Membrane Filtration of Agro-industrial Wastewaters and Isolation of Organic Compounds with High Added Value

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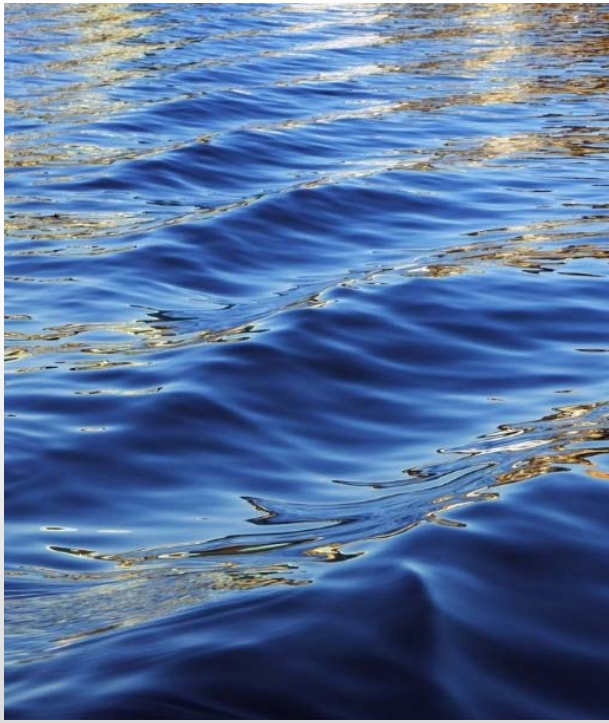
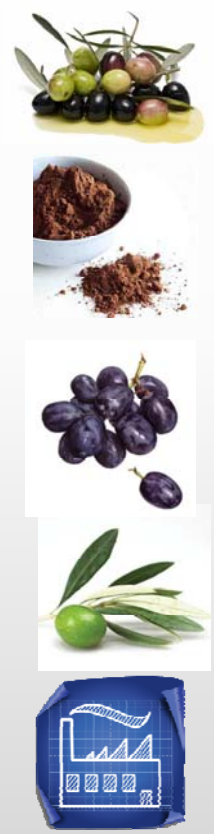


Presentation Outline



- Purification of olive mill wastewater phenols
- Purification of grape marc phenols
- **Purification of olive leaf phenols**
- Preliminary design of phenols purification plant

Scope

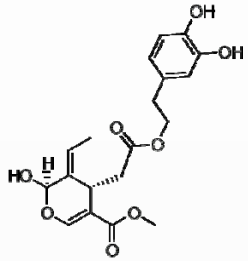
- Large amounts of agricultural byproducts are produced every year, some of them rich in phenolic compounds.
- Phenols are antioxidants with high-added value and positive effects to the human health.
- Their separation for the production of cosmetic products, food supplements etc., is of great interest.
- For this purpose, a combination of solid-liquid extraction, membrane filtration and resin adsorption/desorption following by evaporation is proposed, for the production of phenolic concentrates.
- The final products of the proposed process contain a large percentage of the byproducts' phenolic content, in a small fraction of the initial volume.
- This technique, after modification, can be applied to a variety of phenol-rich byproducts, allowing the operation of phenol separation plant adjustable to local agricultural activities.

Physicochemical Separation Techniques

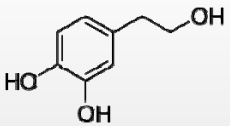
- **Solid-liquid extraction** is the separation of target compounds from a solid matrix through the use of the appropriate solvent.
- **Membrane filtration** is a separation technique that has many applications in chemical process industries.
- **Adsorption** is the selective separation of a solute (adsorbate) from a mixture, which is concentrated on the surface of a solid (adsorbent).
- **Vacuum Evaporation** for the final condensation of the isolated compounds

Olive Mill Wastewater Phenolic Compounds

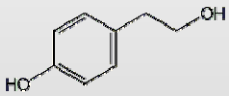
- Olive mill wastewater (OMW) is a byproduct of the three-phase extraction systems during the production of olive oil.
- Because of their partition coefficient, most phenolic compounds of olive fruits end up in the wastewater produced and not in olive oil.
- Oleuropein is the most common phenolic compound of unripe olive fruits, but during maturity it is hydrolyzed to several simpler phenolic compounds like hydroxytyrosol and tyrosol.



