Review TRENDS in Biotechnology Vol.23 No.6 June 2005

Microbial fuel cells: novel biotechnology for energy generation

Korneel Rabaey and Willy Verstraete

Laboratory of Microbial Ecology and Technology (LabMET), Ghent University, Coupure Links 653, B-9000 Ghent, Belgium

Microbial fuel cells (MFCs) provide new opportunities for the sustainable production of energy from biodegradable, reduced compounds. MFCs function on different carbohydrates but also on complex substrates present in wastewaters. As yet there is limited information available about the energy metabolism and nature of the bacteria using the anode as electron acceptor; few electron transfer mechanisms have been established unequivocally. To optimize and develop energy production by MFCs fully this knowledge is essential. Depending on the operational parameters of the MFC, different metabolic pathways are used by the bacteria. This determines the selection and performance of specific organisms. Here we discuss how bacteria use an anode as an electron acceptor and to what extent they generate electrical output. The MFC technology is evaluated relative to current alternatives for energy generation.

Introduction

Microbial fuel cells are not new – the concept of using microorganisms as catalysts in fuel cells was explored from the 1970s [1,2] and microbial fuel cells treating domestic wastewater were presented in 1991 [3]. However, it is only recently that microbial fuel cells with an enhanced power output [4–8] have been developed providing possible opportunities for practical applications.

A MFC converts energy, available in a bio-convertible substrate, directly into electricity. This can be achieved when bacteria switch from the natural electron acceptor, such as oxygen or nitrate, to an insoluble acceptor, such as the MFC anode (Figure 1). This transfer can occur either via membrane-associated components, or soluble electron shuttles. The electrons then flow through a resistor to a cathode, at which the electron acceptor is reduced. In contrast to anaerobic digestion, a MFC creates electrical current and an off-gas containing mainly carbon dioxide.

MFCs have operational and functional advantages over the technologies currently used for generating energy from organic matter. First, the direct conversion of substrate energy to electricity enables high conversion efficiency. Second, MFCs operate efficiently at ambient, and even at low, temperatures distinguishing them from all current bio-energy processes. Third, an MFC does not require gas treatment because the off-gases of MFCs are enriched in carbon dioxide and normally have no useful energy content. Fourth, MFCs do not need energy input for aeration provided the cathode is passively aerated [5]. Fifth, MFCs have potential for widespread application in locations lacking electrical infrastructures and also to expand the diversity of fuels we use to satisfy our energy requirements.

Metabolism in microbial fuel cells

To assess bacterial electricity generation, metabolic pathways governing microbial electron and proton flows must be determined. In addition to the influence of the substrate [7,9,10] the potential of the anode will also determine the bacterial metabolism. Increasing MFC current will decrease the potential of the anode, forcing the bacteria to deliver the electrons through more-reduced complexes. The potential of the anode will therefore determine the redox potential of the final bacterial electron shuttle, and therefore, the metabolism. Several different metabolism routes can be distinguished based on the anode potential: high redox oxidative metabolism; medium to low redox oxidative metabolism; and fermentation. Hence, the organisms reported to date in MFCs vary from aerobes and facultative anaerobes towards strict anaerobes.

At high anodic potentials, bacteria can use the respiratory chain in an oxidative metabolism. Electrons and, concomitantly, protons can be transported through the NADH dehydrogenase, ubiquinone, coenzyme Q or cytochrome [11–13]. The use of this pathway was investigated by Kim *et al.* (2004) [10]. They observed that the generation of electrical current from an MFC was inhibited by various inhibitors of the respiratory chain. The electron transport system in their MFC used NADH dehydrogenase, Fe/S (iron/sulphur) proteins and quinones as electron carriers, but does not use site 2 of the electron transport chain or the terminal oxidase. Processes using oxidative phosphorylation have regularly been observed in MFCs, yielding high energy efficiencies of up to 65% [6]. Examples are consortia containing *Pseudomonas aeruginosa*, *Enterococcus faecium* [7] and *Rhodoferax ferrireducens* [14]. An overview of different bacterial species and their (putative) electron transport pathway is given in Table 1.

If the anode potential decreases in the presence of alternative electron acceptors such as sulphate, the electrons are likely to be deposited onto these components. Methane production has repeatedly been observed when

Corresponding author: Verstraete, W. (willy.verstraete@UGent.be).
Available online 25 April 2005

