



**STUDIES ON MFC FOR BIOELECTRICITY GENERATION FROM  
BACILLUS MEGATERIUM ORGANISM USING COPPER ELECTRODE**

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**Abstract**

Industrial effluents are causing a steady decline in the ecological system. Toxic chemicals are present in the effluents, which in turn pollute ground water and cause new diseases. The costs of treating these industrial effluents are rising. Reduction, Recycle, and Reuse (RRR) using microbial fuel cells is the goal of the current study, which aims to reduce the toxicity of industrial waters. Microbial Power module tests were done in cluster tasks for domestic waste water using *Bacillus megaterium* organism to increment oxygen levels furthermore, decline harmfulness. The boundaries contemplated are plan of MFC, NaCl focus, Agar fixation, fructose measurements, pH, COD, Body, DO, TSS, TDS, Sulfates, Chlorides and Power age.

**Keywords:** DO, pH, COD, BOD, Treatment Efficiency, Microbial Fuel Cell.

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**Introduction**

India's energy crisis is getting worse every year because the cost of energy keeps going up and because more and more non-renewable energy sources are running out. A lot of research has been done to find a potential, usable, and reusable hotspot for energy production due to the desire for a fuel substitute. We must limit our use of petroleum products and reduce the pollution they cause in order to build a reasonable world. By treating waste water, these two things can be grown together. The recent surge in population paved the way for the industrial revolution and revealed the secrets of energy sources that are running out and driving up living costs each day. The scientific community has selected a number of alternative power production technologies to reduce power consumption and use. Microbial power modules can possibly at the same time treat squander water for Reuse and to create power. This study focuses primarily on the performance of dual chambered MFCs that are biocatalyzed with aerobic activated sludge and a strain of *Bacillus subtilis* instead of continuously powered by actual household waste water.

MFCs are bio-electro synthetic frameworks that create power by Oxidation of natural (or) in natural substrates catalyzed by microorganisms. Compared to the mixed cultures in activated

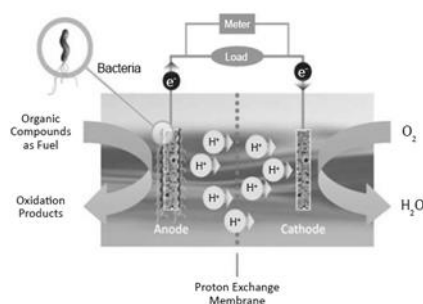
sludge, domestic electrogenic microbe *Bacillus subtilis* as a single pure culture was able to produce power and remove COD effectively. MFCs were able to digest COD in waste water with a removal efficiency of 90% when using single-culture inoculums and 84% when using mixed-culture inoculums [1]. Through the oxidation of organic matter, a MFC uses bacteria to generate electricity. Microbes fit for power age have been improved from homegrown waste water, sea dregs, creature squanders and a high-impact sewage muck [2, 3]. Because MFCs are able to generate electricity by capturing the electrons produced by microbiological processes, they hold the promise of a new, long-lasting energy source, particularly for the waste water industry [4]. Microbial power module innovation has seen huge progressions and upgrades in execution throughout the course of recent many years. Investigation into the creation of environmentally friendly power has zeroed in on MFCs [5].

Microbial fuel cells directly convert the chemical energy that is contained in an organic bioconvertible substrate into electrical energy through the mediation of exoelectrogenic bacteria that serve as the catalyzer of the half-reaction of substrate oxidation [6, 7]. Dual chamber MFCs are typically still under investigation when the specific objective is to utilize the cathodic reduction semi reaction for the removal of nutrients from waste water or organic-free ground water [8, 9]. Enacted slime and other conventional natural processes for treating waste water use energy. Aerobic treatment methods also leave behind a lot of solids that need to be treated and disposed of, which is expensive. Anaerobic processes, which use less energy, and process optimization are important options for reducing operational costs because these processes use a lot of energy. An additional advantage of including MFCs for waste water treatment is the potential for diminished solids creation appeared differently in relation to fiery cycles. Every year, thousands of tons of kitchen and municipal waste water are produced worldwide. Oxygen-consuming and anaerobic organic treatment processes are used to reduce the natural heap of these waste waters.