Oleuropein

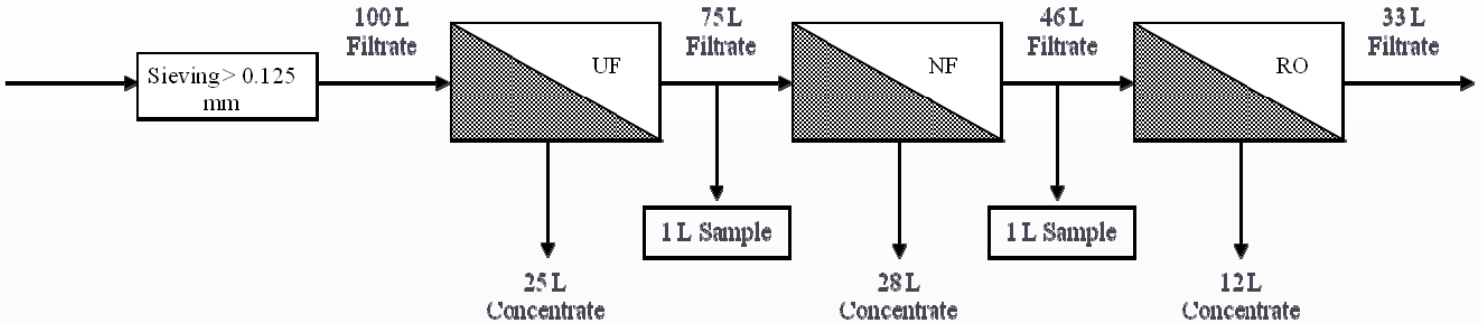


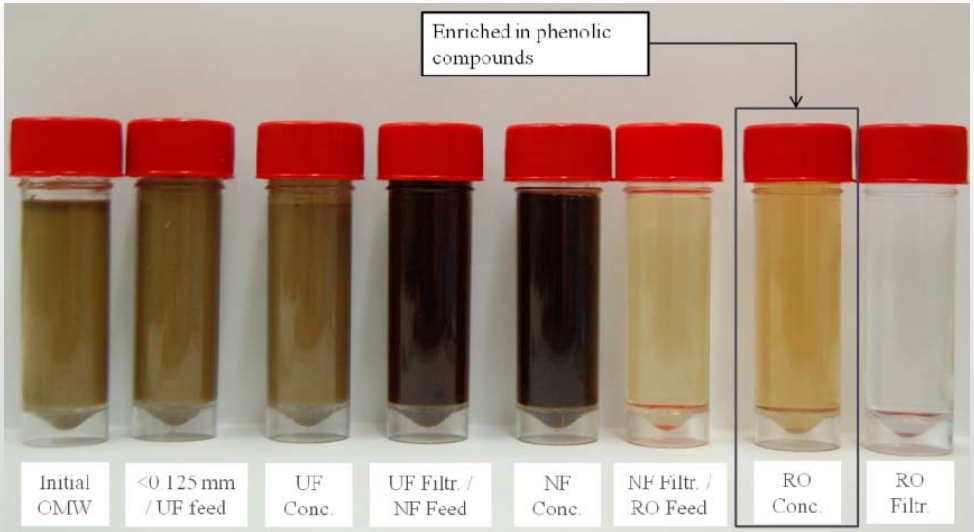
Hydroxytyrosol



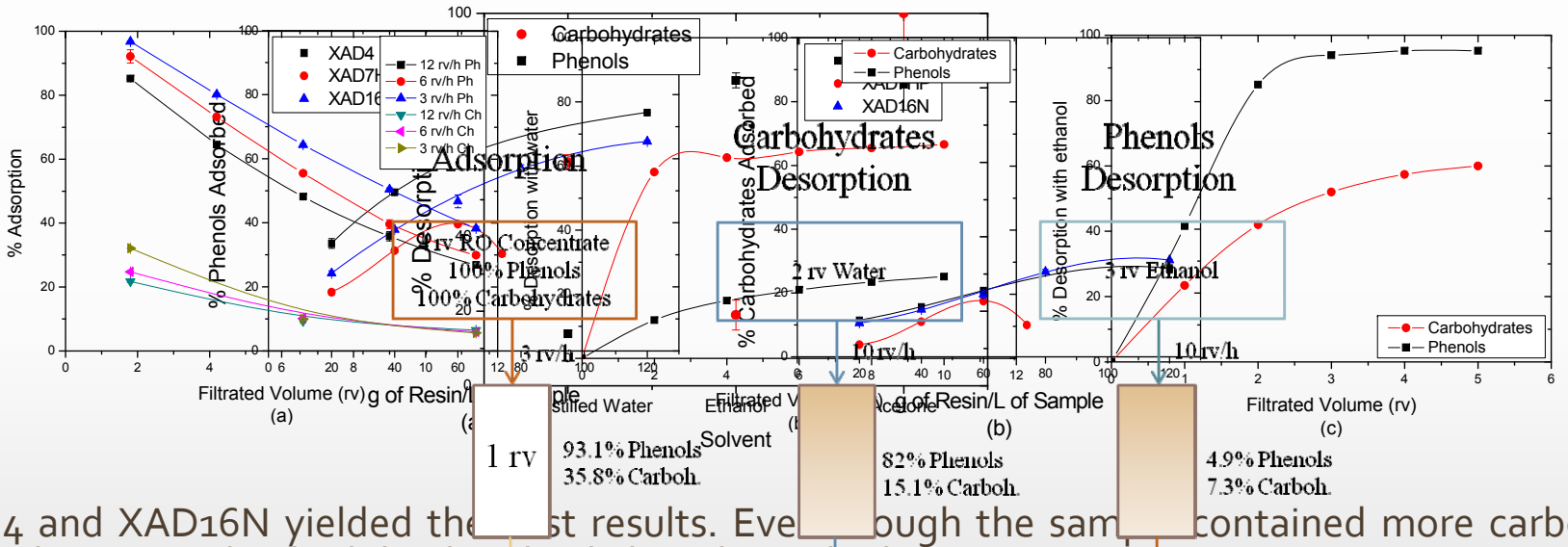
Tyrosol

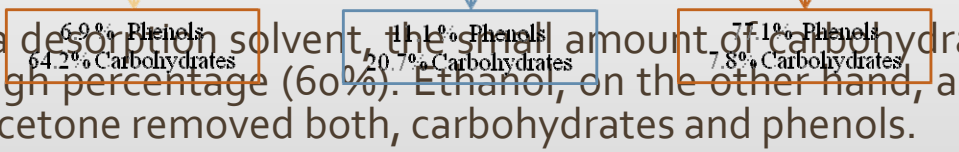
Membrane Filtration of OMW





Resin Adsorption/Desorption of OMW RO_c





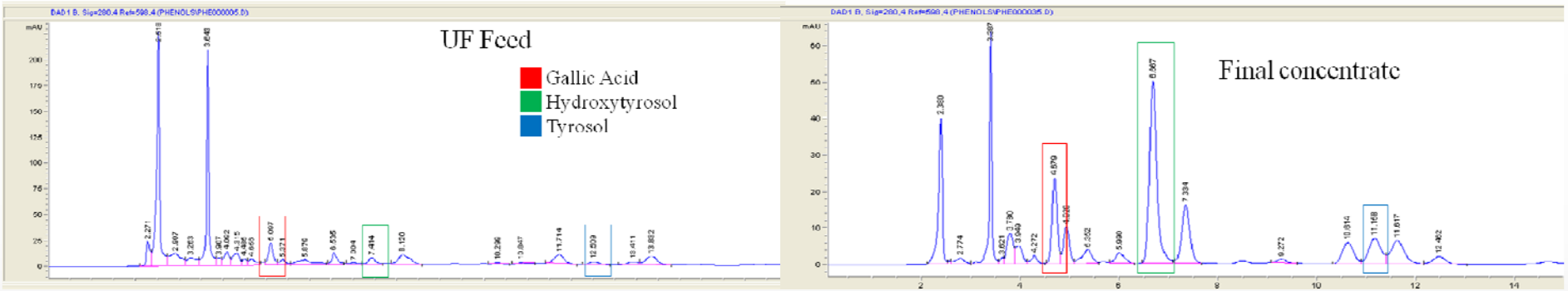
- XAD4 and XAD16N yielded the best results. Even though the sample contained more carbohydrates than phenols, resins adsorbed the dissolved phenols at a higher percentage.
- When water was used as a desorption solvent, the small amount of carbohydrates that was adsorbed on the resin was desorbed at a high percentage (60%). Ethanol, on the other hand, almost selectively removed the adsorbed phenols, while acetone removed both, carbohydrates and phenols.
- Kinetic experiments allowed the optimization of flow rates and total volume of treated sample before the resin surface was saturated.

Final Concentrate of OMW Phenolic Compounds

- After carbohydrates removal via the proposed resin process, the **distillation under vacuum** (-0.95 bar, 55 °C) of the resin ethanolic effluent resulted to a final phenol concentration of 378 g/L in gallic acid equivalents in the distillation residue.

	Initial OMW	RO concentrate	Ethanolic resin effluent	Distillation residue
Volume, mL	16700	2000	1500	9
Phenols, g/L	2.64 *±0.04*	2.09 *±0.02*	2.36 *±0.01*	**377.50** *±8.34*
