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Advances in Biohydrogen Production: Techniques, Challenges, and Future Prospects

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ABSTRACT

Progressing to sustainable and renewable energy sources is essential for addressing climate change and reducing reliance on fossil fuels. Biohydrogen production, which is the production of hydrogen via biological processes, can be one solution since it provides high-value energy and environmental benefits. This review aims to provide summary information about the three basic strategies involved in biohydrogen production: dark fermentation, photo fermentation, and microbial electrolysis cells (MECs). Dark fermentation is the fermentative conversion of organic matter via anaerobic microbes into hydrogen and other products. While this approach is simple and could work with diverse waste substrates, scalability is limited due to low hydrogen yields and the need for substrate pretreatment. Photosynthetic bacteria employ light to transform organic substrates into hydrogen. Photosynthetic bacteria use light to convert organic substrates into hydrogen, indicating that this process could complement dark fermentation. However, its dependence on light and low efficiency present significant challenges. Microbial electrosynthesis (MEC) converts hydrogen into energy by reducing it from carbon dioxide and using electroactive bacterium combined with an external voltage to produce gas from organic matter that is well-aligned but complex with wastewater treatment while also facing costs that make them highly operational. These technologies are likely to see improvements in genetic engineering, new designs for reactors, and the use of them with other industrial processes to improve productivity and profitability. Ongoing research and development are essential to realize large-scale, practical methods for biohydrogen production. This review covers the potential of biosynthetic and anaerobic approaches in this field.

Keywords: Biohydrogen; dark fermentation; photo fermentation; microbial electrolysis cells; sustainable energy

1. Introduction

Hydrogen is a high energy content clean fuel that could replace fossil fuels. One promising method of producing hydrogen is through biohydrogen production, which involves biological processes to generate hydrogen. This approach offers a sustainable and eco-friendly solution [1]. Biohydrogen production can briefly be defined as the biological generation of hydrogen gas. The method could be one of the most significant and promising sustainable energy sources since there are types and ways in industrial applications along with biohydrogen production techniques. Many kinds of methods can be applied to produce biohydrogen. Dark fermentation (DF), a biohydrogen production process, is widely employed because it can use various waste materials and has a simple design [2]. It is low-cost, offers higher hydrogen production, and has no adverse environmental harm. Nevertheless, dark fermentation's hydrogen yield, by-product management, and process efficiency still have much to improve [3].

Photo fermentation (PF) is a process in which organic materials, such as organic acids, are transformed into hydrogen using photosynthetic bacteria at illuminated conditions [4]. In contrast, dark fermentation involves the conversion of organic substrates, including waste materials, into hydrogen without the presence of light. Dark fermentation typically uses anaerobic bacteria to break down carbohydrates into hydrogen and organic acids, whereas photo fermentation relies on photosynthetic bacteria to convert these organic acids into hydrogen [5]. While this method yields a superior rate of hydrogen production compared to dark fermentation, it requires specialized light and is ineffective with natural solar energy. Another option for biohydrogen production is Microbial Electrolysis Cells (MECs). MECs utilize organic matter for the microbially aided production of hydrogen using electroactive bacteria in the presence of an external voltage [6]. It has a high hydrogen yield and integrates well with wastewater treatment. However, this system's high operational costs and complex nature are a huge roadblock. The advantages of DF, PFs, and MECs can be explored jointly to find an alternative approach to improve their drawbacks. As a result, the integration of these methods gives rise to a hybrid process, which ultimately leads to augmentations in biohydrogen output [7]. These approaches do not need any special protective conditions or complicated processes, so they are economically friendly and require less work power [8]. These approaches will enhance the research process to augment biohydrogen production efficiency and yield [9].

Biohydrogens play an essential role in the advancement of sustainable energy knowledge. Researching more and developing biohydrogen production processes is an option for cleaner and less hazardous energy sources. This solves the global energy shortage and contributes to sustainability and environmental protection by minimizing fossil fuel consumption and greenhouse release. The evolutionary development of biohydrogen might yield novel functionalities and pathways to complement and supplement other renewable energy technologies, contributing to future science in sustainable energy systems. This article will review the status of technology in biohydrogen, key processes, limitations for the valorization of biowaste, and prospects for addressing areas that will enhance this sustainability.

2. Methods of Biohydrogen Production

2.1 Dark Fermentation

Microorganisms conduct dark fermentation, a biological process that converts organic substrates into hydrogen without light. This is an easy technique relative to other biohydrogen production methods with several benefits and the reuse of waste materials [2]. Some key procedural features are that microorganisms use anaerobically to obtain hydrogen, and it is instilled within the metabolic pathway [3]. Various microbes, particularly anaerobes, assist in the breakdown of organic molecules and hydrogen generation. These organisms ferment these organic substrates to even smaller compounds, and hydrogen is one of the most abundant end products [10]. It also generates other by-products, such as organic acids or carbon dioxide.

