



DE LA RECHERCHE À L'INDUSTRIE

***CHLORELLA VULGARIS*, A PROMISING FEEDSTOCK FOR STARCH-BASED BIOPLASTICS**

Compadre A., Six A., Dubreuil C., Dimitriadis-Lemaire A., Delrue F., Alvarez P., Fleury G., Mailley S., Sassi J.F.

CEA, CEA Tech Région Sud - Provence-Alpes Côte d'Azur, F-13108 Saint Paul lez Durance, France

Commissariat à l'énergie atomique et aux énergies alternatives - www.cea.fr

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- ▶ CEA is involved in 2 European projects **SEALIVE**^[1] and **Nenu2PHAr**^[2] with a common objective: the development of new sustainable chains, aiming the creation of **greener** and **cost-effective plastics**.

- ▶ **Starch** is seen as a promising plastic **feedstock** since it can...

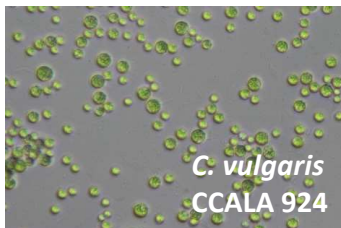
... directly integrate **plastic blends**^[3]



... be degraded into monomeric glucose to **feed PHA producing bacteria**^[4]



- ▶ **Microalgae** are **promising starch producers** since they **do not require arable lands** while having the advantage of **capturing atmospheric CO₂**.



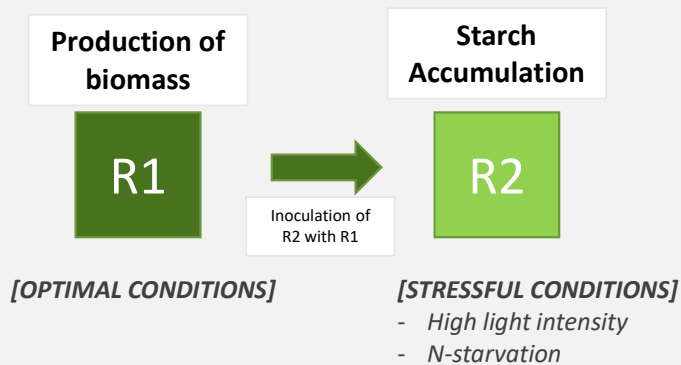
- ▶ ***Chlorella vulgaris*** is known to be an **excellent starch producer** (up to 60 %DW)^[5] and a robust microalga, which can be cultivated at **industrial scale**.



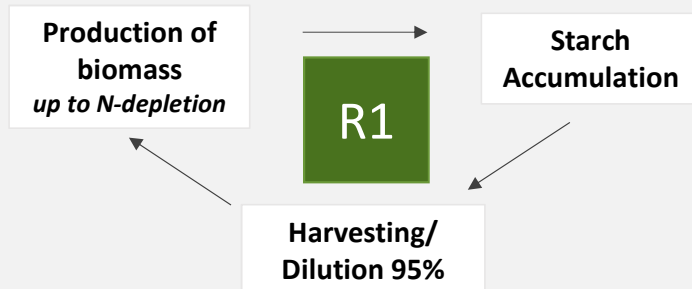
Background: *C. vulgaris* CCALA924 is known for producing starch under high light intensity and N-starvation^[5]

Objective: Development and validation of an optimized strategy for starch accumulation up to a pre-industrial scale

2-reactors-strategy (2R-S)

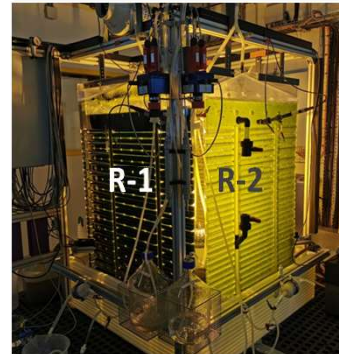


1-reactor strategy (1R-S)



Pilot Scale

2x 25L Flat Panel PBRs

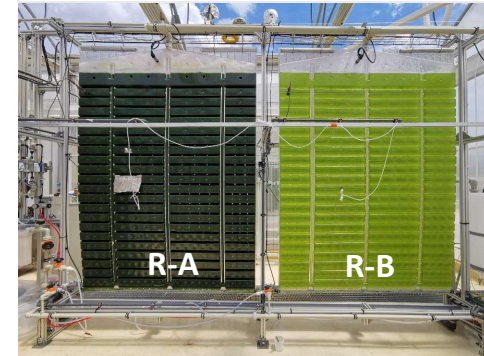


Artificial Light

- T set point = 25 °C
- 200 nL/h gas flow (2% CO₂)
- Light intensity 240 μE/m²/s
- 20h:4h light:dark

Semi-industrial Scale

2x 180L Flat Panel PBRs



Natural Light

- T_{max} set point = 30 °C
- 1500 nL/h gas flow (2% CO₂)
- Natural light (9h:15h L:D) (+ optional artificial light of 30 μE/m²/s (R-A) or 185 μE/m²/s (R-B))

