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Review Article

A systematic literature review on the potential of marine microalgal biohydrogen and value-added products production: Pathways, strains, and future prospects

S Gupta[a], P Chelak[a], S Mishra[b], M Das[b], A Abhinav[d], R Gupta[a,c] & M K Jha*[,a,c]

[a]Centre for Energy and Environment, Dr B R Ambedkar National Institute of Technology, Jalandhar, Punjab – 144 008, India

[b]Biochemical conversion division, Sardar Swaran Singh National Institute of Bio-Energy, Kapurthala, Punjab – 144 603, India

[c]Department of Chemical Engineering, Dr B R Ambedkar National Institute of Technology, Jalandhar, Punjab – 144 008, India

[d]Thermochemical Conversion Division, Sardar Swaran Singh National Institute of Bio-Energy, Kapurthala, Punjab – 144603, India

*[E-mail address: jhamk@nitj.ac.in]

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Microalgal biohydrogen production from marine strains is a promising renewable energy source, leveraging high photosynthetic efficiency and CO_2 sequestration. In this review, a Systematic Literature Review (SLR) of 174 recent research articles (2023 – 2025) quantitatively and qualitatively evaluates marine microalgal biohydrogen production. The study reveals that among various pathways, dark fermentation with strains such as *Chlorella vulgaris* yields up to 190.9 mL H_2/g-VS (Volatile Solids) at pH 5.5 and 35 °C, closely paralleled by *Nannochloropsis oceanica* at 183.9 mL H_2/g-VS, while *Chlorella saccharophila* achieves 169.3 mL H_2/g-VS under similar conditions. Another fermentative pathway for biohydrogen production, photofermentation using *Rhodobacter* sp. produced 5.24 mL H_2/L/h at pH 7.0 and 25.6 °C. High-carbohydrate strains such as *Tetraselmis suecica* (31.72 – 32.06 %) and *Isochrysis galbana* (up to 32.05 %) exhibit superior fermentative potential, and process enhancements, such as Fe_3O_4/TiO_2 nanoparticle pretreatment, can increase hydrogen yields by up to 53 %. However, biophotolysis with *Tetraselmis subcordiformis* reaches 162 cm³ H_2/L, whereas microbial fuel cells achieve 9.8 mL H_2/L/day with *Scenedesmus* sp., showcasing the potential of different pathways in biohydrogen production. This review concludes that systematic pathway selection, hybrid intensification, and the exploitation of high-yield microalgal strains are essential for transitioning marine microalgal biohydrogen from promising research to practical, eco-friendly, and scalable energy solutions in the emerging green hydrogen economy.

[**Keywords**: Anaerobic fermentation, Bioenergy, Biorefinery, Metabolic engineering, Microalgal biotechnology, Photofermentation]

Introduction

Hydrogen, generated through biological processes, is considered biohydrogen. Biohydrogen as a fuel has a high energy density of 120 to 142 MJ/kg, providing additional benefits by promoting resource sustainability and creating new economic prospects[1]. Current hydrogen production methods mainly rely on fossil fuels, raising concerns about greenhouse gas emissions and environmental impact[2]. Earlier projections suggested that hydrogen could supply 10 % of the global energy market by 2025. However, as of 2025, hydrogen accounts for less than 1 % of the global energy supply, with the vast majority still produced from fossil fuels, and green hydrogen representing a very small fraction[3,4]. Currently, global hydrogen production exceeds 1 billion cubic meters per day, with steam methane reforming accounting for nearly half (48 %) of that production. Petroleum-based sources contribute approximately 30 % to hydrogen production, primarily through naphtha reforming processes[5]. Coal contributes about 18 % of hydrogen, primarily through coal gasification.

Although water splitting electrolysis is a smaller player at 3.9 %, this clean method using renewable electricity holds significant promise for the future[6]. Microalgal biohydrogen production has recently gained significant interest[7]. Marine microalgae are a diverse group of microorganisms that flourish in marine conditions with salinity levels of around 30 – 40 parts per trillion[8]. They possess high growth rates and carbon dioxide (CO_2) sequestration capabilities, and, hence, are a viable feedstock for biohydrogen production[9]. The carbohydrate content in these microorganisms is important for biohydrogen production because it serves as a substrate for fermentation reactions[10]. Microalgal biomass may be considered a substitute for fossil fuels due to its short doubling time and zero greenhouse gas emissions.

Microalgae offer several benefits over other biological sources, such as reduced energy requirements and no need for high-energy pretreatment methods, since they do not have lignin composition[11]. Fermentative processes require minimal pretreatment to expose the reducing sugars, since algal biomass comprises complex carbohydrates, proteins, and lipids, which cannot be used directly[12]. Hence, to hydrolyse the complex polymers into simple fermentable sugars, various pretreatment techniques, such as enzymatic hydrolysis, sonication, microwave irradiation, etc., are employed[13]. Apart from this, alkali pretreatment, often using sodium hydroxide, is preferred for its ability to delignify biomass, swell cellulose, reduce crystallinity, and increase the internal surface area of microalgal cells, thus improving carbohydrate availability for fermentation. This method causes less sugar degradation compared to acid treatments and is considered cost-effective. Optimal pretreatment conditions (pH 5.5 – 7.0, temperature 30 – 37 °C) improve hydrogenase activity, which is vital for biohydrogen production efficiency[14].

