The possibility of using olive leaves as a source of polyphenols in the light of circular economy

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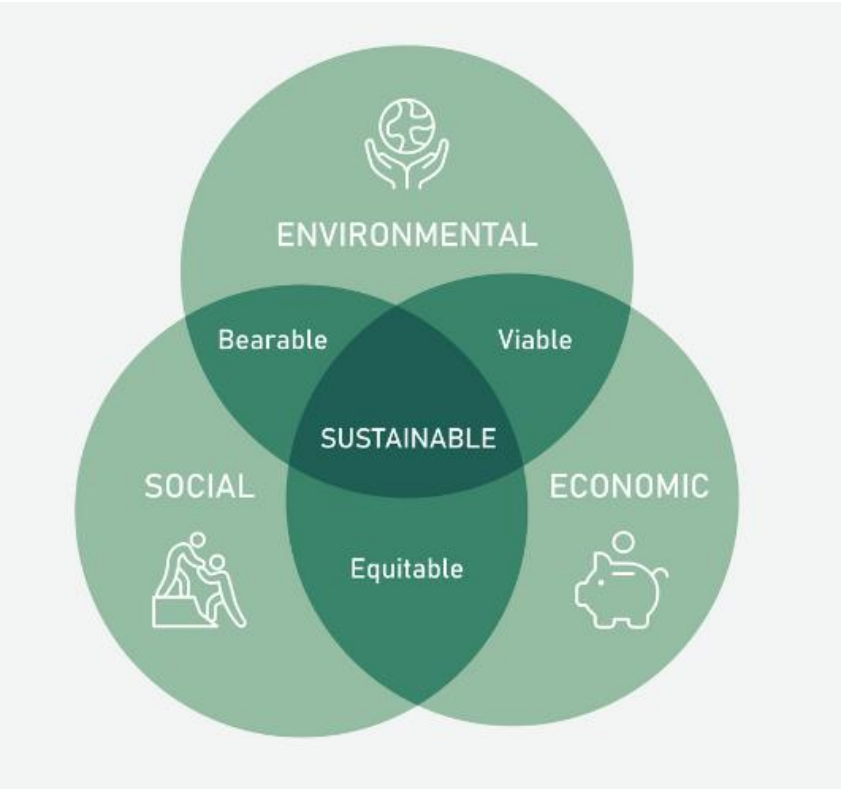
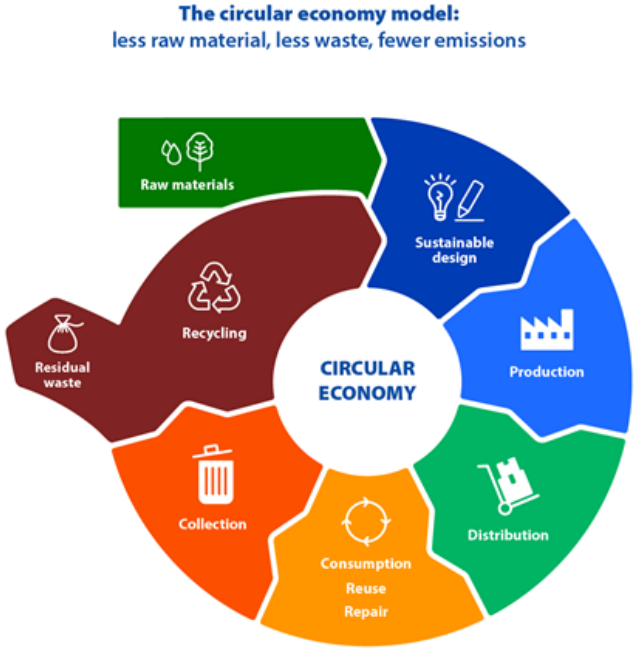
SUSTAINABILITY



Environmental sustainability focuses on:
Responsible use of natural resources
Better management of waste
Minimising carbon footprint

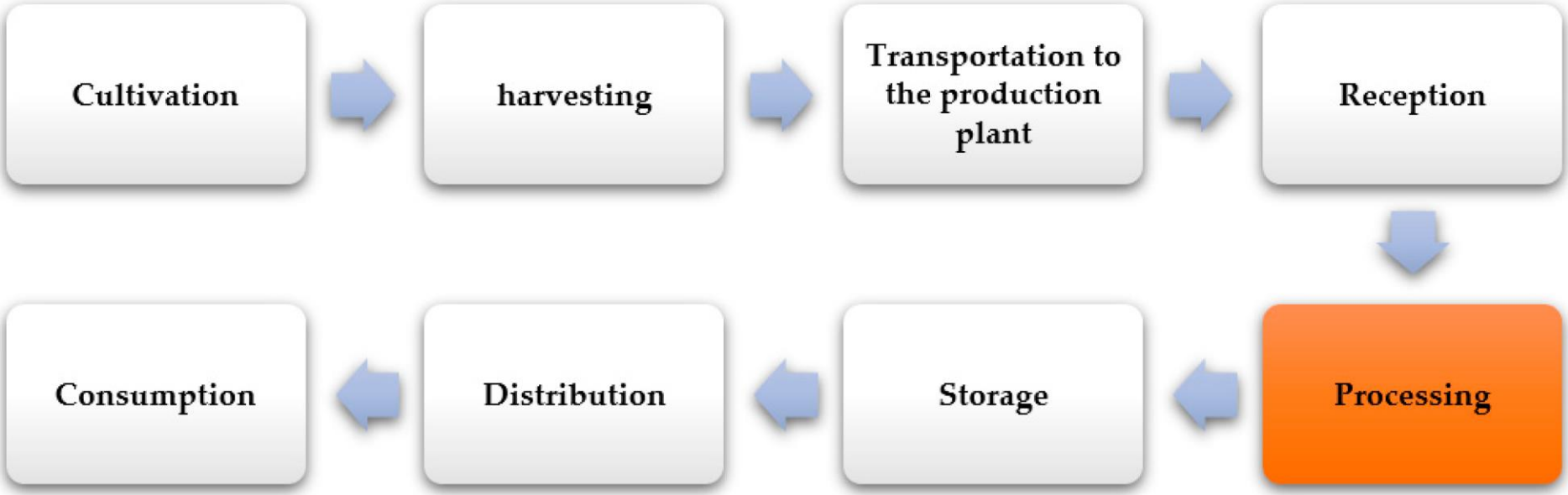
Social sustainability focuses on:
Environmental justice
Employee wellbeing
Education

