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REVIEW ARTICLE

Overview on Electricigens for Microbial Fuel Cell

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Abstract: Microbial fuel cell is a bio-electrochemical system that drives a current by using bacteria and mimicking bacterial interactions found in nature. The electricigens and its activity have important influence on the power generation capacity and organic matter degradation ability of MFC system. The common types of electricigens and their application situation in microbial fuel cell are summarized in this paper, the basic characteristics and electron transfer mechanism of some major microorganisms are described, so as to provide some ideas for electricigens' selection, transformation and optimization of a variety of strains to improve the power generation capacity and organic matter degradation ability of MFC system.

Keywords: Degradation ability, Electricigens, Microbial fuel cell, Power generation capacity, Wastewater treatment.

1. INTRODUCTION

Depletion of conventional energy sources and also its negative effect on environment has led many researchers to look for alternative energy sources. Developing carbon-neutral renewable energy sources is an important research area for alternative power systems.

Microbial fuel cell (MFC) is a kind of biological chemical reaction device that can directly convert the chemical energy stored in organic matter into electrical energy by using microbial metabolic activity. MFC produces electricity while purifying wastewater and cause hardly any pollution to the environment during the power generation process. It is regarded as a new wastewater treatment process and a new green power generation technology which has the advantages of high efficiency, low energy consumption, clean and environmental protection [1 - 4].

Electricigens are a kind of microorganism which can completely oxidize the organic matter by using the electrode as the sole electron acceptor. They are essential for the bioelectrochemical processes in MFCs [5]. A typical MFC is a two-chambered system containing an anode chamber and a cathode chamber separated by an ion permeable membrane [6, 7]. Electricigens in the anode chamber oxidize organic matter to release electrons to the anode electrode. Electrons donated to the anode flow to the cathode through electrical wires, where they are reunited with the protons generated in the anode chamber and combine with oxygen or other electron acceptors to form reduced product.

Various types of microbes have been used in MFCs. A major challenge to the development of MFC as a novel electricity generating technology is harnessing the linkage of electricigens to the electrode substrate *via* electron transport. To prove up the species distribution, growth characteristics, metabolic characteristics and electricity mechanism of electricigens will contribute to the development of MFC.

This paper makes a simple overview on some common electricigens for MFCs.

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2. COMMON TYPES OF ELECTRICIGENS

2.1. *Escherichia coli*

Escherichia coli (*E. coli*) is one of the gram negative bacteria that can ferment most sugars, and it is facultative anaerobic. *E. coli* produces a large quantity of indole in a stationary phase, which functions as an extracellular signal to control diverse aspects of bacterial physiology. In MFC, indole was utilized as a carbon source in a mixture of culture and enhanced power generation in an MFC [8]. After domestication, power production capacity of *E.coli* increased significantly. *E coli* has been used as bacterial strain in many MFCs [9 - 13]. *E coli* can self-mediate the extracellular electron transfer to electrode in fuel cell through the electrochemically activated excretion of electron shuttling molecules, and it can catalyze the glucose oxidation in fuel cell anode in the absence of any artificial electron mediators [14]. Researches show that MFCs catalyzed by the evolved or engineered *E. coli* deliver much higher power density, reduce operation expense and eliminate toxic effects of artificial mediators [15].

E. coli has the advantages of easy access, easy cultivation, low cost, safety, and metabolizing a variety of substrates. The use of *E. coli* as the electricigens of MFC has great application prospects.

2.2. *Shewanella*

Shewanella are well known for the diversity of terminal electron acceptors they can reduce and are one of the primary families of bacteria used in MFCs [16], and their capacity to release electron shuttling out of cells, which help them transfer electrons to electrodes without direct contact. *Shewanella* are facultative gram-negative anaerobes inhabiting a wide variety of niches in nature where they degrade diverse organic compounds and utilize a broad spectrum of electron acceptors for growth under anaerobic conditions.

