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Green Electricity Production from Living Plant and Microbial Fuel Cell

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ABSTRACT

Plant Microbial Fuel Cell (P-MFC) is the one in which plants and bacteria are present to convert solar energy into green electricity. The principle idea is that plants excrete rhizodeposits from roots, mostly in the form of carbohydrates, and the bacteria convert these rhizodeposits into electrical energy via the fuel cell. In our project, we used Asparagus fern and Clorophytom plants with double chambered and single chambered design of P-MFC. The copper plate and carbon rods were used as electrode with copper wire as current collector. The maximum power output of 10 mW/m^2 was achieved by connecting four P-MFCs in series. The average voltage reading 0.7 V was achieved per plant. This technology can be used as renewable source of energy with advancements.

Keyword: Bacteria, Green electricity, Microbial fuel cell, Rhizodeposits, Renewable energy

I. INTRODUCTION

Increased energy demand and utilization is anticipated in coming days. The possible attributable factors for the increased demand include ever increasing population, industrialization and development of smart and hi-tech cities. Furthermore, energy generation and utilization is directly linked with the nation's economic welfare. For making the developing and underdeveloped countries developed, economy of that country has to be strong. And to secure stable and increased economy, the energy requirement of the country should be strategically worked upon.

Regardless of diminishing reserves of fossil fuels, geographical and political constrains in harnessing energy from fossil fuels and the associated environmental pollution, still approximately over 67% of the electricity is produced from coal, oil or natural gas. Significant research is ongoing in harnessing energy from renewable and sustainable sources like solar, wind, water, nuclear and biomass. Worldwide statistics have shown that 13.4% energy is obtained from nuclear reactors, 16.2% energy from hydropower and wind and 3.3% energy from solar and biomass [1]. But these renewable sources also have their drawbacks. Solar can't give the energy during the rainy season, and the devices used for the production and conversion of chemical energy into electrical energy are very expensive. Solar panel performance depends on light intensity. This makes it both weather dependent and daylight dependent Alternative electricity generation technologies are sometimes debated for their environmental performance, even though they are renewable. Solar panels contain scarce metals and wind turbines need the rare earth metal neodymium for construction. Especially mining of these metals causes a lot of environmental pollution, thus the environmental performance of wind and solar power is debatable [1, 2]. Hydropower is generally considered environmental friendly based on the materials used, but debate is ongoing about the loss of natural areas due to

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submergence upstream of the dam. The energy produced from the wind energy is very low. Availability of electricity is affected mainly by two different factors *viz*, source availability and storage. An electricity generation technology or source can be limited geographically or is weather dependent, time dependent or otherwise limited in availability. If the electricity production from a source is weather dependent but peaks in demand coincide with peaks in production, no problems arise. In practice, however, this is not the case with solar power. Wind power is weather dependent and geographically limited in application because open fields without too many buildings is needed. Hydropower is geographically limited to the course of the river and might in some cases be season dependent for its flow. Thus for the easily available and sustainable source of energy we have rely on biomass. All the problems stated above can be avoided by using the plant microbial fuel cell (P-MFC) [1, 3, 4].

P-MFCs have gained a lot of attention in recent years as a mode of converting organic waste including wastewaters into electricity. P-MFCs are the major type of bio electrochemical systems which convert biomass spontaneously into electricity through the metabolic activity of the symbiotic microorganisms. P-MFC is considered to be a promising sustainable technology to meet increasing energy needs, especially using wastewaters as substrates, which can generate electricity. The aim of the P-MFC is to transform solar energy into electrical energy through oxidation of rhizodeposits by electrochemically active bacteria. Photosynthesis in plants occurs in its leaves whereby the solar energy is used to fix carbon dioxide in the form of carbohydrates. Depending on plant species, age, and environmental conditions upto 60% of the net fixed carbon can be transferred from its leaves to the roots. The plant root system produces and releases different types of organic compounds into the soil, which includes exudates of sugars, organic acids, polymeric carbohydrates, enzymes, lysates of dead cell materials, and gases like ethylene and CO_2. Summation of these released products by plants is termed as plant rhizodeposits while the process is called as rhizodeposition. The produced rhizodeposits accounts approximately 40% or even more of the plant's photosynthetic productivity. Utilization of these abundantly produced rhizodeposits, as energy rich substrates for production of electricity is the underlying concept of P-MFC. This energy source seems to be an opt alternative for harnessing the energy [2, 5]. The biochemical reactions taking place in P-MFC are as mentioned below in equation 1 and 2 and the schematic representation of fuel cell comprising of plants is as depicted in Figure 1.