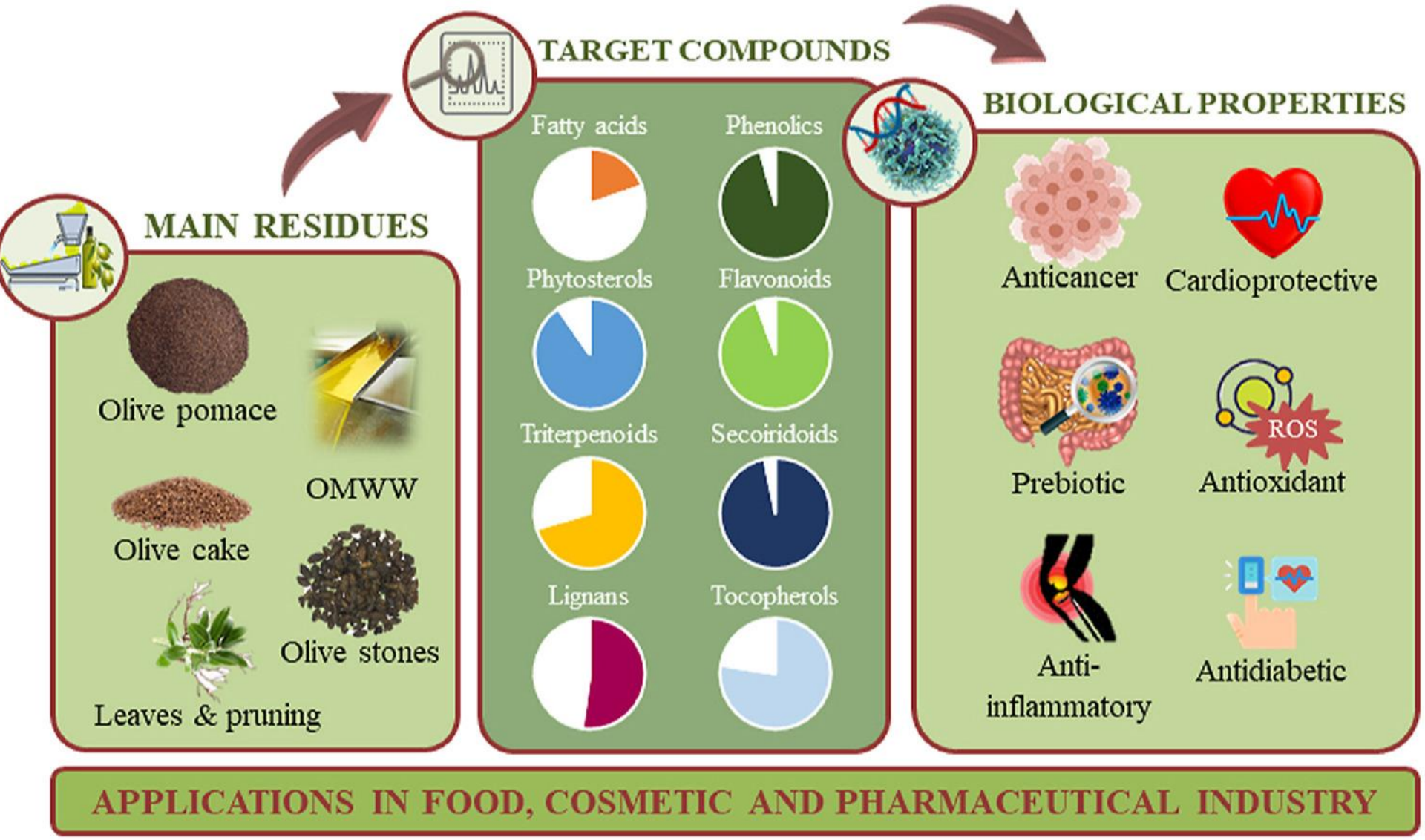


Economic sustainability focuses on:
Balancing profits with ethics
A circular economy
Job creation and stability

https://www.europarl.europa.eu/

Food waste in agri food chain





olive oil mill waste water

https://doi.org/10.1016/j.tifs.2021.09.007

Olive leaves

By-product in olive growing.
Olive leaves represent 10 % of the mass of olive tree.
On average, we get 25 kg of olive leaves per year on one pruned tree.

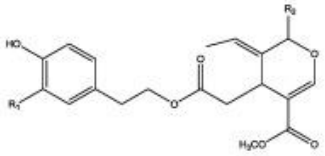
Applications:
- in cosmetics
- nutrition
- pharmaceutical industry

Lowering of total and esterified cholesterol in the blood
hypoglycemic effect, antioxidant, antimicrobial and antifungal effect

Oleuropein inhibits lipoxygenase, modulates signaling pathways, suppresses inflammation

Polyphenolic in olive tree leaves

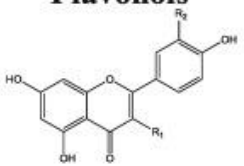
Secoiridoids



Oleuropein (R_1=OH,R_2=Glc)
Ligstroside aglycone (R_1=H,R_2=OH)
Oleuropein aglycone (R_1=OH,R_2=OH)

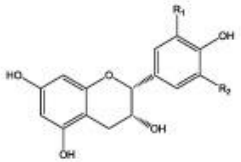
Flavonoids

Flavonols



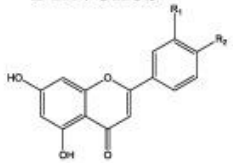
Quercetin (R1=OH,R2=OH)
Isorhamnetin (R1=OH,R2=OCH3)
Rutin (R1=Rut,R2=OH)

Flavanols



Catechin (R1=OH, R2=H)
Gallocatechin (R1, R2=OH)

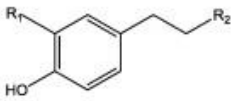
Flavones



Apigenin (R_1=H,R_2=OH)
Luteolin (R_1,R_2=OH)

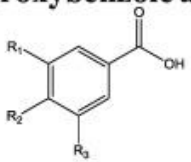
Simple phenols

Phenylethanoids



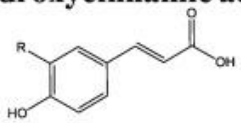
Tyrosol (R_1=H, R_2=OH)
Hydroxytyrosol (R_1, R_2=OH)

Hydroxybenzoic acids



p-hydroxybenzoic acid (R1,R3=H,R2=OH)
Gallic acid (R1,R2,R3=OH)

Hydroxycinnamic acids

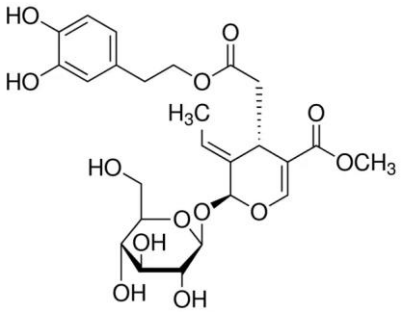


p-coumaric acid (R=H)
Caffeic acid (R=OH)
Ferulic acid (R=OCH3)

https://doi.org/10.1016/j.foodres.2015.09.011

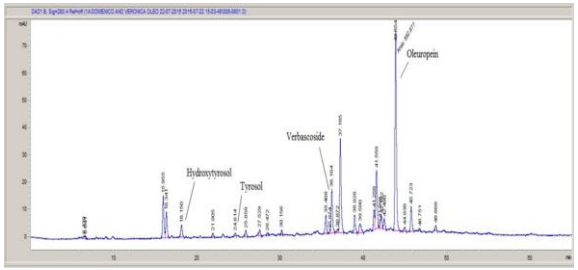
Preparation of extracts rich in phenolic compounds