Dark fermentation is the most suitable because it is relatively specific and can utilize a wide range of substrates and feedstock that contribute to waste management. The provision of fermentable sugars taps lignocellulosic biomass, another common substrate with possibly ubiquitous availability, offers an attractive basis aligning with sustainable resource usage via waste recycling [11]. Carbohydrate-rich feedstocks for hydrogen production can include industrial

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and agricultural wastes such as crop residues and food waste. Additionally, wastewater, although not primarily carbohydrate-rich, can also be utilized depending on its composition [12]. This by-product contains large amounts of organic matter, which can be used to reduce waste and generate energy [13]. Dark fermentation considers sewage sludge and other organic wastes as by-products of different processes (e.g., beer lees, sake lees), which can be transformed synergistically with industrial systems through improved resource recovery efficiencies [14]. Certain plant materials containing high cellulose, like wheat straw, rice straw, cassava, and sugarcane bagasse, are generally used because they provide a localized source of feedstock near the agricultural areas [15]. Additionally, glycerol, a by-product of biodiesel production, is a simple sugar alcohol easily fermented by microorganisms, showcasing the potential of dark fermentation to create value from industrial by-products [16]. Fig. 1 shows the diagram illustrating the functioning of the dark fermentation process. The process used dark fermentative bacteria with hydrolyzed starch waste plus nutrients as the substrate. The feedstock undergoes pretreatment to enhance hydrolysis and improve biodegradability. During dark fermentation, the bacteria convert the organic material into hydrogen and volatile fatty acids (VFAs), which are short-chain organic acids produced during fermentation.

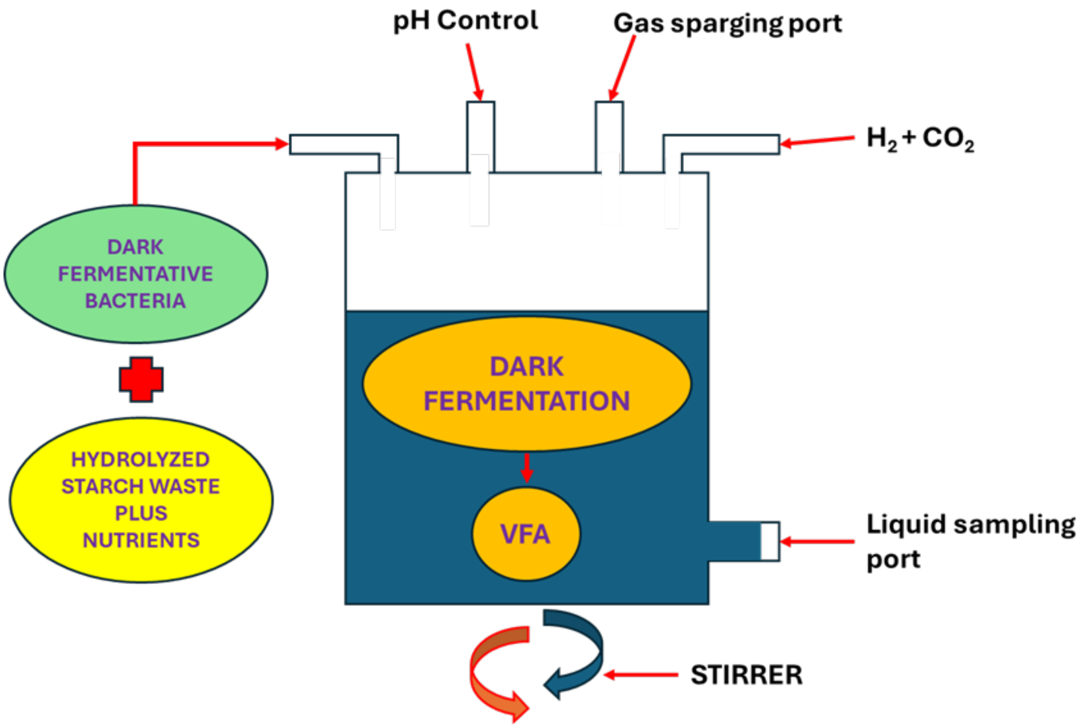

Fig. 1: Diagram illustrating the functioning of the dark fermentation process

The applications of dark fermentation extend beyond just hydrogen production. Primarily, it is used for renewable energy production, generating hydrogen as a clean energy carrier that can be utilized in fuel cells or directly as fuel, aiding the transition to renewable energy sources [17]. Additionally, dark fermentation is an effective waste management tool that utilizes various waste streams as substrates, thereby reducing the environmental impact of organic waste disposal [18]. Integrating dark fermentation into biorefinery systems allows for deriving multiple valuable products from biomass, maximizing resource utilization [19]. Furthermore, dark fermentation can be applied in wastewater treatment, where it simultaneously treats organic-rich wastewater and produces hydrogen as a valuable by-product. Different feedstocks or substrates can be used in dark fermentation, including carbohydrate-rich materials like starch, cellulose-based materials, and various organic wastes.

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2.2 Photo Fermentation

During photo fermentation, organic substrates are converted to hydrogen gas and other by-products in the existence of light by photosynthetic microorganisms, mainly purple non-sulfur bacteria (PNSB) [4]. Photo fermentation depends on light energy to process hydrogen production, which occurs in the presence of light, while dark fermentation happens without the existence of light. Photo fermentation is characterized by the light dependence and capture of light energy by the photosynthetic apparatus of the microorganisms to produce hydrogen. Although photo fermentation needs light, it can only occur in anaerobic conditions, which is vital for microorganisms to produce hydrogen as a metabolic end-product [20]. Organisms from different groups, especially PNSB, are involved in the photoconversion of organic substrates to hydrogen. They are microorganisms that can metabolize organic substrates, often organic acids or simple sugars, forming more simple precursors, one of which is hydrogen [4]. In photo fermentation, hydrogen production is mainly mediated by the nitrogenase enzyme; this enzyme typically catalysis nitrogen fixation but can also generate hydrogen in specific environments [21]. The photo fermentation process in operation is exemplified by Fig. 2. In particular, photoreactors operate in batch mode to provide an oxygen-free environment for the anaerobic activity of purple non-sulfur bacteria (PNSB) on organic compounds, such as organic acids and simple sugars, resulting in hydrogen production.