Carbohydrates, g/L	12.34 *±0.49*	14.96 *±0.03*	3.84 *±0.01*	293.92 *±1.28*

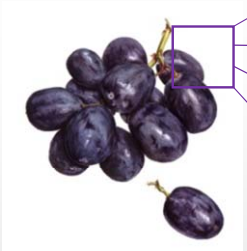
HPLC Analysis of Simple Phenols

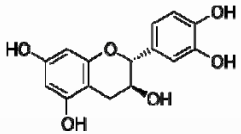


- The only phenolic compounds detected out of the ones tested were gallic acid, hydroxytyrosol (HT) and tyrosol with HT being the dominant phenol.
- No phenols were in detectable levels in the RO filtrate. The membrane process purified the low-molecular-weight phenols through sieving of all the compounds according to their molecular weight.
- The resin process further purified the phenols from the rest of the low-molecular-weight compounds according to their polarity.
- After vacuum distillation, the phenolic compounds appear to withstand the heat process and the final concentration of HT obtained in the distillation concentrate is around 85 g/L.

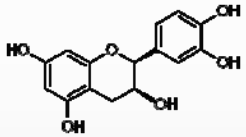
Grape Marc Phenolic Compound

- Grape cultivation is one of the most important agricultural activities in the world with most of the produced grapes used in winemaking.
- In the winemaking process a significant amount of solid byproducts is produced originating from the skin and seeds of grapes after the juice extraction.
- Although part of the phenolic content of the grapes is transferred to the juice and later wine, the solid byproducts are rich in phenols, allowing the production of phenol-rich extracts

