

Biophotolytic Hydrogen Production

Jim Barber ^a, Marko Boehm ^a, Steven Burgess ^a, Klaus Hellgardt ^b,
Geoffrey C Maitland ^b, Peter J Nixon ^a, Bojan Tamburic ^b, Fessehaye W Zemichael ^b
^a Department of Biology and ^b Department of Chemical Engineering, Imperial College London, SW7 2AZ

Introduction

The green alga *Chlamydomonas reinhardtii* has the ability to photosynthetically produce molecular hydrogen under anaerobic conditions. It offers a biological route to renewable and decarbonised hydrogen production from two of nature’s most plentiful resources – sunlight and water. Hydrogen has the potential to provide safe, clean, secure and affordable energy that can be used to power vehicles, homes, factories and even electronic equipment. Algal hydrogen production does not generate any toxic or polluting bi-products and could offer value-added products derived from algal biomass. The main costs of the process are the mineral nutrients required for algal growth and the material costs associated with building a photobioreactor (PBR).

Genetic Approaches to Improve H₂-production in the Green Alga *C. reinhardtii*

Background

When *C.reinhardtii* is deprived of sulphur, photosynthetic O₂ production decreases below the level of respiratory O₂ consumption, leading to an anaerobic environment. Taking advantage of this observation and by separating the growth and H₂ production phases, the extreme O₂ sensitivity of the Fe-hydrogenase of *C. reinhardtii* can be overcome and sustained photo-biological H₂ production can be realised. Under anaerobic conditions, electrons from a mixture of residual photosystem II (PSII) activity and of the fermentation of endogenous starch reserves (Fig. 1) can be fed to the hydrogenase and result in the production of molecular H₂.

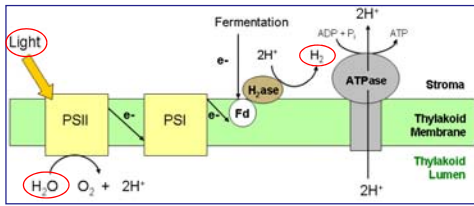


Fig.1 Scheme of the photosynthetic electron transport chain during anaerobic H₂ production in *C. reinhardtii*. PSII, photosystem II; PSI, photosystem I; Fd, Ferredoxin; H₂ase, [Fe]-hydrogenase.

Strategies

- Knock-down of competing fermentative pathways (Fig.2) utilising a novel artificial microRNAi (amiRNAi) technology to increase the flux of electrons to the hydrogenase.
- Constitutive expression of *C. reinhardtii* genes to (A) lower internal cellular O₂ levels and (B) increase the electron flow towards the hydrogenase.
- Screening of already available photosynthetic, mitochondrial and CO₂ requiring mutants for elevated H₂ production (Fig.3)

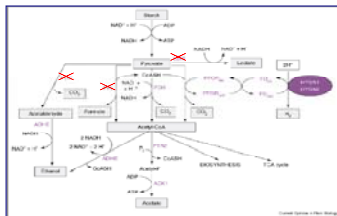


Fig.2 Diagram of the putative anaerobic fermentative pathways in *C. reinhardtii*. PDC, pyruvate decarboxylase; PFL, pyruvate formate lyase; PDH, pyruvate dehydrogenase; PFOR, pyruvate ferredoxin oxidoreductase; LDH, lactate dehydrogenase; ADHE, alcohol dehydrogenase. amiRNAi targets are crossed in red.

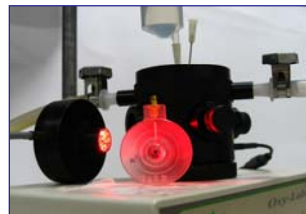


Fig.3 Clark electrode setup. A modified Clark electrode with reversed polarity will be used for the rapid determination of H₂ production rates of genetically engineered *C. reinhardtii* strains.

Conclusion

This project links genetic approaches to reactor design and engineering, demonstrating the power of using an integrated, cross-disciplinary approach to address the challenge of carbon-free H₂ production. Improvements in H₂ production efficiency and bioreactor design may allow hydrogen to fulfil its potential as the sustainable fuel of the future.

Photobioreactor Design

Design a reliable, cheap, continuous and fully automated PBR system that meets the requirements of algal growth, sulphur deprivation and H₂ production

Growth

- Control the light intensity, pH, agitation and temperature of the system (Fig.4)
- Minimise the risk of contamination by using filters and sterilisation procedures

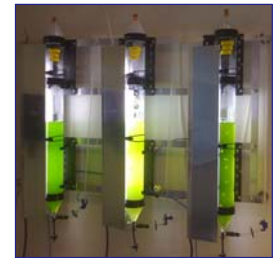


Fig.4 AquaMedic® culture reactors facilitate *C.reinhardtii* growth



Fig.5 Sartorius® photobioreactor used to investigate growth and H₂ production kinetics

Sulphur deprivation

- Cycle the algal growth medium by:
- Extracting a pallet of algal cells by centrifugation or ultrafiltration
 - Heavily diluting the growing culture with a sulphur replete medium
 - Sulphur content control

H₂ Production

Measurement

- Gas phase H₂ production measured by water displacement
- Reversible Clark electrode measures relative dissolved oxygen/hydrogen content
- H₂ permeable membrane (Fig.6) connected to a vacuum system used in conjunction with an amperometric H₂ sensor to quantify and collect dissolved hydrogen



Fig.6 H₂-permeable membrane

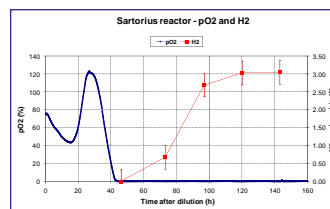


Fig.7 H₂ production commences once anaerobic conditions are established

Optimisation

- Better understanding of kinetic parameters (Fig.5)
- Light intensity and light penetration through the culture
- Temperature, agitation, pH
- Mineral nutrient requirements
- H₂ leak-tightness
- Initial optical density (cell thickness) of the culture