

Agronomical Plant Extracts & Essential Oils

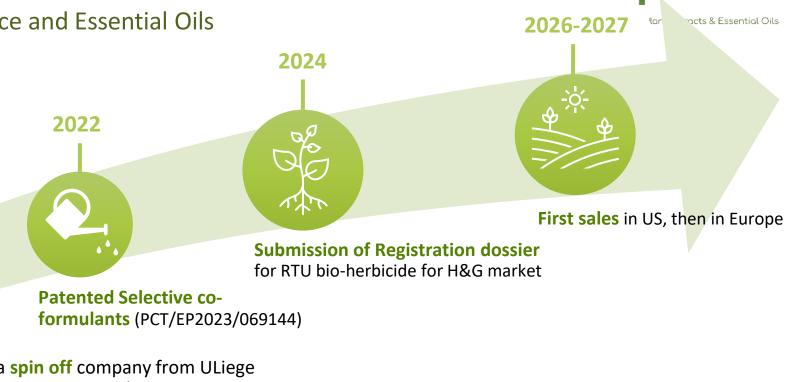
Developing sustainable Agriculture,

Putting Science and Agronomy at the heart of our activities

Mission

Shifting towards sustainable agriculture and gardening by bringing innovative bio-pesticides based on Science and Essential Oils

2021





2018

Creation of APEO, a **spin off** company from ULiege **secured** from private investors and grants

Patented natural Formulation (patent# WO2019238948A1)

Selection of **Essential oils** for their fungicidal and herbicidal properties

Company Information

APEO Team Who/Background





A management combining Science and Business



Arnaud Malerbe, CEO

25+ years managing business development teams and PMI in biosolutions for agriculture. International experience: Mexico, Brasil, Spain, Haiti, Europe, South Africa and USA



Pr. Haïssam Jijakli, co-founder & CSO

33 years' experience in biocontrol Full Professor at ULiège/Gembloux, Phytopathology lab, Creation of 4 Spin-Offs and 10 patents



Simon Dal Maso, co-founder & Technical director

8 years experiences Accelerated Management Program/Solvay Business School 2018

The dedicated team and the right support for our stage of development

Experienced managers in the key functions and external support :

- Regulatory
- Technical development
- Supply and ops
- Business development
- Finance and administration



Dynamic young and hybrid R&D team (APEO & ULiège researchers and technicians)

All committed with the APEO Values and project



CONFIDENTIAL



From research to first development



- More herbicide-resistance of weeds
- Retailer and consumer reluctance to chemical residues and public concern for environmental safety
- Limitation and withdrawal of authorized active ingredients (Glyphosate, Diquat,...)
- Development of novel practices (e.a. Mechanical weeds killing) or products (Pelargonic acid)

Pre-selection considering plant protection market and EO market



 Among 3000 EOs, pre-selection of 91 EOs for fungicidal and herbicidal activities based on :

- Literature
- Composition
- Majority of the chemical families (Terpens, phenol, alcohol)
- OCost
- Availability

Pre-selection considering plant protection market and EO market



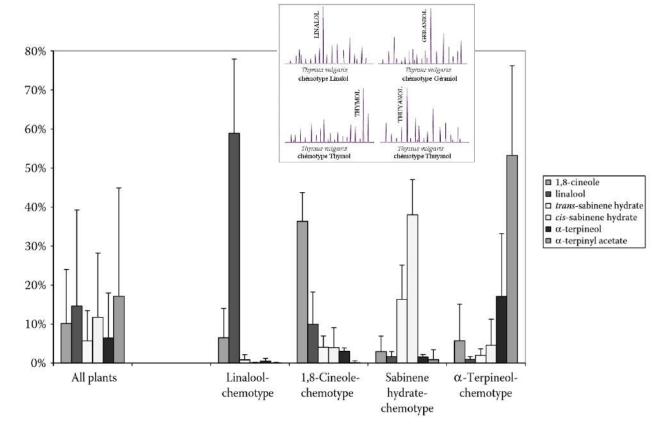
Agronomical Plant Extracts & Essential Oils

