



## RESEARCH ARTICLE

### The optimization of growth parameters in a anodic chamber of a microbial fuel cell

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## ABSTRACT

The pollution is a result of drastically rising fossil fuel usage due to increased global population and industrialization. Today's world needs to replace fossil fuels with new and renewable energy sources. Microbial Fuel Cells (MFCs) are devices that convert the chemical energy to direct electricity by microbial metabolic activity. Microbial growth was actualized at the anodic chamber of MFC and must be controlled carefully by microbial methods. The main aim of this study is optimizing the growth parameters of microorganisms in two-chambered MFC with optical density, dried and wet weight of microorganisms that were grown at the anode chamber of MFC. Moreover, the Chemical Oxygen Demand (COD) values of medium were determined at fed cycles of MFC. The total volume of MFC was 0.7 L and the connection was made with the salt bridge between anode and cathode compartments. The anode chamber was used for bacterial growth that was taken from Akkaya Dam slime, was fed with molasses medium at 5 days intervals. The OD values were determined with a spectrophotometer at 600 nm, COD values were determined with the standard method and wet-dry weight also determined as a function of the incubation period.

**Keywords:** COD removal, growth parameters, microbial fuel cells (MFCs), microbial growth,  $q_m$  value

## 1. INTRODUCTION

The usage of fossil fuels has been increasing day by day, resulting in problems such as the greenhouse effect, global warming, and environmental pollution. Renewable and sustainable energy use is recommended as one of the most important solutions to get rid of the growing energy and environmental crisis. As a renewable energy source, alternative energy production methods are being investigated for electricity production through bacterial metabolism in microbial fuel cell and hydrogen cells. The main aim of microbial fuel cells (MFCs) is bioremediation of different wastes with microbial activity and during this process can provide electricity [1 – 9].

A biofuel cell is a device that directly converts the chemical bond energy to the electricity by microbial metabolism or enzymes. Chemical energy can convert the biocatalytic oxidation of organic or inorganic compounds into electrical energy by combining the oxidation of an oxidant at the interface between the anode and cathode. Microbial fuel cells are new biotechnological systems that use microorganisms as

catalysts while converting the chemical bond energy directly into electrical energy. In addition to these obvious benefits, they are likely to be used for wastewater treatment during bioelectricity generation and are therefore one of the areas of interest for researchers [10 – 15].

The fermented or non-fermented organic compounds in MFC might affect the power production of MFC. In microbial fuel cells, the microbial community development changed by the substrate which was used in anodic chamber, directly. In addition, the substrate used in the fuel cell also changes the strength and columbic efficiency of MFCs. Many different carbon sources that have simple (glucose, acetate, butyrate) or complex structure (starch, food processing wastes, synthetic or real wastewater, etc.) would be used as substrate in MFCs. Molasses produced in the sugar production process; animal feed, green manure or various fermentation processes in industrial use with low cost is preferred as raw material. Decolorization or remediation of molasses is a very difficult process due to the molasses high organic matter content, biological and chemical oxygen demand and dark brown melanoid pigment. On the other hand, molasses might

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be used as organic substrate for supported microbial growth in anodic chamber of MFC [12, 13, 16 – 18].

The performance of MFCs (voltage or power generation, removal of pollutants) is affected by microbial growth in microbial fuel cells. The microbial growth is depended to bacterial culture, substrate, temperature, and MFC cell type and these factors affected power production in MFCs. In studies related to microbial fuel cells, COD monitoring should be performed in order to determine the power efficiency as well as the treatment rate of wastewater. In addition, the degradation of the substrate that used in MFCs and reducing the Chemical Oxygen Demand (COD) is depending on the microbial growth at the anodic chamber of MFC [10 – 15].

Microbial communities have direct relationship with the usefulness of the substrate, pollutant removal and electricity generation of a MFC. It is understood from the literature that the biomass and microbial growth analysis achieved importance because of determine quality and standard property of the microbial community. The most important parameter affecting COD removal rate is the structure of microbial community developing in MFCs [4, 13, 19, 22 – 24]. Researchers need to determine the microbial growth parameters because of usage in the MFCs. The main aim of this work is to determine the microbial growth parameters such as optical density and microbial biomass in the incubation period. Moreover, the wet and dry weight of the microbial community will be given information about not only biological conditions but also biodegradation or bioremediation capacity for organic substrates or organof the MFC. The COD removal of microbial culture in the anodic chamber of MFC also determined and the COD removal for a dry weight of microbial culture was calculated from COD values.

