A Review of Suitable Substrates for Hydrogen Production in Microbial Electrolysis Cells

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Abstract. Microbial electrolysis cells (MECs) represent a renewable hydrogen production technology that offers the possibility of converting wastewater to hydrogen through a bioelectrochemical process. Particularly, the MEC substrate has a significant effect on the performance of MECs, and in this review, the performances of over 30 substrates examined since 2015 are summarized and compared. It was evident that popular MEC substrates include dark fermentation effluents, pyrolysis products, and raw wastewaters. Additionally, the different MEC substrates investigated yielded different MEC performances, indicating that further studies are required before MECs can become a mature technology for up-scale applications.

1. Introduction
Microbial Electrolysis Cells (MECs) represent a booming technology through which hydrogen gas can be produced from organic matter such as wastewater. The advantage of hydrogen is that it is a highly efficient energy carrier associated with little or no pollution, making it an environmentally friendly and sustainable energy source [1]. Hydrogen has the highest potential for application in transportation and industrial production as a mainstream energy source, and its application in this regard can drastically reduce emissions. Additionally, unlike the hydrogen produced from fossil fuels, the hydrogen produced using MECs is renewable [2]. However, despite their great potential, MECs are still a developing technology with several limitations that need to be overcome before implementations of up-scale real-world applications.

MECs, simultaneously discovered by two laboratories, represent a bioelectrochemical system (BES) that allows the production of hydrogen from biomass [3, 4]. Typically, an MEC consists of an anode and a cathode chamber even though single-chamber MECs have been developed by eliminating the membrane between the two electrode chambers [5]. In MECs, electrochemically active bacteria (EAB) grows on the anode surface, consuming the substrate. The electrons and protons released by the EAB are then transferred to the cathode, where the protons are reduced and hydrogen is released. However, given that the formation of H₂ is spontaneous, a small applied voltage of approximately 0.2–0.8 V is typically required [6]. This voltage is still significantly lower than that required for water electrolysis, which is approximately 1.2 V [7]. Equations of the oxidation of acetate and the formation of hydrogen gas at the anode and cathode are shown subsequently [4], and a typical MEC is illustrated in Figure 1.

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\text{Anode: } C_2H_4O_2 + 2H_2O = 2CO_2 + 8e^- + 8H^+ \\
\text{Cathode: } 8H^+ + 8e^- = 4H_2
\]
Figure 1. A typical microbial electrolysis cell (MEC).

The production of hydrogen using MECs is considered a renewable energy production technology, given that organic wastes are used as substrates from which hydrogen is released [6]. However, the specific substrate used has a significant effect on the performance of a given MEC. Also, owing to the variety and complexity of substrates, the performance of an MEC can be unpredictable and inconsistent, thereby limiting its real-world application. Thus, a summary of the different substrate types that have been investigated since 2015 and a comparison of their performances can be conducive to guide researchers in the selection of substrates for MEC development. However, no comprehensive review has been conducted in this regard since the last review conducted by Kadier et al. in 2014 [8]. Therefore, the objective of this paper is to provide a complete review of the performances of MECs with respect to the different types of substrates that have been tested thus far for MEC hydrogen production.

2. MEC Substrates
MECs offer the possibility of producing hydrogen from a variety of substrates. Based on the results of previously reported studies, a summary of different substrate types is presented in this review, and the performances of the MECs with which they are associated are compared in terms of several performance metrics including applied voltage, hydrogen production rate (HPR), and energy efficiency, which is based on electrical input, recovery rates, and removal rates.

2.1. Fermentation Effluents
Volatile fatty acids (VFAs) present in dark fermentation effluents are organic pollutants that require treatments. In one study, the performances of different VFA loads were examined in two MEC structures [9]. The maximum HPR and organic removal rate were 81 mL/L/D and 85%, respectively. Additionally, another study in which the MEC performances of three VFAs—acetate, butyrate, and propionate—were investigated revealed that the acetate exhibited the highest hydrogen production performance [10]. At an applied voltage of 1.0 V, butyrate showed an HPR that was approximately 35% that of acetate; also, findings demonstrate that propionate was ineffective and unsuitable as an MEC substrate for hydrogen production. Another study also suggested that acetate is the most suitable VFA for use as an MEC substrate [11]. The MEC performances of the products of sludge treated via thermal hydrolysis, thermophilic acidogenic fermentation, and mesophilic acidogenic fermentation were compared. The results obtained showed that the sludge resulting from the thermal hydrolysis process (THP) is most suitable as a substrate for MEC treatment. Particularly, the sludge resulting from the THP process had a lower VFA content than that resulting from thermophilic fermentation; however, it had a higher acetate content, which led to the superior MEC performance. These findings led to the conclusion that acetate is the preferred VFA for biodegradation, followed by butyrate and propionate, based on the extent to which they are consumed in the substrate.

