

# UNIDAD DE VIGILANCIA TECNOLÓGICA E INTELIGENCIA COMPETITIVA

## Microalgas

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Juan Ignacio Gori

Fernando Carlos Zelaschi

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En este boletín se presentan las publicaciones, patentes y noticias de interés del segundo trimestre del año 2018 pertenecientes a la rama Bioproductos del árbol de categorías.

## PUBLICACIONES

### **An endogenous microRNA (miRNA1166.1) can regulate photobio-H<sub>2</sub> production in eukaryotic green alga *Chlamydomonas reinhardtii*.**

Biotechnol Biofuels. 2018;11:126

Authors: Wang Y, Zhuang X, Chen M, Zeng Z, Cai X, Li H, Hu Z

#### Abstract

**Background:** Hydrogen photoproduction from green microalgae is regarded as a promising alternative solution for energy problems. However, the simultaneous oxygen evolution from microalgae can prevent continuous hydrogen production due to the hypersensitivity of hydrogenases to oxygen. Sulfur deprivation can extend the duration of algal hydrogen production, but it is uneconomical to alternately culture algal cells in sulfur-sufficient and sulfur-deprived media.

**Results:** In this study, we developed a novel way to simulate sulfur-deprivation treatment while constantly maintaining microalgal cells in sulfur-sufficient culture medium by overexpressing an endogenous microRNA (miR1166.1). Based on our previous RNA-seq analysis in the model green alga *Chlamydomonas reinhardtii*, three endogenous miRNAs responsive to sulfur deprivation (cre-miR1166.1, cre-miR1150.3, and cre-miR1158) were selected. Heat-inducible expression vectors containing the selected miRNAs were constructed and transformed into *C. reinhardtii*. Comparison of H<sub>2</sub> production following heat induction in the three transgenic strains and untransformed control group identified miR1166.1 as the best candidate for H<sub>2</sub> production regulation. Moreover, enhanced photobio-H<sub>2</sub> production was observed with repeated induction of miR1166.1 expression.

**Conclusions:** This study is the first to identify a physiological function of endogenous miR1166.1 and to show that a natural miRNA can regulate hydrogen photoproduction in the unicellular model organism *C. reinhardtii*.



## Anaerobic phototrophic processes of hydrogen production by different strains of microalgae *Chlamydomonas* sp.

FEMS Microbiol Lett. 2018 May 01;365(9):

Authors: Vargas SR, Santos PVD, Giraldi LA, Zaiat M, Calijuri MDC

### Abstract

Hydrogen is an abundant element and a non-polluting fuel that can be biologically produced by microalgae. The aim of this research was to investigate biological hydrogen production by *Chlamydomonas reinhardtii* (CC425) and *Chlamydomonas moewusii* (SAG 24.91) by direct biophotolysis in batch cultures. Strains were cultivated in TAP growth medium (pH 7.2) in two phases: in the first stage, cultures were maintained in an aerobic condition until the middle of the exponential phase; in the second stage, the biomass was transferred to closed anaerobic photobioreactors under sulfur deprived. Gas chromatography and Gompertz model were used to measure the hydrogen production and hydrogen production rate, respectively. We noticed that maximum hydrogen production by biomass of *C. reinhardtii* was  $5.95 \pm 0.88 \mu\text{mol mg}^{-1}$  and the productivity was  $17.02 \pm 3.83 \mu\text{mol L}^{-1} \text{h}^{-1}$ , with hydrogen production five times higher than *C. moewusii*, approximately, though, *C. moewusii* obtained a higher ethanol yield compared to *C. reinhardtii*. The hydrogen production method, with the cultivation of strains in two different phases and sulfur deprivation, was effective for obtaining of biohydrogen for *Chlamydomonas*; however, it depends on the species, strain and growth conditions.

## Effect of carbon limitation on photosynthetic electron transport in *Nannochloropsis oculata*.

J Photochem Photobiol B. 2018 Apr;181:31-43

Authors: Zavřel T, Szabó M, Tamburic B, Evenhuis C, Kuzhiumparambil U, Literáková P, Larkum AWD, Raven JA, Červený J, Ralph PJ

### Abstract

This study describes the impacts of inorganic carbon limitation on the photosynthetic efficiency and operation of photosynthetic electron transport pathways in the biofuel-candidate microalga *Nannochloropsis oculata*. Using a combination of highly-controlled cultivation setup (photobioreactor), variable chlorophyll a fluorescence and transient spectroscopy methods (electrochromic shift (ECS) and P700 redox kinetics), we showed that net photosynthesis and effective quantum yield of Photosystem II (PSII) decreased in *N. oculata* under carbon limitation. This was accompanied by a transient increase in total proton motive force



and energy-dependent non-photochemical quenching as well as slightly elevated respiration. On the other hand, under carbon limitation the rapid increase in proton motive force (PMF, estimated from the total ECS signal) was also accompanied by reduced conductivity of ATP synthase to protons (estimated from the rate of ECS decay in dark after actinic illumination). This indicates that the slow operation of ATP synthase results in the transient build-up of PMF, which leads to the activation of fast energy dissipation mechanisms such as energy-dependent non-photochemical quenching. *N. oculata* also increased content of lipids under carbon limitation, which compensated for reduced NAPDH consumption during decreased CO<sub>2</sub> fixation. The integrated knowledge of the underlying energetic regulation of photosynthetic processes attained with a combination of biophysical methods may be used to identify photo-physiological signatures of the onset of carbon limitation in microalgal cultivation systems, as well as to potentially identify microalgal strains that can better acclimate to carbon limitation.

### **Characterization of light-dependent hydrogen production by new green microalga *Parachlorella kessleri* in various conditions.**

J Photochem Photobiol B. 2017 Oct;175:207-210

Authors: Gabrielyan L, Hakobyan L, Trchounian A

#### **Abstract**

Nowadays, hydrogen (H<sub>2</sub>) production by green microalgae seems to be a very perspective, as stocks of water and solar energy are practically inexhaustible and renewable. The aim of this study was the optimization of conditions (organic carbon sources and lighting regime), which can provide light-dependent H<sub>2</sub> production by green microalga *Parachlorella kessleri* RA-002 newly isolated in Armenia. The results indicated that carbon sources and lighting regimes affected H<sub>2</sub> production. In the presence of used carbon sources H<sub>2</sub> production was observed, but the highest yield of H<sub>2</sub> was obtained in the presence of acetate. It was 2-fold higher than the H<sub>2</sub> yield determined in the presence of glucose. The increase of H<sub>2</sub> production might be connected with the stimulation of H<sub>2</sub>-producing enzyme - [Fe]-hydrogenase synthesis. The data obtained show that acetate can be used as an effective carbon source in H<sub>2</sub> production. H<sub>2</sub> production by microalga (in the presence of acetate and glucose) was enhanced by 1.5-2.5-fold in comparison with continuously illuminated algal cells, when *P. kessleri* was illuminated during 24h, and then was moved in the darkness. H<sub>2</sub> yield increase is possible due to hydrogenase activation and the creation of anaerobic conditions. This study was significant to find out available effective substrates and optimal lighting regime to provide with light-dependent H<sub>2</sub> production by microalgae.



## Compartmentalisation of [FeFe]-hydrogenase maturation in *Chlamydomonas reinhardtii*.

Plant J. 2017 Jun;90(6):1134-1143

Authors: Sawyer A, Bai Y, Lu Y, Hemschemeier A, Happe T

### Abstract

Molecular hydrogen ( $H_2$ ) can be produced in green microalgae by [FeFe]-hydrogenases as a direct product of photosynthesis. The *Chlamydomonas reinhardtii* hydrogenase HYDA1 contains a catalytic site comprising a classic [4Fe4S] cluster linked to a unique 2Fe sub-cluster. From in vitro studies it appears that the [4Fe4S] cluster is incorporated first by the housekeeping FeS cluster assembly machinery, followed by the 2Fe sub-cluster, whose biosynthesis requires the specific maturases HYDEF and HYDG. To investigate the maturation process in vivo, we expressed HYDA1 from the *C. reinhardtii* chloroplast and nuclear genomes (with and without a chloroplast transit peptide) in a hydrogenase-deficient mutant strain, and examined the cellular enzymatic hydrogenase activity, as well as in vivo  $H_2$  production. The transformants expressing HYDA1 from the chloroplast genome displayed levels of  $H_2$  production comparable to the wild type, as did the transformants expressing full-length HYDA1 from the nuclear genome. In contrast, cells equipped with cytoplasm-targeted HYDA1 produced inactive enzyme, which could only be activated in vitro after reconstitution of the [4Fe4S] cluster. This indicates that the HYDA1 FeS cluster can only be built by the chloroplastic FeS cluster assembly machinery. Further, the expression of a bacterial hydrogenase gene, CPI, from the *C. reinhardtii* chloroplast genome resulted in  $H_2$ -producing strains, demonstrating that a hydrogenase with a very different structure can fulfil the role of HYDA1 in vivo and that overexpression of foreign hydrogenases in *C. reinhardtii* is possible. All chloroplast transformants were stable and no toxic effects were seen from HYDA1 or CPI expression.



## **The dual effect of a ferredoxin-hydrogenase fusion protein in vivo: successful divergence of the photosynthetic electron flux towards hydrogen production and elevated oxygen tolerance.**

Biotechnol Biofuels. 2016;9(1):182

Authors: Eilenberg H, Weiner I, Ben-Zvi O, Pundak C, Marmari A, Liran O, Wecker MS, Milrad Y, Yacoby I

### **Abstract**

**BACKGROUND:** Hydrogen photo-production in green algae, catalyzed by the enzyme [FeFe]-hydrogenase (HydA), is considered a promising source of renewable clean energy. Yet, a significant increase in hydrogen production efficiency is necessary for industrial scale-up. We have previously shown that a major challenge to be resolved is the inferior competitiveness of HydA with NADPH production, catalyzed by ferredoxin-NADP(+)-reductase (FNR). In this work, we explored the in vivo hydrogen production efficiency of Fd-HydA, where the electron donor ferredoxin (Fd) is fused to HydA and expressed in the model organism *Chlamydomonas reinhardtii*.

**RESULTS:** We show that once the Fd-HydA fusion gene is expressed in micro-algal cells of *C. reinhardtii*, the fusion enzyme is able to intercept photosynthetic electrons and use them for efficient hydrogen production, thus supporting the previous observations made in vitro. We found that Fd-HydA has a ~4.5-fold greater photosynthetic hydrogen production rate standardized for hydrogenase amount (PHPRH) than that of the native HydA in vivo. Furthermore, we provide evidence suggesting that the fusion protein is more resistant to oxygen than the native HydA.

**CONCLUSIONS:** The in vivo photosynthetic activity of the Fd-HydA enzyme surpasses that of the native HydA and shows higher oxygen tolerance. Therefore, our results provide a solid platform for further engineering efforts towards efficient hydrogen production in microalgae through the expression of synthetic enzymes.

## **Evaluation of the ethanol tolerance for wild and mutant *Synechocystis* strains by flow cytometry.**

Biotechnol Rep (Amst). 2018 Mar;17:137-147

Authors: Lopes da Silva T, Passarinho PC, Galriça R, Zenóglío A, Armshaw P, Pembroke JT, Sheahan C, Reis A, Gírio F

### **Abstract**

Flow cytometry was used to evaluate the effect of initial ethanol concentrations on cyanobacterial strains of *Synechocystis* PCC 6803 [wild-type (WT), and ethanol



producing recombinants (UL 004 and UL 030)] in batch cultures. Ethanol recombinants, containing one or two metabolically engineered cassettes, were designed towards the development of an economically competitive process for the direct production of bioethanol from microalgae through an exclusive autotrophic route. It can be concluded that the recombinant *Synechocystis* UL 030 containing two copies of the genes per genome was the most tolerant to ethanol. Nevertheless, to implement a production process using recombinant strains, the bioethanol produced will be required to be continuously extracted from the culture media via a membrane-based technological process for example to prevent detrimental effects on the biomass. The results presented here are of significance in defining the maximum threshold for bulk ethanol concentration in production media.

### **Marine algal carbohydrates as carbon sources for the production of biochemicals and biomaterials.**

Biotechnol Adv. 2018 May - Jun;36(3):798-817

Authors: Cesário MT, da Fonseca MMR, Marques MM, de Almeida MCMD

#### **Abstract**

The high content of lipids in microalgae (>60% w/w in some species) and of carbohydrates in seaweed (up to 75%) have promoted intensive research towards valorisation of algal components for the production of biofuels. However, the exploitation of the carbohydrate fraction to produce a range of chemicals and chemical intermediates with established markets is still limited. These include organic acids (e.g. succinic and lactic acid), alcohols other than bioethanol (e.g. butanol), and biomaterials (e.g. polyhydroxyalkanoates). This review highlights current and potential applications of the marine algal carbohydrate fractions as major C-source for microbial production of biomaterials and building blocks.

### **Pentoses and light intensity increase the growth and carbohydrate production and alter the protein profile of *Chlorella minutissima*.**

Bioresour Technol. 2017 Aug;238:248-253

Authors: Freitas BCB, Cassuriaga APA, Morais MG, Costa JAV

#### **Abstract**

High concentrations of carbon, which is considered a necessary element, are required for microalgal growth. Therefore, the identification of alternative carbon



sources available in large quantities is increasingly important. This study evaluated the effects of light variation and pentose addition on the carbohydrate content and protein profile of *Chlorella minutissima* grown in a raceway photobioreactor. The kinetic parameters, carbohydrate content, and protein profile of *Chlorella minutissima* and its theoretical potential for ethanol production were estimated. The highest cellular concentrations were obtained with a light intensity of  $33.75\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ . Arabinose addition combined with a light intensity of  $33.75\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  increased the carbohydrate content by 53.8% and theoretically produced  $39.1\text{mL}\cdot 100\text{g}^{-1}$  ethanol. All of the assays showed that a lower light availability altered the protein profile. The luminous intensity affects xylose and arabinose assimilation and augments the carbohydrate content in *C. minutissima*, making this microalga appropriate for bioethanol production.

### **Comparison of red microalgae (*Porphyridium cruentum*) culture conditions for bioethanol production.**

Bioresour Technol. 2017 Jun;233:44-50

Authors: Kim HM, Oh CH, Bae HJ

#### **Abstract**

Microalgae biomass are useful resources in biofuel production. The objective of this study was to evaluate bioethanol production in response to *Porphyridium cruentum* culture conditions. Enzymatic hydrolysis of seawater *P. cruentum* (SPC) and freshwater *P. cruentum* (FPC, 1% substrate loading, w/v) resulted in glucose conversion yields of 89.8 and 85.3%, respectively, without any pretreatment. However, FPC hydrolysate was more efficiently converted to ethanol about 7.1% than SPC hydrolysate. The comparison of separate hydrolysis and fermentation (SHF) and simultaneous saccharification and fermentation (SSF) showed that SSF processing is a superior method for bioethanol production from both SPC and FPC. Though SSF processing (5% substrate loading, w/v) in a 500-mL twin-neck round bottom flask, we achieved ethanol conversion yields of 65.4 and 70.3% from SPC and FPC, respectively, after 9h. These findings indicate that *P. cruentum* can grow in freshwater conditions and is an efficient candidate for bioethanol production.

