

**University of Massachusetts - Amherst**

---

**From the Selected Works of Kelly Nevin**

---

April 28, 2010

## Microbial Fuel Cells, A Current Review

Ashley E. Franks

Kelly Nevin, *University of Massachusetts - Amherst*



SELECTEDWORKS™

Available at: [http://works.bepress.com/kelly\\_nevin/60/](http://works.bepress.com/kelly_nevin/60/)

Review

## Microbial Fuel Cells, A Current Review

Ashley E. Franks \* and Kelly P. Nevin

Department of Microbiology, University of Massachusetts, Amherst, MA 01002, USA;

E-Mail: knevin@marlin.bio.umass.edu

\* Author to whom correspondence should be addressed; E-Mail: aefranks@microbio.umass.edu;  
Tel.: +1-413-575-9782; Fax: +1-413-577-4660.

Received: 19 January 2010; in revised form: 13 March 2010 / Accepted: 24 March 2010 /

Published: 28 April 2010

---

**Abstract:** Microbial fuel cells (MFCs) are devices that can use bacterial metabolism to produce an electrical current from a wide range organic substrates. Due to the promise of sustainable energy production from organic wastes, research has intensified in this field in the last few years. While holding great promise only a few marine sediment MFCs have been used practically, providing current for low power devices. To further improve MFC technology an understanding of the limitations and microbiology of these systems is required. Some researchers are uncovering that the greatest value of MFC technology may not be the production of electricity but the ability of electrode associated microbes to degrade wastes and toxic chemicals. We conclude that for further development of MFC applications, a greater focus on understanding the microbial processes in MFC systems is required.

**Keywords:** microbial fuel cell; extracellular electron transfer; conductive biofilm

---

### 1. Introduction

It is well recognized that alternative sources of energy are urgently required. Current reliance on fossil fuels is unsustainable due to pollution and finite supplies. While much research is being conducted into a wide range of energy solutions, it does not appear that any one solution alone will be able to replace fossil fuels in its entirety. As such it is likely that a number of different alternatives will be required, providing energy for a specific task in specialized ways in various situations. The

discovery that bacteria can be used to produce electricity from waste and renewable biomass [1-3] has gained much attention. Recently the increased interest in microbial fuel cell (MFC) technology was highlighted by the naming of *Geobacter sulfurreducens* KN400, a bacterial strain capable of high current production, as one of the top 50 most important inventions for 2009 by Time Magazine [4]. This list was also populated with other energy related devices such as solar shingles, smart thermostats and energy reducing light bulbs, further stressing the importance currently placed on energy.

The discovery that microbial metabolism could provide energy in the form of an electrical current [5,6] has led to an increasing interest and a dramatic rise in the number of publications in the field of MFC research. These systems are very adaptable and hold much promise to provide energy in a sustainable fashion but major improvements are required if widespread applications will be feasible. This review is unable to examine the entire field of MFC research in detail but hopes to highlight some important points regarding research in the field and recent important advances. Due to the sheer number of papers currently published regarding MFCs we hope that omission of many articles will not cause offence to their authors. This review article will examine MFC's currently in use, potential future applications and the limitations to implementing those applications. We suggest methods for improving the current output of a MFC. We also examine MFC applications in which microbes accept electrons from an electrode instead of donating them. This review will hopefully highlight some of the potential of and limitations to MFC technology implementation.

## 2. Current Applications for Microbial Fuel Cells

The first practical devices to be powered by MFC technology were reported in 2008 [7]. Meteorological buoys capable of measuring air temperature, pressure, relative humidity, water temperature, and transferring data via real-time line of sight radio frequency telemetry were exclusively powered by benthic MFCs. Benthic MFCs generate power through the microbial oxidation of organic substrates in anoxic marine sediments coupled to reduction of oxygen in the overlying water column. Electrons are generated from the metabolism of the naturally occurring microorganism in the marine sediments. As such, benthic MFCs do not require the addition of any exogenous microorganisms or electron shuttles [1,8,9]. Two separate benthic MFCs have been used to varying effect, the first, a prototype had a mass of 230 kg and a volume of  $1.3 \text{ m}^{-2}$ , and could sustain 24 mW or the equivalent of 16 alkaline D-cell batteries per year [7]. This initial design consisted of 7 sub units and required extensive manipulation of the marine sediment for deployment. A second design was developed that had a mass reduced to 16 kg, a volume of  $0.03 \text{ m}^{-2}$  and sustained 36 mW or the equivalent of 26 alkaline D-cell batteries per year [7] (Figure 1). This design required little manipulation of the sediment and is deployable by a single person. The meteorological buoys obtained their entire power from the benthic MFC allowing them to operate continuously and independently from the need to replace batteries. Benthic MFCs have been operated for several years with no decrease in power output. The authors estimated that a benthic MFC could provide power indefinitely at the same power levels and the same cost as a deep sea power and light enclosed lead acid battery could deliver for one year [7].

**Figure 1.** Leonard (Lenny) Tender standing next to a benthic MFC before deployment. Graphite plates (in the yellow casing) are deployed in the marine sediment with a graphite brush cathode in the overlaying water column.



Researchers have proposed methods to increase the power output from benthic MFCs, and thus broaden the devices the benthic MFCs are capable of powering. Since power is derived from the metabolism of the microorganisms in the sediment, the available organic substrate may be one of the limiting factors to higher levels of power production. The addition of insoluble slowly degrading organic substrates, such as chitin or cellulose, to the sediments has resulted in power increases [10–12]. These substrates are an appealing source as they are readily available and inexpensive. The increase in power density and longevity of the power output is affected by the particle size of the insoluble organic substrate [13]. Higher power densities could be reached, but for a shorter period of time, by using smaller sized chitin particles. Power increases from the addition of organic substrate implies the potential to electively increase power levels of benthic MFCs for a certain period of time, but not being sustainable in the long term unless organic substrate is continually introduced. This also could lead to sediment MFCs being situated in areas of high organic flux, such as underneath fish farms or sites of agricultural run off, which could provide an increase in organics for oxidation of the MFCs. To power devices requiring higher power levels than the MFCs are currently capable of producing, MFCs have been linked to charge capacitors to provide brief bursts of increased power which is slowly recharged by the MFC [14,15]. These have the advantage that charge can be stored and supplied intermittently at a level higher than that of the MFC. These studies are still lab

based and have not been incorporated into field-based trials but offer the potential to power a wider range of devices with a benthic MFC.

A limiting factor to general MFC use is the high cost of materials, such as the nafion membrane commonly used in laboratories as a proton permeable membrane. Attempts are currently underway to produce low cost MFCs constructed from earthen pots for use in India [16]. By removing the proton permeable membrane, utilizing locally produced 400 ml earthen pots, stainless steel mesh cathodes and a graphite plate anode, each MFC unit could be produced for US \$1. The earthen pot MFCs used sewerage sludge as an initial inoculum and experiments were conducted using acetate as a carbon source. While producing low levels of power, these devices could potentially be incorporated in large numbers into oxidation ponds for the treatment of concentrated wastewater while generating power. In areas where off grid applications are required, even low power MFC devices may prove useful. The World Bank has provided funding to a company named Lebone (<http://www.lebone.org/>) to start trials with MFC technology to provide energy to isolated communities. Initial trials will be based in Tanzania and attempt to provide power for high efficiency LEDs and battery powered devices. Current applications are all limited to low power level devices. If power can be increased, or cells engineered for specific applications, then a large range of potential applications have been speculated to be possible.

### 3. Potential Applications for Microbial Fuel Cells

The use of an anode as a final electron acceptor by bacteria has led to the possibility of a wide range of applications. It should be noted that many of these envisaged applications are not currently feasible and require significant improvements if they are to become viable technologies [17-19]. One of the most active areas of MFC research is the production of power from wastewaters combined with the oxidation of organic or inorganic compounds. Studies are demonstrating that any compound degradable by bacteria can be converted into electricity [20]. The range of compounds include, but by no means limited to, acetate [21,22], glucose [23], starch [24], cellulose [25], wheat straw [26], pyridine [27], phenol [28], *p*-nitrophenol [29] and complex solutions such as domestic waste water [30,31], brewery waste [32], land fill leachate [33], chocolate industry waste [34], mixed fatty acids [35] and petroleum contaminants [36]. Within these systems less biomass is also generally produced than their equivalent aerobic processes and without the need for an energy intensive aeration process less energy is required [7]. MFCs for the large scale treatment of wastewaters still face problems of scale up from laboratory experiments and slow rates of substrate degradation.

