IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)
e-ISSN: 2279-0853, p-ISSN: 2279-0861.Volume 18, Issue 10 Ser.5 (October. 2019), PP 60-67
www.iosrjournals.org

Enhancement of Bioelectricity Production using Different Chamber Design, Catholytes and Varied Electrode in MFCS

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Abstracts: *Bioelectricity generation through Microbial Fuel Cells (MFCs) technology is an alternative source of energy generation to fossil fuel which has been shown to have negative environmental impact. Microbial degradation of biomass could possibly be the most renewable means of generating electrical and thermal energy. This study was designed to evaluate different factors to enhanced bioelectricity generation in MFCs which include: architectural design of MFCs chamber, selected catholytes, and different local with foreign electrode, different mediators and the use of different proton exchange membrane for bioelectricity generation. Different yeast species were isolated from cassava waste water, whey waste water, human urine and rabbit dung using spread plate method. These isolates were identified using Analytical profile index (API). Results obtained revealed the identity of the isolated yeast species as Candida famata, Candida hellenical. Candida tropicalis, and Saccharomyces cerevisia (using API method). Out of the four wastes used, cassava processing wastewater gave the highest bioelectricity potential and was subsequently used as substrate for further study. Among different types of electrode evaluated, galvanized plate exhibited the highest potential for bioelectricity generation with 936.53mV. There was no significant difference (p-value>0.05) between galvanized plate and an imported electrode (graphite rod) in their ability to generate electricity. There was also no significant difference (P value >0.05) in the voltage generated using different catholytes (potassium ferric cyanide 932.54mV, Methylene blue 925. 23mV and neutral red 936.53mV).*

Key words*: Bioelectricity, Catholytes, Electrode, Microbial fuel cell (MFC) and Renewable biomass*

Date of Submission: 23-09-2019 Date of Acceptance: 12-10-2019

I. Introduction

According to the World Energy Council (2013), fossil fuels combustion causes a lot of damage to the climate, environment and human health, and this has given rise to unbearable inequalities, wars and political tensions at international levels. The current climate change threat by greenhouse gas emissions from the combustion of fossil fuels has necessitated a search for alternative non-polluting, reliable, renewable and sustainable sources of energy such as solar energy and its derivatives. The use of biomass in the generation of bioelectricity through Microbial Fuel Cells (MFCs) technology could possibly be the most renewable means for producing electrical and thermal energy. This technology is cable of using organic wastes of domestic or industrial origin for the production of renewable energy by converting the chemical energy in the bonds into electrical energy. Renewable biomass may therefore, become an important source of electricity in future, for therein exist the possibility of extracting electric current from a wide range of soluble or dissolved complex organic wastes and renewable biomass (Singh *et al.,* 2010). The benefits of bio-fuel are enormous, which include sustainable and renewable fuels, decrease carbon dioxide in the atmosphere and turning the problem of waste into source of energy.

Essentially any organic material can be used as electron donors in the microbial fuel cell technology. Microbial Fuel Cell technology could be installed to waste water treatment plants. The active microorganisms in the anaerobic anode compartment of such MFC would utilize waste materials in the treatment plant to generate bioelectricity for the plant. There are many advantages to be derived from this technology such as; clean and an efficient method of energy production; it is a more efficient way of energy combustion than the standard combustion engines (Kim *et al*., 2006). However, it is a challenging technology because of its current situation of comparatively low energy output but with high cost input. The utilization of noble metal catalyst and ion exchange membrane contributes to the large proportion of this MFC configuration expense (Du *et al.,* 2011).

In Microbial Fuel Cells (MFCs), the nature of substrates to be used is very important. The quality of substrate determines the rate of bioelectricity generation (Logan *et al.,* 2012). MFCs rely on living biocatalysts to facilitate the movement of electrons through the system instead of the traditional chemically catalyzed oxidation of a fuel at the anode and reduction at the cathode. The anode-respiring microorganism is able to oxidize substrates (organic pollutants), releasing the chemically bonded energy in the form of electric energy. Microbial fuel cell (MFC) technology is a new opportunity for a sustainable production of electrical energy by converting chemical energy to electrical energy by biodegradation of biomass using microorganisms as biocatalysts (Moqsud *et al*., 2007). In MFCs, microorganisms oxidize organic compounds and produce electrons at the anode which pass through the external circuit to the cathode where the final electron acceptor is reduced. It is known that many prokaryotic microorganisms could utilize oxygen for their growth through respiratory pathways in aerobic conditions or through fermentative pathways in anaerobic conditions. Yeast can grow in both aerobic and anaerobic environments and are veritable instrument for bioelectric generation because they are easily activated, inexpensive and do not need sterile conditions because they tolerate different environmental conditions (Walker *et al.,* 2006).

