THE TECHNOLOGY OF MICROBIAL FUEL CELLS: A PROMISING APPROACH TOWARDS SIMULTANEOUS AND SUSTAINABLE WASTEWATER TREATMENT AND BIOELECTRICITY GENERATION

Musa Bishir[1,2,3*], Catalina Rodriguez Correa[1], Maciej Pawel Olszewski[1,2], Saleh Alhaji Ado[3], Marium Tariq[1], Andrea Kruse[1]

[1]Department of Conversion Technologies of Biobased Resources, University of Hohenheim, Germany
[2]Universität Hohenheim Verwaltung, Unterer Lindenhof, Germany
[3]Department of Microbiology, Ahmadu Bello University, Nigeria

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ABSTRACT

Microbial Fuel Cell (MFC) is a dual-edged technology which combines wastewater treatment with power generation. The chemical energy present in the organic matter of wastewater is usually transformed into electricity in MFCs through the action of electroactive bacteria on electrodes which act as catalysts. The MFC technology has piqued the interest of numerous researchers since it has the unique capability for wastewater treatment while concurrently generating some bioelectricity. When compared with traditional energy sources, MFCs have various advantages over other wastewater treatment processes, such as the trickling filter method and the commonly employed activated sludge process. These advantages include but are not limited to low energy requirements, low or no production of hazardous products, and mild reaction conditions. As a result, this wastewater treatment process is both cost-effective and long-term. The various designs of MFCs, electrode materials, and various wastewaters (substrates) that electrogenic bacteria could utilize are discussed in this work. The review gave succinct information on the components of microbial fuel cells, their operations as well as the stability and sustainability of the fuel cell systems.

Keywords: *microbial fuel cell, wastewater, substrate, electrodes, electroactive bacteria, bioelectricity*

INTRODUCTION

The global energy demand is steadily rising, and fossil fuels currently meet 80% of this demand. By 2050, experts have predicted that this demand will have risen by a factor of two compared to current levels [1]. Furthermore, the increased use of fossil fuels not only contributes to the depletion of finite resources but also has negative environmental consequences, which are already visible in the form of rising pollution levels and climate change around the world. As a result, major efforts are being made to produce bioenergy using alternative methods based on renewable resources, which will result in considerable reductions in carbon emissions to the environment [2]. The development of environmentally friendly ways for waste management and remediation, along with addressing climate change issues are crucial for sustainable development [3].

This framework requires lower energy treatment of wastewater, especially domestic and industrial wastewater. To address these aspects, an emerging bio-electrochemical concept and evolving technology are MFC. Fuel cells are electrochemical systems in which spatial separation of the oxidation and reduction reactions takes place to transform the chemical energy of a fuel into electricity. An MFC is, therefore, a bio-electrochemical device in which the natural metabolic activities of electroactive are harnessed to generate electricity by oxidizing organic materials into electrons. As opposed to the conventional fuel cell systems, which utilize fuel sources such as hydrogen, and primary alcohols (ethanol, methanol etc.), MFC principally produces electricity using biodegradable organic substrates via anaerobic oxidation. Microorganisms can convert several different chemicals that are biodegradable in nature into energy and other by products, such as CO_2 and water. MFC systems benefit from the energy that microorganisms provide, a medium for them to develop and carry out their metabolic functions. [4].

MFC technology is gaining attention and could be highly adaptable for sustainable wastewater treatment because; (1) as opposed to chemical fuel cells, microbial fuel cell systems are cost-effective because it operates under ambient temperature and pressure [5]; (2) It allows direct retrieval of energy; (3) better quality of effluent and low environmental footprints since both electrochemical and biological processes are combined [6]-[7]; (4) different types of wastewaters can be used as substrates in MFC. Hence, MFC shows the feasibility of treating wastewater by decreasing the Chemical Oxygen Demand (COD) with simultaneous electricity generation [8]. There are several factors upon which the performance of MFC systems depend these factors include, among others, the species of microorganisms involved and their type of metabolism, MFC design (single or double chamber), substrate type and its concentration,

