

Conversion of lignocellulose residue obtained from biorefinery stream to electricity by microbial fuel cell

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Abstract. In general, lignocellulose biorefinery has the main functions to fractionate biomass compositions and convert them to value-added products. However, leftover organic compounds in output streams are mixed with large amounts of wastewater becoming the cost and burden for treatment. Therefore, to close the loop of circular economy, this review paper explores the potential of microbial fuel cells (MFCs) as a sustainable and efficient way to convert lignocellulose residue, a byproduct of biorefinery processes, into electricity. Lignocellulose residue is a complex mixture of carbohydrates and lignin that is often difficult to dispose of properly. By using MFCs, this waste material can be converted into valuable energy while reducing the environmental impact of its disposal. The paper covers the different types of MFCs, their working principles, and their potential application in lignocellulose residue conversion. It also discusses the factors that affect the performance of MFCs, including substrate availability, electrode material, and reactor design. Additionally, the paper reviews the current state of research in this area, highlighting recent advances and identifying areas for future exploration. Overall, this review paper demonstrates the promise of MFCs as a sustainable and innovative approach to converting lignocellulose residue into electricity.

Keyword. Circular economy, Lignocellulose biomass, Microbial fuel cell, Wastewater

1 Introduction

Lignocellulose is the fundamental structural component of plant biomass and is comprised of cellulose, hemicellulose and lignin. It is the most abundant organic material on Earth and it is estimated to be produced globally more than 1.3 billion tons per year. Engaged with agricultural activities, improper management of lignocellulose residues, which are discarded with no use after harvesting season, can lead to the emission of greenhouse gases, the depletion of soil quality, and the contamination of water resources [1]. Therefore, the circular economy model is proposed to be a solution by converting lignocellulose residues to be the raw materials for productions of value-added products to motivate the social and industrial movement to this campaign. Biorefining process adapts to the circular economy model by converting lignocellulose biomass to various types of products such as biofuels, bioenergy, platform chemicals, biochemical and biopolymers [2].

On one hand, lignocellulose biorefinery is a solution to reduce the release of biomass waste to environment in manageable facility or process, on another hand, this process generates large amounts of wastewaters containing organic and inorganic residues. For example,

after pretreatment of mixed vegetable waste with organic acids, such as citric acid, oxalic acid, and acetic acid, about 5-6 liters of water needed to be used for washing organic acids from 5 g biomass to remove inhibitors for enzymatic saccharification and microbial fermentation [3]. It is estimated that a typical biorefinery process requires input water about 4–10 times more than the amount of generated biofuel [4]. Therefore, several attempts have been done to close the loop of circular economy model of lignocellulose biorefinery by integrate various technologies or units, such as anaerobic digester, photocatalytic, and microbial fuel cell.

The use of microbial fuel cells (MFCs) is a promising technology for converting lignocellulose residue into electricity, thereby providing a sustainable and efficient way to utilize this waste material. By harnessing the natural abilities of microorganisms to oxidize organic matter and produce electrical energy, MFCs offer a novel approach to the management of lignocellulose residue [5]. The use of MFCs is not limited to the conversion of lignocellulose residue, they have also been applied in other areas, such as wastewater treatment, bioremediation, and biosensors. Furthermore, their potential applications extend beyond the production of electricity, with some researchers exploring the use of

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MFCs for the production of chemicals and fuels [6]. Due to these wide ranges of applications, the numbers of publications related to MFCs indexed in SCOPUS are doubling-increased from 701 papers in 2013 to 1417 papers in 2022 (Figure 1). In the same period of time, the trend of biorefinery research with application of MFCs has been growing significantly from 10 to 198 papers suggesting the potential of MFC technology. This review paper aims to explore the current state of research on the use of MFCs for lignocellulose residue conversion, covering the different types of MFCs, their working principles, and their potential application in this area. The paper also discusses the factors that affect the performance of MFCs, such as substrate availability, electrode material, and reactor design. Finally, the paper highlights recent advances in this field and identifies areas for future exploration.

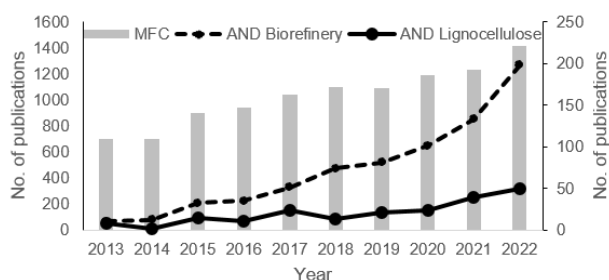


Fig. 1. Numbers of publications indexed in SCOPUS database during 2013 – 2022 related to MFCs, biorefinery and lignocellulose.

