	-ve	-ve	-ve	Gamma
Shigella sp.	-ve rods	A/AN	+ve	-ve	-ve	-ve	-ve	Beta
Staphyococcus sp.	+ve cocci	A/AN	+ve	-ve	+ve	+ve	-ve	Beta
Streptococcus sp.	+ve cocci in chain	A/AN	- ve	-ve	+ ve	-ve	-ve	Alpha

4.0 Conclusion

High conductivity in Abattoir wastewater was responsible for higher electricity generated from it. High microbial counts in the Abattoir wastewater was also responsible for the generation of higher electricity generated from it. The BOD level of the abattoir wastewater signifies that it is somewhat polluted and organic matter is present which led to high concentrations of decomposing bacteria. Wastewater has a lot of endogenous bacteria which are capable of generating electricity. Electricity generated from waste water is low compared to the amount generated by non-renewable sources hence newer means of harvesting electrons from the microbes is encouraged.

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