International Journal of Current Research and Review
DOI: http://dx.doi.org/10.31782/IJCRR.2019.11174

Research Article

IJCRR
Section: Life Sciences
Sci. Journal Impact Factor: 5.385 (2017)
ICV: 71.54 (2015)

Effect of Methylene Blue Addition as a Redox Mediator on Performance of Microbial Fuel Cell Using Mud Sediment of River Ala

Olotu T. M.[1,2*]; Adegunloye D. V.[2], Ekundayo F. O.[2]

[1]Deparment of Microbiology, Faculty of Science, Adeleke University, P.M.B 250, Ede, Osun State, Nigeria; [2]Department of Microbiology, School of Sciences, Federal University of Technology Akure, P.M.B 704, Akure, Ondo state, Nigeria.

ABSTRACT

Microbial fuel cells (MFCs) are also bioreactors that convert chemical energy stored in the bonds of organic matters into electricity through biocatalysis of microorganisms. Mud sediment of various depths (surface water, mud surface, 50cm, 100cm and 150cm) of River Ala were used in a double chamber microbial fuel cell (MFC) to generate electric current and comparative studies of the methylene blue mediator and mediator-less chamber were carried out. Microbial analyses, physiochemical analysis of the sediment were analyzed using standard methods. River Ala surface has the highest bacteria count of 2.4 x 10-5 and AL100cm has the lowest of 0.48 x 10-5 while AL100cm had the lowest fungi count of 0.2 x 10-6. The pH of sediment ranged from 7.52 to 6.52 and organic matter content 3.67 to 1.83(%). The mud surface has the highest conductivity and salinity content of 740 (µS) and 359 (ppm) respectively. The current and voltage readings obtained from of the methylene blue mediator chamber were slightly higher than that of the mediator-less chamber. Current 0.5 (mA) at only depth 50cm was observed in mediator-less chamber while 0.4 (mA) were common occurrences at depth 50cm and depth 100cm at the methylene blue mediator chamber; voltage readings of 0.3(V) only occurred depth 50cm in the mediator-less chamber while 0.3 (V) were observed at both depth 50cm and 100cm at the methylene blue mediator chamber. The low current and voltage reading were as a result of the high resistance it's generated and its low organic matter content. It is also a confirmation that the mediator used has an impact in the current and voltage generated in microbial fuel cell.

Key Words: Sediment, Microorganisms, Electric current, Methylene blue and Microbial fuel cell

INTRODUCTION

Electricity is an essential element in our daily life, something we cannot live without literally, we would die without it. From the simplest form of living organism to the complicated human body, electrical force governs every single physiological process. Bio-electricity is vital in storing metabolic energy and providing signals to other cells which influence growth, regeneration and communication (Levin and Stevenson, 2012). In 2005, 66% of that electricity was generated from coal, petroleum and natural gas and was responsible for 10.9 Gt (41%) of world energy-related CO_2 emission (Brandt, *et al.*, 2007). Depending on the region, your electricity could come from the dirtiest coal burning plant, a high risk nuclear facility, or a hydro electrical dam, which, although pollution free, still deteriorates the local geological and ecological systems. The human-induced greenhouse effect as a result of fossil fuel reliance has become an increasingly controversial issue in many countries since the 1960s. The fast depletion of fossil fuel due to intensive extraction and usage is widely believed to be associated with the atmospheric CO_2 concentration increase from 275 ppm to 397 ppm in the last two centuries. As a result, development of sustainable energy technologies which can continue providing society with energy-derived benefits without further environmental

Corresponding Author:
Olotu T. M., Deparment of Microbiology, Faculty of Science, Adeleke University, P.M.B 250, Ede, Osun State, Nigeria.
E-mail: olotutitilayomabel@gmail.com

ISSN: 2231-2196 (Print) **ISSN:** 0975-5241 (Online)
Received: 01.08.2019 **Revised:** 19.08.2019 **Accepted:** 29.08.2019

destructions is highly desirable. A series of green energy solutions, such as solar, wind and biomass energy, have been introduced in the hope of preventing the impending global environmental crisis (Brandt, *et al.*, 2007). Microbial fuel cell (MFC) is a bio-electrochemical cell which utilizes electrogenic bacteria to oxidize a variety of substrates including acetate, glucose, volatile fatty acids and inorganic substances such as sulfides and nitrite, to form electrical current(Faraghi and Ebharimi, 2012; Rabaey, et al., 2006). Through the oxidation process electrons and protons are generated at anode and recombined at the cathode to produce water (Logan *et al.,* 2006) MFC consists of two compartments: an anaerobic anode and aerated cathode compartments which are separated by a proton exchange membrane or salt bridge (Sharma *et al.,* 2010; Higgins *et al.,* 2013). Microbial fuel cells (MFCs) are also bioreactors that convert chemical energy stored in the bonds of organic matters into electricity through biocatalysis of microorganisms (Davis and Yarbrough, 1962; Moon *et al*., 2006). A typical MFC chamber has the anodic and cathodic chamber and is separated by a proton exchange membrane (PEM) (Wilkinson, 2000; Gil *et al.*, 2013) which allows transport protons while blocking oxygen and other compounds. Microbes in the anodic chamber degrade organic matters and produce electrons, protons and carbon dioxide. Electrons and protons produced by microbes are then transported to the cathodic chamber via external circuit and a proton exchange membrane (PEM), respectively. In the cathodic chamber, protons and electrons react with oxygen to form water. Because the terminal electron acceptor (*i.e*., oxygen) is kept away from the anodic chamber, electrons are allowed to pass through the external load to generate electricity (Park, *et al.*, 2000; Du *et al*., 2007). A variety of bacteria can produce a modicum of electricity in an MFC if a mediator is used to facilitate the transfer of electrons between the bacterial cells and the anodic surface used in the system, while many other bacteria have been found to possess the ability to transfer electrons from fuel (substrate) oxidation to a working electrode without a mediator (Logan, 2009). Direct electron transfer from anaerobic anode chamber to its surface had shown to take place at low efficiency. Electron transfer efficiencies in MFCs can be improved using a suitable electron mediator. Most biological fuel cells use electron mediator component to improve the power output of the cell. It has been reported that mediators are artificially added to anode chamber, such as MB, neutral red (NR), thionin, ferricyanide, humic acid or methyl viologen. The presence of artificial electron mediators is essential to improve the performance of MFCs (Park and Zeikus, 2000). This experiment is to determine microbial population and identification of microorganisms in various depths of the mud sediment of River Ala, to compare the methylene blue mediator microbial fuel cell chamber with the mediator-less chamber in current and voltage generation. It will also determine of the pH, organic carbon, conductivity, ionization potential and salinity of the mud sediments in relation to current generation.

