Linking life and technology to shape the future

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Ohmic heating – a sustainable technology for the extraction of bioactive compounds.

José A. Teixeira

CEB - Centre of Biological Engineering

University of Minho

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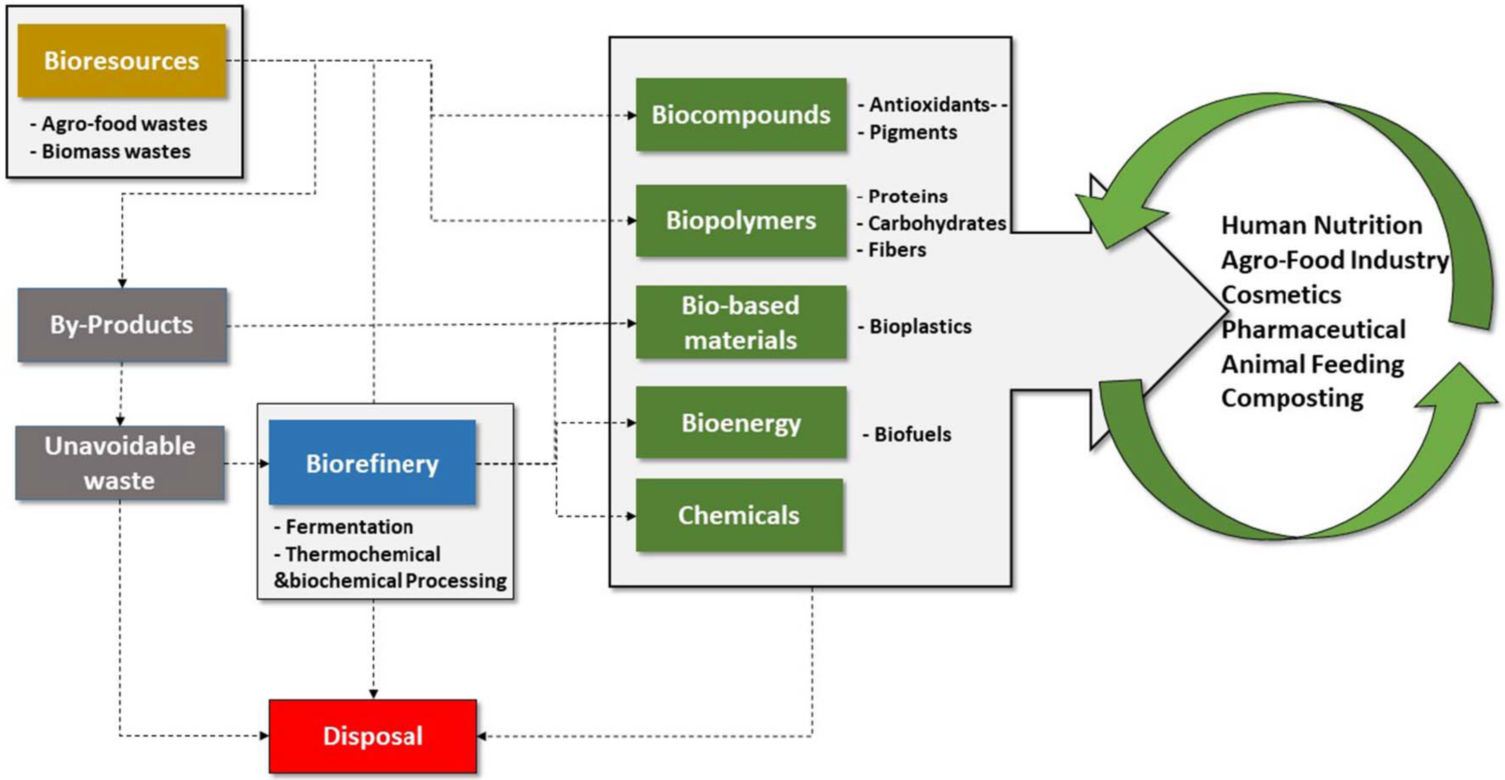
Make your process *greener*

Green materials
concepts and practices need to be introduced

BY-PRODUCTS VALORIZATION CHAIN



Byproduct valorization chain

Rocha, C.M.R., Bioresource Technology (2018), https://doi.org/10.1016/j.biortech.2018.01.068

PRINCIPLES FOR OBTAINING EXTRACTS

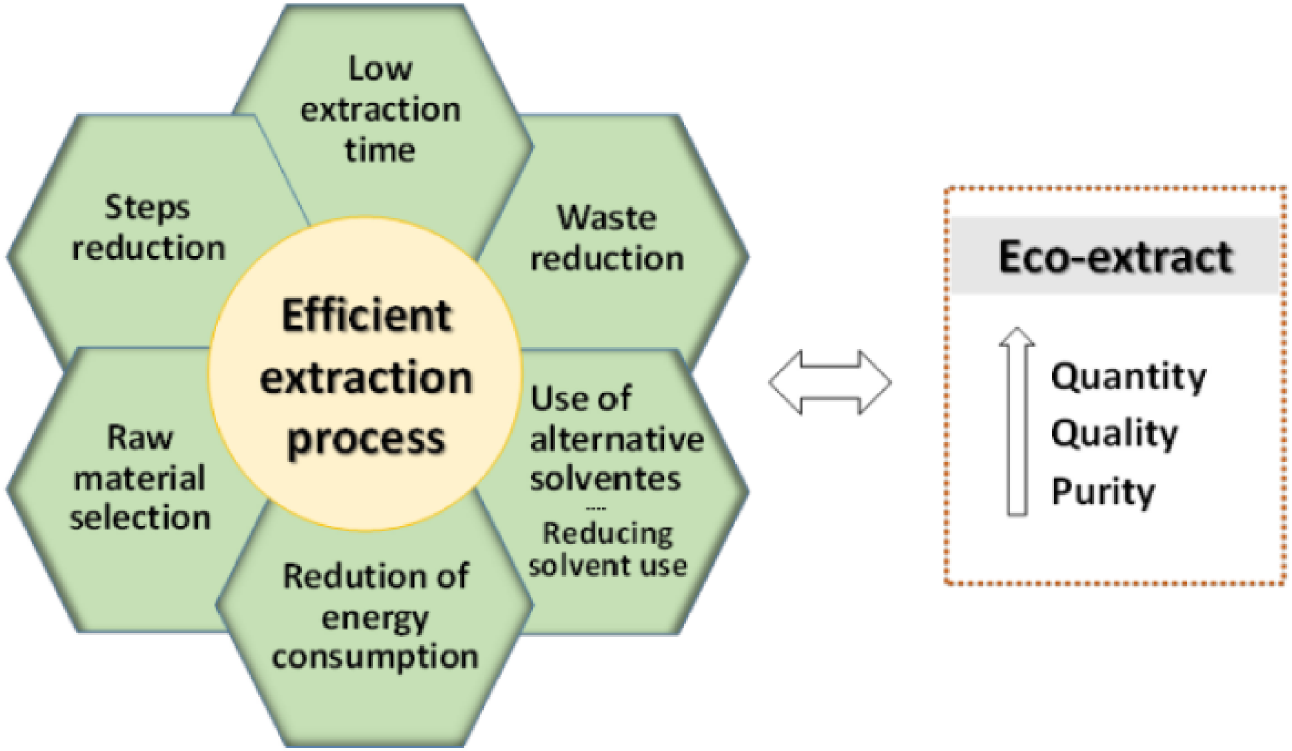


Figure 3. Principles of efficient process for obtaining natural extracts. Adapted from Chemat et al. [8].

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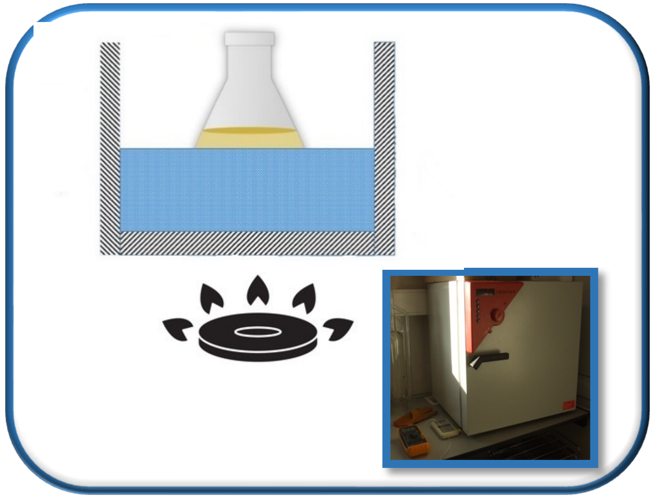
Traditional solvent solid-liquid extraction of natural compounds

- Correct choice of solvents –depending on the target solute’s solubility and polarity;
    - ✓ Solvents with different polarity/hydrophobicity, such as dichloromethane, ethanol, methanol, water



- Use of heat

and/or

- Use of agitation to:
    - ✓ Increase solubility (by removing concentrated solution from the solid surface;
    - ✓ Increase mass transfer (increase “diffusion”).

Extraction of natural compounds

Aims:

- High extraction yields;
- High concentration in the target compound(s) (e.g. bioactives)
- Minimum damage to the target compound(s) – avoid oxidation and/or thermal degradation

The choice of the solvent (and thus the extraction efficiency) depends on the:

- Polarity of the targeted compound
- Molecular affinity between solvent and solute
- Mass transfer
- Use of co-solvent
- Environmental safety
- Human toxicity
- Financial feasibility

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ALTERNATIVE EXTRACTION TECHNOLOGIES

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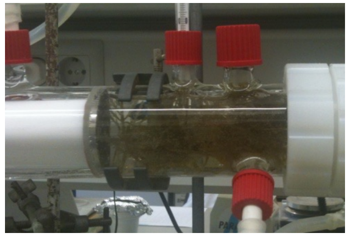
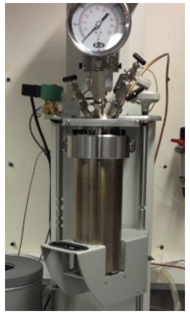
- **Electric (ohmic heating)**
- **Pulsed electric fields**
- Accelerated solvent extraction
- Subcritical water extraction
- Microwave-assisted (MAE)
- Ultrasounds-assisted (UAE)
- Supercritical fluid extraction

- Greener/alternative solvents – ionic liquids/deep eutectic solvents, surfactants, bioethanol, ethyl lactate
- Enzyme digestion
- Extrusion
- Mixed approaches



Electric fields-based technologies

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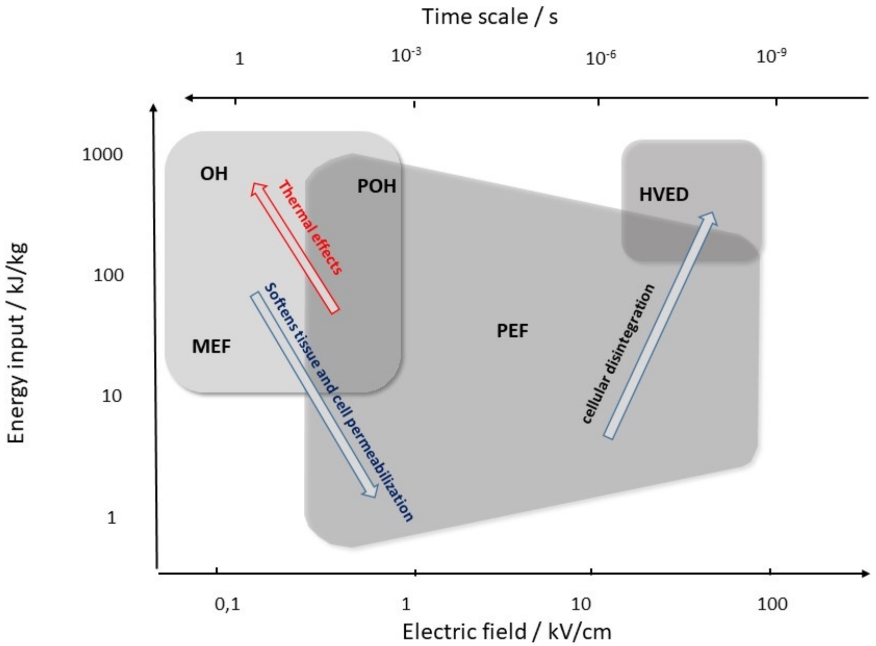