Shewanella have become one of the most important electricity producing microorganisms due to their respiratory type diversity [17]. Bacteria of the groups Shewanellaceae and Geobacteraceae are classic models in MFC research because of the breadth of knowledge about their metabolism and versatility [18]. *Shewanella* can generate nano wire to transfer the electron to the electrode directly, so the anode has a high conductivity even if multilayer cells biofilm accumulate on it [19].

Shewanella fuel cell is the earliest available mediator-less microbial fuel cell. In 1999, Kim *et al.* used *Shewanella* in MFC and realized electricity production in the absence of exogenous mediators, which lift the curtain on researching the mediator-less MFC [20]. The importance of *Shewanella* as potential bioelectrochemical catalysts for MFC technology continues to drive research into the adaptability of *Shewanella* for power production. Various *Shewanella* strains were developed as electrogenic biocatalysts in MFCs, and intensive studies were conducted to understand the mechanism of electron transfer to electrode using *Shewanella oneidensis* MR-1 as a model microorganism [21].

2.3. *Enterococcus faecalis*

Enterococcus faecalis (*E. faecalis*) is generally defined as a class of gram positive bacteria isolated from feces and used for isolation and identification of nitrogen sodium. *E. faecalis* is facultative anaerobic. Xie separated a strain of *E. faecalis* from acclimation mangrove sediment, and its characteristics of electricity generation were investigated. The experimental results show that the electricity generation capacity of MFC with *E. faecalis* is higher than that of MFC with *Shewanella* [22]. Cui studied the characteristics of electricity generation by *E. faecalis*, and validated the optimum growth temperature, and showed that the contact of bacteria and electrode is the essential factor of electricity production [23]. In Zhang *et al.*'s study, *E. faecalis* was employed in microbial fuel cells, bacterial biofilms formed by *E. faecalis* were investigated with respect to electricity production, and showed that trace riboflavin was essential for transferring electrons in the absence of other potential electron mediator. *E. faecalis* can resist the external environment, and has strong resistance and adaption to temperature, even can resist many kinds of antibiotics [24].

2.4. *Geobacter*

Geobacter (*G.*) is a very important kind of electricigens, and it is gram-positive anaerobes. Amongst many known bacteria which can produce electricity, the most successful as of today are the *Geobacter* species [25]. Researches show that if a platinum or graphite electrode is inserted into seawater sediments and anther electrode linked together with it is inserted in water with rich dissolved oxygen, a continuous current will produce. This is because *Geobacter* is highly enriched on the electrode [26 - 28]. Some *Geobacter* species also showed direct electron transferring properties [29], they can completely oxidize electron donor by using electrode as the only electron acceptor [30]. So far, *Geobacter* which was found to be able to use electrode as the only electron acceptor contains *G. Metallireducens, Desulfuromonas acetoxidans, G. sulfurreducens, G. psychrophilus and Geopsychrobacter electrodiphilus* [31]. Because the testing of the complete genome sequence of *G. sulfurreducens* has been completed, *G. sulfurreducens* is generally used as a model strain for microbial fuel cell research.

2.5. *Sulfate Reducing Bacteria*

Sulfate-rich waters are often found around the world as waste products from many mining, industrial processes or natural chemical reaction. The *sulfate reducing bacteria* (*SRB*) can convert sulfate in anaerobic environment. Under anaerobic conditions, dissimilatory *SRB* use sulfate as a terminal electron acceptor for the degradation of organic compounds, and the potential possibilities for conversion and storage of the energy obtained by *SRB* are more and more attractive [32]. *SRB* are heterotrophic bacteria that consume organic compounds as carbon and energy sources. A few studies proposed the use of *SRB* to remove sulfate from wastewater with MFC [33]. Zhao *et al.* have developed a MFC for removal of sulfur-based pollutants and for simultaneous wastewater treatment and electricity generation [34], and they also have clarified that the generation of electricity in MFC using *SRB* is mainly from the oxidation of biologically produced sulfide to elemental sulfur [35].