During the acetogenesis phase of fermentation, the facultative bacteria convert simple sugars into organic acids (propionic acid, acetic acid, and butyric acid) by secreting extracellular enzymes[15]. During this process, bacteria generate carbon dioxide (CO_2), hydrogen, and acetate[16]. Microalgae utilise 1.83 kg of carbon dioxide in photosynthesis to produce one kg of algal biomass[17]. This review discusses the potential of marine microalgae strains as feedstock for biohydrogen production and for the production of value-added products from residual biomass. A systematic literature review has been conducted to understand the topic in detail, particularly in the Indian region, and it shows that only 174 research articles have been published from India in this research area. This demands further research and potential applications in this field to harness the maximum efficiency of marine microalgal biomass for bioenergy and biorefinery.

Materials and Methodology

The Systematic Literature Review (SLR) was conducted using the Web of Science (WoS) database to identify appropriate research articles on marine microalgae and biohydrogen production published in India during 2023 – 2025. The search was conducted using the keywords "Marine microalgae" OR "Marine algae" AND "Biohydrogen" OR "Hydrogen" including publications from inception until March 20, 2025 (Fig. 1). The keyword was selected based on the most frequently used keywords in the area. The initial search returned 12,741 records, which were filtered by excluding 727 review articles and selecting only research articles, reducing the dataset to 12,014. Further, research articles from 2023 to 2025 led to additional exclusions of 10,082 documents, bringing the number down to 1,932 articles. Filtering for the engineering research category removed 762 articles, bringing the total to 1,170. Finally, all open-access articles were excluded, and the selection focused on publications from India, resulting in final dataset of 174 research articles that met the predefined inclusion and exclusion criteria for this systematic review. This step-by-step filtering ensured a targeted dataset for assessing recent advances in biohydrogen production from marine microalgae in the engineering field. The collected data were analysed

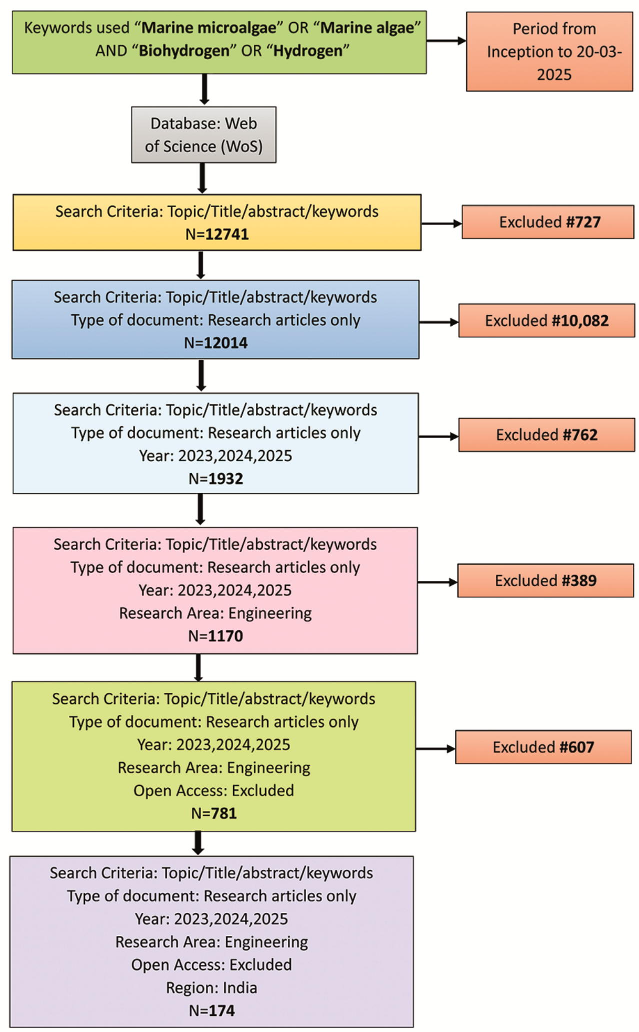

Fig. 1 — Methodology of Systematic Literature Review (SLR) representing search criteria, timeline, research area, region, inclusions and exclusions

using RStudio software, and the visualisations were prepared to depict the prevailing research trends (Fig. 2). Additionally, a word cloud of the most relevant keywords in these studies is shown in Figure 3.

Marine microalgal strain in biohydrogen production

Algae and cyanobacteria hold immense potential for sustainable hydrogen production. Over 70 species of algae and cyanobacteria across more than 30 genera have been explored for their potential in biohydrogen production. That is a significant number, demonstrating the widespread interest and diverse possibilities within these microorganisms[18]. Marine algae are found in the salty water of seas and oceans.

Isochrysis galbana, Chaetoceros calcitrans, *Tetraselmis suecica*, *Nannochloropsis oculata*, *Aphanocapsa* spp., *Dunaliella tertiolecta*, *Chlorella vulgaris*, *Synechococcus elongatus*, *Spirulina platensis*, *Scenedesmus* sp., *C. noctigama*, *C. pulsatilla*, *C. vectensis*, *Chlorella ovalis*, *C. pyrenoidosa*, *C. vulgaris*, *Chlorococcum* sp., *Pseudokirchneriella subcapitata*, *Cladophora glomerata*, *Rhizoclonium* sp., *Chlamydomonas* sp., *Anabaena* sp., *Chlamydomonas reinhardtii*, *Spirulina* sp., *Nostoc* sp., *Desmodesmus subspicatus*, *C. cf. obesa*, *Spirogyra*, *Clostridium beijerinckii*, *Ulothrix*, *C. rapa*, *C. Euryale*, *Clostridium pasteurianum*, and *Enterobacter* sp. are some of the documented marine microalgal strains that have the potential to be implemented for biohydrogen and other value-added products production because of their biochemical compositions (Fig. 4; Table 1)[8,19,20]. The major advantages and limitations of using marine microalgal biomass as feedstock for biohydrogen production are presented in Table 2.