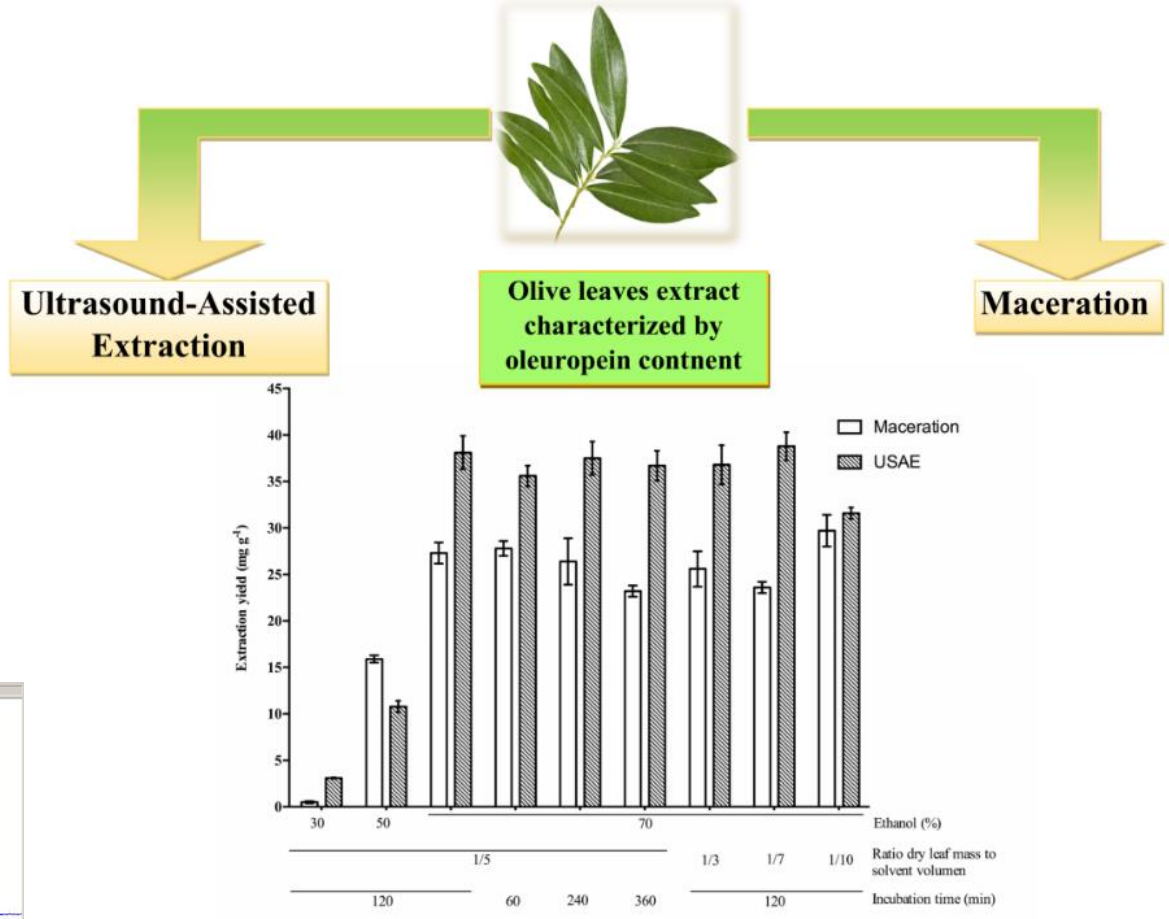
Preparation and analysis of olive leaf extracts
Optimization of the extraction process



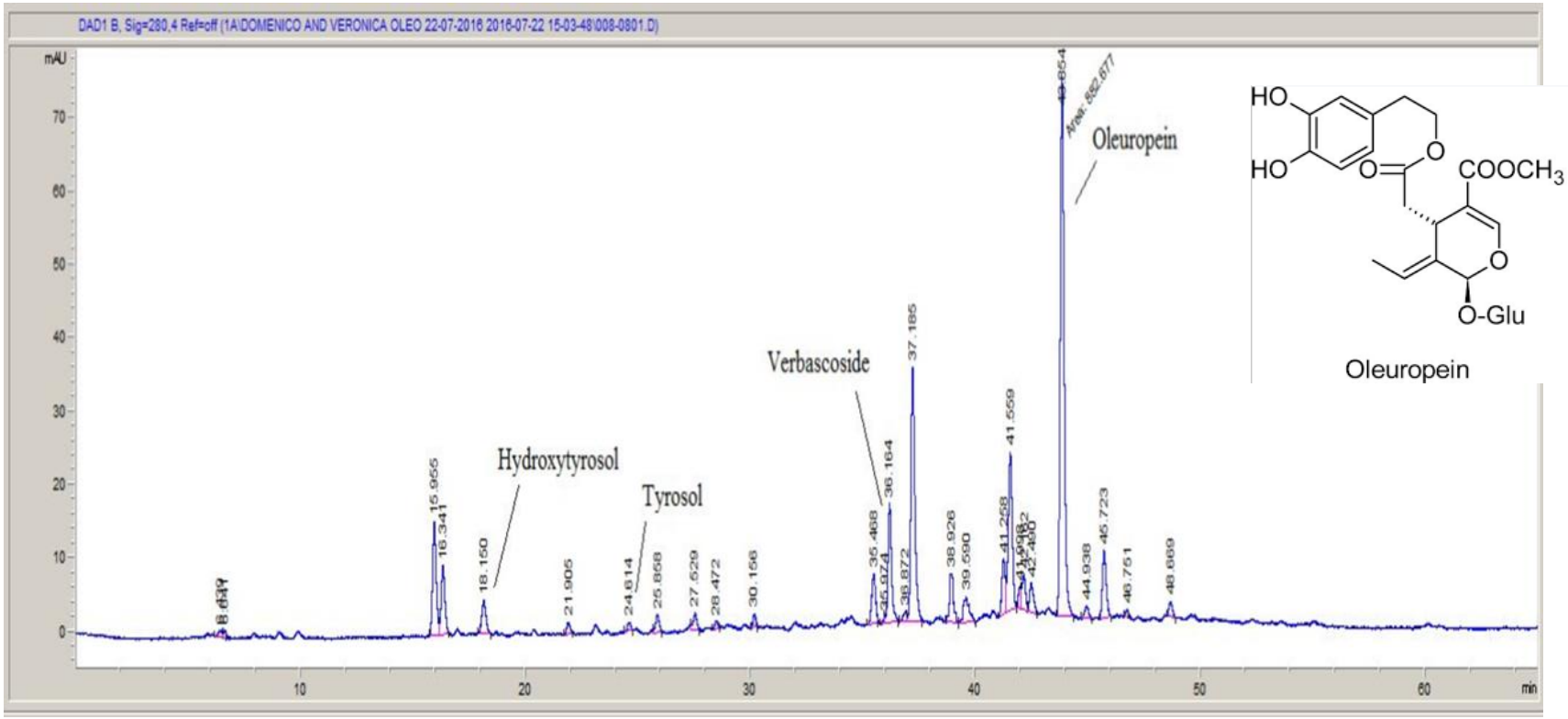
Ethanol extracts were prepared using ultrasound, lyophilized and stored.

