

BIOACTIVE-NET

Assessment and dissemination of strategies for the extraction of bioactive compounds from tomato, olive and grape processing residues

Project No. FOOD-CT-2006-43035

CibusTec - Fiera di Parma,

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BIOACTIVE-NET

"*Assessment and dissemination of strategies for the extraction of bioactive compounds from tomato, olives and grape processing residues*"

SPECIFIC SUPPORT ACTION (SSA) - European Commission (6FP)

TTZ, TCA, AINIA

AMITOM, ANFOVI, CCAE, PEZA UNION, VIGNAIOLI PIEMONTESI

Duration: ***24 months***

Start date: 1st November 2006

End date: 31st October 2008

Total eligible costs: 589.354,48 €

OBJECTIVES

- Create a board Information Platform

- Implement dissemination workshops in the South European Countries

- Strengthen the European market on natural ingredients

- Increase competitiveness of the European food industry

- Increase use of bioactive compounds in the European diet

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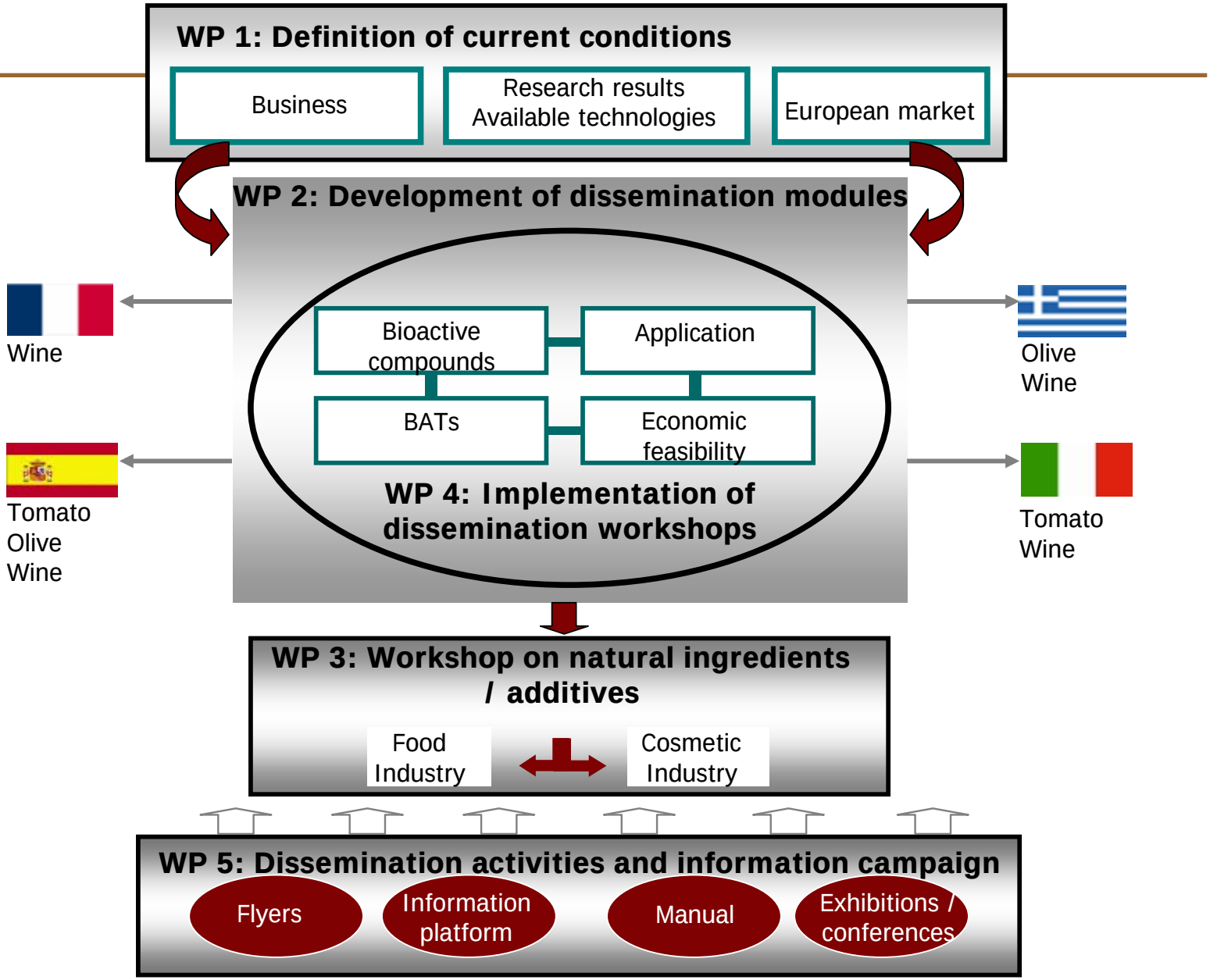

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Bioactive compounds in tomato processing solid wastes

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Tomato processing

- The processing of tomato consist of **different treatment of the tomato fruit** depending on the final product desired:
  - Tomato puree
  - Concentrate
  - Tomato Pulp
  - Whole Peeled Tomatoes
  - Tomato sauces
  - Ketchup
  - Tomato juice
  - Dehydrated tomato

Source: www.erikarathje.ca

Source: www.alibaba.com

Source: www.hormel.com

- During all these process steps, **solid wastes are produced**.

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Tomato processing wastes

- The tomato processing generates following wastes:

peels **seeds** **Waste water**

(2% in weight of fresh tomato)

POMACE

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Tomato processing wastes:

Pomace

- In Italy
- In Spain

tomatoes processed yearly

4.400.000 tons | 1.650.000 tons

99.440 tons (yield=0,0226)*

47.190 tons (yield=0,0286)*

pomace produced yearly

*ton pomace /ton tomato

Source: BIOACTIVE-NET questionnaires

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Tomato processing wastes:

Current destination of the residue

- The by-products produced during the tomato transformation process are defined as **Secondary Raw Materials**

- **Council Directive 96/25/EC**, legislate the **re-use** in particular of “tomato pulp obtained by pressing tomatoes Solanum lycopersicum Karst. during the production of tomato juice” **for animal feeding**

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Tomato processing wastes:

Current destination of the residue

- **Is it possible to extract more added value from the tomato processing residues?**

- **Can the health benefits from tomato be obtained from tomato wastes?**

The solution may be the bioactive substances present in tomatoes solid wastes…

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Bioactive compounds

What is a bioactive compound?

A compound is considered bioactive if it has **interaction with** or **effect on** any **cell tissue in the human body**

The effect might be beneficial or adverse.

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Bioactive compounds

The level of bioactive compounds is affected by:

- **Fruit variety**
- **Ripening stage**
- **Agronomic conditions**
- **Post-harvest manipulation**
- **Processing**

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Bioactive compounds in tomato

The main bioactive compounds present in tomato processing residues are:

Lycopene

Enzymes

Tomato fibre

Tomato seed oil

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Lycopene:

	Amount of extractable molecule	*Metabolic known effects*	*Sector of interest*
Lycopene (the most important tomato carotenoid)	From 5 to 50 mg/kg (even if it is possible, through the genetic improvement, to obtain as far as 200 mg/kg of Lycopene)	Antioxidant effect Protection against UV induced damage Protection against degenerative diseases Decrease cardiovascular diseases risk Immune-stimulant effect Cancer risk reduction (e.g. prostate)	High quality food **integrator** (additive, antioxidant and **colouring E160d**) Pharmaceutical product Cosmetic product (**skin maintaining**)

