NEW GREEN ALTERNATIVES FOR THE EXTRACTION OF BIOACTIVES FROM NATURAL SOURCES

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CIAL

CSIC
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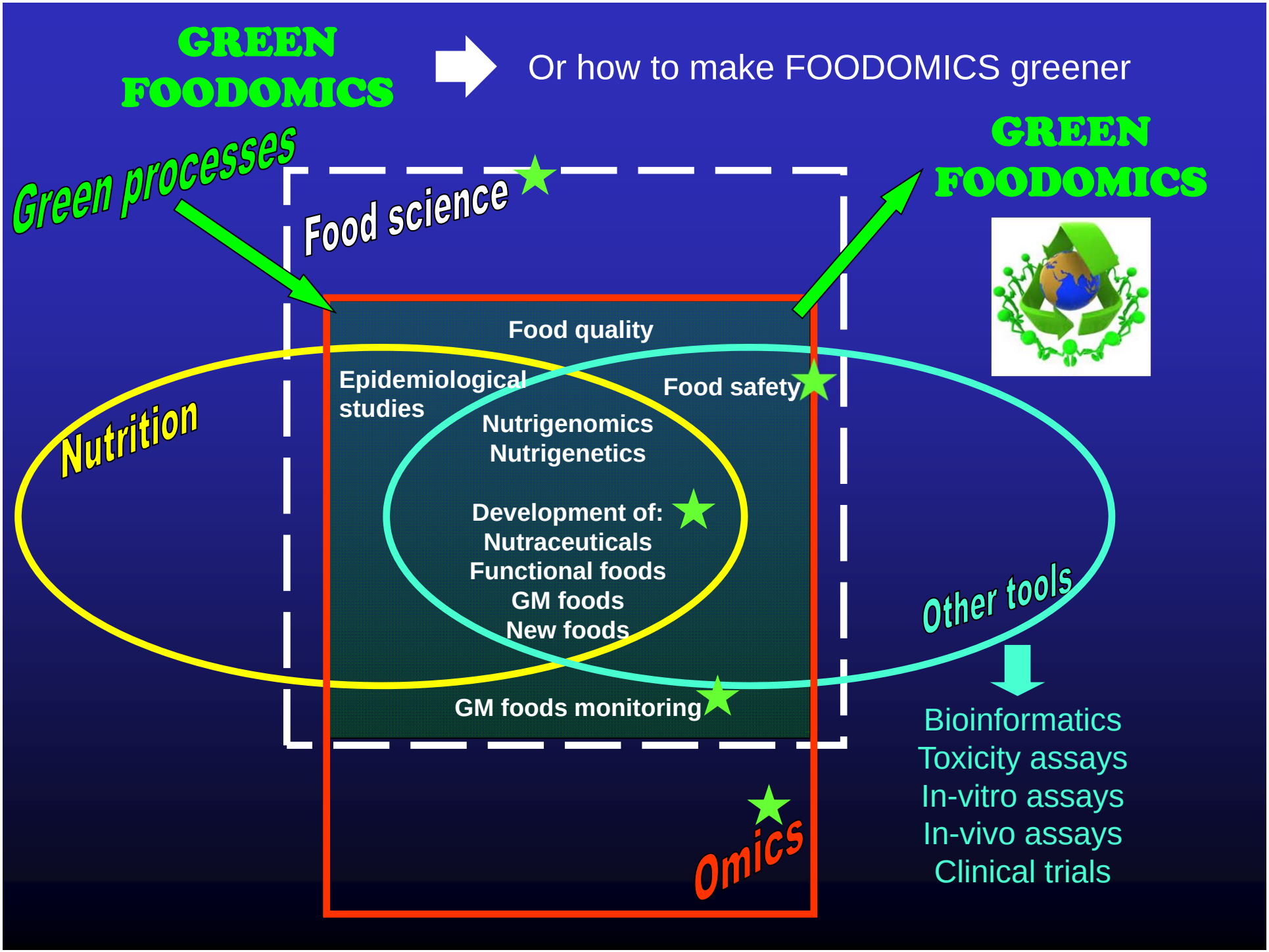
http://www.cial.uam-csic.es/pagperso/foodomics


GREEN FOODOMICS
Or how to make FOODOMICS greener
GREEN FOODOMICS
Green processes
Food science
Food quality
Epidemiological studies
Food safety
Nutrition
Nutrigenomics
Nutrigenetics
Development of:
Nutraceuticals
Functional foods
GM foods
New foods
Other tools
GM foods monitoring
Omics
Bioinformatics
Toxicity assays
In-vitro assays
In-vivo assays
Clinical trials