2.6. *Rhodoferax ferrireducens*

Rhodoferax ferrireducens (*R. ferrireducens*) is able to metabolize sugar into electricity conversion, and conversion efficiency can reach above 80%. *R. ferrireducens* is a facultative gram-negative anaerobic iron reducing microorganisms, which can directly transfer electrons to the electrode without requirement of a catalyst [36]. Compared with other MFCs, the most important advantage of *R. ferrireducens* MFC is that it transforms sugars into electrical energy effectively, thus greatly promotes the practical application process of microbial fuel cells. Liu *et al.* selected *R. ferrireducens* as electricigens, and studies the effective degradation of the sugar microbial biomass and the electricity generation performance of the MFC. Through the recycling experiment and biomass utilization experiment, the MFC constructed by *R. ferrireducens* is verified to be similar to an ideal two rechargeable battery [37]. Li *et al.* also constructed a MFC by using *R. ferrireducens* as electricigens, and experiments showed that the *R. ferrireducens* MFC has the advantages of normal temperature power generation, good cycle and so on [38].

2.7. *Saccharomyces cerevisiae*

Saccharomyces is widely distributed in nature as a kind of fungus. According to its distribution, Saccharomyces suits to grow in acidic environment with high sugar, has a strong resistance to osmotic pressure, a good resistance to high concentrations of sulfate, and resistance to high ammonia nitrogen. Thus MFC using *Saccharomyces* as electricigens is promising for treating high concentration organic wastewater. In MFC, *Saccharomyces Cerevisiae* is the most used *Saccharomyces.* Fatemi *et al.* used *Saccharomyces cerevisiae* as a pure culture in MFC [39]; Mathuriya *et al.* demonstrated the comparative electricity production capacity of *Saccharomyces Cerevisiae* and *Clostridium acetobutylicum* in two chambered MFC using artificial wastewater, and drew a conclusion that both *Saccharomyces cerevisiae* and *Clostridium acetobutylicum* are good candidate for bioelectricity production *via* MFC technology [40]; Rossi *et al.* found that the rate of substrate consumption by *Saccharomyces Cerevisiae* in the anaerobic compartment of a dual chambered MFC presented a great potential to generate electrons in a microbial fuel cell [41]; Arbianti *et al.* testified that microbial cultures of *Saccharomyces cerevisiae* can be used to produce electrical energy alternatives in MFC system, and also found that the media glucose yeast extract (GYE) is the most optimum medium for growth of *Saccharomyces cerevisiae* [42]; Gunawardena's research also show that *Saccharomyces cerevisiae* performed favorably as the bio-catalyst in a glucose powered microbial fuel cell [43]. In Rahimnejad's experiments, several microorganisms such as *Pseudomonas putida*, *Saccharomyces cerevisiae*, *Lactobacillus bulgaricus*, *Escherichia coli* and *Aspergillus niger* were cultured in an anaerobic chamber for the generation of electrons. The rate of substrate consumption in the anaerobic compartment indicated that *Saccharomyces cerevisiae* had the great potential to generate electrons [44].

2.8. *Pseudomonas aeruginosa*

Pseudomonas aeruginosa is widely distributed in nature, and is one of the most common bacteria in the soil. *Pyocyanine* excreted by *Pseudomonas aeruginosa* can be used as the electronic intermediaries for anodic reaction. *Pyocyanine* can be reduced by bacteria, and the reduced *Pyocyanine* will transfer electrons to the electrode to oxidize. The process will continue to cycle, and then electron transfer between the electrode and the thalli is achieved [45]. When using *Pseudomonas aeruginosa* to degrade quinolone, the MFC is more effective than the conventional anaerobic culture method [46]. Study clearly indicates that *Pseudomonas aeruginosa* can metabolize chicken feathers as a source of carbon and nitrogen, and chicken feathers can be successfully employed for electricity generation using MFC technology [47]. The optimal operating conditions of MFC with *Pseudomonas aeruginosa* F026 as electricigens are derived by experiments as: soluble starch as substrate, running temperature 35°C, and pH was neutral and alkaline [48], and tests with pure cultures have shown that the addition of these compounds in MFCs can increase power [49].