 0167-7799/$ - see front matter doi:10.1016/j.tibtech.2005.04.008

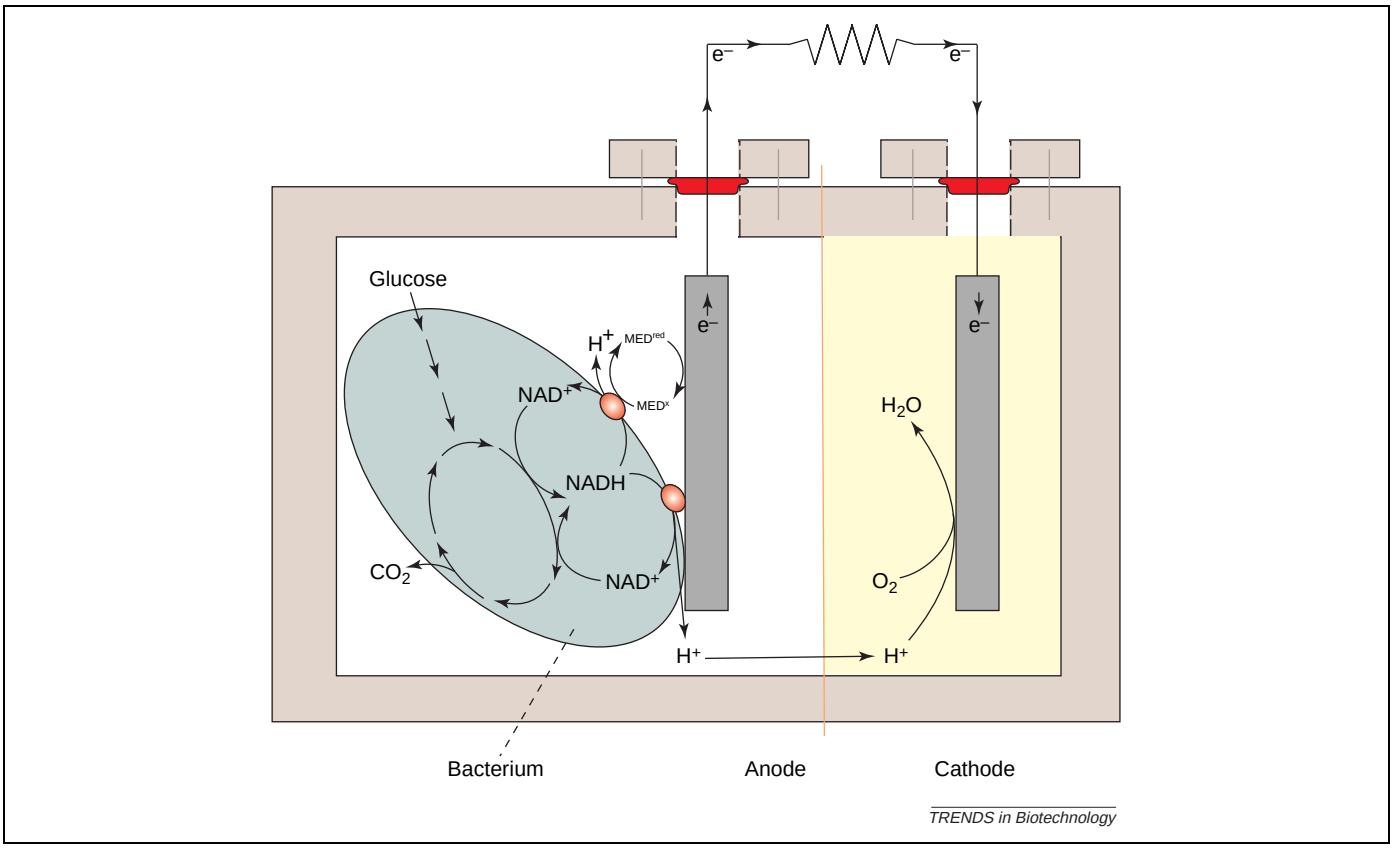

Figure 1. The working principle of a microbial fuel cell. Substrate is metabolized by bacteria, which transfer the gained electrons to the anode. This can occur either directly through the membrane or via mobile redox shuttles. MED, redox mediator; Red oval, terminal electron shuttle in or on the bacterium.

the inoculum was anaerobic sludge [10,15], indicating that the bacteria do not use the anode. If no sulphate, nitrate or other electron acceptors are present, fermentation will be the main process when the anode potential remains low. For example, during fermentation of glucose, possible reactions can be: $C_6H_{12}O6 + 2H_2O \rightarrow 4H_2 + 2CO_2 + 2C_2H4O_2$ or $C_6H_{12}O_6 \rightarrow 2H_2 + 2CO_2 + C_4H_8O_2$ [16]. This shows that a maximum of one-third of a hexose substrate electrons can theoretically be used to generate current, whereas two-thirds remain in the produced fermentation products such as acetate and butyrate [17]. The one-third of the total electrons are possibly available for electricity generation because the hydrogenases, which generally use the electrons to produce hydrogen gas, are often situated at places on the membrane surface [18] that are accessible from outside by mobile electron shuttles [19] or that connect directly to the electrode. As repeatedly observed, this metabolic type can imply a high acetate or butyrate production. Several organisms that are known to produce fermentation products and belong to the genus *Clostridium, Alcaligenes, Enterococcus*, have been isolated from MFCs [7,20]. This pathway is further substantiated by the significant hydrogen production observed when MFC enriched cultures are incubated anaerobically in a separate fermentation test [7].

Fermentation products such as acetate can be oxidized at low anode potential by anaerobic bacteria such as *Geobacter* species, which is capable of withdrawing electrons from acetate in MFC conditions [62].

Table 1. Bacterial species identified in microbial fuel cells and their possible metabolism and pathway of electron transfer

Metabolic type	Transfer type	Examples of organisms	Terminal bacterial electron shuttle	Added redox shuttle	Refs
Oxidative metabolism	Membrane-driven	*Rhodoferax ferrireducens*	Unknown		[14]
		Geobacter sulfurreducens	89 kDa c-type cytochrome[a]		[61]
		Aeromonas hydrophila	c-type cytochrome[a]		[23]
	Mediator-driven	*Escherichia coli*	Hydrogenase	Neutral red	[18]
		Shewanella putrefaciens	Quinones[a]		[28,55–58]
		Pseudomonas aeruginosa	Pyocyanin, phenazine carboxamide		[31]
		Erwinia dissolvens	Unknown	Fe(III)CyDTA (an iron chelator)	[59]
		Desulfovibrio desulfuricans	S^{2-}		[60]
Fermentative metabolism	Membrane driven	*Clostridium butyricum*	Cytochromes[a]		[20]
	Mediator driven	*Enterococcus faecium*	Unknown	Pyocyanin	[31]

[a]Putative.

This metabolic variation, together with the observed redox potential data, provides insight into microbial 'electrodynamics'. A MFC, operated at low external resistance, will initially generate low current during biomass build-up, and hence have a high anode potential (low MFC cell potential). The result is a selection towards facultative aerobes and anaerobes (Box 1 and 2). Upon growth of the culture, the metabolic turnover rate, and hence the current, will increase. The now moderate anode potential will favour lower redox facultative anaerobes. However, strict anaerobes will still be hampered by the redox potential in the anode compartment and possibly also by the possible intrusion of oxygen through the membrane [4]. When a high resistance is used, the potential of the anode will be low, even at small current levels. In that case, one will select for low redox facultative anaerobes and strict anaerobes, limiting the possibilities for bacterial selection.

Anodic electron transfer mechanisms in MFC

The electrons to be diverted towards the electrode need a physical transport system for extracellular electron transfer. This can either occur through the use of soluble electron shuttles [2,21] or through membrane-bound electron shuttling compounds [62,22].

The oxidative, membrane-associated electron transfer is thought to occur through compounds that belong to the respiratory chain. Bacteria known to use this pathway are for example *Geobacter metallireducens* [62], *Aeromonas hydrophila* [23] and *Rhodoferax ferrireducens* [14]. The main requirement for a component to act as an electron gateway seems to be the steric accessibility [19] (physical contact between electron donor and acceptor). The potential of the gateway in relation to the anode will determine whether the gateway is actually used (an electron will not be transferred to a more reduced electrode).

