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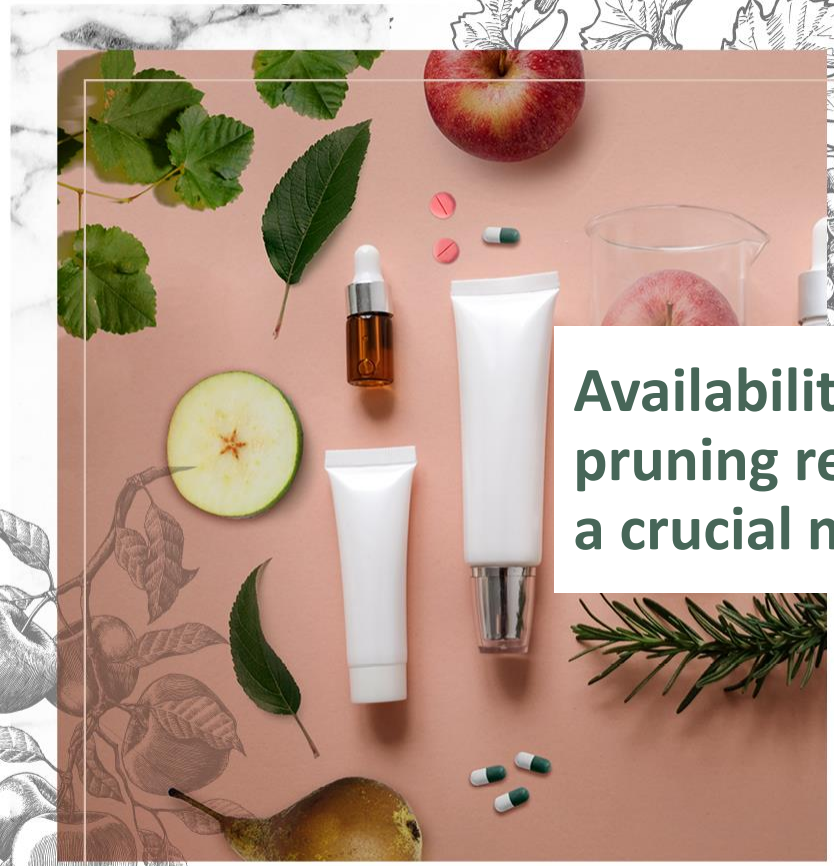

Availability and distribution of pruning residues: a crucial matter

Pierre-Louis Bombeck,
Valbiom

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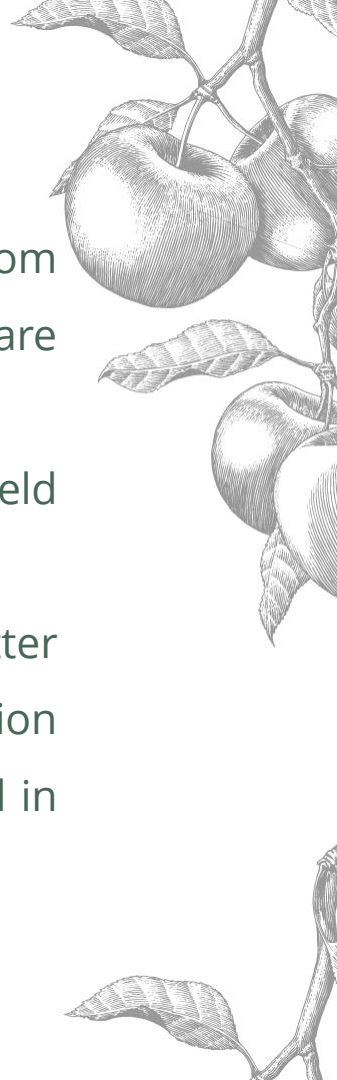
ABOUT valbiom

- Non-profit association based in Gembloux (BE)
- Valbiom supports you in implementing sustainable solutions in the bio-based economy

From biomass residues to fertilizers

- Pruning residues from orchards and vineyards are generally poorly valued
- Removed from the field and/or burned
- One of our goals : a better valorization by extraction and the return to the field in the form of fertilizers



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Raw material for extraction process

How much, when and where are the agriwastes side streams for the sourcing ?

Steps

Sourcing availability (data) :

- Areas of orchards and vineyards
- Selecting the most promising sources
  - ➔ Apple trees, pear tree and vines pruning residues
- Potential production / ha
- Total potential (economy of scale)

Samples collection :

- Selected orchards and vineyards
- Collection campaign
  - 2019, 2020, 2021
- Samples for
  - Extraction and others analysis
  - Data on yield of pruning residues

Sourcing : raw material availability

Action	Source	
Surface of orchards	Statistics	Per country/ for total region
Potential yield of cuttings	Research done	Per country
	Interview (online)	Min 100 replicants
	Interview (in person)	20 / country – key figures
		E.g. Holland : NFO
	Research	Actual measurements

Most promising: apple, pear & grapes

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Samples collection

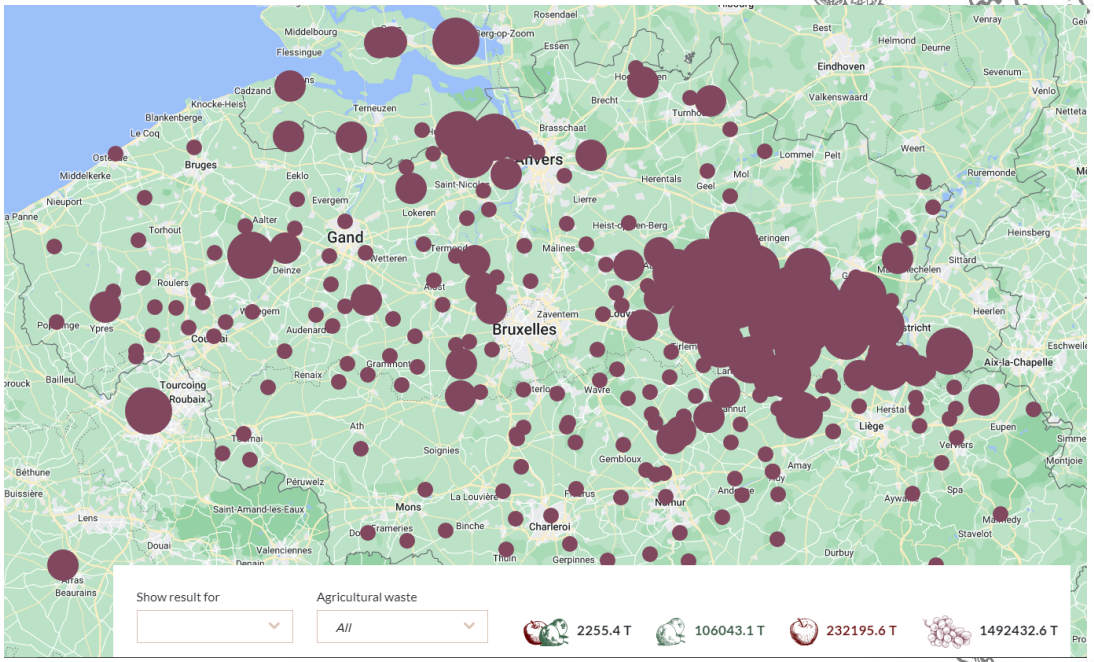
Steps

Mapping :

- ‘Hot spots’
- Geolocation
- Total potential by geographic region (localization opportunities)

Geolocation of biomass side streams

- By type and origin
  - Apple tree pruning
  - Pear tree pruning
  - Vine stalks / branch
- By quantity
- By region



➔ Connect potential users with a new source of raw material

Map available on the AgriWasteValue website www.agriwastevalue.eu

Impacting parameters

- **Temporary availability :**
  - A few months a year (November-March), which can fluctuate from year to year
- **Availability at harvest site:**
  - Within each orchard/vineyard, left between rows of trees, rarely grouped together
  - Mean amounts of pruning residues
    - Vine: 1,71 t DM/ha
    - Appel tree: 3,04 t DM/ha
    - Pear tree: 2,74 t DM/ha
- **Storage conditions :**
  - Left on the ground after pruning, subject to bad weather
- **Transport :**
  - Only 50-60 % of dry matter

Conclusion

- Pruning residues available in quantity and various location, with “hot spots” due to regional cultural specificities
- Other important parameters than just "where?" and "how much?“
- Logistic is the key

Project under the program
Interreg North-West Europe
European Regional Development Fund

With the financial support of the European Regional Development Fund and Wallonia
EUROPEAN UNION
European Regional Development Fund
Wallonie

Thank you

Pierre-Louis BOMBECK
pl.bombeck@valbiom.be

AgroParisTech Innovation
celabor
Cosmetic Valley France
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eurasanté Invest for Success
PFI Biotechnology
Université de Reims Champagne-Ardenne
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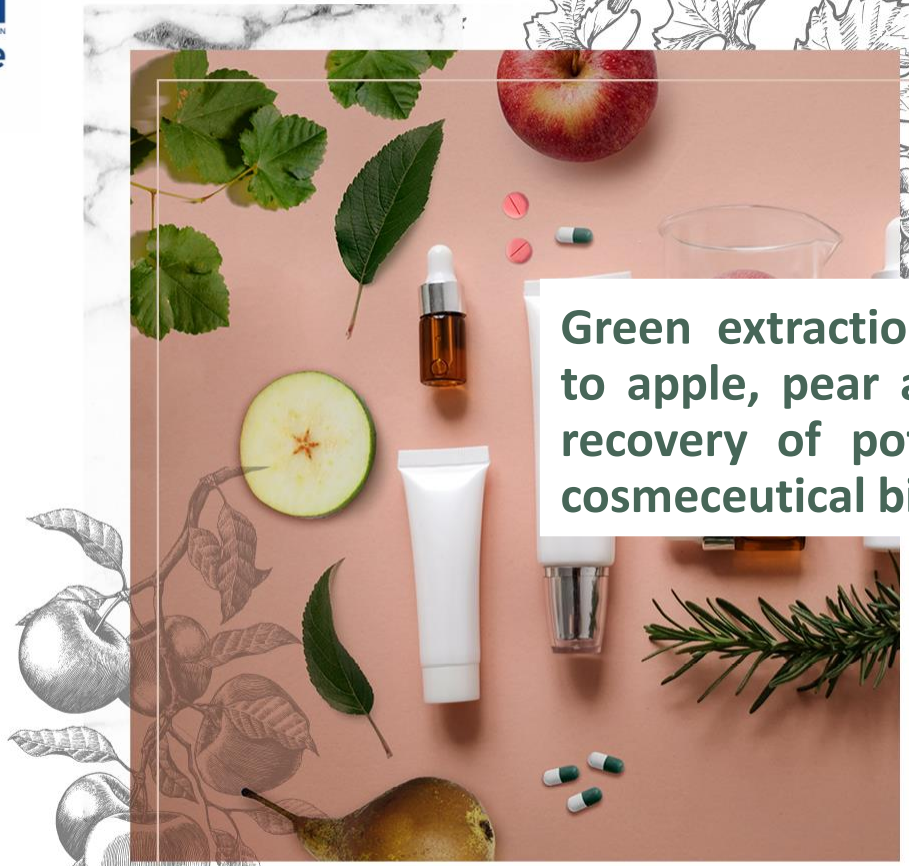

Green extraction methodologies applied to apple, pear and vine residues for the recovery of potential nutraceutical and cosmeceutical bioactive ingredients

Job Tchoumtchoua

R&D Project Leader

CELABOR scrl

Is a Belgian scientific and technical center accredited by the Walloon region. CELABOR is offering scientific and technical support to companies involved in all sectors of the **circular-economy** and **bioeconomy** including agri-food, green processes, packaging, textile and environment.

Four departments in the heart of the bioeconomy sectors

Food technologies

Environment

Biomaterials Packaging & Textile

Biomass valorisation Platform

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Biomass valorization Platform - Extraction department

The “Extraction” department is equipped with a **technological platform** (350 m^2) unique in Wallonia boasting an **ATEX zone**, a test hall constituted of laboratory and pilot extraction machinery.




Three pilot plants **Supercritical Fluid Extractors SFE-CO_2** - 2x 6L/batch + 1 lab-scale equipment

Pilot-scale **Subcritical Water Extraction** -6L/batch

Conventional **solvent extraction** – 60-350L

ATEX

Ultrasounds & Microwave Assisted Extraction - 25-50L UAE, 2-5L MAE

Pilot-scale **Pulsed Electric Field** - liquid 350 L/h & solid 0,5L/batch

Lab and pilot-scale **membrane separation** – Ultra-, Nano-filtration

Pilot-scale post-treatment equipment - Freeze-dryer, Spray-dryer, Evaporator, Centrifuge, 135L-High Pressure processing for debacterisation

Purification platform - Centrifugal partition Chromatography CPC, MPLC, Prep-HPLC

Advanced **analytical lab** - UPLC-MS, GC-MS, ICP-MS, HPLC-DAD-ELSD

More than 20 years of experience

Research Programs & Collaborative Projects

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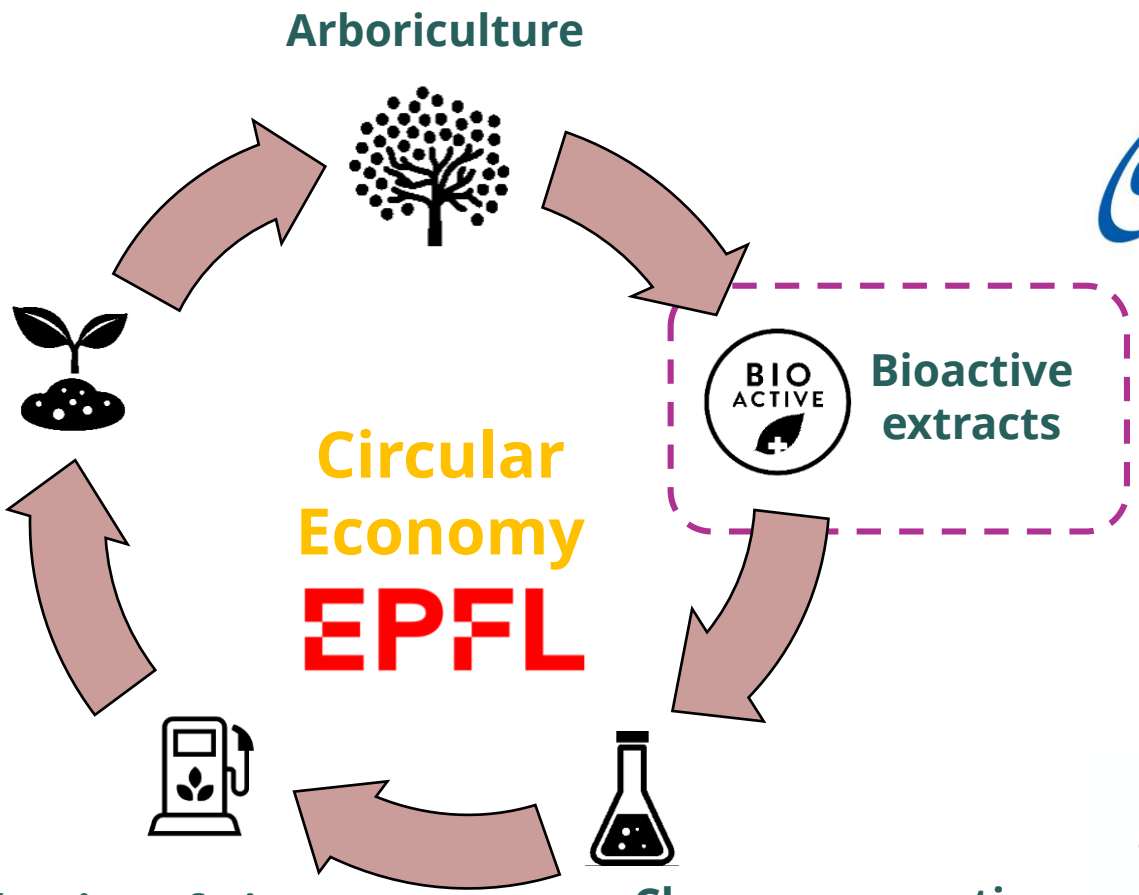
Delphy

celabor

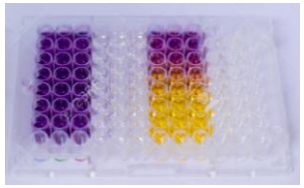


PFI
Germany

Université de Reims Champagne-Ardenne

AgroParisTech
Innovation

Global approach



Collection & pretreatment of agri-residues

Green extraction process

Post-treatment, concentration, drying

Evaluation of biological properties

Quantification of bioactives metabolites

Collection

Vine prunings
11 varieties

- *Régent*
- *Cabernet noir*
- *Dornfelder*
- *Chardonnay*
- *Souvignier gris*
- *Solaris*
- *Johanniter*
- *Muscaris*
- *Pinot noir*
- *Pinot gris*
- *Pinot meunier*

Apple prunings
10 varieties

- *Jonagold*
- *Jonagored*
- *Braeburn*
- *Golden*
- *Gala*
- *Elster*
- *Novajo*
- *Bisquet*
- *Rouge Duret*
- *Saint Remy*
- *Capitaine*

Pear pruning
5 varieties

- *Adams*
- *Conférence*
- *Doyenné*
- *Plant de Blanc*
- *Doyenné du comice*

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Safety analysis

Regulation (EC) 1881/2006, 19.12.2006 modified by **Regulation (UE) 2018/290**, 26.02.2018

- ✓ Heavy metals and metalloids
- ✓ Mycotoxins: Aflatoxins
- ✓ Polycyclic aromatic hydrocarbons (PAHs)
- ✓ Pepticides residues

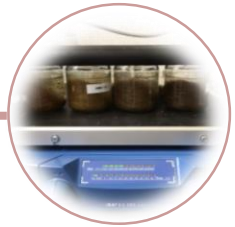
- 4 regulated regulated aflatoxins were determined by **HPLC fluorescence:** Aflatoxines **B1**, **B2**, **G1**, **G2**

- 4 regulated regulated PAHs were determined by **GC-MS:** Benzo[a]anthracene-**BaA**, Benzo[a]pyrene-**BaP**, Benzo[b]fluoranthene-**BbF**, Chrysene-**CHR**

- 6 regulated heavy metals and metalloids were determined by **ICP-MS**: Arsenic-**As**, Cadmium-**Cd**, Chromium-**Cr**, Nickel-**Ni**, Lead-**Pb**, Mercury-**Hg**

Extraction

Agri-residues



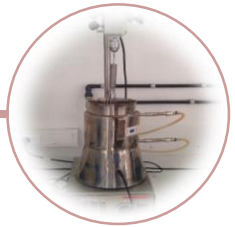
Maceration

- Temperature : 0 - 50°C
- Duration : 15 min - 4H
- Cycles : 1 ou 2
- Solvents : EtOH70%, H_2O
- Ratio : 1/5 - 1/10

Accelerated solvent extraction (Extraction by pressurized solvents)

- Pressure : 100 bars
- Temperature : 120°C
- Duration : 10 min x 2 cycles
- Solvents covering large polarity : EtOAc, EtOH70%, H_2O

Initial Screening

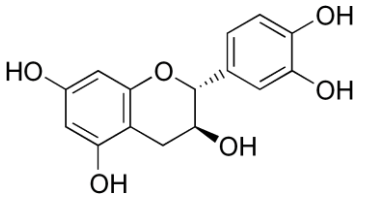


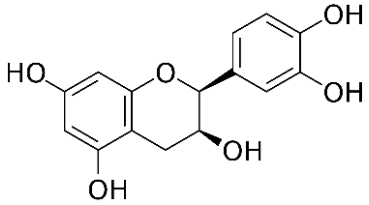
Ultrasounds Assisted Extraction

- Temperature : 0 - 50°C
- Duration : 30 min - 1H
- Cycles : 1 ou 2
- Solvents : EtOH70%, H_2O
- Ratio : 1/5 - 1/10