2 Lignocellulosic biorefining process and its output streams

Lignocellulose is a complex material that is resistant to degradation, and its disposal can lead to environmental and economic issues. Additionally, the transportation and disposal of lignocellulose residue can be costly and can consume significant amounts of energy due to its bulky shape and low-density characteristic. The biorefinery process of lignocellulose biomass typically involves four main steps, including pretreatment, hydrolysis, fermentation and product separation (Figure 2) [7]. The bottleneck of the process to convert lignocellulose biomass to value-added products, such as biofuels and biochemical, is the hydrolysis due to the recalcitrant structure of biomass that functions as the

protector of plants in nature from environmental stresses and pathogen attacks [8].

Pretreatment involves breaking down the lignocellulosic material into smaller components to increase their accessibility for downstream processes, especially enzymatic hydrolysis by cellulase enzyme. Currently, there are many pretreatment methods that are categorized as chemical, biological, physical and combined methods. The most common pretreatment methods include steam explosion, acid hydrolysis, and alkaline treatment [9]. It is estimated that the pretreatment cost could be 40-60% of the whole process. Hydrolysis uses enzymes or acid catalysts to break down the cellulose and hemicellulose into simple sugars, such as glucose and xylose, which can be used as feedstocks for productions of various chemical and biological processes (e.g. fermentation). The final step in the biorefinery process involves separating and purifying the desired products from the fermentation broth using various physical and chemical processes [10].

In production process of cellulosic ethanol or second generation ethanol (Figure 2), the combination of fractionation and reaction in each step produce various output streams containing different products, which are influenced by the conditions of each biorefining step. For example, strong acid pretreatment, such as hydrochloric or sulfuric acid, leads to the degradation of glucose and dehydration of xylose to 5-Hydroxymethylfurfural (HMF) and furfural, respectively. The harsh condition of pretreatment can further promote productions of various degraded chemicals from hemicellulose, such as organic acids (e.g. acetic acid, formic acid, levulinic acid, furoic acid), aldehydes, uronic acids. The lignin could be also degraded to various phenolic compounds, for example, coniferyl aldehyde, ferulic acid, benzoic acid [11]. Although many of these by-products are conceptually proved to be candidates for productions of platform chemicals or potential intermediates compounds for downstream industries [12]. Nevertheless, considered to maximize the cellulosic ethanol yield, the more by-product formation, the less ethanol yield. Based on this scheme of biorefining process, these pretreatment by-products are mostly fractionated to be the liquid fraction, or mostly called “pretreatment liquor”. Additionally, pretreated biomass in solid fraction is usually needed to be washed to remove the chemical residues used in pretreatment. In a previous study, it is demonstrated that

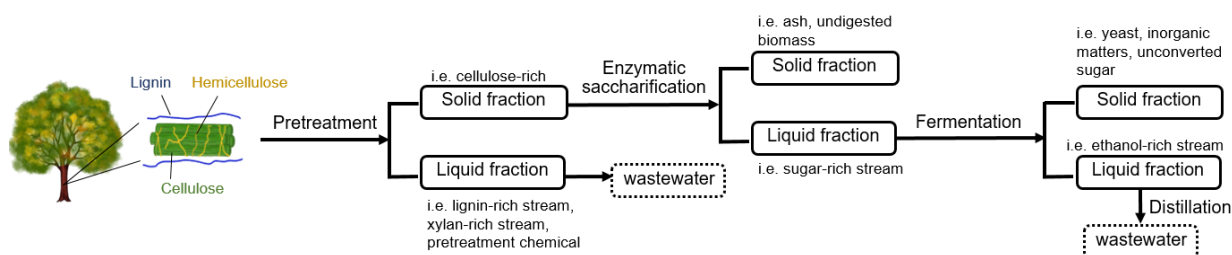


Fig. 2. Process diagram of lignocellulose biorefinery and ethanol production

the remaining pretreatment chemicals, such as ionic liquid, reduce the cellulase activities up to more than 60% when about 10-20% concentration of ionic liquid remains [13-14]. Therefore, a large volume of wastewater is generated during biomass solid washing (Figure 2). In the next step, pretreated biomass is hydrolyzed by cellulase enzyme, and the liquid fraction stream of hydrolysate is enriched with sugar monomers, especially hexose and pentose, which are further fermented by *Saccharomyces cerevisiae* to produce ethanol. The ethanol stream as an output from fermenter is proceeded to distillation column and spent liquid of distillates is enriched with organic compounds. Collectively, the output liquid streams of cellulosic ethanol contain various chemical by-products that are the sources of Chemical oxygen demand (COD) and Biochemical Oxygen Demand (BOD) values when they are released to environment.

