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PORTO

Agrofood Byproducts as a Source for New Food Ingredients for Sustainable and Healthier Diets

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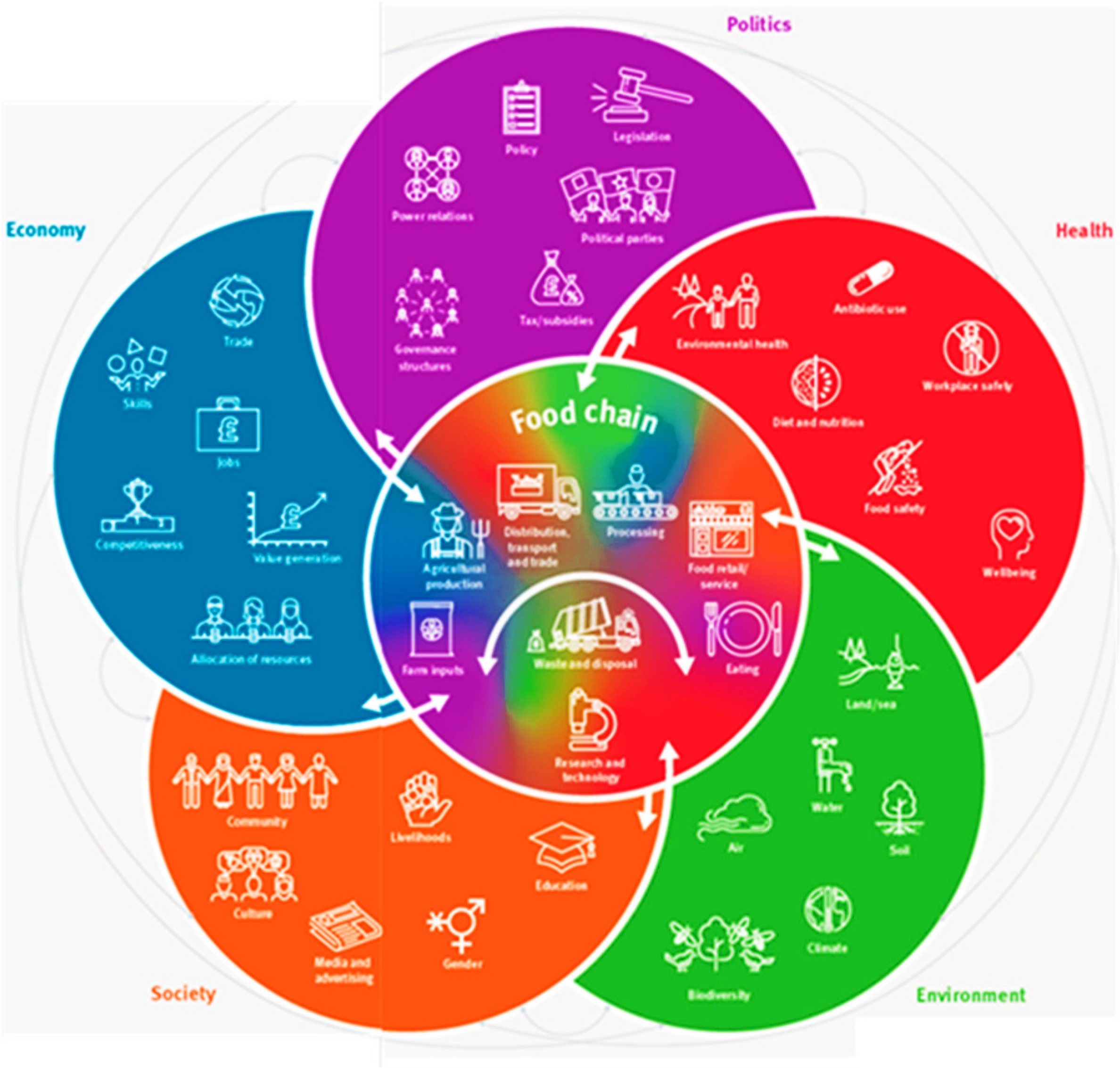
PORTO

AGENDA

- Challenges and Needs in the 21st Century
- Agrofood Byproducts and Their Potential as New Food Ingredients
- Nutritional and Functional Benefits
- Processing Techniques Methods
- Examples of Plant Byproducts and Losses
    - Melon, Lemon, Soy meal and Okara, Tomato, Cruciferous Vegetables, Olive Pomace and Okara

Challenges and needs in the 21st century



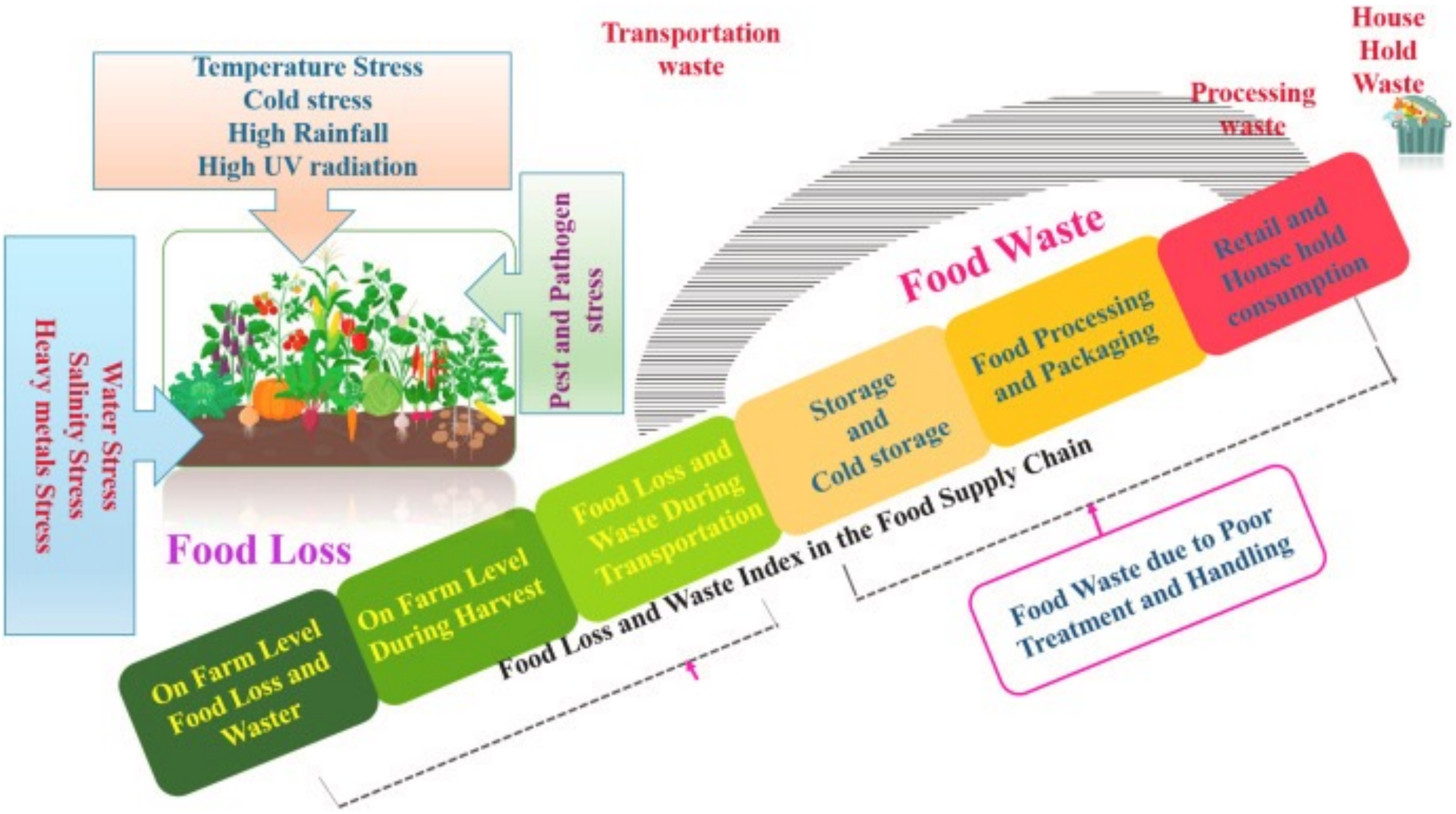
Parsons, K.; Hawkes, C.; Wells, R. Brief 2. What is the food system? A Food policy perspective. In *Rethinking Food Policy: A Fresh Approach to Policy and Practice*; Centre for Food Policy: London, UK, 2019

Challenges and needs in the 21st century

Today around 100 million tonnes of food are wasted annually in the EU

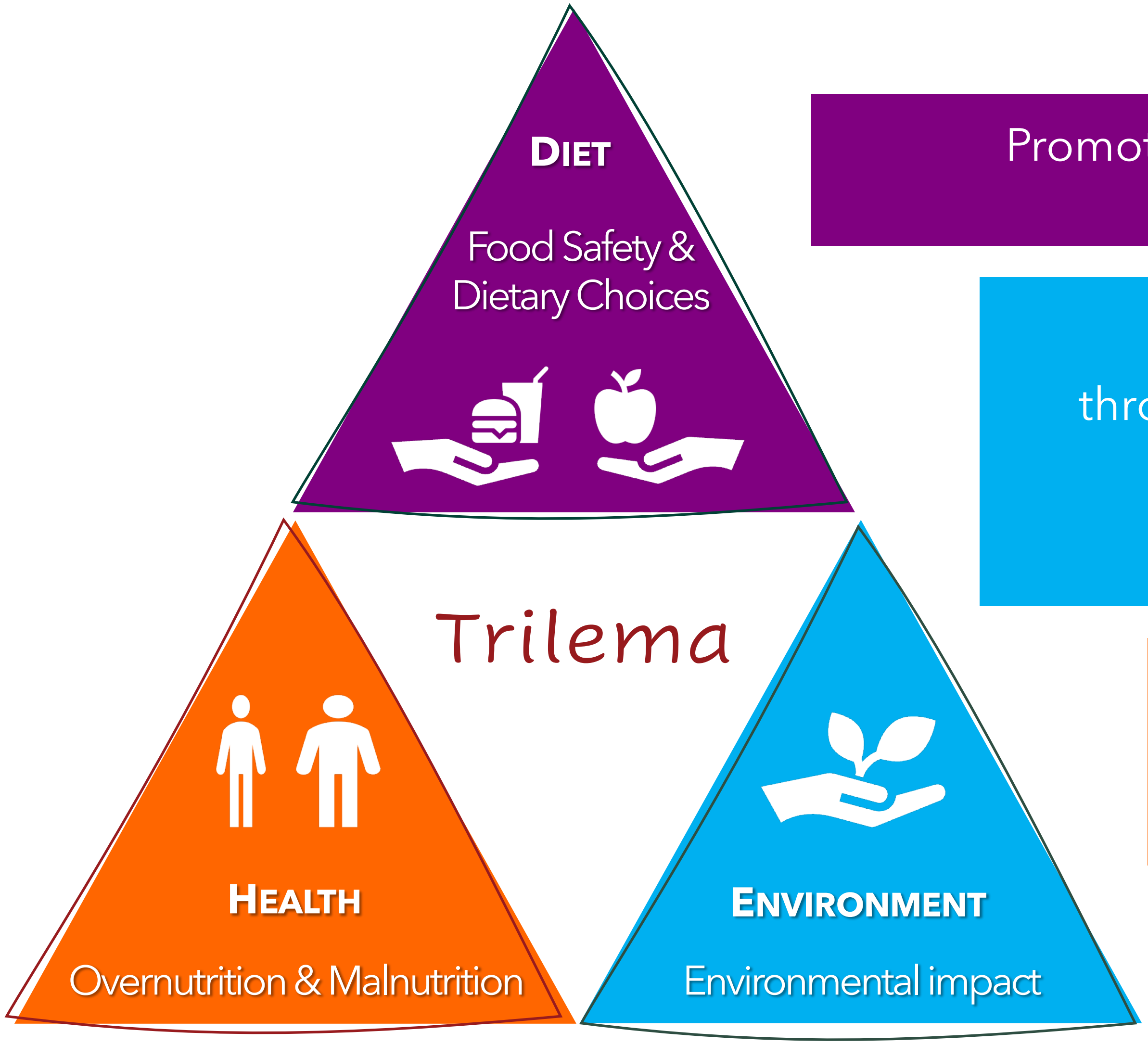
European Commission
50% Food losses and waste by 2030

Challenges and needs in the 21st century



Bioresource Technology 343 (2022) 126151

Challenges and needs in the 21st century



Promote food and nutrition reforms that support diets – Healthy food in optimal quantities

Build policies that support food security for populations throughout their lifetime, including specialized nutrition care for those at risk of or suffering from malnutrition – the foundation for a healthier global population

Develop food systems with adequate production & distribution and, at the same time, long-term sustainable food production

JOURNAL OF THE ACADEMY OF NUTRITION AND DIETETICS 120, 8 (2020)

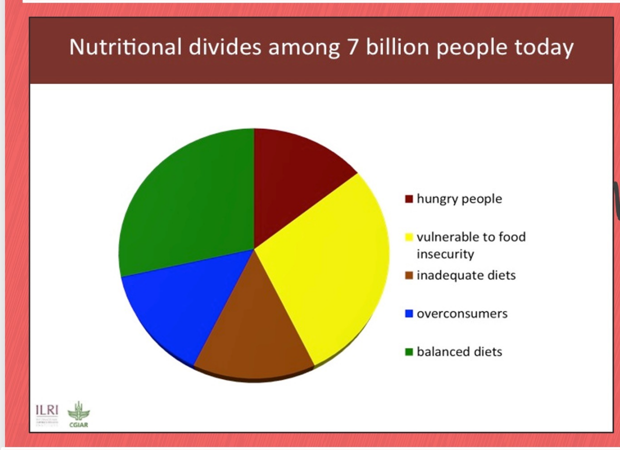
Clean label

- Natural ingredients: no artificial flavors, artificial colors, artificial preservatives or synthetic additives
- Simplicity: less chemicals and recognizable ingredients that do not have any artificial chemicals
- Transparency: information on how ingredients are sourced and how products are manufactured
- Minimal processing: processing using techniques that consumers don't understand to be artificial

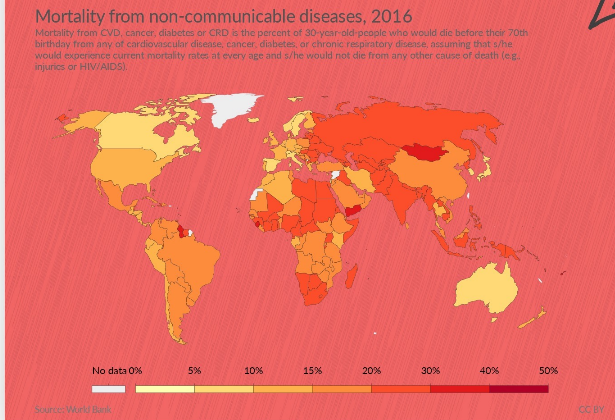
Agrofood byproducts and their potential as new food ingredients

Issues

Dietary Inadequacy



Non-Communicable Diseases



Prevention of

Fortified / Enriched foods

Dietary supplements

Consistent with SDG2

Consistent with SDG12

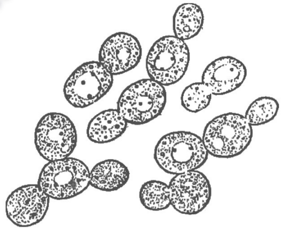
Up-Cycling

- Increase of fiber availability
- Increase of antioxidant activity
- Increase of bioactive compounds
- Decrease of anti-nutritional factors

Food By-Products and Wastes are…
Issue & Solution

Potential sorces of:
- Fiber
- Proteins
- Minerals
- Vitamins
- Bioactive compounds

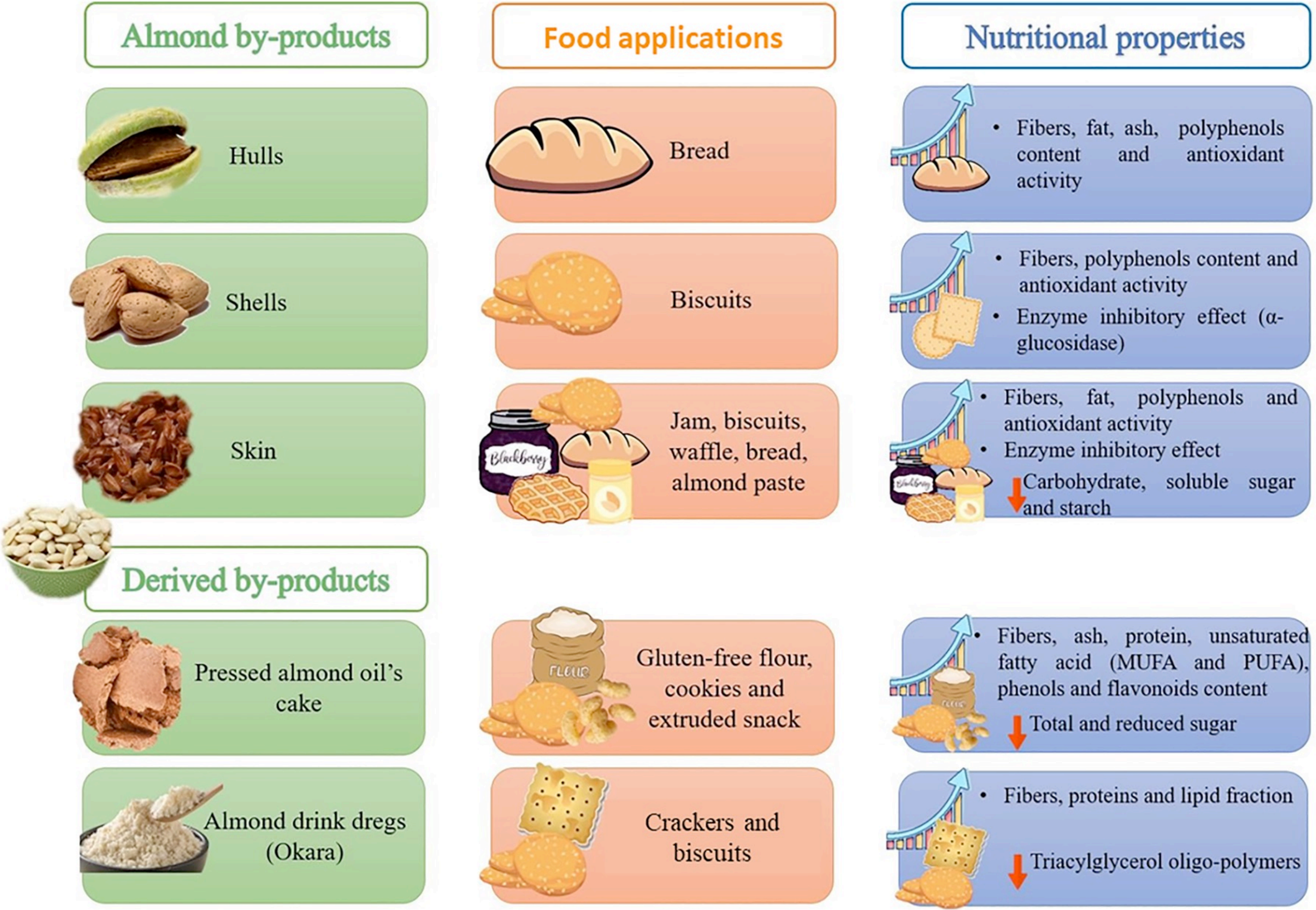
Bioprocessing



CATOLICA CBQF · INTERNATIONAL CONFERENCE ON SUSTAINABLE FOODS

Sustainable and Health-Protecting Food Ingredients from Bioprocessed Food by-Products and Wastes. Sustainability 2022, 14, 15283. https://doi.org/10.3390/su142215283

Lacivita et al. Discover hidden value of almond by-products: Nutritional, sensory, technological and microbiological aspectsFuture Foods 10 (2024) 100398