*Corresponding author.
E-mail address : musa.bishir@uni-hohenheim.de; bishirmusa73@gmail.com

type of electrode material used, anode type and operating conditions, cathode type and pH buffer as well as the proton exchange membrane. This review, therefore, summarized the various components of microbial fuel cells and aimed at providing concise literature to fill knowledge gaps, especially on the stability and sustainability of the microbial fuel cell systems, which is hardly discussed in most review reports on MFCs.

MFC Design and Components

Typically, an MFC set-up has two chambers (anodic and cathodic chamber) and is known as a single or dual-chamber design. A single-chambered fuel cell, also known as an air-cathode MFC, does not have a separate cathode chamber and the cathode is exposed to the air directly. The two-chambered MFC usually operates in the water-cathode-air mode, whereas the single-chamber MFC operates in the air-cathode mode. A Proton Exchange Membrane (PEM) or a Gas Diffusion Layer (GDL) separates the anodic chamber from the air-exposed cathode in a single chamber MFC. This enables passive oxygen delivery to the electrode. By completing the circuit with an external conductive wire, electrons are transmitted to the cathode [9]. A PEM such as Nafion, Ultrex, or a plain agar-salt bridge [10], separates the anode and cathode compartments in dual-chamber fuel cells. This allows the transfer of protons from the anode to the cathode while inhibiting oxygen diffusion into the anode chamber during cathode aeration. Aside from these two typical configurations (Figures 1a and b), MFC design and structure have undergone several changes.

The anodic chamber typically holds the inoculum/biocatalyst, usually electroactive bacteria, and organic substrate as carbon and electrons source, which are assimilated by the electroactive bacteria for growth and energy production, along with the generation of electrons and protons. The cathode chamber, on the other hand, simply contains a buffer of phosphate salts and, occasionally, a chemical mediator of electron transfer such as the ferrous cyanide (Fe $(CN)_2$) [11]. Instead of oxygen, biological agents like methane-producing bacteria function as the terminal electron acceptor in the bio-cathode. Electron mediators are chemical substances that operate as catalysts by speeding up the transport of electrons from the donor to the acceptor. However, using these mediators is not always necessary, depending on the species of bacteria involved in the MFC process. This is because in some bacteria, such as the *Shewanella*, direct electron transfer is possible through direct contact of the membrane with the anode, while others must first attach to the anode via attachment pili, also known as nanowires so that they can effectively transfer the electrons to the anode electrode. Other species, however, transfer electrons indirectly to the anode via electron mediators [12]. When compared to the mediator-free MFC, this increases the cost of the MFC operation.

An ideal, sustainable, and nontoxic electron acceptor is required to achieve a higher power density with no interference to the microbial colonies or other system components. Similarly, oxygen as an oxidizing agent is preferred to simplify the MFC operation, which works as an ideal electron acceptor without causing any toxicity to the system [9],[13].

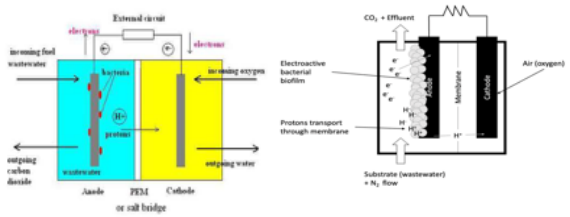

Figure 1 (a) Schematic diagram of a two-chambered MFC with permission from UK Essays [14] (b) Schematic representation of a single chamber MFC (adapted from Sevda et al. [15])

MFC Working Principle

Microbial metabolism (anabolism and catabolism) of the biodegradable substrate is a natural phenomenon both in aerobic and anaerobic environments. In MFC, microorganisms oxidize the available organic substrate as fuel and take part in a redox reaction (oxidation-reduction reaction) to produce protons and electrons in the form of reducing equivalents such as FAD+, FMN+, and NAD+). This helps to produce energy during respiration as Adenosine Triphosphate (ATP) [16].