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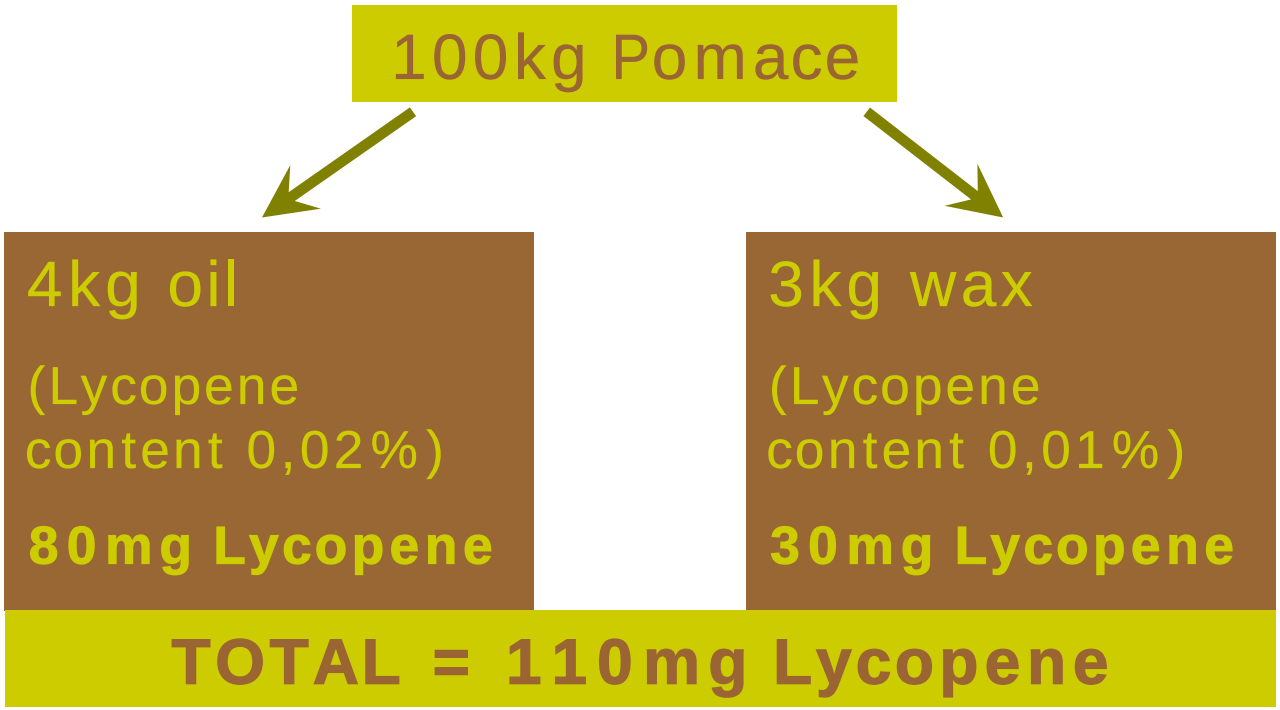
Lycopene:

Availability in tomato processing residues



*Yields obtained during the TOM project, using Super Critical Fluid extraction

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Tomato fibres:

	Amount of extractable molecule	***Metabolic known effects***	***Sector of interest***
Tomato Fibres	Not defined	- Positive effects during the mastication mechanisms - Reduce the caloric contribution of foods - Induce satiety sensation - Reduce the glycemia - Reduce the cholesterol - Tie the toxic substances - Stimulate the digestive processes - Increase the time of intestinal transit - Favour the fermentative processes in the colon	Food integrator (as additives or in dietetic foods) Food industry: as viscosity modifier (in soup and sauces)

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Tomato fibres:

Availability in tomato processing residues

100kg Pomace

↓

75kg Tomato fibre

*Yields obtained during the TOM project, using Super Critical Fluid extraction

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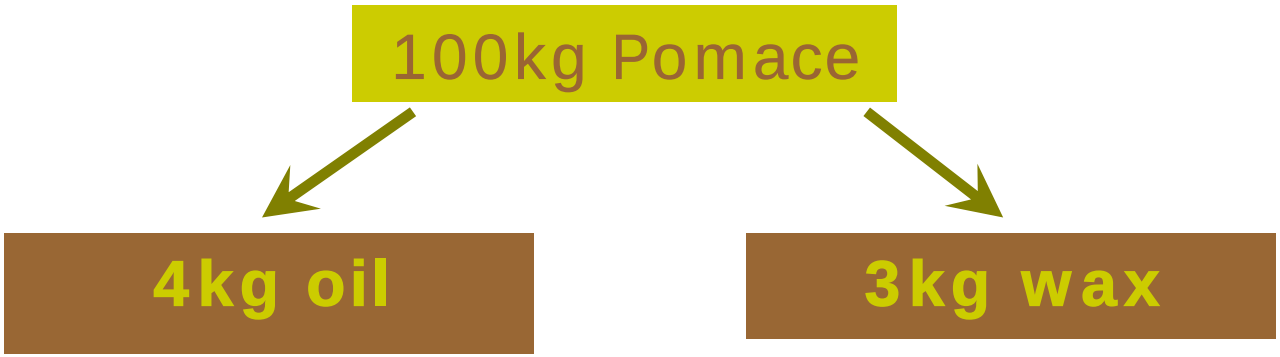
Tomato seed oil:

Availability in tomato processing residues



- **Vegetable fats and oils** are substances derived from plants that are composed of triglycerides. Nominally, oils are liquid at room temperature, and fats are solid; a dense brittle fat is called a wax.

*Yields obtained during the TOM project, using Super Critical Fluid extraction

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Tomato seed oil:

- 75% of unsaturated fatty acids
- good source of the essential linoleic fatty acid.

Metabolic known effects:

- Tomato seeds oil acts as:
  - -vascular protector
  - -emollient

Applications:

- Food additive rich in poly-unsatured fatty acids
- cosmetic additive

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Enzymes:

	Amount of extractable molecule	***Metabolic known effects***	***Sector of interest***
Enzymes:	Not defined		Food industry applications
Pectin Methyl Esterase		Aggregating for vegetables	
Polygalacturonase		Disgregating of pectines	

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Bioactive compounds in olive processing wastes

Source: www.juntadeandalucia.es

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Olive Processing:

Olive oil production

- Olive oil production is divided into three activity fields:
  - -Oil mills
  - -Refineries
  - -Plants where the oil cake is processed

- During all the processing steps, **wastes are produced**.

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Olive processing wastes

- The wastes obtained from the olive oil milling process are:

Alperujo

Source: www.juntadeandalucia.es

Oil washing water

Pomace

Source: www.kangarooblue.com

alpechin

Source: www.unex.es

Waste water

Source: www.alabastercorp.com

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Olive processing wastes:

Source: BIOACTIVE-NET questionnaires

355.000 tons olive oil produced in **Greece** yearly

81.650 tons pomace produced yearly

(yield of 0,23t pomace / t tomato)

177.500 tons alpechin produced yearly

(yield of 0,5t margine / t tomato)

600.000 tons olive oil produced in **Spain** yearly

408.000 tons pomace produced yearly

(yield of 0,68t pomace / t tomato)

270.000 tons alpeorujo produced yearly

(yield of 0,45t alpeorujo / t tomato)

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Olive processing wastes:

Current destination of the wastes

- General EU Legislation on wastes:

Directive 2006/12/EC

-Member States shall take the necessary measures to **ensure that waste is recovered or disposed of without endangering human health** and without using processes or methods which could harm the environment.

-Such directive is **applicable to any substance** or which the holder discards or intends or is required to discard.