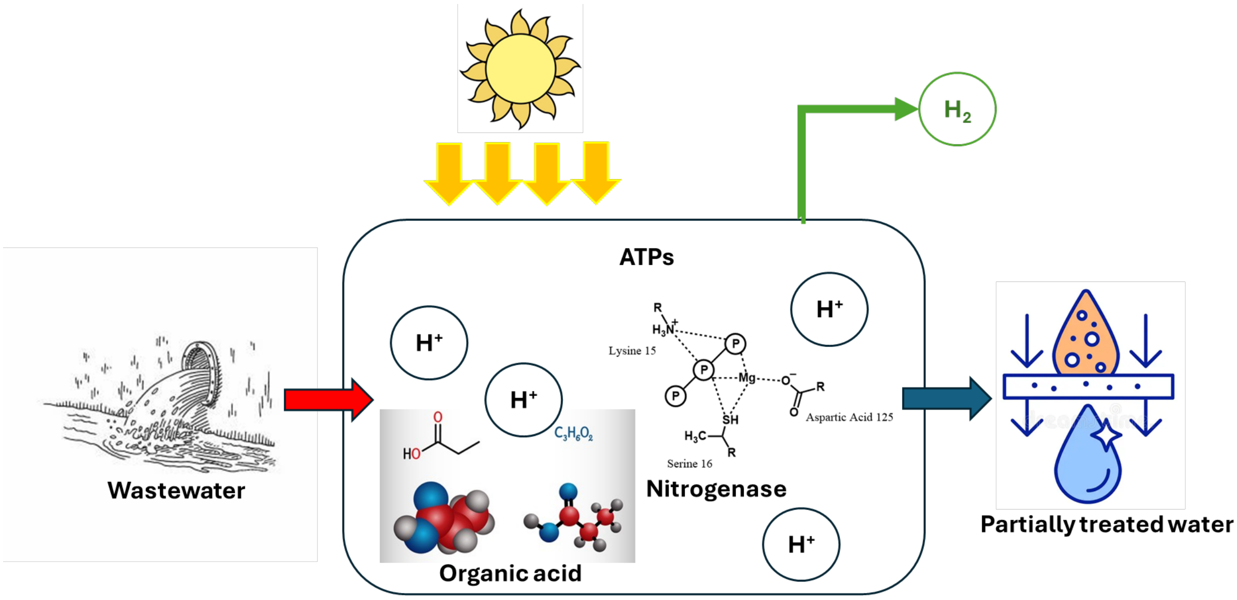

Fig. 2: A diagram showing how the photo-fermentation process works

In photo fermentation, commonly used substrates for photosynthetic bacteria are volatile fatty acids such as acetic acid, butyric acid, and lactic acid, which are often considered to be by-products of dark fermentation processes, meaning that these two processes could complement each other [22]. Other substrates include simple sugars such as glucose and fructose [23]. Moreover, many industrial and agricultural wastewaters are organic-rich and can be photo-fermentation substrates [24]. Similarly, since effluents resulting from dark fermentation processes yield unconverted sugars and organic acids, they are also excellent substrates for photo fermentation, allowing further integration of both [24].

Photo fermentation not only has the application of producing hydrogen. The first one, mainly relevant to renewable energy production, generates hydrogen as a clean and high-density energy carrier that can be used in fuel cells or directly as a fuel, supporting the shift toward renewables [25]. In addition, photo fermentation is also used in wastewater treatment, whereby organic-rich wastewater is treated while hydrogen (a vital fuel choice) is generated as a side-product. The process can also be combined with dark fermentation to increase hydrogen yield in a two-stage process, where the organic acids produced by dark fermenters are used as substrates [2]. Carbon dioxide fixation is possible in some photosynthetic bacteria allocated for photo fermentation, which may help carbon sequestration efforts [26]. Lastly, the efficiencies of photo fermentation can also produce some other high-value products (e.g., biodegradable plastics-polyhydroxy butyrate (PHB)), which shows its versatility for producing high-value products [27].

2.3 Microbial Electrolysis Cells (MECs)

MECs exploit microorganisms to catalyze the conversion of organic matter into hydrogen gas and other value-added products in bioelectrochemical systems [28]. MECs are based on principles opposite to MFCs; bioelectrochemical conversion uses metabolic activity to convert the chemical energy stored in organic compounds into electricity and produce hydrogen gas or other products [29]. This process occurs under anaerobic conditions, which are crucial for the microorganisms to produce hydrogen as a metabolic by-product. Unlike Microbial Fuel Cells (MFCs), MECs require a small external power input to overcome thermodynamic barriers and drive hydrogen production or other desired products [30]. MECs utilize electrodes (anode and cathode) to facilitate the transfer of electrons from the microorganisms to the final electron acceptor [31]. Fig. 3 shows the MEC's functioning, which includes an anode, electrogenic microbes, a membrane, a cathode, and a power supply.