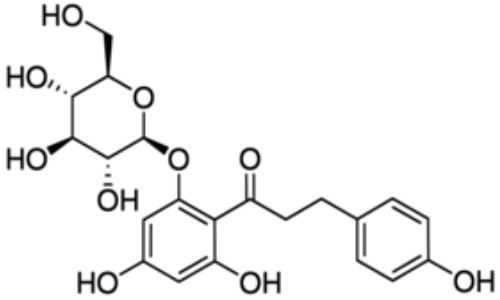
Determination of bioactive metabolites

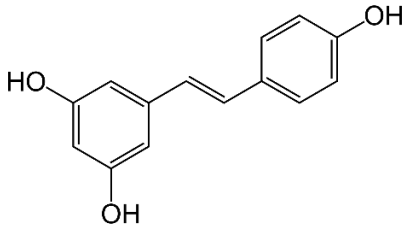
- 25 standard polyphenols (DDB Celabor)
- Quantification by LC-MS/MS
  - *Rapid*
  - *Sensible*
  - *Robust*





ACQUITY UPLC® **BEH Shield RP18** columns





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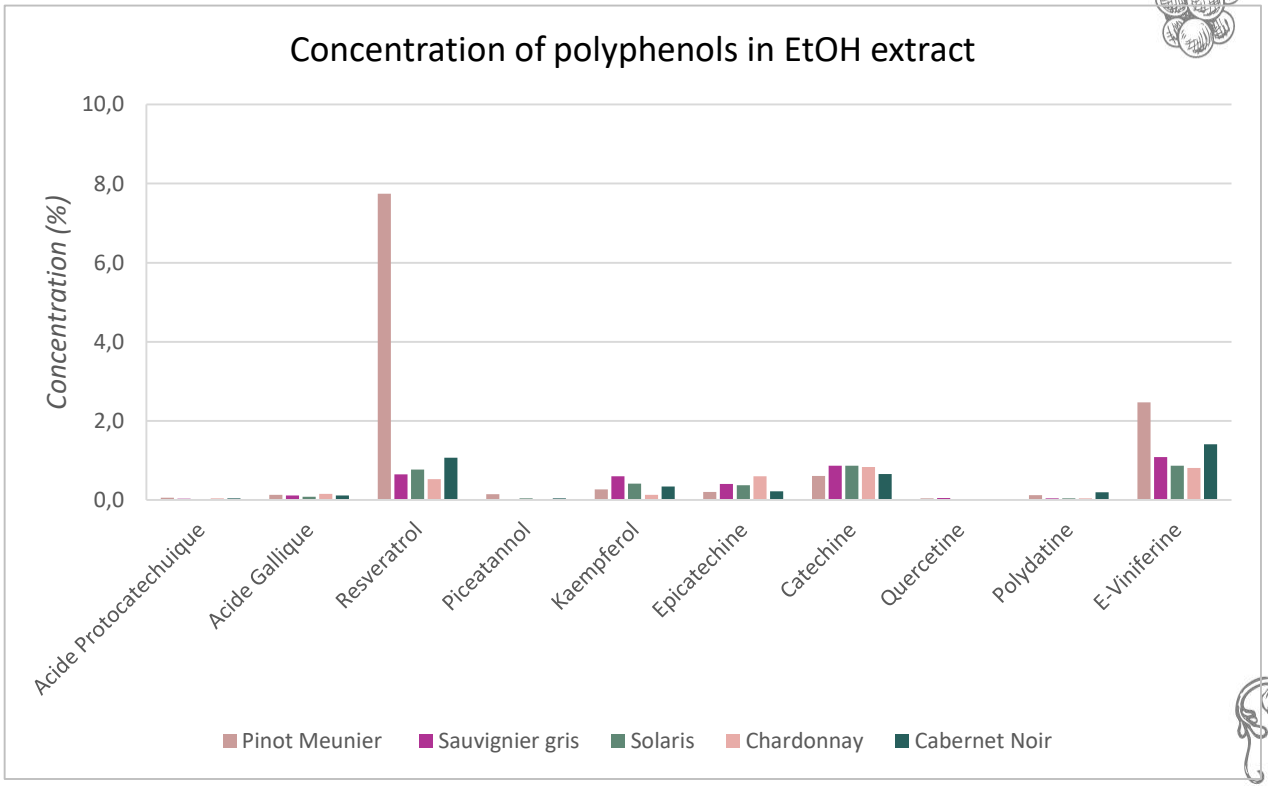
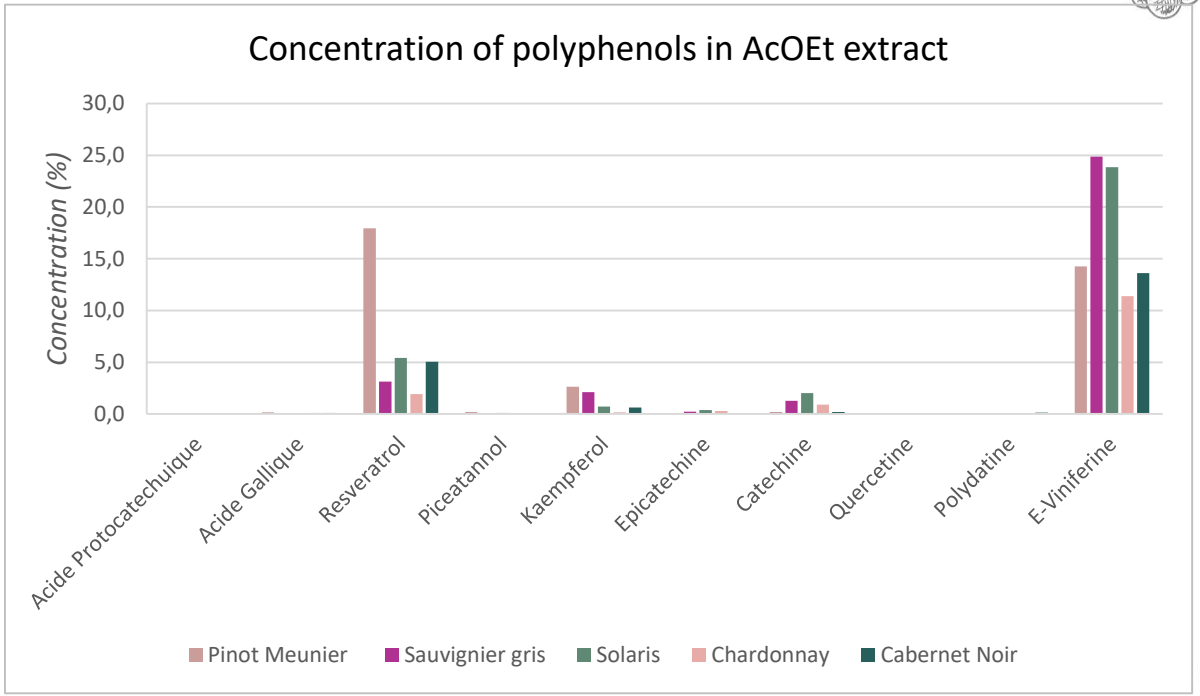
Bioactive metabolites of vine residues



Bioactive metabolites of vine residues



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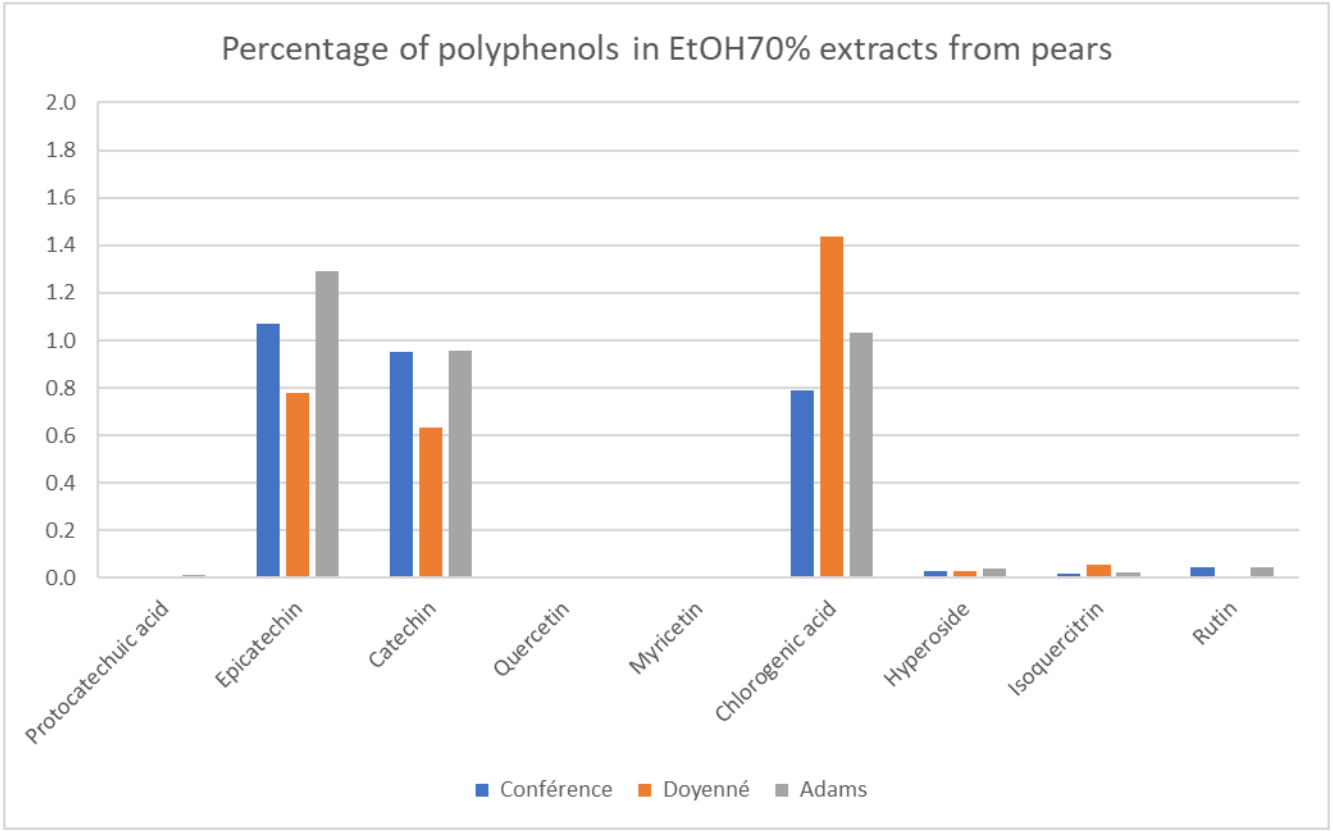
Bioactive metabolites of pear residues



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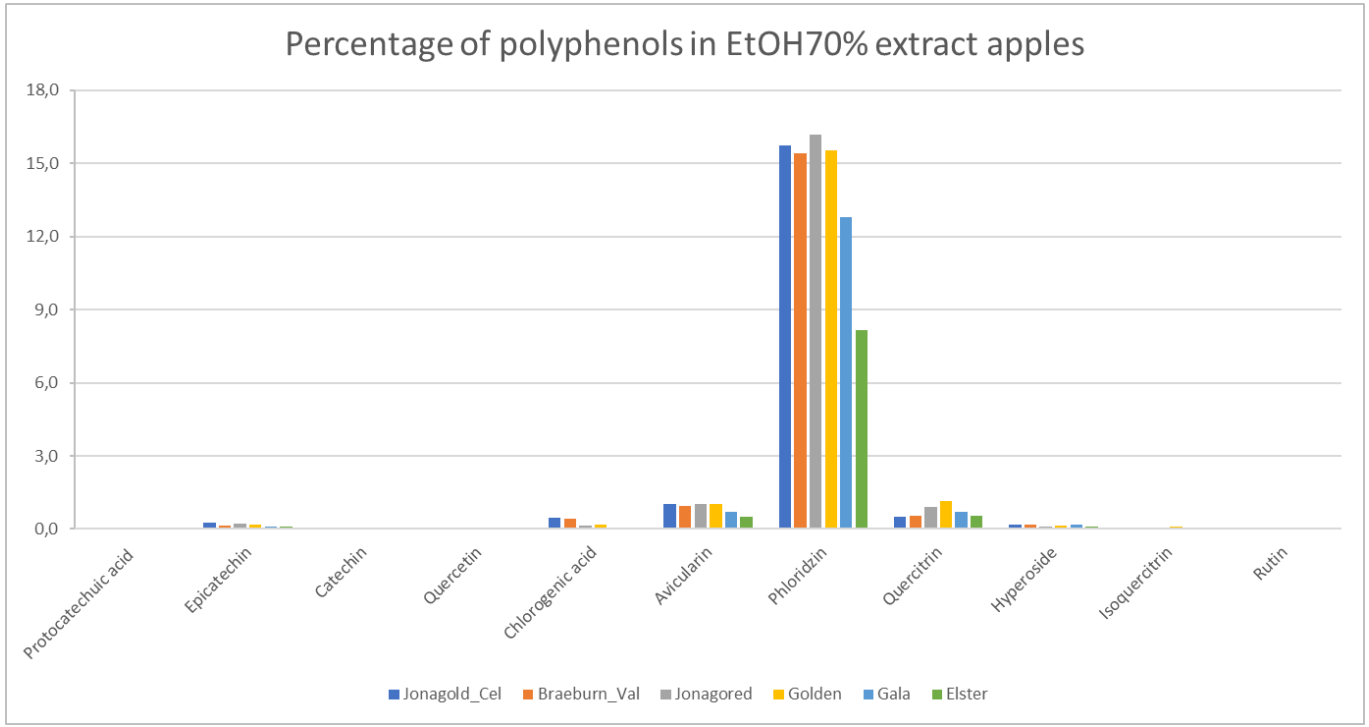
Scale up extracts from pear residues

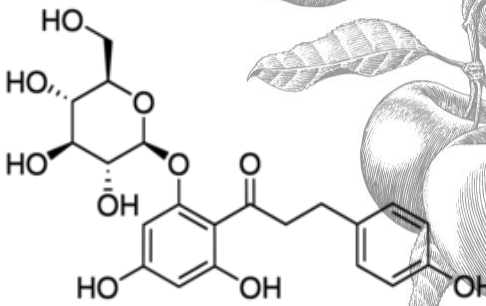
	Sub-critical Water Extraction	Pressurised EtOH70% Extraction
Mass yield	**16,3%**	**8,1%**
Total phenolic content (Folin)	**155,45** mg EGA/g	**187,08** mg EGA/g
Arbutin concentration (LC-MS/MS)	**6,4%**	**8,2%**
Recovery of arbutin by the process	**99,3%**	**57,5%**
Antioxydant activity (FRAP)	**0,97** mmol eq Fe(II)/g	**1,11** mmol eq Fe(II)/g
Antioxydant activity (DPPH)	**20** mg EGA/g	**65** mg EGA/g
Anti-tyrosinase activity @500ppm	**65,1%**	**74,5%**

- **Sub-critical Water extraction yields to a better extraction of arbutin**
- **Pressurized EtOH70% extraction technique yields to a more active extract on both anti-oxidant and anti-tyrosinase activities**

Bioactive metabolites of apple residues



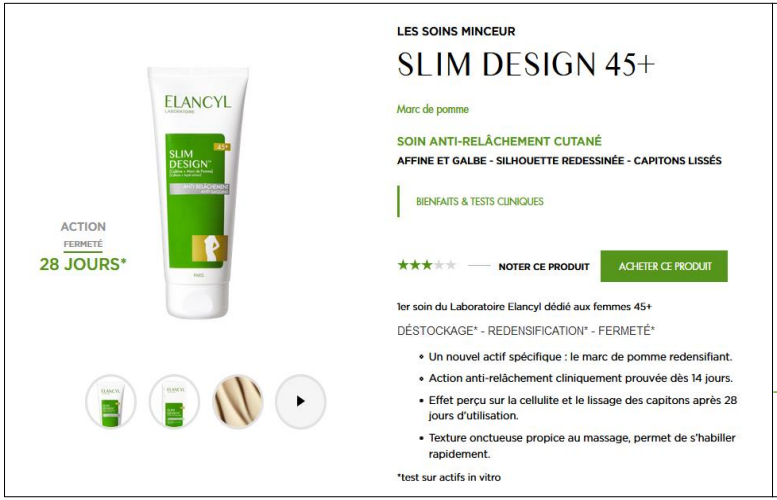
- **Phloridzine:**

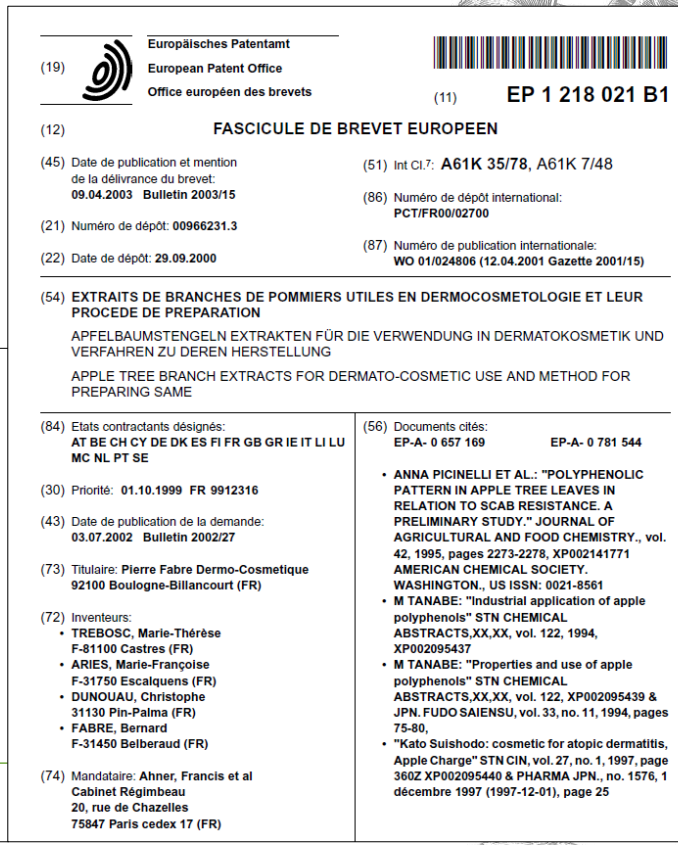


Dihydrochalcone utilisé dans la cosmétique et l'agro-alimentaire

Phloridzin potential in Cosmetics

- Several patents
- Several apple by-products studied





(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) EP 1 218 021 B1

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(45) Date de publication et mention de la délivrance du brevet:
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(21) Numéro de dépôt: **00966231.3**

(22) Date de dépôt: **29.09.2000**

(51) Int Cl.[7]: **A61K 35/78**, A61K 7/48

(86) Numéro de dépôt international:
PCT/FR00/02700

(87) Numéro de publication internationale:
WO 01/024806 (12.04.2001 Gazette 2001/15)

(54) **EXTRAITS DE BRANCHES DE POMMIERS UTILES EN DERMOCOSMETOLOGIE ET LEUR PROCEDE DE PREPARATION**

APFELBAUMSTENGELN EXTRAKTEN FÜR DIE VERWENDUNG IN DERMATOKOSMETIK UND VERFAHREN ZU DEREN HERSTELLUNG

APPLE TREE BRANCH EXTRACTS FOR DERMATO-COSMETIC USE AND METHOD FOR PREPARING SAME

(84) Etats contractants désignés:
AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE

(30) Priorité: **01.10.1999 FR 9912316**

(43) Date de publication de la demande:
03.07.2002 Bulletin 2002/27

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(56) Documents cités:
EP-A- 0 657 169 **EP-A- 0 781 544**

- **ANNA PICINELLI ET AL.: "POLYPHENOLIC PATTERN IN APPLE TREE LEAVES IN RELATION TO SCAB RESISTANCE. A PRELIMINARY STUDY." JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY., vol. 42, 1995, pages 2273-2278, XP002141771 AMERICAN CHEMICAL SOCIETY. WASHINGTON., US ISSN: 0021-8561**
- **M TANABE: "Industrial application of apple polyphenols" STN CHEMICAL ABSTRACTS,XX,XX, vol. 122, 1994, XP002095437**
- **M TANABE: "Properties and use of apple polyphenols" STN CHEMICAL ABSTRACTS,XX,XX, vol. 122, XP002095439 & JPN. FUDO SAIENSU, vol. 33, no. 11, 1994, pages 75-80,**
- **"Kato Suishodo: cosmetic for atopic dermatitis, Apple Charge" STN CIN, vol. 27, no. 1, 1997, page 360Z XP002095440 & PHARMA JPN., no. 1576, 1 décembre 1997 (1997-12-01), page 25**

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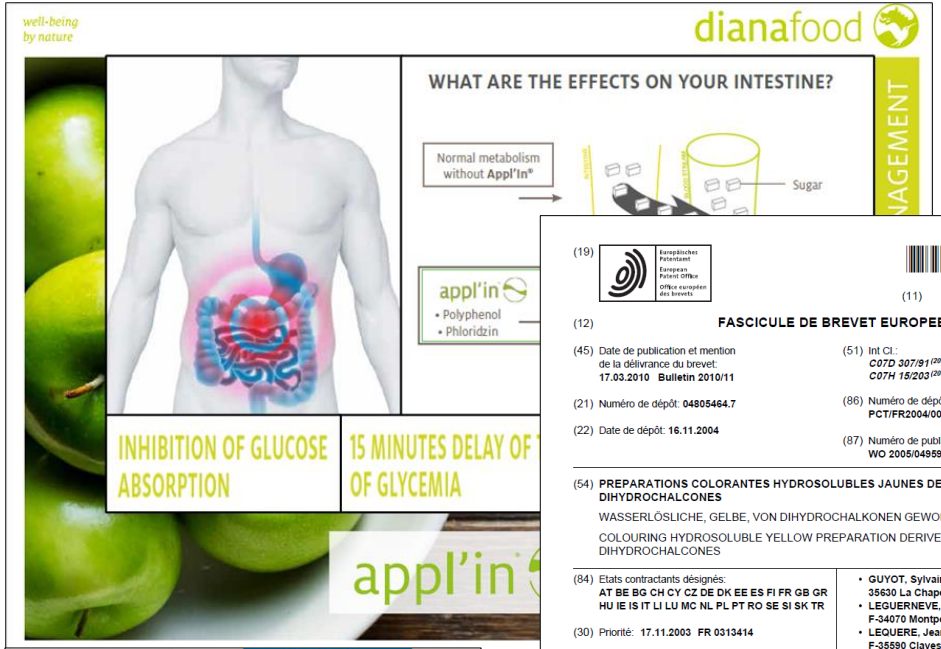
Phloridzin potential in Nutraceutics



(19) Europäisches Patentamt / European Patent Office / Office européen des brevets (11) EP 1 6

(12) **FASCICULE DE BREVET EUROPEEN**

(45) Date de publication et mention de la délivrance du brevet: **17.03.2010 Bulletin 2010/11**

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(22) Date de dépôt: **16.11.2004**

(87) Numéro de publication internationale: **WO 2005/049598 (02.06.2005 Gazette 2005/22)**

(54) **PREPARATIONS COLORANTES HYDROSOLUBLES JAUNES DERIVEES DES DIHYDROCHALCONES**

WASSERLÖSLICHE, GELBE, VON DIHYDROCHALKONEN GEWONNENE FÄRBEZUBEREITUNG

COLOURING HYDROSOLUBLE YELLOW PREPARATION DERIVED FROM DIHYDROCHALCONES

(84) Etats contractants désignés: **AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HU IE IS IT LI LU MC NL PL PT RO SE SI SK TR**

(30) Priorité: **17.11.2003 FR 0313414**

(43) Date de publication de la demande: **02.08.2006 Bulletin 2006/31**

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(56) Documents cités:
EP-A- 0 773 226 **EP-A- 0 850 948**
FR-A- 2 822 466

US 20030003120A1

(19) **United States**

(12) **Patent Application Publication** (10) **Pub. No.: US 2003/0003120 A1**
Gaudout et al. (43) **Pub. Date: Jan. 2, 2003**

(54) **PHLORIDZIN-RICH PHENOLIC FRACTION AND USE THEREOF AS A COSMETIC, DIETARY OR NUTRACEUTICAL AGENT**

(76) Inventors: **David Gaudout**, Fougeres (FR); **Denis Megard**, St Brice En Cogles (FR); **Claude Inisan**, Rennes (FR); **Christian Esteve**, Cosse Le Vivien (FR); **Frederic Lejard**, Arradon (FR)

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(21) Appl. No.: **10/105,040**

(22) Filed: **Mar. 22, 2002**

(30) **Foreign Application Priority Data**

Mar. 23, 2001 (FR) 0103968

Publication Classification

(51) **Int. Cl.**[7] **A61K 6/00**
(52) **U.S. Cl.** **424/401**

(57) **ABSTRACT**

A phenolic fraction of fruit obtained from the Rosaceae family and also to the process for the specific extraction of this fraction. The phenolic fraction is rich in dihydrochalcones (phloridzin and phloretin) and may be used as a cosmetic, dietary, or nutraceutical preparation.

Patent from INRA about the extraction of POP (Produit d'Oxydation de la Phlorizine), a yellow die

Scale-up extraction of apples

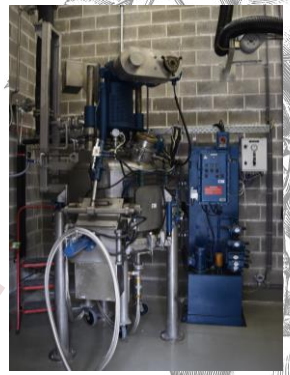
Lab-scale-1
(5 - 20 gr)

Lab-scale- 2
(200 gr)

75 litres

Semi-pilot scale
(4500 gr)

Pilot scale
(30 kg)



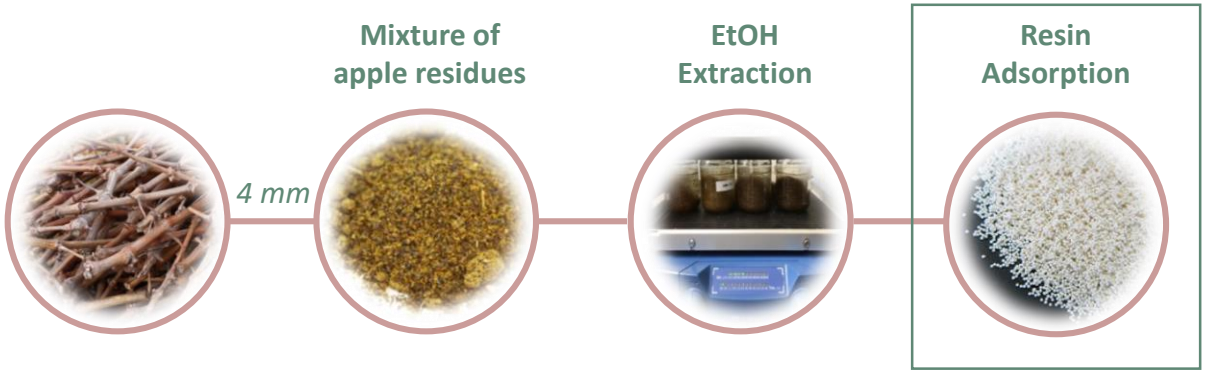
350 litres

Study of the scale-up

	Lab Scale - 1 (20 gr)	Lab Scale - 2 (200 gr)	Semi pilot Scale (4500 gr)	Pilot Scale (4500 gr)
Extraction yield	5,4 %	11,3 %	13,2 %	14 %
Concentration in Phloridzin	15,6 %	19,7 %	20 %	34 %

- The extraction yield increased as we go to higher scale
- Moreover, phloridzin content increased also in higher extraction scale

Bioactive concentration



	Initial extract	Enriched extract
Extract mass recovered	100 g	47.3 g
Phloridzin content	20% →	49%
Phloridzin recovered in concentrated extract		100%

- Total recovery of the bioactive (Phloridzin)
- High concentrated extract in Phloridzin

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European Regional Development Fund

Extracts characterization

	Initial extract	Enriched extract
Total Phenolic content	**160** mg EGA/g	**385** mg EGA/g
Anti-oxidant activity (DPPH)	**41** mg EGA/g	**75** mg EGA/g
Anti-oxidant activity (FRAP)	**0.46** mmol eq Fe(II)/g	**1.25** mmol eq Fe(II)/g

After resin adsorption

Conclusions

- ✓ Agricultural residues (apples, pears and vine) represent a promising source of ingredient for cosmetics and nutraceutics
- ✓ Environnement friendly technologies allow the extraction of these molecules
- ✓ However, the legislation on Novel Foods reste is a major limitation on the exploitation of these results

Project under the program
Interreg North-West Europe
European Regional Development Fund

With the financial support of the European Regional Development Fund and Wallonia
EUROPEAN UNION
European Regional Development Fund
Wallonie

Thank you

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ICMR Institut de Chimie Moléculaire de Reims

Enzymatic modifications of natural extracts of agricultural by-products:

to produce new bioactive molecules and enhance their biological properties for use in cosmetics

Final event of the AgriWasteValue project

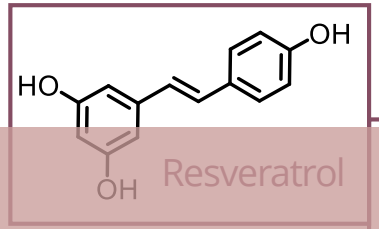
Laurène Minsat

PhD student

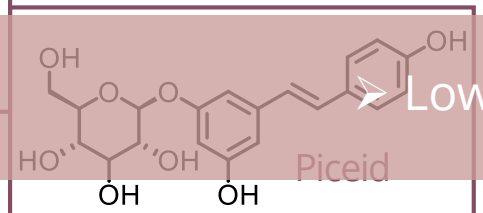
Supervisors: Pr Jean-Hugues Renault, Pr Florent Allais

Natural extracts : a source of molecule of interest

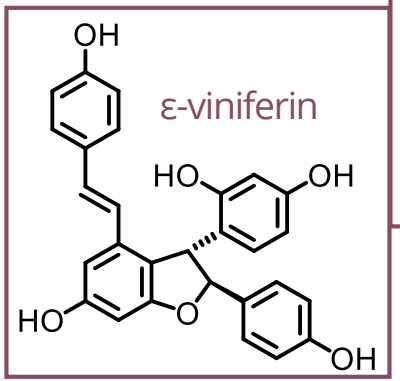
- Several phenolic compounds with diverse biological properties



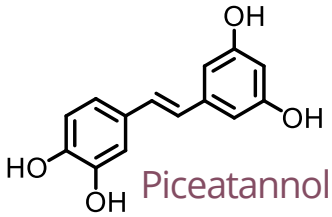
Resveratrol



Piceid

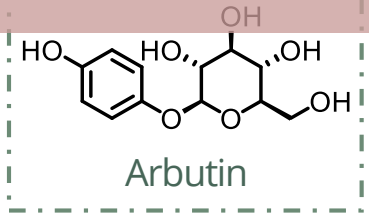


ε-viniferin

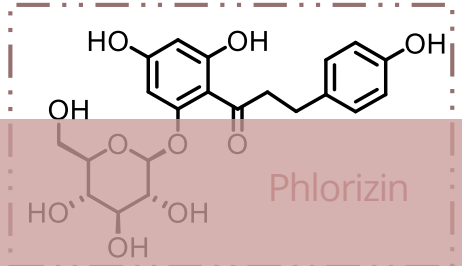


Piceatannol

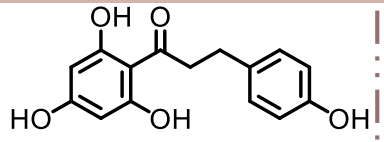
- Low water solubility



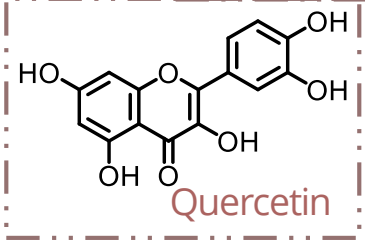
Arbutin



Phlorizin



Phloretin



Quercetin

Enzymatic *O*-glycosylation

To increase the water solubility of hydrophobic molecules

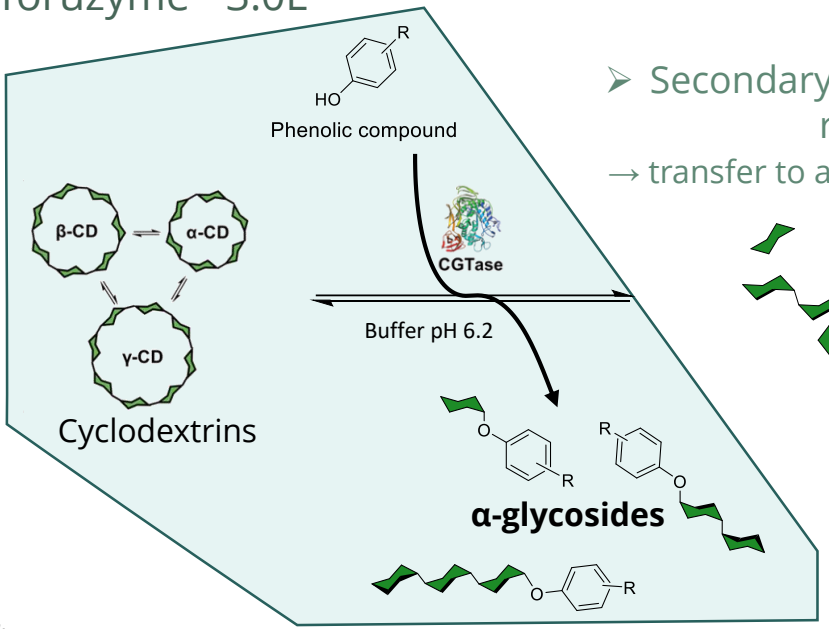
❖ Formation of a covalent bond between a carbohydrate (“donor”) and another molecule (« acceptor »)

❖ Use of a commercial CGTase: Toruzyme® 3.0L[1]

Advantages:

❖ Use of β-CD as substrate → solubilization of phenols (no use of co-solvent)

❖ Production of α-glycosides (≠ natural β-glycosides) → new properties[2]



➢ Secondary activity: coupling reaction

→ transfer to an acceptor site (OH)

Free carbohydrates

Source/scheme inspiration : © Sophie Beeren/ Technical University of Denmark

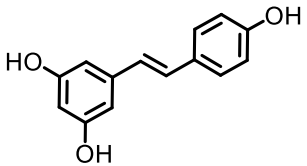
[1] *T. Marié thesis . « Extraction et fractionnement de stilbénoïdes issus de cultures cellulaires de vigne ». Université de Reims Champagne-Ardenne, 2019*

[2] *Torres et al, 2011 https://doi.org/10.1002/adsc.201000968*

Enzymatic *O*-glycosylation

on resveratrol

❖ T. Marié's PhD work, « Extraction et fractionnement de stilbénoïdes issus de cultures cellulaires de vigne ». Thesis. ICMR -URCA/URD ABI, 2019.

