PhD IN AGRI-FOOD SCIENCES, TECHNOLOGIES AND BIO-TECHNOLOGIES

UNIMORE
UNIVERSITÀ DEGLI STUDI DI MODENA E REGGIO EMILIA

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AGRI-FOOD PRODUCTS AND BY-PRODUCTS AS A SOURCE OF BIOACTIVE COMPOUNDS: EXTRACTION, CHARACTERIZATION AND BIOLOGICAL ACTIVITIES

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State of the art

Agricultural waste

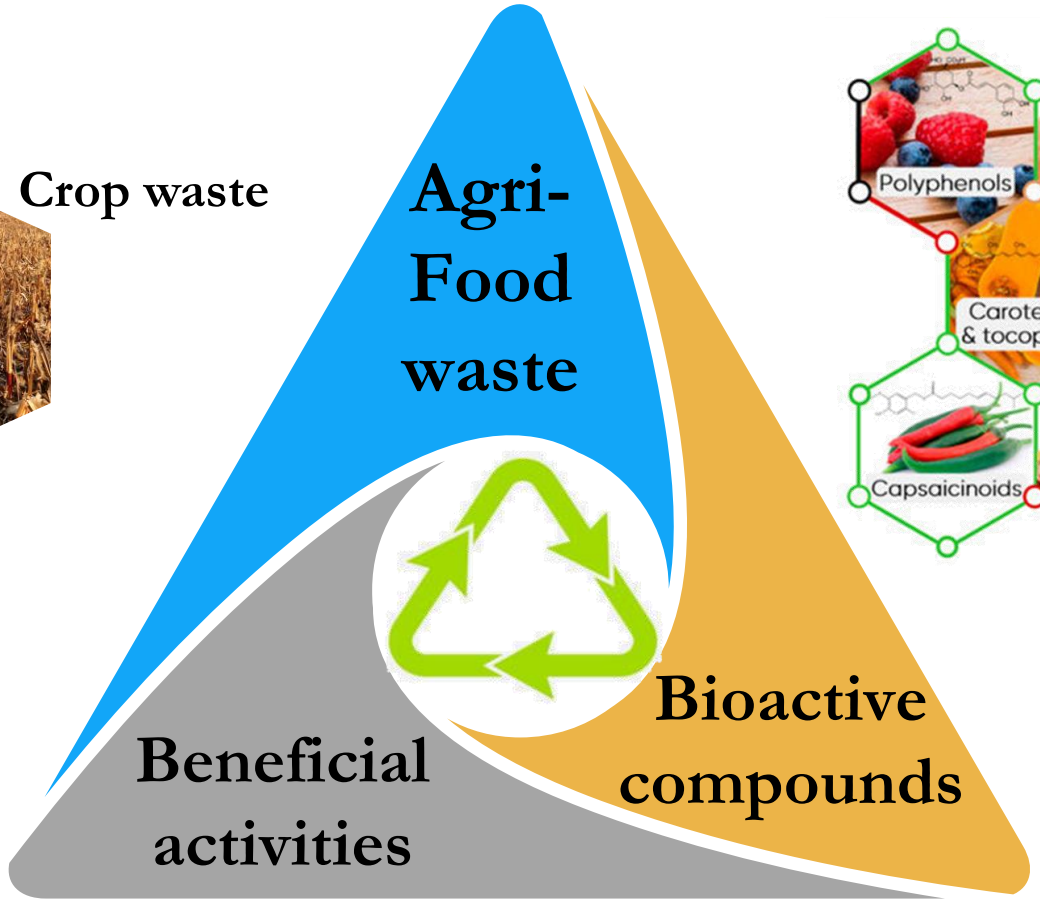
Crop waste

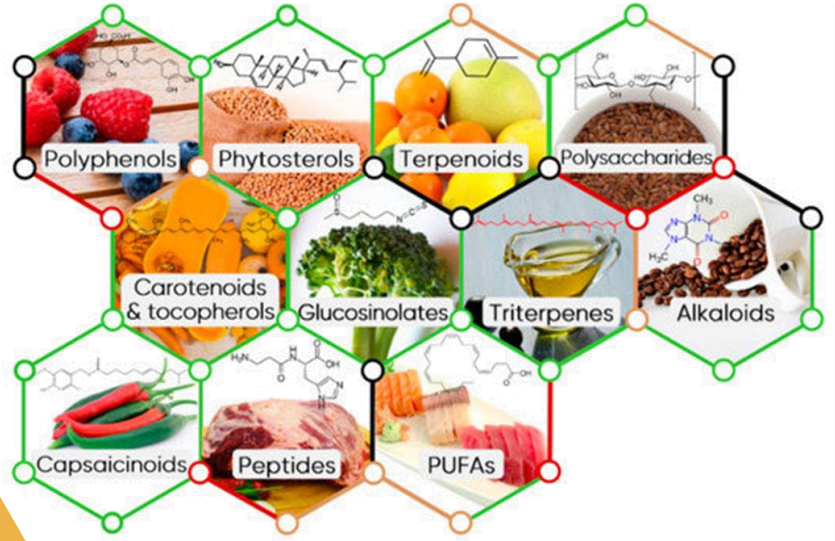
Fruit and vegetables processing industries