## 2. MATERIALS AND METHODS

### 2.1. Microbial growth medium

In the anode chamber, molasses was used to provide bacterial growth. In the anode and cathode chamber, a 50 mM phosphate buffer solution was used, and 0.13 g L<sup>-1</sup> KCl was added to buffer to increase the conductivity of the solution. 50 mL L-1 of molasses solution was added to 1.0 g L<sup>-1</sup> (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> and 0.5 g L<sup>-1</sup> KH<sub>2</sub>PO<sub>4</sub> 50 mM 1 L phosphate buffer and the pH of the medium was settled at 7 [25]. In the cathode chamber, ferricyanide was used as the last electron acceptor to capture the electrons produced by the bacterial metabolism. 50 mM [K<sub>3</sub>Fe(CN)<sub>6</sub>] was added to 50 mM 1 L phosphate buffer solution [12, 20].

### 2.2. Microorganism source and growth conditions

The sediment sludge of Akkaya Dam was used as an initial source of microorganism. The growth of microorganisms determined as a function of optical density and biomass. Incubation conditions were conducted with an orbital shaker. The MFC was mixed at 50 rpm shaker incubator at 35°C. The optical density values were measured with a spectrophotometer at 600 nm. The biomass weight values were determined

as milligrams, microorganism biomass cleaned with saline solution three times and weighed by balance. After determining wet weight the samples were dried at 60°C for 24 hours and dry weight of samples weighed by balance [26].

### 2.3. MFC construction

In the study, glass MFC was used for voltage generation at total volume as 750 mL. 300 mL anode compartment was filled with molasses medium. The connection between anode and cathode compartment was made with a 150 ml salt bridge that was prepared with agar agar and a saturated KCl solution. The anode compartment fed with fresh molasses medium every 48 hours and sparged N<sub>2</sub> gaseous to ensure the anaerobic conditions [22].

### 2.4. Determination of chemical oxygen demand

The organic matter pollution was determined with the COD removal, which was determined with the standard potassium dichromate technique before and after the feeding of MFC at all the incubation period [21]. The ΔCOD referred to wasted COD and calculated by subtracting the COD values at the beginning and the end of the feeding period.

$$\Delta\text{COD} = \text{COD}_{\text{beginning}} - \text{COD}_{\text{end}} \quad (1)$$

The removed COD amount was calculated with the dry weight of microbial cells at the given formula for every feeding period [25].

$$qm = \frac{\Delta\text{COD}}{\text{Dry weight}} \quad (2)$$

## 3. RESULTS AND DISCUSSION

### 3.1. The microbial growth parameters

The microbial growth parameters such as optical density, wet and dry weight of biomass were determined to decide about the usefulness of the microbial community for removal of pollutants or power generation in a MFC. In this work the microbial growth parameters obtained from the incubation period were shown in the graphs below. The obtained data and equations are used to determine microbial growth at five days intervals of the incubation period in the anodic chamber of MFCs. The charts below were showed the correlation between the optical density and wet weight (Fig 1), and optical density and dry weight (Fig 2) of the microbial community that was grown at the anodic chamber.

The increase of microbial growth was slow at first feeding periods, on the other hand after 4<sup>th</sup> feeding period the increase of microbial growth was determined fastly. The optical density increase determination is a good way to set the wet weight of microbial biomass (Fig 1).

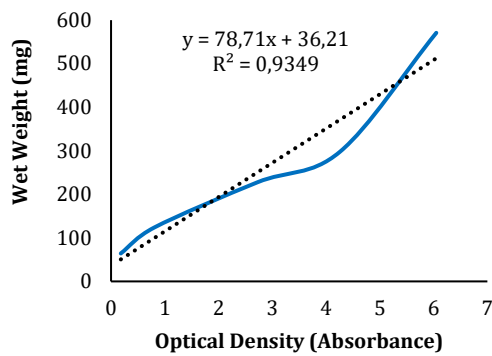


Fig 1. The optical density vs wet weight of biomass at 5 days interval feeding periods