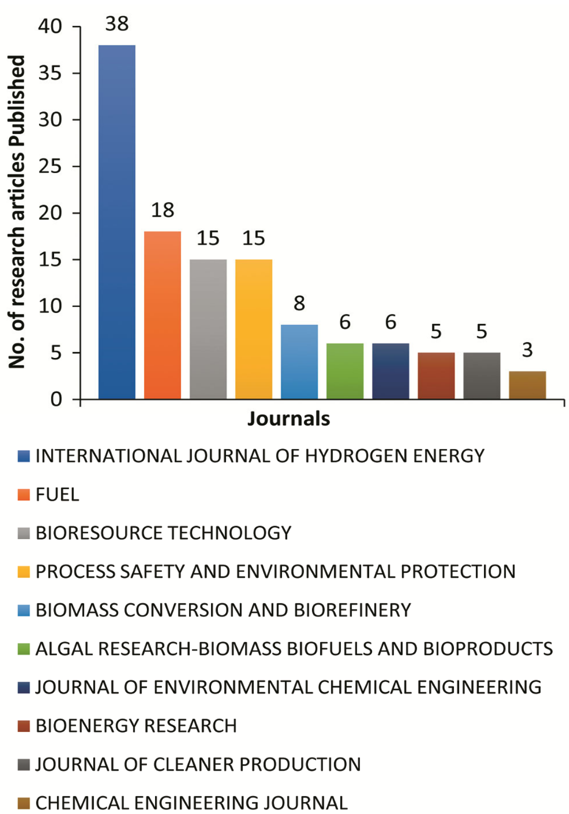

Fig. 2 — Distribution of published research articles on marine microalgal biohydrogen production and allied topics across leading scientific journals (2023 – 2025)

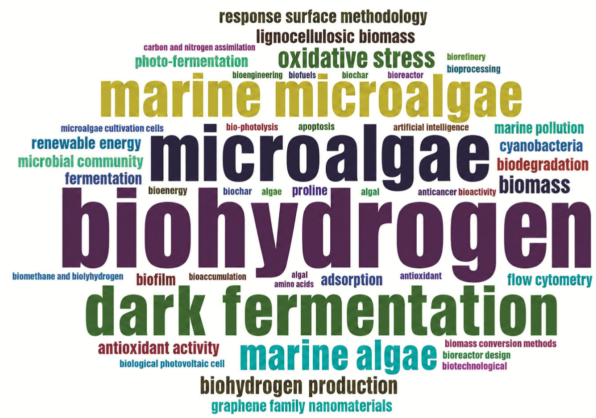

Fig. 3 — Word cloud illustrating the most frequently used keywords in research on marine microalgal biohydrogen production from 2023 to 2025

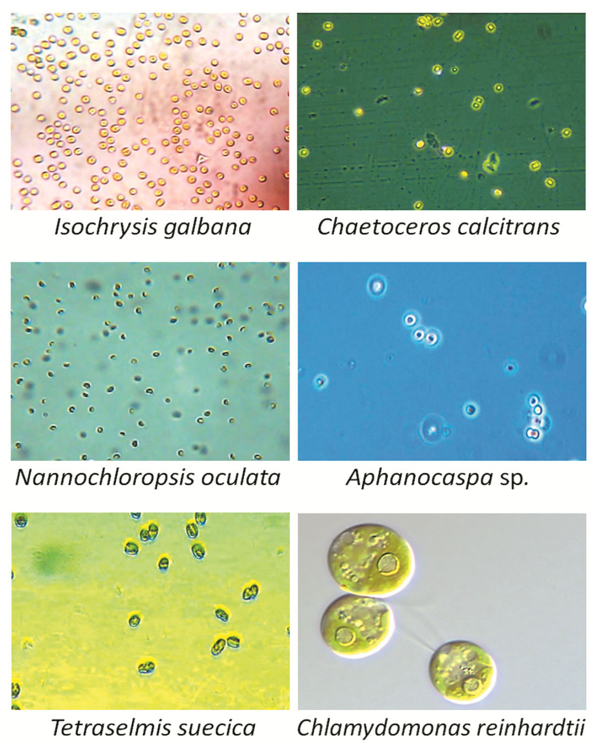

Fig. 4 — Potential marine microalgal strains in biohydrogen and value-added products production

Table 1 — Biochemical composition of different microalgal strains

Microalgal strain	Carbohydrate (%)	Protein (%)	Lipid (%)	Reference
Chlorella proteinucleus	21	50	10	Yu *et al.*[87]
Chlorella vulgaris	11	32	18	Rodrigues *et al.*[88]
Chlorella pyrenoidosa	14.87	57.30	NA	Cheng *et al.*[89]
S. obliquus UTEX 393	32.05±0.40	45.20±0.80	18.20±0.06	Singh *et al.*[90]
S. obliquus, *C. vulgaris*, *O. tenuis*	32.39	41.97	11.61-11.96	Zhang *et al*[91]
Chlorella and *Phormidium*	28	33	43	Choudhary *et al.*[92]
Scenedesmus sp., *Chlorella* sp., or *Oscillatoria tenuis*	20.29-25.93	30.35-36.73	20.10-23.20	Zhang *et al.*[93]
N. granulata	10.89 ± 0.11	32.7	57.33 ± 0.09	Dong *et al.*[94]

Table 2 — Advantages and disadvantages of marine microalgal feedstock for biohydrogen production

