



Green Wastes as a Bio-Anode in Microbial Fuel Cell for Wastewater Treatment: A Review

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Abstract

This review explores how an efficient carbonaceous anode can be developed from green waste through simple thermal carbonization processes at relatively low temperatures. These bio-derived carbon materials, when integrated into microbial fuel cells (MFCs), exhibit promising structural and electrochemical properties that support biofilm formation and efficient electron transfer. Analytical methods typically used in material science often reveal partial graphitic features and porous textures, both of which are beneficial for microbial colonization and electrochemical activity. Compared to conventional materials such as carbon felt, green waste-derived electrodes have shown the potential for enhanced performance due to their tunable surface chemistry and increased surface area. Moreover, microbial analysis of biofilms developed on such anodes frequently identifies electroactive bacterial species, some of which may be previously uncharacterized, especially in region-specific environments. These findings underscore the significance of utilizing sustainable materials in MFCs and exploring novel microbial communities capable of extracellular electron transfer in complex and nutrient-rich wastewaters. This review consolidates current research findings on the development and performance of green waste-derived bio-anodes for MFCs.

Keywords: Bio-Anodes; Sustainable Energy; Microbial Fuel Cells (MFCs); Biomass; Electroactive Bacteria; Extracellular Electron Transfer; Sugarcane Waste water

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1. Introduction

The contemporary world faces two of the most significant challenges: the increasing demand for energy and the urgent need to combat water pollution. Addressing these issues is essential for promoting a healthy lifestyle and sustaining a stable, eco-friendly environment. Consequently, the development and advancement of green energy technologies, alongside environmental conservation efforts, have become pivotal focal points of global attention more than ever.

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Renewable energy sources and advanced technologies have increasingly assumed a vital role in replacing conventional fossil fuels, contributing to improved environmental sustainability (Yaqoob, Parveen, et al. 2020).

Microbial fuel cells (MFCs) present a promising dual-purpose technology: they oxidize organic pollutants in wastewater using microorganisms, thereby generating electricity while simultaneously treating the water. (Yaqoob, Ibrahim, and Guerrero-Barajas 2021).

The utilization of natural resources provides an environmentally friendly approach to producing sustainable bioenergy from organic waste. Plant waste is the most commonly used natural precursor for carbon-based anodes. Typically, the direct carbonization of plant materials at high temperatures results in porous carbon structures. Additionally, heteroatoms present in plants (e.g., nitrogen, phosphorus, sulfur, silicon, etc.) act as natural dopants during the preparation process, leading to self-doped materials that can enhance the bioactivity of the resulting anodes. Furthermore, water evaporation and the release of byproduct gases during the graphitization of macromolecules in the carbonization process contribute to the formation of a porous carbon structure, significantly increasing its surface area (Zhang, He, Yang, Sun, Li, Han, Zhao, Shi, Feng, and Tang 2018). Figure 1 illustrates the working mechanism of microbial electrolysis cells, which utilize plant waste as a source of carbon and energy, where bacteria are employed to catalyze electrochemical reactions within the cell. (Source: U.S. Department of Energy, Office of Science—Biological and Environmental Research (BER)).

Moreover, harnessing energy from wastewater, which contains approximately nine times the energy required for its treatment, is an efficient approach to offset the energy consumption involved in the wastewater treatment process, for example (the United States consumes approximately 3–5% of the total electricity generated for wastewater treatment) (LOGAN 2008; Gao, Scherson, and Wells 2014). Many researchers have highlighted the significance of green waste as a valuable resource for developing sustainable materials. Various studies have focused on its potential applications in energy production and environmental technologies due to its abundance, low cost, and eco-friendly nature.

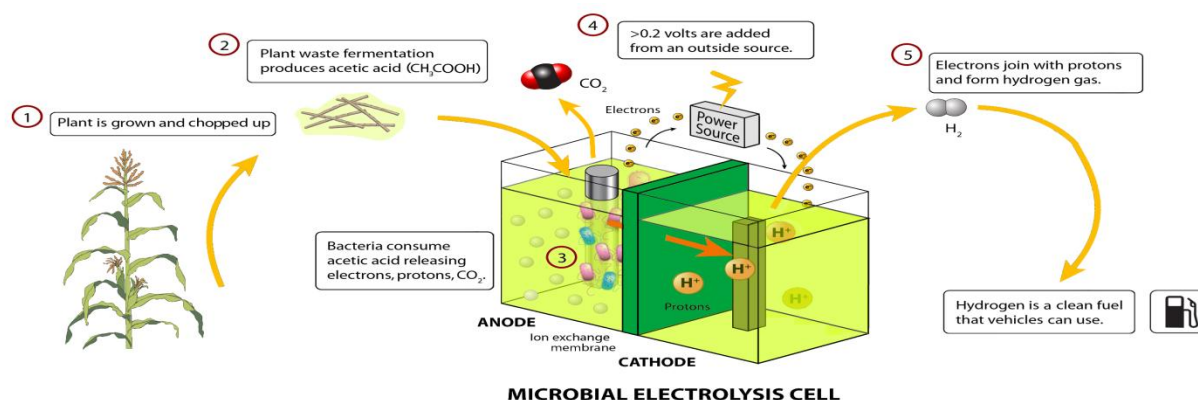


Figure (1). Microbial Electrolysis Cell Using Plant Waste

(Source: U.S. Department of Energy, Office of Science—Biological and Environmental Research (BER)).