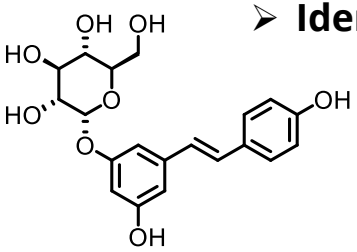


Resveratrol

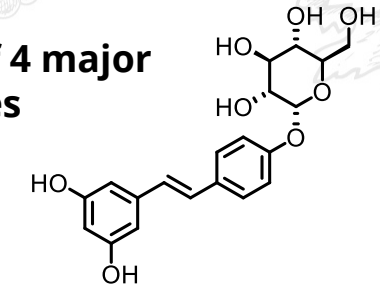
1. Encapsulation in the β-CD

2. Glycosylation by Toruzyme® 3.0L

➢ **Identification of 4 major glycosides**



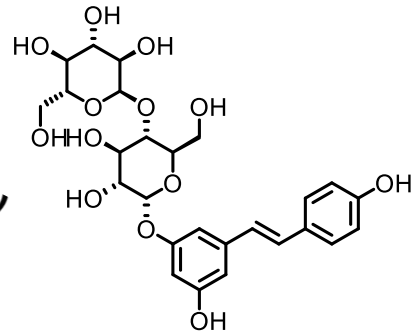
3-*O*-α-D-glucosyl-resveratrol



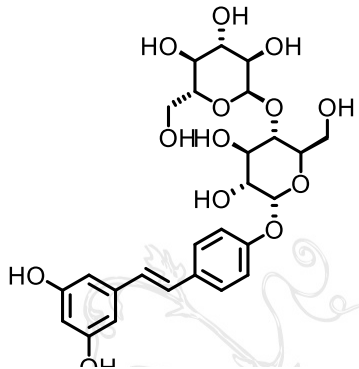
4'-*O*-α-D-glucosyl-resveratrol

➢ Increased solubility (x65)
Compared to resveratrol

➢ New surfactant properties



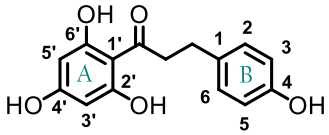
3-*O*-α-D-maltosyl-resveratrol



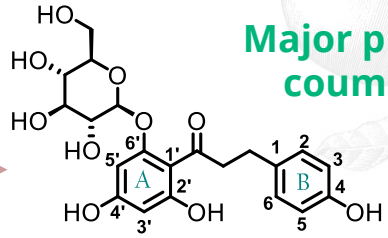
4'-*O*-α-D-maltosyl-resveratrol

Enzymatic *O*-glycosylation

Study on apple tree molecules



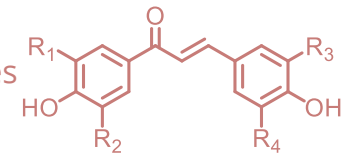
Phloretin



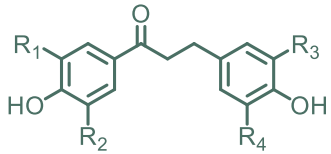
Phlorizin

Major phenolic coumpond

Chalcones



Dihydrochalcones



R_1 ; R_2 ; R_3 ; R_4 = H ou OH ou OMe

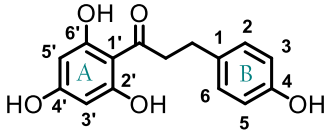
1 To facilitate the study

2 To produce potentially more active new molecule

Minsat *et al.*, *Antioxidants*, 2021

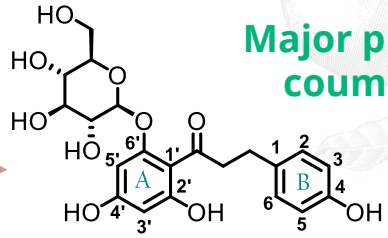
Enzymatic *O*-glycosylation

Study on apple tree molecules



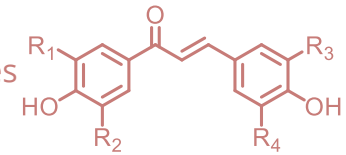
Phloretin

Major phenolic coumpond

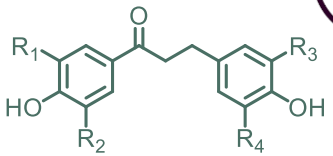


Phlorizin

Chalcones

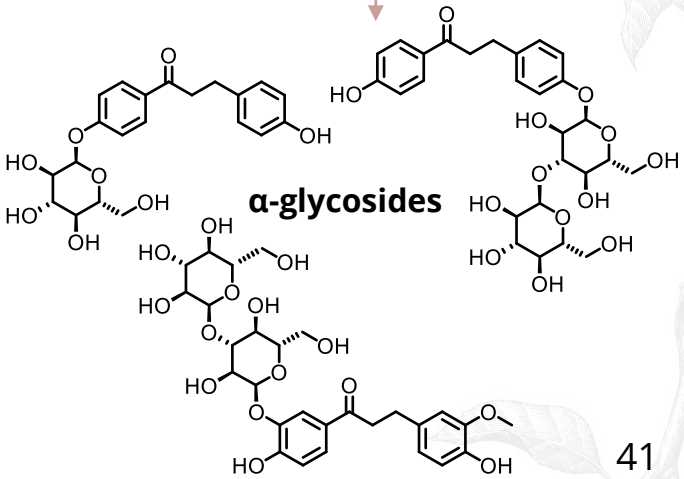


Dihydrochalcones



R_1 ; R_2 ; R_3 ; R_4 = H ou OH ou OMe

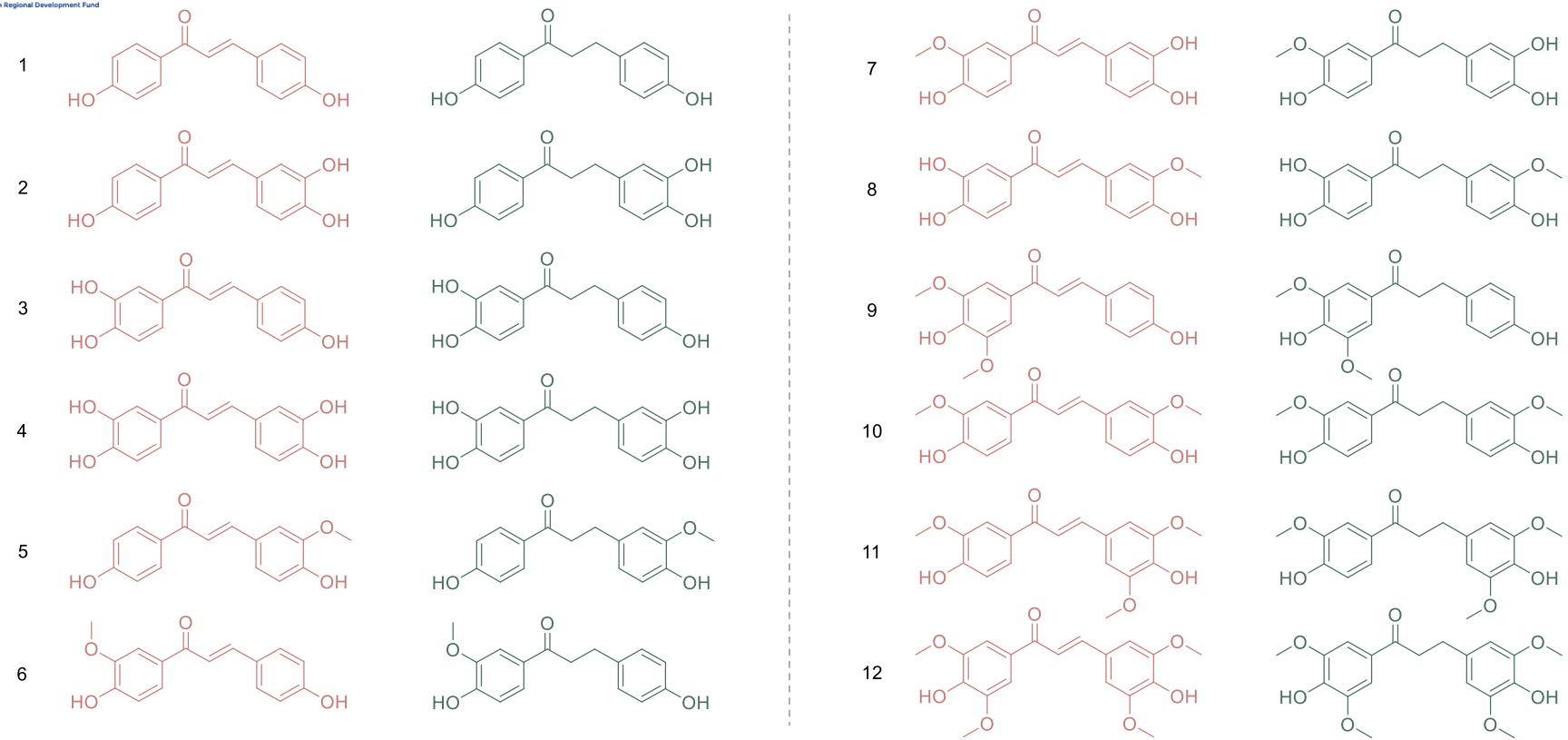
Enzymatic glycosylation



α-glycosides

Minsat *et al.*, *Antioxidants*, 2021

Development on phloretin analogues

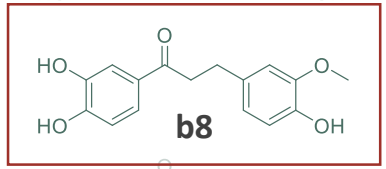


Series a : chalcones
Series b : dihydrochalcones

Development on phloretin analogues

⇩

Molecules with only 2 OH
To evaluate the impact of the double bond



⇩

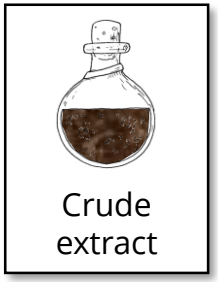
➢ Promising results for DPPH and anti-tyrosinase tests

Series a : chalcones
Series b : dihydrochalcones

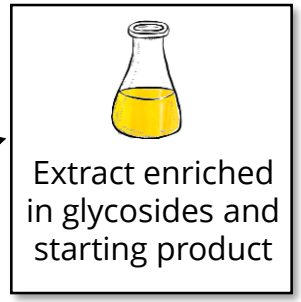
Comprehensive strategy of purification

Purification on resin Amberlite® XAD 7

Injection

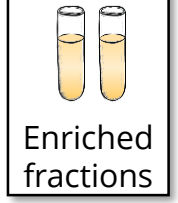


Crude extract



Extract enriched in glycosides and starting product

Fractionation by CPC

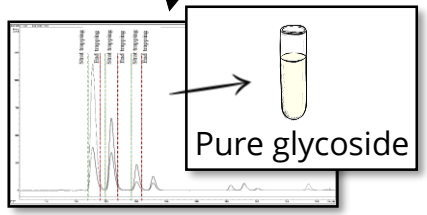


Enriched fractions

Injection

(Semi-)preparative HPLC

HPLC profil acquisition

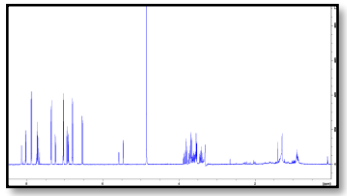


UV chromatograms

Evaporation

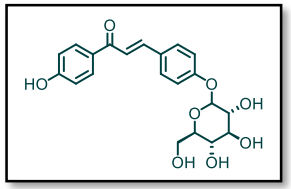
NMR system (600 MHz with cryo-probe)

NMR profil acquisition



NMR spectra

Structural elucidation



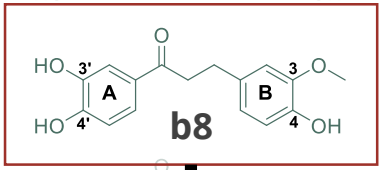
Glycosides of phloretin analogues – Overview

5 new compounds :
- **2 monoglycosides**
- **3 diglycosides**

- Regioselectivity: B ring
- Double bond isomerization
- Diglycosides: α(1→4) linkage

6 new compounds :
- **2 monoglycosides**
- **4 diglycosides**

- Regioselectivity: B ring
- Diglycosides: α(1→3) linkage



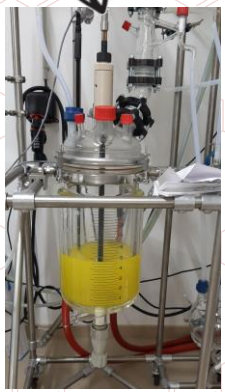
7 new compounds :
- **3 monoglycosides**
- **4 diglycosides**

- Regioselectivity: A ring (position 14)
- Diglycosides: α(1→4) linkage
- No diglycosides in position 7

Diglycosides → only monodesmosides (2 glucoses on the same OH)

Series a : chalcones
Series b : dihydrochalcones

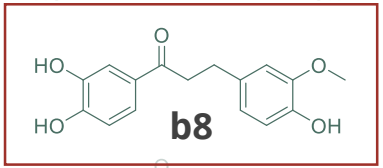
Glycosides of phloretin analogues



10L reactor

- Stop by addition of HCl to reach pH 2

➡ Enzyme denaturation



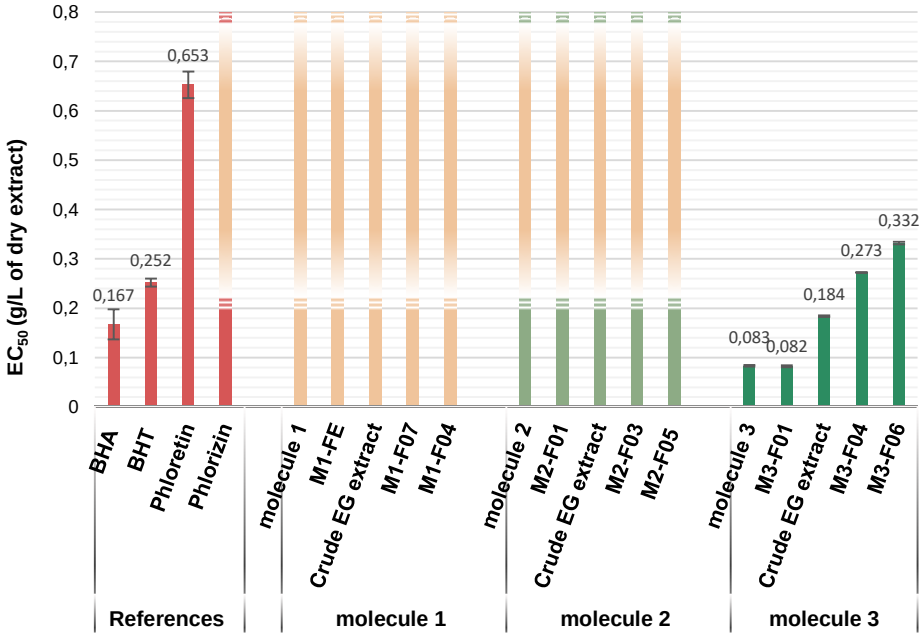
Round-bottom flask

- Stop by addition of MeOH (1 to 1 V/V) and thermal shock (ice bath)

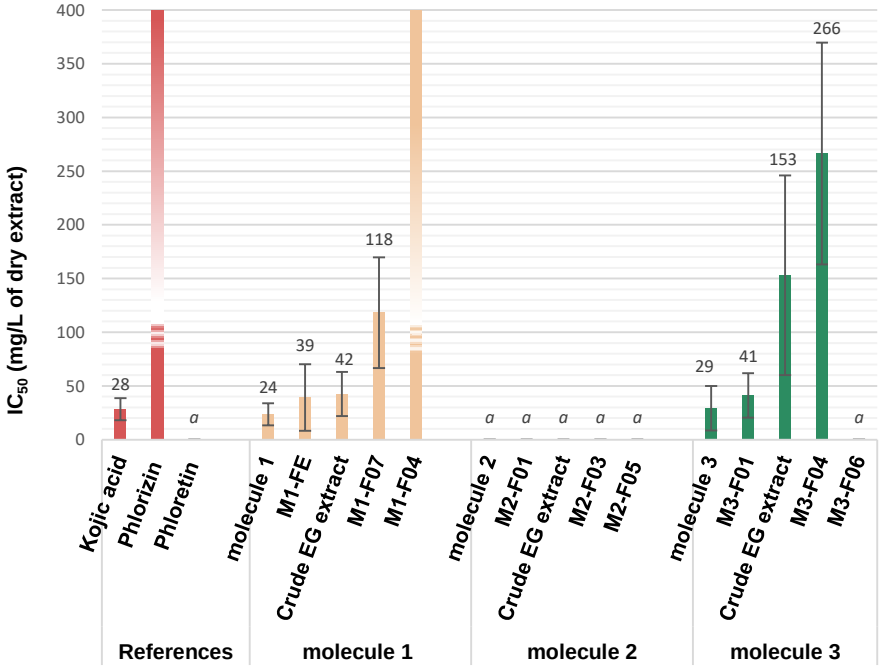
⬇ Enzyme inactivation

Evaluation of biological activities

DPPH test results
Antiradical properties

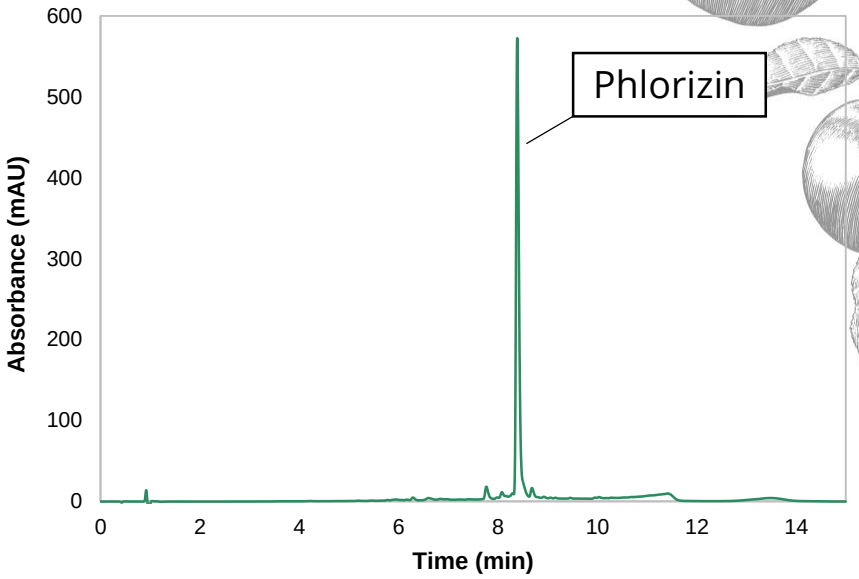


Inhibition of tyrosinase test results
Depigmenting properties



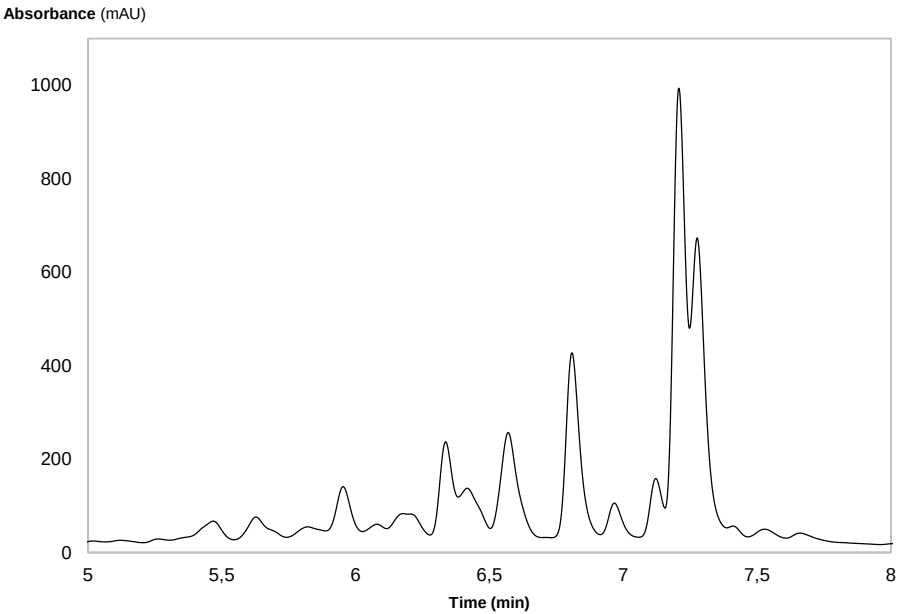
➢ Still interesting activities after glycosylation

Transposition on apple tree wood extracts

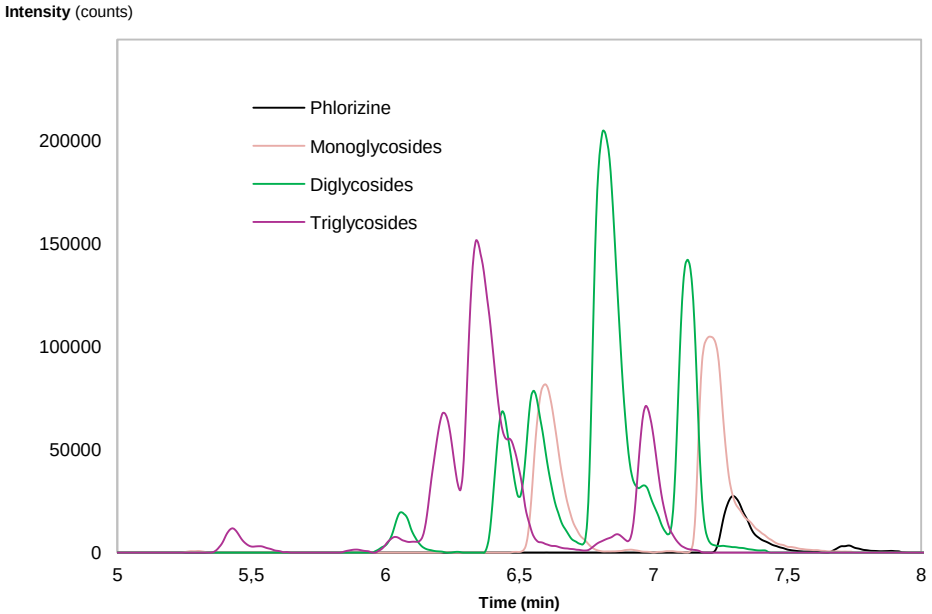


O-glycosylation of apple tree extract

UV chromatogram (280 nm) after 1h15 of reaction

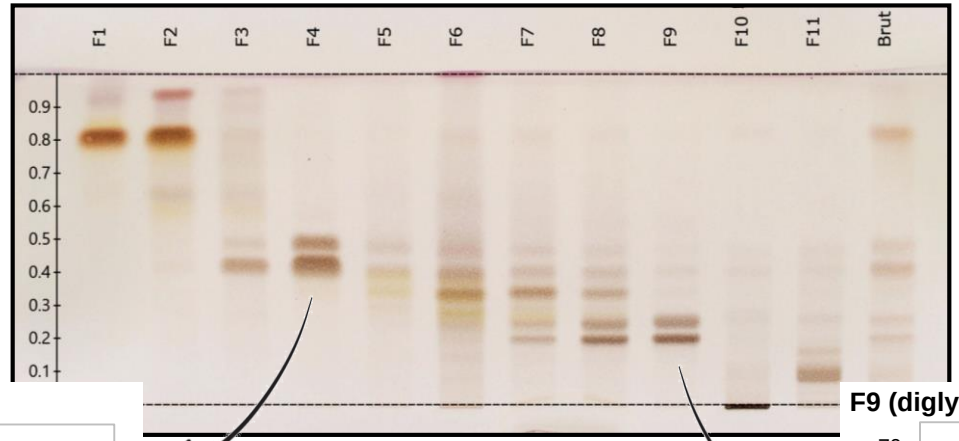


Extracted Ion Chromatograms (EIC)