Many fermentative organisms identified in MFCs possess a hydrogenase, for example *Clostridium butyricum* [20] and *Enterococcus faecium* [7]. Hydrogenases could be directly involved in electron transfer towards

Box 1. Bacterial potential for electricity generation

Bacteria gain energy by transferring electrons from a reduced substrate at a low potential, such as glucose, to an electron acceptor with a high potential, such as oxygen. An overview of common reactions is given in Table I (based on information in [53]). The energy gained can be calculated as: $\Delta G = -n \times F \times \Delta E$ [with n the number of electrons exchanged, F Faraday's constant (96485 Coulomb/mol) and ΔE the potential difference between electron donor and acceptor]. If bacteria derive reducing equivalents from glucose in the form of NADH, and subsequently shuttle electrons from NADH to oxygen (not taking into account potential decreases between NADH and the final bacterial electron shuttle), the potential difference is ~1.2 V [$\Delta E = (+0.840V) - (-0.320V)$], and the energy to be gained (2 electrons per molecule of NADH) $\Delta G = -2 \times 10^2$ kJ/mol. If the electron acceptor is sulphate, the potential difference decreases to ~100 mV, yielding a ΔG of $\sim 2 \times 10^1$ kJ/mol. The amount of energy available for the bacteria to grow is very low in that case. In a MFC no oxygen is present. If an anode is available with a higher potential than, for example, sulphate present in the feed stream, the energetic gain will be much higher for bacteria that can deliver to the anode. Hence, the anode will become the preferred electron acceptor. For more information regarding bacterial energy conservation see [16].

Table I

Redox reaction	E'_0 (mV)
$2H^+ + 2e^- \rightarrow H_2$	−420
$Ferredoxin(Fe^{3+}) + e^- \rightarrow Ferredoxin(Fe^{2+})$	−420
$NAD^+ + H^+ + 2e^- \rightarrow NADH$	−320
$S + 2H^+ + 2e^- \rightarrow H_2S$	−274
$SO_4^{2-} + 10H^+ + 8e^- \rightarrow H_2S + 4H_2O$	−220
$Pyruvate^{2-} + 2H^+ + 2e^- \rightarrow Lactate^{2-}$	−185
$FAD + 2H^+ + 2e^- \rightarrow FADH_2$	−180
$Fumarate^{2-} + 2H^+ + 2e^- \rightarrow Succinate^{2-}$	+31
Cytochrome $b(Fe^{3+}) + e^- \rightarrow$ Cytochrome $b(Fe^{2+})$	+75
$Ubiquinone + 2H^+ + 2e^- \rightarrow UbiquinoneH_2$	+100
Cytochrome $c(Fe^{3+}) + e^- \rightarrow$ Cytochrome $c(Fe^{2+})$	+254
$NO_3^- + 2H^+ + 2e^- \rightarrow NO_2^- + H_2O$	+421
$NO_2^- + 8H^+ + 6e^- \rightarrow NH_4^+ + 2H_2O$	+440
$Fe^{3+} + e^- \rightarrow Fe^{2+}$	+771
$O_2 + 4H^+ + 4e^- \rightarrow 2H_2O$	+840

Box 2. Energy available for electricity generation

The amount of energy (Joules) gained out of an electrochemical process can be calculated based on power output and process duration: $E = P \times t$, with P the power (Watts) and t time (s). The power depends both on the voltage V and the current I: $P = V \times I$. The latter factors are linked by the fuel cell resistance, by Ohm's law $V = I \times R$ in which R represents the resistance (Ohm). The voltage over the resistance (V) can be described as [33]: $V = E^0 - \eta_a - \eta_c - I \times R$ with E^0 maximum cell voltage, η_a and η_c overpotential losses at the electrodes and $I \times R$ the loss owing to electrolyte resistances. Hence, what is measured over the fuel cell will be lower than the attainable voltage. In practice, the maximal open circuit potentials (potential observed when no current is running through the MFC electrical circuit) observed are of the order of 750–800 mV [53]. Upon closure of the electrical loop, this voltage decreases significantly, mainly because of the so-called overpotentials, which are potential losses owing to electron transfer resistances and internal resistances (see Figure I). Three kinds of overpotentials can be defined: activation overpotentials, ohmic losses and concentration polarization [41]. For MFCs, the activation overpotential appears to be the major limiting factor (see also 'Parameters defining MFC performance' in [41]). This overpotential is largely dependent on the current density flowing through the anode, the electrochemical properties of the electrode, the presence of mediating compounds and the operational temperature [41,54].

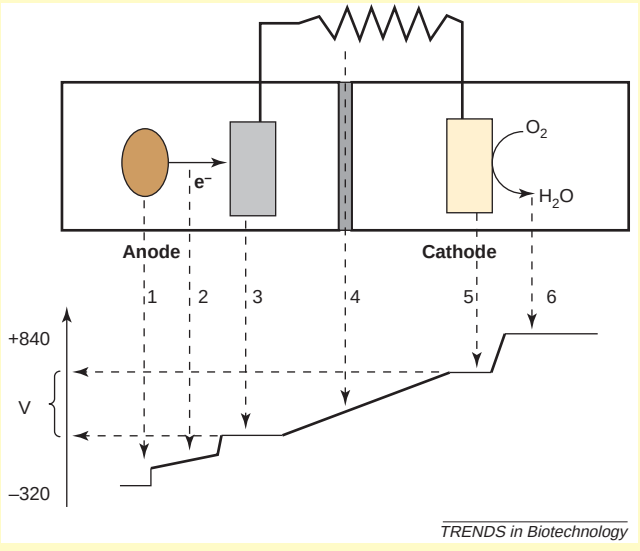

Figure I. Potential losses during electron transfer in a MFC. 1. Loss owing to bacterial electron transfer. 2. Losses owing to electrolyte resistance. 3 Losses at the anode. 4. Losses at the MFC resistance (useful potential difference) and membrane resistance losses. 5. Losses at the cathode. 6: Losses owing to electron acceptor reduction.