This review aims to summarize the potential of green waste as an alternative resource for environmentally friendly and eco-friendly energy production and water treatment. It seeks to contribute to climate change mitigation by reducing reliance on conventional, polluting energy sources and by transforming organic waste into a valuable energy source instead of allowing it to decompose and emit harmful greenhouse gases like methane. The study also promotes the use of renewable and low-cost energy sources to reduce the environmental burden of agricultural waste accumulation. Additionally, it focuses on enhancing wastewater treatment efficiency using these waste materials, aiming to develop an integrated model that supports environmental protection, clean energy generation, and circular economy practices, particularly suited for rural and low-resource areas.

2. Definition and Working Principle

Bio-electrochemical systems (BESs) are emerging technologies capable of addressing both clean energy generation and wastewater treatment needs. Among these systems, microbial fuel cells (MFCs) are particularly notable for utilizing electroactive bacteria to convert organic matter into electricity (Gul and Ahmad 2019). These cells operate by transforming the chemical and biological energy in organic compounds, including those found in wastewater, into electrical energy. A typical MFC comprises key components such as an anode, a cathode, an organic feedstock, a permeable membrane, and microbial agents.

The cell's overall efficiency is influenced by various operational and material factors. Organic compounds serve as either electron donors at the anode—oxidized through microbial activity—or as electron acceptors at the cathode (He,

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Minteer, and Angenent 2005). Microbial fuel cells utilize electrogenic bacteria to degrade organic substrates in wastewater, releasing electrons that can be harvested as electrical energy. These microorganisms are fundamental to the electrochemical reactions that power the system.

Research has categorized several groups of microorganisms—including Proteobacteria, Firmicutes, Fungi, Acidobacteria, and Algae—that are capable of generating bioelectricity during their metabolic processes. Some other species have also demonstrated similar capabilities (Yaqoob, Khatoun, et al. 2020). Bioelectrodes are crucial in determining the performance of MFCs. They serve as the solid surface upon which electroactive bacteria can proliferate, leading to the formation of biofilms and facilitating the exchange of electrons. To ensure these characteristics, the physical and chemical characteristics, along with the structural design of the electrodes, play a key role in regulating electron movement at the interface between biological systems and inorganic materials, as well as in maximizing the surface area that supports the adherence and proliferation of electroactive microorganisms. (Palanisamy et al. 2019).

2.1. Advantages and Disadvantages of MFCS

Compared to conventional wastewater treatment, MFCs offer distinct technical benefits such as high conversion efficiency, ambient operating conditions, and potential for energy recovery. These characteristics make them suitable for integration into decentralized wastewater management systems and environmental remediation platforms. They have promising potential in applications such as heavy metal treatment, harmful algal biomass treatment, water desalination, and bio-electro-Fenton technology. Additionally, MFCs can operate effectively under mild conditions, such as ambient temperature and pressure. [(Zhao et al. 2015), (Logan and Rabaey 2012)] Despite significant advancements in the efficiency and applicability of MFCs, their large-scale implementation is still limited by factors such as low power density, high-cost electrode materials, a limited number of electricigens on the electrodes, and the inefficiency of extracellular electron transfer (EET), which restricts the output current density (Alipanahi and Rahimnejad 2018).

2.2 Types of MFCS

Microbial fuel cells can be categorized into several types based on their design, operational mode, and electrode placement. These include single-chamber MFCs, double-chamber MFCs, stacked MFCs, and mediator-based or mediator-less MFCs. Each type offers distinct advantages and limitations, making them suitable for specific applications and environments. Understanding the underlying principles and variations of these MFC types is crucial for advancing their scalability and practical deployment. Table 1 presents a comparison between single-chamber and double-chamber microbial fuel cells (MFCs) in terms of structure, ease of fabrication, efficiency, and applications, highlighting the key differences and the rationale for choosing each configuration.

Table 1. Comparison of Single- and Double-Chamber Microbial Fuel Cells

Aspect	Single-Chamber MFC	Double-Chamber MFC
Structure	One chamber, exposed cathode	Two chambers, membrane-based
Ease of Fabrication	Easy	Complex
Cost	Lower cost	Higher cost
Power Generation Efficiency	lower	Higher
Microbial Activity Control	Weak	Good
Oxygen Leakage	High	Low
Suitable Applications	Sensors, simple uses	Research, wastewater, energy

Electrodes are critical components in microbial fuel cells (MFCs), playing a fundamental role in the electron transfer process that drives electricity generation. The efficiency and performance of MFCs are largely determined by the properties and types of electrodes used. Selecting appropriate electrode materials and designs is essential for optimizing power output, enhancing microbial attachment, and minimizing resistance within the system. In MFCs, electrodes are typically categorized into two main types: anodes and cathodes, each with distinct roles. The anode facilitates the oxidation of organic matter by microorganisms, enabling the transfer of electrons to the circuit. In contrast, the cathode serves as the site for the reduction reactions, often involving oxygen or other electron acceptors. Both electrodes must be designed to support high conductivity, chemical stability, and compatibility with the microbial environment.

Electrode design plays a crucial role in the performance of microbial fuel cells (MFCs). One key distinction in electrode technology is the difference between 2D (two-dimensional) and 3D (three-dimensional) electrodes. Both types have their advantages and are used based on the specific requirements of the MFC system, such as power density, microbial attachment, and the efficiency of electron transfer. Table 2 illustrates the comparison between 2D and 3D electrodes in terms of performance and application in microbial fuel cells (MFCs).