Palm oil mill effluent coupled with dark fermentation and an MEC exhibited high hydrogen performance with an HPR of 7.81 L/L/D at an external voltage of 0.7 V [12]. This finding indicates a threefold increase in performance, relative to that associated with dark fermentation effluents without palm oil mill effluent addition. Sugar beet juice was also examined in a dark fermentation and MEC coupled system [13]. The observed efficiency was relatively low and indicated that a higher butyrate
utilization rate is required to improve the performance. Cassava starch processing wastewater was used in a two-step fermentation process and an MEC for hydrogen production [14]. With an external voltage of 0.6 V, the hydrogen yield was 182 ml per g of COD, and the overall hydrogen recovery rate was 78%.

In another study, Satinover et al. found that the product of the fermentation of pretreated corn stover, at an organic loading rate of 30 g/L/D, exhibited the highest HPR (20.2 ± 2.0 L/L/D) [15]. Additionally, its hydrogen recovery and COD removal rates were 74.6 and 60.4%, respectively, and its anodic coulombic efficiency was 68.5%. The treatment process for the substrate consisting of approximately 50% sugar and acetate, which are easily biodegraded, included acidification, enzymatic hydrolysis, and ethanol fermentation [16].

2.2. Industrial Wastewater
To simulate complex industrial wastewater, various substrates have been examined [17]. In a particular study, dairy industry, potato industry, and biodiesel industry wastewaters, predominantly consisting of milk, starch, and glycerol, respectively, were investigated as MEC substrates, and a mixture of these three wastewaters as an MEC substrate was also investigated. The mixed substrate exhibited the highest MEC performance in terms of current density, HPR, cathodic gas recovery, and purity of the produced hydrogen. Additionally, milk exhibited the highest individual substrate performance, indicating the potential of dairy industry wastewater as an MEC substrate. The superior performance of the mixed substrate can be ascribed to the increased biodegradability. In another study, three substrates, including rice straw waste, vegetable waste, and a mixture of both, were processed through a simultaneous saccharification and fermentation and used as MEC substrates [18]. The mixture exhibited superior performance, compared with those of the individual substrates. The energy efficiency based on electrical input of the mixture reached 215.33%, and the HPR was 2.46 ± 0.07 mmol/L/D. These results also suggest that mixed substrates may be more suitable for microbial metabolism.

The MEC performances of dark fermentation effluents resulting from six types of industrial wastewaters, including cheese, fruit juice, paper, sugar, fruit processing, and spirits processing wastewaters, were explored [19]. Fruit juice wastewater exhibited the best performance owing to its degradability. Wastewaters are more advantageous compared with extract-concentration substrates in that they can be directly treated for energy production. A maximum COD removal rate of 78.5 ± 5.7% and a hydrogen yield of 1608 ± 266.2 ml Hydrogen/g of COD consumed was recorded.

The use of cheese whey, a dairy industry by-product, as an MEC substrate has also been investigated in a one-chamber MEC [20]. The effluent treated via anaerobic digestion exhibited better performance than that treated via fermentation or the raw cheese whey. The cathodic hydrogen recovery rate of this effluent was 63%, and its energy efficiency based on electrical input was 116.6%. However, its coulombic efficiency was only 31.8%, which is significantly lower than that of the cheese whey resulting from dark fermentation (92.7%), which also resulted in the production of a significant amount of methane. Another study in which raw cheese whey was used as an MEC substrate showed a recorded HPR of 0.8 L/L/D and a coulombic efficiency of 49.8 ± 8% [21].

