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Article

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Review article

The electrochemical perspective of bioelectrocatalytic activities in microbial electrolysis and microbial fuel cells

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ABSTRACT

Transforming organic waste directly into electricity or indirectly into sources of hydrogen fuel is credible through exoelectrogen microorganisms grown on the anode or cathode that catalyze electrochemical reactions. In this review, we discuss the origin of the electrochemical kinetic in both microbial fuel cells (MFCs) and microbial electrolysis cells (MECs) that are utilized to produce energy from waste through either directly by producing electric energy, or indirectly through hydrogen gas production, respectively. The concept of utilizing electrochemical techniques of cyclic voltammetry, chronoamperometric and derivative cyclic voltammetry to study the interfacial kinetics of exoelectrogenic bacteria and characterize biofilms are described. Additionally, we discuss the influence of various parts of electrochemical cells on bioelectrocatalytic processes, i.e. system design, electrolyte properties, anode and cathode materials. Thus, the necessity of optimizing parameters impacting the efficiency, rate, bacteria enrichment, and system implementations for improved biofilm performance are briefly discussed along with the figures of merit.

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1. Introduction

It is said that as much as 10 times the energy required for the treatment of municipal wastewater may be recoverable from it (Shizas and Bagley, 2004; Fan et al., 2012). One of the key sustainable technologies that are useful for retrieving energy

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from wastes is via the use of exoelectrogen microorganisms (or exoelectrogens), which are characterized as extracellular electron transferors (EETs), i.e., capable of donating or accepting electrons (Hernandez and Newman, 2001). Based on this principle, dual benefits are obtained: treatment of wastewater and production of energy from waste (Zhang et al., 2019). Thus, as with organisms involved in the biological remediation of waste, EETs digest soluble organic or inorganic entities from streams to produce metabolism products such as alcohol, methane, or hydrogen. The production of these metabolites from waste is of great value from the environmental and energy conservation points of

view. Hence, due to the importance of these electron transfer reactions, electrochemists have utilized some of the natural bacterial processes for energy conservation at anode and/or cathode compartments (Rittmann, 2008). A schematic of the exchange of electrons at donor and acceptor EETs is simplified in Fig. 1. Growing acceptor EETs on cathode has been used as a potential alternative to platinum for catalyzing some cathodic reactions because of its low cost and acceptable microbial catalytic activity (Jeremiasse et al., 2010; Rowe et al., 2017; Rimboud et al., 2017). Microorganisms that can produce hydrogen such as *Desulfovibrio vulgaris* (Guiral-Brugna et al., 2001; Lojou et al., 2002) are found in a variety of environments and contain hydrogenases that catalyze the reversible reaction ($2\text{H}^+ + 2\text{e}^- \leftrightarrow \text{H}_2$) (Schwartz et al., 2013). *Methanosarcina* bacteria, that are abundant in the environment, also grow on cathode and use electrons for the reduction of carbon dioxide to methane (Deppenmeier, 2004).

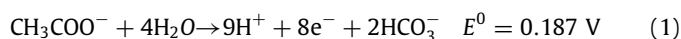
Donor EETs that grow on the anode are referred to as anodic respiring bacteria (ARB). ARBs, which include a variety of microorganisms, such as dissimilatory iron-reducing bacteria (DMRB) (e.g., *Shewanella* and *Geobacter*). ARBs oxidize soluble organics in the waste stream and donate electrons within a closed electrochemical circuit to the final electron acceptor port (Deutzmann and Spormann, 2017).

Exploiting the principle of using ARB to utilize organic wastes as feed substrates and at the same time to generate energy will promote an environmentally benign technology. The energy of the electrons can be utilized for electricity generation in a microbial fuel cell (MFC) (Paitier et al., 2017; Kim et al., 2002) or for hydrogen gas production in a microbial electrolysis cell (MEC) (Yasri and Nakhla, 2017a). Both systems, collectively are referred to hereafter as MXCs, and share similarities in the anodic process as well as EETs. These systems are considered among the electrochemical technologies, and in particular, the bioelectrochemical technology for microbial energy generation. Published literature supports the effectiveness and the environmental compatibilities of the MXCs for a variety of organic remediation as well as a wide range of applications, e.g. remote monitoring for water quality (Parkhey and Mohan, 2019), power productions (Xin et al., 2019), renewable source of energy (Kokabian et al., 2018; Chen et al., 2019), CO_2 reduction (Sánchez et al., 2019), conversion of waste streams to valuable resources via biosynthesis (Wu et al., 2019; Katuri et al., 2018; Reddy and Sun, 2019). Moreover, researchers have started working on genetically engineered biofilms (Li et al., 2018; Angelaalincy et al., 2018; Davidov et al., 2019) and hybrid systems (Tee et al., 2018), hoping for a real new breakthrough of the system. In this regard, the fundamentals and applications of MFCs (Slate et al., 2019; Santoro et al., 2017) and MECs have been reviewed recently and so are some important aspects of active biofilms and biocatalysts in MECs (Babauta et al., 2012a; Hasany et al., 2016; Rathinam et al., 2019). Other related reviews on the subject include the type of anode materials and feeding substrates used to grow biofilms (Angelaalincy et al., 2018; Ghasemi et al., 2013; Kadier et al., 2014; Azeredo et al., 2017), the type of cathodes used to reduce oxygen in MFCs and to produce hydrogen in MECs (Ben Liew et al., 2014; Kundu et al., 2013). Unfortunately, however, these reviews do not focus on the electrochemical understanding of MXCs. The electrochemical principles and concepts of MXCs systems have scarcely detailed in the literature. Therefore, in this review, we focus on the electrochemical systems that mostly depend on ARBs for energy production from waste. Taking an electrochemist's perspective of MXCs, we have sought to establish a firm foundation for the understanding of concepts, highlight the importance of each part of the system and the future trends for application. We hope this review will provide a deeper insight into the electrochemical theory and models for MXCs.

2. MFC and MEC

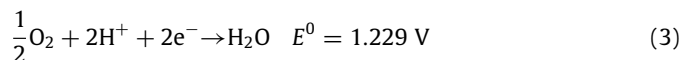
The basic principles of MFC and MEC systems are depicted in Fig. 2. Though there are a variety of cell designs reported in the literature, for comparison purposes we will consider MEC and MFC cells that are constructed similarly with an anode, a cathode, electrolyte, and an electrical circuit. From an electrochemical point of view, the main difference between the two systems is in how they contribute to energy by allowing hydrogen production in MECs and utilizing the reduction of dissolved oxygen to produce electric current in MFCs (Harnisch and Schroder, 2010; Pandit and Das, 2018).

Considering the oxidation of acetate at anode surface (Eq. (1)) in neutral conditions similar to the inoculum found widely in MFC and MEC systems (i.e. pH = 7 in 5 mM bicarbonate), the theoretical equilibrium potential required to oxidize 1 g/L (16.9 mM) of acetate can be calculated via the Nernst equation (Eq. (2)) as about −300 mV. Although the Nernst potential should not depend on the anode material, the presence of biofilm may change the kinetics of the reaction and hence derives an additional overpotential (which is not considered in this calculation). In addition, the concentration of species in the electrolyte media are not constant, thus the calculated value of the Nernstian potential may shift from the aforementioned theoretical value due to changes in the species concentrations in the presence of the biofilm at the electrode surface, or possibly the reaction pathway may change to a different redox potential. These various possibilities will be discussed further in Section 4.



$$\begin{aligned} E_{\text{An}} &= E_{\text{An}}^0 - \frac{RT}{8F} \ln \frac{[\text{CH}_3\text{COO}^-]}{[\text{HCO}_3^-]^2 [\text{H}^+]^9} \\ &= 0.187 - \frac{\left(\frac{8.31 \text{ J}}{\text{molK}}\right) (298.15 \text{ K})}{8 \left(\frac{96500 \text{ C}}{\text{mol}}\right)} \ln \frac{[0.0169]}{[0.005]^2 [10^{-7} \text{ M}]^9} \\ &= -0.300 \text{ V} \end{aligned} \quad (2)$$

Considering, on the other hand, the reactions at the cathode side, the conditions for MFCs and MECs in this chamber are different; i.e., dissolved oxygen is available in the cathodic chamber for MFC system but not for MEC. Thus oxygen reduction reaction (Eq. (3)) will occur in MFC, whereas, hydrogen production occurs in MEC (Eq. (5)). In the MFC system, considering also a neutral pH (= 7) and an oxygen saturated catholyte media (an oxygen partial pressure of $p_{\text{O}_2} = 0.21 \text{ atm}$), the cathodic Nernst potential is calculated to be about 805 mV (Eq. (4)). Thus, the MFC's equilibrium cell voltage corresponds to about 1.105 mV (i.e. $E_{\text{e,cell}} = [0.805 \text{ V}] - [-0.300 \text{ V}]$).



$$\begin{aligned} E_{\text{Ca}} &= E_{\text{Ca}}^0 - \frac{RT}{nF} \ln \frac{1}{[\text{O}_2]^{1/2} [\text{H}^+]^2} \\ E_{\text{Ca}} &= 1.229 - \frac{\left(\frac{8.31 \text{ J}}{\text{molK}}\right) (298.15 \text{ K})}{2 \left(\frac{96500 \text{ C}}{\text{mol}}\right)} \times \ln \frac{1}{[0.2]^{1/2} [10^{-7} \text{ M}]^2} \\ &= 0.805 \text{ V} \end{aligned} \quad (4)$$

For a deoxygenated cathodic chamber in the MEC system, hydrogen will be produced. The calculated voltage from Nernst equation (Eq. (6)) at neutral pH (= 7) is −414 mV. Thus, the corresponding equilibrium cell voltage is about −114 mV ($E_{\text{e,cell}} = [-0.414 \text{ V}] - [-0.300 \text{ V}]$). The reaction, in this case, is not spontaneous, i.e., external energy is required. Hence, biofilm formation is almost universally performed at a constant applied potential

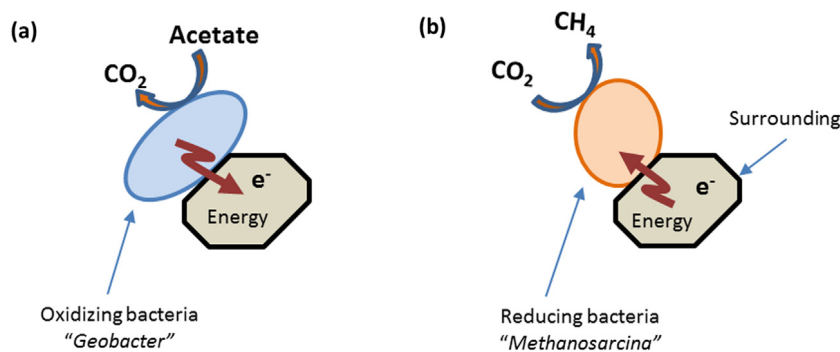


Fig. 1. Schematic of exchange of electrons at EET donor (a) and acceptor (b).

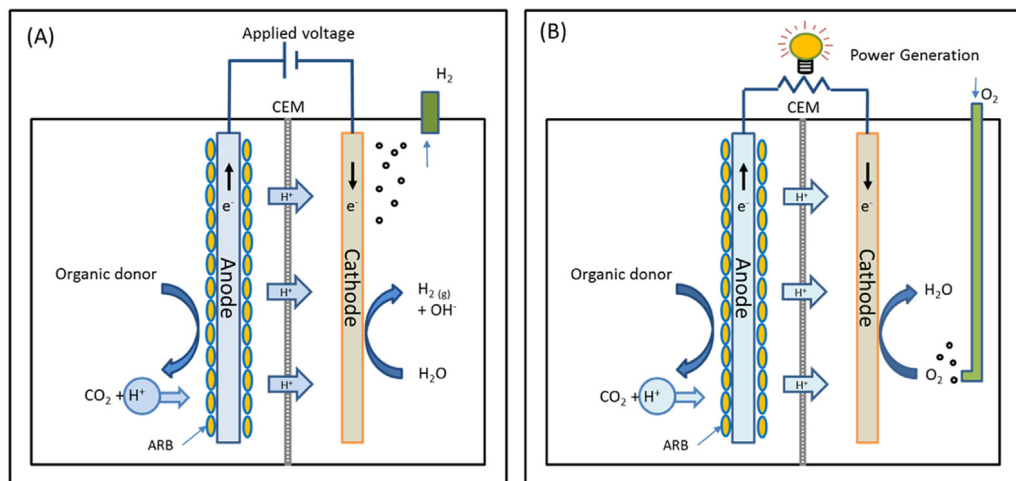


Fig. 2. Schematic of (A) MEC and (B) MFC systems consisting of two-compartment cells of anolyte and catholyte separated by cation exchange membrane (CEM). MEC produces H₂ by combining substrate utilization reaction at the anode by ARBs under small applied potential and MFC produces electric current by combining two half-cell reactions of substrate utilization at the anode and oxygen reduction at the cathode.

(−0.6 V to +0.5 V vs. standard hydrogen electrode, SHE) to produce H₂ in MEC systems (Rimboud et al., 2014).

$$2\text{H}^+ + 2\text{e}^- \rightarrow \text{H}_2 \quad E^0 = 0.000 \text{ V} \quad (5)$$

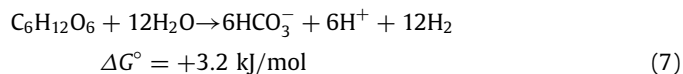
$$E_{\text{Ca}} = E_{\text{Ca}}^0 - \frac{RT}{nF} \ln \frac{\text{H}_2}{[\text{H}^+]^2} \quad (6)$$

$$E_{\text{Ca}} = 0.0 - \frac{\left(\frac{8.31 \text{ J}}{\text{molK}}\right) (298.15 \text{ K})}{2 \left(\frac{96500 \text{ C}}{\text{mol}}\right)} \times \ln \frac{1}{[10^{-7} \text{ M}]^2} = -0.414 \text{ V}$$

According to these calculations of the total Nernstian cell potentials, MFC is a galvanic cell capable of energy generation at a maximum cell voltage of 1.105 V [$E_{\text{e, cell}} = (0.805 \text{ V}) - (-0.300 \text{ V})$]; and MEC is an electrolytic cell requiring external energy input at a minimum cell voltage of 0.114 V [$E_{\text{e, cell}} = (-0.414 \text{ V}) - (-0.300 \text{ V})$], with the production of gaseous hydrogen at the cathode. Besides, it is worth noticing that in the aforementioned calculation we have not included the resistance encountered within the MXC (ohmic loss, activation overpotential and concentration overpotential, which may vary from system to another). Thus, hydrogen production will require a voltage higher than the calculated $E_{\text{e, cell}}$ (ranging between 0.2–0.8 V). Thus, MECs can be considered as modified MFCs, which are known as bio-catalyzed electrolysis cells or bio-electrochemically assisted microbial reactors (Pandit and Das, 2018; Roy and Pandit, 2019; Liu et al., 2005a). Considering, however, hydrogen as a green and sustainable source of energy that releases no greenhouse gases upon combustion (Edwards et al., 2007), the use of MEC is considered a sustainable option for H₂ production (Venkata Mohan and

Pandey, 2019) that utilizes substrates such as wastes and/or other biomass for energy production (Wang and Ren, 2013).

Studying, on the other hand, substrates other than acetate, e.g., glucose (Eqs. (7)–(9)), the conversion reactions are the fermentation to acetate or butyrate are favored (negative ΔG° values) rather than complete hydrogen evolution (Eq. (7); nonspontaneous reaction). To understand the advantages of MEC system over fermentation and its variations, we compare here the energy requirement for oxidation of some simple waste products (e.g., glucose, butyrate, and acetic acids) when they are utilized as feed substrates in both MECs and fermentation processes. Theoretically, the maximum yield for complete oxidation of 1 M of glucose in an MEC is 12 M of H₂ (Eq. (7)), whereas, in the fermentation process it is 4 M of H₂ if acetate is formed (Eq. (8)) or 2 M of H₂ if butyrate is formed (Eq. (9)). We should note that oxidation of glucose and its fermentation products (acetate and butyrate) (Eqs. (7), (10), (11)) are not directly converted to H₂ without an external energy input (free energy values of reactions are not spontaneous under standard conditions). Therefore, the use of MEC to oxidize these chemicals will theoretically yield more H₂ and more energy than what can be obtained with fermentation. The increase in H₂ production is perhaps due to the ability of MECs to use various bacteria and wider sources of organic matter compared to fermentation (Lu et al., 2012).



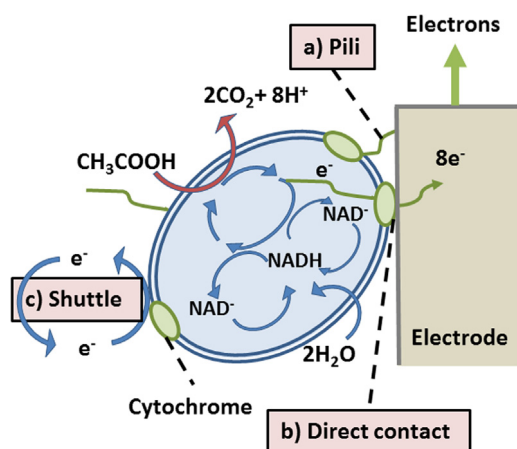
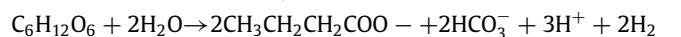
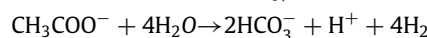


Fig. 3. Illustration of EET mechanisms employed by microorganisms at the electrode–biofilm interface. (a) use of cell appendages for electron transfer (pili), (b) direct electron transfer, and (c) electron transfer mediated by electron shuttle compounds (With modification from Carmona (2012)).

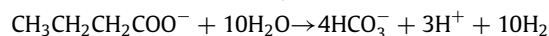
$$\Delta G^\circ = -206.3 \text{ kJ/mol} \quad (8)$$



$$\Delta G^\circ = -256.8 \text{ kJ/mol} \quad (9)$$



$$\Delta G^\circ = +104.6 \text{ kJ/mol} \quad (10)$$



$$\Delta G^\circ = +257.3 \text{ kJ/mol} \quad (11)$$

Fermentation is a simple process, in both reactor conditions and design; however, considering MXCs many parts and parameters should be of importance. In MXCs, the construction of all the system components (i.e., electrodes, membrane, media condition, inoculum sources, cell design) contribute to the better performance of the process. Henceforth, the role of each component in improving the bioelectrocatalytic performance of the system will be discussed.