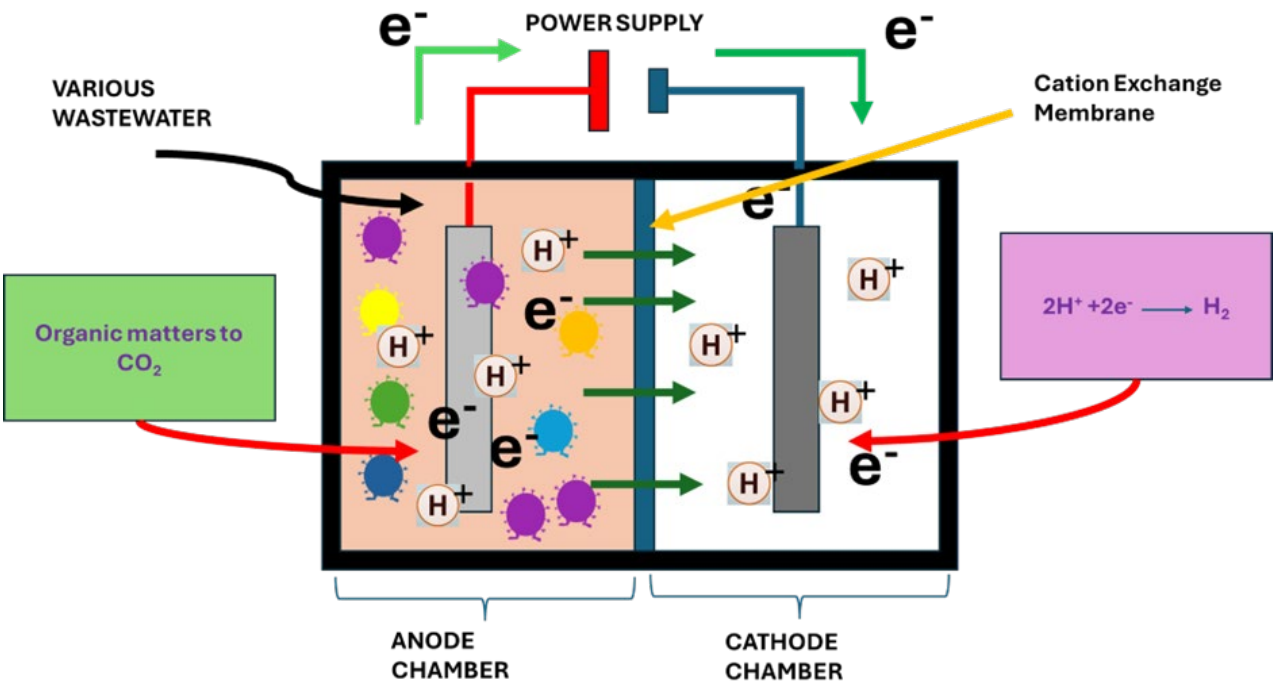

Fig. 3: Diagram illustrating the functioning of the Microbial Electrolysis Cell process

A typical Microbial Electrolysis Cell (MEC) consists of several key components: the anode, where electrochemically active microorganisms oxidize organic substrates and transfer electrons to the electrode; the cathode, where reduction reactions occur, typically producing hydrogen gas from protons; the electrolyte solution, which contains the organic substrate and facilitates ion transport between the electrodes; a proton exchange membrane or ion exchange membrane, which separates the anode and cathode chambers while allowing proton transfer; and an external power supply, which applies a small voltage to overcome thermodynamic barriers and drive the desired reactions [32].

The microbial community is one of the most significant factors affecting Microbial Electrolysis Cells' (MECs) performance. Exoelectrogens are electrochemically active microorganisms (e.g., species of Geobacter, Shewanella) that can transfer electrons to the anode [33]. Despite the crucial role of these exoelectrogens, complex microbial communities that play an essential role in substrate degradation and the MEC's overall performance are often inoculated with MECs

[34]. Furthermore, the biofilms turned on an anode surface are prerequisite for electron transfer and the performance of MECs [35].

Microbial electrolysis cells (MECs) have a broad substrate range and the ability to use many different types of organic substrates; this adds the dimension of flexibility and potential usefulness to their application. MEC substrates include a wide range of domestic, industrial, and agricultural wastewater [36]. Moreover, volatile fatty acids and other organic acids are frequently utilized substrates that confer abundant organic material for microbial activity [34]. Other suitable substrates include simple sugars and more complex carbohydrates, as they provide the mobilizable carbon needed for the metabolisms of electroactive microorganisms [37]. After pretreatment, lignocellulosic biomass can be used as a substrate, expanding the spectrum of feedstocks accepted for MECs [37]. Simple sugars typically do not require pretreatment, whereas complex sugars and lignocellulosic materials do. The capability to use a variety of substrates makes MECs an exciting technology for sustainable energy generation and waste management.

Applications of MECs are up-and-coming. Mainly, MECs are applied for hydrogen gas production as a clean energy carrier. MECs are also widely used in wastewater treatment, as they can treat organic-rich wastewater and produce valuable products (H_2) at the same time [37]. MECs also have applications in resource recovery, where MECs are used to recover valuable resources like nutrients or metals from waste streams. MECs can co-produce energy and (e.g., nitrogen) nutrient recovery from several wastewater streams [38]. They can be applied to pollutant removal, methane, hydrogen peroxide production, and resource recovery [39]. Microbial fuel cells (MFCs) are integrative with MEC and share fundamentals like MFC. To expand its functionality, the pair can be complemented with further technologies like anaerobic digestion or other microbial electrochemical systems [39]. However, though MECs have limitations in scaling up, they have advantages over conventional treatment methods, such as low environmental impact and lower infrastructure costs [38]. Furthermore, nutrient reclaim from waste streams can enable sustainable microalgal cultivation for high-value biorefineries and assist with crucial economic and environmental issues [40].

