GREEN EXTRACTION OPTIMIZATION OF PHENOLIC COMPOUNDS FROM OLIVE POMACE BY PRESSURIZED ETHANOL-WATER

KATSINAS N.[1], COCERO M.J.[1], BENTO DA SILVA A.[6,7], ENRIQUEZ-DE-SALAMANCA A.[2,3], FERNANDEZ N.[4], BRONZE M.R.[4,5,6], RODRIGUEZ ROJO S.[1]

[1]High Pressure Processes Group, Dpt. of Chemical Engineering and Environmental Technology, University of Valladolid (UVa), Spain. [2]Institute of Applied Ophthalmobiology (IOBA), UVa,Valladolid, Spain. [3]Biomedical Research Networking Center in Bioengineering, Biomaterials and Nanomedicine (CIBER-BBN), Madrid, Spain. [4]Instituto de Biologia Experimental e Tecnológica (iBET), Oeiras, Portugal. [5]Instituto de Tecnologia Química e Biológica da Universidade Nova de Lisboa (ITQB NOVA), Oeiras, Portugal. [6]Faculty of Pharmacy, University of Lisbon (FFULisboa, Lisbon, Portugal. [7]Faculdade de Ciências e Tecnologia da Universidade Nova de Lisboa (FCT NOVA), Caparica, Portugal.

1st Greenering International Conference, Virtual, 15–17 February 2021

1 INTRODUCTION

Olive Pomace (OP) is the main by-product of the olive oil industry and poses a great environmental threat due to its high organic load and phenolic content. At the same time, it can be considered as a source of valuable bioactive compounds → **high content in simple phenols (i.e. Hydroxytyrosol – HT) and secoiridoids (i.e. Oleuropein – OL, Oleacein – OLE)** with numerous biological activities reported, such as **antioxidant, anti-inflammatory, anti-carcinogenic, etc**.

Furthermore, 2-3% of the OP weight consists of **lipids/non-polar compounds**, from which the commercialized "OP oil" is produced. A defatting pre-treatment can not only contribute to a better phenolic extraction, but also add an extra value to the olive oil industry. Also, after the recovery of the lipophilic and phenolic components, the remaining waste can have plenty of **further potential applications in a biorefinery framework, i.e. bioenergy, agronomic uses, etc.**

2 METHODS & RESULTS

DEFATTING PRE-TREATMENT

1. **Freeze drying:** OP with ca. 3% moisture

2. **Conventional extraction**

0.5 g OP/mL *n*-hexane

30ºC, 15 min, under stirring

VS

Supercritical cCO_2 extraction ($scCO_2$)

30 MPa, 60ºC,

10.5 kg CO_2/h for 3h

3. **Residual oil recovery**

Conventional:

2.4±0.5% of the dry basis

$scCO_2$:

2.40±0.15% of the dry basis

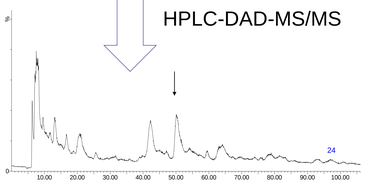
Similar oil recovery → $scCO_2$ selected, as a green technique

PHENOLIC EXTRACTION

Conventional solid-liquid extraction

0.5 g $scCO_2$ defatted OP/mL 50% EtOH-H_2O, 70ºC, 1h, under stirring

HPLC-DAD-MS/MS



OLE: most abundant polyphenol

VS

Pressurized Liquid Extraction (PLE)

Central composite design (CCD;24 experiments):

- Five levels (±1, ±a, 0)
- 10 center points

Three main parameters selected:

- **Temperature (T):** 65.0-185.0ºC
- **EtOH% in H_2O:** 8.0-92.0%
- **Solid/Liquid ratio (S/L):** 0.2-0.8 g $scCO_2$ defatted OP/mL solvent

Responses:

- Extract richness in OL, OLE and HT
- ORAC Antioxidant Activity (AA)

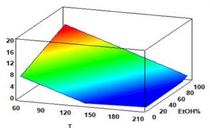
Dry Extract (DE) Characterization

HPLC-DAD

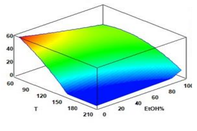
ORAC AA

Optimal PLE conditions: comparison with conventional

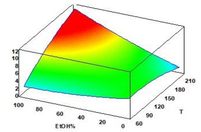
OL	OLE	HT	AA
475% increase	373% increase	428% increase	89% increase: 8.9 vs 4.7 $mmol_{TE}/g_{DE}$
13.8 vs 2.4 mg_{OL}/g_{DE}	52 vs 11 mg_{OLE}/g_{DE}	9.5 vs 1.8 mg_{HT}/g_{DE}	



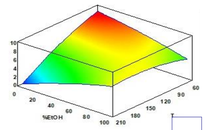
T=66.4ºC
EtOH%=92.0%
S/L=0.8 g/mL



T=66.1ºC
EtOH%=19.3%
S/L=0.8 g/mL



T=183.9ºC
EtOH%=90.0%
S/L=0.8 g/mL



T=66.1ºC
EtOH%=8.0%
S/L=0.8 g/mL

Pearson correlation with OL, OLE & HT

Positive correlation with OLE (r=0.634) statistically significant (P<0.01)

3 CONCLUSIONS

- Two sustainable techniques were combined, producing highly concentrated phenolic extracts in 3 times shorter extraction time (from 1h to 20min) and 1.6 times fewer solvent volume (from 0.5 to 0.8 g OP/mL solvent), and transforming an environmentally hazardous agro-industrial by-product to an extract of great biological interest.
- $scCO_2$ was able to remove the remaining oil similarly to the conventional method, excluding toxic organic solvents. The PLE CCD system achieved a clear enhancement of the DE richness, using exclusively green solvents. Different conditions were found to optimize each response, establishing a selective, scalable and effective process.

EU, H2020 Marie Sklodowska-Curie Actions ITN "IT-DED3" (grant agreement No 765608); UVa, postdoctoral contract (Soraya Rodríguez Rojo); Financial support from Fundação para a Ciência e Tecnologia (FCT) and Portugal 2020 to the Portuguese Mass Spectrometry Network (Rede Nacional de Espectrometria de Massa RNEM; LISBOA 01 0145 FEDER 402 022125).

Poster ID: D12001