The ability of the MFC microbial communities to degrade a wide range of environmental pollutants may be more valuable than production of electricity itself in certain settings, especially when the MFC technology can be used for environmental clean up *in situ*. *Geobacter* species have been shown to be important in the anaerobic degradation of petroleum components and landfill leachate contaminants in ground water [37-41]. The oxidation of the contaminant is linked to the reduction of Fe (III). The oxidation and reduction process can be increased through the addition of Fe(III) chelators or electron shuttles [42-44] to promote increased transfer of electrons between cells and insoluble Fe(III) oxides. Contaminants often persist in the environment due to a lack of suitable electron acceptors [45-49] and the addition of chelators, electron shuttles or other electron acceptors in the subsurface environment is

not feasible. Pure cultures of *Geobacter metallireducens* are able to oxidize benzoate [1], and toluene [50] using an electrode as the final electron acceptor. Providing an electrode to hydrocarbon-contaminated soils accelerated the rates of toluene, benzene and naphthalene degradation [50]. The use of an electrode as an electron acceptor in the soil is attractive, as the microbes responsible for degradation will co-localize with the contaminant at the graphite anode. Once in position the electrode can provide a continuous long-term electron sink for the degradation of the harmful environmental contaminants. In this setting the electrons produced by the microbes in the form of current is irrelevant when compared to the increased rates of bioremediation. Likewise experiments have shown that MFCs may potential be able to remove fermentation inhibitors which accumulate in process water after the pretreatment of cellulosic biomass [51]. The removal of the inhibitors allows for increased fermentation product yields while providing small amounts of energy.

An unusual application for MFC technology is to power implanted medical devices using glucose and oxygen from blood. An implanted MFC could provide power indefinitely and negate the need for surgery to replace batteries. Abiotic fuel cells based on noble metal catalysts and activated carbon have been demonstrated to produce energy from blood glucose *in vitro* and *in vivo* [52,53]. Fuel cells based on enzymatic catalysts have also been shown to operate under physiological conditions but still require much improvement to become viable [54,55]. Interest has also been expressed in using human white blood cells as a source of electrons for an anode [56]. Experiments using white blood cells in phosphate-buffered saline solution with a ferric-cyanide cathode produced a low current level of 1–3  $\mu\text{Acm}^{-2}$  but it could not determine if electron transport to the anode was through a direct or indirect process [57].

#### 4. Microorganisms in A Microbial Fuel Cell

In its most basic form, a MFC is a device that uses microorganisms to generate an electrical current through the oxidation of organic material (Figure 2). Microorganisms in the MFC metabolize organic substrates and extracellularly transfer electrons to an electrode surface. The oxidation of the organic material liberates both electrons and protons from the oxidized substrate. Electrons are transferred to the anode and then to the cathode through an electrical network. The protons migrate to the cathode and combine with the electron and a catholyte, a chemical such as oxygen, which is reduced at the cathode surface. As such, an electrical current is generated in a fashion similar to a chemical fuel cell, but with microbes acting as a catalyst on the anode surface. Catalysts generally increase the rate of a reaction without being changed by or receiving energy from the reaction they catalyze. The microbes in a MFC are not true catalysts since they obtain energy from the oxidation of the substrate to support their own growth and create an energy loss. Microbes in a MFC may gain all the energy and carbon required for cellular growth from the oxidation of the complex organic material and as such MFC technology has been considered self-sustaining [58]. As long as conditions remain favorable for current production by the anode-associated microbes, a MFC has the potential to produce electricity indefinitely.

A diverse range of microorganisms are found in association with electrodes in MFC systems, especially when an environmental inoculum is used to seed the MFC [1,59–62]. A general term for bacteria associated with a surface is a biofilm. It is likely that not all of the organisms associated anode

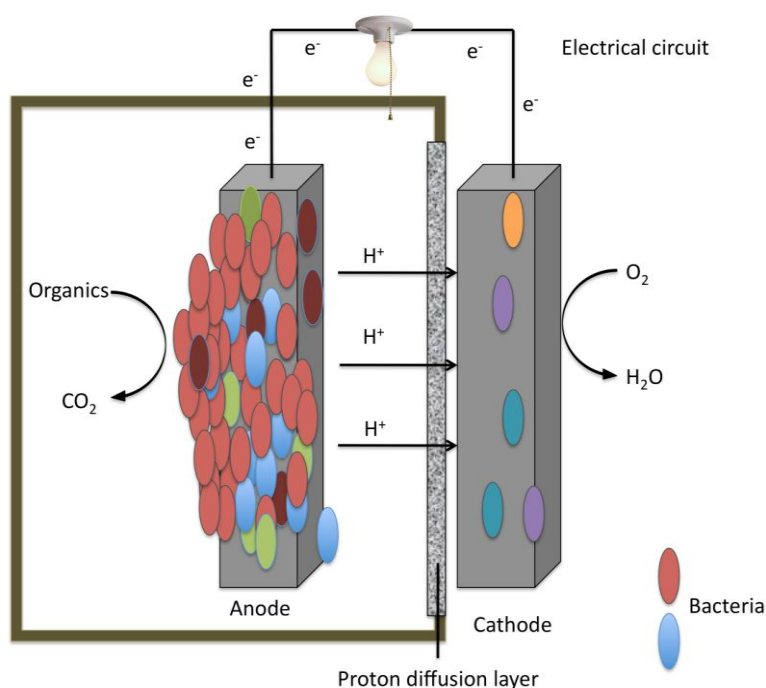
biofilm interact directly with the anode but may interact indirectly through other members of the electrode community. For example, *Brevibacillus* sp. PTH1 was found to be an abundant member of a MFC community. Power production by *Brevibacillus* sp. PTH1 is low unless it is cocultured with a *Pseudomonas* sp. or supernatant from a MFC run with the *Pseudomonas* sp. is added [63]. Pure cultures capable of producing current in a MFC include representatives of the Firmicutes [64] and Acidobacteria [65], four of the five classes of Proteobacteria [59,66–72] as well as the yeast strains *Saccharomyces cerevisiae* [73] and *Hansenula anomala* [74]. These organisms interact with an anode through a variety of direct and indirect processes producing current to varying degrees.

One common measure of MFC efficiency, coulombic efficiency, is a measure of the number of coulombs recovered as electrical current compared to the theoretical maximum number of coulombs recoverable from the organic substrate added to the system. The coulombic efficiency of the MFC is dependent, in part, on the microorganisms that are carrying out the oxidation and the organic carbon from which the electrons are derived [75–77]. This is due to the different metabolic pathways utilized by different microorganisms and the mechanisms by which the microorganisms transfer electrons to the anode. To gain the highest theoretical amount of energy from an organic substrate, the substrate needs to be completely oxidized to carbon dioxide with efficient transfer of electrons to the electrode. Without the complete oxidation of the organic substrate, energy is lost from the system in the form of unoxidized substrate. For example, studies found that *Shewanella oneidensis* that did not completely oxidize the organic substrate lactate in a MFC, leaving electrons unutilized as waste products such as acetate, had a coulombic efficiency of about 56.2% [78]. When microorganisms in the MFC system are capable of completely oxidizing the organic substrate to CO<sub>2</sub> higher coulombic efficiencies have been reported. Bacteria reported to be capable of the complete oxidation of an organic substrate in a MFC system include: *Geothrix fermentans* (94% coulombic efficiency oxidizing acetate) [65]; *Geobacter* species (approaching 100 % coulombic efficiency oxidizing acetate or 84% oxidizing benzoate) [1,21,22]; and *Rhodospirillum rubrum* (83% coulombic efficiency oxidizing glucose) [72]. Reported coulombic efficiencies can vary greatly when environmental inoculums are used, such as wastewater, with a maximum of 65–89 % being reported after microbial enrichment [79].

A number of other metrics have been proposed to measure MFC efficiency and for the comparison of MFC technology with other organic-matter-to-energy converting technologies. These include energy-capture efficiency, voltage efficiency, mass-transfer efficiency and process energy efficiency. Detailed discussions of these metrics are outside the scope of this review. Readers interested in discussion of MFC efficiency, and comparisons of MFC efficiency with technologies for generating bioethanol and anaerobic digestion to methane are directed to Rittmann *et al.* 2008 [80].

A major limitation to the MFC system is the reduction of molecular oxygen by the cathode. Various metals have typically been used to catalyze the cathodic reaction [81,82] but reduction of oxygen at the cathode is currently an important limiting factor in a MFC. To reduce these cathodic limitations researchers have increased the cathode to anode ratio [83] and used biological catalysts [84], which will be discussed later.

**Figure 2.** A diagram of a MFC containing a graphite anode acting as an electron acceptor for anaerobic microbial oxidation of organic compounds separated by a proton diffusion layer from an aerobic graphite cathode. Oxygen is combined with electron and protons at the cathode forming water. The reduction of oxygen at the cathode can be either an abiotic or biotic process.



Depending on the energy loss due to the cathode reaction and bacterial metabolism, a voltage of typically 0.3–0.5 V can be obtained from organic substrates such as glucose or acetic acid [19]. Anodes in these systems are usually small, generally in the range of  $cm^2$ , but projected power densities are reported on the scale of mW per  $m^2$ . Scaling in a MFC does not seem to be as easy as increasing the surface area of the anode and cathode. Problems associated with internal resistance, diffusion of substrate and protons are encountered. As recently highlighted [19], there has been initial success in increasing power output of MFC systems but the power outputs of MFCs are now plateauing. Another major draw back for many of the MFCs used by researchers is that the pumps, stirring bars and temperature control for the systems consume more power than the MFC itself generates. While these limitations need to be overcome to move from the laboratory to industrial large-scale MFC applications, some low power applications are being successfully deployed.

