

AN OVERVIEW OF ENERGY GENERATION USING MICROBIAL FUEL CELLS

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Abstract. Microbial fuel cell remains one potent replacement for conventional fossil fuels. Its application holds enormous capacity in managing waste while producing energy. Microbial fuel cells use microbial metabolism from micro-organisms to create an electrical current from chemical energy from variety of dissolved biomass. The involvement of microbial consortia is necessary since the microbial fuel process has high versatility and benefit for sustainable energy production. However, significant advancements are needed before broad scale implementations can be achieved. Waste treatment facilities use microbial fuel cells to decompose organic waste materials. Microbial fuel cells have been improved upon seeing that microbial communities are malleable and are used to make new changes. This review presents an overview of the microbial fuel cell highlighting its performance, shortcomings and introducing progress in using selected microbial community. Possible optimization and tailored development for the future of the microbial fuel are equally captured.

Key words: microbial fuel cell; micro-organisms; biogas; substrate.

INTRODUCTION

The majority of people on earth rely on pricey appliances to light their homes and utilize fossil fuels for various household tasks that release toxic emissions into the atmosphere. Many people run the risk of everyday exposure to pollutants and potential fire hazards by using generators to power their buildings and homes [41]. Access to steady and reliable energy is almost impossible as it applies to several billions of people in today's world. An eco-friendly, safe and affordable source of power such as microbial fuel cell would help to solve this problem. The unique ability of bacteria to produce electricity through the use of biocatalysts and organic materials as substrate (food for the microorganisms) has been harnessed by the breakthrough technology known as the microbial fuel cell. As bacteria consume the substrate, chemical energy is converted to electrical energy [8, 72].

Using microbes' metabolic processes to transform organic materials into useful energy, a microbial fuel cell (MFC) generates current. This device operates on the premise of producing electricity through microbial catabolic processes [89]. MFCs also known as bioreactors, are devices that oxidize organic and inorganic substances and generate currents by micro-organisms as catalyst [61].

Microorganisms have most oftentimes proven to be a gift of nature; from bio-fertilizers to being used in metabolite production, microbes have greatly aided humanity. Microorganisms have been found to have the ability to survive at very extreme places of the earth and can equally respire with or without oxygen. Chemical energy buried in biomass material transforms into electrical energy as micro-organisms pump out electrons [36]. This indicates that microorganisms can generate electrical energy and there are a handful of bacteria which can carry out this crucial function; however, this energy product needs to be harnessed from these minute lives. By generating electrons and

transferring them across their cell membranes, bacteria use extracellular electron transfer to make this power [113, 115].

The MFCs are an environmentally friendly technology that produce heat, energy and biological oxygen demand sensors among other recycled and renewable products. Microbial fuel cells are potential new technologies meant for environmental remediation with sustainable energy production. The core idea behind MFC functioning is using microorganisms' metabolic processes to generate electrical current. Microbes, primarily bacteria, and organic matter, generally waste materials, are employed as substrates (electrolytes) in MFCs, where biological reactions produce electrons. Contaminants can be expunged at the cathode as an electron acceptor by reduction or as electron donors oxidized at the anode by microbiological catalysis. Certain contaminants can act as electron mediators at either the anode compartment or cathode. Applications of MFCs include desalination, bioremediation, wastewater treatment, environmental monitoring, and the production of renewable energy.

According to the MFC's operating principle, microbes oxidize the biodegradable substrate anaerobically at the anode, producing carbon dioxide in addition to electrons and protons. At the cathode chamber, these protons and electrons are depleted once they mix with oxygen to create water. In an attempt to create energy, electrons move via external circuit meanwhile the protons travel via the exchange membrane. In order to keep potential for producing electricity alive, oxygen must be continuously consumed.

Gaining an understanding of the fundamental components of MFC systems is necessary to maximize their functionality and increase their usefulness. Through the catalytic activity of microorganisms, chemical energy embedded in organic materials transforms into electrical energy in the MFCs.

MFC can be used to treat wastewater while maintaining the production of bioelectricity and achieving zero liquid effluent discharge (ZLED), free of strong organic pollutants.

With a few minor modifications, MFCs can be used to produce byproducts like hydrogen (H_2) as a bioelectricity option [85]. MFC provides a dual benefit of waste management and bio-electricity generation due to its usage of organic material like wastewater [14, 72]. Soil, activated sludge, freshwater sediment, marine sediment and wastewater are amongst fuel sources where bacteria that participate in electron transfer have been found thus far [84, 106].

Seeing that microorganisms have properties to breakdown organic matter, MFCs can then make use of a several organic sources as fuels instead of depending on the catalysts of metals [72]. More so, its performance further relies on operational factors such as microbial diversity, wastewater type, anodic potential, electrode material, reactor type and configuration [45, 72]. More so, it hinges on function parameters as time [6, 23], ionic strength [40], temperature [94], external resistance, pH [40] and concentration [75].

STRUCTURE OF THE MICROBIAL FUEL CELL

Exoelectrogens, or organisms that can release electrons after consuming biological matter, live at the anode. The anode holds the electron transport location and microbial metabolism. Carbonaceous substrates like as carbon paper, graphite felt, and carbon cloth are frequently utilized as anodes. According to Logan [58], these materials offer a conductive surface that facilitates electron transmission and bacterial attachment (biofilm) at the anode. The cathode chamber consists of electron acceptor like oxygen or other oxidants (permanganate, triiodide, hydrogen peroxide) that can be reduced with the help of the cathode. It anticipates the anode to supply electrons. High catalytic activity materials, including manganese dioxide, carbon nanotubes, or platinum, are used to speed up the oxygen reduction reaction (ORR) and increase output of electricity [75].

The polymer-electrolyte membrane (PEM), or the proton exchange membrane, as it is also known, separates cathode from anode compartments in the microbial fuel cell [99]. This membrane keeps solutions from directly mixing while facilitating proton

transport between the cathode and anode compartments. Protons produced from bacterial metabolism must be transported from anode to cathode. Because of their chemical stability and proton conductivity, materials including polyvinyl alcohol (PVA), sulfonated polyether ether ketone (SPEEK), and nafion are frequently used [59]. Ion movement between the anode and cathode section is facilitated by an electrolyte. It permits the exchange of ions between the electrodes and preserves electrical neutrality. In MFC systems, saline solutions, potassium ferricyanide, and phosphate buffer solution are commonly utilized as electrolytes [59]. Below is Figure 1 showing a microbial fuel cell:

Carbon dioxide, electrons and protons are generated at the anode; the electrons travel to the electrode during the oxygenation of organic materials [32]. Then protons generated at the anode move to the cathode via the proton exchange membrane with the electrical circuit, located outside. Oxygen which is a typical oxidant is reduced at the cathode. Microorganisms that are electrochemically (biologically) active are found on the anode side, leaving the cathode side abiotic. Exoelectrogens (bacteria) help to break down biomass material to produce electrons which migrate to the electrical circuit by the cathode side allowing substrates to make protons and electrons. As electrons move via the electrical circuit to the cathode pole, hydrogen ions migrate towards the cathode region fusing with the oxygen molecules at the internal circuit to create water [104, 110]. When the anode and cathode electrodes join, the circuit makes it easier for electrons generated in microbial metabolism to move. Generated electrical current can be stored for later use or used to power other equipment.

According to Logan [59], 0.3 - 0.5 voltage range is normally attained as fuel sources like glucose and acetic acid are used; it however depends on the amount of energy received by the bacterial species in question and the energy lost at the cathode. Electromotive force, also known as maximum theoretical cell voltage, controls voltage produced by an MFC, while the rate of substrate bio-degradation influences the current it generates [89]. Iron-reducing bacteria including *Shewanella* sp., *Geobacter* sp., and *Escherichia coli* are some of the model species that are now utilized in MFCs. For the purpose of nutrient cycling, these bacteria may break down organic materials like iron oxides that are present in both soil and sediments [89].

