

# Biotreatment of Slaughterhouse Wastewater Accompanied With Sustainable Electricity Generation in Microbial Fuel Cell

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## ABSTRACT

This study aimed to investigate the performance of microbial fuel cell (MFC) for simultaneous bioremediation of slaughterhouse wastewater and sustainable power generation. For the first time, an integrated system of tubular type microbial fuel cell (MFC) was used in this study. The MFC consisted of three concentric Plexiglas tubes; the inner tube was the anaerobic anodic compartment, the mid tube was the aerobic biocathodic chamber, and the outer tube act as an aerobic bioreactor for extended nitrification process. The MFC system was connected to a complementary external anaerobic bioreactor for denitrification process. The microbial fuel cell was inoculated with freshly collected activated sludge and was continuously fueled with simulated slaughterhouse wastewater. Results revealed that the removal efficiency of the chemical oxygen demand (COD) was up to 99%, and the power generation was  $165 \text{ mW/m}^2$ . Also, results demonstrated that maximum removal of  $\text{NO}_3^-$  via the denitrification process in the final effluent was 94.7% when the initial concentration of  $\text{NO}_3^-$  in the effluent of the extended bioreactor was  $15.2 \text{ mg/L}$ . Approximately; complete recovery of nitrogen gas was obtained in the complementary external anaerobic bioreactor. These results indicated that MFC could be a promising approach for slaughterhouse wastewater bioremediation and renewable power generation.

**Key words:** Microbial Fuel Cell, Slaughterhouse Industry, Wastewater, Biomass, Power Generation.

## 1. INTRODUCTION

Microbial fuel cells (MFCs) are devices that use bacteria as a catalyst for converting the organic

matters into electricity. An MFC typically consists of anaerobic anodic and aerobic cathodic chambers separated by ion conducting separators. In MFCs, microorganisms on the anode surface oxidize substrates and generate electrons and protons [1]. These electrons are transferred to the anode and pass towards the cathode via an external circuit to generate electricity. The generated protons diffuse through the solution across the separator towards the cathode. The electrons, protons and oxygen combine to form water on the cathodic electrode [2]. Previously reported studies have mentioned the importance of large surface area of anode to achieve high power generation. However, a greater extent of bacterial growth should assist in a higher electron transfer to the anode [3]. On the other hand, when wastewater happens uncontrolled, it oozes through to streams, rivers, and subsoil water causing quality deterioration of these media [4]. Slaughterhouse wastewater has been classified as industrial waste, the category of agricultural and food industries. The agro-industrial wastewaters such as slaughterhouses, fisheries, and seed oil processing are very strong in terms of contaminants concentrations and those significantly contribute to the overall pollution load imposed on the environment. In food industry, the meat industry is one of the largest producers of organic wastes. The wastewater generated from slaughterhouse industry generally contain high organic load and the treatment is primarily carried out using two major types of biological methods; aerobic and anaerobic. However, due to high strength, it is infeasible to treat some slaughterhouse wastewater using aerobic biological processes [5, 6]. Effluent from slaughterhouses and packing houses are usually heavily loaded with solids and floatable materials (fats), blood, manure, and a variety of organic compounds that generated from proteins. The composition of effluents depends on the type of production and facilities. The slaughterhouse

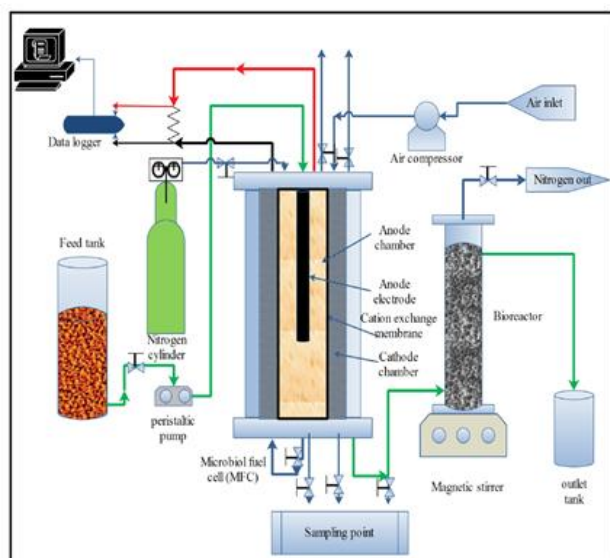
wastewater has high strength, in terms of biochemical oxygen demand (BOD), and chemical oxygen demand (COD), suspended solids (SS), nitrogen and phosphorus, as compared with domestic wastewater [6, 7]. As reported by Wiesmann et al. [8], the typical values of BOD, COD, and nitrogen in slaughterhouse wastewater are 1900, 2579-6650, and 80 mg/L, respectively. Wastewaters from slaughterhouses have been classified by the EPA as one of the harmful waste streams to the environment. The discharge of untreated slaughterhouse wastewater contributes to greatly degrading the aquatic environment.