## 5. Electrical Interactions between Microbes and Electrodes

Cyclic voltammetry has been regularly exploited to investigate the electrochemical interactions of mixed or pure cultures with an anode [59,85–88]. These techniques can differentiate direct and indirect electron transfer process that occur within the anode-associated biofilms. Cyclic voltammograms have been used to interpret the electron transfer process occurring within an anode associated biofilm, which can vary depending on the stage of biofilm development, the biofilm community and power generation by the anode associated biofilm [59,88–92]. The cyclic voltammograms can be quite



complex, for example when *G. sulfurreducens* was examined using cyclic voltammetry the complexity of the voltammogram was increased at lower scan rates increasing the distinguishable redox systems from 2 to 4 or more [89]. As cyclic voltammetry has been shown to only probe the biofilm closest to the electrode, the usefulness of this technique in probing the bulk biofilm has been questioned due to significant potential gradient that maybe present in the biofilms [93]. Cyclic voltammetry cannot be used as a stand-alone method to determine the physiochemical nature of the mediator involved in the electron transfer process [94]. Many different bacterial species, including *Shewanella* species [95], *Geothrix fermentans* [96] and *Pseudomonas* species [85,97], have been identified to produce electron mediators but their use in high current producing MFCs has been questioned compared to bacteria that can directly transfer electrons to the electrode [58].

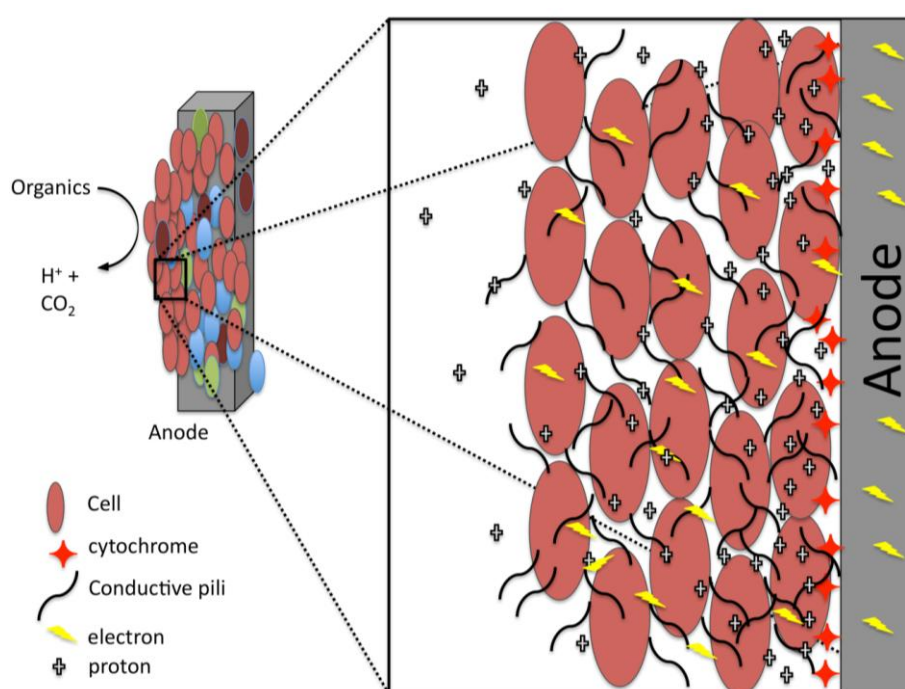
Likely mechanisms of direct and indirect electron transfer processes have been identified from studies of dissimilatory metal reducing bacteria that use insoluble iron oxides as a terminal electron acceptor [85,98,99]. Three main pathways for electron transfer have been identified and include: the production of electron shuttles [85], direct contact of outer surface c-type cytochromes [21] and long range contact via electrical conductive pili or nanowires [98] (Figure 3). The advantages of direct electron transfer and the use of pili to form a conductive biofilm has over the production of shuttles has been extensively reviewed elsewhere [18,19,58,100–102].

One of the most extensively studied microorganisms capable of high current densities in a MFC is *G. sulfurreducens*. This organism has become a model for bacterial processes in a MFC since: it is representative of *Geobacter* species commonly enriched for from environmental samples using a MFC [8,21,103]; pure cultures of *G. sulfurreducens* have been found to produce near or greater than maximum power of mixed species biofilms [22,104]; the full genome sequence is available [105] and is amenable to genetic manipulation [106]; whole genome microarray analysis is available [107]; and a genome-based *in silico* metabolic model is available [107]. Furthermore, *G. sulfurreducens* belongs to class of microbes referred to as electricigens, a term used to describe microbes that conserve energy to support growth by completely oxidizing organic compounds to carbon dioxide with direct electron transfer to the anode of the MFC [58]. Other proposed terms for microorganisms that can transfer electrons to an electrode include: anodophiles [108], exoelectrogens [109], electrogenic microorganisms [110], anode-respiring bacteria [76], and electrochemically active bacteria (EAB) [111]. Electricigens have many advantages in a MFC such as: high coulombic efficiency due to the complete oxidation of the organic substrate with transfer of electrons to the electrode; long term stability associated with the conservation of energy for maintenance and growth from the electron transfer to the anodes; and direct electron transfer to the anode by the bacteria negating the need for the addition of any exogenous or production of electron mediators [18].

Genome scale gene expression studies [112–114] and electrochemical analysis [115] indicate that cells in direct contact with the anode in a MFC interact through c-type cytochromes on the outer cell surfaces. Interestingly *G. sulfurreducens* biofilms can form biofilms greater than 50  $\mu\text{m}$  thick. All the cells in the thick biofilm are metabolically active [116] and contribute to current production [22,117]. Gene expression studies have suggested that the production of microbial nanowires is important for long-range electron transfer through the *G. sulfurreducens* biofilms (Figure 3) [117]. Modeling studies have also predicted that high current production by a thick anode associated biofilm is only feasible if the bacterial biofilm is conductive [118,119]. The production of a conductive biofilm is highly unusual

as most biofilms act as insulators [120–122]. A major breakthrough in the field would be the measurement and determination of the conductivity, and the components responsible of conductivity, of a bacterial biofilm in a MFC.

**Figure 3.** In current producing *Geobacter sulfurreducens* biofilms cells close to the electrode are proposed to transfer electrons via membrane bound cytochromes, where as cells furthest from the electrode are able to use a conductive nanowire network for long range electron transfer to the electrode. Oxidation of the organic substrate throughout the biofilm leads to an accumulation of protons within the biofilm. The proton concentration is closet at the anode surface and can be greater then ten fold when compared to the bulk fluid.



An important point recently highlight is the general misconception that the bacteria on the electrode gain energy from the electron shuttle or the electrode directly itself [101]. The bacterial cells gain energy from the pumping of protons across the inner membrane to form a proton gradient, which drives the formation of ATP from ADP through ATPase. As such, extracellular electron transfer acts to move the electron to the anode surface but does not gain any further energy for bacterial growth. It is the creation of a proton gradient that drives the synthesis of ATP and provides metabolic energy for the bacterium.

The production of high current densities by microorganisms in a MFC is highly artificial with no natural equivalent in the environment. Attempts to improve current production through genetic engineering have met with little success [123]. Over production of cytochromes or the microbial nanowires did not lead to an increase in power production, likewise the creation of an ATP drain, predictive by the metabolic model to increase in metabolism [124], did not increase current production. This observation highlights that current production by bacteria in a MFC is a complex process that is

highly regulated and requires more than changes in a few genes or changes in bacterial respiration rates to increase current production. Adaptive selection within a MFC has met with more success at producing a strain capable of increased current density [125]. Over a 5 month period a MFC was operated at a low potential and led to the isolation of the variant of *G. sulfurreducens* named KN400 capable of an 8 fold increase in power density. Of interest is that this strain produces a thinner biofilm with less outer surface cytochromes but an abundance of nanowires providing further insight into the complex process of electron transfer at high current densities [125].

## 6. Proton Inhibition in Microbial Fuel Cell Biofilms

The oxidation of organic material produces both protons and electrons. The electrons are removed instantaneously via the conductive biofilm and the electrical circuit of the MFC. The larger protons have to migrate out of the biofilm to the cathode. This occurs at a much slower rate and may cause a bottleneck inhibiting power production. For every electron produced in the form of current, a proton is also produced within the biofilm (Figure 3). Using a modified MFC and a fluorometric pH sensitive dye, a proton gradient was observed across the biofilm between the anode surface and the bulk fluid [116]. The specific production of current by the microbes in the MFC caused a ten-fold increase in proton concentration, equivalent to 1 pH unit. A decrease in the pH of the bulk fluid of a MFC has been shown to decrease power production [83,126]. Modeling studies predicted that proton accumulation would lead to zones of metabolic inactivity within the biofilm [127], but metabolic staining indicated activity throughout the entire biofilm [128,129].