II. Materials and Methods

Four different samples (whey waste water, cassava waste water, human urine and rabbit dung) were used for this study. Whey waste water was collected from a Fulani settlement at Aduramigba in Osogbo, cassava processing waste water was collected from a local fufu producer at Ofatedo in Osogbo, human urine was from a student donor in Osogbo and rabbit dung was collected from an animal house at Mercy-land unit of LAUTECH campus, Osogbo.

One (1) gram of the rabbit dung was weigh into a 10ml universal bottle, emulsified with 5ml sterile distilled water and made up to 10ml with sterile distilled water to form the stock solution. Ten millimeters (10 ml) each of human urine, cassava waste water and whey waste water were put into different universal bottles and label appropriately to serve as stock solutions for each of these samples. The stock sample solutions were serially diluted to the power of 10^{-7} dilution and plated on potato dextrose agar using spread plate method. Chloramphenicol antibiotics were added to the medium to inhibit bacterial growth. The plates were incubated at 37 ºC for 24h. Organisms with different cultural morphological features were selected and sub-cultured on separate plates to obtain pure cultures. Isolated organisms were maintained on PDA slant at 4^0C and later identified with Analytical profile index (API).

There are different possible configurations for a microbial fuel cell chamber. In this study however, two different designs of a two chamber MFC were used, according to the design of Ghasemi *et al*. (2011). It consisted of two 250 ml bottles, connected by a tube containing a cation exchange membrane (Salt Bridge) as shown on (Plate 1). Spent battery rods were used as electrodes both at the cathode and anode units. The constructed chamber was used for bioelectric generation and its performance noted. In the second type of microbial fuel chamber constructed, a proton exchange membrane was used to link the two subunits together instead of the PVC salt bridge used in the first design. For this design, two 100 ml bottles were connected with Nafion 117 (proton exchange membrane) instead of salt-bridge used in the first design (Plate 2). The proton exchange membrane (Nafion 117) was pre-treated before being used. It was boiled in 30% hydrogen peroxide, washed in de-ionized water and 0.5 M H_2So_4 and stored in de-ionized water until used.

Also, the same waste materials (Whey wastewater (WW), Human urine (HU), Cassava processing water (Cass) and Rabbit dung (RT)) were screened as probable substrate to be used in this study. The electrodes in the two subunits were of great importance in the chamber connected with copper wire, then join to a resistor to complete the circuit. Three local electrodes and one foreign electrode were compared. The cathode subunit of the MFC system contained electron acceptors (catholytes). The effect of different catholyte materials was evaluated. The three catholytes investigated were; methylene blue, neutral red and potassium ferric- cyanide. These materials were used differently as catholyte in an MFC and the rate of energy generation monitored (Table 1).

The anode subunit of the designed MFC with Nafion 117 proton exchange membrane also contained materials known as mediators (electron donors). Two percent (2%) solution of sodium acetate and phenol solution were assessed as possible mediators (electron donors) at the anode subunit. These two solutions were used differently as mediator materials in an MFC system to determine their effect on bioelectricity generation.