2018

(European Union)

2337 million tonnes





Antioxidant

Anti-inflammatory

Antimicrobial

Aim of the work

Prunus domestica
L. var d'Ente

MONTERÉ

"COOPERATIVA MODENESE ESSICCAZIONE FRUTTA"

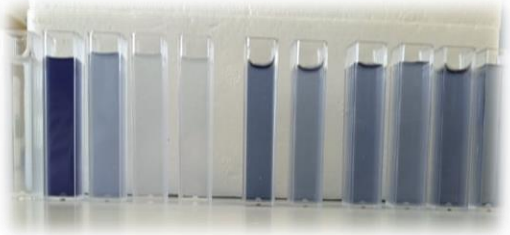
Prune by-product

- Estimation of the chemical composition (polyphenols, sugars, minerals …)
- Reduction of sugar content by fermentation
- Formulation of a nutritional supplement in order to develop a product useful for intestinal function

Chemical composition

Phenolic and flavonoid content

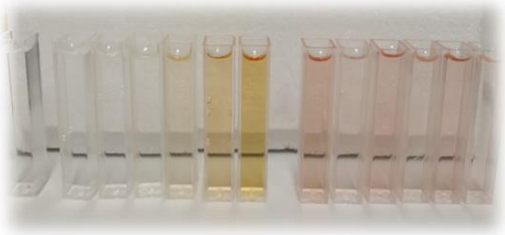
Folin-Ciocalteu assay



Total phenolic content

TPC

Aluminum chlorate assay



Total flavonoid content

TFC

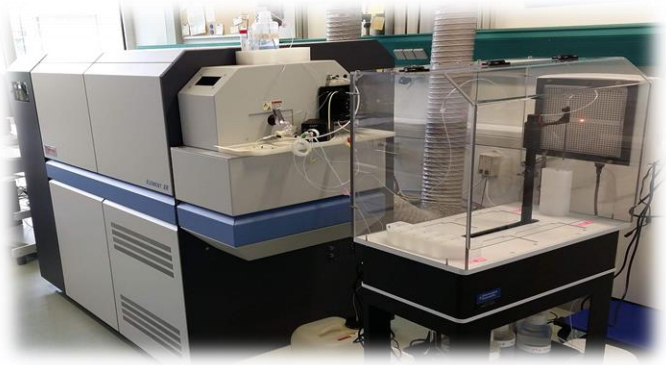
Water / Pectin / Moisture

Freeze-drying → **Water content**

Precipitation (EtOH) → **Pectin content**

Oven drying → **Moisture content**

Metal content



ICP-MS

Chemical composition

Water (%)	73.97±2.75
Pectin (%)	14.80 ±0.53
Moisture (%)	9.67 ±1.37
TPC (mg GAE/g)	3.32 ±0.66
TFC (mg QE/g)	0.06 ±0.03

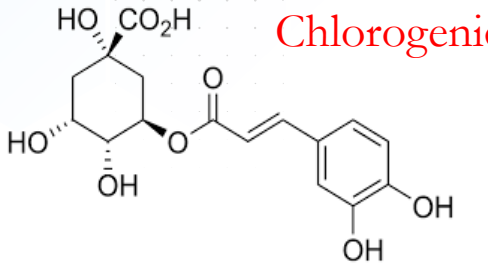
Metals (mg/100g)			
K	583.79 ±82.16	**Ni**	0.65 ±0.50
Ca	45.33 ±18.48	**W**	0.48 ±0.33
Mg	27.15 ±9.79	**S**	0.34 ±0.19
Na	25.58 ±18.50	**Mn**	0.19 ±0.12
Zn	24.31 ±8.84	**Li**	1.50 ±1.28
B	2.2 ±0.73	**Pb**	0.14 ±0.08
Fe	0.97 ±0.46	**Cr**	0.02 ±0.01
Cu	0.84 ±0.53		