A novel technique that allowed spatial transcriptional profiling of the current producing biofilm indicated that there was no significant difference in metabolism between the cells closest to the anode and those furthest from the electrode surface [129]. Gene expression patterns suggest that the cells closest to the anode surface, while metabolically active, expressed a range of proteins commonly associated with environmental stress, expected to be a result of the proton accumulation. Interestingly the cell furthest from the anode did not express any genes commonly associated with electron acceptor limitation. Combined with the observation that these cells are metabolically active it seems that the biofilm thickness is limited due to unknown factors. Another interesting observation is that biofilms produced by *G. sulfurreducens* are heterogeneous with pillar structures [116] whereas the high current density strain KN400 produces more power from a thinner, but more homogeneous biofilm [125]. This also indicates that the way the cells are packed in a biofilm is more important than just producing thicker biofilms.

## 7. Cathode Interactions

The reduction of oxygen at the cathode of MFCs is recognized as one of the current bottlenecks in power production [19,130–132]. Oxygen is an ideal acceptor for use in a MFC due to its high oxidation potential, low cost and formation of water as its waste product. Plain graphite/carbon electrodes are a commonly used electrode material due to cost and performance. Oxygen reduction occurs at a very slow rate on the surface of carbon electrodes and leads to a high over potential, thought to be one of the most limiting factors in high current density MFCs [133]. To overcome this

electrode overpotential, laboratory based MFC systems commonly use potassium ferricyanide as an electron acceptor [22,87,108,125]. The use of potassium ferricyanide leads to a low overpotential of the carbon cathode, and allows the MFC to work close to open circuit potential. The use of ferricyanide outside the laboratory is not practical due to toxicity and insufficient reoxidation by oxygen requiring the catholyte to be regularly replaced [87,134]. Furthermore the use of catalysts, such as platinum or other non-precious metals [81,82], may also be cost inhibitory for large-scale applications and not suitable for long-term applications. In the search for a suitable catalyst for oxygen reduction in a MFC system research is investigating the use of a biocathode.

Microorganisms can accept electrons from an electrode and reduce organic and inorganic compounds. Initial studies demonstrated that *Geobacter* species could utilize an electrode as a sole electron source [135]. A negatively poised electrode enriched for *Geobacter* species from sediment accompanied by a corresponding reduction of nitrate [135]. Quite interestingly pure culture studies with further *Geobacter* species showed biological reduction of nitrate and fumarate, as well as reduction of insoluble U(VI) to U(IV), with an electrode species acting as the sole electron donor [136]. Microbes capable of manganese oxidation have also been demonstrated to increase current by two orders of magnitude by cycling manganese on a cathode. Manganese used as a final electron acceptor is reduced and precipitated on the electrode as Mn(IV), the manganese oxidizing organisms oxidize the Mn(IV) to Mn(II) which in turn can be reduced once again by the electrode [137]. The use of a biocathode has recently gained interest in the treatment of wastewater. The use of a biocathode offers the simultaneous removal of organic compounds at the anode link to nitrogen removal at the cathode [138].

Biocathodes using oxygen as a final electron acceptor have been used to improve the performance of marine MFCs with stainless steel electrodes. The performance of a wet air cathode inoculated with a consortium of sludge and sediment microbes was increased through the interactions of the microorganisms on the cathode surface [131,139]. The use of a biocathodes has also been reported to reduce charge transfer resistance of the cathode from 188 to 17 ohms [140] and 40.2 to 12 ohms [141]. Microbial community analysis of the biocathode has identified a wide range of organisms present in the anode community including representatives of the divisions: *Betaproteobacteria*, *Bacteroidetes*, *Alphaproteobacteria*, *Chlorobi*, *Deltaproteobacteria*, *Actinobacteria*, and *Gammaproteobacteria* [141]. Not surprisingly, since this MFC was set up to investigate nitrate removal at the cathode, bacteria with the ability to reduce nitrate dominated the microbial community. When oxygen was used as a final electron acceptor then *Bacteroidetes* isolates have been found to dominate the community [84]. Pure isolates have only just been isolated from these communities but show significant improvements when used individually on the electrodes. How these bacteria interact with the electrode surface and the mechanisms for accepting electrons from the electrode so far remain unknown.

The ability of microbes to accept electrons from an electrode has potential in bioremediation and bioproduction. *G. lovleyi* and *Anaeromyxobacter dehalogenans* have been shown to reduce chlorinated compounds with as electrode as the sole source of electrons [142,143]. Field trials have also shown that electrons provide by an electrode can improve uranium removal in contaminated sites [127]. Bioremediation using electrodes as electron donors has several advantages over traditional methods including positive selection of bacterial strains and reduced competition for electron donor [135]. While these process described are all of interest due to applications associated with bioremediation, it

has yet to be seen if this process can be used for commercial applications, such as the modification or production of commodity chemicals [136].

## 8. Summary

MFCs are a promising technology for the production of electricity from organic material and wastes. Currently limited applications are possible because of low MFC power output. An understanding of the microbiology of the current producing process is required before further advances in power output are possible. Two major problems that need to be addressed is proton accumulation within the biofilm and over potential at the cathode. Of interest are some current application of MFCs where current production is not the major advantage, but wastewater treatment or bioremediation using a cathode or anode maybe much more promising then the electrical production of the MFC itself.

## Acknowledgements

The authors wish to thank Lenny Tender, NRL, Washington for providing the picture of the benthic microbial fuel cell. The authors are supported by the Office of Science (BER), U.S. Department of Energy, Cooperative Agreement No. DE-FC02-02ER63446 and Office of Naval Research Award No. N00014-07-1-0966. The authors wish to thank the reviewers for their constructive comments.

## References and Notes

1. Bond, D.R.; Holmes, D.E.; Tender, L.M.; Lovley, D.R. Electrode-reducing microorganisms that harvest energy from marine sediments. *Science* **2002**, *295*, 483–485.
2. Kim, H.J.; Park, H.S.; Hyun, M.S.; Chang, I.S.; Kim, M.; Kim, B.H. A mediator-less microbial fuel cell using a metal reducing bacterium, *Shewenella putrefaciens*. *Enzyme Microb. Technol.* **2002**, *30*, 145–152.
3. Kim, H.J.; Hyun, M.S.; Chang, I.S.; Kim, B.H. A microbial fuel cell type lactate biosensor using a metal-reducing bacterium, *Shewanella putrefaciens*. *J. Microbiol. Biotech.* **1999**, *9*, 365–367.
4. Time. The 50 Best Inventions of 2009. Available online: [http://www.time.com/time/specials/packages/article/0,28804,1934027\\_1934003\\_1933965,00.html](http://www.time.com/time/specials/packages/article/0,28804,1934027_1934003_1933965,00.html) (accessed on 25 March 2010).
5. Potter, M.C. Electrical effects accompanying the decomposition of organic compounds. *Proc. R. Soc. Lond. B* **1911**, *84*, 260–276.
6. Potter, M.C. On the difference of potential due to the vital activity of microorganisms. *Proc. Univ. Durham Phil. Soc.* **1910**, *3*, 245–249.
7. Tender, L.; Gray, S.; Groveman, E.; Lowy, D.; Kauffma, P.; Melhado, R.; Tyce, R.; Flynn, D.; Petrecca, R.; Dobarro, J. The first demonstration of a microbial fuel cell as a viable power supply: Powering a meteorological buoy. *J. Power Sourc.* **2008**, *179*, 571–575
8. Tender, L.M.; Reimers, C.E.; Stecher, H.A.; Holmes, D.E.; Bond, D.R.; Lowy, D.A.; Pilobello, K.; Fertig, S.J.; Lovley, D.R. Harnessing microbially generated power on the seafloor. *Nat. Biotechnol.* **2002**, *20*, 821–825.