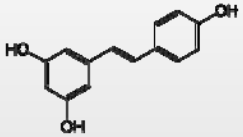




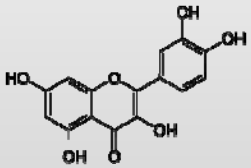
Catechin



Epicatechin

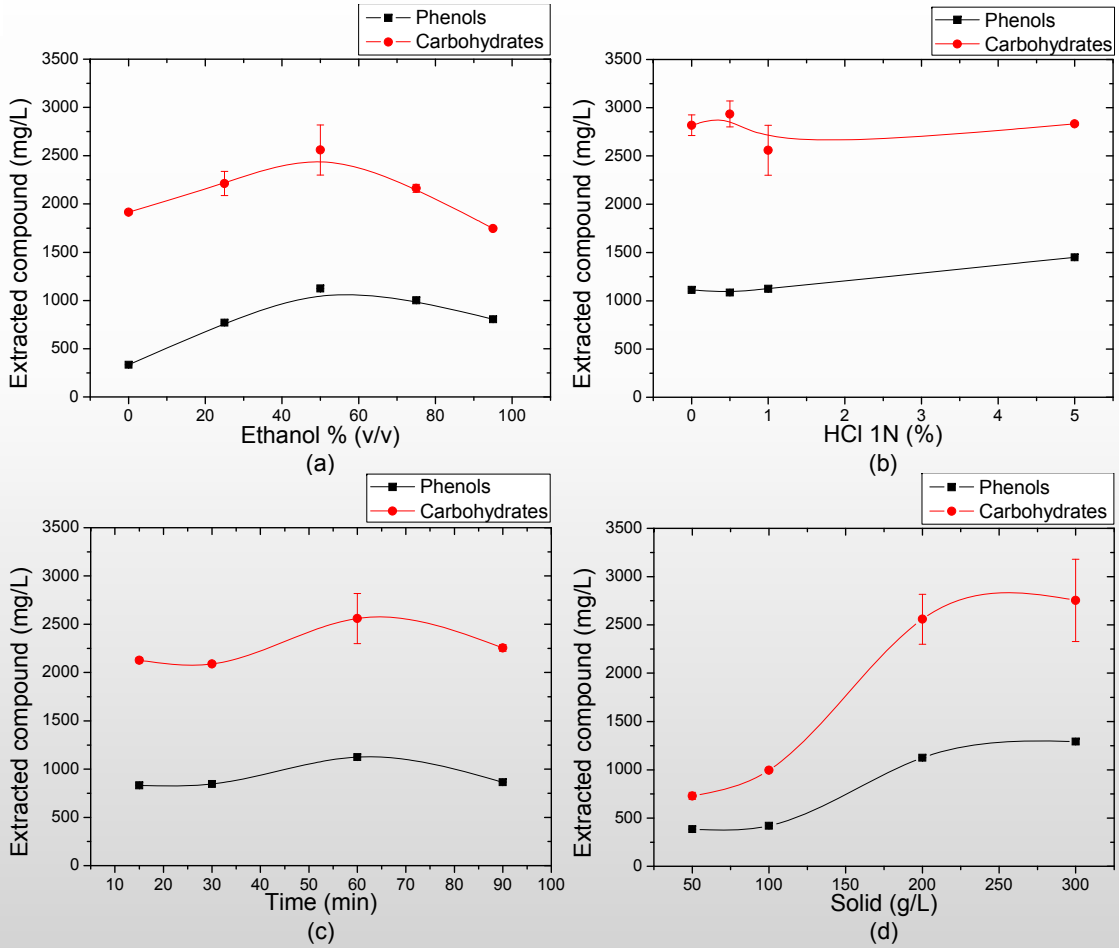


trans-Resveratrol



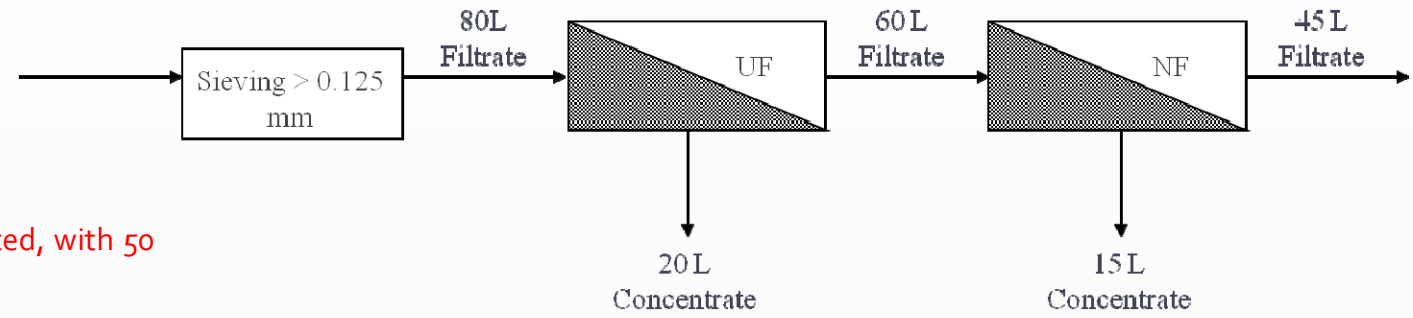
Quercetin

Extraction of Grape Marc Phenolic Compounds

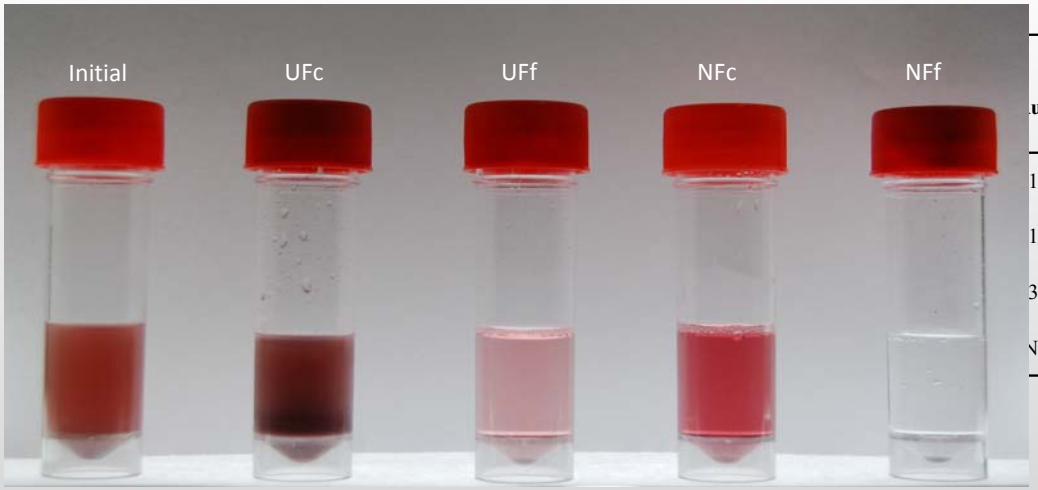


Optimum extraction conditions	
Ethanol %	50
HCl 1N %	1
Duration	15 min
Solids/Solvent	200 g/L
Double extraction	