Chemical composition

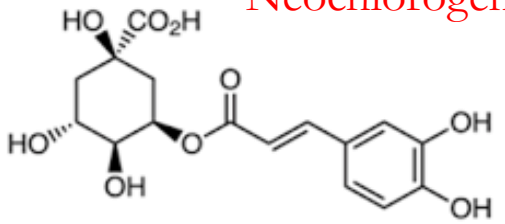
Phenolic compounds

LC – ESI - MS

Chlorogenic acid



Chlorogenic acid

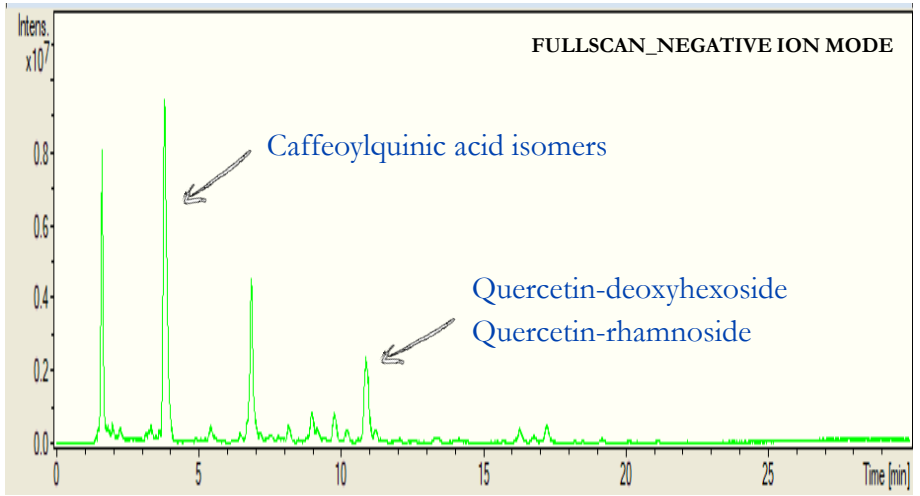


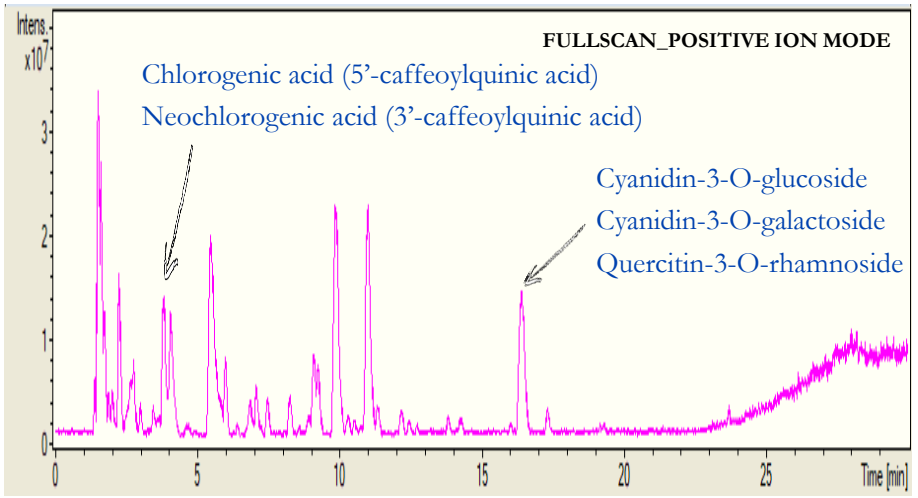
Neochlorogenic acid

HPLC / DAD

Chlorogenic acid

0.53 ±0.12 mg/g FW

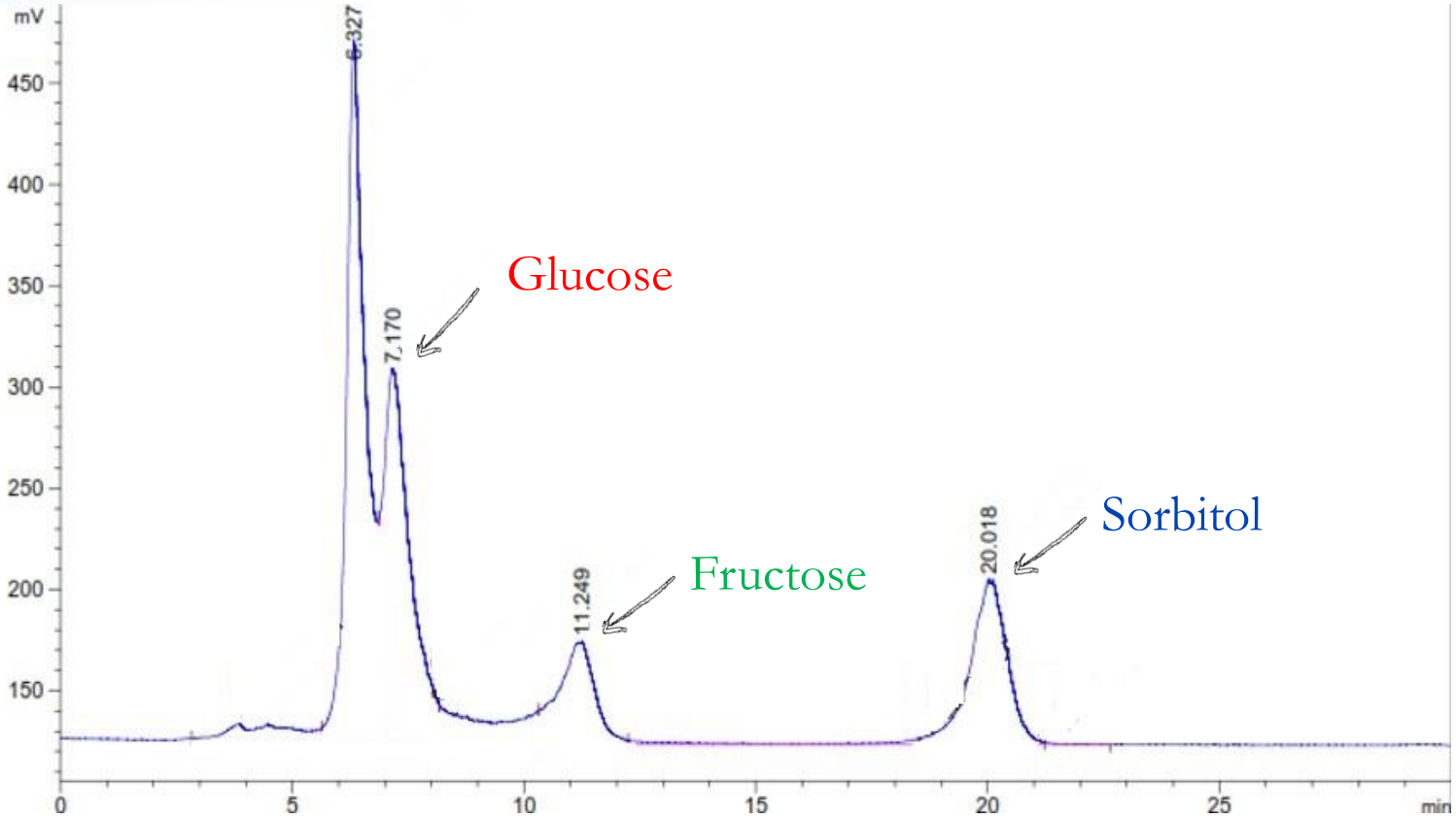




Chemical composition

Sugar content

HPLC/ELSD



Sorbitol

44.04 ±9.50 mg/g FW

Fructose

125.03 ±38.24 mg/g FW

Antioxidant activity

DPPH assay

Total antioxidant Activity (TAC)

Ethanolic extract

196.13 ±67.96 mg AAE/100 g FW

AAE: Ascobic acid equivalent

Cosmulescu et al. (2012)

Plum fruit

Fruit	22.17 - 92.65	mg AAE/100 g FW
Skin	163.62 - 972.74	mg AAE/100 g FW
Pulp	10.38 - 38.17	mg AAE/100 g FW