Table 2. Performance of MFCs based on both axenic (single bacterial species) and mixed culture systems[a]

	Substrate	Electrode type	Redox mediated	I (current, mA)	P (mW/m^2)	P (W/m^3)	Refs
Axenic cultures							
Proteus vulgaris	Glucose	Glassy carbon	X	0.8	4.5	18	[21]
Erwinia dissolvens	Glucose	Woven graphite	X	0.7	0.27[b]	n.a.[e]	[59]
Proteus vulgaris	Glucose	Glassy carbon	X	0.7	85	9.0	[25]
Shewanella putrefaciens	Lactate	Woven graphite		0.04	0.00032	0.08	[58]
Geobacter sulfurreducens	Acetate	Plain graphite		0.4	13	0.35	[61]
Rhodoferax ferrireducens	Glucose	Plain graphite		0.2	8	0.25	[14]
		Woven graphite		0.57	17	1.7	[14]
		Graphite foam		0.4514	33	0.96	[14]
Pseudomonas aeruginosa	Glucose	Plain graphite		0.1	88	8.8	[31]
Escherichia coli	Lactate	Woven graphite[c]	X	3.3	1.2	7.6	[37]
		Plain graphite[c]	X	2.6	91	3.6	[37]
Mixed cultures							
Mixed, saltwater	Acetate	Plain graphite		0.23	10	n.a.[e]	[63]
	S2-/acetate	Plain graphite		60[d]	32	n.a.[e]	[64]
Mixed consortium, batch	Glucose	Plain graphite		30	3600	216	[6]
Activated sludge	Wastewater	Woven graphite		0.2	8	1.6	[10]
	Lactate	Woven graphite[b]	X	11	5.3	34	[37]
		Plain graphite[b]	X	2.6	788	32	[37]
	Wastewater	Woven graphite		4.85	26	1.6	[4]
	Glucose	Woven graphite		0.9	494	13	[4]
Mixed consortium, continuous	Sucrose	Granular graphite		6.2	23	47	[43]
	Glucose	Granular graphite		5.4	18	37	[43]
	Acetate	Carbon paper		1.27	506	13	[52]
	Butyrate	Carbon paper		0.46	305	7.6	[52]

[a]No surface data available, value in absolute mW.
[b]Power output was calculated as average power output where possible because peak power outputs are less representative.
[c]Mediator immobilized in/on electrode matrix.
[d]Data as mA/m^2 anode surface.
[e]n.a, unsufficient data available, or not applicable.

electrodes. Recently, this possibility of electron transfer was suggested by McKinlay and Zeikus [18], however this was in combination with a mobile redox shuttle. They showed that hydrogenases have a role in reducing neutral red at the bacterial surface.

Bacteria can use soluble components that physically transport the electron from an (intra)cellular compound, which becomes oxidized, to the electrode surface. In many studies, redox mediators such as neutral red [24], thionin [25,26] and methyl viologen [2] were added to the reactor. The addition of these mediators often seemed to be essential [27]. However, bacteria can also produce redox mediators themselves, which can occur in two ways: through the production of organic, reversibly reducable compounds (secondary metabolites) and through the generation of oxidizable metabolites (primary metabolites).

The first method was suggested for many bacteria, such as *Shewanella putrefaciens* [28–30] and *Pseudomonas aeruginosa* [31,32]. It was recently shown that these microbial mediators influence the performance of an MFC [31] or more generally interfere in extracellular electron transfer [32]. Inactivation of the genes responsible for mediator production in a *Pseudomonas aeruginosa* MFC isolate reduced the current generation with a factor of 20. The redox mediators produced by one bacterium can be used by other bacterial species to reach the electrode.

The second way that bacteria can produce redox mediators – that is through primary metabolites – uses metabolites such as H_2 and H_2S as mediators. Schröder and coworkers [8,33] used *E. coli* K12 for generation of hydrogen gas, which was re-oxidized at a poly-anilin protected-platinum catalyzed electrode submerged in the bioreactor. In this way they obtained current densities of up to 1.5 mA/cm^2 (A, Ampere), which had not been attained previously. Similarly, Straub and Schink [34] addressed the reduction of sulphur by *Sulfurospirillum deleyianum* to sulphide, which was subsequently re-oxidized by iron to more oxidized intermediates.

Parameters defining the performance of MFCs

The power that can be generated in a microbial fuel cell is dependent on both biological and electrochemical processes (Box 2).

The substrate conversion rate

This depends on the amount of bacterial cells, the mixing and mass transfer phenomena in the reactor, the bacterial kinetics (μmax, the maximum specific growth rate of the bacteria, and Ks, the bacterial affinity constant for the substrate), the biomass organic loading rate (g substrate per g biomass present per day) [6], the efficiency of the proton exchange membrane for transporting protons [4,35] and the potential over the MFC.

Overpotentials at the anode

Generally, when the open circuit potential (OCP) of MFCs is measured, this OCP is in the order of between 750 mV to a reported maximum of 798 mV [5]. Parameters influencing the overpotentials are the electrode surface, the electrochemical characteristics of the electrode, the electrode potential, and the kinetics together with the mechanism of the electron transfer and the current of the MFC.

Overpotentials at the cathode

Similar to the losses observed at the anode, the cathode exhibits significant potential losses. To remediate this, several researchers have used hexacyanoferrate solutions [6,36,37]. However, hexacyanoferrate is not completely re-oxidized by oxygen in the air, and should be considered as an electron acceptor rather than a mediator [45]. To be sustainable, MFC cathodes preferably should be open-air cathodes [38–41].

The proton exchange membrane performance

Most MFC studies thus far applied Nafion™ (Dupont; http://www.dupont.com) proton exchange membranes (PEMs). However, Nafion™ membranes are sensitive to (bio)fouling by ammonium, for example. The best result was obtained using an Ultrex (Membranes International; http://www.membranesinternational.com) cation exchange membrane [7]. Liu *et al.* (2004) omitted the membrane, using pressed carbon paper as the separator. However, although this omission significantly decreased the MFC internal resistance [4] this type of separation provoked growth at the cathode based on anolyte constituents and allows poisoning of the cathode catalyst [41]. No data are as yet available regarding the stability of these carbon paper-cathode systems during periods longer than a few days [4].

Internal resistance of the MFC

This is dependent on both the resistance of the electrolyte between the electrodes and by the membrane resistance (Nafion™ has the lowest resistance). For optimal operation, anode and cathode need to be as close together as possible [41]. Also proton migration significantly influences resistance-related losses [42]; adequate mixing could minimize these losses.

