



2023 Newsletter



Microalgae Production Systems

Microalgae are some of nature's finest examples of solar energy conversion systems, transforming carbon dioxide into complex organic molecules through photosynthesis.

Editorial

The production of microalgae as a source of renewable fuel lived its golden years during the 2000s. The high photosynthetic efficiency of microalgae as well as their potential to grow on non-arable land and without the need for potable water attracted enormous interest on the part of the scientific community. Such interest ended up permeating the business world that saw microalgae production as a promising industry: green gold. But all expectations placed on microalgae as a renewable fuel source collapsed by the end of the decade. What seemed like a panacea turned out to be the downfall of many companies that lost their interest in the long-awaited industry of microalgae biofuels. Companies require profit in the short term but, like any other technology that starts from a lab-flask, its large-scale implementation implies overcoming a series of drawbacks, which sometimes means a long way to go.

One of the main drawbacks in this case lies in the fact that tamed microalgae grow in isolation, but in large-scale systems they need to deal with a multitude of pathogens and diseases that reduce their productivity. PRODIGIO project aims to address this issue by developing early warning systems. These systems, which are common and very successful in many other industries, will allow us to predict when the system is going to fail, giving us time to apply countermeasures. The ultimate goal is to increase the stability of the biological communities involved in the production of microalgae biomass and its subsequent conversion into biogas. The PRODIGIO consortium, made up of world experts in the fields of process engineering, ecogenomics of microalgae and anaerobic digestion systems, and mathematical modelling, aims to renew commercial interest in microalgae as a source of biofuel and thus contribute to building a healthier planet.

Below we present the progress of the activities planned in the PRODIGIO project as well as some intermediate results.

Progress on microalgae production systems

Failure tests in photobioreactors



Raceway reactors for microalgae biomass production located at the IFAPA facilities near the UAL campus.

Microalgae-based processes are determined by environmental and operational variables such as light, temperature, pH, dissolved oxygen, carbon dioxide and nutrient concentration (mainly nitrogen and phosphorous). These parameters not only determine microalgal activity but also influence the rest of the microbial communities in the systems. Any change in the operating conditions will affect the dynamic populations, promoting or inhibiting the growth of microbial groups, strongly determining the success of the industrial biological process.

During the first stage of the project, three reactors widely used in the industrial production of microalgae, known as raceway reactors, were operated under controlled conditions in the laboratory to study the role of temperature in the stability of microalgal cultures. The main objective of the tests was to determine how its progressive increase promotes changes in the ecology of the systems, which can lead to system failure.

In parallel with this activity, two identical external raceway reactors fed with freshwater plus chemical fertilizers, and with primary wastewater from the UAL were operated in continuous mode for nine months under environmental conditions. During the study period, three times per week samples were collected from the systems to perform metagenomic analyses by ICM-CSIC (Barcelona, Spain) and chemical fingerprint analyses by AWI (Germany) to “catch” a possible system collapse and its potential sources.

This complex network of experimental data, together with the experience of researchers in the field of study, will open the gates to elucidate the biological keys to the success of microalgal biotechnology.

Modelling the failure of microalgae production

The overall goal of this activity is to analyse the mechanisms underlying the productivity decreases in microalgal PBRs, identify early warning signals and evaluate, from the whole set of identified signals, those that meet the specified criteria of scalability, reliability, and affordability.

During the 1st two years of the project, the work on this activity was centered on collecting and assembling all available data from routine measurements, MetaOMICs and chemical fingerprint analyses.

In addition, a model of microalgae production in a photobioreactor is currently being developed from scratch. The model simulates a steady state system with a microalgae, a group of remineralisers (bacteria) and a predator. The model is relatively simple, and its ecological complexity is being increased by applying perturbations to the system. Possible perturbations include sudden changes in temperature, light, and adding a pathogen. The effect of the pathogen would be to increase the mortality of the microalgae. Different increases in mortality, i.e. pathogenic levels, are being simulated. The end result of these simulations will be the time series of the population dynamics and their effect on the total biomass generated in the bioreactor.