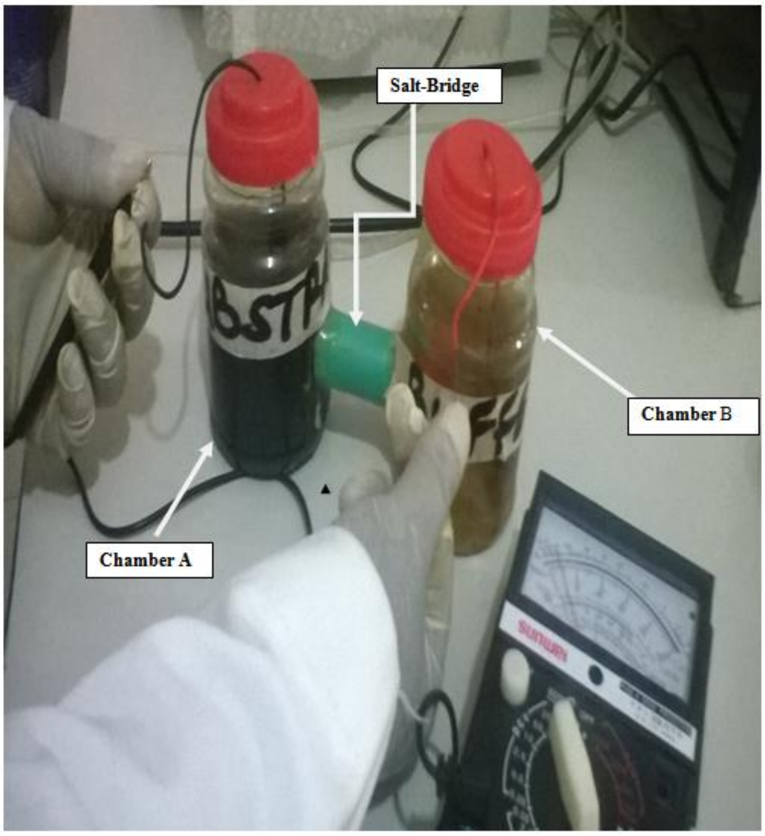

Plate 1: Two Chamber Microbial Fuel Cells separated by Salt-bridge

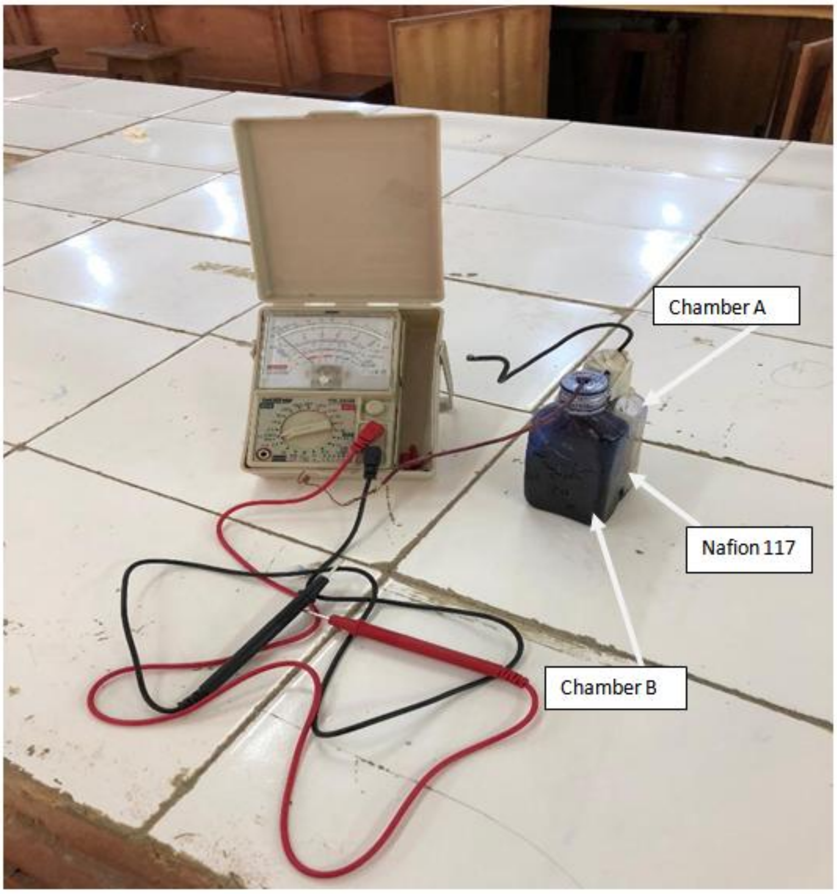

Plate 2: Two Chamber Microbial Fuel Cells separated by Nafion 117

III. Results

Table 1: Morphological, cultural and biochemical characteristics of yeasts isolates

