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Energy from Plants and Microorganisms: Progress in Plant–Microbial Fuel Cells

Huan Deng,[a, b] Zheng Chen,[b] and Feng Zhao*[a]

Plant–microbial fuel cells (PMFCs) are newly emerging devices, in which electricity can be generated by microorganisms that use root exudates as fuel. This review presents the development of PMFCs, with a summary of their power generation, configurations, plant types, anode and cathode materials, biofilm communities, potential applications, and future directions.

1. Introduction

Microbial fuel cells (MFCs) produce electrical energy through the degradation of substrates (pollutants) by living microorganisms.[1] In addition to power generation, MFCs can be used to carry out specific tasks, such as treating waste water and monitoring pollutants.[2] MFCs have also been developed to accommodate various conditions and, thus, have expanded their applications. One example is a sediment microbial fuel cell (SMFC), in which the anode is embedded in the sea floor or freshwater sediment while the cathode is located in overlying, oxygen-rich water (Figure 1a).[3,4] Substrates in the sediment act as a fuel for the anode reaction. An SMFC has been used to power hydrophones to track marine animals for three months.[5] Plant–microbial fuel cells (PMFCs) are emerging derivative devices of MFCs. Organic substrates are mostly supplied by the root deposits of aquatic plants in PMFCs and they are degraded by using bacteria on the anode surface to generate electricity. The classical configuration of a PMFC is shown in Figure 1b. The anode is submerged in a support matrix near the plant roots to obtain the exudates as fuel, and the cathode position is similar to that of SMFCs.

The superiority of PMFCs is sustainability as the carbon substrate flow is achieved through continuous input by roots. Considering that root exudates account for approximately 20% of photosynthesized carbohydrates,[6] PMFCs carry out the conversion of solar energy into electrical power. The conversion efficiency (*CE*) of solar energy into electrical energy can be estimated by using Equation (1).

$$CE = Pe \times Rp \times Ra \times Er \quad (1)$$

The theoretical maximum limit of photosynthetic efficiency (*Pe*) is estimated to be 4.6 and 6.0% for C3 and C4 plants, respectively.[7] However, these limits are hardly achieved. The conversion efficiency in the growth phase comes within 70% of these limits, thus 3.2 and 4.2% are assigned to the *Pe* of C3 and C4 plants, respectively.[8] There is a 20% ratio of root deposit to photosynthesized carbohydrates (*Rp*),[6] 30% rhizodeposit availability for microorganisms (*Ra*),[9] and 9% microbial fuel cell energy recovery (*Er*).[10] Our estimations of the *CE* for C3 and C4 plants are 0.017 and 0.022%, respectively.

The average annual solar irradiation on a horizontal surface (*As*) is approximately 170 $W m^{-2}$, and an average growth season (*Gs*) is assumed to be six months.[11] The prospective annual gross PMFC outputs are $As \times Gs \times CE = 1266$ and 1638 $kWh ha^{-1}$ for C3 and C4 plants, respectively.

Increasing the *Er* value is a promising way to improve the *CE* value. To illustrate the factors that affect the *Er* of PMFCs, Equation (2) describes the relationship between *Er* (denoted as Er_{PMFC}) and power density:

$$Er_{PMFC} = \frac{\text{power density} \times \text{anode area}}{As \times \text{leaf area} \times Pe \times Rp \times Ra} \quad (2)$$

Improvement of the leaf area, *Pe*, *Rp*, and *Ra* is limited for a fixed plant type, and *As* cannot be changed too much at a specific location. However, the power density can be improved by adopting a suitable PMFC configuration, high performance electrode, catalyst, and microorganism.[12]

Studies on PMFCs are still in their infancy; the earliest papers addressing PMFCs were published in 2008.[10,11] Current studies show that PMFCs with freshwater plants can generate a maximum power density ranging from several to dozens of $mW m^{-2}$, whereas that with the seawater plant *Spartina anglica* (*S. anglica*) can generate a maximum power density of 222 $mW m^{-2}$.[13] With the aim of introducing PMFC techniques in this paper, we seek promising ways to improve the performance and to broaden their applications. The character of current PMFCs is analyzed, progress is addressed, and a possible future development is proposed.