MFC is extraordinary because it uses microorganisms to decompose organic substrate for their proliferation and development of biofilms, along with transferring the electrons produced to the surface of the anode. Microbial biofilms are essential as biocatalysts to carry out electrochemical redox reactions. In principle, the electrons delivered to the anode's surface by the microbial cytochromes or redox-active proteins are carried to the cathode via an external circuit (such as copper wire) to produce electricity [17]. The generated protons are simultaneously transferred to the cathode via the proton exchange membrane [9]. Usually, oxygen is continuously sparged into the cathode chamber as the terminal electron acceptor for the reduction of electrons along with the reaction of electrons with the proton and oxygen (oxygen reduction reaction) to form metabolic water as a final and clean product [13],[18].

Polarisation studies do the performance examination and characterization of the fuel cell. The polarisation curve is obtained by plotting a graph of change in current density against the change in cell voltage and power density, which resulted from the change in applied external resistance. A lower resistance generates a higher current by increasing the electron flow over the circuit by lowering down the potential differences between the anode and the cathode. However, the increase in current and voltage results in higher power output (P = IV). The resistance at which the power output is maximum indicates that the internal resistance is equal to the external resistance. Furthermore, the resistance at which the highest current and voltage are obtained gives the highest power is known as a cell design point. Therefore, an MFC operation above this point performs efficiently, while below this point causes instabilities due to the higher current and lower voltage [8],[16].

Electroactive Bacterial Strains Used in MFC

Due to their ability to participate in numerous intracellular catabolic events with the help of electrodes, electrochemically active bacteria play a crucial role in MFC operation. Some of these bacteria can directly transfer the electrons produced during substrate decomposition from the bacterial cells to the anode electrode [19].

Direct Electron Transfer (DET) and Mediated Electron Transfer (MET) are the two methods by which electron transfer from the microbial cell to the anode (MET) takes place. The DET does not require any mediators or redox species to transfer electrons; it works by the direct physical contact of the microbial cell to the anode, such as cytochromes and nanowires. On the other hand, MET requires a mediator for the transfer of electrons, and this requirement for a mediator depends mainly on the type of electroactive bacteria used (i.e. if the bacteria lack conductive pili) and on the conductivity of the wastewater used as a substrate since the electron mediators reduce the internal resistance thereby increasing the electrical conductivity of the substrate. These mediators could be exogenous or naturally excreted by the microorganism. Microbes that are distant from the anode excrete electron shuttlers, such as 4-naphthoquinone, phenazine-1-carboxamide, pyovirdin, pyocyanine, and 2-amino-3-carboxyl-1. The most studied and suitable natural mediators in the literature include phenazines, quinines, and phenoxazines [20]-[21]. MFC's most studied microbial strains include *Geobacter sulfurreducens*, *Pseudomonas aeruginosa* and *Shewanella spp*. This is because of their electrochemical activities and electron transfer mechanisms [16].

Shewanella oneidensis MR-1 is a widely studied strain because of its catabolic activities and its use in MFC for the current generation. It is a facultative bacterium that can survive in aerobic and anaerobic environments [22]. This bacterium belongs to the class *Gammaproteobacteria*. *S. oneidensis MR-1* is of particular interest because of its performance for bioremediation in anaerobic contaminated environments, especially with heavy metals such as Lead, Uranium, and Iron [23]. These bacteria can utilize various substrates, including lactate, pyruvate, and formate. These bacteria are self-mediators, hence metabolize the substrate and transfer the electrons from their inner cell to the external electrode (electron acceptor) via c-type cytochrome proteins MtrC and OmcA located in their outer membrane [24]. Furthermore, *S. oneidensis* secretes electron shuttles compounds known as flavins that aid Extracellular Electron Transfer (EET) [25].