3 MFC: components and design

The use of microbial fuel cells (MFCs) has been growing rapidly in recent years due to their potential as a sustainable and efficient technology for the production of electricity from organic waste materials. Comparing to the traditional treatment method for wastewater face several challenges that limit their effectiveness. One of the main challenges is the high energy consumption required for the treatment process. Traditional methods rely on aeration and mixing to promote microbial growth and degradation of pollutants, which requires significant amounts of electricity and power. In addition, traditional treatment methods are limited in their ability to remove certain types of pollutants such as microplastics, pharmaceuticals, and other emerging contaminants [15]. Another challenge of traditional treatment is the production of large volumes of sludge, which can pose

challenges for disposal and management. The sludge contains high levels of nutrients and organic matter, which can contribute to eutrophication and other environmental problems if not properly managed. In addition, traditional methods are susceptible to operational issues, such as clogging, odors, and corrosion, which can lead to decreased efficiency and increased maintenance costs [16].

MFCs use microorganisms to catalyze the oxidation of organic matter and generate electrical energy, making them an attractive option for the treatment of organic waste and the generation of renewable energy. In an MFC, microorganisms are attached to an electrode and consume organic matter in the wastewater, producing electrons that flow through an external circuit to produce electrical power. The potential of MFCs for wastewater treatment lies in their ability to not only treat the wastewater but also generate electricity as a byproduct. MFCs have demonstrated high removal efficiencies for pollutants such as organic matter, nutrients, and heavy metals, and have been shown to remove emerging contaminants, such as pharmaceuticals and microplastics [17]. One of the main advantages of MFCs over traditional treatment methods is their lower energy consumption. Unlike traditional treatment methods that require significant amounts of electricity for aeration and mixing, MFCs rely on microbial metabolism to drive the treatment process. This reduces the energy demand of the treatment process and makes MFCs more sustainable and cost-effective. Additionally, MFCs produce lower volumes of sludge compared to traditional methods, which can reduce the costs and environmental impacts associated with sludge disposal [18].

The MFC system consists of two compartments, an anode and a cathode, separated by a proton exchange membrane (Figure 3). Microorganisms, especially

