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Deployment of Fermentative Biohydrogen Production for Sustainable Economy in Indian Scenario: Practical and Policy Barriers With Recent Progresses

Vinayak V. Pathak[1,2] · Shamshad Ahmad[1] · Arya Pandey[1] · Vineet V. Tyagi[3] · D. Buddhi[4] · Richa Kothari[1]

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Abstract Depleting fossil fuel reserves and its significant contribution to greenhouse gas emission have created energy crisis and environmental degradation. Therefore, it is necessary to develop alternative fuels with a proper policy framework to support research and development. The potential biomass resource of India such as agricultural products, lignocellulosic waste biomass, industrial waste, and food processing waste has been extensively investigated by Indian researchers via fermentative biohydrogen production. The impact of key factors lowering fermentative biohydrogen yield can be reduced by the intervention of recent advancement in fermentative biohydrogen production such as combined fermentation process, optimized trace metal application, and pH control. A policy dealing with bioenergy promotion should adopt a market pull approach to promote bioenergy as a people-friendly technology. The present review provides recent advances in fermentative biohydrogen production process as well as practical and policy-related barriers in way of biohydrogen energy generation and promotion.

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✉ Richa Kothari
kothariricha21@gmail.com

1 Bioenergy and Wastewater Laboratory, Department of Environmental Science, Babasaheb Bhimrao Ambedkar University, Lucknow, UP, India

2 Department of Environmental Science, Alpine Institute of Management and Technology, Dehradun, UK, India

3 Department of Energy Management, Shri Mata Vaishno Devi University, Katra 182320, Jammu & Kashmir, India

4 Department of Renewable and Sustainable Energy Studies, Suresh Gyan Vihar University, Jaipur, Rajasthan, India

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Introduction

Major energy reforms around the globe have been stimulated due to fluctuation in energy prices, energy scarcity, and environmental pollution due to consumption of conventional fuels. Evidence from several scientific studies clearly reported changes in climatic variables due to global warming. The consequences of this global threat were well recognized; despite this fact, various developing countries such as India largely depend on fossil fuel to run its industrial and transportation sectors. The share of carbon dioxide in total greenhouse gas emissions is projected to increase by double from 14 % in 2000, and total emission would raise to 80 % by 2050 [1]. Thus, there is a need for adaptation of bioenergy under a sustainable economic approach, which is expected to provide a solution to the double challenge of environmental restoration and energy security. Explorations of such energy alternatives are the need in the present, which have the potential to meet the energy demand and supply gap. Hydrogen is the most abundant element in the universe that has a potential to serve as an excellent fuel due to its high heat of combustion (122 kJ/g) with no by-products of pollutant nature (Table 1) [2, 3]. Hence, hydrogen as an option among the other alternatives has emerged as a viable alternative, which has been well explored by recent studies.

Developing countries like India have implemented strategic policies to reduce the carbon emission under the national action plan on climate change. Studies have reported that cumulative emission of greenhouse gasses from developing countries contribute up to 75 %. Thus, significant reduction target could not be achieved without the effort of these fast-growing economies (http://www.cfr.org/climate-change/global-climate-change-regime/p21831). Therefore,

Table 1 Assessment of energy and carbon emission for commercially viable fuels [12, 13]

Fuel type	Energy/unit mass (MJ/kg)	Energy/vol (MJ/l)	Carbon emission (kg C/kg fuel)
Hydrogen (gas)	120	2	0
Hydrogen (liquid)	120	8.5	0
Coal (anthracite)	15–19	–	0.5
Natural gas	33–50	9	0.46
Diesel	42.8	35	0.9
Biodiesel	37	33	0.5
Ethanol	21	23	0.5

future vision for global bioenergy market would be heavily reliant on biohydrogen to provide a long-term sustainability for economic development as a future fuel with zero pollution. In this context, researchers across the globe have developed a keen interest in the development of hydrogen (H_2) energy. However, conventional hydrogen production methods such as steam methane reforming, auto-thermal reforming, partial oxidation, splitting of water were found as less economical as it involves high cost and energy input; thus, sustainable and cost-effective routes for H_2 production can be achieved through biological ways [4, 5] (Table 2). Hydrogen is an intermediate product during the process of anaerobic digestion, a process currently in practice at a technical level for methane generation [6–10]. On the other hand, biological hydrogen production processes are mostly operated at ambient temperatures and pressures, thus less energy intensive. These processes are not only environment-friendly, but they also lead to open a new avenue for the utilization of renewable energy resources which are inexhaustible [11, 12]. Hydrogen was identified as a potential alternative to fossil fuels among all other renewable energy sources due to its carbon neutral and high-energy value properties [12, 13].