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Bioactive compounds in Olive

The main bioactive compounds present in olive processing residues are:

Polyphenols

Hydroxytyrosol

Oleuropein

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Polyphenols:

Description

- subdivided into tannins, and phenylpropanoids such as lignins and flavonoids

- Present in berries, tea, beer, wine, olive oil, chocolate/cocoa, walnuts, peanuts, yerba mate, and other fruits and vegetables (high levels in fruit skins)

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Polyphenols:

Metabolic known effect

- antioxidant characteristics
- reduce the risk of cardiovascular disease and cancer

Applications

- cosmetic industry
- health food products (biscuits, bakery products, dietetic products)
- nutraceuticals

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Hydroxytyrosol and Oleuropein:

	Amount of extractable molecule	***Metabolic known effects***	***Sector of interest***
Hydroxytyrosol (polyphenol)	Polyphenols concentrations in oil oscillate from 100 to 1000 mg/kg	Antioxidant and inhibiting towards pro-inflammatory oxygenasic enzymes	Food (as natural additives alternatively to synthesis compounds), chemical, pharmaceutical and cosmetic industries
Oleuropein (Polyphenol)		Antioxidant and antibacterial properties	Food, chemical, pharmaceutical and cosmetic industries

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Bioactive compounds in grape processing wastes

Source: www.ace76.blogia.com

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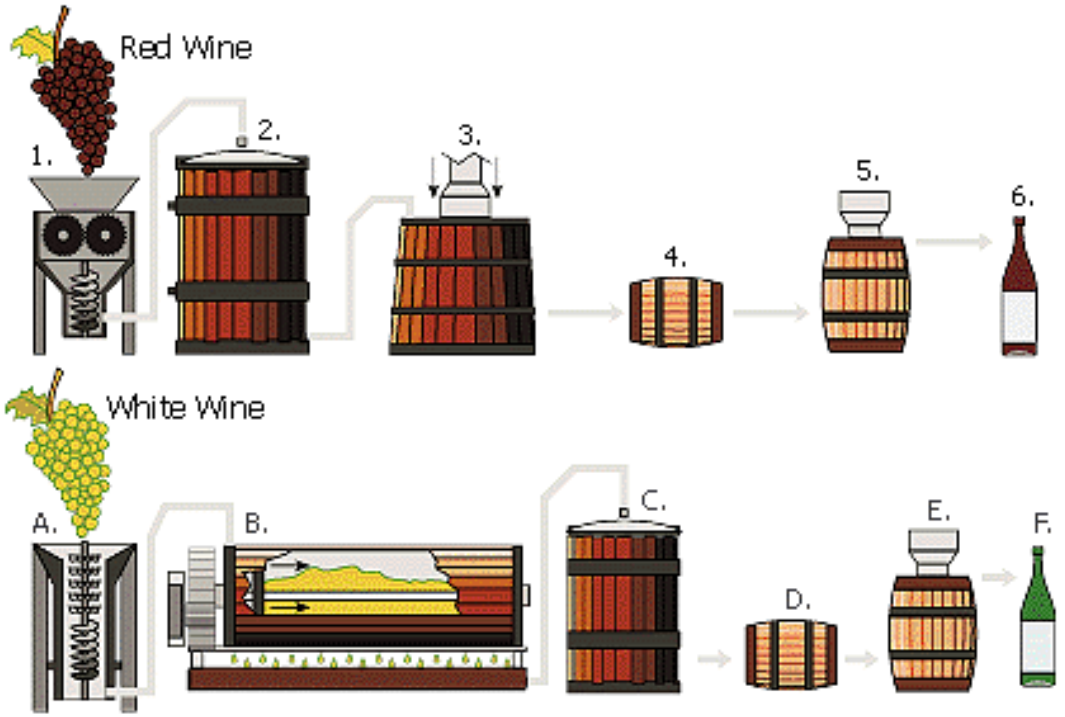
Grape Processing:

Wine production

- Wastes are produced during all the production steps of wine production !



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Grape processing residues

Source: www.cibercentros.jcyl.es

Grape stems

(MASHING)

Waste water

(WASHING of the equipment)

Pomace

(PRESSING)

Lees

(FERMENTATION)

Filter cake

(CLARIFICATION)

- The solid components form the **dregs of grapes**.

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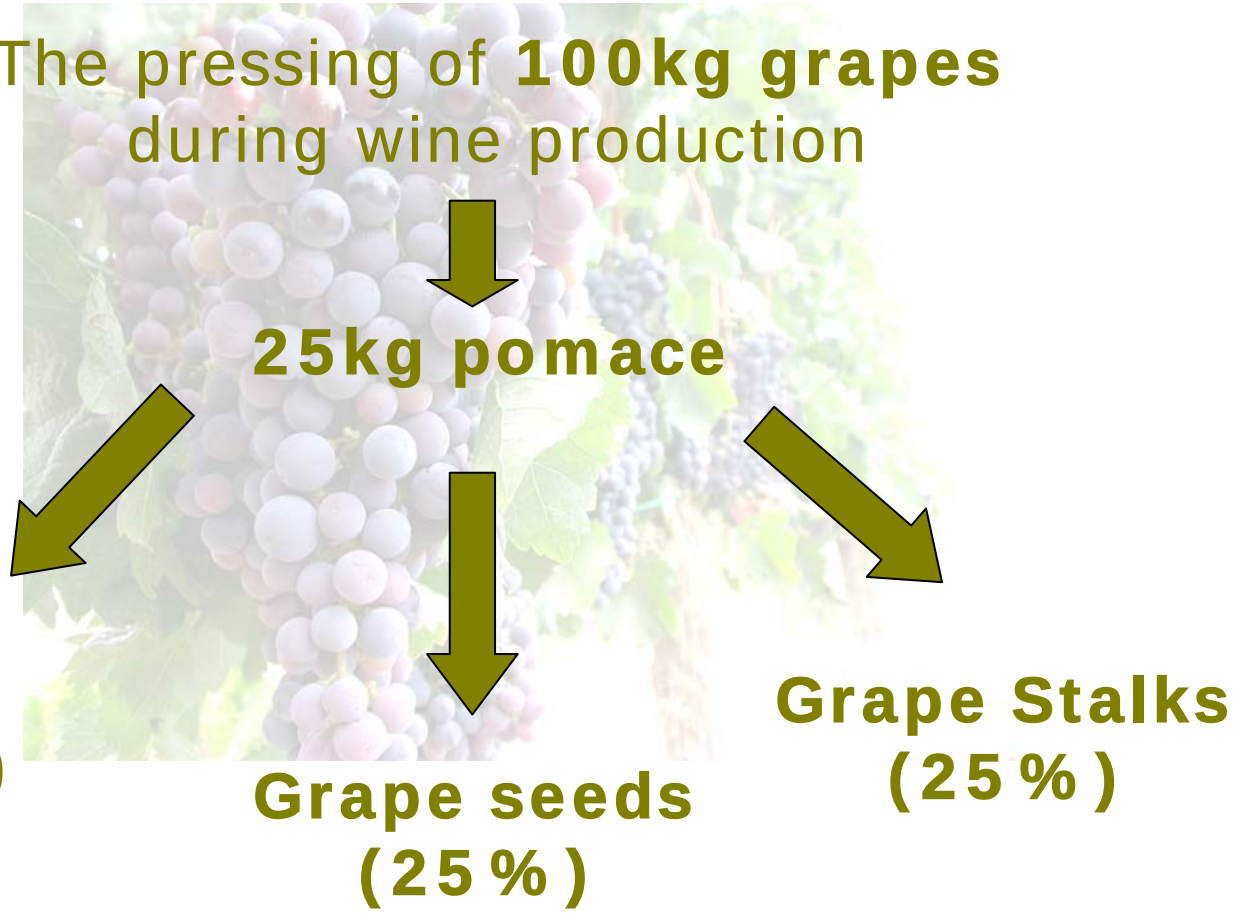
Grape processing wastes



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Grape processing wastes:

Source: BIOACTIVE-NET questionnaires

GRAPE STEMS produced yearly			
570.000 tons	90.133 tons	259.000 tons	206.800 tons

Grapes processed yearly for the production of wine			
France	Greece	Italy	Spain
7.605.000 tons	521.000 tons	7.400.000 tons	4.700.000 tons

1.400.000 tons	88.000 tons	740.000 tons	409.000 tons
POMACE produced yearly			

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Grape processing wastes:

Filter cake produced yearly

68.445 tons	24.000 tons	133.200 tons	42.300 tons

Grapes processed yearly for the production of wine

France	Greece	Italy	Spain
7.605.000 tons	521.000 tons	7.400.000 tons	4.700.000 tons

335.000 tons	27.000 tons	392.000 tons	517.000 tons

Lees produced yearly

Source: BIOACTIVE-NET questionnaires

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Grape processing wastes:

Current destination of the wastes

- General EU Legislation on wastes:
Directive 2006/12/EC

-Member States shall take the necessary measures to **ensure that waste is recovered or disposed of without endangering human health** and without using processes or methods which could harm the environment.

