Abstract

In this experiment, a batch mixed culture microbial fuel cell with Shewanella putrefaciens sp. 200 as the primary inoculant (cell 1) was tested under aerobic and anaerobic conditions under nitrogen gas. In the microbial fuel cell with Shewanella putrefaciens sp. 200 as catalysis in the 1% lactate solution can prove an open circuit potential up to -0.514V vs. Ag/AgCl 4M at pH= 7.5 STP. The ideal potential for the cell is around -0.610V, so the efficiency of this cell is 84.26%. A reference non-sterile cell containing growth medium can provide the open circuit potential to 0.03V vs Ag/AgCl 4M at pH=7.

Introduction

Microbial fuel cells (MFC) are systems that take advantage of certain microorganisms as catalyst to create a potential and drive a current from reactions in media. The goal of this experiment is to make a new microbial fuel cell with Shewanella Putrefaciens as catalyst in a media with lactate. This type of MFC are one of the currently successful applications would be using microbial fuel cells to treat waste water as well as make electric power.

We are interesting in improving the efficiency of the potential that the cell made by changing the lactate concentration and temperature. The experiments are mentored by digital simulation of cyclic voltammograms and fine voltmeter. In the measurement, MFC will be treat as working electron, and the reference electron we chose Ag/AgCl (4M KCl sat. with AgCl filling solution) which has -0.222V vs. normal hydrogen electrode (NHE).

For this experiment, we did two runs to make sure the results are repeatable and actuate. All the experiments are under nitrogen to eliminate the influence of oxygen in liquid to change
the data. The first run, we were focusing on testing the whether lactate is the reason to make
electric, second run we were not only prove lactate is one of the important reason that make
electric but also trying to improve the efficiency by varying the concentration of the lactate in
medium.

Experiment

This experiment analyzed the performance of two non-sterile microbial fuel cells. Cell
performance was determined by the voltammograms and the open circuit potentials. Data used to
compare the two cells came from the first three days of operation.

Preparation

Three-Compartment Cells

Each cell is a three compartment cell. All three cells are filled with about 100 g of growth
medium prepared for *Shewanells putrefaciens* sp. 200. However, each cell has different amount
of Sodium Lactate: 0 mL, 3 ml. and 6 ml. where it acts as a nutrient for *Shewanells putrefaciens*’s
growth. All three cells have a working electrode that is made of carbon cloth and have a
roughness factor of about 60. The cells with 0 ml. and 3 ml. of Sodium Lactate used graphite
rods as counter electrode and Ag/AgCl 4M as a reference electrode, which were placed in their
respective compartment. Even through the cell with 6 ml of Sodium Lactate has the similar
counter electrode; it has a calomel reference electrode. All measurement were at room
temperature (about 20° C) with a a model 173 analog potentiostat, parc model 175 universal
programmer and an HP 7045A x-y chart recorder. In addition, all three cells were sealed and had
Argon gas streaming into the medium with *Shewanells putrefaciens* for the first thirty minutes of
the operation. After thirty minutes, the cells had Argon gas streaming on top of the medium. It is
important to note that unlike other microbial fuel cells in recent publications, the microbial fuel cells used in this experiment did have air cathodes or a barrier between the working and counter electrode compartment.

*Bacteria*

The three-compartment cells were inoculated with bacteria directly off of the petri dish.

**Voltammograms**

Voltammograms were made using the following settings: Upper Limit: 0.5V vs. Ag/AgCl 4M, Lower Limit: -0.6V vs. Ag/AgCl 4M, Scan Rate: 10 mV/sec, Working Electrode: carbon cloth

**Experiment**

**For the first experiment**

Cell 1

Cell one was assembled and inoculated on July 5, 2011 using sterilized growth medium made for Shewanella *putrefaciens* sp.200 prepared on June 28, 2011. Cell one was left under open circuit conditions when the potentiostat was not cycling. Cell one was deairated by bubbling nitrogen gas in the growth medium for 20 minutes while the solution was stirred. At the end of the 20 minutes, the nitrogen tube was removed from the liquid and allowed to flow at the surface to minimize oxygen diffusion. Voltammograms were made forty minutes for the next six hours the first day. During the second and third days of operation, voltammograms were made roughly every two hours. The growth medium was deairated with nitrogen when signs of an aerobic environment were seen (oxygen reduction in voltammograms, cell growth in solution).

