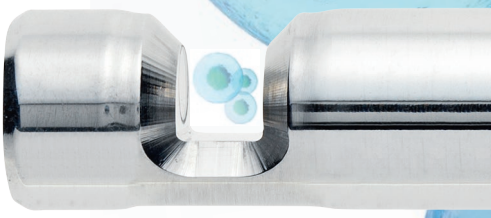
Continuous Cyanobacteria - Limnospora (Spirulina) Monitoring in the MELiSSA Loop using Dencytee



Drift free long term biomass monitoring for better understanding of process dynamics

Industry Segment: R&D for extraterrestrial systems
Application Field: Life support equipment
Hamilton Products: Dencytee, Retractex B HyCIP, pH and DO Arc sensors, ArcAir software

Introduction

MELiSSA (Micro Ecological Life Support System Alternative) is an international collaborative effort of *15 partners* led by European Space Agency (ESA) focused on the development of a *Regenerative Life Support System to support long-term Space Missions*. Technologies as those studied in MELiSSA, will contribute to make possible the presence of humans on a planet like Mars.

The goals of MELiSSA are the production of food, recovery of water and regeneration of the atmosphere, with a concomitant use of wastes, i.e. CO_2 and organic wastes, using light as a source of energy.

Point of Measurement

In a context of biological life support system, long-term continuous bioreactor operation enables the constant production of oxygen and food to meet with human requirements. In the MELiSSA Pilot Plant a photobioreactor where *Limnospira indica* (previously known as *Arthrospira/ Spirulina*) is cultivated aims to continuously produce oxygen and food to partially cover these human needs. In this scenario, the use of robust, reliable monitoring, and control sensor technology is mandatory. Parameters such as pH, pO_2, and optical density are necessary to ensure the best bioreactor performance. But special attention must be focused on optical density (OD). In the MELiSSA photobioreactor the optical density of cells is an output from two main manipulated variables: dilution rate (D) and light intensity. Online measurement of biomass concentration is essential to react to any bioreactor perturbation and to monitor the culture status. Additionally, it allows the fine-tuning of manipulated variables to reach a target optical density to optimize the culture performance for O_2 and biomass production.

Online monitoring of biomass was performed in a 180 day experiment where the aim was to characterize and understand the bioreactor performance under different regimes of light and dilution rate. A Dencytee sensor was used for online turbidity monitoring. It was installed in the bioreactor through the retractable housing Retractex B HyCIP, which enables the retracting of the sensor for cleaning and sterilization if needed (Figure 1).

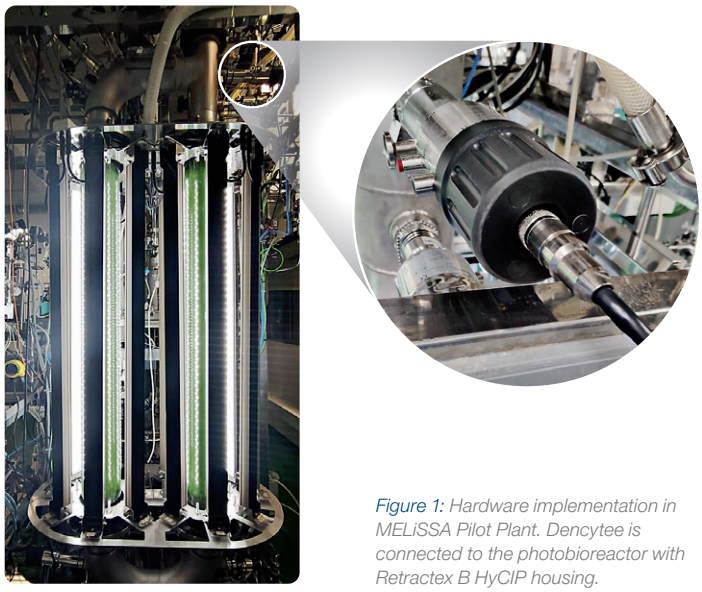


Figure 1: Hardware implementation in MELiSSA Pilot Plant. Dencytee is connected to the photobioreactor with Retractex B HyCIP housing.