Mixed cultures have also been shown to function better in MFCs than isolated pure cultures [26], with the added benefit of substrate adaptability. Thus, even abrasive materials like lignocellulose and chitin could be used to generate power [27]-[28]. Furthermore, such a mixed population would be more resilient to environmental changes in temperature, organic loading rate, and other parameters, with the possibility of metabolic pathways and/or population shifts that would facilitate adaption to the new environment. Because the influent cannot be sterilised and local changes of environmental elements are unavoidable in the plant, the mixed microbial anode biofilm formed from the wastewater influent can be exploited through natural or ecological selection for MFC applications in wastewater treatment [29].

Feedstocks Employed as Substrates in MFCs

The substrates are one of the most important chemical parameters impacting power generation in MFCs [30]. MFCs may generate energy from various of substrates, ranging from pure molecules like glucose to complicated mixes of organic matter like wastewater, as shown in Table 1. The type of MFC substrate utilized significantly impacts electron delivery and overall MFC performance. Because of the dual benefits of transforming undesired organic waste into a valuable product through energy generation, wastewater is being examined as a potential substrate for the MFC. The monomers of carbohydrates, fatty acids, and amino acids are found in almost all intricate and high molecular weight wastewaters [31]. Some of the substrates include domestic wastewater, dairy wastewater, pharmaceutical wastewater, food processing water, biogas digestate, kitchen-based waste, industrial wastewater, and commercial wastewater [16],[32]. The main goal of the various wastewater treatment methods has been to eliminate contaminants before releasing them into the environment to protect human health. The performance of MFCs with diverse wastewaters is being studied in connection to many metrics such as Columbic Efficiency (CE), Chemical Oxygen Demand (COD), maximum voltage output, and power density dependent on substrate concentration [4].

The Activated Sludge Process (ASP) has been the mainstay of wastewater treatment for the previous century. However, because of the high energy input demand, it is quite limited. According to Venkata et al. [8], the amount of electricity required to deliver oxygen in ASPs in the United States accounts for about 2% of the entire US electricity consumption. Furthermore, today's waste management focuses on energy reuse and recovery, which has resulted in fresh perspectives on how to deal with these streams. The performance of microbial fuel cell systems reported by previous researchers in terms of the power outputs generated from various substrates is presented in Table 1.

Electrodes Employed in MFCs

Electrodes are indispensable in the production of exo-electrogenic biofilms as well as the electrochemical processes that improve MFC efficiency and performance [33]. High surface area, high conductivity, cost-effectiveness, biostability, and biocompatibility are all characteristics of an excellent electrode [34]. Research shows that waste biomass is vastly available at a low cost and is recyclable with a great carbon source after a thermal process known as pyrolysis to produce biochar in the absence of oxygen. This can be a potential material for making electrodes with equal or better electrochemical properties than fossil-based electrodes [35]. Carbon-based electrodes typically meet these requirements while also being less expensive [36]. The surface area and porosity of the carbon material used for electrode preparation are normally improved through activation by physical means, including steam, carbon dioxide or air. Similarly, the activation can also be achieved chemically using chemical agents such as hydroxides and

carbonates of potassium, Zinc Chloride, or both [37]. Interestingly, activated carbon can be made from various carbonaceous materials, including wood, corncob, rice husk, coal, coconut shell and other agricultural by-products through pyrolysis and then physically or chemically activated [38] as shown in Figure 2.