MATERIALS AND METHODS

Collection of sample used in the Microbial fuel chamber

1kg mud sediment sample were collected at the various depths of the mud surface, depth 50cm, depth 100cm, depth 150cm and the surface water. Samples were collected aseptically in clean containers and transported to Microbiology laboratory of Federal University of Technology, Akure.

Microbiological and physiochemical analyses of the mud sediment samples

Microbial population and identification was determined for each samples (mud surface water, mud surface, depth 50cm, depth 100cm and depth 150cm) and for the control (which was soil sample of area close to the river). The microorganisms which were mainly bacteria, fungi and yeast were isolated using nutrient agar, centrimede agar, mannitol salt agar, salmonella- shigella agar, marconkey agar, eosin methyl blue and potato dextrose agar. The physiochemical parameters determined were pH using a Jenway's pH meter, Conductivity using digital Conductivity Mettler Toledo M400 measuring meter and Salinity using electrical conductivity using a conductivity bridge .Organic matter of the soil was done according to the method of (Skotnikov, 1998) and mineral matter was determined using atomic Absorption Spectrophotometer according to (Bhargava and Raghupathi, 1993).

Construction for the various mud sediment depths

1.5 liter size transparent plastic bottles made up the cathode and anode chamber, hole of 2cm in diameter were bored at each side of the bottles. Polyvinyl chloride (PVC) pipe of dimensions 5 cm length and 2 cm diameter made up the agar salt bridge. Each container was surface sterilized with 70% ethanol before introduction of its content. The salt bridges to be used was prepared prior to collection of the mud sample and kept from contamination before use. Each salt bridge was then attached to the each anode and cathode bottle using an epoxyl gum as according to (Parkash, 2016).

Composition of anodic and cathode chamber

Mud sediment of 1litre size of the various samples (mud surface water, mud surface, depth 50cm, depth 100cm and depth 150cm) was introduced into the mediator and mediator-less anode chamber and the cathode chambers was filled 1L of NaCl solution which was made up 7.5g of Nacl in 100ml of water (Parkash, 2016).

Composition of salt bridge

The salt bridge solution was prepared according to the methods of (Parkash, 2016) by dissolving 3% agar in 1M NaCl. The solution was first subjected to heat for blending, which in return gave a clear solution of agar solution and was poured into each PVC pipe which was properly sealed with foil paper and was kept at 25^0C for 2hrs for solidification.

Addition of methylene blue as mediator

30mls of mehylene blue was added to the each anode mediator chamber where 10mls was added daily for 72hrs as according to (Zuhri *et al.,* 2016).

Measurement and collection of data calculation

Readings obtained for current, voltage and resistance was obtained.

RESULT AND DISCUSSION

Figures 1-3 shows the bacteria, fungi and yeast population isolated from mud sediment of River Ala at various depths. Microorganisms were isolated at varies depth including control, surface water, mud surface, 50cm, 100cm and 150cm depth. Generally, microbial population of the various depths decreases as the depth increases. ALS has the highest bacteria count of 2.4 x 10^{-5} and AL100cm has the lowest of 0.48 x 10^{-5} while AL100cm had the lowest fungi count of 0.2 x 10^{-6} and ALC (Control) had the highest of 1.6 x 10^{-5}. Also there were no growth observed at depth 150cm for fungi count and in yeast count at depth ALC, AL50, AL100 and AL150. Table1 shows the arrays of bacteria, fungi and yeast isolated from the various depths of surface water to depth 150 cm.

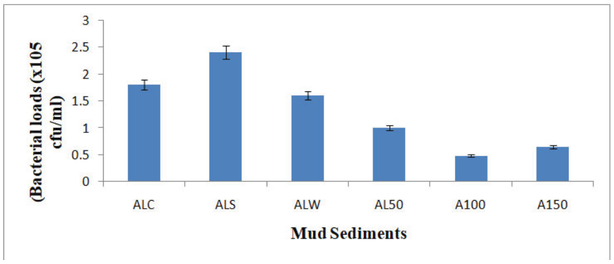

Figure 1: Bacterial load of Ala River at different depths.

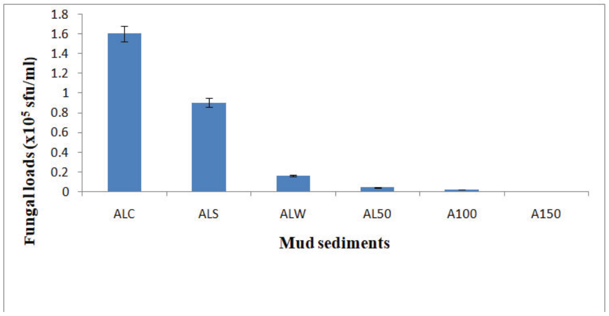

Figure 2: Fungal load of Ala River at different depths.