9. Reimers, C.E.; Tender, L.M.; Fertig, S.; Wang, W. Harvesting energy from the marine sediment-water interface. *Environ. Sci. Technol.* **2001**, *35*, 192–195.
10. Rezaei, F.; Richard, T.L.; Logan, B.E. Enzymatic hydrolysis of cellulose coupled with electricity generation in a microbial fuel cell. *Biotechnol. Bioeng.* **2008**, *101*, 1163–1169.
11. Rezaei, F.; Richard, T.L.; Logan, B.E. Analysis of chitin particle size on maximum power generation, power longevity, and Coulombic efficiency in solid-substrate microbial fuel cells. *J. Power Sourc.* **2009**, *192*, 304–309.
12. Rezaei, F.; Richard, T.L.; Brennan, R.A.; Logan, B.E. Substrate-enhanced microbial fuel cells for improved remote power generation from sediment-based systems. *Environ. Sci. Technol.* **2007**, *41*, 4053–4058.
13. Rezaei, F.; Xing, D.; Wagner, R.; Regan, J.M.; Richard, T.L.; Logan, B.E. Simultaneous cellulose degradation and electricity production by *Enterobacter cloacae* in a microbial fuel cell. *Appl. Environ. Microbiol.* **2009**, *75*, 3673–3678.
14. Donovan, C.; Dewan, A.; Heo, D.; Beyenal, H. Batteryless, wireless sensor powered by a sediment microbial fuel cell. *Environ. Sci. Technol.* **2008**, *42*, 8591–8596.
15. Dewan, A.; Donovan, C.; Heo, D.; Beyenal, H. Evaluating the performance of microbial fuel cells powering electronic devices. *J. Power Sourc.* **2009**, *195*, 90–96.
16. Behera, M.; Jana, P.S.; Ghangrekar, M.M. Performance evaluation of low cost microbial fuel cell fabricated using earthen pot with biotic and abiotic cathode. *Bioresour. Technol.* **2009**, *101*, 1183–1189.
17. Lovley, D.R. Microbial energizers: Fuel cells that keep on going. *Microbe* **2006**, *1*, 323–329.
18. Lovley, D.R.; Nevin, K.P. Electricity production with electricigens. In *Bioenergy: Microbial Contributions to Alternative Fuels*; Wall, J., Harwood, C., Demain, A., Eds.; ASM Press: Washington D.C., USA, 2008; pp. 295–306.
19. Logan, B.E. Exoelectrogenic bacteria that power microbial fuel cells. *Nat. Rev. Microbiol.* **2009**, *7*, 375–381.
20. Pant, D.; Van Bogaert, G.; Diels, L.; Vanbroekhoven, K. A review of the substrates used in microbial fuel cells (MFCs) for sustainable energy production. *Bioresour. Technol.* **2009**, *101*, 1533–1543.
21. Bond, D.R.; Lovley, D.R. Electricity production by *Geobacter sulfurreducens* attached to electrodes. *Appl. Environ. Microbiol.* **2003**, *69*, 1548–1555.
22. Nevin, K.P.; Richter, H.; Covalla, S.F.; Johnson, J.P.; Woodard, T.L.; Orloff, A.L.; Jia, H.; Zhang, M.; Lovley, D.R. Power output and coulombic efficiencies from biofilms of *Geobacter sulfurreducens* comparable to mixed community microbial fuel cells. *Environ. Microbiol.* **2008**, *10*, 2505–2514.
23. Kim, N.; Choi, Y.; Jung, S.; Kim, S. Effect of initial carbon sources on the performance of microbial fuel cells containing *Proteus vulgaris*. *Biotechnol. Bioeng.* **2000**, *70*, 109–114.
24. Lu, N.; Zhou, S.G.; Zhuang, L.; Zhang, J.T.; Ni, J.R. Electricity generation from starch processing wastewater using microbial fuel cell technology. *Biochem. Eng. J.* **2009**, *43*, 246–251.
25. Ren, Z.; Steinberg, L.M.; Regan, J.M. Electricity production and microbial biofilm characterization in cellulose-fed microbial fuel cells. *Water Sci. Technol.* **2008**, *58*, 617–622.

26. Zhang, Y.; Min, B.; Huang, L.; Angelidaki, I. Generation of electricity and analysis of microbial communities in wheat straw biomass-powered microbial fuel cells. *Appl. Environ. Microbiol.* **2009**, *75*, 3389–3395.
27. Zhang, C.; Li, M.; Liu, G.; Luo, H.; Zhang, R. Pyridine degradation in the microbial fuel cells. *J. Hazard. Mat.* **2009**, *172*, 465–471.
28. Luo, H.; Liu, G.; Zhang, R.; Jin, S. Phenol degradation in microbial fuel cells. *Chem. Eng. J.* **2009**, *147*, 259–264.
29. Zhu, X.; Ni, J. Simultaneous processes of electricity generation and p-nitrophenol degradation in a microbial fuel cell. *Electrochem. Comm.* **2009**, *11*, 274–277.
30. Liu, H.; Logan, B.E. Electricity generation using an air-cathode single chamber microbial fuel cell in the presence and absence of a proton exchange membrane. *Env. Sci. Tech.* **2004**, *38*, 4040–4046.
31. You, S.J.; Zhao, Q.L.; Jiang, J.Q. Biological wastewater treatment and simultaneous generating electricity from organic wastewater by microbial fuel cell. *Huan Jing Ke Xue* **2006**, *27*, 1786–1790.
32. Feng, Y.; Wang, X.; Logan, B.; Lee, H. Brewery wastewater treatment using air-cathode microbial fuel cells. *App. Microbiol. Biotechnol.* **2008**, *78*, 873–880.
33. Gávez, A.; Greenman, J.; Ieropoulos, I. Landfill leachate treatment with microbial fuel cells; scale-up through plurality. *Bioresour. Technol.* **2009**, *100*, 5085–5091.
34. Patil, S.A.; Surakasi, V.P.; Koul, S.; Ijmulwar, S.; Vivek, A.; Shouche, Y.S.; Kapadnis, B.P. Electricity generation using chocolate industry wastewater and its treatment in activated sludge based microbial fuel cell and analysis of developed microbial community in the anode chamber. *Bioresour. Technol.* **2009**, *100*, 5132–5139.
35. Freguia, S.; Teh, E.H.; Boon, N.; Leung, K.M.; Keller, J.; Rabaey, K. Microbial fuel cells operating on mixed fatty acids. *Bioresour. Technol.* **2009**, *101*, 1233–1238.
36. Morris, J.M.; Jin, S. Feasibility of using microbial fuel cell technology for bioremediation of hydrocarbons in groundwater. *J. Environm. Sci. Health A: Tox./Hazard. Subst. Environm. Eng.* **2008**, *43*, 18–23.
37. Lin, B.; Braster, M.; van Breukelen, B.M.; van Verseveld, H.W.; Westerhoff, H.V.; Roling, W.F.M. *Geobacteraceae* community composition is related to hydrochemistry and biodegradation in an iron-reducing aquifer polluted by a neighboring landfill. *Appl. Environ. Microbiol.* **2005**, *71*, 5983–5991.
38. Rooney-Varga, J.N.; Anderson, R.T.; Fraga, J.L.; Ringelberg, D.; Lovley, D.R. Microbial communities associated with anaerobic benzene degradation in a petroleum-contaminated aquifer. *Appl. Environ. Microbiol.* **1999**, *65*, 3056–3064.
39. Roling, W.F.M.; van Breukelen, B.M.; Braster, B.L.; van Verseveld, H.W. Relationships between microbial community structure and hydrochemistry in a landfill leachate-polluted aquifer. *Appl. Environ. Microbiol.* **2001**, *67*, 4619–4629.
40. Lovley, D.R.; Baedeker, M.J.; Lonergan, D.J.; Cozzarelli, I.M.; Phillips, E.J.P.; Siegel, D.I. Oxidation of aromatic contaminants coupled to microbial iron reduction. *Nature* **1989**, *339*, 297–299.