2.3. Other Substrates
Crude glycerol is a by-product of biodiesel production that can be used as an MEC substrate for hydrogen production [22]. Generally, it is considered to have little value, and to increase its value, treatment is often required. During this experiment, an alkaline MEC with an external voltage in the range 0.8–1.2 V was used, and the results obtained showed that it is possible to produce hydrogen from crude glycerol (HPR = 0.46 L/L/D). A relatively high cathodic hydrogen recovery rate (85%) was also observed. The findings of this study led to the conclusion that under such specific conditions, crude glycerol and acetate have similar MEC performances.

Reportedly, landfill leachate treatment, coupled with an MEC, can also produce hydrogen [23]. Specifically, landfill leachate is a toxic pollutant that can be highly damaging, meaning that its further treatment using various technologies is required. To determine its MEC performance, experiments were performed using a membrane-less single-chamber MEC at an applied voltage of 0.8 V. The landfill
Leachate was mixed with diary industry wastewater, and this mixture was used as an MEC substrate. Landfill leachate concentrations in the range of 2–16% were examined, and the stabilized HPR was 15 mL/L/D. The study showed that leachate treatment is possible through MEC; however, higher loads resulted in a decline in performance. Another study on the use of landfill leachate as an MEC substrate for hydrogen production exhibited an HPR of 0.148 L/L/D at an external voltage of 1.0 V [24]. This study also reported high hydrogen yield, excellent pure hydrogen concentration, and stability, following several months of operation. Further examination of the biofilms showed that anodic microbes were enriched.

Neutralized bio-oil organic phase (NBOOP), which can be used as an MEC substrate, is a mildly acidic product resulting from the neutralization of the acids in bio-oil via capacitive deionization [25]. Its use as an MEC substrate exhibited a maximum HPR of 4.3 L/L/D, and high conversion rates for organics were also observed. The study also showed that increasing the organic loading rates resulted in an increase in the current density.

Switchgrass pyrolysis yields an aqueous stream that can be fed to an MEC [26]. Thus, a maximum HPR of 4.4 L/L/D was reported. However, the coulombic efficiency and the overall energy efficiency were 54 and 48%, respectively. Additionally, the hydrogen yield was only approximately 50% even though significantly higher efficiencies were reported at different loading rates. Reportedly, guayule and willow from pyrolysis also exhibit appreciable hydrogen production rates [27]. Willow, which has higher levels of degradable organics, showed an HPR of 5.0 ± 1.0 L/L/D, as well as higher efficiencies and recovery rates at various loading rates than guayule. The COD associated with the use of switchgrass pyrolysis products was significantly higher than those associated with guayule and willow. Another pyrolysis product, pine sawdust, also exhibited a high HPR of 5.8 ± 0.18 L/L/D [28]. However, organic loading rates beyond 10 g/L/D resulted in decreased performances.

After hydrothermal liquefaction treatment, swine manure wastewater can be used as an MEC substrate for hydrogen production [29]. This substrate, which has an organic concentration (TCOD) of 37366.7 ± 351.2 mg/L, has a relatively low pH and high conductivity. The study showed overall high efficiencies across various loading rates and applied voltages, with the organic removal rate exceeding 90% and an HPR of 168 mL/L/D. Extremely high VFA removal rates were also observed, indicating high efficiency. The by-product of the hydrothermal liquefaction of cornstalk can also be used as an MEC substrate [30]. It showed a relatively low HPR of 3.92 mL/L/D at an applied voltage of 1.0 V.

When real urban wastewater was used as an MEC substrate in a larger-scale MEC experiment, it exhibited good performance with respect to hydrogen production [31]. The MEC had a volume of 130 L and was considered a pilot experiment for upscale operations. The observed HPR reached 4.2 L/L/D, and the hydrogen gas purity exceeded 95%. Further, the real urban wastewater substrate outperformed synthetic urban wastewater, which contained glucose or glycerol. Furthermore, no buffer or pH control was performed during this study, indicating high feasibility for actual applications.

Several other experiments have been conducted using more varieties of substrates for MEC hydrogen production. Corncob treated using a two-step process showed a higher internal resistance and a lower hydrogen production rate than sodium acetate and xylose fermentation broth [32]. Makgeolli wastewater fed with acetate showed a hydrogen production rate of 1.55 L/L/D [33]; hydrogen was the main gas product, with a purity above 90%. Experiments involving dewatered sludge yielded a disappointing overall performance [34]. The HPR was only 0.027–0.038 L/L/D, and the cathodic hydrogen recovery was only 15.56–20.05%. Further, a study on food waste dark fermentation effluent showed a maximum HPR of 0.685 L/L/D and a cathodic hydrogen recovery rate above 93% [35]. Furthermore, the liquid fraction from pressed municipal solid waste was investigated as an MEC substrate [36]. However, even at an external voltage of 3.0 V, the HPR was only 0.38 ± 0.09 m³/m³/D.