Hydrogen production by biological means is advantageous since the energy requirement and investment cost is low. Biological hydrogen production involves fermentative hydrogen production by anaerobic bacteria and photobiological hydrogen production by photosynthetic bacteria, cyanobacteria, and green algae [14, 15]. Adaptation of hydrogen energy as a main stream of energy economy for developing country like India requires extensive research and development as well as demonstration projects with the development of appropriate storage technology. In this way, Indian government has taken serious initiatives to support research and development for the development of biohydrogen-based energy generation. The present review provides an overview of a different bioprocess for hydrogen production and their associated challenges with suggestive measures based on recent studies. Government efforts in the form of the policy framework and program initiatives are also reviewed to provide barriers and policy suggestion in a way to build up biohydrogen economy.

Bioprocesses for Hydrogen Gas Production

Biohydrogen can be produced by following various bioprocesses such as (1) biophotolysis of water, (2) dark-fermentative hydrogen production during the acidogenic phase of anaerobic digestion of organic matter, and (3) combined dark/photofermentative production of hydrogen. Some studies on microbial fermentative hydrogen production were also investigated under control cell metabolic pathway to enhance hydrogen-producing efficiency at a microbial molecular level [9–14, 16]. There are various technological factors (substrate composition, operating parameters, intermediate inhibitory products, metal inhibition, etc.) in the way of commercialization of fermentative biohydrogen production unique to each type of fermentative biohydrogen production processes.

Dark Fermentation

The dark fermentation process is characterized by degradation of an organic substrate by anaerobic bacteria in an environment with the absence of light and oxygen to produce biohydrogen, and by breakdown and conversion of complex organic compounds such as carbohydrate-rich materials. These organic polymers were initially hydrolyzed into sugar molecule, which undergoes a series of chemical reactions to produce biohydrogen [17]. The fate of the dark fermentation process for the quantity of hydrogen produced depends on the bacteria involve in the process and formation of acids. Theoretically, the dark fermentation process of 1 mol glucose yields 4 mol H_2 by acetate pathway and 2 mol H_2 through butyrate pathways, respectively [18]

$$C_6H_{12}O_6 + 2H_2O \rightarrow 2CH_3COOH + 2CO_2 + 4H_2 \quad (1)$$

$$C_6H_{12}O_6 + 2H_2O \rightarrow 2CH_3CH_2COOH + 2CO_2 + 2H_2 \quad (2)$$

$$\text{Pyruvate} + \text{CoA} + 2\,\text{Fd(ox)} \rightarrow \text{acetyl-CoA} + \text{Fd(red)} + CO_2 \quad (3)$$

$$2H^+ + \text{Fd(red)} \rightarrow H_2 + \text{Fd(ox)} \quad (4)$$

Researchers have used numerous microflora to carry out biohydrogen production using different feedstocks.

Table 2 Summarized remarks with advantages and disadvantages of hydrogen production processes

