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Effects of mediator producer and dissolved oxygen on electricity generation in a baffled stacking microbial fuel cell treating high strength molasses wastewater

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ABSTRACT

The effects of Pseudomonas aeruginosa, pyocyanin, and influent dissolved oxygen (DO) on the electricity generation in a baffled stacking microbial fuel cell (MFC) treating high strength molasses wastewater were investigated. The result shows that the influent chemical oxygen demand (COD) of 500–1000 mg l⁻¹ had the optimal substrate-energy conversion rate. The addition of a low density of *P. aeruginosa* (8.2 mg l⁻¹) or *P. aeruginosa* with pyocyanin improved the COD removal and power generation. This improvement could be attributed to the enhancement of electron transfer with the help of redox mediators. Influent DO at a concentration of up to 1.22 mg l⁻¹ did not inhibit the electricity generation. Large proportions of COD, organic-N and total-N were removed by the MFC. The MFC effluent was highly biodegradable. Denaturing gradient gel electrophoresis analysis shows that the added pyocyanin resided in the MFC for up to 14 days. An analysis of anode voltage reveals that microbial proton transport to the cathode was importantly responsible for the internal resistance.

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Introduction

The fermentation industry usually generates a large volume of high strength molasses wastewater, which has a high chemical oxygen demand (COD) and contains high concentrations of nitrogen pollutants. The discharge of untreated molasses wastewater often causes serious water pollution [1].

The microbial fuel cell (MFC) is a promising technology for treating organic wastewater and recovering energy; it uses microorganisms as the biocatalyst to convert organic

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pollutants to electricity [2–4]. A typical MFC consists of an anaerobic anode chamber, a cathode chamber, and a proton exchange membrane (PEM), which allows proton transfer from the anode chamber to the cathode chamber and prevents the diffusion of oxygen from the cathode chamber to the anode chamber [5,6].

Several factors can affect the performance of an MFC, including the types of wastewater, substrate concentrations, solution pH, hydraulic retention time, the degraders, mixing and the electrode materials [2,5,7]. Mechanical agitation promotes mixing of the solution, but consumes energy. To save energy, baffles are extensively used in anaerobic reactors to increase mixing. Recently, baffles have been used in MFCs for mixing the solution [7–9]. Graphite felt can be used as baffle. It is a porous material, and has the advantages of a high specific area and a low electrical resistance. Microorganisms, such as electrochemical active bacteria, can attach to, and grow on, the surface of carbon felt and directly transfer electrons to the anode, promoting the generation of electricity [10,11].

Mixed consortia from anaerobic sludge are often used as an inoculum in the anode chamber. The anaerobic mixed consortia are metabolically diverse [2,12]. Electrochemically active bacteria can attach themselves to the anode and directly transfer microbial electrons to the anode through the membrane-bound electroactive enzymes, such as C-cytochromes or bacterial nanowires [2,12].

However, some degraders cannot transfer electrons to the anode [12]. *Pseudomonas aeruginosa* reportedly can produce soluble endogenous redox mediators, such as phenazine pyocyanin, which can be used by *P. aeruginosa*, itself, and other bacterial species to facilitate microbial electron transfer [4,13,14]. Several studies have investigated the effects of adding *Pseudomonas* sp. to generation of electricity. However, most of such studies have been conducted in synthesis solutions [4,6,15,16]. Studies of the effects of *P. aeruginosa* on the treatment of real molasses wastewater using an MFC are lacking. Generally, the anode chamber of an MFC must be maintained under anoxic and anaerobic conditions to prevent the capture of microbial electrons by dissolved oxygen (DO) [2]. However, wastewaters can contain various levels of DO. Studies of the influence of DO on MFC performance are limited [17].

In this study, the effects of *P. aeruginosa*, pyocyanin, and influent DO on the performance of MFC in treating real molasses wastewater were studied. Anaerobic mixed consortia that were obtained from a winery (central Taiwan) was used as inoculums in the anode chamber. Denaturing gradient gel electrophoresis (DGGE) was employed to identify the distribution of inoculums and *P. aeruginosa* in the MFC. Polarization analysis and cyclic voltammetry (CV) were used to analyze the influence of different parameters on the bioelectrochemical characteristics of MFCs.

Materials and methods

Chemicals and bacterial strains

Anaerobic mixed consortia that were acquired from a winery (Fortune Brewery International Co., Taiwan) were used as inoculums in the anode chamber. The mixed consortia samples were transported to our laboratory and inoculated in an air cathode-type MFC with a volume of 5 liter at room temperature around 30 °C for pre-culture. The MFC system was flushed with gaseous nitrogen to maintain the solution under anoxic conditions (with a DO concentration of less than 1.0 mg l^{-1}). One milliliter of each of various condensed molasses fermentation solubles (Vedan Enterprise Corp., Taichung, Taiwan) was used as the sole carbon source in a long-term acclimation culture to obtain mixed microbial consortia that effectively degraded molasses. Phosphate buffer was used to maintain the solution pH of 7. Each liter of the medium/anolyte contained K₂HPO₄ 1.75 g, KH₂PO₄ 2.145 g, NH₄Cl 10 mg, MgCl₂·6H₂O 100 mg, CaCl₂ 45 mg, FeCl₃·6H₂O 1 mg,



Fig. 1 – The configuration of baffled stacking carbon-felt microbial fuel cell. S: solution, A: anode.

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