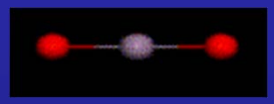
CHALLENGES IN THE DEVELOPMENT OF GREEN EXTRACTION PROCESSES

FAST

SELECTIVE

GREEN

COST-EFFECTIVE



HIGH YIELDS

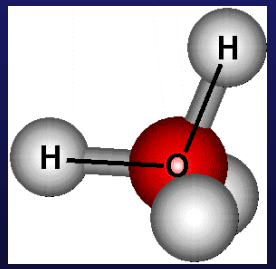
SUSTAINABLE

EFFICIENT

ECOLOGICAL

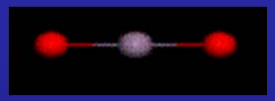
SUSTAINABLE

Without organic solvents

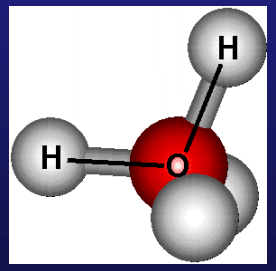


CHALLENGE

EXTRACTION BIOACTIVE COMPOUNDS FROM NATURAL SOURCES THROUGH GREEN PROCESSES

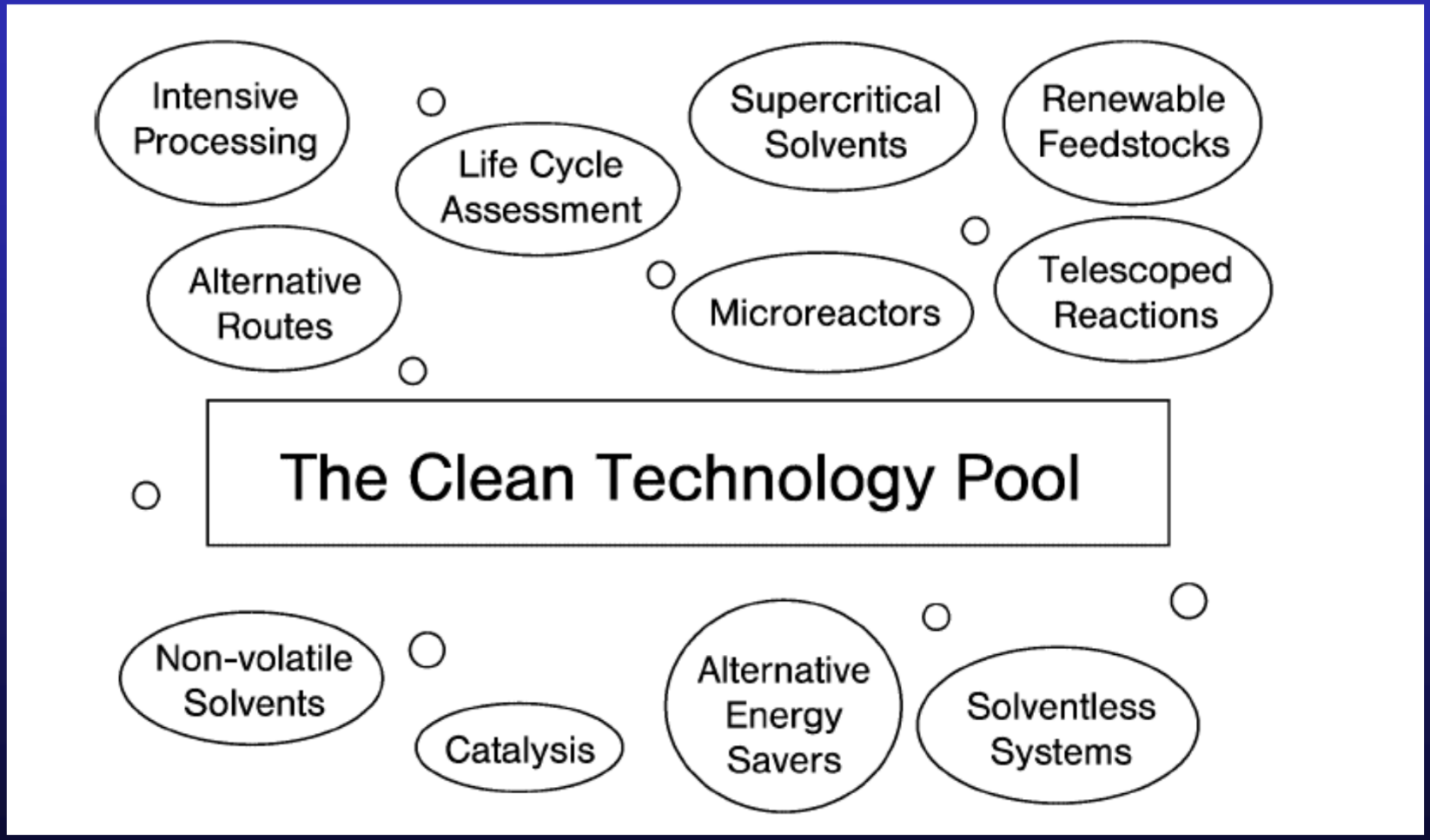


INTENSIFICATION OF PROCESSES

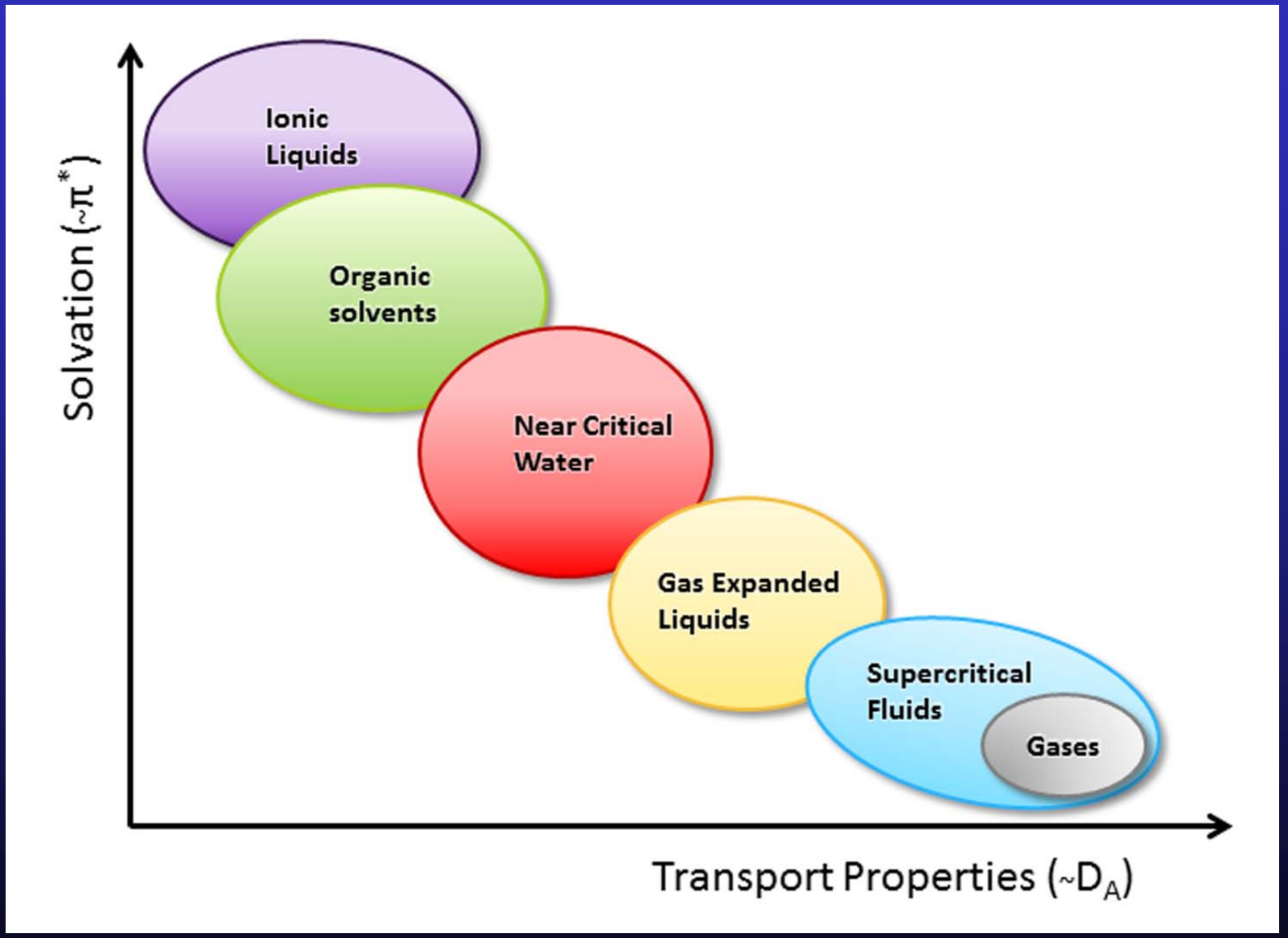


PRESSURE
TEMPERATURE
MICROWAVES
ENZYMES

Environmentally Friendly Technologies



SOLVENTS FOR SUSTAINABLE CHEMICAL PROCESSES



Green Chem, 2014, 16, 1034

SOME EXAMPLES OF NATURAL SOURCES AND GREEN PROCESSES STUDIED AT FOODOMICS LAB

J. of Supercritical Fluids 100 (2015) 86–96

Contents lists available at ScienceDirect

The Journal of Supercritical Fluids

journal homepage: www.elsevier.com/locate/supflu

Copaifera langsdorffii supercritical fluid extraction: Chemical and functional characterization by LC/MS and in vitro assays

Ana R.M. Costa[a,b], Luís A.P. Freitas[a], J. Mendiola[b], E. Ibáñez[b,*]

[a] Faculdade de Ciências Farmacêuticas de Ribeirão Preto, Núcleo de Apoio à Pesquisa em Produtos Naturais e Sintéticos, Universidade de São Paulo, Via do Café, s/n, Campus USP, Ribeirão Preto 14040-903, SP, Brazil
[b] Bioactivity and Food Analysis Department, Food Research Institute (CIAL-CSIC), Campus de Cantoblanco, Calle Nicolás Cabrera, 9 Cantoblanco, 28049 Madrid, Spain

Journal of Chromatography A, 1288 (2013) 1–9

Contents lists available at SciVerse ScienceDirect

Journal of Chromatography A

journal homepage: www.elsevier.com/locate/chroma

Enrichment of antioxidant compounds from lemon balm (*Melissa officinalis*) by pressurized liquid extraction and enzyme-assisted extraction

T.L. Miron[a], M. Herrero[b,*], E. Ibáñez[b]

[a] Faculty of Food Science and Engineering, "Dunărea de Jos" University, Domnească 111, 800201 Galaţi, Romania
[b] Laboratory of Foodomics, Institute of Food Science Research (CIAL-CSIC), Nicolás Cabrera 9, Campus Cantoblanco, 28049 Madrid, Spain

G Model
SUPFLU-3370; No. of Pages 9

ARTICLE IN PRESS

J. of Supercritical Fluids xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

The Journal of Supercritical Fluids

journal homepage: www.elsevier.com/locate/supflu

Optimization of rutin isolation from *Amaranthus paniculatus* leaves by high pressure extraction and fractionation techniques

Paulius Kraujalis[a], Petras Rimantas Venskutonis[a], Elena Ibáñez[b,*], Miguel Herrero[b]

[a] Kaunas University of Technology, Department of Food Science and Technology, Radvilėnų pl. 19, LT-50254, Kaunas, Lithuania
[b] Foodomics Laboratory, Institute of Food Science Research (CIAL, CSIC), Nicolás Cabrera 9, Campus UAM Cantoblanco, 28049 Madrid, Spain

Amaranthus paniculatus

SOME EXAMPLES OF NATURAL SOURCES AND GREEN PROCESSES STUDIED AT FOODOMICS LAB

JOURNAL OF FUNCTIONAL FOODS 11 (2014) 293–303

Available at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/jff

Two-step sequential supercritical fluid extracts from rosemary with enhanced anti-proliferative activity

Andrea del Pilar Sánchez-Camargo [a], *Alberto Valdés* [a], *Giuseppe Sullini* [a,b], *Virginia García-Cañas* [a], *Alejandro Cifuentes* [a], *Elena Ibáñez* [a,*], *Miguel Herrero* [a]

[a] *Laboratory of Foodomics, Institute of Food Science Research (CIAL, CSIC), Calle Nicolas Cabrera 9, Madrid 28049, Spain*

[b] *Dipart. di Scienze del Farmaco e dei Prodotti per la Salute (S.C.I.F.A.R.), University of Messina, Viale Annunziata, Messina 98168, Italy*

JOURNAL OF FUNCTIONAL FOODS 15 (2015) 429–439

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/jff

Rosemary polyphenols induce unfolded protein response and changes in cholesterol metabolism in colon cancer cells

Alberto Valdés, Giuseppe Sullini, Elena Ibáñez, Alejandro Cifuentes, Virginia García-Cañas *

Laboratory of Foodomics, Institute of Food Science Research (CIAL, CSIC), Calle Nicolás Cabrera 9, Madrid 28049, Spain