### **Experimental Procedure**

The anode and cathode chambers of the dual chambered MFC were constructed using airtight plastic bottles each 1.5 litres in volume. On each bottle, a PVC pipe was connected to a side opening with a radius of 1 cm at a height of 9 cm from the bottom (roughly in the middle). The 100 grams of agar and 100 grams of sodium chloride (NaCl) salt were heated in a 1000 millilitre water bath before being poured into a PVC pipe and sealed at one end with a plastic cap and cello tape. The agar was left undisturbed to cement. During MFC operation, the PVC pipe containing the salt-agar mixture served as a salt bridge, assisting in the proton transfer mechanism. It was attached between the two bottles with epoxy material.

Copper, 10 cm tall; electrodes (diameter = 0.5 cm) were utilized. In the MFC setup, the distance between the two chambers remained constant at 25, 20, 15, 10, and 5 cm. Copper wires were utilized to associate the anodes to the circuit. An external resistance (R) of 10, 47, 220, 500, and 1000 was connected, and a digital multimeter was used to take readings. In order to keep an eye on how the MFC's biodegradation process was progressing, the sample that was collected was analysed in accordance with standard techniques. Numerous boundaries are utilized to decide squander water characteristics. pH, TSS, TDS, BOD, COD, DO, chlorides and sulfates, among other parameters, were analysed in this study to evaluate the MFC's effectiveness.



**Figure 1: Schematic diagram of Microbial Fuel Cells**

The waste water test is investigated for at regular intervals and its different boundaries are assessed. During the activity, voltage and current is likewise checked by utilizing a multimeter.

### **Characterization of Synthetic waste water sample:**

#### **Estimation of TSS:**

Fill a funnel with 50 milliliters of water and a previously weighed Whatman No. 1 channel paper and channel. After being weighed once more, the filter paper is dried in an oven at 105(+/-) 50C.

#### **Estimation of TDS:**

The filtrate is gotten beforehand is taken into a weighed china dish and warmed on a burner till the water is dissipated. The china dish is heated for one hour at 105 degrees Celsius (+/- 50 degrees Fahrenheit) until all of the water has evaporated, at which point it is collected and weighed.

#### **Estimation of BOD:**

##### **Preparation of dilution water:**

A diffusion tube is used to aerate water by passing compressed air through it until it is completely saturated. Dilution water should be used to reduce the sample's volume to 300 milliliters. Two bottles are used to collect the diluted sample, which is filled to the neck. After five days of incubation, the dissolved oxygen level in one bottle can be determined immediately.

##### **Determination of dissolved oxygen (DO):**

- 1) In a bottle with a stopper, a predetermined amount of water—say, 250 milliliters—is taken without contacting air. Using a pipette, add 0.2 milliliters of  $MnSO_4$  solution to it, dipping the end well below the water's surface. 2.
- 2) Additionally, add 2 milliliters of the alkaline iodide-azide solution. Shake the bottle well with the stopper. Allow the brown  $MnO(OH)_2$  precipitates that have formed to settle.
- 3) At the point when some piece of the fluid beneath the stoppered is clear, add 2ml of concentrated  $H_2SO_4$  with the assistance of a pipette.
- 4) Mix with the stopper until the precipitate has completely dispersed. Iodine's signature brown color is produced.
- 5) Move 100 ml of the above arrangement in a 250 ml cup with a pipette. Until the sample solution turns a pale yellow color, titrate the liberated  $I_2$  with a standard sodium thiosulfate

solution.

- 6) The solution will turn blue if 2 ml of the starch solution is added. Titration should continue until the blue color fades. To obtain a second reading, repeat.
- 7) Volume of the water test taken for titration =100ml.

#### **Estimation of COD:**

Take reflux carafe and add to it 0.4gms of  $H_2SO_4$  and 20 ml of test. Mix well, and if necessary, dilute to a suitable level.  $K_2Cr_2O_7$ , 0.25N, add 10 milliliters. Add 30 milliliters of Concentration slowly after dropping a pumice stone.  $H_2SO_4$ - $AgSO_4$  reagent. Connect the flask to the condenser and thoroughly mix the contents. 2 hours of reflux. Cool the condensers and rinse them out. Weaken the combination to 150 ml by adding refined water. Titrate the indicator with N/10 ferrous ammonium sulphate solution after adding three drops of Ferro until the color changes from green to wine red. Note the end point. Carry out a similar technique with 'Clear' utilizing refined water rather than the example.