Performance data

There is a clear discrepancy between results expressed in power per anode surface and power per unit of MFC reactor volume. Table 2 provides the most important results reported to date with MFCs. Most studies expressed power output as mA/m^2 respectively and mW/m^2 of electrode surface, as derived from descriptions of conventional catalytic fuel cells. The latter might be sufficient for chemical fuel cells but the nature of MFCs is different because the catalysts (bacteria) have specific requirements and occupy a certain volume in the reactor thus decreasing free space and pore size. Every study refers to a specific combination of reactor volume, proton-exchange membrane, catholyte, organic loading rate and anode surface. Comparison of these data is difficult at this point. From a technical point of view, it is useful to express the performance of the reactors in terms of $Watts/m^3$ of anode compartment volume (liquid) as a benchmark. This unit enables comparison of all tested reactors, not only within the existing studies but also with other existing bioconversion technologies.

There is a notable discrepancy between coulombic and energetic efficiency of reactors. The coulombic efficiency is calculated based on the amount of electrons transferred in relation to the amount of electrons theoretically delivered by the substrate. Energetic efficiency also implies the energy of the electrons transferred, incorporating both voltage and current. As can be seen in Table 2, the relationship between MFC current and power is not always unequivocal. Emphasis needs to be put on the electron transfer rate at a certain potential, and the fine-tuning of the operational parameters such as the resistance. Taking this parameter issue into account, whether maximal coulombic efficiency (e.g. for wastewater treatment) or energetic efficiency (e.g. for small scale batteries) is the ultimate goal must be determined. The power outputs thus far observed vary from mW/m^2 up to several W/m^2 electrode surface.

Optimization

Biological optimization implies the selection of suitable bacterial consortia and the bacterial adaptation to the optimized reactor conditions. Although the selection of the bacterial inoculum will largely determine the rate of enrichment, it does not determine the structural outcome of this procedure. Based on a mixed anaerobic–aerobic sludge inoculum and using glucose as feed, seven-fold increases in bacterial substrate to electricity conversion rates were observed after three months of microbial adaptation and selection [6]. Much faster increases were noted when larger anode surfaces were available for bacterial growth [43].

Batch systems will allow for accumulation of organisms that can produce soluble redox mediators [29,30,44]. Continuous systems select for biofilm-forming species that can either use the electrode directly by growing onto it, or transfer electrons through the biofilm matrix using mobile shuttling molecules [31].

Technological optimization can occur through the addition of soluble redox mediators to a batch anode: redox mediators have been added to MFCs and improved electron transfer consistently [2,25,45]. The selection of these mediators has so far been empirical, and generally a low mediator potential, in the order of -300 mV or more-reduced, was assessed as favourable. Redox mediators with a potential enabling bacteria to have a sufficiently high turnover rate in relation to the electrode should be selected, taking into account whether high coulombic or high energetic efficiency is the objective.

Several researchers have developed improved anode materials, by impregnating them with chemical catalysts. Park and Zeikus [37] used manganese modified kaolin electrodes, yielding power outputs up to $788\ mW/m^2$. Increasing the specific surface of the anode will allow for a lower current density (which in turn decreases the activation overpotential) and a higher biofilm surface. However, there is a distinct limit to this because small pores can become clogged rapidly by bacteria. Bacteria isolated from the food supply can die off and hence decrease the active surface of the electrode before lysis. Decreasing the activation overpotentials and the internal resistance will most strongly affect the power output. Some examples of existing reactor designs are depicted in Figures 2 and 3.

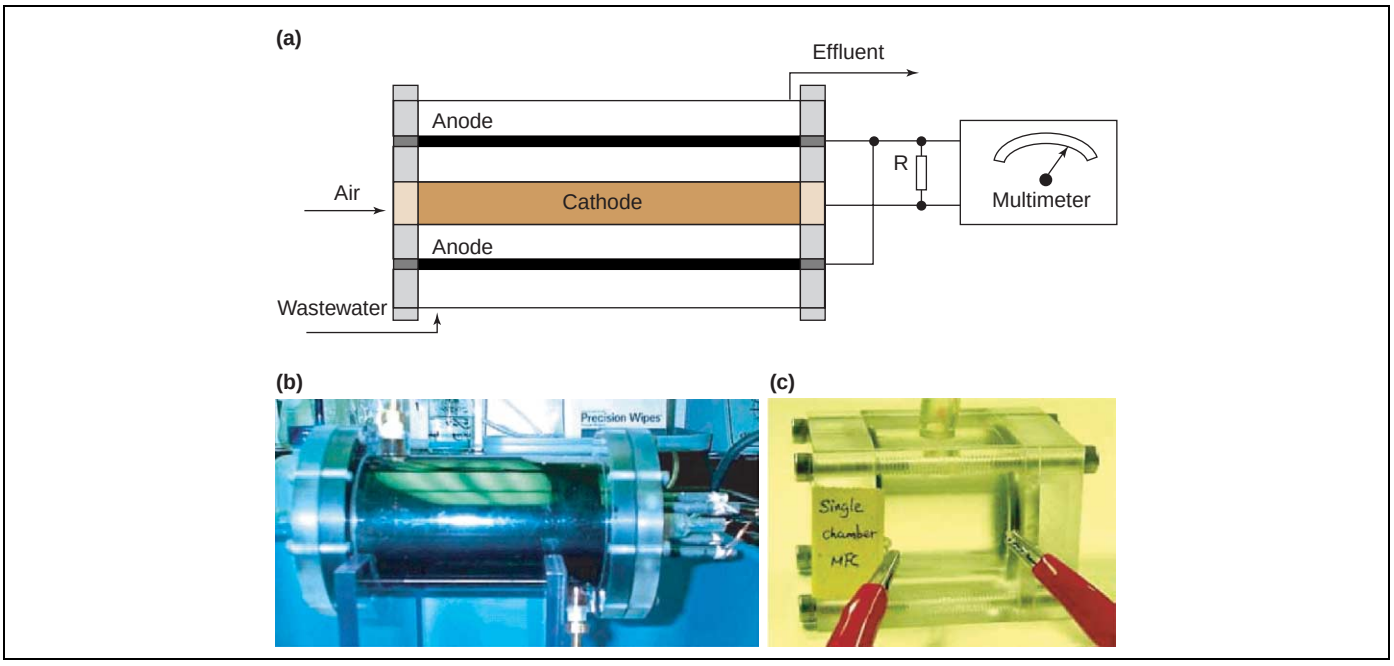

Figure 2. Microbial fuel cells (MFC) for the treatment of wastewater. (a) Tubular MFC with inner cathode compartment [5]. (b) Photograph of the set-up of drawing (a). (c) Single chamber MFC, where cathode and anode are at opposite ends of the reactor chamber [4]. Reprinted with permission from [5]. Copyright 2004 American Chemical Society.