Anode Reaction: $C_6H_{12}O_6 + 6H_2O \rightarrow 6CO_2 + 24H^+ + 24e^-$ Equation 1

Cathode Reaction: $6O_2 + 24H^+ + 24e^- \rightarrow 12H_2O$ Equation 2.

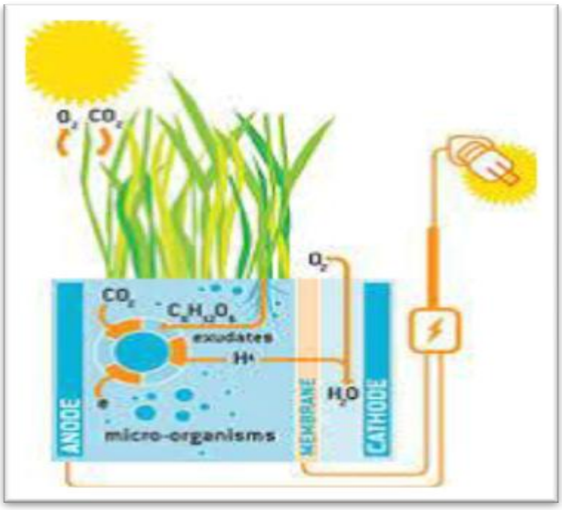

Figure 1: Schematic Representation of P-MFC in power generation [6].

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Firstly it tackles the major problem of environmental pollution and geographical limitation associated with non renewable fossil fuels; as well as the process is not limited by seasonal variation, day and night cycle or dependence on use of any rare and toxic elements.

Reviewing the pros and cons of all the renewable sources of energy production, we in our study attempted to generate electricity using P-MFC.

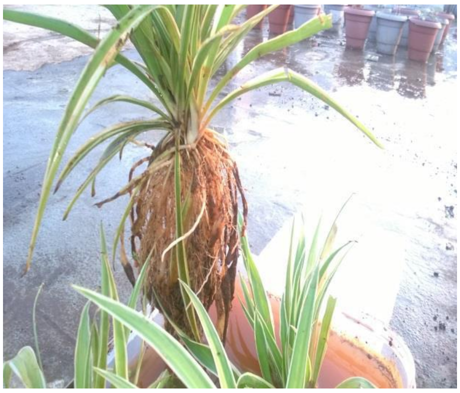
II. MATERIALS AND METHODS

2.1. Materials

Agar, NaCl, KCl, sodium potassium tartarate, dinitro salicylic acid, H_2SO_4, H_2O_2, n-napthol solution, isopropanol, peptone, NaOH, beef extract, yeast extract, glucose, Fehling reagents A and B, crystal violet, iodine solution, acetone, safranin, distilled water ,cedar wood oil, etc were procured from Himedia Labs Pvt. Ltd., Mumbai.

2.2. BIOLOGICAL SPECIMEN

Two plants belonging to the family Asparagaceae *viz*, *Chlorophytum inornatum* and *Asparagus fern* were employed in our study (Figure 1). They were procured from local nursery.



Chlorophytum inornatum

Asparagus fern

Figure 2: *Chlorophytum* and *Asparagus* plants

2.3. Methods and Construction of P-MFC set up

2.3.1. Double Chambered P-MFC

Two containers of size 35 cm × 25 cm approximately were taken and they were connected with each other with the help of a salt bridge. The copper plates were added to the bottom of both the compartments. Copper wire was wounded or fixed on the copper plate and taken outside of compartment by making small hole at the bottom side of the container. In the anode compartment, the plants having fibrous roots (*Asparagus fern)* and soil and some drainage water was added. Cathode compartment was containing only demineralized water. The anode compartment was covered with help of plastic so that the less oxygen condition was maintained in the compartment. Both the compartments were connected to the external circuit having external resistance with the help of copper wire. The voltage readings were taken with the help of multimeter. After few days for the proper growth and development of plants, plant growth hormones were added [3].

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Single Chambered P-MFC

Single chambered P-MFC was very much similar in construction with the double chambered unit with exception of presence of cation exchange membrane in single chambered P-MFC, bottom situated anode was and top located cathode [3].

2.3.2. Salt Bridge Formation

2% agar was taken in the 0.1 N solutions of NaCl / KCl. Then the solution was heated up to 100 ^{o}C and stirred simultaneously till agar completely dissolves. These solutions were added into the plastic or glass tube covered from one end and the solution was left undisturbed until solidification of agar [5].