[a] Dr. H. Deng, Prof. F. Zhao
Key Laboratory of Urban Environment and Health
Institute of Urban Environment
Chinese Academy of Sciences
1799 Jimei Road, Xiamen, P.O. 361021 (P.R. China)
E-mail: fzhao@iue.ac.cn

[b] Dr. H. Deng, Dr. Z. Chen
Research Center for Eco-Environmental Sciences
Chinese Academy of Sciences
1799 Jimei Road, Xiamen, P.O. 361021 (P.R. China)

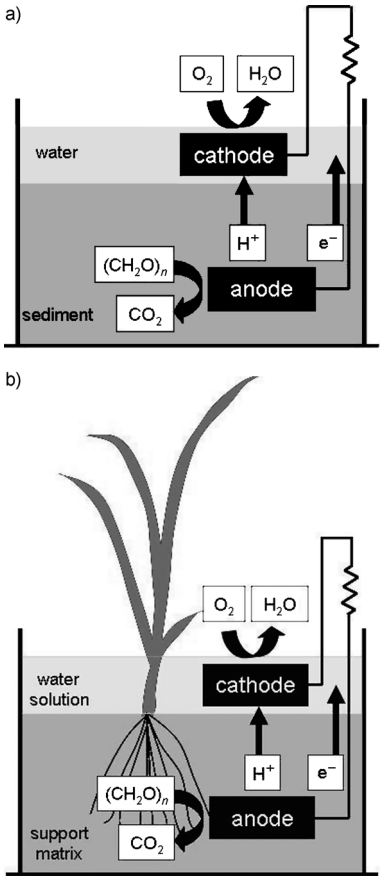

Figure 1. a) Electricity production in sediment MFCs. Electrons are produced at the anode from the anaerobic degradation of substrates by bacteria in the sediment and transferred through an external circuit to the acceptors (e.g., oxygen) at the cathode, while a charge-balancing number of proton/ions are transferred between the anode and cathode. b) Electricity production in PMFCs. The organic substrates are mostly from root exudates, and electricity is generated by the anaerobic degradation of the root exudates by bacteria.

2. Plants and Substrates

To date, aquatic plants are the only choices for PMFCs, and these include reed manna grass (*Glyceria maxima*),[11] rice plant,[14,15] *S. anglica*, *Anisogramma anomala*, and *Arundo donax*.[13,16] Selecting the most suitable plant is a promising way to increase electricity output. Aquatic plants are sensitive to solution conductivity, for example, rice growth is negatively affected at conductivities under 0.6 $S\,m^{-1}$.[17] The conductivity of the electrolyte can be improved by an increase in salinity when using seawater plants in PMFCs.[13] In addition, rice has to be harvested after four months; utilizing perennial aquatic plants in the system will decrease the installation costs for the electrodes and elongate power generation.

Root deposits comprise water-soluble exudates: secretions, lysates, gases, and mucilage. Within exudates, carbohydrates, carboxylic acids, and amino acids are highly degradable to microorganisms[18] and, therefore, the most responsible for electron donation. Bacilio-Jiménez et al. tested the composition of carbohydrates, carboxylic acids, and amino acids in rice root exudates and found that glucose comprises approximately 90% (mol/mol) of the total amount of components.[19] Glucose is one of the most suitable substrates for MFCs, and glucose-fed MFCs generate the highest power density.[20] However, coulombic efficiency in glucose-fed MFCs is the lowest (15%) compared to that in acetate-fed (72%), butyrate-fed (43%), and propionate-fed MFCs (36%). A possible reason is that glucose can be anaerobically degraded by non-electricity-generating bacteria, and thus a large proportion of electrons are not transferred to the anode.

In addition to root deposits, soil is another source of electron donors, if it is used as the support matrix. Inorganic elements in soil can generate electrons by anaerobic respiration or chemical processes. The reactions that generate electric power include i) the chemical oxidation of microbially produced reductants, such as humic acids, iron(II), and sulfur compounds, ii) the microbial oxidation of sulfur to sulfate,[21] iii) the oxidation of ammonia to nitrite or nitrate by ammonia-oxidizing bacteria,[22] iv) the conversion by self-supporting bacteria of carbonate into organic carbon, which is further degraded to generate electrons.

Aquatic plants survive anoxic conditions by supplying their root systems with oxygen from the atmosphere. The transportation of oxygen in the plant is facilitated by a tissue named aerenchyma.[23] The oxygen transport rates per day in different plants varies from several to dozens of mg of oxygen per g of root biomass.[24,25] For rice plants, the released oxygen dramatically increases the potential within a distance of 4 mm from the root tip.[26] It may hamper the performance of PMFCs as electrons are generated through anaerobic respiration in the anode region. Considering that the released oxygen can be used as an electron acceptor, Chen et al. set up a PMFC (Figure 2b) with a cathode in the rhizosphere to utilize the released oxygen from the roots as the electron acceptor while an anode was located beneath the root.[27] This type of PMFC can act as an in situ biosensor to monitor the oxygen concentration in the soil.