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2D electrodes typically consist of flat, planar surfaces, which can be made from various materials such as graphite, carbon cloth, or other conductive materials. These electrodes are the traditional type used in MFCs and are known for their simplicity and ease of fabrication. Advantages: It is easy to fabricate and cost-effective, has uniform current distribution across the surface, and is suitable for simple laboratory-scale MFC designs. Limitations of 2D: Limited surface area, which restricts microbial colonization and electron transfer, and Low power output compared to 3D electrodes due to the relatively small surface area available for microbial activity and electron exchange.

3D electrodes offer a more complex design, featuring a porous or structured surface that increases the overall surface area available for microbial colonization and electron transfer. These electrodes often consist of materials such as carbon nanotubes, graphite foams, or conductive polymers. Advantages: Significantly increased surface area compared to 2D electrodes, leading to enhanced microbial attachment and greater electron transfer capacity. Higher power density due to the larger area for microbial activity and better conductivity. Improved efficiency in larger-scale MFCs, especially in wastewater treatment applications. Limitations of 3D: More complex fabrication processes may increase the cost. Higher resistance due to the increased complexity and potential for reduced flow of electrons through the porous structure.

Table (2). Comparison of 2D and 3D Electrodes in MFCs

	2D Electrodes	3D Electrodes
Surface Area	low	High porous
Power Density	Low	High
Electron Transfer	Limited	Enhanced
Fabrication Cost	Low	High
Application	small-scale	large-scale
Efficiency	Moderate	High

It is worth noting that most anode materials developed so far have been designed in two-dimensional (2D) forms. However, the relatively low surface area and the absence of appropriately sized pores to support microbial growth in these flat electrodes remain significant challenges for scaling up 2D electrodes. Additionally, the formation of thick biofilms hinders the diffusion of organic substrates from the outer to the inner surface, adversely affecting the colonization of beneficial microorganisms. As a result, three-dimensional (3D) porous electrodes have garnered increasing attention (Yuan et al. 2013).

3. Factors Affecting the Efficiency of Microbial Fuel Cells (MFCs)

Several factors influence the efficiency of microbial fuel cells (MFCs), each playing a direct role in determining the system's overall energy output from organic matter. These factors can be summarized as follows:

3.1. Electrode Material and Structure:

The efficiency of microbial fuel cells (MFCs) is highly sensitive to variations in electrode design and composition. Changes in material porosity, electrical resistance, or surface chemistry can significantly impact microbial colonization dynamics and electron transfer rates. For instance, electrodes with insufficient surface complexity may limit bacterial attachment and biofilm development, leading to lower current output. Similarly, materials with poor conductivity or surface defects can increase internal resistance and disrupt the flow of electrons. Thus, optimizing electrode parameters is essential not only for maximizing performance but also for maintaining operational stability under real-world wastewater conditions.

3.2. Substrate Type and Concentration (Organic Matter):

The type of organic substrate added to the system greatly affects electrochemical performance. Readily biodegradable materials, such as glucose, are more efficient than complex compounds. The substrate concentration must be kept within an optimal range; low concentrations may not provide sufficient energy, while high concentrations can cause microbial inhibition or imbalance in the pH levels within the cell.

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3.3. Microbial Community:

The efficiency of the system is highly dependent on the type of microorganisms present. Certain species, such as *Geobacter* and *Shewanella*, are known for their excellent electron transfer capabilities, which significantly improve system performance. Moreover, biofilm formation is a crucial factor, as dense and stable biofilms enhance the interaction between the bacteria and the electrode surface.

3.4. Internal Resistance:

Internal resistance encompasses several components, including ohmic resistance, which depends on the distance between electrodes and the conductivity of the medium; charge transfer resistance, which is influenced by the electrode surface properties; and mass diffusion resistance, which relates to the ease of movement of substances (substrates or products) within the system. These resistances directly impact the voltage output and, consequently, the overall efficiency of the cell.

3.5. Environmental Conditions:

Operating conditions must be controlled to maintain high efficiency. pH level is particularly influential, with a neutral value around 7 being optimal for microbial activity. Moderate temperatures (25–35°C) support bacterial metabolism and energy production. In contrast, oxygen leakage into the anode chamber negatively affects performance, as it consumes electrons before they can reach the electrode.

3.6. Membrane Properties:

In systems utilizing a membrane to separate the anode and cathode chambers, proton exchange efficiency is a key factor in maintaining ion balance. However, membrane fouling or clogging over time may lead to performance degradation. Designs that omit the membrane can reduce system cost and internal resistance but may compromise selectivity between the electrode reactions.

3.7. Reactor Design:

The engineering design of the reactor significantly affects performance. Whether the reactor is configured as a single- or dual-chamber system influences oxygen crossover and internal resistance; other factors such as flow rate, reactor size, and orientation also directly impact the overall operational efficiency and long-term stability of the system.

4. MFCs Anodes

The anode is a critical component in microbial fuel cells (MFCs) and plays a central role in facilitating the generation of electricity. It serves as the site where microorganisms oxidize organic substrates, such as glucose, acetate, or wastewater, releasing electrons as a byproduct of their metabolic processes. These electrons are then transferred to the anode, initiating the electron flow that powers the MFC.

To ensure efficient performance, the anode must meet several key requirements, including high conductivity, biocompatibility, and a large surface area to promote microbial attachment and electron transfer. The material and design of the anode significantly influence the efficiency of the MFC, with commonly used materials including carbon-based substances like graphite, carbon cloth, and carbon paper, and advanced materials such as carbon nanotubes or conductive polymers.