HPLC-DAD detection of olive leaf extrac

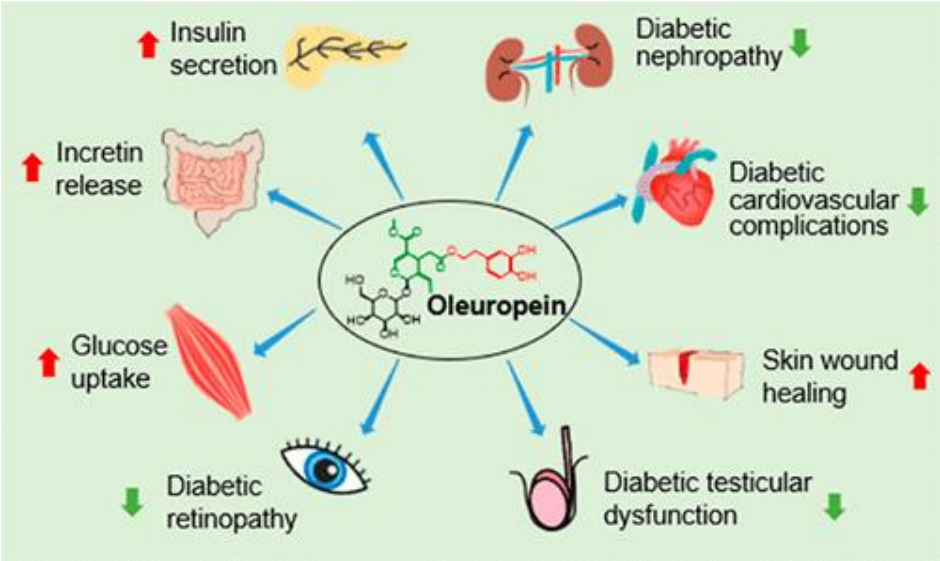




OLIVE LEAF EXTRACT



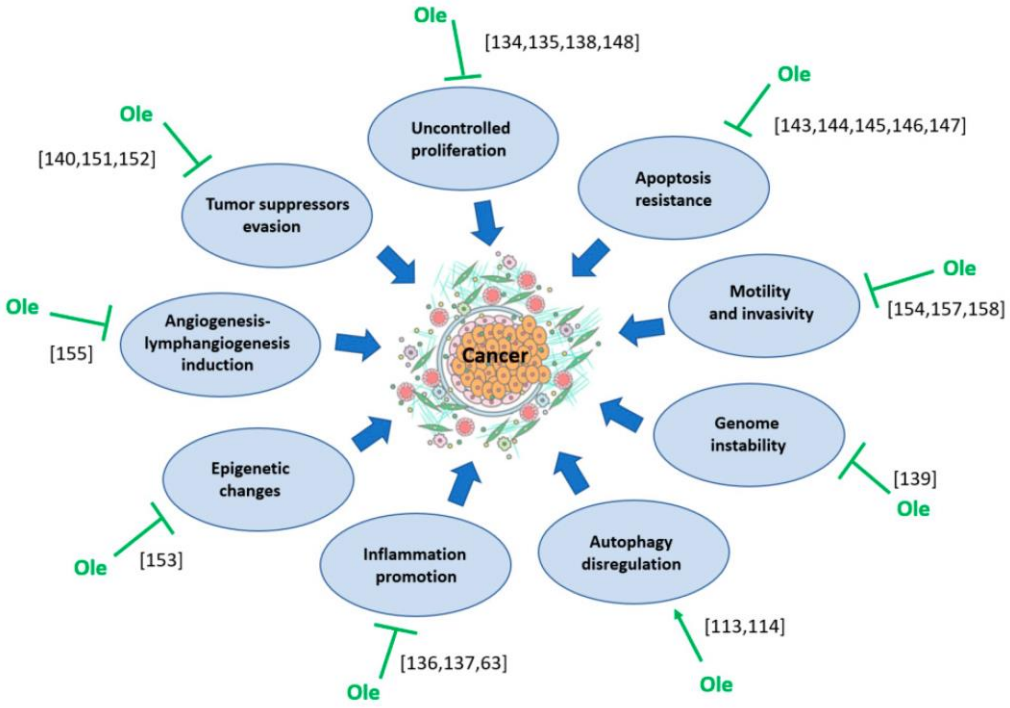
HPLC chromatograph of olive leaf extract



Efficacy and Mechanisms of Oleuropein in Mitigating Diabetes and Diabetes Complications

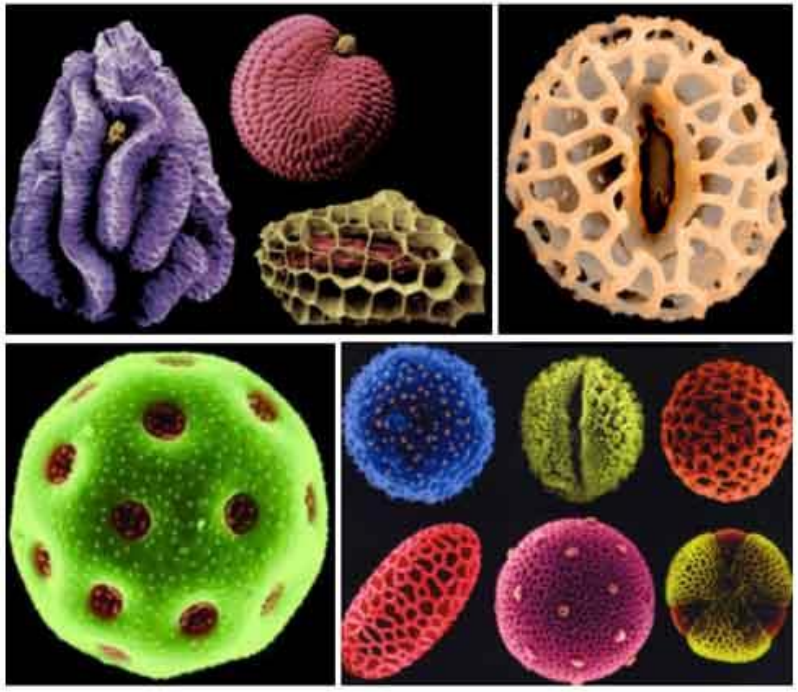
Journal of Agricultural and Food Chemistry **2021** *69* (22), 6145-6155
DOI: 10.1021/acs.jafc.1c01404

Effect of oleuropein (Ole) on the factors contributing to cancer development.



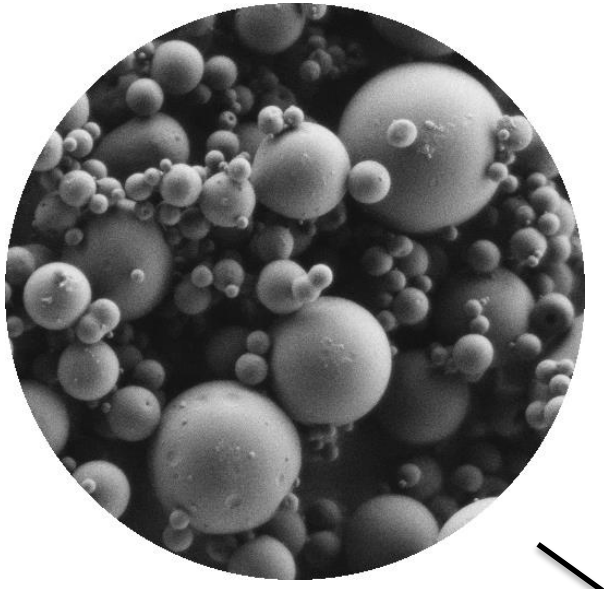
Antioxidants 2019, 8(12), 578; https://doi.org/10.3390/antiox8120578