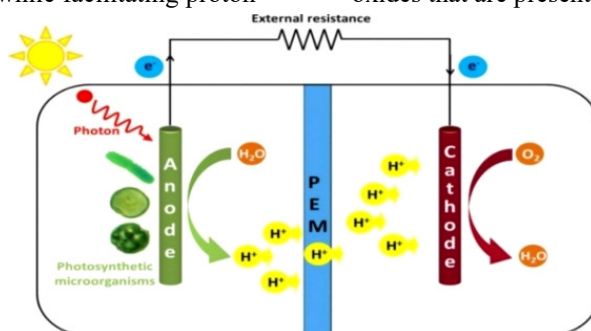


Figure 1. Anode and cathode chamber are set apart by the proton exchange membrane in a microbial fuel cell (culled from Cao *et al.* 2019 [13]).

KEY COMPONENTS INFLUENCING MICROBIAL FUEL CELL OPERATION

When enhancing efficiency as it has to do with a microbial fuel cell certain factors are involved.

The make-up of Electrode

A material needs a variety of characteristics in order to function well as an electrode. Ideally, it must have a large surface area that generates increased current densities, be mechanically stable, inexpensive, and display advantageous electrochemical properties [18, 89, 106]. This electrochemistry research to create new materials to replace platinum has become increasingly expensive because of its expanding world demand in many sectors and uses. Furthermore, it has demonstrated antibacterial qualities that inhibit *Escherichia coli* by resulting in the production of cisplatin by a platinum electrode during electrolysis [31]. Non-corrosive metals and carbon materials also satisfy requirements for cost and performance and also appear to be preferable for use as base electrode materials in MFC setups [10, 62, 89].

For stable biofilm formation, the anode material regulates the adherence of bacteria to its surface. Stable biofilm formation is required for effective electron transfer. It should also exhibit improved electron conductivity and be biologically active with the electroactive microorganisms. The anode material should also possess certain characteristics like chemical stability, mechanical strength, and good corrosion resistance [21]. Conversely, it may lead to more fouling, which may limit its long-term applicability. Consequently, when choosing suitable materials for anodes, consideration should be given to their durability and insensitivity to changes in pH and operating conditions.

According to Yaqoob *et al.* [107] rougher electrode surfaces generated much more power densities compared to its smoother counterpart, as two glassy carbon plates are polished to same roughness in order of magnitude of 10 nanometers each; this occurred after 275 hours of testing [107]. Surface roughness is an important factor that should be taken into cognizance when selecting the materials for electrodes. Research has indicated that when the microbial cell size is more compact, the microbial adhesion ability gets stronger; more so, the more the biomass and the electrode surface roughness, the greater the battery generation activity [114]. The efficiency of electron transfer can be impacted by material modifications to the electrode surface. The usage of a polydimethylsiloxane composite as the cathode combined with a metal current collector mesh can significantly increase MFC's capacity for power generation, with a coulomb efficiency of above 80%. According to experiments, the highest nitrate nitrogen removal rate of MFC using poly-neutral red modified cathode (CPNR-MFC) was 0.040 kg/m^3 , being 14.29% greater than the control [114].

High mechanical strength and electrical conductivity are essential characteristics of electrode

materials [102]. The electrode materials of the anode and cathode chamber must be inherently conductive, non-corrosive and fouling-free.

Anodic electrodes are a necessary component of an MFC and are critical to increasing its efficiency. Thus, when designing a standard anode in an MFC, appropriate anode material should be considered. It should be inexpensive, have a large surface area, be highly conductive and not corrode. These include carbon-based materials, metal complexes and oxides, and other substances. Graphite felt, activated carbon, granular graphite, and other carbon-based materials are also utilized in the biocathode formation process [60, 61].

Compact materials, like carbon-based electrodes, carbon-based electrode materials (plates or rods) are promising components of anode electrodes for the development of biocompatible and active microorganisms that can successfully produce energy from complex substrates because of their affordable cost, simplistic operation, extremely good pore stability [56, 59] and also large surface area and porosity [22]. The total power outputs of carbon felt and carbon brushes were high at 2437 and 2110 mW/m^2 (90% COD elimination) [56, 57, 59].

According to Roy *et al.* [84], a series of researches on the suitability of anode materials have shown and proven that the main ways to improve MFC performance are through a highly porous structure, large specific surface area, mechanical strength, stability, and conductivity. Even though carbon-based materials are frequently utilized as anodes, carbon electrodes that have been pre-treated with nitrogen can achieve even higher power densities [56, 57, 59].

More so, Nanomaterials, such as carbon nanomaterials, have a lot of advantageous qualities that have been observed, like promotion of molecule adsorption, which makes them potentially perfect for usage as electrode materials in MFCs, also having a large surface area and an improved electron transport. Many carbon electrode materials like carbon meshes, clothes, felts and graphite rods have been studied for usage under MFC conditions [19, 68, 70]. In MFCs, the electrodes that are used need to be compatible with the bacteria that survive there.

In an MFC arrangement, electrodes with high surface areas and rough surface feel are considered to be the optimal surface property due to the fact that it aids in the retention time of the bacteria on the periphery [34]. Most importantly, it is widely known that the surface area and roughness of an electrode are significant factors in the production of electrical current. Wang *et al.* [99] noted in their research that the MFC's volumetric power density increased when the total anodic surface was increased inside a chamber with set volume [98].

With the support of exogenous mediums like permanganate solution, the cathode electrode helps to complete the electron flow and reduce the treated wastewater to a basic cleaner effluent that is free of

high levels of organic or biodegradable pollutants as well as possibly particulate or non-biodegradable pollutants. Even though cathode electrodes have advanced significantly, they still have shortcomings that require regular catholyte maintenance, including high cost, microbial surface toxicity, and inadequate re-oxidation [22]. Research into more enticing materials to boost MFC power output has been prompted by these factors.

Giving biofilm formation a larger surface area can boost the efficiency of biocathodes. According to Roy *et al.* [84] biocathodes also improve performance by lowering internal resistance. Because of their benefits, biocathodes are receiving more attention. Research is being done to identify biocathodes that can enhance MFC performance at a lower cost.

Biocathodes' microbial metabolism can be used to eliminate undesirable substances or create beneficial chemicals. As a biosensor for chemical oxygen demand (COD) real-time monitoring in tubular membrane bioreactors (TMBRs), the biocathode MFC was created. With the help of the MFC–TMBR integrated system, the straightforward and small bio-cathode MFC biosensor for TMBR shown encouraging potential for direct and affordable COD online monitoring, opening up new applications for MFC-based biosensors. In recent times, it has been discovered that sophisticated electrochemical methods like impedance spectroscopy and cyclic voltammetry (CV) are useful in comprehending the processes involved in the creation of electrochemically active biofilms (EABs), cell adhesion to the electrode surface, diffusion coefficients, and variations in the MFC's internal resistance [87].

Low-cost biocathodes made of graphite fiber brushes with microbial biofilm growth originating from the dynamic membrane and percolating anolyte were recently developed by Chouler *et al.* [20]. Ieropoulos *et al.* [37] tested a patented multi-panel air cathode for treating household wastewater with an 85 L SCMFC.

This cathode allowed the welding of several smaller cathodes into a single metal sheet. Anode consisting of 22 graphite fiber brushes and separator made of glass fiber allowed for a maximum power density of 0.605 W/m^3 with 80% COD removal and 27% CE. A 255 L MFC prototype was also used to test the same electrode module [30].

Electrodes enhanced with nanoparticles produce more electrical current than electrodes without them. Hence, in their study, Alatraktchi *et al.* [1] used *Geobacter sulfurreducens* as inoculum and a magnetic nanoparticle containing electrode, over a pliant carbon paper, to boost current production in MFC [1]. In the same vein, Kumar *et al.* [47] used nanoparticles to optimize electron transfer mechanism and thereby raised the conductivity of the biofilm electrode by 1.88 times against a standard electrode of carbon paper [47]. However, without a catalyst, a platinum or platinum-coated cathode can produce more power than a normal cathode [72].

