Algemene gegevens	
Nummer	AF-EU-13007
Titel	FUEL4ME
Topsector (A&F of T&U)	A&F
Projectleider (onderzoek)	Dorinde Kleinegris
Contactpersoon overheid	Cor Wever
Status (lopend of afgerond)	Lopend
Type onderzoek (F, T of V)	T
Werkelijke startdatum	01-01-2013
Werkelijke einddatum	01-01-2017
Korte omschrijving inhoud	The 4-year FUEL4ME project will develop a sustainable chain for continuous biofuel production using microalgae as a production platform, thereby making 2nd generation biofuels competitive alternatives to fossil fuels.

Highlights:

FUEL4ME: Future European league for microalgal energy

Project context and objectives

The 4-year FUEL4ME project will develop a sustainable chain for continuous biofuel production using microalgae as a production platform, thereby making 2nd generation biofuels competitive alternatives to fossil fuels. This will be achieved by:
1) Transforming the current 2-step process for algal lipid production into a continuous 1-step process with high lipid content (production process);
2) Development of a continuous downstream process using all components of the algal biomass (conversion process);
3) Integration of production and conversion process.

After setting up a proof of concept within controlled indoor conditions, the continuous process will be tested outdoors under real production conditions in four different regions (NL, IL, IT ES). Simultaneous with research on biomass production, a continuous downstream process will be developed. Finally the whole process (both biomass production and conversion into biofuel) will be integrated and subjected to an economic analysis and life cycle analysis.

FUEL4ME aims 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 (ω3 fatty acids). This way optimal use of biomass results in simultaneous production of food and fuel.

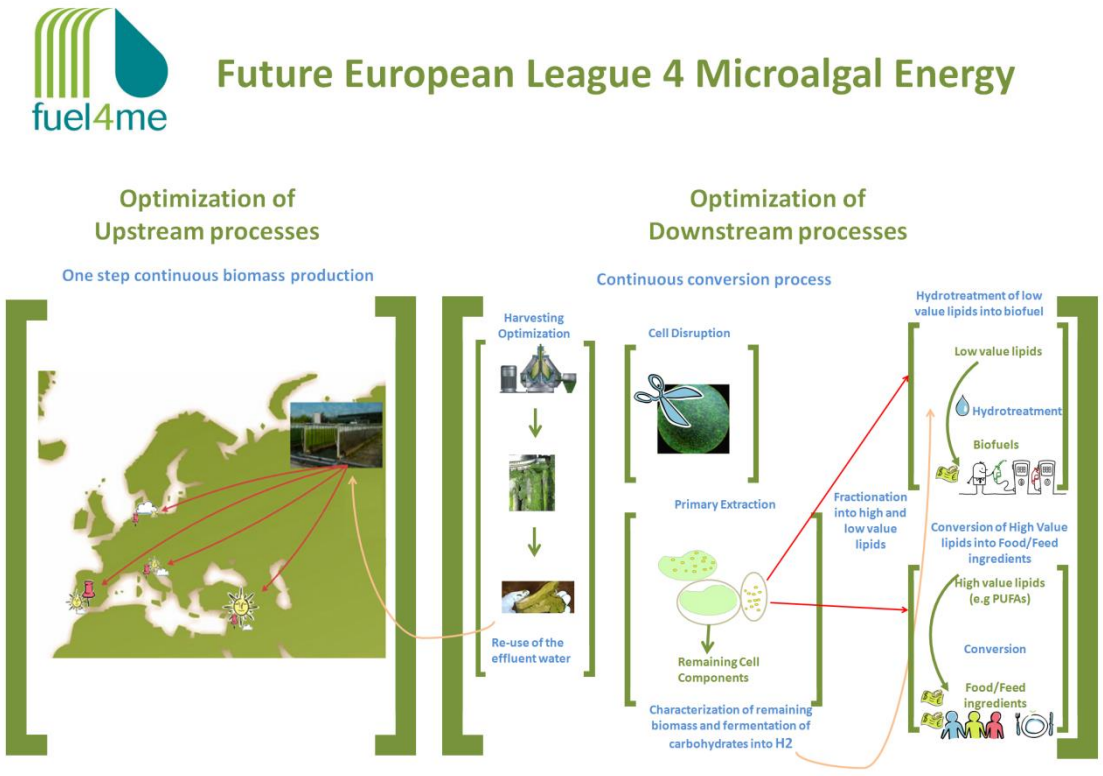

Figure 1. Schematic project overview

Main results achieved so far

The kick-off meeting was organised 5 – 6 March 2013 where total project overview, scientific dependencies, project obligations, reporting, meetings, legal and financial aspects and also the importance to act as a team were intensively discussed and agreed upon. In the course of the project, regularly project meetings have been organised with the consortium partners, and also with our scientific advisory board to discuss progress of the project.