Encapsulation technologies



http://www.world-mysteries.com/science-mysteries/cosmic-blueprints

Encapsulation of bioactive compounds



Separation of different active compounds

Homogenic distribution

Mask unpleasent taste

Controled release

Changed physicochemical properties of compound

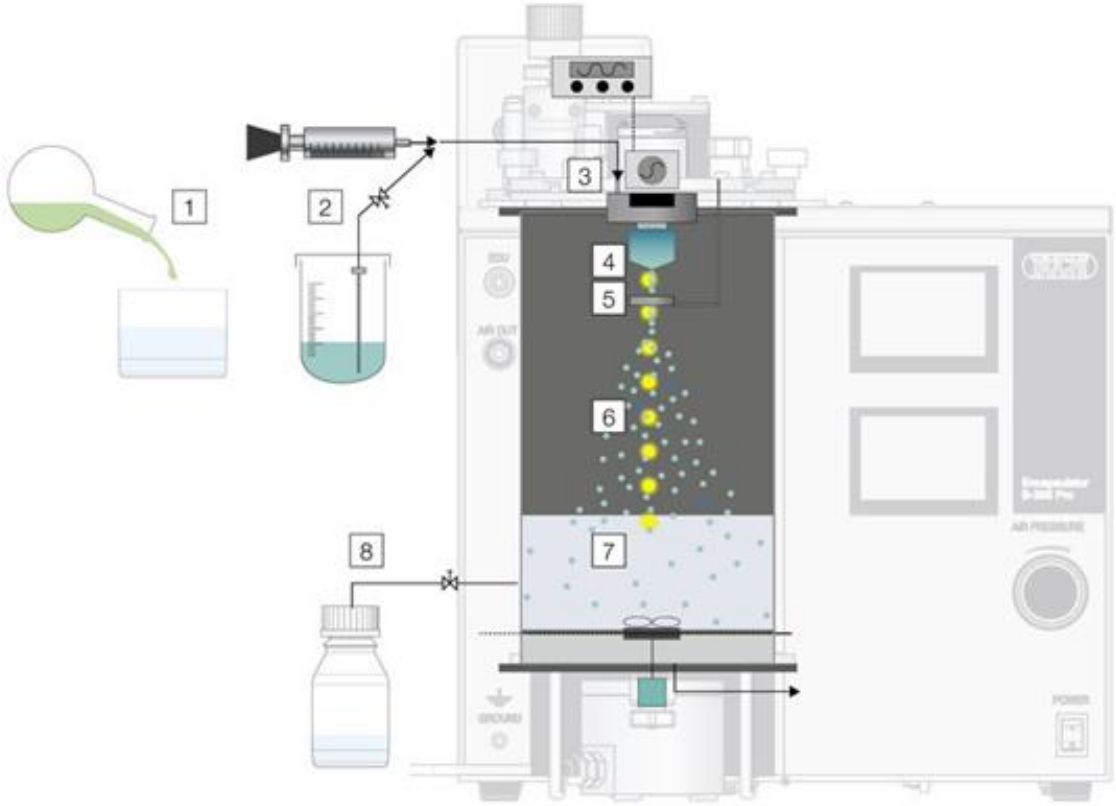
Decrease the evaporation

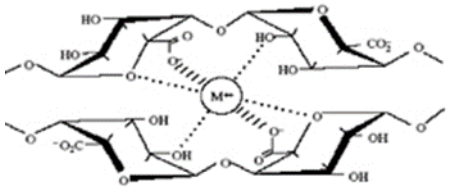
Protection against environmental factors

Preparation of extracts rich in phenolic compounds

Encapsulation of oleuropein and olive leaf extracts in alginate capsules and characterization.

Microencapsulator B-B-395 Pro, Büchi





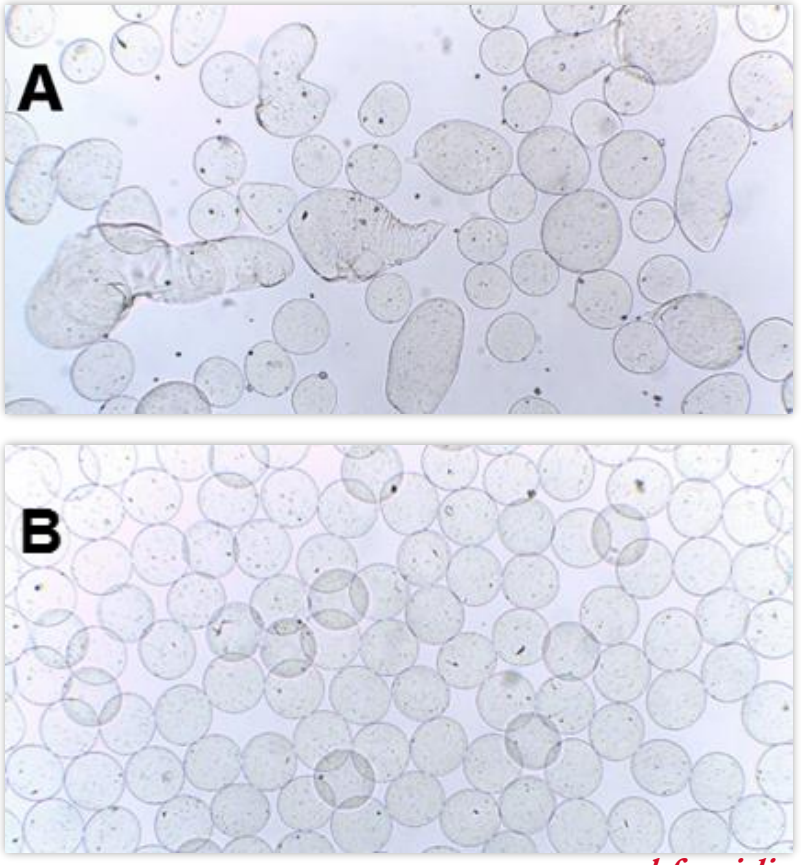
Preparation of extracts rich in phenolic compounds

Encapsulation of oleuropein and olive leaf extracts in alginate capsules and characterization.

Encapsulation efficiency	
oleuropein (pure substance) 1.8 %	olive leaf extract 4.2 %

	Release	
T(°C)	Oleuropein	Extract
25	100 %	100 %

(A) Non uniform size and non-spherical shape of capsules prepared under suboptimal conditions;
(B) Uniform size and spherical shape of capsules prepared under optimal conditions

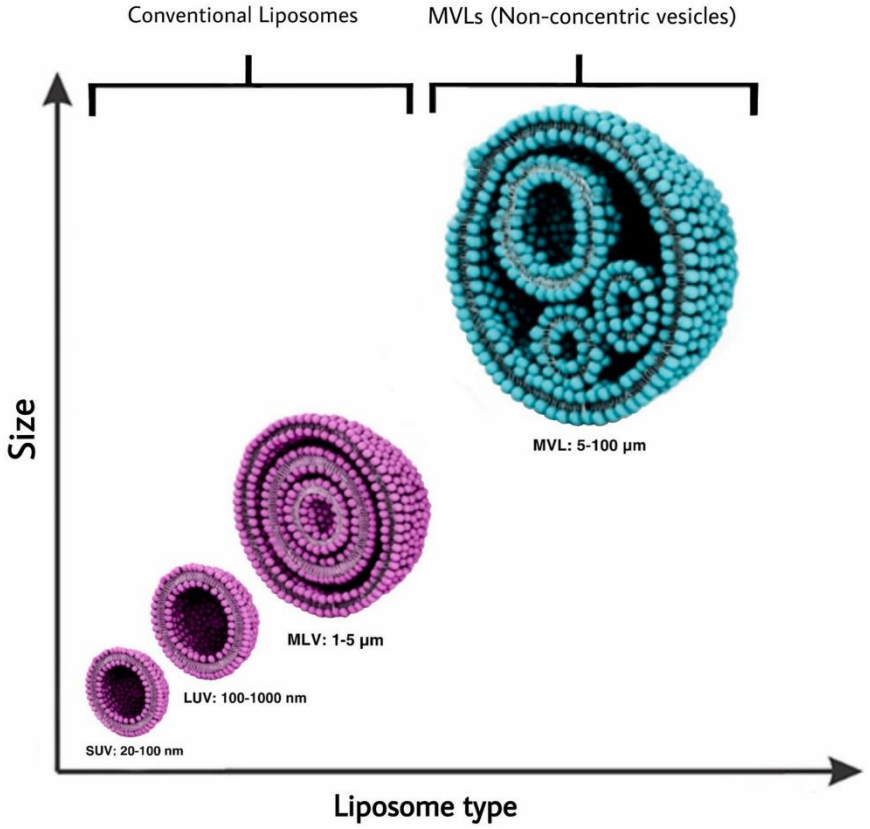


www.bf.uni-lj.si

Conclusion:
Encapsulation of olive leaf extract in alginate capsules is not sufficiently effective.
The stability of the encapsulated olive leaf extract cannot be analyzed because oleuropein is immediately released from the prepared capsules.
The development of new functional products will take place with the encapsulated extract from proliposomes.