Chemotype

Chemotype = intraspecific variation Chemotype of *Thymus vulgaris*



Selection of our suppliers according to the right chemotype and its stable composition



Source graph: Baser, K. H. C., & Buchbauer, G. (2015). Sources of essential oils. Handbook of Essential Oils: Science, Technology, and Applications, Second Edition, p.52. https://doi.org/10.1201/b19393

Selection under greenhouse conditions



Preselection of 22 EOs for herbicidal action and test on:

- Monocotyledons: grass (Festuca 70%, Lolium 30%)





- Dicototyledons : *Urtica dioica, Chenopodium, Papaver, Trifolium incarnatum*

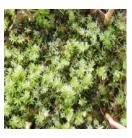








- Bryophyte



- Pteridophyte



Selection under greenhouse conditions



Activity against

	Type of major peak	Monocotyledon	Dicotyledon	Mosses	Horsetail
EO1	Aromatic	+++	++++	++++	++++
EO2	Aromatic	++++	++++	undertermined	undertermined
E03	Terpenic	++++	+	undertermined	undertermined



Efficacy of EO1



EO2



EO3



Untreated



Untreated



Efficacy of EO1

Importance of the formulation



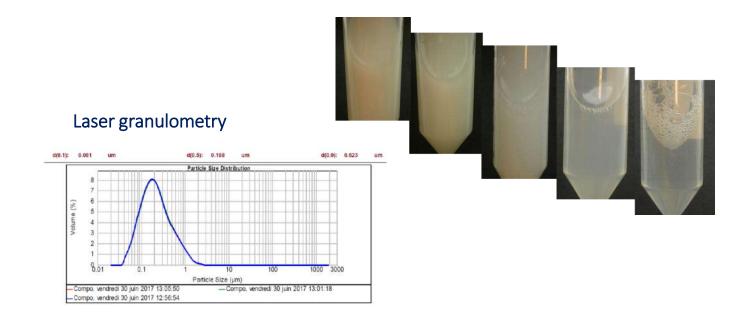
- Influences the modes of action
- Protects the EOs against the environment
- Influences the stability

And it strongly affects the efficacy

Formulation for stable emulsion



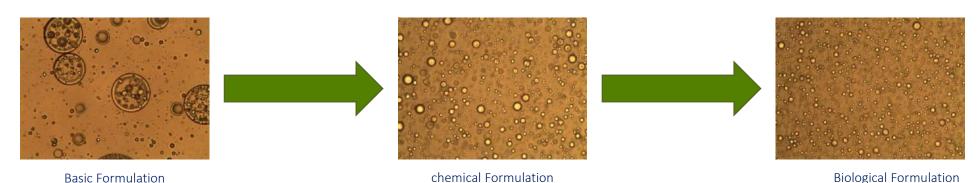
Туре	Droplet size	appearence	Stability
Marco emulsion	2-20μm	Milky white	Instable
Mini emulsion	0.1-0.3μm	Bluish white	Several week
Micro emulsion	<0.1µm	translucent	Several months



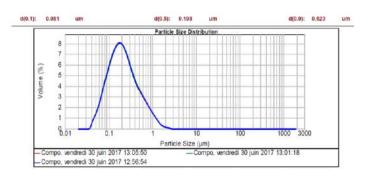
Formulation from chemical to biological adjuvants