This study aimed to design, construct, and evaluate the performance of a tubular type MFC for simultaneous biotreatment of simulated slaughterhouse wastewater, nitrogen recovery, and power generation.

## 2. MATERIALS AND METHODS

### Design of lab scale microbial fuel cell

An integrated MFC system consisted of tubular type microbial fuel cell surrounded by aerobic bioreactor and connected to an external anaerobic bioreactor was set up as given in Figs. 1 and 2.



**Fig. 1** Schematic diagram of the MFC system

The tubular dual-chamber MFC consisted of three concentric Plexiglas cylinders. The internal, mid, and external cylinders represent the anodic section, cathodic compartment, and the extended aerobic bioreactor, respectively. The anodic compartment was used for COD removal, whereby, the cathodic section was used for nitrification process ( $\text{NH}_4^+$  transformation to nitrite and nitrate).



**Fig. 2** Scheme of the MFC system

The extended aerobic bioreactor was utilized for further nitrification process and complete transformation of  $\text{NH}_4^+$  to nitrite and nitrate. An external cylindrical reactor was integrated with the MFC system and was used as an anaerobic bioreactor for denitrification process and nitrogen recovery.

### Inoculum and substrate

MFC inoculated with mixed culture (activated sludge) collected from the aeration tank in a local sewage treatment plant, Baghdad (Iraq). The MFC was fueled with simulated slaughterhouse wastewater having the characteristics given in Table 1.

Simulated wastewater was prepared by dissolving 1.278 g/L of sodium acetate ( $\text{CH}_3\text{COONa}$ ) and 0.191 g/L of ammonium chloride ( $\text{NH}_4\text{Cl}$ ) as a nitrogen source in mineral salt media (MSM) which was prepared by dissolving the following constituents in distilled water; 0.56 g/L  $(\text{NH}_4)_2\text{SO}_4$ , 0.20 g/L  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , 15 mg/L  $\text{CaCl}_2$ , 1 mg/L  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ , 20 mg/L  $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ , 0.42 g  $\text{NaHCO}_3$ .

### Analytical analysis and methodologies

Chemical oxygen demand (COD), biological oxygen demand (BOD), dissolved oxygen (DO), pH,  $\text{NO}_3^-$ ,  $\text{NH}_4^+$ ,  $\text{SO}_4^{2-}$ ,  $\text{Cl}^-$ ,  $\text{PO}_4^{3-}$ ,  $\text{NO}_3^-$  TDS and electrical conductivity tests were carried out by the researchers on a daily basis. The tests were conducted every day in accordance to the procedures outlined in the Standard

Methods [9] and the ASTM Manual of Water and Environmental Technology, D1426-92.

**Table. 1** Quality of slaughterhouse wastewater

Parameters	Units	Average concentration
COD	mg/l	1000
NH <sub>4</sub> <sup>+</sup>	mg/l	50
SO <sub>4</sub> <sup>-2</sup>	mg/l	70
Cl <sup>-</sup>	mg/l	49.0
TDS	mg/l	501
PO <sub>4</sub> <sup>-3</sup>	mg/l	3.07
pH	mg/l	6.7
EC	μS/cm	1100

### Process operation

To start up and operate the MFC, one liter of the activated sludge was placed in the anode compartment, and was sparged with nitrogen gas for a period of 10 min to maintain anaerobic environment. During the enrichment period, the anode was periodically fed by the mineral salt media (MSM) solution. After 45 days, the simulated slaughterhouse wastewater was continuously fed to the anodic chamber of MFC at a flow rate of 0.38 ml/min to achieve hydraulic retention time (HTR) of 30 h. Nitrogen gas was purged into the wastewater feed tank to eliminate oxygen content in wastewater.