Rocha et al., Bioresource Technology (2018), https://doi.org/10.1016/j.biortech.2018.01.068

For innovative processing:

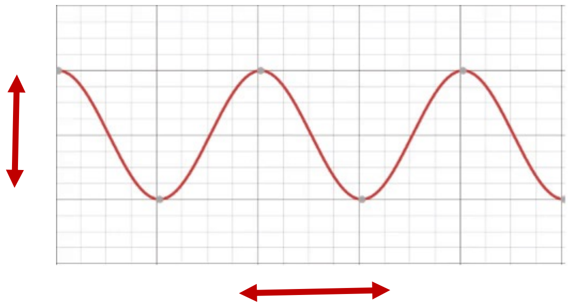
- Enhancement of food quality attibutes
- Optimization of process efficiency
- Reduction of spoilage agents by inactivation of target microorganisms
- Enhancement of extraction yield and selectivity
- Process sustainability

Electric field-based technologies

- Pulsed Electric Fields (PEF)
- High Voltage Electric Discharges (HVED)
- Ohmic Heating (OH)
- Pulsed ohmic heating (POH)
- Moderate electric fields (MEF)

Electric parameters

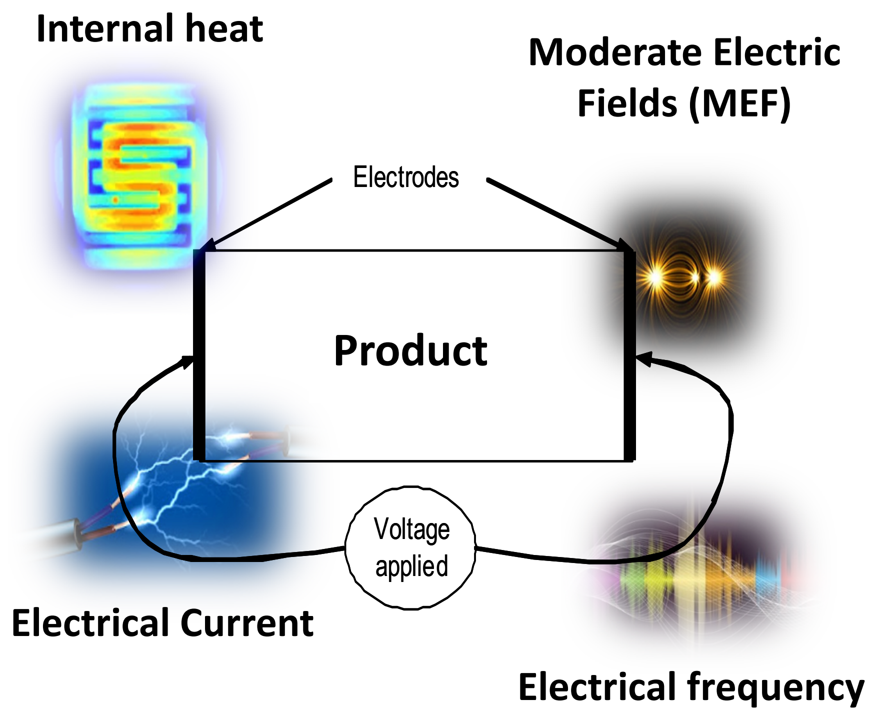
Voltage

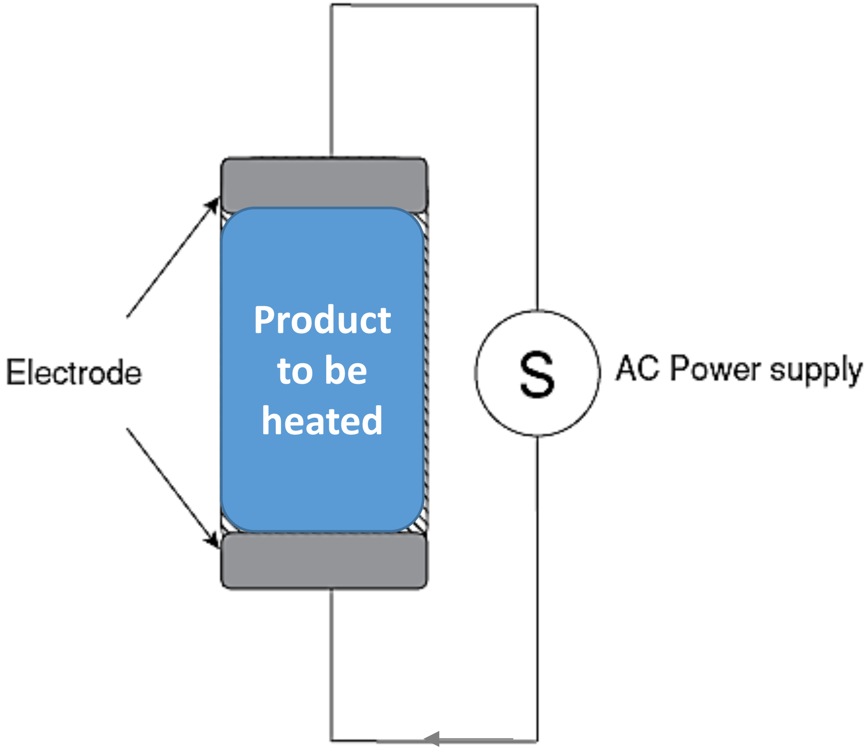


Frequency

ELECTRIC FIELDS - OHMIC HEATING

Ohmic heating - a thermal process where heat is generated directly inside of food products





- Moderate Electric Fields (MEF) range from 1 to 1000 $V.cm^{-1}$

ELECTRIC FIELDS - OHMIC HEATING

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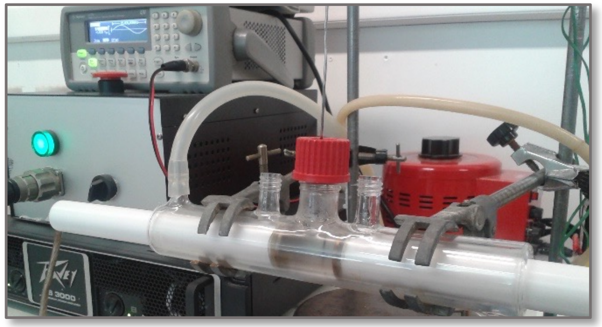
Electric heating, Ohmic heating, Joule heating

- Process where in an electric current is passed through materials with the primary of heating them
- Heating is generated inside the material to be heated (Joule effect) - the heating process does not depend on heat transfer between phases and interfaces, allowing uniform heating and extremely fast heating rate
- No need of boilers/heat exchangers
- It allows heating of large particulates and viscous fluids at comparable rates as long as their conductivity remains similar
- OH also has an effect on cell wall permeabilization



- ❑ Environmentally friendly technology (~ 95% efficient)

- ❑ Relationship between fundamental electric quantities:
  - Electrical current (I);
  - Voltage (V);
  - Resistance (R) or electrical conductivity of food material (σ) .

EXTRACTION OF ADDED-VALUE COMPOUNDS FROM AGRICULTURAL BY-PRODUCTS

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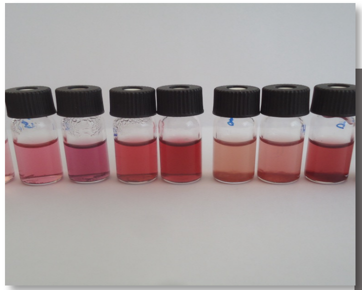


Biotechnological, cosmetics and Food Applications

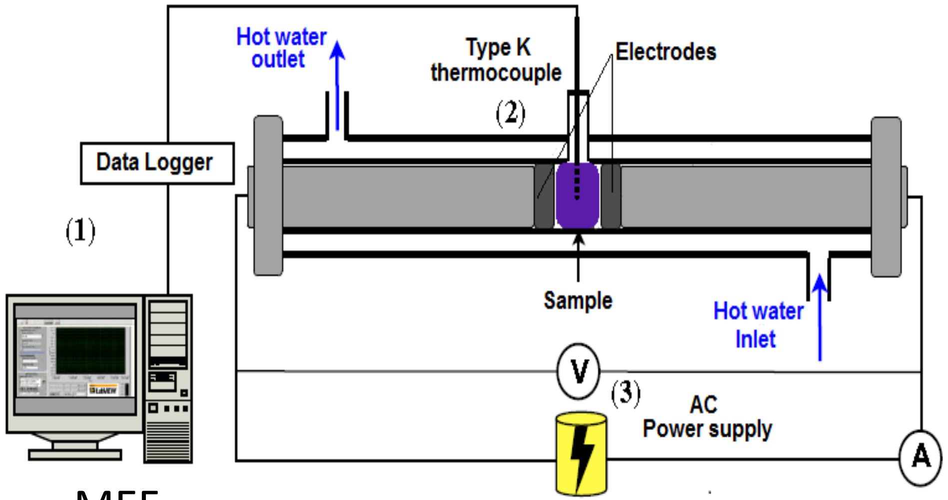
Research at UMinho

- Fruits
- Tomato
- Grapes skins, seeds and stalks
- Chestnut
- Juçara fruit juice
- Olive oil
- Pine bark
- Coloured potato

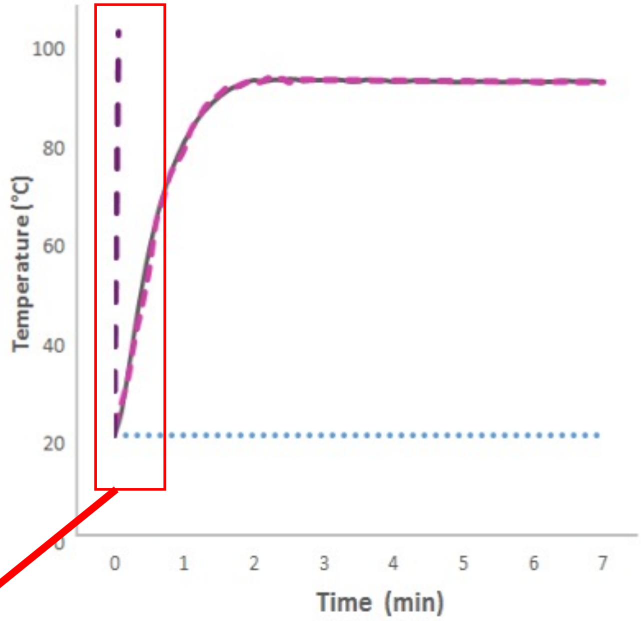
- **Purple potatoes (Solanum tuberosum L. var. Vitelotte)**

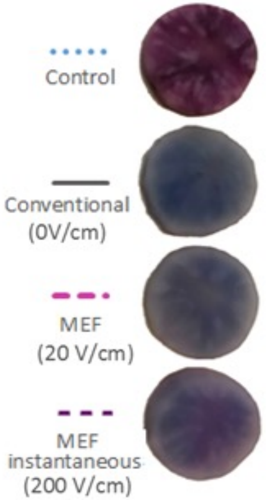
1) Ohmic Heating at different temperatures and moderate electric fields - MEF

