PROJECT FINAL REPORT

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1 Executive summary

FUEL4ME: Future European league for microalgal energy

The EU funded FUEL4ME (FUture European League 4 Microalgal Energy) project was established to evaluate microalgae as potential sustainable source for second-generation biofuels that can compete with fossil fuels. To realise this an increase in the scale of microalgae production needed to be matched with a simultaneous decrease in production costs. The FUEL4ME project has achieved significant decreases in production costs of algal lipids, but the production costs for algal biodiesel are still (more than) an order of magnitude too high to make this process commercially attractive in the short term. However, microalgae as source for high-value components such as omega-3 fatty acids for application in food and feed products, has proven to be realistic and cost-effective using microalgae as production organism.

FUEL4ME aimed at exploiting algae's unique ability to produce high value biomass and lipids efficiently by photosynthesis. Such biomass and lipids could form an excellent starting material for the sustainable production of biofuels and other products such as animal feed. Moreover, microalgae, including the target algae of this project (*Nannochloropsis oceanica* and *Phaeodactylum tricornutum*), do not compete with food crops for land and freshwater as they are grown in sea, saline or other marginal water on unproductive dryland.

FUEL4ME investigated in detail the molecular and metabolic mechanisms governing lipid accumulation in two microalgae species and demonstrated enhanced lipid accumulation by metabolic engineering. Furthermore, we compared the current two-step batch nitrogen starvation production process for microalgal lipids with a newly developed continuous one-step nitrogen limitation process and optimised lipid production under different growth conditions. Researchers developed the various steps of the downstream processing chain, which involved harvesting, cell disruption, lipid extraction and fractionation and hydro-treatment of the lipids to create biofuel. In addition, biorefinery steps to valorise high value molecules such as omega-3 fatty acids, or high quality algae protein, were developed and demonstrated. They were shown to be successfully applicable to microalgae and are now ready to be used in commercial processes, especially for high-value applications in Food & Feed.

The consortium designed and set up three pilot plants for outdoor microalgae production based in Italy, the Netherlands and Israel, respectively, and one demonstration facility in Spain. At outdoor pilot scale it was shown that although with lower lipid content, the one-step N-limitation process had comparable lipid productivity to the traditional N-starvation batch process, but requires further testing for prolonged periods on a large scale.

To determine the actual state of the technology as well as study how key parameters influence the sustainability of the FUEL4ME integrated process a life-cycle assessment (LCA) study was performed. The main influences upon sustainability were cultivation and harvesting, electricity demand especially for PBR cooling, sources of freshwater and carbon dioxide, and suitable land. FUEL4ME has addressed some of these parameters by improving productivity of cultivation, high efficiency thermoregulation of the photobioreactors, and improved harvesting efficiency, including integrated water and resource re-use in cheap desert land. However, further major improvements, will be needed to make biofuel production with microalgae fully economic and environmentally sustainable. Currently the process is best suited for the production of high value products such as polyunsaturated fatty acids (PUFAs), and a promising biorefinery approach showed to have a strongly improved economic balance. We believe that FUEL4ME's long-term innovation strategy, with initial focus on high value products, will result in economically feasible and environmentally sustainable microalgae-based products. This will ensure a further decrease in production costs and an increase in the scale of production. However, microalgae as source for biofuels seems only viable when the majority of the

microalgal biomass is commercialized as high-value chemicals/commodities and the biofuels are a co-product instead of the main product. Both for microalgae cultivation and downstream processing FUEL4ME has provided an excellent opportunity for industrial partners to conduct pilot tests of their technologies, improving them and demonstrating their use within the microalgae field. This has helped to achieve more reliable and scalable industrial solutions.. Additionally, FUEL4ME has generated highly skilled professionals with expertise in algal microbiology and microalgae cultivation and processing systems. By developing knowledge and skills as well as sustainable valorised products from microalgae, the project has made a valuable contribution to Europe's research capacity and bioeconomy. Furthermore, this eco-friendly process has the capacity to reduce dependence on fossil fuels.

2 Summary description of project context and objectives

FUEL4ME: Future European league for microalgal energy

Project context and objectives

The aim of the 4-year FUEL4ME project was to develop a sustainable chain for continuous biofuel production using microalgae as a production platform, thereby making 2^{nd} generation biofuels competitive alternatives to fossil fuels. Main focus points were:

- Transforming the current 2-step process for algal lipid production into a continuous 1-step process with high lipid content (production process);
- Development of a continuous downstream process using all components of the algal biomass (conversion process);
- Integration of production and conversion process.



Future European League 4 Microalgal Energy

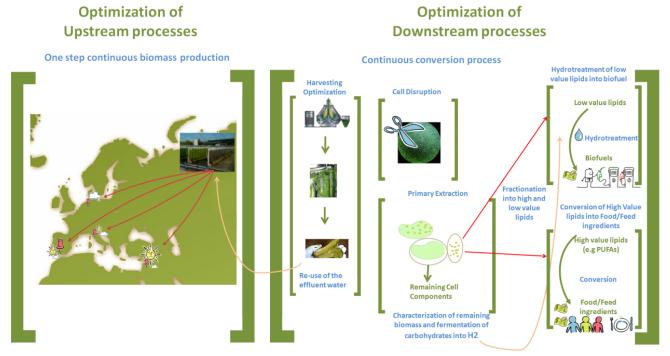


Figure 1. Schematic project overview

FUEL4ME aimed to exploit one the unique strengths of algae: the ability to produce lipids using energy from photosynthesis. These lipids form excellent starting material for the production of bulk products; the largest fraction of the lipids will be used for the production of biofuel (NExBTL) and a smaller fraction will be used for food and feed components (ω 3 fatty acids). This way optimal use of biomass results in simultaneous production of food and fuel.

Production process

In order to produce biofuels from microalgae, it is important to have significant algal production capacity. However, this capacity is not yet available. Two important reasons for the lack of significant algae production are:

- High costs; with the current knowledge and systems, it should be possible to produce biomass in a reactor of 100 hectare at a cost price of 5 € kg⁻¹.² To make microalgae truly interesting as a source of biofuels and able to compete with fossil fuels the scale of production needs to increase at least 3 orders of magnitude, with a concomitant decrease in the cost of production by a factor of 10.³
- Negative energy balance; in the current situation, more than 24% of the production costs are energy costs.⁴ The energy balance is negative or neutral for most systems, with an energy input equal or larger than the energetic output. It is evident that, to make microalgae attractive as a source of biofuels, the energy balance should be positive. A net energy ratio (NER = energy output / fossil energy input) of over two is an ambitious yet achievable goal required to achieve the renewable fuels standards in terms of fossil fuels input, and GHG savings.⁵

Both high costs, GHG balance and negative energy balance can be addressed by developing and improving the technology behind biomass and lipid production. Currently, production of lipids from microalgae is a two-step process. First, the microalgae are grown in batch cultures and, when a certain biomass concentration is obtained, stress in the form of nitrogen starvation is applied upon the algae to induce lipid accumulation. Consequently, the growth rate decreases resulting in low lipid productivity. Within FUEL4ME, we have developed a one-step continuous process. We know the basic principles to trigger lipid accumulation and to produce biomass continuously. However, to facilitate lipid production, higher biomass and lipid productivity may be achieved continuously in a single step process. First, we have worked on the in depth understanding of lipid metabolism in three microalgae species (*Phaeodactylum tricornutum, Nannochloropsis oceanica*, and *Acutodesmus obliquus*). Next, at lab scale a proof of principle concept for a one-step lipid production process was developed.

After proof of concept within controlled indoor conditions, the continuous process was tested outdoors under real production conditions in four different regions (NL, IL, IT – small scale test plants; ES – large scale demonstration plant) with different climates and two microalgae strains with different optimal growth temperatures (*Phaeodactylum* and *Nannochloropsis*) to investigate optimal growth conditions (temperature, sunlight, scale), the effect of optimal growth temperature on the energy balance and the robustness and reliability of the production process.

Conversion process

Simultaneous with research on biomass production, a continuous downstream process was developed. The conversion process consists of different steps (harvest, cell disruption, primary extraction, fractionation, conversion, hydrotreatment). For each step, the most promising technologies presently used in other industrial sectors, were applied to microalgal biomass. Where necessary, adaptations have been made. Important criteria for the selection of the technologies were low operational costs and low energy consumption. It is important to find the right balance between costs and energy consumption. In order to lower the operational costs of the total conversion process, we have designed a continuous conversion process

² Ruiz et al. (2016) Energy Environ. Sci. 9: 3036-3043

³ Wijffels RH, Barbosa MJ (2010) Science 329:796-799

⁴ Wijffels RH, Barbosa MJ, Eppink MHM (2010) Biofuel Bioprod Bior 4:287-295

⁵ Xu L, Brilman DWF, Withag JAM, Brem G, Kersten S (2011) Biores Technol 102:5113-5122

that uses all components of the algal biomass. Besides biofuels, high-value lipids (PUFAs) are valorised. From the remaining biomass, hydrogen is produced which is used in the conversion process of lipids into biofuel.

<u>Integration of production and conversion processes</u>

In order to facilitate the continuous production of biofuels from microalgae, the continuous process for algal biomass production with high lipid content has been coupled to the continuous downstream process to convert the biomass into biofuel, high-value lipids and hydrogen that is used within the biofuel conversion process. We have established guidelines describing the process requirements and parameters (costs, energy consumption, characteristics of intermediate products, etc.), ensuring the right fit between the different process steps and providing the information required to design a fully integrated process chain. Finally the whole process (both biomass production and conversion into biofuel) was integrated and subjected to a sustainability assessment, including environmental, economic and social elements based on the whole value chain in a life cycle perspective. The results have been used to guide the technical development and future implementation in the desired direction and to be able to determine economic feasibility and environmental sustainability.

Partnership

In order to achieve the overall aim and the specific objectives, an excellent consortium has been established. The multidisciplinary team includes partners who are leading in their work field. Specialists from both algae production and algae conversion were brought together. The consortium consisted of a powerful mix of established research organisations and universities, small and medium enterprises (55% of total partners) and large-scale industry. These are: Wageningen Food & Biobased Research, Wageningen Plant Research, Wageningen University, Ben Gurion University of the Negev, Fotosintetica & Microbiologica S.r.l., Norsker Investigationes, Proviron, Evodos B.V., Cellulac, Feyecon Development and Implementation B.V., Neste, JOANNEUM RESEARCH Forschungsgesellschaft mbH and IDconsortium SL. (see also http://www.fuel4me.eu)

Final results and their potential impact and use

Understanding and improving lipid metabolism

~Omics analyses of *Phaeodactylum tricornutum* have identified genes, proteins and metabolites that are differentially expressed under nitrogen replete vs. nitrogen limited conditions. Key biological pathways include nitrogen and amino acid metabolism, fatty acid metabolism, membrane and neutral lipid metabolism and pigment biosynthesis. Proteomics analysis resulted in identification of novel lipid-droplet-associated proteins in *P. tricornutum*. Preliminary results suggest that overexpression of two of these proteins may result in increased lipid production in *P. tricornutum*. Finally, lipid droplet formation, fusion and movement seem to be controlled by the microtubule system, as indicated by inhibitor studies.

Next to insight in the lipid metabolism, the work in this project has brought forth Mapomics, a new tool for integrative transcriptomics, proteomics and metabolomics analysis. Mapomics has been implemented as a Galaxy server instance for the support of FUEL4ME and future research focussed on omics integration.

The insights and tools gathered in this task will be used in future projects aimed at targeted metabolic engineering for improved lipid productivity in microalgae.

Comparison of batch versus continuous process

Traditionally, two-step batch mode cultivation (Nitrogen starvation) is adopted for the production of microalgal lipids. In this cultivation mode, biomass is first grown under optimal conditions, after which the

cells are induced to accumulate lipids by nitrogen starvation. For the first time, we have shown that production of microalgal lipids in continuous nitrogen limitation mode is, on average, in same range of the lipid productivities achieved in batch mode (both in lab and outdoor). Total fatty acid contents were generally higher in batch mode, especially when batch reactors were harvested well-beyond the moment at which maximum lipid productivity was achieved.

Varying results were obtained for robustness of either cultivation mode (*i.e.* in some cases batch was more robust, while in other instances continuous mode was more robust). Costs could not be properly assessed at the tested scale. However, in view of the comparable productivities achieved, costs (of entire process including downstream processing) should in the end be decisive for ultimately adopting one of the two cultivation modes. For this, further improvement and upscaling of both cultivation technologies is needed to improve, properly assess and finally compare the process economy. Key point for this analysis is also to take into account the compatibility of the cultivation technology with the downstream processing chain.

Conversion process

Researchers developed the various steps of the downstream processing chain, which involved harvesting, cell disruption, lipid extraction and fractionation and hydro-treatment of the lipids to create biofuel. They were shown to be successfully applicable to microalgae, have been demonstrated at industrial scale and are now ready to be used in commercial processes, which are not only limited to the production of biofuels.

<u>Integration of production and conversion processes</u>

A sustainability assessment, integrating environmental, economic and social aspects of the whole value chain, was carried out to determine the actual state of the technology as well as how key parameters influence the sustainability of the FUEL4ME integrated process. The assessment of the FUEL4ME integrated process includes the comparison to a reference system with the substituted conventional products. The conventional product for HVO (hydrotreated vegetable oil) is diesel, PUFAs would substitute high value lipids from fish oil, fertilizer would substitute synthetic mineral fertilizer; and for hydrogen, the conventional product is hydrogen from natural gas. The reference system provides the same services and products as the FUEL4ME integrated process with conventional products. Due to the state of technology and availability of data a modelling of a possible future full scale FUEL4ME integrated process with TRL 9 was developed to describe a possible future commercial HVO production from microalgae by giving guiding values and sustainability indicators for some key technological, economic and environmental data. The main influences upon sustainability were cultivation and harvesting, electricity demand, sources of freshwater and carbon dioxide, and suitable land. The research has addressed some of these parameters by improving productivity of cultivation, and harvesting efficiency, including water and resource re-use in cheap desert land. Further improvements, however, need to be made to make the process of biofuel production with microalgae fully economic and environmentally sustainable. The FUEL4ME integrated process could be economic viable and environmental sustainable because of the current TRL level of the FUEL4ME integrated process, and the future possible technology improvements in a long-term perspective. FUEL4ME believes that the opportunities and long term innovation strategy, with a stronger focus on high value products, will result in economically feasible and environmentally sustainable microalgae-based products.

3 Description of main science and technology results

WP 1 - Fundamental research and enabling technologies: genes, metabolism, biochemical aspects, bioprocess

Overall objectives of WP 1

- 1. Understand lipid metabolism in two microalgae species (*Phaeodactylum tricornutum* and *Nannochloropsis oceanica*). NB. After previous EU-review, *Acutodesmus obliquus* was added.
- 2. Develop proof of principle concept for a one-step lipid production process at lab scale

Key findings and conclusions:

1.1 Mapomics, a new tool for integrative transcriptomics, proteomics and metabolomics analysis was developed (figure 2).

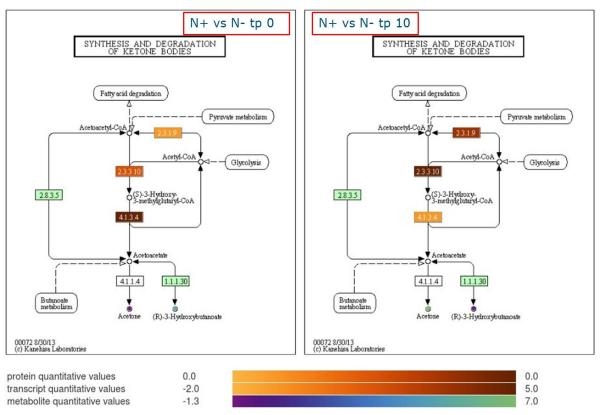


Figure 2. Pathway quantification and visualization by Mapomics. Expression of transcripts, proteins and metabolites are compared between nitrogen replete N+) and deplete (N-) growth conditions at different time points. Log fold changes as indicated in the legend below the plots are shown as coloured heat maps onto the nodes of specific KEGG pathways.

1.2 *P. tricornutum,* cultivated in continuous mode in either nitrogen replete or limited condition, exhibits differential expression of genes, proteins and metabolites that relate to key biological pathways, such as nitrogen and amino acid metabolism, fatty acid metabolism, membrane and neutral lipid metabolism and pigment biosynthesis.

