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# Comparison of synthetic medium and wastewater used as dilution medium to design scalable microbial anodes: Application to food waste treatment

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## HIGHLIGHTS

- Bioanodes were designed by replacing synthetic medium by costless wastewater.
- Using wastewater ensured 50% of the current density obtained in synthetic medium.
- Bioanodes differed in biofilm structures and redox charge contents.
- Current densities were directly correlated to the enrichment in Geobacteraceae.
- Wastewater is a suitable medium to design bioanodes for food waste treatment.

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## ABSTRACT

The objective was to replace synthetic medium by wastewater as a strategy to design low-cost scalable bioanodes. The addition of activated sludge was necessary to form primary bioanodes that were then used as the inoculum to form the secondary bioanodes. Bioanodes formed in synthetic medium with acetate 10 mM provided current densities of  $21.9 \pm 2.1 \text{ A/m}^2$ , while bioanodes formed in wastewater gave  $10.3 \pm 0.1 \text{ A/m}^2$ . The difference was explained in terms of biofilm structure, electrochemical kinetics and redox charge content of the biofilms. In both media, current densities were straightforwardly correlated with the biofilm enrichment in *Geobacteraceae* but, inside this family, *Geobacter sulfurreducens* and an uncultured *Geobacter* sp. were dominant in the synthetic medium, while growth of another *Geobacter* sp. was favoured in wastewater. Finally, the primary/secondary procedure succeeded in designing bioanodes to treat food wastes by using wastewater as dilution medium, with current densities of  $7 \pm 1.1 \text{ A/m}^2$ .

## 1. Introduction

The treatment of the domestic and industrial wastes that are produced by developed countries consumes huge amounts of energy. Abating the organic matter to a level that permits its disposal in natural environments generally involves aerobic processes, which consume considerable amounts of electrical energy for aeration (Miksch et al., 2013). Nowadays, much research focuses on extracting the chemical energy contained in so much wasted organic matter in order to treat it and, even better, to generate a positive energy balance. Microbial electrochemical technologies (METs) may be the tools that will make this dream a reality (Wang and Ren, 2013). They would offer the great advantages of abating organic matter in anaerobic conditions, thus saving the cost of aeration, and converting the chemical energy of organic compounds directly into electrical energy (microbial fuel cells), hydrogen (microbial electrolysis cells) or other services (microbial desalination cells, for example).

All these applications are based on the implementation of microbial bioanodes that use electrochemically active microorganisms able to oxidize the organic compounds and to transfer the electrons produced to the electrode material. Lab studies have had considerable successes in bioanode design by working in well-controlled conditions, in synthetic medium and with acetate as substrate (Rimboud et al., 2014). Current densities of  $5 \text{ A/m}^2$  have been obtained on graphite electrodes (Liu et al., 2008) and even more with sophisticated electrode designs, e.g.  $80 \text{ A/m}^2$  with stainless steel foam (Ketep et al., 2014) and up to  $67 \text{ A/m}^2$  with layered corrugated carbon (Baudler et al., 2014).







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A few bioanodes have also been successfully designed to treat real raw effluents (Pant et al., 2010) but current densities remain modest, for example 0.18 A/m<sup>2</sup> has been reported with urban wastewater (Rodrigo et al., 2007), 2 A/m<sup>2</sup> with brewery wastewater (Feng et al., 2008), 3 A/m<sup>2</sup> with chocolate industry wastewater (Patil et al., 2009), and 4–6 A/m<sup>2</sup> with paper mill effluent (Ketep et al., 2013). So far, the gap is large between the performance reached in synthetic media and that obtained in raw effluents.

Designing METs for the treatment of food wastes should be of great economic interest because food wastes are generated abundantly in concentrated form and need to be treated before disposal (Pant et al., 2013). In 2010, more than 34 million tons of food waste was generated in the United States, of which less than 3% was recovered and recycled, according to the U.S. Environmental Protection Agency (Li et al., 2013). Despite the high economic interest, studies that have investigated bioanodes able to treat food wastes remain rare (Table 1).

Food wastes are generally high in soluble COD and consequently need to be diluted to be oxidized by bioanodes. For this purpose, synthetic medium, drinking water or deionized water has generally been used. To our knowledge, only one study (Tenca et al., 2013) has overcome the problem by using food processing water with a fairly low COD (1.8 g/L) that did not require dilution. If the objective is to treat large quantities of wastes, it will not be possible to scale up large-sized METs that require huge amounts of phosphate or other buffer salts and the addition of micro-nutriments and vitamins. The cost would be too high, the release of salt would induce serious new environmental problems or require costly downstream separation and re-cycling processes. Using tap water would be less costly, but unfortunate in a world that should be increasing its efforts to preserve drinking water. Pant et al. (2013) have proposed a clever solution to this critical issue by using domestic wastewater as a dilution medium. The current densities obtained were modest  $(65 \text{ mA/m}^2)$  but the idea is surely interesting enough to deserve further investigation. Actually, if efficient bioanodes could be formed by diluting food wastes, or any other concentrated wastes, in wastewater, it would open up a cost-free method that could be easily scaled-up to large sized industrial METs.