### **Production and purification of amylolytic enzymes for saccharification of microalgal biomass.**

Bioresour Technol. 2017 Feb;225:134-141



Authors: Rodrigues ÉF, Ficanha AMM, Dallago RM, Treichel H, Reinehr CO, Machado TP, Nunes GB, Colla LM

#### Abstract

The aim of this study was the production of amylolytic enzymes by solid state or submerged fermentations (SSF or SF, respectively), followed by purification using chemical process or microfiltration and immobilization of purified enzymes in a polyurethane support. The free and immobilized enzymes obtained were used to evaluate enzymatic hydrolysis of the polysaccharides of *Spirulina*. Microfiltration of the crude extracts resulted in an increase in their specific activity and thermal stability at 40°C and 50°C for 24h, as compared to extracts obtained by SSF and SF. Immobilization of polyurethane purified enzyme produced yields of 332% and 205% for the enzymes obtained by SF and SSF, respectively. Free or immobilized enzymes favor the generation of fermentable sugar, being the application of the purified and immobilized enzymes in the hydrolysis of microalgal polysaccharides considered a promising alternative towards development of the bioethanol production process from microalgal biomass.

### **Comparative evaluation of chemical and enzymatic saccharification of mixotrophically grown de-oiled microalgal biomass for reducing sugar production.**

Bioresour Technol. 2016 Mar;204:9-16

Authors: Pancha I, Chokshi K, Maurya R, Bhattacharya S, Bachani P, Mishra S

#### Abstract

For the commercialization of microalgal based biofuels, utilization of de-oiled carbohydrate rich biomass is important. In the present study, chemo-enzymatic hydrolysis of mixotrophically grown *Scenedesmus* sp. CCNM 1077 de-oiled biomass is evaluated. Among the chemical hydrolysis, use of 0.5M HCl for 45 min at 121°C resulted in highest saccharification yield of 37.87% w/w of de-oiled biomass. However, enzymatic hydrolysis using Viscozyme L at loading rate of 20 FBGU/g of de-oiled biomass, pH 5.5 and temperature 45°C for 72 h resulted in saccharification yield of 43.44% w/w of de-oiled biomass. Further, 78% ethanol production efficiency was achieved with enzymatically hydrolyzed de-oiled biomass using yeast *Saccharomyces cerevisiae* ATCC 6793. These findings of the present study show application of mixotrophically grown de-oiled biomass of *Scenedesmus* sp. CCNM 1077 as promising feedstock for bioethanol production.



## **A state of the art of metabolic networks of unicellular microalgae and cyanobacteria for biofuel production.**

Metab Eng. 2015 Jul;30:49-60

Authors: Baroukh C, Muñoz-Tamayo R, Steyer JP, Bernard O

### **Abstract**

The most promising and yet challenging application of microalgae and cyanobacteria is the production of renewable energy: biodiesel from microalgae triacylglycerols and bioethanol from cyanobacteria carbohydrates. A thorough understanding of microalgal and cyanobacterial metabolism is necessary to master and optimize biofuel production yields. To this end, systems biology and metabolic modeling have proven to be very efficient tools if supported by an accurate knowledge of the metabolic network. However, unlike heterotrophic microorganisms that utilize the same substrate for energy and as carbon source, microalgae and cyanobacteria require light for energy and inorganic carbon (CO<sub>2</sub> or bicarbonate) as carbon source. This double specificity, together with the complex mechanisms of light capture, makes the representation of metabolic network nonstandard. Here, we review the existing metabolic networks of photoautotrophic microalgae and cyanobacteria. We highlight how these networks have been useful for gaining insight on photoautotrophic metabolism.

## **Biological potential of microalgae in China for biorefinery-based production of biofuels and high value compounds.**

N Biotechnol. 2015 Dec 25;32(6):588-96

Authors: Li J, Liu Y, Cheng JJ, Mos M, Daroch M

### **Abstract**

Microalgae abundance and diversity in China shows promise for identifying suitable strains for developing algal biorefinery. Numerous strains of microalgae have already been assessed as feedstocks for bioethanol and biodiesel production, but commercial scale algal biofuel production is yet to be demonstrated, most likely due to huge energy costs associated with algae cultivation, harvesting and processing. Biorefining, integrated processes for the conversion of biomass into a variety of products, can improve the prospects of microalgal biofuels by combining them with the production of high value co-products. Numerous microalgal strains in China have been identified as producers of various high value by-products with wide application in the medicine, food, and cosmetics industries. This paper reviews microalgae resources in China and their potential in producing liquid biofuels (bioethanol and



biodiesel) and high value products in an integrated biorefinery approach. Implementation of a 'high value product first' principle should make the integrated process of fuels and chemicals production economically feasible and will ensure that public and private interest in the development of microalgal biotechnology is maintained.

### **Vortex fluidic mediated direct transesterification of wet microalgae biomass to biodiesel.**

Bioresour Technol. 2018 Jul 05;266:488-497

Authors: Sitepu EK, Corbin K, Luo X, Pye SJ, Tang Y, Leterme SC, Heimann K, Raston CL, Zhang W

#### **Abstract**

A bottleneck in the production of biodiesel from microalgae is the dewatering and lipid extraction process which is the dominant energy penalty and cost. A novel biodiesel production platform based on vortex fluidic device (VFD)-assisted direct transesterification (DT) of wet microalgal biomass of *Chloroparva pannonica* was developed and evaluated. Fatty acid extraction and fatty acid to FAME conversion efficiencies were used at different parameter settings to evaluate performance of the processing technology in confined and continuous mode. A response surface method based on Box-Behnken experimental design was used to determine the effects of water content, the ratio of biomass to methanol and residence time in the VFD. Average extraction efficiencies were 41% and conversion efficiencies >90% with the processing technology showing a broad tolerance to parameter settings. The findings suggest that VFD-assisted DT is a simple and effective way to produce biodiesel directly from wet microalgal biomass at room temperature.

### **Effects of Bacteria on the Growth of and Lipid Accumulation in *Chlorella pyrenoidosa* Cultivated in Municipal Wastewater.**

Huan Jing Ke Xue. 2017 Oct 08;38(10):4279-4285

Authors: Tu RJ, Jin WB, Han SF, Chen HY

#### **Abstract**

Cultivating microalgae using municipal wastewater can treat wastewater and recover algal biofuel as an energy source. Wastewater provides necessary nutrients such as nitrogen, phosphorus, and water for microalgal growth. Due to the



complexity of the components of municipal wastewater and the complex symbiotic and antagonistic relationship between microalgae and bacteria, it is necessary to select the suitable dominant bacterial species that can promote the microalgae to achieve high lipid production and algal biofuel production using municipal wastewater. Based on the microalgal growth and lipid production, we selected Photosynthetic bacteria and W4 bacteria from 13 different types of bacteria and analyzed the microbial community structure of the municipal wastewater at the end of the microalgal culture cycle. Laboratory test results showed that the amount of lipid production by Photosynthetic bacteria and W4 was 0.114 g·L<sup>-1</sup> and 0.113 g·L<sup>-1</sup>, which is 22.58% and 21.50% higher than the production by the control group, respectively. According to the gas chromatography (GC) analysis of the lipids, Photosynthetic bacteria and W4 bacteria exerted a relatively low influence on the composition of fatty acids of *Chlorella pyrenoidosa* but increased the content of monounsaturated fatty acids that improve the grade of biodiesel. The results of the analysis of microbial community structure of the municipal wastewater showed that Photosynthetic and W4 bacteria reduced the richness and diversity of bacterial communities and have the potential to become the dominant bacterial community.

### **Effects of temperature and its combination with high light intensity on lipid production of *Monoraphidium dybowskii* Y2 from semi-arid desert areas.**

Bioresour Technol. 2018 Jun 15;265:407-414

Authors: He Q, Yang H, Hu C

#### **Abstract**

Temperature and light intensity are important environmental factors influencing microalgae for biodiesel production. The aim of present work was to study the effects of temperature (15 °C, 25 °C, and 35 °C) and its combination with high light intensity (HL, 400 μmol photon m<sup>-2</sup> s<sup>-1</sup>) on lipid production of *Monoraphidium dybowskii* Y2 which was isolated from desert. The results demonstrated that algal growth was only inhibited at 15 °C. Promoted lipid content and decreased Fv/Fm were observed in 15 °C and 35 °C. Cellular carbohydrate, protein conversion and membrane lipid (MGDG, DGDG and SQDG) remodeling contributes for lipid accumulation. Stress combined temperatures with HL are benefit for lipid production, especially desired neutral lipid productivity all exceed 40 mg L<sup>-1</sup> d<sup>-1</sup>. Fatty acids compositions of C16:0 and C18:1 were further promoted under 15 °C or 35 °C combined with HL. Thus, *M. dybowskii* Y2 will well-adapted to outdoors cultivation for biodiesel production.



## **<sup>13</sup>C Metabolic Flux Analysis of Enhanced Lipid Accumulation Modulated by Ethanolamine in *Cryptocodinium cohnii*.**

Front Microbiol. 2018;9:956

Authors: Cui J, Diao J, Sun T, Shi M, Liu L, Wang F, Chen L, Zhang W

### **Abstract**

The heterotrophic microalga *Cryptocodinium cohnii* has attracted considerable attention due to its capability of accumulating lipids with a high fraction of docosahexaenoic acid (DHA). In our previous study, ethanolamine (ETA) was identified as an effective chemical modulator for lipid accumulation in *C. cohnii*. In this study, to gain a better understanding of the lipid metabolism and mechanism for the positive effects of modulator ETA, metabolic flux analysis was performed using <sup>13</sup>C-labeled glucose with and without 1 mM ETA modulator. The analysis of flux distribution showed that with the addition of ETA, flux in glycolysis pathway and citrate pyruvate cycle was strengthened while flux in pentose phosphate pathway was decreased. In addition, flux in TCA cycle was slightly decreased compared with the control without ETA. The enzyme activity of malic enzyme (ME) was significantly increased, suggesting that NADP<sup>+</sup>-dependent ME might be the major source of NADPH for lipid accumulation. The flux information obtained by this study could be valuable for the further efforts in improving lipid accumulation and DHA production in *C. cohnii*.

## **Maximizing CO<sub>2</sub> biofixation and lipid productivity of oleaginous microalga *Graesiella* sp. WBG-1 via CO<sub>2</sub>-regulated pH in indoor and outdoor open reactors.**

Sci Total Environ. 2018 Apr 01;619-620:827-833

Authors: Wang Z, Wen X, Xu Y, Ding Y, Geng Y, Li Y

### **Abstract**

Carbon dioxide (CO<sub>2</sub>) and pH are two interdependent factors that greatly impact the growth and lipid accumulation of microalgae. However, the effects of these two factors are usually studied separately. The use of exogenous CO<sub>2</sub>, such as flue gas derived, to regulate pH in the large-scale cultivation of microalgae provides an ideal means for combining CO<sub>2</sub> biofixation and biodiesel production. In this study, the CO<sub>2</sub> biofixation and lipid production of oleaginous microalga *Graesiella* sp. WBG-1



was explored for four pH levels regulated by exogenous 15% CO<sub>2</sub> (flue gas concentration) in 10L circular culture ponds and 5m<sup>2</sup> open raceway reactors. Results revealed that pH8.0-9.0 was the optimum pH for CO<sub>2</sub> fixation and lipid production, attaining the highest CO<sub>2</sub> fixation rates of 0.26gL<sup>-1</sup>day<sup>-1</sup> and 18.9gm<sup>-2</sup>day<sup>-1</sup>, respectively, lipid contents of 46.28% and 32.38%, and lipid productivities of 64.8mgL<sup>-1</sup>day<sup>-1</sup> and 3.14gm<sup>-2</sup>day<sup>-1</sup>. A positive correlation between CO<sub>2</sub> utilization efficiency and pH in open reactors was also suggested in this research, and thus provides direction for screening of CO<sub>2</sub> fixation by microalgae. The present study provides an excellent strategy for coupling CO<sub>2</sub> fixation and lipid production via microalgae in large-scale cultivation.

### **A *Leptolyngbya*-based microbial consortium for agro-industrial wastewaters treatment and biodiesel production.**

Environ Sci Pollut Res Int. 2018 Jun;25(18):17957-17966

Authors: Tsolcha ON, Tekerlekopoulou AG, Akrotos CS, Antonopoulou G, Aggelis G, Genitsaris S, Moustaka-Gouni M, Vayenas DV

#### **Abstract**

A mixed cyanobacterial-mixotrophic algal population, dominated by the filamentous cyanobacterium *Leptolyngbya* sp. and the microalga *Ochromonas* (which contributed to the total photosynthetic population with rates of less than 5%), was studied under non-aseptic conditions for its efficiency to remove organic and inorganic compounds from different types of wastes/wastewaters while simultaneously producing lipids. Second cheese whey, poplar sawdust, and grass hydrolysates were used in lab-scale experiments, in photobioreactors that operated under aerobic conditions with different initial nutrient (C, N and P) concentrations. Nutrient removal rates, biomass productivity, and the maximum oil production rates were determined. The highest lipid production was achieved using the biologically treated dairy effluent (up to 14.8% oil in dry biomass corresponding to 124 mg L<sup>-1</sup>) which also led to high nutrient removal rates (up to 94%). Lipids synthesized by the microbial consortium contained high percentages of saturated and mono-unsaturated fatty acids (up to 75% in total lipids) for all the substrates tested, which implies that the produced biomass may be harnessed as a source of biodiesel.



## **Extraction of astaxanthin from microalga *Haematococcus pluvialis* in red phase by using Generally Recognized As Safe solvents and accelerated extraction.**

J Biotechnol. 2018 Jul 09

Authors: Molino A, Rimauro J, Casella P, Cerbone A, Larocca V, Chianese S, Karatza D, Mehariya S, Ferraro A, Hristoforou E, Musmarra D

### **Abstract**

Solvent Extraction was tested to extract astaxanthin from *Haematococcus pluvialis* in red phase (HPR), by investigating effects of solvents, extraction pressure and temperature. Astaxanthin isomers were identified and quantified in the extract. The performances of acetone and ethanol, Generally Recognized As Safe (GRAS) solvents, were explored. Negligible effect of pressure was found, while with increasing extraction temperature astaxanthin recovery increased till a maximum value, beyond which thermal degradation seemed to be greater than the positive effect of temperature on extraction. Furthermore, to maximize the extraction yield of astaxanthin, mechanical pre-treatment of HPR biomass was carried out and several extraction runs were consecutively performed. Experimental results showed that after the mechanical pre-treatment the astaxanthin recovery strongly increased while a single extraction run of 20 minutes was sufficient to extract more than 99% of total astaxanthin extracted. After pre-treatment, maximum recovery of about 87% was found for acetone (pressure = 100 bar; temperature = 40 °C; total time = 60 minutes).