MFC: sustainable core technology

Waste-driven applications require mainly significant removal of the waste substrate. Currently, when applying conventional aerobic treatment, ~1 kWh of energy is needed for oxidation per kilogram of carbohydrate present. For example, treatment of domestic wastewater represents an aeration energy cost of ~0.5 kWh per m^3, amounting to an energy use of the order of 30 kWh per capita per year (about €3 energy cost per capita per year). To address this issue, several technologies were developed, particularly for high-strength wastewaters. Most widespread in this context is the Upflow Anaerobic Sludge Blanket reactor, in which methane is produced, particularly when treating concentrated industrial wastewater. UASB reactors typically handle highly digestable wastewaters at a loading rate of 10–20 kg COD per m^3 reactor per day, and have (with a combustion engine as converter) overall electrical efficiencies of up to 35% [46], implying a power output of a 0.5–1 kW/m^3 reactor. The efficiency is mainly determined by energy losses during combustion of the biogas. Higher efficiencies might be possible in the future owing to the development of chemical fuel cells that oxidize the methane more efficiently than those currently available [47].

A battery converting a qualitative substrate with positive market value, such as glucose, will have as a primary goal high energetic efficiency (Box 3). Although the power density of the MFCs in comparison with, for example, methanol-driven FCs is considerably lower, the versatility in terms of safe substrates is an important asset of this technology.

Overall, as a matter of reference, the capital expenditure (Capex) for energy recovery from biomass by means of high rate anaerobic digestion is in the order of 1 million € per MW capacity installed [48]. The latter value is also valid for energy production from fossil fuel by conventional combustion processes, by wind turbines and by chemical fuel cells [49]. Hence, the processes are in a competitive area. Microbial fuel cells currently do not reach power outputs of this level. Loading rates of 0.1–10 kg chemical

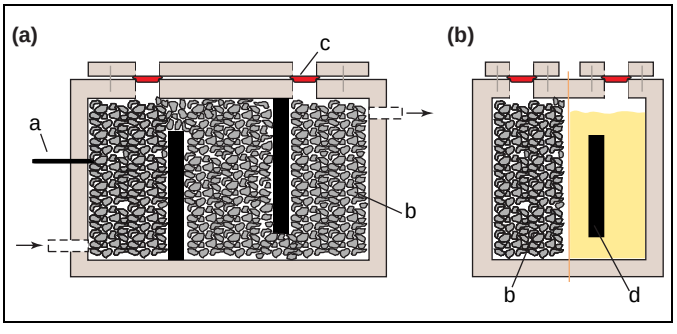

Figure 3. A flow-through microbial fuel cell for continuous treatment of liquid streams, as used by Rabaey *et al.* [43]. (a) Side view of the anode. (b) Front view of the MFC. Labels: a, contacting rod; b, granular graphite electrode matrix; c, sampling port; d, cathode in catholyte solution.

Box 3. Biomass as fuel for MFCs

Biomass has an energetic value, whether it is considered as foodstuff, energy crop or waste (in which case the value is generally negative). On average 1 kg of sugar, as a model component, contains 4.41 kWh of energy or potentially 13×10^6 Coulombs of charge. This 1 kg of sugar also represents 1.06 kg chemical oxygen demand (COD). Out of 1 kg carbohydrates, one can currently produce 0.5 L ethanol, 1.2 m^3 H_2 gas, 0.36 m^3 CH_4 gas or 0.5 m^3 biogas. On average, these processes yield ~1 kWh of useful energy. In the EU, 1 kWh is worth up to €0.16.

Because the production of this 1 kg of sugar costs about €0.25 and the market value approximates €1, using sugar to drive batteries is not a process feasible at large scale. However, much biomass is available on the market for low or negative prices. Although the intrinsic quality of this 'waste-biomass' is lower, the energy yield might still be sufficient to allow energy recovery by means of the MFC process.

oxygen demand (COD) per m^3 reactor per day can be expected that can, in practice, provide a power output between 0.01–1.25 kW/m^3. For a granular bed in a stacked MFC the capex cost based on materials as presented by Tsuchiya and Kobayashi [49], assuming a cost of €4000 per m^3 of electrode compartment, and 1 kW power output per m^3 anode, is estimated to be at a level 10 times that of the abovementioned energy producing processes. Even if the future material costs for MFCs decrease to the same extent as the material costs for chemical fuel cells, MFCs still need important breakthroughs to become economically competitive. However, their overall applicability and potential to operate at ambient temperatures is still largely unexplored. Moreover, waste-driven MFCs produce less excess biomass than aerobic wastewater treatment facilities [50]. Whereas for an aerobic treatment process the observed growth yield is ~0.4 g biomass formed per g organic substrate consumed, this yield is theoretically only 0.077 for anaerobic fermentation to methane [51]. Owing to the nature of the MFC process, the yield will be in between the two types of metabolism. Observed growth yields vary between 0.07 and 0.22 in glucose-fed MFCs [6]. As the sludge treatment cost of wastewater treatment facilities can amount up to €500 per ton dry matter [51], this quantitative reduction can have considerable implications on the economic balance of the process.

Efficient design and operation can create a platform technology, applicable in diverse fields without substantial modification. Aside from the economical aspect, the MFCs profile themselves as a sustainable core technology (i.e. basic technology adaptable to a wide variety of applications). They convert a wide array of electron donors with effective energy generation at low and moderate temperatures, even when the electron donor is provided at low concentrations. No existing technology today can match these criteria.

Conclusions

Microbial fuel cells are evolving to become a simple, robust technology. Certainly in the field of wastewater treatment, middle term application can be foreseen at market value prices. However, to increase the power output towards a stable 1kW per m^3 of reactor, many technological improvements are needed. Provided the biological understanding increases, the electrochemical technology advances and the overall electrode prices decrease, this technology might qualify as a new core technology for conversion of carbohydrates to electricity in years to come.