The activity related to the definition of the time series of chemical, biochemical and microbiological data (including metabarcodes and metagenomes) generated during the first sampling campaign (April 2021 to January 2022) of the microalgae production systems were successfully processed and sent to the National Taiwan University on 24 August 2022. This partner and members of our team at ICM-CSIC are carrying out empirical dynamic modelling analyses from which the first interactomes of the bioreactors (i.e. topologies of the interaction networks and strength of interactions) will be generated.

Progress on anaerobic reactor systems

Failure tests in anaerobic reactors



Bench-scale anaerobic digesters located at the IMDEA-energy facilities.

The overall goal of this activity is to generate time-series data from perturbation experiments in continuous-flow anaerobic reactors (ARs) for subsequent identification of early warning signals, and quantification of threshold values and warning times.

In the last 12 months, IMDEA Energy has evaluated the microalgae anaerobic digestion failure against 5 different perturbances: (1) long-lasting shock of stepwise organic loading rate (OLR) increase, (2) long-lasting shock of microalgae composition change, (3) salinity intrusion shock, (4) pesticides perturbation and (5) antibiotics perturbation.

Firstly, IMDEA Energy has investigated the impact of long-lasting shocks on process performance.

The ARs subjected to a long-lasting OLR increase from 1.5 to 7.0 g COD L⁻¹ d⁻¹ did not show a sudden biogas production decline. However, the methanogenic stage exhibited greater damage than those that occurred in short-term shocks

since high concentration of metabolites were detected and a long process recovery time was observed.

An opposite trend was identified when a long-lasting microalgae composition change was applied to ARs. In this case, the process was able to successfully deal with this perturbation since no variations in biogas production were observed.

Secondly, IMDEA-E has investigated the impact of the presence of some inhibitors such as salinity, pesticides and antibiotics.

The results evidenced that the presence of inhibitors greatly damages the whole AD process, including not only methanogenesis but also acetogenesis, acidogenesis and hydrolysis were inhibited. Overall, these results showed that each perturbation should be individually evaluated to elucidate the process behavior since the AD response cannot be extrapolated.

In the last months, IMDEA sent biological samples to the partners located at NMBU (Norway) as well as the filtered samples to AWI partners (Germany) to perform chemical fingerprinting analysis. The samples will be evaluated to establish a relationship between chemical and biological results upon AD shocks.

Modelling the failure of anaerobic digestion

“Modelling the failure of an anaerobic reactor” has the goal of detecting signatures responsible, or at least indicative of reactor failure. These involve chemical parameters of reactor performance (methane yield) but also metabolic shifts within the microbial community living inside the reactor. Three experiments aim to investigate the failure including 1) high organic overload, 2) high-protein biomass, and 3) the presence of chemical pesticides.

NMBU analyzes the 16S microbiome structure as well as perform in-depth analysis of select timepoints during the reactor perturbations using state-of-the-art metagenomics and metaproteomics techniques, to pinpoint active microorganisms and the enzymes and pathways they utilize. So far, we have

analyzed the first dataset of organic overload and are in the process of analyzing the second dataset for high-protein biomass.

Preliminary results have shown that reactor methane production significantly decreases as a response of the high-organic, and this corresponds to a shift in the methane-producing archaea. During steady-state, Methanotrix is the most abundant and active methanogen, while during the high-organic load, Methanotrix seems to perish, and in the reactor-recovery phase, methane production is taken over by Methanosarcina. Thus, the decreasing abundance of Methanotrix may serve as a signature of reactor failure. This will be further investigated using empiric dynamic modelling in early 2023.

Sustainability assesment of Prodigio from environmental, social and cost perspectives

The overall goal of this activity is to evaluate and compare the potential environmental, economic and social impacts of energy production from microalgal biogas with and without the early-warning technologies developed in PRODIGIO, and to identify the most sensitive aspects to be considered for future commercial application with a life cycle perspective.

To evaluate environmental aspects using Life Cycle Assessment (LCA), ARMINES has developed a set of parameterized models in Python programming language using [LCA-specific libraries Brightway2](#) and [lca algebraic](#). The parameterized models require several variable input parameters that serve to estimate, through mass and energy balances, the life cycle inventory on

consumed elementary substances and emitted pollutants, as well as the resulting environmental impacts.