3. Exoelectrogenic microorganisms

Exoelectrogenic microorganisms (Logan, 2009a) are bioelectrocatalytically active species that grow in MXCs and transfer electrons to the anode port without the need for exogenous mediators. These microorganisms are usually grown in anaerobic condition on the anode surface as electron acceptors and utilize substrates (organic and inorganic) in the effluent as electron donors. Babauta et al. (2012a) and Hasany et al. (2016) reviewed various factors impacting the activity of biofilms and biocatalysts in microbial electrochemical systems. We found that to understand the electrochemical concept of the MXCs system, it is necessary to examine from an electrochemical perspective the main factors and methods impacting the system. In this section, we will discuss the EET mechanisms, inocula cultivation, acclimation, substrate variation, and enrichment of microorganisms that influence the electrochemical catalytic activity of biota in MXCs to help understand the concept of exoelectrogenic microorganisms.

Simplified mechanisms of EET digesting acetate ions, as an example of electron donor are depicted in Fig. 3 (Carmona, 2012), which are based on three proposed pathways: (1) direct electron transfer through microbial outer membrane cytochromes, (2) electron shuttle molecules, mediated by the grown biofilm, and (3) via a solid conductive protein appendages produced by bacteria and shaped like nanowires or pili (Rathinam et al., 2019;

Logan, 2009a; Torres et al., 2010; Lovley, 2008; Amit Kumar et al., 2012; Lienemann et al., 2018). Many detailed investigations on the type and structure of the nanowires, specifically those performed by the Malvankar research group at Yale University (USA), reveal recently that the structures are assembled by micrometer-long polymerization of the hexaheme cytochrome, with hemes packed within $\sim 3.5\text{--}6 \text{ \AA}$ of each other (Wang et al., 2019a). This structure explains the significant capability of biofilm grown on electrode to conduct electrons toward remote collectors. Moreover, in the surrounding media, the electrochemical gradient is considered the main driving force for the diffusion of the basic cellular functions, including chemo-osmotic transport and ATP synthesis (Amit Kumar et al., 2012). EET mechanisms are not mutually exclusive within a species nor in one pathway. For example, pure *Shewanella oneidensis* culture can transfer electrons through the interiorly produced riboflavins that can function as electrons shuttle. Pure culture of *Geobacter sulfurreducens* also has an outer membrane cytochrome to grow nanowires that can conduct electrons through a $50 \text{ }\mu\text{m}$ thick anodic biofilm, however, do not produce flavins or other mediators (i.e. pyocyanin, melanin, or quinones) (Nevin et al., 2008). In mixed culture biofilms, however, due to the variety of biota involved and their secretions, it is impossible to distinguish the electron transfer mechanisms involved (Zhen et al., 2017). Elucidation of the mechanism of EET requires working with model microorganisms (pure strains), under controlled conditions, or via simulation (Cereda et al., 2014).

Substrates which have been exploited as feed in MXCs are varied, both in number and implementations, with special interests usually focused on waste containing organics for bioenergy production (Zhang et al., 2019; Pant et al., 2011). The major substrates that have been utilized include wastewaters, fermentable and non-fermentable organic materials (Kadier et al., 2014; Pant et al., 2010). Pant et al. (2010) and Kadier et al. (2014) reviewed various substrates used in MFCs and MECs systems, respectively. However, for benchmarking newly developed or designed operational conditions, acetate is commonly used as carbon source, that is considered as an end product of several metabolic pathways and an inertness molecule toward alternative microbial conversions (i.e. fermentations) (Biffinger et al., 2008). It has been reported, for both of MFCs (Liu et al., 2005b) and MECs (Yang et al., 2015) systems, that the power generation and hydrogen production with the acetate-fed system are higher than those produced with butyrate, propionate, and glucose. Very recently, Yang et al. (2015) compared the performance of three common fermentation products of acetate, butyrate and propionate as MEC's substrates in terms of H_2 and power output. Acetate-fed MEC showed the highest hydrogen production rate ($0.53 \text{ m}^3/\text{m}^3\cdot\text{day}$), current density (6.0 A/m^2) and Coulombic efficiency (87%), followed by butyrate ($0.18 \text{ m}^3/\text{m}^3\cdot\text{day}$, 2.5 A/m^2 , 72%), and propionate ($0.072 \text{ m}^3/\text{m}^3\cdot\text{day}$, 1.6 A/m^2 , 51%). They attributed the variation in efficiency to the ease of acetate utilization by ARB in MEC, while butyrate and propionate could not be oxidized to the same degree. The use of more complex substrates resulted in lower utilization rate and efficiency. For example, Montpart et al. (2015) evaluated the efficiency of a single-chamber MEC fed with synthetic wastewater containing carbon sources of different complexity, such as glycerol, milk and starch. Although their results indicated that milk-fed MEC is the highest in hydrogen production rate ($0.086 \text{ m}^3/\text{m}^3\cdot\text{day}$), current density (4.0 A/m^2 calculated from data in Montpart et al. (2015)) and Coulombic efficiency (52%), but these values are still far less than those obtained with acetate.

Biofilm formation on electrode starts immediately after adding inocula to the substrate medium, that contains electron donors (Sultana et al., 2015), and closing the electrochemical circuit

in which this growing phase indicated as lag-phase (or start-up phase). Cultivation of ARB biofilms is preferably conducted at steady state in a fed-batch system, as the flowing or movement of inocula lengthens the start-up of the acclimatization process (Yi et al., 2009). The importance of the start-up phase has been reviewed by the literature of Kumar et al. (2017). Their review revealed the relations between the start-up phase and efficient bioenergy generation and adequate long-term operation. The factors that impact the start-up phase and hence reflect on the efficiency of the total process include inoculum selection, enrichment, operating conditions and cell architecture (Kumar et al., 2017). They summarized that to obtain as high process efficiencies as possible, cell design, taking into account electrode materials, interfacial charge-transfer resistances, and membranes must all be of primary concern to aid start-up and subsequent steady-state operation. Indeed, we proved in a recent study on the impact of interfacial charge transfer on the start-up of bio-electrochemical system inoculated from mixed culture (Yasri and Nakhla, 2017b) that both anodic interfacial conductivity and natural attraction of material present on the electrode surface can be important factors to initiate attachment of microorganisms on the surface (Yasri and Nakhla, 2017b). We evaluated the initial acclimatization behavior of ARB on various anode surfaces doped with conductive (Fe_3O_4), semi-conductive (FeS) or non-conductive (CaS) interface. Our work demonstrates that when dealing with common species of ARB (e.g., Fe, Ca, S), the interfacial charge transfer is not among the factors which enhance EET. During start-up, the attraction of biota to grow on the iron-containing interface was high, however, in the subsequent growth stages, the electrochemical kinetic study indicated suppression of the bacteria-produced mediators on iron-containing anodes which reduces their electrochemical activity. However, electrode interface containing nonconductive species (e.g., CaS) shows higher electrochemical efficiency than a conductive iron-containing anode that may naturally attract bacteria to behave differently (Yasri and Nakhla, 2017b).

Moreover, the electrical output in MXCs is independent of the concentration of the substrate used, i.e. increasing the concentration of electron donor substrate does not increase the electric current output (Yasri and Nakhla, 2017a; Liu et al., 2005b; Yang et al., 2015; Strycharz et al., 2011). For example, Liu et al. (2005b) reported that the voltages generated in an MFC system using acetate at different concentrations (from 80 mg/L to 800 mg/L) was nearly stable at around 0.45 V. Yang et al. (2015) noted that the current density in an MEC system also remained the same upon changing the influent acetate concentration from 1600 mg/L to 800 mg/L. This concentration independent behavior reflects the basic concept of the limiting catalytic activity to produce a limiting current in MXCs, i.e., the current generated by ARB is the sum of the metabolism rates of bacteria which is associated with substrate utilization, biomass synthesis, respiration, and decay. The current output depends only on the type of the cultivated bacterium, interface materials on the electrode, and the type of utilized substrate.

The phenomena of limiting current density in bioelectrochemical cells is well established in the literature (Yang et al., 2015), for example, Oh and Logan (2005) noted that the limiting current density remained nearly constant by doubling the amount of propionate substrate from 0.26 mM to 0.53 mM. The limiting current is dependent on the amount of bacteria grown on the electrode and also correlated with the type of bacteria as well as the type of electronically conductive strain present in the biofilm. An important study performed by Yi et al. (2009) found that the growth of electrically conductive microbial nanowires in the biofilm (in the presence of KN400 strain) reached a higher limiting current of 7.6 A/m² than non-conductive strain (1.4 A/m²).

Moreover, continuous investigation in this domain by Wang et al. (2019a) reveals that wild-type OmCS filaments show 100-fold greater conductivity than other filaments from a ΔomCS strain, emphasizing the significance of conductive strains in biofilms. On the other hand, different electrode materials will show different limiting current density (Yasri and Nakhla, 2016), these mostly depend on the type of electrode interface to provide an attracting surface for biotic species to attach. For example, although Indium-Tin oxides (ITO) film shows a highly conductive surface electrode, the modification of the surface with three-dimensional conductive nanowire networks produce higher limiting current density as compared to the plain ITO which was attributed to the enhanced electron transfer via mediator molecules in the surface structures of the electrode (Zhao et al., 2010). The type of substrate also impacts the limiting current density, which has been attributed to the rate at which bacteria utilize the substrate (Yang et al., 2015). Moreover, the amount of mediator excreted by biofilms is equally important in limiting the current. For example, Marsili et al. (2008a) reported a drop of 80% in current produced via *S. Oneidensis* MR-1 on graphite electrode following the replacement of a spent growth medium with fresh medium. They presume that this drop in the limiting current is due to the presence of flavins in the spent medium which acts as redox mediators, impacting the current production.

When the substrate is consumed, the current will reduce, hence, the current generation can be boosted again by adding or dosing the substrate (Yasri and Nakhla, 2017a, 2016); however, in some cases, it will be necessary to partially or completely replace the medium (Zhu et al., 2012; Chen et al., 2012). Thus, successive additions of an inoculum are necessary in the first batches, and then only the addition of electron donor substrate (Yasri and Nakhla, 2017a). Following the growth of biofilms, their stability can be determined by obtaining a constant current during successive addition of the same substrate (Pasternak et al., 2018; Ramírez-Vargas et al., 2018). However, to maintain a stable current output the substrate concentration is usually kept above a threshold corresponding to the required limiting current output (Torres et al., 2007).

Although mixed culture biofilms are generally cultivated from the primer inoculum containing mixed communities that coexist in natural (sludge, sediments, biological treatment plants, etc.), purifying these types of biofilm can be approached by eliminating or washing off non-active species, e.g., non-exoelectrogens microorganisms. This can be performed by transplanting biofilm from one MFC system to another, which also helps with the enrichment of microorganisms. Secondary biofilms obtained via transplanting usually enables the growth of fewer varieties of culture in the biofilm (Mahmoud et al., 2018). On the other hand, some experimental designs require bacterial enrichment to promote specific species in the inoculum (Saratale et al., 2017). Considering the fact, however, that waste stream which may be fed to MXCs will contain a variety of complex species, efficient MXCs will require acclimation of bacterial to this media. In this regard, efforts were made to adapt organism growing on anode with various strains to enhance the efficiency. Yi et al. (2009) found that adaption of KN400 strain associated with the abundance of electrically conductive microbial nanowires was more effective in current production (7.6 A/m²) than strain DL-1 (1.4 A/m²), which is non-motile and flagella. Furthermore, adaption of organisms at various conditions were made to crop robust exoelectrogens that are capable of enhancing the production of current density and facilitating the operation in extreme cultivating conditions of pH (Zhuang et al., 2010; Yoho et al., 2014; Liao et al., 2014), potential (Torres et al., 2009), temperature (Tkach et al., 2017) and polluted stream (oil, grease, or saline conditions) (Liao et al., 2014; Badalamenti et al., 2013;

Table 1

Examples of recalcitrant waste treated via Bioelectrochemical systems.

Substrate	Culture condition	Anode type/applied voltage	Reactor/other conditions	Influent COD (mg/L)	COD Removal efficiency (%)	CE (%)	Ref.
Dyes (methyl orange)	Municipal wastewater	Graphite brush/0.8 V cell	Membraneless single-chamber MEC/acetate as co-substrate & A TiO ₂ -coated photocathode	50 300	98 76	114 80	Liu et al. (2005b)
Pesticides (Atrazine)	Municipal wastewater	Graphite fiber brushes/−0.2 V vs SHE	Membraneless single-chamber MFC or MEC/ 1.0 g/L acetate as co-substrate	50 µg/L	74/MFCs 85/MECs		Pant et al. (2011)
Insecticide (DEET (N,N-Diethyl-meta-toluamide))	Municipal wastewater	Graphite fiber brushes/−0.2 V vs SHE	Membraneless single-chamber MFC or MEC/ 1.0 g/L acetate as co-substrate	50 µg/L	28/MFCs 39/MECs		Pant et al. (2011)
Furanic mixture ^a	Working MFC	Porous carbon felt /0.6 V cell	H-type MEC provided with cation exchange membrane/ No co-substrate	1200 mg/L	57 ± 10 based on SCOD	44–69	Biffinger et al. (2008)
Petroleum refinery wastewater	Mixed culture with refinery wastewater	Carbon cloth with carbon coating/318 mV cell (Batch)	Sandwich type MFC/ No co-substrate	95% oil content	84.4	2 ± 0.8	Yang et al. (2015)
Produced water from oil field processing	Anaerobic sludge acquired from the same treated wastewater produced from a desalination unit of oilfield	Graphite-varnished stainless steel mesh/330 mV cell	MXC (MEC or MFC)/salinity 40000 ppm	250–700 produced water	90%	0.2	Yi et al. (2009)

^aFuranic compounds mixture are: furfural, 5-hydroxymethyl furfural, syringic acid, 4-hydroxybenzoic acid and Vanillic acid.

Cheng et al., 2010). In this respect, exoelectrogens acclimatize in extreme conditions are useful to treat recalcitrant and persistent wastes such as pesticides (Mu et al., 2009; Werner et al., 2015), insecticide (Werner et al., 2015), heavy metals (Luo et al., 2014), furanic (Zeng et al., 2015), dyes (Hou et al., 2017), phenolic (Zeng et al., 2015) compounds, and wastewater products from petroleum (Srikanth et al., 2016) and natural gas (Stoll et al., 2015; Jain et al., 2017; Ghasemi Naraghi et al., 2015; Fakhru'l-Razi et al., 2009) operations. Recalcitrant wastes are usually incompatible with typical MXCs systems (Nevin et al., 2008; Zhen et al., 2017; Zhuang et al., 2010; Yoho et al., 2014; Liao et al., 2014; Torres et al., 2009; Tkach et al., 2017; Badalamenti et al., 2013; Cheng et al., 2010; Huang et al., 2011) thus acclimatizing bacteria in extreme conditions that simulate the natural existence of the treated effluent will produce biofilms that can bear with the acclimatization conditions and capable of utilizing recalcitrant wastes as substrates (Mohanakrishna et al., 2018). Examples of recalcitrant waste that have been treated via bioelectrochemical systems grown at extreme conditions are presented in Table 1. Products such as water from oil and natural gas processing [250–700 mg COD/L, 40 000 ppm salinity], furanic mixture [1200 mg COD/L], pesticides or insecticide [50 µg/L] and textile dyeing [50–300 mg COD/L] have been treated at reasonable rates when biofilms pre-acclimatized with the same pollutants as substrates in the mixed cultures of bacteria. Moreover, thermophilic and halophilic exoelectrogens have been cultivated from extreme natural conditions of pH, temperature and saline conditions such as seawater (Zhang et al., 2011).

The enrichment processes can be classified into three main categories: (1) substrate, (2) physical or (3) chemical. Substrates are feed sources for ARB, existing in the effluent and are converted or oxidized within the MXCs system. Substrate-related enrichment can be performed by feeding the inoculum with specific nutrients (Cercado-Quezada et al., 2010) or substrates (Yang et al., 2015) that enhance the growth of some species but not others. For examples, Zhang et al. (2013) found that adding iron

salts to mixed culture inoculum enrich the dominant *archaeobacterium* in the anode biofilm. Kadier et al. (2014) discussed substrates used in MECs, including simple to complex sources such as methanol-rich industrial molasses and waste streams from refinery, food processing, winery, dairy, swine, and domestic operations. Among these, use of complex substrates helps in obtaining a stable electrochemically active microbial community; whereas, use of simple substrates (e.g. acetate, or glucose) typically leads to microbial communities that degrade easily but improve the H₂ production rate.

Altering the physical conditions such as temperature (Patil et al., 2010), sonication (More and Ghangrekar, 2010), dissolved oxygen concentration (i.e. partial aerobic or anaerobic conditions) (Cercado-Quezada et al., 2010), electrode potential (Cereda et al., 2014; Torres et al., 2009; Nam et al., 2011), the application of intermittent energy input (Cho et al., 2019), electrode type (Yong et al., 2012), material doped on anode (Yasri and Nakhla, 2016), may promote species to tolerate the applied conditions. On the other hand, inhibiting some microorganisms can enable biological enrichment in MXCs by adding chemicals (Theivasanthi and Alagar, 2011) or modifying the chemical conditions of the anolyte chamber, e.g., pH (Patil et al., 2011), alkalinity (Yasri and Nakhla, 2017a, 2016), adding antibody or fungicides (Fenner et al., 2006). For instance, Babauta et al. (2012b) noted that the pH inside the anolyte compartment is an important factor affecting the intensity of current output, type of bacterial growth, and the total electron transfer process, which is also tied to proton transfer within the biofilm. They concluded: (1) pH varied within the biofilm during different growth phases, (2) pH is not always a limiting factor for a biofilm, and (3) biofilms respire in a unique internal environment, so that variation of pH and redox potential are associated only with the biofilm.

The pH dependency of the microbial community is due to the complex biological processes within bioelectrochemical systems (Patil et al., 2011), which has not been fully explained. Studies of ARB grown from municipal waste stream indicates

that the bioelectrocatalytic activity was suppressed at pH lower than 3.0 and higher than 11.0 (Patil et al., 2011). However, in principle the anode-biofilm generates hydronium cations; thus, the access of hydroxyl anion around biofilm will kinetically favor withdrawal of hydronium cations generated from the biofilm to accelerate the metabolic processes. Similarly, the withdrawal of hydronium cations explains the limitation of performance associated with proton transport out of the biofilm at lower pH (Zhao et al., 2014). This interpretation can justify the lower activity of biofilm at low pH (due to lack of OH⁻) but not at high pH (> 11.0), which may be due to the low tolerance of the biofilm to high pH environment. To the best of our knowledge, real-time monitoring of the pH gradient at the biotic/abiotic interface, and pH within the biofilm have not been investigated; however, such an approach using pH indicator during metabolism could be a subject of future studies, which may provide insight into these pH effects (Jin and Kirk, 2018).