As biosensors, the electrical output of MECs can be utilized; in these systems, organic content and certain water compounds may be detected, which are cost-effective features like real-time response [39,41]. Wang et al. [41] reported a MEC-based BOD sensor with high sensitivity for determining BOD concentrations between 10-500 mg/L. Micro-microbial electrochemical sensor with integrated bioanode and biocathode was developed by Chu et al. [42] to detect formaldehyde in the aqueous phase. A microscale MFC biosensor was then constructed by Fei and Ren [43], which performed high-throughput bioanalysis to allow for a sigmoid relationship between the applied doses (i.e., formaldehyde concentrations at different levels from 1×10^{-6} to 3×10^{-3} g/L) and dose responses to be established. Further, microbial electrochemical systems (MECs) are a promising new tool for water quality monitoring for pollutant detection and environmental change monitoring [39]. In addition, MECs can combine other bioelectrochemical systems with renewable energy technologies to improve overall efficiency and product [44]. The versatility of MECs provides the potential for a broad range of sustainable energy and environmental management applications.

3. Challenges in Biohydrogen Production

Dark fermentation represents an encouraging strategy for biohydrogen generation from lignocellulosic feedstock, providing a sustainable replacement of fossil fuels [45-46]. However, many issues prevent it from becoming more widespread. These are low yield and productivity [46], the requirement of proper biomass pretreatment methods for enhancing lignin-depolymerizing efficiency [3], and scale-up barriers from lab to industry level [45]. Researchers have investigated several approaches to mitigate these challenges, including the co-location of dark fermentation and microbial electrolysis cells [17], design optimization of bioreactors, and metabolic engineering [3]. Moreover, adding additives, especially iron-based materials, appears to enhance the pretreatment of biomass and biohydrogen production [3]. Overcoming these constraints may provide a more economically feasible and efficient pathway toward biohydrogen production, thereby enhancing a sustainable circular bioeconomy and less dependence on fossil fuels [46].

Nevertheless, various limitations associated with the photo-fermentation of biohydrogen may hamper its broad-scale applications. This approach has, however, some drawbacks concerning low light conversion efficiency and small hydrogen yields [20,47]. Furthermore, the enzymes' oxygen sensitivity and limited ability to use complex carbohydrates also result in low yields [20]. Researchers are investigating possible solutions, including improving light transfer

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efficiency and enzyme activity through genetic engineering, immobilization technology, and nanotechnology [20,47]. Combining dark and photo-fermentation can potentially enhance hydrogen yield [47-48]. Despite these challenges, photo-fermentation is still attractive due to its higher efficiency, lower cost, and environmental impact [20,24]. However, more studies are required to achieve the best conditions for the most effective biohydrogen production and its subsequent commercialization [48].

Microbial Electrolysis Cells (MECs) also meet several challenges that impede large-scale application and commercialization. The high electrode and membrane costs, high internal resistance, methanogenesis, and membrane/cathode biofouling are listed as some of these challenges [30]. Due to this, however, design limitations and scale-up issues represent real challenges for practical applications [49]. Researchers have investigated different approaches to overcome these difficulties, such as optimizing reactor designs, upgrading anode materials, and coupling MECs with other bioelectrochemical systems [39]. Nanomaterials are, indeed, addressing many technological challenges thanks to their peculiar physiochemical properties [50]. MEC can also be improved by optimizing operational parameters like organic loading, ionic strength, hydraulic retention time, and applied voltage [30]. MECs are still considered promising technologies for sustainable biohydrogen production and wastewater treatment systems, but they have these limitations. Additional investigation is required to improve and strengthen the performance/ applicability of MEC.

4. Future Prospects

There is a significant potential for dark fermentation, photo fermentation, and Microbial Electrolysis Cells (MECs) in the future, and they have been very much researched areas. Genetic engineering of the organisms during dark fermentation is being explored to enhance microorganism hydrogen production and inhibition tolerance [51]. Furthermore, there is growing interest in the combination of dark fermentation with other processes (e.g., photo–fermentation or MECs) to improve overall hydrogen yield and process performance [52]. Designs for new reactors that optimize substrate consumption, product separation, and the overall process [53]. In addition, investigations for suitable substrate combinations and pretreatment processes contribute to the efficiency of the process and make it economically feasible [54]. Dark fermentation can also be included with more elaborate biorefinery designs that generate multiple commercial items from lignocellulosic biomass feedstocks.

It is also used in combination with photo fermentation to enhance light-dependent hydrogen production of photosynthetic bacteria and ameliorate light-utilization efficiency and tolerance to inhibition conditions [55]. Moreover, researchers are investigating the coupling of photo fermentation with other processes, such as dark fermentation or MECs, to enhance hydrogen production and efficiency [56]. Various designs of photobioreactors are being proposed to enhance light penetration, substrate consumption, and product separation [57]. Studies of artificial light sources such as long-wavelength LEDs are conducted to improve the efficiency of lights for continuous operation without being affected by outdoor light [58]. Future directions may target incorporating photo fermentation into larger biorefinery designs and designing resilient outdoor systems that could function efficiently across environmental variations.

Researchers are focusing on developing novel electrode materials for MEC technology that have improved conductivity, biocompatibility, and durability [59]. Microorganisms capable of transferring electrons and utilizing more diverse substrates are potential targets of genetic engineering efforts to achieve this [60]. It has also been reported that integrating MECs with other bioelectrochemical systems or renewable energy technologies (such as solar and wind) can boost the overall stability, efficiency, and product yield of a whole bioprocess [61]. New reactor designs are in progress to improve substrate consumption, product separation, and process performance [53]. The scope of economically valuable products generated in MECs is being extended beyond hydrogen production through research. Another line of research in MEC studies has been the application of artificial intelligence and machine learning for process optimization and control.