G Model

ARTICLE IN PRESS

J. of Supercritical Fluids xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

The Journal of Supercritical Fluids

journal homepage: www.elsevier.com/locate/supflu

Supercritical antisolvent fractionation of rosemary extracts obtained by pressurized liquid extraction to enhance their antiproliferative activity

A.P. Sanchez-Camargo, J.A. Mendiola, A. Valdés, M. Castro-Puyana, V. García-Cañas, A. Cifuentes, M. Herrero, E. Ibáñez*

J. of Supercritical Fluids xxx (2015) xxx–xxx

SOME EXAMPLES OF NATURAL SOURCES AND GREEN PROCESSES STUDIED AT FOODOMICS LAB

Food and Chemical Toxicology 50 (2012) 1817–1825

Contents lists available at SciVerse ScienceDirect

Food and Chemical Toxicology

journal homepage: www.elsevier.com/locate/foodchemtox

Use of advanced techniques for the extraction of phenolic compounds from Tunisian olive leaves: Phenolic composition and cytotoxicity against human breast cancer cells

Amani Taamalli [a,b,c], David Arráez-Román [b,c], Enrique Barrajón-Catalán [e], Verónica Ruiz-Torres [e], Almudena Pérez-Sánchez [e], Miguel Herrero [d], Elena Ibañez [d], Vicente Micol [e], Mokhtar Zarrouk [a], Antonio Segura-Carretero [b,c,*], Alberto Fernández-Gutiérrez [b,c]

[a] Laboratoire Caractérisation et Qualité de l'Huile d'Olive, Centre de Biotechnologie de Borj Cedria, BP 901, 2050 Hammam-Lif, Tunisia
[b] Department of Analytical Chemistry, Faculty of Sciences, University of Granada, Granada 18071, Spain
[c] Research and Development of Functional Food Centre (CIDAF), Health Science Technological Park, Avda. DelConocimiento s/n, 18100 Granada, Spain
[d] Department of Bioactivity and Food Analysis, Institute of Food Science Research-CIAL (CSIC-UAM), Nicolás Cabrera 9, Campus Cantoblanco, 28049 Madrid, Spain
[e] Institute of Molecular and Cell Biology, University Miguel Hernández, Avda. Universidad s/n, 03202 Elche, Spain

Int. J. Mol. Sci. **2014**, *15*, 16270-16283; doi:10.3390/ijms150916270

OPEN ACCESS

International Journal of

Molecular Sciences

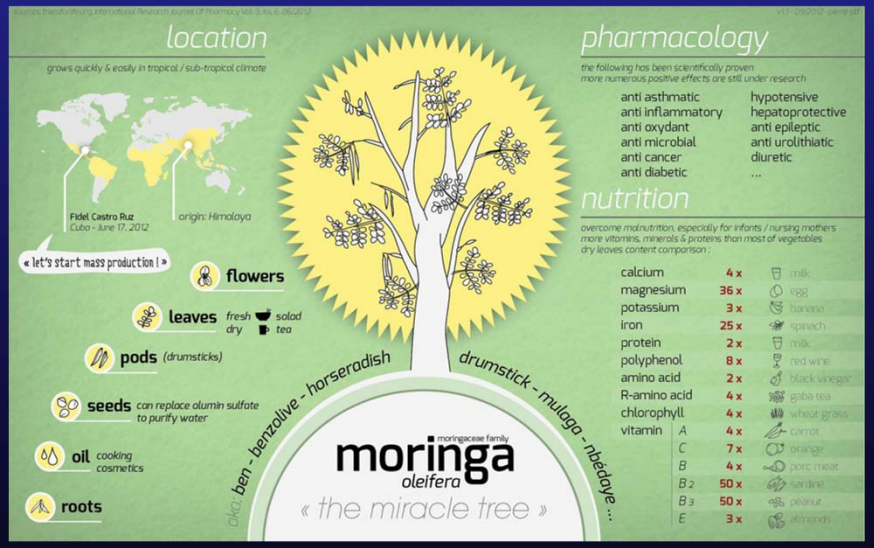
ISSN 1422-0067

www.mdpi.com/journal/ijms

Article

Recovering Bioactive Compounds from Olive Oil Filter Cake by Advanced Extraction Techniques

Jesús Lozano-Sánchez [1,2,†], María Castro-Puyana [3,4,†], Jose A. Mendiola [3], Antonio Segura-Carretero [2], Alejandro Cifuentes [3,*] and Elena Ibáñez [3]



PROYECTO DE TESIS DOCTORAL EN EL EXTRANJERO

Alumna: Simone Mazzutti

Coordinador: Dra. Sandra R. S. Ferreira

Universidad de destino: Instituto de Investigación en Ciencias de la Alimentación

Orientadora en el extranjero: Dra. Maria Elena Ibáñez Ezequiel

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1 TÍTULO:

Obtención de extractos bioactivos de *Plantago major* y *Plantago lanceolata* con actividad antioxidante mediante el empleo de extracción con agua subcrítica

ALGAE- WHY?

- Thousands of species, some unkown or unexplored, with a great diversity
- Algae live in complex habitats submitted to extreme conditions (salinity, nutrients, radiation, etc.), they must adapt rapidly to the new environmental conditions to survive, producing a great variety of secondary (biologically active) metabolites, with structures that cannot be found in other organisms
- Possibility of collecting (harvesting) different species throughout the year and depending on the geographic position.
- Important natural reactors: can be used at large scale to produce active compounds.
- Economic Importance
- Biorefinery concept

COMPARISON BETWEEN PRESURIZED LIQUID EXTRACTION AND MICROWAVE ASSISTED EXTRACTION OF BIOACTIVE COMPOUNDS FROM THE MICROALGA *PHAEODACTYLUM TRICORNUTUM.*

B. Gilbert-López, A. Barranco, M. Herrero, A. Cifuentes, E. Ibáñez

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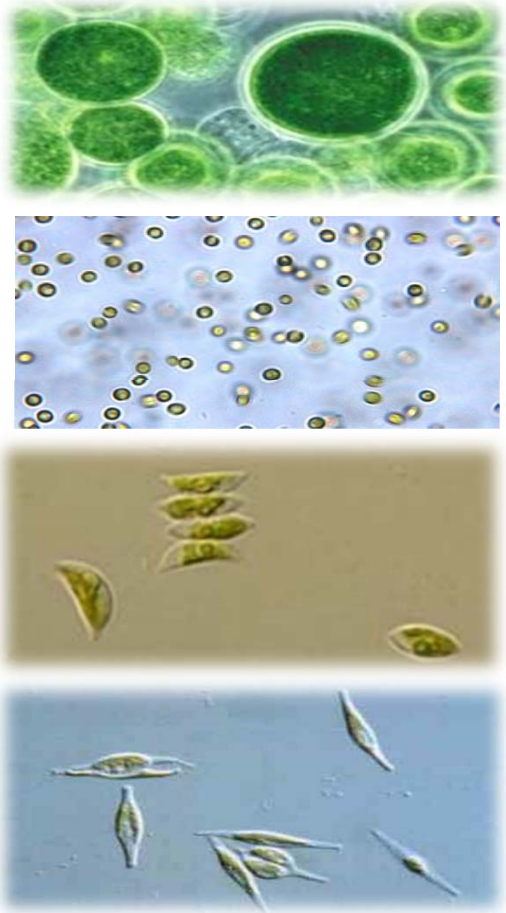
http://www.cial.uam-csic.es/pagperso/foodomics/

http://miraclesproject.eu/

EU project **http://miraclesproject.eu/**

Multi-product **I**ntegrated bio**R**efinery of **A**lgae: from **C**arbon dioxide and **L**ight **E**nergy to high-value **S**pecialties: **MIRACLES**



SEVENTH FRAMEWORK PROGRAMME

MIRACLES
SPECIALTIES FROM ALGAE

fitoplancton marino

MIRACLES
SPECIALTIES FROM ALGAE

***P. tricornutum* culture**

↓

High pressure treatment (1200 bar) to break the cell wall

↓

Freeze-drying

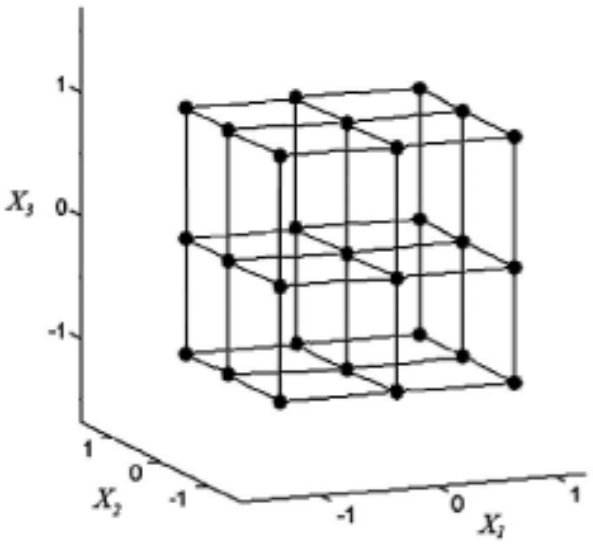
↙ ↘

Microwave Solvent-Assisted Extraction (MAE)