4. Electrochemical concept of ARB

Understanding the principles and behavior of electrochemical interface reactions between biotic entities in biofilm, substrate utilization, and the surface of the electrode are important to continue advancements in MXC systems. Electrochemical studies for MXCs include voltammogram analysis of the interface reactions (Strycharz et al., 2011; Richter et al., 2009), the performance of the biofilm on electrodes during start-up (Paitier et al., 2017; Yasri and Nakhla, 2017b), growth (Choi and Sang, 2016; Rodrigues and Rosenbaum, 2014; Yoho et al., 2015) and starvation (Zhu et al., 2014), as well as studying of their kinetic parameters (Torres et al., 2010, 2008).

The relationship between current, ARB, and the electrode has been well established, indicating that the electron generation is a result of biocatalytic utilization of substrates and electron transfer via various EETs to the electrode (Lovley, 2008; Richter et al., 2009; Reguera et al., 2005). However, electrons flowing within MXCs containing both of biotic and abiotic phases will encounter resistance, leading to voltage losses within the cell. The total voltage loss is the sum of losses at the interface between the biofilm and the electrode (anode), the ohmic losses in the electrode, the biofilm, the electrolyte medium, the membrane, and the cathode overpotential (for H₂ production or O₂ reduction) (Zhen et al., 2017). Hence, an efficient electrochemical system would require minimization of potential losses; whereas, tuning of the potential losses at the biofilm/electrode interface is imperative.

Voltammetric studies of the anodic electrode–biofilm interface are not very common. Generally, the behavior of local current that is generated as a result of electron exchange at an electrode–solution interface is characterized via the Butler–Volmer equation, which describes the current–voltage response in electrochemical systems (Torres et al., 2008). However, in the case of biofilm grown on an anode, both biotic and abiotic processes must be considered on the electrode surface, including the metabolism associated with both conductive and nonconductive microorganisms (Torres et al., 2008). Thus, the kinetic behavior of this type of anodes, which is attributed to substrate utilization via both metabolism and the electron transfer process, will deviate from the abiotic system and hence will also deviate from the conventional Butler–Volmer equation.

Various kinetic models have been proposed to evaluate the kinetic responses of substrate utilization (biotic) and the electrochemical electron transfer (abiotic) processes. Among these, two important modified versions of Monod model will be discussed hereafter; (1) the Butler–Volmer–Monod model that describes the relationship between overpotentials of the metabolized substrate (Hamelers et al., 2011), and (2) Monod–Nernst model that

describes the rate of substrate metabolism and its link with species concentration and the redox potential (Torres et al., 2008).

In developing Butler–Volmer–Monod model, Hamelers et al. (2011) proposed a two-step process: (1) the first step involves intercellular biochemical utilization reaction that promotes conversion of organic and electron generation, where the rate depends on enzyme kinetics that is described via the Monod equation; and (2) the second step involves the extracellular electron transfer from biofilm to the electrode and is described via the conventional electrochemical electron transfer kinetics of the Butler–Volmer equation. The efficiency of process conversion is typically lower than 100% due to losses at the bio-anode and can be quantified via the anode polarization curve (i.e. potential as a function of the current density). Based on this argument, Hamelers et al. (2011) developed a basic Butler–Volmer–Monod model (Eq. (12)) that described the kinetics of microbial metabolism based on a combination of enzyme kinetics (Monod) and electrochemical kinetics (Butler–Volmer). This polarization model was found to fit the measured anode-biofilm polarization curves, and it was found that the apparent Monod constant of an anode-biofilm is dependent on the anode overpotential (Hamelers et al., 2011):

$$j = j_{lim} \left(\frac{1 - e^{\frac{F}{RT} \eta_{anode}}}{K_M \cdot e^{-(1-\alpha) \frac{F}{RT} \eta_{anode}} + K_2 \cdot e^{\frac{F}{RT} \eta_{anode}} + \left(\frac{K_M}{S} + 1\right)} \right) \quad (12)$$

where F is Faraday's constant, R is the universal gas constant, T is the operating temperature, η_{anode} the anode overpotential, j is current density (A m⁻²), j_{lim} is the limiting current density (Am⁻²), α is the charge transfer coefficient of the anode reaction, K_M is the substrate affinity constant (describing the effect of substrate on the biochemical conversion), and S is the substrate concentration (Zhao et al., 2014; Hamelers et al., 2011).

Based on the Butler–Volmer–Monod model, Zhao et al. (2014) developed a mathematical model for an MFC system with air cathode at various period of growth levels of the anode-biofilm. They were able to model the measured polarization curves and provide insights into the limiting physical, chemical and electrochemical phenomena and their effects on cell performance. For example, the MFC data demonstrated that the performance of the cell is primarily limited by cathode electrochemical kinetics, and that anode biofilms with longer growth time will lead to higher limiting current and less polarization losses under the same operating current. They were also able to simulate the overpotentials developed from ohmic, cathodic, and anodic losses. They noted that the overall ohmic overpotentials were relatively small, and that the cathode overpotential is dominant at both low and high current density region, and for short (110 h) and long (160.5 h) growth periods of anode-biofilm. Compared to the electrode overpotential, the overall ohmic overpotentials are relatively small. Thus, the limitations of performance are bounded to cathodic electrocatalytic performance and proton transport from the biofilm. Thus, they concluded that the limitation can be mitigated by using a high buffer concentration (Zhao et al., 2014).

The Nernst–Monod model considers the biofilm as an electron generator via substrate utilization and the anode is the final electron acceptor (Torres et al., 2008). The Nernst equation is commonly utilized in electrochemistry to describe the relationship between species concentration and the redox potential at the electrode–solution interface. In biology, however, the Monod model is used to describe the kinetics of biological growth and its dependence on the concentration of substrates (Wang et al., 2010a). Kato Marcus et al. (2007) combined the two to propose the Nernst–Monod model (Eq. (13)), which describes the kinetic behavior of bacteria (current production) under an electrochemical potential.

$$j = j_{lim} \left(\frac{1}{1 + \exp\left[-\frac{F}{RT}(E - E_K A)\right]} \right) \quad (13)$$

where: j = current density (A m^{-2}); j_{lim} = limiting current density (A m^{-2}), which is the maximum current density reached at the applied conditions; R = ideal gas constant; F = Faraday constant; T = absolute temperature (303.15 K); and E_{KA} = catalytic potential at which $j = \frac{1}{2} j_{\text{lim}}$.

Ideally, the voltammetric response of ARB grown at anode interface should follow the sigmoidal Nernst–Monod model (Fig. 4A) (Torres et al., 2008; Kato Marcus et al., 2007). However, the experimental responses of biofilm anode vary from ideality (Fig. 4B) (Torres et al., 2010), which will be discussed later after describing some of the experimental setups in the following section.

Generally, to obtain an ideal voltammogram, a steady state condition has to be reached and a slow potential variation with a slow scan rate is usually applied. At this slow variation in potential condition at the interface, the electrocatalytic activity is independent of the scan direction and the ARB reach a stable metabolic status (Marsili et al., 2008b). Hence, in a bioelectrocatalytic investigation, a linear-scan cyclic voltammetry (LSCV) with a slow scan rate of about 1 mVs^{-1} , is considered a powerful technique. At this LSCV condition, the catalytic response (current) can be measured at a steady-state condition of ARB as a function of the anode potential (Torres et al., 2010).

The prime postulation of the Nernst–Monod model is that the biofilm anode catalyzes the interfacial electron exchange process and hence controls the kinetics as well as the current generation. Thus, the lower the potential loss at the biofilm/electrolyte interface during current flow, the higher the efficiency of the electrochemical system. The potential loss at both the biofilm/electrolyte and the electrode/biofilm interfaces are anode potential losses (APL). Thus, from an electrochemical point of view, the evaluation of APL is of prime importance for the development of MXC systems. However, the nature of APL, its relationship to anode biofilm performance and the method of calculation are not clearly defined in the literature.

The APL (η_{anode}) is associated with the electrocatalytic features of bio-active sites, and ideally, the APL should be small so that the electrode potential is as close as possible to the redox potential of the substrate being oxidized. That is, the amount of potential (energy) losses by bacteria during utilization of substrate and EET as well as electron transfer to anode surface should be as low as possible for the process to be efficient. Torres et al. (2010) defined the APL as the losses in potential due to extracellular electron transfer, and electron transfer to the electrode surface, indicating that the lower the APL the higher the transfer of energy, and consequently a more efficient process. Thus, from an electrochemical point of view, the main goal during process optimization of MXCs is to reduce potential loss (i.e. close to the redox potential of the substrate being utilized) and to increase the biologically active surface area to obtain a maximum current output. Whereas, the practical goal in MXCs is to find a bioelectrocatalytic system that can utilize specific organic (or waste) to produce high current density at low cell potential (Torres et al., 2010; Grattieri and Minteer, 2018).

Fig. 4B represents an ideal experimental LSCV curve (scan rate of 1 mVs^{-1}) (Torres et al., 2010) for a system containing ARB biofilm that utilizes acetate as substrate and produces a limiting current density (j_{lim}) of about 0.15 A m^{-2} . The obtained steady-state conditions can be distinguished from the similarity in the forward and backward scan, whereas, the APL η_{anode} can be measured by calculating E_{donor} : for acetate as electrons donor in this case, vs. SHE. When the anode potential is equal to E_{donor} the biofilm will not have enough energy to transfer electrons to the electrode, hence no catalytic current will be observed, i.e. $j = 0$. Increasing the anode potential to a more positive value, the metabolic activity of ARB will increase, and the electron-exchange between biofilm and electrode will take place, hence the utilization process of the substrate will catalyze the current to reach

a maximum value (limiting current). The interfacial impedance may restrict the current flow and APL will take place. Thus, the potential window between the E_{donor} and the point where the current starts to increase (onset potential) can be considered the value of APL (mV).

The Nernst–Monod equation is a kinetic description of a bioelectrocatalytic system that describes the ideal j - E trend for a pure culture and electron transfer pathways (Lee, 2018). Anode-biofilms grown from mixed culture media may not obey the Nernst–Monod model. Although the j - E voltammogram may show a sigmoidal shape, the curve may not fit the Nernst–Monod model, and the interpretation of the kinetic data becomes complicated (Yasri and Nakhla, 2016). The deviation of experimental data from the Nernst–Monod model can arise for several different reasons, including the composition of electrode material, the type of the culture grown on the electrode and the existence of several EET pathways, which may occur at various electrode potentials. For example, in a previous work, we observed a range of different sigmoidal j - E voltammograms grown from the same inoculum with the same substrate (acetate), but with different electrode compositions. A carbon anode doped with magnetite (Fe_3O_4) produced sigmoidal-shaped cyclic voltammogram with a series of convex shapes indicating multiple EET pathways (Fig. 5A). Fitting the voltammogram (red line) with the Nernst–Monod model (black line) shows a significant deviation (Yasri and Nakhla, 2016; Yoho et al., 2014, 2015). However, the voltammogram can be characterized using the derivative cyclic voltammogram (DCV) which revealed several steps from the series of convex sections (red curves in Fig. 5B) (Yoho et al., 2015), indicating multiple EET pathways. These convex sections indicate a series of electron transfer processes occurring at different potentials. Multiple derivatives of the Nernst–Monod equation fit well with each convex curve in the forward section of the DCV (dashed lines, Fig. 5B) (Yasri and Nakhla, 2016).

The derivative of the sigmoidal curve is described by a convex shape. Thus, in the case of Nernst–Monod equation, the variation of dj/dE (A V^{-1}) versus anode potential (V) is convex and the value of E_{KA} can be found from the position of the peak. Applying this approach to an experimentally measured DCV by fitting the Nernst–Monod equation at each convex point will help deconvolute the curve and distinguish multiple EET pathways (Yasri and Nakhla, 2016; Yoho et al., 2014, 2015). The derivative of an experimental LSCV curve will generate, during the forward and backward scans, multiple convex and concave curves, that may be associated with multiple EET pathways (Yasri and Nakhla, 2016), i.e. each pair of opposing convex/concave couples corresponds to an EET pathway. In our earlier investigation (Yasri and Nakhla, 2016), as well as the study performed by Yoho et al. (2015), the forward scan (oxidation) of the DCVs were deconvoluted by fitting with multi Nernst–Monod derivatives to provide an overall electron exchange response. In these two studies, the midpoint potential (E_{KA} , V) of each convex curve in the forward DCV scan were fitted to the Nernst–Monod equation using the corresponding E_{KA} value and adjusting the current and assuming $n = 1$. Considering $n = 1$, we can infer that each Nernst–Monod fit will correspond to an EET pathway with one electron transfer process. Thus, by fitting multiple curves, the existence of multiple EET pathways can be established, and the differences in the potential may be due to the variation in ARB type or responses to the surrounding conditions (Yasri and Nakhla, 2016; Yoho et al., 2015).

Discussion related to the variation of pathway-potentials can be derived from the aforementioned APL, that is the higher the potential windows between the redox potential of the substrate being oxidized and the associated potential (i.e. the E_{KA} of the convex section), the higher the APL. Consider the DCV in Fig. 5B,

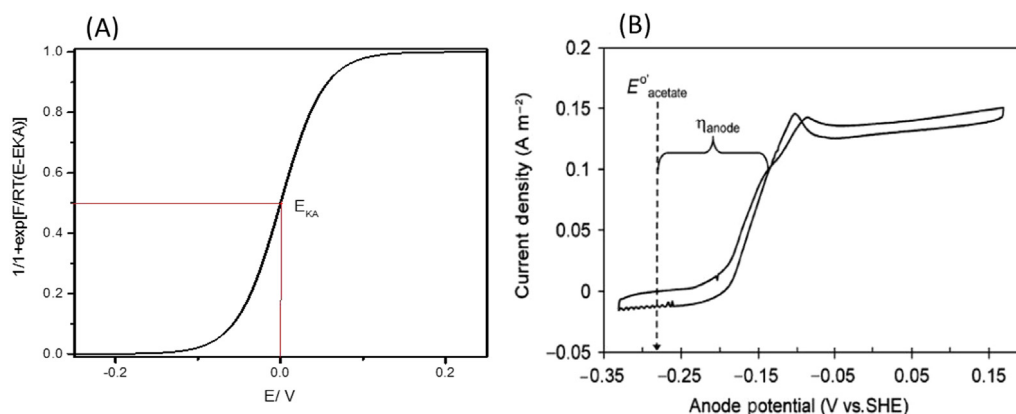


Fig. 4. (A) Ideal plot of the Nernst–Monod model for $E_{KA} = 0$ V and $T = 30$ °C, and (B) Low-scan cycle voltammetry of ARB biofilm with acetate substrate. Reproduced with permission from Torres et al. (2010).

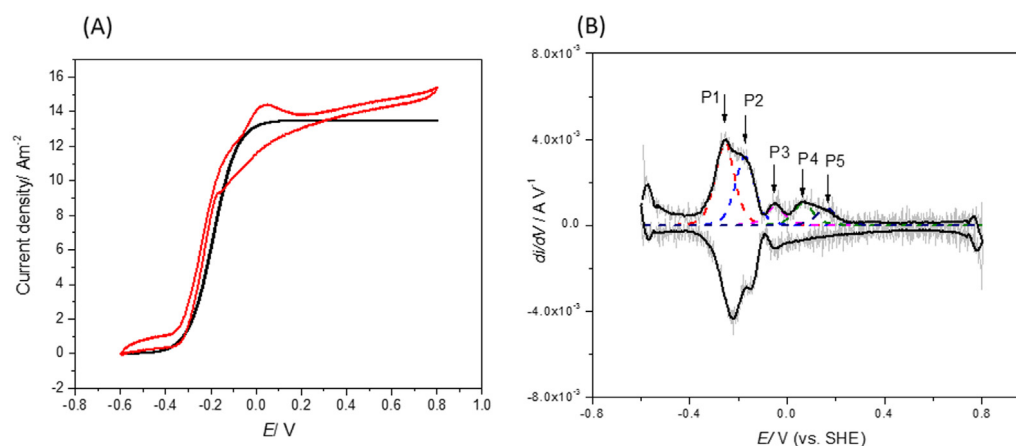


Fig. 5. (A) Cyclic voltammogram (CV) of mixed culture inoculum (red line) on graphite anode doped with trace magnetite (Fe_3O_4), recorded at 1 mV s^{-1} after day 8 of biofilm growth. Adapted from Yasri and Nakhla (2016), the black curve shows the Nernst–Monod model fitted to experimental data. (B) Derivative CVs suggest multiple electron transport pathways identified by arrows; dashed lines represent the Nernst–Monod fit of each pathway, reproduced with permission from Yasri and Nakhla (2016). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

recorded from a mixed culture biofilm fed with acetate and grown on an activated carbon electrode doped with Fe_3O_4 (Yasri and Nakhla, 2016). In this case, the DCV shows a series of four convex sections each at a different potential (E_{KA}), corresponding to four EET pathways or biocatalytic potentials. A higher E_{KA} value indicates a larger energy consumption for substrate reaction and vice versa. Thus, pathway 1 in our example (Fig. 5B) corresponds to a lower biocatalytic potential and lower APL (Torres et al., 2010; Yasri and Nakhla, 2016), which corresponds to a higher catalytic activity and efficiency for utilization of the substrate (Torres et al., 2010; Yasri and Gunasekaran, 2017). In this respect, further investigations are needed to improve the catalytic activity of the system and reduce the internal energy loss. Detailed voltammetric studies in the absence of substrate (starvation condition) (Zhu et al., 2014) are needed to investigate the underlying electron transfer mechanism (Strycharz et al., 2011). Thus, it is essential to perform basic studies on electrodes with biofilms containing a single microbial strain, using voltammetric methods to gain a fundamental understanding of the behavior of anode biofilms.

5. Anode system

Among the main factors affecting the performance of MXC systems are anode materials and their configuration (Baudler et al., 2015; Wagner et al., 2012; He et al., 2015). Physical confinement or localization of entities such as enzymes, cellular

organelles, animal and plant cells without destroying their ability to functionalize is a difficult task. This confinement is known as biological immobilization, which is performed to limit free transportations of biota in the surrounding environment while retaining their catalytic activity.

Conductive carbonaceous materials that satisfy the aforementioned physical properties are favored for ARB immobilization on the anode (Logan et al., 2008; Logan, 2010a), including carbon cloth, carbon fibers, carbon felt, carbon rods, carbon paper, carbonized cardboard (Chen et al., 2012; Massazza et al., 2015) and granular activated carbon (GAC) (Logan, 2009b) (See Fig. 6).