Anode material for an MFC reactor must be non-corrosive, affordable, highly conductive, non-fouling, and possess a large specific surface area in order to fulfill its intended purpose as an electron receiver for current [87]. MFCs with copper anodes have a high-power generation capability and of 2 W/m^3 power density at minimum, as relayed by a study comparing the usage of corrosive metal anodes in MFCs [58].

Furthermore, graphite sheets or rods make the simplest anode electrode materials since they are affordable, manageable, and have a specified surface [72]. Vitreous carbon can be used to create a greater surface area since it has cross-linked varied pore diameters, encapsulated carbon, and granule grades. To prevent clogging, high porosity must be maintained in every situation.

In a study conducted using a stacked MFC with a fed-batch operating time of 48 hours, 97% COD was successfully removed from the wastewater while achieving a maximum power density of 1.7 mW/m^3 .

Table 1. Advantages and disadvantages of different types of Anode and Cathode materials [5, 15, 33]

Anode Electrode	Advantages	Disadvantages
Carbon nanotube (CNT)	Large surface area High mechanical capacity Stability Electrical conductivity	Clogging High operational cost Complex synthesis procedure
Graphene	Good electrical conductivity High mechanical strength Large surface area Biocompatibility Increased electron mobility	Complex synthesis procedure
Metal Oxide	Reduction in internal resistance Improved biocompatibility	Expensive for large-scale implementation
Cathode Electrode	Advantages	Disadvantages
Air-cathode and aqueous air-cathode	Simple architecture. Cathodes can be modified using affordable items such as activated carbon or HNO_3 to improve performance. Recycling of catholyte is not needed.	Performance of aqueous air-cathode limited by the solubility of oxygen. Oxygen crossover. Use of catalyst can lead to additional cost. Biofouling of the cathodes.
Biocathode	Inexpensive and sustainable. Protection against catalyst poisoning. Reduction in internal resistance	Lower power output. Fluctuation of pH

Compact electrodes made of granular activated carbon (GAC) were used in the experiment [22]. When graphite plates were utilized for both electrodes, remarkable MFC performance was achieved, yielding a power density of 1771 mW/m^2 [22]. Furthermore, utilizing incapacitated GAC on the cathode, 90% oxidative removal of tetracycline hydrochloride by reactive oxygen species was accomplished.

Carbon and metal nanoparticles with unique combinations and diverse dimensional structures have attracted a lot of interest [22]. The use of a bio-cathode to produce electricity and remove carbon and nitrogen was thoroughly investigated.

Microbes

Numerous anaerobic and photosynthetic microbes are being used as electron donors and acceptors in MFC operation. They consist of several anaerobic bacteria as well as *Scenedesmus armatus*, *Desulfovibrio desulfuricans*, *Rhodospirillum rubrum*, *Klebsiella pneumoniae*, *Pseudomonas fluorescens*, *Thiobacillus ferrooxidans*, *Geobacter metallireducens*, *Leptothrix* and *Phormidium* sp. Certain organisms have undergone genetic engineering to yield exponential increases in both current production and sustainable biomass generation when compared to strains of the natural existing type.

Archaeobacteria

Numerous archaea can endure harsh conditions, like high salinity and temperature, which put a great deal of stress on microorganisms. Under some circumstances, they might act as electricigens in MFCs. In the anode of an MFC, two halophilic archaea species - *Haloferax volcanii* and *Natrialba magadii* - were evaluated as electricigens. For *H. volcanii* and *N. magadii*, the maximal power density and current density without any exogenous mediators were $11.87/4.57 \text{ } \mu\text{W/cm}^2$ and $49.67/22.03 \text{ } \mu\text{A/cm}^2$, respectively. The maximum power density for both archaea was further enhanced when neutral red was introduced as the electron mediator, and this power output was considerably greater than *Escherichia coli* under the same conditions [13].

Acidobacteria

Physiologically varied acidophilic bacteria are known as acidobacteria. They are able to use a broad range of substrates and can be found in a number of situations. This phylum's members displayed electrochemical activity in some cases. The reduction reaction in the electrode was facilitated by the production of electron mediators by the iron-reducing bacterium *Geothrix fermentans*. The current generation rate in the *G. fermentans*-based MFC was 0.6 mA and the electron recovery was 97% when the operating conditions were optimized. From an MFC fed acetate, two representatives of the acidobacteria genus *Arcobacter* were identified. Approximately 90% of the population in the MFC was responsible for producing a maximum power density of 296 mW/L. These strains were obtained in their pure cultures and were

selectively linked to the anode of MFCs, with negative potentials ranging from -200 to -300 mV [13].

Cyanobacteria

Green sources of bioenergy generation, photosynthetic microorganisms are called cyanobacteria. A lot of research has been done in the last several years on the use of cyanobacteria in MFCs. Known as photosynthetic MFCs (PMFCs), these cyanobacteria-based bioelectrochemical systems use light as their power source to oxidize water, which produces electricity. Various cyanobacteria species have been assessed for their potential as PMFC electricigens. Using *Synechocystis* PCC-6803 as a model cyanobacterium, a dual chamber PMFC was built. This PMFC had a consistent power output, reaching a maximum power density of 72.3 mW/m^2 . Without the need for external feedstocks, PMFCs employing *Spirulina platensis* as the biocatalyst could run at high open circuit voltage. This PMFC achieved a maximum power density of 6.5 mW/m^2 . *Nostoc* sp. ATCC 27893 was a recently isolated cyanobacterium that was also used in a PMFC's anode, producing current and power densities of 250 mA/m^2 and 35 mW/m^2 , respectively. A notable increase in power generation capacity was seen upon the addition of 1,4-benzoquinone as the electron mediator (maximum current density of 2300 mA/m^2 and peak power density of 100 mW/m^2) [13].

Firmicutes

Firmicutes can withstand extreme environments and have robust cell walls. They could always be kept apart from other cultures in the MFC anode; but because electrons must travel through the cell wall to reach the anode, firmicutes have comparatively less electrochemical activity. One firmicute that has been successfully isolated and used in MFCs is *Clostridium butyricum*. There is a broad pH and temperature range over which this stringent anaerobe can thrive. Ten hours after inoculation, the maximum current of 0.22 mA was generated; this current gradually dropped as the logarithmic growth phase began. The same genus *C. beijerinckii* could also produce energy from cheap substrates like flour and molasses, with a current density of $1\text{-}3 \text{ mA/cm}^2$. After more *C. beijerinckii* mutagenesis, the best mutant generated a maximum power density of 79.2 mW/m^2 in an MFC with methylviologen serving as the electron mediator and glucose as the carbon source. After being isolated from a thermophilic MFC, the firmicute *Thermococcus* sp. strain JR was shown to be the predominant strain within the electricigens community. Using acetate as the substrate, it produced an average current of 0.42 mA, or almost 70 % of the total electricity produced by the community.

Due to their well-established safe status, rapid growth rate, and distinct genetic background, yeasts make excellent candidates for use as electricigens. *Saccharomyces cerevisiae* was also examined for energy generation in groundbreaking research. Despite continuing to generate less power than bacterial MFCs,

yeast MFCs are gaining more attention. For instance, by exhibiting glucose oxidase on its cell surface, an engineered strain of *S. cerevisiae* with good electrochemical activity was created. Compared to unaltered yeast, the MFC displayed increased power output and current density. *S. cerevisiae* was employed as the electricigen in a recent work to assess how much electricity was produced and how substrates degraded under various redox circumstances. *Candida melibiosica*, a different strain of yeast, was also used as an MFC biocatalyst. Because of its strong phytase activity and ability to generate electricity without the need for external electron mediators, *C. melibiosica* was identified as an electricigen [13].

In studying the operability of MFCs it is vital to understand that mixed bacteria MFCs are more resistant to changes in the external environment than pure bacteria. At the same time, the bacteria can work together to better the electrical performance and ensure the MFCs remain stable, thereby offering significant benefits towards the actual manufacturing of MFC in the future. Since mixed cultures can adapt to different nutrients and can withstand stress, they are more advantageous than pure cultures. Due to the existence of various types of bacteria, including electronics and the ability to provide a high-power density, the application of rich and diversified sources of bacteria, such as activated sludge, wastewater, soil, or sediments, in mediator-less MFCs is more favorable in wastewater treatment [13].