Processes	Advantages	Disadvantages	Remarks
Thermal processes			
Natural gas reforming	Most viable means of hydrogen production in the present scenario due to the presence of potential infrastructure support	Capital cost is high for the establishment	Improved catalyst efficiency and reduction in process cost are required; development of low cost, efficient; separation/ purification mechanism is also needed
Bio-derived liquid reforming	Futuristic fuel with existing infrastructure	High capital cost Feedstock quantity and quality parameters affect the process	Low-temperature and liquid-phase catalysts are needed. Characterization of biomass is required
Coal and biomass gasification	Low-cost fuel technology	Feedstock impurities with carbon content affect the system's efficiency. Capital cost is high	Feedstock storage, preparation, and handling are the major hurdles. Emissions control measures are also required
Thermochemical production routes	Clean and sustainable route for energy production using solar and nuclear energy in integration with chemicals	Long-term technology but durable and potential	Developments of thermochemical storage and heat transfer devices are needed
Electrolytic processes			
Water electrolysis (splitting water using electricity)	Pollution-free fuel cell device can be used for electricity generation	The solar system can provide a better efficient system, but high cost is involved	Improved photocatalyst with multifunctional materials at low cost to assure uniform quality production designs is required
Photolytic processes			
Photoelectrochemical hydrogen production route	Clean and sustainable technology with low-temperature requirements	Solar-based technologies require a high cost	Long-term technology for sustainable development
Biological processes			
Direct biophotolysis	An integrated approach for H_2 production using sunlight with water	Light intensity should be high, and O_2 acts as an inhibitor in the reaction	More R&D is needed for efficient functioning of microorganism for long-term development.
Indirect biophotolysis	Use of blue-green algae for hydrogen production from water	Uptake of hydrogenates is removed	A zero-cost method for the development of microbes using wastewater as a nutrient resource.
Photofermentation	Different ranges of light spectrum can be optimized to enhance the yield. A wide spectral energy can be used by photosynthetic bacteria	Nitrogenase enzymes get inhibited in the presence of a small amount of O_2. Light conversion efficiency is low	The development of an efficient bioreactor is required
Dark fermentation	In the light-independent process, several metabolites are produced as a by-product, and various substrates can be used	Relatively low H_2 yield; the process becomes thermodynamically unfavorable	
Two-stage fermentation	Relatively high H_2 yield; first-stage metabolites can be converted to H_2 and CH_4	Requires continues light source in the dark + photofermentation process and pH control in anaerobic digestion processes	

Compared to mixed microflora, higher hydrogen yield was observed with pure bacterial strains; however, this practice is not feasible for large-scale operations. In fact, the dark fermentation process involves a variety of bacterial populations for hydrolysis before the fermentation process. The estimated biohydrogen yield in the practical situation was lower than 4 mol/mol glucose.

Photofermentation

The photofermentation process shows the dependency of light to produce biohydrogen as it is mediated by some phototrophic bacteria (like *Rhodobacter sphaeroides*, *Rhodopseudomonas capsulate*, *Rhodopseudomonas palustris*, and *Rhodospirillum rubrum*) by utilizing the organic carbon of feedstock/substrates. These bacteria lack the enzymes like nitrogenase and hydrogenase to ferment the substrate into biohydrogen, carbon dioxide, and organic acids. Besides this, these bacteria lack photosystem II which helped in eliminating O_2 present in the system and maintained anaerobic conditions throughout the process [19].

$$C_6H_{12}O_6 + 12H_2 + \text{light energy} \rightarrow 2H_2O + 6CO_2 \quad (5)$$

$$(CH_2O)_2 \overset{NADPH}{\rightarrow} \text{ferredoxin} \rightarrow \text{nitrogenase} \rightarrow H_2 \quad (6)$$

Combined Dark and Photofermentation Processes

The combination of dark and photofermentation processes for biohydrogen production is termed as the combined process. According to the theoretical estimation, about 12 mol/mol glucose can be achieved by this integrated process. Various authors have studied the efficacy of the combined fermentation process such as the observed combined fermentation process using a transparent substrate, i.e., glucose [10]. The spent from the dark fermentation (DF) process was transformed by photofermentation (PF) using *R. sphaeroides* in a column photobioreactor for generating higher hydrogen yield. The effect of different variables such as the ratio of dark and photofermentation bacteria, light intensity, and light/dark cycle has also been studied in the combined fermentation process that is also investigated. Among different variables, pH control in the single-stage combined fermentation process is found highly useful for biohydrogen production. At optimum pH (7.5), the biohydrogen production was increased from 0.966 to 2.502 L H_2/L medium [20]. The combined fermentation process is also considered as a cost-efficient process due to the larger exploitation of stored chemical energy.