1.3 Three *P. tricornutum* lipid-droplet-associated proteins (identified by proteomics analysis of isolated lipid droplets) were overexpressed in *P. tricornutum*, fused to Green Fluorescent Protein, using a starvation induced promoter. All three fusion proteins were found associated to lipid droplets after nitrogen starvation. Preliminary results suggest that overexpression of LD7 and LDA may result in increased fatty acid production, while LDE might actually inhibit fatty acid accumulation under nitrogen starvation. Finally, lipid droplet formation, fusion and movement seem to be controlled by the microtubule system, as indicated by inhibitor studies. (Figure 3)

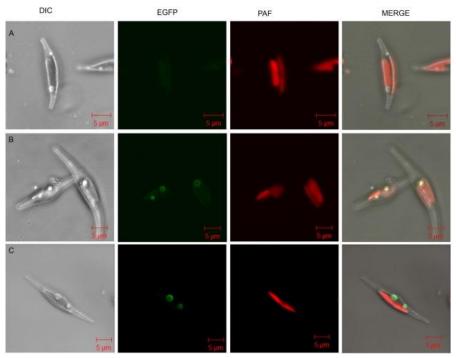


Figure 3. Green Fluorescent Protein fused to Lipid Droplet protein (EGFP-HOGP) associates with lipid droplets when expressed in *Phaeodactylum tricornutum*: Confocal micrographs of WT and two different transformant lines (C8 and C3) under conditions of Lipid droplets (LD) formation show green fluorescence signals (the green rings in rows 2 and 3) associated with LDs that are identified as grey-white balls in the pictures to the left. DIC: differential interference contrast microscopy; EGFP: EGFP-HOGP fusion protein fluorescence; PAF: chlorophyll fluorescence; merge: overlay of DIC with EGFP showing that GFP fluorescence exactly overlays with LDs. Top row: Wild type cells. Middle and bottom rows: two different clones expressing EGFP-HOGP.

- 1.4 Overexpression of a fusion protein (of Green Fluorescent Protein and an oil globule protein from *Haematococcus pluvialis*) increased TAG accumulation in *P. tricornutum* by over 50%.
- 1.5 In contrast to what is generally acknowledged, synchronized cell growth does not require major transitory energy storage molecules in *A. obliquus*, but the presence of such molecules does improve photosynthetic efficiency (figure 4).

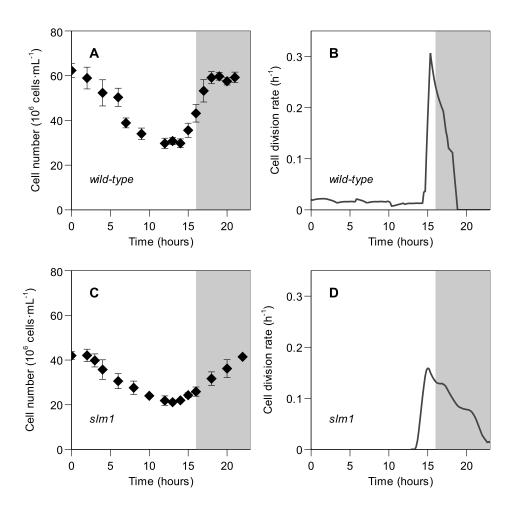


Figure 4 Cell number and Cell division rate for *A. obliquus* wildtype and starchless mutant (slm1). Light:dark (LD) cycles were applied with 16h of light and 8h of darkness (indicated by the shaded areas). The wild-type accumulates starch during the day, and consumed it during the night. It was always hypothesized that starch fuels cell division at night, but the starchless mutant also showed synchronized cell division to LD cycles. It therefore appears that starch is not required for synchronized cell division to LD cycles. We were not able to find any other temporary carbon source that fuels cell division at night for the starchless mutant.

- 2.1 Technical feasibility shown for continuous (one-step) lipid production with *P. tricornutum*, *A. obliquus* and *N. oceanica*.
- 2.2 Optimal batch and continuous mode cultivation conditions were determined for lipid production with *P. tricornutum*, *A. obliquus* and *N. oceanica*, to ensure fair comparison of both cultivation modes.
- 2.3 It depends on the species tested whether batch starvation or continuous cultivation under nitrogen limitation has highest TAG production at lab scale. For *A. obliquus* lower TAG content and TAG yield on light were observed for the continuous mode (table 1), whereas for *N. oceanica* higher TAG content and TAG yield on light were observed for the continuous mode. For *P. tricornutum*, the TAG yield on light was highest for continuous mode, while the TAG content was highest in batch mode.

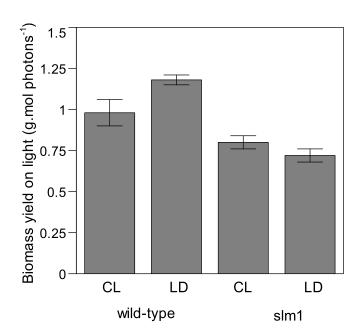


Figure 5 Biomass yield on light for *A. obliquus* wild-type and starchless mutant (slm1) for both continuous light (CL) and light:dark (LD) cycles. The wild-type shows improved photosynthetic efficiency under LD cycles. Contradictory, the starchless mutant does not benefit from the LD cycles. We concluded that starch is not required for synchronized cell division (Figure), but does lead to improved photosynthetic efficiencies.

Table 1 Comparison of maximum yield TAG yield on light with corresponding TAG content and productivity for batch and continuous cultivation with *A.obliquus* wt and slm. The TAG productivity and yield on light were calculated over the entire cultivation period, including inoculum production. Standard deviation was found to always be lower than 5%. *Data interpolation showed that maximum TAG yields of light of 0.14 g TAG·mol_{ph}-1 for the wt and 0.16 g TAG·mol_{ph}-1 for the slm1could be reached under continuous nitrogen limitation.

	Batch		Continuous	
	Wild type	Slm1	Wild type	Slm1
Maximum TAG yield on light (g·mol _{ph} -1)	0.16	0.20	0.081	0.115
Maximum TAG productivity (g·L ⁻¹ ·day ⁻¹)	0.19	0.26	0.110	0.157
TAG content at maximum productivity (% of dry weight)	23	44	16	33

2.4 Regardless of the species used, the differences between both cultivation modes are in such a range that other factors, such as operational costs and compatibility with the downstream process, will likely become decisive in adopting either batch or continuous mode.

WP 2 - Translation to outdoors and production

Overall objectives of WP 2

• The overall objective of WP2 is to translate the developed (WP1) one-step process to outdoors, thereby achieving a robust and reliable production process, continuous year round lipid production under different climates and weather conditions, and oil production of constant quality. WP2 is closely linked to WP3 for the production and delivery of the biomass for downstream processing activity and to WP5 for providing data for LCA.

Key findings and conclusions:

1) Three pilot plants and a demonstration plant were designed, build and operated. The pilot plants were built in Italy, Israel and The Netherlands, whereas the demonstration plant was built in Spain (figure 6).

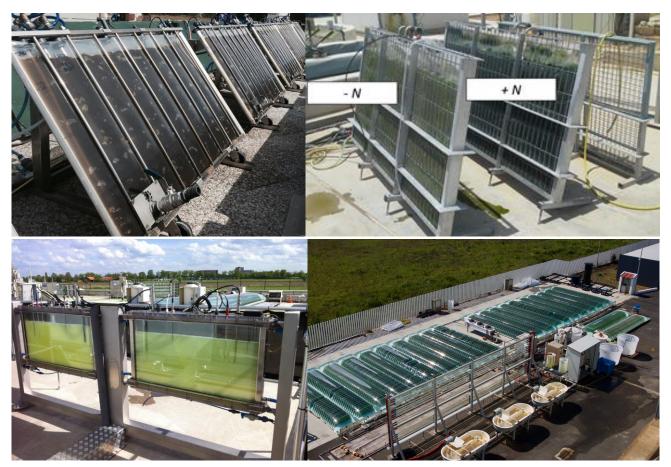


Figure 6. The three pilot plants designed, build and operated within the FUEL4ME project at 3 locations: upper left: flat panel photobioreactor (GWP II) at F&M, Italy; upper right: flat panel photobioreactor model reactors at BGU, Israel; bottom left: flat panel photobioreactor at AlgaePARC/Wageningen UR, The Netherlands; bottom right: demonstration plant in Huelva, Spain.

- 2) Research activity mainly focused on *N. oceanica* as it resulted the most promising strain in terms of TFA productivity and content with respect to *P. tricornutum*. *P. tricornutum* also presented recurring contamination and culture crashes that made difficult to cultivate it outdoors.
- 3) Outdoors, *N. oceanica* TFA productivity (g m⁻² ground d⁻¹) under a semi continuous N-limitation regime showed to be as productive as N-starvation and in some cases even better.
- 4) TFA productivity was inversely related with the amount of nitrogen supplied to the cultures. A clear effect of the available light on TFA productivity was observed, resulting the highest in Israel (IL) and the lowest in The Netherlands (NL). Having performed outdoor tests at different sites (NL, IT and IL) it was possible to evaluate which location better performed in terms of TFA productivities. By comparing TFA productivities, both per m² of occupied ground and m² of panel surface, obtained in the three pilots under comparable nitrogen supply regime (starvation and limitation) and period of the year, it is clear as solar radiation available played a fundamental role (figure 7, figure 8, table 2)

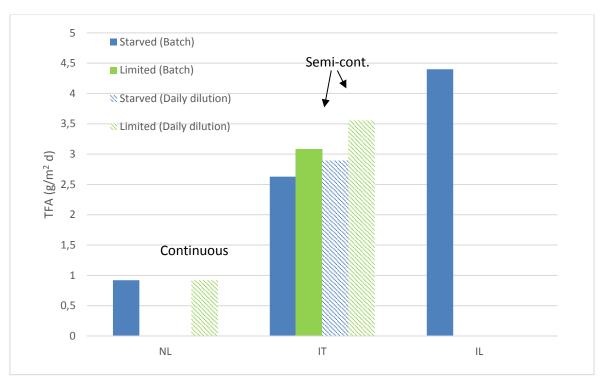


Figure 7. TFA productivities (g m⁻² ground d⁻¹) for the three pilots in a comparable period of the year. NL (April to June - $10 \text{ MJ/m}^2 \text{ d}$); IT (Average of May and September – $21 \text{ MJ/m}^2 \text{ d}$); IL (June – $30 \text{ MJ/m}^2 \text{ d}$)

Table 2 Average monthly solar radiation (MJ/m²day) for horizontal surface (source: http://re.jrc.ec.europa.eu/pvgis)

	NL (Wageningen)	IT (Florence)	IS (Sde-Boker)
January	2.4	5.4	12.0
February	4.5	8.6	14.9
March	9.5	13.2	20.9
April	15.5	17.6	23.4
May	18.0	22.1	26.9
June	19.4	24.7	29.8
July	18.4	26.1	29.0
August	15.1	22.4	26.8
September	11.0	16.5	23.1
October	6.5	10.6	18.8
November	3.0	6.2	13.9
December	1.9	4.7	11.6

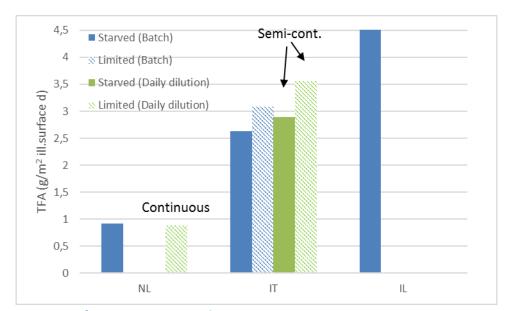


Figure 8. TFA productivities (g m $^{-2}$ illuminated surface d $^{-1}$) for the three pilots in comparable period of the year. NL (April to June - 16,MJ/m 2 d - 75 days); IT (Average of May and September : 21 MJ/m 2 d - 8 days); IL (May - 26.4 MJ/m 2 d - 9 days)

- 5) The highest TFA content with *N. oceanica* (% d.w.) has been obtained under N-starvation, when N-limitation was applied TFA content was generally lower.
- 6) Continuous N-limitation was found to be much more stable and easy to maintain with respect to starvation in The Netherlands (WFBR, figure 9). This finding is in contrast to what obtained by F&M (Italy) when daily dilution (semi-continuous) was applied, in fact TFA productivity of daily diluted culture under N-limitation was not stable and decreased with time. A higher stability could be obtained by reducing nitrogen limitation at the expenses of TFA content and productivity.

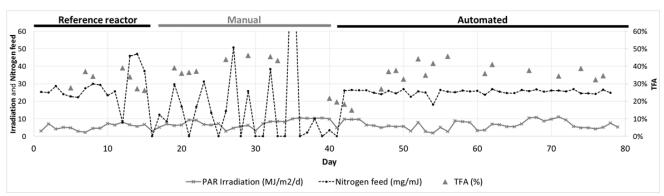


Figure 9: Continuous outdoor N-limitation cultivation with 3 different strategies: reference reactor, manual addition and automated addition for a total period of ~3 months.

- 7) Inputs (energy and materials) and outputs (biomass and TFA content and productivity) were collected in the pilots, and at F&M and WFBR the data were used to elaborate an energy and mass balance for a 1-ha and 100-ha scale plants. These data were then used by WP5 partner for the LCA analysis. Mass and energy balances results were reported in details in the following deliverables: D2.13; D2.15 and D2.17. In order to standardize as much as possible the energy inputs data for the LCA, a methodology for the extrapolation of the data, from the pilot plant scale to the 1-ha and 100-ha plant, was elaborated. This mainly focused to the identification of reference efficiency values, characteristic for each type of pumps and blowers. The activity was performed by all WP2 partners, particularly with the contribution from BIT/NIS and the outcome is reported in D2.7.
- 8) Biomass for downstream processing was produced. A first batch (48 kg) of frozen paste of *N. oceanica* was produced from 11.03.2016 to 10.04.2016 in 20 ProviAPT reactors (D2.14). During the FUEL4ME General Assembly held in Olhão in April 2016, the problem of the lack of large amount of lipid-enriched *Nannochloropsis* biomass for the downstream process was addressed. During the meeting, the quotations received for the production of the biomass outside the consortium were evaluated and considered too expensive to be afforded. Moreover the availability of good lipid-enriched biomass was not certain. As a possible solution, the partner F&M, proposed the use of the biomass produced during the EU FP7 project BIOFAT, in which F&M participated. In fact, despite the use of different strains, the two projects investigated the same algal species: *Nannochloropsis oceanica* sp.2 in the FUEL4ME project and *Nannochloropsis oceanica* F&M-M24 in the BIOFAT project. Thus, the lipid enriched *Nannochloropsis oceanica* F&M-M24 biomass produced in Camporosso Pilot Plant for the purposes of BIOFAT (collection of data on Pilot Plant performances) was harvested and stored in a frozen form and it was made available for FUEL4ME project.

WP 3 – Downstream processing and conversion of oils to biofuels

Overall objectives of WP 3

 The development and integration of an innovative and continuous downstream process for conversion of microalgae into biofuels with consistent volume and quality

Key findings and conclusions:

- 1) Harvesting of *Nannochloropsis* and *Phaeodactylum* was successfully demonstrated at F&M in Italy with the Evodos type 10. For both microalga the maximum separation efficiency was 95%, and the time required for reaching this separation efficiency was 45 min and 80 min for *Phaeodactylum* and *Nannochloropsis*, respectively. The harvested biomass paste contained 26% solids for *Nannochloropsis* and 22% for *Phaeodactylum*. The prototype centrifuge worked at a maximum input flow of 537 l.h⁻¹ for *Phaeodactylum*.
- 2) Improved Evodos 25 was developed with automated discharge system. Harvesting of *Nannochloropsis* was successfully demonstrated at NIS facility in Huelva-Spain (figure 10). Separation efficiencies above 92% in combination with capacity up to 1200 l.h⁻¹ was achieved and an algae paste with concentration above 20% with less than 1% loss was obtained (figure 11).