According to studies, the most abundant microbial flora on the surface of the anode can be enhanced by mixed microbial flora MFC, which increases power output by approximately six times compared to pure MFC [104]. Yang *et al.* [105] invented an algal biofilm microbial fuel cell (ABMFC), a continuous flow device that can handle complicated and variable effluent. In the wastewater, the removal rates of chemical oxygen demand, total phosphorus and total nitrogen were 80.2%, 91.5% and 96% respectively. ABMFC's maximum power density of 62.93 mW/m² was rated higher than that of MFC's with 52.33 mW/m². The understanding of the development and metabolism of algae consortium improved the work and consequently brought to light a method for treating wastewater and recycling energy. Research is being focused on substituting catalysts and electron acceptors at the cathodic compartment with bio-cathodes, which can enhance cathodic processes to reduce high expenses connected with their use [39, 77, 99]. It is crucial to note that investigation has been done on both aerobic and anaerobic bio-cathodes, depending on the type of terminal electron acceptor used at the cathode pole [57]. *Thiobacillus ferrooxidans* is one instance of an aerobic bio-cathode; this has been seen to replenish ferric ions, that function as electron mediators in the cathode compartment [60, 89].

Geobacter metallireducens is an additional illustration of an anaerobic bio-cathode. It can reduce nitrate (to nitrogen) and oxidize ammonia, which

results in removal of nitrogen in an MFC setup [24, 89]. Because bio-cathode is both economical and biocompatible, it can be used for waste treatment and heavy metal removal with equal benefits. Through the installation of bio-cathodes, costly building supplies and perhaps dangerous chemicals are avoided, as is the requirement to recycle and properly dispose of them [30, 79].

Polymer-electrolyte membrane

Polymer-electrolyte membrane (PEM) is found in the center of the MFC's cathode and anode chambers. It has an impact on the MFC's output power, bias loss concentration, and internal resistance [49]. This is so that protons can move quicker across the anode to the cathode due to the charged side walls of voids [72]. PEM is interesting in that it lets protons or hydrogen ions through, which are in charge of producing electricity. The membrane affects the fuel cell's current generation by raising the internal resistance of the MFC and facilitating ion diffusion [72, 83].

It is feasible for protons generated at the anode to pass via a liquid solution and then over to the PEM [78]. In MFC, PEMs are usually used as separators due to their beneficial qualities, which include their selective permeability to protons, low internal resistance, high conductivity to cations and ability to tolerate prolonged periods of inactivity without adversely affecting the MFC [80]. At the cathode, protons and water can combine with the electrons to form oxygen [89].

PEMs are primarily for maintaining anode and cathode solutions in a manner that permits ion transfer. Additionally, it makes it easier for protons to go to the cathode when electrons are transmitted by means of an external circuit. Because PEM regulates the flow of protons across the anode to the cathode compartment, it equally assists in the cell's ability to produce energy [86]. The concentration polarization effect on the PEM film may have an impact on the movement, which may result in less energy being produced in the system. In accordance with Elhenawy *et al.* [26], the PEM regulates both the substrate flow and the oxygen diffusion towards the anode chamber in an equal manner. Nafion is the most frequently employed material for PEM, followed by Ultrex CMI [72].

pH

The pH of MFC is one of the key elements that governs both cathode response and microbial anodic activity. One of the critical elements in MFC efficiency that can impact microorganisms' metabolic activity is the pH of the anodic portion [61]. According to a particular investigation, methanogen activity causes lowering pH to increase organic waste removal efficiency [61]. Proton buildup causes the electrolyte in the cathode compartment to increase in pH hence making the anode to acquire a greater acidity. Therefore, low energy production from a reduction in pH in the anode compartment inhibits activation of electrically active microorganisms (EAM) [38]. Although it has been said that the pH at the anolyte can

be controlled by using a pH buffer (phosphate or bicarbonate, pH 7.0), it could equally increase operating expenses and make desalinating wastewater or phosphorus removal more difficult [16].

According to Zhang *et al.* [111], ideal pH range for the acetate-charged air cathode MFC to produce the most energy is 8 to 10. For the best conditions for cell growth, an anodic microbiological reaction typically requires a pH of neutrality, whereas a cathode reaction benefits from a low alkaline pH. The achievement of high potential difference and coulombic efficiency (CE) in MFC is contingent upon the maintenance of an optimum pH in the electrolyte. In addition to H^+ , other cations that can travel to the cathode include Na^+ and K^+ . However, the mass transport of H^+ is sluggish, and its accumulating at the anode results in anodic acidification, which severely restricts the amount of power that can be generated inside an MFC [66, 82].

One of the key elements in MFCs that influences cathodic reactions as well as anodic microbial activity is pH.

Proton buildup in the cathode chamber would result in electrolyte alkalization and anolyte acidification. As a result, reduced power production from lowering pH in the anode chamber brought on by an increase in proton concentration suppresses the activation of EAMs. Conversely, it raises the pH in the cathode chamber, which prevents the oxygen reduction process (ORR) from occurring [38]. While it has been claimed that pH at the anolyte can be controlled by using pH buffers such phosphate or bicarbonate (pH 7.0), doing so may result in higher operational expenses, the need for effluent desalination, or an increased load of phosphorous removal [16].

It was discovered that a higher anodic pH would result in a higher elimination of chemical oxygen demand (COD) and enhance MFC performance. The ideal pH level is highly dependent on the kind of microbe. It was investigated how pH affected the anodic microbial community's and MFC's performance. The main communities under acidic pH circumstances were *Acinetobacter*, *Comamonas*, *Variovorax*, and *Simplicispira*, and the results demonstrated a faster elimination of COD under these conditions. At $pH \leq 5.0$, anodic biofilm broke and the number of cells significantly dropped [17]; also, MFC failure occurred at pH 4.0 as a result of alterations in the makeup of the microbial population.

However, when the pH was further readjusted to 7.0, the MFCs were able to resume producing electricity at their ideal level [16]. The ideal pH range for an air-cathode MFC powered by acetate to provide the most power was 8–10. For optimal cell growth, the anodic microbiological reaction often required a neutral pH; in contrast, the cathodic reaction was better suited for a weakly alkaline pH [17].

Furthermore, suitable pH ranges for the bacteria in the MFC responsible for energy production lie slightly higher at the cathode pole or even neutral and within a range 6 to 8 at the anode chamber. pH lowers at the

anode compartment distorting bacterial growth due to the formation of protons [97]. The high pH at the cathode chamber lowers current generation. Reduced oxygen and increased current from the MFCs are made possible by the low pH [51]. The low pH of the anolyte impedes bacterial activity, grossly affecting biofilms formation and power output of the MFC.

At low anodic pH, high quantities of protonation are observed. This suggests that the cathode chamber accumulates a lot of protons, which lowers the power density. Through the reduction of electricity, high pH at the anode chamber affects COD removal from wastewater treatment process. It is interesting to note that the anode's species and the cathode's electron donor affect the favorable and varied pH values between the two. Additionally, the cathode redox reaction rate and bacterial metabolism are influenced by the pH in relation to the electrolyte [97]. In summary, increased power output is achieved by microbial growth when the pH is between 6 and 9.

Temperature

Temperature has an impact on MFC performance, which in turn has an influence on microbial metabolism of microbes, thermodynamics and mass transfer. It has been demonstrated that MFC operates best at a 25–30°C temperature range [72]. Raising the temperature will cause more bacterial activity, which will improve the MFC's output power [90]. Consequently, a number of studies have demonstrated that temperature has a role in the initial generation of biofilms when using MFC [29].