Reduction of sugar content

Prune by-product

+ H_2O

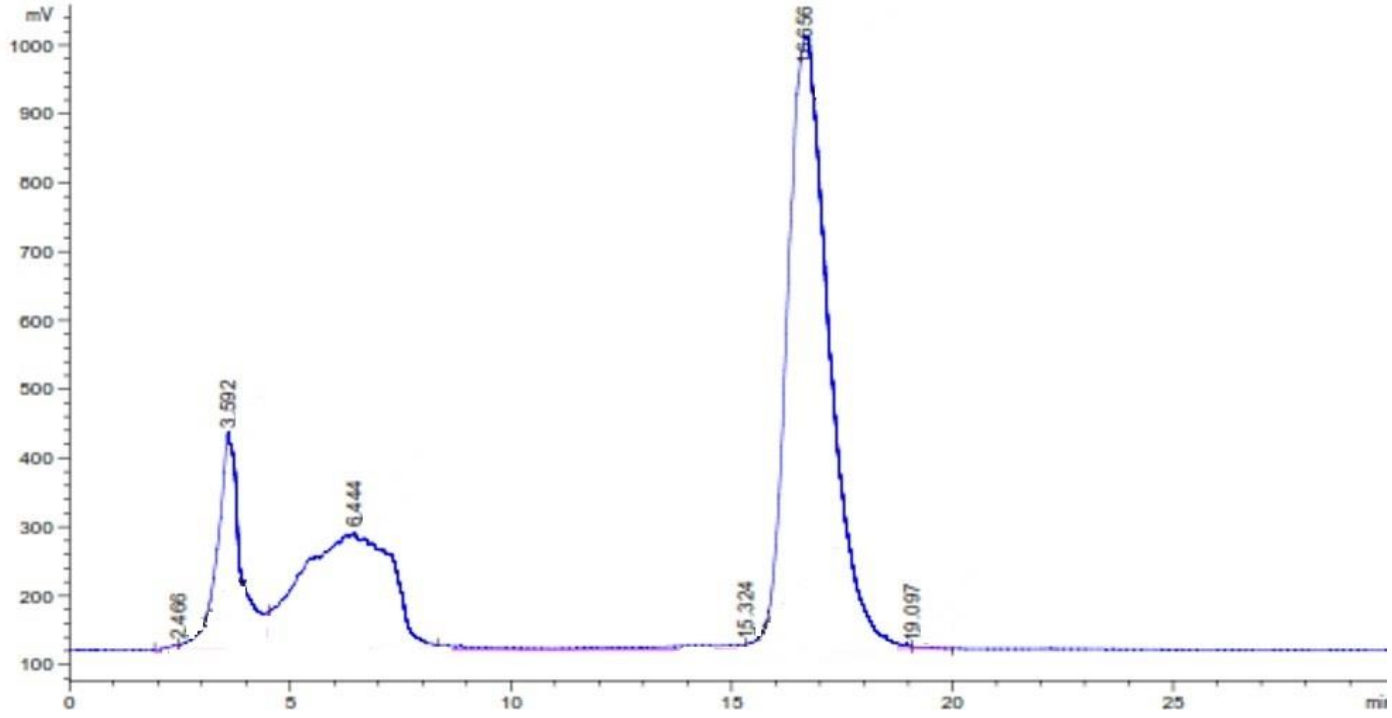
Fermentation (24h / room Tº)

Yeast (*Saccharomyces cerevisiae*)

Centrifugation
Filtration

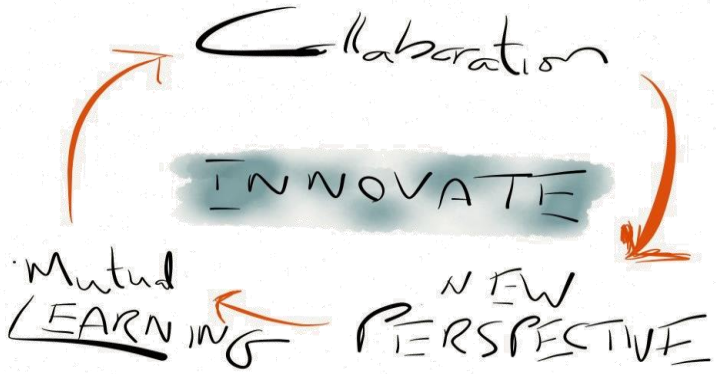
HPLC/ELSD

Fermentation



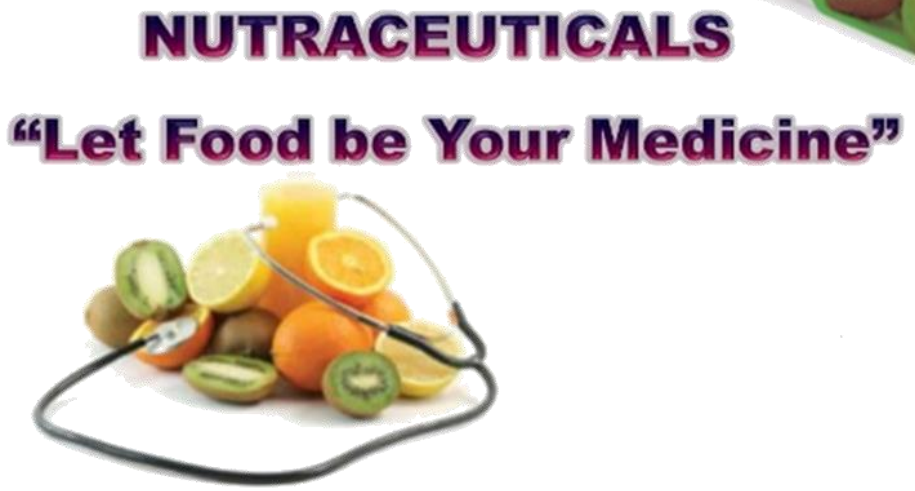
Sorbitol	77.45 ±12.04	mg/g FW
Fructose	12.97 ±2.19	mg/g FW

