

Designing Microbial Communities For Enhanced Biohydrogen Production

INTRODUCTION

Phototrophic microbial communities – groups of tiny organisms whose energy for growth comes from light – play a significant role in global primary production by absorbing carbon dioxide and nitrogen gas. With the growing challenges of energy demands and environmental concerns, researchers are exploring scientifically designed (synthetic) phototrophic communities as a promising alternative to traditional energy generation methods. These consortia can efficiently convert CO₂ and N₂ gases, along with water and solar energy, into bioenergy products, offering a potential solution to today's energy and sustainability problems.

In this context, the development of synthetic phototrophic communities has attracted increased attention due to their ability to divide tasks among different species, allowing them to function more efficiently and remain stable. However, challenges remain, particularly in maintaining balance among strains and ensuring stable performance in environments that do not replicate the complex natural conditions in which these consortia typically thrive.

To address these challenges, recent PROMICON studies have focused on how cyanobacteria interact with purple nonsulfur bacteria (PNSB). These bacteria, including *Rhodospseudomonas palustris* (*R. palustris*), have shown potential in producing biohydrogen and lipids by capturing nitrogen in oxygen-free environments. Nevertheless, a key limitation is that they need a carbon-based food source (e.g., acetate) to produce energy. A promising approach to overcome this issue involves growing *R. palustris* with cyanobacteria, which can pull carbon dioxide from the air and turn it into the organic carbon that *R. palustris* needs to thrive.

EVIDENCE AND ANALYSIS

To help turn greenhouse gases into sustainable energy and promote a circular bioeconomy, researchers in the international EU-funded PROMICON project successfully built and controlled a light-powered microbial community that produces hydrogen (H₂) and fatty acids by capturing carbon and nitrogen from the air. This highlights the potential of controlling and optimising phototrophic communities for sustainable bioenergy production. The full extent of the work and results described below are available in Pan, M. et al. (2025) and Roussou, S et al. (2025).

- Elemental balance confirmed that **the consortium successfully captured carbon and fixed nitrogen.**
- The strategy of controlling light cycles helped maintain low oxygen levels in the system, **allowing *R. palustris* to use nitrogen, even though oxygen is produced by *Synechocystis*.**
- When infrared light was added to the light cycles, **the production of hydrogen (H₂) and fatty acids**, particularly C16 and C18, **increased significantly.**
- Proteomic analysis showed that the microbes exchanged acetate and **regulated their metabolic activities based on light.**
- **Infrared light boosted the production** of proteins involved in nitrogen fixation, carbohydrate processing, and transport in *R. palustris*, while **constant white light promoted the production of photosynthesis-related proteins** in *Synechocystis_acs*.

This study explores biotechnology that fixes atmospheric N₂ and CO₂, offering a sustainable path toward a net-zero emissions economy. However, challenges remain. Inactivating hydrogenases could prevent the consumption of produced H₂, and continuous reactors with selective membranes might improve performance. Long-term stability needs further testing.

POLICY IMPLICATIONS

The development and application of synthetic phototrophic microbial consortia for bioenergy production holds significant policy impacts, particularly in the context of the EU's sustainability and climate goals. The bioeconomy is stipulated by several EU policies and initiatives, including the **Bioeconomy Strategy** and **the Circular Economy Action Plan**.

- Phototrophic microorganisms hold great promise for capturing solar energy and converting greenhouse gases into sustainable energy carriers, providing an alternative solution to the increasingly fierce energy challenge.
- By harnessing the power of genetic engineering and light regulation, these synthetic microbial communities have the potential to revolutionise bioenergy production by enhancing the production of both biofuels and valuable chemicals through carbon capture.
- Such fatty acids-derived biofuels (e.g., via transesterification and esterification) even have a higher energy density and are more compatible with current infrastructure when compared to other forms of renewable energies.
- This process offers a sustainable alternative to traditional energy sources and directly supports key EU policies aimed at reducing carbon emissions and promoting renewable energy.
- Providing funding for genetic engineering and biotechnological research to optimise microbial strains for hydrogen production.
- Supporting the development of advanced bioreactors with selective membranes to enhance continuous H₂ and O₂ removal, improving efficiency and scalability.
- Establishing regulatory incentives and funding for long-term studies on N₂ and CO₂ fixation to ensure the viability of microbial systems for industrial applications.

SUSTAINABILITY AND LEGACY

The methodology, results and implications of the described work are openly available at <https://doi.org/10.1021/acs.est.4c08629> and <https://doi.org/10.1016/j.ymben.2025.01.008>.

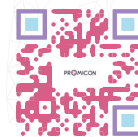
PROJECT OBJECTIVES AND METHODOLOGY

The overall objective of the PROMICON project is to learn from nature how microbiomes function in order to steer their phenotypes towards the production of biopolymers, energy carriers, drop-in feedstocks, and antimicrobial molecules. The project will achieve this by developing novel analysis and modeling approaches targeting essential key species in productive microbiomes, as well as whole microbiomes. PROMICON also aims to establish a standardised platform for obtaining quantitative single-cell data and connected coherent OMICS and Meta-OMICS data sets for complex microbiomes.

The methodology involves the analysis and modeling of both essential key species in productive microbiomes and whole microbiomes using a combination of data mining tools, mechanistic process models, and machine learning and deep learning approaches. The project also employs synthetic biology and systems metabolic engineering to optimise and assemble bacterial farmers, producers, and stabilisers, providing optimal production of target metabolites. The project focuses on developing sustainable bioproducts that can contribute to the circular economy, thus aligning with the objectives of the EU 2018 bioeconomy strategy.

REFERENCES

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2. Roussou, S., Pan, M., Krömer, J. O., & Lindblad, P. (2025). Exploring and increased acetate biosynthesis in *Synechocystis* PCC 6803 through insertion of a heterologous phosphoketolase and overexpressing phosphotransacetylase. *Metabolic Engineering*, 88, 250-260. <https://doi.org/10.1016/j.ymben.2025.01.008>



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This project receives funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101000733. Views and opinions expressed are however those of the author(s) only and do not necessarily reflect those of the European Union or the European Commission. Neither the EU nor the EC can be held responsible for them.