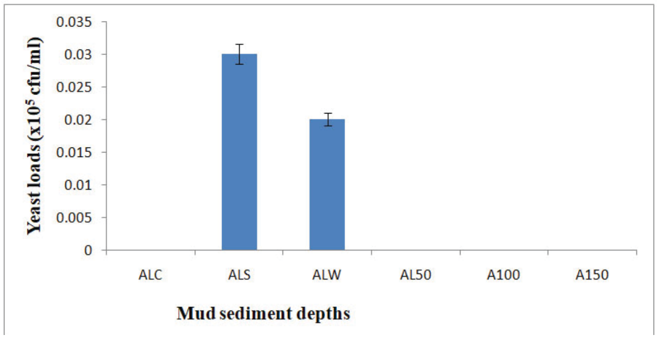

Figure 3: Yeast load of Ala River from different depths
KEYS: ALC- Ala control, ALS- Ala mud surface, ALW- Ala surface water, A50- Ala 50cm depth, A100- Ala 100cm depth, A150- Ala 150cm depth

Table 1: Microorganisms isolated from Mud sediment of River Ala

Depth	Bacteria	Fungi
Control	*Escherichia coli, Staphylococcus aureus*, and *Salmonella cholerasuis*	*Penicillium notatum, Aspergillus niger*
Surface water	*Escherichia coli, Salmonella cholerasuis, Pseudomonas aeruginosa, Enterobacter aerogenes, Staphylococcus aureus, Enterococcus faecalis*	Nil
Surface mud	*Salmonella cholerasuis, Pseudomonas* aeruginosa, *Micrococcus* luteus and *Shigella flexneri*	*Penicillium notatum Trichoderma viride* and *Saccharomyces cerevisae*
Depth 50	*Escherichia coli, Pseudomonas* aeruginosa, and *Bacillus* megaterium,	*Penicillium notatum*
Depth 100	*Staphylococcus* aureus, *Pseudomonas* aeruginosa and *Bacillus* megaterium	Nil
Depth 150	*Erwinia carotovora* and *Clostridium* tetani	Nil

Figure 4 shows the Conductivity, Ionization potential, Organic matter content and Salinity content of River Ala at various depth of surface water, depth 50cm, depth 100cm and 150cm. The pH values of all mud were observed to be slightly acidic to neutral. Ala surface water (W), surface mud (0cm), 50cm, 100cm and 150cm depth records 7.52, 5.59, 5.50, 5.47 and 6.52 respectively. Organic matter content of mud at various depth were quite low, surface water (W), surface mud (0cm), 50cm, 100cm and 150cm depth recorded

2.12, 1.83, 3.69, 2.09 and 3.67 (%) respectively. For the ionization potentials depth 0cm has the highest ionization potential of 52Mev, surface water (W), surface mud (0cm), 50cm, 100cm and 150cm depth recorded 29, 52, 47, 24 and 32 (Mev) respectively. In the Conductivity readings, it was observed that depth 0cm has the highest conductivity and depth 100cm had the lowest. Surface water (W), surface mud (0cm), 50cm, 100cm and 150cm depth recorded 740,172, 119, 209 and 56 (μS) respectively. Salinity content of the various depths shows that surface water has the highest salinity content while depth 150cm had the lowest. Surface water (W), surface mud (0cm), 50cm, 100cm and 150cm depth recorded 359, 114, 189, 107 and 32 (ppm) respectively.

Tables 1 to 6 are showing the readings of current, voltage and resistance obtained from the mediator-less chamber and the mediator chamber across the various mud sediment depths. The current readings generally increase in the first four days of the experiment and decreases as the numbers of days of the experiment increases. Current readings for the mediator microbial fuel chambers were generally slightly higher than the mediator-less chamber. The mediator-less chambers readings ranged from 0.484 to 0.019 (mA) and mediator chambers readings ranged from 0.369 to 0.012 (mA) for surface water to depth 150cm. Plates 1 and 2 are showing the pictures of the microbial fuel cell chambers during the experiment.

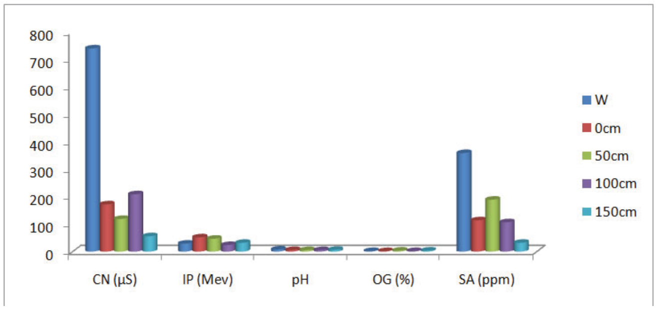

Figure 4: Conductivity, Ionization potential, Organic matter content and Salinity content of River Ala.