2.9. Some other Electricigens

Some other electricigens used in MFC can also be found in reports. For example, the MFC based on the *Fe(III)-reducing bacteria* has the advantages as there is no need of any additional medium, a variety of organic electron donors can be used as fuel, higher energy conversion efficiency [50]; the *Lactobacillus bulgaricus* MFC system produced quantifiable amounts of electricity [51]; *Ochrobactrum anthropic* produced current using a wide range of substrates, including acetate, lactate, propionate, butyrate, glucose, sucrose, cellobiose, glycerol, and ethanol [52]; the mechanism of electron transfer in an MFC system was studied by using *Klebsiella pneumoniae* as biocatalyst [53]; *Geopsychrobacter electrodiphilus* may serve as important model organism for further elucidation of the mechanisms of microbe-electrode electron transfer in sediment fuel cells [54], *Clostridium butyricum* can produce electricity by using starch, molasses, glucose and lactic acid [55].

The basic characteristics of some common electricigens for MFC are shown in Table **1**.

Table 1. Basic characteristics of some common electricigens.

Electricigens	Aerobic/anaerobic	Gram staining	Electron transfer
E. coli	facultative anaerobic	negative	direct
Shewanella	anaerobic	negative	indirect
E. faecalis	facultative anaerobic	positive	direct
Geobacter	anaerobic	positive	direct
SRB	anaerobic	negative	direct
R. ferrireducens	facultative anaerobic	negative	direct
Saccharomyces cerevisiae	anaerobic	/	direct
Pseudomonas aeruginosa	aerobic	negative	direct

3. ELECTRON TRANSFER MECHANISM OF ELECTRICIGENS

3.1. Electron Transfer from the Inside to the Outside of the Cell

The electron transfer from the cell to the extracellular involves two steps: (1) electron transfer from the cytoplasma membrane to the outer membrane of the cell; (2) Electron transfer from the outer membrane to the extracellular electron acceptor. Most of the electricigens, such as *Geobacter*, can directly oxidize small molecules organic acids by dehydrogenase on its membrane and transfer the released electrons to the electron acceptor on the membrane. Some other electricigens, such as *R. ferrireducen*, *Shewanella*, can oxidize organic compounds such as sugars to generate reducing power (NADH); subsequently, the electrons are transferred from NADH to the electron transport chain (metabolic respiratory chain) under the action of NADH dehydrogenase, which is then released to the outside of the cell by the respiratory chain. At present, NADH dehydrogenase, quinones, flavine enzyme and cytochrome C on the cell membrane are all key components of the electron transport system, and play an important role in the electron transfer process from the cell to the extracellular medium [56].

3.2. Extracellular Electron Transfer

It is generally regarded that there are two extracellular electron transfer (EET) mechanisms: direct electron transfer or indirect electron transfer.

3.2.1. Direct Electron Transfer

The direct electron transfer (DET) takes place *via* a physical contact of the bacterial cell membrane or a membrane organelle with the fuel cell anode, with no diffusional redox species being involved in the electron transfer from the cell to the electrode [57, 58]. This typically occurs over short distances and requires cells to be in close proximity to the electrode. The experimental results presented evidence for direct electron transfer by a gram-positive bacterium

Geobacter sp. [59]. The electron produced by the metabolism process can be transferred to the electrode through the cytochrome only if the bacteria that grow on the surface of the electrode can contact cytolemma to the surface of the electrode [60]. Electronic medium is a kind of inorganic or organic small molecule that can be involved in the oxidation and reduction system. When the medium is in a state of oxidation, it can be used as the electron acceptor for cell to send electrons to the electrode surface. Nano wires (Nanowire) can directly contact with solid iron oxide and pass the electrons from the bacteria to Fe (III). At present, the nanowires with electronic transmission functions have been found in many kinds of bacterial strain such as *Shewanella* [61], *Geobacter* [62, 63]. It is speculated that the nanowire is a widespread phenomenon in microorganisms.