According to Obileke *et al.* [72], 30°C is often much more favorable in order for MFC to obtain higher power production. This is so since bacterial biofilm is most catalytically active between the previously specified temperatures. According to reports, there are certain ideal growth temperatures for microorganisms: psychrophiles (10–15°C), mesophiles (25–40°C), thermophilic (50–84°C) and hyperthermophilic (80–110°C). Microbial operations decrease when temperatures are very low, and in most situations, MFC eventually becomes inactive [38]. High coulombic efficiency (CE) can be achieved by MFCs using psychrophilic electroactive microorganisms (EAMs) even at low temperatures [75]. On the other hand, temperature fluctuations may result in distinct microbial populations within the MFC's anode compartment [66].

Tkach *et al.* [96] understudied generation of pure and mixed isolates in MFCs at 10°C and 5°C. The data showed that the mixed isolates of MFC functioned better at 10°C compared to pure culture (*Shewanella* sp.), yielding a maximum power output voltage range of 550–565 mV, which is about 15% more than that of *Shewanella* single species. The maximum density was 465.3 mW/m² (68.7 mW/m²) when compared to the pure culture. The type of anodic EAMs and the properties of the biomass in MFCs determine the temperature effect. According to reports, bacteria can grow in four categories of ideal growth temperatures:

mesophiles ($25-40^{\circ}\text{C} < 45^{\circ}\text{C}$), thermophiles ($50-85^{\circ}\text{C}$), hyperthermophiles ($80-113^{\circ}\text{C}$), and psychrophiles ($10-15^{\circ}\text{C} < 20^{\circ}\text{C}$) [93]. Nonetheless, the majority of the identified EAMs fall into the mesophilic group. Microbial reactions slow down at very low temperatures, and finally, in most situations, MFCs cannot function [38]. On the other hand, MFCs containing psychrophile EAMs can achieve significant CE while operating at moderate temperatures. Temperature adjustments between 20 and 55°C were used to assess the impact of temperature on the performance of dual-chambered mediator-less MFC [65].

The COD elimination effectiveness of 84% was found to be highest at 40°C working temperature. The effects of different operating temperatures on the performance of MFC were investigated with an adsorption system (MFC-AHS) using palm oil mill effluent as a substrate. It was discovered that 35°C was the ideal operating temperature for such a system. Additionally, the results showed that while the maximum PD followed a polynomial function, the maximum current density increased linearly with temperature at a rate of $0.1772 \text{ mA/m}^2/^{\circ}\text{C}$ [65]. Single-chambered and dual-chambered MFC operation at various temperatures between 4 and 35°C . The temperature was found to be a critical factor in both the removal of COD and the production of bioelectricity.

At 4°C , the maximum power density (PD) of 15.1 mW/m^3 reactor was obtained, and at 35°C , the maximum PD of 174 mW/m^3 reactor was obtained, yielding a 58% removal of COD [65].

External resistance

One of the most prevalent factors influencing MFC that directly affects start-up time is external resistance. When the right external resistance is selected, while operating an MFC, losses within the system are reduced [61]. As external resistance decreases, MFC performance - removal efficiency and output current - both rise [11, 22]. One of the most prevalent factors influencing MFC that directly affects start-up time is external resistance [64]. Start-up time reduces by increasing resistance.

In one study, for example, a decrease in external resistance resulted in a three-day startup time reduction to 0.6 days. This indicated that at higher resistances, bacteria proliferate more quickly [93, 109]. As soon as the operation commences, output current rises as the resistance falls. It is significant to note that at reduced resistances startup process becomes difficult because they impede the rate at which microorganisms develop and evolve. Furthermore, low external resistance results in a thin and compact biofilm, and higher external resistance also enhances biomass yield [109].

To get a higher current generation, it is therefore preferable to begin running an MFC at high resistance and progressively reduce resistance as operation continues [61]. According to Buitrón's research, power production and efficiency decreased when the resistance was changed from the adaption resistance

[12]. As resistance varies, so does microbial and metabolic diversity [112].

Organic substrates

Substrates that are organic are also vital. The MFC method can be applied to a variety of substrates to enable energy production, including complicated mixtures from materials found in wastewater or pure chemicals [27]. Since low MFC yields are the result of the organic substrate's insufficient ability to supply energy to the bacterial population, its stability could have an influence on the amount of energy generated.

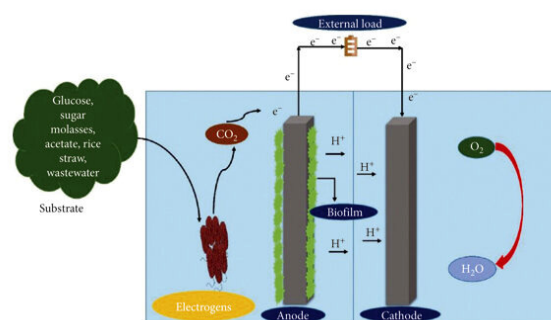


Figure 2. Systematic picture of MFC (culled from Fadzli *et al.* [27])

Acetate is often used as a substrate when a carbon source is needed to activate electroactive bacteria. Acetate therefore stands as a perfect yardstick for recent MFC materials, reactor prototypes or operating conditions because of its inherent capacity to transform microorganisms, in processes such as fermentation at room temperature and also methanogenesis [44, 48]. Four different substrate type of power generation and columbic efficiencies were recently differentiated [27]. The most amount of columbic efficiency (72.3%) was obtained from MFC fed with acetate, succeeded keenly by butyrate, propionate then glucose with percentages of 43, 36 and 15 respectively. More so, the cell's half-life can equally be affected by the choice of substrate [27]. Many different types of substrates, ranging from straightforward chemicals to intricate blends of organic materials, have been used in MFCs. Pure substrates including proteins, cellulose, cysteine, glycine, glycerol, butyrate, acetate, and lactate were utilized in certain instances. When used as an electron donor by dissimilatory iron-reducing bacteria, acetate is a non-fermentable substrate that can produce up to 66% more energy than butyrate. Wastewater is one type of substrate that MFCs can process and is rich and sustainable. Various reports have been published on the direct generation of electricity from complex organic wastewater. These include wastewater from pig farms, rice mills, tanneries, slaughterhouses, molasses, refineries, wineries, breweries, chemical industries, and wastewater with high sulfur content. Another element that needs to be taken into account is the substrate concentration. Power output increased from 0.2 to 1.2 W/m^3 when substrate concentration was increased from 100 to 850 mg/L . The power output remained unchanged at high concentrations of $1000 - 1500 \text{ mg/L}$ [87].

Table 2. shows some substrates in MFC with performance (culled from Fadzli *et al.* 2021 [27]).

Type of organic substrate	Volume	Inoculum	Type of MFC	Power density (mA/cm ²)
Carboxymethyl cellulose	1 g/L	<i>Clostridium</i> sp. <i>G. sulfurreducens</i>	Double chambered MFC of ferrocyanide catalyst having graphite plates as electrodes.	0.05
Cellulose	4 g/L	<i>Enterobacter cloacae</i>	MFC with U tube having cathode and anode carbon fibers.	0.02
Furfural	6.8 mM	Microorganisms from an MFC's ferricyanide-cathode anode	Single-chambered MFC having cathode and anode carbon paper.	0.165
<i>Chlorella vulgaris</i>	2500 mg/L	Wastewater treatment facility	Platinum cathode and graphite brush anodes in air-cathode MFC (25 mL).	0.20
Sucrose	2674 mg/L	Septic tank anaerobic sludge	MFC in two chambers with ferricyanide catholyte and braided graphite anode (7 cm ²).	0.19

Lignocellulosic materials made from agricultural leftovers are an ideal feedstock from producing cheap electricity since they are readily available and renewable [106]. Sadly, they cannot be used directly by microbes in MFC energy production until they have been broken down into reduced substances like monosaccharides [106]. Every monosaccharide that is directly formed from the hydrolysis of lignocellulosic material poses as a respectable energy source in MFC.

Energy production from cellulose requires a microbial population that can perform cellulolytic and exoelectrogenic activities [46]. Operations such as acidic steam-exploded hydrolysis procedures, which separate hemicellulose into soluble sugars, in order to supply samples for their analysis were used to produce biomass from maize stover waste in MFC power generation [27].