Advantages of microalgal biomass as feedstock	Disadvantages of microalgal biomass as feedstock
High photosynthesis rate, and higher growth rate compared to plants (10 – 50 times).	Requires temperature maintenance (25 – 30 °C)
CO_2 sequestration	Microbial contamination
Cultivation on non-arable land	Cultivation requires high nutrients and water
High tolerance for SOx and NOx	Energy intensive drying and harvesting process
High carbohydrate content in algal biomass that supports fermentation and increases H_2 production	Low biomass concentration that makes harvesting costly
Lacks lignin content and requires milder pretreatment to release fermentable sugar	Low biomass yield and productivity
Algal biorefinery	Energy-intensive cultivation

These strains exhibit significant variations in their biochemical profiles, which directly influence their potential for hydrogen production and other biotechnological applications[21]. The protein content varies across these strains, with *Isochrysis galbana* containing 16.25 – 22.70 %, *Chaetoceros calcitrans* 20.86 – 26.86 %, *Tetraselmis suecica* 8.9 – 13.61 %, *Nannochloropsis oculata* 18.14 – 22.68 %, and *Aphanocapsa* sp. 8.15 – 21.42 %, depending on the culture conditions[22]. Lipid accumulation is also a crucial factor for biofuel applications, with *N. oculata* exhibiting the highest lipid content (24 – 32 %) and *C. calcitrans* the lowest (2 – 9.22 %) under different salinity and media conditions. Carbohydrate content, which is essential for fermentation-based hydrogen production, is highest in *T. suecica* (31.72 – 32.06 %) and ranges from 29.15 – 32.05 % in *I. galbana*, 16.44 – 23.81 % in *C. calcitrans*, and 29.62 – 31.56 % in *Aphanocapsa* sp.[23].

Fatty acid profiles of these microalgae also complement their implementation for biohydrogen and biofuel production. Saturated fatty acids like palmitic acid (C16:0) and stearic acid (C18:0) are found in different concentrations, with *N. oculata* having the highest content of palmitic acid (6.2 mg/g). Monounsaturated and polyunsaturated fatty acids like oleic acid (C18:1) and linoleic acid (C18:2) in these strains make them more suitable for bioenergy applications[24]. Microalgal hydrogen production pathways primarily depend on nitrogenase and hydrogenase enzymes that operate under anaerobic and light-dependent conditions. Growth parameter optimisation, such as salinity, light intensity, and nutrient availability, adversely affects biomass accumulation and hydrogen production[25]. Different growth media, such as Conway and f/2, which vary in nutrient composition and salinity levels, have been shown to influence the biochemical composition of marine microalgal strains. These media affect the accumulation of key biomolecules, such as proteins, lipids, and carbohydrates, thereby impacting the overall metabolic profile and productivity of the microalgae[26].

Apart from biohydrogen, these microalgal strains also have value-added uses in bioplastics and pharmaceuticals because of their bioactive compounds. *Tetraselmis suecica*, for example, is very effective as an antibacterial against *Vibrio harveyi* and *Staphylococcus aureus*, with a maximum inhibition zone of 20 mm[27]. In addition, the presence of essential minerals like sodium, potassium, calcium, and phosphorus in these microalgae makes them a good candidate for nutritional and nutraceutical purposes, including the synthesis of essential amino acids, dietary fibre, and omega-3 and omega-6 fatty acids[23]. The integration of these marine microalgae into bioreactor systems for large-scale biohydrogen production is also an active area of research to