Pressurized Liquid Extraction (PLE)

Microwave Solvent-Assisted Extraction (MAE)

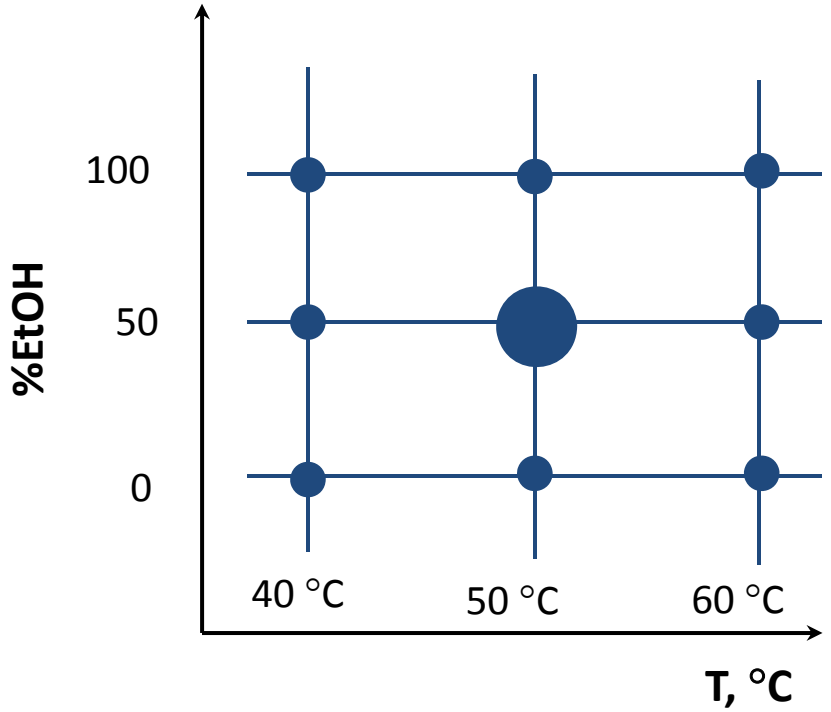
Factorial 3^3
4 replicates in central point



Factor	*Range*	
%EtOH	0-100	**%**
Temp	30-170	**°C**
Time	2-20	**min**

Pressurized Liquid Extraction (PLE)

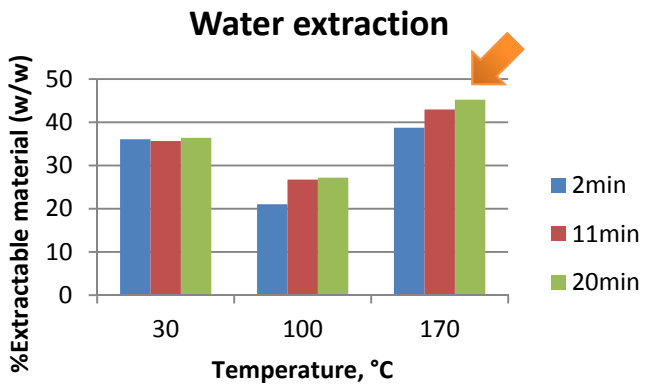
Factorial 3^2
4 replicates in central point

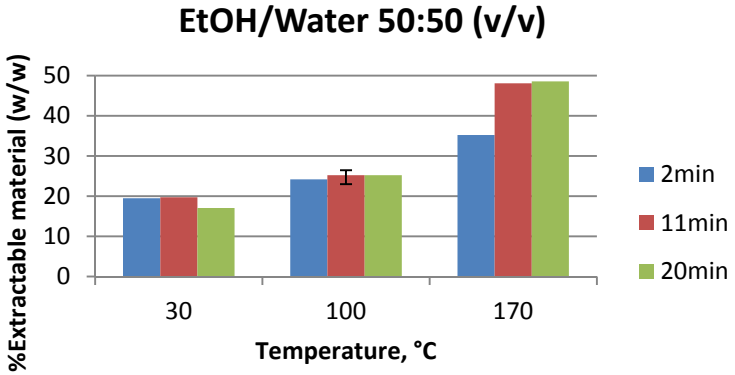


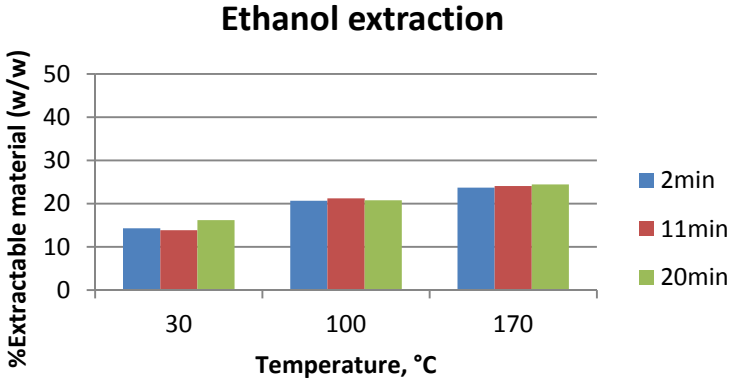
Factor	*Range*	
%EtOH	0-100	**%**
Temp	50-170	**°C**

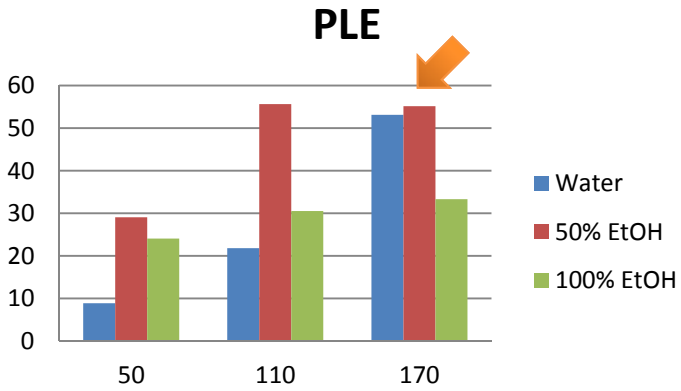
MAE

Yield









Experimental Design PLE 3^2

Optimum values proposed:

Extraction time:	**20 min**
Temperature:	**170 ºC**
%EtOH:	**40%**

Yield: 61.8975 %(w/w)

Experimental Design MAE 3^3

Optimum values proposed:

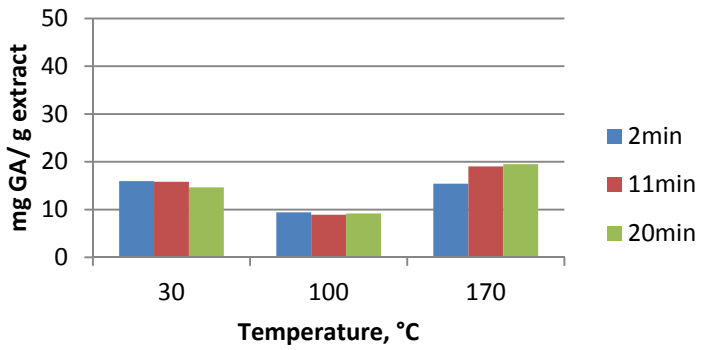
Extraction time:	**20 min**
Temperature:	**170 ºC**
%EtOH:	**0%**

Yield: 46.5207 %(w/w)