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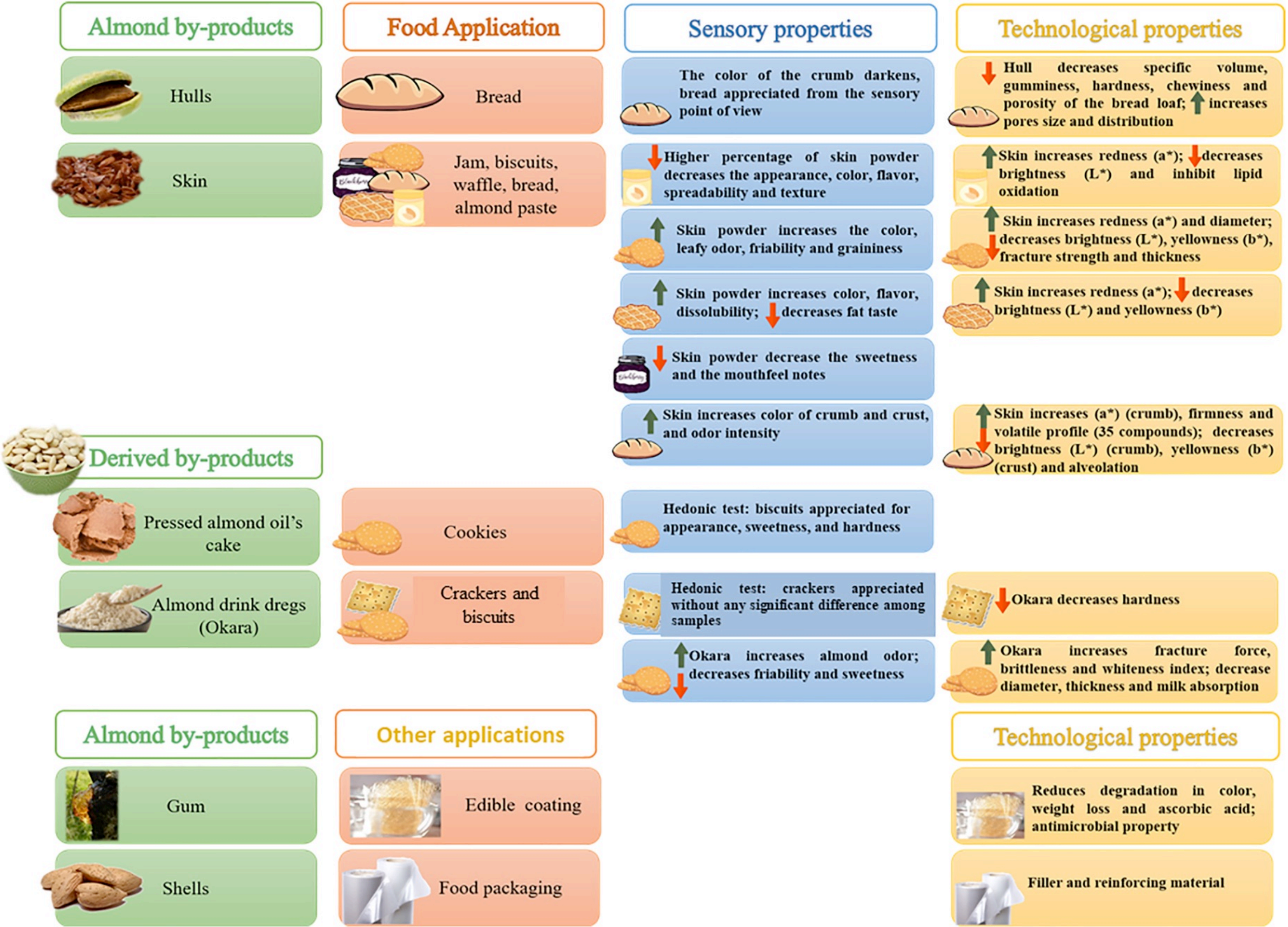
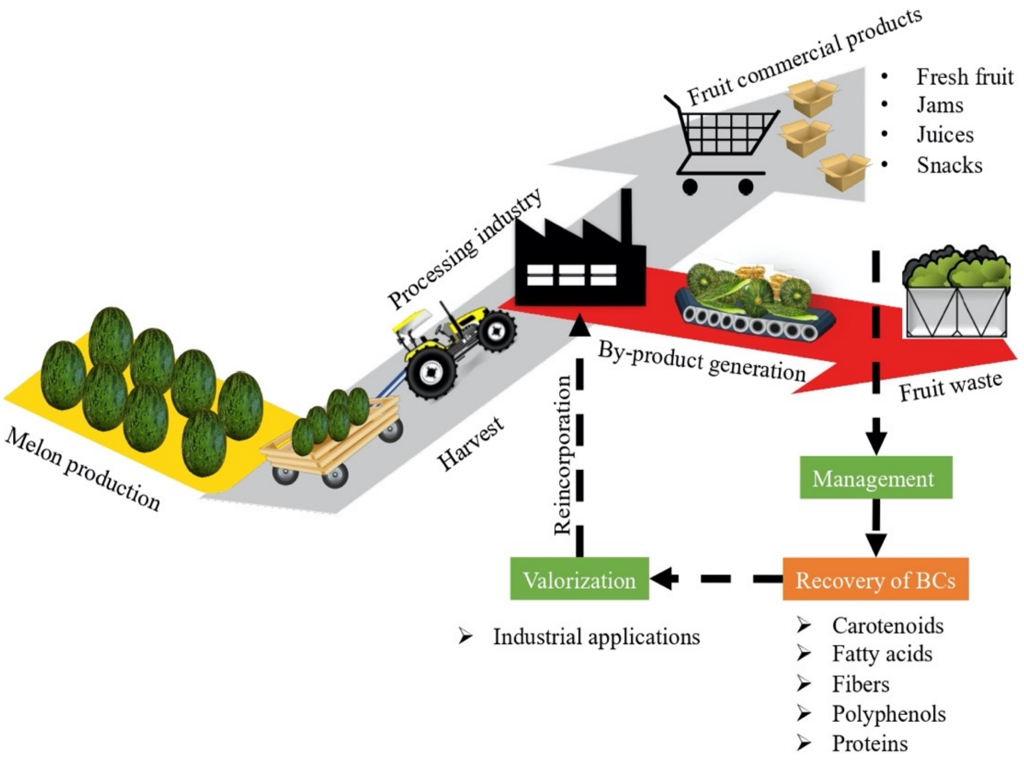


VALORIZATION OF AGRO-INDUSTRIAL FOOD BY-PRODUCTS: AN OPPORTUNITY TOWARDS BIOACTIVES

Circular Economy

Agri-Food Cycle



ELSEVIER
Journal of Environmental Management
Volume 299, 1 December 2021, 113571

Valorisation of food agro-industrial by-products: From the past to the present and perspectives

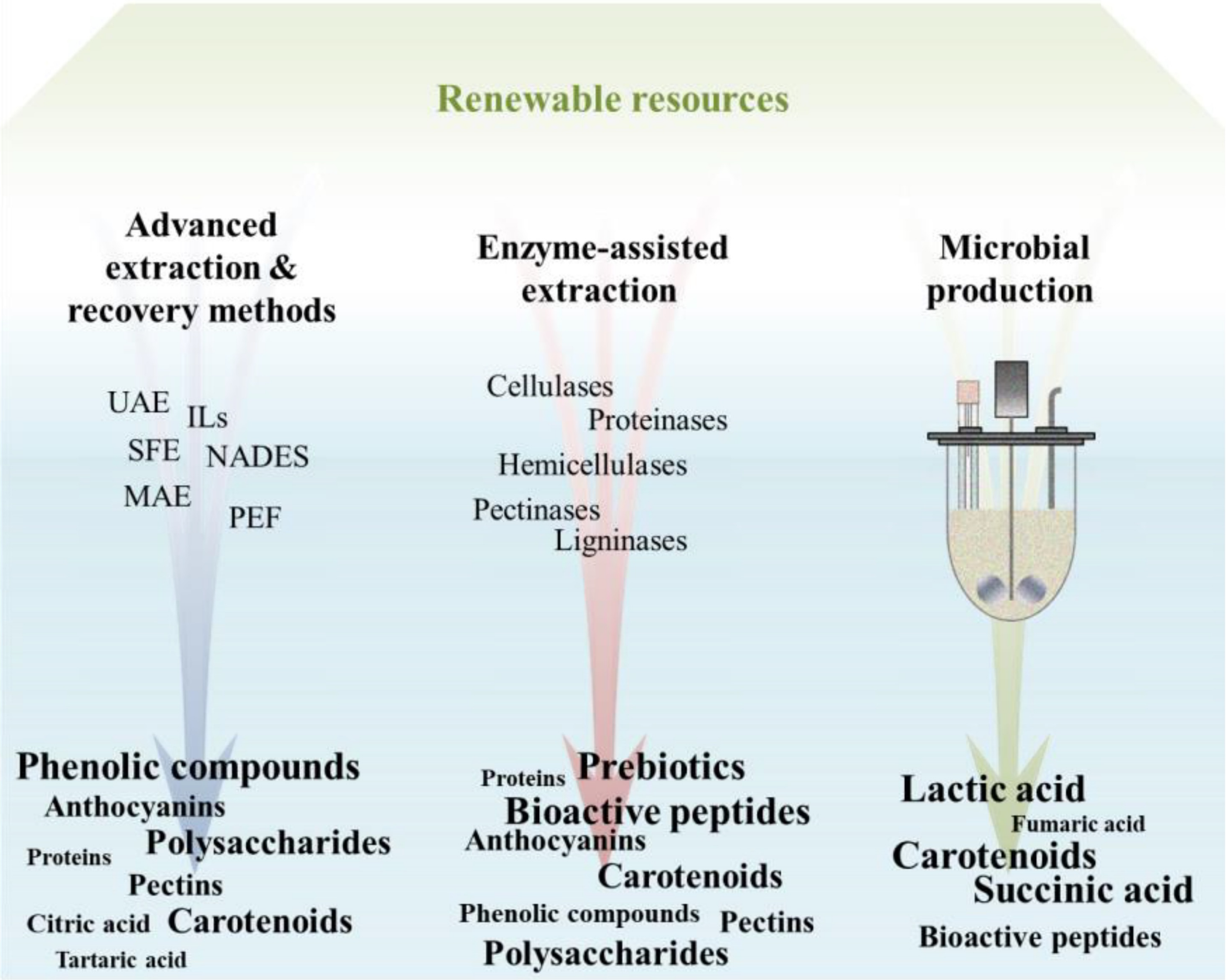
Ricardo Gómez-García [a, b], Débora A. Campos [a], Cristóbal N. Aguilar [b], Ana R. Madureira [a], Manuela Pintado [a]

BIOACTIVE COMPOUNDS

Bioactive compounds have the ability to interact with one or more components of living tissue by providing a **wide range of potential effects** and are derived from plant, animal, or other sources such as microorganisms, which are "**generally regarded as safe** (GRAS)"

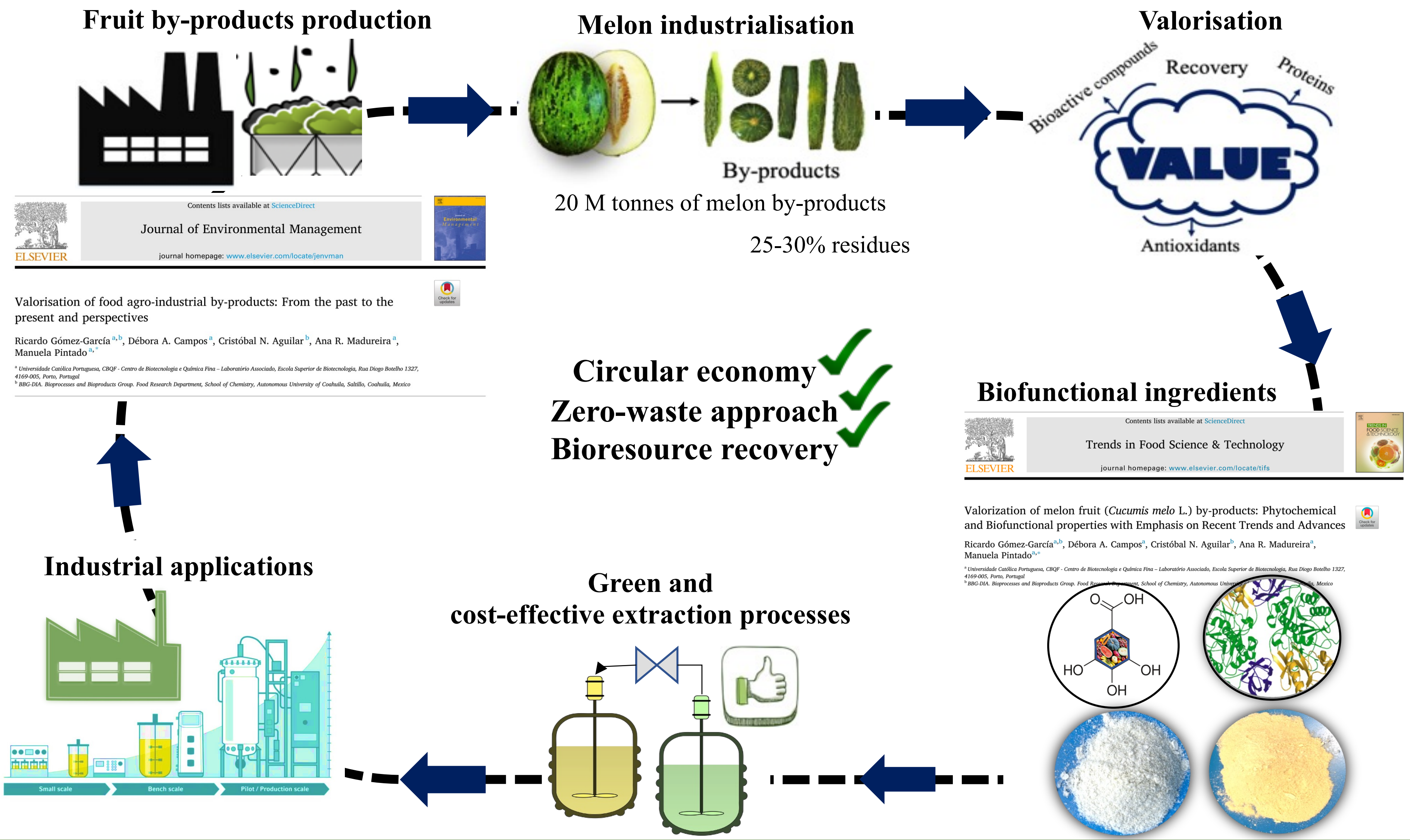
Various biological and functional activities such as antioxidant, anti-inflammatory, antidiabetic, anticancer, antiviral, and antitumor activities.

K. Banwo et al. Food Bioscience 43 (2021)

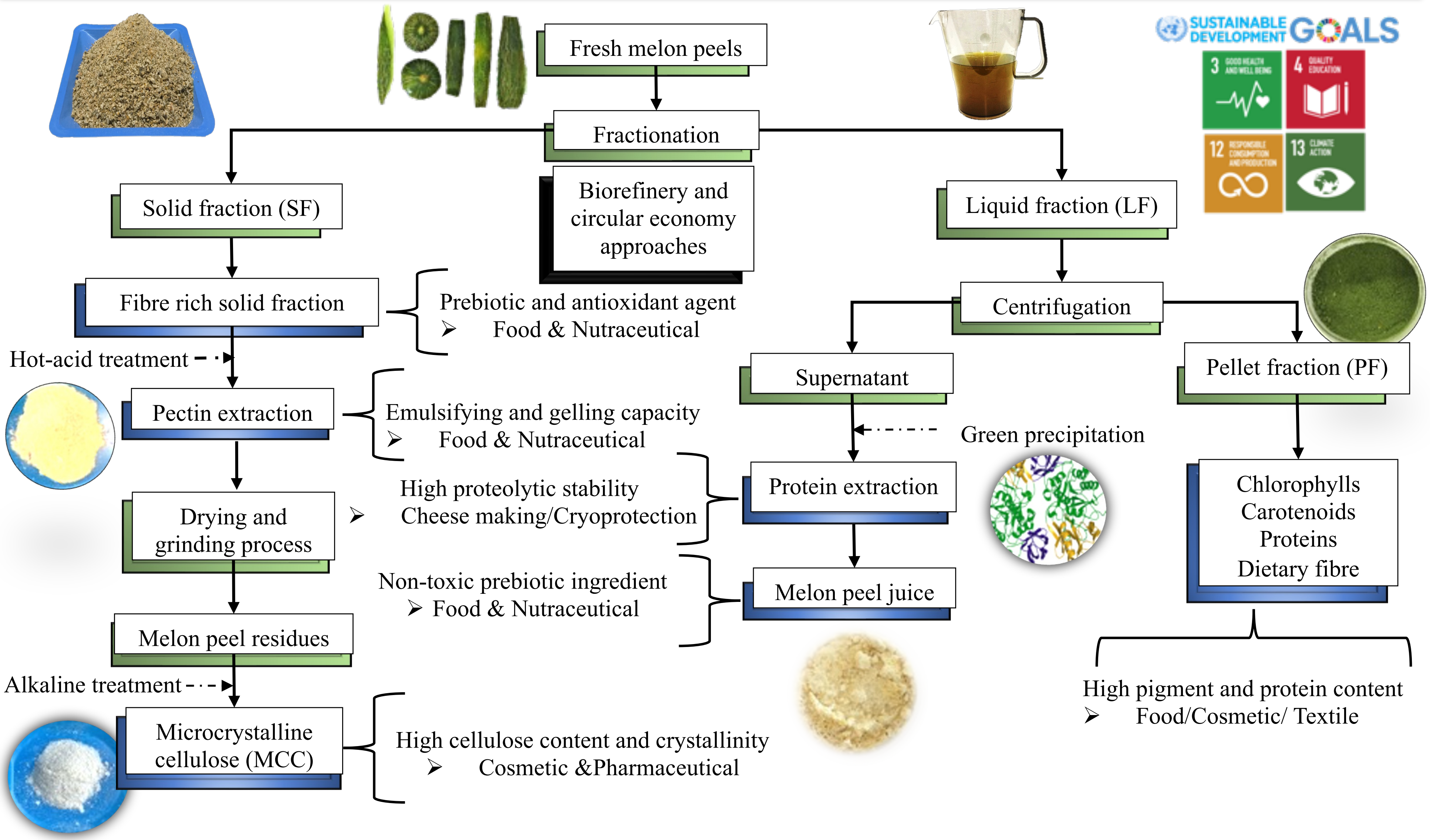


Sustainable Food Systems: The Case of Functional Compounds towards the Development of Clean Label Food Products, Foods 2022, 11, 2796. https://doi.org/10.3390/foods11182796

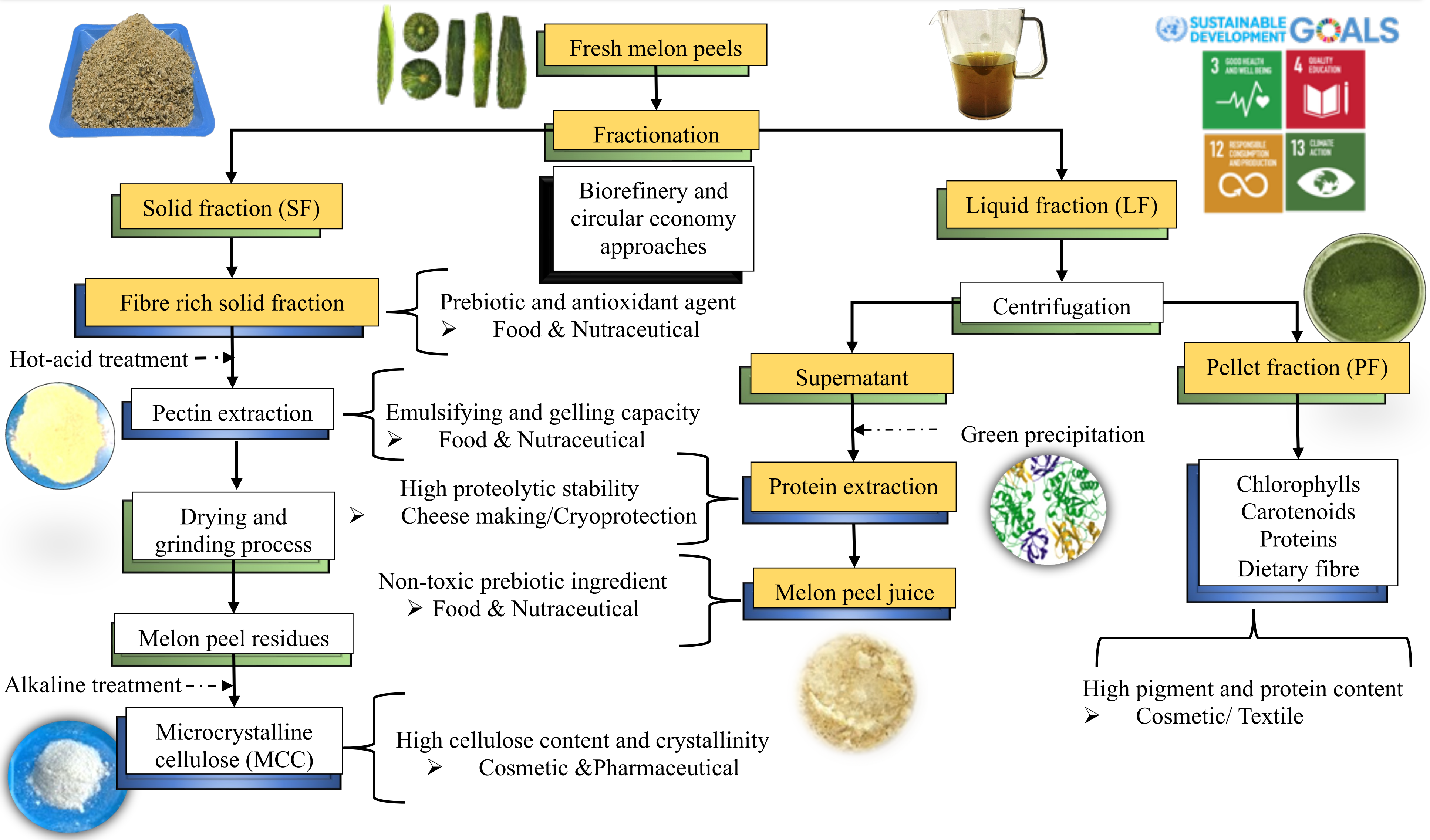
The Melon (*Cucumis melo* L.) by-products



Biorefinery multifunctional foods ingredients



Biorefinery multifunctional foods ingredients

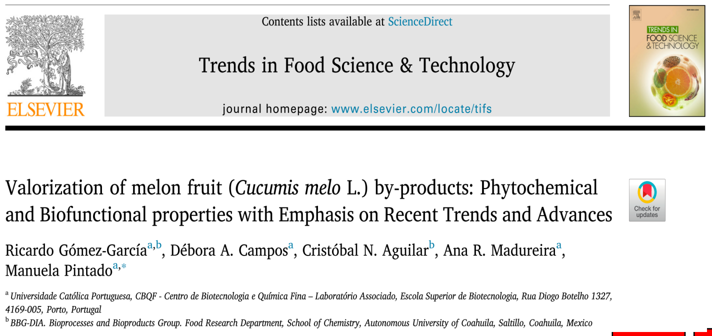


In vitro gastrointestinal tract (GIT) evaluation

Melon peel by-products a functional food ingredients

Solid Fraction **(SF)**

Liquid Fraction **(LF)**


Contents lists available at ScienceDirect

Trends in Food Science & Technology

journal homepage: www.elsevier.com/locate/tifs

Valorization of melon fruit (*Cucumis melo* L.) by-products: Phytochemical and Biofunctional properties with Emphasis on Recent Trends and Advances