Materials utilized as anodes in MFCs alternate solid electron acceptors include traditional carbonaceous materials such as carbon paper, carbon cloth, carbon fibre brush, and carbon mesh due to their superior biocompatibility and chemical stability. Novel carbon materials, such as biochars, carbon nanotubes, conductive polymer and graphite, are also in use. On the other hand, the cathode electron acceptor is commonly oxygen, with metals or polymers acting as catalysts. Cheng et al. employed carbon cloth with Pt as the cathode of an MFC and reported a fourfold increase in power density after modification [39]. However, research into biocathode MFCs that use microorganisms as catalysts is gaining traction. Compared to typical cathode MFCs, the bio-cathode MFC has several advantages, including inexpensive construction and operating costs, avoidance of efficiency loss from the chemical catalyst, and long-term stability due to the self-duplication of the microorganisms utilized as biocatalysts.

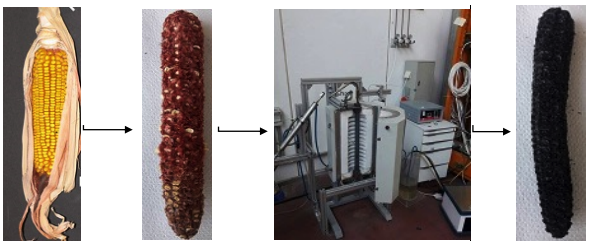

Figure 2 Pyrochar Production from Corncob Biomass by Slow Pyrolysis (Adapted from Musa et al. [40])

Table 1 MFC performance relative to configuration, substrate, electrode, and electron acceptor (Adapted from Flimban et al. [9])

Types	Substrates	Electrode		Electron Acceptor	Power Density
		Anode	Cathode		
Single chamber	Glucose	Graphite carbon fibre brush	Pt (30%) coated carbon cloth	Oxygen	2.4 W m^{-2}
Single chamber	Acetate-amended Wastewater	Graphite fibre brush	Activated carbon catalyst on Stainless	Oxygen	1.1 W m^{-2}
Double chamber	Glucose	Carbon paper	Carbone cloth	Permanganate	0.12 W m^{-2}
Double chamber	Glucose	Graphite plate	Graphite plate	Hexacynoferrate	4.3 W m^{-2}
Stacked	Sodium acetate	Granular activated carbon	Granular activated	Oxygen	50.9 W m^{-3}
Stacked	Neat undiluted urine	Untreated carbon fibre veil	Coating activated carbon paste on polytetrafluorethylene	Oxygen	0.8 W m^{-3}

Proton Exchange System (PES) in MFCs

The proton exchange systems influence internal resistance and concentration polarisation loss in MFC systems, which in turn influence the MFC's power output. Nafion (DuPont, Wilmington, Delaware) is well-known for its highly selective proton permeability. A sulfonated synthetic polymer with specific ionic characteristics, the Nafion membrane is a proton-exchange membrane that is commercially accessible. Despite researchers' efforts to find less expensive and more durable alternatives, Nafion remains the best option. Even with Nafion, however, negative side effects of other cations transport are unavoidable during MFC operation.

Because concentrations of cations such as Na^+, K^+, NH^{4+}, Ca^{2+}, and Mg^{2+} in the anolyte and catholyte are much higher than proton concentrations in the anolyte and catholyte, transportation of cation species other than protons by Nafion dominates the charge balance between the anodic and cathodic chambers in an accumulative batch system [41]. This indicates that Nafion and other Proton Exchange Membranes (PEMs) utilized in MFCs are cation-specific membranes rather than proton-specific membranes. The ratio of PEM surface area to the system volume influences the power output. The PEM surface area significantly impacts the maximum power output if the power output is below a crucial threshold. Over a wide range, the internal resistance of the MFC reduces as the PEM surface area increases [42]. The performance of PEM and a salt bridge in an MFC inoculated with *G. metallireducens* was compared [43].

MFC Stability

MFC operation's process stability or process management could save a failed process, especially in biological systems that take a long time to react, such as wastewater treatment [44]. The MFC system's control improves process performance by monitoring quantifiable factors and employing a control plan to optimize operating conditions. Although monitoring and control of MFC systems are still in their early stages, it has a lot of promise. The substrate concentration, cell potential, and power density are critical variables utilized to anticipate MFC's dynamic and real-time feedback control. Changes in parameters like dilution rates, substrate loading and flow, aeration, and electrical loading can all affect these parameters [44]. Interestingly, MFCs have been used to monitor bioactivity, such as in BOD sensing [45].