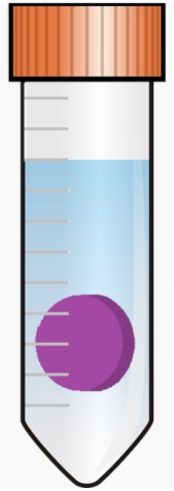
2) Extraction in distilled water



MEF





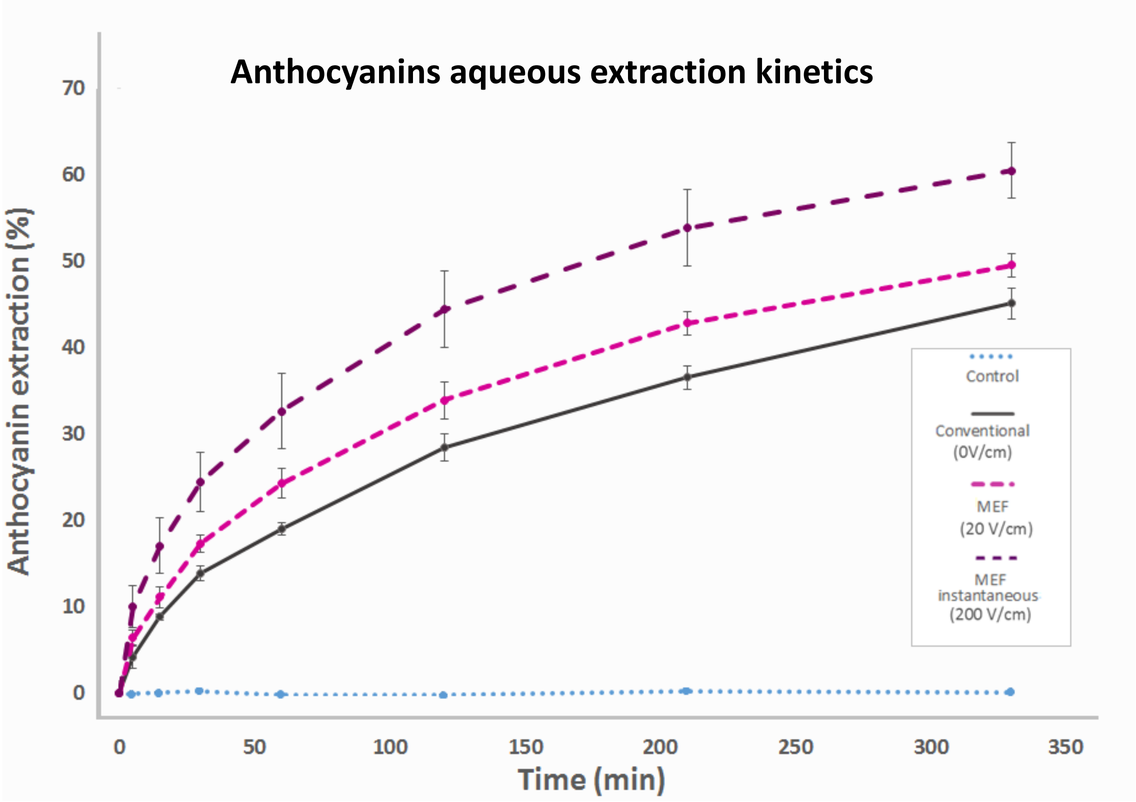


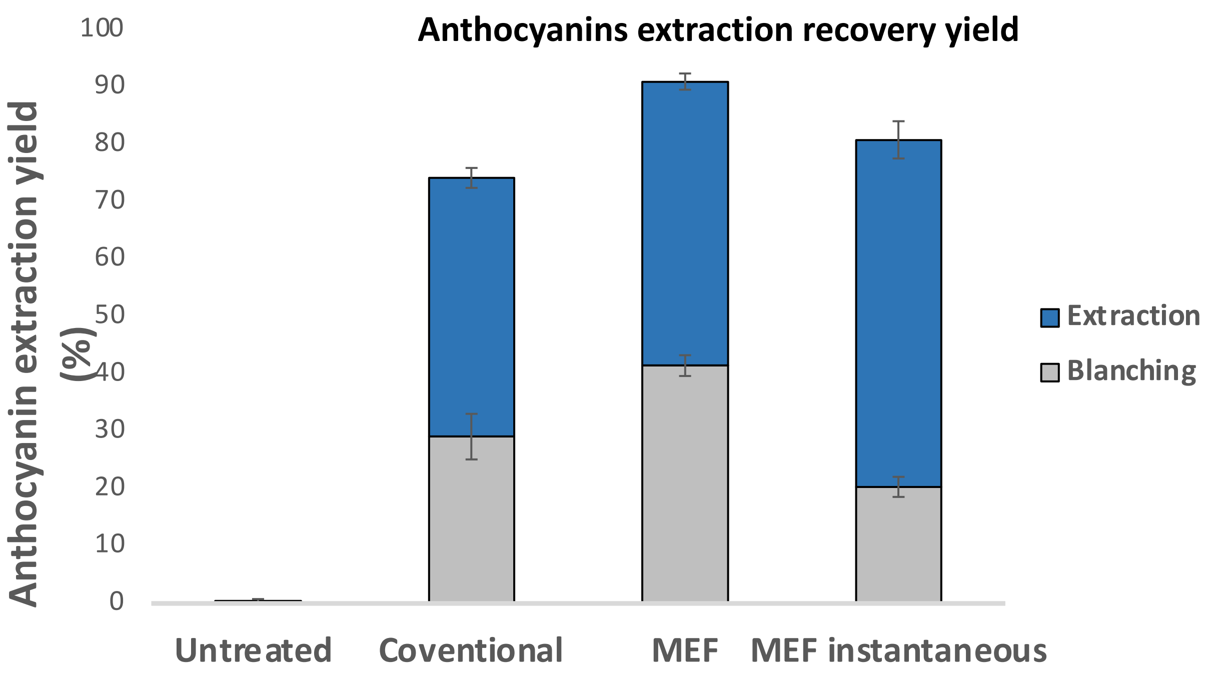
MEF instantaneous (100 °C at 200 V/cm for 1 s)

Pereira et. al. LWT - Food Science and Technology 74 (2016) 493-503

Extraction of bioactive compounds from purple potato

- Maximum anthocyanins and phenolic contents were determined by ensuring full cellular rupture (by freezing and mechanical mashing)





Pereira et. al. LWT - Food Science and Technology 74 (2016) 493-503

Extraction of bioactive compounds from Fruits and vegetables

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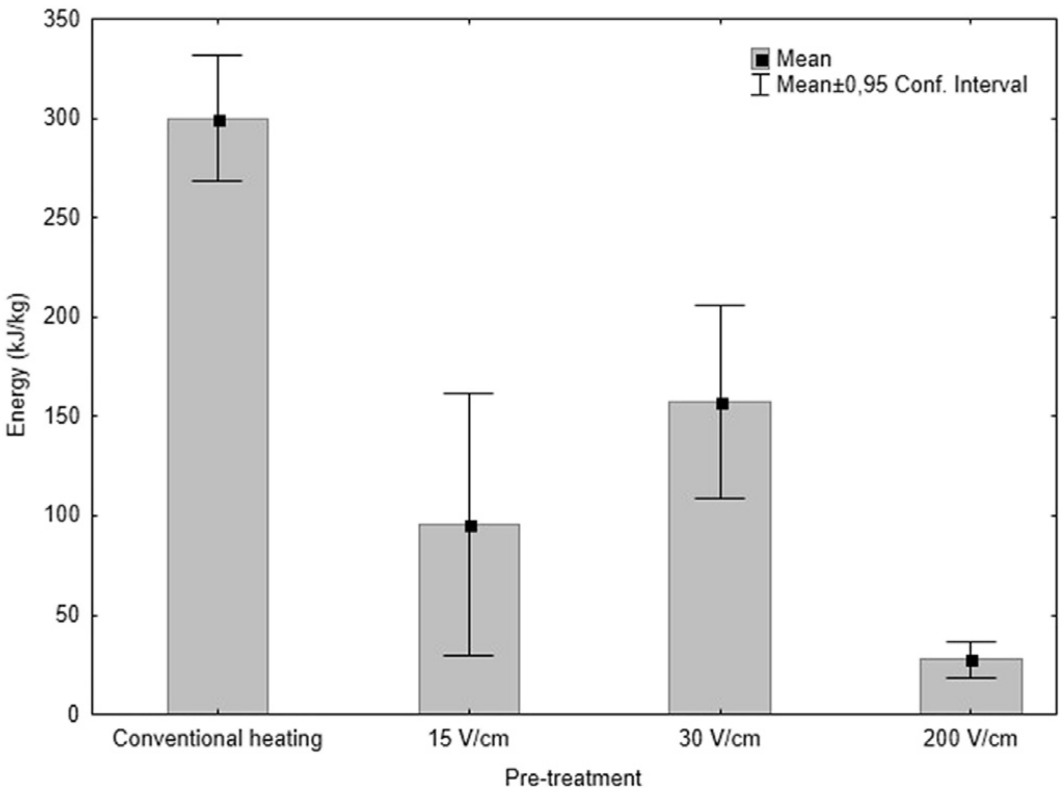
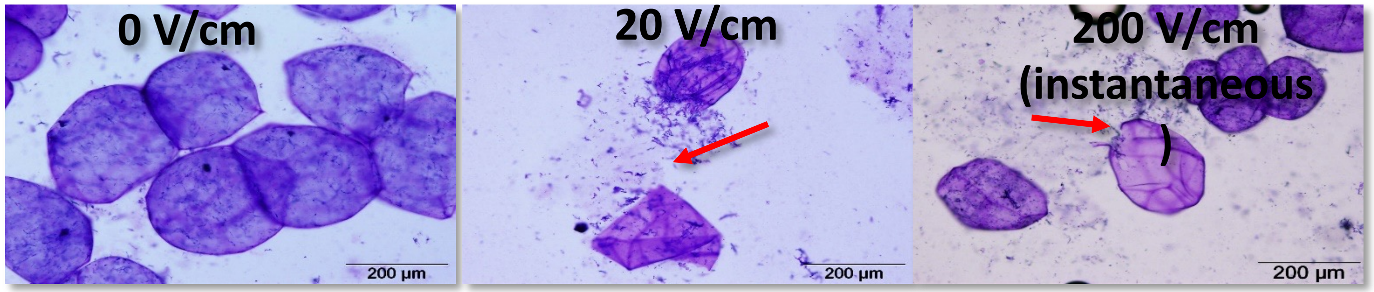


Fig. 7. Total energy consumption of conventional and MEF pre-treatments; conventional heating, MEF at 15 and 30 V/cm were performed at 90 C for 5 min, while MEF at 200 V/cm was performed at 100 C for 1 s. Column spreads correspond to confidence interval of 95%.