Table 2: Current Readings of River Ala at Various Depths

DAYS	AW(mA)	AS (mA)	A50 (mA)	A100 (mA)	A150 (mA)
1	0.0965 ± 0.0185^{a}	0.1103 ± 0.011^{b}	0.1903 ± 0.0574^{a}	0.0535 ± 0.0072^{b}	0.171 ± 0.0451^{a}
2	0.0593 ± 0.011^{a}	0.253 ± 0.1246^{c}	0.037 ± 0.0105^{a}	0.2005 ± 0.1178^{c}	0.0388 ± 0.0098^{a}
3	0.0405 ± 0.0116^{a}	0.1878 ± 0.0031^{d}	0.0975 ± 0.029^{a}	0.2213 ± 0.1077^{d}	0.3285 ± 0.2121^{a}
4	0.0293 ± 0.0044^{a}	0.1845 ± 0.0874^{e}	0.4838 ± 0.4024^{a}	0.041 ± 0.0112^{e}	0.0635 ± 0.0092^{a}
5	0.0575 ± 0.0101^{a}	0.069 ± 0.0085^{f}	0.0775 ± 0.006^{a}	0.021 ± 0.0037^{f}	0.0571 ± 0.0118^{a}
6	0.084 ± 0.0192^{a}	0.0408 ± 0.0145^{g}	0.0833 ± 0.0075^{a}	0.053 ± 0.012^{g}	0.0675 ± 0.0038^{a}
7	0.0668 ± 0.0194^{a}	0.0243 ± 0.0054^{h}	0.0568 ± 0.016^{a}	0.0335 ± 0.0075^{h}	0.0688 ± 0.0195^{a}
8	0.064 ± 0.0202^{a}	0.0458 ± 0.0107^{i}	0.2045 ± 0.1256^{a}	0.0193 ± 0.0053^{i}	0.0678 ± 0.0189^{a}
9	0.092 ± 0.0148^{a}	0.0333 ± 0.0096^{j}	0.1043 ± 0.0029^{a}	0.1223 ± 0.0935^{j}	0.0835 ± 0.0125^{a}
10	0.18 ± 0.1074^{a}	0.1263 ± 0.0744^{k}	0.1043 ± 0.0029^{a}	0.0395 ± 0.0107^{k}	0.2143 ± 0.1019^{a}
11	0.0545 ± 0.0111^{a}	0.086 ± 0.0110^{l}	0.431 ± 0.3197^{a}	0.0793 ± 0.0039^{l}	0.0433 ± 0.0124^{a}
12	0.0295 ± 0.0037^{a}	0.152 ± 0.0791^{m}	0.107 ± 0.0472^{a}	0.2595 ± 0.0886^{m}	0.0848 ± 0.0103^{a}
13	0.023 ± 0.0021^{a}	0.193 ± 0.0588^{n}	0.0911 ± 0.0346^{a}	0.0818 ± 0.0281^{n}	0.0638 ± 0.0145^{a}
14	0.0198 ± 0.0036^{a}	0.228 ± 0.0114^{o}	0.0528 ± 0.0096^{a}	0.0625 ± 0.0251^{o}	0.0775 ± 0.027^{a}

Table 3: Current Readings of River Ala at Various Depths with Methtylene Blue

DAYS	AWM (mA)	AS M (mA)	A50M (mA)	A100M (mA)	A150 M (mA)
1	0.124 ± 0.0144^{a}	0.2208 ± 0.1181^{a}	0.168 ± 0.0327^{b}	0.3685 ± 0.0822^{b}	0.1973 ± 0.0377^{a}
2	0.052 ± 0.0139^{a}	0.0795 ± 0.0142^{a}	0.0698 ± 0.0126^{c}	0.2173 ± 0.0866^{c}	0.065 ± 0.0079^{a}
3	0.052 ± 0.0129^{a}	0.1693 ± 0.0721^{a}	0.3113 ± 0.0573^{d}	0.3478 ± 0.1239^{d}	0.0488 ± 0.01^{a}
4	0.067 ± 0.0078^{a}	0.1998 ± 0.1052^{a}	0.1303 ± 0.0356^{e}	0.0853 ± 0.0057^{e}	0.064 ± 0.0212^{a}
5	0.0913 ± 0.0168^{a}	0.0825 ± 0.0179^{a}	0.0803 ± 0.0076^{f}	0.0775 ± 0.003^{f}	0.077 ± 0.0104^{a}
6	0.0928 ± 0.0088^{a}	0.049 ± 0.0078^{a}	0.0633 ± 0.0144^{g}	0.0703 ± 0.007^{g}	0.0928 ± 0.0064^{a}
7	0.0648 ± 0.0224^{a}	0.1928 ± 0.1003^{a}	0.0855 ± 0.0108^{h}	0.0685 ± 0.0155^{h}	0.0565 ± 0.0119^{a}
8	0.1513 ± 0.0525^{a}	0.177 ± 0.1117^{a}	0.056 ± 0.0171^{i}	0.0535 ± 0.0076^{i}	0.0688 ± 0.0103^{a}
9	0.1448 ± 0.0690^{a}	0.1503 ± 0.0866^{a}	0.2078 ± 0.1099^{j}	0.0588 ± 0.0112^{j}	0.2903 ± 0.1819^{a}
10	0.1755 ± 0.1008^{a}	0.1923 ± 0.1149^{a}	0.028 ± 0.0102^{k}	0.038 ± 0.0111^{k}	0.1318 ± 0.0097^{a}
11	0.1245 ± 0.0847^{a}	0.0915 ± 0.0180^{a}	0.034 ± 0.0064^{l}	0.0693 ± 0.0067^{l}	0.107 ± 0.0166^{a}
12	0.0295 ± 0.0060^{a}	0.13075 ± 0.0454^{a}	0.0243 ± 0.0109^{m}	0.2455 ± 0.0975^{m}	0.0758 ± 0.0121^{a}
13	0.0428 ± 0.0114^{a}	0.029 ± 0.0083^{a}	0.0183 ± 0.0036^{n}	0.037 ± 0.0054^{n}	0.0353 ± 0.003^{a}
14	0.0235 ± 0.0055^{a}	0.019 ± 0.0098^{a}	0.0120 ± 0.005^{o}	0.0178 ± 0.0025^{o}	0.0338 ± 0.0065^{a}