In the first reporting period (January 2013 – June 2014) work has started on fundamental research and enabling technologies: understanding the genes, metabolism, biochemical aspects of the chosen strains (*Nannochloropsis oceanica* and *Phaeodactylum tricornutum*). In the second reporting period (July 2014 – June 2015) more knowledge on the formation and migration of intracellular lipid globules was generated. This knowledge should enable us to understand and finally improve the lipid productivity of our process. Furthermore, the proof of principle concept for a one-step lipid production process was developed. In the second reporting period we have shown that an optimized one-step process with *P. tricornutum* resulted in an 8-fold higher TAG yield on light (88 mg/mol PAR-photons) and a similar TAG content (27% of dry weight) compared to a two-step process with full nitrogen depletion, at same incident light intensity. Further optimization of the one-step process for *P. tricornutum* seems limited mainly by the capacity of the organism itself (i.e. to remain photosynthetically efficient during nitrogen limitation). In work package 2, where the translation from outdoors to production will be made, detailed engineering of the pilot plants was performed. Flat panel photobioreactors were installed in Israel and Italy and the Netherlands and a larger demonstration plant consisting of ProviAPT photobioreactors has been installed in Spain. At the smaller pilot plants experiments

have been performed under various nitrogen sufficient and limited conditions with both *Nannochloropsis* and *Phaeodactylum*, and in batch and semi-continuous mode. These trials allowed for the collection of input data for our life cycle analysis (WP 5), next to of course understanding and input for process optimization of lipid production with both algae species. Furthermore, biomass has been produced for downstream processing experiments (WP 3) both in Italy and in Spain. Harvesting with the Evodos Type 10 of *Nannochloropsis* and *Phaeodactylum* was successfully demonstrated. The tests demonstrated that > 95% of the algae can be harvested at highest capacity of the pilot scale Evodos. Preliminary tests were done with supersonic flow processing equipment for *Nannochloropsis* cell disruption to achieve the maximum lipid release. The first experiments on extraction with supercritical carbon dioxide and subsequent characterization of fatty acids have been performed. Further tuning of process conditions improved the recovery of the omega 3 PUFAs significantly (from 30% to more than 80%) and the purity was increased from 50% to more than 85%. A further fractionation of the raffinate resulted a distillate free of contaminants with an increase of EPA content.
Objective of WP 5 is to perform a sustainability assessment, including environmental, economic and social elements based on the whole value chain in a life cycle perspective. For the assessment of the FUEL4ME integrated process a modelling approach was developed to describe a possible future commercial HVO production from microalgae by giving guiding values for some key technological and economic data. Three cases with a production capacity of 100 kt/a HVO and coproducing PUFA are modelled. For the modelling of algae oil production, targets, characteristics, data and assumptions were defined with strong interaction of project partners. These targets give possible framework conditions for a future commercial algae oil production.
In WP 6, concerning communication and exploitation, a communication and dissemination plan, website, newsletter, roll-up, project brochure have been established and are continuously updated. Moreover, a stakeholder list has been issued and an industrial board and an online advisory service have been set up. Partners have been very active in disseminating the scientific results in various conferences and other meetings.

Expected final results and their potential impact and use
These key outcomes from this 4-year initiative will foster the production of sustainable biofuels in an economically, socially, and environmentally manner and to alleviate possible problems regarding competition with food in the bioenergy field:

- A sustainable process for biofuel production from microalgae demonstrated at pilot scale.
- Optimisation of lipid productivity in an integrated microalgae cultivation process based on photobioreactors
- Improvement in the extraction of oil from microalgae and conversion into biofuel.
- Commercial viability of the process by valorisation of the residual biomass
- Enabling a sustainable approach that can be replicated elsewhere across Europe.