Characteristic	**Yeast isolates**			
	1	**2**	**3**	**4**
Colour	Cream	Cream	Cream	Cream
Shape	Oval	Oval	Oval	Oval
Germ tube test	+ve	+ve	+ve	+ve
Lactophenol blue	+ve	+ve	+ve	+ve
Urease	+ve	+ve	-ve	-ve
Catalase	+ve	+ve	-ve	+ve
Oxidase	-ve	-ve	+ve	+ve
Sugar Fermentation				
D-Galactose	+	+	+	+
Cycloheximide	_	_	+	+
D-Saccharose	+	+	+	+
N-Acetyl-Glucossamine	_	+	+	+
Lactic acid	+	+	+	+
L –arabinose	_	+	+	_
D-Celiobiose	_	+	+	+
D-Raffinose	+	+	+	_
D-Maltose	+	+	+	+
D-Trehalose	+	+	+	+
Potassium 2-Keto Gluconate	_	+	_	+
Methyl α D- Gluconate	+	+	_	+
D Sorbitol	_	+	+	+
XYL	_	+	+	+
D- Ribose	_	_	+	_
Glycerol	_	+	+	_
L-Rhamnose	_	+	+	_
Palatinose	+	+	+	+
Erythritol	_	+	_	_
D-Melibiose	_	_	_	_
Sodium Glucuronate	_	+	+	_
D-Melezitose	_	+	_	+
Potassium Gluconate	_	+	+	+
Levulinic acid	_	_	_	_
D- Mannose	_	+	+	+
D- lactose	_	+	_	_
D-innositol	_	_	+	_
Glucosamine	+	+	+	+
Escolin ferric citrate	_	+	+	_
GLN		+	+	+
Probable identity	***Saccharomyces Cerevisiae***	***Candida famata***	***Candida hellenica***	***Candida Tropicalis***

When wastes were screened for probable substrate to be used in the study, Cassava processing water (Cass) has the highest potential, generating 0.890mV of electric energy. Whey waste water, rabbit dung and human urine generated 0.645mV, 0.644mV and 0.600mV of electric energy respectively with the use of salt-Bridge as proton exchange membrane. Subsequent experiments were done using cassava waste water. As shown in this figure, the rate of energy generation decreased with increase in the fermentation time.

Microbial fuel cells (MFCs) of two different architectural designs were evaluated for their bio-energy generation in this study. The two MFC designs investigated were; (a) microbial fuel cell with salt-bridge (Figure 1) and (b) microbial fuel cell with Nafion 117 as (PEM) (Figure 2**).** Result obtained shows that the structural properties of an MFC affect its overall performance. The highest bioelectric energy generated by the first design was 0.891V while the second design generated 1.074V of electrical energy. Subsequent experiments were therefore conducted using the modify design.

Records have shown that the type of electrode used in the construction of an MFC affects the rate of bioelectricity generation of the system. In this study, four electrode materials (graphite rods (imported and served as control), and aluminum plate, galvanized plate and zinc plate (all locally procured) were investigated to determine their potentials in bioelectricity generation. Among the locally procured materials assessed, galvanized plate electrode elicited the highest bioelectricity generation potential (926.67V) while zinc had the least with 285.89V. This finding was true irrespective of the organisms used as inoculums in the process (Tables 2 – 4). A comparison of the control (graphite rod) with galvanized plate shows that there was no significant difference between them (p value > 0.05)