Bioelectrochemical Process

It is the advanced approach for biohydrogen production which involves microbial electrolysis cells (MECs). In this process, oxidation of complex organic compound is carried out by the bacteria present at an anode and produces protons, carbon dioxide, and electrons. Transportation of proton takes place through proton exchange membrane, and electrons are transported via an external circuit to the cathode. Upon supplement of 0.5 to 0.9 V, electrons combine with protons on the cathode and produce H_2 gas (http://www.batteryuniversity.com/parttwo-52.htm). Microbial fuel cell application for biohydrogen production is a promising approach that involves aerobic and anaerobic treatments using bacteria as a catalyst. A range of organic substrates such as domestic wastewater, animal wastewater, and wasted sludge can be utilized as a feedstock in this device [21, 22]. Recently, Zhang et al. [23] observed the impact of methane inhibitor on microbial electrolysis. Under repress condition of methane maximum hydrogen production, 8.4 ± 0.2 mol H_2/mol glucose was achieved with an applied voltage of 0.8 V. Researchers have observed a cyclic voltammetric test for anodic biofilm and analyzed that electron transfer in MECs is mainly due to biofilm-bound redox compounds.

Indian Scenario on Hydrogen: R&D and Roadmap

Currently, India is importing about 122 MT of oil per year and investing a significant share of its economy to support the transportation sector. An alternative like hydrogen energy can replace conventional fuel, and it would reduce the dependency from suppliers. The Ministry of New and Renewable Energy (MNRE) has been extensively supporting the research and development to promote hydrogen energy by operating an annual budget of US$100 million. Different types of hydrogen fuel cell of varying capacities have been developed and managed by Bharat Heavy Electricals Ltd. (BHEL), Tata Energy Research Institute (TERI), the Central Electro Chemical Research Institute (CECRI), Hydrogen Energy Center (HEC), Banaras Hindu University (BHU), and the Indian Institute of Science (IISc). Researchers have also been successful in the biological production of biohydrogen from organic effluents, and a large-scale bioreactor of 12.5 m^3 capacity was developed in Chennai [24]. Efforts are also underway to utilize significant amounts of hydrogen produced as a by-product in many industries such as the chloralkali industry.

India is the founder member of International Partnerships on Hydrogen Economy (IPHE) and implemented various demonstration programs for hydrogen in motorcycles and three wheelers, power-generating units, catalytic combustion, air conditioning, and biological production of hydrogen from organic wastes at the pilot plant scale and bagasse at the laboratory scale at global level [25]. A remarkable achievement by the MNRE was made in 2008–2009 by the establishment of compressed hydrogen natural gas distribution station in Dwarka, India. MNRE-supported programs such as (a) hydrogen production through the biological route, (b) hydrogen storage in hydrides and carbon materials, and (c) development and demonstration of hydrogen-fueled internal combustion engines for vehicles are the focused initiatives of the government in the direction of biohydrogen production. Research, development, and demonstration (RD&D) activities on different types of fuel cells (an electrochemical device that converts the chemical energy of hydrogen directly into electricity and heat without combustion) have also been supporting a broad-based RD&D program on different aspects of hydrogen energy technologies [26].

The MNRE had recognized the importance of hydrogen power source, and the National Hydrogen Energy Road Map (NHERM) was approved by the National Hydrogen Energy Board (NHEB) in 2006 [27]. The roadmap on hydrogen energy has different aspects of hydrogen energy, which include production, storage, transportation, applications, safety, standards and codes, capacity building, and awareness. The roadmap has highlighted hydrogen production as a critical area of focus in addition to the existing methods of hydrogen production based on steam methane reformation, production of hydrogen from coal gasification, nuclear energy, and biomass, biological, and renewable energy. In the area of hydrogen storage which includes gaseous, liquid, and solid-state storage, various goals concerning the efficiency of storage, useful cycle life, compactness, and cost to be achieved by 2020 have been identified in the roadmap. The hydrogen energy board has set a target of one million vehicles and 1000 MW of power-generating capacity based on hydrogen energy by 2020. The board has proposed two major initiatives: namely Green Initiative for Future Transport (GIFT) and Green Initiative for Power Generation (GIP). The renewable energy ministry of India has allocated Rs. 25,000 crore (US$6 billion) to promote the use of hydrogen in India by 2020 [27].