## **Induced High-Yield Production of Zeaxanthin, Lutein, and $\beta$ -Carotene by a Mutant of *Chlorella zofingiensis*.**

J Agric Food Chem. 2018 Jan 31;66(4):891-897

Authors: Huang W, Lin Y, He M, Gong Y, Huang J

### **Abstract**

Natural resources of zeaxanthin are extremely limited. A *Chlorella zofingiensis* mutant (CZ-bkt1), which could accumulate high amounts of zeaxanthin, was generated and characterized. CZ-bkt1 was achieved by treating the algal cells with a chemical mutagen followed by a color-based colony-screening approach. CZ-bkt1 was found to consist of a dysfunctional carotenoid ketolase, leading to the accumulation of zeaxanthin rather than to its downstream ketocarotenoid astaxanthin. Light irradiation, glucose, NaCl, and nitrogen deficiency all induced CZ-



bkt1 to accumulate zeaxanthin. CZ-bkt1 accumulated zeaxanthin up to  $7.00 \pm 0.82$  mg/g when induced by high-light irradiation and nitrogen deficiency and up to  $36.79 \pm 2.23$  mg/L by additional feeding with glucose. Furthermore, in addition to zeaxanthin, CZ-bkt1 also accumulated high amounts of  $\beta$ -carotene ( $7.18 \pm 0.72$  mg/g or  $34.64 \pm 1.39$  mg/L) and lutein ( $13.81 \pm 1.23$  mg/g or  $33.97 \pm 2.61$  mg/L). CZ-bkt1 is the sole species up to date with the ability to accumulate high amounts of the three carotenoids that are essential for human health.

### **An evidence of C16 fatty acid methyl esters extracted from microalga for effective antimicrobial and antioxidant property.**

Microb Pathog. 2018 Feb;115:233-238

Authors: Davoodbasha M, Edachery B, Nooruddin T, Lee SY, Kim JW

#### **Abstract**

Fatty acid methyl esters (FAME) derived from lipids of microalgae is known to have wide bio-functional materials including antimicrobials. FAME is an ideal super-curator and superior anti-pathogenic. The present study evaluated the efficiency of FAME extracted from microalgae *Scenedesmus intermedius* as an antimicrobial agent against Gram positive (*Staphylococcus aureus*, *Streptococcus mutans*, and *Bacillus cereus*) Gram negative (*Escherichia coli* and *Pseudomonas aeruginosa*) bacteria and Fungi (*Aspergillus parasiticus* and *Candida albicans*). The minimal inhibitory concentration (MIC) for the gram negative bacteria was determined as 12-24  $\mu\text{g mL}^{-1}$ , whereas MIC for gram positive bacteria was 24-48  $\mu\text{g mL}^{-1}$ . MIC for the fungi was as high as 60-192  $\mu\text{g mL}^{-1}$ . The FAME profiles determined by gas chromatography showed 18 methyl esters. Among them, pharmacologically active FAME such as palmitic acid methyl ester (C16:0) was detected at high percentage (23.08%), which accounted for the bioactivity. FAME obtained in this study exhibited a strong antimicrobial activity at the lowest MIC than those of recent reports. This result clearly indicated that FAME of *S. intermedius* has a strong antimicrobial and antioxidant property and that could be used as an effective resource against microbial diseases.



## An anti-inflammatory effect of red microalga polysaccharides in coronary artery endothelial cells.

Atherosclerosis. 2017 Sep;264:11-18

Authors: Levy-Ontman O, Huleihel M, Hamias R, Wolak T, Paran E

### Abstract

**BACKGROUND AND AIMS:** Polysaccharides (PSs) produced by the red microalga *Porphyridium* sp. were reported to exhibit anti-inflammatory bioactivities in the human skin. The primary goal of the present research was to assess whether PSs attenuate inflammatory processes by interfering with tumour necrosis factor- $\alpha$  (TNF- $\alpha$ )-induced inflammation, in human coronary artery endothelial cells (HCAECs).

**METHODS:** Functional and inflammatory markers were quantified in TNF- $\alpha$ -stimulated HCAECs, with and without pre-treatment with PSs. The expression/activation of these markers was assessed by Western immunoblotting and a luciferase reporter assay. NO levels were measured using the Griess method and intracellular reactive oxygen stress (ROS) was determined with the fluorescent probe 2',7'-dichlorodihydro-fluorescein diacetate (H2DCFDA).

**RESULTS:** The TNF- $\alpha$ -induced up-regulation of inter-cellular adhesion molecule 1 (ICAM-1) and vascular cell adhesion molecule 1 (VCAM-1), nuclear factor kappa-light-chain-enhancer of activated B cells (NF- $\kappa$ B) translocation, as well as I $\kappa$ B degradation were significantly attenuated in cells pre-treated with PSs. In addition, PSs were able to inhibit NF- $\kappa$ B activation as well as TNF- $\alpha$ -induced oxidative stress in HCAECs. Endothelial function was also improved, as measured by increased nitric oxide (NO) formation and decreased endothelin (ET-1) protein expression.

**CONCLUSIONS:** This is the first report that demonstrates the anti-inflammatory effect and vaso-relaxing property of red microalgae PSs in a HCAEC-TNF- $\alpha$  induced system. This study lays the foundation for basic research concerning the PS mode of action in biochemical processes involving endothelial dysfunction, and it also holds potential for applied research, possibly promoting the use of PSs as a therapeutic agent or food additive to improve vascular health.



## **Growth stimulation and synthesis of lipids, pigments and antioxidants with magnetic fields in *Chlorella kessleri* cultivations.**

Bioresour Technol. 2017 Nov;244(Pt 2):1425-1432

Authors: Bauer LM, Costa JAV, da Rosa APC, Santos LO

### **Abstract**

This study aimed at applying different intensities (30 and 60mT) and exposure times (24 and 1hd-1) of MF to cultures of *Chlorella kessleri* and evaluated their effects on cell growth, proximate composition of biomass, pigment production and antioxidant activity. The condition of 60mT for 1hd-1 stimulated biomass concentration of 83.2% by comparison with control culture (CC). Besides stimulated 13.7% lipid synthesis, 38.9% chlorophyll a and 59.1% chlorophyll b, 25.0% total carotenoids and antioxidants up to 185.7%. Thus MF application is an excellent alternative to stimulate cell growth and high biotechnological interest biocompounds.

## **In vitro evaluation of *Spirulina platensis* extract incorporated skin cream with its wound healing and antioxidant activities.**

Pharm Biol. 2017 Dec;55(1):1824-1832

Authors: Gunes S, Tamburaci S, Dalay MC, Deliloglu Gurhan I

### **Abstract**

**CONTEXT:** Algae have gained importance in cosmeceutical product development due to their beneficial effects on skin health and therapeutical value with bioactive compounds. *Spirulina platensis* Parachas (Phormidiaceae) is renowned as a potential source of high-value chemicals and recently used in skincare products.

**OBJECTIVE:** This study develops and evaluates skin creams incorporated with bioactive *S. platensis* extract.

**MATERIALS AND METHODS:** *Spirulina platensis* was cultivated, the aqueous crude extract was prepared and in vitro cytotoxicity of *S. platensis* extract in the range of 0.001-1% concentrations for 1, 3 and 7 d on HS2 keratinocyte cells was determined. Crude extracts were incorporated in skin cream formulation at 0.01% (w/w) concentration and in vitro wound healing and genotoxicity studies were performed. Immunohistochemical staining was performed to determine the collagen activity.

**RESULTS:** 0.1% *S. platensis* extract exhibited higher proliferation activity compared with the control group with 198% of cell viability after 3 d. Skin cream including 1.125% *S. platensis* crude extract showed enhanced wound healing effect on HS2 keratinocyte cell line and the highest HS2 cell viability % was obtained with this concentration. The micronucleus (MN) assay results indicated that *S. platensis*



extract incorporated creams had no genotoxic effect on human peripheral blood cells. Immunohistochemical analysis showed that collagen 1 immunoreactivity was improved by increased extract concentration and it was strongly positive in cells treated with 1.125% extract incorporated skin cream.

CONCLUSIONS: The cell viability, wound healing activity and genotoxicity results showed that *S. platensis* incorporated skin cream could be of potential value in cosmeceutical and biomedical applications.

### **Inhibitory effect of ethanol extract of *Nannochloropsis oceanica* on lipopolysaccharide-induced neuroinflammation, oxidative stress, amyloidogenesis and memory impairment.**

Oncotarget. 2017 Jul 11;8(28):45517-45530

Authors: Choi JY, Hwang CJ, Lee HP, Kim HS, Han SB, Hong JT

#### Abstract

Oxidative stress and neuroinflammation is implicated in the pathogenesis and development of Alzheimer's disease (AD). Here, we investigated the suppressive possibility of ethanol extract of *Nannochloropsis oceanica* (*N. oceanica*) on memory deficiency along with the fundamental mechanisms in lipopolysaccharide (LPS)-treated mice model. Among several extracts of 32 marine microalgae, ethanol extract of *N. oceanica* showed the most significant inhibitory effect on nitric oxide (NO) generation, NF- $\kappa$ B activity and  $\beta$ -secretase activity in cultured BV-2 cells, neuronal cells and Raw 264.7 cells. Ethanol extract of *N. oceanica* (50, 100 mg/kg) also ameliorated LPS (250  $\mu$ g/kg)-induced memory impairment. We also found that ethanol extract of *N. oceanica* inhibited the LPS-induced expression of iNOS and COX-2. Furthermore, the production of reactive oxygen species (ROS), malondialdehyde (MDA) level as well as glutathione (GSH) level was also decreased by treatment of ethanol extract of *N. oceanica*. The ethanol extract of *N. oceanica* also suppresses I $\kappa$ B degradation as well as p50 and p65 translocation into the nucleus in LPS-treated mice brain. Associated with the inhibitory effect on neuroinflammation and oxidative stress, ethanol extract of *N. oceanica* suppressed A $\beta$ 1-42 generation through down-regulation of APP and BACE1 expression in vivo. These results suggest that ethanol extract of *N. oceanica* ameliorated memory impairment via anti-inflammatory, anti-oxidant and anti-amyloidogenic mechanisms.



## **Effects of butanol on high value product production in *Schizochytrium limacinum* B4D1.**

Enzyme Microb Technol. 2017 Jul;102:9-15

Authors: Zhang K, Chen L, Liu J, Gao F, He R, Chen W, Guo W, Chen S, Li D

### **Abstract**

Schizochytrium is a microalgae-like fungus and is widely used for producing docosahexaenoic acid (DHA). It is also a promising source of squalene and carotenoids. However, few fermentation strategies are available in enhancing squalene and carotenoid content in Schizochytrium. This study showed that butanol addition had multiple effects on Schizochytrium limacinum B4D1. First, butanol addition altered the lipid content of cells. Second, 6g/L of butanol decreased the proportion of DHA by nearly 40%. Third, the squalene content increased 31-fold in the presence of 6g/L butanol. Finally, cells accumulated more carotenoids upon butanol addition. Specifically, when cells were treated with 8g/L butanol, the astaxanthin content increased to 245 times than that of the untreated control. These results are helpful for the commercial exploitation of Schizochytrium in producing squalene and carotenoids.

## **A heptameric peptide purified from *Spirulina* sp. gastrointestinal hydrolysate inhibits angiotensin I-converting enzyme- and angiotensin II-induced vascular dysfunction in human endothelial cells.**

Int J Mol Med. 2017 May;39(5):1072-1082

Authors: Heo SY, Ko SC, Kim CS, Oh GW, Ryu B, Qian ZJ, Kim G, Park WS, Choi IW, Phan TT, Heo SJ, Kang DH, Yi M, Jung WK

### **Abstract**

In this study, a marine microalga *Spirulina* sp.-derived protein was hydrolyzed using gastrointestinal enzymes to produce an angiotensin I (Ang I)-converting enzyme (ACE) inhibitory peptide. Following consecutive purification, the potent ACE inhibitory peptide was composed of 7 amino acids, Thr-Met-Glu-Pro-Gly-Lys-Pro (molecular weight, 759 Da). Analysis using the Lineweaver-Burk plot and molecular modeling suggested that the purified peptide acted as a mixed non-competitive inhibitor of ACE. The inhibitory effects of the peptide against the cellular production of vascular dysfunction-related factors induced by Ang II were also investigated. In human endothelial cells, the Ang II-induced production of nitric oxide and reactive oxygen species was inhibited, and the expression of inducible nitric oxide synthase



(iNOS) and endothelin-1 (ET-1) was downregulated when the cells were cultured with the purified peptide. Moreover, the peptide blocked the activation of p38 mitogen-activated protein kinase. These results indicated that this *Spirulina* sp.-derived peptide warrants further investigation as a potential pharmacological inhibitor of ACE and vascular dysfunction.

### **Community analysis of pigment patterns from 37 microalgae strains reveals new carotenoids and porphyrins characteristic of distinct strains and taxonomic groups.**

PLoS One. 2017;12(2):e0171872

Authors: Serive B, Nicolau E, Bérard JB, Kaas R, Pasquet V, Picot L, Cadoret JP

#### **Abstract**

Phytoplankton, with an estimated 30 000 to 1 000 000 species clustered in 12 phyla, presents a high taxonomic and ecophysiological diversity, reflected by the complex distribution of pigments among the different algal classes. High performance liquid chromatography is the gold standard method for qualitative and quantitative analysis of phytoplankton pigments in seawater and culture samples, but only a few pigments can be used as robust chemotaxonomic markers. A major challenge is thus to identify new ones, characteristic of a strain, species, class or taxon that cannot be currently identified on the basis of its pigment signature. Using an optimized extraction process coupled to a HPLC de-replication strategy, we examined the pigment composition of 37 microalgae strains, representative of the broad taxonomic diversity of marine and freshwater species (excluding cyanobacteria). For each species, the major pigments already described were unambiguously identified. We also observed the presence of several minor unidentified pigments in each chromatogram. The global analysis of pigment compositions revealed a total of 124 pigments, including 98 pigments or derivatives unidentified using the standards. Absorption spectra indicated that 35 corresponded to chlorophyll/porphyrin derivatives, 57 to carotenoids and six to derivatives having both spectral signatures. Sixty-one of these unidentified or new carotenoids and porphyrin derivatives were characteristic of particular strains or species, indicating their possible use as highly specific chemotaxonomic markers capable of identifying one strain out of the 37 selected. We developed a graphical analysis using Gephi software to give a clear representation of pigment communities among the various phytoplankton strains, and to reveal strain-characteristic and shared pigments. This made it possible to reconstruct the taxonomic evolution of microalgae classes, on the basis of the conservation, loss, and/or appearance of pigments.