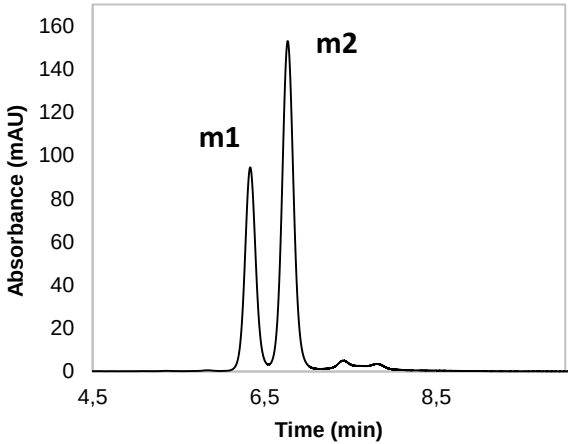


Identification of phlorizin glycosides

1 Purification by CPC

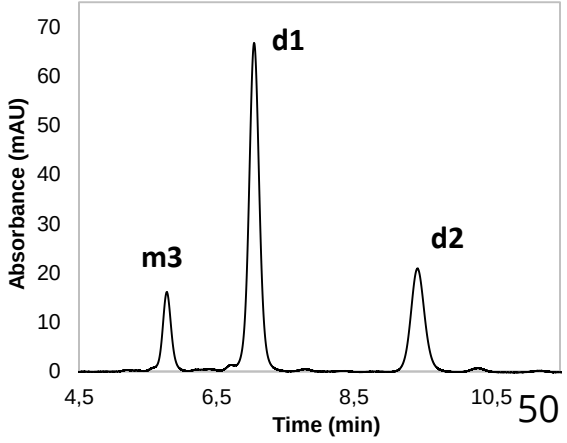


F4 (monoglycosides)

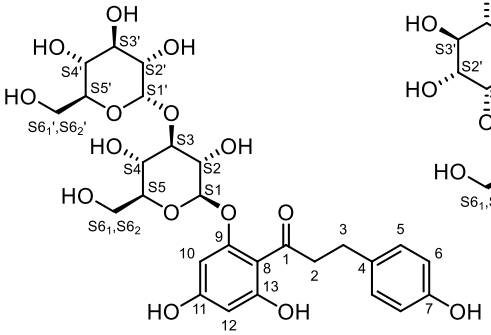


2 Purification by preparative HPLC

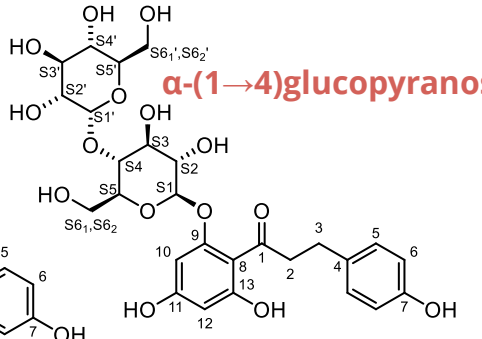
F9 (diglycosides)



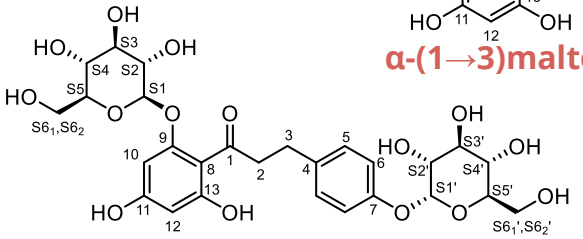
Identification of phlorizin glycosides



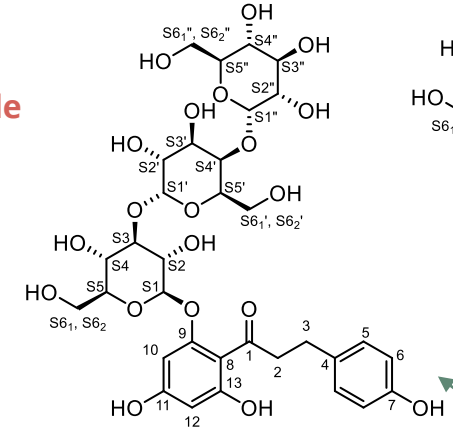
α-(1→3)glucopyranoside



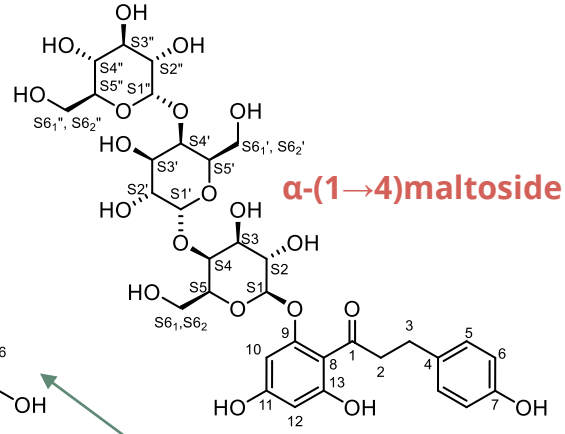
α-(1→4)glucopyranoside



4-*O*-α-glucopyranoside

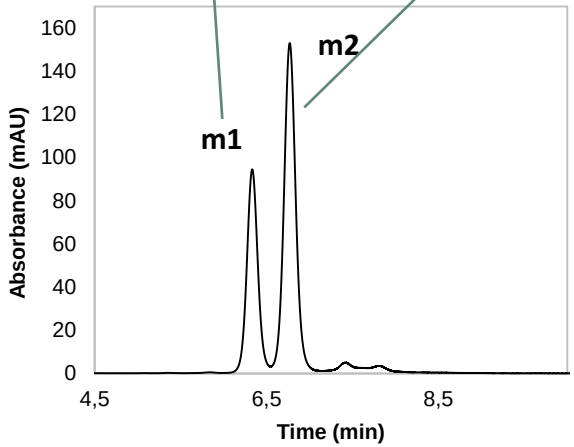


α-(1→3)maltoside

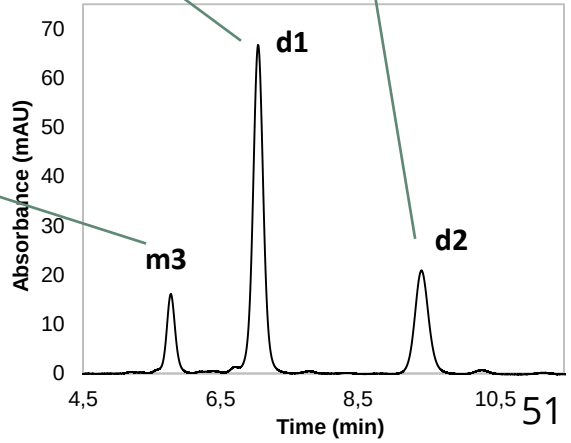


α-(1→4)maltoside

F4 (monoglycosides)

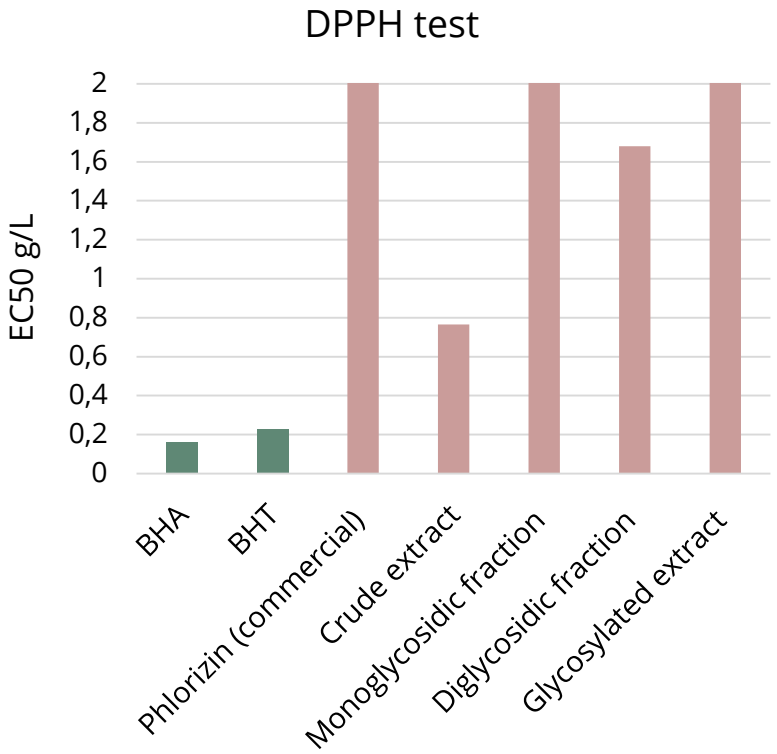


F9 (diglycosides)



2 Purification by preparative HPLC

Evaluation of biological activity



- ✓ BHA and BHT were used as references
- ✓ No activity observed for phlorizin
- ✓ Good activity for the crude extract of apple tree wood
- ✓ A low activity observed for the diglycosidic fraction

Transposition on pear tree wood extracts

- Low concentration of phenolic compounds in the extract
- Identification of **arbutin** monoglycosides by LC-MS analysis but in very low quantities
- Problems encountered to purify and isolate the compounds of interest
- Unsuitable valorization way

Transposition on vine extracts

- Large diversity of phenolic compounds in the extract
- Protocol developed on apple tree molecules not applicable at this stage
- Problems encountered for the production of glycosides
- Identification of 2 **resveratrol** monoglycosides
- Suitable valorization way with some adjustments

Conclusion

☑ Succesful development of a comprehensive methodology for enzymatic modification / purification on model molecules at **large scale (10g of product)**

☑ Succesful transposition on apple tree extracts

☒ Methodology modifications are required to be applicable to vine extracts

☒ Methodology not applicable to pear tree extracts

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Thank you for your attention

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Université de Reims Champagne-Ardenne
valbiom

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European Regional Development Fund

B
R
E
A
K

By-products & residues from viticulture & arboriculture : promising natural & local ingredients!

Next conferences
4pm

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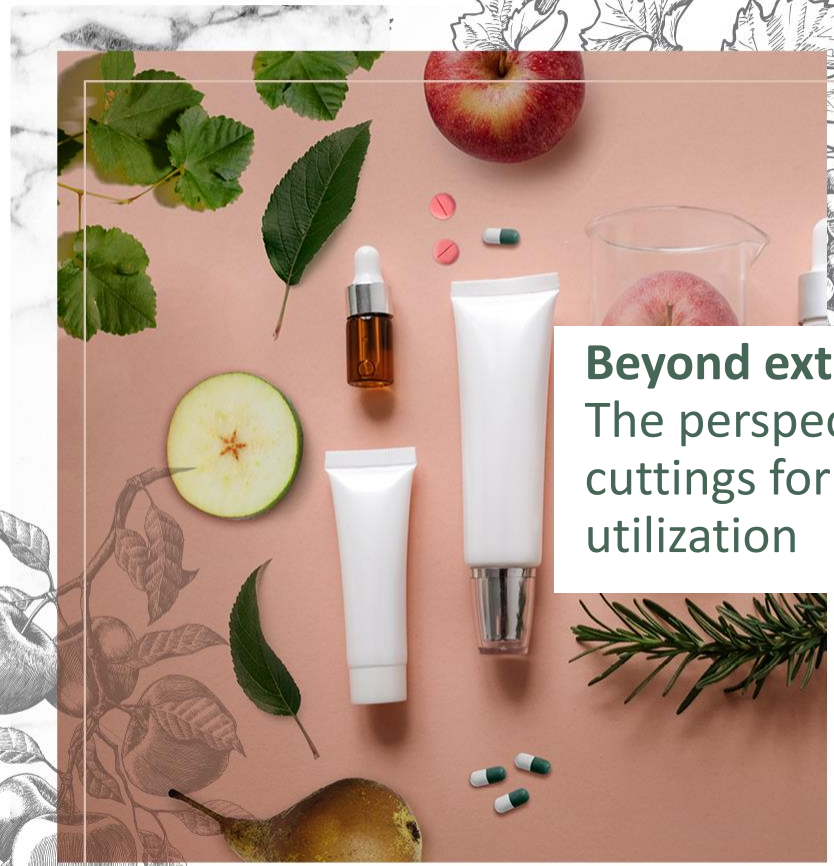
AgriWasteValue
European Regional Development Fund



Beyond extractives

The perspective of tree and vineyard cuttings for a material and energetic utilization

Final event of the AgriWasteValue project

Speaker: Dr. Patrick Ballmann

What's the goals for this specific part of the project?

- Demonstration of a potential way to use more than the extractives
- The production of bio solvents like ethanol by microbial fermentation
- Provision of electricity and heating energy by biogas production for the whole process
- Increase the whole value of the wooden tree biomass

What's left in the extracted biomass?

Component	Before Extraction	After Extraction
Extractives	16.2 %	6.1 %
Cellulose	32.4 %	37.1 %
Hemicellulose	16.9 %	20.7 %
Lignin	33.6 %	34.2 %

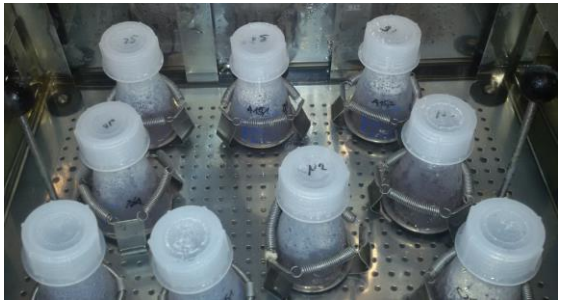
- Cellulose and hemicellulose contained enough sugars for a fermentation process
- Pretreatment process necessary!

Developing of a treatment strategy

Based on the pretreatment process of wheat straw

Thermal **P**ressure **H**ydrolysis (TPH)

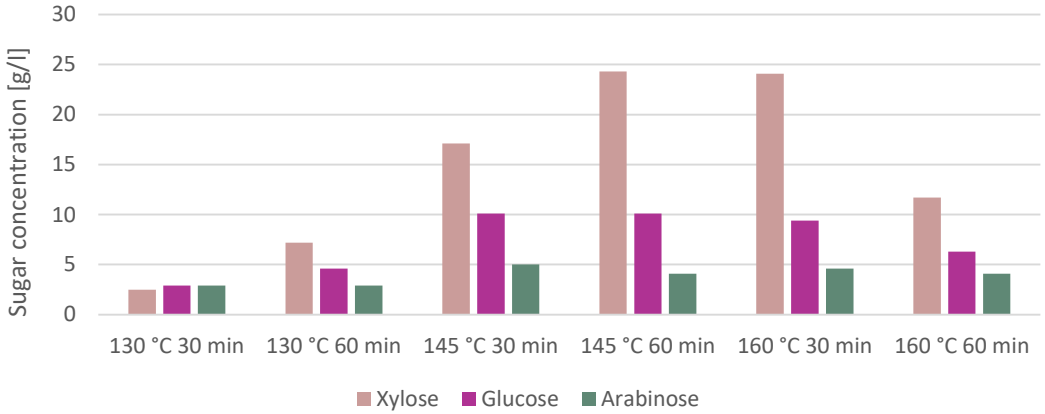
Enzymatic **H**ydrolysis (EH)



Sugar solution for bio solvent production!

Getting the optimal conditions for both process to maximize the sugar release!

Results of the pretreatment process



- Best results with 160 °C and 30 min

Enzyme dosage	Glucose-concentration [g/l]	Glucose recovery[%]
30 FPU/ g DM	40,3	79,4
45 FPU/ g DM	42,5	82,4
60 FPU/ g DM	48,3	90,7

- Good glucose recovery
- But high enzyme demand

High enzyme demand necessary → General high costs for pretreatment

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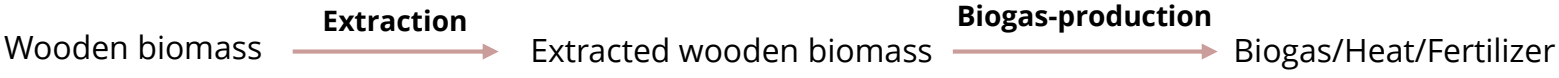
What does it mean for a potential bio solvent production?