At the same time, an air compressor with a maximum flow rate of 10 ml/min was connected to the cathode compartment to supply oxygen in a continuous manner. The up flowrate was controlled at 3 ml/min. Oxygen concentration was monitored continuously in the anodic compartment, and the absence of oxygen was observed indicating that the flow of oxygen from the cathodic to the anodic compartment was negligible and the anode compartment can be considered as anoxic. Also, the pH of the solution in the MFCs was monitored continuously and adjusted to 7-7.2 using 1M HCl or NaOH solution. The MFC integrated system was operated at ambient temperature range of 28-36 °C. The cathodic chamber was bio-catalyzed with aerobic mixed culture including nitrifying bacteria to oxidize the ammonium ions which passed from the anodic section to nitrate ions. The effluent from the cathodic chamber entered the external

cylinder which served as aerobic bioreactor for further oxidation of NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub><sup>-</sup> to NO<sub>3</sub><sup>-</sup>. In this aerobic bioreactor, almost all the remaining ammonium ions were converted to nitrate. Then the NO<sub>3</sub><sup>-</sup>-loaded effluent from the external cylinder was fed to the anaerobic bioreactor for the denitrification process. The released nitrogen gas was collected by syringe and the recovered nitrogen was analyzed by Gas Chromatograph (Packard Models 438A, Packard instrument company, USA). Daily effluent samples were analyzed for determination of ammonium, nitrate and COD concentrations. The MFC system was continuously operated for 120 days.

### 3. RESULTS AND DISCUSSION

Performance of MFCs mainly depends on the nature and the composition of wastewater. However, there are many factors that may influence the nitrogen removal and recovery in MFC such as oxygen, pH, C/N ratio, microbial community composition, reactor design, materials and electricity generation.

In the present investigation, the performance of the suggested MFCs was evaluated in terms of COD, ammonium and nitrate removal efficiency and power generation.

#### Chemical Oxygen Demand (COD) Removal

MFC was continuously operated for 120 days achieving a maximum COD removal efficiency of 99% as given in Fig. 3. Fast removal of COD was observed after 1 day of MFC start up. This could be attributed to the fact that the inoculation of MFC was favorable to the constituents of the slaughterhouse wastewater.

This observed maximum removal efficiency of COD in the current study was higher than the previously reported values in the range of (62%–92%) for the efficiency of COD removal from other types of industrial wastewaters in MFCs [10-15].

Nevertheless, the source and type of substrate, type and concentration of inoculum, geometric design of microbial fuel cell (MFC), type of electrodes, and other parameters highly affect the organics removal efficiency. However, the overall efficiencies observed for COD removal potentially indicate an effective wastewater treatment process.