Microscopy



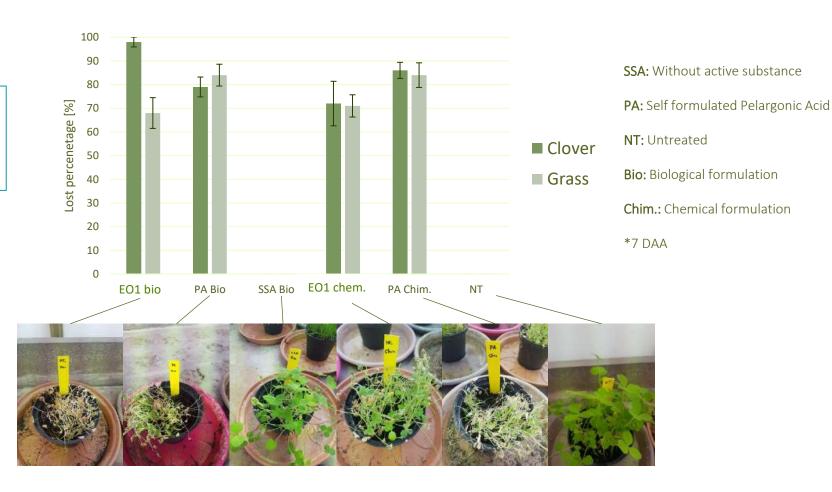
Laser granulometry



Formulation from chemical to biological adjuvants



Efficacy on clover and grass



Improvement and validation of the biological formulation with efficacy trials at different scales







Patented formulation





From development to first products

Characteristics of EO1 - Marketing



- ✓ EO1 is a unique novel biosourced biocontrol herbicide active ingredient based on essential oil
- ✓ As such, EO1 has a general positive perception form the public and essential oils are also known in other positive applications (medical, para-medical, cosmetic,...)
- ✓ RTU (Ready to use) and 5 x concentrated formulations are fully from natural organic ingredients
- ✓ PLEASANT ODOR
- ✓ Fully, steadily biodegradable
- √ (Will be) OMRI certified and certified as biocontrol
 - ✓ All ingredients are OMRI certified

Efficacy trials under field conditions 2019



June 2019 in Belgium

Sown weeds in fields (4 monocotyledons and 12 dicotyledons)

Application of EO1 with final formulation

2 applications (10 days between applications)