Microbial consortium

Electron mediators for MFCs are produced by a broad range of bacteria, consisting and *Pseudomonas* species, *Shewanella* species and *Geothrix fermentans* [13, 27]. *Geobacter sulfurreducens* is one of the most well-researched electrigenes that can achieve large current densities in an MFC. It's interesting to note that *G. sulfurreducens* creates biofilms thicker than about 50 μm . Most of the cells in the packed biofilm are engaged in metabolism successfully adding to the generation of current, according to [35, 43]. According to expression of gene studies, *G. sulfurreducens* forms microbial nanowires, which are crucial for long-distance electron transfer through its biofilms; see figure 3.

A biochemical and distribution system for lactate transport was built in the strain of *Klebsiella pneumonia* employed in a bioengineered microbial consortium that was isolated from the hydrolysis of corn stalks by expressing the genes for lactate dehydrogenase (ldhD) and lactate transporter (ldP) [50]. When the phosphotransacetylase (pta) and alcohol dehydrogenase (adhE) genes were removed, *K. pneumoniae* was able to convert biomass hydrolysate to lactate as fuel for the production of electricity, thereby blocking the ethanol and acetate pathway [50].

Shewanella oneidensis uses the altered *K. pneumonia* to ferment the hydrolysate biomass into lactate, as fuel. Additionally, a *Bacillus subtilis* heterogeneous flavins production pathway was expressed in *S. oneidensis* to improve its efficiency to

transfer electrons extracellularly. When it came to producing power from biomass hydrolysate, the genetically modified microbial consortia demonstrated excellent efficiency. It has been demonstrated that microbial communities created through genetic engineering are very effective in producing energy from organic hydrolysates [50]. Lin *et al.* [54] improved the synthesis and transport of flavins of *Shewanella oneidensis*, thereby increasing its extracellular electron transfer (EET) efficiency [56]. McNulty *et al.* [63] concentrated on expanding the substrate to generate power for MFC systems. By forming a synthetic consortium and utilizing modified strains of *Methanosarcina acetivorans*, *Paracoccus denitrificans* and *Geobacter sulfurreducens*, they were able to convert methane into energy [52, 63].

The co-production of electricity and hydrogen (H_2) has become quite interesting for MFC application since both energy products can be generated in a single operated MFC, considerably lowering the cost of reactor and operation [66]. The rapid metabolism of xylose in MFC was achieved by isolating and acclimating a new strain of exoelectrogenic yeast called *Cystobasidium slooffiae* JSUX1. This strategy allowed for bioelectricity production and biohydrogen simultaneously, with electrons being harvested from xylose- H_2 fermentation [65]. Wheat straw has also been studied and tested to be a potential fuel for power

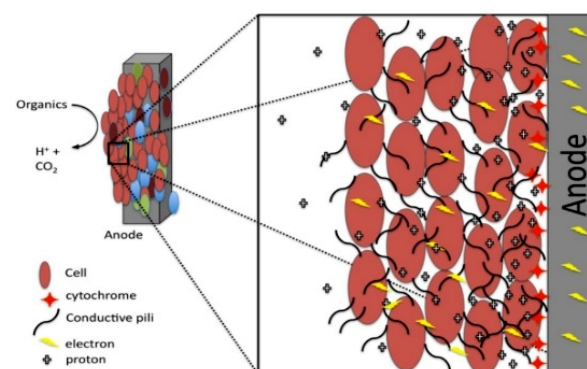


Figure 3. shows the potential of a *Geobacter sulfurreducens* biofilm to transport electrons across membrane-bound cytochromes when it is in close proximity to the electrode, while the cells that are closest away from the electrode can employ a conductive nanowire network to commute electrons along longer distances to the electrode. Protons build up inside the biofilm as a result of the organic substrate's oxidation throughout the biofilm (culled from Frank and Nevin, 2010 [28]).

generation involving MFC, with an intent to generate electricity through microbial communities utilizing biomass from the straw [73].

Molecular biology techniques have made it possible to develop microbial consortia which feed on organic materials as fuels for electrical generation, as well as to elucidate the pathways for the steps involved in electron transfer. Thus far, diverse technical strategies have been employed, such as random techniques rational design (like the engineering of metabolic processes, bacterial pili, and heterologous gene expression), and new designs like hybrid MFC-enzyme based fuel cells, bacterial surface show of redox proteins, and yeast surface display of redox proteins have all been examined for the genomic engineering of an optimal biocatalysis in MFCs [19].

Mixed microorganisms of electrogenic and electrophilic origin contribute to current generation in MFCs more effectively than pure isolates of bacteria [57]. There is a great deal of promise in using single and mixed community biofilms in MFC; such consortia as fungi-like yeast and bacteria are involved. *Geobacter* strain was used along-side sodium acetate as a microbial consortium and fuel source respectively [42]. The system worked with a cathode made of platinum at a voltage of 0.6 V - 1.1 V. Due to its improved design, hydrogen production efficiency increased to 91.80 [42].

A separate study that employed the fungus *Trametes versicolor* in conjunction with *S. oneidensis* showed increase in power generation using bacterial-fungal combination that hitherto increased power production, harnessing a power density as high as 0.78 W/m³ [25]. According to de Dios *et al.* [25], electrons form the filamentous network of *T. versicolor* and attract the bacteria, eventually sticking to it. Moreover, *T. versicolor* is capable of producing oxidative enzymes that function as an oxidoreductase mechanism, moving electrons from donor to acceptor [25]. It is obvious that diverse biofilm communities, capable of producing power through several processes, will be essential to the improvement of MFCs [53].

The growth and behavior of electroactive biofilms can be bettered by the application of synthetic biology, selective growth condition control and bioengineering techniques [4, 17, 54].

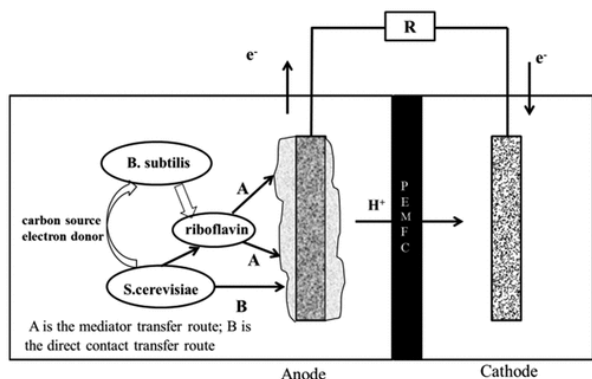


Figure 4. Detailed diagram of the anode electron transfer route of mixed culture in MFC (culled from Ren *et al.* [81])

Earlier MFC Performance Analysis

According to Idris *et al.* [36], the MFC can reach 0.153 V and 0.202 V respectively, whether the air pump is attached or not; this is sequel to the findings of Barua *et al.* [7], who produced an output of 0.202 V. It proved beneficial even when the voltage supply was cut off because of decreased bacterial activity. More so, a microbial complex that was enhanced for the production of energy utilizing organic waste was isolated using a single and dual chamber MFC. *Bacillus licheniformis* produced nominal voltage range of 0.93 - 0.95 V, according to the study's findings [7]. How degradable organic matter is, its temperature and lack of hazardous compounds, all influence how efficient the workability of a MFC will be.

Chouler *et al.* [20] performed research with human urine as the organic substrate intending to create small-scaled but effective MFC for energy production. This study examined effects of two catalysts with various biomass origins as well as the impact of length of electrode. Chouler *et al.* [20] concluded by mentioning that adding an oxygen reduction reaction catalyst made of biomass increased the MFC's electrical density to 1.95 W/m³ and its power density to 0.580 W/m³ from 0.053 [20]. Undiluted pee has the potential to be used as the main feedstock for small-scale MFC stacks and other MFCs, as demonstrated by Ieropoulos *et al.* [37]. The findings demonstrated that when 48 tiny scales were linked together in a stack and powered by urine, the power density rose to 1.5 W/m³ [37].