- Under recent conditions the cost for the complete pretreatment is too high
- Whole process currently not economical feasible
- New valorization path → No bio solvent production
- Using the extracted biomass directly for the biogas production



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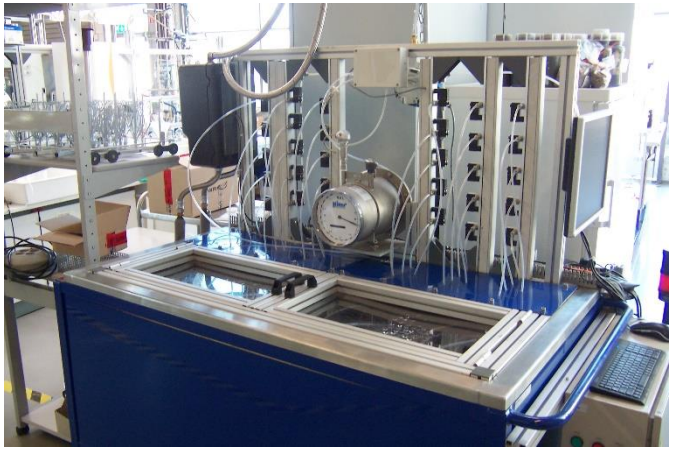
Evaluation of the biogas potential

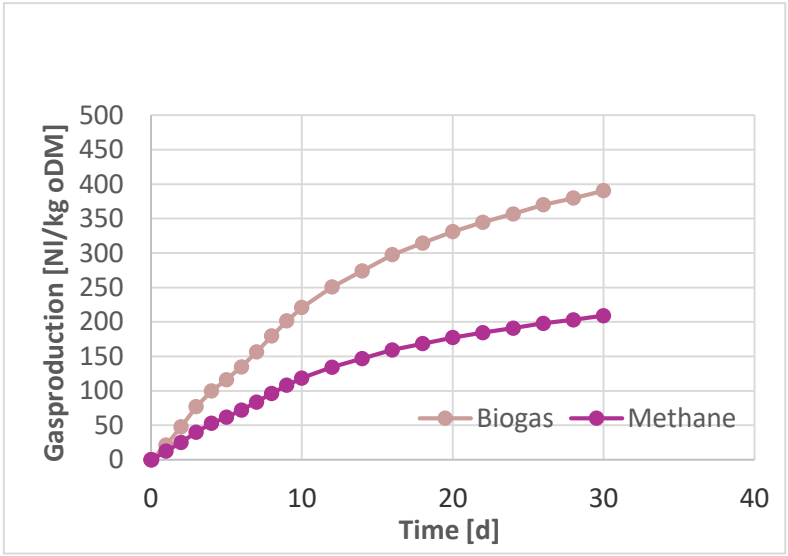
Using static biogas tests with various biomass samples

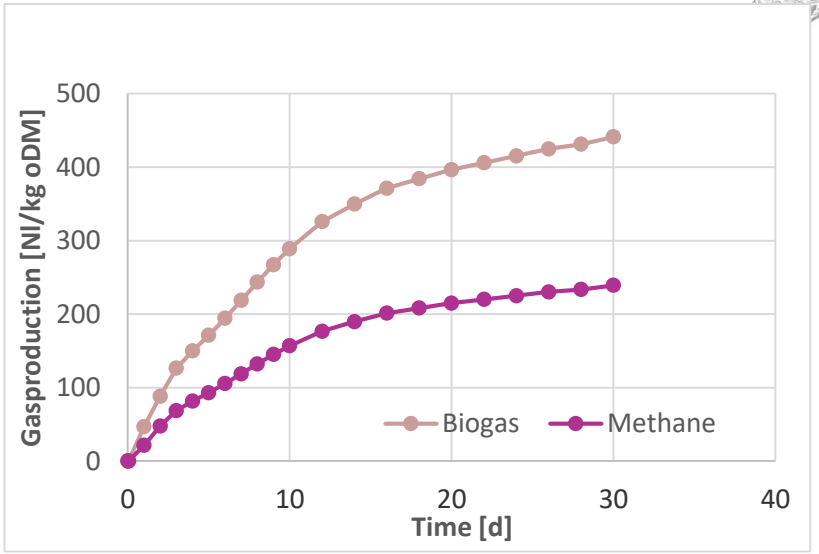


Mixing biomass with seed sludge and measure the gas production over 4 weeks

Results of the static biogas test

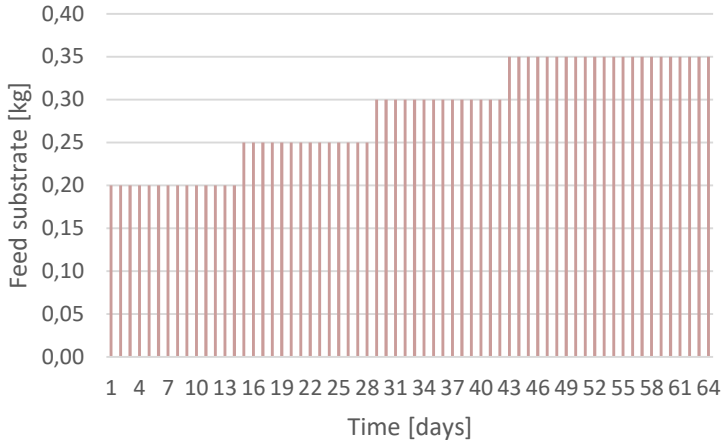
Comparison between extracted (left) and not extracted biomass (right)





A little bit more biogas from the unextracted biomass, but promising results overall

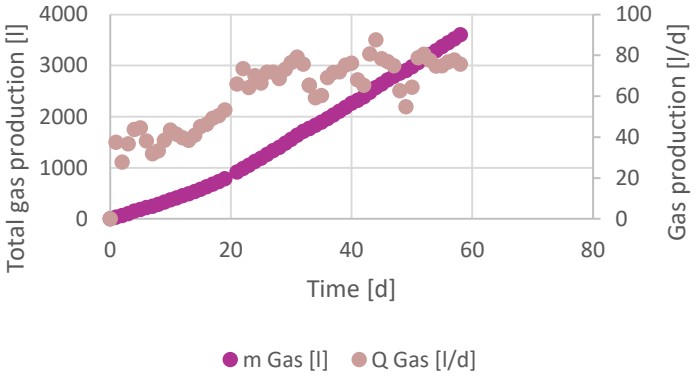
Up-scaling in the dynamic biogas test

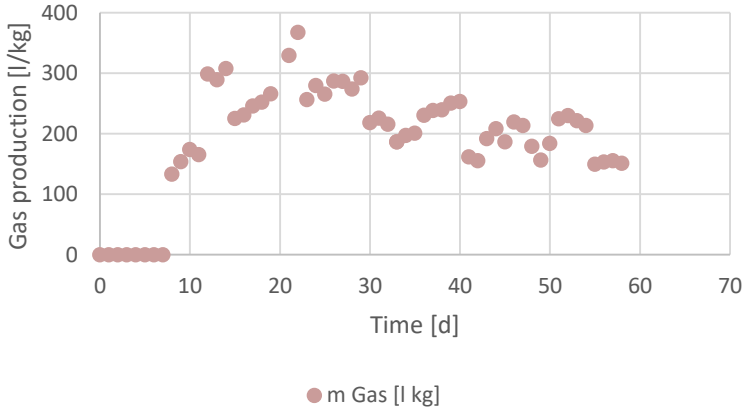


- 100 L reactor with active stirring and feeding
- Active sampling for analytical measurements possible
- Measurement of biogas production
- Actually 3-4 months duration → In our case shorter time
- Biomass had a very high dry matter content (over 95 %), therefore mixing with water!

Results of the dynamic biogas test

Mixed results → Good overall biogas production, but specific biogas production not so good





- Stirring problems → Small heavy particles concentrated on the bottom of the reactor
- Sampling was very difficult

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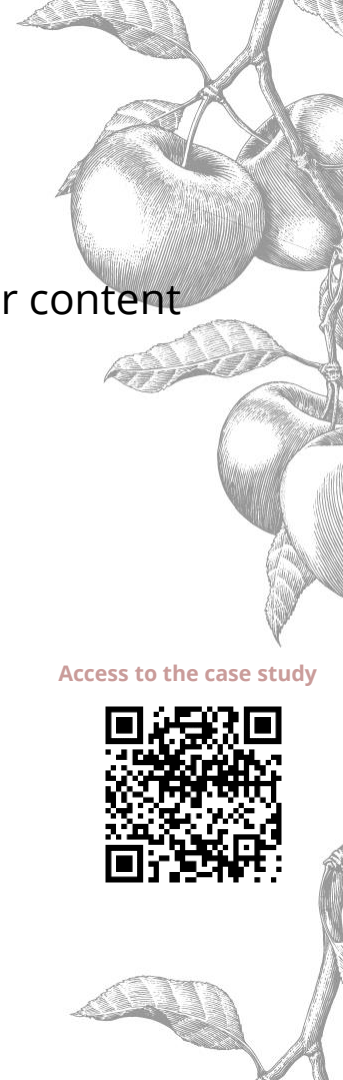
What does it mean for a potential biogas production?

- Using the extracted biomass as substrate alone difficult → High dry matter content
- Stirring problem must be resolved!
- Potential solution for both problems: dry fermentation process!
- Critical factor: need enough biomass 5.000 – 10.000 t per year!
- Using as a co-substrate with other biomasses like maize or grass silage
- A case-study was made by the project partner Valbiom!
- Sending the residues to project partner Delphy for fertilizer experiments



Access to the case study

Project under the program
Interreg North-West Europe
European Regional Development Fund

With the financial support of the European Regional Development Fund and Wallonia
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Thank you for your attention

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To orchards cuttings residues to fertilizers : closing the loop

Cor Van Oers,
Delphy

The trial 2022 (WP T3: Fertilizertest)

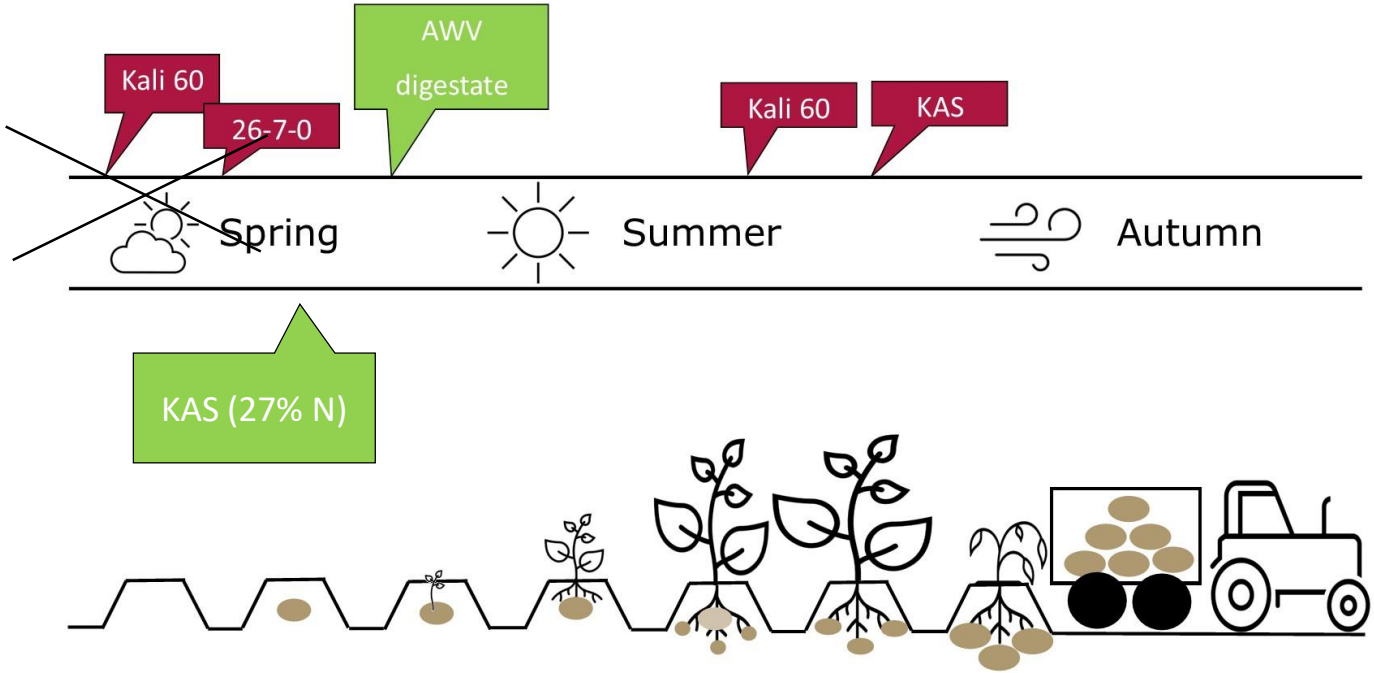
Research question

→ What is the value of the residues of orchards cuttings after processing use of cosmetics?

Field trial		
In 4 repetions, x objects		
Crop	potato	
Variety	Frieslanders	
Soil	clay	
Location	Netherlands	

Nutrient analyses

Parameter/Component		
Dry matter content		13,2%
Total nitrogen amount	Total N	3,243 kg/ton
Ammonium nitrogen	NH4+	1,28 kg/ton
Calcium	Ca	17,58 kg/ton
Potassium	K	15,46 kg/ton
Phosphorous	P	7,62 kg/ton
Acetic acid	Azijnzuur	0,043 kg/ton
Sodium	Na	0,698 kg/ton



	Before planting 01-03-2022		Before planting 27-03-2022		Before making ridges 26-04-22		Plants touch each other		Largest tubers are 1cm	
A (standard)	250 kg/ha	Kali 60	750 kg/ha	26-7-0			250 kg/ha	Kali 60	200 kg/ha	KAS
B (AWV)			500 kg/ha	KAS	4,5 ton/ha	AWV	350 kg/ha	Kali 60	350 kg/ha	KAS
C (80% working coefficient)	200 kg/ha	Kali 60	375 kg/ha	26-7-0	200 kg/ha	KAS	200 kg/ha	Kali 60	200 kg/ha	KAS

Analyses

- Growth
- Leaf and soil nutrient analyses
- Yield (kg/ha and sorting)
- Quality and nutrient content tubers

Things we ran into during applying:

- You have to mix it during applying
- You need more than 20 tonnes to apply by machine in the field

Growing season

- Planting: 28th of March
- Applying digestate: 26th of April
- Harvest: 8th of August

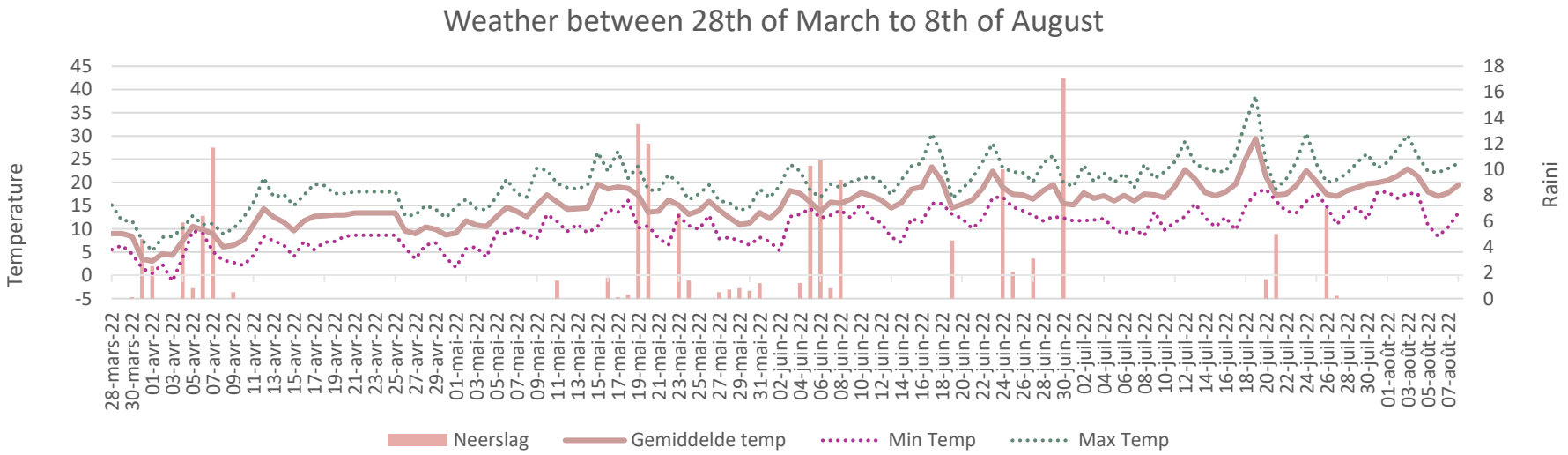


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Photo 08-06-2022

Interreg North-West Europe
AgriWasteValue
European Regional Development Fund

Photo 18-07-2022

Observations during season

- No differences in growth between objects.
  - (see photo 08-06-2022)
- No differences in blooming or in dying off
  - (see photo 18-07-2022)

Leaf and soil content

Soil content	Nitrogen (g/kg)	Potassium (g/kg)	Calcium (g/kg)
A (Standard)	140	83	285
B (AWV)	150	67	224
C (80%)	142	63	182

Leaf content	Nitrogen (g/kg)	Potassium (g/kg)	Calcium (g/kg)
A (Standard)	55	52	29
B (AWV)	50	52	29
C (80%)	53	55	30

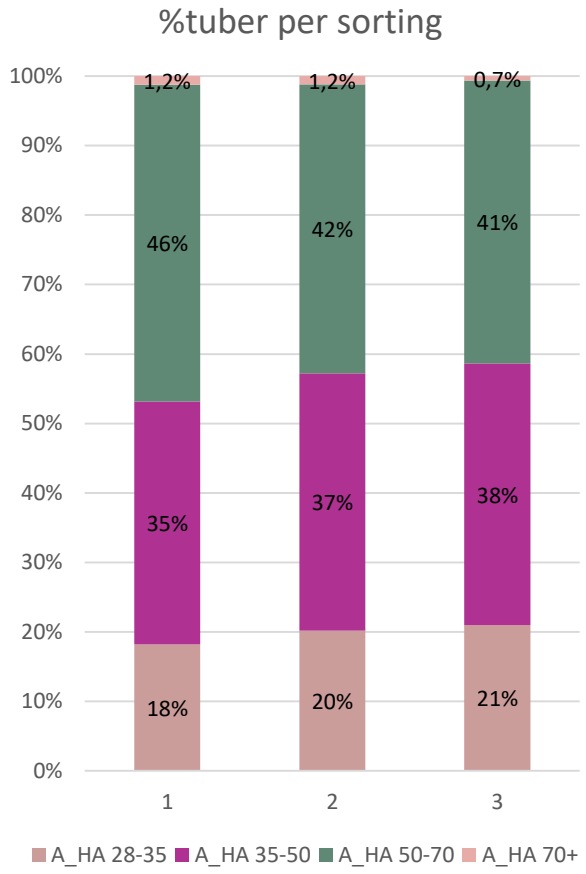
Tuber content

Tuber content	Nitrogen (g/kg product)	Potassium (g/kg product)	Calcium (g/kg product)
A (Standard)	0,70	5,129	0,74
B (AWV)	0,70	5,078	0,74
C (80%)	0,63	5,069	0,83

Yield

	Aantal /ha
A (Standard)	340.556
B (AWV)	345.278
C (80%)	339.167

	Kg/ha	Kg/ha 28-35	Kg/ha 35-50	Kg/ha 50-70	Kg/ha 70+
A (Standard)	34.647	2042	8933	22.447	1225
B (AWV)	33.603	2436	9508	20.419	1239
C (80%)	32.342	2408	9653	19.639	642



Quality

	Under water weight	% Starch
A (Standard)	423	16,8
B (AWV)	435	17,4
C (80%)	425	16,9

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Conclusion

- Only one year of open field trial
- Growing season was hard for the potatoes to grow
- The residues gives the same yield as normal fertilizer
- The only difference is the under water weight

Delphy

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With the financial support of the European Regional Development Fund and Wallonia
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Thank you

Cors VAN OERS

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valbiom

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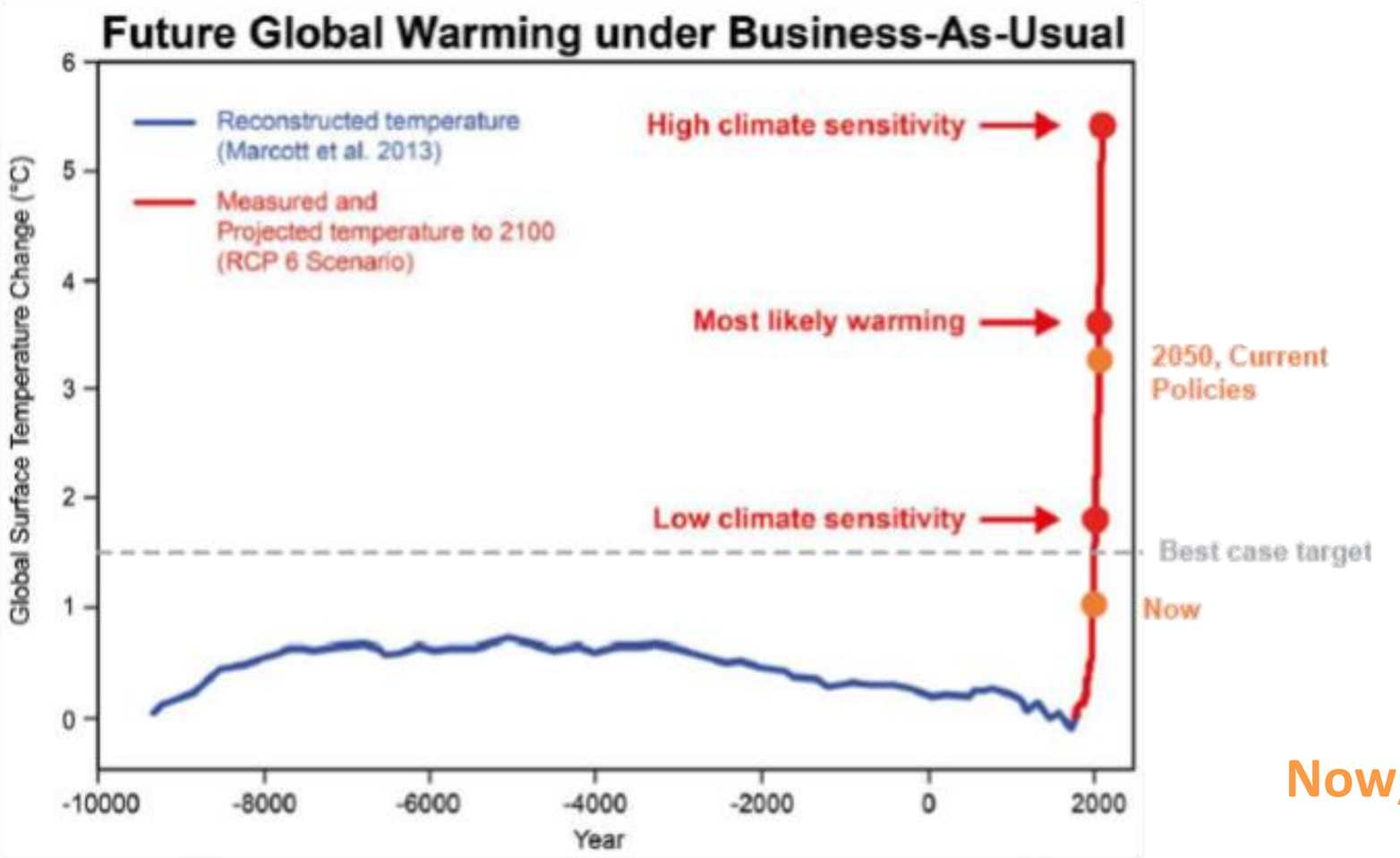
Green Solving Attitude