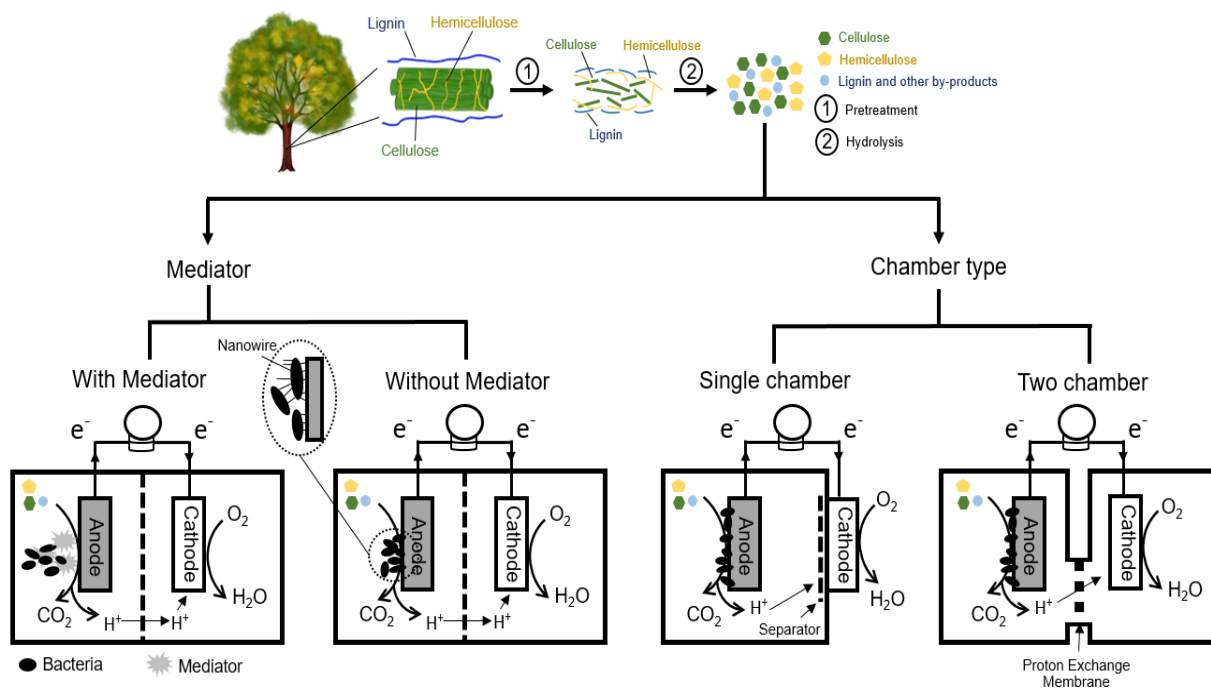


Fig. 3. Various MFC design for application in lignocellulose biorefinery

bacteria, are attached to the anode, where they oxidize organic matter and produce electrons and protons. The electrons flow from the anode through an external circuit to the cathode, where they combine with protons and oxygen to form water [17]. The conversion of organic matter into electrical energy in an MFC involves two main processes: microbial metabolism and electron transfer. Microbial metabolism is the process by which microorganisms break down organic matter into simpler compounds, releasing energy in the form of electrons and protons. The electrons and protons produced by microbial metabolism are then transferred to the anode and the cathode, respectively, through electron transfer mechanisms. Electron transfer in MFCs occurs through two main pathways: direct transfer and mediated transfer (Figure 3). In a direct transfer, electrons are transferred from the microbial cells to the anode through conductive pili or other extracellular electron transfer structures, so-called nanowire. This process is often facilitated by excreted electron shuttles such as flavins, quinones, and humic substances. Mediated transfer, on the other hand, involves the use of a mediator that acts as an intermediate electron carrier between the microbial cells and the anode. The mediator accepts electrons from the microbial cells and then transfers them to the anode, however, these mediator chemicals have high cost and toxicity, such as thionine, methyl viologen, methyl blue, humic acid and neutral red [19].

Currently, MFCs can be configured in various ways to optimize their performance for specific applications. The most common configurations of MFCs are the single-chamber MFC (SC-MFC) and the two-chamber MFC (TC-MFC) (Figure 3). The SC-MFC is a simple and compact design that consists of a single chamber containing the anode and the cathode. The chamber is filled with wastewater that serves as the substrates for the microorganism metabolisms. The anode and cathode are usually separated by a proton exchange membrane (PEM) to prevent direct contact between the electrodes and to maintain the electrochemical potential difference between the anode and cathode. The main advantage of the SC-MFC is its simplicity and low cost. However, its performance is often limited by the low power output and relatively low treatment efficiency compared to the TC-MFC [20]. On the other hand, the TC-MFC consists of two separate chambers, one for the anode and one for the cathode. The chambers are connected by a salt bridge or a PEM that allows for the transfer of ions and maintains the electrochemical potential difference between the anode and cathode. The wastewater or nutrient solution is fed into the anode chamber, and air or oxygen is fed into the cathode chamber to provide the necessary electron acceptor for the reduction of protons and electrons. The TC-MFC has several advantages over the SC-MFC, including higher power output, improved treatment efficiency, and better control over the operating conditions. The separation of the anode and cathode also allows for the recovery of valuable byproducts, such as hydrogen gas and organic acids [21].

The choice of MFC configuration depends on the specific application and the desired performance properties, such as power output, treatment efficiency,

and cost-effectiveness. As MFC technology continues to advance, new configurations and designs are likely to emerge, further expanding the potential applications of this promising technology. The performance of MFCs is affected by several factors that need to be carefully optimized to achieve optimal treatment efficiency and power output. The main factors that affect MFC performance are its electrode. The anode material is an essential component of the MFC and can significantly affect its performance. The choice of anode material depends on its conductivity, surface area, and biocompatibility with the microbial community. Common anode materials include carbon-based materials, metal oxides, and conductive polymers. Additionally, the cathode material also affects the reduction of oxygen or other electron acceptors. Common cathode materials include graphite, carbon cloth, and metal catalysts such as platinum. In addition to the electrode, the microbial community in MFCs plays a critical role in the degradation of pollutants and the generation of electrical power. The selection of the appropriate microbial community depends on the wastewater composition and the desired treatment efficiency and power output [22]. Different microbial communities, such as bacteria, archaea, and fungi, have been used in MFCs for wastewater treatment.

