

**GREEN ELECTRICITY PRODUCTION BY EPIPREMNUM AUREUM AND
BACTERIA IN PLANT MICROBIAL FUEL CELL**

NEGAR DASINEH KHIAMI

UNIVERSITI TEKNOLOGI MALAYSIA

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NEGAR DASINEH KHIAMI

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This thesis is dedicated to my parents, Nader and Ashraf, who have always loved me unconditionally and whose good examples have taught me to work hard for the things that I aspire to achieve.

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ABSTRACT

Due to high energy demand worldwide, finding an alternative renewable and sustainable energy source is of great interest. Plant microbial fuel cell (P-MFC) is one of the most promising methods to generate green energy. In P-MFC, a plant is placed into the anode compartment. Mutual interaction between plant root rhizodeposits and bacterial community present in biofilm format at the vicinity of the rhizosphere area in plant root could be utilized to generate electricity. Indeed, in P-MFC, bacteria metabolize rhizodeposits into electrons and protons. These electrons could be then converted into green electricity. In this work, *Epipremnum aureum*, was selected as the studied plant species. Measurement of electricity generation by this specific species was conducted for 20 days. The open circuit voltage (OCV) was measured at 195 mV and the maximum power density was 0.85 $\mu\text{W}/\text{cm}^2$. Five isolated bacterial strains from the graphite felt surface found on the anode were screened by nine biochemical tests such as catalase, TSI (triple sugar iron agar), gelatin and etc.

ABSTRAK

Oleh kerana permintaan tenaga yang tinggi di dunia, mencari alternative sumber tenaga boleh diperbaharui merupakan satu bidang yang sangat menarik. Sel bahan api mikrob (MFC-P) adalah salah satu kaedah yang paling berpotensi untuk menjana tenaga hijau. Di dalam P-MFC, tumbuhan ditempatkan ke dalam petak anod. Interaksi bersama di antara rhizodeposits tumbuhan dan komuniti bakteria (bio-film) di sekitar rizosfera menghasilkan proton dan elektron. Elektron yang terhasil ini kemudiannya ditukarkan menjadi tenaga elektrik. Di dalam projek ini, sejenis sepsis pokok keladi, telah dipilih sebagai tumbuhan kajian, dan pengukuran penjana elektrik menggunakan spesies ini telah dijalankan selama 20 hari. Maksimum voltan litar terbuka (OCV) yang diukur bernilai 195 mV dan ketumpatan kuasa maksimum sebanyak $0.85\mu\text{W}/\text{cm}^2$ telah diperolehi. Lima jenis bakteria telah dipencilkan daripada permukaan anod dan telah disaring untuk 9 ujian biokimia seperti katalase, TSI (tiga kali ganda agar besigula), gelatine dan sebagainya.

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LIST OF ABBREVIATION AND SYMBOLS

°C	-	Degree Centigrade Celsius
CO ₂	-	Carbon dioxide
g	-	Gram
hr	-	Hours
H ₂ O	-	Dihydrogen oxide
Kg	-	Kilogram
mg	-	Milligram
mg/L	-	Milligram/Liter
min	-	Minute
ml	-	Milliliter
NA	-	Nutrient Agar
O ₂	-	Oxygen
Ω	-	Ohm
s	-	second
M	-	Molarity
cm	-	centimeter
L	-	liter
P-MFC	-	Plant microbial fuel cell
MFCs	-	Microbial fuel cells
P _t	-	Platin
SEM	-	Scanning electron microscope
mA.m ⁻²	-	Mili ampere per square meter
mV/m ²	-	Mili volt over per square meter
HG	-	Hoagland solution
S-MFC	-	Plant-assisted Sediment-MFCs