#### **Estimation of Sulphates:**

Take 150 ml test in measuring glass and make it acidic with HCl. Add the barium chloride solution slowly while stirring the solution until it reaches boiling point. Add this until all of the precipitation is gone. Digest the precipitate for two hours at 90oC. Use filter paper to filter the solution out. Utilizing an  $AgNO_3$  solution, wash the precipitate in warm distilled water until there is no color change and the wash water is free of chloride. The filter paper should be dried and precipitated for 30 minutes in a muffle furnace at 750<sup>0</sup> C. Cool the precipitate and weigh it with a crucible.

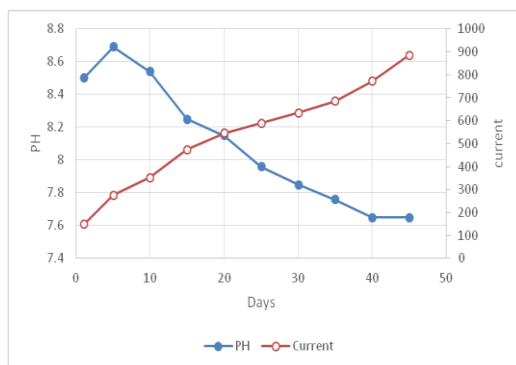
#### **Estimation of Chlorides:**

Take 50 ml of test in a funnel shaped carafe; Add three drops of the  $K_2Cr_2O_7$  indicator to this. And use a burette to take  $AgNO_3$  solution. After being treated with an  $AgNO_3$  solution, the sample takes on a yellowish to reddish brown precipitate. To obtain a second reading, repeat.

#### **Results and Discussion:**

##### **Effect of pH**

A pH meter was used to settle the pH of the waste model, which was domestic waste water. Figure 2 depicts the model's pH range taken over a standard number of days [10, 11].

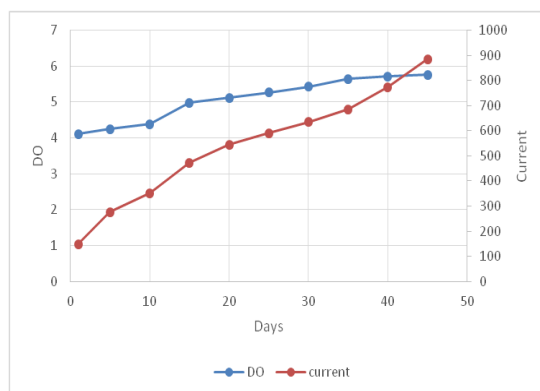


**Figure 2: Variation of pH and Current with time**

The acidity of the waste water sample is due to the addition of dextrose, which microorganisms use as food. The pH was brought down from 8.5 to 7.65, which is in line with BIS guidelines for healthy levels. In the meantime, the processing of living things constantly produces weak, harmful mixtures and maintains their intracellular pH.

### **Effect of Dissolved Oxygen:**

Figure 3 depicts the variation of dissolving oxygen over time. The results show that crumbled oxygen increased from 4.12 mg/L to 5.76 mg/L. The decrease in BOD and COD levels in the waste water sample and aeration are to blame for the increase in dissolved oxygen [12, 13].

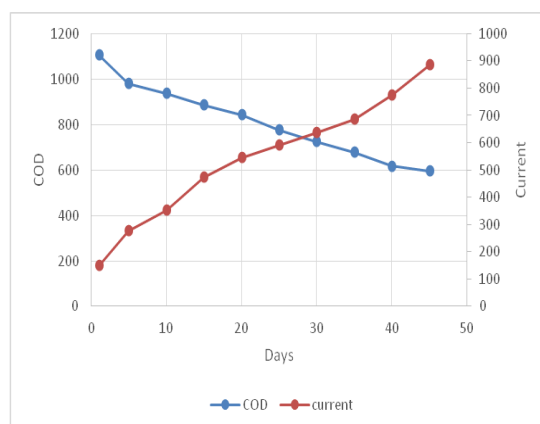


**Figure 3: Variation of Dissolved oxygen with time and current**

### **Effect of Chemical oxygen demand (COD)**

Figure 4 shows the COD of household waste water at various time scales. The results show that COD has decreased from a fundamental level of 1106.36 mg/L to 595.36 mg/L as the organisms in the waste water have grown and degraded the organic matter in the waste water sample.