Among these, carbon brush (Logan et al., 2007) and carbonized cardboard (Chen et al., 2012) have high microbial electrical efficiency, which may be due to their high surface area combined with low ohmic resistance. In this respect, increasing surface area by using a packed bed anolyte compartment filled with GAC may increase the volume of the biofilm and hence the number of ARB reactive sites. However, although some reports indicate an improvement in MXCs performance when using GAC as a three-dimensional (3-D) electrode (Pant et al., 2010; Wang et al., 2010b; Liu et al., 2014; Wang et al., 2019b), some reports indicate that the performance was not significantly better than a similarly designed MEC reactor using sandwich type electrodes (anode and cathode are only separated by a membrane (Yasri and Nakhla, 2017a)). For example, the hydrogen yield ($3.6 \text{ mol H}_2/\text{mol acetate}$), hydrogen recovery efficiency (90%), and COD

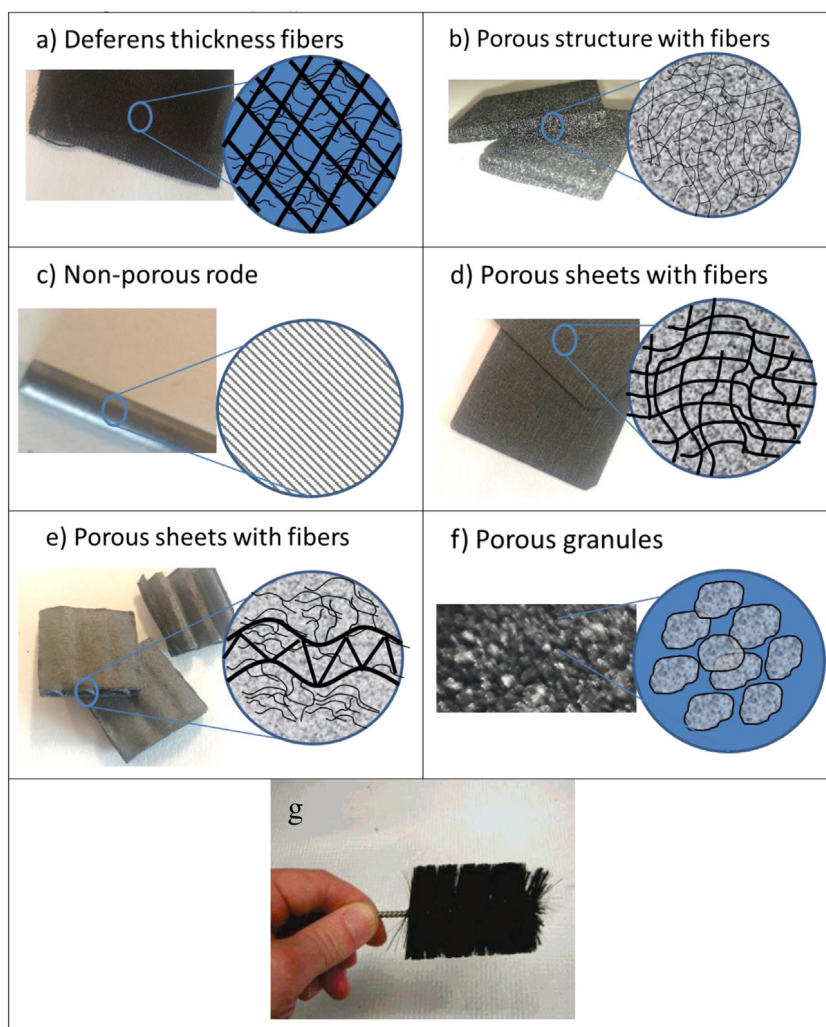


Fig. 6. Carbonaceous material used as ARB anode materials; (a) carbon cloth, (b) carbon felt, (c) carbon rods, (d) carbon paper, (e) carbonized cardboard, (h) granular carbon, and (g) carbon brush (a–h images taken by the author, whereas; g reproduced with permission from Logan et al. (2007)).

removal rate (80%) for an MEC using a packed GAC (operating as a 3-D electrode) bed between the electrodes, were similar to those of a conventional sandwich-type MEC (Yasri and Nakhla, 2017a). This finding emphasizes the importance of anode structure, and cell design, since a packed bed of GAC is used to increase the surface area of anode, the GAC particles are not connected effectively to the electrode (current collector) or have loose connections which enables current disconnection and hence current destruction from the freshly formed biofilm on the surface of the GAC. On the other hand, carbon electrode materials such as carbonized cardboard or carbon brushes that have high surface area accompanied with macrostructure, are typically well connected to the core of the electrode (or current collector) and are able to establish rapid growth of biofilm with immediate connection to the electrode body with less destruction. For comparison, the average current density reported in literature using a carbon anode with an acetate substrate can range between 5–15 A m^{-2} (Baudler et al., 2015), however, with one layer of carbonized corrugated cardboard Chen et al. achieved a much higher current density (Chen et al., 2012) of 70 A m^{-2} . With three and six corrugated cardboard layers, the current density increased further to 200 A m^{-2} and 390 A m^{-2} , respectively.

Other factors that may affect the immobilization of microorganisms on the anode such as cell design, applied voltage, anode charge transfer resistance, electrolyte conductivity (ion transfer

through the electrolyte), pH, substrate types, and electrode porosity, have also been extensively studied (Wang et al., 2010b). Of these factors, the electrode arrangement is important in designing an MXC system and reducing the ohmic voltage drop (Li et al., 2014). For example, Liang et al. (2011) separately placed two anodes, either on one side or both sides of a cathode within a membraneless MEC. They found that the MEC with anodes on both sides of the cathode improved current and H_2 production rate by 72% and 118%, to yield 621 A m^{-3} and 5.6 $\text{m}^3 \text{H}_2/\text{m}^3$ per day (based on the anode volume), respectively, as compared with an MEC that contains only one anode and one cathode whose yield is 360 A m^{-3} and 2.55 $\text{m}^3 \text{H}_2/\text{m}^3$ per day.

Research on the effect of applied voltage on the performance of MXCs has been inconclusive. Commault et al. (2013) reported the effect of anode potential on the type of *Geobacter* strain grown in MECs. Zhu et al. (2014) proposed that the same strains will regulate their EET pathways to acclimatise to the anode potentials. To understand the effect of potential, electrochemical investigations should be performed on biofilms grown from various pure cultures. For example, voltammograms should be obtained and analyzed to determine how the applied potential affects the catalytic performance of various bioelectrochemical systems (see also the discussion in Section 4, Electrochemical concept of ARB).

Anode pre-treatment is important as it can be used to alter the porosity, and interfacial conductivity of the anode material, as

well as to dope particles that can encourage the growth of biofilm on the anode (Yasri and Nakhla, 2016; Paul et al., 2018). Considering the porosity of the anode surface (Sleutels et al., 2011, 2009a; Dhand et al., 2014), an increase of porosity typically results in an increase in the number of biocatalytic sites within the pores and hence increasing the electrochemically active surface area (Dhand et al., 2014). Moreover, various approaches have been used for chemical modification of the surface of an electrode to attract microbial growth on an electrode surface. For example, Cheng and Logan (2007) reported that treating an anode surface with ammonia at a high temperature (700 °C, 5% ammonia gas for one hour) improved the adhesion of microorganisms to the anode and resulted in faster MEC start-up for H₂ production (Cheng and Logan, 2007). Several chemical pre-treatments of carbon including: ammonia, trace metals, phosphate etc. have resulted in increased attachment of microorganisms on the anode and enhancing the start-up of the system (Zhang et al., 2014a). On the other hand, increasing catalytic activity of the interface is thought to occur via variation of the conductivity of the electrode interface, which has been achieved by modifying the anode surface with particles such as iron (Xu et al., 2012) or conductive polymer (Liu et al., 2015). Much work has been done using conductive polymers or nanocomposites for enhancing the power output of ARB. For example, a conductive poly(3,4-ethylenedioxythiophene (PEDOT) film showed superior biocatalytic performance when coated on carbon felt (CF) anode and inoculated in palm oil mill waste to catalyze acetate metabolism (Kang et al., 2015). However, the role of the conductivity of the electrode surface in enhancing the growth and the performance of anode biofilms is unclear. For example, carbon electrodes derived from natural fibers of corn stem of 94 Ω interfacial charge transfer resistance (Karthikeyan et al., 2015) outperformed anodes coated with carbon nanotubes-textile of lower interfacial charge transfer resistance (30 Ω) (Xie et al., 2011), or even lower resistance of graphene nanoparticles (22 Ω) (Xie et al., 2012). Almost all studies reported in the literature suggest that microbes use conductive minerals as conduits of electrons, resulting in efficient electron transfer behavior and better catabolism (Kato et al., 2012). Thus, considering the electrical conductivity of the electrode current feeder, since metals are highly conductive and rigid they should outperform carbon materials for ARB growth. However, although silver is more conductive than copper, Baudler et al. (2015) found that, using acetate as substrate, copper is comparable to the benchmark graphite but outperformed silver, gold, nickel, cobalt, titanium and stainless steel, with a maximum current density of 15 Am⁻². Thus, in light of the aforementioned examples, it is unclear if the attraction of biota to the anode is due to a natural affinity or due to the conductivity of the anode material. Thus, the question, “how critical is anode conductivity to ARB growth?” remains unanswered.

Similarly, electrolyte conductivity (ion transfer through the electrolyte) is also considered to play an important role in reducing the ohmic voltage drop, and is also thought to affect the performance of ARB by inducing cultivation, stimulating catabolism, and consequently improving the current density (Zhao et al., 2010; Baudler et al., 2015; Kang et al., 2015; Kato et al., 2012; Li et al., 2011; Tang et al., 2015; Gnana kumar et al., 2014; Luckarift et al., 2012; Rousseau et al., 2013; Lacroix et al., 2014). However, research on the effect of electrolyte conductivity on the performance of the bioelectrocatalytic system has also been inconclusive. For example, Vereea et al. (2014) found that the rate of H₂ production in a MEC system increased from 0.13 to 0.82 m³H₂/m³ per day when the electrolyte conductivity increased from 7.5 mS/cm to 15 mS/cm, which was attributed to the increased electron shuttling between the biofilm and the electrode surface (Rousseau et al., 2013; Lacroix et al., 2014; Vereea et al., 2014). For verification, the electron shuttling is an electrode interface process that is taking place between biofilm and abiotic

electrode and depend on the electrode matrix as well as the type of biofilm. However, apart from this one paper by Vereea et al. (2014), we have found no other evidence to attribute the change in efficiency with the electrolyte conductivity to the enhancement of electron shuttling between the biofilm and the electrode surface. Indeed contradictory results were obtained by Liu et al. (2011), who found that the initial current production from acclimatized biofilm using wastewater took longer when conductive phosphate buffered saline (PBS) was added to the media. The acclimatisation period of biofilm increased when the concentration of conductive PBS was increased from 149 h at 25 mM PBS, to 251 h at 50 mM PBS, and 526 h at 100 mM PBS. Hence, it may be concluded that conductive media may contain species that enhance or slow the microbial metabolism and thus affect the ARB performance, rather than simply contribute to the conductivity. Metal sulfides, especially those containing Fe, Ca, K, or Mg, which are not highly conductive but play a major role in improving the biological activity, are found at the active centers of a wide variety of redox and catalytic species (Lapinsonnière et al., 2012; Bertini et al., 1994; Picot et al., 2011). The presence of these compounds may enhance the biotic performance independently of their effect on electrolyte conductivity. These species include simple soluble electron-transfer mediators (e.g., Fe(II)/(III), sulfate/sulfide redox couple (Schroder, 2007), and Ca²⁺ ions (Fitzgerald et al., 2013)), membrane-bound components of electron-transfer chains, and some complex metalloenzymes (Ghasemi et al., 2013; Lapinsonnière et al., 2012; Bertini et al., 1994; Schippers, 2004; Amend et al., 2004; Weber et al., 2006; Lovley and Phillips, 1986). Considering the fact, however, that the interfacial electron transfer process involves neutral and indistinguishable biological species, the biocatalytic interaction remains inscrutable. The origin of interfacial interaction requires further investigation.

6. Cathode system

Generally, reduction of dissolved oxygen in MFC and hydrogen production in MEC occur at the cathode surface, which is located in the catholyte chamber; however, tuning both the medium and the electrochemical conditions are required to favor the desired reaction. For example, considering the case that both the oxygen reduction and hydrogen evolution reactions can occur on carbon electrodes, the potential for H₂ evolution on a graphite electrode reaches −420 mV vs SHE (Logan et al., 2008) in pH 7 PBS, and the potential for dissolved O₂ reduction ranges between −56 mV to −556 mV vs SHE (Nissim and Compton, 2013). Based on these potentials, simultaneous O₂ reduction and H₂ evolution can take place, but controlling the conditions of the media will favor one reaction over another. Changing the conditions can be performed by controlling the O₂ concentration in the catholyte chamber. Air saturation in the catholyte chamber will favor O₂ reduction at a lower potential value; whereas, the elimination of O₂ will allow H₂ production, provided there is sufficient overpotential at the cathode surface.

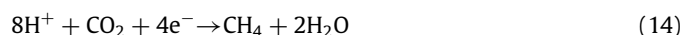
The voltage losses due to ohmic resistance of the electrolyte and interfacial electron transfer at the cathode (for O₂ reduction or H₂ production) contribute significantly to the potential loss in MXC systems. To reduce the reaction overpotential, Pt is often used as an electrocatalyst doped on carbon, which is commercially available as a powder and the electrode can be prepared by mixing with a binder (Cheng et al., 2006). Although Pt is the most active electrocatalyst for reduction of hydrogen or oxygen under the conditions used in MXCs, the possibility of deactivation by constituents present in wastewater (such as sulfide) should also be taken into account. Alternatives to costly Pt considered include other metals, alloys, non-metallic doped carbon materials, or a reducing biofilm cathode (Rozendal et al., 2008).

Kundu et al. (2013) reviewed the types of cathode material, catalysts and modifications that are suitable for the H_2 generation in MEC. Ben Liew et al. (2014) reviewed non-Pt abiotic catalysts for oxygen reduction reaction in MFCs. Moreover, Harnisch and Schroder (2010), analyzed the different cathode tasks in MXC systems and discussed the catalytic processes. In an MEC system, among the many materials used as cathodes, stainless steel, Ni alloys, and MoS_2 -doped or Pd-doped nanoparticles on carbon cathodes, have been found to be very effective for H_2 evolution (Hou et al., 2014; Tenca et al., 2013). On the other hand, among the carbon supported electrocatalysts materials for reduction of O_2 in MFC systems manganese oxide (Li et al., 2010), composite compounds (e.g., metal macrocyclic compounds such as iron phthalocyanines or cobalt tetramethoxyphenylporphyrins) (Antolini, 2015), nitrogen-doped carbon materials (Yang et al., 2016; Singh et al., 2019), and electroconductive polymers (Gnana kumar et al., 2014) have proven to have good electrocatalytic activity.

Recently, there has been increasing interest in developing metal-free cathodes to reduce cost and to minimize the environmental impact of MXCs (Fogel and Limson, 2016). Metal electrocatalyst materials may leach and contaminate the media. However, composites have been developed using carbon materials doped with nitrogen (Zhang et al., 2014b), phosphate (Munoz et al., 2010), sulfur, or their mixtures (Ghasemi et al., 2013; Zheng et al., 2014) and used as cathode material for both of O_2 reduction and H_2 production. The concept of doping foreign non-metallic species on carbon substrate changes the electron density on the substrate via the introduced doped moieties that donate or withdraw electrons. The heteroatoms in the graphitic structure thus enhance the catalytic activity of the carbon by favoring adsorption/desorption processes of reactants and products, and thus enhance some redox reactions at the electrode interface (Liang et al., 2012; Abrol et al., 1988).

7. Separator and membrane

Some reducing bacteria favorably grow on the cathode, such that a membrane-free MXC may lead to side reactions and reduce the efficiency of the system. The incorporation of a membrane in the case of MEC improves the purity of hydrogen and in MFC prevents mixing anaerobic biota at the anode with the aerobic conditions at the cathode. In MECs the membrane helps prevent the growth of electron acceptor bacteria present in the inoculated mixed culture and isolates the electron donor species in the anolyte chamber. Yin et al. (2016) demonstrated that *Geobacter* (oxidizing bacteria) and *Methanosarcina* (reducing bacteria) could coexist in the biofilm or grow at the anode and cathode, respectively. The coexistence of reducing bacteria in MECs may lead to the production of products such as methane (Eq. (14) (Bajracharya et al., 2015; Jadhav et al., 2019)) and will impact the efficiency of the cell.



The membrane used in MXCs is normally either a cation exchange membrane (CEM) or an anion exchange membrane (AEM) (Logan, 2008). In CEM, cations are free to exchange with other cations diffusing into the membrane, or to migrate under the influence of an applied electric field. Thus, the use of CEMs allows transport of H^+ and other cations such as Na^+ , K^+ , NH_4^+ , Ca^{2+} , and Mg^{2+} toward the catholyte chamber. Whereas, in AEM the exchange groups are typically anions that permits transport of anions such as OH^- , PO_4^{3-} , Cl^- , SO_4^{2-} , and anionic complexes (Chaudhary et al., 2000) toward the anolyte chamber.

Studies performed by Kim et al. (2007) compared the power generation using different cation, anion, and ultrafiltration membranes in a two-chambered MFC, demonstrated the effectiveness of many types of membranes. However, the use of an AEM produced the largest power density (up to 610 mW/m^2) and Coulombic efficiency (72%). In a similar investigation, Sleutels et al. (2009b) showed a better hydrogen recovery of a two-compartment MEC system when separated by an AEM ($2.1 \text{ m}^3 \text{ H}_2 \text{ m}^{-3} \text{ d}^{-1}$) as compared with the CEM ($0.4 \text{ m}^3 \text{ H}_2 \text{ m}^{-3} \text{ d}^{-1}$). The investigation was based on potential losses and partial resistances for acetate utilization for a cell voltage of 1 V. They attributed the better performance to lower internal resistance with AEM ($192 \text{ m}\Omega \text{ m}^2$) compared to the CEM configuration ($435 \text{ m}\Omega \text{ m}^2$).