Ricardo Gómez-García[a,b], Débora A. Campos[a], Cristóbal N. Aguilar[b], Ana R. Madureira[a], Manuela Pintado[a,*]

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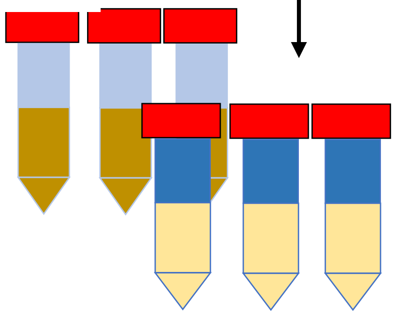
[b] BBG-DIA. Bioprocesses and Bioproducts Group. Food Research Department, School of Chemistry, Autonomous University of Coahuila, Saltillo, Coahuila, Mexico



Sample suspensions

Mouth
α-amylase (100 U/mL)

200 rpm | 37 ºC
2 min | pH 5-6

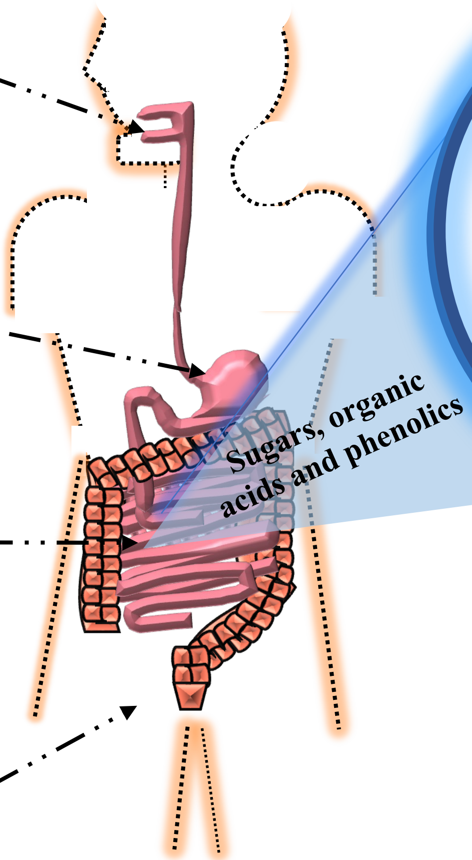
Stomach
Pepsin (25 mg/mL)

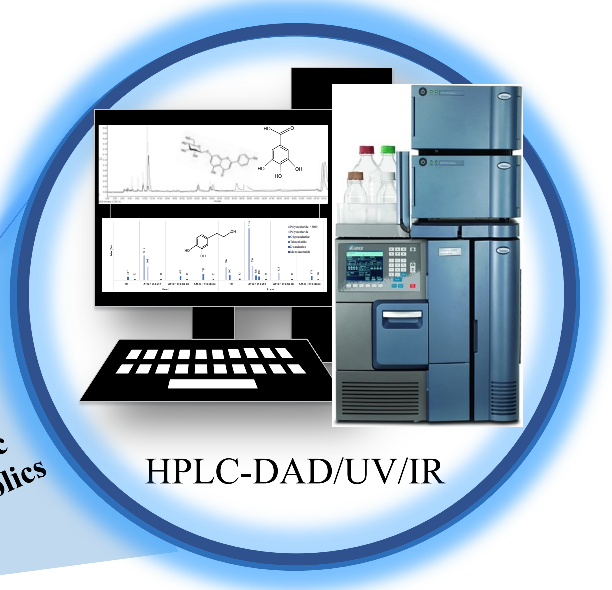
100 rpm | 37 ºC
120 min | pH 2

Small intestine
Pancreatin & Bile salts
(0.25 mL/mL)

60 min | 40 rpm
pH 6.5

Large intestine





HPLC-DAD/UV/IR

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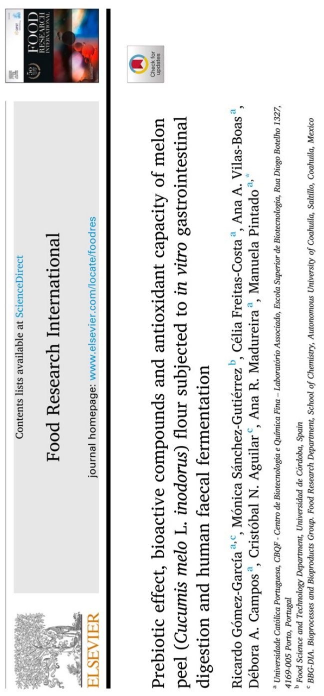
Gastrointestinal tract impact on melon peel flour – solid fraction



Phenolic content (TPC) and Antioxidant activity (AA)

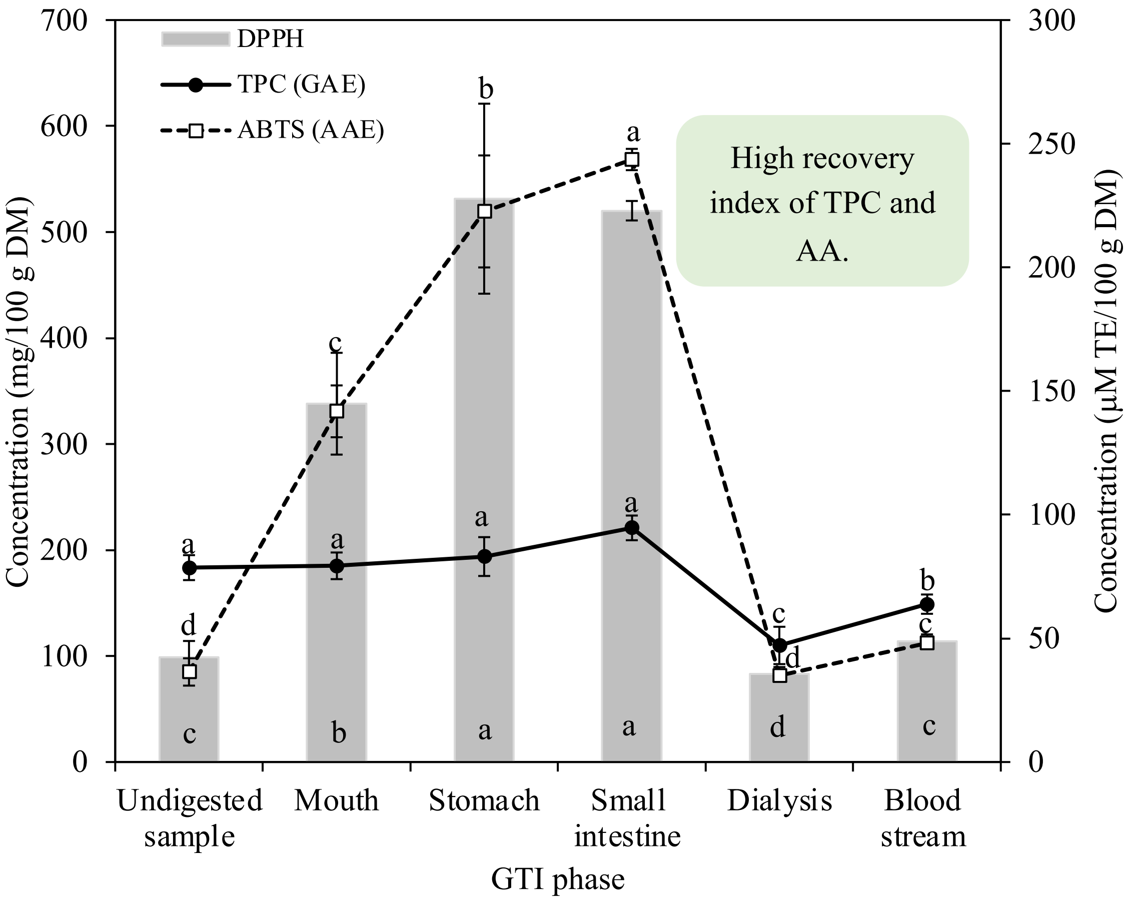


Figure 10. Total phenolic content (TPC) and antioxidant activity stability after each phase of the *in vitro* gastrointestinal tract (GIT).

Accessibility index (ACI %)	
TPC (mg GAE/100 g DM)	67.51 ± 4.49[a]
DPPH (µM TE/100 g DM)	21.94 ± 6.23[b]
ABTS (mg AAE/100 g DM)	19.73 ± 3.41[a]

Good accessibility index particularly **TPC**

Accessibility index (ACI %)	
Gallic acid	85.62 ± 0.05[a]
Hydroxytyrosol	UQ
Tyrosol	97.29 ± 0.06[a]
Chlorogenic acid	88.13 ± 0.05[a]
4-hydroxybenzoic	82.63 ± 0.42[a]
Caffeic acid	84.88 ± 1.20[a]
Luteolin-6-glycoside	13.28 ± 2.26[f]
p-coumaric acid	47.95 ± 16.78[c]
Ferulic acid	83.56 ± 0.12[a]

High accessibility of phenolics particularly **Tyrosol** and **Chlorogenic acid**

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INTERNATIONAL CONFERENCE ON SUSTAINABLE FOODS
Achieving the Sustainable Development Goals

Positive modulation of faecal microbial communities

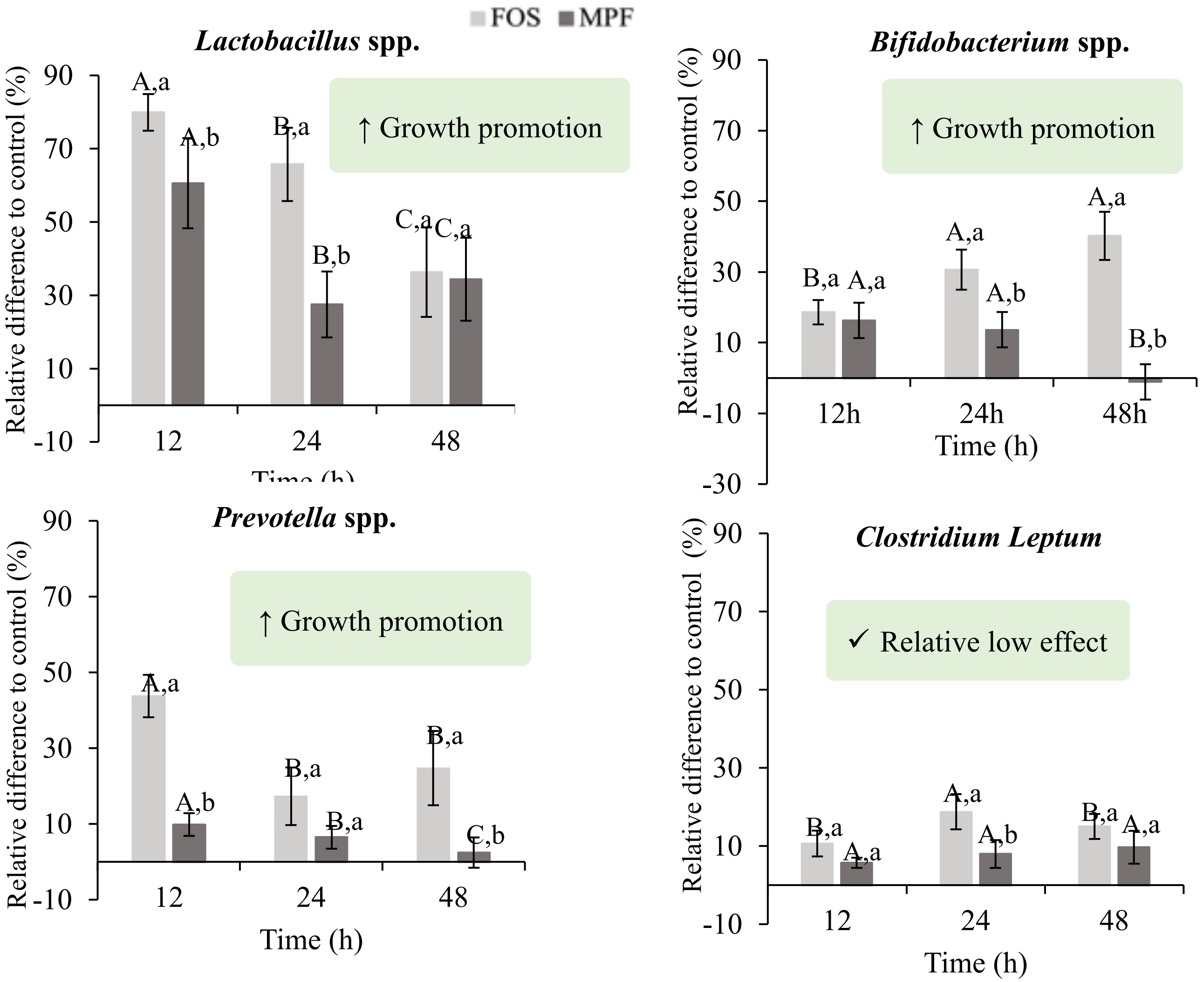
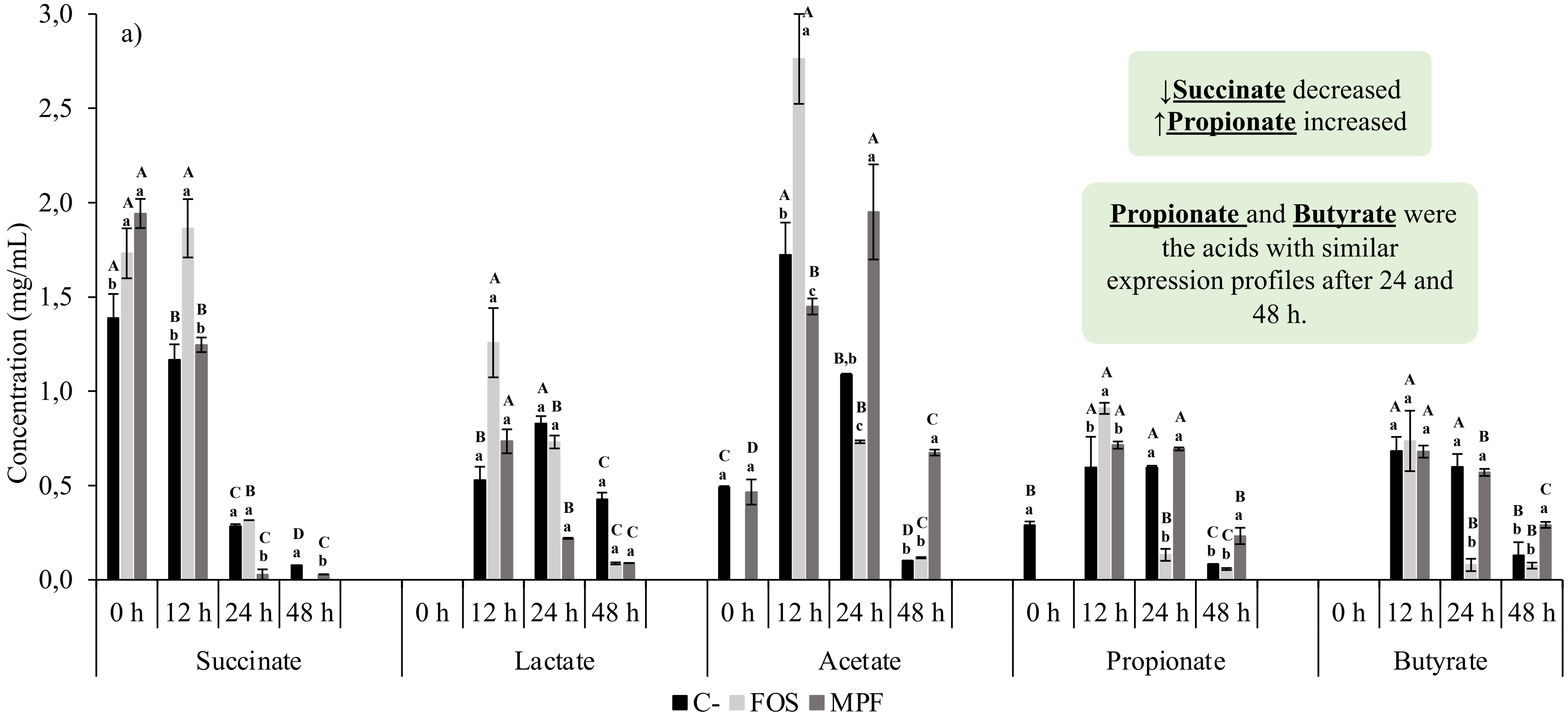


Figure 11. Relative differences to negative control throughout *in vitro* fecal fermentation.

Short-chain fatty acids (SCFA)

Production of **Acetate** > **Propionate** > **Butyrate**

Acetate was the SCFA with the highest concentration for MPF and FOS.



↓**Succinate** decreased
↑**Propionate** increased

Propionate and **Butyrate** were the acids with similar expression profiles after 24 and 48 h.

Figure 12. Concentration variation of organic acids and b) pH through the *in vitro* feacal fermentation. C-: negative control; FOS: positive control (2% w/v); MPF: melon peel flour (2% w/v).

The Lemon by-products

PRIMA
PARTNERSHIP FOR RESEARCH AND INNOVATION IN THE MEDITERRANEAN AREA

MEDI SMART

120 million tons of Citrus production per year

Lemons and limes are responsible for 15.9 million tons

Lemon Waste

- Industrial processes exploit only 45% of the total fruit weight;
- Peel (flavedo; 27%), pulp (albedo and endocarp; 26%), and seeds (2%), constitute a disposal rest.

Lemon Wastes in Frutas Tereso company:

- 14.2%, which represents 49 700 kg/year.

Valorisation of this by-product: using a circular economy approach

foods MDPI

Review

Functional Ingredients and Additives from Lemon by-Products and Their Applications in Food Preservation: A Review

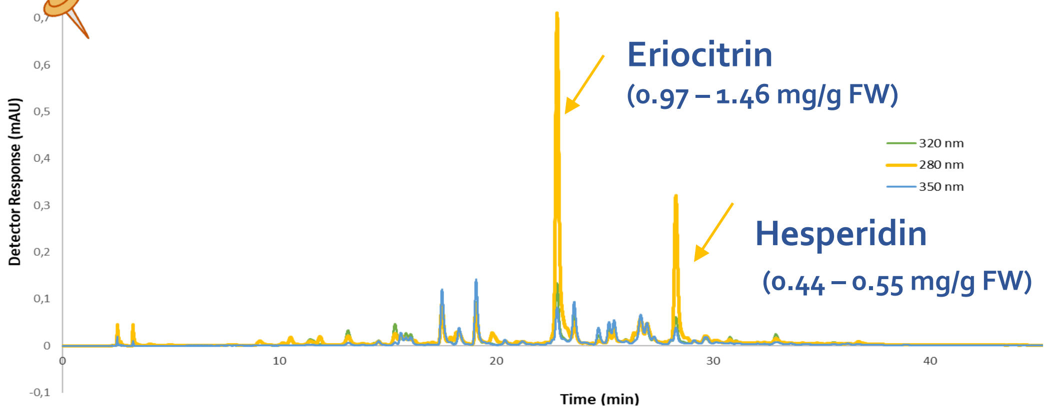
Daniela Magalhães, Ana A. Vilas-Boas, Paula Teixeira and Manuela Pintado *

Development and characterization of LP fractions

Citrus limon from Algarve (Portugal)

3 LOTs; Variety: Eureka; Caliber: 4/5;

Phenolic compounds identification and quantification



Nutritional composition

Component	LOT 1	LOT 2	LOT 3
Moisture (% w/w)	82.71 ± 0.19	79.93 ± 0.19	85.16 ± 0.06
Expressed in dry basis (% w/w)			
Ash	4.38 ± 0.02	4.23 ± 0.27	3.66 ± 0.03
Protein	5.35 ± 0.08	5.36 ± 0.05	5.39 ± 0.05
Lipids	1.04 ± 0.07	1.09 ± 0.03	0.91± 0.03
Carbohydrates	89.06*	89.12*	89.89*
Total Dietary Fiber	41.16	43.21	40.76

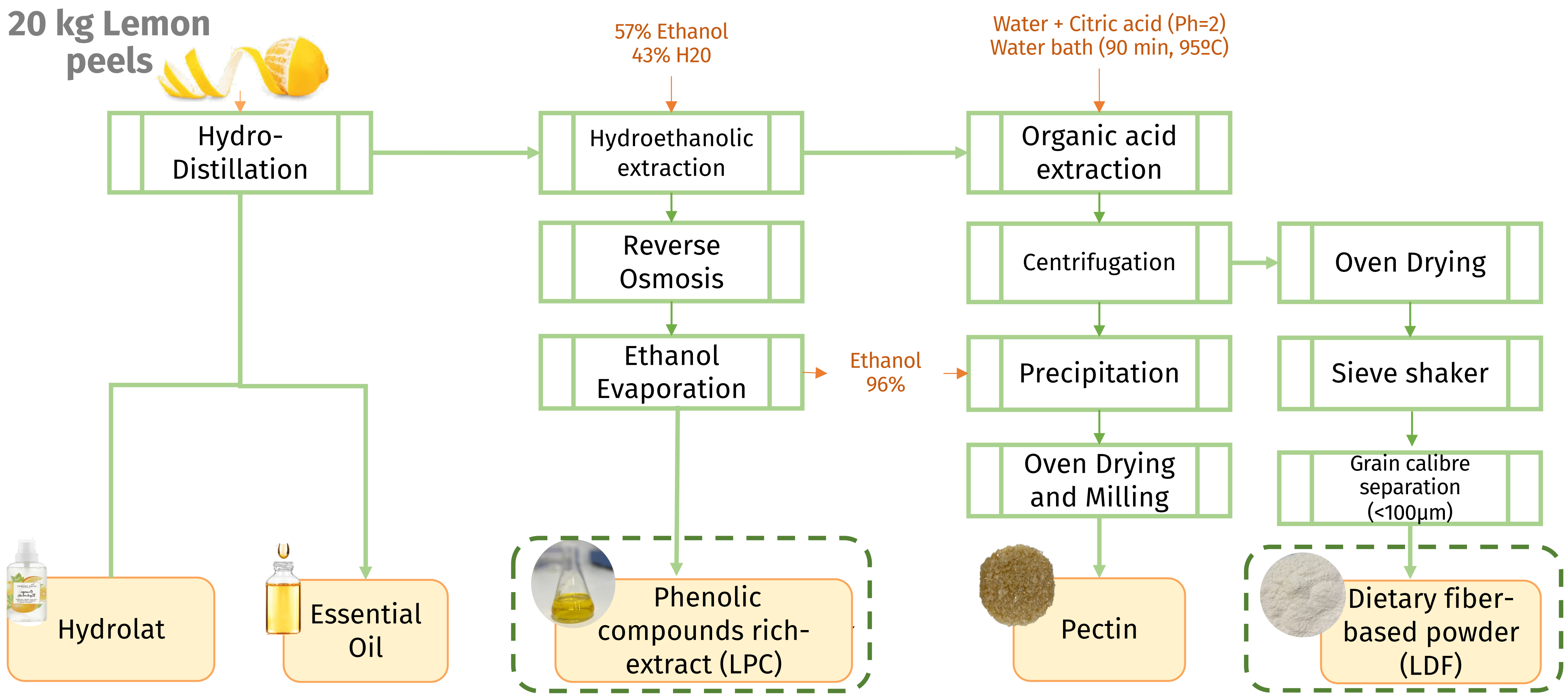
*Total carbohydrates content obtained by difference

Antioxidant activity

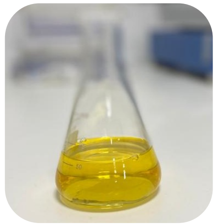
	CONCENTRATION		
PARAMETER	**LOT 1**	**LOT 2**	**LOT 3**
Folin-Ciocalteu (mg GAE/g FW)	2.30 ± 0.26	2.31±0.26	2.26±0.23
ABTS Assay (mg AAE/g FW)	2.33 ± 0.08	2.08±0.07	2.08±0.07
DPPH Assay (mg TE/g FW)	0.85 ± 0.10	0.77±0.06	0.73±0.06
ORAC Assay (umol TE/mL FW)	57.82±3.16	54.36±3.95	52.12±2.01

Abb.: gallic acid equivalent, GAE; Ascorbic acid equivalent, AAE; Trolox equivalent, TE; Fresh weight, FW.