Figure 10. ProviAPT demonstration plant in Huelva, Spain at partner NIS



Figure 11. Left: The Evodos 25 with automated discharge system; Middle: the spiral plate technology; right: the harvested paste

- 3) A membrane filtration unit has been developed for the demo plant at Biotopic in Huelva-Spain for capacities up to 600 litres/ hour. The membrane system was used for re-cycling of effluent water. During the test with effluent water an average flow 600 litre/hour with effluent water from a *Nannochloropsis* harvesting process with Evodos algae harvester was obtained and no fouling observed. By using the re-cycling system a yield of 97.6% was achieved.
- 4) A continuous high throughput cell cracking system with capacity of 50-60L/min of algae biomass was built and the system was able to facilitate mechanical cell disruption before further downstream processing and the experience of using SoniqueFlo for microalgae provided data for sustainability assessment that identified options for further up-scaling, implementation and commercialization of the SoniqueFlo technology for cell disruption (figure 12).



Figure 12. Small (left) and large (right) rig of the Soniqueflo by Cellulac, used for cell cracking

- 5) FeyeCon developed analysis method for characterization of oils, fatty acids and type of lipids (phospholipids, glycolipids and triacylglycerol). The results showed the strain and method of cultivation has significant effect on types of lipids and fatty acid profile. The lipids were extracted by using supercritical carbon dioxide technology and yield up to 46% was achieved from TAG-rich biomass. The effect of starvation on extraction yield was studied and it showed that the yield of fatty acids extraction in starved cells was 25% lower which might be due to stronger cell wall.
- 6) A process for extraction of lipids by using ethanol as solvent was developed and optimized and the extract was successfully trans-esterified. The ethyl esters were fractionated by using supercritical carbon dioxide fractionation column and a separation up to 78% was achieved.
- 7) Several batches of biomass were extracted by using supercritical CO₂. The effect of pressure, temperature, solvent to feed ratio, density and time was studied and extraction yield above 80% was achieved. For the TAG-rich *Nannochloropsis* more than 95% of the TAG was extracted by using only CO₂ as solvent. The extracted oil has high free fatty acid content which is due to hydrolysis of lipids during storage and shipment. Free fatty acid content of oil was reduced to 1% by using CO₂ refining column.
- 8) The fatty acid analysis of biomass and extracted oil showed the LC-PUFAs (EPA) remains mainly in biomass due to polarity of the molecule that EPA is attached to. The EPA content of extracted oil is below 8% which is too low to be separated by using fractionation technologies.
- 9) The esterified fatty acids including EPA were fractionated successfully by using supercritical carbon dioxide as solvent (figure 13). The fractionation process parameters including feeding rate, pressure, temperature, reflux ration and solvent to feed ratio were optimized and a fraction of LC-PUFA rich raffinate with concentration of above 70% and a low value fraction of short chain fatty acids in ester form was obtained. The process were performed in continuous mode and the robustness was tested in a 60 hours non-stop running. The process showed good stability during the process.
- 10) Neste used three algae oil samples including 2.7 L of extracted oil from TAG-rich *Nannochloropsis*. The analysis results showed all samples were suitable for NEXBTL process in terms of impurities. However, chlorine level in sample 3 was high. Oil sample 3 was pre-treated in the laboratory according to general procedure available at commercial NEXBTL plants. And pre-treated algae oil was tested in microreactor-scale at Neste to produce NEXBTL products. The microreactor testing simulated the process conditions in existing commercial NEXBTL-plants. Testing was done with blend of 20 % algae oil with 80 % of regular NEXBTL feedstock. Blending ratio was determined by chlorine content of the sample. The results showed measured properties of samples containing algae oil are similar with sample of typical NEXBTL feed.

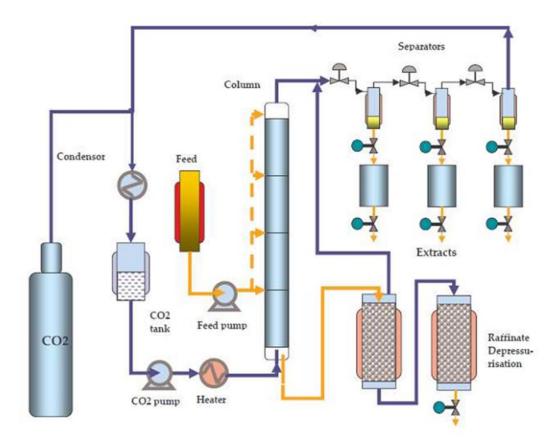


Figure 13. The fractionation process based on supercritical CO₂ extraction

- 11) De-oiled biomass was analysed. Components of value in the biomass residue were the carbohydrates (26%) and proteins (30%). The protein fraction of the biomass residue might have application as feed. Carbohydrates was used for the production of hydrogen (H₂) which is subsequently applied in hydrotreatment of the algal lipids for the production of biofuels. For fermentative H₂ production the biomass residue was enzymatically hydrolysed (by cellulase activity) thereby converting polysaccharides (mainly glucan) to soluble sugars. The fermentability of the hydrolysate with a sugar concentration of 18 g/l (a mixture of glucose, galactose, oligomeric galactose and mannitol) was tested using two strains of extreme thermophilic hydrogen producing bacteria. Next the H₂ yield and productivity were determined using the same thermophiles in fermentations under controlled conditions in a bioreactor. The H₂ potential of the algal biomass residue was 3.7 g of H₂ per kg of dry matter. This is circa 50% of the maximal feasible amount. Further optimization of carbohydrate conversion efficiency and H₂ fermentation performance is necessary.
- **12)** In conclusion it is shown all the process steps to work: harvesting, re-use of water, cell disruption, extraction and further fractionation of lipids, conversion of low-value lipids into biofuel, quality of high-value lipid fraction after fractionation, fermentability of remaining biomass into hydrogen.

WP 4 – Demonstration at pilot scale

Overall objectives of WP 4

 Demonstrate the capability of the optimised process (cultivation and downstream processing) at pilot scale under representative industrial conditions, based on the technology and methodologies developed in the previous WPs.

Key findings and conclusions

- 1) The integration of all steps from biomass production in continuous mode to harvesting, re-use of effluent water, cell disruption, extraction of triglycerides, conversion of triglycerides to biofuel, fractionation of high value fatty acids from low value fatty acids and conversion of carbohydrates in biomass to H₂ by fermentation were performed based on optimized conditions in WP2 and WP3.
- 2) Each process step has been performed with industrial-like equipment.
- 3) Continuous cultivation: 20 reactor units were used for cultivation of TAG-rich microalgae in continuous mode and total harvest from the system during the production period was 40.2 kg DW with an average biomass concentration of 2.3 g/L. Daily average dilution rate was 5 %.
- 4) Harvesting: *Nannochloropsis* was harvested by modified Evodos 25 with an efficiency of >90%, capacity > 6 m³/day, dry weight >20% and with minimized paste losses <1% of the total volume.
- 5) Re-use of effluent water: Average flow 600 litre/hour, effluent water from a *Nannochloropsis* harvesting process with Evodos algae harvester, No fouling observed and a yield of 97,6%, was obtained.
- 6) Cell cracking and extraction of TAGs in demonstration scale: by proper pre-treatment 40kg of lipid-rich *Nannochloropsis* (produced by EU project BIOFAT) was extracted by using CO₂ as solvent and more than 90% of TAG was extracted. The free fatty acid content of extract was reduce to below 1% by using CO₂ column and co-extracted water and solid particles were separated from the extracted oil.
- 7) Fractionation of fatty acids: LC-PUFAs were fractionated by using CO_2 as solvent and purity of 72% was achieved. The process was performed in batch and continuous mode and a 60 hours run was carried out in order to evaluate the robustness.
- 8) Conversion of low value TAGs to fuel: the extract was further purified and tested in micro-reactors to simulate the NEXBTL process. The results showed the algal oil can be used for biofuel production.
- 9) Conversion of biomass to hydrogen (H_2) for the hydro-treatment step: the sugars present in biomass after extraction of oils was converted to H_2 . A production of 7.6 g of H_2 per kg should be feasible by an optimized carbohydrate conversion of 90% and a H_2 yield of 75%.

WP 5 – Sustainability assessment of integrated process

Overall objectives of WP 5

Assess the sustainability of a continuous production and conversion process based on the work
performed in WPs 1-4, including environmental, economic and social assessment based on the whole
value chain in a life cycle perspective. The results were used to guide the technical development and
future implementation in the desired direction and to be able to determine economic feasibility and
environmental sustainability.

Key findings and conclusions

The key findings and conclusions are on sustainability assessment of FUEL4ME integrated system, modelling of a full scale commercial plant; and technology assessment. The results are described below.

Sustainability assessment of FUEL4ME integrated process

The sustainability assessment integrates environmental, economic and social aspects of the whole value chain of the FUEL4ME integrated process and is assessed in comparison to the reference system (Figure 13). Within the life cycle sustainability assessment the methodologies Cost assessment, Life Cycle Analyses (LCA) and social Life Cycle Analysis (s-LCA) were used and specifically adapted to the considered algae concepts. A list of indicators was identified: 14 environmental indicators, 14 economic indicators and 11 social indicators. Due to the relevance of the indicators especially for the FUEL4ME integrated process and the availability of data for their assessment the most applicable indicators for the assessment were identified, e.g.:

- Economic: e.g. production costs, annual costs, value added, employment, market aspects;
- Environmental: e.g. global warming potential, primary energy demand;
- Social: e.g. employment, supplier relationships.

Social indicators were described in a qualitative way.

The sustainability assessment of the FUEL4ME integrated process includes the comparison to a reference system which supplies the substituted conventional products. The reference system provides the same services and products as the FUEL4ME integrated process with conventional products. In Figure 14 the whole value chain of the FUEL4ME integrated process in comparison with the reference system with conventional products is shown. The conventional product for HVO (hydrotreated vegetable oil) is diesel, PUFAs would substitute high value lipids from fish oil, fertilizer would substitute synthetic mineral fertilizer; and for hydrogen the conventional product is hydrogen from natural gas. All production steps of the value chain are included in the reference system as well as for the FUEL4ME integrated process, e.g. for transportation services the following process steps are included: extraction of crude oil, transport to a refinery, processing in a refinery, distribution and the use in passenger cars.

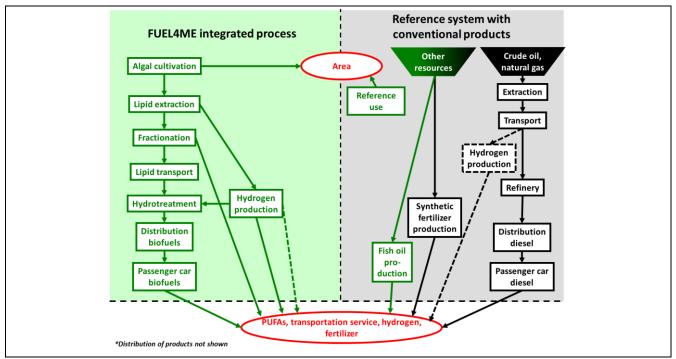


Figure 14: Whole value chain of FUEL4ME integrated process and reference system

Modelling of a full scale commercial and integrated microalgal-based process

Due to the state of technology and availability of data a model of a full scale FUEL4ME integrated process with TRL 9 (Technology Readiness Level) was developed to describe a possible future commercial HVO production from microalgae. The modelling was done using the methodologies LCA, cost assessment, assessment of macro-economic aspects and s-LCA. Three cases with a production capacity of 100 kt/a HVO and coproducing up to 5 kt/a PUFA were modelled. For the modelling targets, characteristics, data and assumptions were defined among project partners (Figure 15). The three main targets are:

- 1) Revenue ≥ costs,
- 2) Reduction of GHG emissions ≥ 60 % for HVO biofuel instead of diesel based on LCA methodology according to Renewable Energy Directive (RED)
- 3) Cumulated fossil primary energy demand in relation to HVO energy content ≤ 30 % (≤ 0.3 MJ_{fossil}/MJ_{biofuel}).

These targets give the possible framework conditions for a future full scale commercial plant. The results of the modelling of a full scale FUEL4ME integrated process are guiding values and sustainability indicators formulated as ranges for the most relevant technical, environmental and economic characteristics. These sustainability indicators show how the three targets are fulfilled simultaneously.

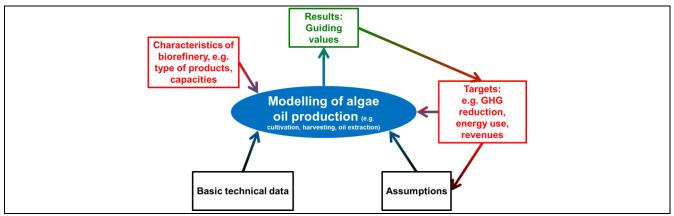


Figure 15: Modelling of an algae oil production

In Figure 16 the GHG emissions for a full scale integrated microalgal-based process (TRL 9) are shown on the left side. This figure provides information on the biochemical and chemical contribution to global warming by GHG emissions (process emissions). Biophysical impacts, displayed as equivalent GHG emissions are assessed within FUEL4ME as well. Biophysical impacts are due to the land use change and therefore the change in albedo, which is a coefficient measuring the reflection of a surface. Darkening the surface (lowering the albedo) causes more solar energy to be absorbed and so contributes to the global warming. The process emissions (biochemical GHG emissions) for the three cases were 18 − 25 CO_{2-eq}/MJ_{HVO}. The three cases required an area for algae cultivation of 4,000 - 15,000 ha to produce the 100,000 t_{HVO} per year. The equivalent GHG emissions due to albedo change are 5 − 19 g CO_{2-eq}/MJ_{HVO}, then the combined effect on global warming in equal emissions are 23 − 44 g CO_{2-eq}/MJ_{HVO} which are still less than emissions of diesel (84 g CO_{2-eq}/MJ). Figure 16 shows on the right side the results of the macro-economic assessment on employment. Expressed in absolute numbers the new algae-based technology leads to approx. 2,000 jobs at expenses of € 127 mio., whereas the reference system using conventional technologies − even if they can produce products cheaper − employment is approx. 1,500 to 2,000 jobs, which remains valid even at high price discrepancies between HVO and conventional fuels.

In FUEL4ME a methodology to assess the social aspects was developed to show at least in which categories and subcategories of the social dimension a company must be careful. Some of the most relevant social "hot spots" concerning the installation of a large algae cultivation system are identified in the category "society". These "hot spots" include the engagement with local citizens, local employment and transparency to foster the acceptance of the new technology. In Table 3 the indicators in the category "Regional corporate citizenship" relevant for regional cooperations are listed. For each indicator four possible situations are described; green – this situation should be reached, red – this situation should not be reached at all.

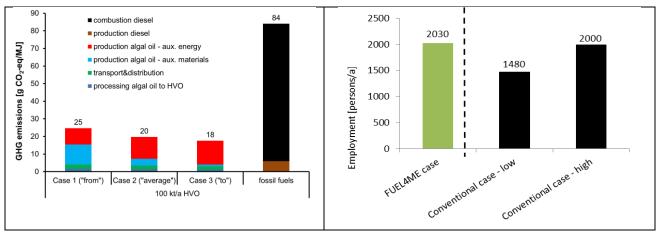


Figure 16: GHG emissions and effect from biophysical impacts (albedo) to GHG emissions in CO₂-equivalents (left side) and employment: impacts of analyzed cases (right side)

Table 3: Example for social indicator - Regional cooperation - of a full scale integrated microalgal-based process

Regional corporate citizenship	Assessment green: where we a red: no aim at all	Unit			
Regional cooperation					
Creation of jobs in the region	commitment to	create jobs in the	done by the company	Hardly no jobs in the	[number of jobs in the region]
Contracts with regional suppliers	contracts with regional suppliers	There is no strong commitment to sign contracts with	Not much effort is done to sign contracts with regional suppliers	There is no commitment for contracts with regional suppliers, the cheapest offer is favoured	[number of contract with regional

Technology assessment

After this modelling of a commercial plant (TRL 9) the actual demonstrated state of technology (actual TRL) of the FUEL4ME integrated process was assessed with following characteristics:

- location of the cultivation: South Europe,
- microalgae species: Nannochloropsis,
- capacity of biofuel production: 10,000 t/a, 100,000 t/a,
- size of one algae cultivation site: 1 ha, 100 ha,
- hydrogen (H₂) surplus: "yes" or "no",
- no protein separation and
- fertilizer production.