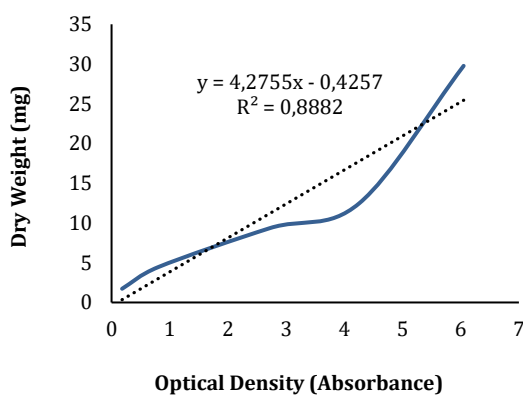
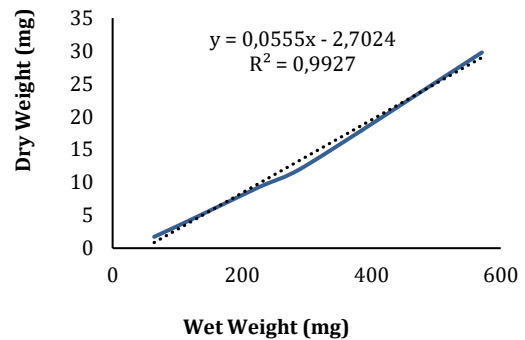


Fig 2. The optical density vs dry weight of biomass at 5 days interval feeding periods

As understood from the results mentioned at Fig 1 and Fig 2 that the microbial growth parameters could give information about biomass in the anode chamber and should be used for understanding the usefulness of the biomass for pollutant removal or power generation in MFCs. In Fig 1 and Fig 2, the determined correlations between optical density, wet and dry weights should be used for calculate the wet and dry weight values based on the increase in optical density for the microbial growth and biomass increase in anodic chamber of MFC.

Fig 3 is shown that the correlation with wet and dry weight of biomass at MFC. The correlation between wet weight and dry weight of microbial biomass was showed that the researcher could use the wet or dry weight of biomass interchangeably. The dry weight of biomass was depended to the wet weight of biomass directly and could be used for calculation of the pollutant removal rate per mg microbial biomass ( $q_m$ ) with these values. Therefore, the optical density values could be used for calculations of biomass weights.



### 3.2. The relationship between dry weight of biomass and COD removal

In Table 1 the biodegradation of organic compounds was determined by COD decrease at the feeding cycle of the anodic chamber of MFC. The  $q_m$  (mg  $\Delta$ COD  $mg^{-1}$  dry biomass) value refers to the COD removal for 1 mg dry biomass. The  $q_m$  values were showed that the maximum COD removal was determined at the 5 days intervals at incubation period and the prolonged incubation did not increase the COD removal for bacterial weight.

The COD removal was demonstrated as  $\Delta$ COD, determined by the given formula at material and method section, at the beginning and end of feeding periods were determined as 1741 and 3305 mg at minimum and maximum level at 7<sup>th</sup> and 5<sup>th</sup> feeding periods sequentially. Moreover, the  $q_m$  values were calculated as 497  $mg\ mg^{-1}$  and 1372  $mg\ mg^{-1}$  as maximum and minimum level at 7<sup>th</sup> and 5<sup>th</sup> feeding periods.

Table 1. The  $\Delta$ COD, dry weight of bacterial cells and  $q_m$  values at the different feeding periods at incubation period

Feeding cycle	$\Delta$ COD (mg)	Dry Weight (mg)	$q_m$ (mg $mg^{-1}$ )
1	2021	2.64	766
5	2496	2.54	983
10	2903	2.76	1051
15	2931	2.67	1096
20	3305	2.41	1372
25	2068	2.65	780
30	1741	3.50	497

#### 4. CONCLUSION

Industrialization has increased rapidly to meet the needs of the growing population and society. A large portion of domestic and industrial energy is derived from fossil fuels. The use of fossil fuels has been increasing day by day, resulting in problems such as greenhouse effect, global warming and environmental pollution. Fossil fuels are expected to run out in the near future. As a renewable energy source, alternative energy production methods such as electricity production through bacterial metabolism, microbial fuel cell and hydrogen cell are being investigated. Microbial fuel cells are new biotechnological systems that use microorganisms as catalysts while converting the chemical bond energy directly into electrical energy. Microbial fuel cells can generate energy at low costs and at the same time produce very little sludge waste. There are many factors affecting bacterial growth and thus electricity production in microbial fuel cells. Bacterial culture, substrate, mediator compound and MFC cell type used are the most important factors affecting power production in MFCs. As it understood from results above mentioned that the microbial growth parameters could give some information about biomass in the anode chamber. On the other hand, researchers must have to define the microorganisms that are found in microbial community.

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