Table 4: Voltage Readings of River Ala at Various Depths

DAYS	AW(V)	AS (V)	A50 (V)	A100 (V)	A150 (V)
1	0.072 ± 0.0072^{a}	0.2135 ± 0.1119^{a}	0.1158 ± 0.0193^{a}	0.1368 ± 0.0185^{a}	0.0458 ± 0.0067^{b}
2	0.1395 ± 0.0808^{a}	0.1823 ± 0.087^{a}	0.0655 ± 0.0091^{a}	0.1321 ± 0.0078^{a}	0.1145 ± 0.0203^{c}
3	0.0683 ± 0.0022^{a}	0.455 ± 0.3584^{a}	0.1713 ± 0.0696^{a}	0.1315 ± 0.0051^{a}	0.1073 ± 0.0211^{d}
4	0.0525 ± 0.0041^{a}	0.089 ± 0.0049^{a}	0.0883 ± 0.0077^{a}	0.0978 ± 0.0105^{a}	0.0778 ± 0.0064^{e}
5	0.0653 ± 0.0072^{a}	0.077 ± 0.0089^{a}	0.0998 ± 0.0095^{a}	0.1308 ± 0.0128^{a}	0.1005 ± 0.0016^{f}
6	0.2625 ± 0.2046^{a}	0.0543 ± 0.0050^{a}	0.087 ± 0.0227^{a}	0.2923 ± 0.1766^{a}	0.0703 ± 0.0034^{g}
7	0.1395 ± 0.0262^{a}	0.0533 ± 0.0046^{a}	0.077 ± 0.022^{a}	0.0763 ± 0.0086^{a}	0.0663 ± 0.0044^{h}
8	0.0888 ± 0.0114^{a}	0.0678 ± 0.0157^{a}	0.3225 ± 0.2033^{a}	0.0613 ± 0.0023^{a}	0.0848 ± 0.0082^{i}
9	0.0905 ± 0.0092^{a}	0.1113 ± 0.0709^{a}	0.1088 ± 0.0034^{a}	0.065 ± 0.003^{a}	0.0868 ± 0.0048^{j}
10	0.2605 ± 0.1686^{a}	0.0778 ± 0.0061^{a}	0.1105 ± 0.0026^{a}	0.0733 ± 0.0046^{a}	0.0995 ± 0.0059^{k}
11	0.332 ± 0.27^{a}	0.0888 ± 0.0089^{a}	0.1078 ± 0.0083^{a}	0.075 ± 0.0049^{a}	0.047 ± 0.0055^{l}
12	0.0413 ± 0.0036^{a}	0.0623 ± 0.0161^{a}	0.1148 ± 0.027^{a}	0.0635 ± 0.003^{a}	0.0573 ± 0.012^{m}
13	0.0373 ± 0.0047^{a}	0.0440 ± 0.0112^{a}	0.0933 ± 0.0174^{a}	0.0438 ± 0.004^{a}	0.0478 ± 0.0097^{n}
14	0.0283 ± 0.0035^{a}	0.0310 ± 0.0060^{a}	0.0655 ± 0.0107^{a}	0.0398 ± 0.0025^{a}	0.038 ± 0.007^{o}

Table 5: Voltage Readings of River Ala at Various Depths with Methylene Blue

DAYS	AW(V)	AS (V)	A50 (V)	A100 (V)	A150 (V)
1	0.0993 ± 0.0359^{a}	0.1278 ± 0.0218^{a}	0.1405 ± 0.0126^{b}	0.172 ± 0.0058^{b}	0.1465 ± 0.0467^{b}
2	0.081 ± 0.0036^{a}	0.074 ± 0.0136^{a}	0.1055 ± 0.0189^{c}	0.1155 ± 0.0105^{c}	0.117 ± 0.0196^{c}
3	0.0725 ± 0.0036^{a}	0.0635 ± 0.0175^{a}	0.197 ± 0.0541^{d}	0.2858 ± 0.1587^{d}	0.067 ± 0.0132^{d}
4	0.0696 ± 0.015^{a}	0.089 ± 0.0122^{a}	0.1043 ± 0.0161^{e}	0.0788 ± 0.0113^{e}	0.0925 ± 0.0129^{e}
5	0.0809 ± 0.0119^{a}	0.074 ± 0.0125^{a}	0.1028 ± 0.015^{f}	0.0858 ± 0.0077^{f}	0.1005 ± 0.0103^{f}
6	0.062 ± 0.0164^{a}	0.067 ± 0.0093^{a}	0.0803 ± 0.0077^{g}	0.0743 ± 0.0049^{g}	0.1158 ± 0.0103^{g}
7	0.061 ± 0.0255^{a}	0.1248 ± 0.0627^{a}	0.086 ± 0.0228^{h}	0.0618 ± 0.0069^{h}	0.0793 ± 0.0125^{h}
8	0.075 ± 0.0076^{a}	0.0775 ± 0.0201^{a}	0.258 ± 0.0759^{i}	0.089 ± 0.0084^{i}	0.0938 ± 0.018^{i}
9	0.0748 ± 0.0178^{a}	0.057 ± 0.0191^{a}	0.124 ± 0.0211^{j}	0.082 ± 0.0123^{j}	0.098 ± 0.0119^{j}
10	0.0763 ± 0.0173^{a}	0.0778 ± 0.0123^{a}	0.0793 ± 0.0123^{k}	0.091 ± 0.0126^{k}	0.1228 ± 0.0199^{k}
11	0.322 ± 0.2748^{a}	0.081 ± 0.0114^{a}	0.0885 ± 0.0425^{l}	0.0753 ± 0.0129^{l}	0.0828 ± 0.0096^{l}
12	0.0265 ± 0.0069^{a}	0.0895 ± 0.0131^{a}	0.062 ± 0.0198^{m}	0.0885 ± 0.0059^{m}	0.077 ± 0.0168^{m}
13	0.0238 ± 0.0049^{a}	0.022 ± 0.0081^{a}	0.0138 ± 0.0036^{n}	0.0188 ± 0.0062^{n}	0.0293 ± 0.0093^{n}
14	0.0158 ± 0.0049^{a}	0.0263 ± 0.0040^{a}	0.0195 ± 0.0042^{o}	0.0258 ± 0.0069^{o}	0.0308 ± 0.0067^{o}