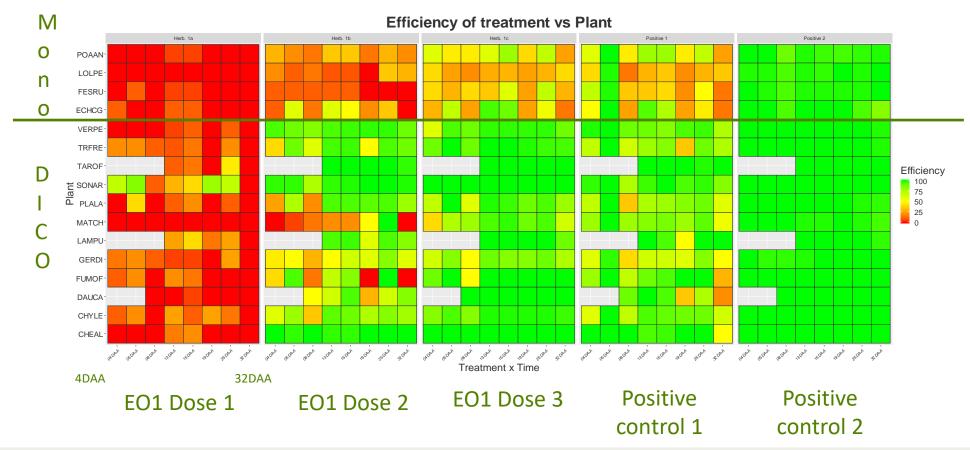
4 replicates



Efficacy trials under field conditions 2019

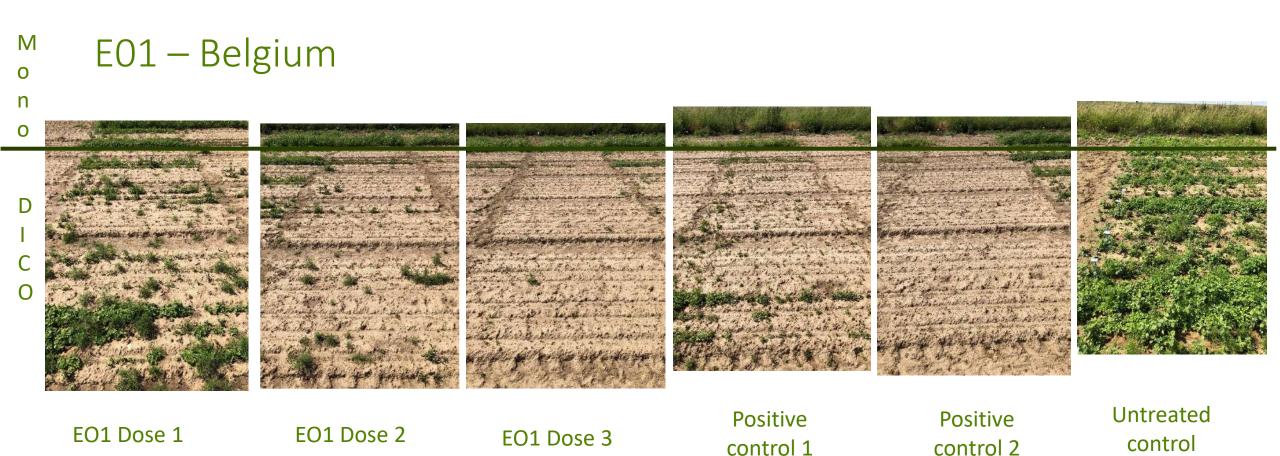


EO1 – Belgium



Efficacy trials under field conditions 2019





Characteristics of EO1 – EFFICACY on permeable soil



Belgium in 2021 Sown weeds in fields

APEO



Fatty Acid



Characteristics of EO1 – EFFICACY after <u>5 years of GEP trials</u>



- ✓ EO1 has a **broader and higher global efficacy against DICOT** than products based on Pelargonic Acid (AP)
 - ✓ EO1 has a higher efficacy on 29/40 species of DICOT
- ✓ EO1 has a global similar efficacy against MONOCOT than products based on AP
 - ✓ EO1 has a higher efficacy on 4/7 species of MONOCOT
- ✓ EO1 has a global similar activity in comparison with US market reference products based on
 - √ Fatty acids + hydrazide maleic (chemical)
- ✓ Residual activity
 - ✓ better than Pelargonic Acid
- ✓ Germicidal action
 - ✓ No regrowth of seeds produced by the treated plant

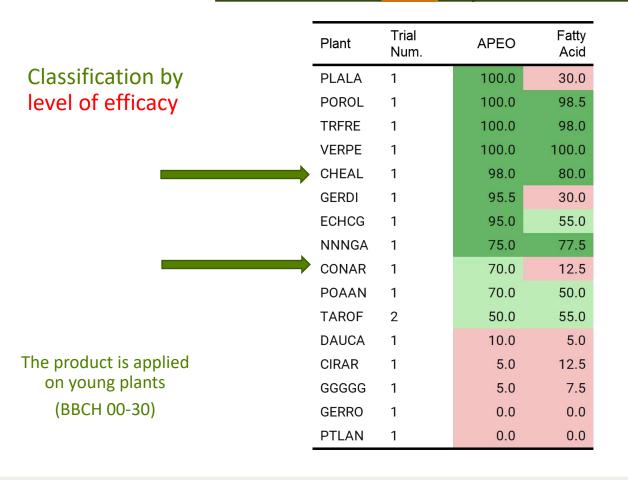
Characteristics of EO1 – EFFICACY on permeable soil QDE

Permeable soil – 21-25 Days after treatment

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	Plant	Trial Num.	APEO	Fatty Acid
	GERRO	1	100.0	12.5
	LAMPU	1	100.0	12.5
	MATCH	3	100.0	75.0
	STEME	1	100.0	15.0
	THLAR	1	100.0	12.5
	VERPE	1	100.0	100.0
	GERDI	2	99.5	75.0
	POROL	2	99.0	90.0
	PLALA	1	97.5	40.0
	TRFRE	2	94.0	52.5
	SETVI	1	92.5	87.5
	CONAR	1	80.0	22.5
	GLNPU	1	80.0	17.5
	MATIN	1	75.0	25.0
	ECHCG	2	75.0	35.0
	POAAN	4	72.5	62.5
	DAUCA	1	60.0	30.0
	NNNGA	2	55.0	70.0
	LOLPE	1	50.0	15.0
	CHEAL	3	30.0	35.0
	HOLMO	1	30.0	10.0
	EPIAD	1	20.0	0.0
	PTLAN	1	17.5	17.5
ILY CONFIDENTI	TAROF	3	12.5	10.0