The assessment of the developed concepts shows that due to the actual state of technology the GHG emissions (biochemical only) and the primary energy demand of the FUEL4ME integrated process (current TRL) are higher as the values for the conventional reference system (TRL 9). The main influencing factors of the GHG emissions are the electricity demand especially needed for cultivation and harvesting, the yearly substitution of plastic films in PBRs, and the energy needed for drying the biomass. Furthermore the actual demonstrated state of technology is economically not feasible. The costs for personnel and for auxiliary energy contribute to a high share of the annual operational costs.

Due to actual demonstrated state of technology (current TRL) further technology development is needed. A comparison using the guiding values of the modelled full scale commercial and integrated FUEL4ME process

was made to analyse the gap between the current TRL and the guiding values for TRL 9. An assessment of the actual state of technology, based on the developed concepts, in comparison to TRL 9 is provided. In Figure 17 the two important parameters for a future full scale commercial microalgal-based process are shown: algal biomass yield $[t_{DM}/(ha*a)]$ and electricity demand $[kWh/t_{DM}]$.

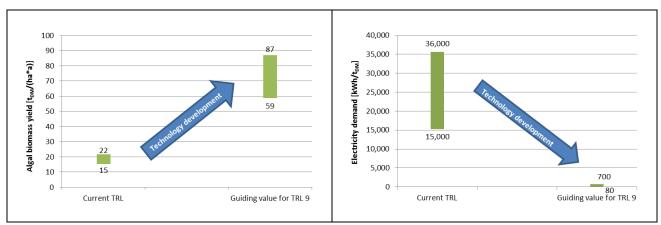


Figure 17: Comparison of the state of technology with the full scale integrated microalgal-based process

In comparison to the FUEL4ME integrated process cultivation parameters are listed for two other FP7 projects (BIOFAT and InTeSusAI) (Table 4). The data for the two other FP7 projects is based on information from project members of BIOFAT (BIOfuels From Algae Technologies) and InTeSusAI (Demonstration of integrated and sustainable microalgae cultivation with biodiesel validation). The parameters include extrapolated biomass production per cultivation ha, depending on the biomass productivity, operational days and irradiation per location of the cultivation.

Table 4: Comparison of cultivation parameters of three FP7 projects

Parameters	Unit	FUEL4ME*	InTeSusAl*	BIOFAT**
Extrapolated biomass production per cultivation ha	[t _{DM} /(ha*a)]	15 - 22	~20	35 - 40
Biomass productivity	[g/(m²*d)]	5 - 7	~6	9 - 13
Operational days	[No.]	300 - 339	300	365
Location/irradiation	[MJ/m2 *d]	Spain (15.5)	Portugal (18,7, Olhão)	Portugal (17)

^{*}cultivation ha includes area for PBRs, inoculum production, equipment

Cellana, a company located in Hawaii, United States, intends to construct and operates commercial facilities to produce Omega-3 EPA and DHA nutritional oils, food/animal feed, and biofuels. Therefore they are cultivation marine microalgae in open ponds.

At the company website (Cellana, 2017) following basic data for microalgae can be found. But it is not stated if these data are general information or based and calculated on their own shown biomass productivities:

- biomass productivity: 50 150 t/(ha*a);
- oil content: 20 50% of dry mass;

^{**}cultivation area includes only area for PBRs

biofuel productivity: 10 – 75 t/(ha*a).

A comparison with FUEL4ME data is difficult, due to the lack of additional data, e.g. biomass productivity per g/(m²*d), operational days, detailed information on the assumed cultivation area – with our without surroundings. Data on costs were not found. Furthermore the production system (open ponds vs. flat panels photobioreactors) and the location (Hawaii vs. Southern Europe) are completely different.

Comparison with other microalgae projects or processes is not very well possible as data is not simply available in the same units and with detailed background information. Furthermore production systems and products are not similar to the FUEL4ME process and calculation methods are not explicitly mentioned to get more detailed information.

FUEL4ME co-organised with the European Algae Biomass Association (EABA) and with the EU projects Miracles, Splash and the Algae Cluster (InteSusAl, BIOFAT and All-gas) the Conference "European Roadmap for an Algae-Based Industry" in April 2016 in Olhão, Portugal. As a result of the conference a Whitepaper (FUEL4ME partners are co-authors) will be published. In this Whitepaper essential needs of the sector will be identified and the direction of the European algae strategic research agenda for the upcoming years will be pointed out. Research areas that need to be addressed in the future are included in the fields of markets, production technology, industrial strains and biorefinery. This information was furthermore used as the basis for the roundtable discussion during the workshop "Algae Research Agenda for 2020 and 2030" held in Madrid 13-15 December 2016 (AlgaEurope 2016 Conference).

In future the FUEL4ME integrated process could become economic viable and environmental sustainable with future possible technology improvements in a long-term perspective. The possible future technology improvements in a large scale operation are expected in following aspects:

- Algae strain improvement:
 - o Higher yields
 - More efficient land use and
 - o Reduction of electricity demand
- Innovation in cultivation technologies:
 - Lower personnel demand due to possible automatization (it seems to be possible to get to 100 – 200 persons per 1,000 ha)
 - Lower electricity demand of PBRs— a possible reduction to 10% of the current TRL could be reachable, more focus should be on reduction of electricity demand, decrease of electricity demand could be possible by optimization, higher productivities and bigger scale of cultivation
 - o Larger scale installations with continuous production
 - Reduction of the high impacts from embodied energy in the cultivation system

The FUEL4ME partners will further work to realize the opportunities and long term innovation strategy with a stronger focus on high value products, to develop economic feasible and environmentally sustainable microalgae-based products in a long term perspective.

Key performance indicators for the FUEL4ME project

In table 5 we show the targets for FUEL4ME and how far we have reached our ambitions, based on the Key Performance Indicator list as determined at the start of the project.

Table 5 Key performance indicators

Key performance indicators	Baseline	Target FUEL4ME	Reached	% reached
	Technica			
TAG yield on light in lab-scale	0.15 g TAG / mol photon	0.3 g TAG / mol photon	0.20 TAG / mol photon	33%
reactors	(TAG = triacylglycerides)			
Outdoor lipid productivity	17 ton lipids ha ⁻¹ y ⁻¹	31 ton lipids ha ⁻¹ y ⁻¹	16 ton TFA ha ⁻¹ y ⁻¹	-7%
Harvesting:	90%	95%	95%	100%
Separation efficiency				
Harvesting:	150	200	200	100%
solids in paste (g L-1)	75	00	0.5	670/
Extraction yield (% of extractable lipids)	75	90	85	67%
Fractionation efficiencies	55% LC-PUFA (EPA and	80% LC-PUFA (EPA and	85% LC-PUFA (EPA and ARA) in	120%
Fractionation emclencies	ARA) in bottom fraction	ARA) in bottom fraction	bottom fraction	120%
	60% C16+C18 in top	72% C16+C18 in top	80% C16+C18 in top fraction	
	fraction	fraction		
Freehooding of C. C. C.			Character have 111	A - l-1 1
Evaluation of fractionated	Non-proven	Algae oil is of such quality	Shown to be possible.	Achieved
algae oils as suitable for NEXBTL		that it can be processed		
NEABIL		to NEXBTL with existing process		
	Economic	<u> </u>		
Costs for biomass production	> 6€ kg ⁻¹	< 2€ kg ⁻¹ *	3.4 € kg ⁻¹	35%
		- 0	Incl. harvesting	
Production cost of biofuel	-	<1€ L ⁻¹ *	Approx. 0.20 € L ⁻¹	Achieved
			If we assume co-products to	
			pay for other processes, excl.	
			harvesting	
	Sustainabil			
Energy use (NER = energy	Biomass production	Biomass production	0.65	23%
content of biomass / fossil	process with a NER of 0.4	process with a NER > 1.5		
energy used) *		> COO/++ 1+++	N. I. I.	AL
GHG reduction (biofuel)	-	≥60%** and ***	No reduction compared to reference system	Not achieved
Cumulated fossil primary		≤0.3 MJ _{CEDfossil} /MJ _{biofuel} **	>20 MJ _{CEDfossil} /MJ _{biofuel}	Not achieved
energy demand (biofuel)		and ***	>20 IVI3CEDTOSSII/ IVI3DIOTUEI	Not acmeved
0, ()	Disseminat			
Number of scientific articles	-	6 for WP 1	Pub: 1/ In prep:7	Achieved
		3 for WP 2	Pub: 0 / In prep: 4	
		1 for WP 5	Pub: 0 / In prep: 1	
Visits to website	Actual figure 3880	20.000	17.000	81%
Twitter: followers and posts	Actual figure Followers:	Followers: 400	Followers:258	49%
	124 Posts 20	Posts 500	Posts: 75	11%
Attendees to final FUEL4ME	-	100	>100	101%
conference				
Number of university courses	-	4	4	100%
on microalgae technology				
organized		4	Many 1.1 official aggregated to	Achious
Visits from industry to the	-	4	Many + 1 official organised to	Achieved
pilot plants			Demo plant	

^{*} calculated for a production plant in a (pre)commercial-scale size based on data achieved in FUEL4ME project (NER calculated without taking embodied energy into account)

^{**} calculated according to RED

^{***} calculated in analogy to calculation of GHG reduction in RED

4 The potential impact and main dissemination activities and exploitation of results

Dissemination of the foreground

Dissemination strategy

The main objectives of communication and dissemination activities have been:

- To enable an effective and fluent communication within the project and between project partners and other actors relevant to FUEL4ME purposes
- To ensure the widest dissemination of project results possible, so they reach their highest potential impact on society
- To raise awareness among stakeholders, general public, the scientific community, industry and public authorities
- To pave the way for the future commercialization and deployment of the technologies and processes being developed in FUEL4ME.

The dissemination and communication strategy intended to establish the proper channels, materials and activities to meet these objectives by:

- Setting up a Dissemination and Communication infrastructure: website, social networks, intranet
- Defining a graphical identity for FUEL4ME project
- Producing effective communication materials: project roll-up, brochure
- Keeping stakeholders informed about project activities and results: e-bulletin, publications on website and social networks.
- Organising and participating in activities to provide the project with opportunities and channels to share their results and interact with different audiences: participation in international conferences, organization of visits to the cultivation plants during the last year of the project
- Identifying relevant stakeholders and their interests in the processes and technologies being optimized in the project, and so better customize the dissemination activities and gather information for exploitation purposes.

More detailed information on the communication and dissemination strategy, channels, audiences and messages can be found on FUEL4ME deliverable *D6.1Communication and dissemination plan*. Table 5 summarizes the main target groups per communication channel.

Table 6. Main target groups per communication channel

Target Group/Communication Channel	EC	Authority	Industry	Research sector	General public
Website	х	x	х	Х	х
Deliverables -restricted	х				
Deliverables -public	х	х	X	х	х
Technical & Scientific Publications			Х	х	
Dedicated workshops, Symposia, etc.	х	х	Х	х	
Trade shows		х	х		
Technical fairs		х	х		
Congresses	х	Х	х	х	
Stakeholder forums		Х	х	х	
Social media			х	х	х
Audiovisual media					х
Posters, flyers, leaflets, etc.		х	х	х	х
Printed & online press					х

Dissemination materials and activities

Website

FUEL4ME website was designed considering the inputs of all partners regarding the sections to be included, the layout and the functionalities. It has received over 16000visits⁶ from all over the world, as shown in figure 17. The number of visits considered in the actual report are the visits that have stayed in the website longer than one minute.

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⁶ Total number of visitors on 23th December 2016

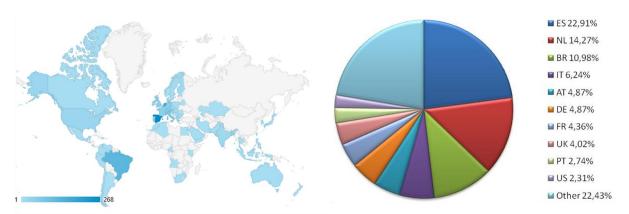


Figure 17: Geographical precedence of FUEL4ME website visitors

The website presents a complete description of project concept, activities, objectives and consortium partners, as well as collection of FAQs and it's updated with news on the progress of the project.

It also serves as a platform to keep stakeholders and FUEL4ME followers informed about the progress of the project through the regular publication of articles, posts and updates on the activities of the different WPs. Besides, dissemination materials and e-bulletins can be downloaded directly from the website. A calendar shows the most important upcoming events of the project.

The website contains a link to an intranet available for FUEL4ME partners, where all project documents from different WPs are shared and managed. The website is also linked to the Facebook and twitter profile of the project. It can be visited at www.fuel4me.eu.

After the end of the project the website will be kept and maintained by IDC for an additional 1 years (31st December 2017) to ensure visibility of the project results after the end of the project.

Brochure and roll-up

FUEL4ME brochure is a 3-folded leaflet containing an overall view on FUEL4ME project partners, activities and goals. 2000 copies have been distributed among the partners, so that they can provide their own contacts with some information about the project.

FUEL4ME roll-up has been designed to be taken to be taken to the congresses and conferences where consortium partners participate, as well as to be displayed in all the events where FUEL4ME is present. One roll-up has been provided to each partner. They are 2 metre high x 1 metre wide, made in polyester supported by an Aluminium structure.

Figure 18 shows the outer and inner sides of the brochure and roll-up.



Figure 18: FUEL4ME brochure and roll-up

In addition to the aforementioned brochure and roll up, a final brochure will be distributed at the end of the project with the goal of disseminating project results to stakeholders, the scientific community, the European commission and other private and public entities (figure 19). Future steps and recommendations to advance in the development of sustainable, scalable process for biofuels from microalgae and to valorise the by-products will be presented in the form of a several page booklet.



Figure 19: FUEL4ME final booklet

Stakeholders' list

With the purpose of ensuring an effective flow of communication within the project, an internal contact lists with all project partners has been set.

FUEL4ME external contact list gathers more than 400 hundred stakeholders from academia, industry, organizations and public entities from sectors ranging from different sectors related to the project activities. All partners have contributed to create this list, providing the contacts from their local and international stakeholders.

FUEL4ME has contacted two related EU-funded projects, SPLASH and MIRACLES, both coordinated by Wageningen UR and has worked together with them to create common algae stakeholders' list with over 1000 contacts. These contacts can be filtered according to:

- Project:
 - o FUEL4ME
 - o MIRACLES
 - SPLASH
- Type of entity:
 - Algae feedstock suppliers
 - Industry

- Human food
- Feed
- Fuels
- Pharmaceutical
- Biotechnological
- Fertilizers
- Bioplastics
- University and R&D
- Public administration
 - EU
 - National
 - Local
- NGOs and associations
- o Press
- Consulting
- EU related projects

E-bulletin

FUEL4ME **e-bulletin** is distributed among FUEL4ME partners, stakeholders and external contacts every six months, informing them about the progress of each WP, the most recent project activities and the upcoming events where the project is participating.

All project Newsletters can be found on the FUEL4ME <u>website</u>. They have been sent to over 400 relevant external entities (public authorities, industrial stakeholders, universities and research centres, NGOs, etc.)

Social networks

FUEL4ME has opened a profile in three social networks: <u>Twitter, Facebook</u> and <u>LinkedIn</u>. The kind of information published is customized to the functionalities and audiences of these different networks.

Twitter and Facebook are used to publish short posts on FUEL4ME latest activities, conferences where the project is presented, links to interviews to project partners, related EU projects, videos on our microalgae production facilities and relevant news and findings in the fields of microalgae biomass production and/or conversion to biofuels, photobioreactors and new innovative cultivation sites.

LinkedIn group is meant to be a forum to invite partners and stakeholders to foster discussion on relevant topics, such as the prospective for the use of biofuels in different fields or its share in the energy mix, as well as to inform them about interesting project results.

Twitter has proven to be a very good channel to be in contact with the main actors in the field of renewable energies, algae biomass production and biofuels. So far, FUEL4ME follows 135 entities, has 145 relevant followers belonging to the mentioned fields. Publications are published on weekly basis.

After the end of the project these profiles will be kept public but with minimal maintenance until they are no longer relevant.



Figure 20: FUEL4ME twitter profile

International Course on Microalgae Process Design

The Bioprocessing Engineering group from Wageningen University, in cooperation with the Graduate School VLAG and BioSolar Cells has organized in July 2013 and 2014 an *International course on Microalgae Process Design. From cells to photobioreactors,* which was also available for summer 2015.