Membrane Filtration of Grape Marc Extract



- 20 kg of grape marc were extracted, with 50 L of extract occurring.
- Prior to membrane filtration, the extract was sieved through stainless steel sieves with final pore diameter 0.125 mm. and ethanol was partly removed through distillation leading to 15 L of residue with 14% v/v ethanol (compared to 50% v/v). The extract was then diluted with water to 80 L.



utin
1.4
1.8
3.0
N/D

Final Concentration of Grape Marc Phenolic Compounds

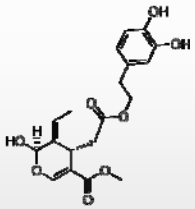
- With the removal of carbohydrates from the NF_C, further concentration can be achieved through evaporation. For this purpose 2.4 L of NF_C were treated with the proposed resin process, leading to the production of 0.64 L of ethanolic effluent that was evaporated under vacuum (0.05 bar, 50 °C). The final concentrate had a volume of 5 mL.

Sample	Volume mL*	Total carbohydrates	Total phenols	Catechin	Quercetin	Epicatechin	Rutin
		mg/L					
Desorbed	4000	1951	3023	65.0	<1	13.0	31.0
Evap.	31	112333	**190850**	**4746**	60	853	381

* The resin process was not applied in all of the NF_C, the volumes presented occurred after scaling of the results.

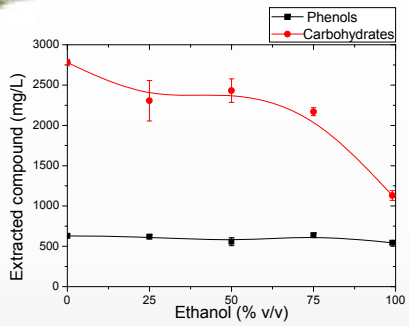
Olive Leaf Phenolic Compounds

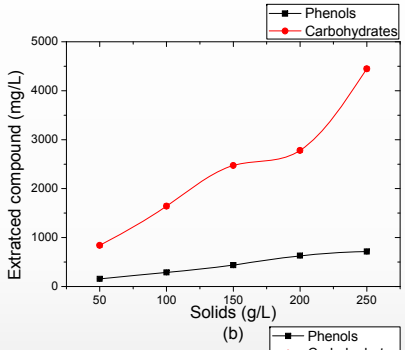
- Olive leaves are a byproduct of olive fruit harvesting and initial stages of olive oil extraction, during their separation from olive fruits.
- Olive leaf extracts have been proven to be rich in phenolic compounds, with the most prominent one being **oleuropein**, which, unlike in the olive fruit, it is not hydrolyzed to simpler phenols.
- Oleuropein can be either bound to a sugar molecule (Oleuropein glycoside) or be present in its free form (Oleuropein aglycon).

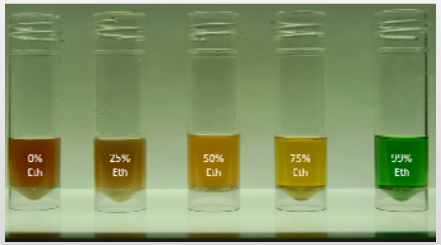


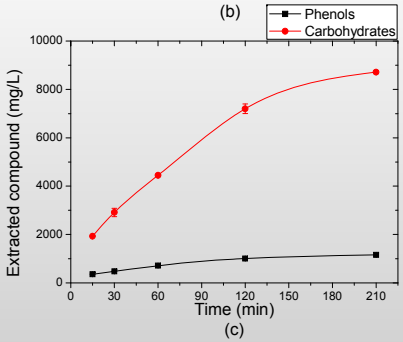
Oleuropein

Extraction of Olive Leaf Phenols



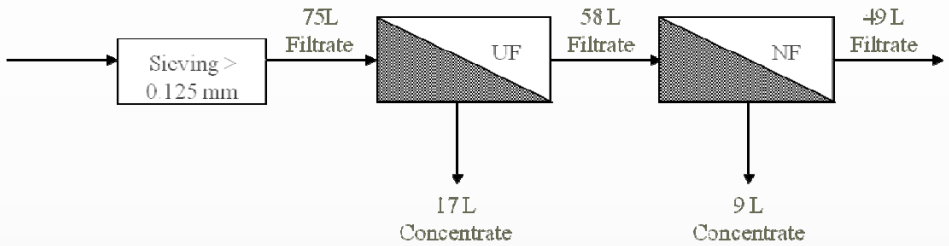




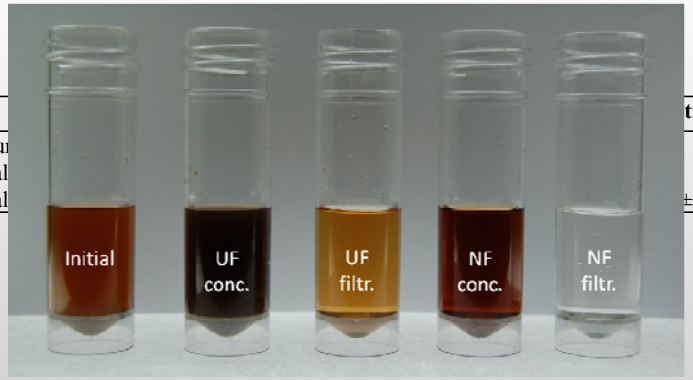


Optimum extraction conditions	
Ethanol %	0
Duration	120 min
Solids/Solvent	250 g/L