Preparation of extracts rich in phenolic compounds

Encapsulation of oleuropein and olive leaf extracts into proliposomes



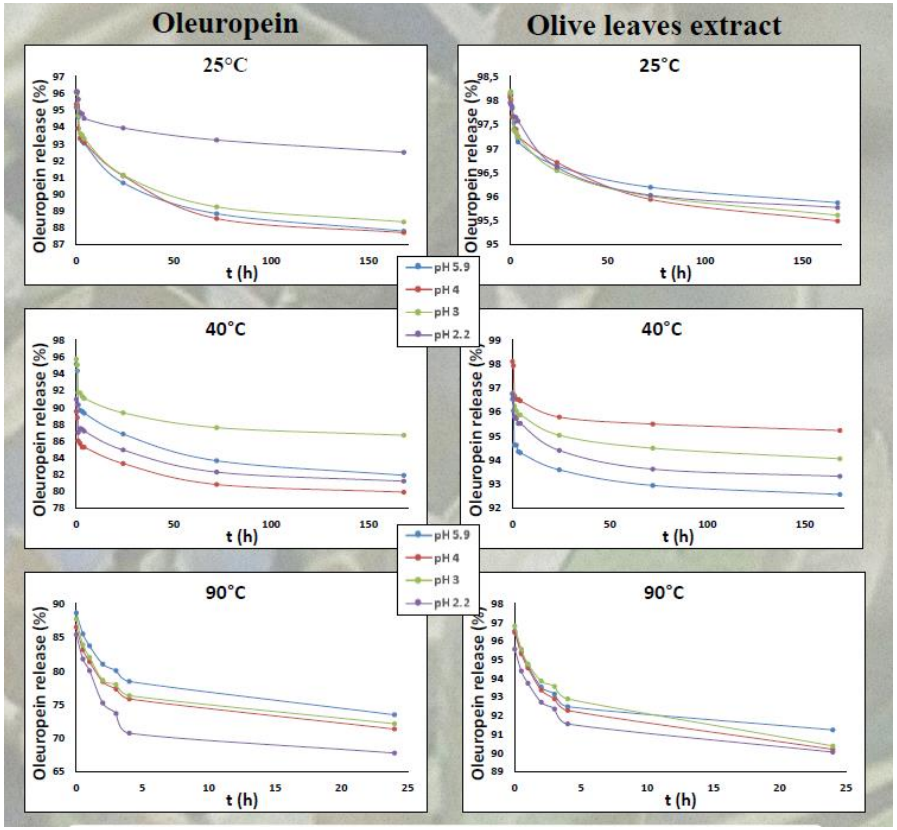
Journal of Drug Delivery Science and Technology 66 (2021) 102842

Preparation of extracts rich in phenolic compounds

Encapsulation of oleuropein and olive leaf extracts into proliposomes

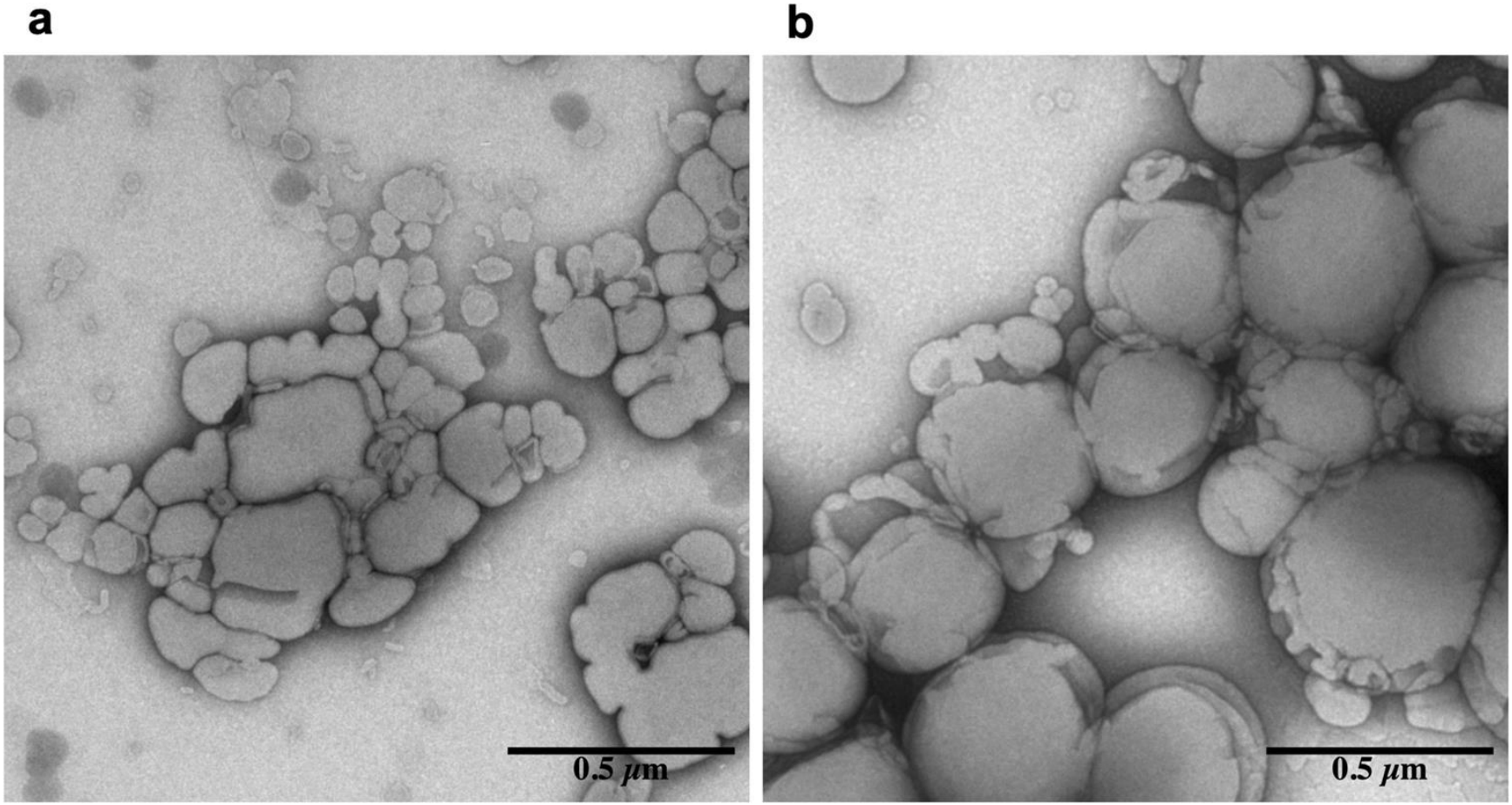
Encapsulation efficiency	
Oleuropein (pure chemical) 75 %	Olive Leaf extract 83 %