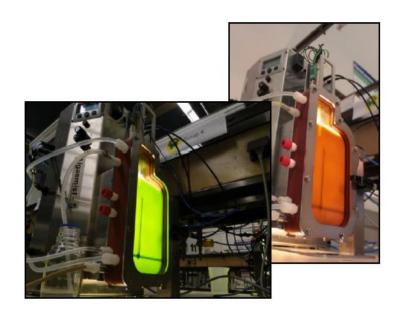
The **aim** of the course is to provide the essential skills for designing optimal microalgae-based production processes, for both research and commercial purposes.

The course is **targeted** at PhD candidates, postgraduate and postdoctoral researchers, as well as professionals, that would like to acquire a thorough understanding of microalgal metabolism and photobioreactor design. An MSc level in bioprocess technology, or alike, is recommended.

Course motivation and background

The waters of the world, oceans, seas, rivers, creeks, lakes and even ice, house a tremendous variety of microorganisms able to use light as the only source of energy to fuel metabolism. These unicellular organisms, microalgae and cyanobacteria, have the potential to produce a variety of products.

Among them are therapeutic proteins, polyunsaturated fatty acids and pigments. In addition to these high-value products, microalgae are regarded as one of the most promising resources for the production of bulk products such as food and feed lipids, food and feed proteins,



base chemicals and energy sources for the industry, and possibly even biofuels. To make economical large-scale production of such bulk products possible, optimal design of bioreactor and cultivation strategy are

essential. This process design has specific demand because sunlight, or artificial light, is used. The figure above shows small scale, fully controlled, photobioreactors containing green microalgae that are cultivated either under optimal conditions (left) or under conditions that stimulate accumulation of an orange pigment (right).

Course contents

Through lectures, digital cases and a photobioreactor practical, the participants will learn: 1) how to describe microalgal metabolism quantitatively, 2) how to apply basic design principles and set up mass/energy balances for photobioreactors, 3) how to cultivate microalgae in fully controlled photobioreactors, and 4) how to integrate all acquired knowledge into optimal production strategies for microalgae biomass or secondary metabolites. The course is composed by lectures, digital classes and practical work and includes a visit to AlgaePARC (www.AlgaePARC.com).

The **programme topics** are the following ones:

- Fundamentals of photoautotrophic growth and light
- Quantifying light-limited microalgae growth
- Predicting productivity of photobioreactors and raceway ponds
- Microalgal metabolism and secondary product formation
- Metabolic (flux) modelling
- Metabolic engineering strategies
- Mass transfer in photobioreactors
- Photobioreactor cultivation strategies for optimal yield
- Going large scale: experiences and challenges

The last edition the course was coordinated by Dr. Packo Lamers (Bioprocess Engineering WUR, and FUEL4ME WP1 leader) and by Chantal Doeswijk, MSc - Graduate School VLAG.

Course lecturers are leading researchers in the fields of microalgae cultivation and give lectures in the course, such as:

- Dr Maria Barbosa,
- Prof Olaf Kruse, Bielefeld University, Germany
- Dr Luc Roef, Proviron, Belgium
- Prof Gabriel Acién-Fernandez, Almería University, Spain
- Dr Mark Michiels, Proviron, Belgium
- Prof René Wijffels, Bioprocess Engineering, Wageningen UR
- Dr Marcel Janssen, Bioprocess Engineering, Wageningen UR
- Dr Dirk Martens, Bioprocess Engineering, Wageningen UR
- Dr Arjen Rinzema, Bioprocess Engineering, Wageningen UR
- Dr Rouke Bosma, Bioprocess Engineering, Wageningen UR
- Dr Dorinde Kleinegris, Food & Biobased Research, Wageningen UR

5 Plan for use and dissemination of foreground

The PUDF summarises the consortium's strategy and concrete actions to disseminate, exploit and protect the foreground generated by a project and should serve as a guideline to the Consortium for the dissemination and exploitation (D&E) activities to be carried out in the context of the FUEL4ME project.

The PUDF consists of these sections:

- Section A provides an insight to the Dissemination strategy, defined in the Dissemination Plan (D6.1) and a description of dissemination activities carried out in the project, including a list of the scientific publications.
- Section B outlines the exploitation strategy for the management of the intellectual property of the
 project results, aimed at identifying the potential exploitable results, assessing on the different
 possibilities for IPR protection and exploring new business opportunities.

The deliverable 6.8 is devoted to present the dissemination and exploitation outputs and also the strategy to follow for each exploitable results. In the following tables, we present a summary of dissemination activities, exploitable results and strategies to follow up with them.



Section A (public)

The following table shows the list of publications made by FUEL4ME. It is important to state that according to special clause in grant agreement nº 39, all partners have made an effort to make publicly available the scientific publications. In the last 2 columns it is reflected whether the public publications are publicly available and the URL to download them.

List of scientific publications

	A1-LIST OF SCIENTIFIC PUBLICATIONS													
No.	Title	Main author	of the conference		Publisher	Place of publication	Year of publication	Relevant pages	Permanent identifiers (if available)/ Website to download the publication	Is open access/ publicly available provided to this publication?				
1	Interactive effects of salinity, high light, and nitrogen starvation on fatty acid and carotenoid profiles in Nannochloropsis oceanica CCALA 804	Alexei Solovchenko, Alexander Lukyanov, Olga Solovchenko, Shoshana Didi Cohen, Sammy Boussiba and Inna Khozin Goldberg	European Journal of Lipid Science and Technology	Volume 116, issue 5	Wiley-VCH Verlag GmbH & Co. KGaA	Weinheim, Germany	2014	635-644	http://www.wile y- vch.de/publish/e n/journals/alpha beticIndex/2114 /	No				



			A	A1-LIST OF SCI	ENTIFIC PUBLIC	ATIONS				
No.	Title	Main author	Title of the periodical or the series// Name of the conference	Number, date or frecuency	Publisher	Place of publication	Year of publication	Relevant pages	Permanent identifiers (if available)/ Website to download the publication	Is open access/ publicly available provided to this publication?
2	Protein Inference Using Peptide Quantification Patterns	Pieter Lukasse, Twan America	Journal of Proteome Research	Volume 13, issue 7	American Chemical Society	Wageninge n, The Netherlands	2014	3191– 3199	http://pubs.acs. org/doi/full/10.1 021/pr401072g	No
3	"Enhancing TAG productivity in Phaeodactylum tricornutum by expression of heterologous lipid droplet protein under nitrogen starvation" (scientific presentation)	Z. Shemesh, S.Leu, I. Khozin- Goldberg, S. Boussiba	EMBO 2014 seminar and workshop Euro- mediterranean microalgae biotechnology	N/A	Research Centre for Agricultural and Food Biotechnolog y of the University of Almería, Spain	Almeria, Spain	2014	N/A	http://fuel4me.e u/index.php?opt ion=com_jdownl oads&task=dow nload.send&id=1 363&catid=8&m =0&Itemid=120	Yes
4	"Expression and localization of Haematococcus pluvialis oil globule protein in Phaeodactylum tricornutum"	Shemesh, Zachor. Leu, Stefan. Khozin-Goldberg, Inna. Boussiba, Sammy	4th Int. CeBiTec Research Conference Advances in Industrial Biotechnology: Prospects and challenges for	N/A	N/A	Biotechnolo gy Center for Interdiscipli nary Research (ZiF), Bielefeld	2014	N/A	N/A	No



			А	1-LIST OF SCI	ENTIFIC PUBLIC	CATIONS				
No.	Title	Main author	Title of the periodical or the series// Name of the conference	Number, date or frecuency	Publisher	Place of publication	Year of publication	Relevant pages	Permanent identifiers (if available)/ Website to download the publication	Is open access/ publicly available provided to this publication?
	(scientific presentation)		the development of algal.			University, Germany				·
5	"Expression and localization of an oil globule protein in Phaeodactylum tricornutum using an endogenous stress-activated promoter" (scientific presentation)	Shemesh, Zachor. Leu, Stefan. Khozin-Goldberg, Inna. Boussiba, Sammy	ILANIT-7th – the Israel Societies for Experimental Biology (FISEB).	N/A	N/A	Eilat, Israel.	2014	N/A	N/A	No
6	Lipid production in Phaeodactylum tricornutum under simulated outdoor conditions	Ilse M. Remmers, Packo P. Lamers, Dirk E. Martens, René H. Wijffels	Website Wageningen UR	N/A	N/A	Wageninge n, The Netherlands	2013	N/A	http://tinyurl.co m/nd67ddv	No



			Α	1-LIST OF SCI	ENTIFIC PUBLIC	CATIONS				
No.	Title	Main author	Title of the periodical or the series// Name of the conference	Number, date or frecuency	Publisher	Place of publication	Year of publication	Relevant pages	Permanent identifiers (if available)/ Website to download the publication	Is open access/ publicly available provided to this publication?
7	Lipid production in Phaeodactylum tricornutum under simulated outdoor conditions (scientific presentation)	llse M. Remmers, Packo P. Lamers, Dirk E. Martens, René H. Wijffels	YAS conference	N/A	N/A	Montpellier , France	2014	N/A	N/A	No
8	Lipid production in Phaeodactylum tricornutum under simulated outdoor conditions (scientific presentation)	Ilse M. Remmers, Packo P. Lamers, Dirk E. Martens, René H. Wijffels	Algae'nChem conference	N/A	N/A	Montpellier , France	2014	N/A	N/A	No
9	Two stage Lipid production in Phaeodactylum tricornutum (scientific presentation)	Ilse M. Remmers, Packo P. Lamers, Dirk E. Martens, René H. Wijffels	NBC15 conference	N/A	N/A	Ede, The Netherland	2014	N/A	N/A	No



	A1-LIST OF SCIENTIFIC PUBLICATIONS													
No.	Title	Main author	Title of the periodical or the series// Name of the conference	Number, date or frecuency	Publisher	Place of publication	Year of publication	Relevant pages	Permanent identifiers (if available)/ Website to download the publication	Is open access/ publicly available provided to this publication?				
10	Strategies for oil and EPA production from Phaeodactylum tricornutum	Liliana Rodolfi1,2, Alessia Guccione1, Niccolò Bassi2, Massimo D'Ottavio2, Giacomo Sampietro2, Natascia Biondi1, Mario R. Tredici1	EABA 1-3 December 2015	N/A	N/A	Lisbon (Portugal)	2015	N/A	N/A	No				
11	Continuous versus batch production of microalgal lipids	llse M. Remmers, Packo P. Lamers, René H. Wijffels	ESBES conference	N/A	N/A	Dublin, Ireland	2016	N/A	http://fuel4me.e u/index.php?opt ion=com_jdownl oads&task=dow nload.send&id=1 388&catid=8&m =0&Itemid=120	Yes				
12	Economic and social impacts of biofuel and omega-3 fatty acids production from microalgae	Steiner, D., Kaltenegger, I., Hingsamer, M., Jungmeier, G.	N/A	N/A	N/A	N/A	2016	N/A	N/A	No				



	A1-LIST OF SCIENTIFIC PUBLICATIONS														
No.	Title	Main author	Title of the periodical or the series// Name of the conference	Number, date or frecuency	Publisher	Place of publication	Year of publication	Relevant pages	Permanent identifiers (if available)/ Website to download the publication	Is open access/ publicly available provided to this publication?					
13	Inducible expression of Haematococcus oil globule protein in the diatom Phaeodactylum tricornutum: Association with lipid droplets and enhancement of TAG accumulation under nitrogen starvation		Algal Research	Volume 18	N/A	N/A	2016	321-331	N/A	No					

Expected publications

	I	Expected Publications A1 b	- LIST OF SCIENTIFIC OF EX	(PECTED PUE	BLICATION	5
No.	Partne r	Title	Main author	Description	Expected Date of Publicatio n	Is/Will open access provided to this publication?
1	WU- WPR	Orchestration of transcriptome, proteome, metabolome and fluxome during lipid synthesis in the diatom Phaeodactylum tricornutum	Ilse M. Remmers, Dirk E. Martens, Twan America, Jan Cordewener, Ric de Vos, Sander Peters, Linda Bakker, René H. Wijffels, Packo P. Lamers,	analyses ongoing and manuscript in preparation	June 2017	
2	wu	Impact of light intensity on lipid productivity in the diatom Phaeodactylum tricornutum	Ilse M. Remmers, Dirk E. Martens, René H. Wijffels, Packo P. Lamers	manuscript in preparation	March 2017	
3	wu	Continuous versus batch production of microalgal lipids	Ilse M. Remmers, A. Hidalgo, B. Brandt, René H. Wijffels, Packo P. Lamers,	Final editing before clearance/ publication	February 2017	yes
4	wu	The impact of starch as temporary energy storage system in synchronized microalgal growth	Graciela M. Leon Saiki, Ilse M. Remmers, Packo P. Lamers, Douwe van der Veen, Dirk E. Martens, René H. Wijffels	Final editing before clearance/ publication	February 2017	yes
5	BGU	Inducible Expression Drives Haematococcus Oil Globule Protein Localization to Lipid Droplets and Enhances TAG Accumulation in the Diatom Phaeodactylum tricornutum under Nitrogen Starvation	Zachor Shemesh, Stefan Leu, Inna Khozin-Goldberg, Shoshana Didi-Cohen, Aliza Zarka, Sammy Boussiba	Final editing before sending to publication	March 2017	
6	BGU	Isolation and characterization of novel promoters for constitutive and inducible expression of transgenes in the biotechnologically important diatom <i>P. tricornutum</i> under variable conditions, including nitrogen starvation	Zachor Shemesh, Stefan Leu, Aliza Zarka, Inna Khozin-Goldberg, and Sammy Boussiba	Writing	Jun-17	
7	WFBR / WU	Continuous nitrogen limitation of <i>N.oceanica</i>	M van der Zwart, DMM Kleinegris, PP Lamers, MJ Barbosa, RH Wijffels	Manuscript in preparation	March 2017	



	E	Expected Publications A1 b	- LIST OF SCIENTIFIC OF EX	(PECTED PUE	BLICATIONS	5
No.	Partne r	Title	Main author	Description	Expected Date of Publicatio n	Is/Will open access provided to this publication?
7	F&M	Oil and EPA production by the diatom Phaeodactylum tricornutum cultivated outdoors in Green Wall Panel (GWP®) reactors	N.A	Writing	N.A	N.A
8	F&M	Oil and EPA production by Nannochloropsis oceanica cultivated outdoor in Green Wall Panel (GWP®) reactors	N.A	Writing	N.A	N.A
9	F&M	Energy and technoeconomic-analysis of lipid production by Nannochloropsis oceanica cultivated in Green Wall Panels (GWP®)	N.A	Writing	N.A	N.A



Participation in international conferences

Partners have presented FUEL4ME activities and results in the following international conferences, seminars and workshops

	A2: List of Dissemination Activities											
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed				
1	Other, Presentation	IDC	CommNet Dissemination Training: Communication Strategy Seminar for EU biotech Projects. FUEL4ME General Project Presentation	13-feb-13	Dublin, Ireland	Industry	50	EU				
2	Conference	EVODOS	9th ECCE Congress European Conference on Chemical Engineering	21-apr-13	The Hague, Netherlands	Scientific Community, Industry	1900	International				
3	Conference, Presentation	EVODOS	European Algae Biomass FUEL4ME General Project Presentation and Progress	24-apr-13	Vienna, Austria	Scientific Community, Industry	300	EU				
4	Conference, Presentation	F&M	3rd Int. Conf. on Algal Biomass, Biofuels & Bioproducts FUEL4ME Project Presentation	19-jun-13	Toronto, Canada	Scientific Community, Industry	NA	International				
5	Workshop	ВІОТОРІС	ALGAENET workshop	03-jul-13	Seville, Spain	Scientific Community, Industry	NA	International				
6	Conference	EVODOS, WFBR	Algae Biomass Summit	03-oct-13	Orlando, USA	Industry	600	International				
7	Workshop, Presentation	JOANNEUM, WU	Austrian Stakeholder Workshop of IEA Bioenergy Task 42 "Biorefining" FUEL4ME Project Presentation	24-0ct-13	Graz, Austria	Scientific Community, Industry	50	International				