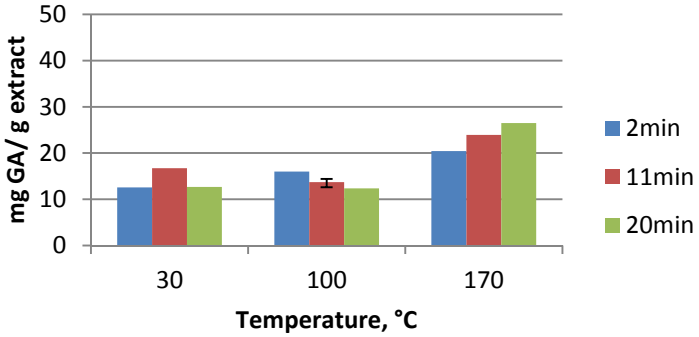
TPC

MAE

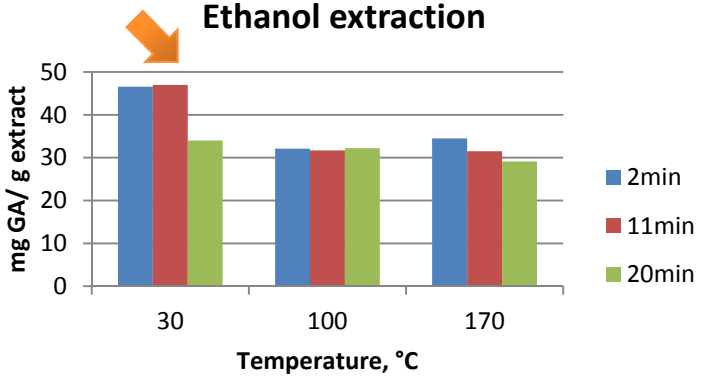
Water extraction



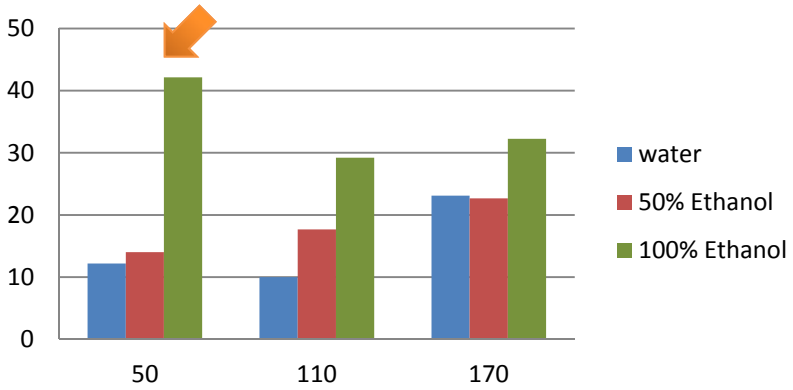
EtOH/Water 50:50 (v/v)



Ethanol extraction



PLE



Experimental Design PLE 3^2

Optimum values proposed:

Extraction time:	**20 min**
Temperature:	**50 ºC**
%EtOH:	**100%**

TPC: 39.5521 mg GA/ g extract

Experimental Design MAE 3^3

Optimum values proposed:

Extraction time:	**2 min**
Temperature:	**30 ºC**
%EtOH:	**100%**

TPC: 44.037 mg GA/ g extract

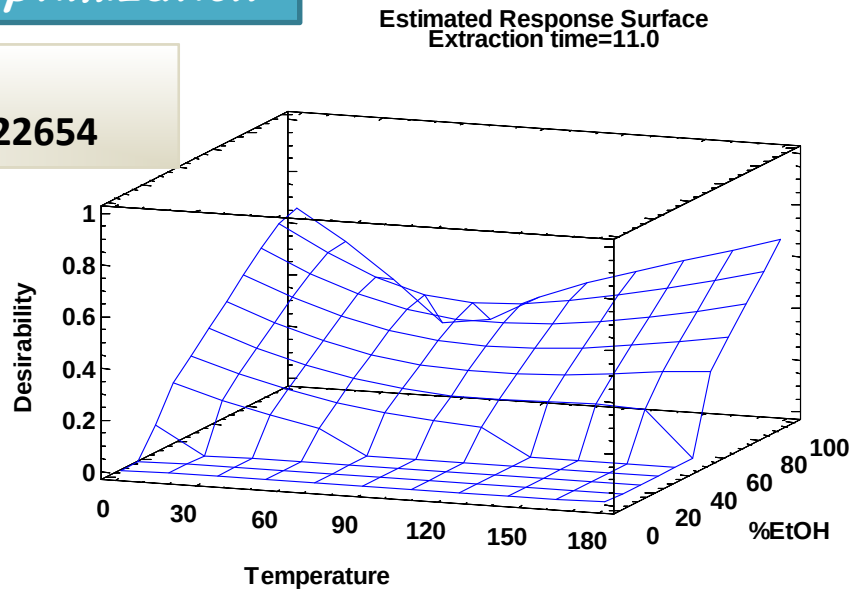
Multiple response optimization

Optimum values proposed:

MAE
Desirability: 0.622654

Extraction time: 5.8 min
Temperature: 170 ºC
%EtOH: 100%

Yield: 28.5715 %(w/w)
TPC: 35.1811 mg GA/ g extract
TEAC: 0.579866 mmol Trolox/g extract
Carotenoids: 24.3037 mg/ g extract

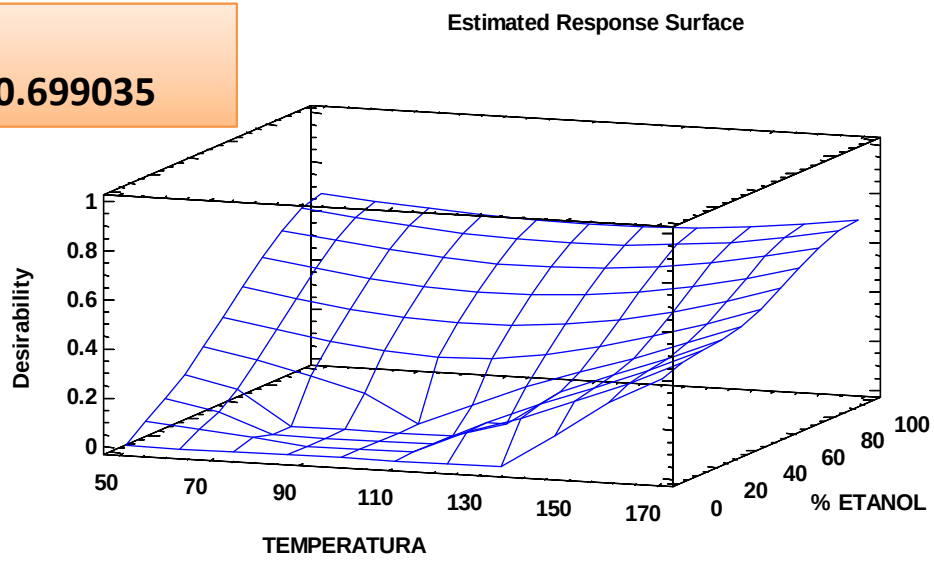


Optimum values proposed:

PLE
Desirability: 0.699035

Extraction time (fixed): 20 min
Temperature: 170 ºC
%EtOH: 97.7%

Yield: 34.357 %(w/w)
TPC: 31.6772 mg GA/ g extract
TEAC: 0.522114 mmol Trolox/g extract
Carotenoids: 30.9267 mg/ g extract



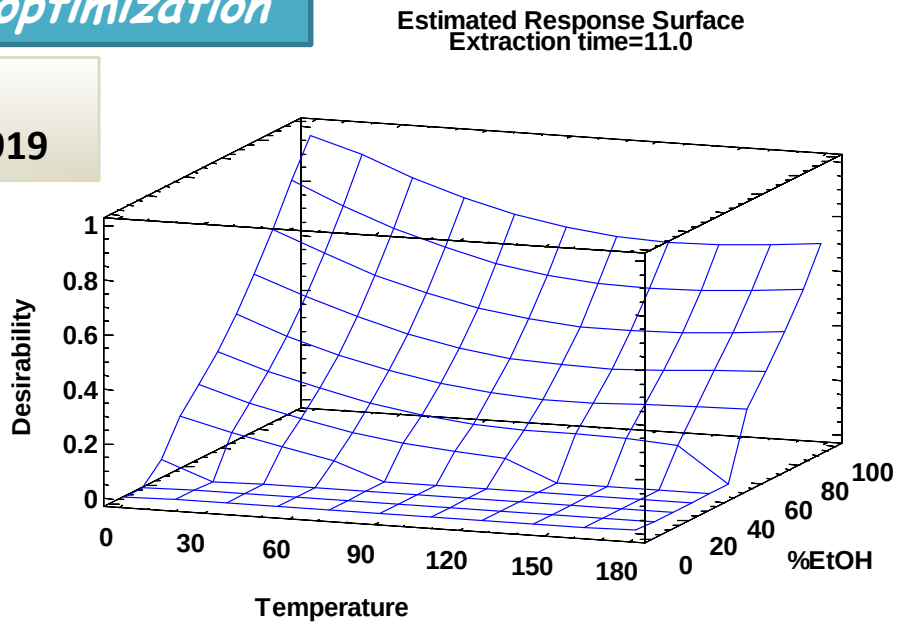
Multiple response optimization

Optimum values proposed:

MAE
Desirability: 0.940919

Extraction time: 2 min
Temperature: 30 ºC
%EtOH: 100%

Yield not considered
TPC: 44.037 mg GA/ g extract
TEAC: 0.5705 mmol Trolox/g extract
Carotenoids: 41.4523 mg/ g extract
Chlorophylls: 54.5781 mg/ g extract