Table 6: Resistance Readings of River Ala at Various Depths

DAYS	AW(Ω)	AS (Ω)	A50 (Ω)	A100 (Ω)	A150 (Ω)
1	2.775 ± 1.2106^{a}	3.2775 ± 1.0501^{a}	2.7725 ± 0.7373^{a}	3.485 ± 0.9807^{a}	4.6425 ± 1.5371^{a}
2	2.09 ± 0.8096^{a}	2.7725 ± 0.7138^{a}	5.4725 ± 1.5767^{a}	2.94 ± 0.9395^{a}	2.92 ± 0.8621^{a}
3	1.8163 ± 0.8421^{a}	3.195 ± 1.0919^{a}	2.345 ± 1.2664^{a}	1.8325 ± 1.0571^{a}	2.705 ± 1.3081^{a}
4	2.435 ± 1.0618^{a}	2.8375 ± 0.9529^{a}	2.1585 ± 0.73^{a}	2.46 ± 0.6688^{a}	2.005 ± 0.7661^{a}
5	3.5775 ± 0.9453^{a}	1.3900 ± 0.3044^{a}	3.1125 ± 0.7218^{a}	3.4475 ± 1.0743^{a}	4.68 ± 0.6428^{a}
6	2.7975 ± 1.3754^{a}	4.1625 ± 1.4944^{a}	3.4425 ± 0.96^{a}	2.9925 ± 1.0692^{a}	4.28 ± 1.535^{a}
7	1.7475 ± 0.9064^{a}	1.3325 ± 0.3597^{a}	1.6675 ± 0.5908^{a}	6.021 ± 1.645^{a}	3.6025 ± 1.3126^{a}
8	2.96 ± 0.9102^{a}	1.8975 ± 0.9224^{a}	3.9625 ± 1.2611^{a}	1.4525 ± 1.0538^{a}	3.6275 ± 1.1905^{a}
9	1.415 ± 0.575^{a}	4.7475 ± 0.9887^{a}	3.535 ± 0.4546^{a}	2.81 ± 1.4552^{a}	4.4575 ± 0.5122^{a}

Table 6: ***(Continued)***

DAYS	AW(Ω)	AS (Ω)	A50 (Ω)	A100 (Ω)	A150 (Ω)
10	1.5575 ± 0.4579^a	1.5775 ± 0.3209^a	3.535 ± 0.4546^a	1.5325 ± 0.2124^a	2.1475 ± 0.7369^a
11	3.075 ± 1.7228^a	2.1775 ± 0.5516^a	4.1975 ± 1.1769^a	3.48 ± 0.5756^a	4.2275 ± 1.5405^a
12	2.470 ± 0.7278^a	3.5375 ± 0.4876^a	3.8525 ± 0.9339^a	2.9975 ± 0.8167^a	4.1875 ± 0.7503^a
13	2.4425 ± 0.7737^a	2.9400 ± 0.300^a	4.0975 ± 0.8334^a	2.0625 ± 0.2319^a	3.405 ± 0.8101^a
14	2.3325 ± 0.7425^a	2.6375 ± 0.0487^a	3.2475 ± 0.978^a	1.8475 ± 0.1918^a	2.965 ± 0.6923^a

Table 7: Resistance Readings of River Ala at Various Depths with Methylene Blue

DAYS	AW(Ω)	AS (Ω)	A50 (Ω)	A100 (Ω)	A150 (Ω)
1	3.510 ± 0.2031^a	4.2375 ± 1.0205^a	3.19 ± 0.9223^a	2.600 ± 0.2755^a	7.7625 ± 3.0939^a
2	3.9375 ± 1.4361^a	1.841 ± 0.6430^a	2.96 ± 0.9743^a	3.4783 ± 1.137^a	4.06 ± 0.8061^a
3	4.7788 ± 1.5812^a	2.0575 ± 0.8292^a	2.727 ± 1.3632^a	2.5503 ± 0.9583^a	3.6375 ± 0.7737^a
4	2.791 ± 0.9614^a	3.2775 ± 1.1553^a	3.49 ± 1.1847^a	2.9975 ± 0.5716^a	5.325 ± 1.9823^a
5	3.11 ± 0.6728^a	2.2378 ± 0.7340^a	3.57 ± 1.4859^a	7.1525 ± 3.0306^a	5.725 ± 0.445^a
6	3.415 ± 0.7762^a	1.3365 ± 0.6212^a	2.853 ± 0.9343^a	3.665 ± 1.3321^a	4.005 ± 0.1281^a
7	4.055 ± 1.6806^a	4.5675 ± 2.3006^a	2.2413 ± 1.2648^a	3.855 ± 1.9047^a	3.1375 ± 1.0006^a
8	3.422 ± 0.8564^a	2.0075 ± 0.8030^a	3.9458 ± 1.5173^a	3.8225 ± 1.233^a	6.605 ± 1.8979^a
9	2.1525 ± 0.922^a	3.7900 ± 1.5820^a	2.265 ± 0.6855^a	2.1375 ± 0.856^a	3.805 ± 0.4768^a
10	3.3375 ± 0.8749^a	2.6325 ± 0.3782^a	4.1875 ± 1.13^a	3.211 ± 0.552^a	4.17 ± 0.694^a
11	3.13 ± 1.2876^a	4.025 ± 0.0035^a	7.4325 ± 2.2224^a	3.675 ± 0.6159^a	5.135 ± 1.2582^a
12	4.431 ± 0.8472^a	2.5325 ± 0.4909^a	4.1255 ± 2.9233^a	5.4525 ± 1.3931^a	4.7925 ± 0.9664^a
13	3.311 ± 0.8173^a	4.1050 ± 1.7632^a	4.5 ± 1.4407^a	5.41 ± 0.9966^a	2.8475 ± 0.8689^a
14	3.703 ± 1.3119^a	3.1618 ± 1.4330^a	3.826± 1.4704^a	4.75 ± 0.5521^a	2.5245 ± 1.6328^a

KEYS: AW-Ala surface water, AS- Ala mud surface, A50- Ala depth 50cm, A100- Ala depth 100cm and A150- Ala depth 150cm; AWM-Ala surface water with methylene blue, ASM- Ala mud surface with methylene blue, A50M- Ala depth 50cm with methylene blue, A100- Ala depth 100cm with methylene blue and A150- Ala depth 150cm with methylene blue.