Anodes in MFCs are typically designed to maximize microbial colonization and optimize the contact between the microorganisms and the electrode surface. Recent advancements in surface modifications, such as coating the anode with biocompatible materials or introducing nanostructures, have shown significant improvements in electron transfer efficiency and overall power output.

4.1 Physical and Chemical Properties of the Anode

Anodes play a critical role in determining the efficiency of MFCs by enabling microbial colonization and electron transfer. Key physical and chemical properties for optimal anode function include: high conductivity, biocompatibility, porosity, and structural durability. Larger surface areas, especially in 3D configurations, provide enhanced biofilm formation and facilitate electron movement across the biofilm-electrode interface.

Due to their lightweight, porous architecture, high surface area, and economic advantages, carbon-based materials have become highly regarded in microbial electrochemical systems (MESs). Studies have shown that these materials support

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microbial growth and biofilm development by creating a conductive environment that facilitates extracellular electron flow. (Yang et al. 2019), (Li et al. 2019).

4.2. Using Natural Waste as an Anode in Microbial Fuel Cells

The utilization of natural waste as an anode material in microbial fuel cells (MFCs) presents an innovative and sustainable approach to enhancing renewable energy production. By repurposing organic waste materials, this method aligns with the principles of circular economy and environmental sustainability. Natural waste, such as agricultural residues, biomass, and processed plant-based waste, offers a cost-effective and eco-friendly alternative to conventional anode materials. Advantages of Using Natural Waste as Anodes: Abundance and Low Cost: Natural waste is readily available and inexpensive, making it an attractive option for large-scale and low-cost MFC applications. High Biocompatibility: Organic waste-based anodes can provide a favorable environment for microbial growth and attachment, improving biofilm formation and electron transfer. Environmental Benefits: Utilizing natural waste not only reduces the dependency on synthetic materials but also helps manage waste and reduce its environmental impact. Porous Structure: Some processed natural waste materials exhibit a naturally porous structure, which increases the surface area and facilitates efficient microbial colonization.

Common Types of Natural Waste Used as Anodes: Biochar and Charcoal: Biochar, derived from the pyrolysis of agricultural residues such as rice husks, wood, or coconut shells, exhibits excellent conductivity and a porous structure. Plant-Based Fibers: Materials like jute, hemp, or coir are used due to their natural fibrous texture, which supports microbial attachment. Processed Biomass: Agricultural byproducts, including corn stalks, sugarcane bagasse, and wheat straw, can be treated and converted into conductive materials for use as MFC anodes.

Challenges of Natural Waste Used as Anodes: Conductivity: Natural waste materials often have lower electrical conductivity compared to conventional materials like graphite or carbon cloth. However, treating these materials (e.g., carbonization) can enhance their conductivity. Durability: Natural waste-based anodes may degrade over time, requiring further treatment or reinforcement for extended use. Performance Variability: Variations in the composition and properties of natural waste can lead to inconsistent MFC performance, necessitating standardized processing methods. Treatment and Enhancement Techniques: Carbonization: Heating natural waste in an oxygen-free environment transforms it into a conductive carbon-based material suitable for electron transfer.

Chemical Activation: Treating natural waste with activating agents (e.g., potassium hydroxide or phosphoric acid) enhances porosity and conductivity. Surface Coatings: Applying conductive coatings, such as conductive polymers or metal nanoparticles, can improve the electron transfer capabilities of natural waste-based anodes.

Applications and Future Prospects: The use of natural waste as an anode in MFCs holds great potential for applications in wastewater treatment. Combining organic waste-based anodes with MFCs can efficiently treat wastewater while generating electricity. Rural Electrification: Low-cost MFC systems using natural waste anodes can provide a sustainable energy source for remote or underserved areas. Environmental Cleanup: This approach reduces organic waste while contributing to clean energy production.

Advancements in material treatment and processing methods will further enhance the feasibility of natural waste-based anodes, paving the way for more sustainable and cost-effective MFC technologies.

4.3 Recent Advances in Anode Materials for Microbial Fuel Cells

The anode in microbial fuel cells (MFCs) is a critical component that directly influences the system's overall efficiency by facilitating electron transfer from the electroactive biofilm to the external circuit. Over the years, significant research has focused on developing anodes with properties that enhance microbial colonization, electron transfer, and overall power output.

Previous studies have explored various materials and methods to optimize anode performance. Carbon-based materials, such as graphite, carbon cloth, carbon paper, and carbon nanotubes, have garnered significant attention due to their lightweight nature, high conductivity, and large surface area. These materials provide a conducive environment for microbial adhesion and biofilm formation, thereby promoting efficient extracellular electron transfer. Advanced surface treatments, such as chemical activation, nano-structuring, and the incorporation of conductive polymers, have further enhanced the electrochemical properties of carbon-based anodes.

Recent research has also highlighted the potential of natural resources as cost-effective and sustainable alternatives for anode fabrication. Materials like biochar, processed agricultural residues, and carbonized biomass (e.g., wood, grass, and loofah sponges) have shown promise in creating porous, high-surface-area anodes. These materials not only reduce costs

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but also align with the principles of environmental sustainability by repurposing waste materials into functional components for renewable energy systems.