New Bioactives – an integral approach



Application of functional ingredients in food models



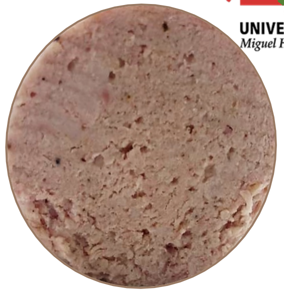
Lemon phenolic compounds (LPC) extract

Lemon dietary fiber (LDF)

***Mortadella* Control**

***Mortadella* LDF (3%)**

***Mortadella* LPC (1%)**



***Mortadella* LDF (3%) + LPC (1%)**

Chemical composition (Day 0)

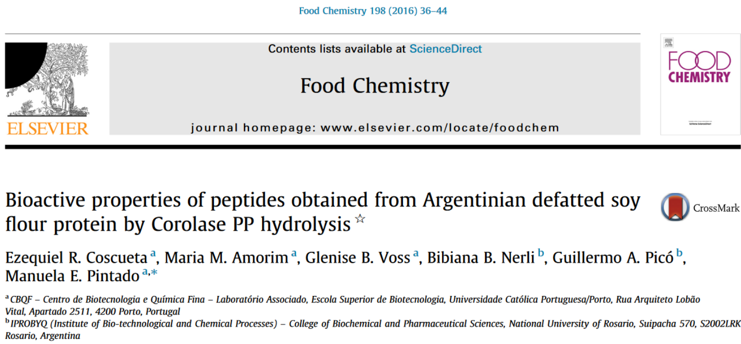
Formulation	Moisture (%)	Fat (%)	Protein (%)	Ash (%)	aw
Control	65.46 ± 0.00	15.05 ± 0.32	15.87 ± 0.75	2.28 ± 0.00	0.959 ± 0.000
LDF	62.67 ± 0.00	14.70 ± 0.12	16.17 ± 0.69	2.17 ± 0.00	0.959 ± 0.000
LPC	65.65 ± 0.00	15.78 ± 0.76	15.27 ± 0.43	1.88 ± 0.00	0.958 ± 0.001
LDF+PC	64.25 ± 0.00	15.10 ± 0.26	15.98 ± 0.09	2.08 ± 0.00	0.958 ± 0.001

Microbiological analysis (Day 0 at Day 28)

- *Enterobacteriacea*, total aerobic bacteria and yeast and moulds were not detected in LDF and LPC reformulated *mortadellas* during 28 days;
- Total aerobic bacteria (2.66 log cfu/g) was deteted at day 28 for *mortadella* control.
- ***LDF and LPC has a stronger antioxidant activity/antimicrobial activity than sodium ascorbate used in mortadella Control.***

- ***Reformulated mortadellas are a good source of fiber, and phenolic compounds;***
- ***Mortadellas with LDF could increase the %protein.***

Defatted soy flour protein


Food Chemistry 198 (2016) 36–44
Contents lists available at ScienceDirect
Food Chemistry
journal homepage: www.elsevier.com/locate/foodchem

Bioactive properties of peptides obtained from Argentinian defatted soy flour protein by Corolase PP hydrolysis ☆

Ezequiel R. Coscueta [a], Maria M. Amorim [a], Glenise B. Voss [a], Bibiana B. Nerli [b], Guillermo A. Picó [b], Manuela E. Pintado [a,*]

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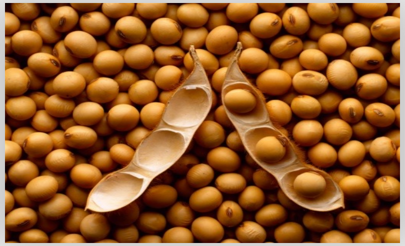
[b] IPROBYQ (Institute of Bio-technological and Chemical Processes) – College of Biochemical and Pharmaceutical Sciences, National University of Rosario, Suipacha 570, S2002LRK Rosario, Argentina

Soybean is one of the most important alternative protein source

Soybean meal is the most important co-product of soybean processing

Low added value **Valorisation**



Generate functional ingredients: Bioactive properties such as antioxidant or antihypertensive properties

Protein hydrolysis

Project BiValBi – Biotechnologies to Valorise the regional Biodiversity in Latin America (Refª PIRSESGA-2013-611493 BI_1)

BIOACTIVE PEPTIDES OBTAINED FROM ARGENTINIAN DEFATTED SOY FLOUR PROTEIN

EXTRACTION

70 °C ,1 h (SPI70)
90 °C, 30 min (SPI90)

Hydrolysis

Protein suspension 1%;
Ratio enzyme:substrate (10 mg enzyme/g SPI).
Enzyme: Corolase PP
Hydrolysis (SPH) 50 °C pH 8: 30 min, 1, 2, 3, 4, 5, 6, 8, 10 y 24 h

GIT

Stomach

pH 2.0, 37 ° C, pepsin at 25 mg / mL, stirred at 130 rpm for 60 min

Intestine

pH 6.5, 37 ° C, for each mL of sample 0.25 mL of pancreatic juice (2 mg / mL of pancreatin solution and 12 mg / mL bile salts diluted in 100mM $NaHCO_3$ solution), stirred at 45 rpm for 90 min

Bioactivity

Antioxidant activity

ABTS (Re et al., 1999)
ORAC (Contreras et al., 2011)

Antihipertensive activity

ACE inhibition (Quiros et al., 2009)

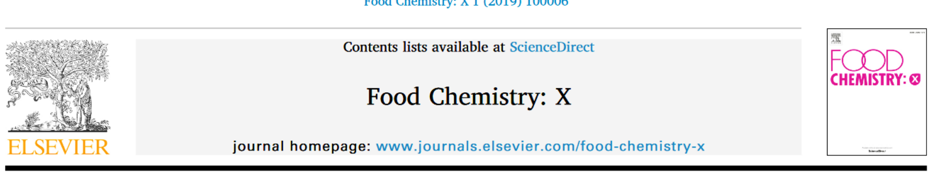
BIOACTIVE PEPTIDES FROM SOY FLOUR PROTEIN – BIOLOGICAL ACTIVITIES

Table 1. Results of degree of hydrolysis and angiotensin-converting enzyme (ACE) inhibitory activity of total hydrolysates at three different times of hydrolysis.a

Time of hydrolysis (h)	SPH	
	Degree of hydrolysis (%)	ACE-inhibitory activity (IC_{50}, µg/ mL)
0	0.90 ± 0.03	>1000
2	5.51 ± 0.31	274.2 ± 2.5*
4	7.33 ± 0.28	221.0 ± 21.6*
10	18.88 ± 1.89	236.5 ± 13.5*

aAnalysis of variance was used to estimate the effects of the ACE-inhibitory activity from every substrate: Tukey's test: *p < 0.001, using hydrolysis at 0h (SPI) as null hypothesis. Values are expressed as average ± standard error (n= 2 for both, degree of hydrolysis and angiotensin-converting enzyme-inhibitory activity).

IMPACT OF GIT


Food Chemistry: X 1 (2019) 100006
Contents lists available at ScienceDirect
Food Chemistry: X
journal homepage: www.journals.elsevier.com/food-chemistry-x

Enzymatic soy protein hydrolysis: A tool for biofunctional food ingredient production

Ezequiel R. Coscueta[a,b], Débora A. Campos[a], Hugo Osório[c,d,e], Bibiana B. Nerli[b], Manuela Pintado[a,*]

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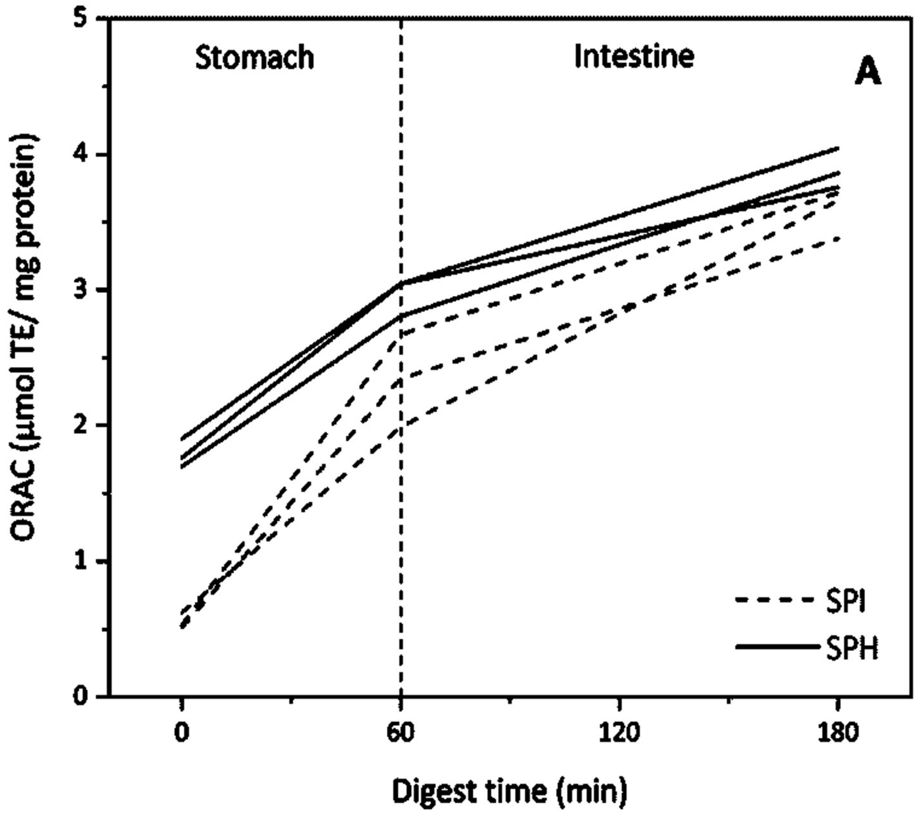
Hydrolyse or not hydrolyse?



Variation of bioactivity during the time of in vitro simulated gastro-intestinal digestion. The lines display the observations of ORAC (A) and iACE (B) for each experimental unit.

Both substrates (SPI and SPH) were observed to present interesting and similar antioxidant properties after GIT

Pre-hydrolysis with Corolase PP does not clearly improve the analysed bioactivities of the intact soy protein isolate.

Food Chemistry: X 1 (2019) 100006

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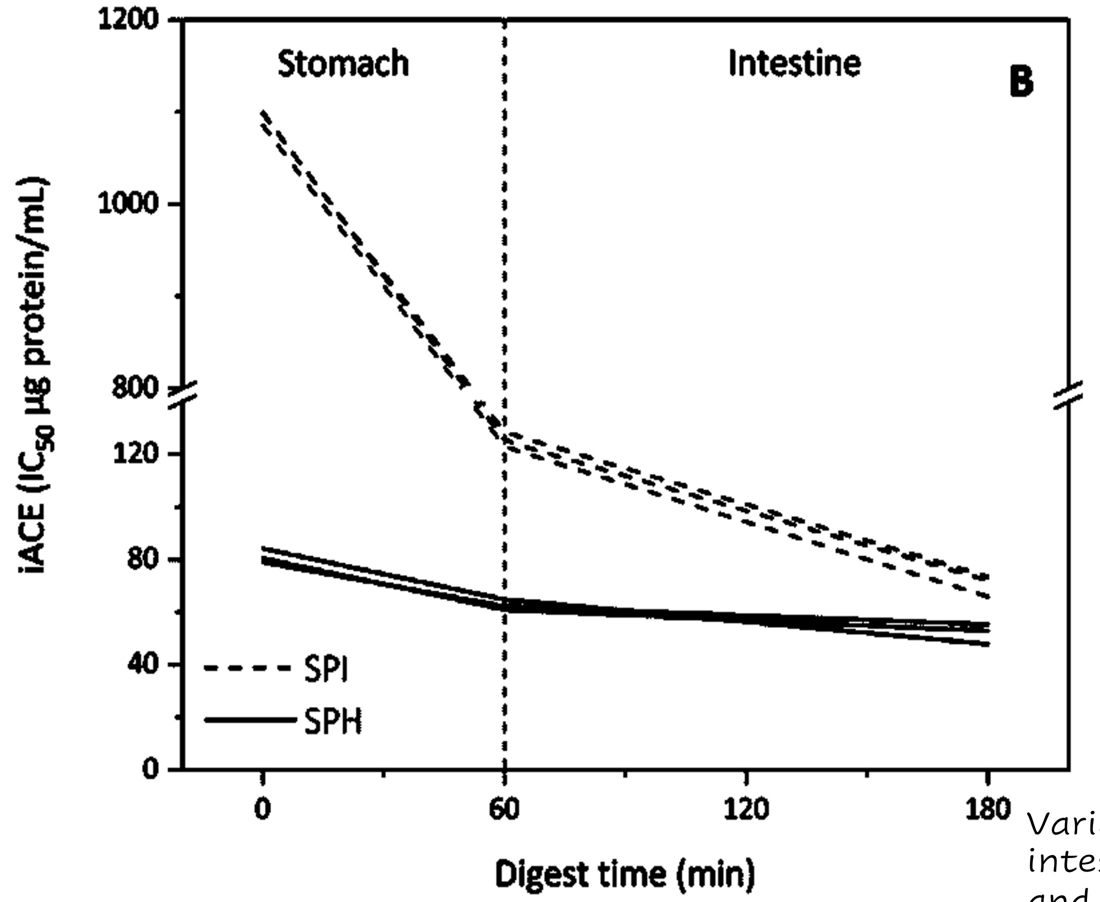
Hydrolyse or not hydrolyse?



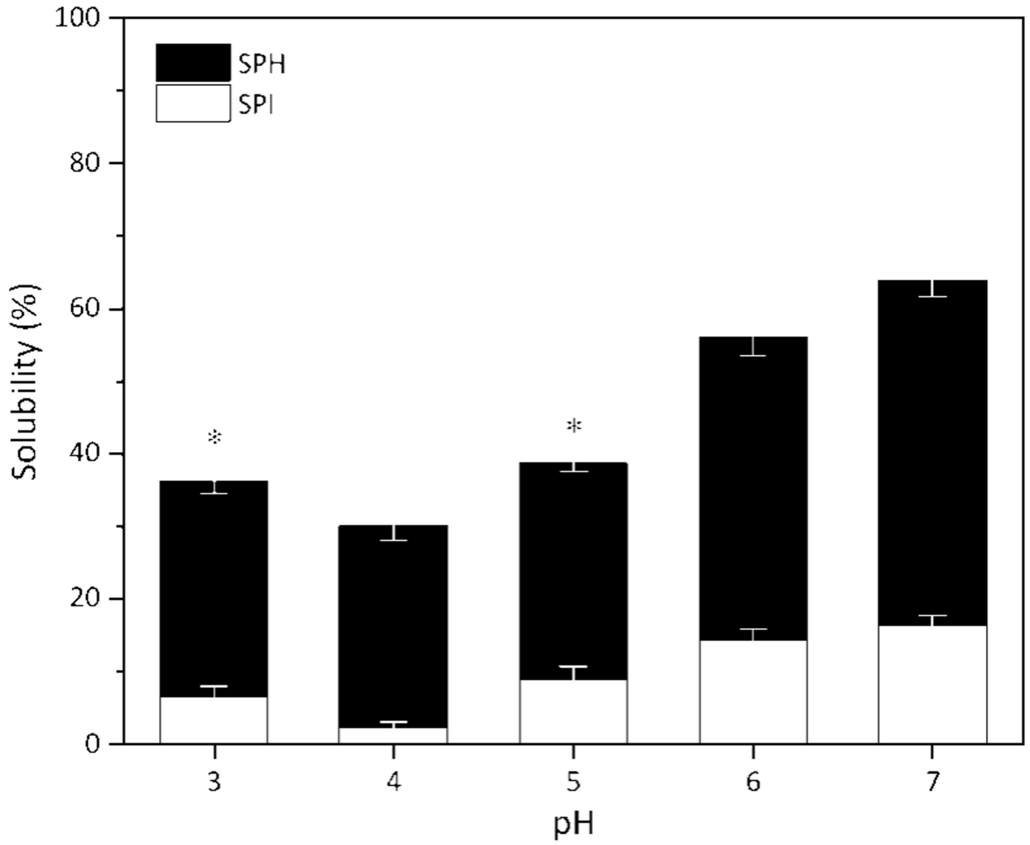
Variation of bioactivity during the time of in vitro simulated gastro-intestinal digestion. The lines display the observations of ORAC (A) and iACE (B) for each experimental unit.

Both substrates (SPI and SPH) were observed to present interesting and similar antihypertensive properties after GIT

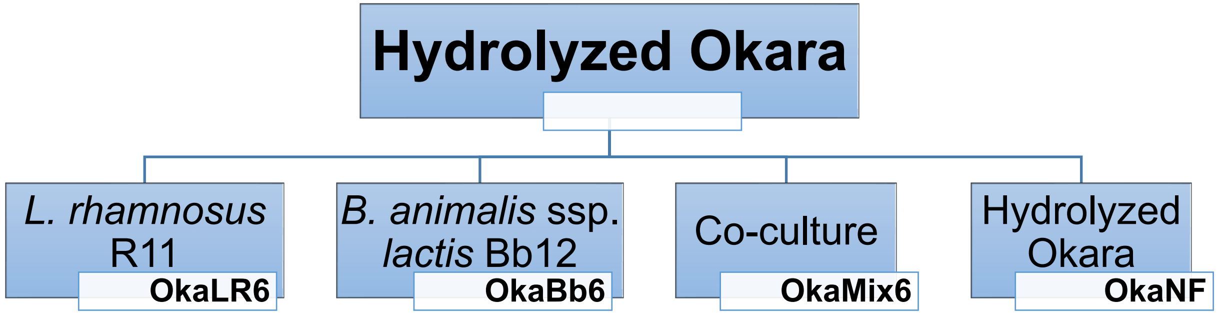
Pre-hydrolysis with Corolase PP does not clearly improve the analysed bioactivities of the intact soy protein isolate.

Hydrolyse or not hydrolyse?

The hydrolysis of soy protein with Corolase PP increases the solubility of the intact SPI between pH 3–7.
At pH 7 the solubility of SPH was approximately 4- fold higher than that of SPI.