RPF	-	Rice Paddy Field
DGGE	-	Denaturing gradient gel electrophoresis
T-RFLP	-	Restriction fragment length polymorphism
$W.m^{-2}$	-	Watt over square meter
Fe_2O_3	-	Iron(III) oxide
CE	-	Columbic efficiency
NADH	-	Nicotinamide adenine dinucleotide
ADP	-	Adenosine diphosphate
ATP	-	Adenosine triphosphate
RVC	-	Reticulated vitreous carbon
CoTMPP	-	Cobalt tetramethylphenylporphyrin
FEPC	-	Iron phthalocyanine
PbO_2	-	Lead dioxide
CEM	-	Cation exchange membrane
PEM	-	Proton exchange membrane
NaOH	-	Sodium hydroxide
H^+	-	Proton
mV	-	Milli Volt
mA	-	Mill ampere
mW/m^2	-	Milli watt per square meter
$W.m^2$	-	Watt per square meter
KNO_3	-	Potassium nitrate
$NH_4H_2PO_4$	-	Ammonium dihydrogen phosphate
$Ca(NO_3)_2$	-	Calcium nitrate
$MgSO_4$	-	Magnesium sulfate
EDTA	-	Ethylenediaminetetraacetic acid
KOH	-	Potassium hydroxide
$FeSO_4.7H_2O$	-	Ferrous Sulfate Heptahydrate
H_3BO_3	-	Boric acid
$MnCl_2.4H_2O$	-	Manganese(II) Chloride Tetrahydrate
$ZnSO_4.7H_2O$	-	Zinc Sulfate Heptahydrate
$CuSO_4.5H_2O$	-	Copper(II) Sulfate Pentahydrate
$H_2MoO_4. H_2O$	-	Molybdic Acid

NaCl	-	Sodium chloride
kΩ	-	Kilo Ohm
DNA	-	Deoxyribonucleic acid
H ₂ O ₂	-	Hydrogen peroxide
NA	-	Nutrient agar
H ₂ S	-	Hydrogen sulphide
TSI	-	Triple sugar iron
NaCl	-	Sodium chloride
μA/cm ²	-	Micro ampere per square centimeter

CHAPTER1

INTRODUCTION

1.1 Background of Study

Excessive emission of greenhouse gases is one of the most critical and important issues in the world. Generation of power with less emission and high efficiency is highly demanding. Introducing sustainable, new and renewable energy could be the best solution to reduce emission of greenhouse gases. Furthermore this is a new challenge between nations to exploit. Recently, fuel cells are considered as a high potential clean energy technology, due to the high energy conversion efficiency through the chemical degradation process. Microbial fuel cells are one of the most studied fuel cells, due to its potential application to generate electricity from wastewater treatment processes. Various types of bacteria and yeast involved in the system have been investigated. The electron transformation mechanism and microorganism behavior have been studied in some articles. (Timmers *et al.*, 2013, Huggins *et al.*, 2014, Xiao *et al.*, 2014, Chen *et al.*, 2014 and, Zhoua *et al.*,2014).

The plant microbial fuel cell (P-MFC) is a bioreactor that generates green electricity from the interaction between microorganisms of rhizosphere and root organic which released compounds such as sugars, organic acids, polymeric

carbohydrates, enzymes, dead cell materials and etc (Strik *et al.*, 2008). Some parts of these organic compounds are then oxidized; donated electrons are then transferred to suitable electrodes which are located at the anode compartment (Yolina *et al.*, 2012). On the other hand, protons are transferred through the membrane and undergo reduction in the cathode chamber producing water. The P-MFCs were primarily implemented by Strik *et al.*, in (2008), and they achieved maximum power production of $67 \text{ mV} \cdot \text{m}^{-2}$ anode surface area. They designed dual-chamber set up for P-MFC which were connected by a membrane (proton exchange membrane), while De Schanphelire, (2008) represents sediment P-MFC without employing membrane between cathode and anode compartments. The scheme of the microbial plant fuel cells in this project is presented in Figure 1.1.