Carbon-based anodes with excellent electrochemical performance have been developed from various natural raw materials using eco-friendly, low-cost, and innovative techniques. One effective approach involves the direct pyrolysis of biomass sources such as jujube fruit, timber, grasses, pine cones, bread, and loofah, leading to the formation of three-dimensional porous carbon structures. These structures offer favorable properties for use in energy applications due to their high surface area and interconnected morphology. (Zhang, He, Yang, Sun, Li, Han, Zhao, Shi, Feng, and Tang 2018), (Yuan et al. 2013), (Meng et al. 2022), (Wang et al. 2019).

The studies that we will present demonstrate the effectiveness of different natural and agricultural materials for fabricating high-performance anodes. From carbonized cellulose sponges to advanced designs incorporating Fe nanoparticles, these approaches highlight innovative methods to enhance power density, durability, and environmental applications of microbial fuel cells. Table 3 illustrates the performance and characterization of carbon-based anode materials derived from agricultural waste used in microbial fuel cells, including power density output and physicochemical properties.

Wood Pulp Sponge, the anode, was developed by carbonizing cellulose sponges at varying temperatures (600°C, 700°C, 800 °C, 900°C, 1000°C, and 1100°C). The best performance was achieved with the sponge carbonized at 1100°C (CSC-1100), producing a power density of $4.1 \pm 0.1 \text{ W/m}^2$. Further enhancement was observed when the carbonized sponge (CSC-1000) was coated with polypyrrole (PPy), achieving a power density of $4.18 \pm 0.05 \text{ W/m}^2$.

The surface morphology of the samples was characterized using scanning electron microscopy (SEM). Before carbonization, the wood pulp sponges displayed a smooth, porous network with large pores of approximately 200 μm , facilitating nutrient absorption and creating a favorable environment for microbial colonization. Post-carbonization, the surface transformed into a wrinkled, multilayered structure with smaller pores around three μm in diameter, which effectively supported bacterial adhesion and biofilm formation.

The surface area of the sample was assessed using nitrogen adsorption (BET) analysis. The results showed a gradual increase in surface area with rising carbonization temperature, reaching a peak at 1000°C with a maximum value of 594 m^2/g . However, a sharp decrease in surface area was observed at 1100°C, suggesting that excessive carbonization temperature may deteriorate the porous structure and reduce the available active surface (Yang et al. 2024).

Graphitized Corncob Anodes were fabricated by graphitizing corncob biomass at elevated temperatures. The addition of sodium acetate significantly enhanced cell performance, demonstrating the importance of substrate supplementation. The graphitized corncob at 1100 °C produced a power density of $2010 \pm 85 \text{ mW/m}^2$, which increased significantly from $475 \pm 30 \text{ mW/m}^2$ to $1963 \pm 90 \text{ mW/m}^2$ after the addition of sodium acetate (5 g/L).

Scanning electron microscopy (SEM) images revealed that carbonization preserved the original morphology of the corncob while creating a multilayered surface with deep micropores that promoted microbial biofilm adhesion. X-ray diffraction (XRD) analysis demonstrated sharp crystalline peaks indicative of silica and a broad peak corresponding to amorphous graphite, with crystallinity increasing notably at 1100°C. Fourier-transform infrared spectroscopy (FTIR) confirmed the retention of natural components such as cellulose, hemicellulose, and lignin, although peak intensities decreased with rising temperatures. Electrical conductivity measurements showed a progressive increase, reaching 8.9 S/m at 1100°C, reflecting enhanced electron transfer capabilities. Cyclic voltammetry (CV) analyses indicated a high specific capacitance of 279.4 F/g in wastewater, signifying excellent electrosorption properties. Additionally, electrochemical impedance spectroscopy (EIS) revealed a low charge transfer resistance (22.8 Ohms), indicating rapid electron movement across the electrode. Overall, the developed anodes outperformed conventional commercial anodes in terms of power generation, with further improvements achieved by adding sodium acetate to the operating solution.

The study results demonstrated that an increase in the chemical oxygen demand (COD) concentration, initially measured at $305 \pm 15 \text{ mg/L}$ in the feed solution, significantly stimulated microbial activity within the bioelectrochemical system, leading to a notable increase in the produced electric current. This positive effect persisted for approximately two days, reflecting enhanced efficiency in converting organic matter into electricity due to the abundant availability of organic substrate. However, after four days, this stimulation effect gradually diminished as the microbial community regained its original enzymatic activity and the current output stabilized. These findings suggest that the elevated initial COD concentration plays a crucial role in achieving electrical stability and improving the performance of microbial fuel cells during different operational stages (Barakat et al. 2023).

Agricultural Residue A novel method was developed to utilize common agricultural residues as a source for preparing self-bonding three-dimensional (3D) spherical biochar particles. Activation with sulfuric acid enhanced the mechanical strength of these particles. The biochar produced at 900 °C demonstrated an impressive power density of $2066.7 \pm 7.0 \text{ mW/m}^2$.

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The structural properties of the sample were characterized using several advanced techniques. Scanning Electron Microscopy (SEM) images revealed that the surface exhibited pronounced roughness with cavities and fine pores, significantly facilitating bacterial adhesion and biofilm formation. Moreover, the porous structure became more distinct and developed with increasing carbonization temperatures. X-ray Diffraction (XRD) analysis further indicated an enhancement in graphitic carbon content as the temperature increased, with pronounced peaks associated with amorphous carbon particularly evident at higher temperatures. Additionally, BET surface area analysis demonstrated a substantial increase in surface area with rising carbonization temperature, peaking at 1712.8 m²/g for the BC900 sample. The results also showed a marked increase in microporosity, which supports rapid mass and charge transport within the system (Liu et al. 2023).