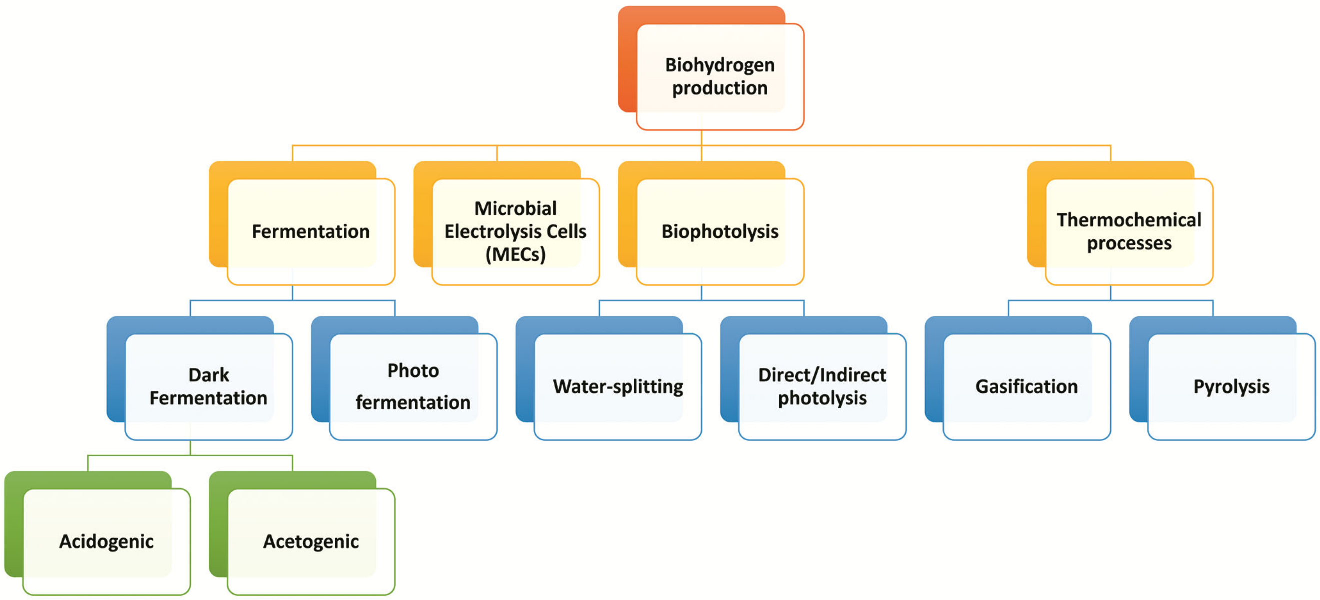

Fig. 5 — Overview of major biohydrogen production pathways and processes

optimise hydrogen yield through metabolic and genetic engineering strategies[28]. Techno-economic studies identify cost-efficient and scaled-up cultivation technologies to commercialise microalgal biohydrogen production[1,29–31].

Among all *Chlorella* species, such as *Chlorella vulgaris*, having a combination of starch and cellulose, makes them rich in carbohydrates, characterised by fast growth rates and effective biohydrogen production, with yields of 6.1 to 31.2 mL H_2/g microalgae[32]. Apart from *Chlorella*, *Chlamydomonas reinhardtii* is well documented for biohydrogen production. This strain is rich in starch, which can be used to produce fermentable sugars for biohydrogen production[33]. *Synechocystis* sp. PCC 6803, a cyanobacterium with high glycogen content, employs both photobiological and fermentative pathways for hydrogen production, providing flexibility and yielding higher H_2[ref. 34]. Further studies are required to identify and optimise algal strains for enhanced hydrogen production potential.

Pathways of biohydrogen production

Different biohydrogen production pathways, including thermochemical, photolysis, and biological pathways, are shown in Figure 5.

Thermochemical pathways

Thermochemical pathways of microalgal biohydrogen production include gasification, pyrolysis, combustion, and liquefaction. Hydrogen is generally produced by hydrocarbon reforming or non-reforming processes like electrolysis and gasification[35]. However, these techniques have many disadvantages, including the heavy reliance on fossil fuels, excessive power demand, and low heat efficiency. Thermochemical biohydrogen production utilises biomass feedstocks such as agricultural waste, forest residues, and wood biomass. Valizadeh & Jang[36] demonstrated biohydrogen production from furniture waste using Ni-loaded ultra-stable Y-type zeolites. Similarly, Pocha *et al.*[37] summarised the potential of various feedstocks for hydrogen production via gasification. In contrast, Arregi *et al.*[38] reported and emphasised that fast pyrolysis in combination with steam reforming is an efficient technique for biohydrogen production using agroforestry biomass wastes. However, Nguyen *et al.*[39] studied hydrogen production from biomass waste via pyrolysis and gasification processes. Further, in their study, the techniques offer flexibility, as the bio-oil and biochar produced can undergo steam reforming to increase hydrogen yield.

Biological pathways

Biological pathways of hydrogen production include biophotolysis (direct/indirect), fermentation (photo/dark; Fig. 6), and anaerobic digestion[33]. Biological techniques are more ecologically benign but have a lower productivity rate than physicochemical approaches. Utilising fermentation for hydrogen production from algal biomass is a

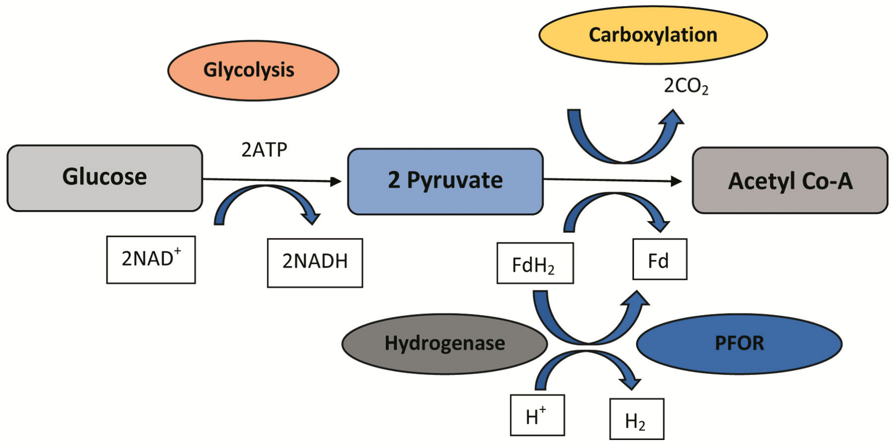

Fig. 6 — Schematic representation of the metabolic pathway of glucose conversion to hydrogen in microalgae during fermentation. NAD: Nicotinamide Adenine Dinucleotide, NADH: Nicotinamide Adenine Dinucleotide (Reduced Form), ATP: Adenosine Triphosphate, PFOR: Pyruvate: Ferredoxin Oxidoreductase, Fd: Ferredoxin, FdH: Ferredoxin Hydrogenase, (H^+): Protons, and (H_2): Molecular hydrogen

well-established technique with potential for scalability[40]. While promising, fermentative hydrogen production from algae still faces challenges such as cost, process optimisation, long-term stability, a lack of efficient microalgal strains with high sugar content, and the inability to protect fermentative conversions from oxygen exposure (Table 3)[41]. Some of the documented solutions to the existing problems include designing optimised systems for efficient algae growth and sunlight utilisation, modifying algae strains to improve hydrogen production or enhance enzymatic robustness, and developing more advanced pretreatment methods to break down the cell wall and facilitate biofuel extraction and conversion. However, significant challenges that hinder the commercial-scale utilisation of biohydrogen production include cost, negative energy balance, low hydrogen yield, and a lack of proper storage facilities[26].