## **Microalgae cultivation in urban wastewater: *Coelastrum* cf. *pseudomicroporum* as a novel carotenoid source and a potential microalgae harvesting tool.**

Bioresour Technol. 2017 Mar;228:210-217

Authors: Úbeda B, Gálvez JÁ, Michel M, Bartual A

### **Abstract**

The aim of this work was to study the optimal growth and high value-added production of the microalgae *Coelastrum* cf. *pseudomicroporum* Korshikov cultivated in urban wastewater. It was observed that *C. cf. pseudomicroporum* grew ideally in this medium, acting as an efficient nutrient starver. Additionally, the obtained biomass increased carotenoid cell content after saltwater stress. The effects of light intensity and salt stress on its growth rate were analysed. The results showed that this alga can grow very fast using wastewater as culture medium, reaching maximum growth rates of  $1.61 \pm 0.05 \text{ day}^{-1}$ , and tolerating strong irradiances. It was also found that under salt-stress this species could accumulate carotenoids (range  $1.73\text{-}91.2 \text{ pg cell}^{-1}$ ). Moreover, a good harvesting efficiency (96.84%) was observed using *Coelastrum* exudates as bioflocculant of *Scenedesmus* sp., so *Coelastrum* exudates could act as a potential bioflocculant for other species.

## **Microstructure and antioxidative capacity of the microalgae mutant *Chlorella* PY-ZU1 during tilmicosin removal from wastewater under 15% CO<sub>2</sub>.**

J Hazard Mater. 2017 Feb 15;324(Pt B):414-419

Authors: Cheng J, Ye Q, Yang Z, Yang W, Zhou J, Cen K

### **Abstract**

The response mechanisms of microalgal mutant *Chlorella* PY-ZU1 cells were investigated in their removal of antibiotic tilmicosin from wastewater under 15% CO<sub>2</sub>. Low concentrations ( $0.01\text{-}2 \text{ mg L}^{-1}$ ) of tilmicosin in wastewater stimulated the growth of microalgal cells, whereas high concentrations ( $5\text{-}50 \text{ mg L}^{-1}$ ) of tilmicosin significantly inhibited cell growth. When initial tilmicosin concentration increased from 0 to  $50 \text{ mg L}^{-1}$ , fractal dimension of microalgal cells monotonically increased from 1.36 to 1.62 and cell size monotonically decreased from  $4.86$  to  $3.75 \mu\text{m}$ . In parallel, malondialdehyde content, which represented the degree of cellular oxidative damage, monotonically increased from  $1.92 \times 10^{-7}$  to  $7.07 \times 10^{-7} \text{ nmol cell}^{-1}$ . Superoxide dismutase activity that represented cellular antioxidant capacity first



increased from  $2.59 \times 10^{-4}$  to the peak of  $6.60 \times 10^{-4}$  U cell<sup>-1</sup>, then gradually decreased to  $2.39 \times 10^{-4}$  U cell<sup>-1</sup>. The maximum tilmicosin removal efficiency of 99.8% by *Chlorella* PY-ZU1 was obtained at the initial tilmicosin concentration of 50 mg L<sup>-1</sup>.

## Enhancing Carbohydrate Productivity of *Chlorella* sp. AE10 in Semi-continuous Cultivation and Unraveling the Mechanism by Flow Cytometry.

Appl Biochem Biotechnol. 2018 Jun;185(2):419-433

Authors: Yuan Y, Liu H, Li X, Qi W, Cheng D, Tang T, Zhao Q, Wei W, Sun Y

### Abstract

Accumulated carbohydrate in microalgae is promising feedstock for bioethanol fermentation. Selection of suitable cultivation conditions in semi-continuous cultivation is critical to achieve a high carbohydrate productivity. In the current study, the effects of macro-nutrient (nitrogen, phosphorus, and sulfur) limitations and light intensity were evaluated for the carbohydrate accumulations of *Chlorella* sp. AE10 under 10% CO<sub>2</sub> conditions. It was shown that nitrogen limitation and high light intensity were effective for improving carbohydrate productivity. The average carbohydrate and biomass productivity in semi-continuous cultivation with 1/4 N medium and 1000 μmol photons m<sup>-2</sup> s<sup>-1</sup> was 0.673 and 0.93 g L<sup>-1</sup> day<sup>-1</sup>, respectively. Sulfur and phosphorus limitations could improve the carbohydrate content but they could not enhance the carbohydrate productivity. The cell cycle progression and chlorophyll a were investigated using flow cytometry (FCM). The results showed that macro-nutrient limitation and high light intensity indeed influenced cell cycle progression and led to the formation of polyploid cells along with the carbohydrate accumulation in a certain range. FCM was rapid and accurate method to investigate the operation conditions why 1/4 N, 2 days as a cycle, and high light intensity were optimal ones. In addition, the remaining high level of photosynthesis activity was also important for achieving a high carbohydrate productivity. Dynamic tracking of carbohydrate accumulation is helpful for establishment of a semi-continuous cultivation for enhancing carbohydrate productivity in microalgae.



## **Efficient conversion of mannitol derived from brown seaweed to fructose for fermentation with a thraustochytrid.**

J Biosci Bioeng. 2018 Feb;125(2):180-184

Authors: Tajima T, Tomita K, Miyahara H, Watanabe K, Aki T, Okamura Y, Matsumura Y, Nakashimada Y, Kato J

### **Abstract**

Macroalgae are a promising biomass feedstock for energy and valuable chemicals. Mannitol and alginate are the major carbohydrates found in the microalga *Laminaria japonica* (Konbu). To convert mannitol to fructose for its utilization as a carbon source in mannitol non-assimilating bacteria, a psychrophile-based simple biocatalyst (PSCat) was constructed using a psychrophile as a host by expressing mesophilic enzymes, including mannitol 2-dehydrogenase for mannitol oxidation, and NADH oxidase and alkyl hydroperoxide reductase for NAD<sup>+</sup> regeneration. PSCat was treated at 40 °C to inactivate the psychrophilic enzymes responsible for byproduct formation and to increase the membrane permeability of the substrate. PSCat efficiently converted mannitol to fructose with high conversion yield without additional input of NAD<sup>+</sup>. Konbu extract containing mannitol was converted to fructose with hydroperoxide scavenging, inhibiting the mannitol dehydrogenase activity. *Aurantiocytrium* sp. could grow well in the presence of fructose converted by PSCat. Thus, PSCat is a potential carbohydrate converter for mannitol non-assimilating microorganism.

## **Evaluation of colour temperatures in the cultivation of *Dunaliella salina* and *Nannochloropsis oculata* in the production of lipids and carbohydrates.**

Environ Sci Pollut Res Int. 2017 Jul 25;:

Authors: Pavón-Suriano SG, Ortega-Clemente LA, Curiel-Ramírez S, Jiménez-García MI, Pérez-Legaspi IA, Robledo-Narváez PN

### **Abstract**

The production of biofuels from microalgae is a promising and sustainable alternative. Its production is determined by the content of lipids and carbohydrates, which is different for each microalgae species and is affected by environmental factors, being lighting one of the principal determining their biochemical composition. The colour temperature (electromagnetic radiation and light spectrum) is a determining factor for the production of lipids and carbohydrates in microalgae.



The aim of this assay was to evaluate the effect of three colour temperatures (6500, 10,000 and 20,000 °K) on the biomass (cel mL<sup>-1</sup>), biomass production and productivity (g L<sup>-1</sup> and g L<sup>-1</sup> day<sup>-1</sup>), lipid and carbohydrate content (%), lipid and carbohydrate production and productivity (mg L<sup>-1</sup> and mg L<sup>-1</sup> day<sup>-1</sup>), composition and content of fatty acids (%) in two microalgae species: *Dunaliella salina* and *Nannochloropsis oculata*. The highest cell density was observed for *N. oculata* in stationary phase in the control ( $83.93 \times 10^6$  cel mL<sup>-1</sup>). However, higher lipid content was obtained in *D. salina* in stationary phase at 10,000 °K (80%), while *N. oculata* showed 67% at 6500 °K. The highest carbohydrate content was 25% in stationary phase for *D. salina* at 20,000 °K. Regarding the production of lipids, *D. salina* reached a maximum of 523 mg L<sup>-1</sup> in exponential phase at 6500 and 10,000 °K. The highest carbohydrate production was 38 mg L<sup>-1</sup> for *D. salina* in exponential phase at 20,000 °K. In both microalgae, 15 different fatty acids were identified; the most abundant was palmitic acid with 35.8% for *N. oculata* in stationary phase at 10,000 °K, while *D. salina* showed 67% of polyunsaturated fatty acids in exponential phase at 6500 °K. In conclusion, the ideal colour temperature for microalgae culture to obtain biofuels should be based on the biomolecule of interest, being necessary to individually evaluate for each species.

### **Current advances on fermentative biobutanol production using third generation feedstock.**

Biotechnol Adv. 2017 Dec;35(8):1049-1059

Authors: Wang Y, Ho SH, Yen HW, Nagarajan D, Ren NQ, Li S, Hu Z, Lee DJ, Kondo A, Chang JS

#### **Abstract**

Biobutanol is gaining more attention as a potential alternative to ethanol, and the demand for fermentative biobutanol production has renewed interest. The main challenge faced in biobutanol production is the availability of feedstock. Using conventional agricultural biomass as feedstock is controversial and less efficient, while microalgae, the third generation feedstock, are considered promising feedstock for biobutanol production due to their high growth rate and high carbohydrates content. This review is primarily focused on biobutanol production by using carbohydrate-rich microalgal feedstock. Key technologies and challenges involved in producing butanol from microalgae are discussed in detail and future directions are also presented.



## **Cultivation of the Marine Macroalgae *Chaetomorpha linum* in Municipal Wastewater for Nutrient Recovery and Biomass Production.**

Environ Sci Technol. 2017 Mar 21;51(6):3558-3566

Authors: Ge S, Champagne P

### **Abstract**

Compared to microalgae, macroalgae are larger in size, thereby imposing lower separation and drying costs. This study demonstrates the feasibility of cultivating macroalgae *Chaetomorpha linum* in different types of municipal wastewaters, their ability to remove nutrient and their biomass composition for downstream biofuel production. Screening experiments indicated that *C. linum* grew well on primary (PW) and secondary wastewaters (SW), as well as centrate wastewater (CW) diluted to less than 20%. In a subsequent experiment, a step feeding approach was found to significantly increase biomass productivity to  $10.7 \pm 0.2$  g AFDW·m<sup>-2</sup>·d<sup>-1</sup> ( $p < 0.001$ ), a 26.5% improvement in comparison to the control with single feeding, when grown on 10-CW; meanwhile, nitrogen and phosphorus removal efficiencies rose to  $86.8 \pm 1.1\%$  ( $p < 0.001$ ) and  $92.6 \pm 0.2\%$  ( $p < 0.001$ ), respectively. The CO<sub>2</sub>-supplemented SW cultures ( $10.1 \pm 0.4$  g AFDW·m<sup>-2</sup>·d<sup>-1</sup>) were 1.20 times more productive than the corresponding controls without CO<sub>2</sub> supplementation ( $p < 0.001$ ); however, similar improvements were not observed in PW ( $p = 0.07$ ) and 10-CW cultures ( $p = 0.07$ ). Moreover, wastewater type and nutrient concentration influenced biomass composition (protein, carbohydrate and lipid). These findings indicate that the application of the macroalgae *C. linum* could represent an effective wastewater treatment alternative that could also provide a feedstock for downstream processing to biofuels.

## **Feasibility of CO<sub>2</sub> mitigation and carbohydrate production by microalga *Scenedesmus obliquus* CNW-N used for bioethanol fermentation under outdoor conditions: effects of seasonal changes.**

Biotechnol Biofuels. 2017;10:27

Authors: Ho SH, Chen YD, Chang CY, Lai YY, Chen CY, Kondo A, Ren NQ, Chang JS

### **Abstract**

**BACKGROUND:** Although outdoor cultivation systems have been widely used for mass production of microalgae at a relatively low cost, there are still limited efforts on outdoor cultivation of carbohydrate-rich microalgae that were further used as feedstock for fermentative bioethanol production. In particular, the effects of



seasonal changes on cell growth, CO<sub>2</sub> fixation, and carbohydrate production of the microalgae have not been well investigated.

**RESULTS:** This work demonstrates the feasibility of using outdoor tubular photobioreactors (PBR) for whole-year-round cultivation of a carbohydrate-rich microalga *Scenedesmus obliquus* CNW-N in southern Taiwan. Time-course profile of the carbohydrate content under nitrogen-deficient conditions was monitored to assess the seasonal changes. The optimal CO<sub>2</sub> fixation rate and carbohydrate productivity were 430.2 mg L<sup>-1</sup> d<sup>-1</sup> and 111.8 mg L<sup>-1</sup> d<sup>-1</sup>, respectively, which were obtained during the summer time. Under nitrogen starvation, the microalgal biomass can accumulate nearly 45-50% of carbohydrates, mainly composed of glucose that accounted for 70-80% of the total carbohydrates in the microalgal cells. This glucose-rich microalgal biomass is apparently a very suitable carbon source for bioethanol fermentation.

**CONCLUSION:** This work shows the feasibility of combining CO<sub>2</sub> fixation and bioethanol production using microalgae grown in outdoor photobioreactors as feedstock. The understanding of the seasonal changes in the carbohydrate productivity makes this approach more practically viable. The novel strategy proposed in this study could be a promising alternative to the existing technology dealing with CO<sub>2</sub> mitigation and biofuels production.