	Release	
T(°C)	Oleuropein	Olive Leaf extract
25	10.9 %	4.3 %
40	17.6 %	6.2 %
90	28.8 %	9.5 %



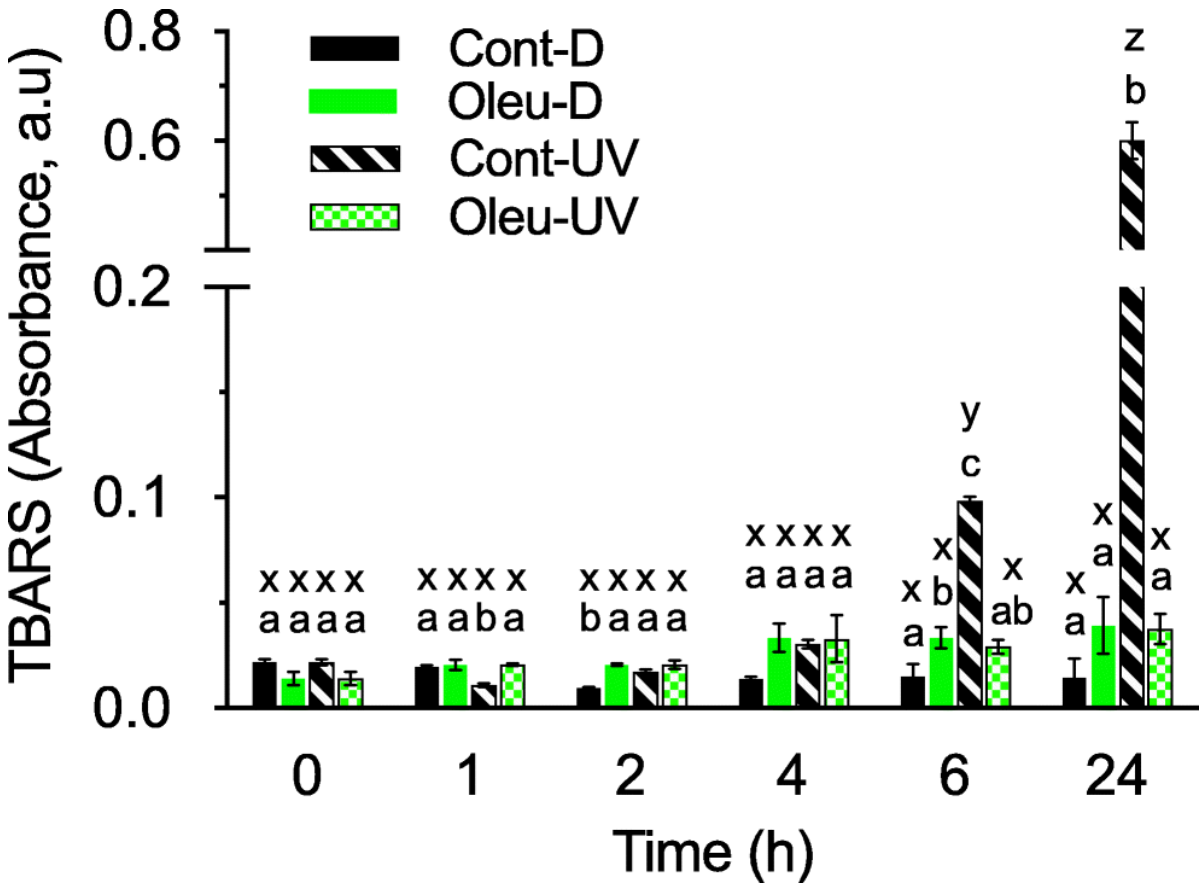
Conclusion:
Enapsulation of olive leaf extract into proliposomes is a promising method, as the capsules are stable and the encapsulation efficiency is high.
It is interesting to note that the encapsulation efficiency of the olive leaf extract is higher than that of the pure one oleuropein, the release is also lower.

Characterization - morphology



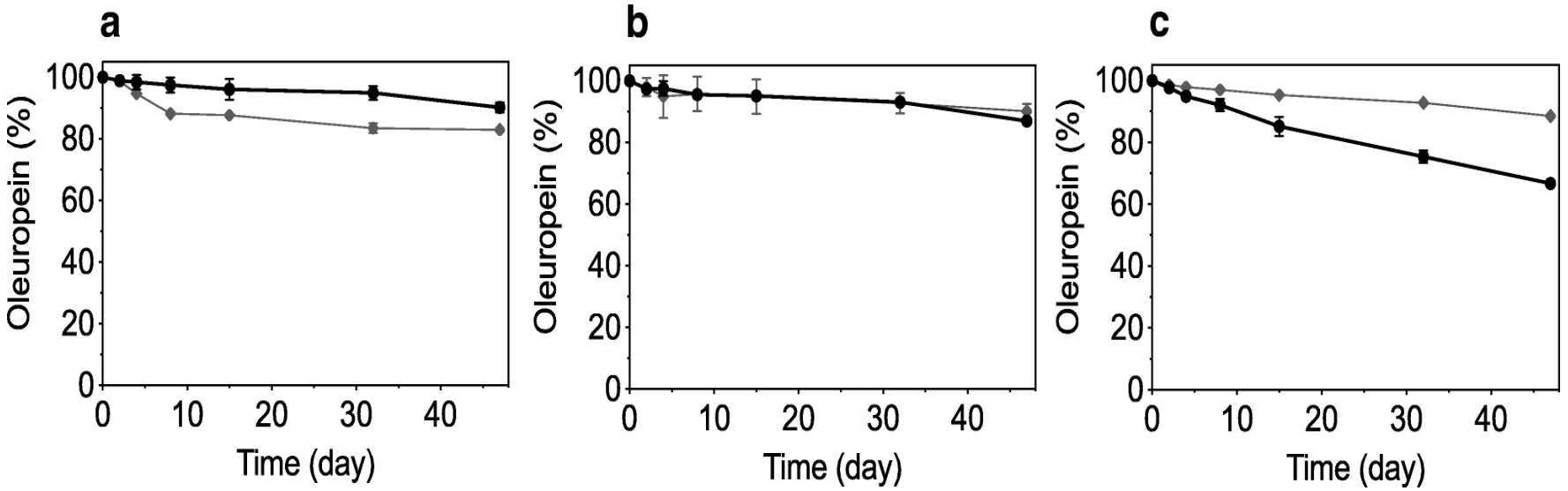
Representative transmission electron micrographs of PL-90 g liposomes without (**a**) and with (**b**) encapsulated OLE
Gonzales Ortega, Food biophysics. 2021

Peroxidation of PL-90 g liposomes with actively encapsulated oleuropein



Peroxidation of PL-90 g liposomes with actively encapsulated oleuropein (Oleu) using the thiobarbituric reacting substances (TBARS) assay following treatment with UV light (UV) and in the dark (D) at 25 °C. Data are means ± standard deviation (n ≥ 02). *Gonzales Ortega, Food biophysics*. 2021

Time courses of oleuropein stability



Time courses of oleuropein stability when added as free OLE (●) or PL-90 g OLE liposomes (◇):
to a commercial lemonade drink (**a**),
and to a 'model drink' of 10 mM sodium citrate/citric acid buffer, pH 2.87, with 1.2 mg mL^{-1} **ascorbic acid** (**b**)
and without the ascorbic acid (**c**), at 5 º C.
Data are means ± standard deviation (n = 03)