The operating conditions of MFCs can significantly affect their performance, including temperature, pH, hydraulic retention time, and external resistance. The optimal operating conditions depend on the specific application and wastewater composition. The characteristics of the wastewater, such as its chemical composition, organic loading rate, and nutrient content, can affect MFC performance. The optimal design and operation of MFCs need to consider the specific wastewater characteristics to achieve optimal treatment efficiency and power output. The spacing between the anode and cathode electrodes can significantly affect MFC performance, with smaller electrode spacing generally leading to higher power output and treatment efficiency. Another important concern is related to substrate composition in wastewater. Inhibitors and toxic compounds, such as heavy metals and antibiotics, can affect the microbial community and reduce MFC performance [23]. Therefore, strategies to mitigate the effects of inhibitors and toxicants should be included, especially pretreatment of the wastewater or using a robust microbial community that can tolerate these substances.

4 Integration of MFC in biorefinery

Lignocellulose is abundant with the compositions of polymeric carbohydrate compounds, especially cellulose and hemicellulose. Following the process scheme of lignocellulose biorefinery (Figure 2), lignocellulose biomass is needed to be hydrolyzed by cellulase enzymes. However, based on the design of MFCs, a cellulose-degrading microbial strain or a consortium, such as *Clostridium cellulolyticum*, could be primary catalysts to convert polymeric lignocellulose substrates to oligomeric or monomeric sugars, which are

subsequently catabolized by other microbes to CO₂ or small organic acids. Several bacterial strains that possess extracellular electron transfer structures, or nanowire, such as *Geobacter sulfurreducens*, *Geobacter metallireducens*, *Enterobacter ludwigii*, *Enterobacter cloacae*, *Shewanella oneidensis* and *Shewanella marisflavi*, are co-culture to improve the electrotransfer capacity to anode and cathode, respectively [24].

The bacterial strains that can function as mediators in MFC could be co-cultured with other microbial consortiums, such as wastewater sludge from food processing industries, wastewater from municipal waste, soil, ruminal secrets. It is reported that when *Geobacter metallireducens* were co-cultured with ruminal contents and pasteurized soil in the FWA-Fe(III)-citrate medium to enrich cellulolytic bacteria. This bacterial culture was filled in anode chamber equipped with graphite anode air-type carbon cloth cathode [25]. The solid form of corncob biomass was used as the substrate for MFC, which continuously produced the current of 0.15 mA and 0.15V for 1300 h. After the addition of pasteurized soil and rumen fluid, the generated maximum currents were increased to 1.38 mA/m³ ad 770 mA/m³, respectively. When the bacterial culture in the anode chamber was augmented by *Geobacter metallireducens*, the power density was maximized to 1170 mA/m³, suggesting that the cellulase-producing bacteria and anode-respiring bacteria can coexist in the presence of solid form lignocellulose substrates [25].

Lignocellulose hydrolysate enriched with sugars could be also used as a substrate for MFC to produce electricity. As a case study, rapeseed straw was pretreated by hydrothermal process and enzymatic hydrolyzed, and its liquid hydrolysate fraction was input in a two-chamber MFC (H-type) for electricity generation [26]. The hydrolysate was fed in an anode chamber equipped with graphite felt anode and mixed with the post-fermentation sludge from the wastewater treatment facility and various formulas of hydrolysis media. The highest electricity was obtained at 58 mW/m² from the utilization of hydrolysate substrate at 150 mg/m³ and the highest COD removal rate was 0.602 g COD/m².d [26]. Another study using wheat straw hydrolysate as the substrate for yeast-type MFC was conducted in a two-chamber type by adding two strains of cellulase- and laccase-producing microbes, *Phlebia floridensis* and *Phlebia brevispora*, in cathode chamber and *Pichia fermentans* in anode chamber [27]. The reducing sugars and phenolic compounds derived from wheat straw were degraded and converted to electricity with a maximum power density of 33.19 mW/m². Combinations of banana peels and wetland sediments (as microbial inoculum) were tested as substrates for MFC using a membrane-less single chamber. When anaerobically fermented banana peels were added into the MFC, the highest current density was obtained at 91.3 mW/m² with the maximum voltage at 146 mV, which is equivalent to the substrate conversion of 13.5 j/g of banana peels. However, this scale of electricity production per gram of substrate was much less than MFC with membrane type [28].