Values followed by the same letters (s) on the same column are not significantly different (P≤ 0.05). Each value represents a mean of four reading

Pictures of the microbial fuel chamber during the experiments

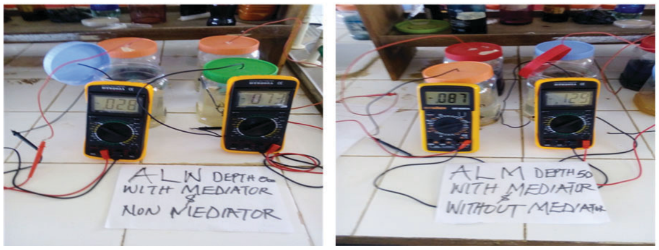

Plate 1: MFC Chamber showing River Ala mud surface and depth 50 cm with mediator and without mediator

Keys: ALWO- Ala mud surface

ALM50- Ala Depth 50 cm

Plate 2: MFC Chamber showing River Ala depth 100 cm and depth 150 cm with mediator and without mediator.

Keys: ALM100- Ala Depth 100 cm

ALM150 cm- Ala Depth 150 cm

Microorganisms are used in MFC to convert organic and inorganic compounds into bioelectricity (Manohar and Mansfeld, 2009). Mud sediments from River Ala was use to generate electric current using microbial fuel cell, the sediment was oxidize by bacteria under anaerobic condition in the anode chamber generating protons and electrons. Microbial population of the various depths decreases as the depth increases (Figure 1 and 2). The decrease in the aerobic bacteria population down the depth might be due to oxygen retention is lower at the lower depths which only permit the growth of only anaerobic organisms. Fungal growth was not observed at the observed at the lower depths of the various river sediments, these findings agrees with findings of (Reddy *et al.,* 2000) who reported that aerobic microbial populations are restricted to zones where oxygen is available and that aerobic organisms become quiescent or die and new inhabitants, largely facultative and obligate anaerobic bacteria take over. (Fischer *et al*., 2002) concluded that Bacterial abundance generally decreases with sediment depth independent of the method used. Majorities of these microorganisms are faecal contaminates which might have accounted for the findings of (Jamieson *et al.,* 2004) that bacteria often show an affinity for sediment attachment as sediments represent a beneficial environment for nutrient, food assimilation and protection from environmental stress such as contaminants and predation. pH of the mud sediments at various depths were slightly acidic to neutral pH. Sediment is also the major site for organic matter decomposition which is largely carried out by bacteria. pH is extremely important, since most of the chemical reactions in aquatic environment are controlled by any change in its value. Anything either highly acidic or alkaline would kill aquatic life. Aquatic organisms are sensitive to pH changes and biological treatment requires pH control (Abowei and Sikoki, 2005). High conductivity and salinity observed at the surface water than other depth might be that more ions that are present at the depth that led to its high conductivity and salinity is a good contributor to salinity. The higher current and voltage generation observed at the early stage of the experiment might be as a result the microorganisms could still get enough organic matter to metabolize in the anaerobic digestion to produce proton and electrons at the anode which at the later weeks the nutrient depleted and causes its reduction as according to the findings of (Pavan *et al.,* 2015) who analyses energy harvested from Kitchen Waste through Two-chamber Microbial Fuel Cell. (Parkash, 2016) in his findings on characterization of voltage and current generated from cow dung using double chambered MFC observed that was a definitive increase in the generated current and voltage from day 1 to day 5 and then a decline in trend is observed from the day 6 downward. The resistance generated by the microbial fuel chamber that was very high might have contributed to the low current and voltage observed which aggress with (Menicucci *et al*., 2006) who revealed that cell voltage of MFC decreases when external resistance increases. (Samrot, *et al.,* 2010) also input that MFCs with lower external resistances resulted in higher anode potentials. Methylene blue mediated MFC current and voltage generated was higher in general observation than mediator-less which is because they are aided by the addition of the methylene which busts their current generation potential which is in accordance with the findings of (Rahim Nejad et al., 2011) on methylene blue as electron promoters in microbial fuel cell. Most microbes are electrochemically inactive because the proteins associated with electron transport are contained within the cell membrane. Mediators can be used to facilitate the transfer of electrons from the microbial membrane to the MFC electrode for these microbes (Kim, *et al.,* 2005). Mediators are preferentially reduced during the metabolic oxidation of organic materials, and the reduced form of the mediator is then re-oxidized at the working electrode (anode), which is maintained at a sufficiently high electric potential. Nearly any bacterium can be used to generate current in a mediated MFC.

CONCLUSION

Mud sediment for the use of generating electricity has proven to be one of the promising technologies through the use of microbial fuel cell and the use of mediator which help to facilitate non electrogenic bacteria to generate electric current; MFC had also proven to be a good cheap alternative to the use of fossil fuel for power generation.