- Microphotographs (400X) of potato cells showing MEF induced rupture. Methylene blue stain (marking cell wall and nucleus material). Red arrows indicate cell wall disruption

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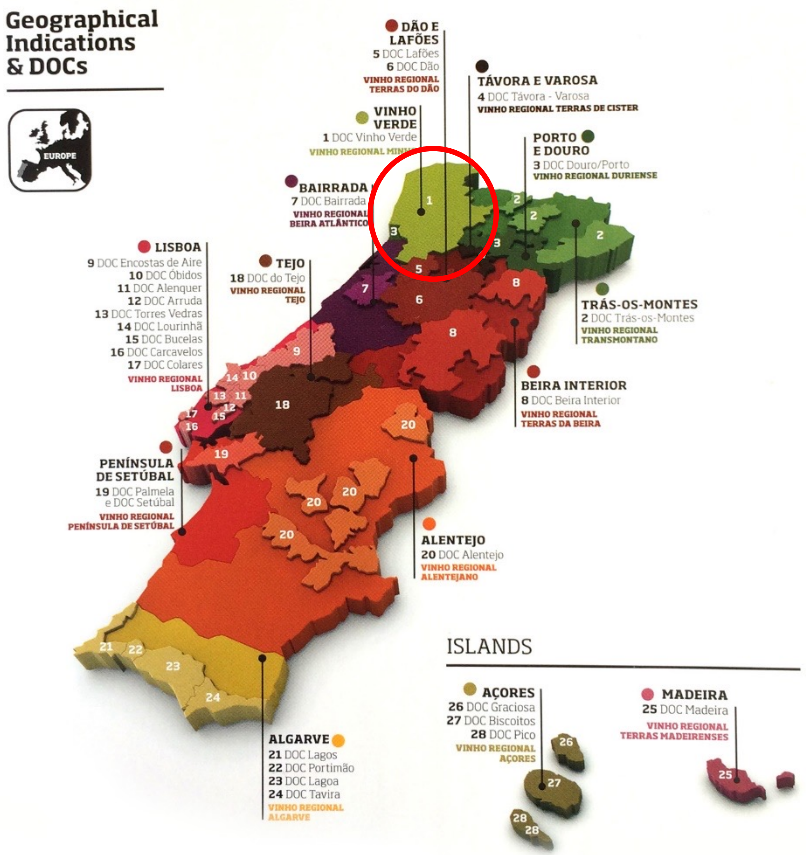
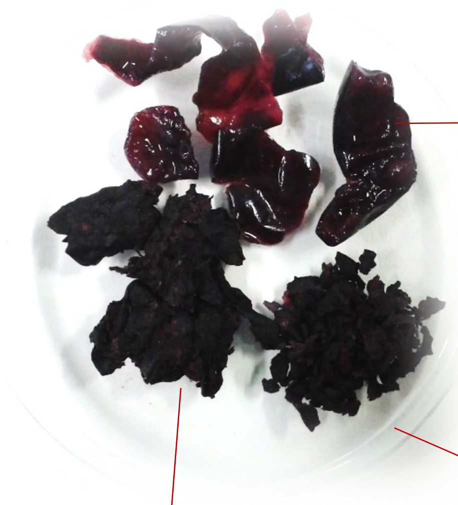
Extraction of bioactive compounds from winemaking residues

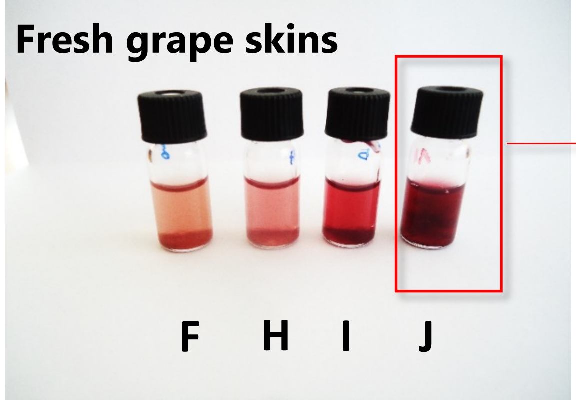
- Grape skins from skins were collected after maceration process for the production of local red Vinho Verde



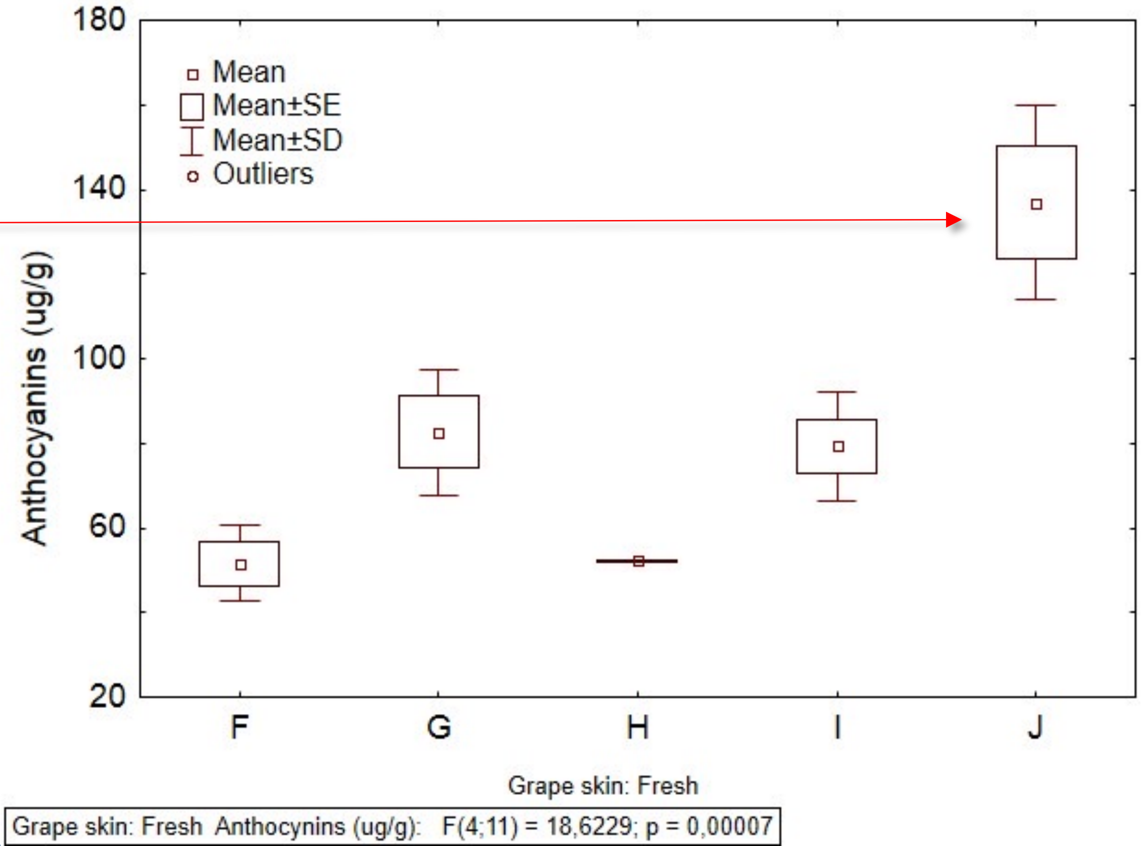


Extraction of bioactive compounds from winemaking residues

- MEF combined with HTST treatments have potential to favor extraction of solutes in grape skins



F: Unheated, 25 °C
G: Control at 40 °C, 20 min
H: Control at 40 °C, 1 min
I: 60 V/cm at 40 °C, 1 min
J: 60 V/cm at 100 °C, 1 s



Extraction of bioactive compounds from Fruits and vegetables

Table 3 – Energy consumption of performed OH pre-treatments and their comparison with other extraction techniques (thermal and non-thermal) using grapes or derived by products.

Treatment	Electrical conditions	Time of treatment	Target temperature/ ºC	Energy input consumption/kJ/kg	Ref
OH40	10 V/cm/25 kHz	20 min	40	21.1 ± 3.3	
OH100	70 V/cm/25 kHz	30 s	100	46.4 ± 14.7	Present study
COV40	–	20 min	40	314	
PEF	0.8 kV/cm	100 ms	–	42	El Darra et al. (2013b)
PEF	5 kV/cm	1 ms	–	53	El Darra et al. (2013b)
PEF	13 kV/cm	10 μs	<30 °C	up to 564	Barba et al. (2015)
PEF	0.4 kV/cm	5 s	45 °C	38.1	El Darra et al. (2013a)
PEF	0.1 kV/cm	210 s	82 °C	178.8	El Darra et al. (2013a)
US	24 kHz	5 min	Na	121	El Darra et al. (2013b)
US	24 kHz	10 min	Na	242	
US	24 kHz	15 min	Na	363	

Pereira, R. N. et al (2020). Using Ohmic Heating effect on grape skins as a pretreatment for anthocyanins extraction. *Food and Bioproducts Processing*. https://doi.org/https://doi.org/10.1016/j.fbp.2020.09.009

VINE PRUNING RESIDUE

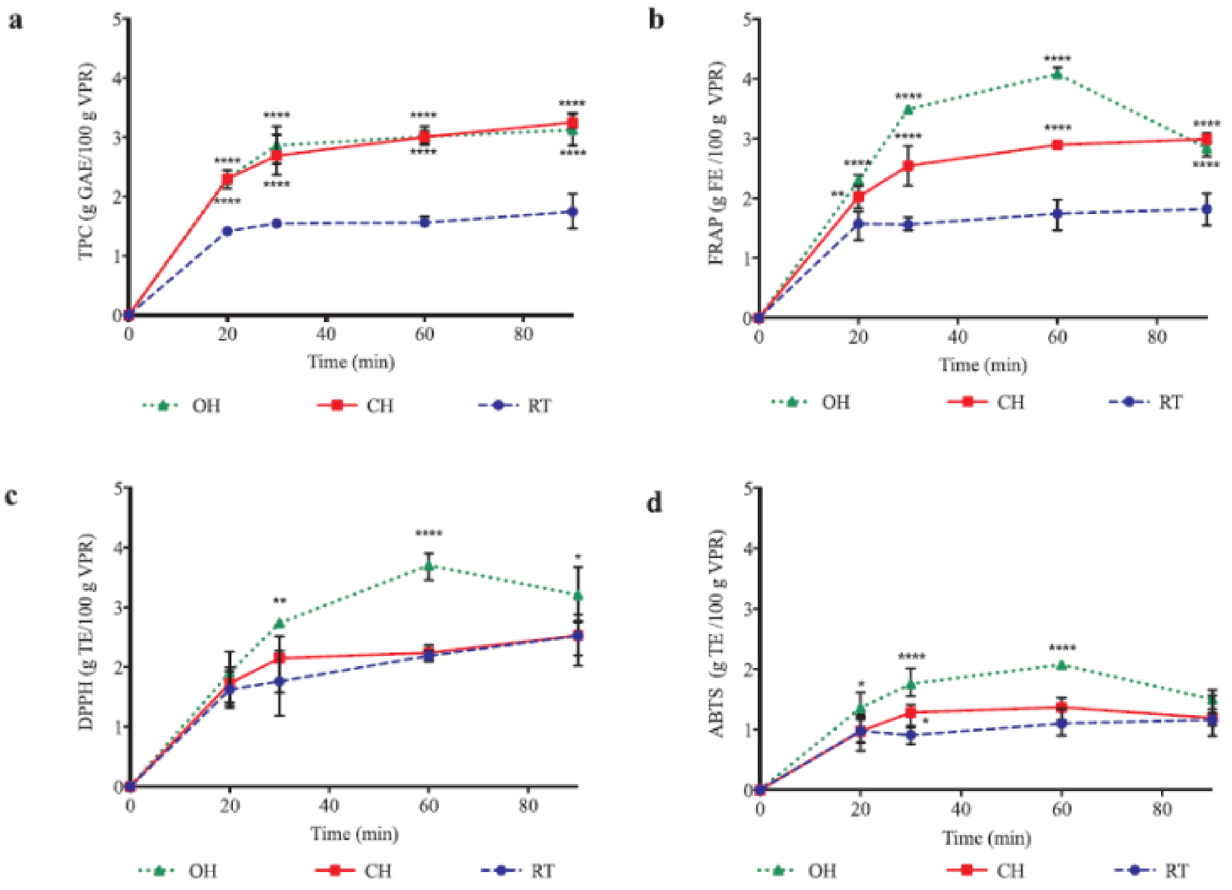


Fig. 1. Extraction of phenolic compounds from vine pruning residue using different extraction times (20, 30, 60 and 90 min) and methods of heating: room temperature (RT), conventional heating (CH) and ohmic heating (OH). (a) Total phenolic compounds; (b) FRAP; (c) DPPH; (e) ABTS. Fixed conditions: solid liquid ratio (1:40 w/v), ethanol concentration (45%) and temperature (80 °C). All experiments were done in triplicate and the results expressed as mean ± SD. *$p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$.

Food Chemistry 316 (2020) 126298

Table 1

Total phenolic compounds (TPC) and antioxidant activity FRAP, DPPH and ABTS of the extracts produced from vine pruning residue by using different methods of heating: room temperature (RT), conventional heating (CH) and ohmic heating at different electric fields (LEF: Low Electric field; IEF: Intermediate Electric field).