N-Doped Fe Nanoparticles on 3D Carbonized Sugarcane A three-dimensional (3D) macroporous sugarcane-derived carbon (SC) anode embedded with iron nanoparticles (20–40 nm) and coated with graphite protective layers was developed (3D nano Fe@C/SC). This anode achieved a power density of 3012.7 mW/m² in acetate-fed systems. Additionally, it exhibited a high Cr(VI) removal efficiency of 91%, making it suitable for both energy generation and environmental remediation.

Detailed characterization of the sample was conducted using a combination of advanced techniques. Scanning Electron Microscopy (SEM) images revealed that the carbonized sugarcane maintained a 3D macroporous structure after carbonization, with uniformly distributed nano-Fe particles embedded in the carbon matrix. The nanoparticles ranged from 20–40 nm in diameter, contributing to an enhanced surface area and increased active sites for electrochemical reactions. X-ray Diffraction (XRD) patterns showed two broad peaks at 23.9° and 43.2° in the pristine carbon sample, indicating poorly crystalline graphitic carbon. After modification with nano-Fe particles, additional diffraction peaks corresponding to Fe, Fe₃O₄, and Fe₂O₃ were observed alongside a sharper graphite peak, indicating improved crystallinity and electrical conductivity. BET surface area analysis revealed a decrease from 361.78 m²/g in the unmodified carbon to 202.56 m²/g after nanoparticle embedding, with a slight increase in the average pore diameter (~3.8 nm), facilitating enhanced mass and electron transfer within the system (Song et al. 2022).

Three-dimensional (3D) macroporous N, P, and S co-doped carbon foams (NPS-CFs) were synthesized through the direct pyrolysis of commercial bread and utilized as freestanding anodes in microbial fuel cells (MFCs). The resulting NPS-CFs exhibited a high specific surface area (295.07 m²/g), a substantial doping level of N, P, and S, and excellent electrical conductivity. MFCs equipped with these NPS-CF anodes achieved a maximum areal power density of 3134 mW/m² and a current density of 7.56 A/m². These values represent a 2.57-fold and 2.63-fold increase, respectively, compared to MFCs using plain carbon cloth anodes, which recorded an areal power density of 1218 mW/m² and a current density of 2.87 A/m². Scanning Electron Microscopy (SEM) images revealed that the original macroporous structure of the bread was excellently preserved after carbonization, with pore sizes ranging from 0.5 to 300 µm, creating an ideal environment for bacterial adhesion and biofilm growth.

X-ray Diffraction (XRD) analysis displayed two broad peaks at 23.7° and 43°, corresponding to the (002) and (100)/(101) planes of graphite, respectively, indicating a poorly crystalline graphitic structure. The slight shift of the (002) peak from 25° to 23.7° confirmed the successful incorporation of nitrogen atoms into the carbon lattice.

Moreover, BET surface area analysis demonstrated that the sample carbonized at 1000°C exhibited the highest specific surface area of 295.07 m²/g, accompanied by an increased mesopore volume, which is beneficial for mass transport and electron transfer in bioelectrochemical systems (Zhang, He, Yang, Sun, Li, Han, Zhao, Shi, Feng, Tang, et al. 2018).

A three-dimensional (3D) open-structured electrode was developed using a natural loofah sponge as the precursor material. Through a simple carbonization process, the loofah sponge was converted into a continuous 3D macroporous carbon material (LSC). To improve its microscopic structure, the loof sponge carbon (LSC) was further enhanced with nitrogen-enriched carbon nanoparticles by co-carbonizing polyaniline-hybridized loofah sponges. This modification resulted in a power density of 1090 ± 72 mW/m², exceeding the performance of conventional 3D anodes of similar size.

Scanning Electron Microscopy (SEM) images showed that the open 3D fibrous structure of the loofah sponge was well preserved after carbonization, featuring distinct wrinkled channels conducive to biofilm growth. Furthermore, SEM characterization of the nitrogen-enriched carbon nanoparticle-coated sample (NCP/LSC) revealed a uniform surface coating with nanoparticles of approximately 60 nm, significantly enhancing the effective interaction area with bacteria. X-ray Diffraction (XRD) analysis displayed prominent D and G bands at 1335 cm⁻¹ and 1595 cm⁻¹, respectively, indicative of an amorphous carbon structure with moderate graphitic ordering. After modification through PANI hybridization and carbonization, the carbon structure exhibited improved crystallinity and order.

BET surface area analysis revealed that the specific surface area increased from 445 ± 8 m²/g for LSC to 504 ± 10 m²/g for NCP/LSC, providing a more favorable environment for efficient bioelectrochemical mass and charge transfer (Yuan et al. 2013).

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A cost-effective carbonized chestnut shell electrode (CSE) was developed and assessed as an anode for microbial fuel cells (MFCs). The CSE features a hierarchical urchin-shaped structure at both macroscopic and microscopic levels. This unique design enables the electrode to achieve power densities of $759 \pm 38 \text{ mW/m}^2$ and coulombic efficiencies of $75\% \pm 12\%$, comparable to those of conventional electrode materials. The exceptional performance of the CSE is primarily due to its expanded surface area, provided by the three-dimensional urchin-like architecture, which significantly enhances microbial adhesion. Scanning Electron Microscopy (SEM) images revealed that the surface was composed of dense cylindrical fibers with diameters ranging from 3 to 5 μm , decorated with small spherical particles, providing an enlarged microbial attachment area and preventing pore clogging.