In addition to be used as a substrate for MFC, lignocellulose biomass is also applied as the electrode for MFC reactors. Water hyacinth biochar was previously produced and applied as an air-type cathode in a single chamber MFC [29]. Water hyacinth biochar was prepared by pyrolysis and its electrochemical property was analyzed to function as a cathode. This biochar cathode was installed in a single chamber MFC that operated in anaerobic condition. The artificial wastewater medium was supplemented with trace element solution and inoculated by sewage sludge, and the maximum power density was obtained at 12.3 mW/m² [29]. Likewise, silver grass-derived activated carbon was prepared to be used as anode in a H-type two-chamber MFC [30]. Silver grass biomass was acid-pretreated and enzymatically hydrolyzed. The anode was prepared on carbon cloth support by adding biomass slurry with a PTFE binder. The anode chamber was inoculated with *Escherichia coli* in the fresh media. Using silver grass activated carbon anode, the maximum voltage reached 790 mV and power density at 963 mW/m², which are much higher than the electricity reported previously for biomass-derived activated carbon [30]. Altogether, these reports suggested the potential of lignocellulose biomass as the substrates and electrodes for MFC. The lists of lignocellulose-based MFC research are shown in Table 1.

Table 1. Integrations of MFCs in different LCB biomass.

Lignocellulose biomass	MFC design	Maximum voltage (mV)	Power generation (mW/m ²)	References
Corn stover powder	Single chamber	380	331	[31]
Corn cob powder	Tubular single chamber	672	7.18	[25]
Corn stover hydrolysate	Single chamber	~500	952	[32]
Rapeseed hydrolysate	Two chamber (H-type)	~470	58	[28]
Wheat straw hydrolysate	Two chamber	504	33.19	[27]
Rice straw compost	Single chamber with air cathode	277	112	[33]
Wheat straw hydrolysate and wastewater	Two chamber	240	123	[34]
Banana peel extracts and wetland sediments	Single chamber	146	91.3	[35]
Bamboo	Two chamber	760	578	[36]
Cashew apple juice	Two chamber	400	31.58	[37]

Although MFCs have been developed and well established in wastewater treatment as a green technology for electricity production. Aforementioned, various types of MFCs are designed and applied to lignocellulose biorefinery by using direct solid biomass, biomass hydrolysate or biomass by-products as substrates. Several limitations of MFCs were discussed, especially low power density and voltage instability. MFCs typically produce low power densities compared to other energy sources. This is due to the slow rate at which microorganisms oxidize organic matter, limiting the amount of electricity that can be generated. This slow metabolism rate of microorganisms could be the result of their sensitivity to changes in environmental conditions, such as temperature, pH, and salinity. Any changes in these conditions can affect the activity of microorganisms and reduce the efficiency of the MFC. Also, due to the activities of microorganisms, the lifespan of microbial cultures is limited due to the saturation of microbial growth. Its lifespan is also limited by the formation of biofilms on electrode and PEM (biofouling) that inhibit the electron transfer between electrodes [38-39]. Specifically, MFC application in lignocellulose biorefinery has the main bottleneck in the hydrolysis of lignocellulose biomass, therefore the bioaugmentation of lignocellulose-degrading microorganisms is necessary. However, most hydrolysis reactions of lignocellulose prefer higher temperatures, especially at 50 °C, whereas most MFCs were operated at room temperature. Therefore, process optimization is needed to be conducted to increase the efficiency of MFCs. Similarly, process integration with other technology, such as hydrogen production, could also increase the feasibility of MFC for further application. However, the prime benefit of lignocellulose-based MFC is the minimization of lignocellulose waste and wastewater produced from biorefinery to release to the environment. Lastly, lignocellulose-based MFC implements the circular economy, which leads toward a sustainable development goal

5 Conclusion

Lignocellulose biomass is an abundant resource for biorefining process to produce various value-added products, especially biofuels and biochemicals. The lignocellulose biorefinery is a multi-step process, which is composed of fractionation and catalytic conversion processes. Based on the process scheme of lignocellulose biorefinery, wastewater and liquid streams containing high organic compounds are produced in large volumes that require treatment before release to the environment. MFCs are a promising technology for the conversion of lignocellulose biomass into electricity as well as a practical method for wastewater treatment. Different MFC designs have been developed and applied to lignocellulose biorefinery, especially single-chamber type and two-chamber type. Based on the production scale of electricity generated from reported MFCs, it is recommended to be used for treatment of liquid streams of lignocellulose biorefinery, which could further

produce value-added by-products, especially organic acid, alcohol and hydrogen to complete the circular economy model.

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