However, the typical pH of wastewater and the inoculating medium of MXCs is about neutral ($\text{pH} \sim 7$) and thus the proton concentration, as well as hydroxide ions, is about 10^{-7} mM (with a maximum of about 10^{-4} mM (Rozendal et al., 2007)), which is much less than the concentration of other cation species in wastewater (typically 4–5 orders of magnitude higher). Thus, by using CEM in MXCs electro dialysis will occur via the transport of cation species present in the anolyte chamber as well as protons through the membrane maintaining at the same time the charge balance within the cell. However, when using an AEM, hydroxide (OH^-) anions generated at the cathode (Eq. (15)) will be transported through the membrane into the anolyte chamber to neutralize the generated protons there. Thus, the incorporation of an AEM will prevent the transport of metal cations to the catholyte chamber (Rozendal et al., 2007) and therefore prevent the change in the composition of the medium surrounding the bacteria in the anolyte chamber, and stabilize the environment for bacteria growth.



Another phenomenon is the pH gradient that can form when a membrane is incorporated between the electrodes in an electrochemical cell. The transport of the charged species to the corresponding electrode will create a pH gradient between the catholyte and anolyte chambers that will negatively impact the MXC performance by increasing the potential loss (Rozendal et al., 2007). Indeed, Rozendal et al. found that a large part of potential losses in an MEC system was associated with a pH gradient across the membrane and that the pH gradient was higher in a CEM incorporated system ($\Delta \text{pH} = 6.4$) as compare with AEM ($\Delta \text{pH} = 4.4$). They link the greater pH gradients in a system incorporating CEM to the possibility of higher transportation of cation species along with hydronium cations toward the cathode chamber. In practice, pH differences across the membrane must be minimized to avoid voltage losses. According to the Nernst equation, each unit difference in pH between the two chambers in MXC will create a reduction of 0.07 V in the total cell voltage according to Eq. (16) (Logan et al., 2008).

$$\Delta \phi_{\text{pH}} = 2.303 \frac{RT}{F} (\text{pH}_{\text{cathode}} - \text{pH}_{\text{anode}}) \quad (16)$$

Thus, the use of an AEM will promote a stable environment for bacteria to grow in the anolyte chamber and reduce acclimatization of reducing bacteria on cathode to ensure pure H_2 production. However, in terms of technical implementation, the presence of a membrane separator creates challenges with regards to cell construction and voltage losses due to increase in internal resistance, lower mass transfer, the possibility of membrane fouling, and pH gradients across the membrane (Rozendal et al., 2007).

An MFC using a membrane electrode assembly (MEA), in which the cathode is directly bonded to the membrane, has also been reported (Choi et al., 2013). Choi et al. investigated the

utilization of MEA in an MFC system using a non-woven paper fabric filter as an alternative to a membrane separator. In this case, the MEA system produced comparable cell voltage (545 mV) and maximum power density (1027 mW/m^3) to those obtained from MFCs with a CEM (551 mV, 609 mW/m^3). However, the ability of the membrane, in this case, to act as a conductive cathode and enable cathodic bacteria to grow on the membrane prevented further development of this system (Choi and Sang, 2016). For more details on the membrane types used in MFCs readers are referred to Logan (2008), who categorized the types of membrane used along with various specifications and industrial manufacturers.

8. Bioelectrocatalytic implementation in practical applications

Although the origin of the bioelectrocatalytic concept goes back to 1911, the utilization of the electrical activity of microorganisms to generate energy in biotechnology has not been exploited in a systematic manner until very recently (Bennetto, 1990). In 1911, Potter (1911) demonstrated that the electrophysiology process can accompany the respiration of biota during biochemical utilization of substrate. Since then, research has progressed to elucidate the chemistry and biochemistry of such phenomena and advanced our understanding of microbial electricity generation. However, this technology has been adopted only modestly at the industrial scale, and much work remains to be done for this technology to be comparable to other renewable energy sources such as solar, wind, and hydropower (García-Gusano et al., 2017). Nevertheless, some reports outline the practicality of implementing the technique for direct/or indirect power generation from wastewater at volumes exceeding those in lab scales for MEC (Zhen et al., 2017; Cusick et al., 2011; Heidrich et al., 2013, 2014; Logan, 2010b) and MFC (Liang et al., 2018; Walter et al., 2018; Jiang et al., 2011; Ge et al., 2015; Zhuang et al., 2012; Dong et al., 2015; Ge and He, 2016; Martinucci et al., 2015).

In his 2010 review, Logan (2010b) discussed the advances and future challenges to bring the MEC technologies out of the laboratory for bioenergy production at larger scales. He described new developments in electrode types, the impact of membranes and separators on the performance, and the results from several pilot-scale tests. The substantial problems associated with scale-up of microbial electrochemical systems are varied and mostly depend on energy losses in the system due to poor performance of the anodic half-cell, variations in the substrates, the high ohmic drop between electrodes, the growth of methanogenic bacteria on the cathode (in the case of membraneless MEC) as well as poor cell designs. Hereafter, we provide some examples from the literature that appeared after Logan's review (Schroder, 2007), which may help in improving the design for better performance. In 2014, Heidrich et al. (2014) used a 100-L MEC system operating at low temperatures with domestic wastewater for 12 months. Although the system was robust, and durable with continuous hydrogen production (an average of 0.6 L/day) over a year of operation, it was not self-sufficient, i.e., only about 48.7% of the electrical energy input was recovered, and COD removal was below expectation. In this, and other similar MECs (Heidrich et al., 2013), the cell design experiences large overpotential, which is correlated to ohmic loss and low efficiency.

To overcome some of the problems, Hahn et al. (2017) modified the cassette-style design to reduce the internal resistance and increase the reactor size by a factor of 16 using six 1 m^2 anodes and municipal wastewater as effluent. This new design outperformed the earlier reactor, and produced about 0.2 L of 93% pure H_2 per day more than the older design with an average COD removal of about 63.5%. Similarly, to reduce the reactor resistance, Cusick et al. (2011) scaled-up an MEC system without

using by installing a stack of 144 electrode-pairs in 24 modules with about 1000 L capacity to treat winery wastewater. Although this type of waste is adequately enriched with organic volatile fatty acids, the efficiency loss was mainly correlated to the growth of a methanogenic biofilm on the cathode and most of the produced gas was converted to methane ($86 \pm 6\%$). Thus, to increase hydrogen recovery, the authors suggested a new design to isolate cathode from inoculum for efficient hydrogen gas production at the cathode. In these cases, a compromise between increasing the resistance of the MEC system by introducing membrane separator and hence reducing the energy efficiency should always be considered to increase the hydrogen purity and reduce the methane production. This compromise is a recurring issue with scale-up of MEC systems. Thus some researchers have preferred to couple the MEC system within an effluent treatment train to provide added value and to enhance other treatment processes, exploiting the advantage of methanogenic growth on the cathodes. For example, Wang et al. (2017) incorporated an MEC system with a bio-methane production technology using an anaerobic sludge blanket. They coupled MEC with a pilot scale up-flow anaerobic sludge blanket system to treat Fischer–Tropsch wastewater. In this system both COD removal and methane production increased, driving a syntrophic metabolism that produced a thermodynamically favored mixture of propionic acid and butyric acid in the effluent. This type of MEC coupling can provide a valuable reference for full-scale MEC application.

Reviews by Dewan et al. (2008) in 2008 and Janicek et al. (2014) in 2014 focused on the scale-up of MFCs for practical applications. The earliest attempt to build a large MFC-scale reactor was by the Advanced Water Management Center at the University of Queensland (Australia) to treat brewery effluent (Logan, 2010b). The reactors of approximately 1 m^3 volume constructed with 12 carbon fiber brush anodes fitted within columns that were covered with graphite fiber brush cathodes. However, not much is known about the performance of this reactor. In 2016, Ge and He (2016) investigated the treatment performance, energy production, and cost of an MFC system consisting of 96 MFC modules and operated in a wastewater treatment plant to treat about 200 L of primary effluent. Despite the effluent fluctuation during more than 300 days of operation, the MFC removed about 75% of COD and 90% of the suspended solids, and generated about 200 mW of power. They also claimed that the generated power from a stack of 96 tubular MFC modules was self-sufficient to drive a 60 W DC pump for catholyte recirculation. However, to evaluate the practical application the capital cost of the MFC system would need to be compared to conventional wastewater treatment processes.

Regardless of these examples, successful demonstration of MFC has yet to be achieved. Progress toward an MFC system with 1000 L volume with long-term (one year) operation at pilot-scale has been recently reported (Liang et al., 2018). Two types of municipal wastewaters of low (average 80 mg L^{-1}) and high (average 250 mg L^{-1}) initial COD concentrations were tested. About 70–90% removal was achieved with both systems to obtain a maximum power density in the range of $7\text{--}60 \text{ W m}^{-3}$ ($0.42\text{--}3.64 \text{ W m}^{-2}$), which is significantly less than the energy produced in the same system with artificial wastewater 125 W m^{-3} (7.58 W m^{-2}). This lower performance may be due to the presence of microbial inhibitors in the real effluent that may be eliminated in a pretreatment step.

Wilkinson (2000) has reported an MFC system that utilized food processing industry waste to power a robot in the same plant. The biocatalyst at the anode was a syntrophic microflora biofilm grown from meat processing residues from the same plant, which metabolize natural substrates to charge storage batteries that were powering a robot and minimizing solid waste

accumulation. Similarly, Ieropoulos et al. (2003) utilized MFC metabolism toward true energetic autonomy in an artificial system. A robot, code-named EcoBot I, was reported and claimed to be the first robot in the world run directly and entirely from bacterial power. In this system, the electrodes of the MFC were directly connected to the robot system via electrolytic capacitors as the source of energy without the use of storage batteries. Subsequent generations of EcoBot-II (Melhuish et al., 2006) and EcoBot-III (Ieropoulos et al., 2010) were modified to address the disadvantages of EcoBot I. EcoBot-III, functioned for about one week, approaching self-sufficiency. The utilized substrates were anaerobic or pasteurized waste. Thus, these examples along with other practical examples found in literature (Escapa et al., 2016) provide evidence of possible implementation of the generated power via bioelectrocatalytic activity for self-sufficient robotic applications.

Other applications, such as on-site bio-analytical applications and self-powered environmental devices have been reviewed by ElMekawy et al. (2018). The review updated the recent research performed on the direct and indirect usage of MFCs to monitor water quality in real-time microbial bioluminescence and BOD-based biosensors. The concept of utilizing MXCs as a sensor is based on the exploitation of the generated electrical current in MXCs via the levels of substrate utilized or other species present in the cultivation media, which are consequently considered the analytes (Parkhey and Mohan, 2019). MXCs, thus, have been studied as biosensors for BOD (Kharkwal et al., 2017), COD (Spurr et al., 2018), toxicity (Zhou et al., 2017), DO (Song et al., 2019), ammonia nitrogen (Zhao et al., 2018; Barua et al., 2018), pathogenic bacteria (ElMekawy et al., 2018) and volatile fatty acids (VFA) (Jiang et al., 2019). Two recent reviews by Parkhey and Mohan (2019) and Jiang et al. (2018) separately discussed the possible utilization of MFCs for water quality sensing systems and the various analytical aspects of MFCs when used as sensors such as stability, repeatability, sensitivity, and selectivity. They concluded that MFC-based biosensors still need to be improved compared with the conventional analytical devices, however, their performance level is acceptable and soon will be approved as a standard analytical technique.

9. Figures of merit

Before conducting any experiment, collecting data, and interpreting the mechanisms involved, it is necessary for researchers to review performance indicators used to assess progress and efficiency of MFC systems and to compare reactor designs and operating conditions. Typically, the cell voltage applied to promote hydrogen production in an MEC system ranges from 0.2 V to 1.0 V, using a direct current power supply, connected in series through a resistor with a data acquisition system to record the voltage drop across the resistor. Ding et al. (2016) explored the impacts of applied voltage on the performance of MEC and the microbial activities under anaerobic conditions. They concluded that voltage higher than 0.8 V led to biological cell rupture, lower growth, and metabolic activity; whereas, COD removal and methane yield increased when increasing the cell voltage from 0 to 0.8 V. For MFC, however, due to the simultaneous production of power and oxygen reduction, the power consumption of the external load was found to impact the start-up of the system. Zhang et al. (2017) found that MFCs connected to a higher external resistor (1000 Ω), i.e. operating with a lower initial current, had a faster start-up than with lower external resistances (250, 50 and 10 Ω). However, MFCs showed better performance when the external resistance was lower, with increased energy output, active biomass, and maximum power density. Thus, they concluded that for MFC start-up, it is feasible to operate at a

higher external resistance (1000 Ω) and then gradually switch to lower external resistances to facilitate higher current.

To estimate the COD (mg/L) distribution in MECs, an assumption has to be made that the only COD sinks are the production of energy (H_2 in MEC or electricity in MFC) and growth of biomass (suspended, attached, and products). Thus, the following overall mass balance for a batch treatment is:

$$COD_{initial} = COD_{output} + COD_{biomass,sus} + COD_{biomass,att} + COD_{final} \quad (17)$$

where $COD_{initial}$ is the initial COD in the anode chamber, COD_{output} is the COD equivalent of the energy output (H_2 in MEC or electricity in MFC), $COD_{biomass,sus}$ is the COD of net accumulated suspended biomass, $COD_{biomass,att}$ is the COD of the net accumulated attached biomass on the anode, and COD_{final} is the amount of COD in the anode chamber at the end of the treatment.

The main indicator of the output from an MXC system is the electrical current (I). However, to compare the performance of reactors, typically, current is normalized to current density (Am^{-2}), if the anode surface area is known, otherwise normalization to reactor volume become conventional in bioelectrochemical systems. One of the main challenges in developing an MXC system is to obtain a high and stable current density with a scalable design. Massazza et al. (2015) obtained a current density of 129 Am^{-2} using an ice-templated titanium-based ceramic anode. A higher current density of 390 Am^{-2} was reported by Chen et al. (2012) using several layers of carbonized corrugated cardboard. Although both reports indicated a stable current density for moderate operating periods, the implementation of scalable MXC system with such high current densities has not been confirmed for energy production from waste. In practice current densities in the range of 5–15 Am^{-2} or up to 30 Am^{-2} have been reported using carbon electrodes and conventional benchtop MXCs (Chen et al., 2011).

A high current density in an MXC system indicates a high catalytic activity of biota and high efficiency i.e. increased H_2 yield in MEC and energy production in MFC systems. As discussed earlier, the efficiency is related to the type and amount of substrate utilized, the origin of the inoculum and the design of MXC system, e.g., anode material, cell design, membrane, etc. The efficiency of a system can be estimated from the amount of substrate utilized and the energy produced, and evaluated using the Coulombic efficiency (η_q , %). Thus, η_q defines the fraction of the degraded organic matter that has been used for energy production considering only the charge passed, and not considering voltage losses. In other words, the η_q value represents the fraction of electrons recovered as current, compared to the total amount of electrons available from the oxidation of the starting organic matter. Accordingly, the relationship between the amount of current recovered from the system and the rate of continuous treatment of COD can be used to calculate η_q , as shown in Eq. (18) (Trapero et al., 2017).

$$I = \frac{\eta_q F q \Delta C_{COD}}{8} \quad (18)$$

where; ΔC_{COD} ($g O_2 L^{-1}$) is the change in the concentration of COD from the inlet to outlet, reflecting the amount of organic matter treated, F ($96485 C mol^{-1}$) is Faraday's constant, q ($L s^{-1}$) is the influent flow rate. The factor 8 in the denominator is a conversion constant used to calculate the number of moles of oxygen consumed for the corresponding change in COD value, which considers the molecular weight of oxygen ($32 g mol^{-1}$) and the number of electrons involved in the oxygen reduction ($4 mol e^-$ per $mol O_2$) (Yasri and Gunasekaran, 2017).

Table 2
Coulombic efficiency and current densities produced in some MXC systems.

Substrate	Culture condition	Reactor/ Anode type/other conditions	Substrate conc. (mg/L)	Current density (A/m ²)	CE (%)	Ref.
Acetate	Pre-acclimated bacteria from MFC	Cube shaped MFC/ Graphite fiber brush anode.	1000	8.0	60	Cercado-Quezada et al. (2010)
Glucose	Pre-acclimated MFC in Na-acetate	Cylindrical chamber MFC/ carbon cloth anode	1200	7.0	28	Chaudhary et al. (2000)
Butyrate	Pre-acclimated from domestic wastewater	Single-chambered MFC/ Carbon paper	1000	0.77	8–15	Kundu et al. (2013)
Na-Acetate	Pre-acclimated from domestic wastewater	Sandwich type two-chambers MEC	1600 mg COD/L	6.16	91	Harnisch and Schroder (2010)
Na-Butyrate				2.57	70	
Na-propionate				1.46	59	
Starch processing wastewater	Starch processing wastewater as inoculum	Sandwiching type two-chamber MFC/ carbon paper/proton exchange membrane	9703 mg COD/L	8.9	8.0	Jadhav et al. (2019)
Cassava mill wastewater	Activated sludge	Two-chambered MFC/ graphite anode/Glass wool separator	16000 mg-COD/L	10	20.0	Logan (2008)

Typical C_E s determined for various MXC systems are presented in Table 2. In general, the C_E values and current densities obtained from conventional volatile fatty acids substrates (e.g., acetate, propionate, or butyrate) are higher than those obtained from complex substrates (such as starch). The utilization of complex substrates requires a microbial community that is capable of breaking down these types of compounds to low-molecular-weight fractions that can be utilized by ARB. For example, when starch- or cassava-containing wastewaters are utilized as substrates, although their hydrolysis products may be good sources of ARB food, electricity generation in MXC system requires a microbial community that can hydrolyze complex starch and cellulose compounds at the same time and has exoelectrogenic activity ([Lu et al., 2009](#); [Kaewkannetra et al., 2011](#)).

Power production in MFC system is usually expressed as power density (W per m² of anode surface area), or power intensity (W per m³ or reactor volume) ([Logan et al., 2006](#)). Early investigations indicated that the power intensity obtained for reactor volumes larger than 1 L is typically less than 1 kW/m³, which for electric production facilities is considered as the threshold for practical industrial application for energy recovery from organic matter ([Pham et al., 2009](#)). In the case of hydrogen recovery in an MEC system, however, the output of a system is usually expressed by the hydrogen yield (mol-H₂/mol-substrate) and the H₂ production rate (HPR) (m³ m⁻³ d⁻¹ normalized to the volume of catholyte, anode or reactor m³) ([Logan, 2008](#); [Logan et al., 2006](#)). The values of the power output and H₂ production in MFCs and MECs, respectively, are dependent on the surface area of both the biofilm anode as well as the cathode. Power output values reported in the literature are mostly estimates based on the normalization to the anode or cathode surface area or their respective compartment volumes, or reactor volume. The assumption when the calculation is based on the anode (volume or surface area), is that the power output is limited by the electrons flux at the anode interface, whereas, when the calculation is based on the cathode (volume or surface area), the proton flux is the limiting factor ([Logan, 2008](#)). Both assumptions can be used to compare reactors of similar volumes to explore materials and understand factors that limit the reactor output ([Logan, 2008](#)). However, to shift the attention to reflects the engineering interest for industrial implementation and scale-up, the calculation also should be shifted to normalize the output to the reactor

volume ([Logan, 2008](#)). In some examples found in literature, moderately high HPR values (>20 m³ m⁻³ d⁻¹) are reported when the calculation is based on normalizing the production rate only to the cathode or anode compartment volumes. For example, [Cheng and Logan \(2011\)](#) reported an HPR of 17.8 m³ m⁻³ d⁻¹ for a reactor using a carbon brush anode of about 6 mL volume and acetate as the substrate, operated with an applied voltage of 1 V. Although the HPR values is high, these are bench-scale reactors and their volumes do not support the feasibility of scale-up and industrial implementation. Recently, a larger reactor with a volume of 1 L has been reported by [Guo et al. \(2017\)](#) that is capable of continuous production of 7.1 m³ m⁻³ d⁻¹ HPR using a novel design with tubular electrodes and acetate as the electron donor, operating at an applied voltage of 1 V. This reactor yielded a promising HPR, but the design has not yet been tested for substrates other than acetate.