	A2: List of Dissemination Activities												
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed					
8	Workshop, Presentation	JOANNEUM	Algen als biogene Ressource – Akteure in Österreich FUEL4ME project presentation	06-nov-13	Wieselburg, Austria	Scientific Community, Industry	25	Austria					
9	Workshop, Presentation	F&M	Workshop on: Scaling Algal Production Technologies for the Kingdom of Saudi Arabia Prof Tredici: Invited Lecturer- Presentation of Production Systems and Products	12-nov-13	KAUST Thuwal, Saudi Arabia	Industry	NA	Saudi Arabia					
10	Conference	WFBR, F&M	EABA Expo and conference	12-nov-13	Florence, Italy	Scientific Community, Industry	NA	International					
11	Congress	EVODOS	International Algae Congress Activities: general Presentation of FUEL4ME project and distribution of brochures and leaflets	05-dec-13	Hamburg, Germany	Scientific Community, Industry, Policy Makers	200	International					
12	Conference, Presentation	JOANNEUM	EnInnov2014 – 13th Symposium Energy Innovation Presentation of FUEL4ME within sustainability of algae energy systems topic	13-feb-14	Graz, Austria	Scientific Community, Industry, Policy Makers	600	International					
13	Conference, Presentation	WU, F&M	Algae'Chem conference F&M participated with Prof Tredici as invited lecturer WU presented progress on WP1	31-mar-14	Montpellier, France	Industry	300	International					
14	Conference, Presentation	WU	YAS conference Presentation 1: WP1 activities of FUEL4ME "Lipid production in <i>Phaeodactylum tricornutum</i> under simulated outdoor conditions"-Ilse Remmers	04-abr-14	Montpellier, France	Scientific community	100	International					



			A2: List of Dissemination Act	ivities				
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed
15	Conference, Presentation	F&M	NUTRA-AFRICA Int. Conference Prof Tredici invited lecturer: Microalgae as Food	11-abr-14	Florence, Italy	Scientific community	NA	International
16	Workshop, Presentation	JOANNEUM, WFBR, WU	European Workshop on Life Cycle Analysis of algal based Biofuels	24-abr-14	Belgium, Brussels	Scientific Community, Industry, Policy Makers	50	EU
17	Conference	F&M	6. European Algae Biomass 2014 Opening, chairing and closing of the conference	07-may-14	Seville, Spain	Scientific Community, Industry	NA	EU
18	Conference, Presentation	wu	Dutch conference on biotechnology Presentation of FUEL4ME project	27-may-14	Ede, Netherlands	Scientific Community	150	Netherlands
19	Conference, Presentation	JOANNEUM	22nd European Biomass Conference and Exhibition Presentation: "Towards a Standard Methodology for the Sustainability Assessment of Energy Systems with Algae – An European Approach in FUEL4ME".	24-jun-14	Hamburg, Germany	Scientific Community, Industry, Policy Makers	NA	EU
20	Conference, Presentation	JOANNEUM	Presentation on FUEL4ME at the Algae Event 2014 organized within the project EnAlgae	25-jun-14	Hamburg, Germany	Scientific community, Industry	100	EU
21	Congress, Presentation	F&M	CHIMALI 2014 - X° Congresso Italiano di Chimica degli Alimenti Prof Tredici Invited Lecturer: Microalgae, can they contribute to a more sustainable future?	08-jul-14	Florence, Italy	Scientific community, Industry	NA	Italy



	A2: List of Dissemination Activities											
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed				
22	Conference, Presentation	F&M	FEBS-EMBO CONFERENCE – Science and Society Session Presentation; Prof Tredici(Invited Lecturer): Food and biofuels from large-scale algae mass cultures: Opportunities and challenges.	31-ago-14	Paris, France	Scientific Community	Na	International				
23	Workshop	JOANNEUM	Algen als biogene Ressource – Forschung in Österreich Participation in workshop	11-sep-14	Duernrohr, Austria	Scientific Community, Industry, Policy Makers	30	Austria				
24	Conference, Presentation	BGU	Algae Biomass Summit 2014 Dr Stefan Leu Lectures on general biofuels research and not specifically focused on FUEL4ME, FUEL4ME support was acknowledged.	29-sep-14	Washington DC (USA)	Scientific Community, Industry	NA	International				
25	Conference, Presentation	F&M	1st EABA and EC Algae Contractors' Conference and 8th International Algae Congress Prof Tredici Food and biofuels from large-scale algae mass cultures: Opportunities and challenges (invited lecture).	02-dic-14	Florence, Italy	Scientific Community, Industry	500	International				
26	Conference, Presentation	BGU	EMBO 2014 seminar and workshop Euro-Mediterranean microalgae biotechnology Presentation: Enhancing TAG productivity in <i>Phaeodactylum tricornutum</i> by expression of heterologous lipid droplet protein under nitrogen starvation (scientific presentation)	03-may-14	Almeria, Spain	Scientific Community	NA	EU				



			A2: List of Dissemination Act	ivities				
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed
27	Conference, Presentation	BGU	4th Int. CeBiTec Research Conference Advances in Industrial Biotechnology: Prospects and challenges for the development of algal. Presentation: Expression and localization of Haematococcus pluvialis oil globule protein in Phaeodactylum tricornutum" (scientific presentation)	21-sep-14	Bielefeld, Germany	Scientific Community	NA	International
28	Conference, Presentation	BGU	ILANIT-7th – the Israel Societies for Experimental Biology (FISEB). Presentation: Expression and localization of an oil globule protein in <i>Phaeodactylum tricornutum</i> using an endogenous stressactivated promoter (scientific presentation)	10-feb-14	Eilat, Israel	Scientific Community	NA	Israel
29	Conference, Presentation	wu	Scientific Presentation at NBC15 conference Presentation: Two stage Lipid production in Phaeodactylum tricornutum Ilse M. Remmers, Packo P. Lamers, Dirk E. Martens, René H. Wijffels	12-abr-14	Ede, Netherlands	Scientific Community	NA	International
30	Workshop	BGU	Solar Fuels Workshop Dr Stefan Leu, and Dr Khozin-Goldberg attended the workshop The lectures were on our general biofuels research and not specifically focused on Fuels4Me, Fuels4Me support was acknowledged.	17-feb-15	Dead Sea, Israel	Scientific Community, Industry	NA	Israel



			A2: List of Dissemination Act	ivities				
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed
31	Symposium, Presentation	F&M	The Royal Swedish Academy of Science Jubilee Symposia. Presentation by Professor M. R. Tredici Food and Biofuels from Microalgae Cultures: Opportunities and Challenges (Plenary lecture).	18-feb-15	Gothenburg, Sweden	Scientific Community, Industry	NA	International
32	Conference, Presentation	F&M	Conferenza su "L'economia circolare per uno sviluppore sponsabilenella produzione di cibo" Presentation by Professor M. R. Tredici- Algal Walls: cibo, combustibili e prodottiutili da pareti fotosintetichenellecittà del futuro	30-abr-15	Firenze, Italy	Scientific Community, Industry	NA	Italy
33	Workshop, Presentation	JOANNEUM	3rd European Workshop "Life Cycle Analysis of Algal based Biofuels & Biomaterials" FUEL4ME WP5 Presentation	11-may-15	Brussels, Belgium	Scientific Community, Industry, Policy Makers	NA	EU
34	Conference	SWR	5th International Conference on Algal Biomass, Biofuels & Bioproducts	10-jun-15	San Diego, USA	Scientific Community, Industry	NA	International
35	Conference, presentation	SWR	International Conference on Microalgae and Biofuels	30-jul-15	Istanbul, Turkey	Scientific Community, Industry	NA	International
36	Conference, presentation	SWR	International Congress and Expo on Biofuels & Bioenergy Biofuels 2015	25-ago-15	Valencia, Spain	Scientific Community, Industry	NA	International



			A2: List of Dissemination Act	ivities				
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed
37	Conference, Presentation	BGU	Algae Biomass Summit 2015 Presentation by Dr Stefan. Leu: The lectures were on our general biofuels research and not specifically focused on FUEL4ME, FUEL4ME support was acknowledged.	29-sep-15	Washington DC (USA)	Scientific Community, Industry	NA	International
38	Poster Presentation	F&M	EABA Strategies for oil and EPA production from Phaeodactylum tricornutum – Liliana Rodolfi	02-dec-15	Lisbon, Portugal	Scientific Community, Industry	NA	International
39	Conference, Presentation	wu	ESBES: The European Society of Biochemical Engineering Sciences Presentation: Continuous versus batch production of microalgal lipids - Ilse Remmers	11-sep-16	Dublin, Ireland	Scientific Community	NA	EU
40	Conference, Presentation	wu	EU Algae Roadmap Presentation: Continuous versus batch production of microalgal lipids - Ilse Remmers	07-apr-16	Olhão, Portugal	Scientific Community	NA	EU
41	Workshop, Presentation	JOANNEUM	Meeting of the Austrian Algae Network, an oral presentation was held on the topic: Pathways of microalgae for the production of biofuels and biomaterials by Maria Hingsamer	17-nov-15	Vienna, Austria	Scientific Community, Industry	35	Austria
42	Conference, Presentation	JOANNEUM	Maria Hingsamer was asked to present at the 12th European Bioethanol and Bioconversion Technology Meeting the topic "Biofuels from Microalgae – Concepts, Modelling and Life Cycle Analysis"	12-apr-16	Detmold, Germany	Scientific Community, Industry	100	EU



			A2: List of Dissemination Act	ivities				
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed
43	Conference, Poster Presentation	JOANNEUM	Attendance of European Biomass Conference (EUBCE): One poster "Modelling and Assessment of Algae Cultivation for Large Scale Biofuel Production Sustainability and Aspects of Up-scaling of Algae Biorefineries" and one oral presentation "Implementing Strategies of Biorefineries in the BioEconomy" in relation to FUEL4ME.	06-jun-16	Amsterdam, Netherlands	Scientific Community, Industry	NA	EU
44	Conference, Poster Presentation	JOANNEUM	European Roadmap for an Algae-Based Industry The topic "BioEconomy with Algae – Life Cycle Sustainability Assessment of an Algae-Based Biorefinery" was presented at Olhão with a poster presentation	07-aug-16	Olhão, Portugal	Scientific Community, Industry, Policy Makers	NA	EU
45	Conference	JOANNEUM	Maria Hingsamer attended the meeting of the Austrian Algae Network, the ongoing work in FUEL4ME was discussed	31-may-15	Wels, Austria	Scientific Community, Industry	NA	Austria
46	Conference, poster presentation	WFBR	Continuous triacylglycerol production by Nannochloropsis oceanica	13-15 dec 16	Madrid, Spain	Scientific community, industry	NA	EU
47	Conference	F&M	1st EABA and EC Algae Contractors Conference and 8 th International Algae Contractors	1-3 December	Florence	Scientific Community, Industry	NA	Italy
48	Poster Presentation	F&M	Liliana Rodolfi attended the meeting Alg'n'Chem'2014	31 March-3 April 2014	Montpelier (France)	Scientific Community, Industry	NA	France
49	Presentation	F&M	The 4 th Asia-Oceania Algae Innovation Summit. Liliana Rodolfi et al. "Energy balance and techno-economic analysis of lipid production by	18-21 September 2016	Wuhan	Scientific Community, Industry	NA	China



	A2: List of Dissemination Activities								
No.	Type of activities	Main Leader	Title	Date/Period	Place	Type of Audience	Size of audience	Countries addressed	
			Nannochloropsis oceanica cultivated in Green Wall Panels (GWP) and raceway ponds at large scale"						
50	Presentation	F&M	European Roadmap for an algae-based industry. "Liliana Rodolfi et al. "One-step vs to step oil production from <i>Nannochloropsis oceanica</i> and Phaeodactylum tricornutum cultivated outdoors in Green Wall Panels"	6-8 April 2016	Olhao	Scientific Community	NA	Portugal	
51	Presentation	F&M	AlgaEurope 2016. Liliana Rodolfi et al. "Lipid production by <i>Nannochloropsis oceanica</i> at large scale: energy balance and technoeconomic analysis"	13-15 December 2016	Madrid	Scientific Community, Industry	NA	Spain	
52	Conference	F&M	International CeBiYec Research Conference	21-24	Bielefeld	Scientific Community, Industry	NA	Germany	



Section B (Confidential⁷ or public: confidential information to be marked clearly)

Part B1

B1: List of Applications for Patents, Trademarks, Registered Designs, etc.								
Confidential Yes/No	Foreseen embargo date	Application reference	Subject or title of application	Applicant				
		WO2016207893A1	Inducible transgene expression and enhanced TAG accumulation by overexpression of DGAT1 or HOGP using endogenous promoters of <i>P. tricornutum</i>					
	Confidential	Confidential Foreseen embargo date	Confidential Yes/No Foreseen embargo date Application reference WO2016207893A1	Confidential Yes/No Foreseen embargo date Application reference Inducible transgene expression and enhanced TAG accumulation by overexpression of DGAT1 or HOGP using endogenous promoters of <i>P. tricornutum</i>				

 $^{^{7}}$ Note to be confused with the "EU CONFIDENTIAL" classification for some security research projects.



				B2: Exploitable Result Identification				
Type of Exploitable Foreground	Description of Exploitable Foreground	Confidential Yes/ No	Foreseen embargo date	Exploitable product or measure	Sector of application	Timetable, commercial or any other use	Patents or other IPR exploitation Strategy	Owner and other beneficiary involved
Commercial Exploitation of R&D results	New promoters for gene expression in <i>Phaeodactylum</i>	No	NA	The result is important for activities after the end of the project. Further development/research is needed. The result will be an input for future projects that could lead to IPR	Research and experimental development on biotechnology	under investigation for commercial use - 3 years	Patented: Inducible transgene expression and enhanced TAG accumulation by overexpression of DGAT1 or HOGP using endogenous promoters of P. tricornutum ad publication submitted	BGU
Commercial Exploitation of R&D results	HOGP over expression in Phaeodactylum	No	NA	The result is important for activities after the end of the project. Further development/research is needed. The result will be an input for future projects that could lead to IPR	Research and experimental development on biotechnology	under investigation for commercial application - 3 years	patented: Inducible transgene expression and enhanced TAG accumulation by overexpression of DGAT1 or HOGP using endogenous promoters of P. tricornutum and published	BGU



				B2: Exploitable Result Identification				
Type of Exploitable Foreground	Description of Exploitable Foreground	Confidential Yes/ No	Foreseen embargo date	Exploitable product or measure	Sector of application	Timetable, commercial or any other use	Patents or other IPR exploitation Strategy	Owner and other beneficiary involved
General Advancement of Knowledge	MAP OMICS tool, a new tool for integrative transcriptomics, proteomics and metabolomics analysis was developed	No	NA	The results will be published, will serve for future scientific research and will also serve as input for future project applications. Moreover, the tool will become publicly available soon (open source structure).	Research and experimental development on bioinformatics	NA	Publication/ Copyright	WPR/ WU
General Advancement of Knowledge	Continuous lipid production and comparison to batch both in lab expermiments at at pilot scale outdoors	No	31.12.2018 - Current work being in process of publication in peer- reviewed journals	The results on various strains and under various conditions, both lab and outdoors will be published in multiple articles and will also serve as input for future project applications	Research and experimental development on biotechnology & microalgae	NA	Publication/ Copyright	WFBR/WU/ BGU/ F&M
General Advancement of Knowledge	Techno economics for pilot real outdoor photo- bioreactors at various locations	No	31.12.2018 - Current work being in process of publication in peer- reviewed journals	The results will be published and will also serve as input for future project applications and for LCAs. The numbers are also used to inform those interested (general public, possible algae-producers and buyers) on the current state of the art.	Research and experimental development on biotechnology & microalgae	NA	Publication/ Copyright	WFBR/ JOANNEUM/ WU/ BGU/ F&M/ BIT/ EVODOS/ PDX/ Feyecon/ NOIL
General Advancement of Knowledge	Improved thermoregulation in outdoor photobioreactors	No	NA	The result will be published as soon as possible	Research and experimental development on biotechnology & microalgae	under investigation for upscaling and commercial use	Patents and publicatios	BGU