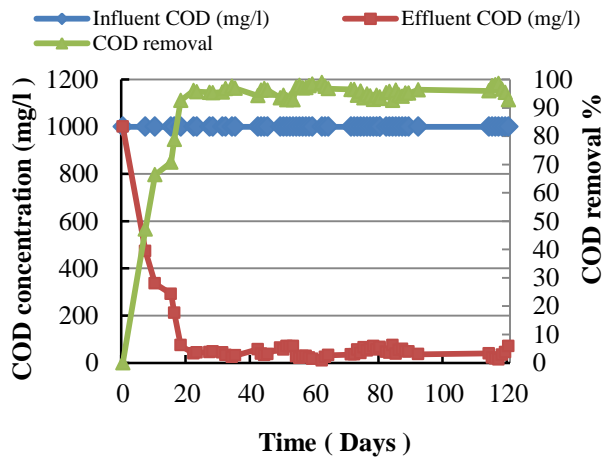


Fig. 3 Profile of COD removal efficiency

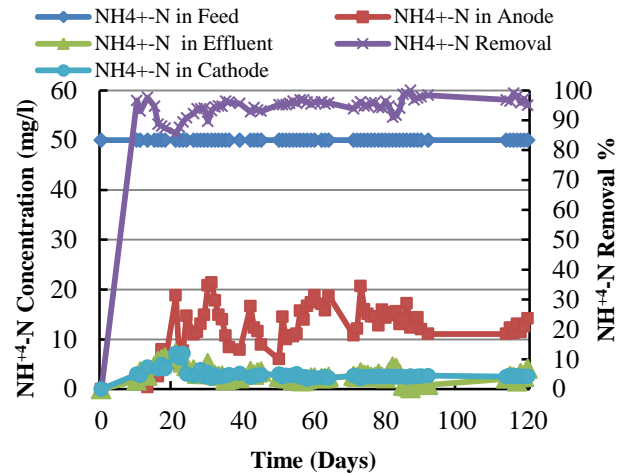


Fig. 4 Profile of ammonium removal in MFC

### Ammonium Removal

The ammonium ions concentrations were examined during the entire period of operation in each part of the MFC-integrated system. The initial ammonium-nitrogen concentration was 50 mg/l, and the removal efficiency of ammonium-nitrogen achieved 97%, after the first 10 days of continuous operation (Fig. 4). The inlet ammonium ions in the anode compartment were diffused through the membrane to the cathodic section in which the nitrification process occurred in the aerated biocathode by a series of bio-electrochemical reactions. This nitrification process was further completed in the extended aerobic bioreactor. After 33 days of continuous operation, the removal efficiency of ammonium-nitrogen was raised from 86% to 95% and the final concentration of ammonium in the effluent was found to be < 0.2 mg/L. These results indicated that the ammonium ions acted as electron donor in the nitrification process and continuously removed in the cathode compartment. The observed removal efficiency of ammonium-nitrogen could be considered comparable to the previously reported values up to 99% [16, 17].

### Nitrate removal

Nitrate was produced from the ammonium oxidation in the biocathode and the extended bioreactor. The nitrate concentration gradually increased from 1.8 to 13.5 mg NO<sub>3</sub>-N /L in the cathode, after 81 days at steady state condition of continuous operation as shown in Fig. 5. The maximum nitrate removal efficiency was 100% after 39 days. These results could be achieved due to ability of the biofilm to transfer electrons from the cathode to the intermediate electron acceptor during nitrification mechanism to increase the yield nitrate ions.

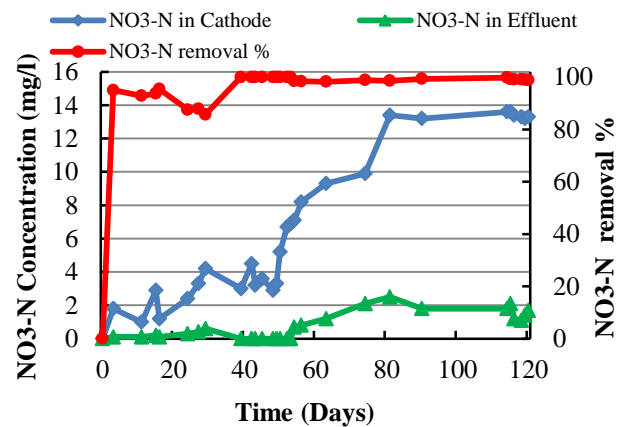


Fig. 5 Profile of NO<sub>3</sub> removal in MFC

However, the overall nitrate ions were reduced to nitrogen gas by the denitrification process by heterotrophic denitrifying bacteria in the external anoxic bioreactor.

### Current and power generation

Power production started on the first day of operation followed by a fluctuated trend of power for a period of 41 days, and then a steady state condition was observed at a maximum power and current densities of 165.22 mW/m<sup>2</sup> and 472 mA/m<sup>2</sup>, respectively (Fig.6). Maximum power and current density of the MFC integrated system suggests that electron transfer was also improved due to efficient oxidation of acetate substrate and the ammonium removal during nitrification process. The high power densities could be due to the better electrochemically active bacteria at high ionic strength, and they represent value of power

and current density was calculated based on surface area of anode.