The limit of the microorganisms found in waste waters' capacity to utilize the carbon source as electron benefactors was demonstrated by the domestic waste water's potential for COD ejection. B.G. found that COD was gone after 30 days of moving [14, 15].



**Figure 4. Variation of COD with time and current**

### Effect of Biochemical oxygen demand (BOD):

The amount of oxygen that an animal will consume while separating common matter in vivacious conditions is known as BOD. Figure 5 depicts the impact of MFC on the BOD of domestic waste water. As a result of continuous aeration and the action of sludge, the BOD level has decreased from 630.39 mg/L to 530.46 mg/L, as shown by the results [16, 17].

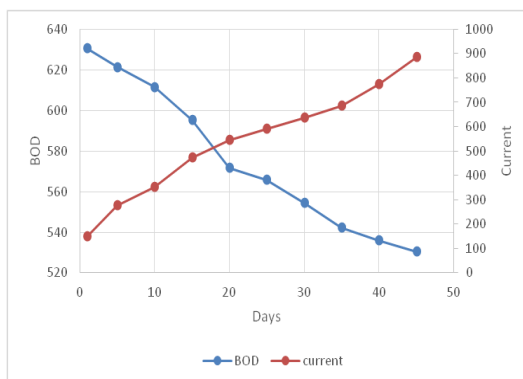


Figure 5. Variation of BOD and current with Time

### Effect of Total Dissolved Solids:

The capacity of the current MFC to clear Total Dissolved Solids was demonstrated. Figure 6 depicts the effect of MFC on the total salted solids of domestic waste water within a typical range. According to exploratory data, separated solids decreased continuously for 30 days during the movement. The domestic waste water test's TDS has decreased to 2410.87mg/L from 2912.31mg/L [18, 19].

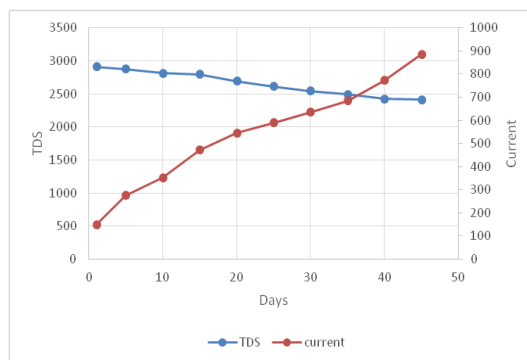


Figure 6: Variation of TDS and current with Time

### Effect of Total Suspended Solids:

Impact of MFC on the evacuation of TSS of the homegrown waste water test is displayed in the figure 7. The exploratory information shows that the extent of TSS in the model has diminished with the sneak past of time from 840.22 mg/L to 386.95 mg/L. The low TSS focus in the MFC reactor can be credited to two reasons. First, the MFC is based on biofilms, so the suspended solid is low because most of the biomass is stored on the electrode, with the occasional biofilm falloff. Another reason is that the MFC's anoxic to aerobic microorganisms produce fewer cells than the activated sludge [20, 21].

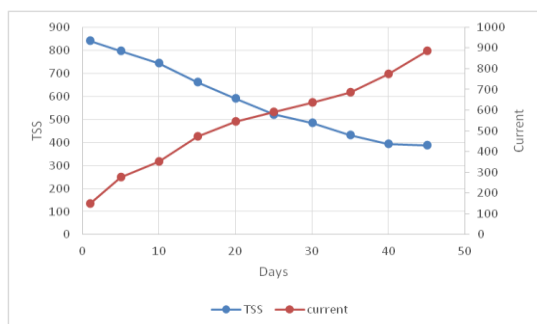


Figure 7: Variation of TSS and current with Time

### Effect of Chlorides:

The figure 8 shows how MFC affected the chlorides of the domestic waste water test. The test results indicate that the concentration of chlorides has decreased from 271.05 mg/L to 165.76 mg/L. The removal of chlorides from the sample may be attributed to the presence of a biodegradable substrate in the waste water sample, which results in microorganisms engaging in competitive inhibition [22, 23].