Runs	TPC	FRAP	DPPH		ABTS	
	g GAE/100 g VPR	g FE/100 g VPR	g TE/100 g VPR	IC_{50}	g TE/100 g VPR	IC_{50}
IEF	3.4 ± 0.1[c]	4.6 ± 0.2[c]	4.1 ± 0.1[d]	0.76[a]	3.1 ± 0.1[c]	0.34[a]
LEF	3.1 ± 0.2[b]	4.1 ± 0.3[b]	3.2 ± 0.1[c]	0.90[b]	1.9 ± 0.2[b]	0.44[b]
CH	3.0 ± 0.2[b]	3.7 ± 0.1[d]	2.7 ± 0.2[b]	0.95[b]	1.9 ± 0.1[b]	0.40[b]
RT	1.2 ± 0.1[a]	1.7 ± 0.2[a]	2.2 ± 0.1[a]	1.25[c]	1.01 ± 0.1[a]	0.94[c]

*The averages followed by the same letters within a column do not differ by the Tukey test ($p < 0.05$). GAE: gallic acid equivalents; FE ferrous equivalents; TE (II): Trolox equivalents. Antiradical activity is expressed as a mean ($n = 3$) of IC_{50} values (g of extract/L of solution).

Table 2

Polyphenolic composition of the VPR extracts (Expressed as mg/100 g VPR) obtained by different methods of heating: room temperature (RT), conventional heating (CH) and ohmic heating at different electric fields (LEF: Low Electric field; IEF: Intermediate Electric field).

Polyphenols (mg/100 g VPR)	IEF	LEF	CH	RT
Phenolics acid				
Gallic acid	2.9[b]	3.5[a]	ND	ND
o-Cumaric acid	15.8[b]	26.5[c]	14.2[b]	6.6[a]
Ferulic acid	46.6[a]	46.1[a]	ND	ND
Ellagic acid	222.9[b]	77.7[a]	ND	ND
Vanillic acid	68.4[bc]	70.3[c]	67.2[b]	31.2[a]
Flavonoids				
Hesperidin	180.3[b]	149.0[a]	ND	ND
Apigenin	384.2[b]	157.5[a]	ND	ND
Quercetin	287.2[bc]	286.8[bc]	281.6[b]	132.8[a]
Taxifolin	23.7[c]	21.8[bc]	19.8[a]	ND
Simple phenols				
HidroxiTyrosol	152.4[c]	151.6[bc]	149.6[bc]	ND
Tyrosol	142.3[c]	139.8[b]	137.1[b]	64.2[a]
Stilbenes				
trans-resveratrol	65.4[a]	137.3[b]	ND	ND

*Where The averages followed by the same letters within a file do not differ by the Tukey test ($p < 0.05$). ND: not detected.

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ACS Sustainable Chemistry & Engineering
Research Article
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pubs.acs.org/journal/ascecg

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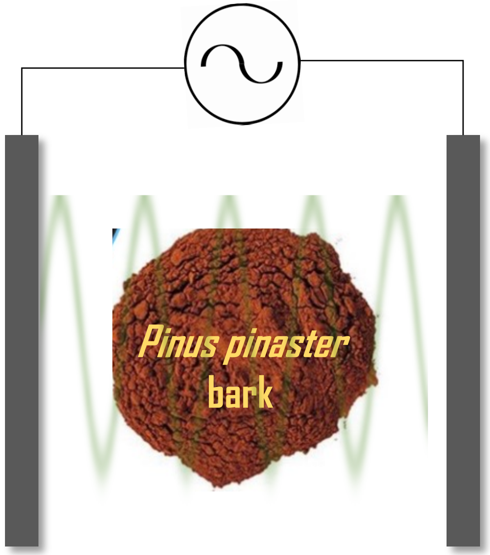
Moderate Electric Fields as a Potential Tool for Sustainable Recovery of Phenolic Compounds from *Pinus pinaster* Bark

Pedro Ferreira-Santos,[†] Zlatina Genisheva,[†] Ricardo N. Pereira,[†] José A. Teixeira,[†] and Cristina M. R. Rocha*[,†]

[†]CEB - Centre of Biological Engineering, University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal



Green Extraction
(Ohmic Heating)

Ohmic heating
Conventional heating

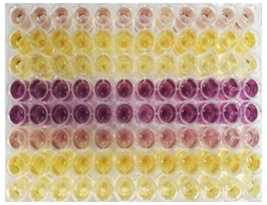
Extraction made at 83 ºC
for 30 min

Water
Ethanol

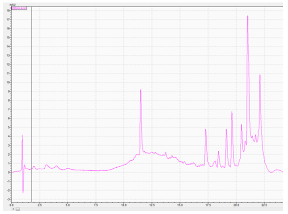
Extraction Yield

Total Phenolic content

Antioxidant activity



Phenolic compounds (UPLC)



Structure (SEM)

Energy consumption

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Extraction of bioactive compounds from pine bark

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REDVALUE
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- Pine bark sample was characterized in terms of proximate analysis (total protein, ashes, carbohydrates, and lipids).

	Outter bark	**Inner bark**
Components	Composition (%)	Composition (%)
***Cellulose*[a]**	**17.39 (±0.37)**	**14.65 (±0.86)**
Hemicellulose	12.31 (±0.20)	8.87 (±0.68)
Xylan	10.92 (±0.19)	7.02 (±0.44)
Arabinan	1.39 (±0.01)	1.85 (±0.24)
Acetyl group	n.d.	n.d.
Klason lignin	**43.25 (±0.24)**	**44.88 (±0.05)**
Soluble lignin	1.60 (±0.01)	1.72 (±0.03)
Ashes	0.87 (±0.00)	2.03 (±0.04)
Protein	1.48 (±0.13)	2.92 (±0.05)
***Extractives*[b]**	**13.16 (±0.15)**	**15.33 (±0.34)**
***Others*[c]**	9.45	9.60

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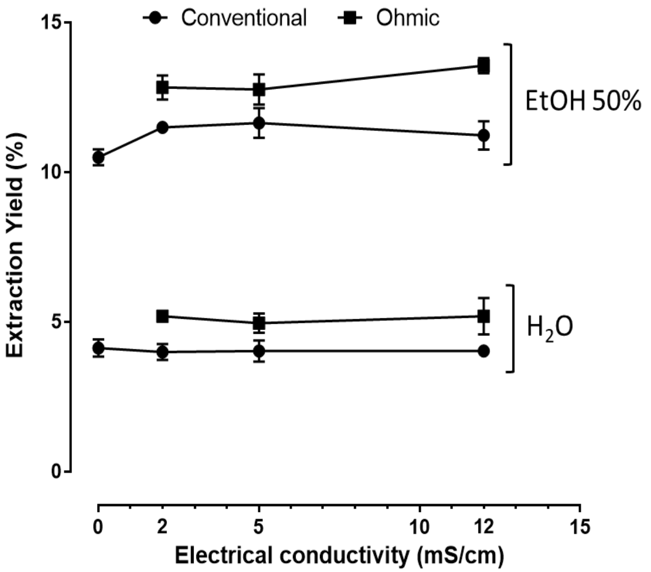
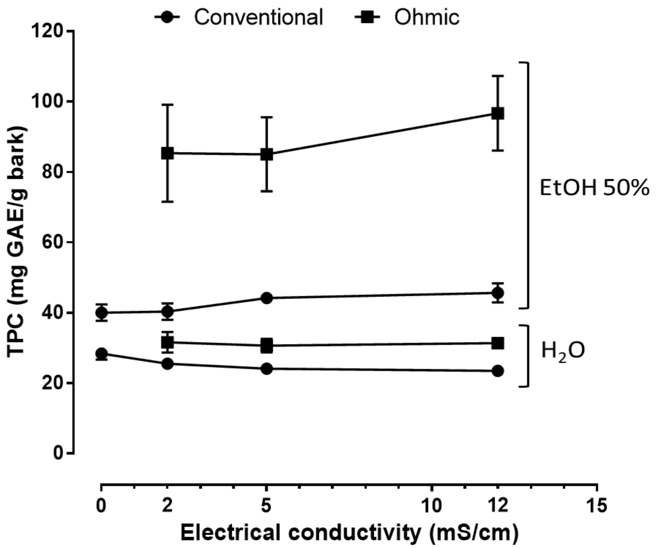


Extraction yields (%), influence of solvent and electrical conductivity using conventional heating and ohmic-assisted extraction methods in *Pinus pinaster* bark



Total phenolic content (TPC) from *Pinus pinaster* bark. Influence of solvent and electrical conductivity using conventional heating and ohmic-assisted extraction methods

Ferreira-Santos et. al. ACS – Sustainable Chemistry & Engineering 7 (2019) 8816–8826

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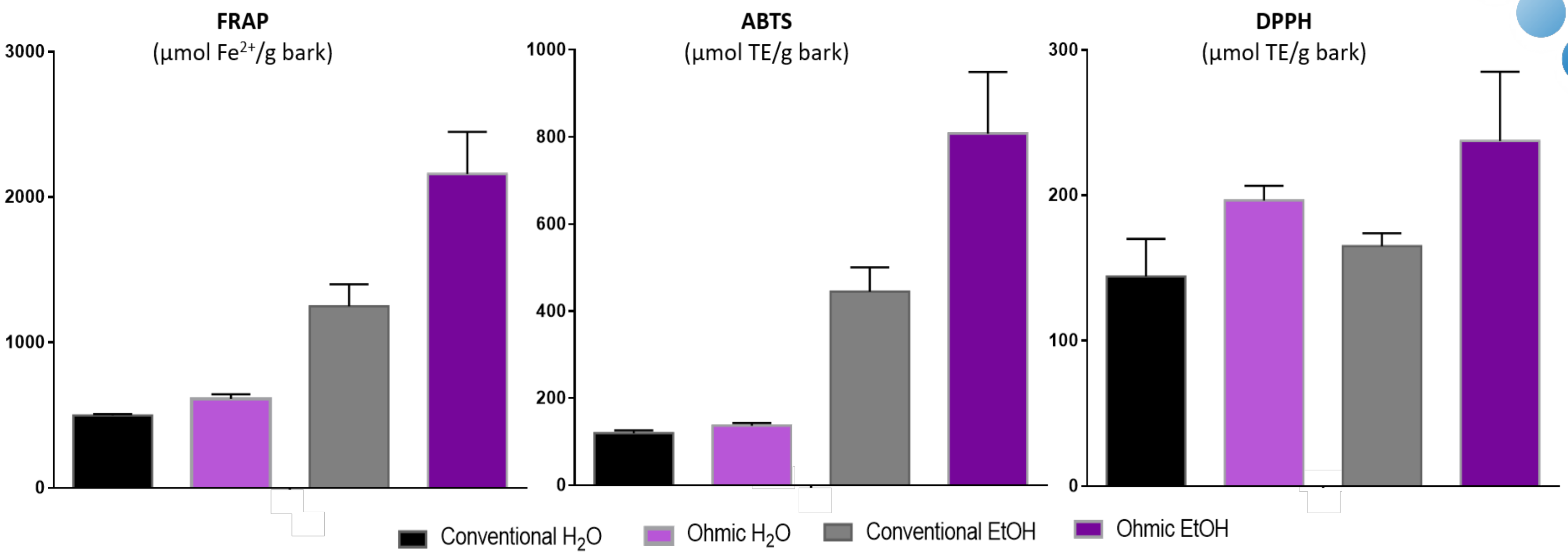

Figure: Antioxidant activity of the extracts obtained from *Pinus pinaster* bark measured by FRAP, ABTS and DPPH methods