MFC-Based Biogas Production

Electroactive methanogen activity in MFCs makes it possible to create biogas, particularly biomethane. Methanogens are microorganisms that, in oxygen-limited conditions, produce methane as a metabolic byproduct. They produce biomethane by using the acetate and carbon dioxide produced during the MFC process [76]. Small voltage must be applied to the electroactive methanogens to increase their activity in order to improve biomethane production with an MFC [71]. Alternative strategies that focus on achieving specific outcomes include choosing methanogenic strains that exhibit greater electroactivity, modifying variables like pH, temperature, and substrate concentration, and choosing electrode materials that have the potential to improve electron transfer [3, 55, 95].

Limitation factors of MFC Technology

The application of microbial metabolism in MFCs presents a promising avenue for producing electricity as a sustainable energy technology. According to Slate *et al.* [89], the frequency of transportation of electron to the anode pole and the electrochemical characteristics of the material are primary inherent factors opposing the power production of MFC setups today. Low energy outputs, short lifespans, and high related costs (mostly because of electrode substances and the utilization of PEMs) are the other known limitations of MFCs. For widespread use, MFCs' existing power densities are unsatisfactory [91]. The

existing limitations of MFCs prevent them from reaching their theoretical power outputs, making the industrial application of this technology difficult.

In one instance, the production of non-conductive debris, such as dormant cells and/or polymeric materials, eliminated the active electrochemical biofilm from the electrode's periphery by employing more porous electrodes, or else it was trapped in a three-dimensional structure. As a result, there was eventually less surface area available, which decreased current generation [9].

There are additional biofilm-related elements that are believed to influence an MFC's performance. Zhang *et al.* [111] used *Geobacter anodireducens* as the main bacterium in a MFC setup. It was noticed that a two-layered biofilm with an outer layer of active cells and an inner layer of inactive cells gradually formed. These results indicated that current generation occurred in the outer layer, while the inactive inner layer persisted as a matrix with electrical conductivity [92].

It could be that the mechanism of electron transfer determines this continuous electrochemical activity since *Geobacter* spp. possess nanowires that have a lively link to the periphery of the electrode, they are recognized as bacteria with unique electrochemical activity. Additional elements that could be detrimental on MFC's power output and effectiveness are the crossing of biomass compounds from the cathode to the anode (and vice versa) and electro-catalysts inactivation, if any are present.

Optimization of MFC Power Output

Determining the gaps in MFC functionality and its various uses has become crucial for configuring a successful MFC system. Researchers are investigating methods to improve the metabolic activity and electron transfer propensity of MFCs, such as optimizing the microbial communities within them [2]. The majority of microorganisms, for instance grow on the electrodes to create biofilms, which are sticky surfaces containing the microbial community and made of extracellular polysaccharides (EPS). Although extracellular polysaccharides physically immobilize bacteria, it allows communication and interaction between cells. In MFCs, this connection between cells facilitates electron transport and electron-electrode contact [4]. Electrons must travel a particular distance to attain the electron acceptor; this determines the efficiency of electron transfer in an inverse manner. Thus, having a good understanding of how biofilms affect electron transfer may help create novel strategies for upgrading the efficiency of microbial fuel cells. Because the internal coating of dormant cells grows within the biofilm layout, biofilm that forms naturally thickens as the culture gets older but remains electrochemically inert all through [92].

As a result, one method that is seen to be encouraging for raising the efficiency of MFCs is inducing lively microorganisms so that more electrically active biofilms will be created [4]. The organism's level of EPS secretion correlates with the

biofilm's density. More microbes are found in a thicker biofilm than in a lighter one. Consequently, as the age of the organisms increases, so does the biofilm's density. It has been discovered that three polysaccharides, Psl, Pel, and alginate polysaccharides, are involved in the production of biofilms. Psl polysaccharides, or EPS, are made from Psl genes in five consecutive steps: microcolony creation, biofilm maturation, dispersion of the biofilm, first attachment, and irreversible attachment [4]. Therefore, elevated development of biofilms by lively microorganisms in MFC could be caused by overexpression of these genes. The Psl polysaccharide functions as a scaffold during the creation of the biofilm matrix by securely adhering to the bacterial cell wall in a helical shape and encouraging robust intercellular connections. The presence of EPS in large quantities can better the working ability of MFCs with microbial biofilms because it plays a crucial function in creation, stability, life of biofilms structural integrity and adhesion property [4].

In a different method, a strong anodic electrochemical biofilm was established using a synthetic fermenter-exoelectrogen possessing a microbial combination of *Escherichia coli* - *S. oneidensis*. In summary, a water loving *S. oneidensis* specie called CP2-1-S1 is utilized to strengthen its adherence to the carbon electrode as the exoelectrogen. To enable flavin-mediated electron transfer, the synthetic riboflavin pathway of *Bacillus subtilis* was revealed in *E. coli*, causing an excess of flavins to be produced. *S. oneidensis* had an advantage over *E. coli* in adhering to the anode surface because of the very hydrophobic interactions between the anode and the flavin overproduction by the engineered *E. coli*. This intentionally produced anodic biofilm had a greater catalytic current (from 0.19 to 1.84 A/m² at 0 V) due to altered microbial community makeup. The MFC with xylose as its substrate was inoculated with modified microbes and yielded a power density 6.8 times greater than the naturally existing coculture-inoculated MFC [105].

Modification by introducing new materials

The introduction of novel anode materials in MFC such as natural polymers, melamine sponges, berl saddles, carbon nanotubes and high capacitance electrode materials has proven successful. The voltage output, start-up, current production, stability, and recovery rate of the sensor's response can all be enhanced by modifying the anode system with natural polymers like polyacrylamide with neutral red (PA + NR) and conductive polymer polypyrrole with positive functional groups poly (pyrrole-alkyl ammonium). The microbial fuel cell can benefit from the wide electrically conductive surface provided by melamine sponges coated with reduced graphene oxide/carbon nanotube (rGO-CNT) sponges, as well as the high porosity that promotes efficient mass transport and electron transfer. The rGO-CNT sponges outperformed conventional carbon- and metal-based anodes in terms

of electrochemical characteristics and durability [91]. Carbon-coated berl saddles are a novel, inexpensive anode material that effectively recovers the electrons generated by bacterial metabolism and promotes the best possible bacterial adhesion. It offers a high void bed and a comparatively large useful contact surface area. It also demonstrated a maximum power density that was around two to three times greater than the results obtained with commercial anode materials.

The new anode materials based on Carbon Nano Tubes (CNTs) exhibit a distinct biofilm morphology and outperform bare gold electrodes in terms of performance. High surface anode treatment of distillery effluent was achieved by interlacing carbon yarn with stainless steel. Because of its unique nanostructure for high electro catalytic activity, which is attributed to improved active surface area of the electrode, strengthened bacterial adhesion, and enhanced interfacial charge transfer between bacteria cells and electrode, the Nickel Oxide (NiO)/carbon cloth anode delivers a maximum power density that is about three times higher than that of the plain carbon cloth anode. The cloth's surface is hydrophobic when it is left untreated, but it becomes extremely hydrophilic when NiO nanoflakes are added [87].

Anode modification with nanocomposite materials is more favorable and improves MFC's ability to generate power. Tin Oxide (SnO_2) stood for endorsing characteristics for MFC anode materials. As anode materials in MFC, nano composites of reduced graphene oxide and tin oxide (RGO/ SnO_2) and multiwalled carbon nanotubes and tin oxide (MWCNTs/ SnO_2) worked well. Maximum power densities of 1421 mW/m^2 , 699 mW/m^2 , and 457 mW/m^2 were demonstrated using MWCNTs- SnO_2 /Glassy Carbon Electrode (GCE), MWCNT/GCE, and bare GCE anodes, respectively. The larger specific area of the MWCNTs and graphene, as well as the synergistic effects of SnO_2 with these materials, were credited with improving the MFC's performance. The nano composite's high conductivity and huge specific surface area significantly enhanced the growth of biofilms and accelerated electron transmission [87].