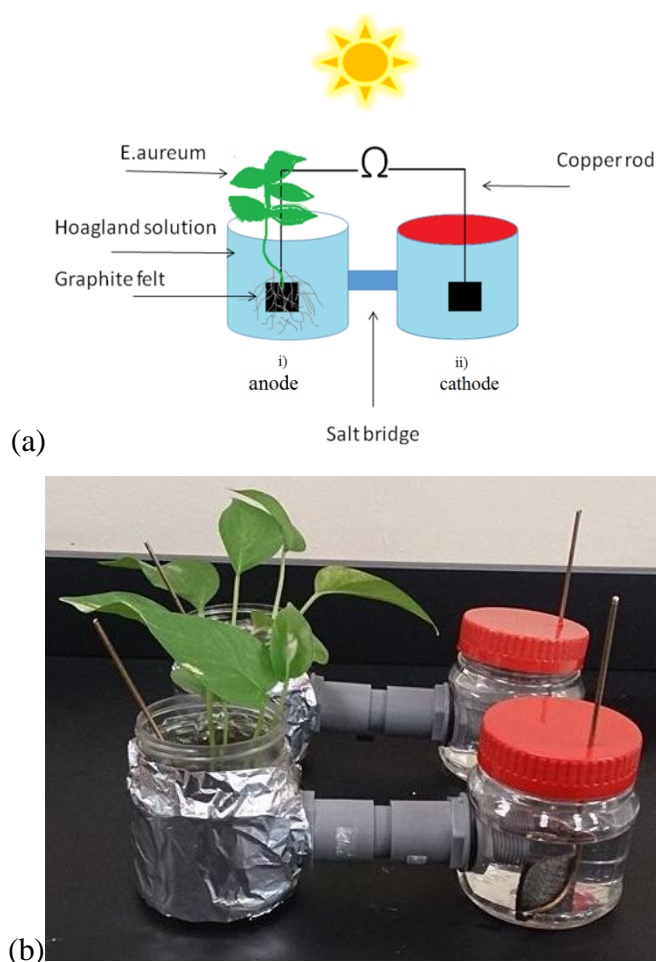


Figure 1.1 Schematic (a) and photograph (b) of dual-chambers Plant Microbial Fuel Cell: two compartments are separated by a salt bridge. Plant and graphite felt placed in the anode compartment.

1.2 Statement of Problem

Although electricity generation by MFCs has increased indefinitely at lab scale, scaling up this system is still a big problem. In addition high cost of proton exchange membrane and its fouling problem is a vital upcoming problem which could lead to the increase of the internal resistance and reduction of power output as well (Hu.,2008). From the energy demand and cost aspect, providing external artificial illumination increase the cost of constructing this system as well (Strik *et al.*, 2008 and He *et al.*, 2009). The biggest disadvantage of MFCs is that based on the constructing condition such as electrode material, configuration design, and temperature and most crucially the feeding substrate the operation period is various (Wang *et al.*, 2009).

This technology besides non compatibility with food production could be united with agricultural products (Helder *et al.*, 2012, Deng *et al.*, 2012 and Hubenova *et al.*, 2012). Therefore this system has the potential to be implemented in inappropriate locations such as green roofs and wetlands for crop production. One of the biggest disadvantages in applying this system is the request for large surface area of electrodes. On the other hand topsoil excavation for integration of this system could hinder the fertility of the soil. Therefore in order to remain the top soil from weakening and also remaining soil fertility aquatic plant could be the better option (Timmers *et al.*, 2013).

A usual problem which normally happens in the MFCs is the pH gradient between the membranes. Due to the degradation of substrates in the anode the pH in the anode convert to the acidic. While in the cathode alkaline by oxygen reduction as well as non-specific permeability of PEM is produced (Harnisch *et al.*, 2009). This problem could be overcome by applying different techniques such as utilizing buffers (Sleutels *et al.*, 2009) and membraneless microbial fuel cell (Hu *et al.*, 2008). However these methods dramatically decline the fuel cell energy recovery

(Rozendalet *et al.*, 2008). Therefore further developments need to be achieved in order to reduce the pH gradient (Harnisch *et al.*, 2009).

1.3 Objectives of Study

Based on Hubenova *et al.*,(2012), microorganisms which inhabit around the rhizosphere of plant roots, are considered to have significant importance to interact with anode in the aquatic MFCs operation. The objectives of this research are:

- 1) To utilize *Epipremnum aureum* plant to generate electricity.
- 2) To observe current generation by different resistors.
- 3) To characterize immobilized bacteria attached on the anode surface.

1.4 Scope of Study

Through this study graphite felt was used as an electrode material in the P-MFC due to its good electrical conductivity, chemical stability, relatively cheap and availability. In addition to graphite felt, other carbon-like materials to improve the efficiency of P-MFCs could also be used. Also, optimizing the cathode and anode chamber pH media to improve the performance of P-MFCs was expected. This aim was achieved by applying various concentration of phosphate buffer. Monitoring current generation between bacteria and plant interaction could achieved by applying various resistors. Presence and activating various species of bacteria with specific characteristics during highest OCV achievement was expected. According to (De Schamphelaire *et al.*, 2010), microbial biofilm on the anode are responsible for the current generation. Characterization of anode attached biofilm by biochemical tests

was done. These bacteria have specific optimum growth temperature. Highest current generation is usually possible when quite a number of bacteria species are available in the form of biofilm on the electrode surface.

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