3.2.2. Indirect Electron Transfer

Extracellular electron transfer achieved through the soluble endogenous redox mediator is regarded as the indirect electron transfer (IET) or mediated electron transfer (MET). There are a lot of endogenous electronic intermediary known, including phenazine, emodin, riboflavin, quinones, melanin *etc*. [64]. For example, *Pseudomonas aeruginosa* can transfer electrons to anode through electronic intermediary pyocyanin and phenazine-1-amide; *Shewanella* can use endogenous lutein as an electron mediator for electron shuttle [65].

Models for electron transfer mechanism are shown in Fig. (**1**) [66].

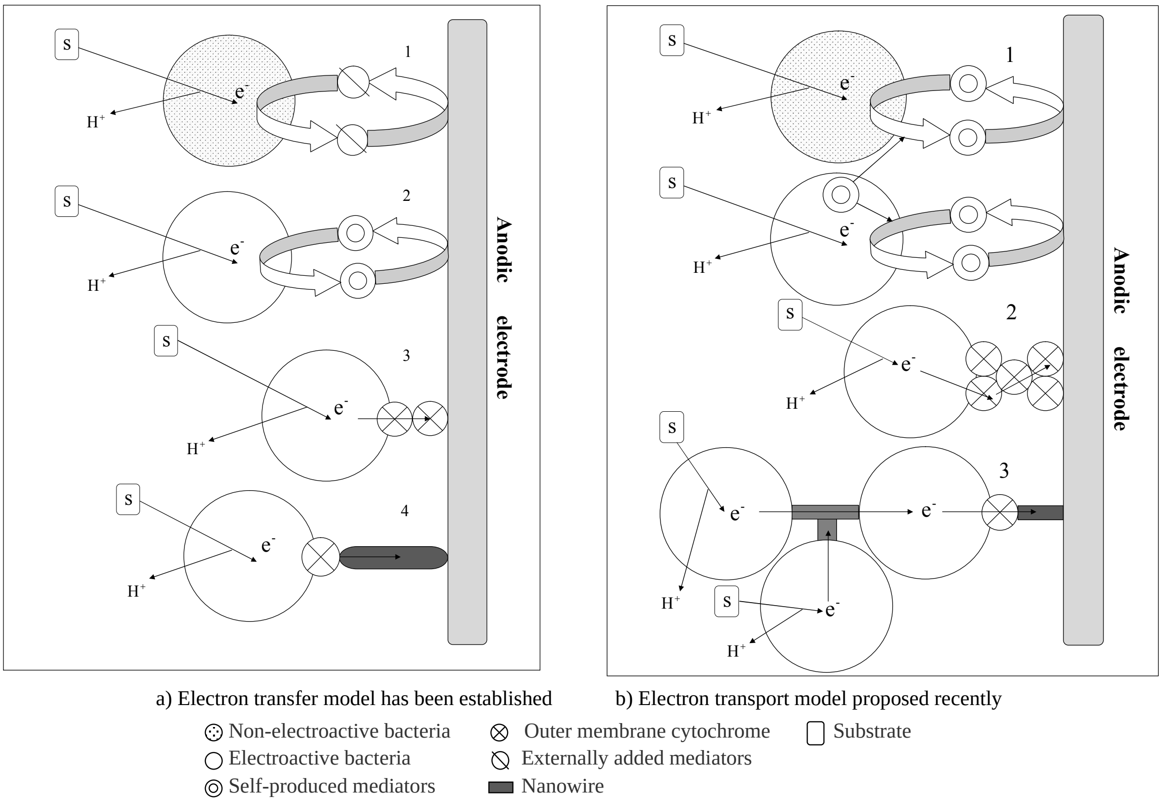

Fig. (1). Models for electron transfer mechanism in anode.

CONCLUSION

The ability of microorganism to produce electricity varies greatly, and every electron transfer path has its advantages and disadvantages. Deep understanding on electricigens and their electron transfer mechanism will help to provide more effective methods to improve the performances of MFCs. The future work will focus on finding or culturing more efficacious electricigens for using in MFC.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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