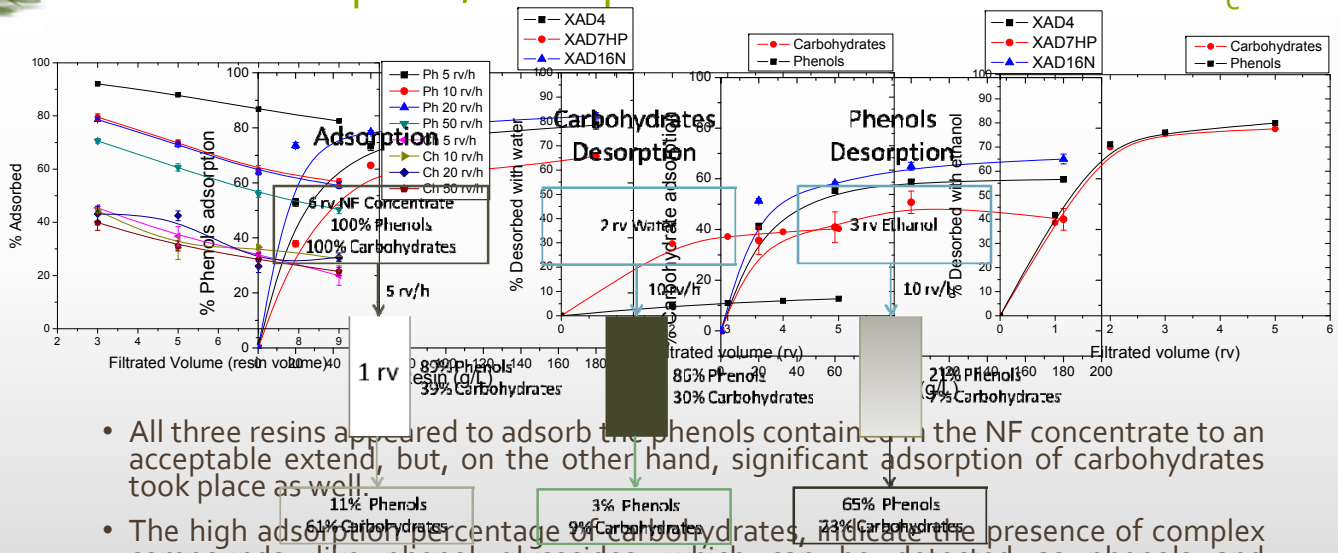
Membrane Filtration of Olive Leaf Extract



		tr.
Volu		
Total		
Total		±24



Resin Adsorption/Desorption of Olive Leaf Extract NF_c



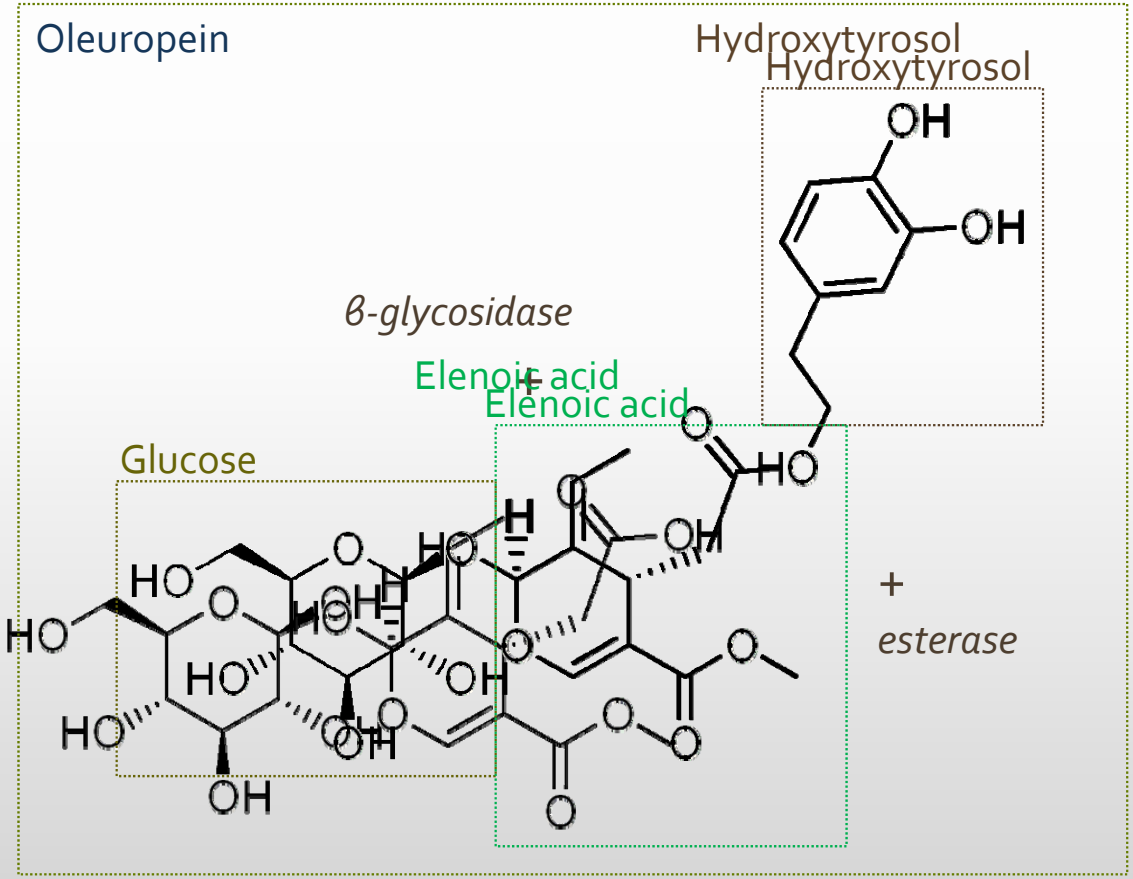
- All three resins appeared to adsorb the phenols contained in the NF concentrate to an acceptable extend, but, on the other hand, significant adsorption of carbohydrates took place as well.
- The high adsorption percentage of carbohydrates, indicate the presence of complex compounds, like phenol glycosides, which can be detected as phenols and carbohydrates.