Double Chambered P-MFC Single Chambered P-MFC Salt Bridge

Figure 3: Experimental Design of types of P-MFC and salt bridge

2.4. Isolation of microbes from P-MFC

The soil associated with the fibrous roots of *Chlorophytum* was collected in a sterile container. The soil sample was suitably diluted using sterile saline and the sample was introduced by spread plate technique onto the surface of nutrient agar plates under strict sterile and aseptic conditions. The plates were then incubated for 48 h at 30 °C in a Bacteriological Incubator (Kumar). The isolates were further purified to homogeneity using streak plate technique.

2.5. Biochemical Characterization of root associated microbes of *Chlorophytum*

The isolate so obtained was further subjected to biochemical tests like Gram's staining, Oxidase test and Catalase test [7].

2.6. Identification tests for carbohydrates

Asparagus plant was placed in the double chambered P-MFC containing waste water and soil for couple of days. Water sample near the rhizodeposits was collected and evaluated for the presence of sugars using standard laboratory tests of carbohydrates. Sample was evaluated by Molisch test, Fehling's test and Dinitro salicylic acid test [8].

III. RESULTS AND DISCUSSION

3.1. Generation of electricity from P-MFC

Electricity can be produced by the symbiotic associations of plants and microbes through a series of ongoing biochemical reactions at cellular level. These biochemical reactions are associated with generation of electrons and

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protons which could be harnessed to produce electricity using the principle of a simple redox reactions taking place in an electrolytic cell [5]. David et al., (2008) reported the generation of electricity by incorporating Reed mangrass; Ueoka et al., (2016) employed rice and paddy plants to produce bioenergy; Timmer's et al., (2010) Moqsud et al., (2014) employed Spartina anglica and grass cutting and leaves mold of rice bran respectively to produce energy. We in our study selected two plants *viz*, *Chlorophytom* and *Asparagus* for generation of green energy [4,5,9,10]. Selection of plants was performed based upon their fibrous roots and ability to grow in presence of large quantity of water [9].

Initially we attempted to evaluate the effect of fuel cell design upon the generation of voltage. *Chlorophytum* was placed in the anode compartment of the fuel cell along with excess water and less quantity of soil. Copper plate was placed in both the compartments and the voltage generated was recorded using a multimeter for a period of 30 days. The results are as depicted in Figure 3 and 4 of double chambered and single chambered fuel cells.

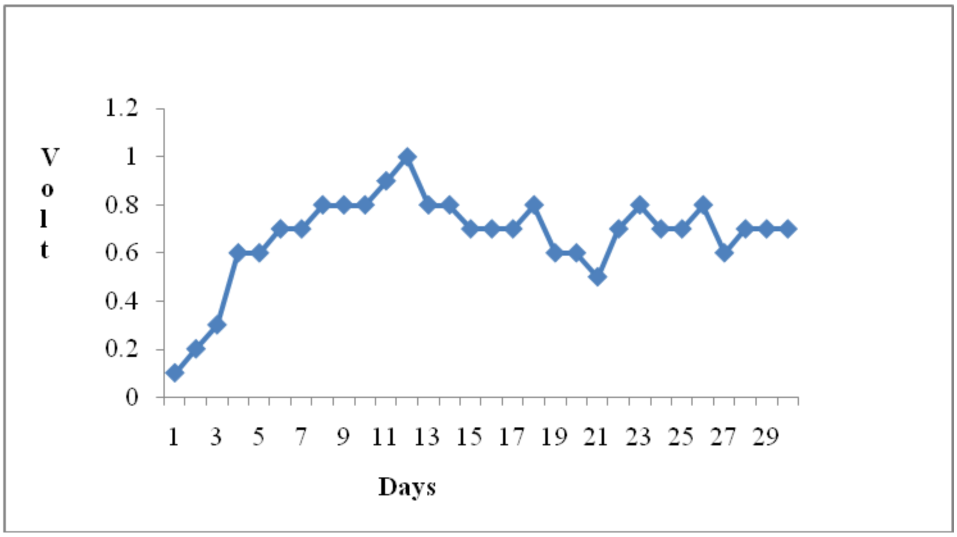

Figure 4: Voltage generation of double chambered P-MFC using *Chlorophytum* plant