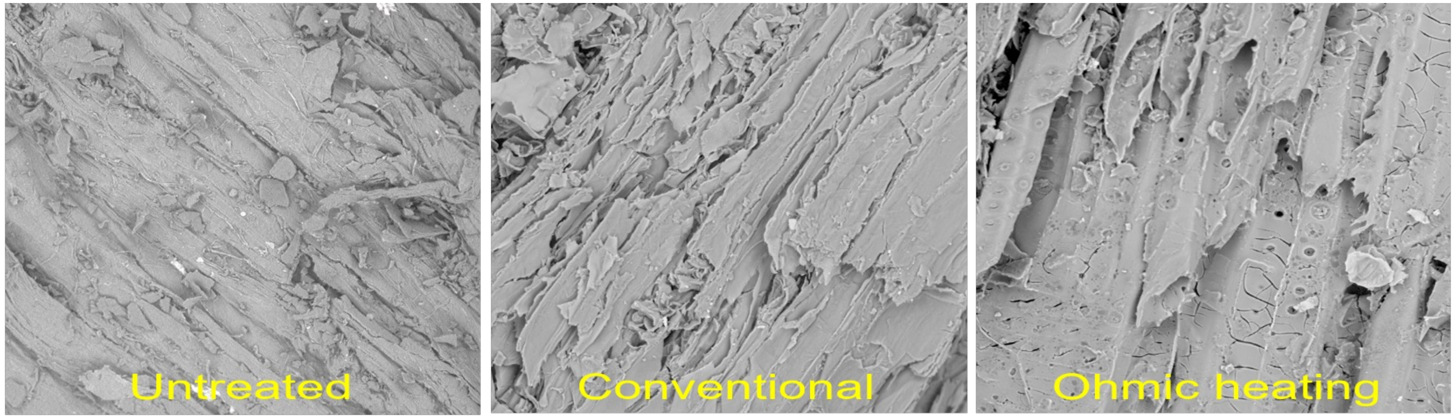

Figure 5. *Scanning electron microscopy images of untreated, conventional heating and ohmic heating treated Pinus pinaster bark*

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Phenolic profile

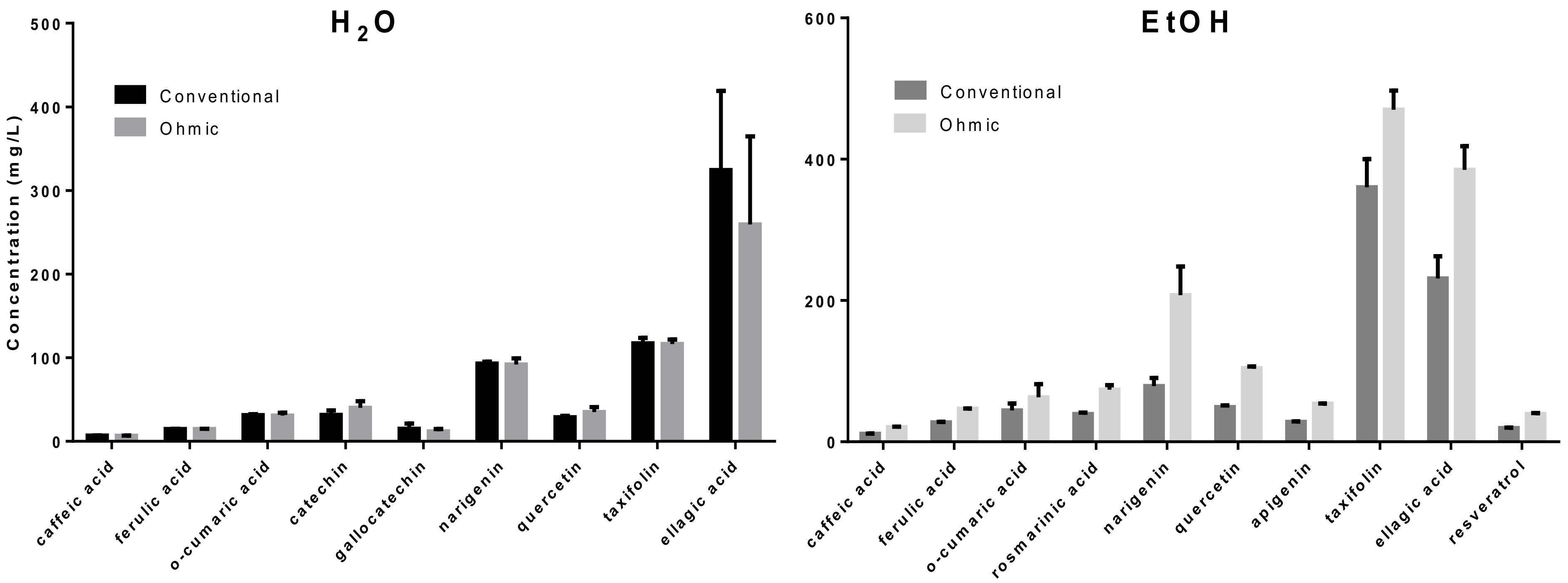

Figure 4. Phenolic compounds profile. Identification and quantification from *Pinus pinaster* bark extracts by UPLC-DAD.

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GREEN EXTRACTION OF SEAWEED´S COMPOUNDS *GRACILARIA VERMICULOPHYLLA*

CENTRE OF BIOLOGICAL ENGINEERING

Clean Technologies

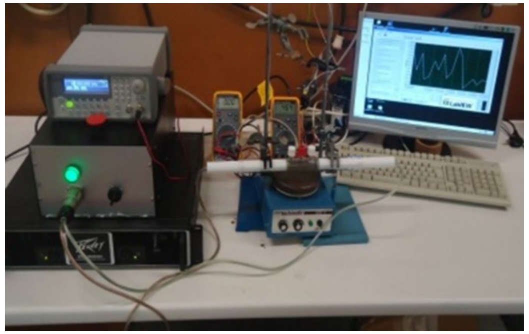
Project: OH20-Ohmic heating for improved green extraction of seaweeds´compounds


Red Macroalgae

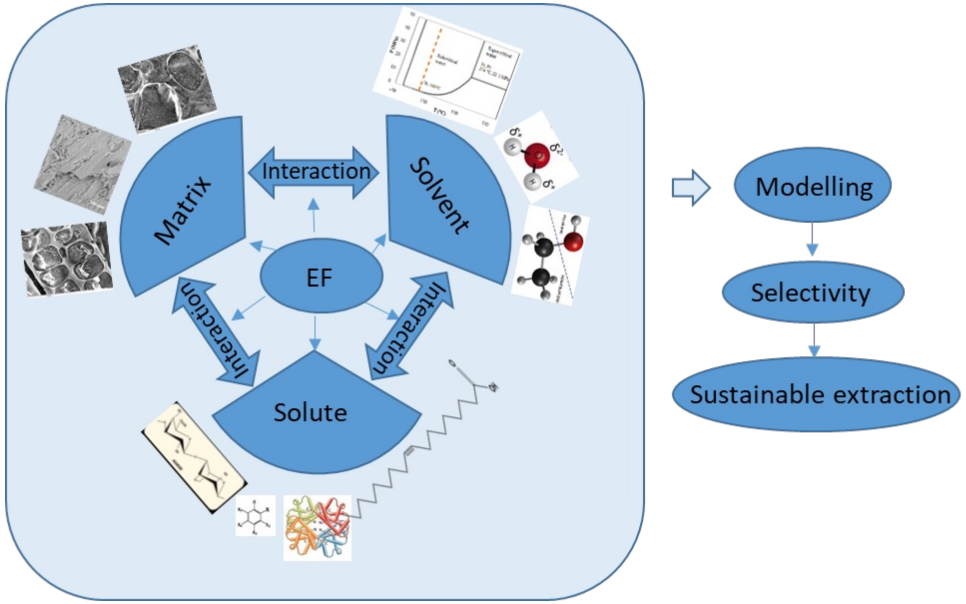

Green Macroalgae


Brown Macroalgae

Ohmic heating



Bioactive compounds
Polysaccharides



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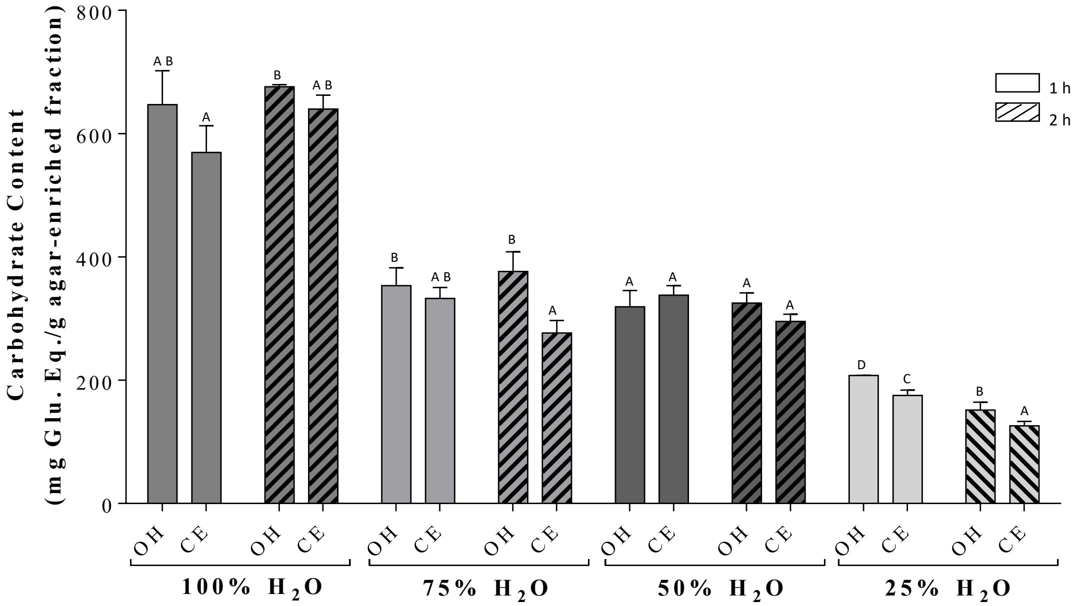
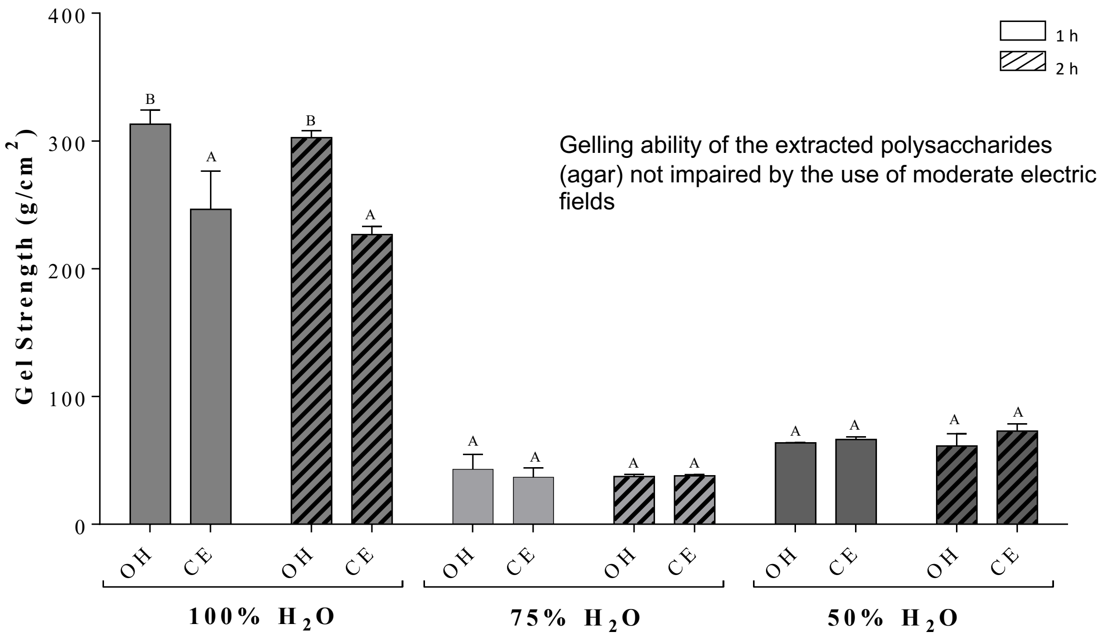
Green Extraction of Seaweed´s Compounds *Gracilaria vermiculophylla*

University of Minho
School of Engineering

Centre of Biological Engineering



Effect of ohmic heating (OH) and conventional extraction (CE) on the carbohydrate content of different solvent ratio at 1 h and 2 h of extraction, expressed in mg glucose equivalent per g of agar-enriched fraction.