Modification by treatment

Rather than the anode surface's concentration of oxygen-containing groups, the shape of the MFC determines its power production. To increase MFC power generation, careful consideration should be given to surface morphology design. Anode materials have had their surfaces modified through electrochemical, acidic, and oxidation processes. The oxidation process gives the carbon felt a rougher surface, makes it more biocompatible, and increases the power output of the MFC. The graphite felt anode's surface was electrochemically treated to promote biofilm growth on the electrode, which increased current output quickly. Strong hydrogen or peptide connections between the carboxyl groups generated on the electrodes and the amide groups of bacterial

components, especially cytochrome c, were the primary cause of biofilm growth on the treated anode [87]. Reduced electrode resistance and start-up time were noted as a result of the carbon cloth anode material's acidic alteration, which also raised the ratio of saturated to unsaturated carbon on the surface. Strong hydrogen or peptide connections between the carboxyl groups generated on the electrodes and the amide groups of bacterial components, especially cytochrome c, were the primary cause of biofilm growth on the treated anode. Reduced electrode resistance and start-up time were noted as a result of the carbon cloth anode material's acidic alteration, which also raised the ratio of saturated to unsaturated carbon on the surface [87].

There has also been an increase in bacterial attachment and electrochemical accessible surface area (ECSA). A micro-porous layer (MPL) was added to the carbon veil (CV) and carbon cloth (CC) anodes, increasing the bacterial population, improving power performance (2.2 and 1.8 times that of the unmodified CV and CC), and enhancing the stability of the MFC [87]. Since microorganisms can act as catalysts to aid in the electron transfer, the use of metal catalysts or artificial electron mediators is avoided, lowering the cost of building and operating MFC. In order to investigate new sustainable technologies for metal treatment, two common oxidized-status metals (Fe (III) and Cr (VI)) were investigated as electron acceptors on cathodes in single chamber microbial fuel cells (SCMFCs). The DNA band patterns of anode biofilms were shown to be identical by microbial community analysis, however cathode biofilms varied according to electron acceptors. The high (>89%) conversion rates of Cr (VI)/Cr (III) and Fe (III)/Fe (II) show that metals have performed well as electron acceptors on cathodes [87].

Magnetic biological effect is another phenomenon that can occur when living microbes are released into a magnetic field. This effect causes a range of biological actions in microbes. Such as DNA, enzymes, and the structural organization of biofilms are impacted by magnetic fields, which modify the metabolism of the microorganism [21]. Higher static magnetic fields suppress the physiological functions of microorganisms, while lowered static magnetic fields promote microbial performance and metabolism [89, 115].

According to Ye *et al.* [108] applying a magnetic field (between 0 and 300 mT) reduced the time requisite for MFCs to start up. It took the lowest amount of 100 mT over a 7-day period to produce an uninterrupted voltage output [108]. Again, Ye *et al.* [108] reported that MFC of 200 mT magnetic field produced highest power density of 1.56 mW/m^2 which was greater than 1.19 mW/m^2 of control setup which was without a magnetic field. However, when exposed to a magnetic field of 300 mT, the MFC generated 0.99 mW/m^2 power density. Therefore, a spectrum of

Table 3. show various electrode materials and their effect on MFC performance

Material	Merits	Demerits	Effect on MFC Performance	References
Carbon nanotubes (CNTs)	It is bio-compatible, has high electrical conductance and high surface area.	It is expensive and hard to manipulate.	Good power output and biofilm formation.	[67]
Conductive Polymers (CPs)	It is affordable and bio-compatible and it's easy to synthesize.	It is susceptible to fouling, unstable and has poor mechanical strength.	Good power output and biofilm formation.	[69]
Metal Oxide (MOs)	Very stable, good catalytic ability and affordable.	Leaching of metal ions, poor bio-compatibility.	Unstable power output, electron transfer depends on composition and type of MOs.	[101]

magnetic fields with optimal intensity depends on the microorganisms used in MFC setups.

It has been demonstrated that using MFCs with greater magnetic fields of 360 mT, had negative impact on cell activity which led to loss in organic material, a decrease in removal of pollutant in the case of ammonia nitrogen (84.6 ± 0.5 mg/L) and a reduction in the maximum voltage (171.8 mV) [114].

The low power output of MFCs can be compensated for by combining them with other renewable energy technologies, such as solar cells or wind turbines, to create hybrid systems that guarantee reliable energy production. For long-term operation and smooth performance, maintaining microbial diversity and stability in MFCs is essential [74].

Leveraging photosynthesis to optimize MFC

The photosynthesis-related electrons are essential for MFC optimization. The photosynthetic electron transport chain (PETC) is used by photosystem I (PS I) and photosystem II (PS II) to transfer electrons. When PETCs and MFCs are combined, an artificial transport chain is formed, linking the plant's photosystems to the MFC anode. According to this promising approach, electrons are produced during photosynthesis and moved to the anode, where they are used by electrogenic bacteria to produce electrical current. The MFC microbes utilize photosynthetic products, such as organic acids and carbohydrates, that are secreted into the rhizosphere as substrates. This approach can improve MFC efficiency and promote a symbiotic relationship between microbes and plants [88].

CONCLUSION

MFC might be used as a substitute in the remediation process to get rid of risky substances from wastes and xenobiotics [31]. MFC is capable of generating energy from a variety of organic resources. Using genetic engineering to enhance MFC system performance has been tested to be a successful strategy. Even if a number of wild-type strains have produced electricity, appropriate techniques must still be used to increase the strains' electrochemical activity. Numerous physical and chemical techniques have been proposed as viable means of improving strains' capacity to enhance power [17, 56, 92].

It has become novel to use MFC in biosensors. According to a study, bacteria are able to recognize the target material on their own and react when the MFC

produces electrical energy, which reflects the analyte concentration through variations in battery voltage and current. As a result, signal converters and outside power limitations can be eliminated, and sensor development can proceed more quickly. The majority of its current applications include biological oxygen demand (BOD) analysis and research, poisonous substance research, microbiological amount and activity, volatile short-chain fatty acid research, and other substance research [103]. An eco-friendly restoration technique called coupling technology treats wastewater and generates bioelectricity at the same time. MFC uses the microbial breakdown of organic waste in sewage to produce energy, whereas CW uses the same process to purify sewage [103].

Microbes must use substrates in order for MFC systems to function. As a result, the choice of substrates must take into account how well a given strain uses a given substrate. Stable conditions both inside and outside the MFC system are necessary for its effective functioning. Appropriate control over operating conditions is necessary to facilitate the scaling up of MFC systems. The primary external component still influencing MFC system performance is temperature. The low temperature may cause MFCs to operate less efficiently and require longer timeframes to start up. Therefore, to obtain steady performance in a wide range of temperatures, MFC systems still require technical improvement. Finding a balance between cost and power generation efficiency, as well as large-scale practical application, is the most pressing solution for MFC's future. Going forward, the following points can be used to improve specific methods to enhance MFC power generation performance: improving the MFC reactor's reaction efficiency by optimizing its structure through the application of heavy metal loading and microbial immobilization technologies, choosing membrane materials that support proton transfer, have a lengthy life cycle, and can withstand microbial migration. The membrane materials' physical and chemical characteristics and the way biofilms are domesticated should be enhanced as processing costs are reduced. Using a combination of bioengineering concepts and screening the newly discovered electric-producing microorganisms in the anode chamber can ensure the battery operates steadily in a variety of scenarios.

Again, to enhance the electrical performance of the battery, the proper external resistance should be

selected in subsequent trials to encourage the anode microorganisms' electron transmission. Finally, integrating immunological and DNA technologies will increase the spectrum of applications for microbial fuel cell sensors.

Reducing energy loss and maintaining high energy production efficiency should be the major goals of MFC operation and maintenance. In order to fully implement MFC technology, several key research areas will need collaboration by engineers and scientists from various disciplines, including environmental, civil, chemical, process and computer science, microbiology, technology, also governmental organizations, and investors.

Conflict of interest. There is no actual or potential conflict of interest in relation to this article.

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