41. Anderson, R.T.; Rooney-Varga, J.; Gaw, C.V.; Lovley, D.R. Anaerobic benzene oxidation in the Fe(III)-reduction zone of petroleum-contaminated aquifers. *Environ. Sci. Technol.* **1998**, *32*, 1222–1229.
42. Lovley, D.R.; Woodward, J.C.; Chapelle, F.H. Rapid anaerobic benzene oxidation with a variety of chelated Fe(III) forms. *Appl. Environ. Microbiol.* **1996**, *62*, 288–291.
43. Lovley, D.R.; Woodward, J.C.; Chapelle, F.H. Stimulated anoxic biodegradation of aromatic hydrocarbons using Fe(III) ligands. *Nature* **1994**, *370*, 128–131.
44. Lovley, D.R.; Coates, J.D.; Blunt-Harris, E.L.; Phillips, E.J.P.; Woodward, J.C. Humic substances as electron acceptors for microbial respiration. *Nature* **1996**, *382*, 445–448.
45. Lovley, D.R.; Chapelle, F.H. Deep subsurface microbial processes. *Rev. Geophys.* **1995**, *33*, 365–381.
46. Lovley, D.R. Potential for anaerobic bioremediation of BTEX in petroleum-contaminated aquifers. *J. Industr. Microbiol.* **1997**, *18*, 75–81.
47. Reddy, C.M.; Eglinton, T.I.; Hounshell, A.; White, H.K.; Xu, L.; Gaines, R.B.; Frysinger, G.S. The West Falmouth oil spill after thirty years: the persistence of petroleum hydrocarbons in marsh sediments. *Environ. Sci. Technol.* **2002**, *36*, 4754–4760.
48. Rogers, S.W.; Ong, S.K.; Kjartanson, B.H.; Golchin, J.; Stenback, G.A. Natural Attenuation of Polycyclic Aromatic Hydrocarbon-Contaminated Sites: Review. *Practice Periodical of Hazardous, Toxic, and Radioactive Waste Management* **2002**, *6*, 141–155.
49. Frysinger, G.S.; Gaines, R.B.; Xu, L.; Reddy, C.M. Resolving the unresolved complex mixture in petroleum-contaminated sediments. *Environ. Sci. Technol.* **2003**, *37*, 1653–1662.
50. Zhang, T.; Gannon, S.M.; Nevin, K.P.; Franks, A.E.; Lovley, D.R. Stimulating the anaerobic degradation of aromatic hydrocarbons in contaminated sediments by providing an electrode as the electron acceptor. *Environm. Microbiol. Rep.* **2010**, doi:10.1111/j.1462-2920.2009.02145.x.
51. Borole, A.P.; Mielenz, J.R.; Vishnivetskaya, T.A.; Hamilton, C.Y. Controlling accumulation of fermentation inhibitors in biorefinery recycle water using microbial fuel cells. *Biotechnol. Biofuels* **2009**, *2*, 7.
52. Kerzenmacher, S.; DucrÈe, J.; Zengerle, R.; von Stetten, F. Energy harvesting by implantable abiotically catalyzed glucose fuel cells. *J. Power Sourc.* **2008**, *182*, 1–17.
53. Kim, H.H.; Mano, N.; Zhang, Y.; Heller, A. A miniature membrane-less biofuel cell operating under physiological conditions at 0.5 V. *J. Electrochem. Soc.* **2003**, *150*, A209–A213.
54. Minteer, S.D.; Liaw, B.Y.; Cooney, M.J. Enzyme-based biofuel cells. *Curr. Opin. Biotechnol.* **2007**, *18*, 228–234.
55. Calabrese Barton, S.; Gallaway, J.; Atanassov, P. Enzymatic biofuel cells for implantable and microscale devices. *Chem. Rev.* **2004**, *104*, 4867–4886.
56. Mingui, S.; Justin, G.A.; Roche, P.A.; Jun, Z.; Wessel, B.L.; Yinghe, Z.; Sclabassi, R.J. Passing data and supplying power to neural implants. *IEEE Eng. Med. Biol. Mag.* **2006**, *25*, 39–46.
57. Justin, G.A.; Zhang, Y.; Sun, M.; Sclabassi, R. An investigation of the ability of white blood cells to generate electricity in biofuel cells. In Proceedings of the IEEE 31st Annual Northeast Bioengineering Conference, Hoboken, NJ, USA, April 2005; pp. 277–278.
58. Lovley, D.R. Bug juice: harvesting electricity with microorganisms. *Nature Rev. Microbiol.* **2006**, *4*, 497–508.



59. Rabaey, K.; Boon, N.; Siciliano, S.D.; Verhaege, M.; Verstraete, W. Biofuel cells select for microbial consortia that self-mediate electron transfer. *Appl. Environ. Microbiol.* **2004**, *70*, 5373–5382.
60. Phung, N.; Lee, J.; Kang, K.; Chang, I.; Gadd, G.; Kim, B. Analysis of microbial diversity in oligotrophic microbial fuel cells using 16S rDNA sequences. *FEMS Microb. Lett.* **2004**, *233*, 77–82.
61. Aelterman, P.; Rabaey, K.; Pham, H.T.; Boon, N.; Verstraete, W. Continuous electricity generation at high voltages and currents using stacked microbial fuel cells. *Environ. Sci. Technol.* **2006**, *40*, 3388–3394.
62. Kim, B.H.; Park, H.S.; Kim, H.J.; Kim, G.T.; Chang, I.S.; Lee, J.; Phung, N.T. Enrichment of microbial community generating electricity using a fuel-cell-type electrochemical cell. *Appl. Microbiol. and Biotechnol.* **2004**, *63*, 672–681.
63. Pham, T.H.; Boon, N.; Aelterman, P.; Clauwaert, P.; De Schampelaere, L.; Vanhaecke, L.; De Maeyer, K.; Hofte, M.; Verstraete, W.; Rabaey, K. Metabolites produced by *Pseudomonas sp.* enable a Gram-positive bacterium to achieve extracellular electron transfer. *Appl. Microbiol. Biotechnol.* **2008**, *77*, 1119–1129.
64. Park, H.S.; Kim, B.H.; Kim, H.S.; Kim, H.J.; Kim, G.T.; Kim, M.; Chang, I.S.; Park, Y.K.; Chang, H.I. A novel electrochemically active and Fe(III)-reducing bacterium phylogenetically related to *Clostridium butyricum* isolated from a microbial fuel cell. *Anaerobe* **2001**, *7*, 297–306.
65. Bond, D.R.; Lovley, D.R. Evidence for Involvement of an Electron Shuttle in Electricity Generation by *Geothrix fermentans*. *Appl. Environ. Microbiol.* **2005**, *71*, 2186–2189.
66. Zhang, T.; Cui, C.; Chen, S.; Yang, H.; Shen, P. The direct electrocatalysis of *Escherichia coli* through electroactivated excretion in microbial fuel cell. *Electrochem. Comm.* **2008**, *10*, 293–297.
67. Zuo, Y.; Cheng, S.; Call, D.; Logan, B.E. Tubular membrane cathodes for scalable power generation in microbial fuel cells. *Env. Sci. Tech.* **2007**, *41*, 3347 – 3353.
68. Zhao, F.; Rahunen, N.; Varcoe, J.R.; Chandra, A.; Avignone-Rossa, C.; Thumser, A.E.; Slade, R.C.T. Activated carbon cloth as anode for sulfate removal in a microbial fuel cell. *Environ. Sci Technol.* **2008**, *42*, 4971–4976.
69. Borole, A.; O'Neill, H.; Tsouris, C.; Cesar, S. A microbial fuel cell operating at low pH using the acidophile acidiphilium cryptum. *Biotechnol. Lett.* **2008**, *30*, 1367–1372.
70. Zhang, K.; Martiny, A.C.; Reppas, N.B.; Barry, K.W.; Malek, J.; Chisholm, S.W.; Church, G.M. Sequencing genomes from single cells by polymerase cloning. *Nat. Biotechnol.* **2006**, *24*, 680–686.
71. Holmes, D.E.; Nicoll, J.S.; Bond, D.R.; Lovley, D.R. Potential role of a novel psychrotolerant *Geobacteraceae*, *Geopsychrobacter electrodiphilus* gen. nov., sp. nov., in electricity production by the marine sediment fuel cell. *Appl. Environ. Microbiol.* **2004**, *70*, 6023–6030.
72. Chaudhuri, S.K.; Lovley, D.R. Electricity generation by direct oxidation of glucose in mediatorless microbial fuel cells. *Nat. Biotechnol.* **2003**, *21*, 1229–1232.
73. Walker, A.L.; Walker, J.C.W. Biological fuel cell and an application as a reserve power source. *J. Power Sourc.* **2006**, *160*, 123–129.