Photofermentation

Hydrogen production in photofermentation primarily occurs via enzyme pathways, such as Pyruvate: Ferredoxin Oxidoreductase (PFOR) and hydrogenases. Nitrogenase, conversely, is the enzyme responsible for nitrogen fixation, converting atmospheric nitrogen (N_2) into ammonia (NH_3)[44]. Nitrogenase activity in hydrogen production is subject to feedback inhibition by various factors, including the presence of ammonia, its product. Some nitrogen-fixing bacteria also produce hydrogen under specific conditions, like iron deficiency or nitrogen limitation[43]. Research in photofermentative biohydrogen production from microalgal biomass is advancing, and significant progress is being made in optimising cultivation, pretreatment, and bioconversion processes. Zhang *et al.*[42] upregulated nitrogenase and hydrogenase enzymes by genetic modification to increase biohydrogen production in microalgal cells. Mechanism of biohydrogen production has been explained in Figure 6. The metabolic pathway of glucose conversion into hydrogen (H_2) has been explained through microbial fermentation, highlighting key enzymatic steps involved in biohydrogen production. The process begins with glycolysis, where glucose is broken down into two molecules of pyruvate, yielding 2 ATP and 2 NADH molecules[45]. Pyruvate then serves as a crucial intermediate and can follow two primary fates. In one pathway, pyruvate undergoes oxidative decarboxylation, catalysed by the enzyme PFOR, to form acetyl-CoA, releasing carbon dioxide (CO_2) in the process. During this reaction, pyruvate is oxidized to acetyl-CoA by pyruvate:ferredoxin oxidoreductase (PFOR), releasing CO_2 and transferring the released electrons to oxidized ferredoxin (Fdox), thereby reducing it to reduced ferredoxin (Fdred)[18]. The reduced ferredoxin subsequently donates electrons to hydrogenase, an enzyme that uses protons (H^+) to produce molecular hydrogen (H_2). This series of reactions forms the basis for biological hydrogen production, using pyruvate as a central metabolite and involving electron transfer via ferredoxin and hydrogenase[46]. Besides genetic modification, different pretreatment processes also enhance overall biohydrogen production. Singh *et al.*[47] studied different pretreatment techniques to break microalgal cell walls and enhance substrate digestibility. Apart from this approach, Masihi *et al.*[48] reported the incorporation of nanoparticles such as Fe_3O_4 and TiO_2 into hybrid dark and photofermentation systems. Nanoparticles enhance the fermentation reactor's light utilisation and microbial activity by acting as photocatalysts[48].

Dark fermentation

Dark fermentative biohydrogen production does not require light and is thus a less energy-consuming process. In most cases, *Clostridium* sp. have been identified in this process because the process yields high hydrogen content and the organisms possess versatile metabolism. A dark fermentation setup using *Clostridium*/*Enterobacter* as the inoculum is shown in Figure 7. According to Gupta *et al.*[49], 9.39 mol H_2/mol sucrose consumed has been produced under optimal conditions. Several factors, such as microbial diversity, substrate type, and operational parameters like pH and temperature, tend to influence the effectiveness of dark fermentation. To enhance hydrogenase activity, mesophilic conditions (30 – 37 °C) and a pH range of 5.5 to 6.5 must be maintained, which may further enhance biohydrogen yield[50]. Additionally, a recent advancement suggests the incorporation of nanoparticles to enhance microbial activity and fermentation efficiency (Tables 3 & 4). Pérez-Barragán *et al.* stated 53 % enhancement of hydrogen production from rice straw hydrolysate utilising iron-based nanoparticles (FeNPs) by improving the metalloenzyme activity[51]. Relatedly, hydrogen productivity has been reported to be improved by 10 % by utilising iron-based nanoparticles in lactate-driven dark fermentation[52]. An alternative approach, as demonstrated by Navarro-Díaz *et al.*[53], is the optimisation of microbial consortia to improve microbial diversity in dark fermentation bioreactors. Two major classes of microbes, namely Obligate anaerobes (such as *Clostridium* and *Desulfovibrio*) and facultative anaerobes (such as *Citrobacter*, *Enterobacter*, *Klebsiella*, and *Bacillus*),

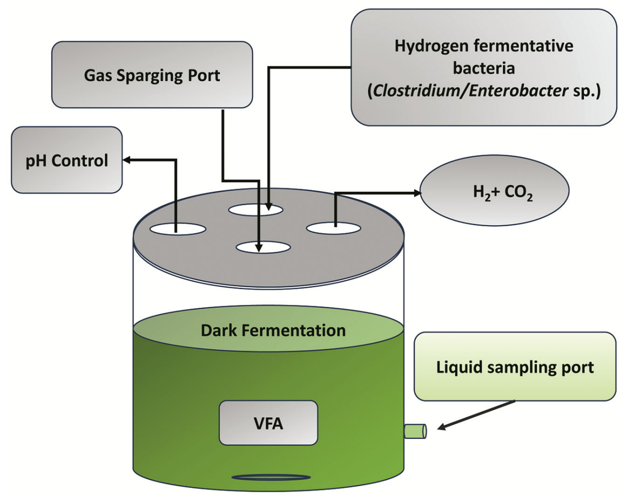

Fig. 7 — Dark fermentation bioreactor setup for hydrogen (H_2) and Volatile Fatty Acids (VFA) production