-Such directive is **applicable to any substance** or which the holder discards or intends or is required to discard.

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Grape processing wastes:

Current destination of the wastes

- **Council Regulation EC/1493/1999**. Article 27

  -Any natural or legal person or group of persons having made wine, shall be required to **deliver for distillation all the by-products of that winemaking** (grapes and pressing of wine lees).

  -The market foresees compulsory distillation of by-products of wine-making.

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Bioactive compounds in grape

The most important bioactive compounds present in the grapes' dregs are:

- Grape seed oil
- Resveratrol
- Quercetin
- Anthocyanins
- Proanthocyanidins

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Grape bio-components

	Amount of extractable molecule	***Metabolic known effects***	***Sector of interest***
Stilbens e.g. Resveratrol (the most important grape polyphenol)	Not defined	Cardiovascular system protection Antioxidant action and photoprotective effect Anticancer action Against Alzheimer	Cosmetic use: - in combination with normal ultraviolet filters as photoprotective effect; - against skin spots As integrator
Anthocyanins (flavonoids)	Not defined	Antioxidant and anti-inflammatory action Against cancer	Cosmetic use (e.g. skin protection after sun-bathe)
Proanthocyanidins	Not defined	Antioxidant activity (very used for skin health)	Cosmetic use: anti-wrinkles Ocular protection (retina)

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Grape bio-components

	Amount of extractable molecule	***Metabolic known effects***	***Sector of interest***
Quercetin (flavonoids)	Not defined	Anti-inflammatory activity Antioxidant activitiy and vitamin C-sparging action May have positive effects in combating or helping to prevent: cancer, prostatitis, heart disease, cataracts, allergies/inflammations, respiratory diseases such as bronchitis and asthma	Mainly used as a food integrator

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From a legislative point of view, natural ingredients are regulated as:
-Food additives (Directive 89/107/EEC) and/or
-Cosmetic products (Council Directive 76/768)

Global market of functional food
= 33 billion US$ (for Europe = 2 billion US$)

Functional food (or nutraceuticals) is any food claimed to have a health-promoting and/or disease-preventing property beyond the basic nutritional function of supplying nutrients.

Global market for “nutricosmetics”
= $1bn (will double over the next five years)

Nutricosmetics are supplements aimed at outward appearance

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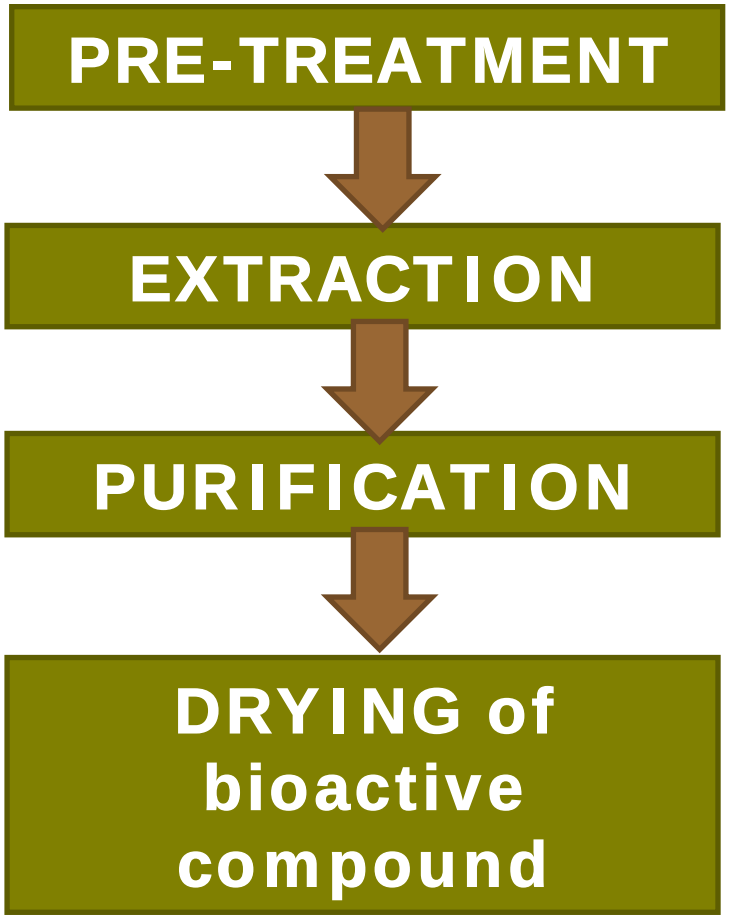
Obtaining of bioactive compounds out of tomato / olive / grape processing wastes

To store and transport the raw material easier

To prepare the raw material for the extraction

To obtain a product with greater purity and therefore greater value added

To store easily the bioactive compounds



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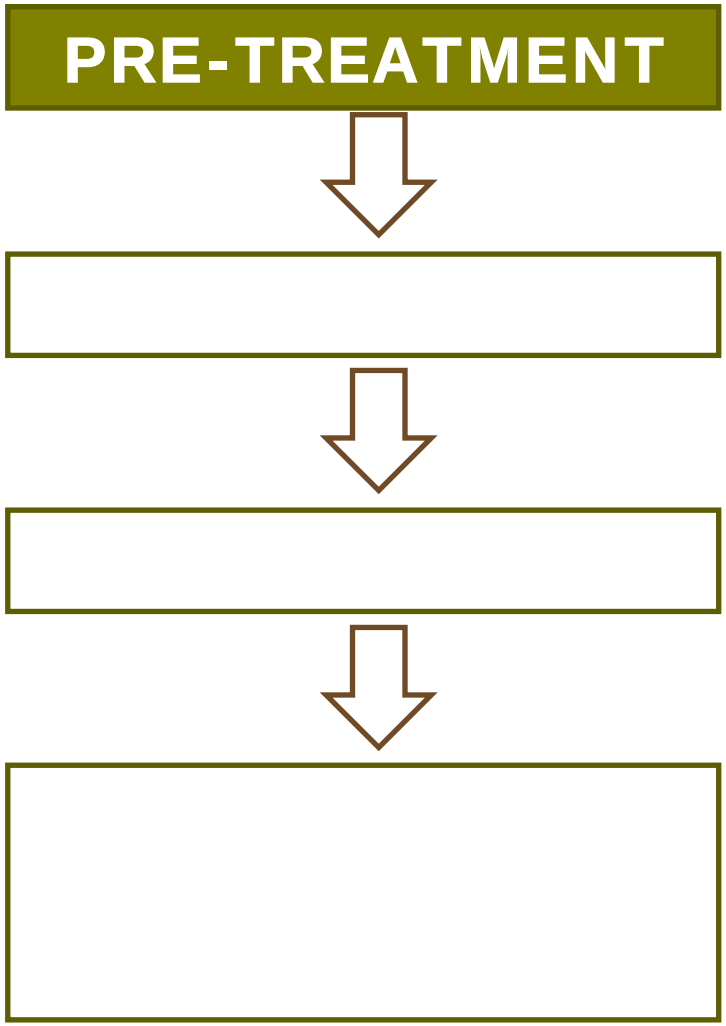
Obtaining of bioactive compounds out of tomato / olive / grape processing wastes

- Draining:
to eliminate a first part of the important amount of water present in the residues
(Tomato pomace ~70% moisture, Olive pomace ~55% moisture, Grape pomace ~50% moisture)