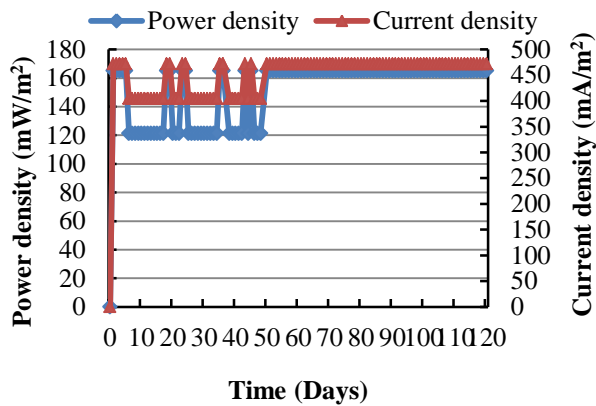


Fig. 6 Profile of current and power generation

Fig.7 illustrates the polarization curve plot. It is obvious from this plot that the maximum power density and current density are 165.22 mW/ m<sup>2</sup> and 472 mA/m<sup>2</sup>, respectively which were obtained at an external resistance of 100 Ω. These results indicated that in spite of the high strength of slaughterhouse wastewater, the removal efficiency and the corresponding power generation were favorable.

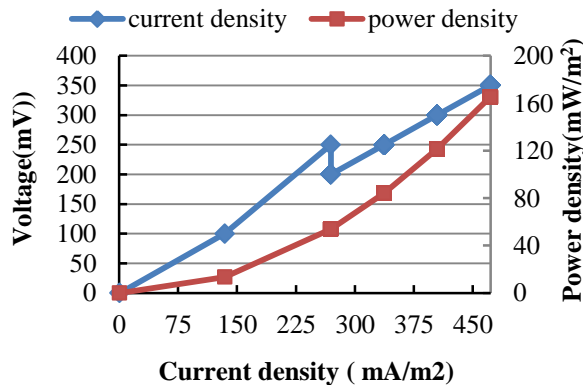


Fig. 7 Polarization curve for the MFC

#### Effect of external resistances

To investigate the effect of external resistances on the current generation, the voltage was recorded with different resistances across the anode and cathode to establish the relationship between the resistance and current. As presented in Fig. 8, at lower external resistance, more COD was removed resulted in a higher current generation.

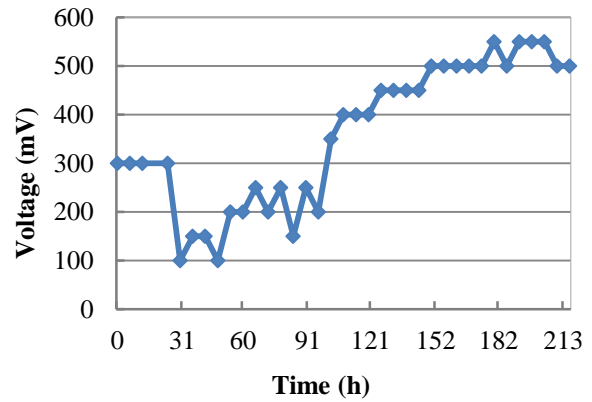


Fig. 8 Profile of voltage with different external resistances for MFC

## 4. CONCLUSION

This study demonstrated and evaluated the performance of a complete system of tubular type mediator-less microbial fuel cell. The MFC was fed with simulated slaughterhouse wastewater for simultaneous wastewater treatment, nitrogen recovery, and power generation. Results revealed that the removal efficiency of the chemical oxygen demand (COD) was up to 99%, and the power generation was 165.22 mW/m<sup>2</sup>. On the other hand, the maximum removal of NO<sub>3</sub><sup>-</sup> via the denitrification process in the final effluent was 94.7% indicating almost complete recovery of nitrogen gas in the complementary external anaerobic bioreactor. These results revealed that MFC could be a sustainable approach for simultaneous slaughterhouse wastewater bioremediation and power generation.

## 5. ACKNOWLEDGEMENT

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