5. Conclusion

Dark fermentation, photo fermentation, and Microbial Electrolysis Cells (MECs) are the methods associated with the potential to develop sustainable hydrogen production and waste management strategies. Due to its versatility in using various substrates (including waste streams), dark fermentation can be considered a promising technology for coupling renewable energy production with waste treatment. Challenges like low hydrogen yield and process stability exist; however, extensive research efforts are currently being made to facilitate more efficient and large-scale applications. Using photo fermentation, organic acids and other by-products of fermentation processes can be utilized for sustainable hydrogen production and wastewater treatment. Its drawbacks include light reliance and low conversion affection, but current research improves its viability. MECs are, therefore, a promising technology for waste-to-hydrogen transformation, merging a waste management technique with renewable energy production. They have problems like low production rates and scaling limitations, but constant research and development are making breakthroughs. With the world looking for sustainable energy solutions, these technologies are expected to be crucial in transitioning towards a circular bioeconomy.

References

[1] Rathi BS, Aravind VD, Ranjith G, Kishore V, Ewe LS, Yew WK, Baskaran R. Sustainability considerations in bio-hydrogen from bio-algae with the aid of bio-algae cultivation and harvesting: Critical review. MRS Energy Sustain, 2024;1:1-26.

[2] Jain R, Panwar NL, Jain SK, Gupta T, Agarwal C, Meena SS. Bio-hydrogen production through dark fermentation: an overview. Biomass Convers Biorefin, 2024;14(12):12699-12724.

[3] Zhao ZT, Ding J, Wang BY, Bao MY, Liu BF, Pang JW, Ren NQ, Yang SS. Advances in the biomass valorization in dark fermentation systems: A sustainable approach for biohydrogen production. Chem Eng J, 2024;448:148444.

[4] Gupta S, Fernandes A, Lopes A, Grasa L, Salafranca J. Photo-Fermentative Bacteria Used for Hydrogen Production. Appl Sci, 2024;14(3):1191.

[5] Goren AY, Dincer I, Khalvati A. Green biohydrogen production from renewable plant-based resources: A comparative evaluation. Process Saf Environ Prot, 2024;185:947-977.

[6] Ndayisenga F, Yu Z, Wang B, Zhou D. Effects of the applied voltage on electroactive microbial biofilm viability and hydrogen production in a recalcitrant organic waste-fed single-chamber membrane-free microbial electrolysis cell performance. Chem Eng J, 2023;469:144002.

[7] El-Sayad RM, Khalil AS. Wastewater treatment and hydrogen production via Microbial electrolysis cells (MECs) and fermentation methods: A comparative review. J Contemp Technol Appl Eng, 2023;2(1):29-41.

[8] Feng S, Ngo HH, Guo W, Chang SW, Nguyen DD, Bui XT, Zhang XY, Ma X, Hoang BN. Biohydrogen production, storage, and delivery: A comprehensive overview of current strategies and limitations. Chem Eng J, 2023;448:144669.

[9] Ghosh A, Koley A, Pal S, Gupta N, Show BK, Nahar G, Balachandran S. Technological Advancement for Biohydrogen Production from Agricultural Waste of In: editor. Technol Adv Biohydrogen Prod Biowaste, 2024:175-221.

[10] Albuquerque MM, Sartor GB, Martinez-Burgos WJ, Scapini T, Edwiges T, Soccol CR, Medeiros ABP. Biohydrogen Produced via Dark Fermentation: A Review. Methane, 2024;3(3):500-532.

[11] Guo J, Zhang Y, Fang J, Ma Z, Li C, Yan M, Qiao N, Liu Y, Bian M. Reduction and Reuse of Forestry and Agricultural Bio-Waste through Innovative Green Utilization Approaches: A Review. Forests, 2024;15(8):1372.

[12] Meena PK, Awale SD, Kumar S, Kumar D, Kumar N. Thermochemical conversion of agricultural residue for the production of hydrogen, methane, and biofuels: A comprehensive overview. Ind Crops Prod, 2024;221:119340.

[13] Shukla KA, Sofian ADABA, Singh A, Chen WH, Show PL, Chan YJ. Food waste management and sustainable waste to energy: Current efforts, anaerobic digestion, incinerator and hydrothermal carbonization with a focus in Malaysia. J Clean Prod, 2024;448:141457.

eISSN Number: 3036-017X

[14] Garduno R, Baranova E, Kinsley C. Valorization of brewery waste slurry with glycerol as co-substrate for hydrogen and butyrate production using dark fermentation. J Chem Technol Biotechnol, 2023;98(8):1975-1985.

[15] Karn A, Thakur S, Shrestha B. Extraction and characterization of cellulose from agricultural residues: wheat straw and sugarcane bagasse. J Nepal Chem Soc, 2022;43(1):93-98.

[16] Elsayed M, Eraky M, Osman AI, Wang J, Farghali M, Rashwan AK, Yacoub IH, Hanelt D, Abomohra A. Sustainable valorization of waste glycerol into bioethanol and biodiesel through biocircular approaches: a review. Environ Chem Lett, 2024;22(2):609-634.

[17] Srivastava P, García-Quismondo E, Palma J, González-Fernández C. Coupling dark fermentation and microbial electrolysis cells for higher hydrogen yield: Technological competitiveness and challenges. Int J Hydrogen Energy, 2024;52:223-239.

[18] Camacho CI, Estévez S, Conde JJ, Feijoo G, Moreira MT. Dark fermentation as an environmentally sustainable WIN-WIN solution for bioenergy production. J Clean Prod, 2022;374:134026.