In a first phase, two static models were developed to obtain LCA results based on average parameters for microalgae culture and biogas production. One of the static models was based on data and mathematical relationships identified in the literature, while the other one was established based on discussions with PRODIGIO partners and their previous experience.

In the second phase, an adaptation was made to convert the static models into a dynamic model, which uses time-series as input data, in order to better represent the effect of variability on the environmental results of the microalgae and biogas production systems over time related to changing surrounding conditions.

Four scenarios have been defined: two scenarios of microalgae cultivation in wastewater with and without early warning systems and two scenarios using clean water and fertilizers. The results obtained to date, corresponding to scenarios without early warning systems, have allowed the cultivation and harvesting stages to be identified as the main relative contributors to the environmental impacts, partly due to the energy requirements of both stages. In the next steps, the LCA results of this project will be compared to other LCA studies in the literature on systems providing the same function (e.g. systems with a wastewater treatment function), to identify potential benefits of microalgae pathways and options for improvement.

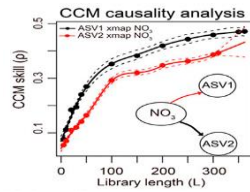
To complete the environmental evaluation with an economic assessment, Aspen simulations are under development. Existing Aspen models will be adapted to consider the same assumptions as those of the environmental model, to obtain Life Cycle Costing results for the same scenarios.

Update C&D and exploitation

News:



The algae-eating microbiome that lives inside an anaerobic reactor



Empirical dynamic modeling for mechanistic understanding and prediction of bioreactors



ICM-CSIC visits Universidad de Almería (UAL) facilities

Scientific publications:

The power of unicellular primary producers

Marine phytoplankton, including cyanobacteria and microalgae,

Realizing algae value chains in arid environments: an Arabian Peninsula perspective

Algae are a promising feedstock for

dominates primary production across two thirds of the earth's surface, sustaining virtually all marine life and exerting a fundamental control over global climate through carbon sequestration into the deep ocean.

the sustainable production of feed, fuels, and chemicals. Especially in arid regions such as the Arabian Peninsula, algae could play a significant role in enhancing food security, economic diversification, and decarbonization.

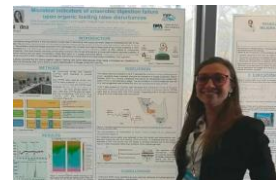
Don't miss our new section "[Stakeholder Platform](#)":

In this new section we will include news and updates on projects and initiatives where PRODIGIO can bring solutions to challenges in the microalgae production industry or anaerobic digestion.

If you are interested in collaborating with PRODIGIO, please register through our "[Get Involved](#)" form.

Related news from Prodigio partners:

[IMDEA Energy has participated in IWA-Young Waters Professionals Conference \(Valencia, Spain\)](#)



[IMDEA Energy is coordinating 2 new projects on the theme of anaerobic digestion](#)



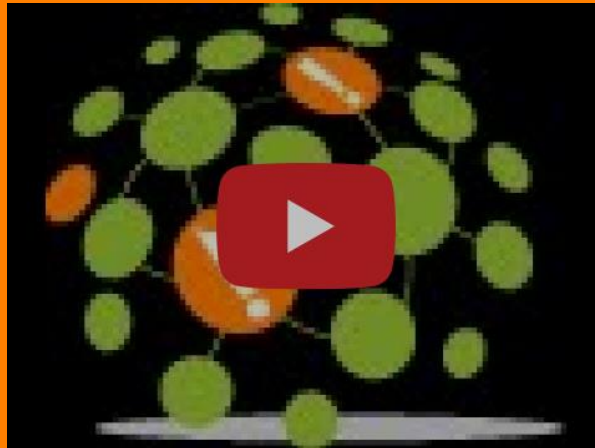
[University of Almeria is a partner in a new horizon europe project about microalgae](#)



Growing sustainable biogas in Europe with microalgae



Don't miss our videos



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