The release of rhizodeposits and oxygen is determined by sunlight,[28] temperature,[18,29] and growth stage.[30] Changes in these factors may lead to a fluctuation of current and voltage in PMFCs. Kaku et al. found that electricity generation was sunlight dependent and artificially shading plants in the daytime inhibited electricity generation[31] because of the reduction in photosynthesis and root exudates. The addition of acetate, which is one of the major root-exuded organic compounds, enhanced electricity generation in the dark.

3. Configuration

To minimize both inert resistance and oxygen diffusion, two-chamber PMFCs have been developed (Figure 2a). In this configuration, a membrane was fixed at the bottom of the anode chamber with one side in contact with the sediment, in which the anode was located near the roots, and the cathode was located on the other side of the membrane.[11,13] However, the membrane is the primary cost for PMFCs.[32]

The principle of PMFCs with a single chamber is that the oxygen concentration declines over the depth of water and

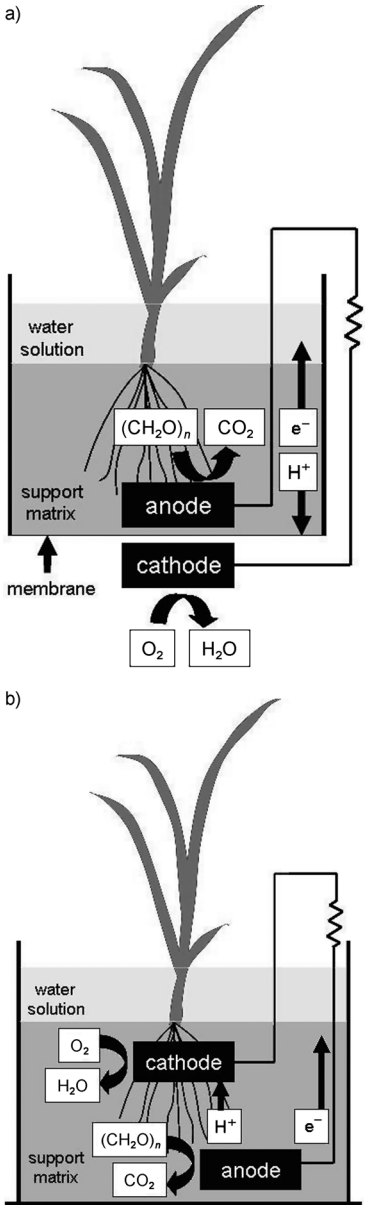

Figure 2. Modified configurations of PMFCs. a) A configuration with a membrane fixed at the bottom with one side in contact with the sediment, in which the anode is located, and the cathode is located on the other side of the membrane. b) Oxygen released from the root can be used as the electron acceptor at the cathode in a single-chamber PMFC.[27]

sediment, and thus a membrane is unnecessary.[33] The support matrix, in which the anode is submerged, is flooded soil or other materials, such as vermiculite or graphite granules.[10,13] Soil blocks the migration of H^+ from the anode to the cathode and the diffusion of the root deposit to the anode surface, which increases mass-transfer resistance and ohmic resistance.[15] There have been two forms of single-chamber configuration: one in which the cathode is located in overlying water to use oxygen from air for reduction reactions (Figure 1 b), and the other in which the cathode is located in the rhizosphere to use oxygen released from the roots as the electron acceptor (Figure 2 b). This membrane-free configuration reduces the capital cost and can decrease mass-transfer and ohmic resistances; hence, it shows a potential to achieve practical implementation in combination with a high performance.

4. Electrodes and Catalysts

Graphite felt is one of the most used materials as electrodes in the lab.[34] Helder et al. used smaller graphite granules as the anode to achieve a higher power output in a PMFC owing to a higher effective surface area, which might increase contact between the microorganisms and graphite, thus inducing lower internal resistance and higher power density.[13] Development in treated anode and cathode materials provides a promising way to increase power generation. The treatment strategy comprises the addition of a functional group or catalyst on the electrode to facilitate electron transfer from the bacteria to the anode surface[35] and from the cathode to the acceptors,[36] and structural alteration of the anode and cathode surfaces to accommodate more bacteria to transport electrons.[37] Scaling up PMFCs is inevitable in the future.[38] Unlike lab studies, in which the performance of the electrode reaction is usually the most emphasized, other properties of electrode materials should also be considered, which include cost, longevity, and sustainability.[39,40]