				B2: Exploitable Result Identification				
Type of Exploitable Foreground	Description of Exploitable Foreground	Confidential Yes/ No	Foreseen embargo date	Exploitable product or measure	Sector of application	Timetable, commercial or any other use	Patents or other IPR exploitation Strategy	Owner and other beneficiary involved
General Advancement of Knowledge	Use of proprietary heat exchanger from F&M	No	NA	Improved process or know-how that will be important for future technology development for F&M after the end of the project.	Biotechnology	NA	NA	F&M
General Advancement of Knowledge	Panel direction optimization for increased light efficiency for production in Israel and Florence	No	NA	Improved know-how that leads to increased production	Research and experimental development on biotechnology & microalgae	NA	Publication/ Copyright	BGU, F&M
General Advancement of Knowledge	Using algae as biocatalysts for converting palmitic to valuable FA's.	No	NA	Improved know-how that leads to high value and sustainable raw materials and products	Research and experimental development on biotechnology & microalgae	NA	Publication/ Copyright	BGU
Commercial Exploitation of R&D results	Use of the ProviAPT (Proprietary photobioreactor) in warm climate	No	NA	Improved process or know-how that will be important for future activities and product development for Proviron after the end of the project.	Biotechnology	NA	NA	BIT/ PROVIRON
Commercial Exploitation of R&D results	Testing of Evodos technology (Evodos25) and performance improvement for harvesting and water recycling	No	NA	Improved process or know-how that will be important for future activities and product development for Evodos after the end of the project.	Biotechnology	NA	NA	Evodos



				B2: Exploitable Result Identification				
Type of Exploitable Foreground	Description of Exploitable Foreground	Confidential Yes/ No	Foreseen embargo date	Exploitable product or measure	Sector of application	Timetable, commercial or any other use	Patents or other IPR exploitation Strategy	Owner and other beneficiary involved
Commercial Exploitation of R&D results	Testing of SoniqueFlo technology developed by Cellulac for cell disruption	No	NA	"The result is important to Cellulac seeking to identify the specific physical hardware design and setting of parameters upon which SoniqueFlo would operate at its most efficient. It would provide the calculation for ratio of energy consumed by the process to energy available from the underlying microalgae cell. The outcome indicating a saving or loss for the commercial exploitation of the process installation, beyond pilot scale, either as part of Cellulac's own production, or licensed to external microalgae producers."	Biotechnology	NA	NA	Cellulac
General Advancement of Knowledge	Results of sustainability assessment (LCA, LCC, sLCA)	No	NA	The applied methodology will be further developed and applied in future projects (international, national). The results will be published in peer reviewed papers.	Research and sustainability assessment	NA	Publication/ Copyright	Joanneum
Commercial Exploitation of R&D results	Mild extraction of TAG and generation of several valuable by- products	No	NA	Results are important for commercial algae production after end of project. Results can help microalgae producers and aquaculture sector	Research and experimental development on biotechnology & microalgae, aquafeed sector	NA	NA	FeyeCon
General Advancement of Knowledge	Analyses of lipid profile, fatty acid profile and impurities of Phaeodactylum and Nannochloropsis oil	Yes	NA	The result is important for activities after the end of the project. Further development/research is needed.	Analyses of oil compositions	NA	NA	SWR/ WU/ F&M/ Feyecon/ Neste



				B2: Exploitable Result Identification				
Type of Exploitable Foreground	Description of Exploitable Foreground	Confidential Yes/ No	Foreseen embargo date	Exploitable product or measure	Sector of application	Timetable, commercial or any other use	Patents or other IPR exploitation Strategy	Owner and other beneficiary involved
General Advancement of Knowledge	Network of universities and companies working with algae	No	NA	Network will be use in collaborating in other projects	Research and experimental development on biotechnology & microalgae	NA	NA	SWR/ WU/ Evodos/ Cellulac/ Feyecon/ BGU/ F&M/ Neste



Results, lessons learned and challenges for the future

As one may conclude from the previous section, no patents, trademarks, registered designs or other IP protection forms have been registered as a direct output of the research carried out, and therefore cannot be attributed directly to the FUEL4ME project. This is not unexpected as there is still a big gap between research and the commercial uptake of 2nd generation biofuels from microalgae that are competitive alternatives to fossil fuels, thus furtherresearch is required to bridge this gap.

As a result, the vast majority of the foreground generated during the project corresponds to general advancements in knowledge in the form of lessons learned and improved know-how leading to cost effective and energy efficient processes. And in the form of rigorous scientific research that can either be published or may serve as input to new R&D projects.

For the FUEL4ME industrial partners the project has served as an excellent opportunity to pilot test their technologies in order to obtain more reliable and scalable industrial solutions for microalgae cultivation and downstream processing.

In addition FUEL4ME has contributed to prepare highly skilled professionals with expertise in algal microbiology and microalgae cultivation and processing systems.

The following paragraphssummarize some of the most outstanding results, lessons learned and future challenges that can be extracted from the FUEL4ME project:

Result 1: Continuous lipid production vs. Batch production

Partners involved: DLO/WU/BGU/F&M

<u>Conclusions</u>: Batch and continuous mode are on average in same range of productivity (in lab and outdoor), while TAG contents were generally higher in batch mode. Varying results were obtained for robustness of either process (i.e. in some cases batch was more robust, while in other instances continuous mode was more robust). Costs could not be properly assessed at the tested scale. However, costs (of entire process including downstream processing) should in the end be decisive for ultimately adopting one of the two cultivation strategies.

<u>Future Challenges</u>: Further development and up scaling of both cultivation technologies is needed to improve, properly assess and finally compare the process economy. Key point for this analysis is to take into account the compatibility of the cultivation technology with the downstream processing chain.

Result 2: Evodos 25 harvesting and water recycling technology testing and optimization.

Partners involved: EVODOS

<u>Conclusions</u>: FUEL4ME project has served for the successful testing and optimization of the EVODOS 25 dynamic settler with Evodos proprietary spiral plate technology. The results of the project show that the integration of an improved splash screen in the fully automated harvester minimized paste losses. The overall system lead to an increase yield, top quality algae paste and high efficiencies (separation efficiency was higher than 90% with a harvesting capacity over 6m3/day for the selected microalgae strains). In addition a water recycling unit was successfully tested.

<u>Future Challenges</u>: More research must be carried out for optimizing the water recycling unit when using the recycled water for cultivation mainly in analyzing performance, yield and the need of adding nutrients. In



addition future testing must be carried out to scale-up the process maintaining productivity and avoiding contamination.

Result 3: SoniqueFlo technology tested for Nannochloropsis cell disruption

Partners Involved: CELLULAC

<u>Conclusions</u>: The FUEL4ME project results indicates strongly that Cellulac's proprietary SoniqueFlo technology has a role to play in improving the economics of commercial scale production using an enzyme based solvent free wet extraction process of algal oils. This process can lead to an estimated 25% decrease in operational costs, less capital expenditure and high energy efficiencies because the solution works directly in raw algae material while still in aqueous solution eliminating the need of de-watering steps and therefore improving energy balance and cost effectiveness. In addition, the solution tested has no organic solvents or toxic waste products and delivers high yields of target oils, sugars and proteins in a readily usable form.

<u>Future Challenges</u>: While the cell disruption technology is ready to be scaled up at commercial scales, the biggest bottleneck is to obtain large volumes and steady supplies of microalgae biomass to attract big industry players.

Result 4: Energy efficient and cost effective lipid extraction using supercritical fluid

Partners Involved: FEYECON

<u>Conclusions:</u> The FUEL4ME project has contributed to the extraction of TAG with very high efficiencies using supercritical fluid (>95%) obtaining a high value and stable biomass after extraction. In addition, very relevant data and cost calculations for scale-up were collected during the project. This acquired knowledge will prove to be useful for future development projects. Lessons learned regarding the effect of the physical properties of the dried biomass on extraction yield and the need for polishing of the extracted oils (drying, FFA removal, deodorization, bleaching, dewaxing) were gathered and shared for future R&D.

<u>Future Challenges</u>: A feasibility comparison must be carried out between dried microalgae and wet microalgae extraction to further understand the relative pros and cons of each method in future technology developments.

Result 5: Energy Efficient and cost effective high and low value fatty acid extraction using supercritical fluid

Partners Involved: FEYECON

<u>Conclusions</u>: As a result of FUEL4ME, novel reactive extraction and separation protocols based on advanced supercritical fluid technology (as opposed to chromatographic methods) have been developed. These protocols and technology application have lead to obtaining high purified PUFAs (>81%), a robust system with stable concentrations of PUFAs and a reduction in FFAs content of oil from 23% to 1% at 50C. In addition, lessons learned regarding the required pre treatments of oil before fractionation (de-waxing, solid removal and bleaching), the improved purification of PUFAs in esterified form at lower pressures (1110-150 Bar) and the possibility to separate FFA from TAG at low temperatures have been recorded and shared for future R&D. <u>Future Challenges</u>: Scaling up with high productivity, cost and energy efficiency in order to meet market demands while ensuring production quality and minimizing product quality variability.



Result 6: Setting up and operation of a demonstration plant

Partners involved: BIT.

<u>Conclusions</u>: A demonstration plant of 1ha was set up and operated in Huelva (South region of Spain) using Proviron technology (ProviAPT). The results show promising potential for scalability and replication. FUEL4ME has lead to increased know-how on the installation, operation and maintenance requirements of ProviAPT technology that will prove to be important for future activities and product development for Proviron and BIT after the end of the project.

<u>Future Challenges</u>: Future research must focus not so much on scalability but on maintaining stable and high productivity and in preventing and control of contamination.

Implementation Plan

Key Aspects

During FUEL4ME , the main role of the exploitation team was to:

- Ensure that background and foreground IP was properly managed during the project and in accordance with the consortium agreement. Acting as a moderator to ensure adherence to the IP consortium agreement clauses and to the PUDF methodology presented in section 2.1 of this document.
- Provide guidance for exploitation strategies to the partners that could fit or be aligned with each
 partner's strategic business goals. Acting as an internal project consulting service providing knowhow on best practices in IP management, external market conditions and sustainability assessment.
 It must be stated, that it was out of the scope of the exploitation team to decide on the strategic
 business goals of each partners but rather to provide input or insights based on the work being
 performed in FUEL4ME.

Contribution of Foreground to Partner Strategic Goals

At the end of the project, one may conclude that these roles have been performed successfully as no IP ownership issues have been raised within the consortium partners and all partners have acquired valuable know how and other intangibles from the project that will shape their strategic goals in some way or another. The following table summarizes how FUEL4ME results and tasks have contributed to the strategic goals of each partner.

Partner	FUEL4ME Contribution to Strategic Goals
DLO	1) New Publications contributing to advance and disseminate publicly new knowledge on microalgae biotechnology 2) Development of highly skilled researchers in the field algal microbiology, cultivation and processing systems 3) Acquired Knowledge, lessons learned and know-how 4) Developed long-term external relationships for future projects 5) Valuable prestige associated with being part of an important transnational project



Partner	FUEL4ME Contribution to Strategic Goals
wu	1) New Publications contributing to advance and disseminate publicly new knowledge on microalgae biotechnology 2) Development of highly skilled researchers in the field algal microbiology, cultivation and processing systems 3) Acquired Knowledge, lessons learned and know-how 4) Developed long-term external relationships for future projects 5) Valuable prestige associated with being part of an important transnational project
BGU	1) New Publications contributing to advance and disseminate publicly new knowledge on microalgae biotechnology 2) Development of highly skilled researchers and professionals in the field algal microbiology, cultivation and processing systems 3) Acquired Knowledge, lessons learned and know-how 4) Developed long-term external relationships for future projects 5) Valuable prestige associated with being part of an important transnational project
F&M	1) New Publications contributing to advance and disseminate publicly new knowledge on microalgae biotechnology 2) Development of highly skilled researchers and professionals in the field algal microbiology, cultivation and processing systems 3) Acquired Knowledge, lessons learned and know-how 4) Developed long-term external relationships for future projects 5) Valuable prestige associated with being part of an important transnational project
ВІТ	1) Increased skills and know-how for future replicability and scalability 2)Increased service quality 3)Developed long-term external relationships for future projects 4)Valuable prestige associated with being part of an important transnational project
EVODOS	1)Testing and optimization of product 2)Increased quality of product 3)Increased access and visibility in alga market 4)Valuable prestige associated with being part of an important transnational project 5)Developed external relationships for future project or cooperations 6)Acquired skills, knowledge and know-how
CELLULAC	1) Uptake of R&D results to backup product benefits 2)Acquired skills, knowledge and know-how 3)Developed long term relationships for future projects 4)Testing and optimization of Product 5)Increased visibility and access to algae market 6)Strengthened collaboration with research institutions
FEYECON	1) Uptake of R&D results to backup product benefits 2)Acquired skills, knowledge and know-how 3)Developed long term relationships for future projects 4)Improved procedures and product quality



Partner	FUEL4ME Contribution to Strategic Goals
NOIL	1) Uptake of R&D results to backup product benefits 2)Acquired skills, knowledge and know-how 3)Developed long term relationships for future projects 4)Improved procedures and product quality
JOANNEUM	1) Acquired skills and knowledge 2) Valuable prestige gained associated to being part of an important transnational research project 3) Built lasting relationships for future project and collaborations 4) Increased quality and diversity of consultancy services 5) New publications
IDC	1) Acquired skills and knowledge 2) Valuable prestige gained associated to being part of an important transnational research project 3) Built lasting relationships for future project and collaborations 4) Increased quality and diversity of consultancy services

Sustainability assessment and inputs for strategic decision-making

JOAENNUM as part of the planned activities for WP5: Sustainability assessment of the integrated process carried out a detailed modelling of a full-scale integrated micro algal-based process with sustainability indicators of the whole value chain in comparison to a conventional reference system including the three dimension economic, environmental and social aspects.

A comparison of the modelled full-scale integrated micro algal-based process with the assessment of to actual state of technology reveals information on key critical parameters impacting the sustainability of the FUEL4ME integrated process.

The main factors that influence sustainability were identified to be cultivation and harvesting, electricity demand, source of CO2, source of water and suitable land.

Some of the conclusions extracted from this analysis clearly underline the need for further technology development to improve economic and environmental sustainability.

This analysis has provided valuable input to all project partners regarding the potential up-scaling of these processes, considering competition for price impacts on food, feed, costs of transportation, capital and operating costs for algal cultivation and conversion, expected final costs of products, and market prices for alternative products or means for supplying equivalent services.

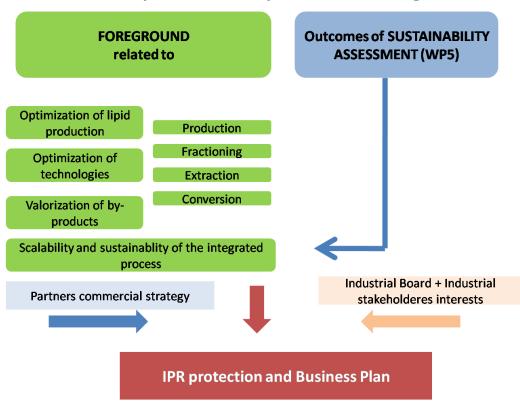
This information will prove to be useful for the project partners to set the grounds for the generation of new business models and apply FUEL4ME project results for commercial exploitation in line with their business strategies. In addition to the sustainability assessment the partners have acquired input from external project stakeholder and the project industrial board and they have received advice from the exploitation team on best practices on IP commercialization strategies while ensuring proper protection of the intangible assets and the foreground generated during the project.

The following diagram illustrates how the generated information and the activities carried out during the project provide valuable input for the project partners for the preparation of business models. Including the



input gathered from the industrial board and industrial stakeholders that is collected in D6.6 Fuel4Me Exploitation Meeting.

FUEL4ME process for the exploitation of the foreground





6 Report on societal implications

Replies to the following questions will assist the Commission to obtain statistics and indicators on societal and socio-economic issues addressed by projects. The questions are arranged in a number of key themes. As well as producing certain statistics, the replies will also help identify those projects that have shown a real engagement with wider societal issues, and thereby identify interesting approaches to these issues and best practices. The replies for individual projects will not be made public.

Α	General Information (completed automa	tically when Grant Agreement number is entered.					
Gra	nt Agreement Number:	308983					
Titl	e of Project:	FUture European League 4 Microalgal Energy					
Na							
	ne and Title of Coordinator:	Lolke Siistma					
В	Ethics						
1.	Did your project undergo an Ethics Review (and/or Screening)?					
•		ress of compliance with the relevant Ethics	No				
	Review/Screening Requirements in the fran	ne of the periodic/final project reports?					
_							
		with the Ethics Review/Screening Requirements					
	•	t Reports under the Section 3.2.2 'Work Progress					
an	d Achievements'						
_							
2.		olved any of the following issues (tick box):					
	SEARCH ON HUMANS						
•	Did the project involve children?						
•	Did the project involve patients?						
•	Did the project involve persons not able to	<u> </u>					
•	Did the project involve adult healthy volunt	eers?					
•	Did the project involve Human genetic mate	erial?					
•	Did the project involve Human biological sa	amples?					
•	Did the project involve Human data collect	tion?					
RES	SEARCH ON HUMAN EMBRYO/FOETUS						
•	Did the project involve Human Embryos?						
•	Did the project involve Human Foetal Tissue	e / Cells?					
•	Did the project involve Human Embryonic S	item Cells (hESCs)?					
•	Did the project on human Embryonic Stem	Cells involve cells in culture?					