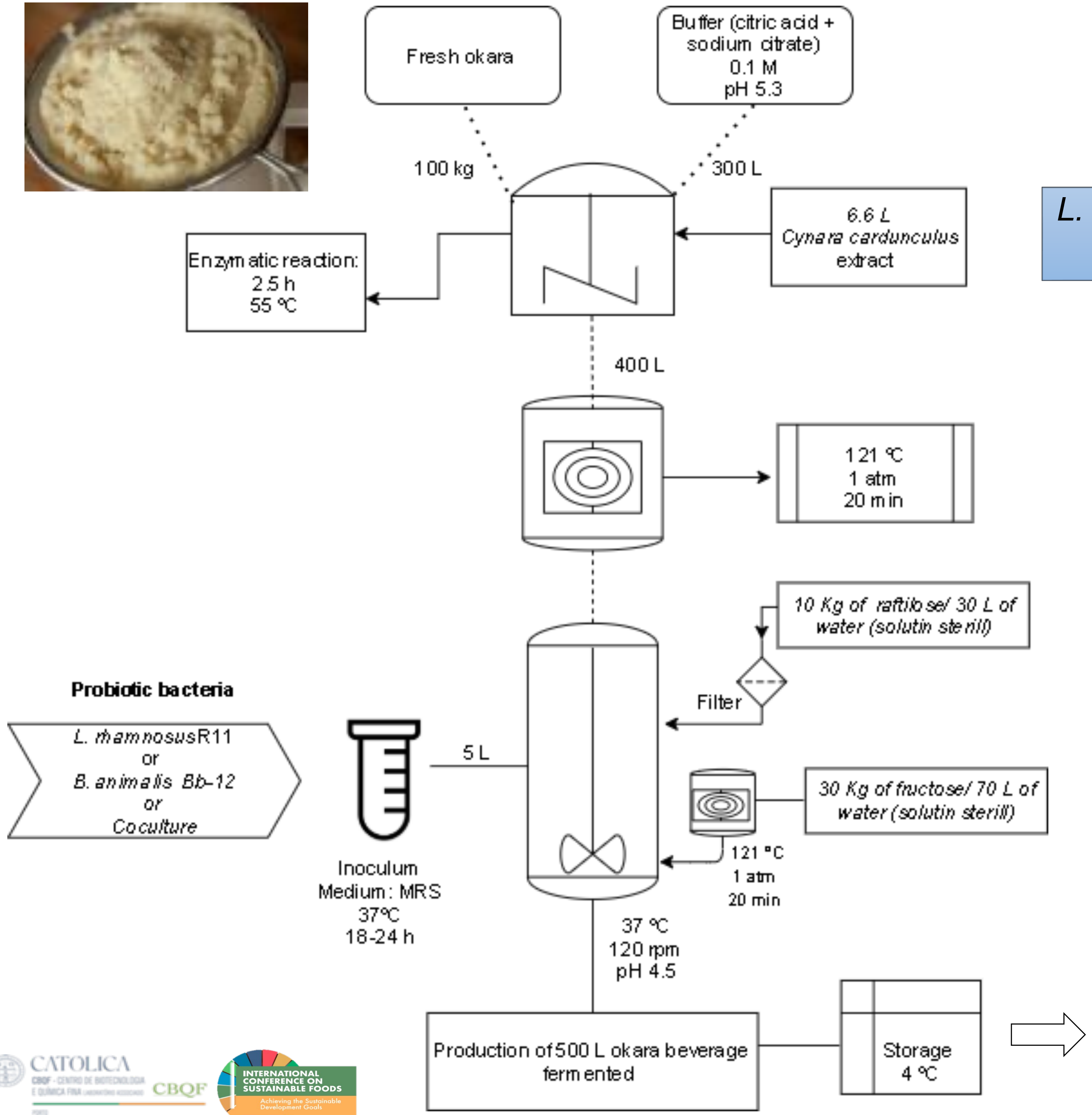


Water solubility profiles of SPI and SPH at different pHs. Bars represent means of three independent experiments. Error bars represent sample standard deviation (SD).

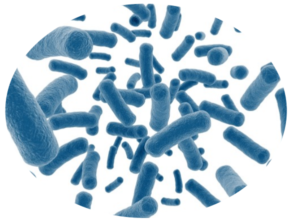


Production of Okara beverage

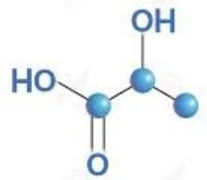
- Prebiotic
- Probiotic



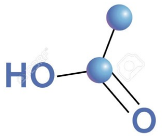
Fermentation of okara



Okara beverage maintained the total viable cells counts(*B. animalis* Bb12 and *L. rhamnosus* R11: 9 log CFU/mL) throughout storage.



⇧ Lactic acid

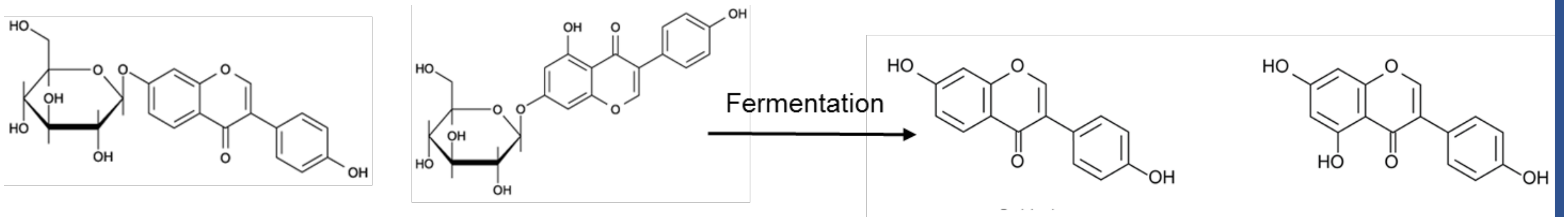


⇩ Acetic acid

Isoflavones

Glycosides

Aglycones



Daidzin

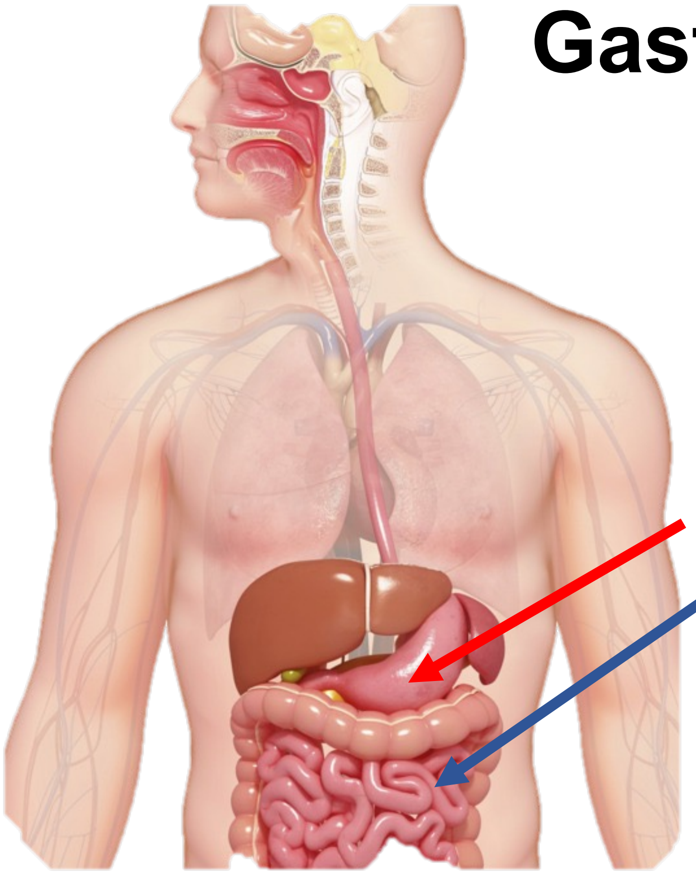
Genistin

Daidzein

Genistein

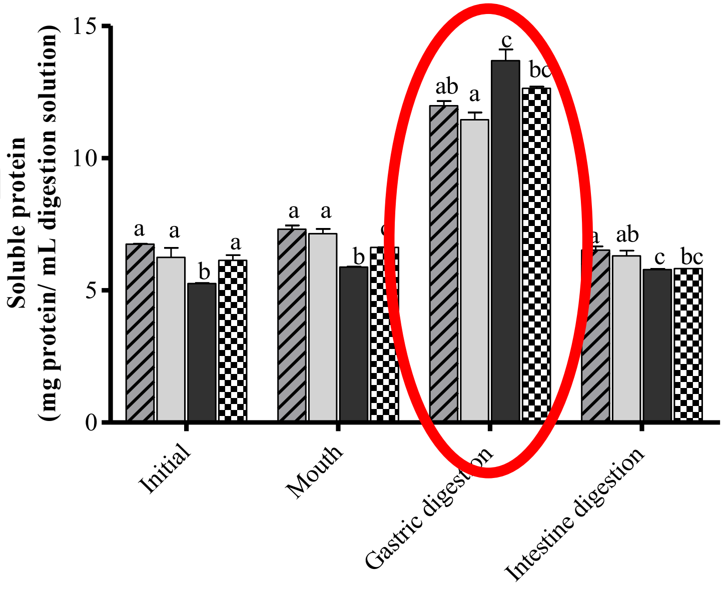
Okara beverage: stability, functionality and impact of gastrointestinal tract

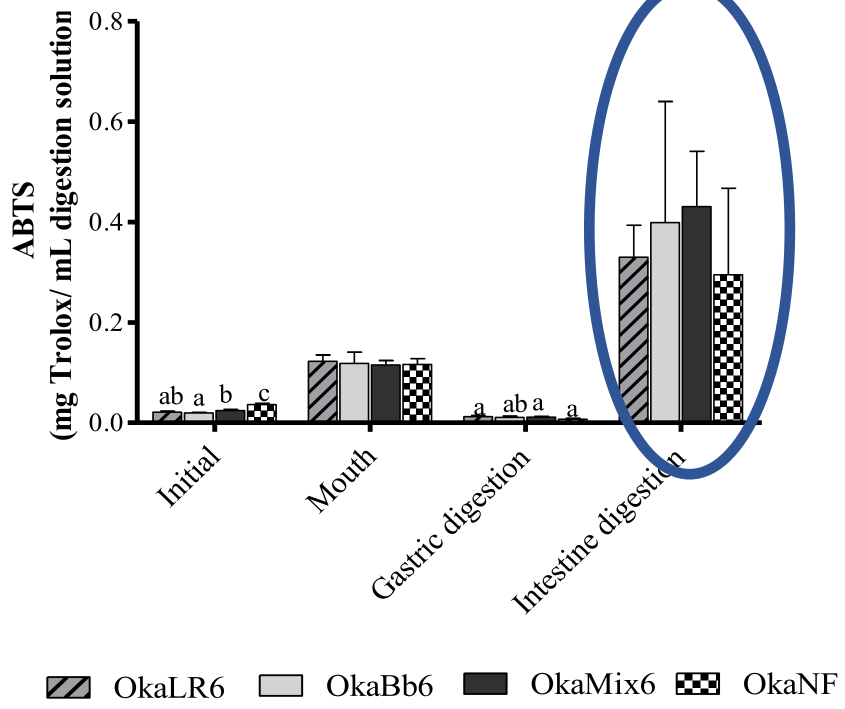
Gastrointestinal digestion



- ✓ Antioxidant activity
- ✓ Ace-inhibitory activity

Okara beverage	Non-digested	Intestine digestion
	IC50 (µg protein/mL)	
OkaLR6	750.83 ± 27.37[Aa]	183.04 ± 6.31[Ba]
OkaBb6	752.21 ± 6.10[Aa]	149.83 ± 22.36[Ba]
OkaMix6	752.37 ± 7.82[Aa]	136.75 ± 12.85[Ba]
OkaNF	442.50 ± 21.02[Ab]	273.54 ± 13.17[Bb]





Tomato byproducts

Impact of extraction method on the Bioactivities of Tomato flour

foods

MDPI

Article

In Vitro Gastrointestinal Digestion Impact on the Bioaccessibility and Antioxidant Capacity of Bioactive Compounds from Tomato Flours Obtained after Conventional and Ohmic Heating Extraction

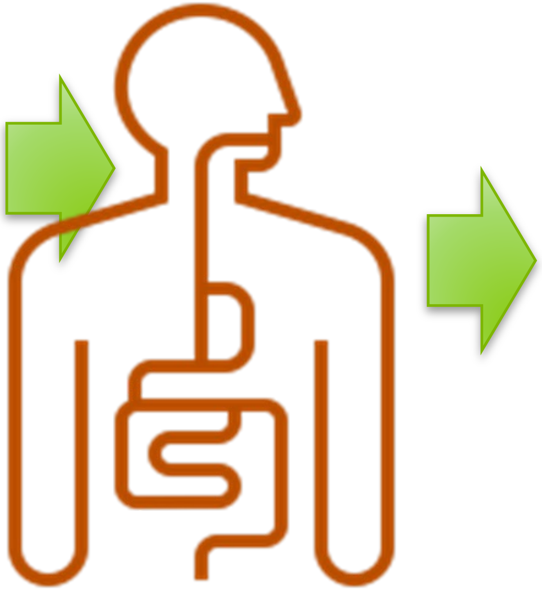
Marta C. Coelho [1,2], Tânia B. Ribeiro [1,3], Carla Oliveira [1], Patricia Batista [1], Pedro Castro [1], Ana Rita Monforte [1], António Sebastião Rodrigues [4], José Teixeira [2] and Manuela Pintado [1,*]

Gastrointestinal tract simulation

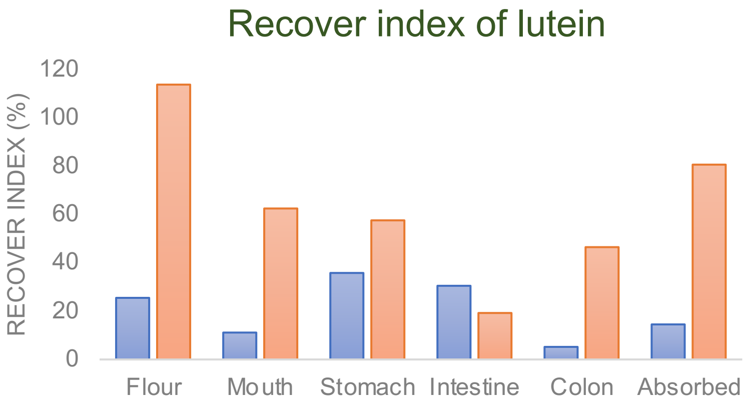
Microbiota modulation

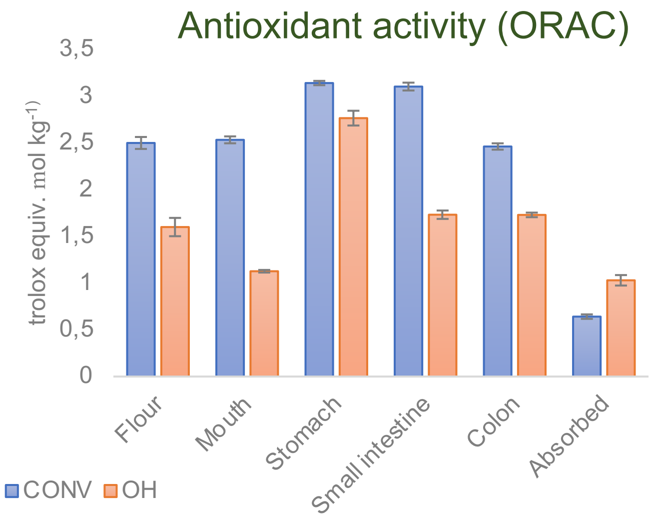
Fraction obtained after ohmic heating extraction

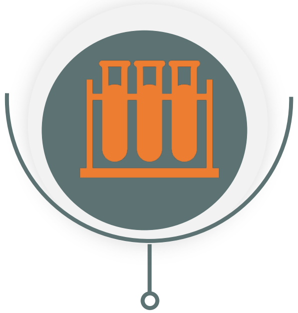
Flraction obtained after conventional extraction



Bioactivities from Tomato flour after gastrointestinal tract





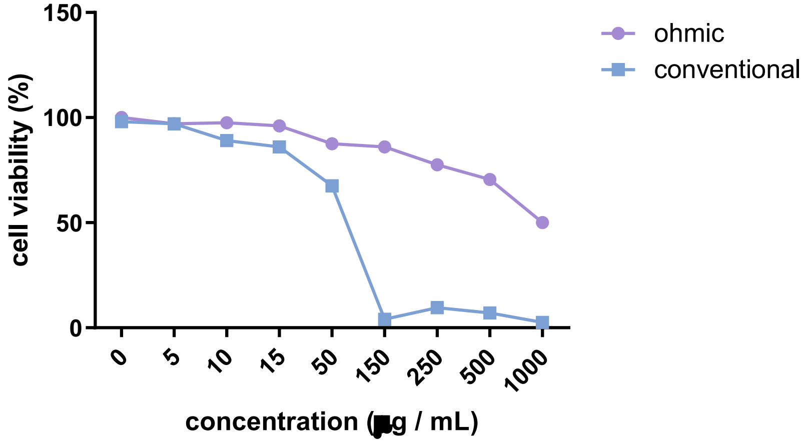


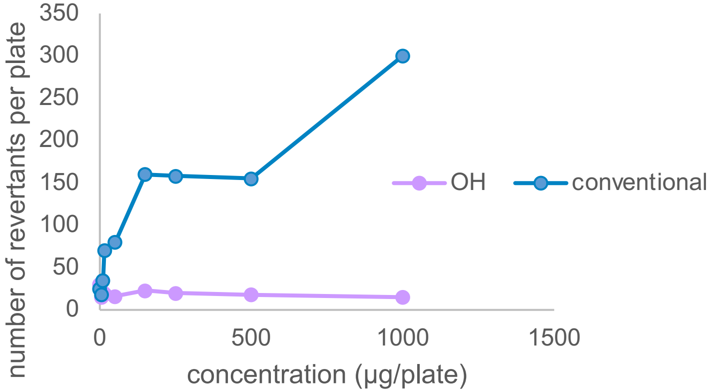
Bioaccessibility

OH increased the bioaccessibility of carotenoids

Lycopene, lutein, and phytofluene;
Lutein present in OH is more absorbed than in CONV

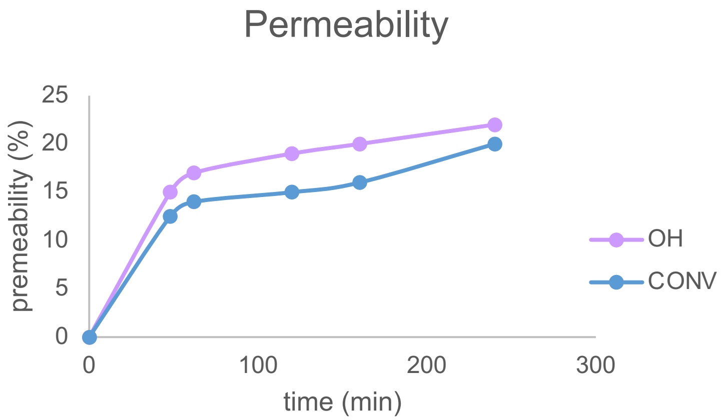
Bioactivities from Tomato flour after gastrointestinal tract





OH impact

SFOH is safer and contains more bioaccessible bioactive compounds than SFCONV



- OH solid fraction is non mutagenic and don't present cytotoxicity
- CONV present mutagenicity

A4. Valorização de Vegetais

- Drying studies have successfully produced new vegetable flours (potatoes, arugula, spinach, parsley, watercress, coriander, carrots and tomatoes), as well as frozen pulp..

Dehydrated and shredded vegetables (powder)

Frozen vegetable pulps UCP

- Impact of freezing and drying on the nutritional and functional profile of watercress, carrots, spinach, arugula and tomato shows high functional and nutritional value for 6 months.

horticulturae MDPI

Article

Development of Frozen Pulps and Powders from Carrot and Tomato by-Products: Impact of Processing and Storage Time on Bioactive and Biological Properties

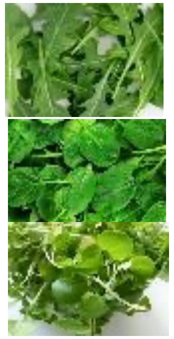
Helena Araújo-Rodrigues [1], Diva Santos [1], Débora A. Campos [1], Modesta Ratinho [2], Ivo M. Rodrigues [3] and Manuela E. Pintado [2,*]

foods MDPI

Article

Impact of processing approach and storage time on bioactive and biological properties of rocket, spinach and watercress by-products

Helena Araújo-Rodrigues[1], Diva Santos[1], Débora A. Campos[1], Suse Guerreiro[2], Modesta Ratinho[2], Ivo M. Rodrigues[3] and Manuela M. Pintado[1,*]



Innovative AGROFOOD products based on vegetable losses

Plant based ham
(Primor/Vitacress)

"Super" spinach spread and mix spread (allergen free)

(Vitacress / UCP)

"Super" functional puree (allergen free)

(Vitacress / UCP)

Plant based spread

(A Poveira)

+ Formulation of dehydrated vegetable bases for aquaculture – see bass

UCP/ESAC/Vitacress/PRIMOR/A Poveira

MOBFOOD

CRUCIFEROUS VEGETABLES

Solution

PHYTOCHEMICALS: A current strategy to prevent and treat chronic diseases, including cancer

Stand out

Brassicaceae

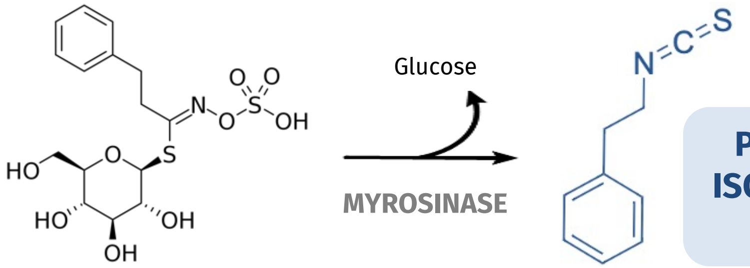
CRUCIFEROUS VEGETABLES have a ⇈ content of **GLUCOSINOLATES (GLs)**

Phytochemicals:
ISOTHIOCYANATES (ITCs)

Watercress
(*Nasturtium officinale*)
"superfood"

GLUCOSINOLATE

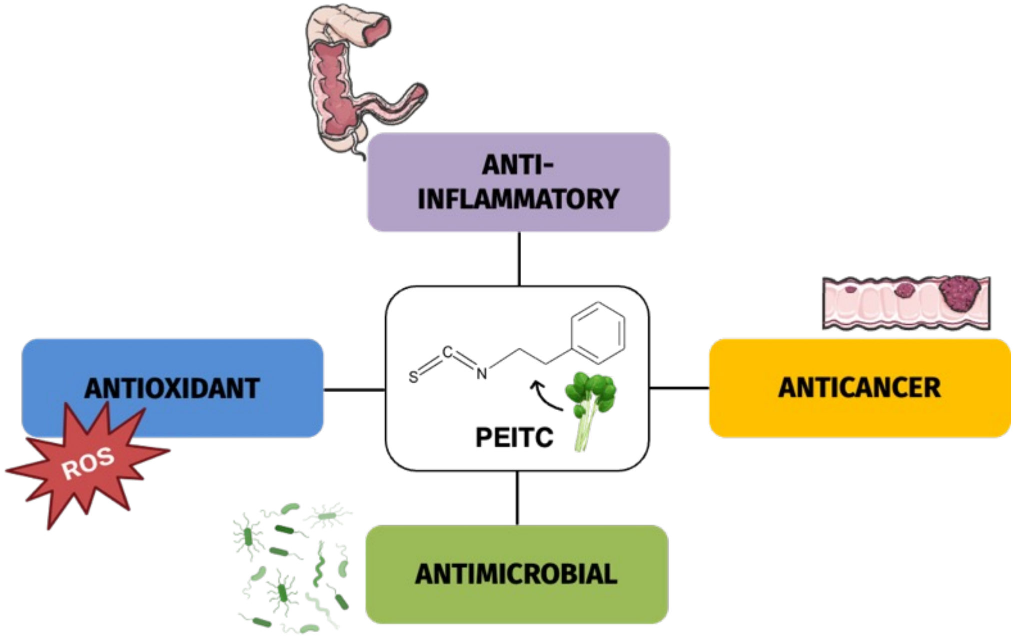
ISOTHIOCYANATE



PHENYLETHYL ISOTHIOCYANATE (PEITC)

Phenylethyl glucosinolate

What are the health benefits of PEITC?