- Drying:
to achieve a moisture of 10% in the residues

- Milling:
to have a common particle size of 2mm

- Homogenization:
to have the same composition throughout the volume to treat



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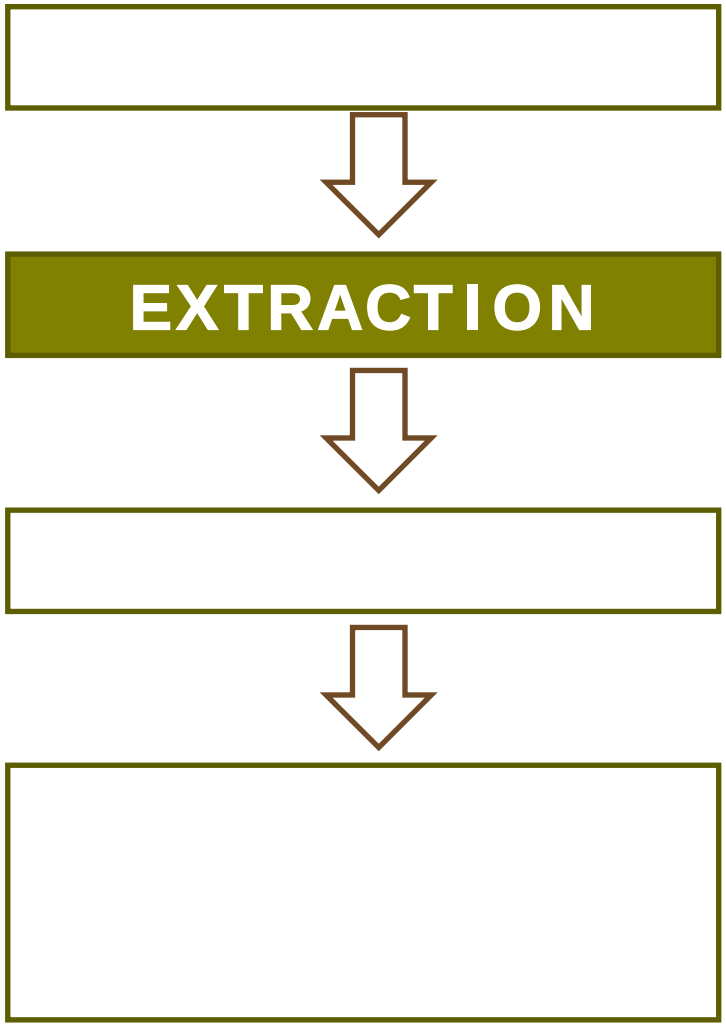
Obtaining of bioactive compounds out of tomato / olive / grape processing wastes

- Conventional solid-liquid extraction
- Supercritical Fluid Extraction (SFE)/SC-CO2 extraction
- Sonicated-assisted extraction (ultrasound)
- Microwave-assisted extraction (MAE)
- Accelerated solvent extraction (ASE)



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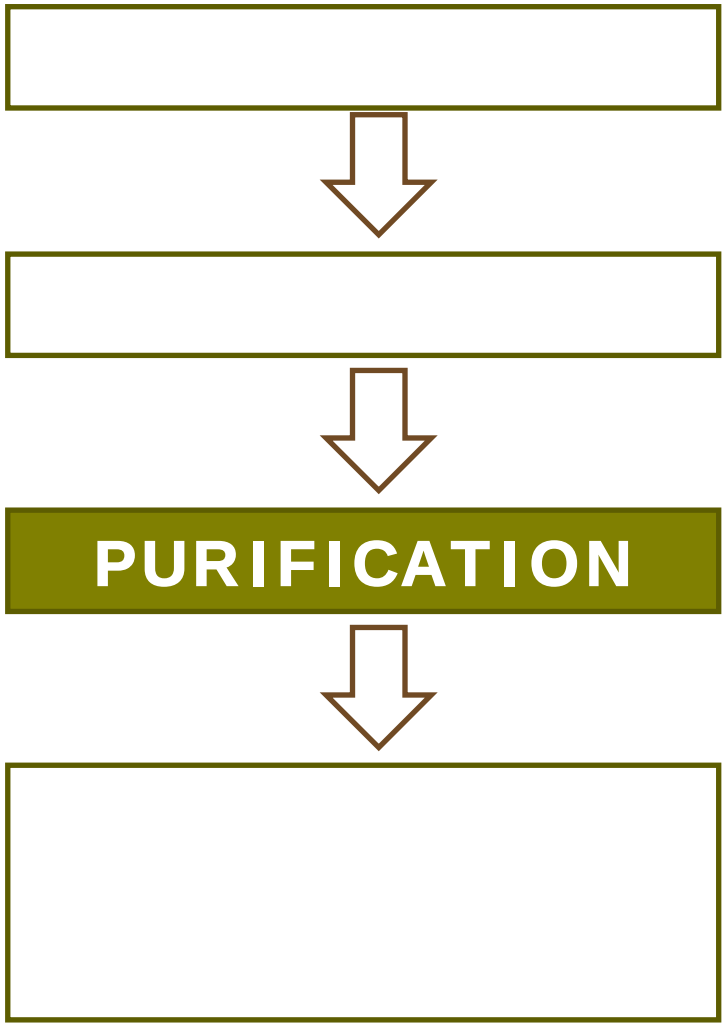
Obtaining of bioactive compounds out of tomato / olive / grape processing wastes

- Chromatographic techniques
  - ✓ partition chromatography
  - ✓ adsorption chromatography
  - ✓ size-exclusion or gel filtration
  - ✓ ion exchange chromatography
  - ✓ affinity chromatography
- Membranes
  - ✓ microfiltration
  - ✓ ultrafiltration
  - ✓ reverse osmosis
- Crystallization



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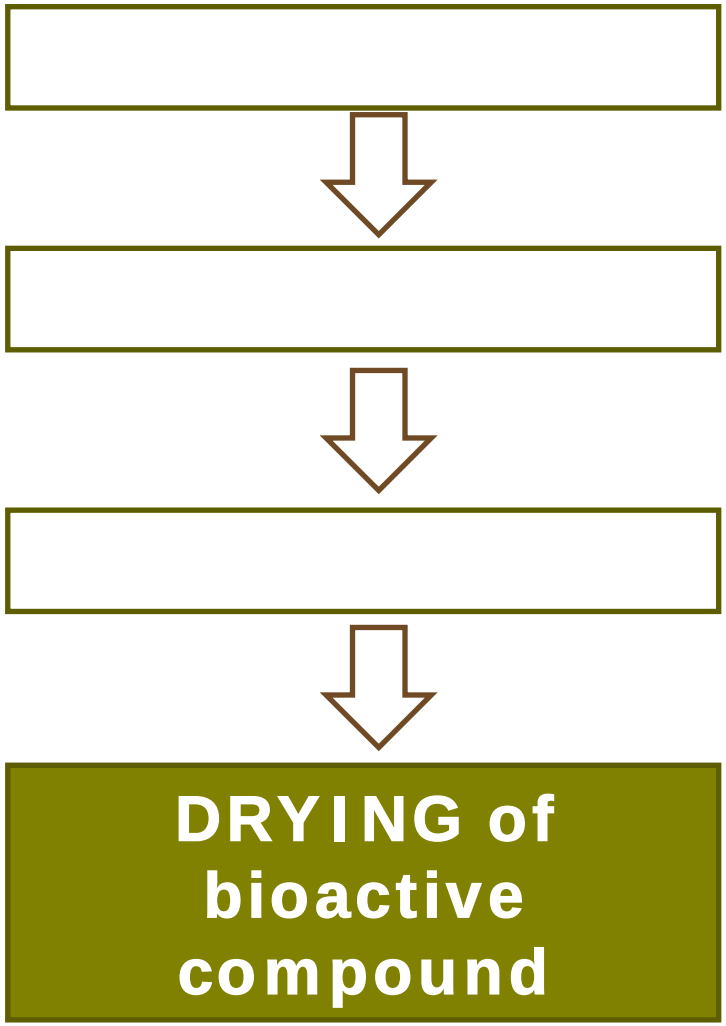
Obtaining of bioactive compounds out of tomato / olive / grape processing wastes

- Freeze Drying
- Spray drying
- Rotary Vacuum drying



- Cost calculation examples for the industrial extraction of bioactive compounds from tomato/olive/grape processing wastes:

  - **Example 1:**
    Supercritical Fluid Extraction (SFE) /SC-CO_2 extraction

  - **Example 2:**
    Solvent extraction

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Assessment of the economic feasibility of the extraction of bioactive compounds from tomato / olive / grape processing wastes