Effect of ohmic heating (OH) and conventional extraction (CE) on agar’s gel strength of different solvent ratio at 1 h and 2 h of extraction, expressed in g per cm^2.

Frequency and electric field were set at 25kHz and 2-8 V/cm and the extraction carried for 1 h and 2 h at 82ºC

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GREEN EXTRACTION OF SEAWEED´S COMPOUNDS *GRACILARIA VERMICULOPHYLLA*

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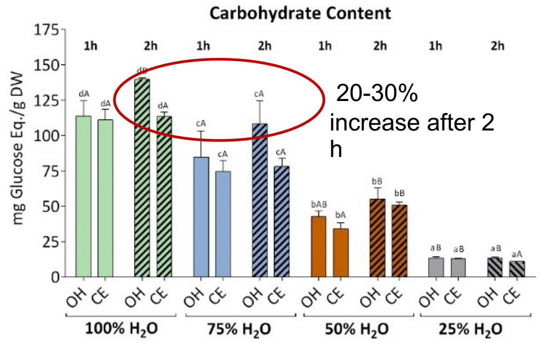
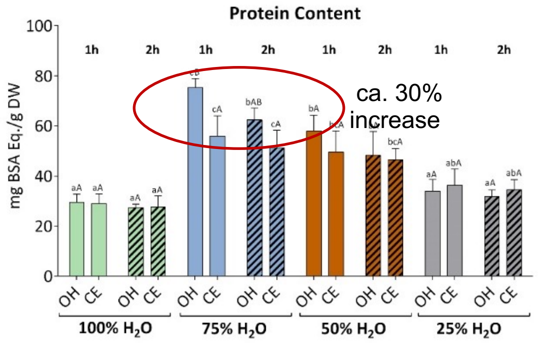
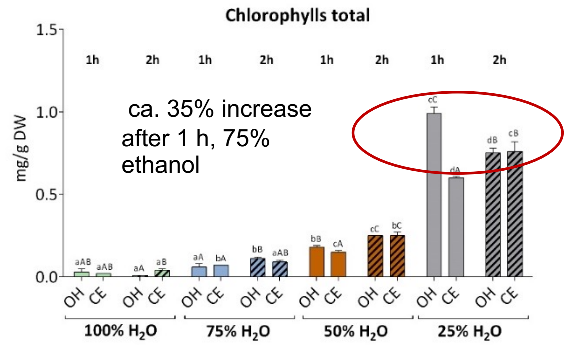
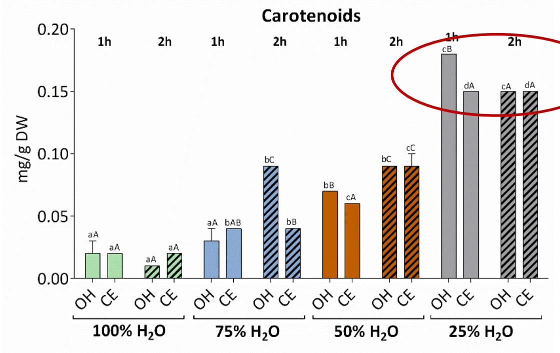


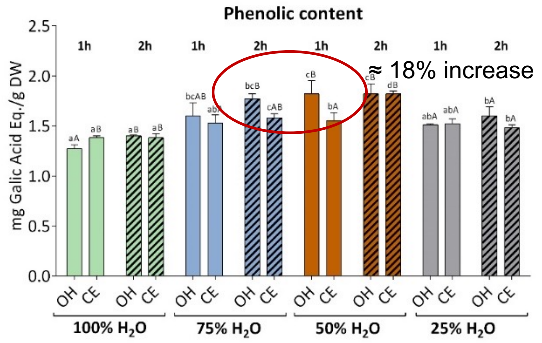


Recovery of different types of compounds from *Gracilaria vermiculophylla*.





20% increase after 1 h, 75% ethanol



OH allowed to enhance the selective action of the solvent leading to higher extraction yields for the compound of interest when the appropriate solvent was used.

A kinetic effect is also observed that accelerates the extraction of some types of compounds

OH is an interesting alternative for extraction as it is more efficient, faster and with reduced energy consumption

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BIOACTIVE COMPOUNDS RECOVERY FROM *SPIRULINA*

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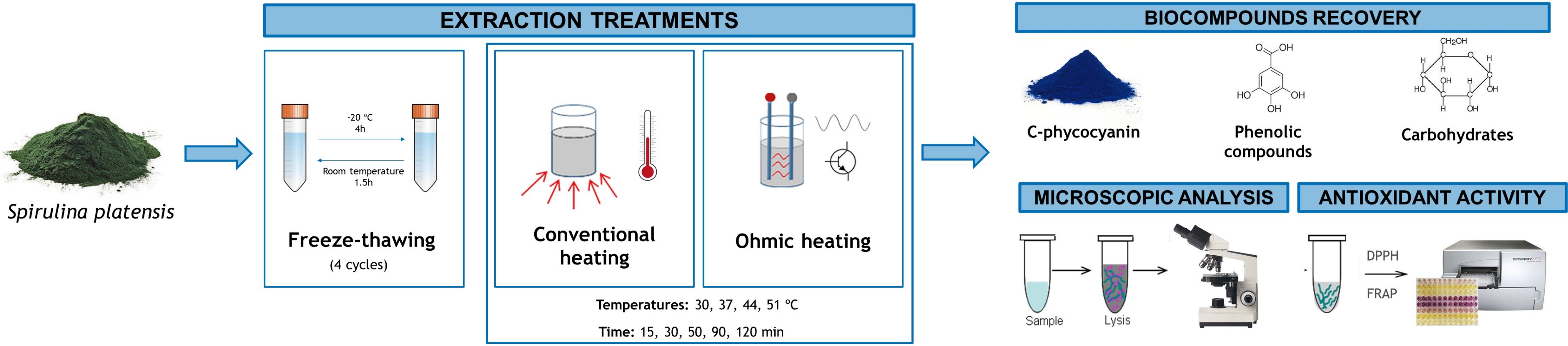

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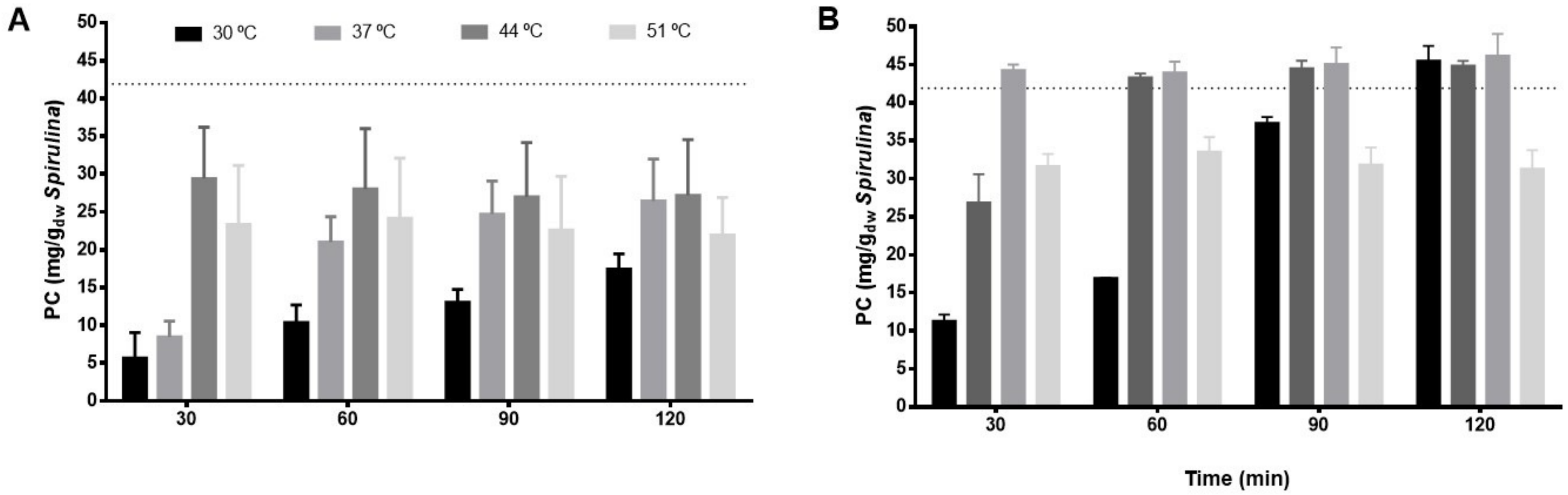
BIOACTIVE COMPOUNDS RECOVERY FROM *SPIRULINA*



C-phycocyanin recovery (mg/g_{dw} of *Spirulina*) for conventional heating **(A)** and ohmic heating **(B)** according to different temperatures (30, 37, 44, 51 ºC) and exposure times (30, 60, 90, 120 min). Error bars represent mean ± SD and the line in the graphs indicates the concentration value obtained using the freeze-thawing method (denaturing temperature of PC – 44ºC)

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Bioactive compounds recovery from *Spirulina*

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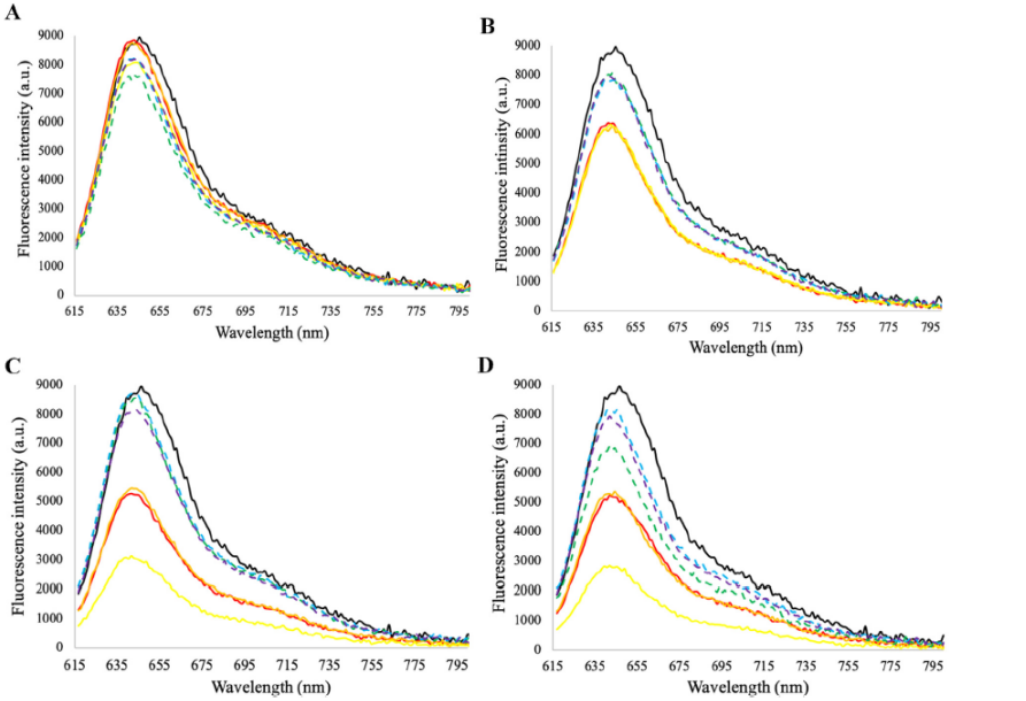

Fig. 1. Fluorescence spectra of C-phycocyanin before (untreated) and after thermal treatments with or without the application of an electric field (conventional and OH, respectively) at different exposure times (15, 30, 60 min) and temperatures (30 °C: A; 37 °C: B; 44 °C: C; 51 °C: D). Excitation wavelength was 609 nm. ▬ Untreated; ▬ Conv 15 min; ▬ Conv 30 min; ▬ Conv 60 min; ••• OH 15 min; ••• OH 30 min; ••• OH 60 min.