Final Concentration of Olive Leaf Phenols

- 1.44 L of NF concentrate were treated with the proposed resin process, leading to the production of 0.72 L of ethanolic effluent that was evaporated under vacuum (0.05 bar, 50 °C). The final concentrate had a volume of 10 mL .

	Volume mL	Total Phenols mg/L	Total Carbohydrates mg/L
NFc	1440	988 ±25	5410 ±37
Desorbed	720	1480 ±1	5260 ±35
Final concentrate	10	97890 ±1230	322333 ±3933

Oleuropein-glycoside



Olive Leaf Extract-Conclusions

- With the proposed method, significant separation of olive leaf phenols was achieved. Firstly optimization of the extraction conditions was carried out, in terms of solvent ethanol percentage, solid/solvent ratio and duration.
- 20 kg of olive leaves were extracted with 80 L of water, and the extract was treated with membrane filtration. In the UF step, the suspended particles were removed, while NF concentrated the majority of the contained phenolic compounds.
- With batch adsorption experiments, XAD16N was proven to have the best adsorption behavior, and was used in resin packed beds to treat a larger amount of NF concentrate. The occurring resin ethanolic product contained 65% of the NF concentrate phenols and 23% of the contained carbohydrates.
- The ethanolic product of the resin process was finally treated with vacuum evaporation, with the finally product containing around *98 g/L phenolic compounds in gallic acid equivalents, compared to 0.5 g/L of the initial extract*.
- This separation, although significant, was affected by the presence of complex phenolic compounds, like oleuropein-glycoside, part phenols and part carbohydrates.

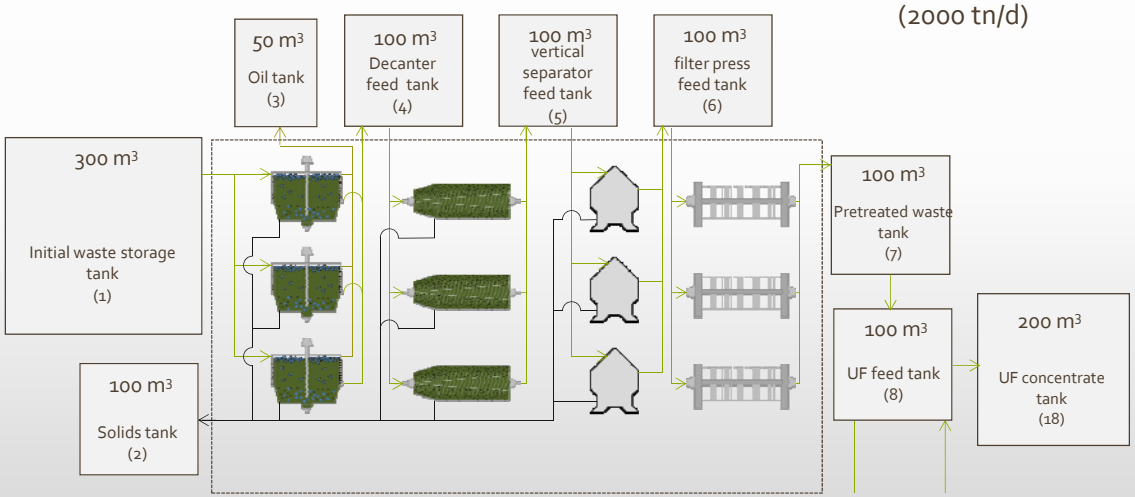
Phenols Purification Plant

- The combination of solid-liquid extraction, membrane filtration and resin adsorption/desorption could be modified and applied to a variety of byproducts rich in phenolic compounds.
- The seasonal nature of these byproducts makes the combination of different byproducts imperative for the continuous operation of the plant.
- The viability of this endeavor strongly depends on the market demand of the high-added value phenolic products and the management of the high volumes of byproducts occurring from the proposed process.
- A preliminary design of such a plant was carried out, based on the results obtained from the treatment of olive mill wastewater.