Oxygen is the most used acceptor of electrons for the cathode reduction reaction in PMFC studies, although a higher power output was obtained when ferricyanide acted as an electron-transfer mediator.[16] However, ferricyanide and other redox mediators, such as thionine, methyl viologen, and neutral red, are toxic and may become pollutants in the environment. Oxygen is the most sustainable electron acceptor because of its inexhaustible availability.[41] Future studies on PMFCs should consider methods to reduce the activation energy by using chemical or biocatalysts. As platinum is expensive and can be easily poisoned by environmental substrates, cheaper catalysts have been intensively studied and applied;[42] phthalocyanine and tetramethylphenylporphyrin[43] have been proposed to replace platinum in MFCs. Biocatalysts, which comprise bacteria developed on the cathode surface, have been reported to decrease the critical oxygen concentration from 5.5–6.6 to 0.1 mg L^{-1} and improve the affinity of oxygen for the cathode.[44]

5. Microbial Community on the Electrode

The formation of a biofilm on the anode surface is essential for the efficient transfer of electrons in MFCs[45] and the reduction of the overpotential of the cathode to facilitate electron transport to the acceptors. Kaku et al. sequenced 16S rRNA DGGE bands that appeared in a PMFC.[31] Their results showed that these sequences on the anode are associated with *Natronocella acetinitrilica*. The uncultured *Beijerinckiaceae* bacterium clone GASP-38KB-490-B06 and the *Rhizobiales* bacterium A48 on the cathode was closely related to *Rhodobacter gluconicum*. As for sequences on the anode, *Natronocella acetinitrilica* belongs to the *Ectothiorhodospiraceae* family and the other two mentioned above are *alpha-proteobacteria*. Previous studies have shown that electricigens mostly belong to *Shewanella*,

Proteobactor, *Pseudomonas*, *Firmicutes*, *Acidobacteria*, and the Fungi kingdom.[14,46,47] Thus, it can be assumed that the uncultured *Beijerinckiaceae* bacterium clone GASP-38 KB-490-B06 and the *Rhizobiales* bacterium A48 transfer electrons to the anode. *Rhizobiales* bacterium A48 may transfer electrons through a ferric redox reaction.[48] *Natronocella acetinitrilica*, which is a heterotrophic bacterium isolated from lake sediment,[49] may not be electricigenic.

Bacteria on the cathode surface can be grouped into two types: i) aerobic microorganisms that use oxygen as the oxidant and assist the oxidation of transition metal compounds, such as those of manganese(II) or iron(II) for electron delivery to oxygen, and ii) anaerobic microorganisms that use compounds, such as nitrate, sulfate, iron, manganese, selenate, arsenate, uranate, fumarate, and carbon dioxide as terminal electron acceptors.[44] *Rhodobacter gluconicum* is an anaerobic *Rhodobacter* sp., and its formation on the cathode surface can be attributed to the submersion of the cathode in water. The role of this bacterium in electron transport is not yet known. However, other *Rhodobacter* sp., such as *Rhodobacter sphaeroides*, have been shown to degrade organic substrates and generate hydrogen under anaerobic conditions.[50]

In a study of rice PMFCs, da Rosa[14] compared microbial communities on anodes in different inoculums (vermiculite, potting soil, and rice field soil) and found that the community composition varied with inoculums. *Desulfobulbus*- and *Geobacter*-related populations on the anode surfaces were closely related to current production in potting soil and rice field soil, respectively. *Delta-proteobacterial Anaeromyxobacter* sp., unclassified *Delta-proteobacteria*, and anaerolineae were also part of the anode biofilm in rice field soil, and these populations might play a role in current production. In rice field soil, bacteria that belong to *Beta-proteobacteria* are relevant to root exudate degradation, whereas the main current-producing bacteria that belong to *Delta-proteobacteria* were not able to assimilate root exudates. This result indicates that the root exudate degrading bacteria may not be directly involved in electricity generation because different pathways are involved in electron transport: direct current generation from organic substrates, indirect oxidation of plant substrates with mediated electron transfer (e.g., sulfide), or yet unknown mechanisms. This study, however, did not illustrate which pathway these populations transport electrons through, that is, direct current generation from organic substrates or indirect oxidation of plant substrates with electron shuttling.

The identification of electricigens requires the isolation of bacteria in a pure culture and a power output test.[51,52] However, probably because of unculturability, only approximately 30 strains have been isolated and shown to generate power in pure culture.[14] Of these strains, most were isolated from MFCs and only a few from SMFCs. In PMFCs with flooded soil as the support matrix, it can be expected that new electricigens will be isolated because soil has a high microbial diversity and different microbial composition.