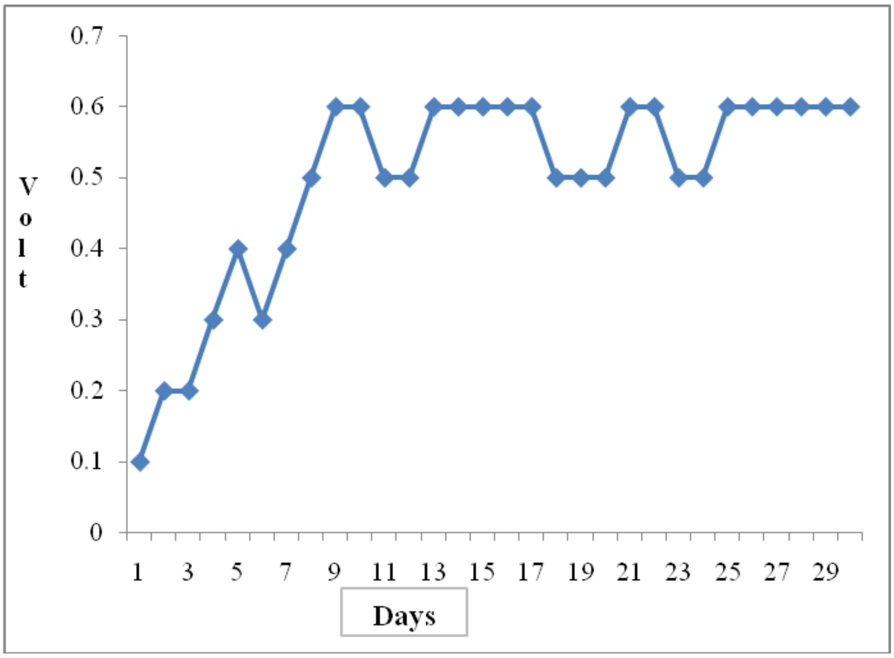

Figure 5: Voltage generation in single chambered P-MFC using *Chlorophytum* plant

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Voltage generated using double chambered P-MFC attained maximum voltage of 0.9 V on 12th day while the voltage kept fluctuating between 0.6 to 0.9 V for the remaining 18 days with the voltage minima of 0.6 V during the last three to four days. In case of single chambered P-MFC, maximum voltage of 0.6 V was attained which was slightly lower than that of the double chambered P-MFC design. Furthermore after 12 days of incubation, the voltage generated seems to fluctuate between 0.4 to 0.6 V. Thus for further studies we decided to incorporate double chambered P-MFC.

Electricity generated using P-MFC is considered to be a function of composition of nutrients present in water, constituents of salt bridge and material of construction of metal used as electrodes in fuel cell [6, 9, 11, 12].

So as to evaluate the effect of constituents present in water upon the generation of electricity we set two assemblies of fuel cells. The construction of assembly in both the variants was same i.e. double chambered copper plate based P-MFC, Chlorophyta except waste water alone was employed in one assembly while in another waste water supplemented with 5% glucose was attempted. Voltage generated was measured for a period of 15 days. In both the cases, it was found that the generation was voltage of 0.9 V was observed during 12th day of study. This implies that added glucose hardly exhibited any advantageous effect. Our results are in confirmation with the results of Barua et al., 2010 who obtained maximum generation of current using biowaste than simpler and homogenous nutrient sources [13].

P-MFC based electricity is a function of size of cathode chamber and type of electrode. Initially we performed our experiments by incorporating smaller plastic trays for constructing the double chambered fuel cell. It was however observed that towards the end of 30 days, the plant had drooped, indicating negligible metabolic processes in the plant cells yielding fluctuating and lower magnitude of voltage from 15 days of experimentation. This could be attributed due overheating of plants cells in smaller sized chamber that could have affected its biochemical reactions resulting into less electricity generation. Strik (2008) has reported the power generation using graphite electrodes in P-MFC and obtained the yield of 67 mW/m^2 [5, 14].

To evaluate the effect of electrode metal upon power generation, we in our study replaced copper electrode with carbon electrode. Carbon is known for its decreased resistance as compared to copper as anticipated yielded generation of higher voltage in almost half of the experimental period. The results of the study are as shown in Figure 6 and 7.