Acknowledgements

The authors wish to thank Peter Aelterman, Peter Clauwaert and Liesje De Schamphelaire for their useful comments. We thank Nico Boon, Vincent Denef, Wim De Windt and Bram Pauwels for critically reading this manuscript and Katleen Braet for the artwork. Financial support of the FWO through grant G.0172.05 is gratefully acknowledged.

References

1 Suzuki, S. (1976) Fuel cells with hydrogen-forming bacteria. *Hospital hygiene, Gesundheitswesen und desinfektion*, 159
2 Roller, S.D. *et al*. (1984) Electron-transfer coupling in microbial fuel-cells.1. Comparison of redox-mediator reduction rates and respiratory rates of bacteria. *J. Chem. Technol. Biotechnol. B Biotechnol.* 34, 3–12
3 Habermann, W. and Pommer, E-H. (1991) Biological fuel cells with sulphide storage capacity. *Appl. Microbiol. Biotechnol.* 35, 128–133
4 Liu, H. and Logan, B.E. (2004) Electricity generation using an air-cathode single chamber microbial fuel cell in the presence and absence of a proton exchange membrane. *Environ. Sci. Technol.* 38, 4040–4046
5 Liu, H. *et al*. (2004) Production of electricity during wastewater treatment using a single chamber microbial fuel cell. *Environ. Sci. Technol.* 38, 2281–2285
6 Rabaey, K. *et al*. (2003) A microbial fuel cell capable of converting glucose to electricity at high rate and efficiency. *Biotechnol. Lett.* 25, 1531–1535
7 Rabaey, K. *et al*. (2004) Biofuel cells select for microbial consortia that self-mediate electron transfer. *Appl. Environ. Microbiol.* 70, 5373–5382
8 Schröder, U. *et al*. (2003) A generation of microbial fuel cells with current outputs boosted by more than one order of magnitude. *Angew. Chem. Int. Ed. Engl.* 42, 2880–2883
9 Lee, J.Y. *et al*. (2003) Use of acetate for enrichment of electrochemically active microorganisms and their 16S rDNA analyses. *FEMS Microbiol. Lett.* 223, 185–191
10 Kim, B.H. *et al*. (2004) Enrichment of microbial community generating electricity using a fuel-cell-type electrochemical cell. *Appl. Microbiol. Biotechnol.* 63, 672–681
11 Champine, J.E. *et al*. (2000) Electron transfer in the dissimilatory iron-reducing bacterium *Geobacter metallireducens*. *Anaerobe* 6, 187–196
12 Beliaev, A.S. *et al*. (2001) MtrC, an outer membrane decahaem c cytochrome required for metal reduction in *Shewanella putrefaciens* MR-1. *Mol. Microbiol.* 39, 722–730
13 Nevin, K.P. and Lovley, D.R. (2002) Mechanisms for accessing insoluble Fe(III) oxide during dissimilatory Fe(III) reduction by *Geothrix fermentans*. *Appl. Environ. Microbiol.* 68, 2294–2299
14 Chaudhuri, S.K. and Lovley, D.R. (2003) Electricity generation by direct oxidation of glucose in mediatorless microbial fuel cells. *Nat. Biotechnol.* 21, 1229–1232
15 Kim, J.R. *et al.* Evaluation of procedures to acclimate a microbial fuel cell for electricity production. *Appl. Microbiol. Biotechnol.* (in press)
16 Thauer, R.K. *et al*. (1977) Energy-conservation in chemotropic anaerobic bacteria. *Bacteriol. Rev.* 41, 100–180
17 Logan, B.E. (2004) Extracting hydrogen electricity from renewable resources. *Environ. Sci. Technol.* 38, 160A–167A
18 McKinlay, J.B. and Zeikus, J.G. (2004) Extracellular iron reduction is mediated in part by neutral red and hydrogenase in *Escherichia coli*. *Appl. Environ. Microbiol.* 70, 3467–3474
19 Park, D.H. and Zeikus, J.G. (2002) Impact of electrode composition on electricity generation in a single-compartment fuel cell using *Shewanella putrefaciens*. *Appl. Microbiol. Biotechnol.* 59, 58–61
20 Park, H.S. *et al*. (2001) A novel electrochemically active and Fe(III)-reducing bacterium phylogenetically related to *Clostridium butyricum* isolated from a microbial fuel cell. *Anaerobe* 7, 297–306
21 Delaney, G.M. *et al*. (1984) Electron-transfer coupling in microbial fuel-cells.2. Performance of fuel-cells containing selected microorganism mediator substrate combinations. *J. Chem. Technol. Biotechnol. B Biotechnol.* 34, 13–27
22 Kostka, J.E. *et al*. (2002) Growth of iron(III)-reducing bacteria on clay minerals as the sole electron acceptor and comparison of growth yields on a variety of oxidized iron forms. *Appl. Environ. Microbiol.* 68, 6256–6262
23 Pham, C.A. *et al*. (2003) A novel electrochemically active and Fe(III)-reducing bacterium phylogenetically related to *Aeromonas hydrophila*, isolated from a microbial fuel cell. *FEMS Microbiol. Lett.* 223, 129–134
24 Park, D.H. *et al*. (1999) Microbial utilization of electrically reduced neutral red as the sole electron donor for growth and metabolite production. *Appl. Environ. Microbiol.* 65, 2912–2917
25 Choi, Y. *et al*. (2003) Dynamic behaviors of redox mediators within the hydrophobic layers as an important factor for effective microbial fuel cell operation. *B. Kor. Chem. Soc.* 24, 437–440
26 Lithgow, A.M. *et al*. (1986) Interception of the electron-transport chain