The energy efficiency (%) from a reactor is usually measured by comparing the total energy recovered to the total energy input. The source of energy input to the system includes both the external electrical energy input (for MECs), and the energy input from the substrate. Thus, a higher energy input to the system will typically be reflected in a lower energy efficiency. For example, the MEC with tubular electrodes reported by [Guo et al. \(2017\)](#) decreased electrical energy efficiency from 209 ± 5% to 149 ± 1% as the voltage increased from 0.55 V to 1.0 V. The efficiency was evaluated here in terms of the electric input. In general, when evaluating the energy efficiency of the MEC in terms of the external electrical energy input only, the values show high efficiency and in most cases exceeding 100%. The efficiency, in this case, compares the combustion energy of the actual hydrogen produced in the system with the equivalent amount of hydrogen produced via water electrolysis when applying the same electric energy input. Thus this is not a true energy efficiency, since the energy of substrate oxidation is not accounted for.

10. Summary

The catalytic activity of biofilm grown on the surface of electrodes in aqueous media has been exploited in electrochemical systems for simultaneous remediation of environmental pollutants and interfacial electron transfer for energy generation. In this review, we have addressed the main electrochemical aspects

of ARBs and their utilization in MECs and MFCs systems. The aim is to draw more attention to theory and models from an electrochemistry perspective, provoking research and innovation into this nascent and synchronized approaches of sustainable power generation and waste remediation. The information provided in this manuscript is summarized and presented to provide the scientific community insight into the electrochemical theory related to these developments.

Following the consideration of the concepts and the viability of both the galvanic MFC and electrolytic MEC systems, the exoelectrogenic microorganisms and their bioelectrocatalytic activity were discussed. The ability of both bioelectrochemical systems to effectively oxidize organic containing waste to extract energy has been demonstrated. Remediation processes accompanied by extracellular electron transfer processes in ARBs utilize a wider sources of organic matter compared to the non-exoelectrogenic fermentation process. Systematic electrochemical approaches to investigate the inoculum interactions at the interface during start-up, growth, or starvation of biofilms can reveal important information related to kinetic parameters and their effects on process limitation and cell performance. Among these, interpretation of Butler–Volmer–Monod, Nernst–Monod, and their derivatives are useful for the investigation of overpotentials developed in various parts of the cell, to distinguish EET pathways of various types of ARB, and the response of ARB to the environmental conditions to improve the catalytic activity of the system and reduce the energy loss.

The review also highlights the importance of each part of the system from current and forthcoming trends in MXCs. Unlike other biological systems, the dominant populations of ARB biofilm usually differ from those that they are originally enriched from, and also depend on the type of substrate and the MXCs conditions. Moreover, differences are also existing between MFCs and MECs due to the intrusion of oxygen through the cathode in MFCs and the release of H_2 gas in MECs.

Careful control of the enrichment process of biofilm e.g. substrate, physical or chemical conditions, can promote a specific stable electrochemically active microbial community that can be utilized in extreme environmental conditions for the treatment of recalcitrant and persistent wastes.

Careful reactor design, choice of electrodes, and systematic control of parameters can lead also to an effective bioelectrochemical process. In MEX, the anode is considered the main component that captures the biological response. Thus, among factors impacting the immobilization of biofilms, (i.e. cell design, applied voltage, interfacial charge transfer, electrolyte conductivity, pH, substrate types, electrode material and porosity), the selection of anode surface with low ohmic loss and supported with compatible materials, most suitable for bacteria growth, are among the important factors to be investigated.

In a closed circuit electrochemical MXCs system, cathodic parts are also important in terms of the system efficiency. Ohmic resistance of the electrolyte and interfacial electron transfer at the cathode contribute significantly to the potential loss in MFC systems. The use of a high cost platinum is unsuitable for MXCs, due to deactivation by constituents present in wastewater (such as sulfide), and alternative choices can be considered on a case-by-case basis.

In term of membranes, although it introduces ohmic loss to the MFC systems, its presence is important to prevent the growth of reducing bacteria on an MEC cathode, and to avoid the bioanode from aerobic contact in MFCs. Moreover, the use of an AEM may be useful for stabilizing the anodic media and has been shown to exhibit better efficiency compared with CEM.

Future developments in electrochemical remediation technologies should be dictated not only by the cost but also by the need to meet the public expectations and satisfying regulatory guidelines. Some moderately successful attempts have been achieved in the implementation of MXCs to generate power and for sensors, e.g. monitoring the level of BOD, inhibitors, DO, pathogenic bacteria and VFAs. Although the performance is acceptable, further improvement is required for commercial viability.

The use of organic wastes as resources for cost-effective energy generation offers significant potential to reduce treatment costs and enhance the sustainability of waste treatment processes. In this regard, microbial electrocatalytic techniques show promise. However, there are challenges to overcome such as the low current density, slow rate of remediation, limited scale-up potential, poor portability etc. Thus, further investment to investigate the concept and innovate the technology for process engineering scale-up and for practical application. Furthermore, the investment in bioelectrocatalytic processes would be more promising if acclimatization of such a system can be integrating to assist remediation of substrates that are difficult to break down or sustainable in the environment e.g. oil by-products or persistent organic pollutants. Thus dual targets can be achieved of sustainable energy production and remediation of such pollutants.

11. List of notations/abbreviations and symbols

Notations/Abbreviations

ACE: Average current efficiency
AEM, Anion exchange membrane
APL: Anode-potential losses
ARB: Anodic respiring bacteria
ATP: Adenosine triphosphate
CEM: Cation exchange membrane
COD: Chemical oxygen demand
CV: Cycle voltammetry
DCV: Derivative cycle voltammetry
DO: dissolved oxygen
EET: Extracellular electron transfer
GAC: Granular activated carbon
HPR, Hydrogen production rate ($m^3 H_2/m^3/d$)
ITO: Indium-Tin oxides
LSCV: low-scan cyclic voltammetry
MEC: microbial electrolysis cell
MFC: microbial fuel cell
PBS: phosphate buffered saline
PEDOT: Poly(3,4-ethylenedioxythiophene)
SCE: Saturated calomel electrode
SCOD: Soluble chemical oxygen demand
SHE: Saturated hydrogen electrode
TCOD: Total chemical oxygen demand
TOC: Total organic carbon
VFA: volatile fatty acids

Symbols

α : charge transfer coefficient
 e_0 : elementary charge of electron (C)
 E : electric potential (V)
 E^0 : equilibrium potential (V)
 E_{An} : anodic potential (V)
 E_{KA} : potential (V) when $j = \frac{1}{2} j_{max}$
 E_{PS} : applied voltage (V)
 E_c : equilibrium cell voltage (V)
 E_{ca} : cathodic potential (V)
 F : Faraday constant (96,485 C mol⁻¹)
 j : current density (Am⁻²)
 j_{lim} : limiting current density (Am⁻²)
 j_{max} : maximum current density (Am⁻²)
 K_M : substrate affinity constant
 n : ion valence
 n_{H2} : Number of moles of the hydrogen (mol)
 ns : number of moles of the substrate utilized (mol)
 p : pressure (atm)
 Q : electric charge (C)
 r_{cat} : cathodic hydrogen recovery (%)
 R : ideal gas constant (8.314 4621 J K⁻¹ mol⁻¹)
 R_{H2} : overall H2 recovery (%)
 R_{ex} : external resistance (Ω)
 t : duration (s)
 T : absolute temperature (303.15 K):
 V_S : solution volume (L)
 W_{H2} : energy content of the hydrogen (J)
 W_R : energy loss on the external resistor (J)
 W_S : energy added by substrate (J)
 W_{in} : energy input to the system (J).
 Y_{H2} : hydrogen yield (mol H₂/mol substrate)
 η_{anode} : anode over potential (V)
 η_{E+S} : energy recovery based on external electrical energy input and substrate input (%)
 η_E : energy recovery based on external electrical energy input (%)
 η_q : Coulombic efficiency (%)
 η_S : energy recovery based on substrate input (%)
 ΔG° : standard free energy change (kJ/mol)
 ΔH : enthalpy change (kJ/mol)
 $\Delta \phi_{pH}$: change in the cell voltage due to pH change (V)
 S : substrate concentration (mol/L)

References

- Abrol, I., Yadav, J.S.P., Massoud, F., 1988. Salt-Affected Soils and their Management. Food & Agriculture Org.
- Amend, J.P., Rogers, K.L., Meyer-Dombard, D.A.R., 2004. Microbially mediated sulfur-redox: Energetics in marine hydrothermal vent systems. *Geol. Soc. Am. Spec. Pap.* 379, 17–34.
- Amit Kumar, K.K., Lens, Piet, Leech, Donal, 2012. Does bioelectrochemical cell configuration and anode potential affect biofilm response? *Biochem. Soc. Trans.* 40, 1308–1314.
- Angelaalincy, M.J., et al., 2018. Biofilm engineering approaches for improving the performance of microbial fuel cells and bioelectrochemical systems. *Front. Energy Res.* 6 (63).
- Antolini, E., 2015. Composite materials for polymer electrolyte membrane microbial fuel cells. *Biosens. Bioelectron.* 69, 54–70.
- Azeredo, J., et al., 2017. Critical review on biofilm methods. *Crit. Rev. Microbiol.* 43 (3), 313–351.
- Babauta, J., et al., 2012a. Electrochemically active biofilms: facts and fiction A review. *Biofouling* 28 (8), 789–812.
- Babauta, J.T., et al., 2012b. pH, redox potential and local biofilm potential microenvironments within *Geobacter sulfurreducens* biofilms and their roles in electron transfer. *Biotechnol. Bioeng.* 109 (10), 2651–2662.
- Badalamenti, J.P., Krajmalnik-Brown, R., Torres, C.I., 2013. Generation of high current densities by pure cultures of anode-respiring *Geobacter* spp. under alkaline and saline conditions in microbial electrochemical cells. *mBio* 4 (3).
- Bajracharya, S., et al., 2015. Carbon dioxide reduction by mixed and pure cultures in microbial electrosynthesis using an assembly of graphite felt and stainless steel as a cathode. *Bioresour. Technol.* 195, 14–24.
- Barua, S., et al., 2018. Anodic performance of microbial electrolysis cells in response to ammonia nitrogen. *J. Environ. Eng. Sci.* 14 (1), 37–43.
- Baudler, A., et al., 2015. Does it have to be carbon? Metal anodes in microbial fuel cells and related bioelectrochemical systems. *Energy Environ. Sci.* 8 (7), 2048–2055.
- Ben Liew, K., et al., 2014. Non-Pt catalyst as oxygen reduction reaction in microbial fuel cells: A review. *Int. J. Hydrog. Energy* 39 (10), 4870–4883.
- Bennetto, H., 1990. Electricity generation by microorganisms. *Biotechnol. Educ.* 1 (4), 163–168.
- Bertini, I., et al., 1994. *Bioinorganic Chemistry*. University Science Books, Mill Valley, CA, USA.
- Biffinger, J.C., et al., 2008. Oxygen exposure promotes fuel diversity for *Shewanella oneidensis* microbial fuel cells. *Biosens. Bioelectron.* 23 (6), 820–826.
- Carmona, A., 2012. Investigation of Electron Transfer Mechanisms in Electrochemically Active Microbial Biofilms (Ph.D. thesis).
- Cercado-Quezada, B., Delia, M.-L., Bergel, A., 2010. Treatment of dairy wastes with a microbial anode formed from garden compost. *J. Appl. Electrochem.* 40 (2), 225–232.
- Cereda, A., et al., 2014. A bioelectrochemical approach to characterize extracellular electron transfer by *Synechocystis* sp. PCC6803. *PLoS One* 9 (3), e91484.
- Chaudhary, A., et al., 2000. Separation of nickel from cobalt using electrodialysis in the presence of EDTA. *J. Appl. Electrochem.* 30 (4), 439–445.
- Chen, S.L., et al., 2011. Electrospun and solution blown three-dimensional carbon fiber nonwovens for application as electrodes in microbial fuel cells. *Energy Environ. Sci.* 4 (4), 1417–1421.
- Chen, S., et al., 2012. Layered corrugated electrode macrostructures boost microbial bioelectrocatalysis. *Energy Environ. Sci.* 5 (12), 9769–9772.
- Chen, X., et al., 2019. Microbial electrochemical treatment of biorefinery black liquor and resource recovery. *Green Chem.*
- Cheng, S., Liu, H., Logan, B.E., 2006. Power densities using different cathode catalysts (Pt and CoTMPP) and polymer binders (Nafion and PTFE) in single chamber microbial fuel cells. *Environ. Sci. Technol.* 40 (1), 364–369.
- Cheng, S., Logan, B.E., 2007. Ammonia treatment of carbon cloth anodes to enhance power generation of microbial fuel cells. *Electrochem. Commun.* 9 (3), 492–496.
- Cheng, S., Logan, B.E., 2011. High hydrogen production rate of microbial electrolysis cell (MEC) with reduced electrode spacing. *Bioresour. Technol.* 102 (3), 3571–3574.
- Cheng, J., et al., 2010. Palm oil mill effluent treatment using a two-stage microbial fuel cells system integrated with immobilized biological aerated filters. *Bioresour. Technol.* 101 (8), 2729–2734.
- Cho, S.-K., et al., 2019. Improved hydrogen recovery in microbial electrolysis cells using intermittent energy input. *Int. J. Hydrogen Energy* 44 (4), 2253–2257.
- Choi, O., Sang, B.-I., 2016. Extracellular electron transfer from cathode to microbes: application for biofuel production. *Biotechnol. Biofuels* 9 (1), 11.
- Choi, S., et al., 2013. Enhanced power production of a membrane electrode assembly microbial fuel cell (MFC) using a cost effective poly [2, 5-benzimidazole] (ABPBI) impregnated non-woven fabric filter. *Bioresour. Technol.* 128, 14–21.
- Commault, A.S., et al., 2013. Influence of anode potentials on selection of *Geobacter* strains in microbial electrolysis cells. *Bioresour. Technol.* 139, 226–234.
- Cusick, R.D., et al., 2011. Performance of a pilot-scale continuous flow microbial electrolysis cell fed winery wastewater. *Appl. Microbiol. Biotechnol.* 89 (6), 2053–2063.
- Davidov, T., et al., 2019. Designing bacterial chemotactic receptors guided by photonic femtoliter well arrays for quantifiable, label-free measurement of bacterial chemotaxis. *ACS Biomater. Sci. Eng.* 5 (2), 603–612.
- Deppenmeier, U., 2004. The membrane-bound electron transport system of methanosaer species. *J. Bioenerg. Biomembr.* 36 (1), 55–64.
- Deutzmann, J.S., Spormann, A.M., 2017. Enhanced microbial electrosynthesis by using defined co-cultures. *ISME J.* 11 (3), 704–714.
- Dewan, A., Beyenal, H., Lewandowski, Z., 2008. Scaling up microbial fuel cells. *Environ. Sci. Technol.* 42 (20), 7643–7648.
- Dhand, V., et al., 2014. Carbon nanospheres synthesized via solution combustion method: their application as an anode material and catalyst for hydrogen production. *Carbon Lett.* 15 (3), 198–202.
- Ding, A., et al., 2016. Impact of applied voltage on methane generation and microbial activities in an anaerobic microbial electrolysis cell (MEC). *Chem. Eng. J.* 283, 260–265.