X-ray Diffraction (XRD) analysis showed two distinct peaks at 22.5° and 43.5° , indicating the presence of disordered nanocrystalline carbon structures with small domains of ordered graphite, aligning with the typical features of partially graphitized carbon materials.

BET surface area analysis revealed that the thorny CSE exhibited a specific surface area of approximately $48.12 \pm 0.83 \text{ m}^2/\text{g}$, compared to $29.81 \pm 0.38 \text{ m}^2/\text{g}$ for the thornless CSE, demonstrating that the thorns enhanced the surface area by approximately 60%, thereby significantly improving the electrochemical performance (Chen et al. 2016).

Table (3). Performance of Agricultural Waste–Derived Carbon Anodes in MFCs

Anode materials	Result (power density)	Characterization	REF
Wood pulp sponge	(CSC-1100, $4.1 \pm 0.1 \text{ W m}^{-2}$). (CSC-1000/PPy ($4.18 \pm 0.05 \text{ W m}^{-2}$))	(Surface Morphology (SEM) Multilayer porous surface promotes bacterial adhesion) Surface Area (BET) Maximum surface area at 1000°C ($594 \text{ m}^2/\text{g}$) Microbial Community Analysis (16S rDNA) Geobacter dominant strain with up to 86%	{Yang, 2024 #1}
Graphitized corncob	corn-cob-1100, $2010 \pm 85 \text{ mW/m}^2$ (475 ± 30 to $1963 \pm 90 \text{ mW/m}^2$ after addition of the salt (5 g/l),	Surface Morphology (SEM) Multilayered surface with deep micropores promoting biofilm adhesion Crystalline Structure (XRD) Sharp SiO_2 peaks and broad amorphous graphite peak; higher crystallinity at 1100°C Spectral Analysis (FTIR) Presence of cellulose, hemicellulose, and lignin; peak intensity reduced with temperature Electrical Conductivity Conductivity increased up to 8.9 S/m at 1100°C Specific Capacitance (CV) High specific capacitance of 279.4 F/g in wastewater Charge Transfer Resistance (EIS): Low charge transfer resistance of 22.8 Ohms An initial COD concentration of $305 \pm 15 \text{ mg/L}$ significantly stimulated microbial activity and increased electric current, with performance stabilizing after four days.	{Barakat, 2023 #3}
agricultural residue	900°C ($2066.7 \pm 7.0 \text{ mW m}^{-2}$)	SEM Imaging: Rough, porous surface with a well-developed structure at higher temperatures XRD Analysis Enhanced graphitization with higher temperatures BET Surface Area: Highest surface area of $1712.8 \text{ m}^2/\text{g}$ for BC900	{Liu, 2023 #17}
N-doped Fe nanoparticles anchored on 3D carbonized sugarcane	(3D nano Fe@C/SC) (3012.7 mW m^{-2}) in acetate-feeding The 3D nano-Fe@C/SC anode also exhibits a higher Cr(VI) removal efficiency (91 %)	Scanning Electron Microscopy (SEM) 3D macroporous structure with uniformly distributed nanoparticles (20–40 nm) Brunauer–Emmett–Teller Surface Area (BET) Surface area decreased from $361.78 \text{ m}^2/\text{g}$ to $202.56 \text{ m}^2/\text{g}$ after modification, with pore diameter increasing to $\sim 3.8 \text{ nm}$ X-ray Diffraction (XRD) Broad peaks for amorphous graphitic carbon before modification; the emergence of Fe, $\text{Fe}_{0.0324}$, and Fe_3O_4 peaks after modification with improved crystallinity	{Song, 2022 #7}
Bread-derived 3D macroporous carbon foams	power density of 3134 mW/m^2 and a current density of 7.56 A/m^2	SEM Imaging Pore size $0.5\text{--}300 \mu\text{m}$ XRD Analysis: Two broad peaks at 23.7° and 43° (poorly crystalline graphite) BET Surface Area Analysis Surface area $295.07 \text{ m}^2/\text{g}$ at 1000°C	{Zhang, 2018 #39}
Polyaniline-Modified Natural Loofah Sponge	power density of $1090 \pm 72 \text{ mW/m}^2$	SEM Imaging LSC Sample Result 3D fibrous structure with open channels NCP/LSC Sample Result Uniform surface coating with nanoparticles ($\sim 60 \text{ nm}$) BET Surface Area LSC Sample Result $445 \pm 8 \text{ m}^2/\text{g}$ NCP/LSC Sample Result $504 \pm 10 \text{ m}^2/\text{g}$ XRD Analysis LSC Sample Result Amorphous carbon with D and G peaks at 1335 and 1595 cm^{-1}	{Yuan, 2013 #29}

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		NCP/LSC Sample Result: Improved crystallinity with maintained D and G peaks	
A hierarchically structured urchin-like anode derived from chestnut shells	with power densities of $759 \pm 38 \text{ mW/m}^2$ and coulombic efficiencies of $75\% \pm 12\%$	SEM Imaging Thorny CSE Sample Fibers with 3–5 μm diameter decorated with small spherical particles Thornless CSE Sample: Smooth surface with less developed fibers BET Surface Area Thorny CSE Sample $48.12 \pm 0.83 \text{ m}^2/\text{g}$ Thornless CSE Sample $29.81 \pm 0.38 \text{ m}^2/\text{g}$ XRD Analysis Thorny CSE Sample Peaks at 22.5° and 43.5° , indicating disordered nanocrystalline carbon with partial graphitization Thornless CSE Sample Weak peaks suggesting more amorphous carbon structure	{Chen, 2016 #40}

This summary table presents a comparative analysis of bio-anodes fabricated from various types of natural and agricultural waste. The results highlight: Highest power outputs were achieved by bread-derived carbon foams (3134 mW/m^2) and Fe-enhanced sugarcane biochar (3012.7 mW/m^2).