Table 3 — Microalgal strains in fermentative biohydrogen production

Hydrogen production method	Algal strain	Wastewater/ Media	Hydrogen Fermenting Bacteria (HFB)	pH/ Temperature (ºC)	Hydrogen yield (mL/g TVS)	References
Dark fermentation	*Nannochloropsis oceanica*	Seawater sparged with flue gas	*Clostridium butyricum*, *Rhodopseudomonas palustris*, *Methanosarcina* and *Methanothrix*	pH: 6.5± 0.1 Temp: 35	183.9	Xia *et al.*[66]
Photo fermentation	-	Sugar plant wastewater	*Rhodobacter* sp. KKU-PS1	pH: 7.0 Temp: 25.6	5.24	Assawamongkholsiri *et al.*[81]
Dark fermentation	*Chlorella* sp.	Pretreated slurries	*Clostridium beijerinckii*, *C. acetobutylicum*, *C. butyricum* and *C. roseum*	pH: 6.0 Temp: 35 ± 3	47.2	Phanduang *et al.*[50]
Dark fermentation	*Microcystis wesenbergii* and *Microcystis aeruginosaare*	Algal bloom suspension	*Clostridium butyricum*	pH: 6.0 ± 0.1 Temp: 35	24.96	Cheng *et al.*[82]
Dark fermentation	*Chlorella vulgaris*	Digested sludge	Mixed culture from digested sludge	pH: 5.5 Temp: 35	190.90	Zhang *et al.*[10]
Dark fermentation	*Chlorella saccharophila* FACHB-4	Modified BG11 medium	Anaerobic activate sludge	pH: 7 Temp: 35	169.30	Mu *et al.*[83]

*TVS- Total volatile solids

Table 4 — Factors affecting biohydrogen yield

Parameter	Description	Effect on biohydrogen production	Ref
Microalgal species & genetic engineering	Genetic, metabolic capabilities, Modified electron flow, and enzyme expression in algae	Optimises enzyme (hydrogenase) activity, Improves hydrogen yield or O_2 tolerance	Zhang *et al.*[84]
Light intensity & type	Solar/artificial, wavelength, illumination period	Drives photosynthesis and H_2 evolution, excessive light may inhibit	Mustafi *et al.*[85]
Temperature	Cultivation/control in bioreactor	Affects enzyme activity and metabolic rates	Com *et al.*[43]
pH Level	Culture medium pH	Optimizes enzyme (hydrogenase) activity	Ananthi *et al.*[24]
Salinity	Salt concentration in medium	Marine species are tolerant; affects growth, hydrogenase	Krishna *et al.*[40]
Nutrient availability	Nitrogen, phosphorus, micronutrients	Supports biomass growth; excess N may lower H_2 yield	Nazarpour *et al.*[86]
Substrate concentration	Sugars, starch, cellulose for fermentation	Drives fermentative hydrogen production	Sambusiti *et al.*[15]
Reactor design	Material, shape, mixing, O_2 removal, surface area	Influences O_2 inhibition, gas exchange, light use	Alkasrawi *et al.*[26]
Algal density & culture phase	Biomass concentration in culture, Stationary *vs* exponential phase	Impacts yield: up to threshold, increasing density = greater H_2 yield, maximum hydrogen yield at early stationary phase; declines with age	Dudek *et al.*[60]
Pretreatment methods	Physical, chemical, enzymatic substrate treatment	Increases substrate accessibility and H_2 yield	Pathy *et al.*[41]

have shown significant results in hydrogen production through dark fermentation[54]**.** The process, remarkably, is more economical than light-dependent photo fermentation and represent a promising route for renewable energy production[55].

Biophotolysis

Biophotolysis utilises photosynthetic pathways of microalgae and cyanobacteria to produce biohydrogen and is a light-dependent biological process. The system harnesses solar energy to split water molecules, ultimately releasing hydrogen gas as a byproduct, and is categorised into direct and indirect biophotolysis[56]. In direct biophotolysis, photosystem-driven electron transfer to hydrogenase enzymes catalyses the reduction of protons to H_2. However, oxygen inhibits the activity of the hydrogenase enzyme; thus, this process is oxygen sensitive[57]. Conversely, indirect biophotolysis production of hydrogen occurs in anaerobic conditions, overcoming this limitation by temporarily storing energy in carbohydrate intermediates[58].

Recent advancements have focused on optimising algal growth by improving hydrogenase activity, particularly regulating oxygen levels[59]. Also, the coupling of biophotolysis with the wastewater treatment process was demonstrated in a study on *Tetraselmis subcordiformis*, which reported a biohydrogen yield of up to 162 cm^3 H_2/L of culture[60]. Engineering approaches, including genetic modifications and semi-synthetic hydrogenases, have also been explored to improve enzyme stability and enhance hydrogen production rates[46]. Despite these developments, large-scale applications remain challenging due to low hydrogen yields and the need for advanced photobioreactor designs. Continued research into optimising biophotolysis efficiency and integrating it with hybrid biohydrogen production systems could facilitate its commercialisation.

Microbial Fuel Cells (MFCs)

Microalgae-based Microbial Fuel Cells (mMFCs) are considered a novel bioelectrochemical approach for hydrogen production, combining oxygenation-dependent photosynthesis with microbial electrogenesis. Here, microalgae serve as biocathodes, providing oxygen for cathodic reactions and sequestering CO_2. According to Mansoor *et al.*[61] improved hydrogen generation by mMFCs-MEC (Microbial Electrolysis Cell) integration has been investigated, with a biohydrogen yield of 9.8 mL/L/day utilising *Scenedesmus* sp. during wastewater treatment. Zieliński *et al.*[62] noted that *Tetraselmis subcordiformis* produced a significant amount of hydrogen in isolated cultures using microbial fuel cell effluents, further suggesting the possible use of wastewater from mMFCs in biohydrogen fermentation. Improved electron transfer and hydrogen yields have also been made possible by recent advancements in electrode material optimisation.