- Dong, Y., et al., 2015. A 90-liter stackable baffled microbial fuel cell for brewery wastewater treatment based on energy self-sufficient mode. *Bioresour. Technol.* 195, 66–72.
- Edwards, P.P., Kuznetsov, V.L., David, W.I.F., 2007. Hydrogen energy. *Philos. Trans. R. Soc. Lond. Ser. A Math. Phys. Eng. Sci.* 365 (1853), 1043–1056.
- ElMekawy, A., et al., 2018. Bio-analytical applications of microbial fuel cell-based biosensors for onsite water quality monitoring. *J. Appl. Microbiol.* 124 (1), 302–313.
- Escapa, A., et al., 2016. Microbial electrolysis cells: An emerging technology for wastewater treatment and energy recovery from laboratory to pilot plant and beyond. *Renew. Sustain. Energy Rev.* 55, 942–956.
- Fakhru'l-Razi, A., et al., 2009. Review of technologies for oil and gas produced water treatment. *J. Hard Mater.* 170 (2), 530–551.
- Fan, Y., Han, S.-K., Liu, H., 2012. Improved performance of CEA microbial fuel cells with increased reactor size. *Energy Environ. Sci.* 5 (8), 8273–8280.
- Fenner, D., et al., 2006. The anti-microbial activity of electrolysed oxidizing water against microorganisms relevant in veterinary medicine. *J. Vet. Med. B* 53 (3), 133–137.
- Fitzgerald, L.A., et al., 2013. *Shewanella frigidimarina* microbial fuel cells and the influence of divalent cations on current output. *Biosens. Bioelectron.* 40, 102–109.
- Fogel, R., Limson, J., 2016. Applications of nanomaterials in microbial fuel cells. In: *Nanomaterials for Fuel Cell Catalysis*. Springer, pp. 551–575.
- García-Gusano, D., Iribarren, D., Garraín, D., 2017. Prospective analysis of energy security: A practical life-cycle approach focused on renewable power generation and oriented towards policy-makers. *Appl. Energy* 190, 891–901.
- Ge, Z., He, Z., 2016. Long-term performance of a 200 liter modularized microbial fuel cell system treating municipal wastewater: treatment energy, and cost. *Environ. Sci. Water Res. Technol.* 2 (2), 274–281.
- Ge, Z., et al., 2015. Energy extraction from a large-scale microbial fuel cell system treating municipal wastewater. *J. Power Sources* 297, 260–264.
- Ghasemi, M., et al., 2013. Nano-structured carbon as electrode material in microbial fuel cells: A comprehensive review. *J. Alloys Compd.* 580, 245–255.
- Ghasemi Naraghi, Z., et al., 2015. Produced water treatment with simultaneous bioenergy production using novel bioelectrochemical systems. *Electrochim. Acta* 180, 535–544.
- Gnana kumar, G., et al., 2014. Conductive polymer/graphene supported platinum nanoparticles as anode catalysts for the extended power generation of microbial fuel cells. *Ind. Eng. Chem. Res.* 53 (43), 16883–16893.
- Grattieri, M., Minter, S.D., 2018. Microbial fuel cells in saline and hypersaline environments: Advancements challenges and future perspectives. *Bioelectrochemistry* 120, 127–137.
- Guiral-Brugna, M., et al., 2001. Electrocatalysis of the hydrogen production by [Fe] hydrogenase from *Desulfovibrio vulgaris* Hildenborough. *J. Electroanal. Soc.* 510 (1–2), 136–143.
- Guo, K., Prévot, A., Rabaey, K., 2017. A novel tubular microbial electrolysis cell for high rate hydrogen production. *J. Power Sources* 356, 484–490.
- Hahn, J., et al., 2017. A switchable linker-based immunoassay for ultrasensitive visible detection of Salmonella in tomatoes. *J. Food Sci.* 82 (10), 2321–2328.
- Hamelers, H.V.M., et al., 2011. Butler–Volmer–Monod model for describing bio-anode polarization curves. *Bioresour. Technol.* 102 (1), 381–387.
- Harnisch, F., Schroder, U., 2010. From MFC to MXC: chemical and biological cathodes and their potential for microbial bioelectrochemical systems. *Chem. Soc. Rev.* 39 (11), 4433–4448.
- Hasany, M., Mardanpour, M.M., Yaghmaei, S., 2016. Biocatalysts in microbial electrolysis cells: A review. *Int. J. Hydrogen Energy* 41 (3), 1477–1493.
- He, Y., et al., 2015. Carbon nanotubes simultaneously as the anode and microbial carrier for up-flow fixed-bed microbial fuel cell. *Biochem. Eng. J.* 94, 39–44.
- Heidrich, E.S., et al., 2013. Production of hydrogen from domestic wastewater in a pilot-scale microbial electrolysis cell. *Appl. Microbiol. Biotechnol.* 97 (15), 6979–6989.
- Heidrich, E.S., et al., 2014. Performance of a pilot scale microbial electrolysis cell fed on domestic wastewater at ambient temperatures for a 12 month period. *Bioresour. Technol.* 173, 87–95.
- Hernandez, M., Newman, D., 2001. Extracellular electron transfer. *Cell. Mol. Life Sci.* 58 (11), 1562–1571.
- Hou, Y., et al., 2014. A 3D hybrid of layered MoS₂/nitrogen-doped graphene nanosheet aerogels: an effective catalyst for hydrogen evolution in microbial electrolysis cells. *J. Mater. Chem. A* 2 (34), 13795–13800.
- Hou, Y., et al., 2017. Accelerated azo dye degradation and concurrent hydrogen production in the single-chamber photocatalytic microbial electrolysis cell. *Bioresour. Technol.* 224, 63–68.
- Huang, L., Cheng, S., Chen, G., 2011. Bioelectrochemical systems for efficient recalcitrant wastes treatment. *J. Chem. Technol. Biotechnol.* 86 (4), 481–491.
- Ieropoulos, I., Melhuish, C., Greenman, J., 2003. *Artificial Metabolism: Towards True Energetic Autonomy in Artificial Life*. Springer Berlin Heidelberg, Berlin, Heidelberg.
- Ieropoulos, I., et al., 2010. EcoBot-III—A robot with guts. In: *ALIFE*.
- Jadhav, D.A., et al., 2019. Suppressing methanogens and enriching electrogens in bioelectrochemical systems. *Bioresour. Technol.* 277, 148–156.
- Jain, P., et al., 2017. Bioelectrochemical approaches for removal of sulfate, hydrocarbon and salinity from produced water. *Chemosphere* 166, 96–108.
- Janicek, A., Fan, Y., Liu, H., 2014. Design of microbial fuel cells for practical application: a review and analysis of scale-up studies. *Biofuels* 5 (1), 79–92.
- Jeremiasse, A.W., Hamelers, H.V.M., Buisman, C.J.N., 2010. Microbial electrolysis cell with a microbial biocathode. *Bioelectrochemistry* 78 (1), 39–43.
- Jiang, Y., Chu, N., Zeng, R.J., 2019. Submersible probe type microbial electrochemical sensor for volatile fatty acids monitoring in the anaerobic digestion process. *J. Cleaner Prod.*
- Jiang, D., et al., 2011. A pilot-scale study on utilizing multi-anode/cathode microbial fuel cells (MAC MFCs) to enhance the power production in wastewater treatment. *Int. J. Hydrogen Energy* 36 (1), 876–884.
- Jiang, Y., et al., 2018. Microbial fuel cell sensors for water quality early warning systems: Fundamentals signal resolution, optimization and future challenges. *Renew. Sustain. Energy Rev.* 81, 292–305.
- Jin, Q., Kirk, M.F., 2018. pH as a primary control in environmental microbiology: 1. Thermodynamic perspective. *Front. Environ. Sci.* 6 (21).
- Kadier, A., et al., 2014. A review of the substrates used in microbial electrolysis cells (MECs) for producing sustainable and clean hydrogen gas. *Renew. Energy* 71, 466–472.
- Kaewkannetra, P., Chiwes, W., Chiu, T.Y., 2011. Treatment of cassava mill wastewater and production of electricity through microbial fuel cell technology. *Fuel* 90 (8), 2746–2750.
- Kang, Y.L., Ibrahim, S., Pichiah, S., 2015. Synergetic effect of conductive polymer poly(3, 4-ethylenedioxythiophene) with different structural configuration of anode for microbial fuel cell application. *Bioresour. Technol.* 189, 364–369.
- Karthikeyan, R., et al., 2015. Interfacial electron transfer and bioelectrocatalysis of carbonized plant material as effective anode of microbial fuel cell. *Electrochim. Acta* 157, 314–323.
- Kato, S., Hashimoto, K., Watanabe, K., 2012. Microbial interspecies electron transfer via electric currents through conductive minerals. *Proc. Natl. Acad. Sci. USA* 109 (25), 10042–10046.
- Kato Marcus, A., Torres, C.I., Rittmann, B.E., 2007. Conduction-based modeling of the biofilm anode of a microbial fuel cell. *Biotechnol. Bioeng.* 98 (6), 1171–1182.
- Katuri, K.P., et al., 2018. Dual-function electrocatalytic and macroporous hollow-fiber Cathode for converting waste streams to valuable resources using microbial electrochemical systems. *Adv. Mater.* 30 (26), 1707072.
- Kharkwal, S., et al., 2017. Development and long-term stability of a novel microbial fuel cell BOD sensor with MnO₂ Catalyst. *Int. J. Mol. Sci.* 18 (2), 276.
- Kim, H.J., et al., 2002. A mediator-less microbial fuel cell using a metal reducing bacterium, *Shewanella putrefaciens*. *Enz. Microbiol. Technol.* 30 (2), 145–152.
- Kim, J.R., et al., 2007. Power generation using different Cation anion, and ultrafiltration membranes in microbial fuel cells. *Environ. Sci. Technol.* 41 (3), 1004–1009.
- Kokabian, B., Ghimire, U., Gude, V.G., 2018. Water deionization with renewable energy production in microalgae–microbial desalination process. *Renew. Energy* 122, 354–361.
- Kumar, G., et al., 2017. Microbial electrochemical systems for sustainable bio-hydrogen production: Surveying the experiences from a start-up viewpoint. *Renew. Sustain. Energy Rev.* 70, 589–597.
- Kundu, A., et al., 2013. An overview of cathode material and catalysts suitable for generating hydrogen in microbial electrolysis cell. *Int. J. Hydrogen Energy* 38 (4), 1745–1757.
- Lacroix, R., et al., 2014. Modelling potential/current distribution in microbial electrochemical systems shows how the optimal bioanode architecture depends on electrolyte conductivity. *Phys. Chem. Chem. Phys.* 16 (41), 22892–22902.
- Lapinonnière, L., Picot, M., Barrière, F., 2012. Enzymatic versus microbial bio-catalyzed electrodes in bio-electrochemical systems. *ChemSusChem* 5 (6), 995–1005.
- Lee, H.-S., 2018. Electrokinetic analyses in biofilm anodes: Ohmic conduction of extracellular electron transfer. *Bioresour. Technol.* 256, 509–514.
- Li, X., et al., 2010. Manganese dioxide as a new cathode catalyst in microbial fuel cells. *J. Power Sources* 195 (9), 2586–2591.
- Li, C., et al., 2011. Effect of conductive polymers coated anode on the performance of microbial fuel cells (MFCs) and its biodiversity analysis. *Biosens. Bioelectron.* 26 (10), 4169–4176.
- Li, X.-H., et al., 2014. Enhanced H₂ production from corn stalk by integrating dark fermentation and single chamber microbial electrolysis cells with double anode arrangement. *Int. J. Hydrogen Energy* 39 (17), 8977–8982.
- Li, M., et al., 2018. Microbial fuel cell (MFC) power performance improvement through enhanced microbial electrogenicity. *Biotech. Adv.* 36 (4), 1316–1327.
- Liang, D.-W., et al., 2011. Enhancement of hydrogen production in a single chamber microbial electrolysis cell through anode arrangement optimization. *Bioresour. Technol.* 102 (23), 10881–10885.
- Liang, J., et al., 2012. Sulfur and nitrogen dual-doped mesoporous graphene electrocatalyst for oxygen reduction with synergistically enhanced performance. *Angew. Chem. Int. Ed.* 51 (46), 11496–11500.

- Liang, P., et al., 2018. One-year operation of 1000-L modularized microbial fuel cell for municipal wastewater treatment. *Water Res.* 141, 1–8.
- Liao, Q., et al., 2014. Electricity generation and COD removal of microbial fuel cells (MFCs) operated with alkaline substrates. *Int. J. Hydrogen Energy* 39 (33), 19349–19354.
- Lienemann, M., et al., 2018. Towards patterned bioelectronics: facilitated immobilization of exoelectrogenic *Escherichia coli* with heterologous pili. *Microbial Biotechnol.* 11 (6), 1184–1194.
- Liu, H., Cheng, S., Logan, B.E., 2005b. Production of electricity from acetate or butyrate using a single-chamber microbial fuel cell. *Environ. Sci. Technol.* 39 (2), 658–662.
- Liu, H., Grot, S., Logan, B.E., 2005a. Electrochemically assisted microbial production of hydrogen from acetate. *Environ. Sci. Technol.* 39 (11), 4317–4320.
- Liu, X., Wu, W., Gu, Z., 2015. Poly (3, 4-ethylenedioxythiophene) promotes direct electron transfer at the interface between *Shewanella loihica* and the anode in a microbial fuel cell. *J. Power Sources* 277, 110–115.
- Liu, G., et al., 2011. Examination of microbial fuel cell start-up times with domestic wastewater and additional amendments. *Bioresour. Technol.* 102 (15), 7301–7306.
- Liu, J., et al., 2014. A microbial fluidized electrode electrolysis cell (MFEEC) for enhanced hydrogen production. *J. Power Sources* 271, 530–533.
- Logan, B.E., 2008. *Microbial Fuel Cells*. John Wiley & Sons Inc., Hoboken, New Jersey.
- Logan, B.E., 2009a. Exoelectrogenic bacteria that power microbial fuel cells. *Nat. Rev. Microbiol.* 7 (5), 375–381.
- Logan, B.E., 2009b. Scaling up microbial fuel cells and other bioelectrochemical systems. *Appl. Microbiol. Biotechnol.* 85 (6), 1665–1671.
- Logan, B., 2010a. Scaling up microbial fuel cells and other bioelectrochemical systems. *Appl. Microbiol. Biotechnol.* 85 (6), 1665–1671.
- Logan, B.E., 2010b. Scaling up microbial fuel cells and other bioelectrochemical systems. *Appl. Microbiol. Biotechnol.* 85 (6), 1665–1671.
- Logan, B.E., et al., 2006. Microbial fuel cells: methodology and technology. *Environ. Sci. Technol.* 40 (17), 5181–5192.
- Logan, B., et al., 2007. Graphite fiber brush anodes for increased power production in air-cathode microbial fuel cells. *Environ. Sci. Technol.* 41 (9), 3341–3346.
- Logan, B.E., et al., 2008. Microbial electrolysis cells for high yield hydrogen gas production from organic matter. *Environ. Sci. Technol.* 42 (23), 8630–8640.
- Lojou, E., et al., 2002. Hydrogenase activity control at *Desulfovibrio vulgaris* cell-coated carbon electrodes: biochemical and chemical factors influencing the mediated bioelectrocatalysis. *Electroanalysis* 14 (13), 913.
- Lovley, D.R., 2008. The microbe electric: conversion of organic matter to electricity. *Curr. Opin. Biotechnol.* 19 (6), 564–571.
- Lovley, D.R., Phillips, E.J.P., 1986. Organic matter mineralization with reduction of ferric iron in anaerobic sediments. *Appl. Environ. Microbiol.* 51 (4), 683–689.
- Lu, L., Xing, D., Ren, N., 2012. Pyrosequencing reveals highly diverse microbial communities in microbial electrolysis cells involved in enhanced H₂ production from waste activated sludge. *Water Res.* 46 (7), 2425–2434.
- Lu, N., et al., 2009. Electricity generation from starch processing wastewater using microbial fuel cell technology. *Biochem. Eng. J.* 43 (3), 246–251.
- Luckarift, H.R., et al., 2012. Facile fabrication of scalable hierarchically structured polymer/carbon architectures for bioelectrodes. *ACS Appl. Mater. Interfaces* 4 (4), 2082–2087.
- Luo, H., et al., 2014. Heavy metal recovery combined with H₂ production from artificial acid mine drainage using the microbial electrolysis cell. *J. Hard Mater.* 270, 153–159.
- Mahmoud, R.H., et al., 2018. Assisting the biofilm formation of exoelectrogens using nanostructured microbial fuel cells. *J. Electroanal. Soc.* 824, 128–135.
- Marsili, E., et al., 2008a. *Shewanella* secretes flavins that mediate extracellular electron transfer. *Proc. Natl. Acad. Sci.* 105 (10), 3968–3973.
- Marsili, E., et al., 2008b. Microbial biofilm voltammetry: Direct electrochemical characterization of Catalytic electrode-attached biofilms. *Appl. Environ. Microbiol.* 74 (23), 7329–7337.
- Martinucci, E., et al., 2015. Energy balance and microbial fuel cells experimentation at wastewater treatment plant Milano-Nosedo. *Int. J. Hydrogen Energy* 40 (42), 14683–14689.
- Massazza, D., et al., 2015. New ceramic electrodes allow reaching the target current density in bioelectrochemical systems. *Energy Environ. Sci.* 8 (9), 2707–2712.
- Melhuish, C., et al., 2006. Energetically autonomous robots: Food for thought. *Auton. Robots* 21 (3), 187–198.
- Mohanakrishna, G., Al-Raoush, R.I., Abu-Reesh, I.M., 2018. Induced bioelectrochemical metabolism for bioremediation of petroleum refinery wastewater: Optimization of applied potential and flow of wastewater. *Bioresour. Technol.* 260, 227–232.
- Montpart, N., et al., 2015. Hydrogen production in single chamber microbial electrolysis cells with different complex substrates. *Water Res.* 68, 601–615.
- More, T., Ghangrekar, M., 2010. Improving performance of microbial fuel cell with ultrasonication pre-treatment of mixed anaerobic inoculum sludge. *Bioresour. Technol.* 101 (2), 562–567.
- Mu, Y., et al., 2009. Nitrobenzene removal in bioelectrochemical systems. *Environ. Sci. Technol.* 43 (22), 8690–8695.
- Munoz, L.D., et al., 2010. Combining phosphate species and stainless steel cathode to enhance hydrogen evolution in microbial electrolysis cell (MEC). *Electrochem. Commun.* 12 (2), 183–186.
- Nam, J.-Y., Tokash, J.C., Logan, B.E., 2011. Comparison of microbial electrolysis cells operated with added voltage or by setting the anode potential. *Int. J. Hydrog. Energy* 36 (17), 10550–10556.
- Nevin, K.P., et al., 2008. Power output and coulombic efficiencies from biofilms of *Geobacter sulfurreducens* comparable to mixed community microbial fuel cells. *Environ. Microbiol.* 10 (10), 2505–2514.
- Nissim, R., Compton, R.G., 2013. Superoxide generation from the reduction of oxygen at the carbon-oil-water triple phase boundary. *Phys. Chem. Chem. Phys.* 15 (28), 11918–11925.
- Oh, S., Logan, B.E., 2005. Hydrogen and electricity production from a food processing wastewater using fermentation and microbial fuel cell technologies. *Water Res.* 39 (19), 4673–4682.
- Paitier, A., et al., 2017. Microbial fuel cell anodic microbial population dynamics during MFC start-up. *Biosens. Bioelectron.* 92, 357–363.
- Pandit, S., Das, D., 2018. Principles of microbial fuel cell for the power generation. In: Das, D. (Ed.), *Microbial Fuel Cell: A Bioelectrochemical System that Converts Waste to Watts*. Springer International Publishing, Cham, pp. 21–41.
- Pant, D., et al., 2010. A review of the substrates used in microbial fuel cells (MFCs) for sustainable energy production. *Bioresour. Technol.* 101 (6), 1533–1543.
- Pant, D., et al., 2011. An introduction to the life cycle assessment (LCA) of bioelectrochemical systems (BES) for sustainable energy and product generation: relevance and key aspects. *Renew. Sustain. Energy Rev.* 15.
- Parkhey, P., Mohan, S.V., 2019. Biosensing applications of microbial fuel cell: Approach toward miniaturization. In: Mohan, S.V., Varjani, S., Pandey, A. (Eds.), *Microbial Electrochemical Technology*. Elsevier, pp. 977–997, Chapter 61.
- Pasternak, G., Greenman, J., Ieropoulos, I., 2018. Dynamic evolution of anodic biofilm when maturing under different external resistive loads in microbial fuel cells. *Electrochem. Perspect. J. Power Sour.* 400, 392–401.
- Patil, S.A., et al., 2010. Electroactive mixed culture biofilms in microbial bioelectrochemical systems: The role of temperature for biofilm formation and performance. *Biosens. Bioelectron.* 26 (2), 803–808.
- Patil, S.A., et al., 2011. Electroactive mixed culture derived biofilms in microbial bioelectrochemical systems: The role of pH on biofilm formation, performance and composition. *Bioresour. Technol.* 102 (20), 9683–9690.
- Paul, D., et al., 2018. Modification of carbon felt anode with graphene oxide-zeolite composite for enhancing the performance of microbial fuel cell. *Sustain. Energy Technol. Assess.* 26, 77–82.
- Pham, T.H., Aelterman, P., Verstraete, W., 2009. Bioanode performance in bioelectrochemical systems: recent improvements and prospects. *Trend. Biotechnol.* 27 (3), 168–178.
- Picot, M., et al., 2011. Graphite anode surface modification with controlled reduction of specific aryl diazonium salts for improved microbial fuel cells power output. *Biosens. Bioelectron.* 28 (1), 181–188.
- Potter, M.C., 1911. Electrical effects accompanying the decomposition of organic compounds. *Proc. R. Soc. Lond. B* 84 (571), 260–276.
- Ramírez-Vargas, C.A., et al., 2018. Microbial electrochemical technologies for wastewater treatment: Principles and evolution from microbial fuel cells to bioelectrochemical-based constructed wetlands. *Water* 10 (9), 1128.
- Rathinam, N.K., Salem, D.R., Sani, R.K., 2019. Biofilm engineering for improving the performance of microbial electrochemical technologies. In: Mohan, S.V., Varjani, S., Pandey, A. (Eds.), *Microbial Electrochemical Technology*. Elsevier, pp. 315–338, Chapter 24.
- Reddy, M.V., Sun, X., 2019. Bioelectrosynthesis of various chemicals and evaluation of their microbiological aspects. In: *Microbial Electrochemical Technology*. Elsevier, pp. 757–776.
- Reguera, G., et al., 2005. Extracellular electron transfer via microbial nanowires. *Nature* 435 (7045), 1098–1101.
- Richter, H., et al., 2009. 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. *Energy Environ. Sci.* 2 (5), 506–516.
- Rimboud, M., et al., 2014. Electroanalysis of microbial anodes for bioelectrochemical systems: basics, progress and perspectives. *Phys. Chem. Chem. Phys.* 16 (31), 16349–16366.
- Rimboud, M., et al., 2017. Different methods used to form oxygen reducing biocathodes lead to different biomass quantities, bacterial communities, and electrochemical kinetics. *Bioelectrochemistry* 116, 24–32.
- Rittmann, B.E., 2008. Opportunities for renewable bioenergy using microorganisms. *Biotechnol. Bioeng.* 100 (2), 203–212.
- Rodrigues, T.D.C., Rosenbaum, M.A., 2014. Microbial Electroreduction: Screening for New Cathodic Biocatalysts. *ChemElectroChem*, p. 1.
- Rousseau, R., et al., 2013. Microbial bioanodes with high salinity tolerance for microbial fuel cells and microbial electrolysis cells. *Electrochem. Commun.* 33, 1–4.