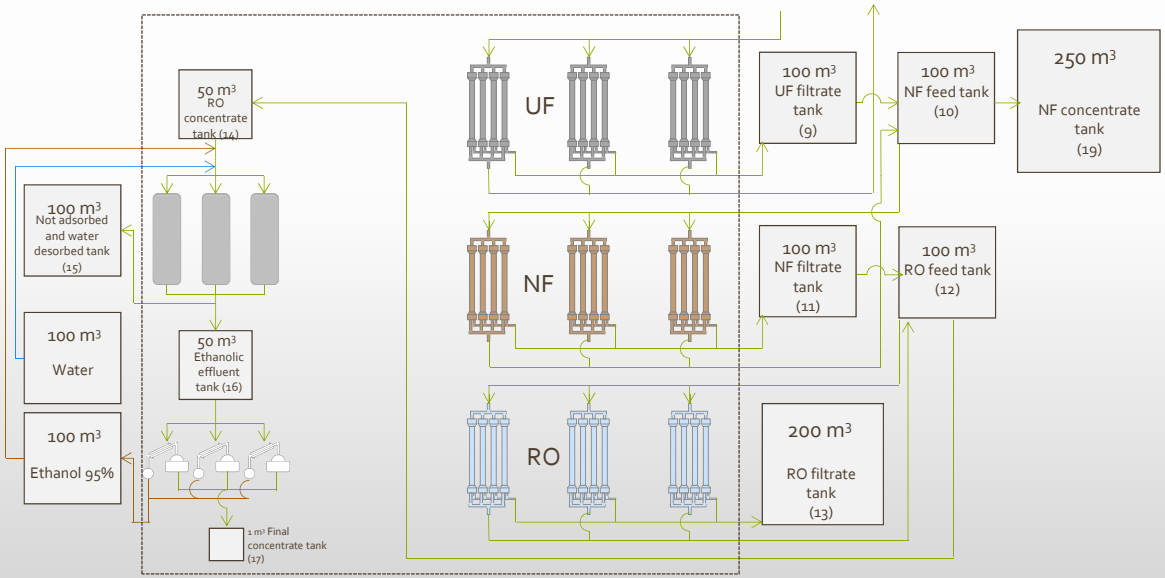
Preliminary Design of Phenols Purification Plant

Pretreatment

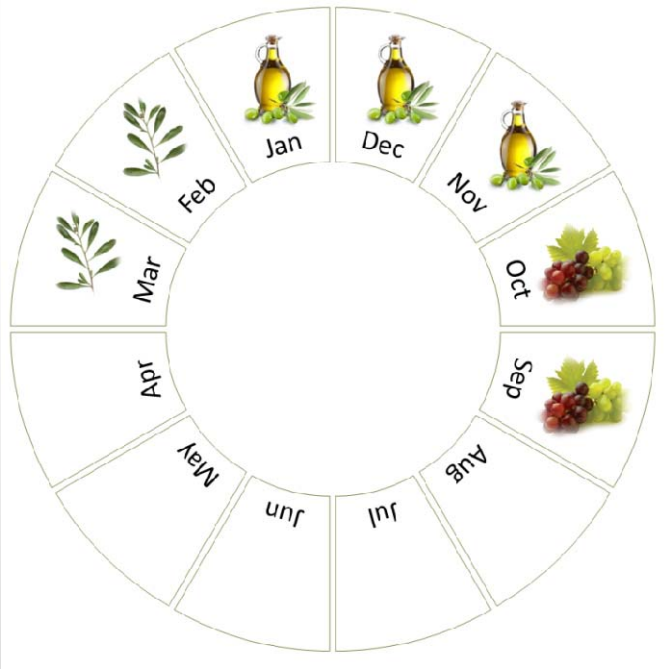
Plant byproduct treatment capacity: 110 tn/h (2000 tn/d)



Preliminary Design of Phenols Purification Plant



Preliminary Design of Phenols Purification Plant



Source	Main product	Harvesting period
Tomato byproducts	Quercetin, Hydroxycinnamic acids and lycopene	May-August
Coffee byproducts	Hydroxycinnamic acids	All year
Citrus byproducts	Hesperidin	November-March
Apple, pear byproducts	Hydroxycinnamic acids	September-January
Strawberry byproducts	Anthocyanins	May-July
Mediterranean aromatic plants (dyctamus, marjoram, vitex, teucrium, rosemary)	Phenolic acids	June-August

General Conclusions

- Solid-liquid extraction, membrane filtration and resin adsorption/desorption were combined for the purification of phenols contained in olive mill wastewater, grape marc and olive leaves.
- For the solid materials examined, correct extraction was crucial for maximizing the phenolic concentration. Moreover, pretreatment of the samples can greatly affect the results, as for example reduction of olive leaf particle size may increase the amount of phenols extracted, with lower extraction durations, or defatting of cocoa powder can prevent the hindrance that was exhibited due to high fat percentage.
- During membrane filtration, the extracted compounds were fractionated according to their molecular weight. In the UF step, the solids contained in the samples were removed. The complex and higher molecular weight compounds were concentrated in the NF step, while low molecular weight at the RO step.

General Conclusions

- After the selective adsorption of the phenolic compounds of the membrane concentrate of interest, water was used to desorb the adsorbed carbohydrates and ethanol for the desorption of phenols.
- During the resin process, the solvent of the phenolic compounds was changed from water to ethanol, facilitating their further concentration through evaporation. The final product of the proposed process contains a large amount of the phenols contained in the initial plant material, in a very small fraction of the initial volume
- Apart from the plant materials examined in this study, the proposed process can be employed for the treatment of any material rich in phenolic compounds. A preliminary design of a treatment plant was presented, that can be adjusted for the extraction of phenols from regional agro-industrial byproducts, with some phenol-rich byproduct proposals as well.

I would like to thank:

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D. P. Zagklis, A. I. Vavouraki, M. E. Kornaros & C. A. Paraskeva, "Purification of olive mill wastewater phenols through membrane filtration and resin adsorption/desorption", *J. Hazard. Mater*. 285 (2015) 69-76.

D. P. Zagklis & C. A. Paraskeva, "Membrane filtration of agro-industrial wastewaters and isolation of organic compounds with high added values", *Water Sci. Technol.* 2014, 69(1), 202-207.