PEITC is a powerful chemopreventive agente and boosts health

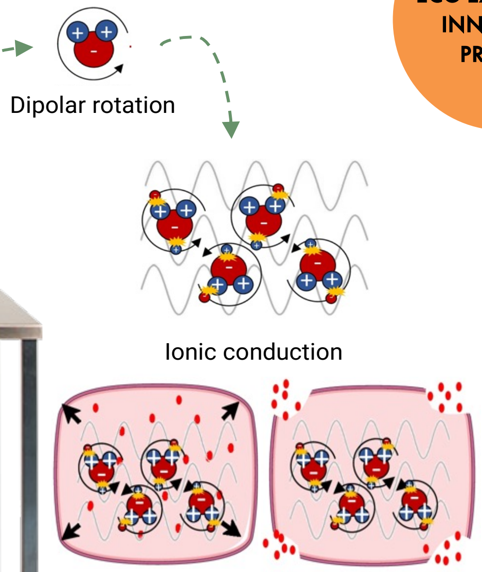
PEITC exhibits high instability, which has jeopardized its industrialisation

How can we extract PEITC?

NEW

For the first time, **Microwave Hydrodiffusion and Gravity (MHG)** was applied as a **green tool** for **PEITC** extraction

https://www.milestonesrl.com



Heating destroys the vegetable cells and transfers the compounds outside the plant material

Adapted from Moraes, D.P. *et al.* *Food Bioprocess Technol* **15**, 1936–1947 (2022)

ECO-EXTRACTION INNOVATIVE PROCESS

Solvent-free

Solvent-free extraction relies on 3 main factors: microwave power, irradiation time, and humidity of the plant material

Scalability

MHG extraction is adaptable to different scales of production.

Rapid Extraction

MHG extraction is faster than conventional methods

Energy Efficiency

Decreases the amount of energy and CO_2 emissions

The leftover biomass can be used for other purposes

Plant biomass can be repurposed after MHG extraction, promoting efficient and sustainable use while preserving biodiversity

Flowchart of the PEITC MHG extraction method

EXTRACTION OPTIMISATION

Watercress

Freezing
-18 °C

Without defrosting

Microwave hydrodiffusion and gravity extraction
1.5 - 2.5 W/g | 10 - 30 min

Solid part (plant biomass) → ZERO WASTE → A potential functional flour

Heating and separation by gravity

Two liquid fractions

1 → 92 - 94 °C → 2

Changing the flask to collect the second fraction

Purple liquid fraction (PLF)

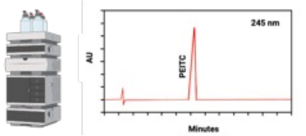
White liquid fraction (WLF)

Phenylethyl Isothiocyanate (PEITC)-rich extract

Total fraction (PLF + WLF)

Hexane | 37 °C | 2h

Identification and quantification of PEITC by HPLC-DAD



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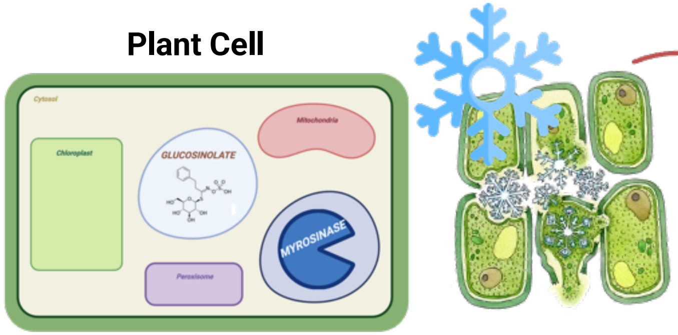
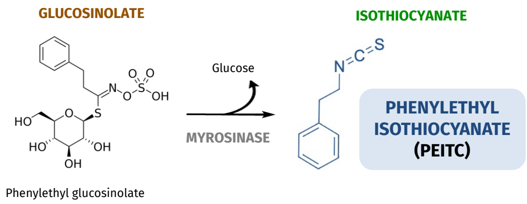
THE KEY NOVELTY

Freezing: a pretreatment to successfully extract PEITC

Freezing fruits and vegetables causes the water inside their plant cells to freeze, which in turn leads to the rupture of cell walls and damage to the cells, resulting in the contents of the cells leaking out

Plant Cell





The effect of freezing on the watercress leaf microstructure was studied using SEM

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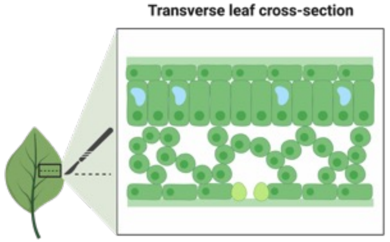
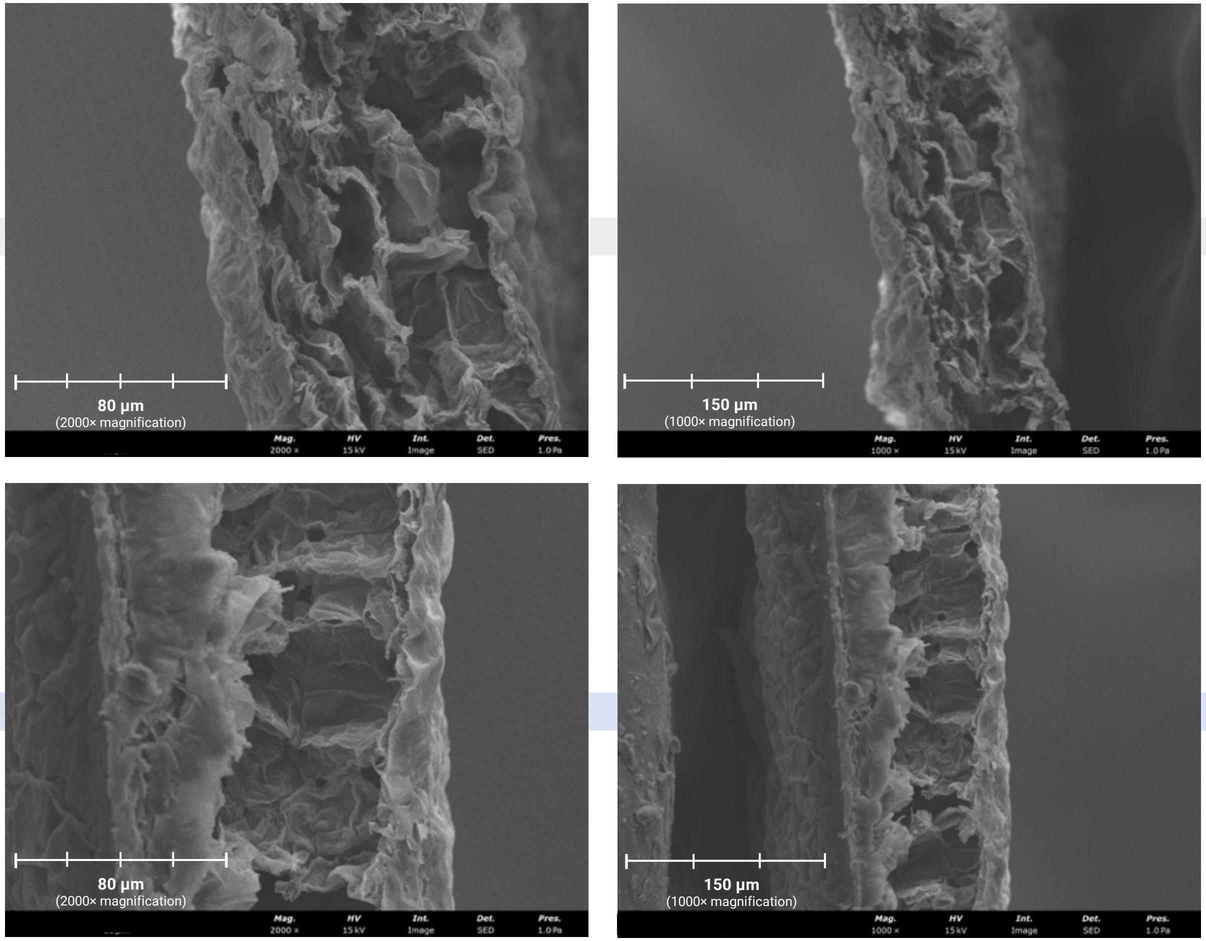
Freezing: The KEY NOVELTY

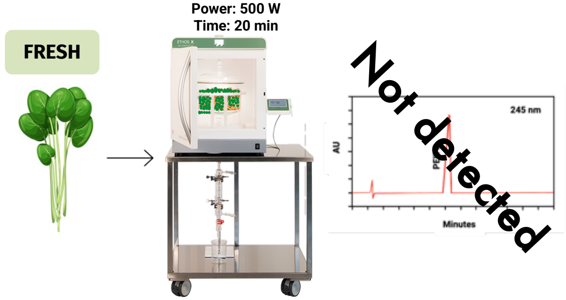


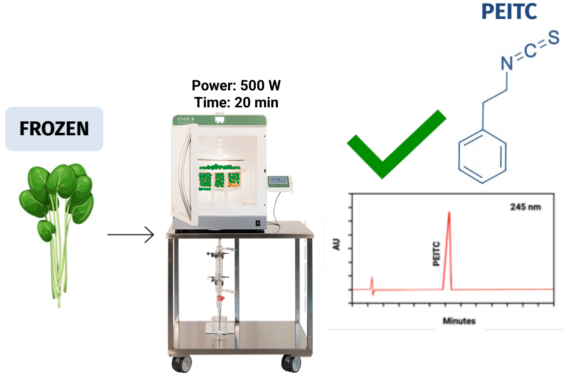
Scanning Electron Microscopy (SEM)

FRESH

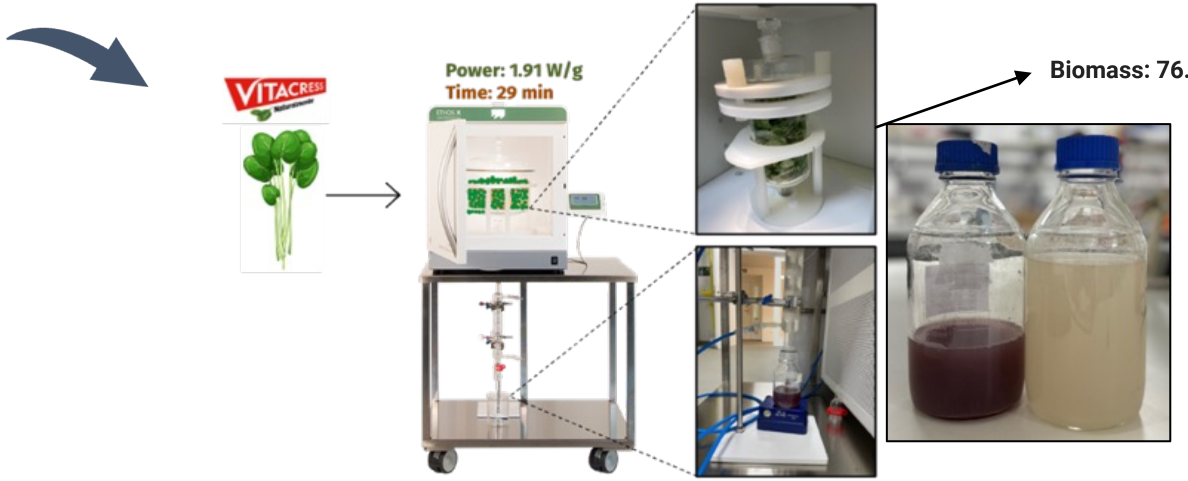
FROZEN







Microstructure of fresh and frozen watercress leaves observed through SEM



	Amount of extracted PEITC µg PEITC g^{-1} WC DB		
	MHG	C_6H_{14} Hexane	CH_2Cl_2 Dichloromethane
Purple Liquid Fraction	166 ± 37	1682 ± 1456 (1) 2300 (2)	3346 (3)
White Liquid Fraction	1652 ± 381		
Total	1818 ± 377		

(1) Coscueta, E. R., et al (2020). Phenylethyl Isothiocyanate Extracted from Watercress By-Products with Aqueous Micellar Systems: Development and Optimisation. *Antioxidants*, 9(8), 698.
(2) Rodrigues, L et al. (2016). Recovery of antioxidant and antiproliferative compounds from watercress using pressurized fluid extraction. *RSC Adv*, 6, 30905–30918.
(3) Farhana, N. et al. (2016). Effects of Temperature and pH on Myrosinase Activity and Gluconasturtiin Hydrolysis Products in Watercress. Trans. *Sci. Technol*, 3, 449–454.

- ✓ **MHG method extracts PEITC as efficiently as hexane and dichloromethane**
- ✓ **Optimising the extraction process of PEITC unlocks its immense potential as a health-promoting agent.**

TASK 1

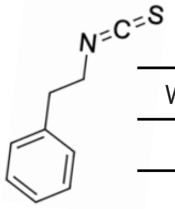
MHG is an **innovative** and **scalable green technique** that allows the **extraction of PEITC from watercress** as an alternative to conventional extraction methods.

GREEN METHOD FOR OBTAINING ISOTHIOCYANATES FROM CRUCIFEROUS VEGETABLES

PATENT APPLICATION SUBMITTED

Brassicaceae

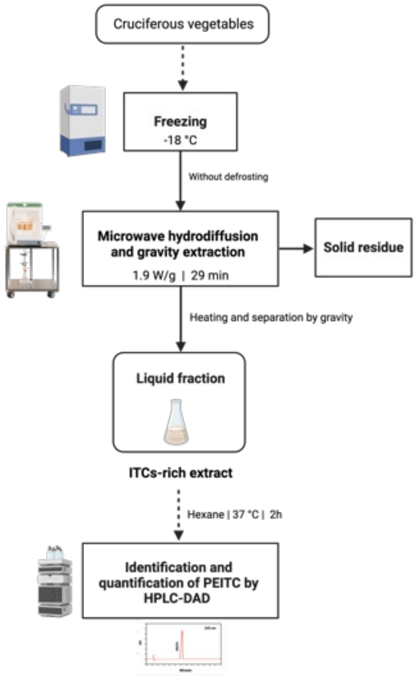
CRUCIFEROUS VEGETABLES



	PEITC content
Watercress	2285–4783 µg/g dry basis
Turnip	80 µg/g fresh weight
Broccoli	65 µg/g dry basis

Karanikolopoulou, S, et al. (2021). Current Methods for the Extraction and Analysis of Isothiocyanates and Indoles in Cruciferous Vegetables. *Analytica*, 2, 93–120.
Ezzat, M, et al.. (2024). Nutritional Sources and Anticancer Potential of Phenethyl Isothiocyanate: Molecular Mechanisms and Therapeutic Insights. *Molecular Nutrition & Food Research*, 68(8), 2400063.

The MHG extraction method was used to extract PEITC from other cruciferous



MHG extract



	PEITC ug g^{-1} dry basis	TPC mg GAE g^{-1} dry basis
Turnip	394 ± 104	1.23 ± 0.10
Turnip greens	Not detected	0.80 ± 0.08
Broccoli	Not detected	0.53 ± 0.06
Watercress	2452 ± 492	3.50 ± 0.60

NEW BATCH

The MHG method should be optimized for each matrix and ITC of interest.

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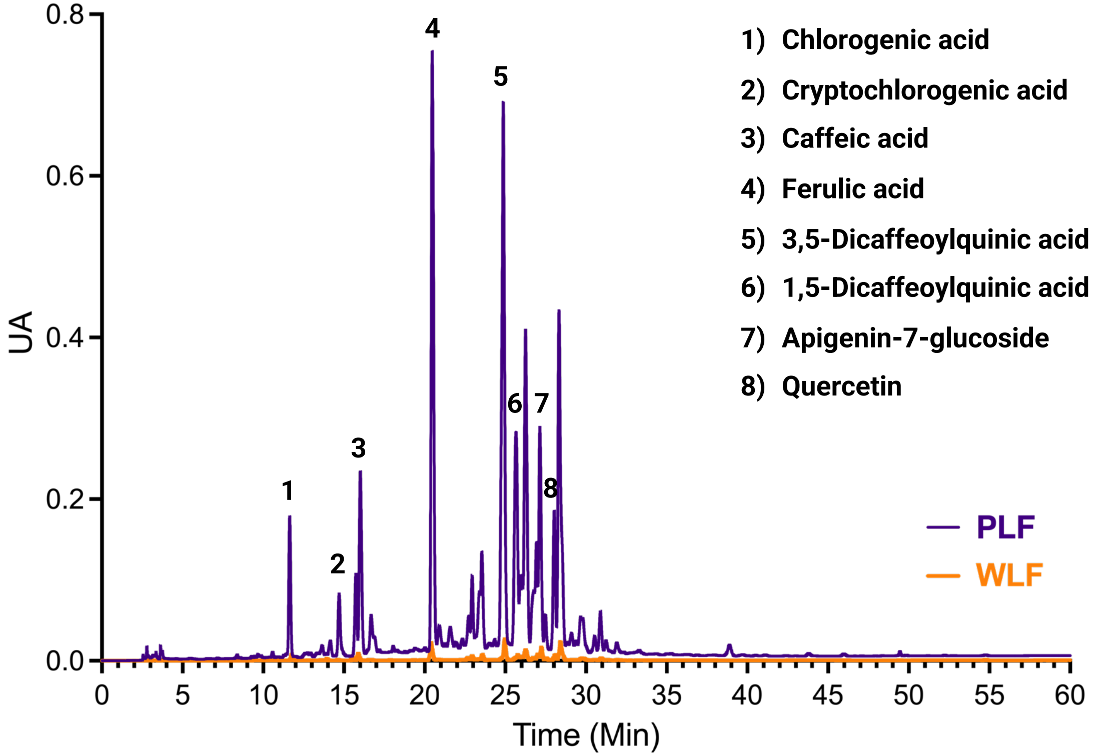
Biological activities

Phenolic compounds profile by HPLC-DAD at 320 nm from a representative replica of optimal extracts

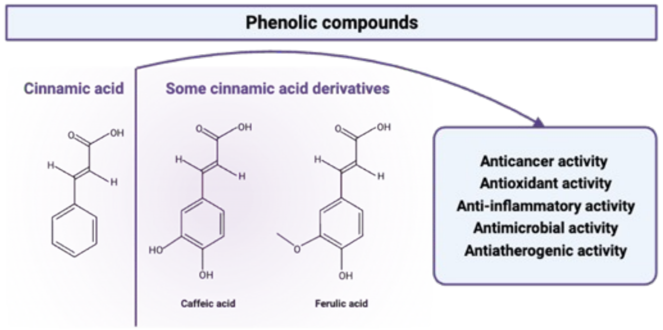


✓ Prominent peaks were identified.