6. Outlook

6.1. Energy supply

We have estimated the capital costs of both, MFCs and PMFCs. Together, the reactor and membrane for MFCs are more than 78% of the total cost (Figure 3). However, PMFCs can operate without membranes and even the reactor is unnecessary when running in wetland. It currently costs approximately 70 USD for operating a 1 m^3 setup of a PMFC in laboratory systems. To achieve practical implementation, the capital costs of PMFCs have to be reduced so that large-scale applications can be targeted. Compared to expensive graphite felt, utilizing dense networks and highly conductive materials (e.g., carbon cloth) for the electrodes is a way to reduce the cost.

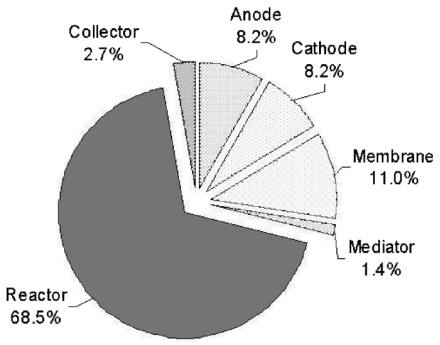

Figure 3. Comparison of the estimated costs of laboratory systems. The following cost assumptions were used: the reactor is constructed from oroglas materials, and operational costs are approximately 250 USD for a 1 m^3 cubic MFC, the anode and cathode (1 m^2, graphite felt) each cost 30 USD, the ion-exchange membrane (1 m^2) costs 40 USD, the collector (titanium wire) costs 10 USD; and the nanocatalyst or mediator (phthalocyanine or ferricyanide) cost 5 USD.

The development of PMFCs may offer energy supplies in remote areas, where power is not available, or serve as an auxiliary energy source. PMFCs can also be developed to power scientific instruments for monitoring and tracking wildlife and as biosensors for testing pollutants in wetlands, such as biochemical oxygen demand.[53]

6.2. Pollutant and greenhouse gas removal

Studies on waste-water MFCs showed that ammonium can be oxidized in the anode chamber[22] and nitrate can be reduced to nitrogen in the cathode chamber,[54,55] which results in the removal of nitrogen pollutants. Therefore, we think that PMFCs could be applied in wetlands to accelerate the removal of nitrate and ammonium concentration in waste water. For PMFCs, the nitrogen conversion is expected to occur near the anode and cathode.

PMFCs can be used to reduce methane emission in wetland and paddy fields. It has been reported that, by submerging an anode in rice field soil, a reduction in methane emissions of up to 50% was observed compared to open circuit controls.[14]

This improvement can be explained by competition with common electron donors such as acetate. We also suppose that some methanotrophs may degrade methane and generate electricity so that methane emission is reduced.

To date, the power generated by PMFCs is not competitive. However, PMFCs represent a new technique to help reduce globing warming as they can decrease methane emissions, which will attract attention and may be a core application in the future, especially in combination with pollutant removal.

6.3. Impact of PMFC operation on the environment

Root exudates are functionally important for plant physiology. Organic acids can improve nutrient availability, and phenolic compounds in root exudates may counteract infection by root pathogens and prevent the microbial degradation of exudate compounds that are involved in phosphorus mobilization. Root flavonoids have been identified as signaling molecules for spore germination and hyphal growth of arbuscular mycorrhiza and ectomycorrhiza.[56] The microbial degradation of these compounds is likely to be accelerated by the introduction of an anode, which acts as an oxidant; thus, it is necessary to investigate the impact of PMFCs on the rhizospheric environment and plant physiology as well as the microbial community.

More diverse communities increase the odds that ecosystem functions remain stable against climate and physiology fluctuation. Increased plant diversity by mixing with shade-requiring, hardy, or evergreen plants can improve the resistance of community electricity against the fluctuation of light intensity and temperature.

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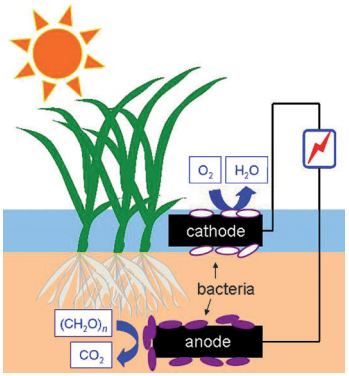
Down to the roots: Plant–microbial fuel cells convert solar energy into electrical power by using microorganisms, which degrade root exudates and pollutants at the anode and pass the electrons to acceptors at the cathode. This setup can provide auxiliary power while reducing the emission of greenhouse gas, that is, methane, from fields.



*H. Deng, Z. Chen, F. Zhao**

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Energy from Plants and Microorganisms: Progress in Plant–Microbial Fuel Cells