3. Conclusion

MEC is a booming technology that has potential as a major source of renewable hydrogen energy. This review, which covers over 30 novel substrates that were examined in the past five years, revealed that complex and diverse substrates result in inconsistent MEC performances. The substrates covered in this
review show that MEC performances varies based on substrate, design, and the startup employed. Dark fermentation effluents and pyrolysis products are two popular substrate sources that were examined, and several untreated substrates also showed hydrogen production ability. We believe that further rigorous studies are required to determine suitable real-world scenarios in which MEC can be used for efficient and sustainable hydrogen production.

References

[1] Balat, M., Potential importance of hydrogen as a future solution to environmental and transportation problems. International Journal of Hydrogen Energy, 2008. 33(15): p. 4013-4029.
[2] Logan, B.E., et al., Microbial Electrolysis Cells for High Yield Hydrogen Gas Production from Organic Matter. Environmental Science & Technology, 2008. 42(23): p. 8630-8640.
[3] Alexander, R.R. and B.C.J. Nico, BIO-ELECTROCHEMICAL PROCESS FOR PRODUCING HYDROGEN. 2004, WAGENINGEN UNIVERSITEIT.
[4] Liu, H., S. Grot, and B.E. Logan, Electrochemically Assisted Microbial Production of Hydrogen from Acetate. Environmental Science & Technology, 2005. 39(11): p. 4317-4320.
[5] Call, D. and B.E. Logan, Hydrogen Production in a Single Chamber Microbial Electrolysis Cell Lacking a Membrane. Environmental Science & Technology, 2008. 42(9): p. 3401-3406.
[6] Lu, L. and Z.J. Ren, Microbial electrolysis cells for waste biorefinery: A state of the art review. Bioresource Technology, 2016. 215: p. 254-264.
[7] Liu, H., et al., Microbial electrolysis: novel technology for hydrogen production from biomass. Biofuels, 2014. 1(1): p. 129-142.
[8] Kadier, A., et al., A review of the substrates used in microbial electrolysis cells (MECs) for producing sustainable and clean hydrogen gas. Renewable Energy, 2014. 71: p. 466-472.
[9] Rivera, I., et al., Hydrogen production in a microbial electrolysis cell fed with a dark fermentation effluent. Journal of Applied Electrochemistry, 2015. 45(11): p. 1223-1229.
[10] Yang, N., H. Hafez, and G. Nakhla, Impact of volatile fatty acids on microbial electrolysis cell performance. Bioresource Technol, 2015. 193: p. 449-55.
[11] Lin, L., et al., Evaluation of sludge liquors from acidogenic fermentation and thermal hydrolysis process as feedstock for microbial electrolysis cells. International Journal of Hydrogen Energy, 2019. 44(57): p. 30031-30038.
[12] Khongkliang, P., et al., High efficient biohydrogen production from palm oil mill effluent by two-stage dark fermentation and microbial electrolysis under thermophilic condition. International Journal of Hydrogen Energy, 2019. 44(60): p. 31841-31852.
[13] Dhar, B.R., et al., Hydrogen production from sugar beet juice using an integrated biohydrogen process of dark fermentation and microbial electrolysis cell. Bioresour Technol, 2015. 198: p. 223-30.
[14] Khongkliang, P., et al., Continuous hydrogen production from cassava starch processing wastewater by two-stage thermophilic dark fermentation and microbial electrolysis. International Journal of Hydrogen Energy, 2017. 42(45): p. 27584-27592.
[15] Satinover, S.J., D. Schell, and A.P. Borole, Achieving high hydrogen productivities of 20 L/L-day via microbial electrolysis of corn stover fermentation products. Applied Energy, 2020. 259.
[16] Borole, A.P., C.Y. Hamilton, and D.J. Schell, Conversion of Residual Organics in Corn Stover-Derived Biorefinery Stream to Bioenergy via a Microbial Fuel Cell. Environmental Science & Technology, 2013. 47(1): p. 642-648.
[17] Montpart, N., et al., Hydrogen production in single chamber microbial electrolysis cells with different complex substrates. Water Res, 2015. 68: p. 601-15.
[18] Zhang, L., et al., Hydrogen production from simultaneous saccharification and fermentation of lignocellulosic materials in a dual-chamber microbial electrolysis cell. International Journal of Hydrogen Energy, 2019. 44(57): p. 30024-30030.
[19] Marone, A., et al., Coupling dark fermentation and microbial electrolysis to enhance biohydrogen production from agro-industrial wastewaters and by-products in a bio-refinery framework. International Journal of Hydrogen Energy, 2017. 42(3): p. 1609-1621.