The purpose of the present work was to help to define a scalable strategy for bioanode design by assessing the suitability of wastewater as a dilution medium. The work was based on the comparison of bioanodes designed in identical conditions in wastewater and in a synthetic medium. Bioanodes were formed in an optimal synthetic solution composed of phosphate buffer, macro-nutriments, trace minerals and vitamins, as commonly used in MET studies. In parallel, the same procedure was implemented in wastewater without any supplementation. The synthetic medium was inoculated with activated sludge and the necessity for the same inoculation with wastewater was discussed. In both cases, the bioanodes were formed in electroanalytical conditions under constant applied potential. The well-controlled electroanalytical conditions allowed the bioanode itself to be characterized by minimizing the interactions and rate-limiting steps that can occur in microbial fuel cells and microbial electrolysis cells (Rimboud et al., 2014). The design procedure consisted of, firstly, forming a primary bioanode that was then used to inoculate a secondary bioanode. This procedure has already been described in the literature and has led to secondary bioanodes displaying twice the current of the primary ones (Liu et al., 2008) and sustaining their performance over long periods (Baudler et al., 2014). This strategy designed in lab conditions with synthetic medium should be of great interest for real effluent because of its effectiveness and its capacity to be reproduced at large scale.

The bioanodes formed in synthetic medium and wastewater were compared by crossing electrochemical measurements, microscopy imaging and analysis of the microbial communities by 16S rRNA pyrosequencing. The comparison was performed with acetate as the substrate and the practical interest of the procedure was then checked with food wastes.

#### 2. Methods

#### 2.1. Electrochemical setup

Sealed vessels (600 mL) served as electrochemical cells that hosted the microbial medium and 3 electrodes. The 3-electrode set-ups consisted of a carbon cloth working electrode of 2 \* 3 cm<sup>2</sup> geometric surface area (Paxitech, Grenoble, France), a saturated calomel reference electrode (SCE, Radiometer Analytical, +0.24 V vs. SHE) and a 2 \* 3 cm<sup>2</sup> platinum grid used as the auxiliary electrode. The anode and cathode were connected to the electrical circuit by a 12-cm-long, 1-mm-diameter platinum wire. The anode (working electrode) was located at around 10 cm from the auxiliary electrode and as close as possible (around 0.5 cm) to the reference electrode. The working electrode was polarized at 0.15 V vs. SCE using a VSP potentiostat (Bio-Logic SA) interfacing with a computer (software EC-Lab) and the current was recorded every 10 min. Chronoamperometry was sometimes interrupted to perform cyclic voltammetry at low scan rate (1 mV/s) in the -0.6 to +0.3 V vs. SCE range.

The reactors were maintained at  $27 \text{ °C} \pm 2 \text{ °C}$  in a water bath and were initially purged with nitrogen for 15 min to remove oxygen. Substrate solutions in reactors were continuously lightly stirred (100 rpm).

#### Table 1

Studies using food wastes as a substrate to power microbial fuel cells or microbial electrolysis cells.

Food waste type	sCOD (gO <sub>2</sub> /L)	Dilution medium	Inoculum	Bioanode type	Configuration	Performances (A/m <sup>2</sup> )	References
Food processing wastewater (Cereal)	8.9	Synthetic medium	Sludge	Toray carbon paper (22.5 cm <sup>2</sup> )	MFC	0.24	Oh and Logan (2005)
Food industry waste (Yoghurt waste)	N/A	Synthetic medium	Anaerobic sludge	Graphite felt (10 cm <sup>2</sup> )	MFC	0.4	Cercado-Quezada et al. (2010)
Canteen food waste	12	Tap water	Anaerobic sludge	Graphite plate (70 cm <sup>2</sup> )	MFC	1.6	Goud et al. (2011)
Food waste from student's cafeteria	27.5	Synthetic medium	Anaerobic sludge	Carbon felt (50 cm <sup>2</sup> )	MFC	0.45	Choi et al. (2011)
Food waste leachate	12.7	Deionized water	Anaerobic sludge	Carbon felt (21 cm <sup>2</sup> )	MFC	0.1	Li et al. (2013)
Fermented reconstituted food waste	13	Domestic wastewater	Compost	Carbon felt (10 cm <sup>2</sup> )	MFC	0.065	Pant et al. (2013)
Food processing wastewater	1.8	No dilution	Acclimated biomass	Graphite fiber brushes	MEC	2	Tenca et al. (2013)

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