Perspectives



- Application of prune by-product into new formulations in order to obtain beneficial nutraceutical derivatives

- Effects on intestinal health



Aim of the work

Lavandula angustifolia

LA

Lavandula x intermedia cv "Grosso"

LI

Distillation process

Lavender by-products

- Determination of the composition in essential oils
- Determination of the composition in phenolic compounds of residual lavender after distillation
- Study of some biological activities

Distillation of lavender

Steam distillation

01

EOs

Essential oils

02

Hyd

Hydrolat / Hydrosol

Distillation waste water

WW

03

Residual lavender

RM

04

Chemical composition of EOs / Hyd

GC-MS / GC-FID analysis

Gas chromatography coupled to mass spectrometer (**GC-MS**)

Gas chromatography coupled to flame ionization detector (**GC-FID**)

- The chemical composition of EOs for the two lavender species did not differ (as expected according to the European Pharmacopoeia)

The highest values were obtained for linalool and linalyl acetate

- Both hydrosols were rich in alcoholic monoterpenes (more than 70% of the whole composition)

The highest values were obtained for linalool, camphor, borneol and α-terpineol

EOs

Compound	*LA*	*LI*
α-pinene	0.54	0.68
camphene	0.32	0.36
sabinene	0.18	0.24
β-pinene	0.56	0.82
Oct-1-en-3-ol	0.14	-
myrcene	1.27	1.52
3-carene	0.10	0.18
α-terpinene	0.26	0.14
limonene	0.65	0.54
1,8-cineole	5.27	7.90
cis-ocimene	1.39	1.65
trans-ocimene	1.27	0.70
gamma terpinene	0.10	0.17
cis linalool oxide	0.15	0.12
trans linalool oxide	0.34	0.33
linalool	31.72	27.61
fenchol	0.36	0.13
camphor	5.68	6.78
borneol	2.47	1.86
lavandulol	0.40	0.27
terpinen-4-ol	1.81	1.08
α-terpineol	0.58	0.61
myrtenal	0.28	0.19
carvone	0.11	0.18
linalyl acetate	33.86	35.63
lavandulyl acetate	3.47	2.91
neryl acetate	0.24	0.29
α-copaene	0.49	0.59
β-caryophyllene	1.56	1.53
α-humulene	1.09	1.29
ar curcumene	0.47	0.58
γ-cadinene	0.20	0.36
Total	**97.36**	**97.07**

Hyd

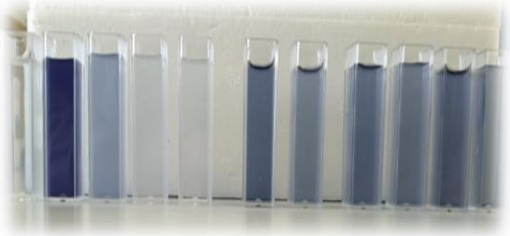
Compound	*LA*	*LI*
α-pinene	0.26	0.17
camphene	0.26	0.24
β-pinene	0.20	0.13
1,8 cineole	2.12	1.80
cis-ocimene	0.57	0.49
cis linalool oxide	4.46	4.62
trans linalool oxide	4.25	4.13
linalool	28.67	32.12
camphor	13.38	15.63
borneol	10.64	10.42
neomenthol	1.96	1.55
lavandulol	1.08	0.92
terpinen-4-ol	5.58	3.94
menthol	0.75	1.00
p-cymen-8-ol	0.74	1.10
α-terpineol	11.18	9.78
myrtenal	0.21	0.25
verbenone	0.44	0.55
nerol	0.33	0.31
citronellol	4.53	2.48
carvone	0.18	0.19
neral	0.15	0.17
piperitone	1.94	1.65
lynalil acetate	0.13	0.10
geranial	0.34	0.39
lavandulyl acetate	0.13	0.10
carvacrol	0.66	0.38
Total	**95.16**	**94.49**

Chemical composition of RM

Phenolic and flavonoid content

Hydroalcoholic extract

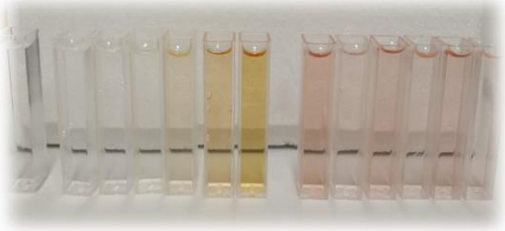
Folin-Ciocalteu assay



Total phenolic content

TPC

Aluminum chlorate assay



Total flavonoid content

TFC

Residual material	*LA*	*LI*
TPC (mg GAE/g)	19.22 ± 4.16	17.06 ± 3.31
TFC (mg QE/g)	1.56 ± 0.21	1.41 ± 0.10