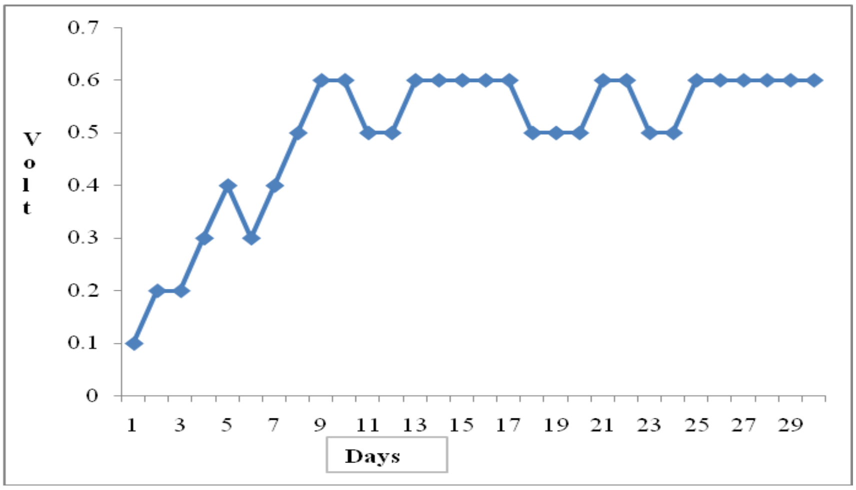

Figure 6: Effect of copper on voltage generation in a double chambered P-MFC

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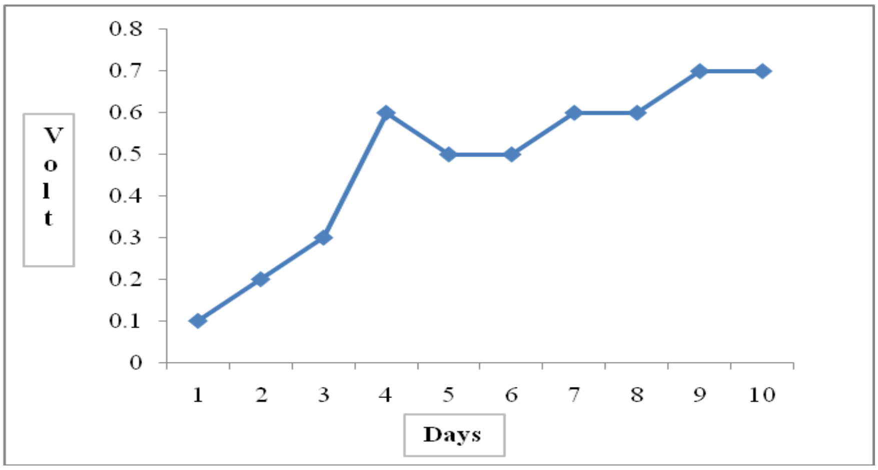

Figure 7: Effect of carbon on voltage generation in a double chambered P-MFC

3.2. Microbial isolation from rhizoids of P-MFC

Microbes associated symbiotically with fibrous roots of the plants in P-MFC were then isolated and characterized to understand the requirement of oxygen. Four isolates were obtained to purity, were then subjected to Gram's staining, catalase test and oxidase test. The results are as illustrated in Table 2. It can be observed that all the cultures are of gram negative nature. Further more from the oxidase and catalase tests it can be concluded that except JNEC 02, all the other three cultures are facultative as they have yielded oxidase test negative and catalase test positive, indicating their ability to grow both under aerobic and anaerobic conditions. In case of JNEC 02, both the catalase and oxidase tests are negative indicating its inability to grow in presence of oxygen resembling it nature similar to strict anaerobe.

Table 2: Biochemical tests of isolates

Tests and Isolates	JNEC 01	JNEC 02	JNEC 03	JNEC 04
Gram's Staining	Negative	Negative	Negative	Negative
Oxidase Test	Negative	Negative	Negative	Negative
Catalase Test	Positive	Negative	Positive	Positive

3.3. Identification of sugars in rhizodeposits

Rhizodeposits are exudates of plants typically composed carbohydrates, lipids, proteins, minerals, etc [2, 5]. So as to ascertain the presence of sugars in the rhizodeposits of the *Chlorophytum* double chambered P-MFC, we performed qualitative tests of sugars like Molisch test, fehling's test and dinitro salicylic acid test and observed for the changes like formation of violet ring, blue to red coloration and development of reddish orange coloration respectively. Thus presence of sugars is confirmed in our study in rhizodepoits. However biochemical conformation of other metabolites requires further tests for protein, lipds, etc to be undertaken.

IV. CONCLUSIONS

Bacteria are able to produce electricity by using plant rhizodeposits as a substrate under limited oxygen condition. We were able to attain the maximum voltage of 0.9 V on 10th day using *Chlorophytum* based P-MFC. Futher

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optimization in terms of area, electrode composition, salt bridge composition, etc can increase the amount of energy generated.

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