Permeable soil – 61-70 Days after treatment



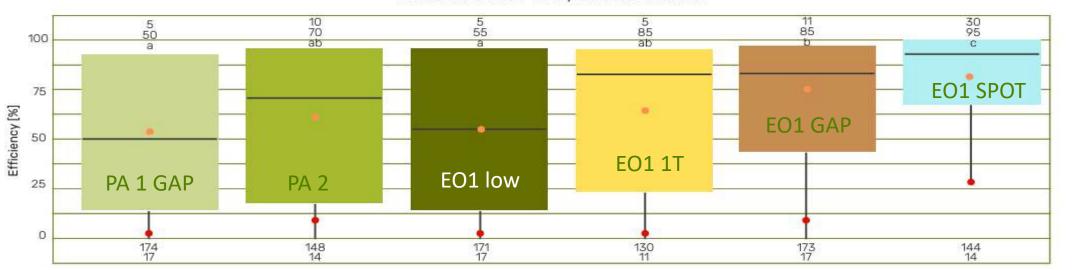
APEO copyright

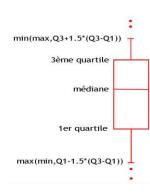
Efficacy Dicots all surfaces 3 years BBCH 30



All surfaces 3 years BBCH 30

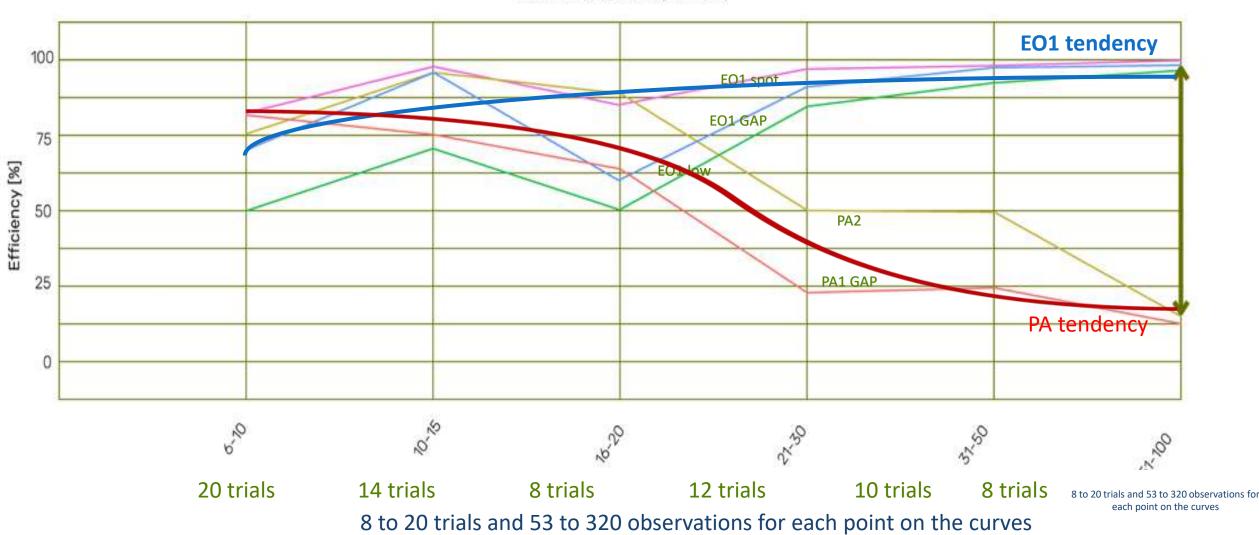
BoxPlot on Dico at 21-30 days after first treatment