### **Pretreatment optimization of the biomass of *Microcystis aeruginosa* for efficient bioethanol production.**

AMB Express. 2017 Dec;7(1):19

Authors: Khan MI, Lee MG, Shin JH, Kim JD

#### **Abstract**

Microalgae are considered to be the future promising sources of biofuels and bio products. The algal carbohydrates can be fermented to bioethanol after pretreatment process. Efficient pretreatment of the biomass is one of the major requirements for commercialization of the algal based biofuels. In present study the microalga, *M. aeruginosa* was used for pretreatment optimization and bioethanol production. Treatment of algal biomass with CaO before acid and/or enzymatic hydrolysis enhanced the degradation of algal cells. Monomeric sugars yield was increased more than twice when biomass was pretreated with CaO. Similarly, an increase was noted in the amount of fermentable sugars when biomass was subjected to invertase saccharification after acid or lysozyme pretreatment. Highest yield of fermentable sugars (16 mM/ml) in the centrifuged algal juice was obtained. 4 Different microorganisms' species were used individually and in combination for converting centrifuged algal juice to bioethanol. Comparatively higher yield of bioethanol (60 mM/ml) was obtained when the fermenter microorganisms were



used in combination. The results demonstrated that *M. arginase* biomass can be efficiently pretreated to get higher yield of fermentable sugars for enhanced yield of bioethanol production.

### **Microalgae cultivation in sugarcane vinasse: Selection, growth and biochemical characterization.**

Bioresour Technol. 2017 Mar;228:133-140

Authors: Santana H, Cereijo CR, Teles VC, Nascimento RC, Fernandes MS, Brunale P, Campanha RC, Soares IP, Silva FCP, Sabaini PS, Siqueira FG, Brasil BSAF

#### **Abstract**

Sugarcane ethanol is produced at large scale generating wastes that could be used for microalgae biomass production in a biorefinery strategy. In this study, forty microalgae strains were screened for growth in sugarcane vinasse at different concentrations. Two microalgae strains, *Micractinium* sp. Embrapa|LBA32 and *C. biconvexa* Embrapa|LBA40, presented vigorous growth in a light-dependent manner even in undiluted vinasse under non-axenic conditions. Microalgae strains presented higher biomass productivity in vinasse-based media compared to standard Bold's Basal Medium in cultures performed using 15L airlift flat plate photobioreactors. Chemical composition analyses showed that proteins and carbohydrates comprise the major fractions of algal biomass. Glucose was the main monosaccharide detected, ranging from 46% to 76% of the total carbohydrates content according to the strain and culture media used. This research highlights the potential of using residues derived from ethanol plants to cultivate microalgae for the production of energy and bioproducts.

### **Outdoor pilot-scale cultivation of *Spirulina* sp. LEB-18 in different geographic locations for evaluating its growth and chemical composition.**

Bioresour Technol. 2018 May;256:86-94

Authors: de Jesus CS, da Silva Uebel L, Costa SS, Miranda AL, de Moraes EG, de Moraes MG, Costa JAV, Nunes IL, de Souza Ferreira E, Druzian JI

#### **Abstract**

This study evaluated whether outdoor cultivation of *Spirulina* sp. in different geographical locations affected its growth and biomass quality, with respect to the chemical composition, volatile compound and heavy metal content, and thermal



stability. The positive effect of solar radiation and temperature on biomass productivity in *Spirulina* sp. cultivated in the northeast was directly related to its improved nutritional characteristics, which occurred with an increase in protein, phycocyanin, and polyunsaturated fatty acid (mainly  $\gamma$ -linolenic) content. The biomass produced in Northeast and South Brazil showed high thermal stability and had volatile compounds that could be used as biomarkers of *Spirulina*, and their parameters were within the limits of internationally recognized standards for food additives; hence, they have been considered safe foods. However, the growth of crops in south Brazil occurred at lower rates due to low temperatures and luminous intensities, indicative of the robustness of microalgae in relation to these parameters.

### **Magnetic field action on outdoor and indoor cultures of *Spirulina*: Evaluation of growth, medium consumption and protein profile.**

Bioresour Technol. 2018 Feb;249:168-174

Authors: Deamici KM, Santos LO, Costa JAV

#### **Abstract**

This study aimed at evaluating whether a magnetic field (MF) affects the growth of *Spirulina* sp. when applied to it at different exposure times in indoor and outdoor culture systems. The effects of MF on chlorophyll content, medium consumption and protein profile were also investigated. In raceway tanks, a 25 mT MF was applied for 24 h or for 1 h d<sup>-1</sup>. MF for 24 h to outdoor assays increased biomass concentration and chlorophyll-a content besides altering the protein profile. Outdoor *Spirulina* growth was higher ( $\sim 3.65$  g L<sup>-1</sup>) than the growth found in indoor assays ( $\sim 1.80$  g L<sup>-1</sup>), while nitrogen and phosphorus consumption was not enhanced by the application of MF. This is the first study that investigated the influence of MF on outdoor microalga assays, and the results showed that MF affected the metabolism of *Spirulina* cultured in raceways, especially when it was grown outdoors in uncontrolled environmental conditions.



## **Enrichment of *Parachlorella kessleri* biomass with bioproducts: oil and protein by utilization of beet molasses.**

J Appl Phycol. 2017;29(4):1735-1743

Authors: Piasecka A, Krzemińska I, Tys J

### **Abstract**

The aim of this study was to determine the suitability of beet molasses, an agro-industrial by-product, as an alternative culture medium component for photoheterotrophic and mixotrophic cultivation of *Parachlorella kessleri*. Application of beet molasses improved microalgal cell growth and modified the biochemical composition of *P. kessleri* biomass. During the addition of molasses to culture media with simultaneous aeration, the maximum biomass productivity, oil and protein productivity, and calorific value were 0.42 g L<sup>-1</sup> day<sup>-1</sup>, 112.56 and 244.95 mg L<sup>-1</sup> day<sup>-1</sup>, and 22.1 MJ kg<sup>-1</sup>, respectively. Under these conditions, the total content of polyunsaturated C16-C18 fatty acids decreased, which was suitable for application in biodiesel. Besides oils and carbohydrates, *P. kessleri* had an ability to synthesize significant amounts of proteins, especially during molasses utilization. This provides a possibility of a wide range of non-fuel applications of *P. kessleri* biomass.

## **A biorefinery for *Nannochloropsis*: Induction, harvesting, and extraction of EPA-rich oil and high-value protein.**

Bioresour Technol. 2017 Nov;244(Pt 2):1416-1424

Authors: Chua ET, Schenk PM

### **Abstract**

Microalgae have been studied as biofactories for almost four decades. Yet, even until today, many aspects of microalgae farming and processing are still considered exploratory because of the uniqueness of each microalgal species. Thus, it is important to develop the entire process of microalgae farming: from culturing to harvesting, and down to extracting the desired high-value products. Based on its rapid growth and high oil productivities, *Nannochloropsis* sp. is of particular interest to many industries for the production of high-value oil containing omega-3 fatty acids, specifically eicosapentaenoic acid (EPA), but also several other products. This review compares the various techniques for induction, harvesting, and extraction of EPA-rich oil and high-value protein explored by academia and industry to develop a multi-product *Nannochloropsis* biorefinery. Knowledge gaps and opportunities are discussed for culturing and inducing fatty acid biosynthesis, biomass harvesting, and



extracting EPA-rich oil and high-value protein from the biomass of *Nannochloropsis* sp.

### **Proteins recovery from wet microalgae using liquid biphasic flotation (LBF).**

Bioresour Technol. 2017 Nov;244(Pt 2):1329-1336

Authors: Phong WN, Show PL, Teh WH, Teh TX, Lim HMY, Nazri NSB, Tan CH, Chang JS, Ling TC

#### **Abstract**

In this work, the extraction of microalgal protein from wet *Chlorella sorokiniana* species using alcohol/salt liquid biphasic flotation (LBF) with the aid of ultrasonication for cell rupturing was proposed. The effect of varying crude feedstock concentration, flotation time, salt type, salt concentration, alcohol type, alcohol concentration, initial volumes of salt and alcohol were investigated. After the optimization process, the highest proportion of protein recovered in the top phase was achieved with 250g/L ammonium sulphate, 60% (v/v) 2-propanol, 1.0VR, initial, 20g/L crude biomass load, 4mm<sup>3</sup>/min air flowrate and 10min of flotation time. The recycling of phase components was introduced to minimize the use of alcohol and salt in the corresponding LBF. It was demonstrated that top phase (alcohol) recycling can achieve increasing performance for three consecutive recycling runs. Under optimized process conditions, the proportion of protein recovered in the top phase was 88.86% for the third recycle run.

### **Assessing the Effect of Pretreatments on the Structure and Functionality of Microbial Communities for the Bioconversion of Microalgae to Biogas.**

Front Microbiol. 2018;9:1388

Authors: Córdova O, Chamy R, Guerrero L, Sánchez-Rodríguez A

#### **Abstract**

Microalgae biomethanization is driven by anaerobic sludge associated microorganisms and is generally limited by the incomplete hydrolysis of the microalgae cell wall, which results in a low availability of microalgal biomass for the methanogenic community. The application of enzymatic pretreatments, e.g., with hydrolytic enzymes, is among the strategies used to work around the incomplete



hydrolysis of the microalgae cell wall. Despite the proven efficacy of these pretreatments in increasing biomethanization, the changes that a given pretreatment may cause to the anaerobic sludge associated microorganisms during biomethanization are still unknown. This study evaluated the changes in the expression of the metatranscriptome of anaerobic sludge associated microorganisms during *Chlorella sorokiniana* biomethanization without pretreatment (WP) (control) and pretreated with commercial cellulase in order to increase the solubilization of the microalgal organic matter. Pretreated microalgal biomass experienced significant increases in biogas the production. The metatranscriptomic analysis of control samples showed functionally active microalgae cells, a bacterial community dominated by  $\gamma$ - and  $\delta$ -proteobacteria, and a methanogenic community dominated by *Methanospirillum hungatei*. In contrast, pretreated samples were characterized by the absence of active microalgae cells and a bacteria population dominated by species of the Clostridia class. These differences are also related to the differential activation of metabolic pathways e.g., those associated with the degradation of organic matter during its biomethanization.

### **Efficient Anaerobic Digestion of Microalgae Biomass: Proteins as a Key Macromolecule.**

Molecules. 2018 May 06;23(5):

Authors: Magdalena JA, Ballesteros M, González-Fernandez C

#### **Abstract**

Biogas generation is the least complex technology to transform microalgae biomass into bioenergy. Since hydrolysis has been pointed out as the rate limiting stage of anaerobic digestion, the main challenge for an efficient biogas production is the optimization of cell wall disruption/hydrolysis. Among all tested pretreatments, enzymatic treatments were demonstrated not only very effective in disruption/hydrolysis but they also revealed the impact of microalgae macromolecular composition in the anaerobic process. Although carbohydrates have been traditionally recognized as the polymers responsible for the low microalgae digestibility, protease addition resulted in the highest organic matter solubilization and the highest methane production. However, protein solubilization during the pretreatment can result in anaerobic digestion inhibition due to the release of large amounts of ammonium nitrogen. The possible solutions to overcome these negative effects include the reduction of protein biomass levels by culturing the microalgae in low nitrogen media and the use of ammonia tolerant anaerobic inocula. Overall, this review is intended to evidence the relevance of microalgae proteins in different stages of anaerobic digestion, namely hydrolysis and methanogenesis.



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## **Enhancing methane production from *U. lactuca* using combined anaerobically digested sludge (ADS) and rumen fluid pre-treatment and the effect on the solubilization of microbial community structures.**

Bioresour Technol. 2018 Apr;254:83-90

Authors: Zou Y, Xu X, Li L, Yang F, Zhang S

### **Abstract**

Methane production by the anaerobic digestion of seaweed is restricted by the slow degradation caused by the influence of the rigid algal cell wall. At the present time, there has been no study focusing on the anaerobic digestion of *U. lactuca* by co-fermentation and pre-treatment with rumen fluid. Rumen fluid can favor methane production from algal biomass by utilizing the diversity and quantity of bacterial and archaeal communities in the rumen fluid. This research presents a novel method based on combined ADS and rumen fluid pre-treatment to improve the production of methane from seaweed. Biochemical methane potential (BMP) tests were performed to investigate the biogas production using combined ADS and rumen fluid pre-treatment at varied inoculum ratios on the performance of methane production from *U. lactuca* biomass. Compared to the control (no rumen fluid pre-treatment), the highest BMP yields of *U. lactuca* increased from 3%, 27.5% and 39.5% to 31.1%, 73% and 85.6%, respectively, for three different types of treatment. Microbial community analysis revealed that the *Methanobrevibacter* species, known to accept electrons to form methane, were only detected when rumen fluid was added. Together with the significant increase in species of *Methanoculleus*, *Methanospirillum* and *Methanosaeta*, rumen fluid improved the fermentation and degradation of the microalgae biomass not only by pre-treatment to foster cell-wall degradation but also by relying on methane production within itself during anaerobic processes. Batch experiments further indicated that rumen fluid applied to the co-fermentation and pre-treatment could increase the economic value and hold promise for enhancing biogas production from different seaweed species.



## **Anaerobic co-digestion of coffee husks and microalgal biomass after thermal hydrolysis.**

Bioresour Technol. 2018 Apr;253:49-54

Authors: Passos F, Cordeiro PHM, Baeta BEL, de Aquino SF, Perez-Elvira SI

### **Abstract**

Residual coffee husks after seed processing may be better profited if bioconverted into energy through anaerobic digestion. This process may be improved by implementing a pretreatment step and by co-digesting the coffee husks with a more liquid biomass. In this context, this study aimed at evaluating the anaerobic co-digestion of coffee husks with microalgal biomass. For this, both substrates were pretreated separately and in a mixture for attaining 15% of total solids (TS), which was demonstrated to be the minimum solid content for pretreatment of coffee husks. The results showed that the anaerobic co-digestion presented a synergistic effect, leading to 17% higher methane yield compared to the theoretical value of both substrates biodegraded separately. Furthermore, thermal hydrolysis pretreatment increased coffee husks anaerobic biodegradability. For co-digestion trials, the highest values were reached for pretreatment at 120 °C for 60 min, which led to 196 mLCH<sub>4</sub>/gVS and maximum methane production rate of 0.38 d<sup>-1</sup>.

## **Qualitative Analysis of Microbial Dynamics during Anaerobic Digestion of Microalgal Biomass in a UASB Reactor.**

Int J Microbiol. 2017;2017:5291283

Authors: Doloman A, Soboh Y, Walters AJ, Sims RC, Miller CD

### **Abstract**

Anaerobic digestion (AD) is a microbiologically coordinated process with dynamic relationships between bacterial players. Current understanding of dynamic changes in the bacterial composition during the AD process is incomplete. The objective of this research was to assess changes in bacterial community composition that coordinates with anaerobic codigestion of microalgal biomass cultivated on municipal wastewater. An upflow anaerobic sludge blanket reactor was used to achieve high rates of microalgae decomposition and biogas production. Samples of the sludge were collected throughout AD and extracted DNA was subjected to next-generation sequencing using methanogen *mcrA* gene specific and universal bacterial primers. Analysis of the data revealed that samples taken at different stages of AD had varying bacterial composition. A group consisting of Bacteroidales, Pseudomonadales, and Enterobacteriales was identified to be putatively responsible



for the hydrolysis of microalgal biomass. The methanogenesis phase was dominated by *Methanosarcina mazei*. Results of observed changes in the composition of microbial communities during AD can be used as a road map to stimulate key bacterial species identified at each phase of AD to increase yield of biogas and rate of substrate decomposition. This research demonstrates a successful exploitation of methane production from microalgae without any biomass pretreatment.

