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Short communication

Microbial fuel cells continuously fuelled by untreated fresh algal biomass

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ABSTRACT

Microbial fuel cells (MFCs) are energy transducers that convert organic matter directly into electricity, via the anaerobic respiration of electro-active microorganisms. An avenue of research in this field is to employ algae as the organic carbon fuel source for the MFCs. However, in all studies demonstrating the feasibility of this principle, the algal biomass has always been pre-treated prior to being fed to MFCs, e.g. centrifuged, dried, ground into powder, and/or treated by acid-thermal processes. The alternative presented here, is a flow-through system whereby the MFCs were continuously fed by fresh algal biomass. The system consisted of i) a culture of *Synechococcus leopoliensis* grown continuously in a photo-chemostat, ii) a pre-digester initiating the digestion of the phototrophs and producing a fuel devoid of oxygen, and iii) a cascade of 9 MFCs, hydraulically and electrically independent. This compartmental system could in theory produce 42 W of electrical power per cubic metre of fresh culture ($6 \cdot 10^5$ cells mL⁻¹).

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1. Introduction

A recently developed avenue in the field of microbial fuel cells (MFCs) is the achievement of self-sustainability by the introduction of photo-autotrophy in the system. MFCs are energy transducers where electroactive microorganisms employ the anode as the electron acceptor for their anaerobic respiration of organic matter [1-4]. Typically, MFC setups comprise two electrodes: an anode and a cathode separated by a proton/cation exchange membrane. The electrons are donated to the anode and flow through an external circuit, before reducing an oxidising agent at the cathode (typically oxygen) combined with the incoming protons that have passed through the exchange membrane. The general idea of photosynthetic MFCs is to have a system whereby the energy comes from light and the carbon from carbon dioxide [5,6]. Phototrophs can also be employed as the catalyst for the cathodic oxygen reduction reaction [7–9], however this is not the scope of the present paper. The aim was to introduce photo-autotrophic microorganisms via a pre-digester, directly into a cascade of 8 MFCs as the carbon source (fuel) for electroactive heterotrophs, without any energy-consuming pre-treatment.

Phototrophs have previously been grown as biocatalysts for electron transfer using an added mediator (e.g. 2-hydroxy-p-

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naphthoquinone) [3]. These phototrophs were selected to produce hydrogen, as an endogenous mediator, which enables the development of a solar driven hydrogen fuel cell [10]. Recently it was shown that algae could serve as fuel for MFCs, either as an internally generated carbon-source, since they were growing within an illuminated anodic compartment [6,11,12], or added as an external source of carbon [13–15]. The incorporation of photo-autotrophy within the MFC itself, even though easily implemented, does not produce high voltage or power compared with more conventional MFCs containing anodophilic bacteria. The important element of distinction from previous work is that whenever algal biomass was employed as an external feedstock, it had always undergone energy-consuming treatments [13–15] such as centrifugation, drying and grinding, and/or acid-thermal treatments.

The aim of this work was therefore to demonstrate that algal biomass could be employed as MFC fuel without the need for energy consuming pre-treatment, by recreating a simplified trophic chain [6], thus maintaining a continuous hydraulic flow and supporting a dynamic steady state [16]. The trophic chain was initiated by oxygenic photosynthesis that fixed organic carbon in a photo-bioreactor. Syntrophic fermenters then initiate digestion and transformation of the photosynthetic biomass into secondary fermentation products (short chain fatty acids) in a pre-digester. The processed digest was then further hydrolysed and utilised by the electroactive organisms within the MFCs [4,6,17]. The aim was to investigate the possibility of running a cascade of MFCs continuously via pre-digestion of fresh algal biomass, rather than create another photomicrobial solar cell, or photosynthetic microbial fuel cell [18].

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2. Material and methods

2.1. Strain and culture media

The anodes were inoculated with activated sludge (Wessex Water, Saltford, UK). The microbial fuel cells (MFCs) were maintained in batch mode for 2 weeks under a 1.5 k Ω load, and subsequently operated under continuous flow.

The strain employed as the primary element of this artificial system was *Synechococcus leopoliensis* (A.591 obtained from Sciento, Manchester, UK). The medium used for the growth of the oxygenic phototrophs in the photo-chemostat was BG-11 [19]. The pH was adjusted to 7.2 prior to autoclaving.