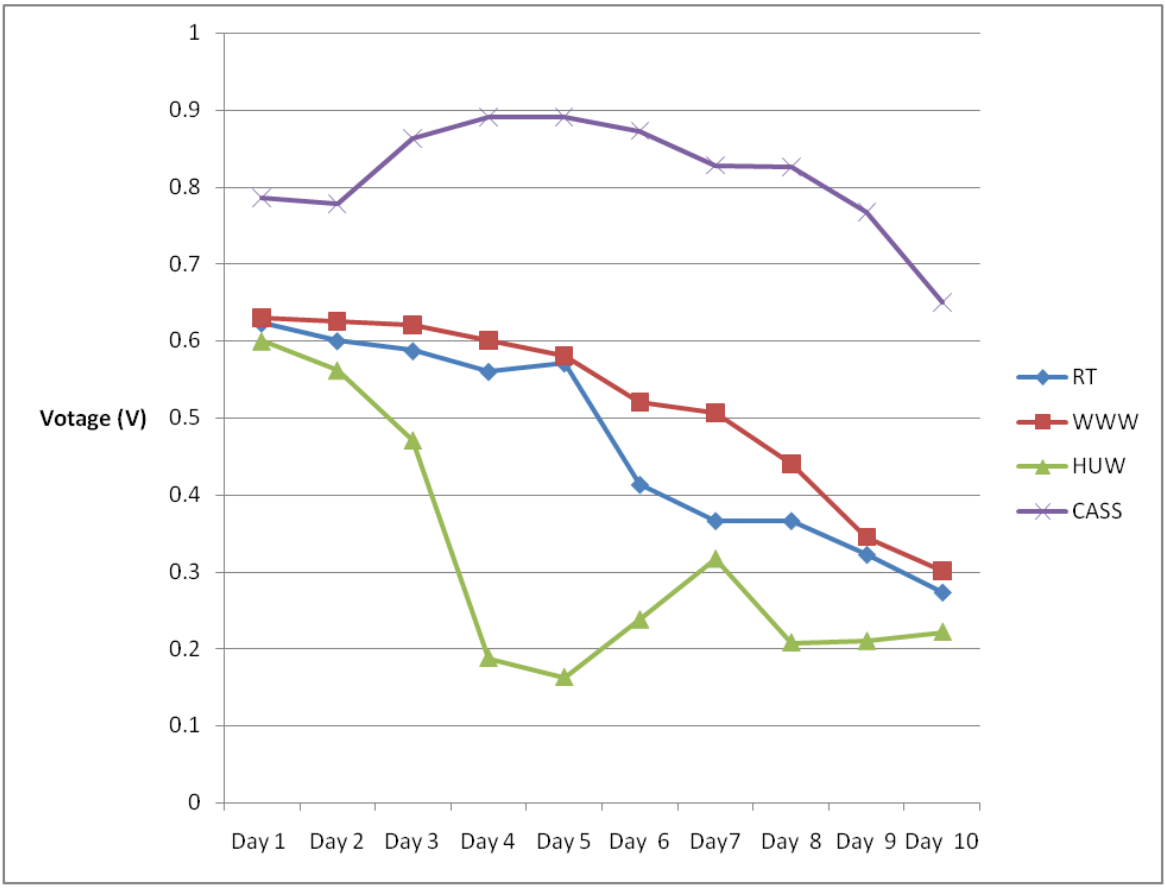

Figure 1: Screening for probable substrate from different waste using MFC Design with Salt bridge

Key:-
RT- Rabbit Dung, HUW-Human urine,
CASS- Cassava processing waste water and WWW- Whey wastewater

Table 2: Generation of bioelectricity with Potassium Ferric cyanide as catholyte

Electrode	*Geotrichum candidum*	*Saccharomyces cerevisiae*	*Candida tropicalis*	Water
Zinc electrode +Acetate	285.89±141.67	625.87±230.18	437.10±202.91	150.14±17.85
Zinc electrode+ phenol	766.30±230.73	587.81±212.72	575.41±210.46	115.37±11.26
0Galvanized electrode +phenol	929.67±201.07	745.47±219.47	764.79±232.79	117.76±7.44
Galvanized electrode +Acetate	916.74±205.65	932.54±216.37	902.19±206.30	280.44±157.22
Alum. Electrode +Acetate	608.34±193.87	479.30±189.67	399.83±183.90	107.53±12.23
Alum. Electrode +Phenol	660.66±214.35	674.06±220.38	661.91±215.12	277.93±277.93
p–value	0.237	0.762	0.523	0.629

Table 3: Generation of bioelectricity with Methylene blue as catholyte

Electrode	*Geotrichum candidum*	*Saccharomyces cerevisiae*	*Candida tropicalis*	Water
Zinc electrode +Acetate	268.43±151.71	312.76±176.47	442.26±204.09	126.99±9.59
Zinc electrode+ phenol	739.66±224.32	709.24±214.46	723.30±214.52	119.66±15.04
Galvanized electrode +phenol	778.33±229.62	774.44±235.85	925.23±208.96	125.13±7.28
Galvanized electrode +Acetate	817.47±237.60	766.31±227.15	789.46±233.98	286.66±160.75
Alum. Electrode +Acetate	608.39±192.94	463.499±193.19	532.29±204.55	142.86±52.01
Alum. Electrode +Phenol	707.86±227.04	696.10±220.38	701.41±2241.53	281.94±181.69
p–value	0.422	0.564	0.658	0.677