Cell 2
Cell two was assembled with the same electrode configuration as cell one. Cell two was also deaerated with nitrogen gas. However, cell two was not inoculated with Shewanella putrefaciens sp.200 and the sterile growth medium in the cell was three weeks old. It is also important to note that the three compartment cell that was used to contain cell two was slightly larger than the three compartment cell used to contain cell 1. Cell two also had a working electrode area of about 7cm$^2$. Cell two was also only operated for three days as unwanted bacteria started to grow. However, a stable open circuit potential was recorded at the end of the first day.

**For the second and third experiment**

**Cell 1**

Cell one was assembled and inoculated on March 23th using sterilized growth medium without sodium lactate made for *Shewanella putrefaciens* sp.200 prepared on March 23th. Cell was lift under open circuit conditions when the potentiostat was not cycling. Cell one was deaerated by bubbling argon gas in the growth medium for 30 minutes while the solution was stirred. By the end of 30 minutes, the argon tube was removed from the solution and allowed to flow at the surface to minimize oxygen diffusion. Voltammograms were made every day since the first day of operation. Because of no concentration of Sodium Lactate, there was no sign of aerobic environment or microbe growth being seen.

**Cell 2 and cell 3**

Cell two and cell three were assembled with the same electrode configuration as cell one. However, unlike cell one, cell two has 3 ml of sodium lactate and cell three has 6 ml of sodium lactate. They were assembled and inoculated on the same day as cell one did. They were also deaerated by argon gas in the growth medium for 30 minutes where the solutions were stirred.
Later after 30 minutes, the argon tube was removed from the solution and allowed to flow at the surface. Same as cell one, voltammograms were performed every day since the first day of operation. There were signs of an aerobic environment that were seen (oxygen reduction in voltammograms, cell growth in solution).

Discussion and results

In the first run of the experiment, we were only focus on getting the lowest open circuit potential (OCU), there is no accurate OCU vs time data. After the successful proved experiment with and without Shewanella putrefaciens sp. 200, we start the second and third run to not only examine the lowest potential of the cells, but also monitor the rate of the changing at potentials.

![Open Circuit Potential](image-url)
As we can tell, the open circuit potentials from the second and third run decrease fast in the beginning, it slowed down and reach to the lowest point around day 10. In the figure 2, we think after day 12, Shewanella *putrefaciens* sp. 200 death rate going much higher since the concentration of lactate limit the energy they need to support their lives.

The cyclic voltammetry graph also indicate the same result. Below are the graphs in third run of the experiment in day 1, day 8, and day 15 respectively.

**Figure 2**

As we can tell, the open circuit potentials from the second and third run decrease fast in the beginning, it slowed down and reach to the lowest point around day 10. In the figure 2, we think after day 12, Shewanella *putrefaciens* sp. 200 death rate going much higher since the concentration of lactate limit the energy they need to support their lives.

The cyclic voltammetry graph also indicate the same result. Below are the graphs in third run of the experiment in day 1, day 8, and day 15 respectively.
Figure 3

Figure 4
We noticed that, at the end of the second day, cell one started to show a new set of oxidation and reduction peaks that occur at approximately the potentials of ferric iron reduction and ferrous iron oxidation.

**Conclusion**

The first run of the experiment clear show that bacteria (Shewanella *putrefaciens* sp. 200) can act as a catalyst to aid the oxidation of a chemical(s). The fuel cell basically reach to the lowest steady state potential in a week and hold in that position for short period of time until the lactate in the solution is run out. In future, we will add lactate after the fuel cell reach to the steady state and examine whether the lactate shortage caused the increasing in open circuit potential from third run.
We also noticed that the solution’s color changed. When the cell reach to the steady state, the solution from pure and transparent to turbid and dark black, one reason is the concentration of the bacteria increased, and more about the color change will be done in future research.