2.2. MFC design and operation

A cascade of 9 MFCs was constructed as shown in Fig. 1, as opposed to a single enlarged MFC of equivalent volume; the cascade is also operated in sequential mode, where the output of one MFC feeds the next one downstream. Such a setup allows for a better utilisation of the organic matter into electricity because of shorter diffusion distances [20,21]. The anodic compartment (4.5 mL) was built in black acrylic material to avoid any development of phototrophic organisms. For the same reason all the tubing used was black ISO-Versinic (3 mm ID; Saint Gobain Performance Plastics, FR). The anodes were made from a 64 cm² sheet of carbon fibre veil (20 g m⁻²) (PRF Composite Materials Poole, Dorset, UK). The cathode employed the same carbon fibre veil but with a 160 cm² total surface area. Both electrodes were folded down to a 3D structure with an exposed surface area of 3.3 cm². The membrane had a surface area of 6.8 cm² and consisted of 2 mm thick terracotta (CTM potter supplies, UK). The water absorption (% of weight) of the terracotta membranes was $9.1\% \pm 0.3\%$ [22]. Tap water was employed as the catholyte with a continuous flow set at 5 mL min⁻¹. Light-tight gas-gap drippers were placed between each MFC to avoid any electrical cross-circuit via fluidic conduction from unit to unit, thus allowing each MFC to be electrically isolated for monitoring purposes. The total volume of the anodic compartment, tube and gas-gaps was approximately 6.5 mL.

2.3. Photo-chemostat design and operation

The photo-chemostat was implemented in order to have a continuous source of fresh algal biomass, as feedstock for the MFCs. Therefore, the optimisation of the growth conditions was not the aim of the present study. The photo-chemostat was a 1000 mL glass vessel with a rubber butyl septum (Glasgerätebau Ochs, Germany). The photochemostat was set on a 12-hour diurnal cycle to simulate light/dark periods typical of natural algal production systems. The 12 h light shift regime consisted of a light dose equivalent for 24 h of 40 μ E m⁻² s⁻¹ \pm 5 μ E m⁻² s⁻¹ (34W; Cool White, Sylvania), incubated at ambient room temperature (23 $^{\circ}C \pm 2 ^{\circ}C$), and under constant agitation. An aquarium pump was constantly pumping air into the vessel through an autoclaved air filter (Midisart® 2000 PTFE 17805; Sartorius). At first, the photo-chemostat was run in batch mode and inoculated with 10 mL of mother culture (5.85×10^3 cells mL⁻¹ final concentration). The algal growth was monitored by cell counts with a haemocytometer (AC1000 Improved Neubauer, Hawksley, UK) and the maximum growth rate calculated in order to select an appropriate input dilution rate for continuous operation as a photo-chemostat by connecting it, through a peristaltic pump (Welco Co., Ltd., Japan), to a 10 L tank of sterile media (BG-11). All tubing and connectors were autoclaved and assembled under sterile conditions. Moreover, to avoid any contamination of the photo-chemostat from downstream MFCs or pre-digester. two sterile anti-grow-back dripping mechanisms were placed at the output of the photo-chemostat. Two identical dripping systems were also introduced between the 10 L tank of sterile media and the photobioreactor to prevent the former from grow-back contamination.

2.4. Pre-digester

In the second phase of the experiment, a pre-digester was added between the photo-chemostat and the cascade of MFCs (Fig. 1). The pre-digester comprised a non-stirred 1000 mL light-tight glass bottle with a rubber butyl stopper, separating the vessel from the outside environment and allowing inlet and outlet tubes from the top. The inlet extended 5 cm into the vessel whilst the outlet tube reached 0.5 cm from the floor of the vessel. The pre-digester had a working volume of 500 mL \pm 15 mL (Fig. 1). The pre-digester was first inoculated with 50 mL of MFC effluent.

2.5. Data capture

The electrical output of each MFC was measured in millivolts (mV) against time using a PicoTech data logger (ADC-24, Pico Technology Ltd.). The voltage was recorded every 2 min. The current *I* in amperes (A) was calculated using Ohm's law, I = V/R, where *V* is the measured voltage in volts (V) and *R* is the known value of the resistor. The power output *P* in watts (W) was calculated as $P = I \times V$.



Fig. 1. Illustration of the in-line system setup. Each MFC is separated from the previous one by an air-gap (not shown) avoiding electrical connection through the anolyte. The pre-digester was introduced in the second phase of the experiment. The catholyte was recycled.

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