From Biomass to Added Value Solutions

EXISTENTIAL THREAT



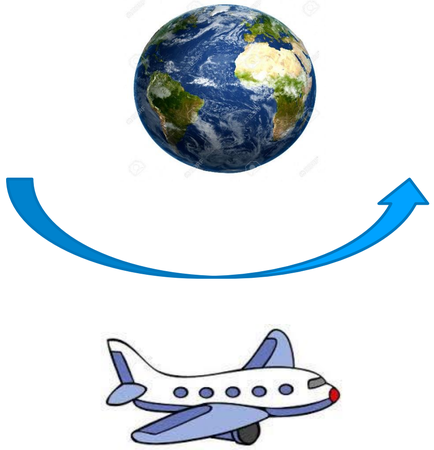
- **Global Warming scenario if we continue with the current model**
- **By 2050 the average temperature can reach + 3°C**
- **Reduction by 40% of UE CO2 emissions**

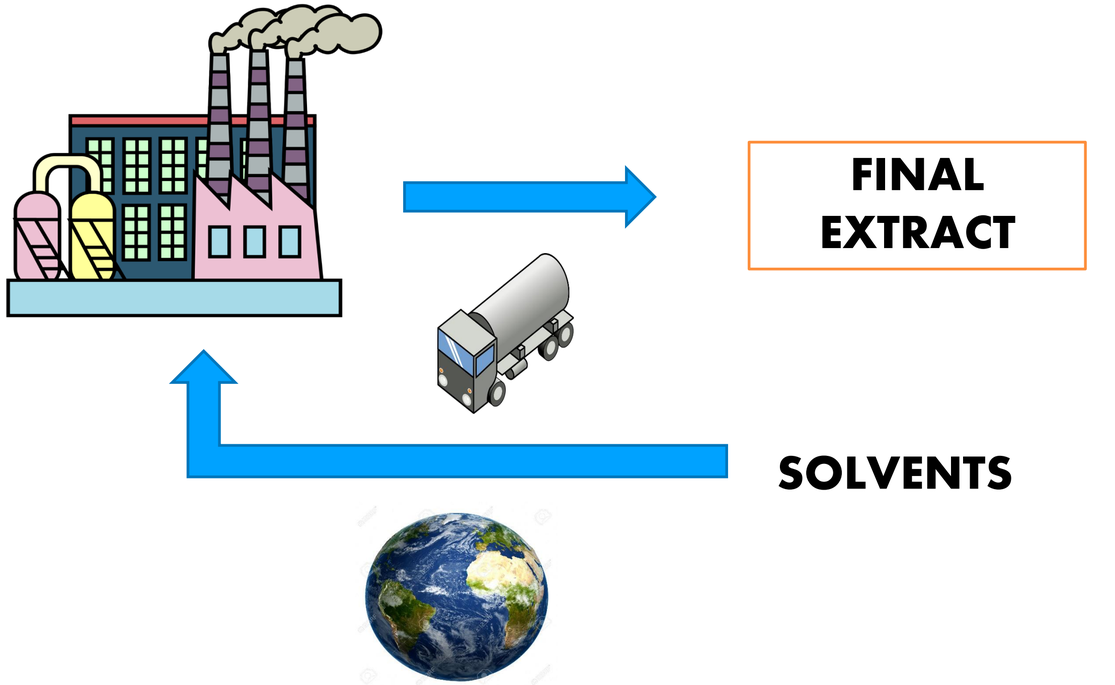
Now, all actions are important

International Panel on Climate Change, Special Report on Global Warming of 1.5°C, 2018

SUPPLY CHAIN
COSMETIC ACTIVES

Many extracts today are coming from exotic origins !





**Does it really make sense to import small quantities of rare flowers from other part of the world by air cargo ?
Carbon footprint ? Traceability ?**

12 PRINCIPLES OF GREEN CHEMISTRY

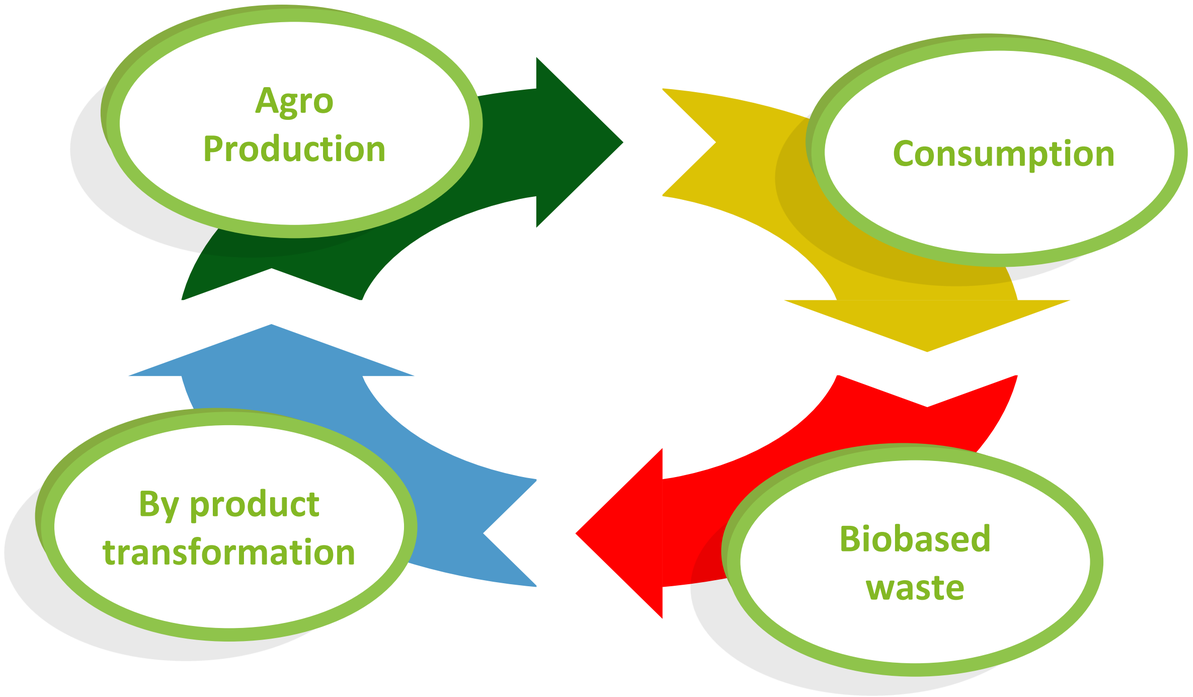
Paul Anastas & John C. Warner 1998

CIRCULAR ECONOMY

The concept of waste doesn’t exist in nature :

« In nature nothing is created, nothing is lost, everything changes.»

Antoine-Laurent de Lavoisier



- **Circular economy principles can offer many opportunities for the full chain of chemical industry to become more ressource efficient**
- **Cosmetic sector :**
  Up Cycling to reduce the carbon foot print of cosmetic industry but performances must be similar

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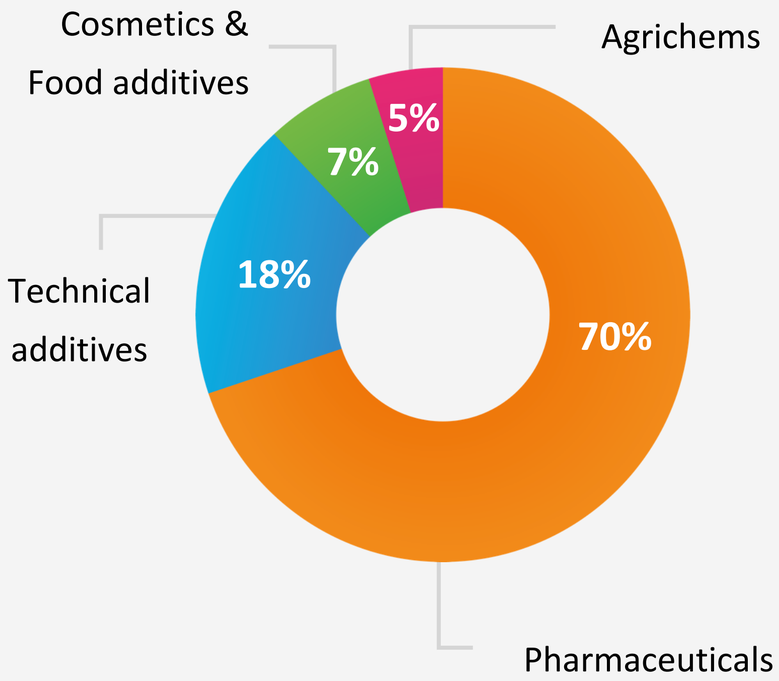
Fine Chemicals for the **Life Sciences** and the **High-Tech Industries**

2021
KEY FIGURES

Acting as a Global Player



€ 236M
revenue
(2021)

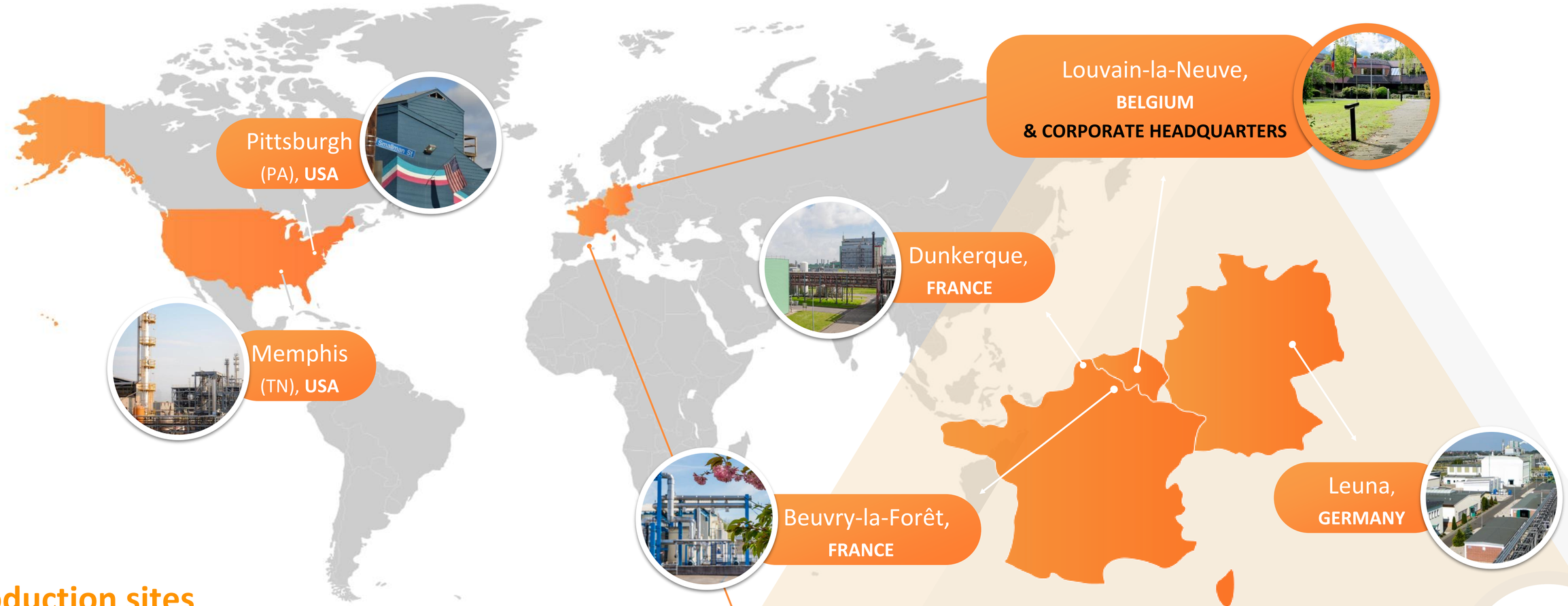
€ 42M
EBITDA
(2021)

91
R&D
resources

915
employees

4 EU & 2 US
sites

WE HAVE EXPANDED GLOBALLY SINCE 2004



6 Production sites
Corporate Headquarters in Belgium

HOW ARE WE ORGANIZED ?

1 | End-Markets Division

Pharma

MINAKEM®

Cosmetics

MINASOLVE®

Agro

MINAGRO®

2 | Niche Technologies Division

Green chemistry

PENNAKEM®

Green extraction

ecoXTRACT®

High Pressure & Polymers

PRESSURE® CHEMICAL

Challenging Chemistry

MINASCENT® TECHNOLOGIES

MINASOLVE :
GREEN SOLVING ATTITUDE

“science without conscience is but the ruin of the soul”
Rabelais

Biobased

Sustainably produced ingredients
for cosmetic application

Process

Eliminate the use or generation
of hazardous substances
Produce less hazardous molecules,
biodegradability

Application

Providing solutions adapted to
customers challenges
linked with our common social and
environnemental values

MINASOLVE EXTRACTION

OUR BIOMASS SOURCES

GREEN CHEMISTRY

>Hemi-cellulose : furfural chemistry

Sugarcane

A-Leen 5

EcoXtract

GREEN EXTRACTION

>Hydrophilic extraction

- Rosemary
- Apple
- Cinnamon
- Cistus Ladaniferus

>Lipophilic extraction

- Raspberry seeds
- Argan
- Plum press cake

PATENTED PROCESS

MINASOLVE A-LEEN 5

PIONEER OF BIOBASED PENTYLENE GLYCOL (A-LEEN 5, 2014)

PENNAKEM®
FURFURAL CHEMISTRY

MINASOLVE®
BIOINGREDIENTS
FOR COSMETIC APPLICATIONS

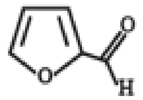
Back Integrated Raw Materials – Full Traceability

PENNAKEM®
RENEWABLE RESOURCE CHEMISTRY

« Pennakem, our US sister company is active in renewable chemistry since the 1940's »

FURFURAL APPLICATIONS

SUGAR CANE BAGASSE
RESPONSIBLE SOURCING

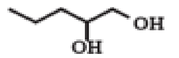


FURFURAL

- Food
- Pharma
- **Cosmetics**
- Rubber
- Agro

FURFURYL ALCOHOL PURIFIED

A-Leen 5



COSMETIC GRADE ODOURLESS

Green Chemistry

- Made from agricultural waste materials
- Sustainable & eco-responsible product
- "Green chemistry" manufacturing process

A-LEEN 5

A BIO-BASED SOLVENT WITH HIGH PRESERVATIVE AND EXTRACTING ABILITIES

INTERESTING PROPERTIES:

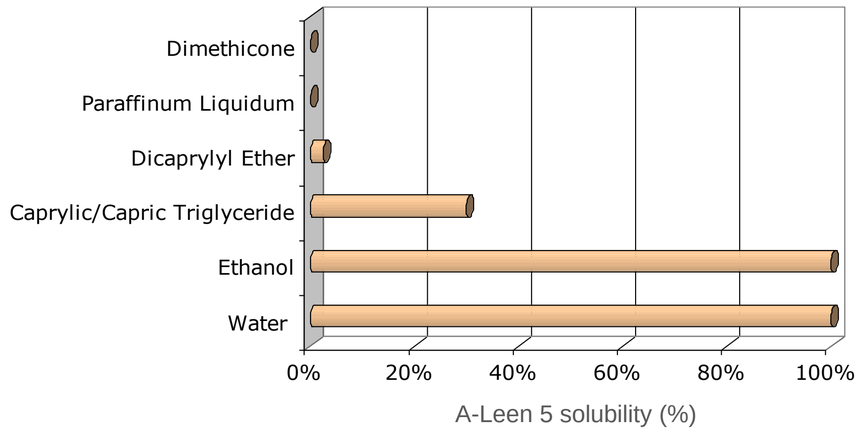
- **Fully adapted to cosmetic use:**
  - ✓ **Preservative** properties when used in water-based formulations
  - ✓ **Emollient, skin humectant, solubilizer**
  - ✓ **INCI listed product** for cosmetic application

NATRUE APPROVED

Approved by ECOCERT RAW MATERIAL
COSMOS APPROVED

USDA CERTIFIED BIOBASED PRODUCT
PRODUCT 100%

- **Interesting physico-chemical properties:**
  - ✓ **Miscible** with water and low alcohols and soluble in polar oils
  - ✓ High flash point (105°C): **safe for handling**
  - ✓ High boiling point (206°C): **low evaporation rate**
  - ✓ Density at 20°C (g/cm^3): 0.966 – 0.976



A-LEEN 5

A BIO-BASED SOLVENT WITH HIGH PRESERVATIVE AND EXTRACTING ABILITIES

INTERESTING PROPERTIES:

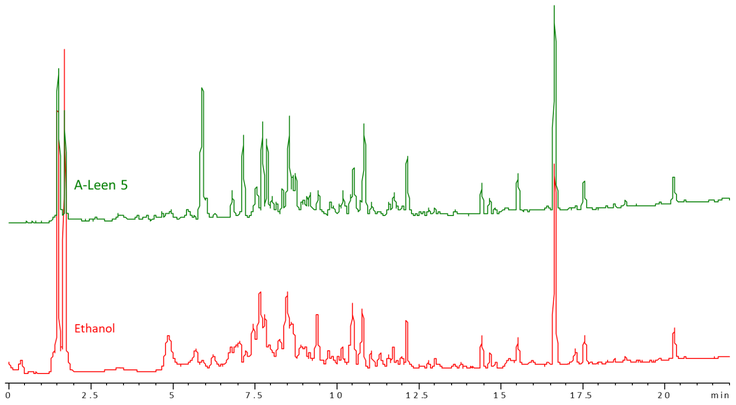
- **Extraction solvent:**

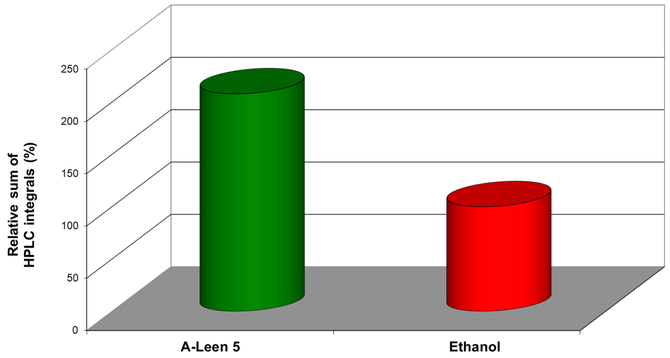
	BP	FP	Classification
1,2-Pentanediol	**206°C**	**110°C**	
MeTHF	**80°C**	**-11°C**	
Acetone	56°C	-18°C	
Cyclohexane	80°C	-18°C	
Dichloromethane	40°C	-	
Ethyl Acetate	77°C	-4°C	
Hexane	69°C	-22°C	
Iso-Propanol (IPA)	83°C	12°C	
Methanol	65°C	12°C	