It is especially relevant for sensor maintenance in long-term operation processes where probe fouling over time may occur.

Figure 2 shows the evolution of off-line OD, off-line cell dry weight (CDW) and the in-line biomass sensor. The optimal performance of the sensor is proved by the excellent correlation between off-line and in-line values no matter the experimental condition. The fitting is observed in low-range absorbance units (A_{750nm}), with 0.4 and 3 AU the minimum and maximum respectively. Changes in biomass concentration are caused by different values in the manipulated variables. A relevant result is that the sensor allows one to follow the dynamics of the process without the need of sampling, which is of significant importance from a control point of view. In most cases there is a good correlation with cell density, but a deviation exists in some conditions. This behaviour is caused by changes in CDW/OD ratio of the cell culture because

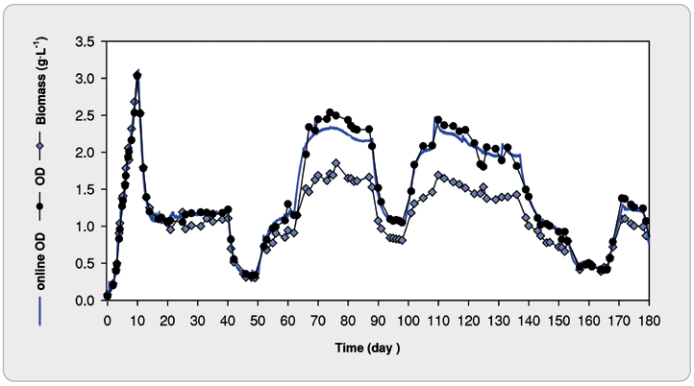


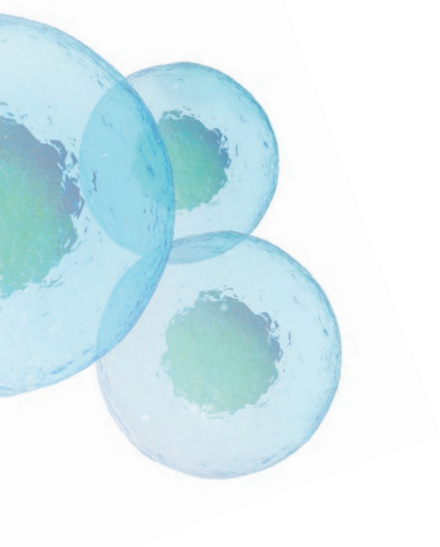
Figure 2: Limnospira indica in-line and off-line monitoring during the 180 days continuous operation. Strong correlation is observed between in-line (Dencytee) and off-line OD. This profile is caused by changes in manipulated variables causing different steady states values for biomass.

of morphology changes, which affect the scattering and absorbance properties of the cells. As the Dencytee sensor is based on absorbance, the changes in scattering properties of the cells are reflected in the measurement. In long-term operation, changes in morphology can occur depending on environmental conditions, which is rare during a short batch cultivation. Therefore, online measurement of OD is helpful to predict morphology variations in the cells.

Advantages of the Hamilton solution

Biomass and oxygen production are the target products of the MELiSSA loop. The Hamilton Dencytee helped to monitor reliable the production of the biomass. No deviations from off-line absorbance data were observed during 180 days culture and only few initial samples were needed to establish a linear correlation between off-line and in-line values. In addition, the sensor clearly showed the dynamics of the culture when exposed to changes in environmental conditions, identifying the point when culture reaches stability for every condition. It gives an idea of the possibilities of this technology in predicting the culture tendency and anticipating potential deviations.

Finally, one of the most important points of the experimental series is that there was no need for maintenance operation and the robustness of the sensor is proved. During the whole process no drift was observed, and no biofilm was formed at the sensor lens. Hence, it is demonstrated that these sensors can be used for long-term operation process (months scale) with a minimum maintenance and without the need of re-calibration.



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MELISSA MICRO-ECOLOGICAL LIFE SUPPORT SYSTEM ALTERNATIVE

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