Artificial neural network modelling was used by Sayed *et al.*[95] to improve mMFC performance, resulting in increased hydrogen generation and bioelectricity. Additionally, Mahmoud *et al.*[63]

designed a biocathode-assisted mMFC in which the synthesis of microalgal biomass, enhanced by light, improved the energy and waste treatment transformation efficiencies. Research is still needed to overcome obstacles such as poor hydrogen recovery, restricted hydrogenase activity, and oxygen inhibition at the cathode[64]. To improve the scaling strategies of microalgae-based hydrogen production systems, future research should focus on developing oxygen-tolerat hydrogenases, optimising reactive configurations, and integrating mMFCs with renewable energy sources.

Marine microalgal strains in value-added product production

Marine microalgal strains, due to their high biochemical diversity, represent a valuable resource for the production of high-value bioproducts that have applications in nutrition, pharmaceuticals, biofuels, and cosmetics[65]. *Nannochloropsis* sp. accumulates 35 – 50 % dry weight as lipids, of which 30 – 50 % is Eicosapentaenoic Acid (EPA), a principal omega-3 fatty acid involved in cardiovascular health, brain function, and anti-inflammatory activity[66]. Similarly, *Isochrysis galbana* accumulates up to 25 % of DHA in total lipids and is a primary source of Docosahexaenoic Acid (DHA), which plays a primary role in neural development and helps prevent the risk of chronic diseases[67]. Some marine diatoms have been found to accumulate lipid content of over 60 % dry weight, making them prime candidates for biodiesel production due to their possible high energy density and sustainability compared to existing fossil fuels[68].

In addition, marine microalgae are an unlimited source of carotenoids, and *Dunaliella salina* can yield up to 14 % of its dry weight as β-carotene, which is mainly used in the nutraceutical and cosmetic industries owing to its potent antioxidant and provitamin A activity[69]. Other microalgae, *e.g.*, *Phaeodactylum tricornutum* and *Isochrysis galbana*, have 1.5 – 2.0 % fucoxanthin, a specific xanthophyll carotenoid that is rich in anti-obesity, anti-inflammatory, and anticancer activities, thereby expanding its use in functional foods and pharmaceuticals[70]. Besides their use in food and health, marine microalgae also possess immense potential for renewable energy production[71]. Among the most promising approaches is biohydrogen production, for which *Chlamydomonas reinhardtii* has been shown to produce up to 400 mL H_2/L of culture under sulfur-starved, light-stimulated conditions[72].

Hydrogen is a green, renewable energy carrier, and biohydrogen production by microalgae is an ecofriendly alternative to hydrogen from fossil fuels[73]. Marine microalgae are also effective producers of sulfated polysaccharides, which have immense applications in medicine and biotechnology[74]. *Porphyridium* species produce sulfated polysaccharides that make up to 25 % dry weight and are reported to possess antiviral, anticoagulant, and immunomodulatory activities of great value for the development of novel therapeutics[75]. Exopolysaccharides from *Spirulina* and *Chlorella* have also been reported to be prebiotic, enhancing gut health and immune function[76].

Marine microalgae have also been found to have sufficient protein content and can serve as an alternative source of protein in foods intended for human use, as well as for animal food[77]. *Spirulina* and *Chlorella* sp. contain 60 – 70 % dry biomass protein content, which is relatively similar to that of conventional protein-rich crops such as soybean[78]. These proteins are also rich in essential amino acids and bioactive peptides with antioxidant, antihypertensive, and antimicrobial activities, contributing to their value in functional foods and pharmaceuticals[79]. Marine microalgae also synthesise new compounds, such as Mycosporine-like Amino Acids (MAAs), which have natural UV-protective functions and are therefore highly sought after in cosmeceuticals and dermatological products to prevent oxidative stress and DNA damage caused by UV radiation[80].

Conclusion

Marine microalgae for biohydrogen production is a sustainable solution with the potential to transform the current bioenergy landscape. The combination of metabolic engineering, upgraded pretreatment methodologies, and advanced reactor designs is being used to further maximise its efficiency. Among the various principal pathways, dark fermentation in carbohydrate-rich strains like *Chlorella vulgaris* and *Nannochloropsis oceanica*, *Clamadomonas* sp. has been reported to produce the highest hydrogen yields when optimised for pH, temperature, etc.; whereas photofermentation, biophotolysis, and microbial electrolysis cells expand the range of sustainable technologies. While marine microalgae offer high productivity, CO_2 sequestration, and renewable biomass, their commercial viability is hindered by low hydrogen production, high culture costs, and inefficient procedures. Biohydrogen production methods have advanced via hybrids, optimised reactor designs, and genetic engineering. Future research should primarily concentrate on enhancing strain efficiency, process economics, and coupling with technologies that connect biohydrogen to carbon capture and wastewater treatment. With further development, microalgal biohydrogen generation might be a major trend-setter in the transition to a hydrogen-based bioeconomy.

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Conflict of Interest

All authors have approved the manuscript and declare that they have no conflicts of interest.

Author Contributions

SG: Conceptualization, methodology, investigation, formal analysis, writing - original draft, writing review & editing. SM: Conceptualization, writing - original draft, writing, review & editing. PC, MD, AA & RG: Writing review & editing. MKJ: Supervision and validation.

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