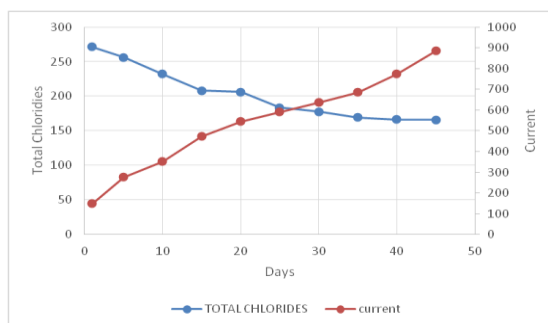


Figure 8: Variation of Total Chlorides and current with Time

### Effect of Sulphates:

The impact of MFC on Sulfates is displayed in the figure 9. The outcomes show decline in Sulfates from 94.30 mg/L to 41.66 mg/L. These outcomes show that the SRB cells in bio-film effectively changed Sulfate over completely to Sulfide in bio-film. Sulfide oxidizing bacteria could use energy from organic matter decomposition to oxidize sulfide with a low Redox potential [24, 25].

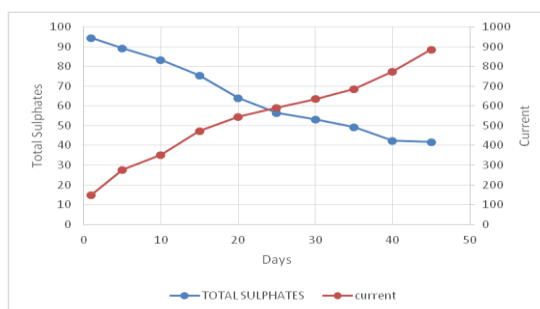


Figure 9: Variation of Total Sulphates and current with Time

### Effect of Total Hardness:

Figure 10 depicts the impact of MFC on the total hardness of the domestic waste water test. The results of the test indicate that the full-scale hardness content has decreased from 93.60 mg/L to 62.69 mg/L. The absence of pure hardness in the model could be attributed to the presence of a biodegradable substrate in the domestic waste water test, citing a genuine microorganism limitation [26, 27].

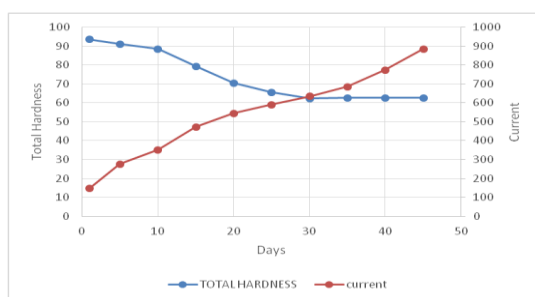


Figure 10: Variation of Total Hardness and current with Time

### Effect of Treatment Efficiency:

Figure 11 depicts the impact of MFC on treatment efficacy. The outcomes plainly show expansion in Treatment Productivity from 6.35 mg/L to 44.59 mg/L [28].

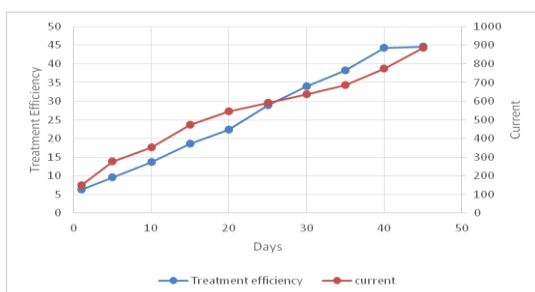


Figure 11: Variation of Treatment Efficiency and current with time

### Effect of Power Density:

The effect of MFC on the domestic waste waster's Power Density is shown in Figure 12. Power Density increased from 197.60 mW/m<sup>2</sup> at the beginning to 596.20 mW/m<sup>2</sup> at the end, according to the findings [29].