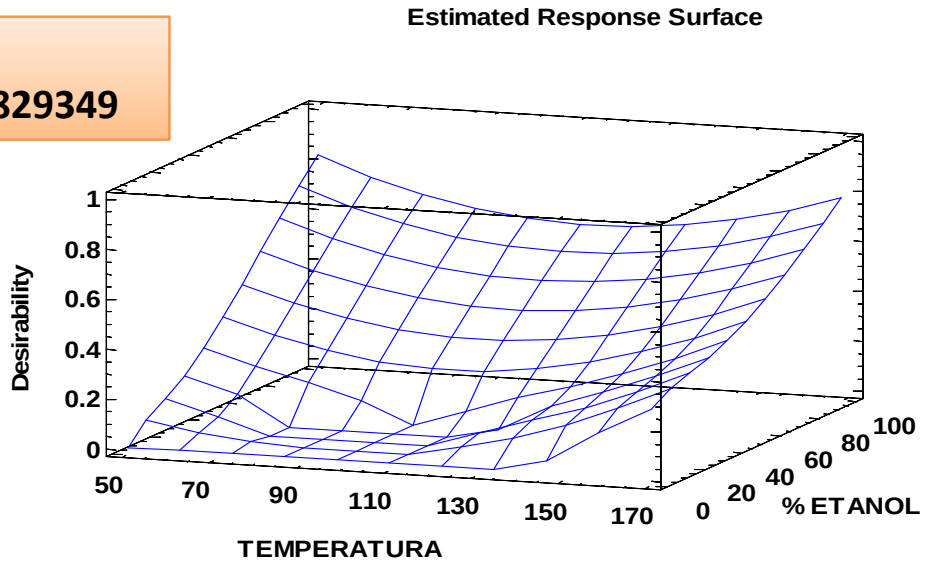


Optimum values proposed:

PLE
Desirability: 0.829349

Extraction time (fixed): 20 min
Temperature: 50 ºC
%EtOH: 100%

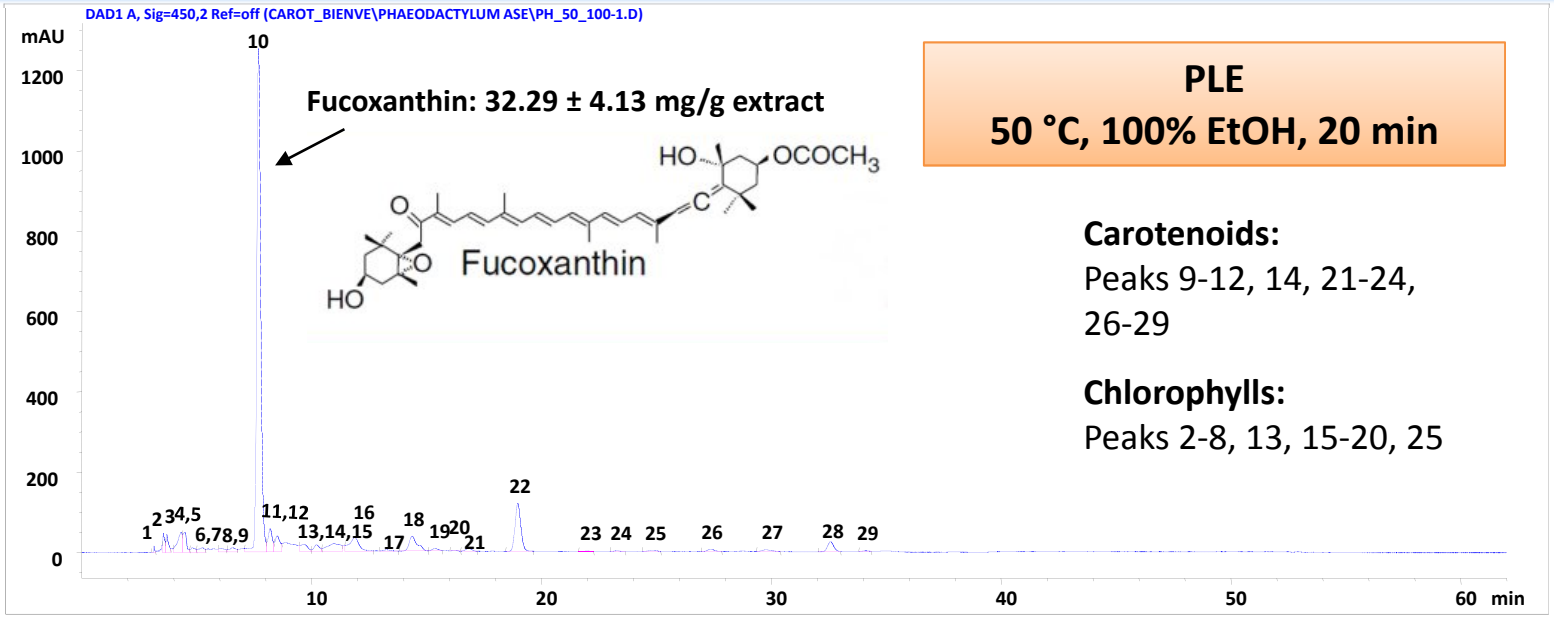
Yield not considered
TPC: 39.5521 mg GA/ g extract
TEAC: 0.591292 mmol Trolox/g extract
Carotenoids: 30.8172 mg/ g extract
Chlorophylls: 42.4847 mg/ g extract



HPLC-DAD CAROTENOIDS AND CHLOROPHYLLS PROFILE

MAE vs. PLE

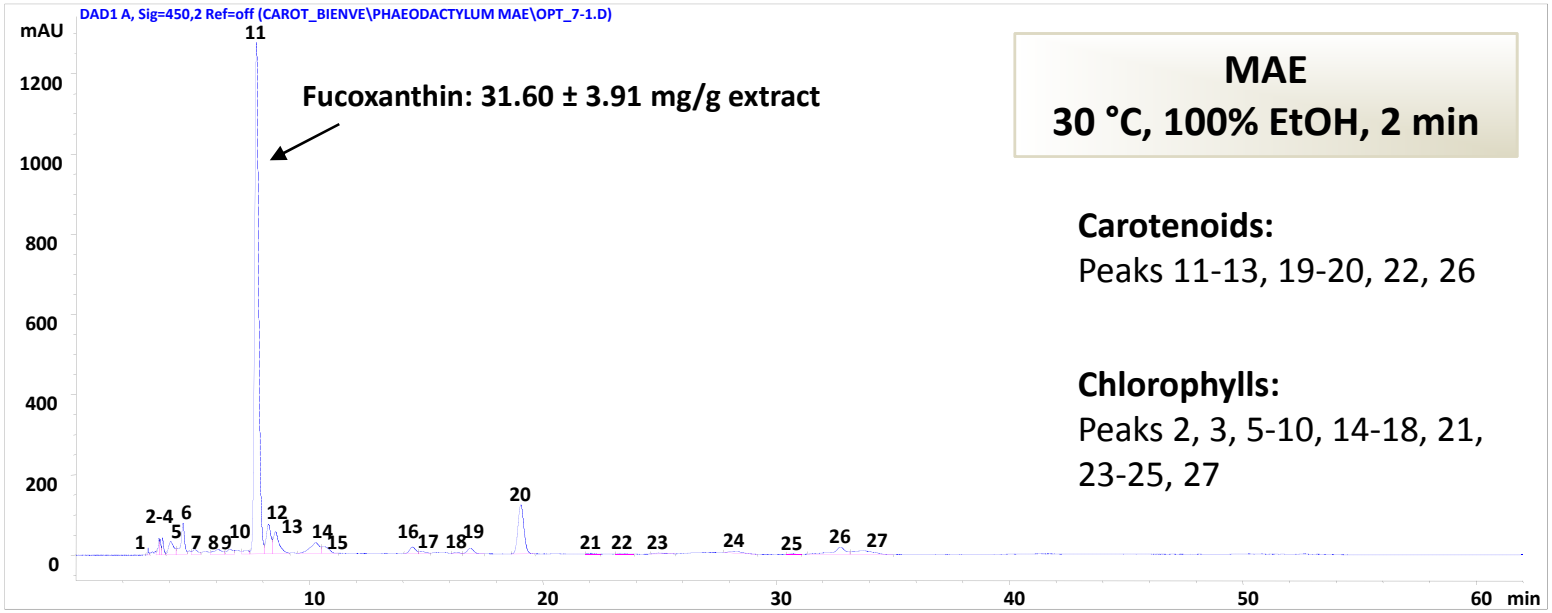
P. tricornutum



PLE
50 °C, 100% EtOH, 20 min

Carotenoids:
Peaks 9-12, 14, 21-24, 26-29

Chlorophylls:
Peaks 2-8, 13, 15-20, 25



MAE
30 °C, 100% EtOH, 2 min

Carotenoids:
Peaks 11-13, 19-20, 22, 26

Chlorophylls:
Peaks 2, 3, 5-10, 14-18, 21, 23-25, 27

Refs: M. Castro-Puyana et al., *Anal. Bioanal. Chem.* (2013), 405, 4607; B. Gilbert-López et al., *Green Chemistry* (2015), 17, 4599-4609