Using OH the PC stability increases when subjected to adverse temperature conditions without affecting its physicochemical properties (fluorescence and secondary structure)

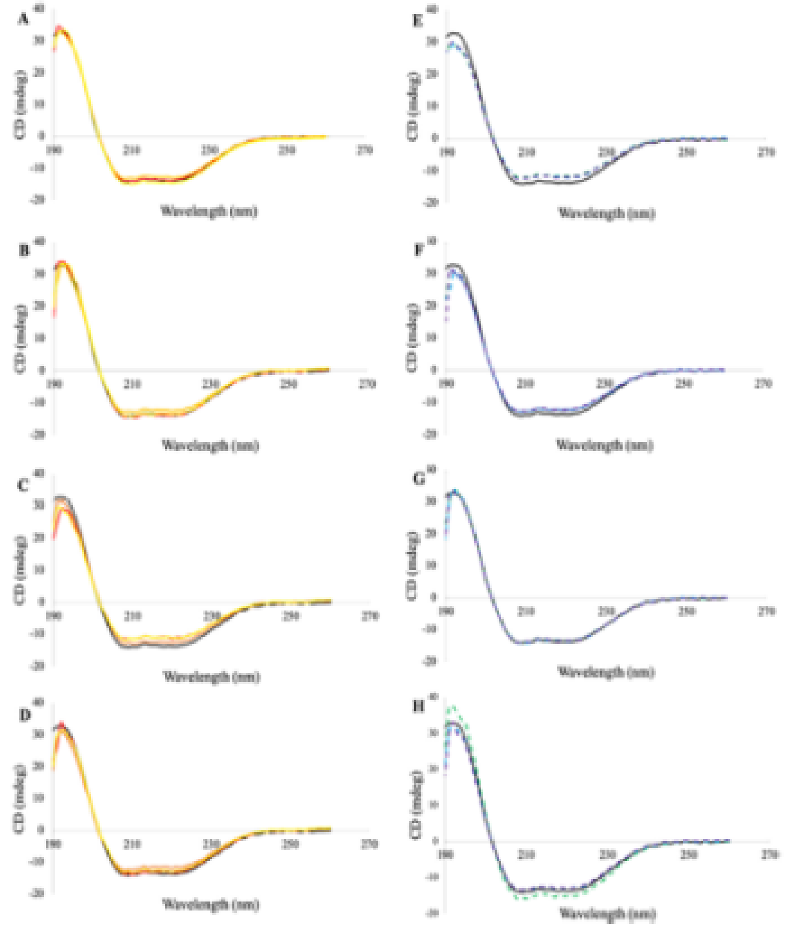

Fig. 2. Circular dichroism spectra of C-phycocyanin before (untreated) and after thermal treatments without or with the application of an electric field (conventional treatment: A, B, C, D and ohmic heating: E, F, G, H) at different exposure times and temperatures (30 °C: A, E; 37 °C: B, F; 44 °C: C, G; 51 °C: D, H). ▬ Untreated; ▬ Conv 15 min; ▬ Conv 30 min; ▬ Conv 60 min; --- OH 15 min; --- OH 30 min; --- OH 60 min.

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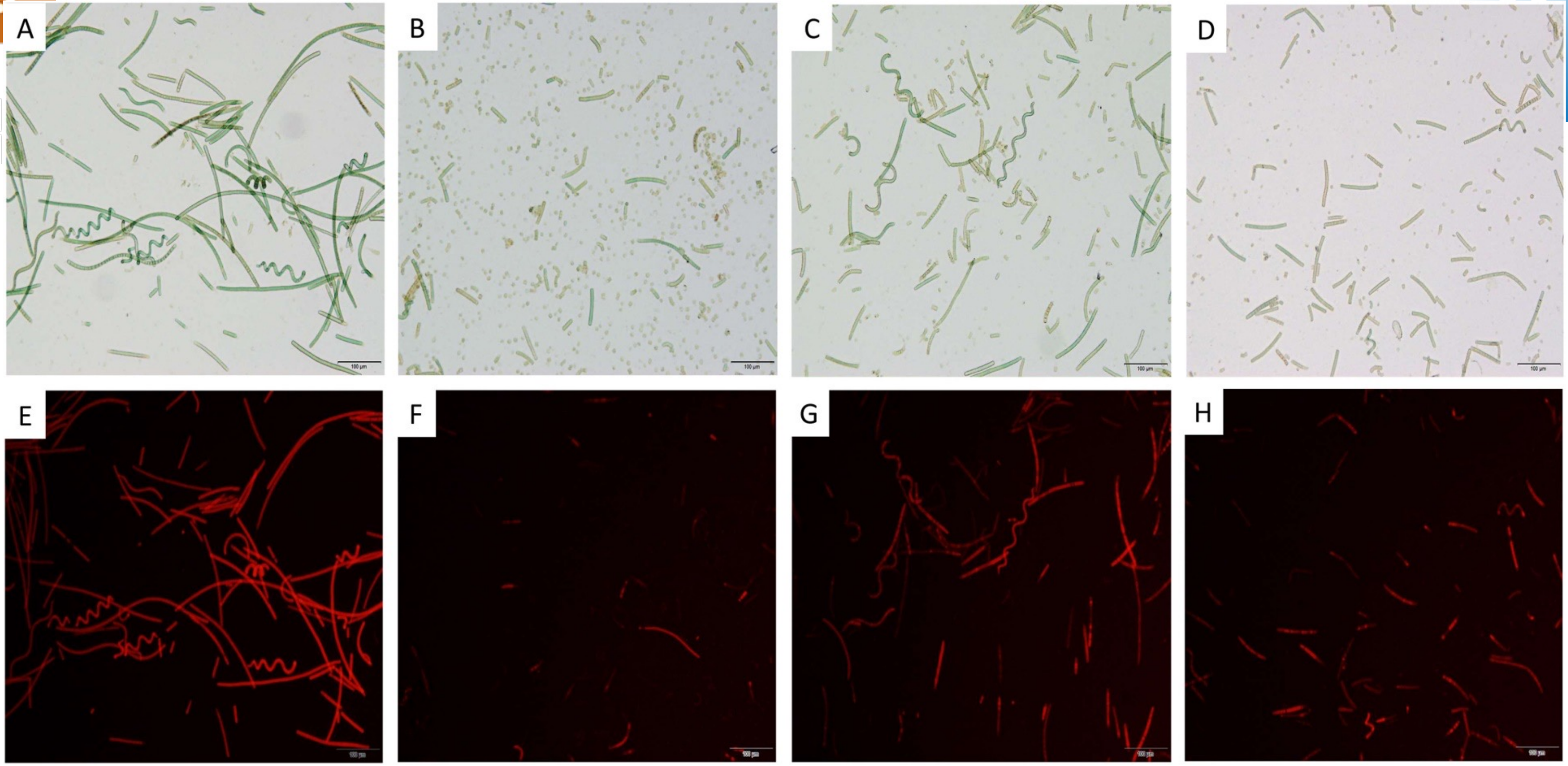

Images of optical (**A**, **B**, **C**, **D**) and fluorescence (**E**, **F**, **G**, **H**) microscopy (100X) of untreated (**A**, **E**), freeze-thawing treated (**B**, **F**), conventional thermal treated (**C**, **G**) and OH treated (**D**, **H**) cells of Spirulina platensis. Red images represent the cyanobacteria autofluorescence using TRITC filter. Scale bar of 100 µm applies to all images

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OH APPLICATIONS – FINAL REMARKS

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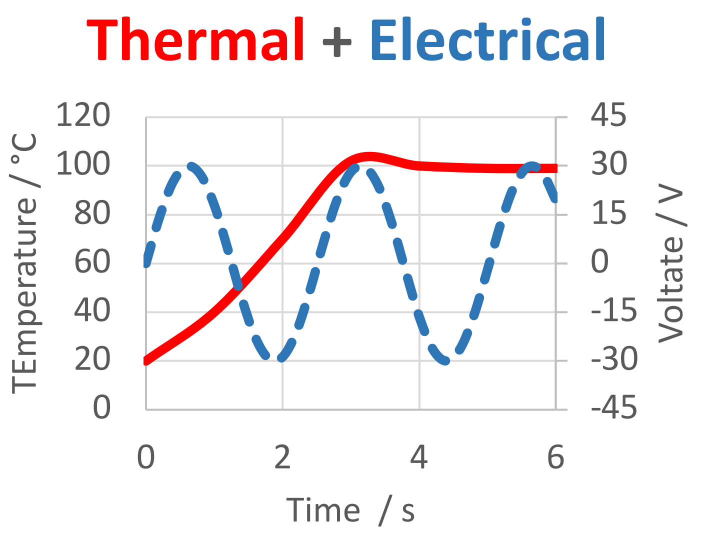
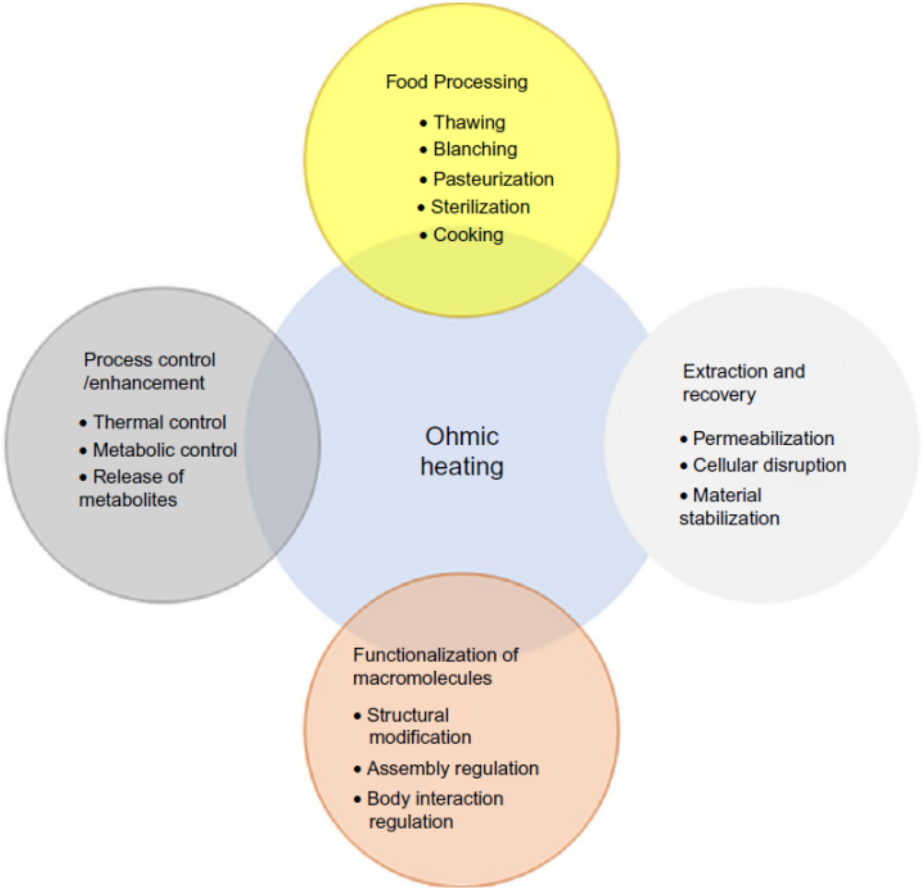
- ✓ Conventional extraction operations can be replaced – less time consuming and more efficient and "clean" technology
- ✓ Synergy of fast internal heating and moderate electric fields on extraction protocols of sensitive bioactive compounds
- ✓ Need of more fundamental knowledge about interaction of electric fields and bioactive molecules
- ✓ Need of more fundamental knowledge about the effect of electric fields in extraction kinetics and solvent selectivity
- ✓ Impact of extraction routes on bioaccessibility of bioactive molecules
- ✓ Interdisciplinary approach combining different competences chemical/biological engineering, biophysics…



OH APPLICATIONS – FINAL REMARKS



R. Rodrigues et al, Ohmic heating – an emerging technology in innovative food processing, Ch 7, in Sustainable Production Technology in Food, Academic Press, 2021