Preparation of extracts rich in phenolic compounds for the preparation of new food products and nutritional supplements

Olive leaves preparation for use in food products

1.
- Olive leaves
- Preparation of extract with 70% ethanol
- Extraction in 3 independent parallels
- HPLC determination of hydroxytyrosol and oleuropein in extracts

2.
- Selection of the most suitable starting material according to the oleuropein content
- Food selection for the preparation of a food product enriched with oleuropein

3.
- Selected liquid/solid yogurts with different fat content ranging from 1.2 to 6.0% fat/100 g of product
- Ground olive leaves were added in amounts of 0.5%, 1% and 3% of the net weight of the food product

Preparation of extracts rich in phenolic compounds

Olive leaves preparation for use in food products

- HPLC results

compound	
Hydroksitirozol (mg/g leaves)	**0.352 ± 0.004**
Oleuropein (mg/g leves)	**37.2 ± 0.1**

- Food products

product	MU liquid yogurt	MU KEFIR	MU yog. wtihout lactose	MU yogurt	Alpsko liquid yogurt	Alpsko creamy yogurt
% fat	1.3	1.5	1.6	3.2	4.0	6.0

- Enrichment with olive leaves

Netto weight of yogurt (g)	Addition of 0.5% olive leaves	Addition 1% olive leaves	Addition 3% olive leaves
500	2.5	5	15
250	1.25	2.5	7.5
180	0.9	1.8	5.4

Preparation of extracts rich in phenolic compounds

Olive leaves preparation for use in food products

+ 3 %

+ 1%

+ 0.5 %

Preparation of extracts rich in phenolic compounds for the preparation of nutritional supplements

Olive leaf preparation for use in food products

Product	Fats (%)	Netto mass (g)	+ 05 % leaves per net weight			+ 1 % leaves per net weight			+ 3 % leaves per net weight		
			I	T	O	I	T	O	I	T	O
MU liquid yogurt 1.3	1.3	500	4.5	4	3.75	4.1	3.75	2.75	3.5	2.6	1
MU KEFIR 1.5	1.5	250	4.7	4.4	4.25	4.2	4.2	4.1	4.1	3.1	1.5
MU yogurt without lactose	1.6	500	4.88	4.5	4.63	4.56	3.93	3.8	3.25	2.38	1.75
MU yogurt 3.2	3.2	500	4.5	4.2	4.55	4.05	4.08	3.98	/*	/*	/*
Alpsko liquid yogurt	4.0	500	4.55	4.58	4.58	4.18	4.08	3.5	/*	/*	/*
Alpsko creamy yogurt	6.0	180	4.67	4.33	4.67	4.17	3.5	3.5	/*	/*	/*

/* - We did not evaluate the products due to the extremely bitter taste of the previously evaluated products

Preparation of extracts rich in phenolic compounds for foods

Olive leaves preparation for use in food products

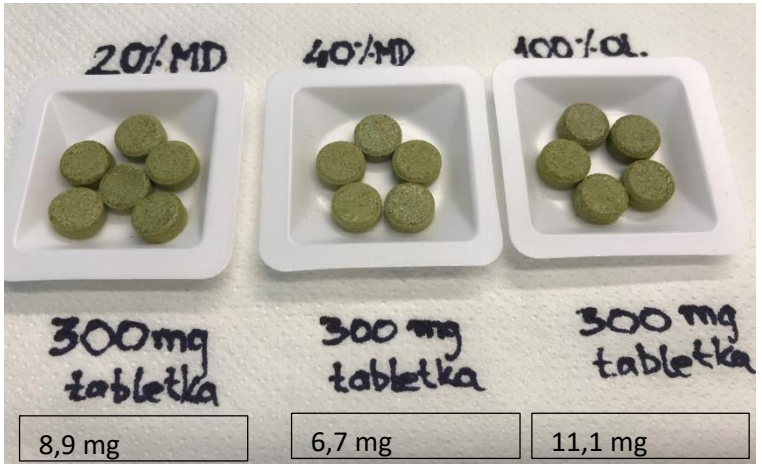
- The added ground olive leaves create from a soft green color to a more intense color. The **colors are natural** and pleasant.
- The added ground olive leaves **do not affect the smell** of the yogurts themselves.
- The amount of ground olive leaves added affects the texture of the product, which is not disturbing at the lowest concentration, but at higher amounts it depends on **the fat content**.
- The added ground olive leaves have the greatest effect on the taste of the yogurt. The taste is slightly bitter, but unobtrusive, or it is masked with the addition of 0.5% olive leaves, with the addition of 1%, the bitter aftertaste is more intense, but still acceptable in some yogurts, and with the addition of 3% olive leaves, the bitter aftertaste is very disturbing , dominant.

Conclusion

- For further development, all products with a 0.5% addition of ground olive leaves are suitable, and with a 1% addition, MU KEFIR, MU yogurt 3.2, Alpine liquid yogurt 4.0 are suitable.

- For the preparation of new products, we recommend more finely ground starting material or **encapsulated extract**.

Nutritional supplements made from olive leaves on the Slovenian market

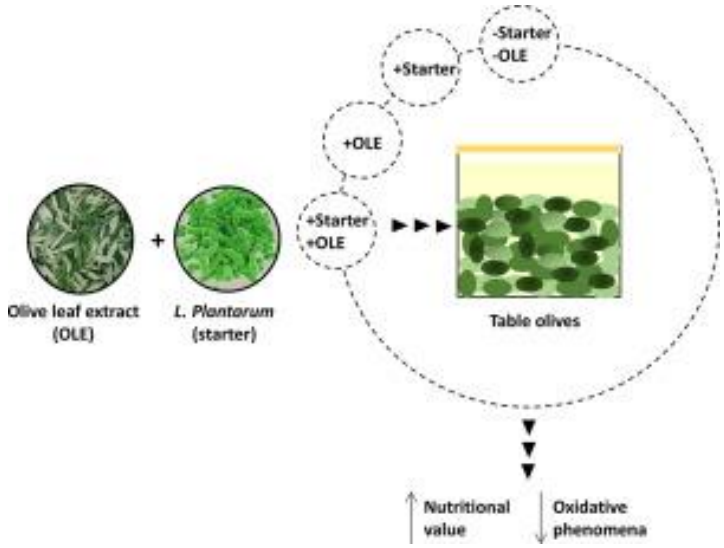


The nutritional supplement in the form of a compressed pill was prepared from ground dry olive leaves and/or with the addition of maltodextrin.

The average weight of one tablet is 300 mg.

All tablets have a diameter of 10 mm and a thickness of 4 mm.

WHAT TO DO NEXT?



Effects of olive leaf extract addition on fermentative and oxidative processes of table olives and their nutritional properties. Food Res Int. 2019 Feb;116:1306-1317

antioxidants

MDPI

Communication

Solid State Fermentation of Olive Leaves as a Promising Technology to Obtain Hydroxytyrosol and Elenolic Acid Derivatives Enriched Extracts

Anna Starzyńska-Janiszewska [1], Carmen Fernández-Fernández [2], Beatriz Martín-García [3], Vito Verardo [3,4,*] and Ana María Gómez-Caravaca [2,4]

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On 21/02/2017, 21/06/2017 and 9/11/2017, project team members presented the draft Guidelines on the handling of olive processing residues to the representatives of the MKGP (B. Butinar, PI, ZRS Koper)

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CRP V4-1621

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THANK YOU!