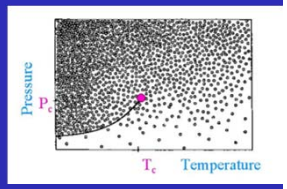
P. tricornutum - MAE vs. PLE

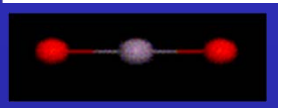
MAE	PLE
Extraction time: 6 min Temperature: 170 °C %EtOH: 100% Yield: 22.20 % TPC: 30.50 mg GAE/g extract TEAC: 0.68 mmol Trolox/g extract Carotenoids: 24.79 mg/g extract Chlorophylls: 52.94 mg/g extract Fucoxanthin: 13.38 ± 0.54 mg/g extract	Extraction time (fixed): 20 min Temperature: 170 °C %EtOH: 97% Yield: 38.78 % TPC: 30.98 TEAC: 0.54 Carotenoids: 16.99 Chlorophylls: 37.84 Fucoxanthin: 14.99 ± 0.56 mg/g extract
Extraction time: 2 min Temperature: 30 °C %EtOH: 100% Yield: 14.51 % TPC: 44.19 TEAC: 0.71 Carotenoids: 41.42 Chlorophylls: 58.61 Fucoxanthin: 31.60 ± 3.91 mg/g extract	Extraction time (fixed): 20 min Temperature: 50 °C %EtOH: 100% Yield: 23.95 % TPC: 38.15 TEAC: 0.59 Carotenoids: 33.96 Chlorophylls: 54.36 Fucoxanthin: 32.29 ± 4.13 mg/g extract

CONCLUDING REMARKS

- ✓ **Similar results** are obtained using either **MAE** or **PLE**.
- ✓ **Ethanol is the best solvent** to obtain extracts with antioxidant activity and rich in fucoxanthin.
- ✓ **High temperature** to increase extraction yield lead to **1/2 concentration** of **fucoxanthin** compared to the extraction at low temperature.

STEP TO THE FUTURE





GREEN PROCESSING PLATFORM

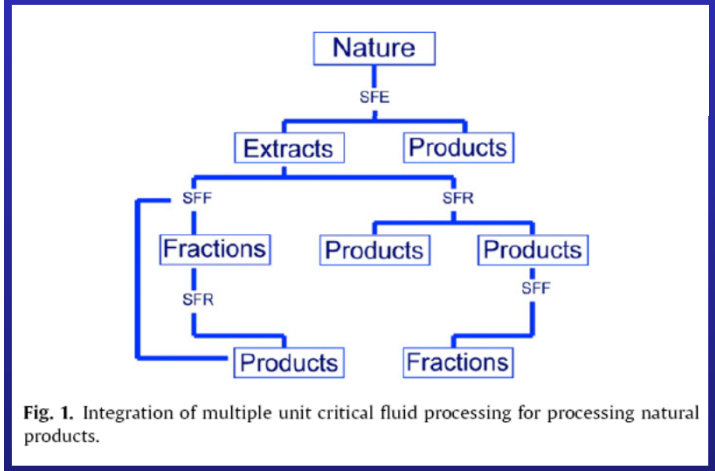
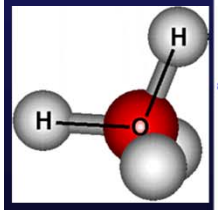
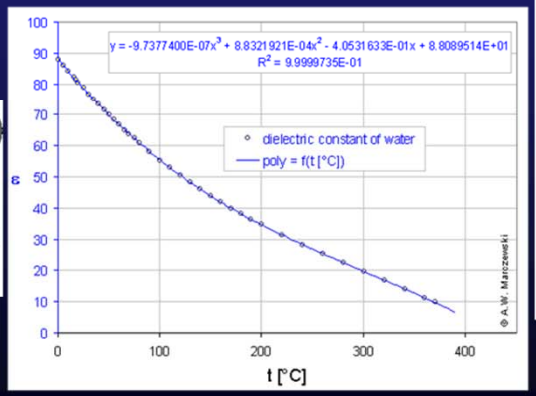
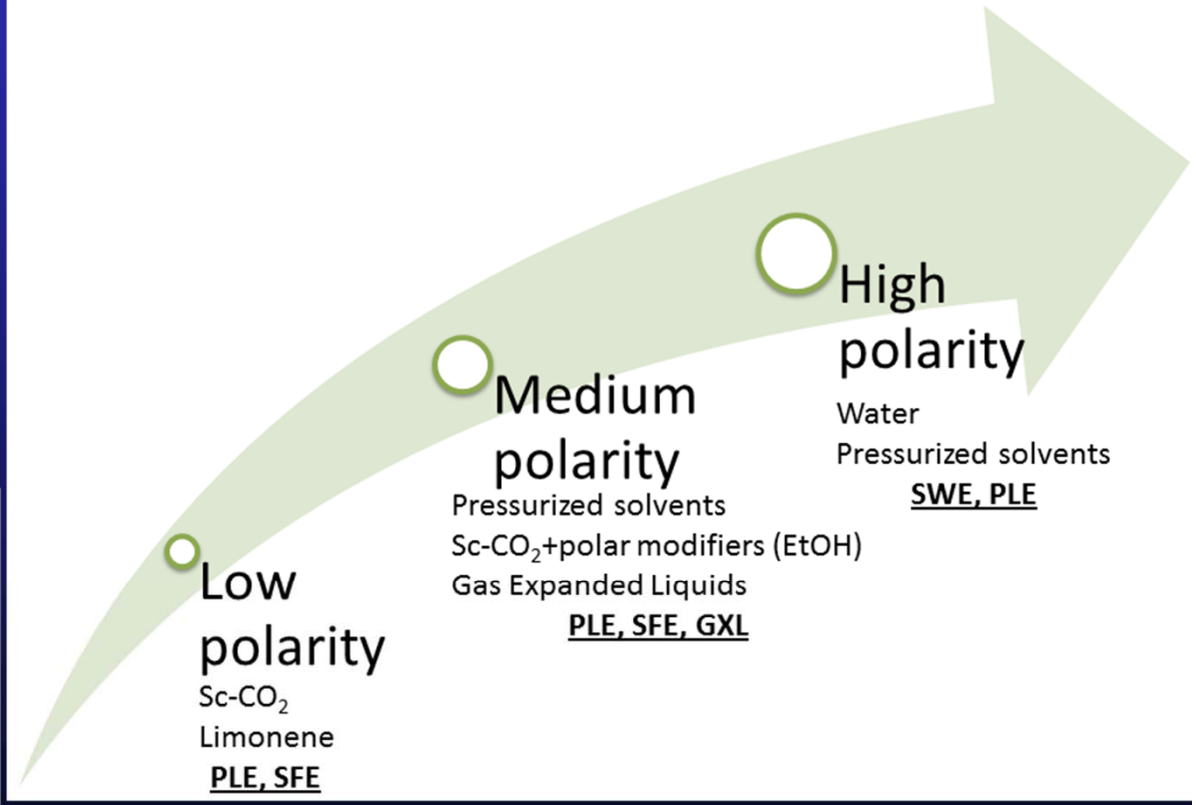


Fig. 1. Integration of multiple unit critical fluid processing for processing natural products.

JerryW. King, Keerthi Srinivas, J. of Supercritical Fluids 47 (2009) 598–610

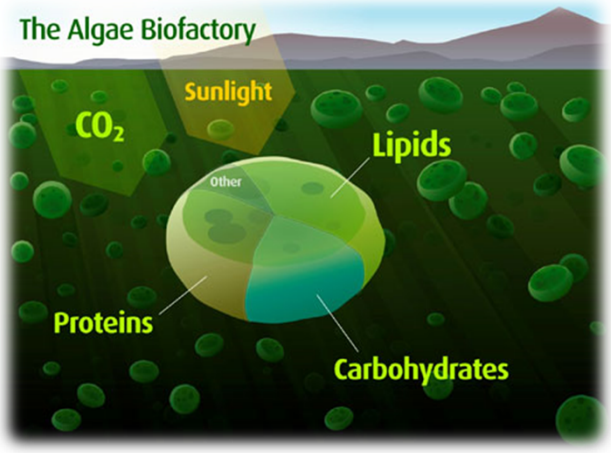






GREEN TECHNOLOGIES FOR THE SEQUENTIAL EXTRACTION OF BIOACTIVE COMPOUNDS FROM THE MICROALGA *ISOCHRYSIS GALBANA*.

B. Gilbert-López, J. A. Mendiola, M. Herrero, A. Cifuentes, E. Ibáñez



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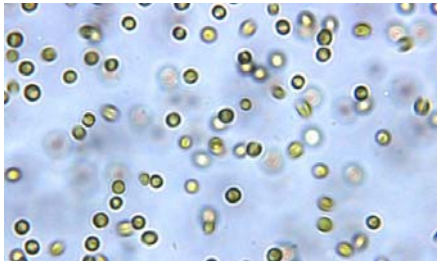
MIRACLES
SPECIALTIES FROM ALGAE



Isochrysis galbana microalga

• *Isochrysis galbana* is a marine flagellated microalga belonging to the phylum Haptophyta.