74. Prasad, D.; Arun, S.; Murugesan, M.; Padmanaban, S.; Satyanarayanan, R.S.; Berchmans, S.; Yegnaraman, V. Direct electron transfer with yeast cells and construction of a mediatorless microbial fuel cell. *Biosen. Bioelectron.* **2007**, *22*, 2604–2610.
75. Freguia, S.; Rabaey, K.; Yuan, Z.; Keller, J. Syntrophic processes drive the conversion of glucose in microbial fuel cell anodes. *Environ. Sci. Technol.* **2008**, *42*, 7937–7943.
76. Torres, C.; Kato-Marcus, A.; Rittmann, B. Kinetics of consumption of fermentation products by anode-respiring bacteria. *App. Microbiol. Biotechnol.* **2007**, *77*, 689–697.
77. Lee, H.S.; Parameswaran, P.; Kato-Marcus, A.; Torres, C.I.; Rittmann, B.E. Evaluation of energy-conversion efficiencies in microbial fuel cells (MFCs) utilizing fermentable and non-fermentable substrates. *Water Res.* **2008**, *42*, 1501–1510.
78. Lanthier, M.; Gregory, K.B.; Lovley, D.R. Growth with high planktonic biomass in *Shewanella oneidensis* fuel cells. *FEMS Microbiol. Lett.* **2008**, *278*, 29–35.
79. Rabaey, K.; Lissens, G.; Siciliano, S.D.; Verstraete, W. A microbial fuel cell capable of converting glucose to electricity at high rate and efficiency. *Biotech. Lett.* **2003**, *25*, 1531–1535.
80. Rittmann, B.E.; Torres, C.I.; Marcus, A.K. Understanding the distinguishing features of a microbial fuel cell as a biomass-based renewable energy technology. In *Emerging Environmental Technologies*; Springer: Berlin, German, 2008; pp. 1–28.
81. Cheng, S.; Liu, H.; Logan, B.E. Power densities using different cathode catalysts (Pt and CoTMPP) and polymer binders (Nafion and PTFE) in single chamber microbial fuel cells. *Env. Sci. Tech.* **2006**, *40*, 364–369.
82. Zhao, F.; Harnisch, F.; Schroder, U.; Scholz, F.; Bogdanoff, P.; Herrmann, I. Application of pyrolysed iron(II) phthalocyanine and CoTMPP based oxygen reduction catalysts as cathode materials in microbial fuel cells. *Electrochem. Comm.* **2005**, *7*, 1405–1410.
83. Nevin, K.P.; Kim, B.C.; Glaven, R.H.; Johnson, J.P.; Woodard, T.L.; Methe, B.A.; DiDonato, R.J.; Covalla, S.F.; Franks, A.E.; Liu, A.; Lovley, D.R. Anode biofilm transcriptomics reveals outer surface components essential for high density current production in *Geobacter sulfurreducens* fuel cells. *PLoS ONE* **2009**, *4*, e5628.
84. Rabaey, K.; Read, S.T.; Clauwaert, P.; Freguia, S.; Bond, P.L.; Blackall, L.L.; Keller, J. Cathodic oxygen reduction catalyzed by bacteria in microbial fuel cells. *ISME J.* **2008**, *2*, 519–527.
85. Rabaey, K.; Boon, N.; Hofte, M.; Verstraete, W. Microbial phenazine production enhances electron transfer in biofuel cells. *Environ. Sci. Technol.* **2005**, *39*, 3401–3408.
86. Sund, C.J.; McMasters, S.; Crittenden, S.R.; Harrell, L.E.; Sumner, J.J. Effect of electron mediators on current generation and fermentation in a microbial fuel cell. *Appl. Microbiol. Biotechnol.* **2007**, *76*, 561–568.
87. Logan, B.E.; Hamelers, B.; Rozendal, R.; Schroder, U.; Keller, J.; Freguia, S.; Aelterman, P.; Verstraete, W.; Rabaey, K. Microbial fuel cells: Methodology and technology. *Environ. Sci. Technol.* **2006**, *40*, 5181–5192.
88. Richter, H.; Nevin, K.P.; Jia, H.F.; Lowy, D.A.; Lovley, D.R.; Tender, L.M. Cyclic voltammetry of biofilms of wild type and mutant *Geobacter sulfurreducens* on fuel cell anodes indicates possible roles of OmcB, OmcZ, type IV pili, and protons in extracellular electron transfer. *Energ. Environ. Sci.* **2009**, *2*, 506–516.

89. Fricke, K.; Harnisch, F.; Schroder, U. On the use of cyclic voltammetry for the study of anodic electron transfer in microbial fuel cells. *Energ. Environ. Sci.* **2008**, *1*, 144–147.
90. Busalmen, J.P.; Esteve-Nunez, A.; Feliu, J.M. Whole cell electrochemistry of electricity-producing microorganisms evidence an adaptation for optimal exocellular electron transport. *Environ. Sci. Technol.* **2008**, *42*, 2445–2450.
91. Zhang, T.; Cui, C.; Chen, S.; Ai, X.; Yang, H.; Shen, P.; Peng, Z. A novel mediatorless microbial fuel cell based on direct biocatalysis of *Escherichia coli*. *Chem. Commun. Camb.* **2006**, 2257–2259.
92. Srikanth, S.; Marsili, E.; Flickinger, M.C.; Bond, D.R. Electrochemical characterization of *Geobacter sulfurreducens* cells immobilized on graphite paper electrodes. *Biotechnol. Bioeng.* **2008**, *99*, 1065–1073.
93. Dumas, C.; Mollica, A.; Feron, D.; Basseguy, R.; Etcheverry, L.; Bergel, A. Checking graphite and stainless anodes with an experimental model of marine microbial fuel cell. *Bioresour. Technol.* **2008**, *99*, 8887–8894.
94. Cheng, K.Y.; Cord-Ruwisch, R.; Ho, G. A new approach for in situ cyclic voltammetry of a microbial fuel cell biofilm without using a potentiostat. *Bioelectrochemistry* **2009**, *74*, 227–231.
95. Marsili, E.; Baron, D.B.; Shikhare, I.D.; Coursolle, D.; Gralnick, J.A.; Bond, D.R. *Shewanella* secretes flavins that mediate extracellular electron transfer. *Proc. Natl. Acad. Sci. USA* **2008**, *105*, 3968–3973.
96. Nevin, K.P.; Lovley, D.R. Novel mechanisms for accessing insoluble Fe(III) oxide during dissimilatory Fe(III) reduction by *Geothrix fermentans* *Appl. Environ. Microbiol.* **2002**, *68*, 2294–2299.
97. Hernandez, M.E.; Kappler, A.; Newman, D.K. Phenazines and other redox-active antibiotics promote microbial mineral reduction. *Appl. Environ. Microbiol.* **2004**, *70*, 921–928.
98. Reguera, G.; McCarthy, K.D.; Mehta, T.; Nicoll, J.S.; Tuominen, M.T.; Lovley, D.R. Extracellular electron transfer via microbial nanowires. *Nature* **2005**, *435*, 1098–1101.
99. Myers, C.R.; Myers, J.M. Localization of cytochromes to the outer membrane of anaerobically grown *Shewanella putrefaciens* MR-1. *J. Bacteriol.* **1992**, *174*, 3429–3438.
100. Lovley, D.R. Microbial fuel cells: Coupling microbial physiologies and engineering approaches. *Curr. Opin. Biotechnol.* **2006**, *17*, 327–332.
101. Lovley, D.R. The microbe electric: Conversion of organic matter to electricity. *Curr. Opin. Biotechnol.* **2008**, *19*, 564–571.
102. Lovley, D.R. Extracellular electron transfer: Wires, capacitors, iron lungs, and more. *Geobiology* **2008**, *6*, 225–231.
103. Holmes, D.E.; Bond, D.R.; O'Neil, R.A.; Reimers, C.E.; Tender, L.R.; Lovley, D.R. Microbial communities associated with electrodes harvesting electricity from a variety of aquatic sediments. *Microbial Ecol.* **2004**, *48*, 178–190.
104. Ishii, S.; Watanabe, K.; Yabuki, S.; Logan, B.E.; Sekiguchi, Y. Comparison of electrode reduction activities of *Geobacter sulfurreducens* and an enriched consortium in an air-cathode microbial fuel cell. *Appl. Environ. Microbiol.* **2008**, *74*, 7348–7355.

105. Methé B.A.; Nelson, K.E.; Eisen, J.A.; Paulsen, I.T.; Nelson, W.; Heidelberg, J.F.; Wu, D.; Wu, M.; Ward, N.; Beanan, M.J.; Dodson, R.J.; Madupu, R.; Brinkac, L.M.; Daugherty, S.C.; DeBoy, R.T.; Durkin, A.S.; Gwinn, M.; Kolonay, J.F.; Sullivan, S.A.; Haft, D.H.; Selengut, J.; Davidsen, T.M.; Zafar, N.; White, O.; Tran, B.; Romero, C.; Forberger, H.A.; Weidman, J.; Khouri, H.; Feldblyum, T.V.; Utterback, T.R.; Van Aken, S.E.; Lovley, D.R.; Fraser, C.M. The genome of *Geobacter sulfurreducens*: Insights into metal reduction in subsurface environments. *Science* **2003**, *302*, 1967–1969.
106. Coppi, M.V.; Leang, C.; Sandler, S.J.; Lovley, D.R. Development of a genetic system for *Geobacter sulfurreducens*. *Appl. Environ. Microbiol.* **2001**, *67*, 3180–3187.
107. Postier, B.L.; DiDonato, R.J., Jr.; Nevin, K.P.; Liu, A.; Frank, B.; Lovley, D.R.; Methe, B.A. Benefits of electrochemically synthesized oligonucleotide microarrays for analysis of gene expression in understudied microorganisms. *J. Microbiol. Methods* **2008** (in press).
108. Park, D.H.; Zeikus, J.G. Improved fuel cell and electrode designs for producing electricity from microbial degradation. *Biotechnol. Bioeng.* **2003**, *81*, 348–355.
109. Logan, B.E.; Regan, J.M. Electricity-producing bacterial communities in microbial fuel cells. *Trend Microbiol.* **2006**, *14*, 512–518.
110. Debabov, V.G. Electricity from microorganisms. *Mikrobiologiya* **2008**, *77*, 149–157.
111. Chang, I.S.; Moon, H.; Bretschger, O.; Jang, J.K.; Park, H.I.; Nealson, K.H.; Kim, B.H. Electrochemically active bacteria (EAB) and mediator-less microbial fuel cells. *J. Microbiol. Biotechnol.* **2006**, *16*, 163–177.
112. Kim, B.C.; Leang, C.; Ding, Y.H.; Glaven, R.H.; Coppi, M.V.; Lovley, D.R. OmcF, a putative c-Type monoheme outer membrane cytochrome required for the expression of other outer membrane cytochromes in *Geobacter sulfurreducens*. *J. Bacteriol.* **2005**, *187*, 4505–4513.
113. Holmes, D.E.; Chaudhuri, S.K.; Nevin, K.P.; Mehta, T.; Methe, B.A.; Liu, A.; Ward, J.E.; Woodard, T.L.; Webster, J.; Lovley, D.R. Microarray and genetic analysis of electron transfer to electrodes in *Geobacter sulfurreducens*. *Env. Microbiol.* **2006**, *8*, 1805–1815.
114. Holmes, D.E.; Mester, T.; O'Neil, R.A.; Perpetua, L.A.; Larrahondo, M.J.; Glaven, R.; Sharma, M.L.; Ward, J.E.; Nevin, K.P.; Lovley, D.R. Genes for two multicopper proteins required for Fe(III) oxide reduction in *Geobacter sulfurreducens* have different expression patterns both in the subsurface and on energy-harvesting electrodes. *Microbiol.* **2008**, *154*, 1422–1435.
115. Busalmen, J.P.; Esteve-Nunez, A.; Berna, A.; Feliu, J.M. C-type cytochromes wire electricity-producing bacteria to electrodes. *Angew. Chem. Int. Ed. Engl.* **2008**, *47*, 4874–4877.
116. Franks, A.E.; Nevin, K.P.; Jia, H.; Izallalen, M.; Woodard, T.L.; Lovley, D.R. Novel strategy for three-dimensional real-time imaging of microbial fuel cell communities: monitoring the inhibitory effects of proton accumulation within the anode biofilm. *Energ. Environ. Sci.* **2009**, *2*, 113–119.
117. Reguera, G.; Nevin, K.P.; Nicoll, J.S.; Covalla, S.F.; Woodard, T.L.; Lovley, D.R. Biofilm and nanowire production leads to increased current in *Geobacter sulfurreducens* Fuel Cells. *Appl. Environ. Microbiol.* **2006**, *72*, 7345–7348.
118. Kato-Marcus, A.; Torres, C.I.; Rittmann, B.E. Conduction-based modeling of the biofilm anode of a microbial fuel cell. *Biotechnol. Bioeng.* **2007**, *98*, 1171–1182.