Table 4: Generation of bioelectricity with Neutral Red as catholyte

Electrode	*Geotrichum candidum*	*Saccharomyces cerevisiae*	*Candida tropicalis*	**Water**
Zinc electrode +Acetate	149.31±9.10	459.59±204.76	758.86±224.99	136.44±19.25
Zinc electrode+ phenol	591.61±212.27	584.90±221.61	757.06±217.07	127.65±16.11
Galvanized electrode +phenol	623.31±235.24	748.67±229.78	741.97±222.50	145.37±23.30
Galvanized electrode +Acetate	753.19±222.93	936.53±215.77	767.60±227.90	135.04±21.08
Alum. Electrode +Acetate	521.84±193.26	519.80±194.23	550.39±186.70	99.77±5.98
Alum. Electrode +Phenol	701.64±218.11	875.89±208.49	737.43±227.91	103.09±1.18
p –value	0.335	0.526	0.980	0.325

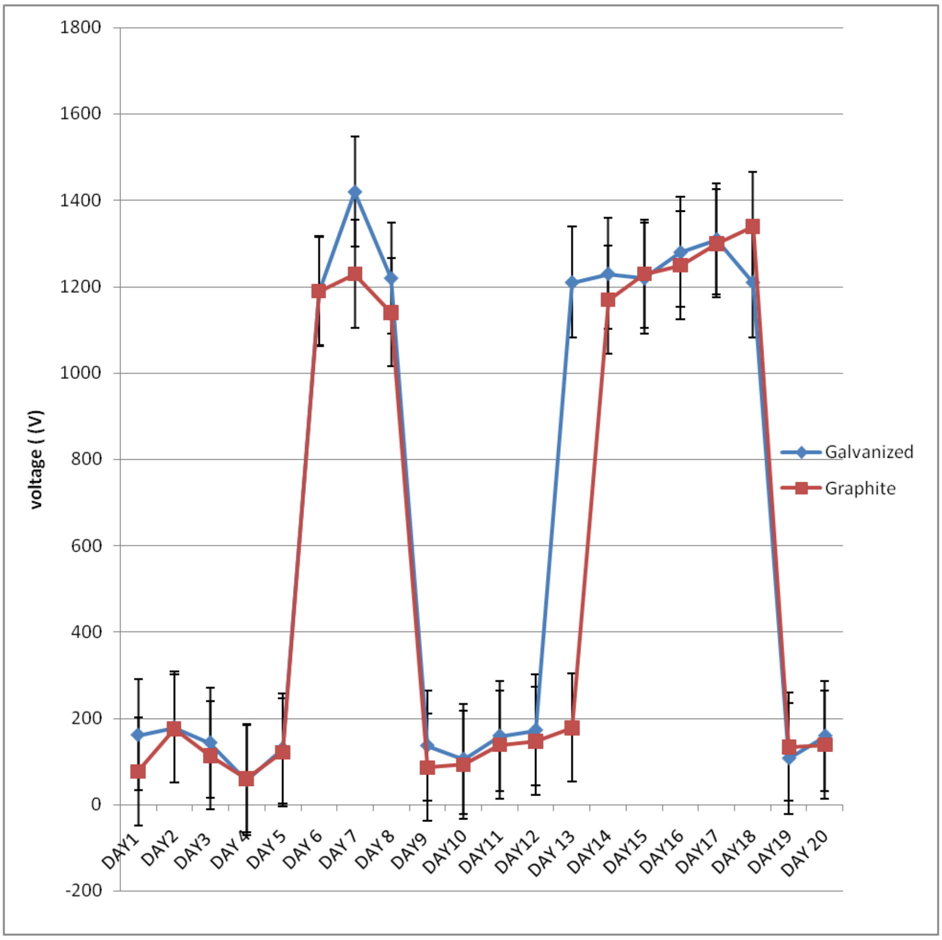

Figure 2: Comparison of two electrodes in MFC's design with Nafion 117 using *Saccharomyces cerevisiae* as inoculums.