### **Waste biorefineries - integrating anaerobic digestion and microalgae cultivation for bioenergy production.**

Curr Opin Biotechnol. 2018 Apr;50:101-110

Authors: Chen YD, Ho SH, Nagarajan D, Ren NQ, Chang JS

#### **Abstract**

Commercialization of microalgal cultivation has been well realized in recent decades with the use of effective strains that can yield the target products, but it is still challenged by the high costs arising from mass production, harvesting, and further processing. Recently, more interest has been directed towards the utilization of waste resources, such as sludge digestate, to enhance the economic feasibility and sustainability of microalgae production. Anaerobic digestion for waste disposal and phototrophic microalgal cultivation are well-characterized technologies in both fields. However, integration of anaerobic digestion and microalgal cultivation to achieve substantial economic and environmental benefits is extremely limited, and thus deserves more attention and research effort. In particular, combining these two makes possible an ideal 'waste biorefinery' model, as the C/N/P content in the anaerobic digestate can be used to produce microalgal biomass that serves as feedstock for biofuels, while biogas upgrading can simultaneously be performed by phototrophic CO<sub>2</sub> fixation during microalgal growth. This review is thus aimed at elucidating recent advances as well as challenges and future directions with regard to waste biorefineries associated with the integration of anaerobic waste treatment and microalgal cultivation for bioenergy production.

### **Acclimation to extremely high ammonia levels in continuous biomethanation process and the associated microbial community dynamics.**

Bioresour Technol. 2018 Jan;247:616-623

Authors: Tian H, Fotidis IA, Mancini E, Treu L, Mahdy A, Ballesteros M, González-Fernández C, Angelidaki I



## Abstract

Acclimatized anaerobic communities to high ammonia levels can offer a solution to the ammonia toxicity problem in biogas reactors. In the current study, a stepwise acclimation strategy up to 10g NH<sub>4</sub><sup>+</sup>-N L<sup>-1</sup>, was performed in mesophilic (37±1°C) continuously stirred tank reactors. The reactors were co-digesting (20/80 based on volatile solid) cattle slurry and microalgae, a protein-rich, 3rd generation biomass. Throughout the acclimation period, methane production was stable with more than 95% of the uninhibited yield. Next generation 16S rRNA gene sequencing revealed a dramatic microbiome change throughout the ammonia acclimation process. *Clostridium ultunense*, a syntrophic acetate oxidizing bacteria, increased significantly alongside with hydrogenotrophic methanogen *Methanoculleus* spp., indicating strong hydrogenotrophic methanogenic activity at extreme ammonia levels (>7g NH<sub>4</sub><sup>+</sup>-N L<sup>-1</sup>). Overall, this study demonstrated for the first time that acclimation of methanogenic communities to extreme ammonia levels in continuous AD process is possible, by developing a specialised acclimation AD microbiome.

## **Co-digestion of chicken manure and microalgae *Chlorella* 1067 grown in the recycled digestate: Nutrients reuse and biogas enhancement.**

Waste Manag. 2017 Dec;70:247-254

Authors: Li R, Duan N, Zhang Y, Liu Z, Li B, Zhang D, Lu H, Dong T

## Abstract

The present investigation targeted on a sustainable co-digestion system: microalgae *Chlorella* 1067 (Ch. 1067) was cultivated in chicken manure (CM) based digestate and then algae biomass was used as co-substrate for anaerobic digestion with CM. About 91% of the total nitrogen and 86% of the soluble organics in the digestate were recycled after the microalgae cultivation. The methane potential of co-digestion was evaluated by varying CM to Ch. 1067 ratios (0:10, 2:8, 4:6, 6:4, 8:2, 10:0 based on the volatile solids (VS)). All the co-digestion trials showed higher methane production than the calculated values, indicating synergy between the two substrates. Modified Gompertz model showed that co-digestion had more effective methane production rate and shorter lag phase. Co-digestion (8:2) achieved the highest methane production of 238.71mL·(g VS)<sup>-1</sup> and the most significant synergistic effect. The co-digestion (e.g. 8:2) presented higher and balanced content of dominant acidogenic bacteria (Firmicutes, Bacteroidetes, Proteobacterias and Spirochaetae). In addition, the archaea community *Methanosaeta* presented higher content than *Methanosarcina*, which accounted for the higher methane production. These findings indicated that the system could provide a practicable strategy for effectively recycling digestate and enhancing biogas production simultaneously.



## PATENTES

### HETEROTROPHIC PRODUCTION METHODS FOR MICROBIAL BIOMASS AND BIOPRODUCTS

**Inventor(s):** SCHURR ROBERT J [US]; KUEHNLE ADELHEID R [US] ± (Schurr Robert J, ; Kuehnle Adelheid R)  
**Applicant(s):** KUEHNLE AGROSYSTEMS INC [US] ± (Kuehnle AgroSystems, Inc)  
**Application number:** US201715640246 20170630 [Global Dossier](#)  
**Priority number(s):** US201715640246 20170630 ; [US201662356896P 20160630](#)

#### Abstract of US2018002711 (A1)

The invention pertains to a method for synthesizing a product of interest by culturing a microalgal cell producing the product of interest in the dark in a culture medium comprising an organic acid as a fixed carbon source, wherein the microalgal cell is a facultative heterotroph. The product of interest can be a microalgal biomass, a pigment, terpene, recombinant molecule, biogas, or a precursor thereof. In an embodiment, the culture medium comprises urea as a primary source of nitrogen. In one embodiment, the microalgal cell belongs to the order Chlamydomonadales. A method of identifying and isolating a microalgal cell having a preferred characteristic that is suitable for synthesis of a product of interest is also provided, the method comprising identifying and isolating a non-mutagenized or recombinant microalgal cell from a microalgal culture using a fluorescence activated cell sorting technique and/or a phototactic response.

### METHOD OF EFFECTIVELY UTILIZING BIOMASS

**Inventor(s):** YU JIANZHONG; XUE MINGXIONG ± (俞建中, ; 薛命雄)  
**Applicant(s):** BEIHAI SPD BIO SCIENCE TECH CO LTD ± (北海生巴达生物科技有限公司)  
**Application number:** CN20161744762 20160828 [Global Dossier](#)  
**Priority number(s):** CN20161744762 20160828

#### Abstract of CN106269799 (A)

The invention relates to the technical field of energy resource environment protection, in particular to a method of effectively utilizing biomass. The method includes: using



a biomass power plant as a core system and one or multiple of biogas fermentation, microalgae breeding and greenhouse vegetable planting as supporting assistant systems to acquire a combined system; utilizing waste generated by each system as production raw materials of other systems or to meet necessary conditions for running other systems; forming a circulating system for waste recycling. The waste generated by each system serves as the raw materials or assistant conditions of other systems, so that discharging of the waste is eliminated or reduced to greatest extent, the objectives of saving energy, reducing discharging, improving efficiency and fully utilizing biomass resources are achieved, and utilization efficiency of biomass energy is improved. Nitrogen element and trace elements are effectively returned into soil, so that consumption of chemical fertilizer and other fertilizer is reduced, and production cost of microalgae, vegetables and grain crops is lowered.

## METHOD FOR SYNCHRONOUSLY ENHANCING YIELDS OF LUTEIN AND CARBOHYDRATES OF AUTOTROPHIC MICROALGAE

**Inventor(s):** XIE YOUPIPING; YANG XUQIU; CHEN JIANFENG; SHEN YING; ZHENG XIANGNAN ± (谢友坪; 阳需求; 陈剑锋; 沈英; 郑向南)

**Applicant(s):** UNIV FUZHOU ± (福州大学)

**Application number:** CN201611030118 20161122 [Global Dossier](#)

**Priority number(s):** CN201611030118 20161122

### Abstract of CN106399111 (A)

The invention relates to a method for synchronously enhancing yields of lutein and carbohydrates of autotrophic microalgae. The method comprises the following steps: inoculating alga seeds into a seed culture medium, culturing to obtain a seed solution, inoculating the seed solution into a photobiological reactor filled with a fermentation culture medium, and culturing. In the culturing process, the initial illumination intensity is controlled at 60-300  $\mu\text{mol}/\text{m}^2/\text{s}$ , and the illumination intensity is enhanced by 50-150  $\mu\text{mol}/\text{m}^2/\text{s}$  every 12 hours until the fermentation finishes; and meanwhile, when the initial nitrogen source concentration starts exhaustion, a nitrogen source with the nitrogen concentration of 2-14 g/L is fed at the constant speed of 1-10 mg/L/h, and the fermentation period is 4-6 days. The biomass concentration of the microalgae cultured by the method can reach 4-9 g/L, the yield of lutein is 20-60 mg/L, and the yield of carbohydrates is 1.4-3.3 g/L. The method has the advantages of short fermentation period, simple production technique and low production cost, and can obviously widen the industrialization prospects for simultaneously producing lutein and biofuels by using microalgae.



## METHOD FOR SPIRULINA MICROALGA CULTIVATION

**Inventor(s):** RUDIC VALERIU [MD]; CEAPURINA LUDMILA [MD]; DENCICOV LIDIA [MD]; DIACON ION [MD]; CAIREAC LEONID [MD] ± (RUDIC VALERIU, ; CEAPURINA LUDMILA, ; DENCICOV LIDIA, ; DIACON ION, ; CAIREAC LEONID, ; РУДИК Валериу, ; ЧАПУРИНА Людмила, ; ДЕНЧИКОВ Лидия, ; DIACON Ivan, ; CAIREAC Leonid)

**Applicant(s):** RUDIC VALERIU [MD]; CEAPURINA LUDMILA [MD]; DENCICOV LIDIA [MD]; DIACON ION [MD]; CAIREAC LEONID [MD] ± (RUDIC VALERIU, ; CEAPURINA LUDMILA, ; DENCICOV LIDIA, ; DIACON ION, ; CAIREAC LEONID, ; РУДИК Валериу, ; CEAPURINA Ludmila, ; DENCICOV Lidia, ; DIACON Ion, ; CAIREAC Leonid)

**Application number:** MD19940000223 19940722

**Priority number(s):** MD19940000223 19940722 ; [SU19894772555 19891222](#)

### Abstract of MD163 (C2)

The invention relates to the biotechnology, particularly to the microalgae cultivation, namely, Spirulina alga- the valuable protein-vitaminic biomass. The aim of the invention consists in increasing the yield of spirulina blue-green microalga. The method consists in the fact that in the nutrient mineral medium there is introduced the coordinative copper compound of copper (II) L- $\alpha$ -alanilate-L-scrinate in quantity of 0,05-0,1 mg/l, after which the microalgae are cultivated at the continuous illumination and mixing in the collection regime. The proposed method provides the increasing of spirulina biomass yield on the 5th day, average 20-40% in comparison with the known method.

## METHOD FOR IMPROVING HYDROGEN PRODUCTION QUANTITY OF PHOTOSYNTHETIC MICROALGAE THROUGH PHYCOMYCETES CO-CULTURE

**Inventor(s):** GE BAOSHENG; XI LIJUN; HE JIAYI; HUANG FANG ± (葛保胜, ; 郝丽君, ; 贺嘉怡, ; 黄方)

**Applicant(s):** CHINA UNIV OF PETROLEUM EAST CHINA ± (中国石油大学 (华东) )

**Application number:** CN201711139565 20171116 [Global Dossier](#)

**Priority number(s):** CN201711139565 20171116



## Abstract of CN107663529 (A)

The invention discloses a method for improving the hydrogen production quantity of photosynthetic microalgae through phycomycetes co-culture. Particularly, green alga and one kind of facultative anaerobic/facultative chemoautotrophic thiomonas intermedia are proportionally mixed for culture; oxygen released by green alga photosynthesis can be consumed by the bacterium respiration effect; the carbon dioxide released through the bacterium respiration effect can be supplied to the green alga for better performing photosynthesis effect, so that the anaerobic characteristics of the whole culture environment can be well maintained. In addition, the balance catalysis capability of the thiomonas intermedia on the sulfur element is utilized; the limited supply of the sulfur element can be realized, so that the normal growth of the green alga can be ensured; the efficient durable hydrogen production can be realized.

### MICROALGAE MEDIUM AND METHOD FOR CULTURING MICROALGAE TO PRODUCE HYDROGEN

**Inventor(s):** SHU LEI; MA WEIMIN; XU XURONG; TANG RUIKANG ± (舒蕾, ; 马为民, ; 徐旭荣, ; 唐睿康)

**Applicant(s):** UNIV ZHEJIANG ± (浙江大学)

**Application number:** CN20171571983 20170713 [Global Dossier](#)

**Priority number(s):** CN20171571983 20170713

## Abstract of CN107267395 (A)

The invention discloses a microalgae medium and a method for culturing microalgae to produce hydrogen. The microalgae medium contains a basic medium and an additive, the additive is DMSO, and the adding amount of DMSO is 0.1%-2% according to a volume ratio. The method comprises the steps of: (1) constructing a microalgae aggregate; and (2) resuspending the microalgae aggregate in the microalgae medium, and performing culture under an illumination condition to produce hydrogen. According to the microalgae medium provided by the invention, a certain amount of DMSO is added into an ordinary medium, when the microalgae medium is applied in culturing microalgae to produce hydrogen, cellular respiration is enhanced, so that more layers of cells in the aggregate are in an oxygen-free environment and participate in hydrogen production, the hydrogenase activity is enhanced, and the hydrogen yield and hydrogen production rate are also effectively improved.



## METHOD FOR PRODUCING METHANE FROM CARBON DIOXIDE BY CO-CULTURE

**Inventor(s):** REVERSO RICCARDO [IT] ± (REVERSO, RICCARDO)  
**Applicant(s):** BIOREWEAL S R L [IT] ± (BIOREWEAL S.R.L.)  
**Application number:** CA20163003829 20161115 [Global Dossier](#)  
**Priority number(s):** [IT2015UB05703](#) [20151118](#) ; [WO2016EP77771](#)  
[20161115](#)

### Abstract of CA3003829 (A1)

A method for producing methane by biological conversion of carbon dioxide, performed by means of a symbiosis between one or more methane-generating bacteria and: (i) one or more hetero-autotrophic cyanobacteria and/or microalgae, or (ii) one or more sulfobacteria and/or acetobacteria, wherein said hetero-autotrophic cyanobacteria and/or microalgae, or said sulfobacteria and/or acetobacteria, produce the molecular hydrogen required for the conversion of carbon dioxide into methane performed by said methane-generating bacteria.