Total phenolic content and antioxidant activity

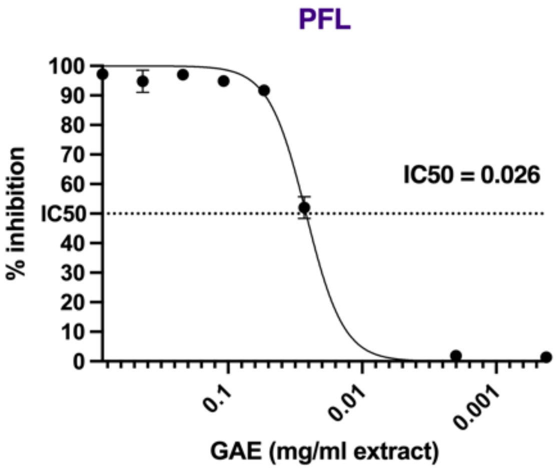
	TPC	ABTS	ORAC
	mg GAE g^{-1} WC DB	µmol TE g^{-1} WC DB	
→ Purple Liquid Fraction	2.04 ± 0.14	11.12 ± 1.38	33.70 ± 4.65
White Liquid Fraction	1.59 ± 0.34	2.99 ± 0.47	7.19 ± 1.19
Total	3.63 ± 0.50	14.26 ± 1.97	40.89 ± 6.51

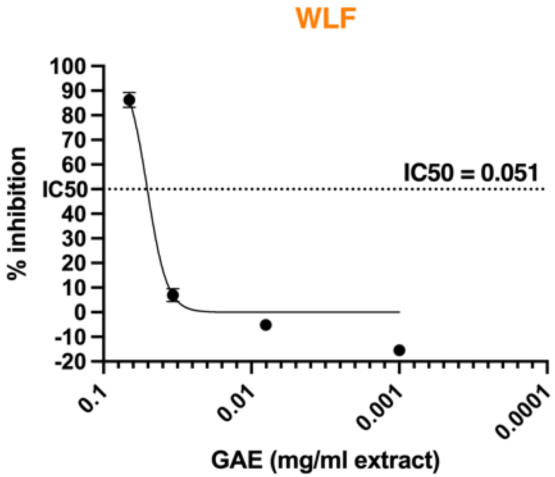
	PLF	WLF	Watercress
Total flavonoid content (µg catechin g^{-1} WC DB)	1477.79 ± 117.49	228.06 ± 45.89	
BCA Protein Assay (g protein $100g^{-1}$ WC DB)	5.14 ± 0.27	0.71 ± 0.13	
Vitamin C (mg ascorbic acid $100g^{-1}$ WC FW)	5.35 ± 0.40	0.22 ± 0.06	43.50 ± 5.48



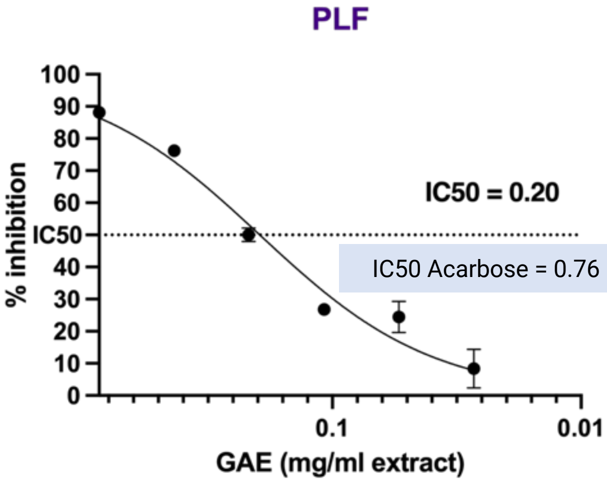
These molecules can synergistically promote the health benefits of PEITC.

Antihypertensive Activity





α-Glucosidase Inhibitory Activity



WFL does not reach the half-maximal inhibitory concentration.

WFL (at 0.07 GAE mg/ml extract)

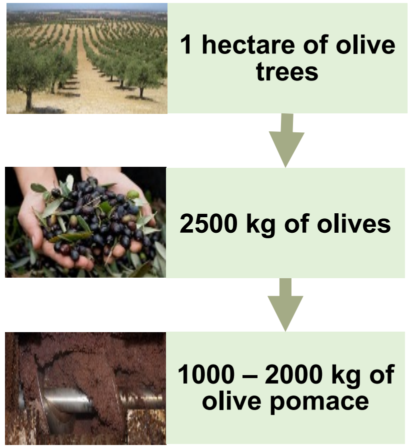
α-Glucosidade Inhibition (%): 33.57 ± 4.57

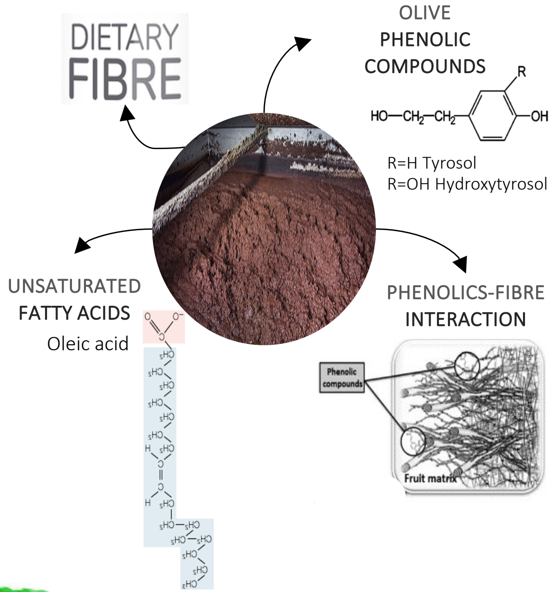
Acarbose inhibition: 87% (at 5 mg/ml)

- **Cruciferous vegetables have indole-3-carbinol,** a phytochemical with **cardioprotective**, **antioxidant**, and **anti-inflammatory** effects.
- Phytochemicals such as phenolic compounds (e.g. flavonoids), alkaloids, and terpenoids can help prevent health issues by controlling postprandial hyperglycemia.

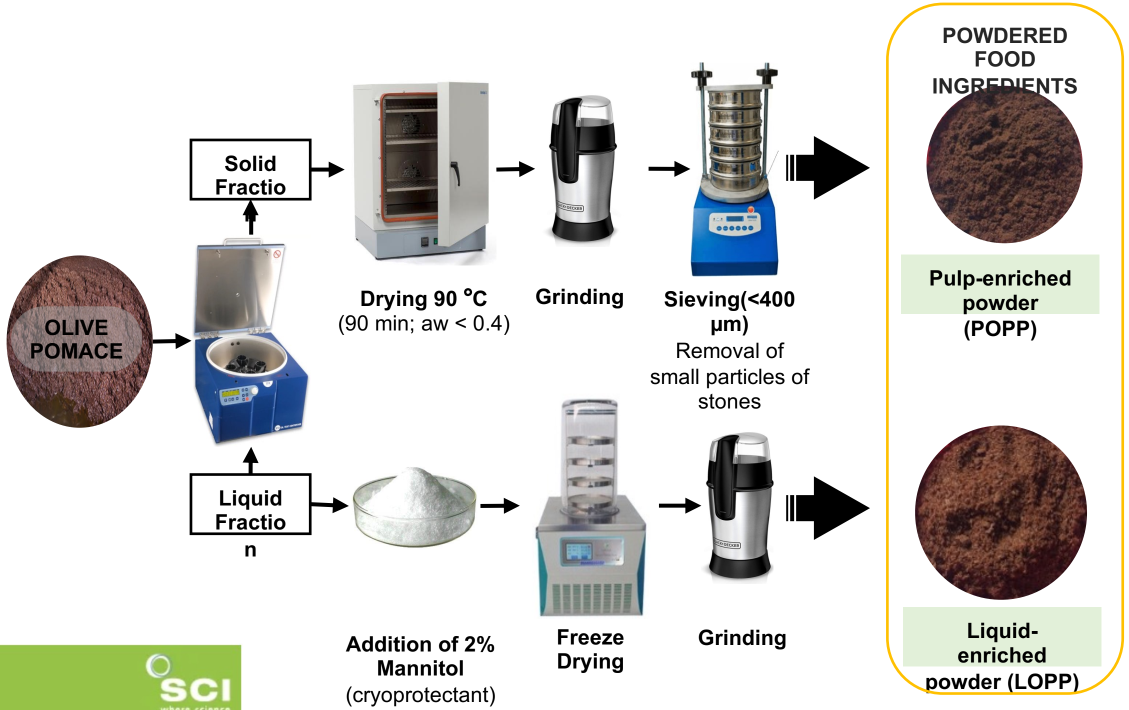
Olive Pomace

Olive Pomace





Olive Pomace Ingredients



Journal of the Science of Food and Agriculture
SCI where science meets business

Research Article

Are olive pomace powders a safe source of bioactives and nutrients?

Tânia Bragança Ribeiro, Ana Oliveira, Marta Coelho, Mariana Veiga, Eduardo M Costa, Sara Silva, João Nunes, António A Vicente, Manuela Pintado

First published: 10 September 2020 | https://doi.org/10.1002/jsfa.10812 | Citations: 3

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PREBIOTIC EFFECTS OF ANTIOXIDANT OLIVE POMACE FIBRE IN THE GUT

ELSEVIER

Food Hydrocolloids

Volume 112, March 2021, 106312

Prebiotic effects of olive pomace powders in the gut: In vitro evaluation of the inhibition of adhesion of pathogens, prebiotic and antioxidant effects

Tânia Bragança Ribeiro [a, b], Célia Maria Costa [a], Teresa Bonifácio - Lopes [a], S Monforte [a], João Nunes [b], António A. Vicente [c], Manuela Pintado [a]

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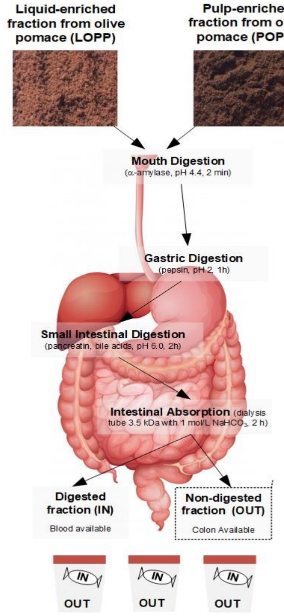
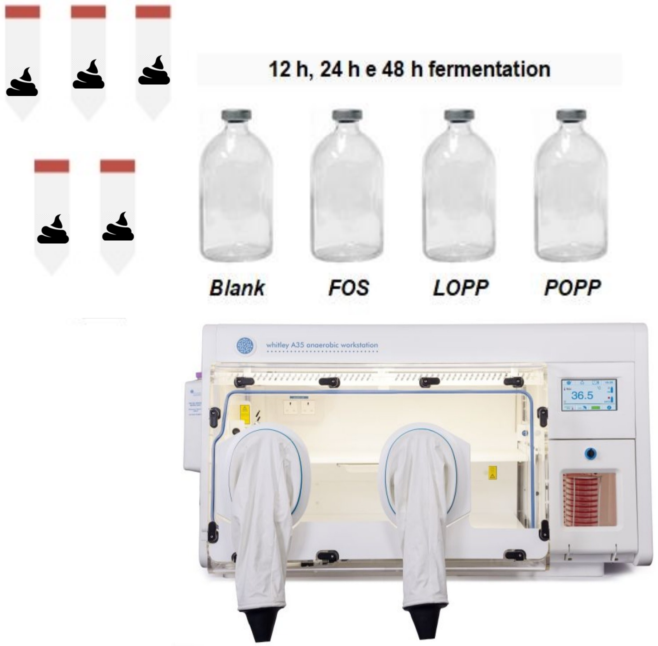
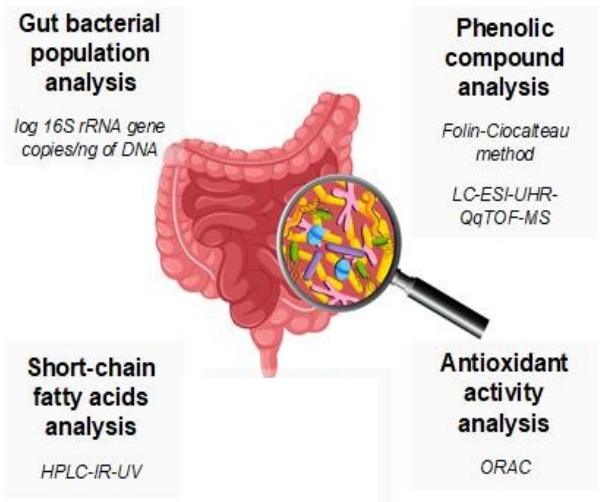
https://doi.org/10.1016/j.foodhyd.2020.106312



Simulated *in vitro* gastrointestinal digestion



***In vitro* large intestinal fermentation model**



Potential prebiotic effects analysis

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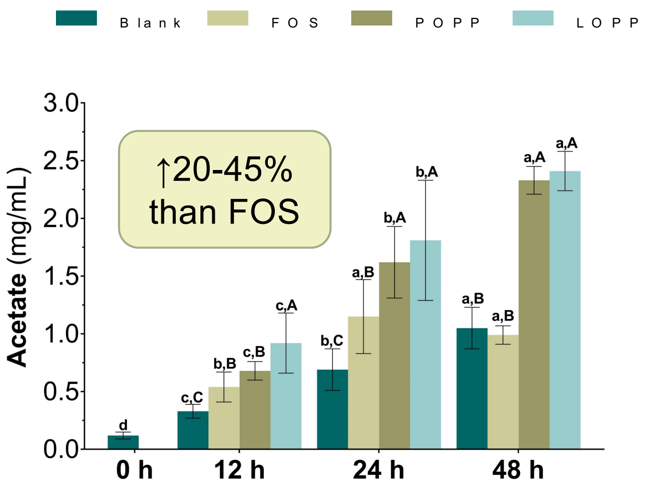
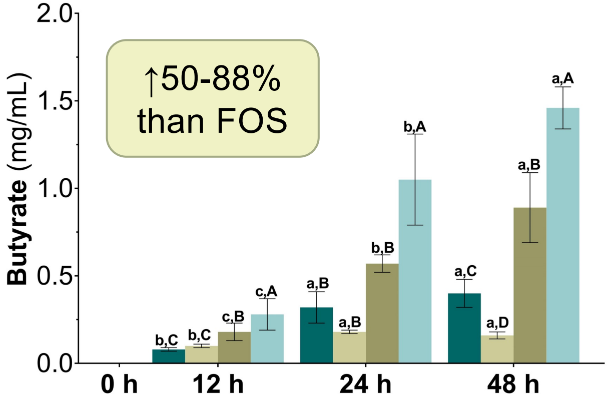
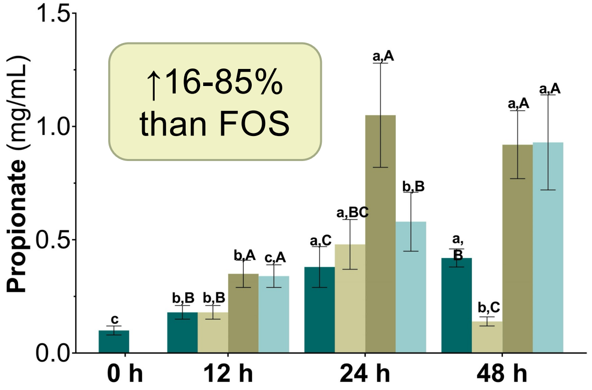
PREBIOTIC EFFECTS OF OLIVE POMACE FIBRE AND PHENOLICS IN THE GUT

Short-chain fatty acid analysis



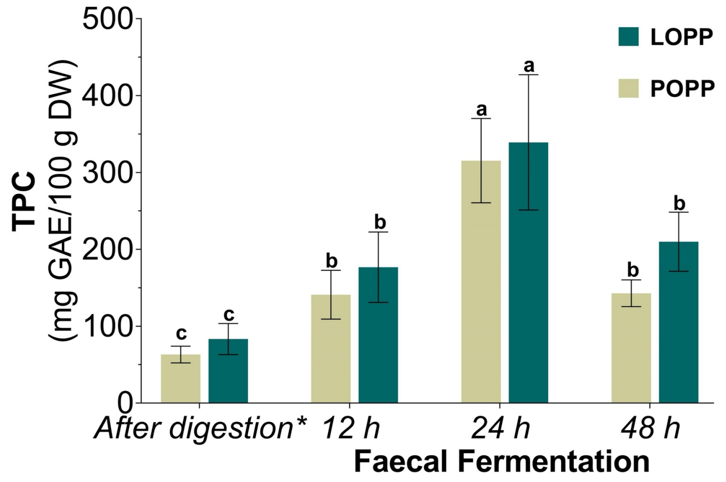




↑ Short-chain fatty acid production by **POPP and LOPP** than **FOS**

Potential prebiotic effect

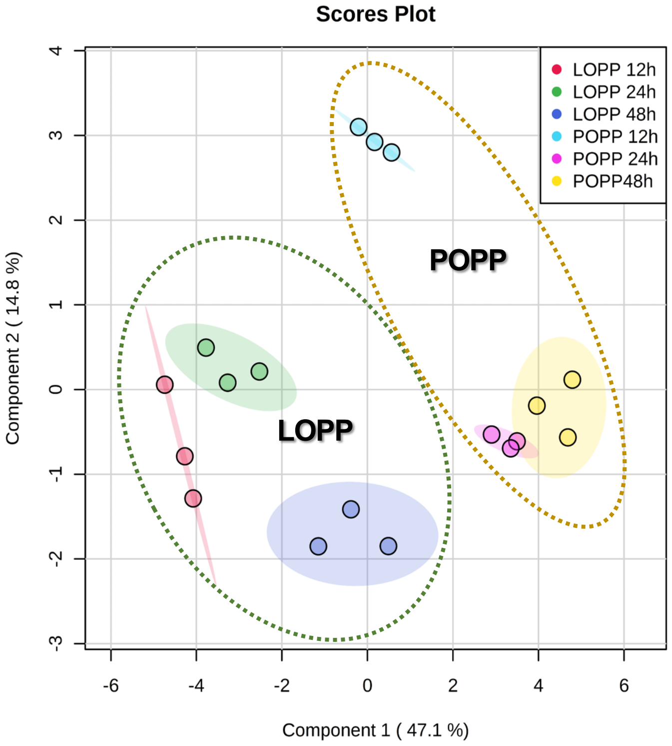
Total phenolic compounds

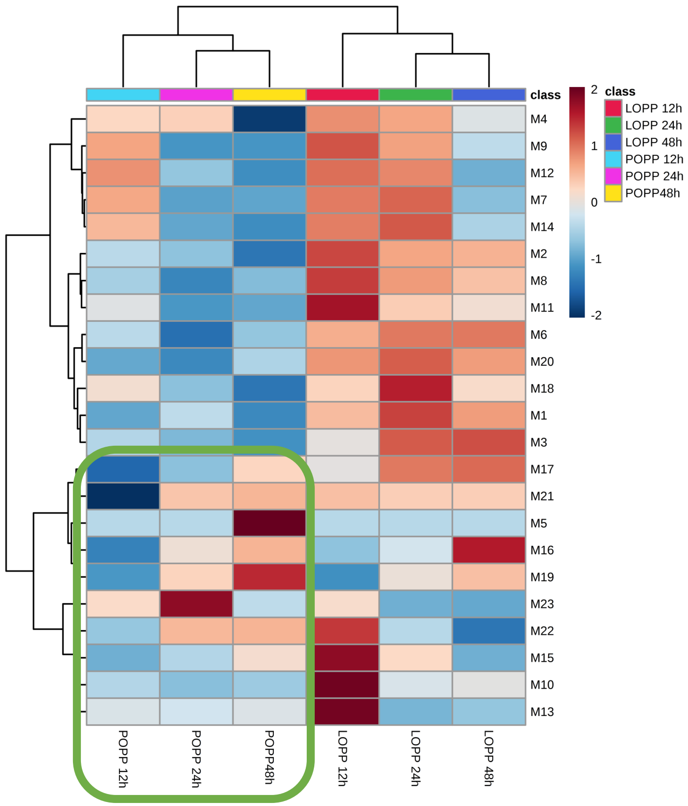


↑ Total phenolic compounds after faecal fermentation

PREBIOTIC EFFECTS OF OLIVE POMACE FIBRE AND PHENOLICS IN THE GUT

Identification of Phenolic Compounds (LC-ESI-UHR-QqTOF-MS)





M5	Vanillyl alcohol
M19	3-Hydroxyphenilpropionic acid
M21	Homovanillic alcohol
M22	Homovanillic acid
M23	Luteolin

LC-ESI-UHR-QqTOF-MS detected:

- ❑Phenolic compounds
- ❑Phenolic acids

ACORNS

Small fruit produced by the *Quercus* spp. trees

Rich in starch and polyphenols

Great potencial application

Production: 401 585 tones/year

20% Food source for wildlife

24% Fattening in livestock

1% Food processing

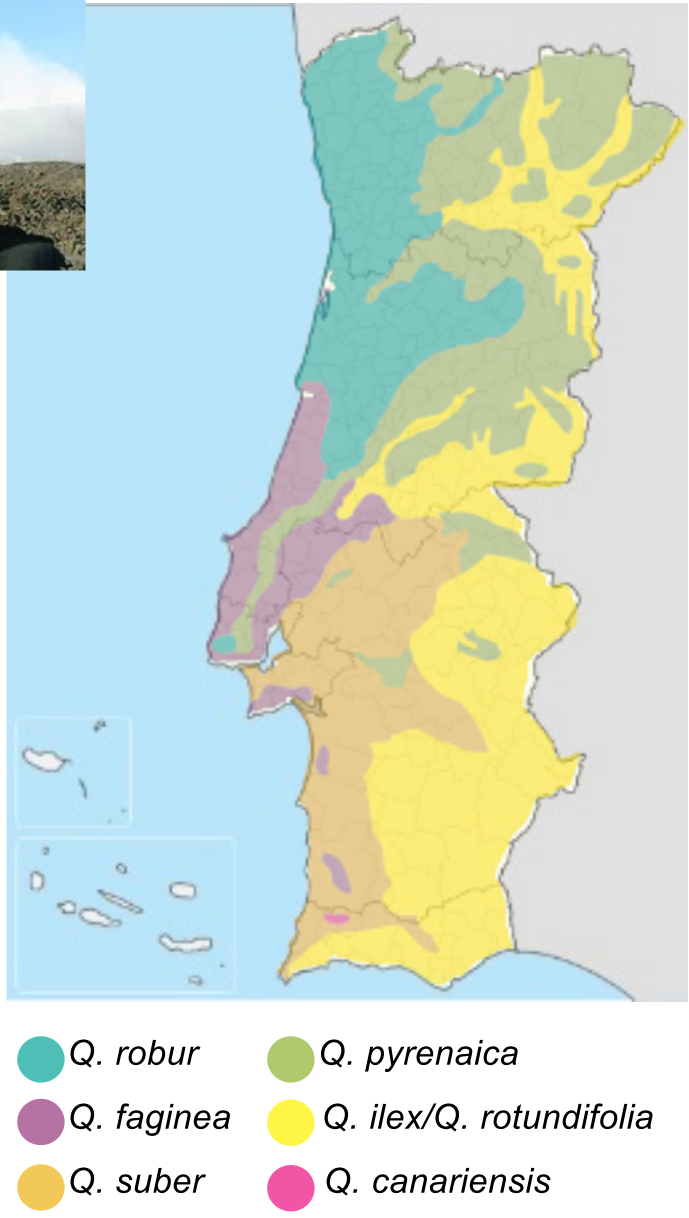
55% Undervalued

79% North

48% Alentejo

STARCH AND POLYPHENOLS VALORIZATION FROM ACORNS

Oakfood



Manuela Pintado, Jorge A. Saraiva, and Elisabete M. C. Alexandre

NUTRITIONAL CHARACTERIZATION

Starch

Dehulling	*Quercus* spp	Total (%, DF)	Resistant (%, DF)	Total digestible (%, DF)	Slowly digestible (%, DF)	Rapidly digestible (%, DF)
Manual	*Q. pyrenaica*	41.9±0.4^{de}	31.0±1.1^{g}	10.9±0.7^{c}	8.9±0.1^{c}	1.9±0.5^{e}
	Q. robur	24.6±1.4^{b}	21.9±1.5^{ef}	2.7±0.1^{a}	0.5±0.0^{a}	2.1±0.1^{b}
	Q. ilex	29.7±0.2^{c}	24.5±0.4^{f}	5.2±0.3^{b}	1.3±0.0^{a}	1.3±0.0^{a}
Thermal (1 min; 100 ºC)	*Q. pyrenaica*	46.4±0.0^{e}	7.5±0.0^{a} ↓	38.9±0.1^{e} ↑	19.0±0.1^{e} ↑	11.6±0.1^{ab} ↑
	Q. robur	22.1±0.3^{ab}	12.8±0.4^{bc} ↓	9.3±0.1^{c} ↑	3.4±0.3^{b} ↑	3.1±0.3^{c} ↑
	Q. ilex	37.0±0.4^{d} ↑	9.4±0.4^{ab} ↓	27.6±0.0^{d} ↑	14.1±1.4^{d} ↑	6.1±0.2^{d} ↑
Drying (30 ºC)	*Q. pyrenaica*	30.1±2.9^{c} ↓	20.2±1.9^{def} ↓	9.9±1.1^{c}	3.9±0.5^{b} ↓	3.4±0.0^{c} ↑
	Q. robur	18.5±0.4^{a} ↓	16.0±0.5^{cd} ↓	2.5±0.1^{a}	0.2±0.1^{a}	2.1±0.0^{b}
	Q. ilex	21.7±1.5^{ab} ↓	18.3±1.7^{de} ↓	3.4±0.2^{a} ↓	0.6±0.0^{a}	1.2±0.0^{a}