Key: Red color - Graphite electrode (Foreign Electrode) Blue color- Galvanized electrode (Local electrode)

Three different dyes were assessed as catholytes in this study, the result obtained revealed that statistically, there were no significant differences ($P > 0.05$) among the three catholytes as shown in Tables 2, 3 and 4 as shown above. Two mediating agents (phenol and acetate) were evaluated for their ability to support the generation of electrical energy in an MFC system. Phenol elicited a higher energy generation than acetate (Tables 2, 3 and 4) above.

IV. Discussion

Using both phenotypic and molecular tools, the identities of the different yeasts isolated from the waste waters used in this study were revealed as *Candida tropicalis, Sacharomyces cerevisiae, Candida hellenicas* and *Candida famata*. However, similar isolates (*Candida tropicalis*, *Sacharomyces cerevisiae* and *Candida albicans*) were isolated by Omemu *et al.* (2007) from waste water and fermented maize.

. Yeasts generally have been implicated in microbial fuel cells as biocatalysts because they are nonpathogenic organisms, easily handled and robust with a good tolerance in different environmental conditions. It is therefore not suprising that the yeast isolates in this study exhibited ability to generate bioelectricity. The report of Mardiana *et al*. (2016) who described how Saccharomyces cerevisiae through oxidation in the presence of Methylene blue generated a maximum current density of 2.3mA /m-2 per cell collaborates the result of this work.

Evaluation of whey waste waste, rabbit dungs, cassava processing waste water and human urine as substrate for microbial generation of bioelectricity reveals that cassava waste water has great potential for bioelectricity generation. Cassava is a root crop that is very rich in carbohydrate, it is therefore believed that its waste water will be rich in carbohydrate based compounds which could have served as carbon source for the growth of the microbial cells. This could possibly account for the high electrical energy generated by it compare to the other wastes used in this study. The finding in this study is in agreement with the report of Olaoye *et al*. (2018), Agarry *et al.* (2016) and Kaewkannetra *et al.* (2011) who used cassava waste water to generate bioelectricity. They recorded maximum bioelecvtricity yield of 0.750mV, 1.771mV, and 275mv respectively in there studies using cassava waste water as their substrates. Adekunle and Raggavan (2017) also used cassava peels extract as substrate in an MFC and generated maximum voltage of 0.687mV.

In this study, an MFC design with agar salt bridge as the electron exchange membrane generated 0.891mV of electrical energy, while the design with Nafion 117 generated 1.074V of electrical energy. This is a significant improvement over what had been reported in a similar study by shahi and Singh (2017) who generated 0.145mV with current of $0.05A/m^2$ and 0.504mV with current density of $0.1A/m^2$ using the two designs as used in this study respectively.

Result obtained in this study on the effect of different catholyte materials (Methylene blue, neutral red and potassium ferric cyanide) on the rate of generation of electricity show that there is no significant difference among them. However, Gunawardena *et al.* (2008) observed improved performance in the rate of electricity generation when Methylene blue and potassium ferric cyanide were used as electron mediators, generating an open circuit voltage of 383.6 ± 1.5mV.

Assessment of different local materials as electrodes in the generation of bioelectricity using microbial fuel cell technology shows that galvanize plate has a high potential, generating 932.54V of electrical energy. This compares favourably with an imported material (graphite rod) which generated 936 34V of energy. In a similar study, Wudneh (2016) evaluated the performance of POCO3, HKO6 and G347 electrodes in the generation of electrical energy with a generation of maximum bioelectricity energy of 540mV from POCO3 electrode.

V. Conclusion

This study has demonstrated that different yeast species and different agricultural waste materials could effectively be deployed in the production of bioelectricity using microbial fuel cell technology. However, in such an MFC system, certain parameters like the electrode type, mediator materials and catholytes, have to be optimised in order to achieve maximum benefit.

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Adekanle M.A. "Enhancement of Bioelectricity Production using Different Chamber Design, Catholytes and Varied Electrode in MFCS." IOSR Journal of Dental and Medical Sciences (IOSR-JDMS), vol. 18, no. 10, 2019, pp 60-67.