[19] Kumar AN, Sarkar O, Chandrasekhar K, Raj T, Narisetty V, Mohan S V, Pandey A, Varjani S, Kumar S, Sharma P. Upgrading the value of anaerobic fermentation via renewable chemicals production: a sustainable integration for circular bioeconomy. Sci Total Environ, 2022;806:150312.

[20] Suresh G, Kumari P, Mohan SV. Light-dependent biohydrogen production: Progress and perspectives. Bioresour Technol, 2023;380:129007.

[21] Mishra P, Krishnan S, Rana S, Singh L, Sakinah M, Ab Wahid Z. Outlook of fermentative hydrogen production techniques: An overview of dark, photo and integrated dark-photo fermentative approach to biomass. Energy Strategy Rev, 2019;24:27-37.

[22] Zagrodnik R, Duber A. Continuous dark-photo fermentative H2 production from synthetic lignocellulose hydrolysate with different photoheterotrophic cultures: Sequential vs. co-culture processes. Energy, 2024;290:130105.

[23] Policastro G, Cesaro A, Fabbricino M. Photo-fermentative hydrogen production from cheese whey: Engineering of a mixed culture process in a semi-continuous, tubular photo-bioreactor. Int J Hydrogen Energy, 2023;48(55):21038-21054.

[24] Putatunda C, Behl M, Solanki P, Sharma S, Bhatia SK, Walia A, Bhatia RK. Current challenges and future technology in photofermentation-driven biohydrogen production by utilizing algae and bacteria. Int J Hydrogen Energy, 2023;48(55):21088-21109.

[25] Akram F, Fatima T, Ibrar R, ul Haq I. Biohydrogen: Production, promising progressions and challenges of a green carbon-free energy. Sustain Energy Technol Assess, 2024;69:103893.

[26] Blessing S, Pisciotta JM. Carbon sequestration: principle and recent advances. Clean Technol Toward Sustain Future, 2023;43.

[27] Sharma R, Solanki P, Chaudhary M, Gupta N, Kaur P. Unveiling the potential of microalgae for bioplastic production from wastewater–current trends, innovations, and future prospects. Biotechnol Sustain Mater, 2024;1(1):10.

[28] Gautam R, Nayak JK, Ress NV, Steinberger-Wilckens R, Ghosh UK. Bio-hydrogen production through microbial electrolysis cell: structural components and influencing factors. Chem Eng J, 2023;455:140535.

[29] Pandya RS, Kaur T, Bhattacharya R, Bose D, Saraf D. Harnessing microorganisms for bioenergy with microbial fuel cells: powering the future. Water-Energy Nexus, 2023.

[30] Sharma A, Mehdi SEH, Pandit S, Eun-Oh S, Natarajan V. Factors affecting hydrogen production in microbial electrolysis cell (MEC): A review. Int J Hydrogen Energy, 2024;61:1473-1484.

[31] Zhang J, Li F, Liu D, Liu Q, Song H. Engineering extracellular electron transfer pathways of electroactive microorganisms by synthetic biology for energy and chemicals production. Chem Soc Rev, 2024.

[32] Liu H Z, Chen T Z, Wang N, Zhang Y R, Li JC. A new strategy for improving MFC power output by shared electrode MFC–MEC coupling. Appl Energy, 2024;359:122677.

[33] Jayathilake C, Piyumika G, Nazeer Z, Wijayawardene N, Rajakaruna S, Kumla J, Fernando E. Recent progress in the characterization and application of exo-electrogenic microorganisms. Antonie Van Leeuwenhoek, 2024;117(1):10.

[34] Li M, Zhang Q, Liu Y, Zhu J, Sun F, Cui M H, Liu H, Zhang TC, Chen C. Enhancing degradation of organic matter in microbial electrolytic cells coupled with anaerobic digestion (MEC-AD) systems by carbon-based materials. Sci Total Environ, 2023;900:165805.