•	• Did the project on human Embryonic Stem Cells involve the derivation of cells from Embryos?						
PRI	VACY						
•	Did the project involve processing of genetic informatio	n or personal data (eg. hea	alth, sexual				
	lifestyle, ethnicity, political opinion, religious or philosop	hical conviction)?					
•	Did the project involve tracking the location or observati	ion of people?					
RES	EARCH ON ANIMALS						
•	Did the project involve research on animals?						
•	Were those animals transgenic small laboratory animals	?					
•	Were those animals transgenic farm animals?						
•	Were those animals cloned farm animals?						
•	Were those animals non-human primates?						
RES	EARCH INVOLVING DEVELOPING COUNTRIES						
•	Did the project involve the use of local resources (genetic	ic, animal, plant etc)?					
•							
education etc)?							
DUAL USE							
Research having direct military use							
				No			
•	Research having the potential for terrorist abuse						
С	Workforce Statistics						
3.	Workforce statistics for the project: Please indicate in worked on the project (on a headcount basis).	the table below the numb	er of people	e who			
Тур	e of Position	Number of Women	Number of	f Men			
Scie	ntific Coordinator	2	1				
Wor	k package leaders	5	2				
Experienced researchers (i.e. PhD holders) 8 9							
PhD Students 4 5							
Oth	er	11	9				
4. How many additional researchers (in companies and universities) were recruited specifically for this project?							
Of which, indicate the number of men:							



D Gender Aspects											
Did you carı	ry out	specific Ge	nder Equal	ity Action	s under the project	:?	Х	Yes No			
Which of	the fol	llowing act	ions did vo	u carry ou	ıt and have affactiv	a wara thay?		1.10			
							•				
		_	_								
	_			•	•						
		•	OVE WOIK-II	ie Dalalice		00700					
							_				
	_										
				consume	ers, users, patients	or in criais, was cr	ie issue o	n gender			
0	Yes-	please spe	cify								
			·								
X	No										
Synergies	with S	Science Edu	ıcation								
	estivals	s and event	ts, prizes/co				s, partici	pation in			
0	No	the pilot and high more that Morever Imagine. work on the by working at that we still	facilities of facilities of school studen 110 tours of the Imaginate of th	the FUEL4 lents arou s were giv negris pa ne Found ear projec rative, sus 14 1-2 gr nselves w technolo won the 1	ME project are local and during tours. In en to the general pricipated as super ation organises a viction to the solutions. Oups that work or hich country and wigical solutions. In 21st price overall in the solution to the country and with the solutions.	ted, we showed notated during the 4 sublic. visor in the high some and the second of the high some and the second of the grand the second of the grand of the gr	nany scho years of F school co ere stud d-world o FBR we s elated to are going roups of	on pupils EUEL4ME Empetion ents can countries supervise opic. The to tackle students			
	Which of Which of Was therefocus of the considered of the consid	Did you carry out Which of the fo Desi Set to Orga Actio Othe Was there a ger focus of the res considered and Yes- X No Synergies with so Did your project science festivals Yes-	Did you carry out specific Ge Which of the following act Design and imp Set targets to a Organise confe Actions to impr Other: Was there a gender diment focus of the research as, focus and addressed Yes- please spectors X No Synergies with Science Edu Did your project involve we science festivals and event and high-more that Morever Imagine. Work on the year students by using that we see the see t	Did you carry out specific Gender Equals Which of the following actions did you Design and implement and a set targets to achieve a get organise conferences and actions to improve work-lift organise conferences and actions to improve work-lift organise conferences and actions to improve work-lift organise considered and addressed? Was there a gender dimension association focus of the research as, for example, considered and addressed? Yes- please specify X No Synergies with Science Education Did your project involve working with science festivals and events, prizes/considered and high-school study and high-school study more than 110 tours. Morever, Dr. D. Klein Imagine. The Imagine work on their final years in the plant of the pl	Which of the following actions did you carry out Design and implement an equal opp Set targets to achieve a gender balance Organise conferences and workshop Actions to improve work-life balance O Other: Was there a gender dimension associated with focus of the research as, for example, consume considered and addressed? Yes- please specify X No Synergies with Science Education Did your project involve working with students science festivals and events, prizes/competitio Yes- please specify Open days were held at the p the pilot facilities of the FUEL4 and high-school students aroumore than 110 tours were giv Morever, Dr. D. Kleinegris pa Imagine. The Imagine Found work on their final year project by working on innovative, sus each year since 2014 1-2 gr students decide themselves we by using microalgae technolo that we supervised won the 2	Which of the following actions did you carry out and how effective Note effection Possign and implement an equal opportunity policy Set targets to achieve a gender balance in the workforce Organise conferences and workshops on gender Actions to improve work-life balance Other: Was there a gender dimension associated with the research content focus of the research as, for example, consumers, users, patients considered and addressed? Yes- please specify X No Synergies with Science Education Did your project involve working with students and/or school pursicence festivals and events, prizes/competitions or joint projects Yes- please specify Open days were held at the pilot facilities. For exithe pilot facilities of the FUEL4ME project are loca and high-school students around during tours. In more than 110 tours were given to the general phorever, Dr. D. Kleinegris participated as super Imagine. The Imagine Foundation organises and work on their final year project within a special content of the possible possible project within a special content of the possible possible project within a special content of the possible possible possible project within a special content of the possible possible project within a special content of the possible project of the possible project within a specia	Which of the following actions did you carry out and how effective were they? Not at all vieffective effective effective were they? Design and implement an equal opportunity policy Organise conferences and workshops on gender Organises on the considered and addressed? Was there a gender dimension associated with the research content Organises or in trials, was the considered and addressed? Yes- please specify X No Synergies with Science Education Did your project involve working with students and/or school pupils (e.g. open day science festivals and events, prizes/competitions or joint projects)? Yes- please specify Open days were held at the pilot facilities. For example at AlgaePA the pilot facilities of the FUEL4ME project are located, we showed in more than 110 tours were given to the general public. Morever, Dr. D. Kleinegris participated as supervisor in the high s Imagine. The Imagine Foundation organises a yearly contest when work on their final year project within a special context: helping thir by working on innovative, sustainable solutions. At Wageningen — each year since 2014 1-2 groups that work on a micro-algae in students decide themselves which country and what problem they a by using microalgae technological solutions. In 2015 one of the great and the great public in the pilot facilities.	Which of the following actions did you carry out and how effective were they? Not at all Very			



9.	Did the project generate any science education material (e.g. kits, websites, explanatory booklets, DVDs)?								
	0	Yes- please specify							
	•	No							
F	Interdisci	plinarity							
10.	Which dis	ciplines (see list below) are involved in y	our pi	oject?					
	•	Main discipline8: 1.5 and 2.3							
	0	Associated discipline ⁸ :	0	Associated discipline ⁸ :					
G	Engaging	with Civil society and policy makers							
11a	Did your p	project engage with societal actors beyon	nd the	research community? (if	•	Yes			
	'No', go	to Question 14)			0	No			
11b	 If yes, did you engage with citizens (citizens' panels / juries) or organised civil society (NGOs, patients' groups etc.)? No Yes- in determining what research should be performed 								
	0	Yes - in implementing the research	_						
	0	Yes, in communicating /disseminating /	using '	the results of the project	1	T			
11c	O Vas								
12.	Did you en organisation	ngage with government / public bodies ons)	r poli	cy makers (including internation	onal				
	0	No							
	0	Yes- in framing the research agenda							
	O Yes - in implementing the research agenda								
	•	Yes, in communicating /disseminating /		the results of the project					
13a	-	roject generate outputs (expertise or so	ientifi	advice) which could be used	by polic	Су			
	makers?								
	0	Yes – as a primary objective (please inc	licate	areas below- multiple answers	possible	e)			
	•	Yes – as a secondary objective (please	indicat	e areas below - multiple answ	er possi	ble)			
	0	No							

⁸ Insert number from list below (Frascati Manual).



13b If Yes, in which fields?								
Agriculture	х	Energy	х	Human rights				
Audiovisual and Media		Enlargement		Information Society				
Budget		Enterprise		Institutional affairs				
Competition		Environment		Internal Market				
Consumers		External Relations		Justice, freedom and security				
Culture		External Trade		Public Health				
Customs		Fisheries and Maritime		Regional Policy				
DevelopmentEconomic and		Affairs		Research and Innovation				
Monetary Affairs		Food Safety		Space				
Education, Training, Youth		Foreign and Security Policy		Taxation				
Employment and Social		Fraud		Transport				
Affairs		Humanitarian aid						



13c If Yes, at which level?								
O Local / regional levels								
O National level								
O European level								
O International level								
H Use and dissemination								
14. How many Articles were published/accepted for reviewed journals?	public	cation	in peer-	12				
To how many of these is open access ⁹ provided?				0				
How many of these are published in open access j	ourna	ls?		0				
How many of these are published in open reposito	ries?			12				
To how many of these is open access not provided?				0				
Please check all applicable reasons for not providi	ng ope	en acce	ess:					
☐ publisher's licensing agreement would not permit pu	ıblishi	ng in a	repository					
no suitable repository available								
no suitable open access journal available	_1							
no funds available to publish in an open access journlack of time and resources	aı							
□ lack of information on open access								
□ other ¹⁰ :								
15. How many new patent applications ('priority fili	ings'\	have h	een made?		1			
("Technologically unique": multiple applications f				ent	_			
jurisdictions should be counted as just one applic			• • •					
16. Indicate how many of the following Intellectual	Prope	erty	Trademark		0			
Rights were applied for (give number in each bo	x).		Registered design	n 0				
			Other		0			
17. How many spin-off companies were created / are project?	e plan	ned as	a direct result of	the	0			
	niosi	N/A						
Indicate the approximate number of a			-					
18. Please indicate whether your project has a poter	ntial in	npact	on employment, i	n com	parison with the			
situation before your project:	.	ln cm	محنه مسيناه مس 9 الميا	d onto	arnricos			
■ Increase in employment, or□ Safeguard employment, or			all & medium-size ge companies	u ente	51 h11262			
Decrease in employment,			• .	t relev	ant to the project			
Difficult to estimate / not possible to	_	1,0110	. S. the above / Ho	C I CIC	rant to the project			
quantify								

⁹ Open Access is defined as free of charge access for anyone via Internet.



19.	fo dir ful	Indicate figure:				
Diff	ficult	to estimate / not possible to quanti	fy			
I	M	edia and Communication to the ge	neral p	oublic		
20.		s part of the project, were any of the	ne ben	eficiaries p	rofessionals in communication	n or media
		Yes	0	No		
21.		d professional media / commu ublic?	nication training /			
22		hich of the following have been us ublic, or have resulted from your p			ate information about your pro	pject to the general
	 ■ Press Release ■ Media briefing □ TV coverage / report □ Radio coverage / report □ Brochures / posters / flyers □ DVD /Film /Multimedia □ Coverage in specialist press □ Coverage in national press □ Coverage in international press □ Website for the general public / internet □ Event targeting general public (festival, conference, exhibition, science café) 					
23	In	which languages are the informati	on pro	ducts for t	the general public produced?	
		Language of the coordinator Other language(s)			English	

Question F-10: Classification of Scientific Disciplines according to the Frascati Manual 2002 (Proposed Standard Practice for Surveys on Research and Experimental Development, OECD 2002):

FIELDS OF SCIENCE AND TECHNOLOGY

1. NATURAL SCIENCES

- 1.1 Mathematics and computer sciences [mathematics and other allied fields: computer sciences and other allied subjects (software development only; hardware development should be classified in the engineering fields)]
- 1.2 Physical sciences (astronomy and space sciences, physics and other allied subjects)

¹⁰ For instance: classification for security project.



- 1.3 Chemical sciences (chemistry, other allied subjects)
- 1.4 Earth and related environmental sciences (geology, geophysics, mineralogy, physical geography and other geosciences, meteorology and other atmospheric sciences including climatic research, oceanography, vulcanology, palaeoecology, other allied sciences)
- 1.5 Biological sciences (biology, botany, bacteriology, microbiology, zoology, entomology, genetics, biochemistry, biophysics, other allied sciences, excluding clinical and veterinary sciences)

2 ENGINEERING AND TECHNOLOGY

- 2.1 Civil engineering (architecture engineering, building science and engineering, construction engineering, municipal and structural engineering and other allied subjects)
- 2.2 Electrical engineering, electronics [electrical engineering, electronics, communication engineering and systems, computer engineering (hardware only) and other allied subjects]
- 2.3. Other engineering sciences (such as chemical, aeronautical and space, mechanical, metallurgical and materials engineering, and their specialised subdivisions; forest products; applied sciences such as geodesy, industrial chemistry, etc.; the science and technology of food production; specialised technologies of interdisciplinary fields, e.g. systems analysis, metallurgy, mining, textile technology and other applied subjects)

3. MEDICAL SCIENCES

- 3.1 Basic medicine (anatomy, cytology, physiology, genetics, pharmacy, pharmacology, toxicology, immunology and immunohaematology, clinical chemistry, clinical microbiology, pathology)
- 3.2 Clinical medicine (anaesthesiology, paediatrics, obstetrics and gynaecology, internal medicine, surgery, dentistry, neurology, psychiatry, radiology, therapeutics, otorhinolaryngology, ophthalmology)
- 3.3 Health sciences (public health services, social medicine, hygiene, nursing, epidemiology)

4. AGRICULTURAL SCIENCES

- 4.1 Agriculture, forestry, fisheries and allied sciences (agronomy, animal husbandry, fisheries, forestry, horticulture, other allied subjects)
- 4.2 Veterinary medicine

5. SOCIAL SCIENCES

- 5.1 Psychology
- 5.2 Economics
- 5.3 Educational sciences (education and training and other allied subjects)
- 5.4 Other social sciences [anthropology (social and cultural) and ethnology, demography, geography (human, economic and social), town and country planning, management, law, linguistics, political sciences, sociology, organisation and methods, miscellaneous social sciences and interdisciplinary, methodological and historical S1T activities relating to subjects in this group. Physical anthropology, physical geography and psychophysiology should normally be classified with the natural sciences].

6. HUMANITIES



- 6.1 History (history, prehistory and history, together with auxiliary historical disciplines such as archaeology, numismatics, palaeography, genealogy, etc.)
- 6.2 Languages and literature (ancient and modern)
- 6.3 Other humanities [philosophy (including the history of science and technology) arts, history of art, art criticism, painting, sculpture, musicology, dramatic art excluding artistic "research" of any kind, religion, theology, other fields and subjects pertaining to the humanities, methodological, historical and other S1T activities relating to the subjects in this group]



7 Project partners and further project details

Partnership

The consortium consists of a powerful mix of established research organisations and universities, small and medium enterprises (55% of total partners) and large-scale industry. These are:

Table 7.1 Partners within FUEL4ME

Participant no.	Participant legal name	Participant short name	Country	Organisation type*
1	Stichting Wageningen Research	WRS	NL	R.O.
2	Wageningen Universiteit	WU	NL	R.O.
3	Ben Gurion University of the Negev	BGU	IL	R.O.
4	Fotosintetica & Microbiologica S.r.l.	F&M	IT	SME
5	Biotopic	BIT	DK	SME
6	Evodos BV	Evodos	NL	SME
7	Pursuit Dynamics PLC	PDX	UK	SME
8	Feyecon Development and implementation BV	Feyecon	NL	SME
9	Neste Oil Corporation	NOIL	FI	Со
10	Joanneum Research Forschungsgesellschaft MBH	JOANNEUM	AT	R.O.
11	IDConsortium SL	IDC	ES	SME
12	Cellulac	Cellulac	UK	SME
14	PROVIRON	PROVIRON	BE	Со
15	Norsker Investigaciones	NIS	ES	SME





Project public website

www.fuel4me.eu

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