Practical and Policy-Related Barriers to Biohydrogen Production

Substrate Composition

Most of the studies have investigated biohydrogen production using simple sugar as a substrate; however, in recent years, various authors have used potential waste substrates for the generation of biohydrogen. The composition of substrate plays a significant role in biohydrogen yield such as that a substrate having high lipid content causes lipid hydrolysis, which is slower than carbohydrate hydrolysis. Hydrolysis of a lipid was inhibited by the accumulation of volatile fatty acid production that causes a decrease of the pH of the medium [28]. Thus, carbohydrate-rich biomass is preferred over such high lipid-containing biomass. In the process of carbohydrate hydrolysis, the hydrolytic bacteria produce simple sugars such as sucrose, glucose, xylose, and hexose, which are further consumed by the anaerobic bacteria to produce biohydrogen. On the other hand, lipid hydrolysis is performed by the lipase enzyme found in some bacteria. Lipid hydrolysis results in the generation of free fatty acids and glycerol that can be hydrolyzed to acetyl-CoA, acetate, and hydrogen form of NADH oxidation during the β-oxidation pathway. Biohydrogen production from proteins involves bacteria that convert proteins into polypeptides and amino acids by protease enzymes. Further, amino acids are broken down to volatile fatty acid, carbon dioxide, and hydrogen. However, there are very few studies that have been done on the use of proteinaceous substrates as biohydrogen production feedstock except by Sun et al. [29] who proposed the pathway for biohydrogen production from protein.

Operating Conditions

Operating parameters such as temperature and pH play a significant role in the fermentative biohydrogen production. Anaerobic bacteria are more sensitive to temperature than the other process variables, depending on temperature, fermentative bacterial ranges from ambient (15–30 °C), mesophilic (30–39 °C), thermophilic (50–64 °C), and hyperthermophilic (>64 °C). Change in temperature ranges profoundly affects the H_2 production rate in general and the consumption of substrate in the process, biohydrogen yield, formation of metabolites in the form of volatile fatty acids, and presence of microbes in the system [30]. Although several studies have been done for biohydrogen production with the variations in temperature, a mesophilic temperature is a more favorable condition in all other aspects of temperature due to its being less expensive as well as technically easy features. Studies revealed that thermophilic and hyperthermophilic bacterial cultures are more proficient in hydrogen production than mesophilic one. The highest yield reported by the thermopiles is 4 mol H_2/mol glucose which is very close to the theoretical yield [23]. In the case of agricultural biomass, it has been recently reported that mesophilic bacteria are unable to use cellulose directly for hydrogen production, and the addition of exogenous cellulase enzyme is required for bacterial hydrolysis. On the other hand, thermophilic anaerobic bacteria efficiently utilize cellulose without the addition of exogenous cellulase. In thermophilic condition, the hydrolysis rate of substrates is also high [31]. After an extensive literature survey, it has been found that among thermophiles, *Thermoanaerobacterium* sp. and, among mesophiles, *Clostridium* sp. and *Enterobacter* sp. are the most popular species of bacteria for hydrogen production.

The significant operating parameters affecting fermentative biohydrogen production is pH. It is a deciding factor for the acidic and alkaline conditions that limit the growth of bacteria and govern the concentration of the solvent in the system. Khanal et al. [32] reported that the optimum range of pH for the hydrogen production is 5.5–6.5, and at this pH, the maximum gas production and the least solvent production occur. Another significance of the pH is its effect on the activity of enzyme [Fe–Fe] hydrogenase as low pH affects this enzyme's activity and inhibits the hydrogen production [32]. Another reason behind the inhibition of hydrogen production at low pH is the presence of some protons generated by the breakdown of organic acids. These ions have the ability to enter in the cytoplasm of bacteria via cell membrane and inhibit their growth.