	Tomato (quantities yearly produced)	**Olives** (quantities yearly produced)	**Grape** (quantities yearly produced)
Minimum hypothesis	2000 tons	500 tons	75 tons
Maximum hypothesis	40000 tons	11000 tons	50000 tons

The minimum hypothesis for the cost calculation considers that the extraction of bioactive compounds is **performed in single factories**

The maximum hypothesis for the cost calculation considers that the extraction of bioactive compounds is **performed by a big extractor** for the processing residues of the whole region

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Example 1:

Supercritical Fluid Extraction

Examples of SFE plants (**ainia**)

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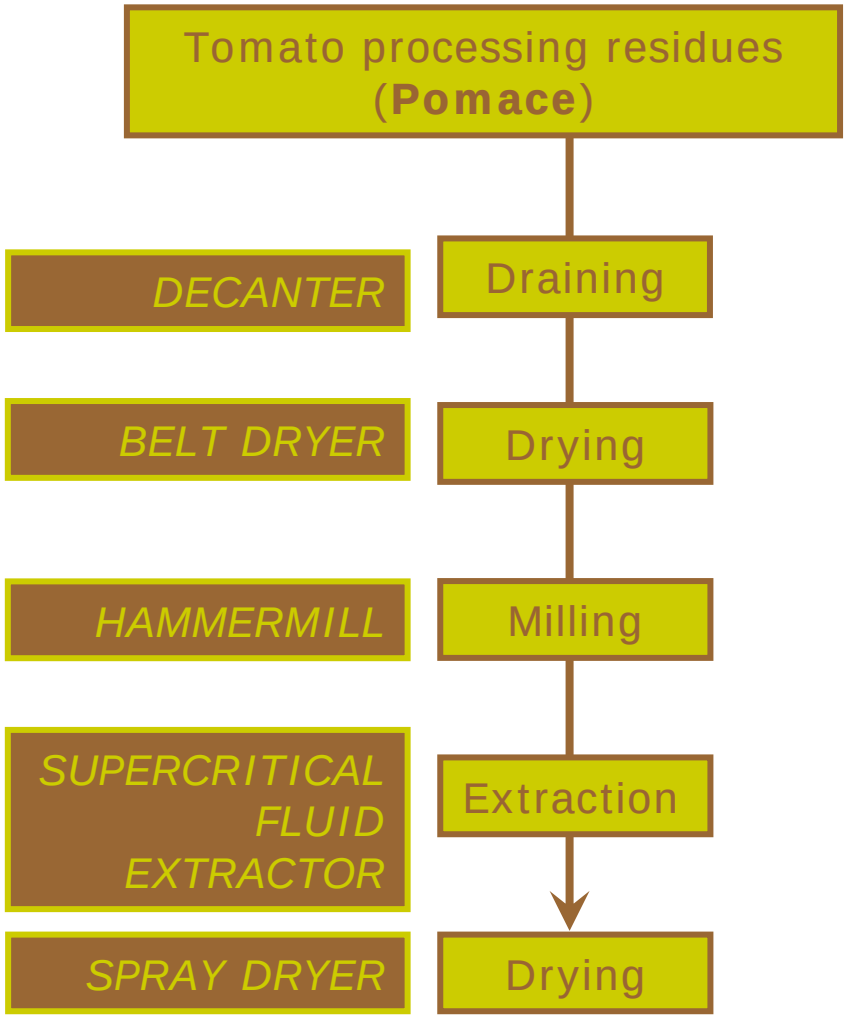
Example 1:

Supercritical Fluid Extraction on Tomato



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Example 1:

Supercritical Fluid Extraction **on Tomato**

Estimated costs:

PRE-TREATMENT

- Initial investment for equipments

-labour costs, energy costs, maintenance & repair, quality control

-EXTRACTION

Initial investment for SFE EXTRACTOR

CO2 consumption (recycling 75%)

-labour costs, energy costs, maintenance & repair, quality control

DRYING

Initial investment for equipments

-labour costs, energy costs, maintenance & repair, quality control

Other costs: supervisory control, average transport needed, raw material transport (0,25€/t/km)

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Example 1:

Supercritical Fluid Extraction **on Tomato**

– TOTAL costs -

TOTAL COSTS yearly	**Minimum Hypothesis** (2.000t/year)	**Maximum Hypothesis** (40.000t/year)
	2.625.668 €	**49.223.770 €**
Costs / kg pomace to treat	*1,31€*	*1,23€*

total initial investment	*4.690.000 €*	*101.650.000 €*

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Example 1:

Supercritical Fluid Extraction **on Tomato**

– Revenues -

Selling prices		**average**
oil 0,02% Lycopene	€/kg	5,00
wax 0,01% Lycopene	€/kg	10,00
fibre	€/kg	4,50
Extraction yield		
oil		4%
wax		3%
fibre		70%

Estimation of the revenues		**Minimum Hypothesis (2.000t/year)**	**Maximum Hypothesis (40.000t/year)**
revenues oil	€	160.000	3.200.000
revenues wax	€	240.000	4.800.000
revenues fibre	€	2.520.000	50.400.000

Estimated revenues	**€**	*2.920.000*	*58.400.000*

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Example 1:

Supercritical Fluid Extraction **on Tomato**

– YEARLY PROFIT -

	Minimum Hypothesis (2.000t/year)	**Maximum Hypothesis** (40.000t/year)
TOTAL REVENUE	2.920.000€	58.400.000€
TOTAL COSTS	2.625.668 €	49.223.770 €
PROFIT PER YEAR	**294.332€**	**9.176.230€**
total initial investment	*4.690.000 €*	*101.650.000 €*

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Example 2:

Solvent Extraction

Conventional extraction laboratory scale plant.
(http://www.pignat.com/default.aspx?idwsgpage=7&idwsglangue=2&idwsgmodulecataloguecategorie=7&idwsgmodulecatalogueproduit=64)

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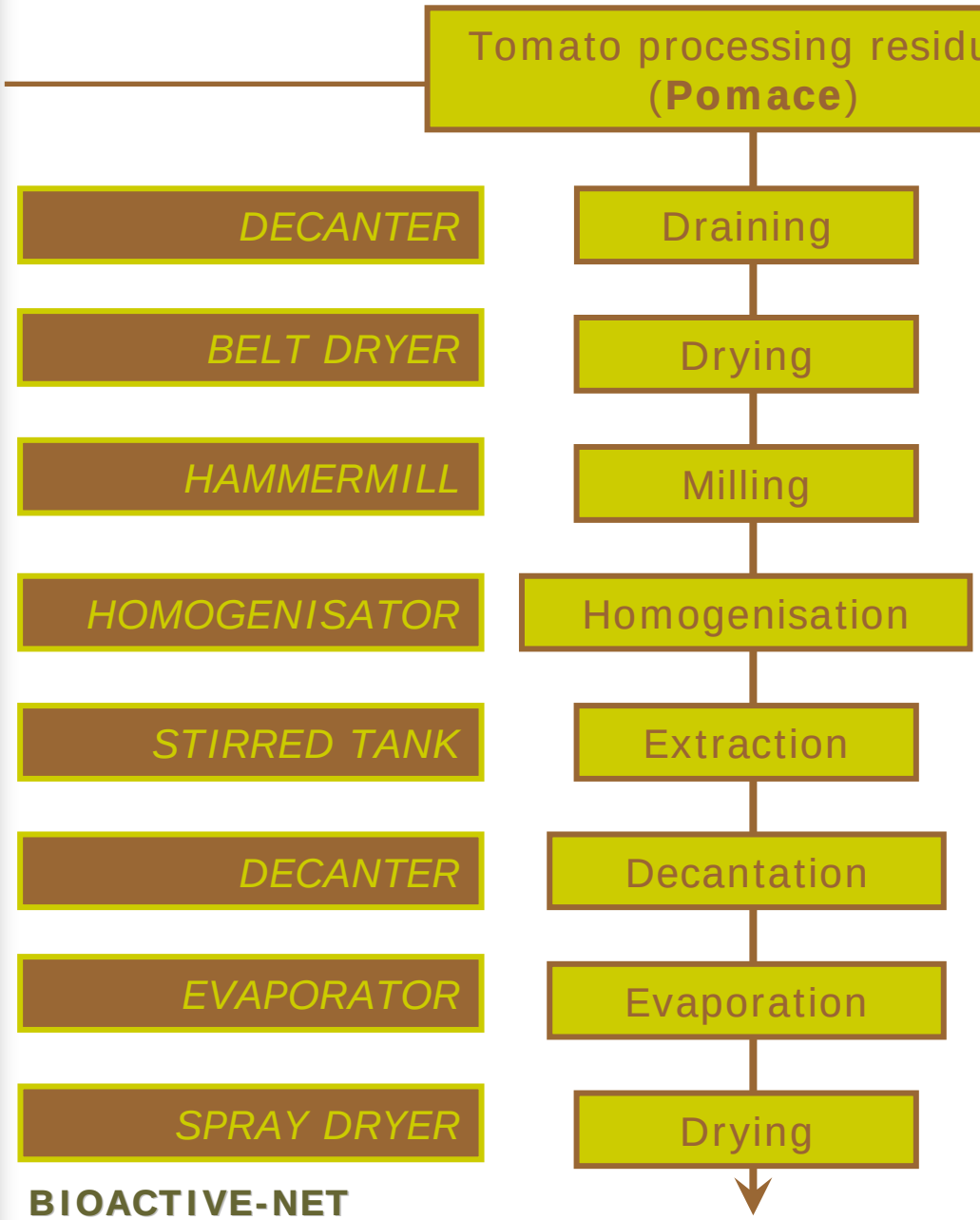
Example 2:

Solvent Extraction



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Example 2:

Solvent Extraction **on Tomato**

Estimated costs:

PRE-TREATMENT

- Initial investment for equipments

-labour costs, energy costs, maintenance & repair, quality control

EXTRACTION

- Initial investment for SFE EXTRACTOR

- CO2 consumption (recycling 75%)

-labour costs, energy costs, maintenance & repair, quality control

DRYING

- Initial investment for equipments

-labour costs, energy costs, maintenance & repair, quality control

Other costs: supervisory control, average transport needed, raw material transport

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Example 2:

Solvent Extraction **on Tomato**

– TOTAL costs -

	Minimum Hypothesis (2.000t/year)	**Maximum Hypothesis** (40.000t/year)
TOTAL COSTS yearly	**2.126.554€**	**35.765.401€**
Costs / kg pomace to treat	*1,06€*	*0,89€*
total initial investment	*1.040.000€*	*2.490.000€*

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Example 2:

Solvent Extraction **on Tomato**

– Revenues -

Selling prices		**average**
Oil price lycopene 0,1%	€/kg	45,00

Extraction yield	
oil 0,02% Lycopene	5%

Estimation of the yearly revenues		**Minimum Hypothesis** (2.000t/year)	**Maximum Hypothesis** (40.000t/year)
revenues oil	€	1.800.000	36.000.000
revenues wax	€	0	0

Estimated revenues	**€**	**1.800.000**	**36.000.000**

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Example 2:

Solvent Extraction **on Tomato**

– YEARLY PROFIT -

	Minimum Hypothesis (2.000t/year)	**Maximum Hypothesis** (40.000t/year)
TOTAL REVENUE	1.800.000€	36.000.000€
TOTAL COSTS	2.126.554€	35.765.401€
PROFIT PER YEAR	**-326.554,28€**	**234.599,12€**
total initial investment	*1.040.000€*	*2.490.000€*

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SYNTHESIS on Tomato:

		Minimum Hypothesis (2.000t/year)	Maximum Hypothesis (40.000t/year)
Supercritical Fluid Extraction	Yearly profit	294.332€	9.176.230€
	Initial investment	*4.690.000€*	*101.650.000€*
	Time needed to recover initial investment	*16 years*	*12 years*
Solvent Extraction	Yearly profit	-326.554€	234.599€
	Initial investment	*1.040.000€*	*2.490.000€*
	Time needed to recover initial investment		*10 years*

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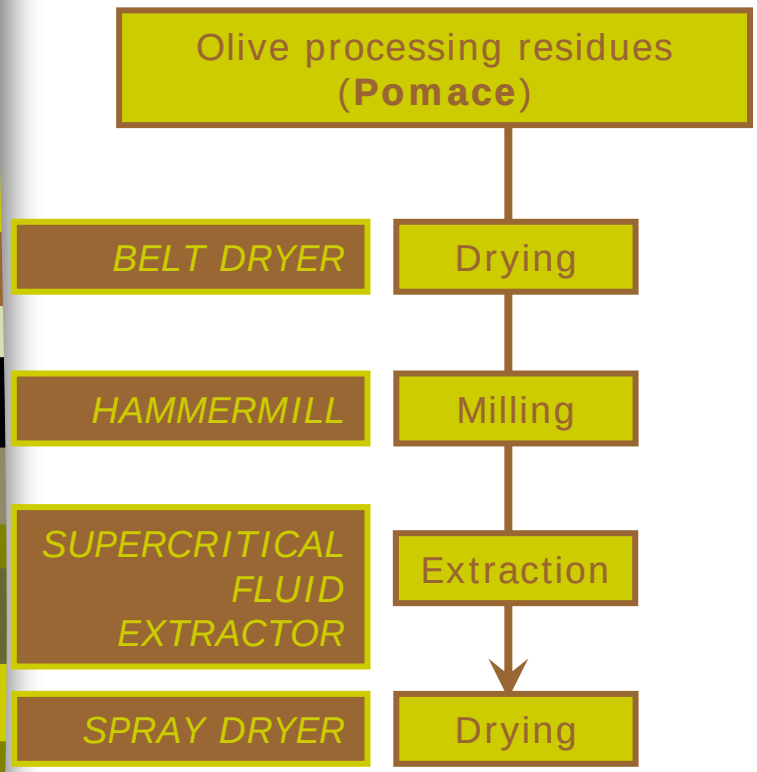
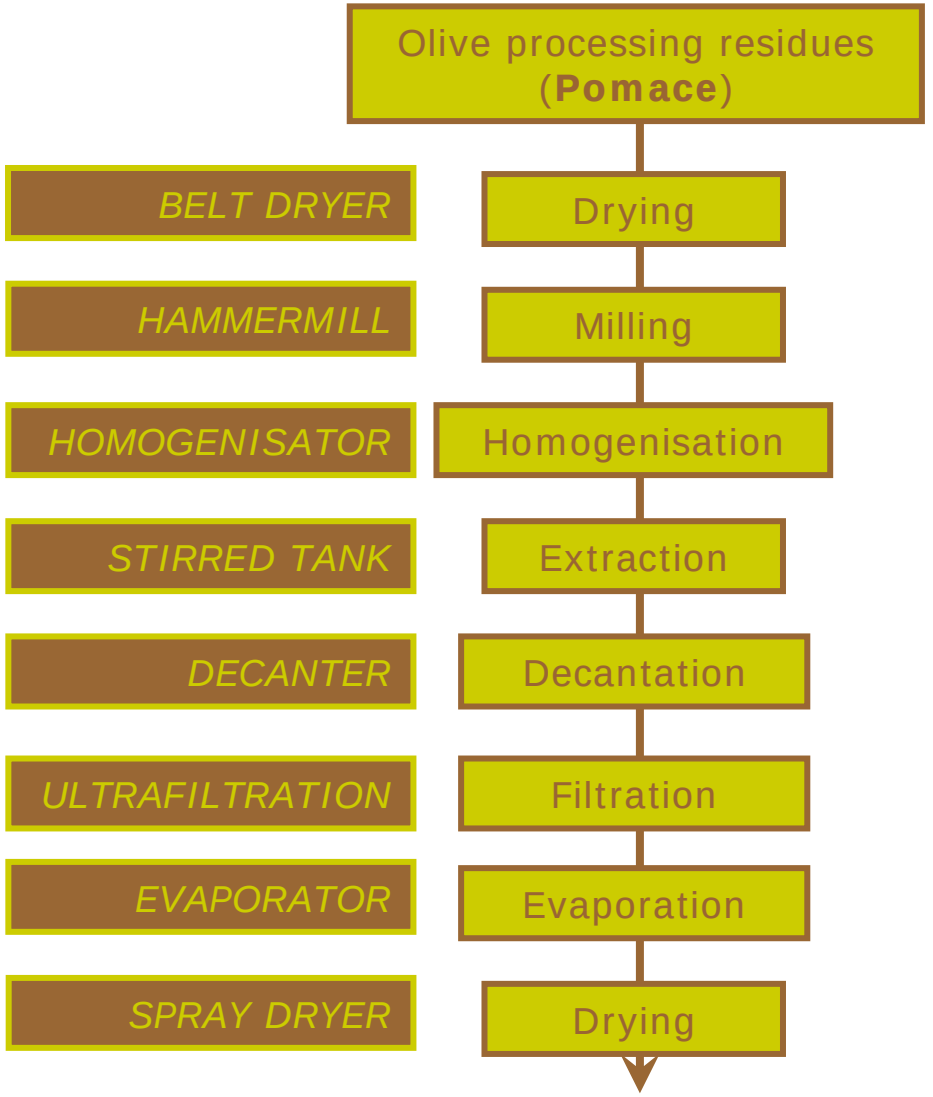
Olives