- Rowe, A.R., et al., 2017. In situ electrochemical enrichment and isolation of a magnetite-reducing bacterium from a high pH serpentinizing spring. *Environ. Microbiol.* n/a–n/a.
- Roy, S., Pandit, S., 2019. 1.2 - Microbial electrochemical system: Principles and application. In: Mohan, S.V., Varjani, S., Pandey, A. (Eds.), *Microbial Electrochemical Technology*. Elsevier, pp. 19–48.
- Rozendal, R.A., et al., 2007. Performance of single chamber biocatalyzed electrolysis with different types of ion exchange membranes. *Water Res.* 41 (9), 1984–1994.
- Rozendal, R.A., et al., 2008. Hydrogen production with a microbial biocathode. *Environ. Sci. Technol.* 42 (2), 629–634.
- Sánchez, O.G., et al., 2019. Recent advances in industrial CO₂ electroreduction. *Curr. Opin. Green Sustain. Chem.* 16, 47–56.
- Santoro, C., et al., 2017. Microbial fuel cells: From fundamentals to applications. A review. *J. Power Sources* 356, 225–244.
- Saratale, G.D., et al., 2017. A comprehensive overview on electro-active biofilms role of exo-electrogens and their microbial niches in microbial fuel cells (MFCs). *Chemosphere* 178, 534–547.
- Schippers, A., 2004. Biogeochemistry of metal sulfide oxidation in mining environments sediments, and soils. *Geol. Soc. Am. Spec. Pap.* 379, 49–62.
- Schroder, U., 2007. Anodic electron transfer mechanisms in microbial fuel cells and their energy efficiency. *Phys. Chem. Chem. Phys.* 9 (21), 2619–2629.
- Schwartz, E., Fritsch, J., Friedrich, B., 2013. H₂-Metabolizing prokaryotes. In: Rosenberg, E., et al. (Eds.), *The Prokaryotes: Prokaryotic Physiology and Biochemistry*. Springer Berlin Heidelberg, Berlin, Heidelberg, pp. 119–199.
- Shizas, I., Bagley, D.M., 2004. Experimental determination of energy content of unknown organics in municipal wastewater streams. *J. Energy Eng.* 130 (2), 45–53.
- Singh, A., et al., 2019. Electrocatalytic activity of functionalized carbon paper electrodes and their correlation to the Fermi level derived from Raman spectra. *ACS Appl. Energy Mater.*
- Slate, A.J., et al., 2019. Microbial fuel cells: An overview of current technology. *Renew. Sustain. Energy Rev.* 101, 60–81.
- Sleutels, T.H.J.A., Hamelers, H.V.M., Buisman, C.J.N., 2011. Effect of mass and charge transport speed and direction in porous anodes on microbial electrolysis cell performance. *Bioresour. Technol.* 102 (1), 399–403.
- Sleutels, T.H.J.A., et al., 2009a. Improved performance of porous bio-anodes in microbial electrolysis cells by enhancing mass and charge transport. *Int. J. Hydrogen Energy* 34 (24), 9655–9661.
- Sleutels, T.H.J.A., et al., 2009b. Ion transport resistance in microbial electrolysis cells with anion and cation exchange membranes. *Int. J. Hydrogen Energy* 34 (9), 3612–3620.
- Song, N., et al., 2019. Development of a sediment microbial fuel cell-based biosensor for simultaneous online monitoring of dissolved oxygen concentrations along various depths in lake water. *Sci. Total Environ.* 673, 272–280.
- Spurr, M.W.A., et al., 2018. Extending the dynamic range of biochemical oxygen demand sensing with multi-stage microbial fuel cells. *Environ. Sci. Water Res. Technol.* 4 (12), 2029–2040.
- Srikanth, S., et al., 2016. Electro-biocatalytic treatment of petroleum refinery wastewater using microbial fuel cell (MFC) in continuous mode operation. *Bioresour. Technol.* 221, 70–77.
- Stoll, Z.A., et al., 2015. Shale gas produced water treatment using innovative microbial capacitive desalination cell. *J. Hard Mater.* 283, 847–855.
- Strycharz, S.M., et al., 2011. Application of cyclic voltammetry to investigate enhanced catalytic current generation by biofilm-modified anodes of *Geobacter sulfurreducens* strain DL1 vs variant strain KN400. *Energy Environ. Sci.* 4 (3), 896–913.
- Sultana, S.T., Babauta, J.T., Beyenal, H., 2015. Electrochemical biofilm control: a review. *Biofouling* 31 (9–10), 745–758.
- Tang, X., et al., 2015. Conductive polypyrrole hydrogels and carbon nanotubes composite as an anode for microbial fuel cells. *RSC Adv.* 5 (63), 50968–50974.
- Tee, P.-F., et al., 2018. Bio-energy generation in an affordable, single-chamber microbial fuel cell integrated with adsorption hybrid system: effects of temperature and comparison study. *Environ. Technol.* 39 (8), 1081–1088.
- Tenca, A., et al., 2013. Evaluation of low cost cathode materials for treatment of industrial and food processing wastewater using microbial electrolysis cells. *Int. J. Hydrogen Energy* 38 (4), 1859–1865.
- Theivasanthi, T., Alagar, M., 2011. Studies of copper nanoparticles effects on micro-organisms. *arXiv preprint arXiv:1110.1372*.
- Tkach, O., et al., 2017. Performance of low temperature microbial fuel cells (MFCs) catalyzed by mixed bacterial consortia. *J. Environ. Sci.* 52, 284–292.
- Torres, C.I., Marcus, A.K., Rittmann, B.E., 2007. Kinetics of consumption of fermentation products by anode-respiring bacteria. *Appl. Microbiol. Biotechnol.* 77 (3), 689–697.
- Torres, C.I., et al., 2008. Kinetic experiments for evaluating the Nernst–Monod model for anode-respiring bacteria (ARB) in a biofilm anode. *Environ. Sci. Technol.* 42 (17), 6593–6597.
- Torres, C.I., et al., 2009. Selecting anode-respiring bacteria based on anode potential: Phylogenetic electrochemical, and microscopic characterization. *Environ. Sci. Technol.* 43 (24), 9519–9524.
- Torres, C.I., et al., 2010. A kinetic perspective on extracellular electron transfer by anode-respiring bacteria. *FEMS Microbiol. Rev.* 34 (1), 3–17.
- Trapero, J.R., et al., 2017. Is microbial fuel cell technology ready? An economic answer towards industrial commercialization. *Appl. Energy* 185, 698–707.
- Venkata Mohan, S., Pandey, A., 2019. Sustainable hydrogen production: An introduction. In: Pandey, A., et al. (Eds.), *Biohydrogen*, second ed. Elsevier, pp. 1–23, Chapter 1.
- Verea, L., et al., 2014. Performance of a microbial electrolysis cell (MEC) for hydrogen production with a new process for the biofilm formation. *Int. J. Hydrogen Energy* 39 (17), 8938–8946.
- Wagner, R.C., Porter-Gill, S., Logan, B.E., 2012. Immobilization of anode-attached microbes in a microbial fuel cell. *AMB Express* 2, 2.
- Walter, X.A., et al., 2018. PEE POWER® urinal II – Urinal scale-up with microbial fuel cell scale-down for improved lighting. *J. Power Sources* 392, 150–158.
- Wang, H., Ren, Z.J., 2013. A comprehensive review of microbial electrochemical systems as a platform technology. *Biotech. Adv.* 31 (8), 1796–1807.
- Wang, L.K., et al., 2010a. *Environmental Biotechnology*, Vol. 10. Springer Science & Business Media.
- Wang, A., et al., 2010b. Key factors affecting microbial anode potential in a microbial electrolysis cell for H₂ production. *Int. J. Hydrogen Energy* 35 (24), 13481–13487.
- Wang, D., et al., 2017. Enhanced treatment of Fischer–Tropsch wastewater using up-flow anaerobic sludge blanket system coupled with micro-electrolysis cell: A pilot scale study. *Bioresour. Technol.* 238, 333–342.
- Wang, F., et al., 2019a. Structure of microbial nanowires reveals stacked hemes that transport electrons over micrometers. *Cell* 177 (2), 361–369, e10.
- Wang, W.-K., et al., 2019b. Self-supported microbial carbon aerogel bioelectrocatalytic anode promoting extracellular electron transfer for efficient hydrogen evolution. *Electrochim. Acta* 303, 268–274.
- Weber, K.A., Achenbach, L.A., Coates, J.D., 2006. Microorganisms pumping iron: anaerobic microbial iron oxidation and reduction. *Nat. Rev. Microbiol.* 4 (10), 752–764.
- Werner, C.M., et al., 2015. Attenuation of trace organic compounds (TOCs) in bioelectrochemical systems. *Water Res.* 73, 56–67.
- Wilkinson, S., 2000. “Gastrobots”—Benefits and challenges of microbial fuel cells in FoodPowered robot applications. *Auton. Robots* 9 (2), 99–111.
- Wu, Z., et al., 2019. Engineering an electroactive *Escherichia coli* for the microbial electrosynthesis of succinate from glucose and CO₂. *Microbial Cell Factor.* 18 (1), 15.
- Xie, X., et al., 2011. Three-dimensional carbon nanotube-textile anode for high-performance microbial fuel cells. *Nano Lett.* 11 (1), 291–296.
- Xie, X., et al., 2012. Graphene-sponges as high-performance low-cost anodes for microbial fuel cells. *Energy Environ. Sci.* 5 (5), 6862–6866.
- Xin, X., et al., 2019. An integrated approach for waste activated sludge management towards electric energy production/resource reuse. *Bioresour. Technol.* 274, 225–231.
- Xu, S.T., et al., 2012. Enhanced performance and mechanism study of microbial electrolysis cells using Fe nanoparticle-decorated anodes. *Appl. Microbiol. Biotechnol.* 93 (2), 871–880.
- Yang, N., Hafez, H., Nakhla, G., 2015. Impact of volatile fatty acids on microbial electrolysis cell performance. *Bioresour. Technol.* 193, 449–455.
- Yang, G., et al., 2016. Bifunctional nano-sponges serving as non-precious metal catalysts and self-standing cathodes for high performance fuel cell applications. *Nano Energy* 22, 607–614.
- Yasri, N.G., Gunasekaran, S., 2017. Electrochemical technologies for environmental remediation. In: Anjum, N.A., Gill, S.S., Tuteja, N. (Eds.), *Enhancing Cleanup of Environmental Pollutants: Vol. 2: Non-Biological Approaches*. Springer International Publishing, Cham, pp. 5–73.
- Yasri, N.G., Nakhla, G., 2016. Electrochemical behavior of anode-respiring bacteria on doped carbon electrodes. *ACS Appl. Mater. Interfaces*.
- Yasri, N.G., Nakhla, G., 2017a. The performance of 3-D graphite doped anodes in microbial electrolysis cells. *J. Power Sources* 342, 579–588.
- Yasri, N.G., Nakhla, G., 2017b. Impact of interfacial charge transfer on the start-up of bioelectrochemical systems. *J. Environ. Chem. Eng.* 5 (4), 3640–3648.
- Yi, H., et al., 2009. Selection of a variant of *Geobacter sulfurreducens* with enhanced capacity for current production in microbial fuel cells. *Biosens. Bioelectron.* 24 (12), 3498–3503.
- Yin, Q., et al., 2016. Enhanced methane production in an anaerobic digestion and microbial electrolysis cell coupled system with co-cultivation of *Geobacter* and *Methanosarcina*. *J. Environ. Sci.* 42, 210–214.
- Yoho, R.A., Popat, S.C., Torres, C.I., 2014. Dynamic potential-dependent electron transport pathway shifts in anode biofilms of *Geobacter sulfurreducens*. *ChemSusChem* 7 (12), 3413–3419.
- Yoho, R.A., et al., 2015. Anode biofilms of *Geobacter ferrihydriticus* exhibit electrochemical signatures of multiple electron transport pathways. *Langmuir* 31 (45), 12552–12559.
- Yong, Y.-C., et al., 2012. Macroporous and monolithic anode based on polyaniline hybridized three-dimensional graphene for high-performance microbial fuel cells. *ACS Nano* 6 (3), 2394–2400.

- Zeng, X., Borole, A.P., Pavlostathis, S.G., 2015. Biotransformation of furanic and phenolic compounds with hydrogen gas production in a microbial electrolysis cell. *Environ. Sci. Technol.* 49 (22), 13667–13675.
- Zhang, Y., et al., 2011. Bioreactor technology in marine microbiology: From design to future application. *Biotech. Adv.* 29 (3), 312–321.
- Zhang, J., et al., 2013. Effects of ferric iron on the anaerobic treatment and microbial biodiversity in a coupled microbial electrolysis cell (MEC) – Anaerobic reactor. *Water Res.* 47 (15), 5719–5728.
- Zhang, J., et al., 2014a. Enhanced performances of microbial fuel cells using surface-modified carbon cloth anodes: A comparative study. *Int. J. Hydrogen Energy* 39 (33), 19148–19155.
- Zhang, B., et al., 2014b. Nitrogen-doped activated carbon as a metal free catalyst for hydrogen production in microbial electrolysis cells. *RSC Adv.* 4 (90), 49161–49164.
- Zhang, L., et al., 2017. Startup performance and anodic biofilm distribution in continuous-flow microbial fuel cells with serpentine flow fields: Effects of external resistance. *Ind. Eng. Chem. Res.* 56 (14), 3767–3774.
- Zhang, Y., et al., 2019. Microbial fuel cell hybrid systems for wastewater treatment and bioenergy production: Synergistic effects, mechanisms and challenges. *Renew. Sustain. Energy Rev.* 103, 13–29.
- Zhao, N., Angelidaki, I., Zhang, Y., 2018. Current as an indicator of ammonia concentration during wastewater treatment in an integrated microbial electrolysis cell-nitrification system. *Electrochim. Acta* 281, 266–273.
- Zhao, Y., et al., 2010. Three-dimensional conductive nanowire networks for maximizing anode performance in microbial fuel cells. *Chem. Eur. J.* 16 (17), 4982–4985.
- Zhao, L., et al., 2014. Modeling of polarization losses of a microbial fuel cell. In: ASME 2014 12th International Conference on Fuel Cell Science, Engineering and Technology Collocated with the ASME 2014 8th International Conference on Energy Sustainability. American Society of Mechanical Engineers.
- Zhen, G., et al., 2017. Microbial electrolysis cell platform for simultaneous waste biorefinery and clean electrofuels generation: Current situation challenges and future perspectives. *Prog. Energy Combust. Sci.* 63, 119–145.
- Zheng, Y., et al., 2014. Toward design of synergistically active carbon-based catalysts for electrocatalytic hydrogen evolution. *ACS Nano* 8 (5), 5290–5296.
- Zhou, T., et al., 2017. Microbial fuels cell-based biosensor for toxicity detection: A review. *Sensors* 17 (10), 2230.
- Zhu, X., Yates, M.D., Logan, B.E., 2012. Set potential regulation reveals additional oxidation peaks of *Geobacter sulfurreducens* anodic biofilms. *Electrochem. Commun.* 22, 116–119.
- Zhu, X., et al., 2014. Microbial community composition is unaffected by anode potential. *Environ. Sci. Technol.* 48 (2), 1352–1358.
- Zhuang, L., et al., 2010. Enhanced performance of air-cathode two-chamber microbial fuel cells with high-pH anode and low-pH cathode. *Bioresour. Technol.* 101 (10), 3514–3519.
- Zhuang, L., et al., 2012. Long-term evaluation of a 10-liter serpentine-type microbial fuel cell stack treating brewery wastewater. *Bioresour. Technol.* 123, 406–412.