## BIOLOGICAL TREATMENT DEVICE AND PROCESS FOR HIGH AMMONIA-NITROGEN PIG-RAISING BIOGAS SLURRY

**Inventor(s):** YANG CHUNPING; LUO LE; YAN ZHOU; QIU LU; PENG ZHIXIONG; WEN SHAN; LIU HAIYANG; ZHOU ZILI; LOU WEI ± (杨春平, ; 罗乐, ; 严洲, ; 仇璐, ; 彭志雄, ; 文珊, ; 刘海洋, ; 周自力, ; 娄伟)  
**Applicant(s):** UNIV HUNAN ± (湖南大学)  
**Application number:** CN20161863975 20160929 [Global Dossier](#)  
**Priority number(s):** CN20161863975 20160929

### Abstract of CN106430820 (A)

The invention discloses a biological treatment device and process for high ammonia-nitrogen pig-raising biogas slurry. A body of the biological treatment device is a runway type photo-bioreactor, an efficient and low-consumption standard treatment process for the high ammonia-nitrogen pig-raising biogas slurry is constructed based on a microalgae, photosynthetic bacteria and plant compound ecological system, and meanwhile, algae bacteria biomasses and plants are harvested. The process comprises the following specific steps: pig-raising wastewater is subjected to solid-liquid separation and is introduced into an anaerobic fermentation tank, produced high ammonia-nitrogen biogas slurry is subjected to pH value regulation and nutritive salt



regulation and is introduced into the photo-bioreactor to be treated, biochemical effluent is treated by virtue of a flocculation settling tank and a disinfecting tank, and clean water reaches the standard and is discharged. The biological treatment device and the biological treatment process have the advantages that the operation is simple, the removal efficiencies of nitrogen and phosphorus are high, the operation cost is low, the separated algae bacteria biomasses can be taken as the raw materials for feeds, biological hydrogen production or biodiesel, and the secondary pollution is avoided.

### MICROALGAE WITH IMPROVED PHOTOTAXIS AND PHOTOSYNTHETIC EFFICIENCY

**Inventor(s):** SIM SANG JUN [KR]; KWAK HO-SEOK [KR]; KIM YOUNG HWAN [KR] ± (Sim Sang Jun, ; Kwak Ho-Seok, ; Kim Young Hwan)

**Applicant(s):** UNIV KOREA RES & BUS FOUND [KR] ± (Korea University Research and Business Foundation)

**Application number:** US201715424866 20170205 [Global Dossier](#)

**Priority number(s):** [KR20160132021 20161012](#) ; [KR20160174591 20161220](#)

#### Abstract of US2018100206 (A1)

The present invention relates to microalgae with improved phototaxis and photosynthetic efficiency, and more particularly, to a microalgae PTS42 (KCTC18499P) mutant of *Chlamydomonas reinhardtii* and uses thereof. In the microalgae PTS42 according to the present invention of which photosensitivity and photosynthetic efficiency are improved due to excellent phototaxis, conversion of carbon dioxide into biomass is excellent as compared to a wild type strain and a maximum photosynthesis rate and lipid accumulation ability are also high, such that the microalgae PTS42 are useful as a photosynthetic strain for constructing a biofuel production process as well as an effect of decreasing carbon dioxide.

### TAILORED OILS PRODUCED FROM RECOMBINANT OLEAGINOUS MICROORGANISMS.

**Inventor(s):** FRANKLIN SCOTT [US]; SOMANCHI ARAVIND; WEE JANICE; RUDENKO GEORGE; MOSELEY JEFFREY; RAKITSKY WALT; ZHAO XINHUA; BHAT RIYAZ ± (Franklin SCOTT, ; Somanchi ARAVIND, ; Wee JANICE, ; Rudenko GEORGE, ; Moseley JEFFREY, ; Rakitsky WALT, ; Zhao XINHUA, ; Bhat RIYAZ)

**Applicant(s):** TERRAVIA HOLDINGS INC [US] ± (TERRAVIA HOLDINGS, INC)



**Application number:** MX20160015902 20120202

**Priority number(s):** [US201161438969P 20110202](#) ; [US201161476691P 20110418](#) ; [US201161484458P 20110510](#) ; [US201161548616P 20111018](#) ; [WO2012US23696 20120202](#)

### **Abstract of MX351063 (B)**

Methods and compositions for the production of oil, fuels, oleochemicals, and other compounds in recombinant microorganisms are provided, including oil-bearing microorganisms and methods of low cost cultivation of such microorganisms. Microalgal cells containing exogenous genes encoding, for example, a lipase, a sucrose transporter, a sucrose invertase, a fructokinase, a polysaccharide-degrading enzyme, a keto acyl-ACP synthase enzyme, a fatty acyl-ACP thioesterase, a fatty acyl-CoA/aldehyde reductase, a fatty acyl-CoA reductase, a fatty aldehyde reductase, a fatty acid hydroxylase, a desaturase enzyme, a fatty aldehyde decarbonylase, and/or an acyl carrier protein are useful in manufacturing transportation fuels such as renewable diesel, biodiesel, and renewable jet fuel, as well as oleochemicals such as functional fluids, surfactants, soaps and lubricants.

### **SCENEDESMUS ACUMINATUS SYNTHESIS STARCH DEFECT-TYPE MUTANT STRAINS AND APPLICATION THEREOF**

No documents available for this priority number.

**Inventor(s):** HAN DANXIANG; SUN WENCHAO; WU MINGCAN; HU QIANG ± (韩丹翔, ; 孙文超, ; 吴明灿, ; 胡强)

**Applicant(s):** STATE DEVELOPMENT & INVEST CORPORATION; CHINA ELECTRONICS ENGINEERING DESIGN INST ± (国家开发投资公司, ; 中国电子工程设计院)

**Application number:** CN20171400736 20170531 [Global Dossier](#)

**Priority number(s):** CN20171400736 20170531

### **Abstract of CN107354147 (A)**

The invention provides the Scenedesmus acuminatus synthesis starch defect-type mutant strains MSA1 and MSA5 obtained through screen mutation; the preservation numbers are CGMCC No. 13864 and CGMCC No.13866, which are preserved in general microbiological center of Chinese microorganism preservation management committee. By screening the non-synthesized starch or starch with little starch synthetic quantity, the defect mutants are synthesized, a heterotrophic and photo-autotrophic type from the alternative mutants is found, and finally the starch-synthesized defect mutant with fast growth rate and large biomass under heterotrophic



culture of neutral grease and triacylglycerol is synthesized under photo-autotrophic condition. The mutant strains MSA1 and MSA5 can rapidly accumulate the total aliphatic acid content under heterotrophic fermentation condition, the starch output is greatly reduced, and the total aliphatic acid content in a final fermentation result is increased by 5-10 times by comparing with a wild type. The conversion rate from a carbon source to grease is greatly increased, the mutant strains can be taken as industrial grade oil-production microalgae, and can be widely used for preparing biodiesel.

## SOROKIN CHLORELLA SYNTHETIC STARCH DEFECT TYPE MUTANT STRAIN AND APPLICATION THEREOF

**Inventor(s):** HAN DANXIANG; WU MINGCAN; SUN WENCHAO; HU QIANG ± (韩丹翔, ; 吴明灿, ; 孙文超, ; 胡强)

**Applicant(s):** STATE DEVELOPMENT & INVEST CORPORATION; CHINA ELECTRONICS ENGINEERING DESIGN INST ± (国家开发投资公司, ; 中国电子工程设计院)

**Application number:** CN20171286674 20170427 [Global Dossier](#)

**Priority number(s):** CN20171286674 20170427

### Abstract of CN107118968 (A)

The invention provides sorokin chlorella synthetic starch defect type mutant strains GT-1-SLM1, GT-1-SLM2 and GT-1-SLM3 through mutation screening. The preservation numbers of the strains are respectively CGMCC No.13861, CGMCC No.13862 and CGMCC No.13863, and the strains are preserved in the China General Microbiological Culture Collection Center. As related channels or a small part of the channels for synthesizing starch through the strains are blocked, the content of starch in cells is remarkably reduced, the grease synthesis velocity is remarkably increased, and the highest grease content in the cells is remarkably increased. By adopting the mutant strains provided by the invention, the microalgae oil generation period can be shortened, so that the risk of disease infection in the culture period can be reduced, and meanwhile as biomasses have relatively high content of grease, later grease extraction and preparation processes can be simplified, and high-quality germplasm resources can be provided for industrialization of microalgae oil production.

## PLANT HORMONE, APPLICATION THEREOF AND METHOD FOR INCREASING OIL CONTENT OF PHAEODACTYLUM TRICORNUTUM

**Inventor(s):** CUI YULIN; QIN SONG ± (崔玉琳, ; 秦松)

**Applicant(s):** YANTAI INSTITUTE OF COASTAL ZONE RES CHINESE



ACADEMY OF SCIENCES ± (中国科学院烟台海岸带研究所)

**Application number:** CN20171804337 20170908 [Global Dossier](#)  
**Priority number(s):** CN20171804337 20170908

#### Abstract of CN107354121 (A)

The invention belongs to the technical field of microalgae organism and specifically provides a plant hormone, an application thereof and a method for increasing oil content of phaeodactylum tricornutum. The salicylic acid is taken as the plant hormone. The salicylic acid has no inhibition to the growth rate, photosynthetic rate and biomass of the phaeodactylum tricornutum and is capable of obviously increasing the neutral oil content in the phaeodactylum tricornutum. The method is simple and feasible, low in cost and high in oil content of target object and can be used as an efficient new method for industrial biological oil production. The method has wide application prospect at the aspect of biodiesel development.

#### APPLICATION OF MELATONIN TO IMPROVEMENT IN OIL-BEARING MICROALGAE OIL CONTENT

**Inventor(s):** YU XUYA; LI DAFEI; ZHAO YONGTENG ± (余旭亚; 李大菲; 赵永腾)  
**Applicant(s):** KUNMING UNIV OF SCIENCE AND TECHNOLOGY ± (昆明理工大学)  
**Application number:** CN20171696781 20170815 [Global Dossier](#)  
**Priority number(s):** CN20171696781 20170815

#### Abstract of CN107475171 (A)

The invention discloses new application of melatonin, namely application of melatonin to improvement in the content of oil in oil-bearing microalgae. Results display that the accumulation of the oil in the microalgae can be efficiently promoted only through adding a small quantity of melatonin; the method is simple, the oil yield is improved, and an effective technical means is provided for the problem of low oil yield existing in the microalgae industrialization process.



## METHOD USING PULSED ELECTRIC FIELD TO ASSIST EXTRACTION OF OIL FROM MICROALGAE

**Inventor(s):** JIN WENBIAO; HAN SONGFANG; YANG QIAN; TU RENJIE ± (金文标, ; 韩松芳, ; 杨倩, ; 涂仁杰)  
**Applicant(s):** HARBIN INST TECHNOLOGY SHENZHEN GRADUATE SCHOOL ± (哈尔滨工业大学深圳研究生院)  
**Application number:** CN20171358924 20170519 [Global Dossier](#)  
**Priority number(s):** CN20171358924 20170519

### Abstract of CN107245371 (A)

The invention relates to a method using a pulsed electric field to assist the extraction of oil from microalgae. The method comprises the following steps: 1, breaking walls: pumping a certain concentration of a microalga solution into the pulsed electric field, and carrying out wall breaking treatment; 2, centrifuging: centrifuging the wall-broken microalga solution obtained in step 1; 3, extracting: taking alga mud obtained in step 2, adding an extraction solvent to the alga mud, and carrying out an extraction reaction for 0.5-30 h; and 4, leaching: adding distilled water to a product obtained in step 3, taking an oil-containing organic phase, completely volatilizing the organic component, and drying the obtained organic phase to obtain crude oil. Microalga cells with the walls being broken by the pulsed electric field have the advantages of high broken wall efficiency, low cost and industrialization realization; and the low toxicity solvent substitutes traditional high toxicity chloroform/methanol, so the extraction efficiency is guaranteed, the extraction process is environmentally-friendly, and the method has a good industrial prospect in the extraction of the microalga oil.

## MICROALGAE CAPABLE OF SIMULTANEOUSLY ACCUMULATING GREASES AND SECRETING AMINO ACIDS AND APPLICATION OF MICROALGAE

**Inventor(s):** WANG QIANG; CHEN WEIXIAN ± (王强, ; 陈为先)  
**Applicant(s):** WUHAN ZAOYOU BIOTECHNOLOGY CO LTD ± (武汉藻优生物科技有限公司)  
**Application number:** CN20171661334 20170804 [Global Dossier](#)  
**Priority number(s):** CN20171661334 20170804



## Abstract of CN107435028 (A)

The invention relates to microalgae capable of simultaneously accumulating greases and secreting amino acids, and also relates to an application of the microalgae in producing microalgae oil and amino acids and also relates to a method for cultivating the microalgae by virtue of FGFS so as to produce the microalgae oil and the amino acids, wherein the microalgae is preserved in China Center for Type Culture Collection on November 24, 2016 with preservation number of CCTCC NO: M2016673. With the application of the microalgae provided by the invention, inorganic nitrogen in a medium is transformed into organic nitrogen, so that a removal rate on the inorganic nitrogen in the medium is effectively improved, and meanwhile, a product added value of algae culture is also improved. The greases accumulated by the microalgae can be extracted and applied as bio-diesel, and algae cells, together with the amino acids in culture liquid, can be used for preparing feed. Unsaturated fatty acid in the algae cells and the amino acids in the culture liquid can be also extracted and used for preparing foods, nutrients or medicines.

### METHOD FOR PREPARING MICROALGAE LIPID THROUGH MIXED CULTURE

**Inventor(s):** SHI WENJING; SUN QIMEI; LIAO SHA; WANG PENGXIANG; LI XIAOSHU ± (师文静; 孙启梅; 廖莎; 王鹏翔; 李晓姝)

**Applicant(s):** CHINA PETROLEUM & CHEM CORP; SINOPEC FUSHUN RES INST PETROLEUM & PETROCHEMICALS ± (中国石油化工股份有限公司; 中国石油化工股份有限公司抚顺石油化工研究院)

**Application number:** CN20161386731 20160605 [Global Dossier](#)

**Priority number(s):** CN20161386731 20160605

## Abstract of CN107460217 (A)

The invention discloses a method for preparing microalgae lipid through mixed culture. The method comprises the following steps: adding a microalgae medium and a mixed microalgae seed solution to a photobioreactor, keeping pH value of a culture system at 8-12, preferably at 9-11, and supplying gas that a CO<sub>2</sub> content is 5-45v%, preferably 10-30v%, wherein the mixed microalgae includes parachlorella kessleri FSH-Y3 and ankistrodesmus sp. SS-B7 which are preserved in China General Microbiological Culture Collection Center respectively on May 26, 2014 and April 15, 2013 with preservation numbers of CGMCC No.9238 and CGMCC No.7478. According to the method provided by the invention, the tolerance and the solubility of the microalgae culture system to high-concentration CO<sub>2</sub> are improved, a carbon fixation efficiency is improved and the yield of the microalgae lipid is obviously improved; and the method is conducive to the production of biodiesel.