eISSN Number: 3036-017X

[35] Li D, Ping Q, Mo R, Guo W, Zhang S, Wang L, Li Y. Revealing synergistic mechanisms of biochar-assisted microbial electrolysis cells in enhancing the anaerobic digestion performance of waste activated sludge: Extracellular polymeric substances characterization, enzyme activity assay, and multi-omics analysis. Water Res, 2024;267:122501.
[36] Muddasar M, Liaquat R, Rahman MZU, Khoja AH, Aslam A, Basit A. Coaxial microbial electrolysis cell for cost-effective bioenergy production and wastewater treatment of potato industry effluent. J Chem Technol Biotechnol, 2023;98(9):2203-2213.
[37] Hemashenpagam N, Selvajeyanthi S, Gunasheela N. Effect of Substrate on the Efficiency of Microbial Electrolysis Cells of In: editor. Effect Substrate Efficiency Microb Electrolysis Cells, 2024:75-91.
[38] Zhai S, Zhang D, Liu W, Wang B, Liang B, Liu C, Zeng R, Hou Y, Cheng HY, Wang A. Microbial electrochemical technologies assisted nitrogen recovery from different wastewater sources: Performance, life cycle assessment, and challenges. Resour Conserv Recycl, 2023;194:107000.
[39] Radhika D, Shivakumar A, Kasai DR, Koutavarapu R, Peera SG. Microbial electrolysis cell as a diverse technology: Overview of prospective applications, advancements, and challenges. Energies, 2022;15(7):2611.
[40] Premaratne M, Nishshanka GKSH, Anthonio RADP, Liyanaarachchi V C, Thevarajah B, Nimarshana P, Malik A, Ariyadasa T U. Resource recovery from waste streams for production of microalgae biomass: A sustainable approach towards high-value biorefineries. Bioresour Technol Rep, 2022;18:101070.
[41] Wang Z, Liao C, Zhong Z, Liu S, Li M, Wang X. Design, optimization and application of a highly sensitive microbial electrolytic cell-based BOD biosensor. Environ Res, 2023;216:114533.
[42] Chu N, Liang Q, Hao W, Jiang Y, Zeng RJ. Micro-microbial electrochemical sensor equipped with combined bioanode and biocathode for water biotoxicity monitoring. Bioresour Technol, 2021;326:124743.
[43] Fei S, Ren H. Determining the Dose–Response Curve of Exoelectrogens: A Microscale Microbial Fuel Cell Biosensor for Water Toxicity Monitoring. Micromachines, 2022;13(10):1560.
[44] Bora A, Mohanrasu K, Swetha T A, Ananthi V, Sindhu R, Chi N T L, Pugazhendhi A, Arun A, Mathimani T. Microbial electrolysis cell (MEC): Reactor configurations, recent advances and strategies in biohydrogen production. Fuel, 2022;328:125269.
[45] Rani P, Yadav D K, Yadav A, Bishnoi NR, Kumar V, Ram C, Pugazhendhi A, Kumar SS. Frontier in dark fermentative biohydrogen production from lignocellulosic biomass: Challenges and future prospects. Fuel, 2024;366:131187.
[46] Ahmad A, Rambabu K, Hasan SW, Show PL, Banat F. Biohydrogen production through dark fermentation: Recent trends and advances in transition to a circular bioeconomy. Int J Hydrogen Energy, 2024;52:335-357.
[47] Cheng D, Ngo HH, Guo W, Chang SW, Nguyen DD, Bui XT, Zhang XY, Ma X, Hoang BN. Enhanced photo-fermentative biohydrogen production from biowastes: an overview. Bioresour Technol, 2022;357:127341.
[48] Qyyum MA, Ismail S, Ni SQ, Ihsanullah I, Ahmad R, Khan A, Tawfik A, Nizami AS, Lee M. Harvesting biohydrogen from industrial wastewater: Production potential, pilot-scale bioreactors, commercialization status, techno-economics, and policy analysis. J Clean Prod, 2022;340:130809.
[49] Murugaiyan J, Narayanan A, Mohamed SN. An overview of microbial electrolysis cell configuration: Challenges and prospects on biohydrogen production. Int J Energy Res, 2022;46(14):20811-20827.
[50] Kim B, Yang E, Kim B, Obaid M, Jang JK, Chae KJ. Recent application of nanomaterials to overcome technological challenges of microbial electrolysis cells. Nanomaterials, 2022;12(8):1316.
[51] Talapko D, Talapko J, Erić I, Škrlec I. Biological hydrogen production from biowaste using dark fermentation, storage and transportation. Energies, 2023;16(8):3321.
[52] Hosseinzadeh A, Zhou JL, Li X, Afsari M, Altaee A. Techno-economic and environmental impact assessment of hydrogen production processes using bio-waste as renewable energy resource. Renew Sustain Energy Rev, 2022;156:111991.
[53] Liu Z, Xue X, Cai W, Cui K, Patil S A, Guo K. Recent progress on microbial electrosynthesis reactor designs and strategies to enhance the reactor performance. Biochem Eng J, 2023;190:108745.
[54] Ali SS, Al-Tohamy R, Elsamahy T, Sun J. Harnessing recalcitrant lignocellulosic biomass for enhanced biohydrogen production: Recent advances, challenges, and future perspective. Biotechnol Adv, 2024:108344.
[55] Liu X, Ren N, Song F, Yang C, Wang A. Recent advances in fermentative biohydrogen production. Prog Nat Sci, 2008;18(3):253-258.

[56] Kanwal F, Torriero A A. Biohydrogen—A green fuel for sustainable energy solutions. Energies, 2022;15(20):7783.
[57] Sirohi R, Pandey A K, Ranganathan P, Singh S, Udayan A, Awasthi MK, Hoang AT, Chilakamarry CR, Kim SH, Sim SJ. Design and applications of photobioreactors-a review. Bioresour Technol, 2022;349:126858.
[58] Mahari WAW, Razali WAW, Waiho K, Wong KY, Foo SS, Kamaruzzan AS, Derek C JC, Ma NL, Chang JS, Dong CD. Light-emitting diodes (LEDs) for culturing microalgae and cyanobacteria. Chem Eng J, 2024:149619.
[59] Park SG, Rajesh P, Sim YU, Jadhav DA, Noori MT, Kim DH, Al-Qaradawi SY, Yang E, Jang JK, Chae KJ. Addressing scale-up challenges and enhancement in performance of hydrogen-producing microbial electrolysis cell through electrode modifications. Energy Rep, 2022;8:2726-2746.
[60] Lee HS, Xin W, Katakojwala R, Mohan SV, Tabish NM. Microbial electrolysis cells for the production of biohydrogen in dark fermentation–a review. Bioresour Technol, 2022;363:127934.
[61] Kadier A, Singh R, Song D, Ghanbari F, Zaidi NS, Aryanti PTP, Jadhav DA, Islam MA, Kalil MS, Nabgan W. A novel pico-hydro power (PHP)-Microbial electrolysis cell (MEC) coupled system for sustainable hydrogen production during palm oil mill effluent (POME) wastewater treatment. Int J Hydrogen Energy, 2023;48(55):21066-21087.

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