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: Wageningen UR – Food & Biobased 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 Oil Corporation, JOANNEUM RESEARCH

Forschungsgesellschaft mbH and IDconsortium SL. (see also http://www.fuel4me.eu)

Project public website
www.fuel4me.eu

Zie voor KOL: de bijlage hieronder

Link naar Kennisonline:
http://www.wageningenur.nl/nl/project/Fuel4Me-1.htm

https://www.wageningenur.nl/en/show/FUEL4ME-Future-European-League-for-Microalgal-Energy.htm

http://www.algaeparc.com/project/3/eu-projects/2/fuel4me

http://www.fuel4me.eu/index.php?option=com_jdownloads&view=category&catid=9&Itemid=258

Conferences and Publications

Event	Date & Venue	Partners attending	Event's Abstract & partners contributions
CommNet dissemination training	12th-13th February, 2013	IDC	CommNet offered a seminar on different guidelines and tips to design an effective communication strategy for EU project on biotechnology. As a part of activities, IDC presented overall goal and objectives of FUEL4ME
9th ECCE Congress European Conference on Chemical Engineering	21st-25th April 2013, the Hague	EVODOS	International conference with around 1900 participants, meeting to discuss about new ideas, innovations and research results on all aspects of chemical engineering, applied biotechnology and process intensification. EVODOS presented FUEL4ME project.

Event	Date & Venue	Partners attending	Event's Abstract & partners contributions
European Algae Biomass	24th April 2013, Vienna	EVODOS	Conference on case study examples of latest technologies in operation in the global algae industry and low-cost production of microalgae, focusing on technical challenges faced when optimising the cultivation of algae, the current and future commercial markets for algae products. Over 200 hundred participants. Evodos presented FUEL4ME project and distributed FUEL4ME leaflets.
ALGAENET workshop	3rd-4th July 2013 , Seville	BIOTOPIC	International workshop with participants from industry and academia on the potential uses of algae for the production of biofuels and products of added value. Mr. Norsker (BIOTOPIC) presented the objective and progress of activities of FUEL4ME project.
Algae Biomass Summit, ABO	30th Sept.-3rd Oct.2013, Orlando	EVODOS, DLO	Around 600 industry professionals from all sectors of the world's algae utilization industries including financing, algal ecology, genetic systems, carbon partitioning, engineering & analysis, biofuels, animal feeds, fertilizers, bioplastics, supplements and foods. A presentation on FUEL4ME was made
Austrian Stakeholder Workshop of IEA Bioenergy Task 42 „Biorefining"	24th Oct. 2013, Graz	JOANNEUM , WU	IEA Bioenergy Task 42 "Biorefining" organized a workshop for industry and research to discuss the future role of biorefining. Presentations from international experts, Austrian industry and research experts have been made. Within these presentations demo plants and research activities in the field of biorefining have been presented. Ms. Maria Hingsamer presented as well the project FUEL4ME.

Event	Date & Venue	Partners attending	Event's Abstract & partners contributions
Algen als biogene Ressource – Akteure in Österreich	6th Nov., Wieselburg	JOANNEUM	Workshop (23 participants) organized by Austrian stakeholders in the field of microalgae. Selected organizations (industry and research) have explained their activities in the field of microalgae. Within the presentation of JOANNEUM RESEARCH, project FUEL4ME was also presented.
International Algae Congress	4-5th Dec. 2013, Hamburg	EVODOS	This congress focused specifically on the potential of micro/macro algae in aquaculture and agriculture. Over 150 scientists, authorities and private entities participated. Evodos presented their activities in FUEL4ME project and distributed FUEL4ME leaflets.
EnInnov2014 – 13th Symposium Energy Innovation	12-14 Feb. 2014 , Graz	JOANNEUM	The symposium energy innovation (600 participants) raises topics concerning application of energy (renewable energy, innovative energy technologies), energy distribution systems, demand-side measures (energy saving, energy efficiency, energy management), regulatory issues and as well the development of the European economic system. Within the presentation "Sustainability of Algae Energy Systems – Modelling and Case Studies" the project FUEL4ME and the concept for the assessment has been presented next to other projects in the field of algae energy systems.
Algae'Chem conference	31st March-3rd April 2014, Montpellier	WU, F&M	International conference with over 300 participants on the future for algae in the industry. WU gave a scientific presentation on the progress of the work of BPE in FUEL4ME WP1
YAS conference	3rd-5th April 2014, Montpellier	WU, F&M	YAS conference gathers young scientist to discuss and share knowledge on algal research. Presentations are given in an informal setting to a group of ~100 PhD students. Ms. Ilse Remmers made a presentation on her activities in FUEL4ME regarding "Lipid production