[20] Rivera, I., et al., Evaluation of various cheese whey treatment scenarios in single-chamber microbial electrolysis cells for improved biohydrogen production. Chemosphere, 2017. 174: p. 168-174.

[21] Rago, L., J.A. Baeza, and A. Guisasola, Bioelectrochemical hydrogen production with cheese whey as sole substrate. Journal of Chemical Technology & Biotechnology, 2017. 92(1): p. 173-179.

[22] Badia-Fabregat, M., et al., Hydrogen production from crude glycerol in an alkaline microbial electrolysis cell. International Journal of Hydrogen Energy, 2019. 44(32): p. 17204-17213.

[23] Rani, G., et al., Batch fed single chambered microbial electrolysis cell for the treatment of landfill leachate. Renewable Energy, 2020. 153: p. 168-174.

[24] Hassán, M., et al., Hydrogen evolution in microbial electrolysis cells treating landfill leachate: Dynamics of anodic biofilm. International Journal of Hydrogen Energy, 2018. 43(29): p. 13051-13063.

[25] Park, L.K.-E., et al., Electrosorption of organic acids from aqueous bio-oil and conversion into hydrogen via microbial electrolysis cells. Renewable Energy, 2018. 125: p. 21-31.

[26] Lewis, A.J., et al., Hydrogen production from switchgrass via an integrated pyrolysis-microbial electrolysis process. Bioresour Technol, 2015. 195: p. 231-41.

[27] Satinover, S.J., et al., Microbial electrolysis using aqueous fractions derived from Tail-Gas Recycle Pyrolysis of willow and guayule. Bioresour Technol, 2019. 274: p. 302-312.

[28] Brooks, V., et al., Hydrogen production from pine-derived catalytic pyrolysis aqueous phase via microbial electrolysis. Biomass and Bioenergy, 2018. 119: p. 1-9.

[29] Shen, R., et al., Microbial electrolysis treatment of post-hydrothermal liquefaction wastewater with hydrogen generation. Applied Energy, 2018. 212: p. 509-515.

[30] Shen, R., et al., Microbial electrolysis cell to treat hydrothermal liquefied wastewater from cornstalk and recover hydrogen: Degradation of organic compounds and characterization of microbial community. International Journal of Hydrogen Energy, 2016. 41(7): p. 4132-4142.

[31] Baeza, J.A., et al., Bioelectrochemical hydrogen production from urban wastewater on a pilot scale. Journal of Power Sources, 2017. 356: p. 500-509.

[32] Yan, D., X. Yang, and W. Yuan, Electricity and H2 generation from hemicellulose by sequential fermentation and microbial fuel/electrolysis cell. Journal of Power Sources, 2015. 289: p. 26-33.

[33] Kim, J.H., et al., Hydrogen Production from Makgeolli Wastewater Using a Single-Chamber Microbial Electrolysis Cell. Bulletin of the Korean Chemical Society, 2020. 41(2): p. 150-155.

[34] Hu, K., et al., Degradation of organics extracted from dewatered sludge by alkaline pretreatment in microbial electrolysis cell. Environmental Science and Pollution Research, 2018. 25(9): p. 8715-8724.

[35] Cardeña, R., I. Moreno-Andrade, and G. Buitrón, Improvement of the bioelectrochemical hydrogen production from food waste fermentation effluent using a novel start-up strategy. Journal of Chemical Technology & Biotechnology, 2018. 93(3): p. 878-886.

[36] Zhen, G., et al., Recovery of biohydrogen in a single-chamber microbial electrohydrogenesis cell using liquid fraction of pressed municipal solid waste (LPW) as substrate. International Journal of Hydrogen Energy, 2016. 41(40): p. 17896-17906.