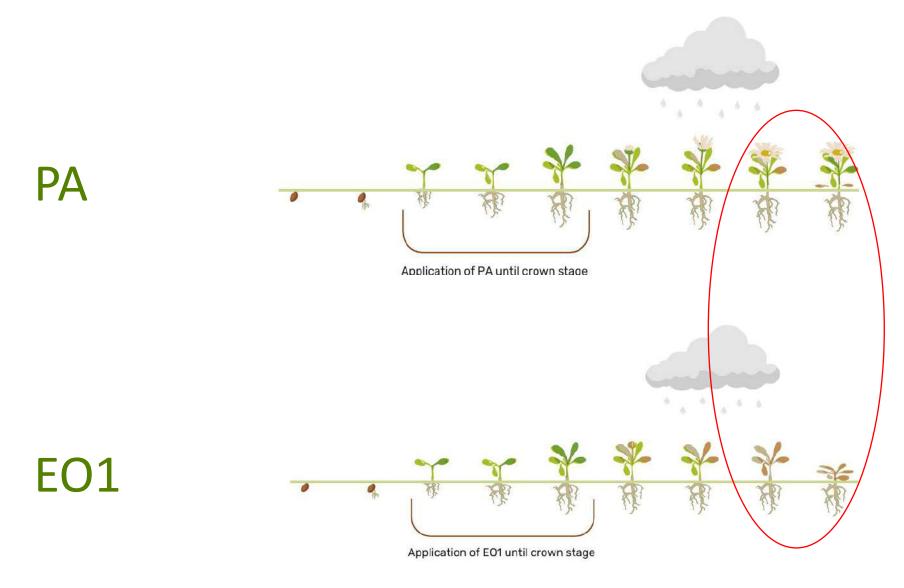
Efficacy, permeable soil, 3 years Dicots BBCH30





Efficacy benchmark EO1 vs PA





Characteristics of EO1 – EFFICACY on permeable soil



Germany in 2021 Natural weeds

APEO



Fatty Acid



Modes of Action of EO1



Contact

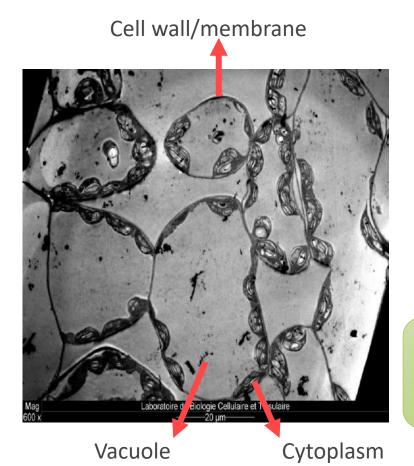
Multisite actions and more modes of actions than AP