in bacteria with hydrophilic redox mediators.1. Selective improvement of the performance of biofuel cells with 2,6-disulfonated thionine as mediator. *J. Chem. Res.* 5, 178–179
27 Park, D.H. and Zeikus, J.G. (2000) Electricity generation in microbial fuel cells using neutral red as an electronophore. *Appl. Environ. Microbiol.* 66, 1292–1297
28 Hernandez, M.E. and Newman, D.K. (2001) Extracellular electron transfer. *Cell. Mol. Life Sci.* 58, 1562–1571
29 Angenent, L.T. *et al*. (2004) Production of bioenergy and biochemicals from industrial and agricultural wastewater. *Trends Biotechnol.* 22, 477–485
30 Rosso, K.M. *et al*. (2003) Nonlocal bacterial electron transfer to hematite surfaces. *Geochim. Cosmochim. Acta* 67, 1081–1087
31 Rabaey, K. *et al.* Microbial phenazine production enhances electron transfer in biofuel cells. *Environ. Sci. Technol.* (in press)
32 Hernandez, M.E. *et al*. (2004) Phenazines and other redox-active antibiotics promote microbial mineral reduction. *Appl. Environ. Microbiol.* 70, 921–928
33 Niessen, J. *et al*. (2004) Exploiting complex carbohydrates for microbial electricity generation - a bacterial fuel cell operating on starch. *Electrochem. Commun.* 6, 955–958
34 Straub, K.L. and Schink, B. (2004) Ferrihydrite-dependent growth of *Sulfurospirillum deleyianum* through electron transfer via sulfur cycling. *Appl. Environ. Microbiol.* 70, 5744–5749
35 Jang, J.K. *et al*. (2004) Construction and operation of a novel mediator- and membrane-less microbial fuel cell. *Process Biochem.* 39, 1007–1012
36 Park, D.H. and Zeikus, J.G. (1999) Utilization of electrically reduced neutral red by *Actinobacillus succinogenes*: Physiological function of neutral red in membrane-driven fumarate reduction and energy conservation. *J. Bacteriol.* 181, 2403–2410
37 Park, D.H. and Zeikus, J.G. (2003) Improved fuel cell and electrode designs for producing electricity from microbial degradation. *Biotechnol. Bioeng.* 81, 348–355
38 Pham, T.H. *et al*. (2004) Improvement of cathode reaction of a mediatorless microbial fuel cell. *J. Microbiol. Biotechnol.* 14, 324–329
39 Min, B. and Logan, B.E. (2004) Continuous electricity generation from domestic wastewater and organic substrates in a flat plate microbial fuel cell. *Environ. Sci. Technol.* 38, 5809–5814
40 Oh, S. *et al*. (2004) Cathode performance as a factor in electricity generation in microbial fuel cells. *Environ. Sci. Technol.* 38, 4900–4904
41 Larminie, J. and Dicks, A. (2000) *Fuel cell systems explained*, John Wiley & Sons
42 Gil, G.C. *et al*. (2003) Operational parameters affecting the performance of a mediator- less microbial fuel cell. *Biosens. Bioelectron.* 18, 327–334
43 Rabaey, K. *et al.* (2005) Continuous microbial fuel cells convert carbohydrates to electricity. *Wat. Sci. Technol.* (in press)
44 Rabaey, K. *et al.* (2004) Bacteria generate current through production and/or use of soluble redox mediators. In *Annual meeting of the American Chemical Society 2004*, American Chemical Society
45 Park, D.H. *et al*. (2000) Electricity production in biofuel cell using modified graphite electrode with neutral red. *Biotechnol. Lett.* 22, 1301–1304
46 Ross, C. *et al.* (1996) *Handbook on Biogas Utilization* (2nd edn), Muscle Shoals, AL: Southeastern Regional Biomass Energy Program
47 Vanherle, J. *et al*. (2004) Biogas as a fuel source for SOFC co-generators. *J. Power Sources* 127, 300–312
48 Kennedy, S. Wind power planning: assessing long-term costs and benefits. *Energy Policy* 33, *1661-1675*
49 Tsuchiya, H. and Kobayashi, O. (2004) Mass production cost of PEM fuel cell by learning curve. *Internat. J. Hydrogen Energy* 29, 985–990
50 Kim, B.H. *et al.* (2004) Microbial fuel cells and beyond. In *Annual meeting of the American Chemical Society 2004*, American Chemical Society
51 Weemaes, M. and Verstraete, W. (2001) Other treatment techniques. In *Sludge into biosolids* (Spinosa, L. and Vesilind, P.A., eds), pp. 364–383, IWA Publishing
52 Liu, H. *et al*. (2005) Production of electricity from acetate or butyrate using a single-chamber microbial fuel cell. *Environ. Sci. Technol.* 39, 658–662
53 Madigan, M.T. *et al*. (2000) *Brock Biology of Microorganisms*, Prentice Hall
54 Rabaey, K. *et al.* (2005) Microbial fuel cells: performances and perspectives. In *Biofuels for fuel cells: biomass fermentation towards usage in fuel cells* (Lens, P.N. *et al.*, eds) (in press)
55 Kim, B.H. *et al*. (1999) Electrochemical activity of an Fe(III)-reducing bacterium, *Shewanella putrefaciens* IR-1, in the presence of alternative electron acceptors. *Biotechnol. Tech.* 13, 475–478
56 Kim, B.H. *et al*. (1999) Direct electrode reaction of Fe(III)-reducing bacterium, *Shewanella putrefaciens*. *J. Microbiol. Biotechnol.* 9, 127–131
57 Kim, H.J. *et al*. (1999) A microbial fuel cell type lactate biosensor using a metal- reducing bacterium, *Shewanella putrefaciens*. *J. Microbiol. Biotechnol.* 9, 365–367
58 Kim, H.J. *et al*. (2002) A mediator-less microbial fuel cell using a metal reducing bacterium, *Shewanella putrefaciens*. *Enzyme Microb. Technol.* 30, 145–152
59 Vega, C.A. and Fernandez, I. (1987) Mediating effect of ferric chelate compounds in microbial fuel-cells with *Lactobacillus plantarum*, *Streptococcus lactis*, and *Erwinia dissolvens*. *Bioelectrochem. Bioenerg.* 17, 217–222
60 Cooney, M.J. *et al*. (1996) Physiologic studies with the sulfate-reducing bacterium *Desulfovibrio desulfuricans*: Evaluation for use in a biofuel cell. *Enzyme Microb. Technol.* 18, 358–365
61 Bond, D.R. and Lovley, D.R. (2003) Electricity production by *Geobacter sulfurreducens* attached to electrodes. *Appl. Environ. Microbiol.* 69, 1548–1555
62 Vandevivere, P. and Verstraete, W. (2001) Environmental applications. In *Basic biotechnology* (Ratledge, C. and Kristiansen, B., eds), pp. 531–557, Cambridge University Press
63 Bond, D.R. *et al*. (2002) Electrode-reducing microorganisms that harvest energy from marine sediments. *Science* 295, 483–485
64 Tender, L.M. *et al*. (2002) Harnessing microbially generated power on the seafloor. *Nat. Biotechnol.* 20, 821–825