Several studies have been carried out on the stability of MFC in terms of stated characteristics, such as MFC designs/configuration, substrate variety, and diversity of the electrogenic microbial community involved. As a result, several promising results in terms

of COD removal, enhanced voltage output and power density, columbic efficiency, and the impacts of substrate types and concentration in the MFC have been obtained.

Understanding the fundamental knowledge of long–term system performance, repeatability, and the development and maintenance of functionally stable microbial communities is the only way to put MFC into practice. Regardless of the sample variability associated with each wastewater, an MFC operated for over 360 days exhibited extremely consistent electrochemical performance and BOD removal rates throughout the hydraulic retention time [46]. On the other hand, phylogenetic investigations of the anode – linked electricity – producing biofilms revealed only a transient fluctuation of the microbial population, which then maintained high biodiversity throughout the year-long trial. This suggests that MFC systems are self-selecting and self-optimizing, allowing them to develop and maintain functional stability despite changes in carbon sources and the regular introduction of microbial competitors like methanogenic bacteria, which redirect carbon to methane production rather than electrons.

MFC Sustainability

The trickling filter method, activated sludge system, and membrane filtration technique are now employed in wastewater treatment and are known to be either cost-prohibitive or energy-intensive [47]. As a result, more effort is being invested into finding innovative wastewater treatment systems that are inexpensive, environmentally friendly, and consume less energy. Similarly, current renewable energy generation technologies, such as hydropower, wind, and solar, fully depend on the local climate and are site-specific. As a result, transmission prices rise with distance [48], making them increasingly difficult to purchase for most of the population, particularly those living in distant areas of underdeveloped and developing countries around the world. As a result, an on-site treatment technology, such as microbial fuel cell technology, is an option for providing a long-term solution to these wastewater treatment concerns.

MFC technology lowers the cost of wastewater treatment while also increasing the long-term viability of the renewable energy produced. This is because electrogenic bacteria drive it and recover electrical energy from organic pollutants in the wastewater while also purifying it for municipal use [47]. MFC is also renowned as a diverse study field for bioremediation, desalination and biohydrogen production, carbon sequestration, and environmental biosensing, in addition to wastewater treatment and electricity generation [49]-[50]. MFC has discovered useful applications of desalination and energy production in remote places that are difficult to achieve with conventional bioenergy methods. MFC technology is, therefore, often recognized as the greatest of all biochemical systems.

Many scientists and engineers throughout the world are interested in this technology. This is because, during MFC operation, by-products such as bio-flocculants, biohydrogen, bioplastics, methane, biosurfactants, bioelectricity, and various additional value-added products for various uses are created. MFCs are expected to be the most sophisticated sustainable wastewater treatment devices that use wastewater as a substrate today [47].

CONCLUSION

The numerous MFC designs, substrate types, microorganism roles, and electrode materials utilized in MFCs for energy generation were highlighted in this review paper. However, novel substrates and other microorganisms are constantly being tested for their possible application in MFCs, and some are showing promise. Simple substrates like acetate and glucose were routinely employed in MFCs a few years ago, but in recent years, researchers have used more atypical substrates intending to utilize waste biomass or wastewater treatment on the one hand and boost MFC output on the other. Similarly, developing new electrode materials has greatly decreased the cost of electrodes. Even so, some of these electrodes, such as graphite, are still quite costly. Finally, using bioenergy technologies like MFCs to generate electricity from renewable and waste biomass helps to alleviate the global energy crisis while ensuring water security. This is not something you will see with other biofuel-producing methods. As a result, MFCs are considered the greatest alternative sustainable and cost-effective bioenergy generation process due to technological advancements, lower costs, and substrate variability.

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