No impact on germination if sowing few days after application

Characteristics of APEO EO1 – MODES OF ACTION

Aproportical Plant Extracts & Essential Oils

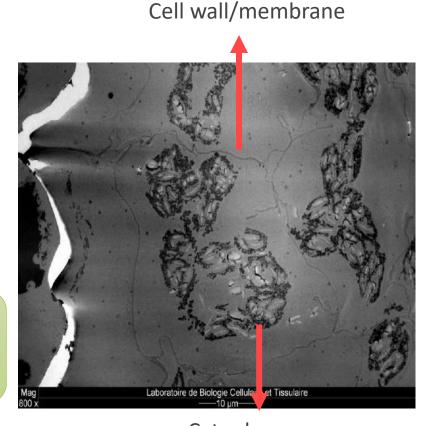
Effect on plant cells



Normal plant cell

Contact

No impact on germination if sowing few days after application



Effect on roots



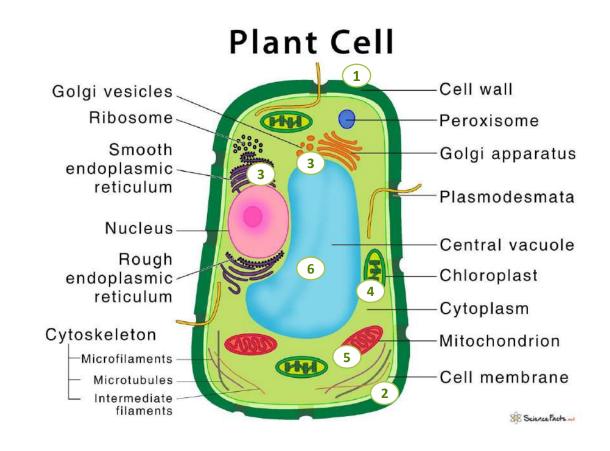
Cytoplasm
Plant cell after application of APEO EO1

Characteristics of APEO EO1 – MODES OF ACTION



Multisite effect

- 1. Inhibition of membrane cell synthesis
- 2. Alteration of membrane proteins,
- Destabilization of lipid organization and electrolyte leakage
- 4. Destabilization of protein synthesis
- 5. Inhibition of photosynthesis (Chloroplast)
- 6. Inhibition of cellular respiration
- 7. Total disappearance of the vacuole



Conclusions for bioherbicides



• APEO E01

- ✓ RTU formulation for H&G, water based
- ✓ Ongoing submission of regisration dossier
- ✓ Ongoing distributorship investigation

RTU herbicide by spraying application



• **APEO E04**

- ✓ One formulation for professional use under development
- ✓ Non-selective weed control
- ✓ Potato leaf kill
- √ Vines shoot control

Suckering test – France 2021







Untreated

Perspectives for bioherbicides



Code	Market	USES	Formulation	First Sales	
APEO EO1	H&G	Non-selective Herbicide on permeable an non- permable soil	RTU	2026	- Herbicides
APEO EO2	H&G	Non-selective Herbicide on permeable an non- permable soil	Concentrated	2027	Tierbicides
APEO EO3	H&G	Algicide on semi and non-permeable soil/material	RTU	2027	Biocide
APEO EO4	PRO	Non-selective Herbicide Grapes, Fruit trees, Potatoes	Concentrated	2028	ĺ
APEO EO5	H&G	Selective Herbicide on permeable an non- permable soil	RTU	2028	– Herbicides
APEO EO6	H&G	Selective Herbicide on permeable an non- permable soil	Concentrated	2028	Herbicides
APEO EO7	PRO	Selective Herbicide Cereals	Concentrated	2030	IJ



From research to first biofungicide development



- Fungi are responsible of yield decrease worldwide
- Chemical fungicide-resistance of fungal populations
- Public is concerned about human health and environmental pollution
- Biofungicides is one of the most dynamic market due to no residue and resistance strategy
- Difficult to control fungal diseases without fungicides

Pre-selection considering plant protection market and EO market



 Among 3000 EOs, pre-selection of 91 EOs for fungicidal and herbicidal activities based on :

- Literature
- Composition
- Majority of the chemical families (Terpens, phenol, alcohol)
- OCost
- Availability

Pathosystem selection



Cultures	Pathogens		
Wheat	Septoria tritici		
	Puccinia striiformis		
	Fusarium graminearum / F culmorum		
Sugar Beet	Cercospora beticola		
	Erysipahe betae		
	Rhizoctonia solani		
Potato	Phytophthora infestans		
	Erwinia carotovora/ E. atrospetica.		
Apple (tree)	Venturia inaequalis		
Apple and Pears	Botrytis cinerea		
	Penicillium expansum		
	Gloesporium perennans		
Strawberry	Xanthomonas fragariae		
	Podosphaera aphanis		
	Botrytis cinerea		

Cultures	Pathogens		
Bean	Colletotrichum lindemuthianum		
Vineyard	Plasmopara viticola (mildiou)		
	Uncinula necator (oïdium)		
Soils	Pythium ultimum		

In vitro screening of best EOs





- 92 extracts (emulsion)
- 2 [EOs]
- =184 objects/pathogen * 8 reps
- =36 plates/pathogen