Event	Date & Venue	Partners attending	Event's Abstract & partners contributions
			in *Phaeodactylum tricornutum* under simulated outdoor conditions" WU gave a scientific presentation on the progress of the work of BPE in FUEL4ME WP1
2nd European Workshop on Life Cycle Analysis of algal based Biofuels	24th April, Brussels	JOANNEUM ,DLO,WU	Kyriakos Maniatis (EC) invited JOANNEUM to a workshop concerning the LCA of algal based biofuels and biomaterials. The approach of different European Projects have been presented and discussed. Ms. Maria Hingsamer presented the project FUEL4ME, especially the activities in the Life Cycle Sustainability Assessment, mainly the Life Cycle Analysis. The methodology (system boundaries, indicators, impacts, functional unit and so on), the FUEL4ME integrated process and future challenges have been presented. Mr. Gerfried Jungmeier was asked to take part at the final panel discussion.
22nd European Biomass Conference and Exhibition	23rd-26th June 2014, Hamburg	JOANNEUM	The EU BC&E is international conference on entire value chain of biomass to conduct business, to network, and to present and discuss the latest developments and innovations. It brings together the leading European researchers, engineers, technologists, standards organisations, policy and decision makers, financing institutions in this field. Ms. Maria Hingsamer gave a presentation: "Towards a Standard Methodology for the Sustainability Assessment of Energy Systems with Algae – An European Approach in FUEL4ME".
Algae Event 2014	25th June 2014, Hamburg	JOANNEUM	The Algae Event 2014 (around 100 participants) was organized within the project EnAlgae and discussed the topics cultivation, conversion methods, products and sustainability. Ms. Maria Hingsamer presented the concept of the FUEL4ME integrated process and gave some information about the Life Cycle Sustainability Assessment.

Event	Date & Venue	Partners attending	Event's Abstract & partners contributions
Dutch conference on biotechnology	27-28th May 2014, Ede	WU	The Dutch conference on biotechnology s a bi-annual conference with the goal to bring together students and researchers from academia, research institute and industry and entrepreneurs active in the field of biotechnology. The number of participants was over 150. The title of the 2014 edition is: Biotechnology by Dutch Design and will highlight scientific and business successes that have their roots in The Netherlands. WU made a presentation of FUEL4ME project on one of the parallel sessions.
Algae as a biogenic resource – Research in Austria (AlgenalsbiogeneRes source – Forschung in Österreich)	11th September 2014, Duernrohr, Austria	JOANNEUM	For Austrian stakeholders in the field of microalgae a workshop has been organized. Selected organizations (industry and research) presented their activities in the field of microalgae. Within the presentation of JOANNEUM RESEARCH the project FUEL4ME has been presented. JOANNEUM RESEARCH distributed FUEL4ME leaflets.
Algae Biomass Summit, ABO	29th Sept.-2rd Oct.2014, An Diego	EVODOS, DLO-FBR	Around 600 industry professionals from all sectors of the world's algae utilization industries including financing, algal ecology, genetic systems, carbon partitioning, engineering & analysis, biofuels, animal feeds, fertilizers, bioplastics, supplements and foods. A presentation by EVODOS on FUEL4ME was made.
Algatec II workshop	11.12.2014, Huelva, Spain	BIT/NIS	The workshop was a final dissemination activity in the FP-7 project Algatec II which examined the use of microalgae in agro-industrial wastewater purification. Microalgae were applied to clean washing water from olive processing. The number of participants were about 30.

Event	Date & Venue	Partners attending	Event's Abstract & partners contributions
AOAIS, South Korea	17th-20th NOV, Seoul	EVODOS	International conference joining the main Asian stakeholders in the production and industrial uses of algae biomass. EVODOS presented FUEL4ME project
EABA Expo and conference	3rd December 2014, Florence	DLO-FBR, F&M	International conference joining the main European stakeholders in the production and industrial uses of algae biomass. DLO and F&M presented FUEL4ME project
3rd European Workshop "Life Cycle Analysis of Algal based Biofuels& Biomaterials"	11th May 2015, Brussels	WU, DLO-FBR	Kyriakos Maniatis (EC) invited JOANNEUM to a workshop concerning the LCA of algal based biofuels and biomaterials. Some LCA-results of different European Projects have been presented and discussed. Gerfried Jungmeier presented the project FUEL4ME, he spoke about "Feasibility of Algae Cultivation for Large Scale Biofuel Production – Aspects of Up-scaling and Infrastructure Integration of Algae Biorefineries."