REFERENCE

1. Levin, M. and Stevenson, C. 2012. Regulation of Cell Behavior and Tissue Patterning by Bioelectrical Signals: Challenges and Opportunities for Biomedical Engineering. *Annual Review of Biomedical Engineering* 14:295-323
2. Brandt, E., Adam R, and Farrell, A. 2007. Scraping the bottom of the barrel: CO2 emission consequences of a transition to low-quality and synthetic petroleum resources, *Clim. Change* 84:241-63
3. Faraghi, N. and Ebharimi, S. 2012. Nitrite as a candidate substrate in microbial fuel cells. *Biotechnology Letters*., 34:1483–1486
4. Rabaey, K., Sompel, V. K.; Maignien, L., Boon, N., Aelterman P. and Clauwaert, P. 2006. Microbial fuel cells for sulfide removal. *Environmental Science & Technology*, 40:5218–5224. (Rabaey *et al*., 2006; Faraghi *et al*., 2012).4;5
5. Logan, B. E., Hamelers, B., Rozendal, R. A., Schrorder, U., Keller, J. and Freguia, S. 2006. Microbial fuel cells: methodology and technology. *Environmental Science and Technology.* 40:5181–5192
6. Sharma, Y., and. Li., B. 2010.Optimizing energy harvest in wastewater treatment by combining anaerobic hydrogen producing biofermentor (HPB) and microbial fuel cell (MFC). *International Journal of Hydrogen Energy*.35: 3789 – 3797
7. Higgins, S.R., Lopez, S. J., Pagaling, E., Yan, T. and Cooney, M. J. 2013. Towardsa hybrid anaerobic digester-microbial fuel cell integrated energy recovery system: An overview of the develop-

ment of an electrogenic biofilm. *Enzyme and Microbial Technology*, 52:344– 351

8. Davis, J.B., and Yarbrough, H.M. 1962. Preliminary experiments on a microbial fuel cell. *Science* 137: 615-616.
9. Moon, H., Chang, I.S. and Kim, B.H. 2006. Continuous electricity production from artificial wastewater using a mediator-less microbial fuel cell. *Bioresource Technology* 97: 621-627.
10. Wilkinson, S. (2000) "Gastrobots benefits and challenges of microbial fuel cells in food powered robot applications. *Autonomous Robots* 9: 99−111.
11. Gil, G.C., Chang, I.S., Kim, B.H., Kim, M., Jang, J.Y., Park, H.S. and Kim, H.J. 2003. Operational parameters affecting the performance of a mediatorless microbial fuel cell. *Biosensensor and Bioelectronics* 18: 327-334.
12. Park, D., Kim, S., Shin, I. and Jeong Y. 2000. Electricity production in biofuel cell using modified graphite electrode with neutral red. *Biotechnol Lett*; 22:1301.
13. Du, Z.; Li, H. and Gu, T. 2007. A state of the art review on microbial fuel cells: A promising technology for wastewater treatment and bioenergy. *Biotechnology Advances* 25: 464-482.
14. Logan, B. E. 2009. Exoelectrogenic bacteria that power microbial fuel cells. *Nature Reviews Microbiology* 7:375-381.
15. Park, D. and Zeikus, J. 2000. Electricity generation in microbial fuel cells using neutral red as an electronophore. *Appl EnvironMicrobiol*; 66:1292
16. Skotnikov, A. 1998. Automated unit for soil sample preparation and processing. *Soil Sci. Plant Anal.*, 29(11–14): 2015–2033
17. Bhargava, B.S. and Raghupathi, H. B. 1993. Analysis of plant materials for macro and micronutrients. In H.L.S. Tandon, ed. Methods of analysis of soils, plants, waters and fertilizers, pp. 49–82. New Delhi, FDCO
18. Parkash, A. 2016. Utilization of Distillery Wastewater for Electricity Generation Using Microbial Fuel. *J. Appl. Emerg. Sci.*, 6(2).
19. Zuhri, F., Arbianti, R., Utami, T. and Hermansyah, H. 2016. Effect of methylene blue addition as a redox mediator on performance of microbial desalination cell by utilizing Tempe wastewater. *International Journal of Technology* 6: 952-961
20. Manohar, A. and Mansfeld, F. 2009. The internal resistance of a microbial fuel cell and its dependence on cell design and operating conditions. *Electrochimica Acta.*54:1664 – 1670.
21. Reddy, K. R., D' Angelo, E. M, and Harris, W. G, 2000. Biogeochemistry of wetlands. In: Summer ME (ed) *Handbook of Soil Science*, pp. G89- G119. Boca Raton, FL: CRC Press
22. Fischer, H., Wanner, S. and Pusch, M. 2002. Bacterial abundance and production in river sediments as related to the biochemical composition of particulate organic matter (POM). *Biogeochemistry* 61:37-55.
23. Jamieson, R., Gordon, R., Joy, D. and Lee, H. 2004. Assessing microbial pollution of rural surface waters: a review of current watershed scale modeling approaches. *Agric Water Manage.* ; 70:1–17.
24. Abowei, J. F. and Sikoki, F. D. 2005. Water Pollution Management and Control, Double Trust Publications Company, Port Harcourt; ISBN: 978-30380-20-16, pp: 236
25. Pavan, S., Kamble, R. Bondre, A., Sumit, L. and Satish, H. 2015. Sathawane Energy Harvesting from Kitchen Waste through Two-chamber Microbial Fuel Cell. *International Journal of dvanced Research in Science, Engineering and Technology*, 2, Issue 11
26. Menicucci, J., Beyenal, H., Marsili, E.; Veluchamy, R. A. and Demir, G. (2006). Procedure for determining maximum sustainable power generated by microbial fuel cells. *Environ Sci Technol*., 40: 1062-1068.
27. Samrot, A., Senthikumar, P., Pavankumar, K., Akilandeswari, G., Rajalakshmi, N. and Dhathathreyan, K. 2010. Electricity generation by *Enterobacter cloacae* SU-1 in mediator less microbial fuel cell. *International Journal of Hydrogen Energy*.35: 7723 – 7729.
28. Rahimnejad, M., Najafpour, G. D., Ghoreyshi, A. A. Shakeri, M. and Zare, H. 2011. Methylene blue as electron promoters in microbial fuel cell. *International Journal Of Hydrogen Energy*. 36:13335 – 13341
29. Kim, J. R.; Min, B. and Logan, B. E. 2005. Evaluation of procedures to acclimate a microbial fuel cell for electricity production, *Appl. Microbiol. Biotechnol.*, 68(1), pp. 23–30, 2005.