Reaction Intermediates

Volatile fatty acids (VFA) are produced in the form of different solvents in a fermentative hydrogen production process. The hydrolysis process provides most of the fatty acids in the acidogenic phase. These acids include acetic acids, propionic acids, isobutyric and butyric acids, lactic acids, and ethanol. Their concentration, distribution, and fractions can be used to monitor the fermentative hydrogen production system [33]. In an anaerobic treatment process, the drop in pH occurs due to either accumulation of volatile fatty acid or excessive generation of CO_2 or both. The identification of volatile fatty acids formed during the process gives valuable information about the type of metabolic pathway followed by the bacteria. The VFA generation in the fermentative hydrogen production process is also affected by change in temperature as at higher temperature (45 °C), the concentration of acetate and butyrate is higher (26–30 %) than the concentration of acetate, propionate, and butyrate (20–25 %) at mesophilic temperature (30–35 °C). Ethanol concentration is also important in the estimation of liquid metabolites, as high ethanol concentration fraction (23–40 %) was achieved at 30–45 °C, which reduces the hydrogen production. Ethanol production consumes electron and favors the propionate formation by directly utilizing H_2, which decreases the yield of H_2 [31, 33].

Limitation by Metals

Trace metal concentration effectively influences the metabolic pathway and enzymatic activity in fermentative bacteria. However, a higher concentration of these metal ions is also responsible for inhibition of the hydrogen production process involved in the dark fermentation process. A high concentration of metal reduces the availability of nutrients and causes the destruction of membrane function. The effect of Fe (iron) and Mg (magnesium) has been explored by several researchers because the presence of both is essential for hydrogenase enzyme. Hydrogenase, which is capable of catalyzing the oxidation of hydrogen or reduction of the proton, can be classified into [Ni–Fe] hydrogenase and [Fe–Fe] hydrogenase. [Ni–Fe] hydrogenase is widely distributed in bacteria, whereas [Fe–Fe] hydrogenase is restricted to some specific bacteria. This [Ni–Fe] hydrogenase is made up of two subunits (one small and one large) and contains one atom of Ni and 12 atoms of Fe/molecule and contains clusters of Fe–S [34]. In the hydrogenase-catalyzed hydrogen production process, electrons are transported through the intra-molecular electron transport chain to the active site where the proton is reduced and hydrogen is produced. Since both Ni and Fe are the fundamental elements of hydrogenase, the concentration of both metals may significantly influence the fermentative biohydrogen production process.

Policy Barriers

Promotion of bioenergy technologies in India follows a technology push approach rather than following a market pull approach, and it causes a lack of interaction between the developer and end users. A traditional innovation theory favors the demand pull approach in which stakeholder demand and economic benefits of the technology are critically evaluated. Thus, institutional framework of India should involve local community while promoting the bioenergy generation technologies. The high cost of biohydrogen energy technology is another key barrier in a way of biohydrogen energy promotion. Lack of financial assistance from the government such as no incentive mechanism in early-stage technology adaptation is another significant barrier in bioenergy technology (BET) promotion. Despite depreciation benefits provided by the MNRE, it has only a marginal impact on technology adaptation. Apart from the institutional barriers, policy framework dealing with BET is a key factor for the downfall of this technology. The pricing conventional fuels tend to favor its consumption as it is supported by huge subsidies; thus, the BET remains as a marginal resource.

Conclusion and Recommendations

The role of hydrogen energy is widely recognized as a sustainable fuel in the future. Investigations carried out in recent years have provided a promising perspective for biohydrogen production. Sustainability in the hydrogen production process can be achieved by following the biological pathway for hydrogen production, i.e., biohydrogen. In India, like other developing countries, biohydrogen production has been no more applicable for industrial level till now and most of the studies are focusing only pure substrate-based lab-scale biohydrogen production by using glucose, cellulose cellobiose, arabinose, starch, xylose, sucrose, and glycerol. However, the use of pure substrates for biohydrogen production is more expensive and not economically viable. Besides pure substrates, many other materials such as industrial wastewater, sludge, municipal solid waste, agriculture waste, and domestic wastewater were found as an economic and efficient substrate. Research and development activity supplemented with appropriate budgetary allocation is required to develop biohydrogen-based economy in India. Though the Indian government has initiated various policies and programs to develop biohydrogen energy, various policy-related barriers have caused failure to achieve the goal of that policy and program. Therefore, an intervention of recent innovations with appropriate policy framework to promote biohydrogen energy generation would be a potential future bioeconomy for India.

Compliance with Ethical Standards

Conflict of Interest Vinayak V. Pathak, Shamshad Ahmad, Arya Pandey, Vineet V. Tyagi, D. Buddhi, and Richa Kothari declare that they have no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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