

**EU Framework Programme for Research and Innovation
H2020-Competitive Low-Carbon Energy
Call topic 11-2014**



www.photofuel.eu

Photofuel - Biocatalytic solar fuels for sustainable mobility in Europe

Deliverable D3.11

**Performance of biocatalysts outdoors in 120L
PBR-scale. Supporting for decision on
achievement of M4**



This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 640720

Editorial	
Deliverable N ^o :	D3.11
Title	Performance of biocatalysts outdoors in 120L PBR-scale. Supporting for decision on achievement of M4
Workpackage:	WP3
Responsible beneficiary:	A4F
Authors:	Diana Gomes, Sara Cabral, Celina Parreira, Tiago Guerra, Sara Badenes, Vitor Verdelho
Contributors:	Diana Gomes, Tiago Guerra, Olaf Kruse, Julian Wichmann, Jonathan Wagner, Klaus Helgardt,
Version:	Final
Due date of deliverable:	< 31/12/2019 >
Version date:	< 06/08/2020 >
Contact:	Tiago.Guerra@algafuel.pt
Dissemination level:	CO-Confidential
Nature:	Report
Review status	WP-leader accepted 12/08/2020
	SC accepted 26/08/2020
	Coordinator submitted 31/08/2020

Publishable Summary

Contamination of 1-butanol cultures at pilot scale have been impossible to avoid and the suspicion remains that some contaminants (e.g. bacteria and fungi) are able to metabolize the fuel product. Thus, a new butanol *Synechocystis* sp. PCC6803 strain was engineered by WP2 partners from University of Uppsala. This strain is capable of assimilating phosphite as single source of phosphorous. Assuming that most of the contaminants use only phosphate as source of phosphorous, this strategy could allow butanol accumulation to proceed to levels similar to the ones obtained at lab scale.

The cultivation of new *Synechocystis* sp. PCC6803 strain was carried out in Unilayer horizontal tubular photobioreactor (UHT-PBR) using continuous operation mode and a cultivation medium containing only phosphite as source of phosphorous. Maximum butanol concentration achieved was higher than in previous assays and culture stability was also improved. However, contaminations were eventually able to proliferate, probably due to the capacity of some contaminants to oxidize phosphite to phosphate.

16S meta-sequencing of culture contaminants was developed by partners from University of Bielefeld in order to understand which bacterial genera are present.

Table of contents

Publishable Summary.....	3
Table of contents	4
1 Deliverable objectives	5
2 Introduction	6
3. Pilot scale production of <i>Synechocystis</i> sp. PCC6803 BuOH-Pt	8
3.1. Culturing conditions	9
3.2. Cultivation results.....	12
3.3. Evaluation of residual phosphate concentration in culture.....	14
3.4. Identification of contaminants present in culture by 16S Meta-sequencing.....	16
4. Conclusions	18
5. References	20