Supercritical Fluid Extraction



Solvent Extraction



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SYNTHESIS on Olives:

		Minimum Hypothesis (500t/year)	Maximum Hypothesis (11.000/year)
Supercritical Fluid Extraction	Yearly profit	179.429 €	13.739.899 €
	Initial investment	*2.410.000 €*	*30.900.000 €*
	Time needed to recover initial investment	*14 years*	*3 years*
Solvent Extraction	Yearly profit	-35.029 €	1.690.861 €
	Initial investment	*1.370.000 €*	*1.840.000 €*
	Time needed to recover initial investment		*2 years*

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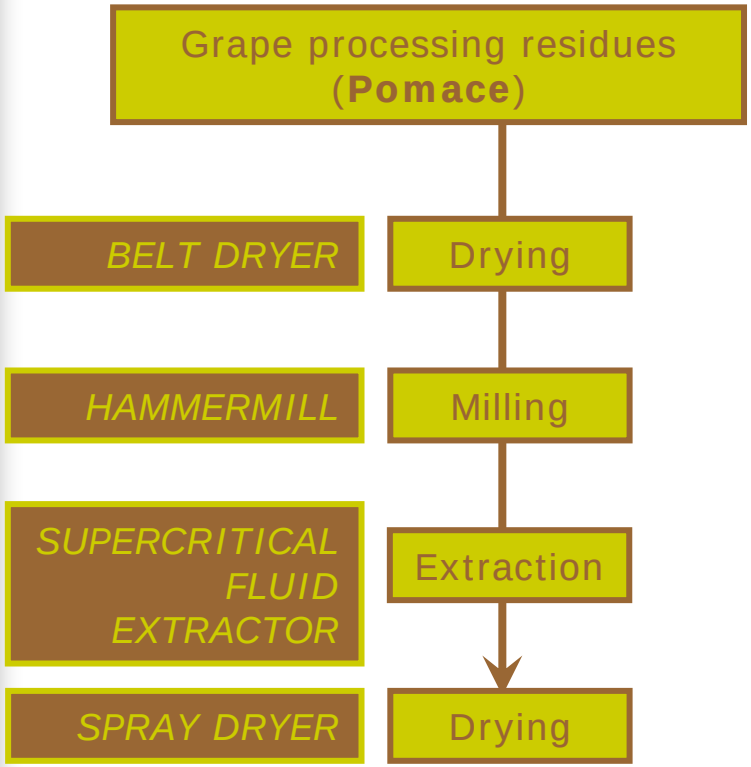
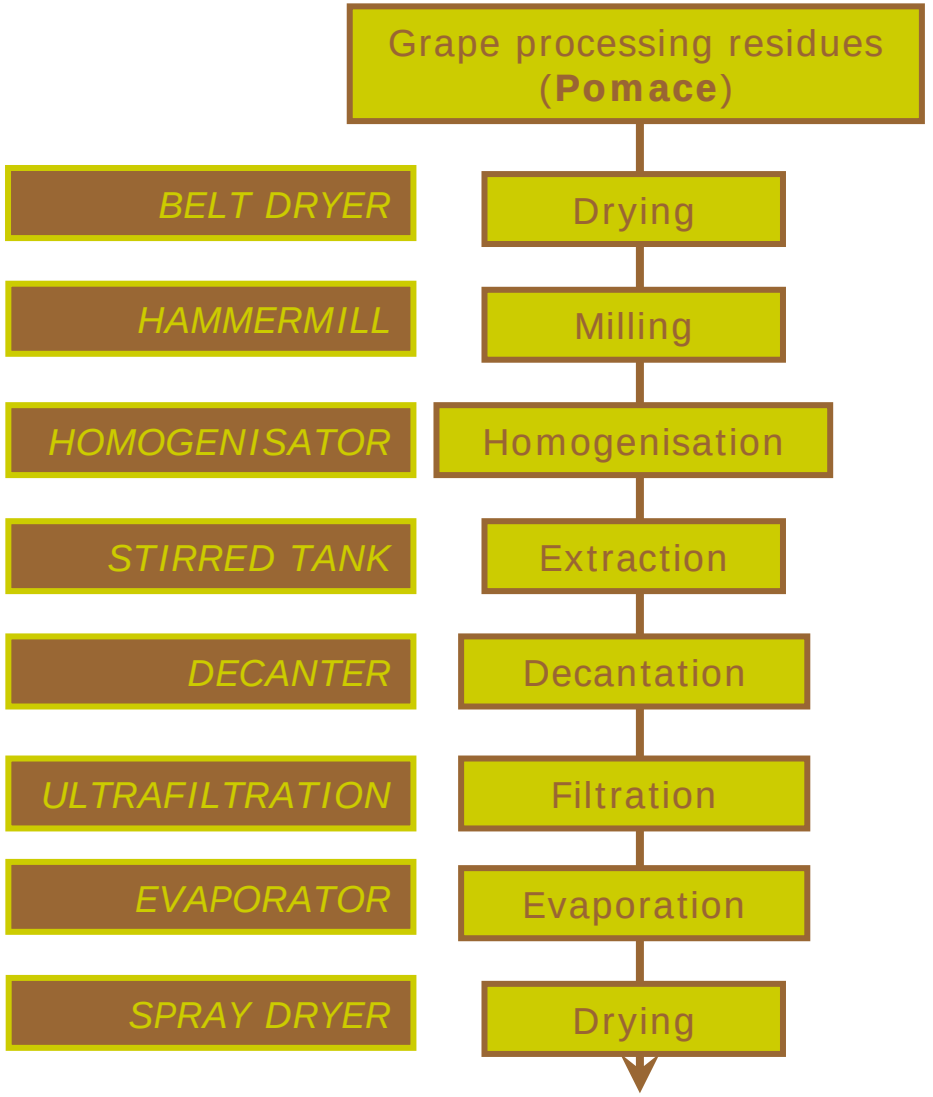
Grape

Supercritical Fluid Extraction



Solvent Extraction



TECNOALIMENTI

SYNTHESIS on Grape:

		Minimum Hypothesis (75t/year)	Maximum Hypothesis (50.000/year)
Supercritical Fluid Extraction	Yearly profit	-58.161 €	69.243.162 €
	Initial investment	*2.410.000 €*	*139.900.000 €*
	Time needed to recover initial investment		*2 years*
Solvent Extraction	Yearly profit	-30.151 €	20.131.800 €
	Initial investment	*1.370.000 €*	*2.940.000 €*
	Time needed to recover initial investment		*1 year*

TECNOALIMENTI

Remarks:

- Working time of 330 days annually
- 24 hours a day

- Costs of sales and marketing, shipping, handling and storing have not been considered
- Recovery costs of solvent have not been considered