The two hydroalcoholic extracts did not show any significant difference in both TPC and TFC

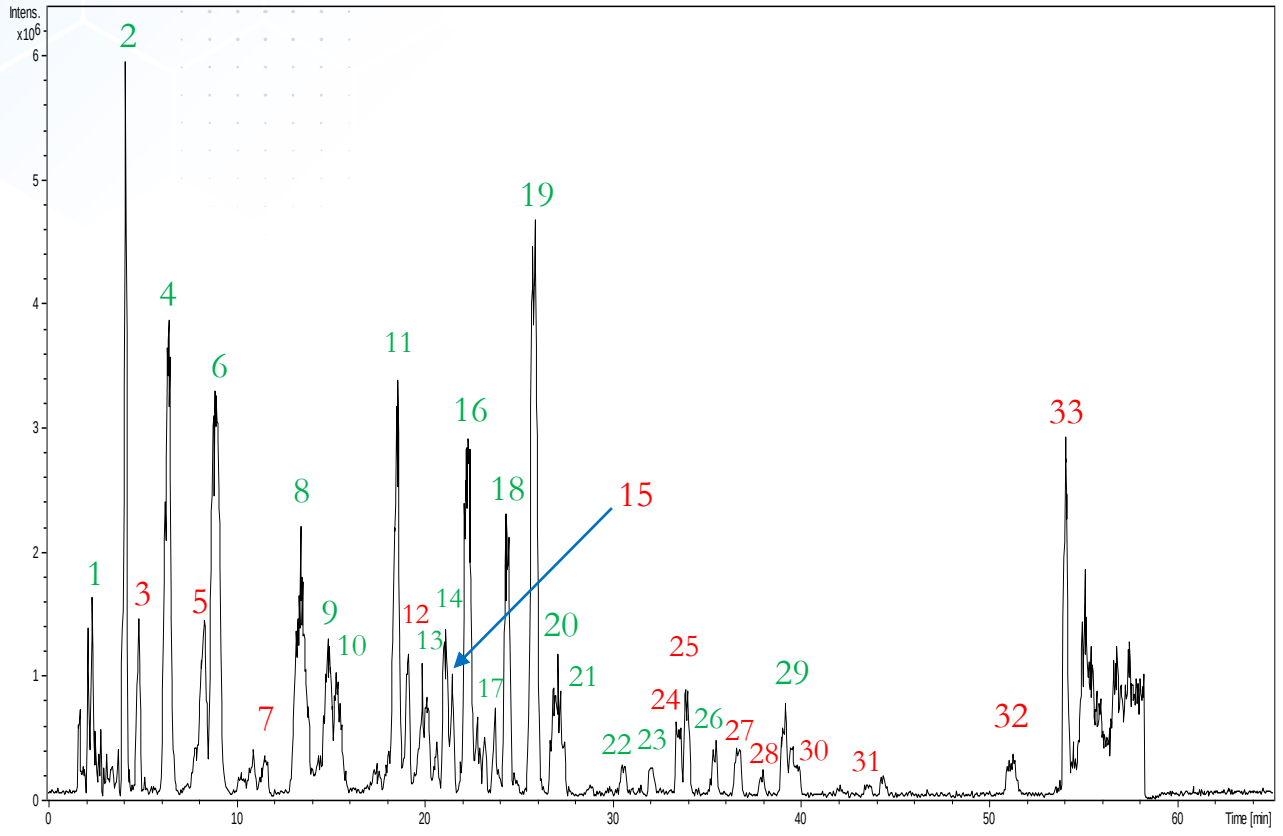
TFC represented about the 8% of TPC in both the lavender extracts

TPC of lavender is strongly affected by the species, the harvest time, the growing conditions of the crops and the age of the plant