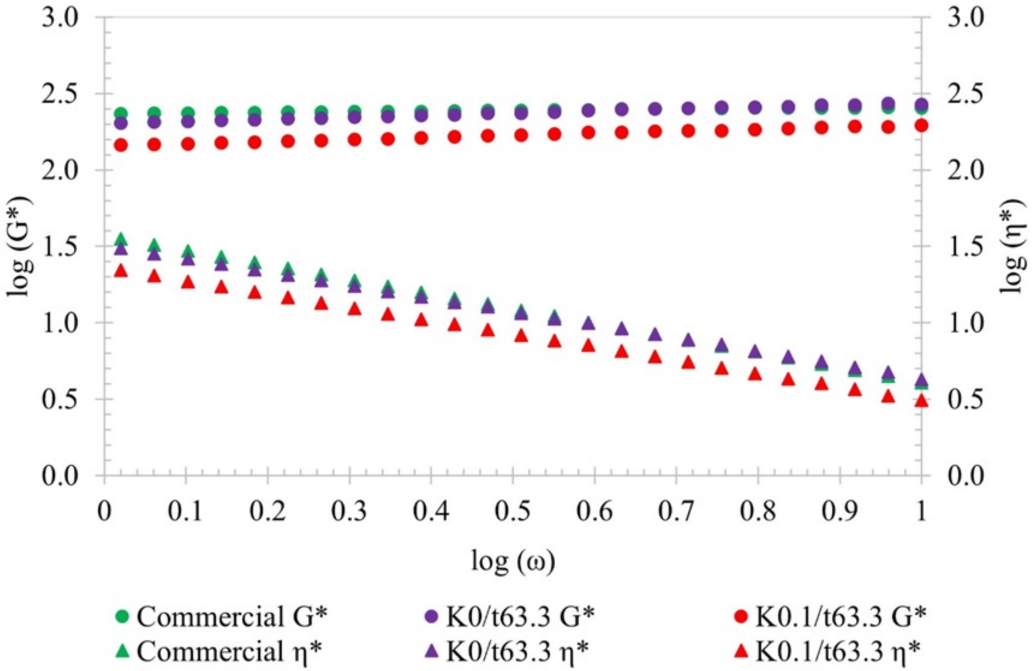
Highest resistant/total starch ratio
Lowest total digestible starch

→ **Manually dehulled *Q. robur***

DF: Dried flour

..BY PEF - PULSED ELECTRIC FIELDS TECHNOLOGY

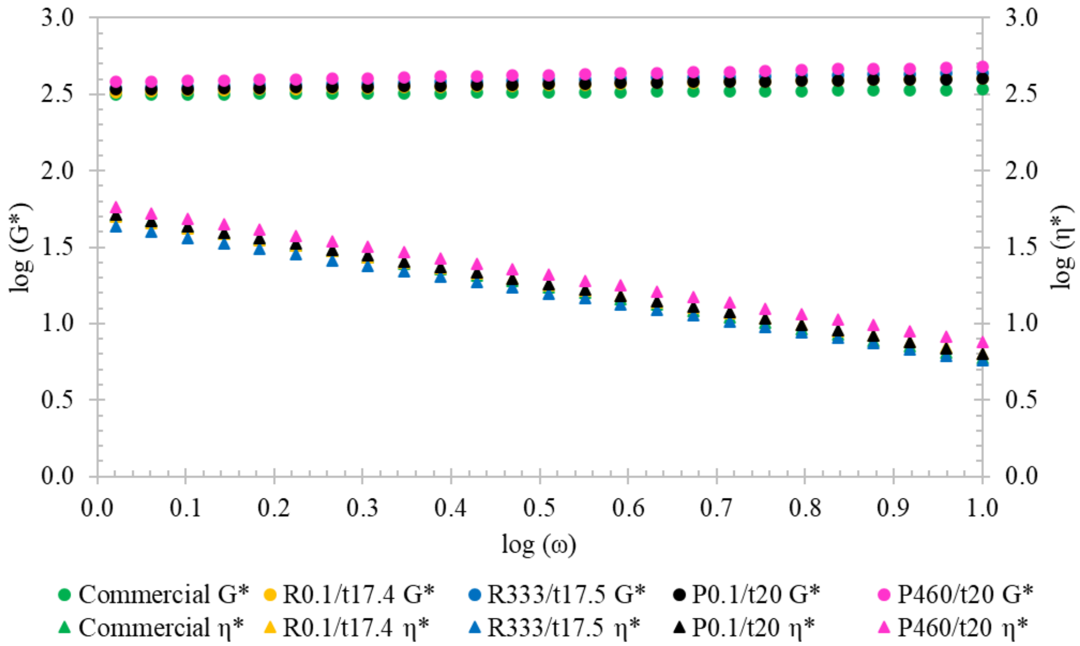
In-vitro digestibility



RDS: Rapidly digestible starch; SDS: Slowly digestible starch; TDS: Total digestible starch; RS: Resistant starch; TS: Total starch

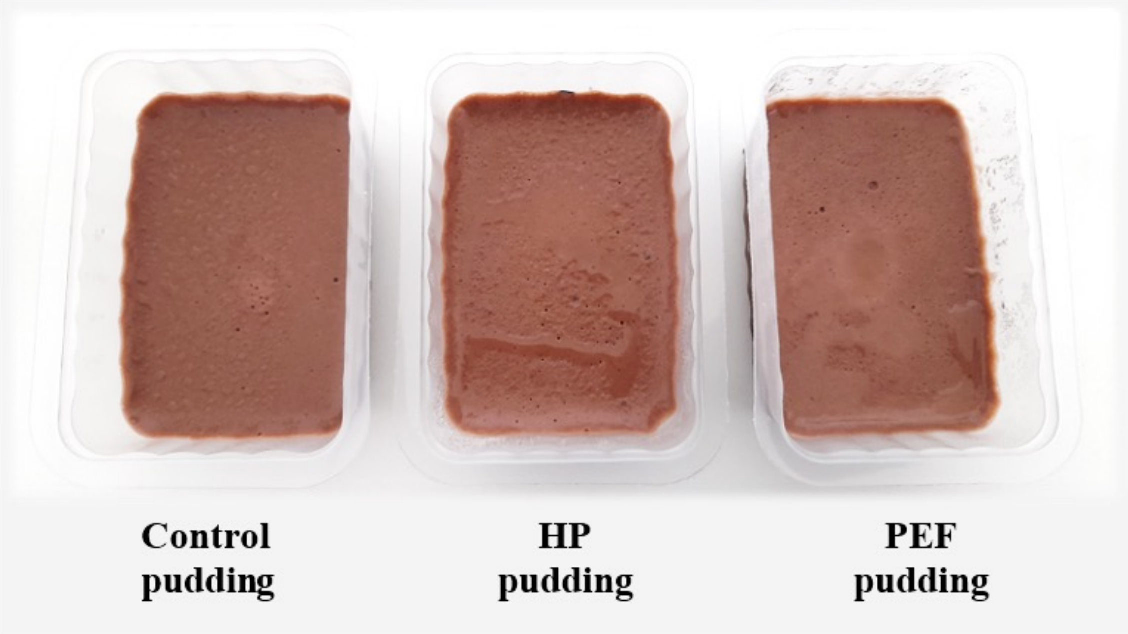
..BY HP - HIGH PRESSURE TECHNOLOGY

In-vitro digestibility

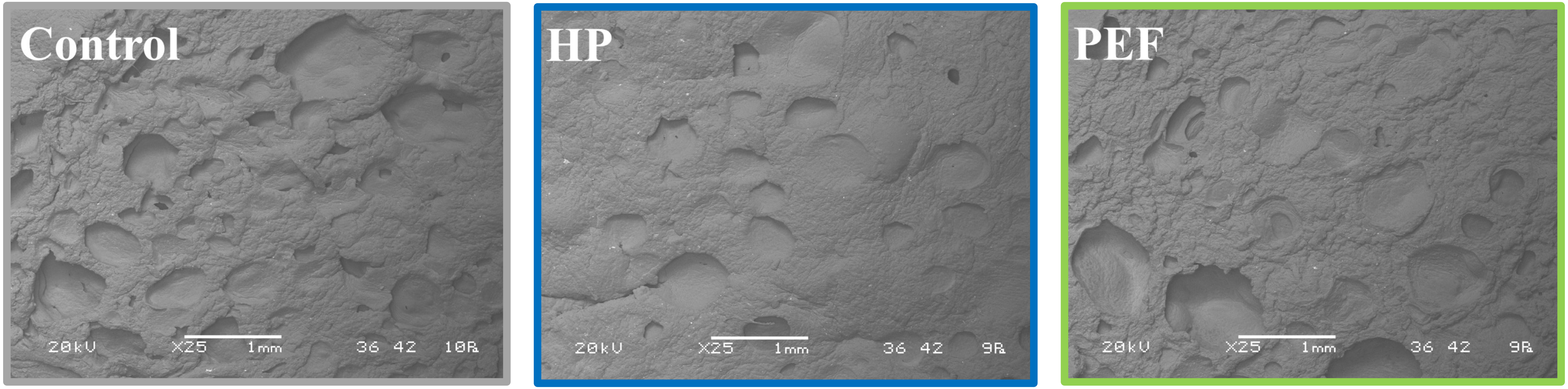


RDS: Rapidly digestible starch; SDS: Slowly digestible starch; TDS: Total digestible starch; RS: Resistant starch; TS: Total starch

FOOD APPLICATION

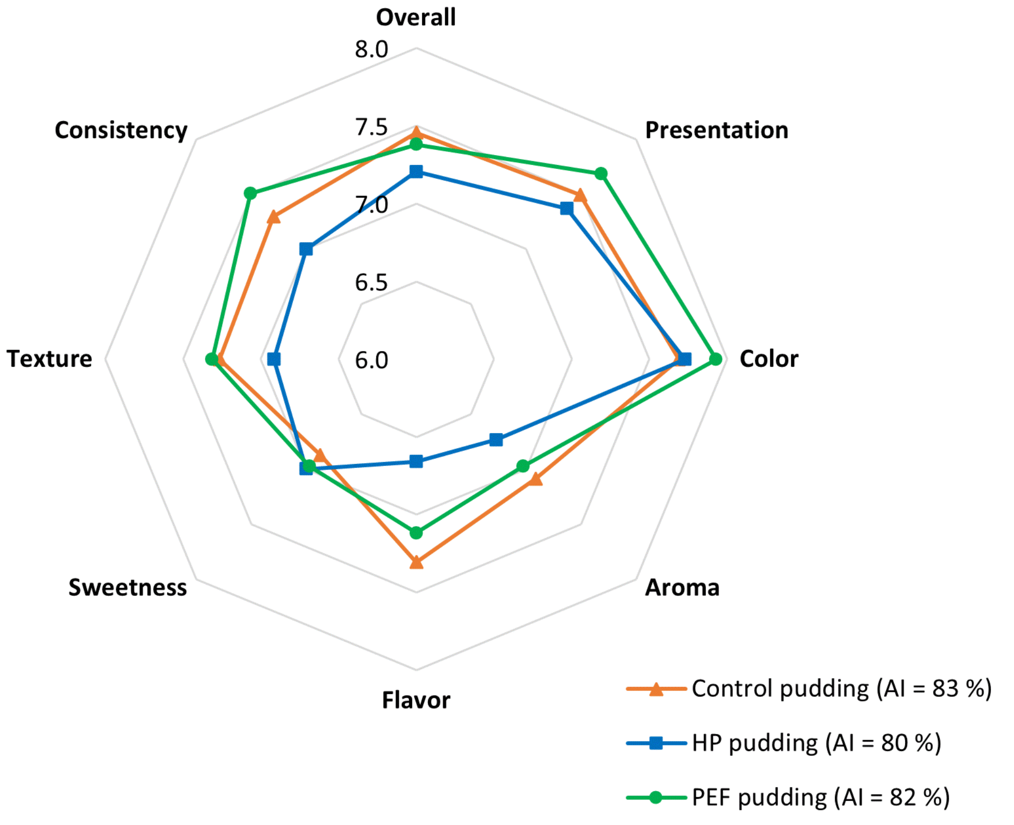


FOOD APPLICATION



FOOD APPLICATION

Sensorial analysis



Legume based systems for soil health and crop valorisation

Extraction and application of cellulose from legume losses in soil

- **Bean**
  - Leaf
  - Stalk
  - Pod
- **Soybean**
  - Pod

André e André de Cima, Marta Vasconcelos, Manuela Estevez Pintado

Composition

(DW)		Leaf bean	Stalk bean	Pod bean	Pod soybean
Moisture (g/ 100g)		14.73 ± 0.44	15.93 ± 0.13	16.37± 0.03	15.94 ± 0.07
Ash (g/ 100g)		1.22 ± 0.07	1.50 ± 0.05	2.08 ± 0.02	1.83 ± 0.12
Protein (g/ 100g)		26.44 ± 0.38	11.64 ± 0.74	18.70 ± 0.13	14.06 ± 0.53
Fiber (g/ 100g)	FND	24.76 ± 0.06	46.51 ± 1.05	45.65 ± 3.55	55.69 ± 0.71
	FAD	11.42 ± 0.40	24.62 ± 0.80	29.21 ± 1.58	34.06 ± 1.24
Sugars (mg/ g)	cellobiose	136.91 ± 9.01	n.d.	n.d.	154.06± 5.40
	glucose	453.48± 12.20	498.72± 20.32	230.45± 17.82	410.87± 31.32
	xylose	214.51± 8.53	287.74± 11.26	279.3± 23.45	307.61± 22.12
	galactose	333.18± 7.80	352.45± 16.96	526.54± 22.87	221.27± 18.70
	arabinose	216.89± 6.79	n.d.	n.d.	n.d.
	mannose	6.35± 1.11	9.79± 1.34	2.31± 0.87	5.82± 0.90
Lignin (g/ 100g)	AIL	18.1± 3.56	27.58± 1.79	33.02± 2.13	37.52± 4.78
	ASL	10.99± 2.42	7.28± 2.10	4.56± 0.54	5.2± 1.34

FND: Neutral fibre detergent; **FAD**: Acid fibre detergent; **AIL**: Acid insoluble lignin; **ASL**: Acid soluble lignin

Cellulose extration from pod bean

Bassani *et al*., (2020)

- **38.5%**

El Halal *et al*., (2015)

- **15.4%**

de Souza Coelho *et al*., (2020)

- **36.4%** **(Bleaching with 30% H_2O_2)**




Germination conditions

Quantity: 40 tomato seeds for each condition

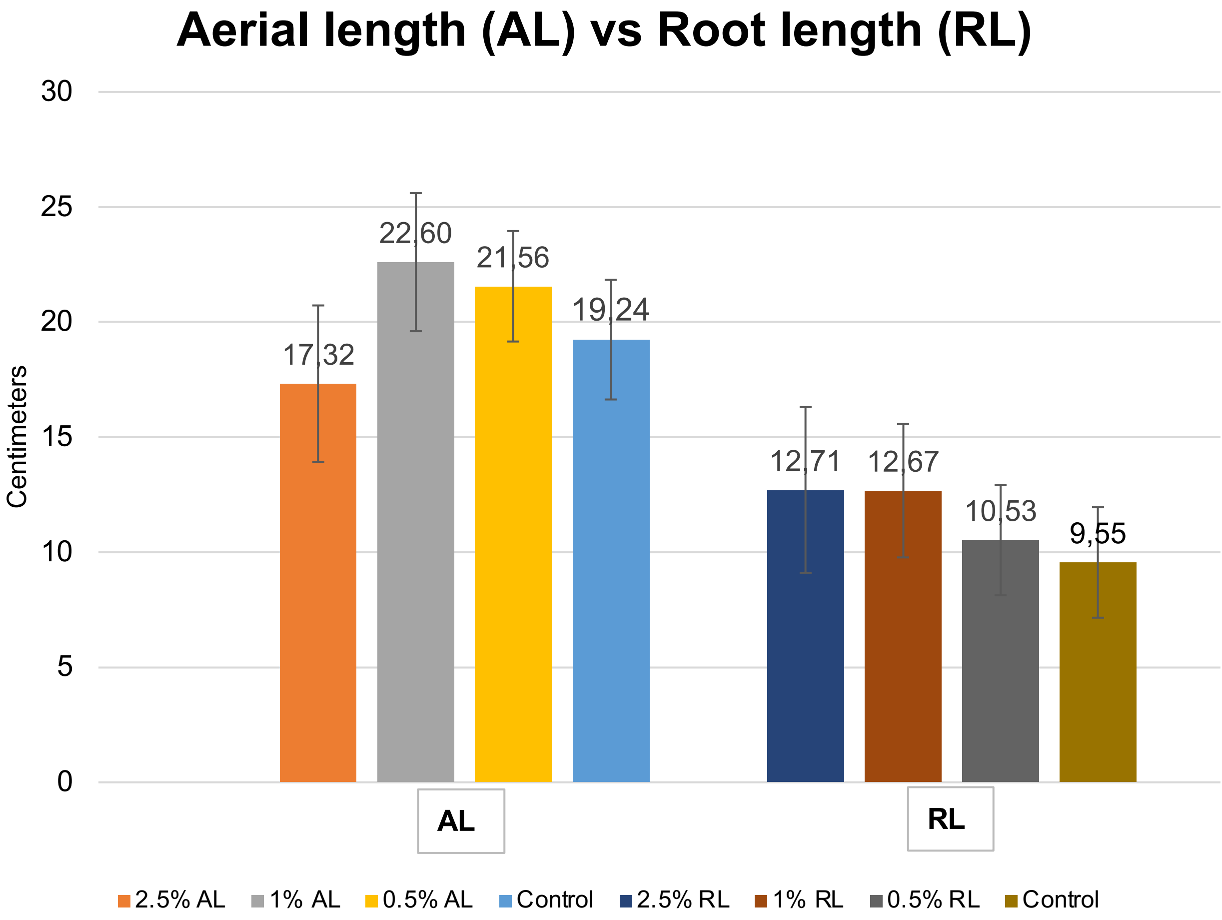
Duration: 4 weeks

Cellulose concentration in soil: control (0%); 0.5%; 1% and 2.5%

Temperature: 25ºC

Light: 24h per 7 days

✓ Throughout time it was noticed that the higher cellulose content (2.5%) had a **better water retention** in soil.



➢ In relation to the control on root length, all concentrations generated a positive effect, with 1% presenting the best results.

TAKE-HOME MESSAGES

- **Resource Optimization**: Using agrofood byproducts reduces waste and environmental impact by converting waste into valuable ingredients
- **Nutritional Benefits**: Agrofood byproducts are rich in nutrients, enhancing the nutritional value of food products.
- **Sustainability**: Incorporating agrofood byproducts supports sustainability by reducing waste and promoting a circular economy
- **Innovation Opportunities**: New food products and ingredients from agrofood byproducts meet consumer demand for sustainable and healthy options
- **Regulatory and Safety**: Bringing products derived from agrofood byproducts to market requires ensuring their safety and regulatory compliance

• Acknowledgments

- **All partners and researchers contributing for this work**

- All funding entities

CATÓLICA FACULTY OF BIOTECHNOLOGY PORTO

Oakfood

PRIMA PARTNERSHIP FOR RESEARCH AND INNOVATION IN THE MEDITERRANEAN AREA

MEDI SMART

COMPETE 2020

PORTUGAL 2020

UNIÃO EUROPEIA Fundo Europeu de Desenvolvimento Regional

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gBIOT

Thank you for your attention

UIDB/Multi/50016/2020