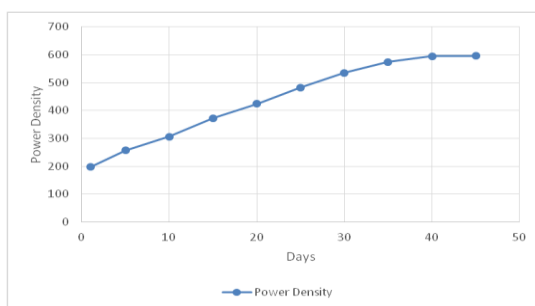


Figure 12: Variation of Power Density with time

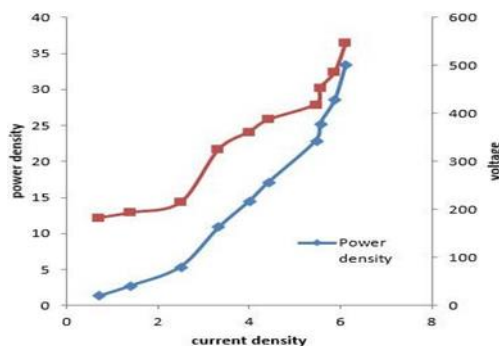


**Table 1: The Physico-Chemical Parameters of Treated & untreated Combination of domestic waste water**

S/N	Parameter	Untreated	Treated	BIS standards
1	Colour	Dark grey	Light Brown	-
2	Temperature	32 c	30 c	-
3	pH	8.5	7.65	6.5-9.0
4	COD, mg/L	1106.36	595.36	250
5	BOD, mg/L	630.39	530.46	50
6	DO, mg/L	4.12	5.76	4-6
7	TDS, mg/L	2912.31	2410.87	2100
8	TSS, mg/L	840.22	386.95	600
9	Chlorides, mg/L	271.05	165.76	600
10	Sulphates, mg/L	94.30	41.66	1000
11	Total hardness	93.60	62.69	200

### Polarization Curves:

The Polarization Curve Depicts the voltage consequence of the power gadget for a particular current Thickness stacking. A potentiostat or galvanostat, which measures the energy component yield voltage and draws the appropriate current from the power source, is frequently used to achieve polarization bends. A multimeter can be used to measure the voltage result of several different kinds of small resistors to compute if a potentiostat is unavailable [30].



**Figure 13: Current and Power Densities during Microbial Fuel cell monitoring**

Changing the circuit weight to choose the open circuit potential and working capacity of electrodes as a part of current is upto 1mA. Considering Ag/AgCl reference terminal the Anodes OCP and working not completely permanently established to be fairly 0.195 volt Figure 13 shows the Mix of the preliminary qualities is upto 0.804. 596.2 Mw/m<sup>2</sup> was the most powerful thickness.

### Effect of resistors:

Indeed, even the power yield was taken a gander at as a piece of the external hindrance. The yield for MFC worked under 10, 47, 220, 500, and 1000 autonomously was modified and noticed. Even at higher current densities, the substrate's lack of use for current generation was demonstrated by the low power yield. This lead was perhaps a prompt outcome of contention for

electron supplier between electrogenic living thing and fermentative and anaerobically breath in typical parts for electron advocate during the central season of anode colonization [31, 32]. Figure 14-18 plots are drawn for no. of days Versus Voltage for different Resistors.

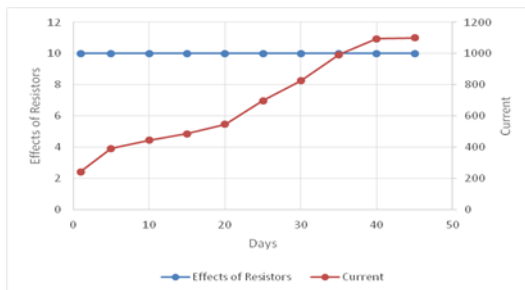


Figure 14: Effect of Resistors (10 Ω)

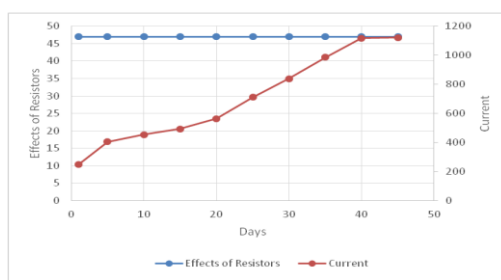


Figure 15: Effect of Resistors (47 Ω)

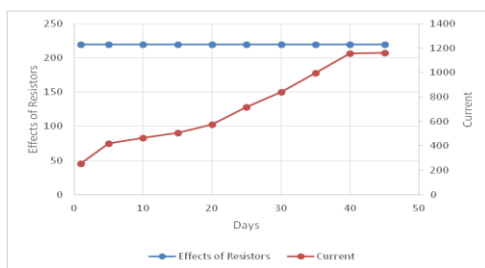


Figure 16: Effect of Resistors (220 Ω)