• Is of substantial interest in aquaculture, principally to feed mollusk larvae, as well as fish and crustaceans in the early stages of growth.

• Its cells are easily assimilated by larval animals because of their small size (4-7 µm) and absence of a tough cell wall.

• *Isochrysis galbana* is a marine microalga well recognized as a natural rich source of **EPA** and **DHA**.

• Other bioactive compounds: **carotenoids** (fucoxanthin) and **chlorophylls**.

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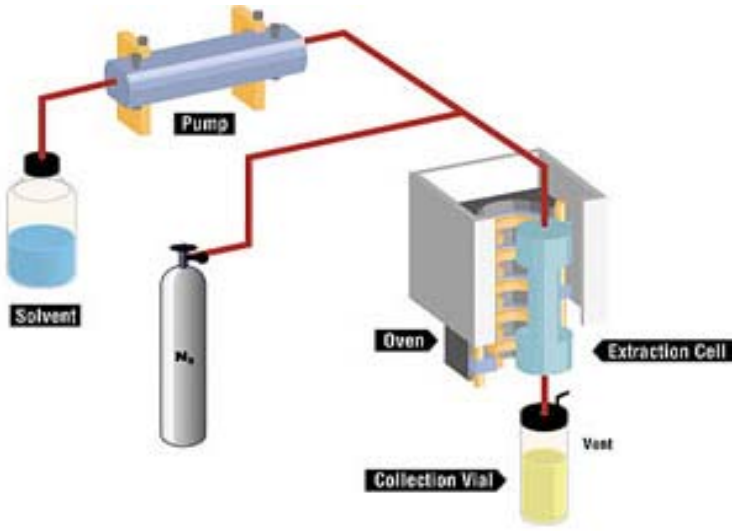
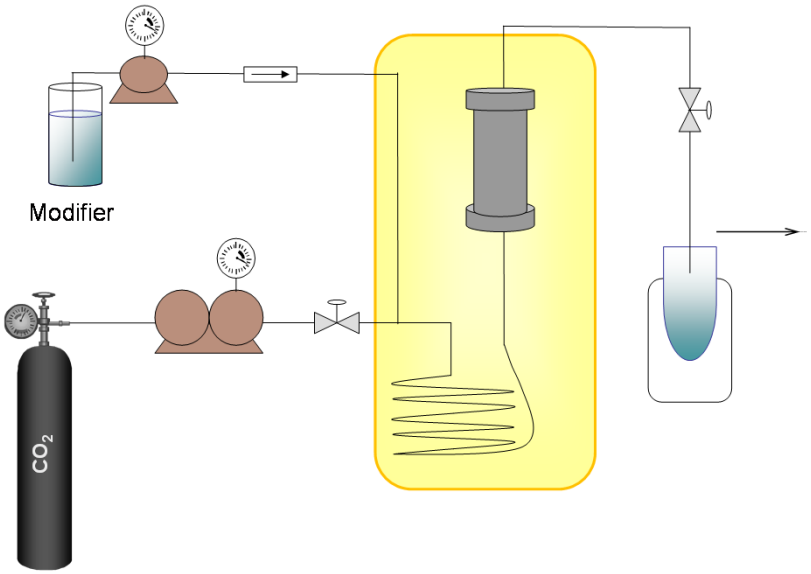
Extraction techniques used

PLE



- Solvents maintained in liquid state (high temperatures and pressures)
- Faster extraction processes
- Low volumes used of solvents

SFE



- CO_2 Extraction
- No oxidative damage
- Solvent-free product collected
- Products not in contact with toxic solvents

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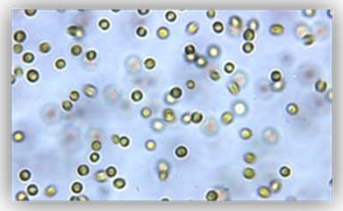


1 $scCO_2$ 300 bar, 50 °C

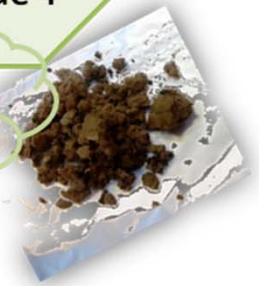
2 CXE $scCO_2$, 70 bar, 50 °C/ 15-45-75% EtOH

3 EtOH 100 bar, 80 °C

4 Water 100 bar, 80 °C

Isochrysis galbana → Residue 1 → Residue 2 → Residue 3 → Residue 4

Exhausted biomass



Extract 1

- Carotenoids
- Non-polar lipids

Extract 2

- Carotenoids
- Chlorophylls
- Mid and high-polar lipids

Extract 3

- Antioxidant activity
- Protein
- Sugars

Extract 4

Green Chemistry

PAPER

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Downstream processing of *Isochrysis galbana*: a step towards microalgal biorefinery†

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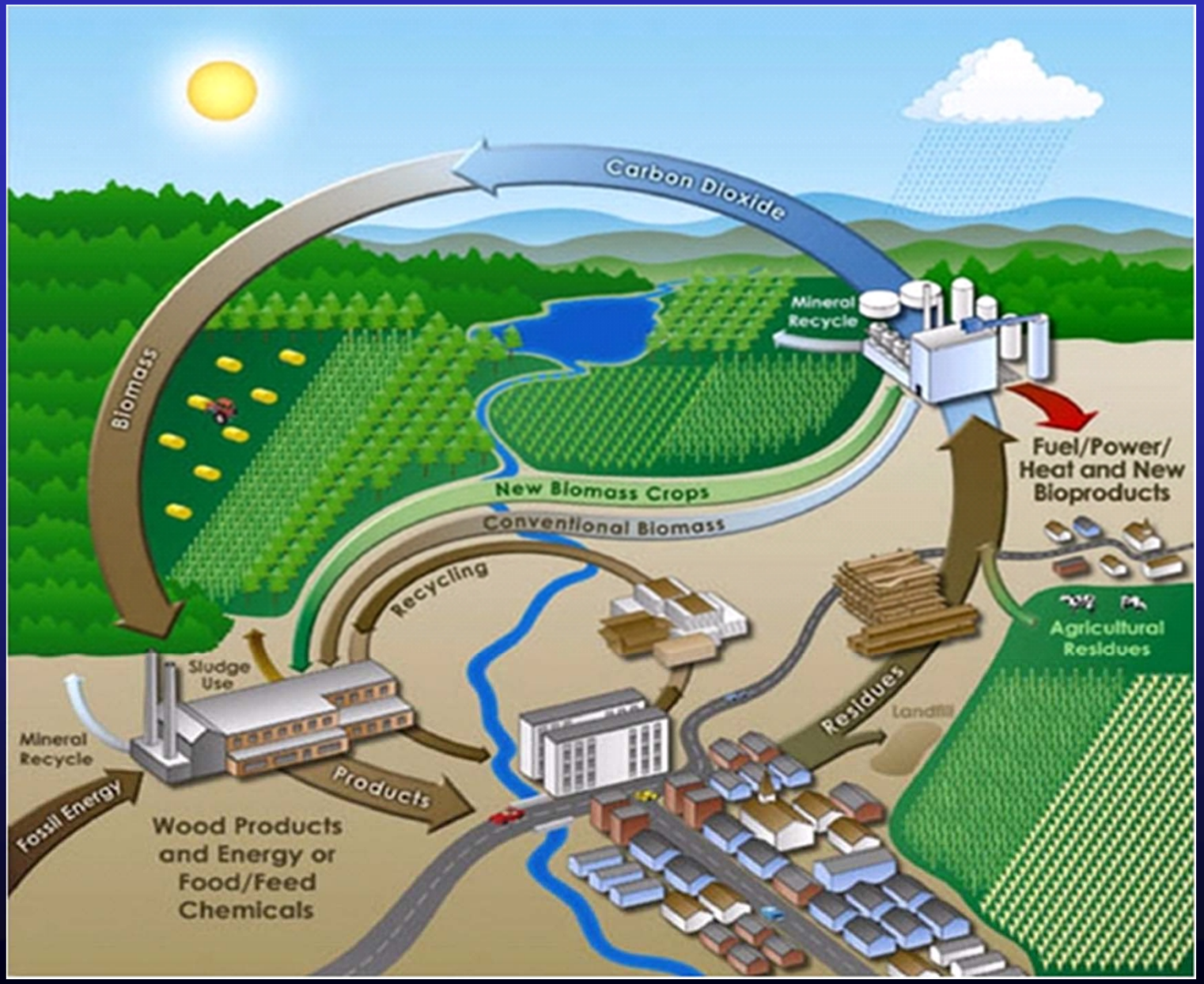
STEP TO THE FUTURE – BIOREFINERY CONCEPT



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GRACIAS!!!