- **High extractant power:**

Example. Extraction of *Chamomilla recutita* (Chamomile flower)

Protocol :
- Maceration
- Solid/liquid ratio: 1/5
- 15 min at 75-80 ° C
- HPLC analysis, RP C_{18}, UV-detection at 210 nm





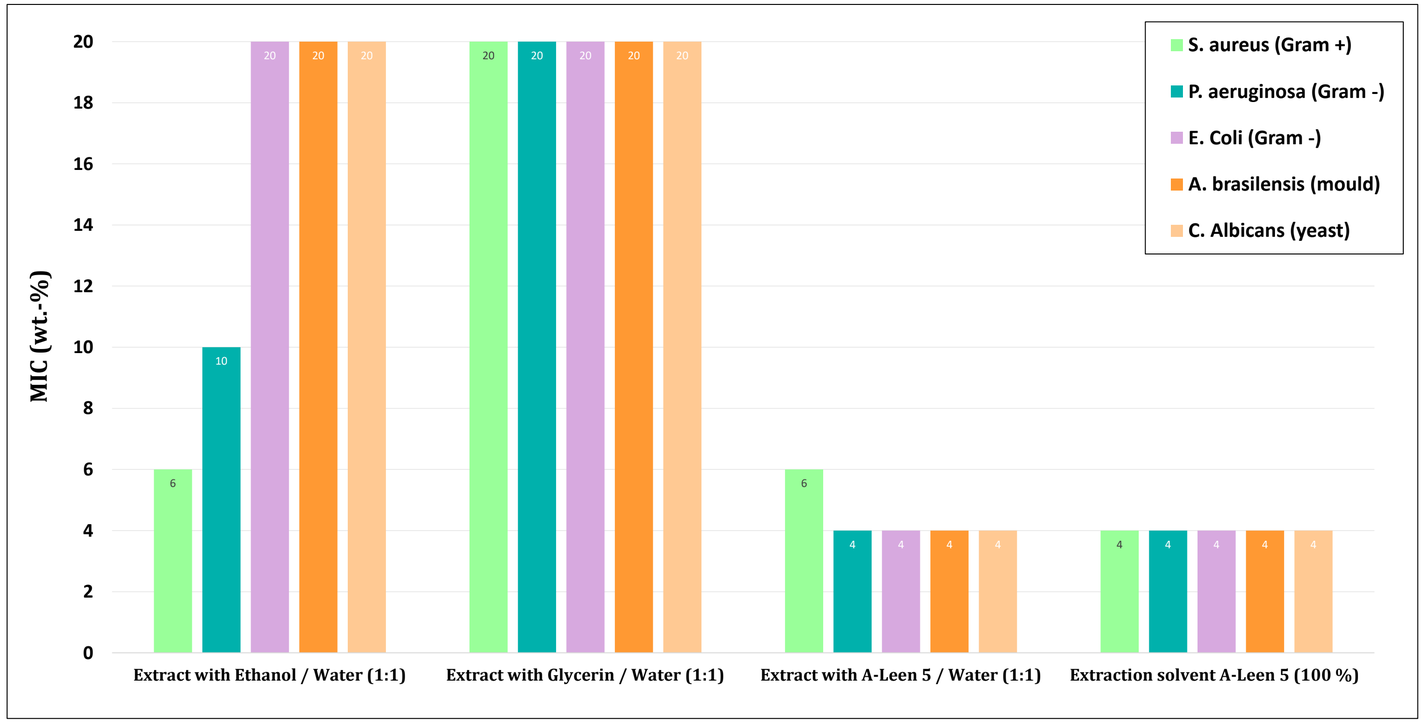
- **Extract profiles are similar**
- **Extraction yield with A-Leen 5 is higher than with Ethanol**

A-LEEN 5

A BIO-BASED SOLVENT WITH HIGH PRESERVATIVE AND EXTRACTING ABILITIES

INTERESTING PROPERTIES:

- **MIC data:**



Protocol

- The extracts obtained with various solvents were diluted with aqueous nutrient solutions, these mixtures were inoculated with different germs.
- The suspensions were incubated and the growth of the germs was visually checked for the occurrence of turbidity.
- The lowest concentration at which no turbidity occurred was designated the minimum inhibitory concentration (MIC).

➢ **Extracts with A-Leen 5 are self-preserved against microbial contamination**

INTEREST IN AGRIWASTE VALUE PROJECT

SIMILAR VALUES TOWARDS MORE NATURALITY AND SUSTAINABILITY

MINASOLVE-GREEN SOLVING ATTITUDE

Local origins to reduce carbon footprint

Diversified biomass sustainably sourced

To valorize **agriwaste** and **sustainable** origins

Partnership with **local farmers**

Perfect traceability for all ingredients involved

Green R&D developments

Innovative eco-friendly processes for higher performance and more respect for nature

Controlled processes

AGRIWASTEVALUE

Objectives:

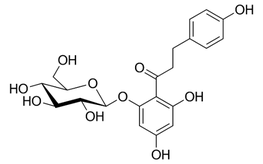
- Developing **local supply chains**
- Valorizing **agricultural residues**

Apple culture

Estimation of total of metric tons available in Europe:

Residues of apple
232195.6 T

- Obtaining **organic, sustainable and local bioactive compounds**



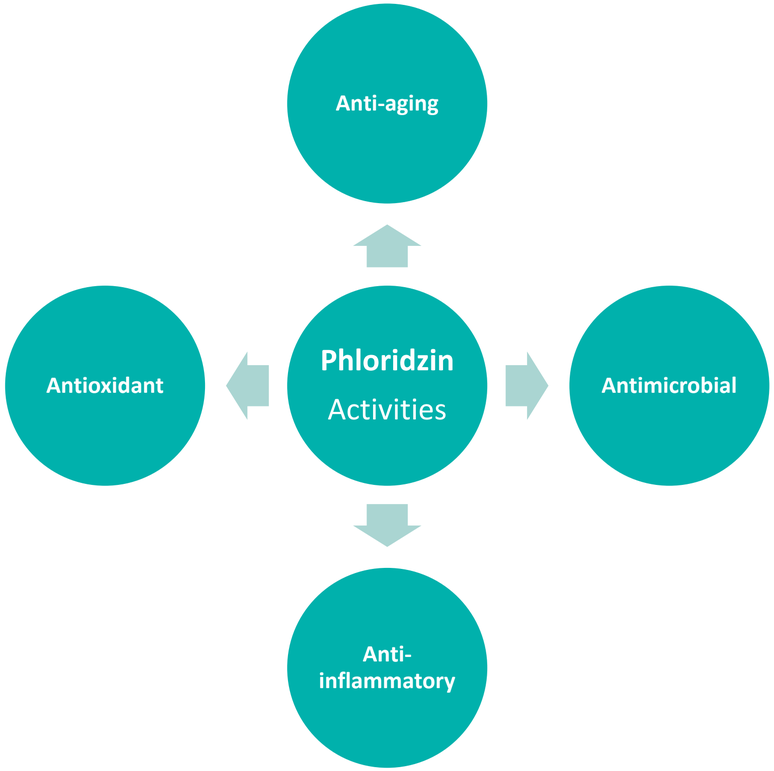
Phloridzin

INTEREST IN AGRIWASTE VALUE PROJECT

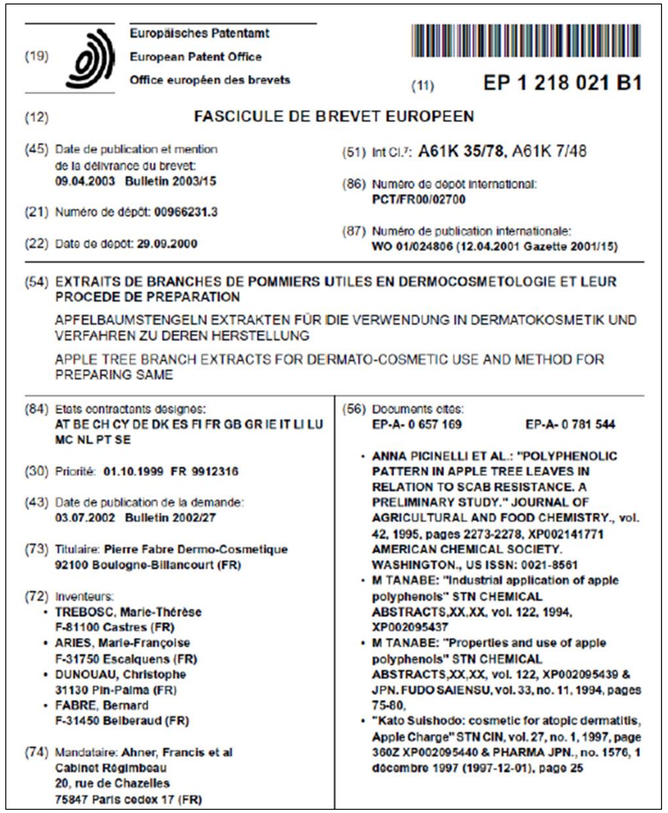
Interreg North-West Europe AGRIWASTEVALUE
European Regional Development Fund

PHLORIDZIN: ACTIVE MOLECULE WITH GREAT INTEREST FOR COSMETIC USE

➢ Interesting biological activities:



➢ Interest from industrials:



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(54) EXTRAITS DE BRANCHES DE POMMIERS UTILES EN DERMOCOSMETOLOGIE ET LEUR PROCEDE DE PREPARATION

APFELBAUMSTENGELN EXTRAKTEN FÜR DIE VERWENDUNG IN DERMATOKOSMETIK UND VERFAHREN ZU DEREN HERSTELLUNG

APPLE TREE BRANCH EXTRACTS FOR DERMATO-COSMETIC USE AND METHOD FOR PREPARING SAME

(84) Etats contractants désignés:
AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE

(56) Documents cités:
EP-A- 0 657 169 EP-A- 0 781 544

- ANNA PICINELLI ET AL.: "POLYPHENOLIC PATTERN IN APPLE TREE LEAVES IN RELATION TO SCAB RESISTANCE. A PRELIMINARY STUDY." JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY., vol. 42, 1995, pages 2273-2278, XP002141771 AMERICAN CHEMICAL SOCIETY. WASHINGTON., US ISSN: 0021-8561
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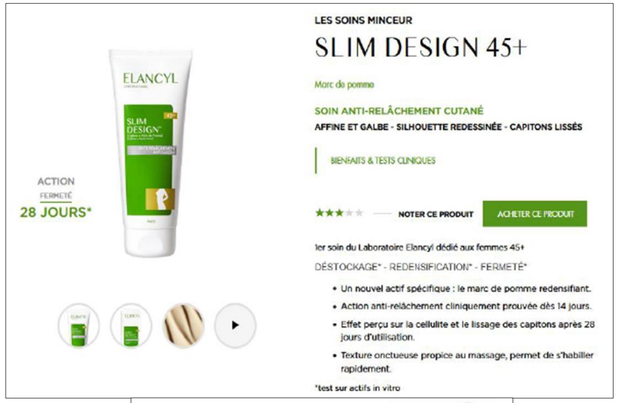
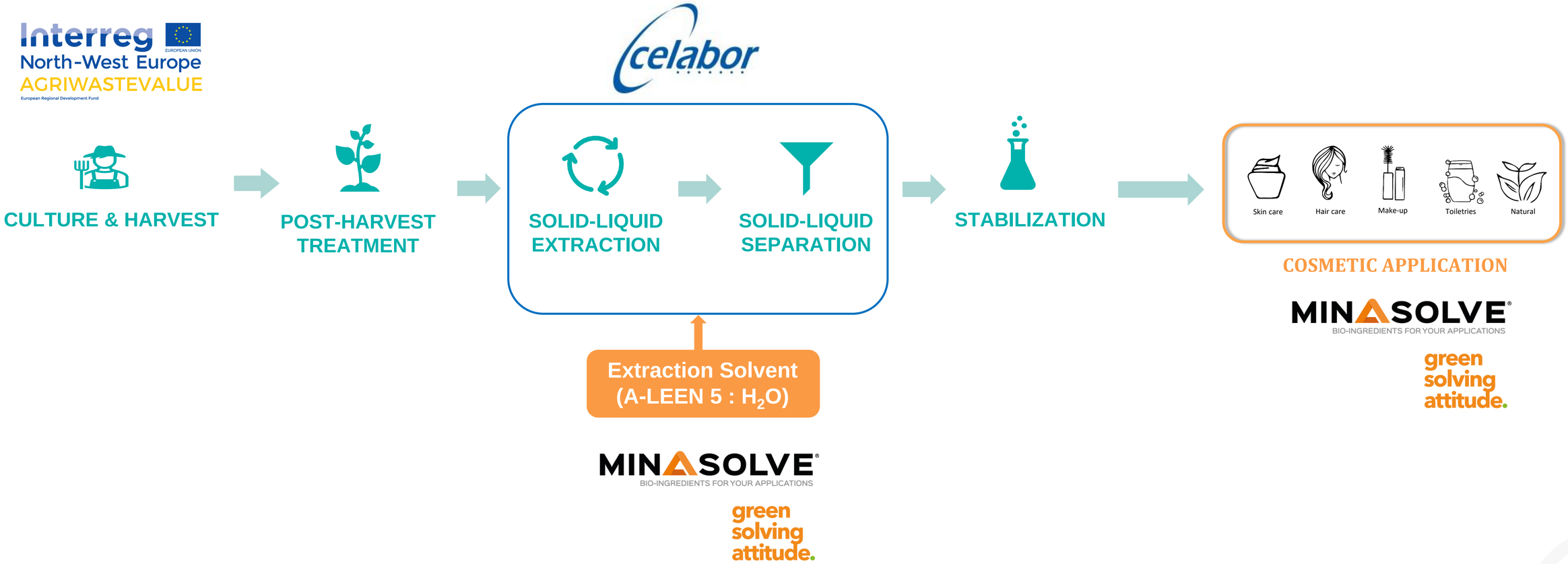


A-LEEN 5 :

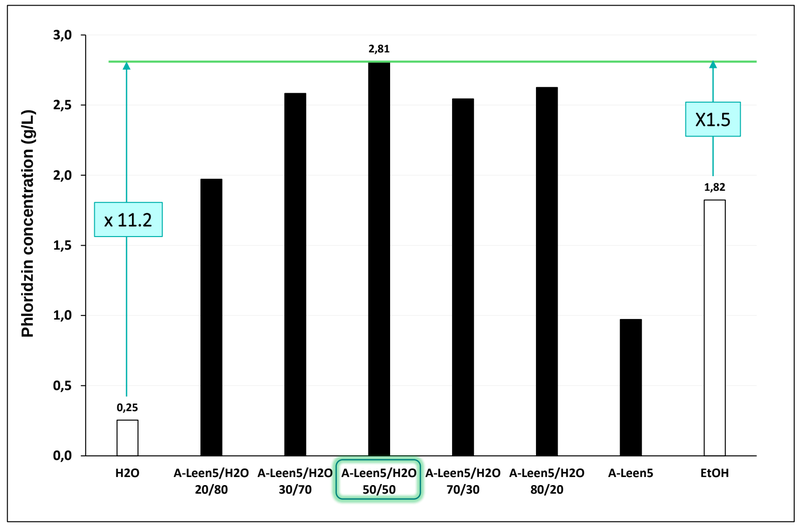
PHLORIDZIN EXTRACTION FROM APPLE RESIDUE



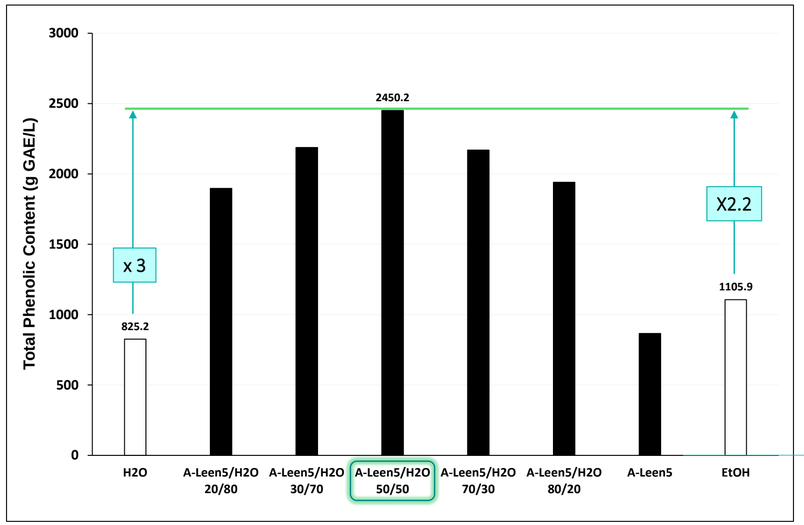
PHLORIDZIN EXTRACTION FROM APPLE RESIDUE

HIGH EXTRACTION PERFORMANCE USING (A-LEEN 5: H_2O) MIXTURE

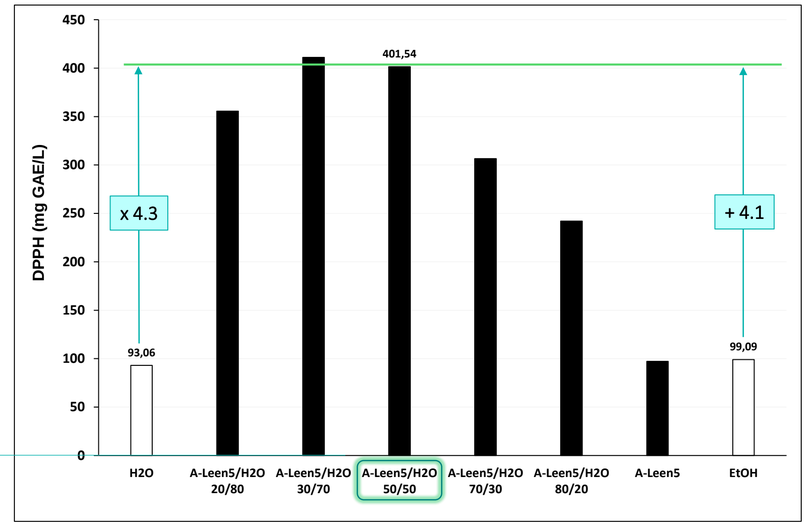
- **Phloridzin extraction performance**



- **Extraction performance of total polyphenols**



- **Antioxidant power**



(A-Leen 5:H_2O) (50:50): **high extractant ability** of **phloridzin** and **total polyphenols**

➔ **Interesting antioxidant power** compared to both aqueous and ethanolic extracts

PHLORIDZIN FOR COSMETIC USE

PERSPECTIVES

CONCLUSION
A-LEEN5 / PHLORIDZIN

- Sourcing Origin
  - Local valuable sourcing / Waste valorization
- Technical Collaboration
  - Process optimization / Performances
- Opportunities
  - New active molecule from 100% Up-Cycled process
  - Duplicate the model to new biomass…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THANK YOU FOR YOUR ATTENTION

Interreg
North-West Europe
AgriWasteValue
European Regional Development Fund

AgriWasteValue

THANK YOU !