Scientific publications

LIST OF SCIENTIFIC (PEER REVIEWED) PUBLICATIONS, STARTING WITH THE MOST IMPORTANT ONES										
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)**	**Is/Will open access 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.wiley-vch.de/publish/en/journals/alphabeticIndex/2114/	No

LIST OF SCIENTIFIC (PEER REVIEWED) PUBLICATIONS, STARTING WITH THE MOST IMPORTANT ONES										
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)**	**Is/Will open access 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	Wageningen, The Netherlands	2014	3191–3199	http://pubs.acs.org/doi/full/10.1021/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 Biotechnology of the University of Almería. Almería, Spain	Almeria, Spain	2014	N/A	N/A	No

LIST OF SCIENTIFIC (PEER REVIEWED) PUBLICATIONS, STARTING WITH THE MOST IMPORTANT ONES										
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)**	**Is/Will open access provided to this publication?**
4	""Expression and localization of *Haematococcus pluvialis* oil globule protein in *Phaeodactylum tricornutum*" (scientific presentation)	Shemesh, Zachor. Leu, Stefan. Khozin-Goldberg, Inna. Boussiba, Sammy	4th Int. CeBiTec Research Conference Advances in Industrial Biotechnology: Prospects and challenges for the development of algal.	N/A	N/A	Biotechnology Center for Interdisciplinary Research (ZiF), Bielefeld University, Germany	2014	N/A	N/A	No
5	"Expression and localization of an oil globule protein in *Phaeodactylum tricornutum* using an endogenous stress-	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	N/A

LIST OF SCIENTIFIC (PEER REVIEWED) PUBLICATIONS, STARTING WITH THE MOST IMPORTANT ONES										
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)**	**Is/Will open access provided to this publication?**
	activated promoter" (scientific presentation)									
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	Wageningen, The Netherlands	2013	N/A	http://tinyurl.com/nd67ddv	Yes
7	Lipid production in *Phaeodactylum tricornutum* under simulated outdoor conditions	Ilse M. Remmers, Packo P. Lamers, Dirk E. Martens, René H.	YAS conference	N/A	N/A	Montpellier, France	2014	N/A	N/A	N/A

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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)**	**Is/Will open access provided to this publication?**
	(scientific presentation)	Wijffels								
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	N/A
9	Two stage Lipid production in *Phaeodactylum tricornutum (scientific presentation)*	Ilse M. Remmers, Packo P. Lamers, Dirk E. Martens, René	NBC15 conference	N/A	N/A	Ede, The Netherland	2014	N/A	N/A	N/A

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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)**	**Is/Will open access provided to this publication?**
		H. Wijffels								

List of Expected Publications until the end of the project:

LIST OF SCIENTIFIC OF EXPECTED PUBLICATIONS						
No.	**Partner**	**Title**	**Main author**	**Description**	**Expected Date of Publication**	**Is/Will open access provided to this publication?**
1	WU-DLO	*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,	experiments finished, analyses ongoing and manuscript in preparation	June 2016	YES
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	experiments finished and manuscript in preparation	February 2016	YES
3	WU	*Dynamics of photosynthesis and carbon partitioning during microalgal lipid production in a continuous cultivation system*	Ilse M. Remmers, A. Hidalgo, B. Brandt, René H. Wijffels, Packo P. Lamers,	experiments ongoing and manuscript in preparation - expected date of publication	July 2016	YES
4	WU	*Effect of day/night cycles on carbon partitioning in Scenedesmus obliquus in nitrogen replete and limited conditions*	Ilse M. Remmers, Graciela M. Leon Saiki, Packo P. Lamers, Douwe van der Veen, Dirk E. Martens, René H. Wijffels	experiments ongoing and manuscript in preparation	November 2016	YES

LIST OF SCIENTIFIC OF EXPECTED PUBLICATIONS						
No.	**Partner**	**Title**	**Main author**	**Description**	**Expected Date of Publication**	**Is/Will open access provided to this publication?**
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 editeing bsfore sending to publication	March 2016	
6	BGU	Isolation and characterization of novel promoters for constitutive and inducible expression of transgenes in the biotechnologically important diatom P. tricornutum under variable conditions, including nitrogen starvation	Zachor Shemesh, Stefan Leu, Aliza Zarka, Inna Khozin-Goldberg, and Sammy Boussiba	Writing	Jun-16	
7	BGU	IBD	Zachor Shemesh, Stefan Leu, Aliza Zarka, Inna Khozin-Goldberg, and Sammy Boussiba	Experiments ongoing	November 2016	