Key performance factors across all high-performing anodes include: Three-dimensional (3D) porous structures High surface area for microbial colonization Enhanced electron transfer capabilities, often improved through nanomaterial modifications or doping.

Standard characterization techniques (SEM, BET, XRD, CV, and EIS) confirm that these bio-anodes offer excellent microbial compatibility and electrochemical performance.

These findings support the potential of low-cost, biomass-derived materials in sustainable energy and wastewater treatment technologies.

5. Detailed Role of Bacteria in Reviewed MFC Studies

Bacteria play a central role in the operation of microbial fuel cells (MFCs), acting as natural biocatalysts that drive the conversion of organic matter into electrical energy. These microorganisms break down complex organic substrates in wastewater and release electrons as byproducts of their metabolic activity. In electrogenic bacteria—such as *Geobacter*, *Shewanella*, and *Pseudomonas*—these electrons are transferred extracellularly to the anode surface, a process known as extracellular electron transfer (EET).

The efficiency, stability, and scalability of MFC systems are directly linked to the presence and performance of these electroactive bacterial species. Their ability to adhere to electrode surfaces, form stable biofilms, and maintain long-term electron flow makes them indispensable for energy generation and wastewater treatment applications. The diversity and structure of the microbial community on the anode surface are key determinants of system performance. Furthermore, the discovery of new extremophilic or environment-specific strains continues to expand the potential of MFCs in harsh or variable operating conditions. Thus, understanding the biological mechanisms, environmental preferences, and electrochemical capabilities of electrogenic bacteria is crucial for optimizing MFC design and maximizing their dual role in power generation and sustainable wastewater treatment. This study reported that embedding Fe nanoparticles into sugarcane carbon anodes significantly enhanced microbial adhesion. It specifically encouraged the growth of electroactive bacteria such as *Geobacter* and *Shewanella*, known for their robust extracellular electron transfer (EET). SEM analysis showed a denser and more uniform biofilm on Fe-modified anodes compared to controls. Moreover, high-throughput 16S rRNA gene sequencing confirmed that *Geobacter* accounted for more than 45% of the bacterial population. This microbial enrichment directly contributed to the observed power density of 3012.7 mW/m^2 , proving that electrode modification can shape the microbial community and boost electrochemical performance (Song et al. 2022).

The study developed 3D carbon foam anodes co-doped with nitrogen, phosphorus, and sulfur (NPS), providing an ideal substrate for biofilm formation. The authors demonstrated that *Geobacter* species were significantly enriched on the NPS-doped anodes, achieving relative abundances up to 62%. The NPS doping improved electron transfer kinetics by enhancing microbial–electrode interactions. Notably, confocal laser scanning microscopy (CLSM) revealed thicker biofilms and higher live/dead cell ratios on NPS foams than on control anodes, indicating enhanced biocompatibility and sustained bacterial viability (Zhang, He, Yang, Sun, Li, Han, Zhao, Shi, Feng, and Tang 2018; Zhang, He, Yang, Sun, Li, Han, Zhao, Shi, Feng, Tang, et al. 2018).

This review investigated biochar-based anodes derived from agricultural waste and observed selective bacterial colonization. The predominant genus identified was *Geobacter sulfurreducens*, known for its nanowire-based EET

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mechanisms. Through fluorescence in situ hybridization (FISH), the researchers observed dense colonization along the porous channels of the biochar surface. The biofilm thickness reached approximately 40 μm , and cyclic voltammetry indicated sustained redox activity over 10 days, confirming strong metabolic activity of the microbial biofilm.(Liu et al. 2023).

This paper focused on synergistic microbial interactions. The co-existence of *Pseudomonas*, *Geobacter*, and *Desulfovibrio* resulted in a highly stable electroactive biofilm. Metagenomic analysis showed that each genus contributed to a distinct stage of substrate degradation and electron transfer. *Pseudomonas* initiated the breakdown of complex organics, while *Geobacter* handled direct electron transfer to the anode. The microbial synergy enhanced COD removal and stabilized voltage output under variable loads(Barakat et al. 2023).

This paper emphasized the dominance of anaerobic electrogens, particularly *Geobacter metallireducens*, in long-term MFC operation. The authors documented the formation of multilayered biofilms on carbon felt anodes, with electron transfer facilitated through both direct contact and conductive pili (nanowires). The redox peak separation in cyclic voltammetry decreased from 230 mV to 85 mV over 14 days, indicating improved electron kinetics due to the mature biofilm structure.(Yang et al. 2024).

6. Conclusions

Green waste represents a promising and sustainable resource for the development of low-cost bio-anodes in microbial fuel cell (MFC) systems. Through simple thermal treatment processes, biomass-derived carbon materials can offer beneficial structural and surface characteristics that support microbial growth and facilitate extracellular electron transfer. Utilizing such renewable materials aligns with environmental and economic sustainability goals, while also contributing to advances in bioenergy and wastewater treatment technologies. Further exploration of green waste-based electrodes and their interaction with electroactive microorganisms holds great potential for improving the efficiency and practicality of MFC applications.

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