119. Picioreanu, C.; Head, I.M.; Katuri, K.P.; van Loosdrecht, M.C.; Scott, K. A computational model for biofilm-based microbial fuel cells. *Water Res.* **2007**, *41*, 2921–2940.
120. Muñoz-Berbel, X.; Muñoz, F.J.; Vigués, N.; Mas, J. On-chip impedance measurements to monitor biofilm formation in the drinking water distribution network. *Sensor Actuat. B: Chem.* **2006**, *118*, 129–134.
121. Dheilly, A.; Linossier, I.; Darchen, A.; Hadjiev, D.; Corbel, C.; Alonso, V. Monitoring of microbial adhesion and biofilm growth using electrochemical impedancemetry. *Appl. Microbiol. Biotechnol.* **2008**, *79*, 157–164.
122. Herbert-Guillou, D.; Tribollet, B.; Festy, D.; KiÈnÈ, L. *In situ* detection and characterization of biofilm in waters by electrochemical methods. *Electrochimica Acta* **1999**, *45*, 1067–1075.
123. Izallalen, M.; Mahadevan, R.; Burgard, A.; Postier, B.; Didonato Jr, R.; Sun, J.; Schilling, C.H.; Lovley, D.R. *Geobacter sulfurreducens* strain engineered for increased rates of respiration. *Met. Eng.* **2008**, *10*, 267–275.
124. Mahadevan, R.; Bond, D.R.; Butler, J.E.; Esteve-Nunez, A.; Coppi, M.V.; Palsson, B.O.; Schilling, C.H.; Lovley, D.R. Characterization of metabolism in the Fe(III)-reducing organism *Geobacter sulfurreducens* by constraint-based modeling. *Appl. Environ. Microbiol.* **2006**, *72*, 1558–1568.
125. Yi, H.; Nevin, K.P.; Kim, B.C.; Franks, A.E.; Klimes, A.; Tender, L.M.; Lovley, D.R. Selection of a variant of *Geobacter sulfurreducens* with enhanced capacity for current production in microbial fuel cells. *Biosens. Bioelectron.* **2009**, *24*, 3498–3503.
126. Torres, C.I.; Lee, H.S.; Rittmann, B.E. Carbonate species as OH<sup>-</sup> carriers for decreasing the pH gradient between cathode and anode in biological fuel cells. *Environ. Sci. Technol.* **2008**, *42*, 8773–8777.
127. Torres, C.I.; Kato Marcus, A.; Rittmann, B.E. Proton transport inside the biofilm limits electrical current generation by anode-respiring bacteria. *Biotechnol. Bioeng.* **2008**, *100*, 872–881.
128. Lee, H.S.; Torres, C.I.; Rittmann, B.E. Effects of Substrate Diffusion and Anode Potential on Kinetic Parameters for Anode-Respiring Bacteria. *Environ. Scienc. Technol.* **2009**, *43*, 7571–7577.
129. Franks, A.E.; Nevin, K.P.; Glaven, R.H.; Lovley, D.R. Microtoming Coupled to Microarray Analysis to Evaluate the Spatial Metabolic Status of *Geobacter sulfurreducens* Biofilms. *ISME J.* **2010**, DOI: 10.1038/ismej.2009.137
130. Logan, B.E.; Regan, J.M. Microbial fuel cells-challenges and applications. *Environ. Sci. Technol.* **2006**, *40*, 5172–5180.
131. Clauwaert, P.; van der Ha, D.; Boon, N.; Verbeken, K.; Verhaege, M.; Rabaey, K.; Verstraete, W. Open air biocathode enables effective electricity generation with microbial fuel cells. *Environ. Scienc. Technol.* **2007**, *41*, 7564–7569.
132. Rismani-Yazdi, H.; Carver, S.M.; Christy, A.D.; Tuovinen, O.H. Cathodic limitations in microbial fuel cells: An overview. *J. Power Sourc.* **2008**, *180*, 683–694.
133. Gil, G.C.; Chang, I.S.; Kim, B.H.; Kim, M.; Jang, J.K.; Park, H.S.; Kim, H.J. Operational parameters affecting the performannce of a mediator-less microbial fuel cell. *Biosens. Bioelectron.* **2003**, *18*, 327–334.

134. Rabaey, K.; Clauwaert, P.; Aelterman, P.; Verstraete, W. Tubular microbial fuel cells for efficient electricity generation. *Environ. Sci. Technol.* **2005**, *39*, 8077–8082.
135. Gregory, K.B.; Bond, D.R.; Lovley, D.R. Graphite electrodes as electron donors for anaerobic respiration. *Env. Microbiol.* **2004**, *6*, 596–604.
136. Gregory, K.B.; Lovley, D.R. Remediation and recovery of uranium from contaminated subsurface environments with electrodes. *Env. Sci. Tech.* **2005**, *39*, 8943–8947.
137. Rhoads, A.; Beyenal, H.; Lewandowski, Z. Microbial fuel cell using anaerobic respiration as an anodic reaction and biomineralized manganese as a cathodic reactant. *Environ. Sci. Technol.* **2005**, *39*, 4666–4671.
138. Tran, H.T.; Kim, D.H.; Oh, S.J.; Rasool, K.; Park, D.H.; Zhang, R.H.; Ahn, D.H. Nitrifying biocathode enables effective electricity generation and sustainable wastewater treatment with microbial fuel cell. *Water Sci. Technol.* **2009**, *59*, 1803–1808.
139. Bergel, A.; Fèron, D.; Mollica, A. Catalysis of oxygen reduction in PEM fuel cell by seawater biofilm. *Electrochem. Comm.* **2005**, *7*, 900–904.
140. You, S.J.; Ren, N.Q.; Zhao, Q.L.; Wang, J.Y.; Yang, F.L. Power generation and electrochemical analysis of biocathode microbial fuel cell using graphite fibre brush as cathode material. *Fuel Cells* **2009**, *9*, 588–596.
141. Chen, G.W.; Choi, S.J.; Lee, T.H.; Lee, G.Y.; Cha, J.H.; Kim, C.W. Application of biocathode in microbial fuel cells: Cell performance and microbial community. *App. Microbiol. Biotechnol.* **2008**, *79*, 379–388.
142. Strycharz, S.; Gannon, S.; Boles, A.; Franks, A.; Nevin, K.; Lovley, D.R. Reductive dechlorination of 2-chlorophenol by anaeromyxobacter dehalogenans with an electrode serving as the electron donor. *Environ. Microbio. Environ. Microbiol. Report* **2010**, *2*, 289–294.
143. Strycharz, S.M.; Woodard, T.L.; Johnson, J.P.; Nevin, K.P.; Sanford, R.A.; Löffler, F.E.; Lovley, D.R. Graphite electrode as a sole electron donor for reductive dechlorination of tetrachlorethene by *Geobacter Lovleyi*. *Appl. Environ. Microbiol.* **2008**, *74*, 5943–5947.