Chemical composition

Phenolic compounds

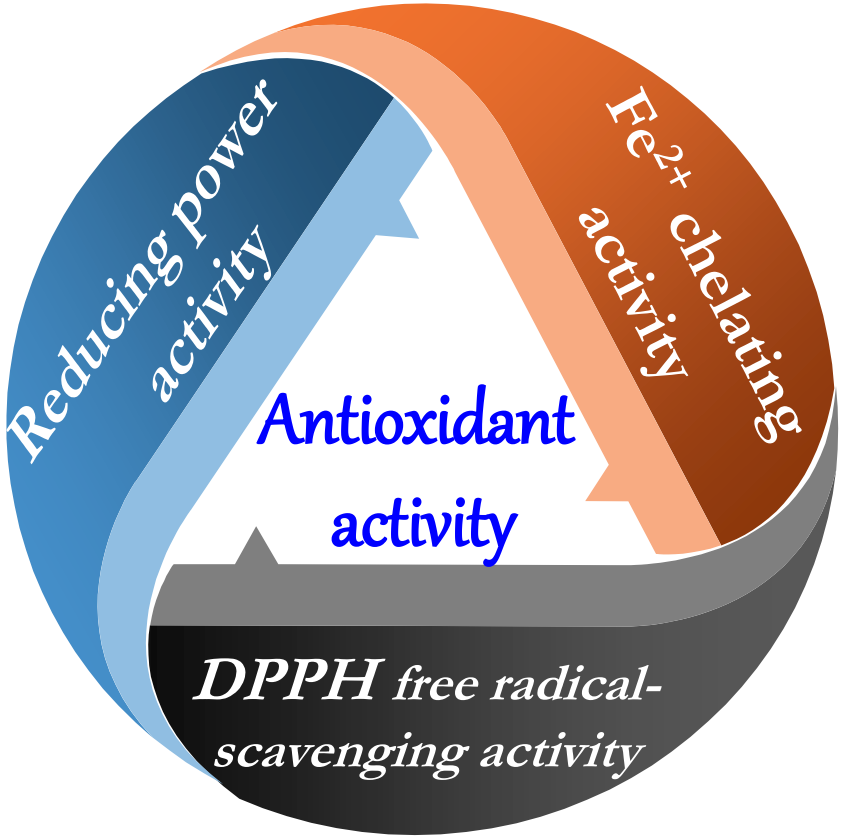
LC – ESI - MS



1. Caffeoyl aspartic acid
2. Danshensu
4. p-coumaric acid hexose
6. Ferulic acid hexose
8. p-coumaric acid hexose
9. Luteolin 7-O-diglucoronide
10. Apigenin 7-O-diglucoronide
11. Ferulic acid hexose
13. Quercetin hexose
14. Luteolin/kaempferol hexose
16. Luteolin 7-O-glucuronide
17. Quercetin 3-O-rhamnoside
18. Apigenin 7-O-glucoside
19. Rosmarinic acid
20. Luteolin 7-O-glucuronide
21. Apigenin 7-O-glucoronide
22. Rosmarinic acid methylester
23. Kaempferol
26. Quercetin hexose
29. Ellagic acid

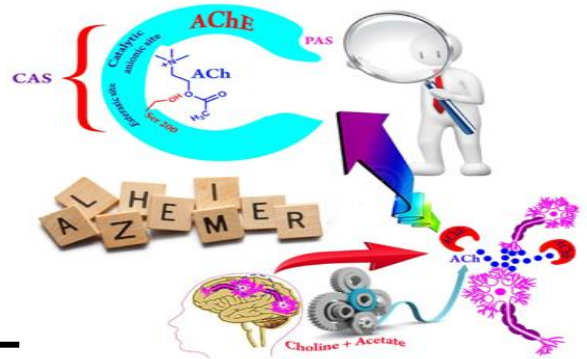
Antioxidant activity

	LA	*LI*
DPPH (mg Teq/g)	94.17 ± 6.29	94.51 ± 2.85
Fe^{2+} chelation (mg EDTAeq/g)	1.49 ± 0.03	2.10 ± 0.13
Fe^{3+} reduction (mg AAeq/g)	77.29 ± 0.05	69.79 ± 0.04



- The two lavenders exhibited similar free scavenging activities
- LA showed a greater capability in reducing ferric ions » Higher total phenolic content for LA
- LI exhibited higher activity in chelating ferrous ions » Polyphenols with more than one chelating site or with greater stability constants of the complex

Anti-cholinesterase / Anti-tyrosinase activities



Sample	LA (mg/mL)	LI (mg/mL)	Galantamine (µg/mL)	Kojic acid (µg/mL)
AChE	5.35 ± 0.47	6.67 ± 0.12	18.83 ± 1.05	-
Tyrosinase	5.26 ± 0.02	6.56 ± 0.16	-	18.13 ± 0.45

- Significant inhibition of the enzymes for both the hydroalcoholic extracts
- LA extracts showed lower IC50 values than LI extracts » Stronger inhibition of AChE and Tyrosinase
- The higher inhibition capacity of LA extract may be related to the highest content of TPC and TFC

Perspectives

03

Study of other biological and pharnmacological activities (antimicrobial, Anti-Inflammatory…)

Correlation between the amounts of phenolic compounds and their influence on the antioxidant, enzymatic activities

02

01

Identification of remaining pics / Quantitative analysis of the identified phenolic compounds

THANK YOU