## METHOD FOR CULTIVATING MICROALGAE USING SEWAGE AND LIQUEFIED FERTILIZER

**Inventor(s):** LEE TAE HO [KR]; PARK SEONG HWAN [KR]; KIM JEONG MI [KR] ± (LEE, TAE HO, ; PARK, SEONG HWAN, ; KIM, JEONG MI)  
**Applicant(s):** PUSAN NAT UNIV IND COOP FOUND [KR] ± (PUSAN NATIONAL UNIVERSITY INDUSTRY-UNIVERSITY COOPERATION FOUNDATION)  
**Application number:** KR20130134450 20131106 [Global Dossier](#)  
**Priority number(s):** KR20130134450 20131106

### Abstract of KR20150052938 (A)

The present invention relate to a composition comprising sewage and a liquid fertilizer as active ingredients for culturing microalgae, and a culture method of microalgae using the same. The culture method of microalgae according to the present invention is economically feasible in terms of using sewage and a liquid fertilizer to culture microalgae and has an effect of remarkably improving lipid productivity as well as the growth rate of *Micractinium inermum* NLP-F014 microalgae. Further, the *Micractinium inermum* NLP-F014 strain has a high content of C16 to C18 fatty acids, thus being suitable for use in a biofuel composition.

## MANIPULATION OF MICROALGAL LIPID METABOLIC PATHWAY BY ALTERING CAMP SIGNALING PATHWAYS

**Inventor(s):** CHOI YOON E [KR]; LEE CHANG SU [KR]; YANG JI WON [KR] ± (최윤이, ; 이창수, ; 양지원)  
**Applicant(s):** NAT UNIV CHONBUK IND COOP FOUND [KR] ± (전북대학교산학협력단)  
**Application number:** KR20130138833 20131115 [Global Dossier](#)  
**Priority number(s):** KR20130138833 20131115

### Abstract of KR20150056174 (A)



The present invention relates to a method for adjusting the microalgae lipid metabolism by treating the chemical substance called 3-isobutyl-1-methylxanthine (IBMX) which affects the cAMP signaling. The present invention adjusts the cAMP signaling process through the IBMX treatment to up-regulate the microalgae lipid metabolism. The signaling can be applied to various types of microalgae instead of one type of strains. The cAMP signaling system adjusting method enables the products of the microalgae lipid metabolism to be industrially useful.

## COAL-FIRED POWER PLANT EFFLUENT GAS CARBON DIOXIDE FILM CAPTURING COUPLING MICROALGAE CARBON SEQUESTRATION PROCESS

**Inventor(s):** YANG ZHENGSAN ± (杨正山)  
**Applicant(s):** YANG ZHENGSAN ± (杨正山)  
**Application number:** CN201711195589 20171124 [Global Dossier](#)  
**Priority number(s):** CN201711195589 20171124

### Abstract of CN107744719 (A)

The invention discloses a coal-fired power plant effluent gas carbon dioxide film capturing coupling microalgae carbon sequestration process, which consists of a carbon dioxide film capturing part and a microalgae carbon sequestration part. Under the condition of enabling the total capturing target of a system to be unchanged, the unit capturing cost of a film capturing system is reduced through reducing the carbon dioxide capturing rate of a film capturing system; the economic benefits of the process are improved through a lipid processing product and a microalgae powder processing product synthesized by the microalgae carbon sequestration part through photosynthesis. Interception gas in the film capturing technology part is used as a carbon source; the breeding cost and difficulty of the algae capable of tolerating high-concentration carbon dioxide and pollutants are reduced; the application range of the microalgae is expanded; the treatment cost of oxynitride and oxysulfide in the smoke gas when coal-fired power plant effluent gas is directly used as a carbon source is reduced.

## METHOD FOR CULTIVATING CHROOCOCCUS BY DOMESTIC- WASTEWATER-DILUTED KITCHEN WASTE ANAEROBIC DIGESTION SOLUTION

**Inventor(s):** PEI HAIYAN; CHENG JUAN; HAN FEI; JIANG LIQUN; NIE CHANGLIANG; ZHANG LIJIE ± (PEI HAIYAN, ; CHENG JUAN, ; HAN FEI, ; JIANG LIQUN, ; NIE CHANGLIANG, ; ZHANG LIJIE)  
**Applicant(s):** UNIV SHANDONG ± (SHANDONG UNIVERSITY)  
**Application** CN2016134390 20160119 [Global Dossier](#)



**number:**

**Priority  
number(s):** CN2016134390 20160119

**Abstract of CN105483015 (A)**

The invention discloses a method for cultivating chroococcus by a domestic-wastewater-diluted kitchen waste anaerobic digestion solution and belongs to the technical field of microalgae biology. The method is characterized in that BG11, domestic wastewater, a digestive fluid and culture solutions diluted by different times are utilized for constant-temperature light culture of chroococcus SDEC-6 and chroococcus SDEC-6 is harvested in the last growth stage. Results show that the biomass, the growth rate, the lipid content and the like of chroococcus SDEC-6 in domestic wastewater and at different dilution times are higher than those of BG11, the biomass and the lipid content, obtained by diluting a culture medium by 100 times, are higher than those of domestic wastewater. In conclusion, a mixed culture solution obtained by the domestic-wastewater-diluted kitchen waste anaerobic digestion solution can serve as a high-quality substitutive medium, promote biomass yield and lower the culture cost and is worthy of popularization.



## NOTICIAS

### **Osaka City University Develops CO2 Consuming Biofuel Cell Utilizing Microalgae Spirulina**

[https://www.japanfs.org/en/news/archives/news\\_id036122.html](https://www.japanfs.org/en/news/archives/news_id036122.html)

Osaka City University announced on April 25, 2018, that it succeeded in developing a new biofuel cell system with the functions of a solar cell and the ability of carbon dioxide conversion. Utilizing the photosynthesis function of spirulina, a biomass of dark-green unicellular microalgae, this solar-light driven biofuel cell generates electricity by consuming CO2 in water as source material and produces formic acid as a byproduct while generating electricity.

### **Israeli biotech company grows unique microalgae to treat fatty liver**

[http://www.xinhuanet.com/english/2018-07/10/c\\_137312870.htm](http://www.xinhuanet.com/english/2018-07/10/c_137312870.htm)

JERUSALEM, July 9 (Xinhua) -- Israeli biotechnology company Algatech, which grows and processes special species of microalgae, has developed a new product for the treatment of fatty liver disease.

## EVENTOS

VII Congreso Nacional de Acuicultura.

Arica, Chile

Septiembre 11-14, 2018.

18° Conferencia Internacional sobre algas nocivas (ICHA 2018)

Nantes, Francia

Octubre 21-26, 2018

Algae Biomass Summit.

The Woodlands Waterway Marriott Hotel & Convention Center

The Woodlands (Greater Houston), Texas

October 14 - October 17, 2018

XI Congreso de Ciencias del Mar MarCuba 2018.

15-19 de octubre. La Habana, Cuba

ALGAE EUROPE Conference

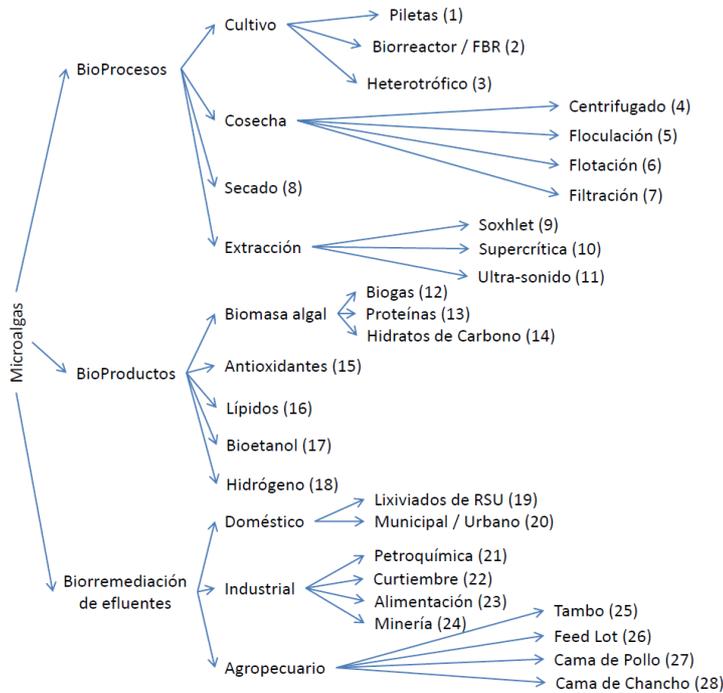
December 4-6, 2018

Amsterdam, the Netherlands.

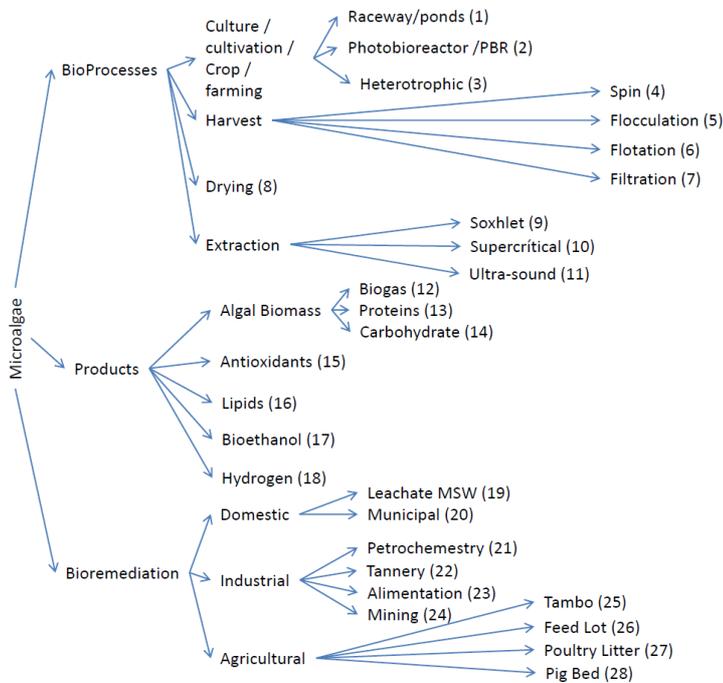


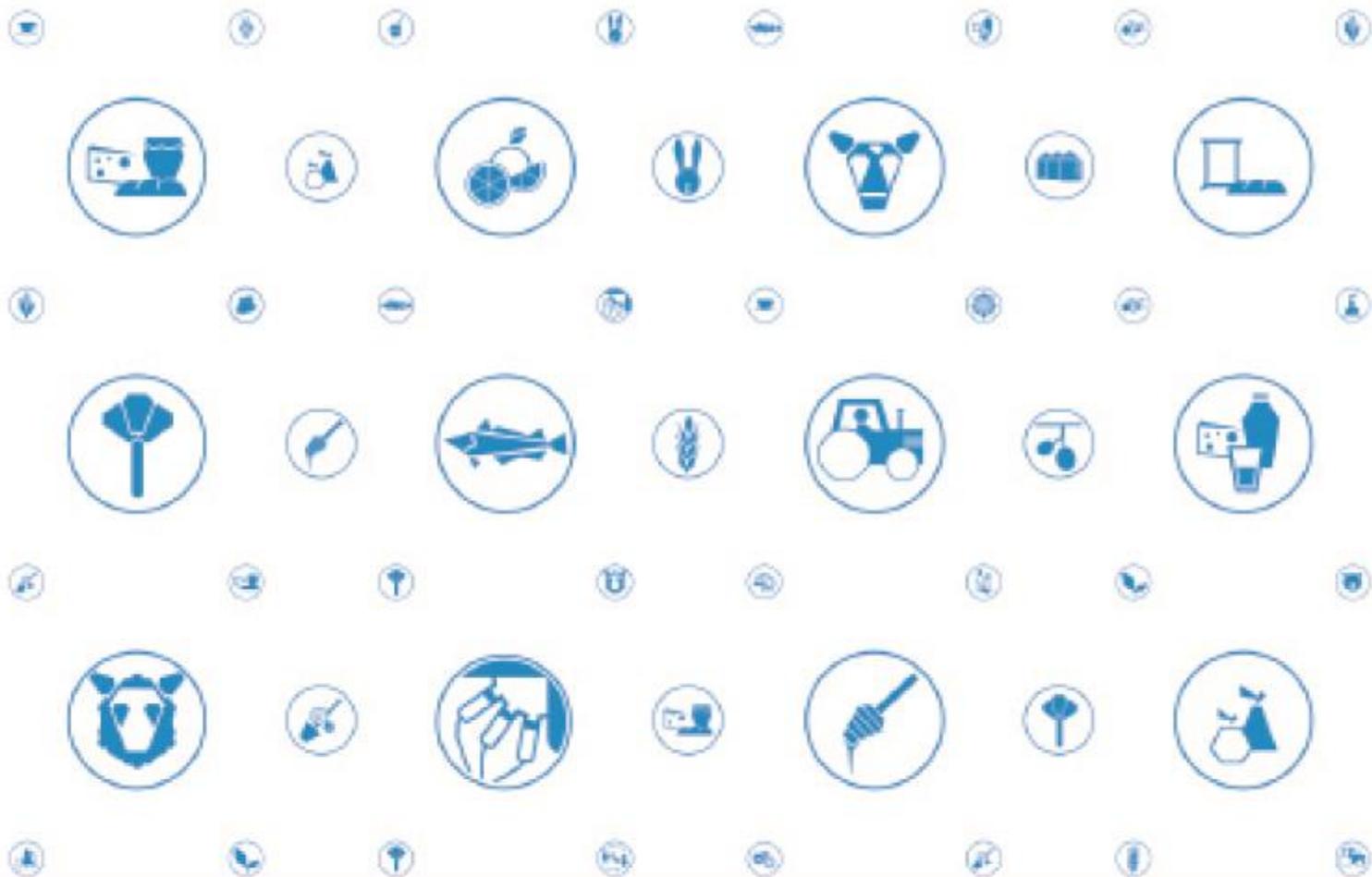
## Árbol de categorías

Español



Inglés





# TITULO

## SUBTITULO