Culture Medium: Adapted Beijrink medium, with and without NaNO₃

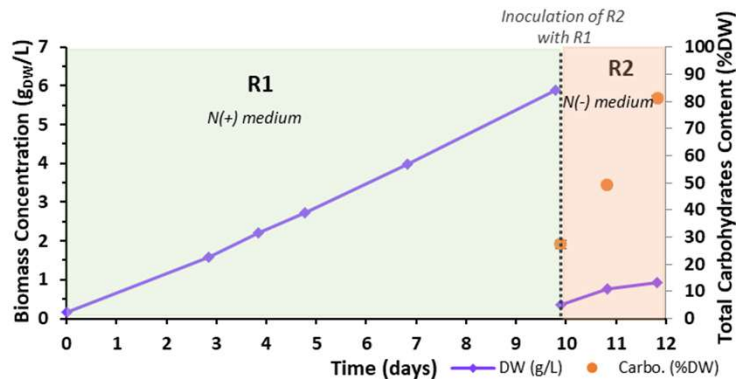
Daily sample analyses :

- OD@880nm + [DW]
- [NO₃⁻], [PO₄³⁻], [SO₄²⁻]
- Carbohydrates content (Dubois method^[6])

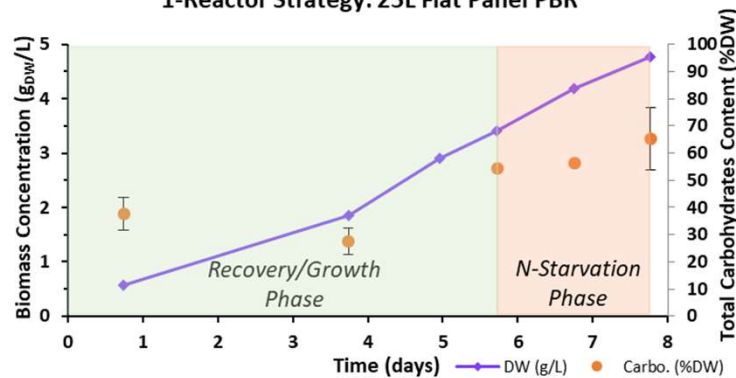
Objective: Optimization of the carbohydrates-enriched biomass production at pilot scale (25 L PBRs), under artificial light: 1-reactor vs 2-reactors strategy



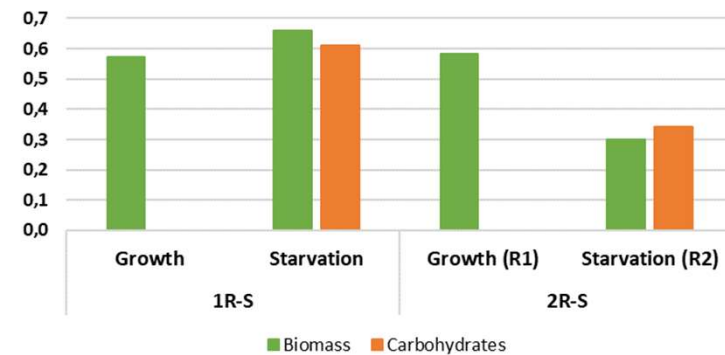
2-Reactors Strategy: 25L Flat Panel PBRs (R1 and R2)



1-Reactor Strategy: 25L Flat Panel PBR

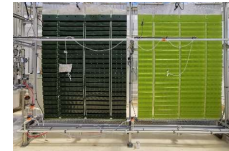


Mean Productivity (g/L/d)