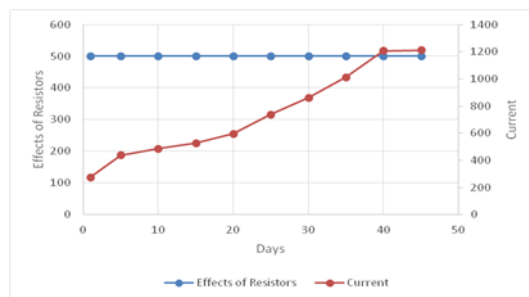


Figure 17: Effect of Resistors (500 Ω)

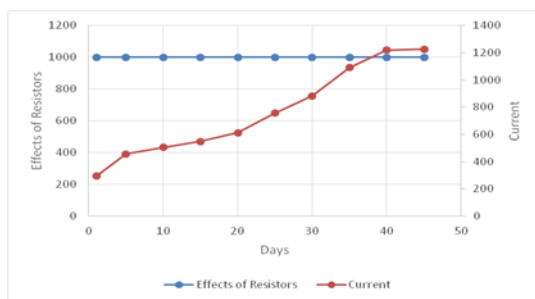


Figure 18: Effect of Resistors (1000  $\Omega$ )

### Coloumbic Efficiency:

The rate at which an electrochemical reaction is carried out by the system is called columbic efficiency. Using the ongoing Estimated over the long term (t) and the hypothetical current based on substance oxygen interest (COD) Evacuation, where  $\delta$  is constant for synthetic oxygen demand (M of O<sub>2</sub> = 32Gm/mole, 4 electrons traded per mole of oxygen, F is the Faraday's steady/96485 C/mole electrons), q is the volume of the medium Chamber, and COD is the adjustment of the compound oxygen interest over the long term [33, 34].

The Coloumbic efficiency was between 0.019 and 38.2, with day 35 seeing the highest value.

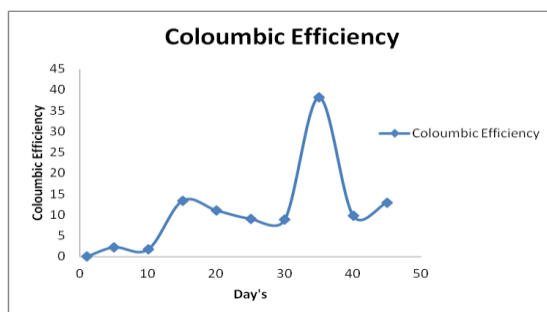


Figure 19: Variation of Coulombic Efficiency with time

### Conclusion

The primary objective of this study was to investigate the biological wastewater treatment methods. In order to improve the quality of waste water, this physical study selected the Microbial Fuel Cell. When contrasted with the underlying qualities of waste water, there is a diminishing in how much TSS, TDS, Body, COD, sulfates and chlorides, oil, and oil. The MFC was effective, more affordable, easy to keep up with, and needn't bother with a gifted laborer. During wastewater treatment, they certainly have the potential to recover energy. They may occupy a market niche in terms of direct wastewater treatment and a standalone power source. It was discovered that the fundamental guiding principle for removing toxicity and producing electricity is the presence of biodegradable compounds in waste water samples.

### Study on domestic waste water using *Bacillus megaterium* organism

The PH decreased from 8.75 to 7.65 at room temperature.

At room temperature, COD decreased from 1106.36 mg/L to 595.36 mg/L.

At room temperature, the Body dropped to 530.46 mg/L from 630.39 mg/L.

At room temperature, DO expand from 4.12 mg/L to 5.76 mg/L.

TDS was decreased from 2912.31 mg/L to 2410 mg/L.

TSS decreased from 840.22 mg/L to 386.95 mg/L. at room temperature, chloride decreased from 271.05 mg/L to 165.76 mg/L, sulfates decreased from 94.30 mg/L to 41.66 mg/L, and total hardness decreased from 93.6 mg/L to 62.69 mg/L.

Treatment efficacy increased from 6.35 mg/L to 44.59 mg/L at room temperature, and power thickness increased from the initial value of 197.60 mW/m<sup>2</sup> to the final value of 596.2060 mW/m<sup>2</sup>.

The colonic efficiency was anywhere from 0.019 to 38.20.

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