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Review

Cyanobacterial hydrogen production – A step towards clean environment

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ABSTRACT

Environmental pollution and exhaustive depletion of non-renewable energy sources demand the exploration of alternate energy sources. Hydrogen has been crowned as future fuel by virtue of its immense potential. Many microorganisms mediate hydrogen production. Cyanobacteria are excellent biological means of hydrogen production. This review highlights the significant progress achieved in cyanobacterial hydrogen production methods. The role of key enzymes catalyzing hydrogen production and the various parameters influencing the path of increased hydrogen productivity has been discussed.

Recent approaches towards enhanced hydrogen production like genetic engineering, alteration in nutrient and growth conditions, entrapment in reverse micelles, combined culture and metabolic engineering have been emphasized. Improvisation in hydrogen production methods mediated by microbes will pave the path for commercialization of molecular hydrogen as environmental friendly energy source.

1. Introduction

Exploration of alternate energy sources is the need of today, considering the environmental pollution and exhaustive depletion of non-renewable energy sources. Hydrogen, the most abundant element in the universe has the potential to serve the purpose of fuel and is ecofriendly. Hydrogen can serve as an excellent fuel because it is renewable, does not pollute the environment by evolving carbon dioxide, liberates large amounts of energy per unit weight in combustion (122 kJ/g) and is easily converted to electricity by fuel cells [1,2]. Numerous virtues of hydrogen have crowned it as future fuel. The photoconversion of water to hydrogen is an excellent solution. Three ways to achieve photoconversion of water to hydrogen are:

(a) The use of photochemical fuel cells,
(b) By applying photovoltaics, or
(c) By photosynthetic microorganisms [3–5].

The production of hydrogen is ubiquitous, natural phenomenon under anoxic or anaerobic conditions. The capacity of certain microorganisms to metabolize molecular hydrogen was discovered at the end of the 19th century [6] and later identified to be catalyzed by a hydrogenase [7]. Microorganisms are capable of producing hydrogen via either fermentation [8,9] or photosynthesis [10–12]. The biological species involved in hydrogen production are green algae, heterocystous and non-heterocystous cyanobacteria, photosynthetic bacteria and fermentative bacteria [13]. Hydrogen production by biological means is advantageous since the

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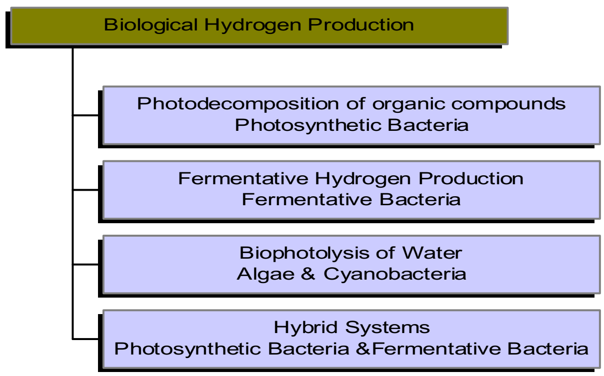
energy requirement and investment cost is low. Biological hydrogen production involves fermentative hydrogen production by anaerobic bacteria and photobiological hydrogen production by photosynthetic bacteria, cyanobacteria and green algae. This review highlights the advancements achieved in enhancing cyanobacteria mediated hydrogen production and the various strategic approaches.



1.1. *Hydrogen production by cyanobacteria*

Cyanobacteria are unique prokaryotes with diverse range of properties. They are potential microbial species for hydrogen production via direct and indirect photolysis [14]. They are ideal microbes for photobiological H_2 production, since they require the simplest nutritional conditions. For example, even the nitrogen and carbon dioxide available in open air is sufficient for their growth while they can use water as reductant and a source of electrons. Moreover, simple mineral salts present in natural water along with sunlight can act as source of energy for them.

Hydrogen production has been studied in a very wide variety of cyanobacterial species and strains. Hydrogen production occurs within at least 14 genera of cyanobacteria, under a vast range of culture conditions [14]. In addition to the advantages of cyanobacterial hydrogen production there are some obstacles namely inhibition of enzymes directly involved in hydrogen production by oxygen, H_2 consumption by an uptake hydrogenase, and an overall low productivity [15]. Cyanobacteria have some unique strategies to overcome some obstacles in the path of hydrogen production.

Cyanobacterial species capable of hydrogen production are categorized into three groups – heterocystous, non-heterocystous and marine (Table 1). Heterocystous cyanobacteria are specialized for nitrogen fixation by possessing unique structures called heterocysts. The heterocyst provides a micro-anaerobic environment suitable for the functioning of nitrogenase since it lacks photosystem II activity, it has a higher rate of respiratory oxygen consumption, and it is surrounded by a thick envelope that limits the diffusion of oxygen through the cell wall [16,17]. In non-Heterocystous cyanobacteria temporal separation between photosynthetic oxygen evolution and nitrogen fixation seems to be the most common strategy adopted by non-heterocystous cyanobacteria.

Cyanobacteria have unique feature of oxygenic photosynthesis. Oxygenic photosynthetic cyanobacteria normally absorb sunlight and store the energy in the form of polysaccharide glycogen and these storage biomolecules are mobilized, as required, to produce the energy needed to drive microbial metabolism. The photosynthetic apparatus is represented by isolated and freely lying photosynthetic lamellae. Chlorophyll *a* and several accessory pigments (phycoerythrin and phycocyanin) are embedded in these photosynthetic lamellae. The major photosynthetic pigment in cyanobacteria is Chlorophyll *a*. They use water as electron donor and oxygen is evolved as by-product. Carbon dioxide is reduced to carbohydrate through Calvin cycle. However, under certain conditions, electrons on the reducing side of PSI can be diverted at the level of ferredoxin (Fd) to another pathway (Fig. 1), employing an [FeFe]-hydrogenase (H_2ase) to evolve H_2 gas. During photosynthetic H_2 production, two protons and two electrons are recombined in a reaction catalyzed by the hydrogenase enzyme to yield one H_2 molecule. The rate of hydrogen production varies from species to species within cyanobacterial strains (Table 2).

2. Role of enzymes in cyanobacterial hydrogen production

In cyanobacteria, hydrogen production is accomplished by the catalytic action of enzymes (Fig. 2). Cyanobacteria use two distinct enzymes to generate hydrogen gas:

Table 1 – Hydrogen producing cyanobacteria.

Heterocystous cyanobacteria	Marine cyanobacteria	Non-Heterocystous cyanobacteria
Anabaena flos-aquae	*Oscillatoria brevis*	*Synechococcus* sp.
Anabaena cylindrica	*Oscillatoria limosa*	*Microcystis* sp.
Anabaena variabilis	*Oscillatoria* sp. Miami BG7	*Gloebacter* sp.
Anabaena azollae	*Calothrix scopulorum*	*Synechocystis* sp.
Anabaena sp. PCC 7120	*Calothrix membranacea*	*Aphanocapsa montana*
Nostoc muscorum	*Cyanothece* 7822	*Gloeocapsa alpicola* CALU 743
Nostoc linckia	*Anabaena cylindrica* B-629	*Chroococcidiopsis thermalis* CALU 758
Nostoc commune		*Mycrocystis* PCC 7806
Anabaenopsis circularis		Microcoleus chthonoplasts