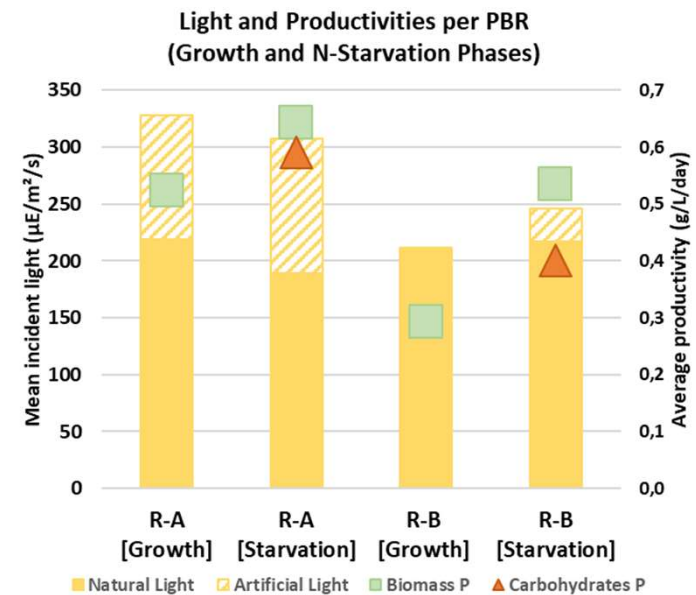
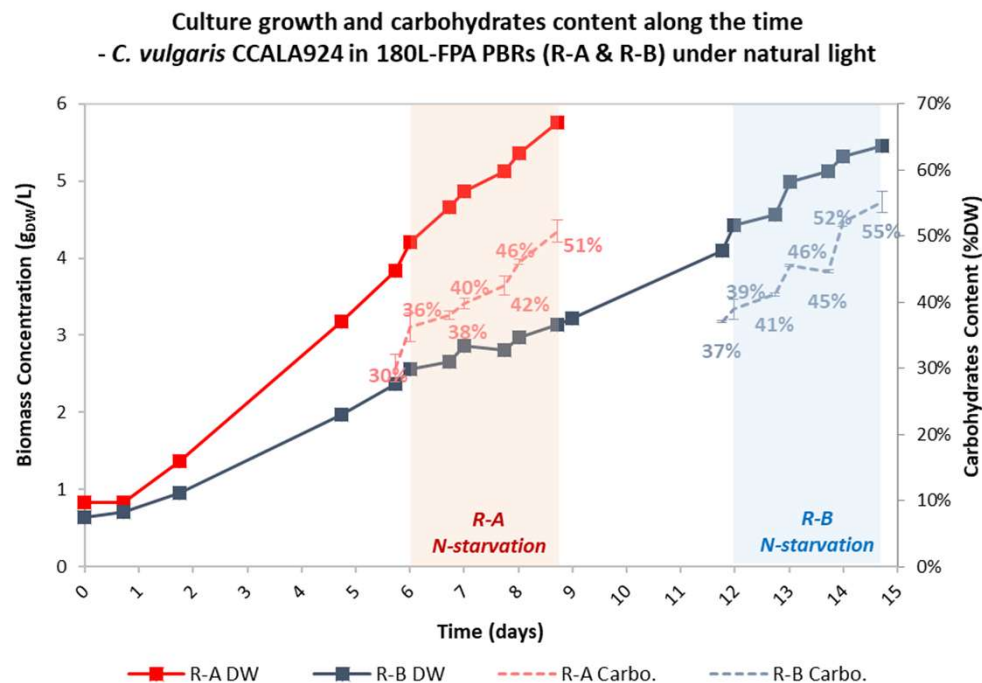


Decision factor	2R-S	1R-S
Biomass concentration	✗	✓
Carbohydrates content	✓	✗
Carbohydrates productivity	✗	✓
Resources (man-power, utilities, etc.)	✗	✓

Objective: Validation of the 1-reactor strategy at pre-industrial scale (180L PBRs) under natural light and with additional artificial light



Autumn 2021
 South of France
 (greenhouse)



► Artificial light crucial to boost biomass productivity and to compensate the lack of natural light in the starvation phase in R-A

Conclusions

- **Total carbohydrate** content raised up to a range of **65-80 %_{DW}** for both strategies at **pilot scale** under **artificial light**, confirming that **nutrient-deprivation** plays a crucial role on starch accumulation.
- **1R-S was selected** for the upscaling due to its **good productivity**, and versatility to operate with the available **utilities** and **man-power**.
- Results were validated at **semi-industrial** scale under natural light (up to **55%_{DW}** carbohydrates content). **Extra artificial light** showed to be **essential** to improve the system performance in Autumn/Winter season.

Ongoing and future work

- Production process under natural light is under a continuous optimization;
- Different **starch purification** schemes are under development, in order to effectively include *C. vulgaris* feedstock into the production of biodegradable bio-based plastics.





Thank you for your attention



Ana Compadre

 ana.compadre@cea.fr

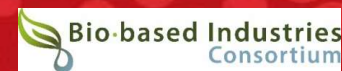
 [linkedin.com/in/anacompadre](https://www.linkedin.com/in/anacompadre)



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REFERENCES

- [1] SEALIVE Project, <https://sealive.eu/>
- [2] Nenu2PHAR project, <https://nenu2phar.eu/>
- [3] Jiang T., et al. (2020) Starch-based biodegradable materials: Challenges and opportunities, *Advanced Industrial and Engineering Polymer Research*, 3: 8-18. <https://doi.org/10.1016/j.aiepr.2019.11.003>.
- [4] Jiang, G., et al. (2016) Carbon Sources for Polyhydroxyalkanoates and an Integrated Biorefinery, *International Journal of Molecular Sciences*, 17(7), 1157. <https://doi.org/10.3390/ijms17071157>.
- [5] Brányiková, I., et al. (2011), Microalgae—novel highly efficient starch producers. *Biotechnol. Bioeng.*, 108: 766-776. [doi:10.1002/bit.23016](https://doi.org/10.1002/bit.23016).
- [6] Dubois M, Gilles KA, Hamilton JK, Rebers PA, Smith F. (1956) Colorimetric Method for Determination of Sugars and Related Substances. *Analytical Chemistry*. 28: 350-6