- •Measure of OD /24 h
 - •120h
- •Measure of OD /2 h
 - •24h

In vitro screening of best EOs





Phytotoxicity screening

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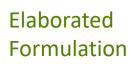
- Importance of the formulation
 - Concentration of EOs
 - Formulation

Basic Formulation











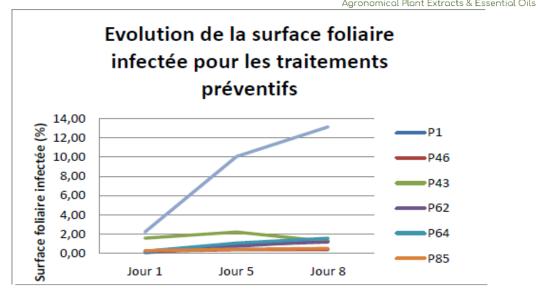




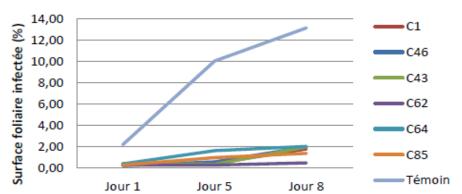
In vivo screening of best Eos on sugar beet



- Set up
 - Preventive treatment
 - 2-3 Hours before inoculation
 - By spraying EOs on plants
 - Curative treatments
 - 24 hours after inoculation
 - By spraying EOs on plants



Evolution de la surface foliaire infectée pour les traitements curatifs



In vivo screening of best Eos on strawberry



- Set up
 - Curative treatments
 - After natural inoculation
 - By 2 successive spraying treatments with 1 week interval



Control

1rst treatment



2nd treatment



In vivo screening of best EOs

Plant	Pathogen	Efficacy
Sugar beet	Cercospora beticola	© ©
	Erysiphae betae	©©
	Rhizoctonia solani	☺
Potato	Phytophthora infestans	◎ (◎)
Apple(tree)	Venturia inaequalis	©©
Strawberries	Podosphaera aphanis	©©
	Xanthomonas fragariae	☺
Apple-Pear	Botrytis cinerea	⊗(©)
	Penicillium expansum	⊗(©)
	Gloesporium perennans	⊕(◎)
Wheat	S. tritici, P. striiformis. Fusarium sp.	(2)
Soil	P. Ultimum	©©

From research to first biofungicide development



Undergoing Post-doc





Undergoing PhD



Undergoing PhD



ALLO COPYLISHE

PhD starting in Sept 2024

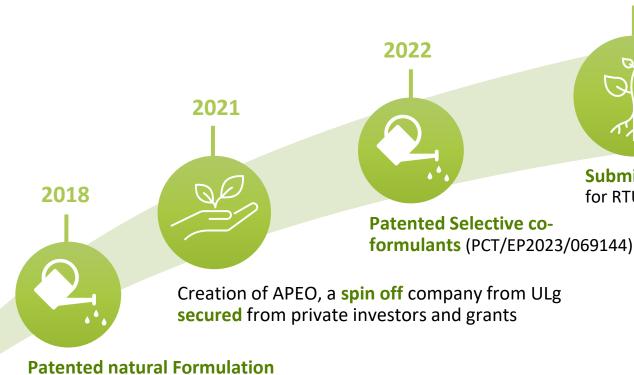
Perspectives for biofungicides



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APEO EO7	PRO	Selective Herbicide Cereals	Concentrated	2030	
APEO EO8	PRO	Fungicide against Potato mildew	Concentrated	2029]
APEO EO9	PRO	Fungicide against Apple scab	Concentrated	2029	- Fungicides
APEO EO10	PRO	Fungicide against Septoriose on wheat	Concentrated	2030	
APEO EO11	PRO	Fungicide against Cercospora leaf spot on sugar beet	Concentrated	2030	

Thank you for your attention





Submission of Registration dossier for RTU bio-herbicide for H&G market

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Website

2024

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Selection of **Essential oils** for their fungicidal and herbicidal properties

(patent# WO2019238948A1)

2011