



Essential oils as alternative for sustainable management of *Phytophthora infestans* causing potato late blight disease : From *in vitro* to *in vivo* experiments



Florian MARTINI

Teaching assistant, PhD student

Department of Chemistry for sustainable food and
environmental systems

Clément BURGEON, Jérôme MUCHEMBLED, Eric GONTIER,
Marie-Laure FAUCONNIER & Haïssam IJAKLI





01

Introduction



Potato and late blight disease

Solanum tuberosum
L., 1753



Perennial plant grown for its tubers rich in carbohydrates



1st non-cereal grown worldwide for human consumption



Late blight disease represents on potato an annual loss of \approx \$6B

Phytophthora infestans
(Mont.) de Bary, 1876

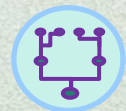
Eucaryotic microorganism belonging to Oomycota



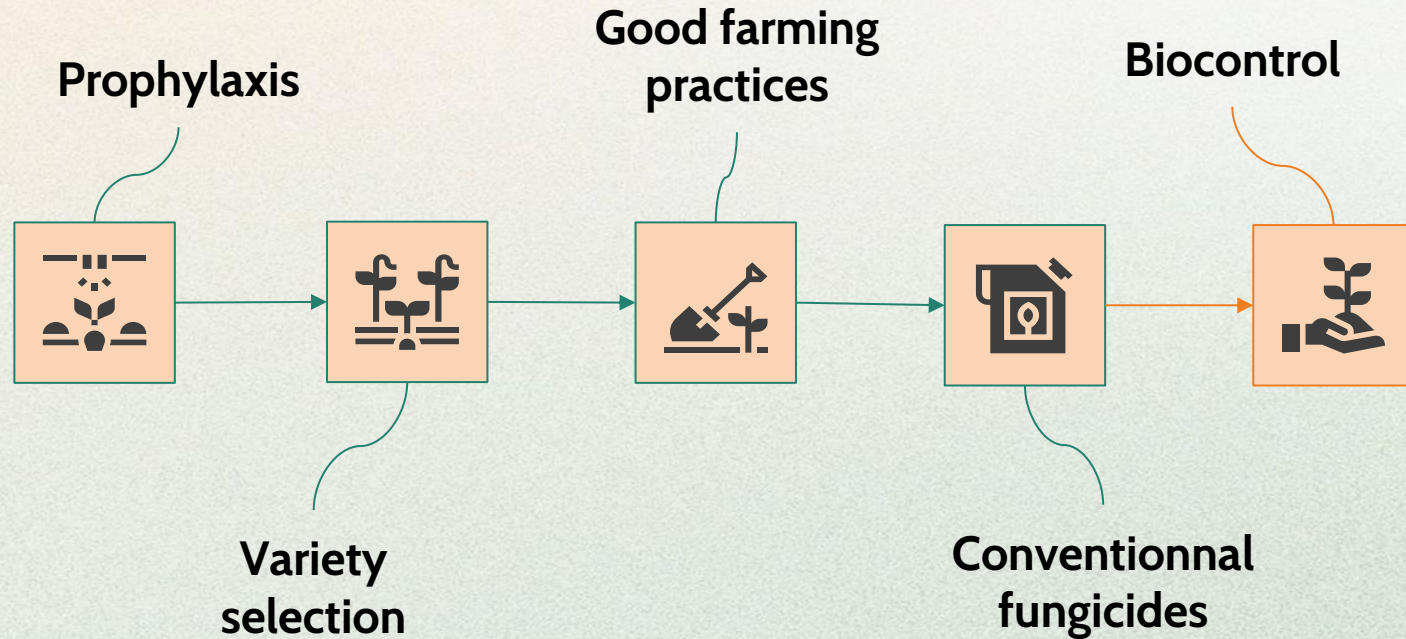
Cellulosic cell wall and membrane without ergosterol



Phylogenetically closer to Rhodophyta than Fungi

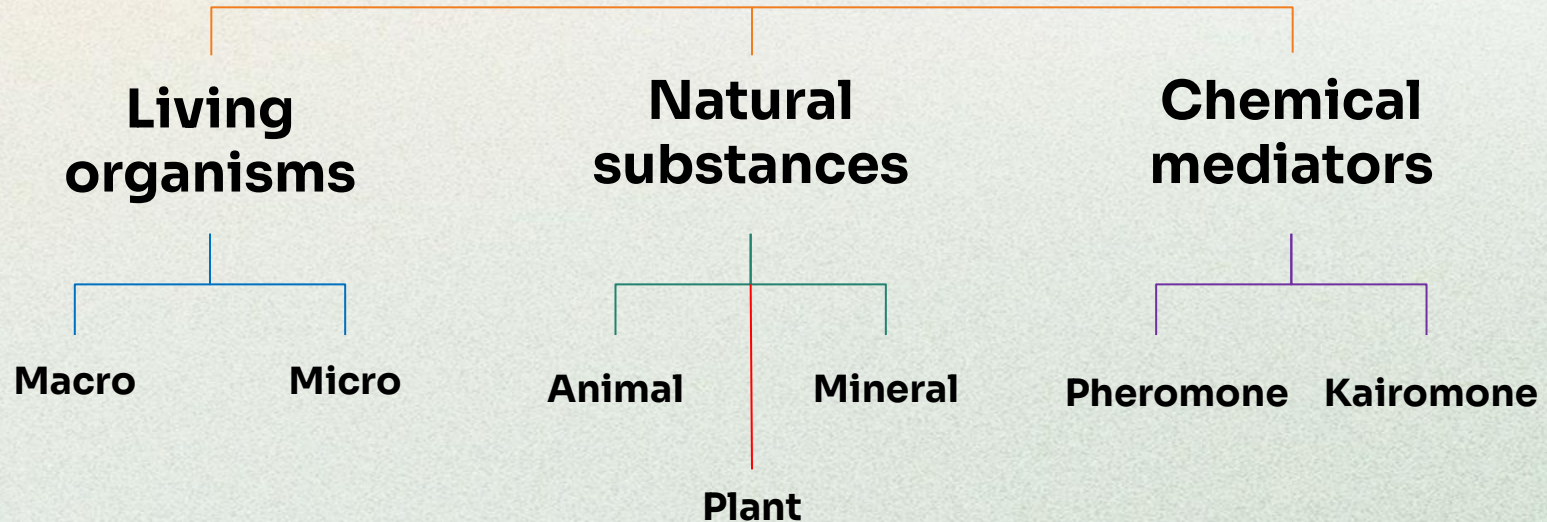


Current management of phytopathogens

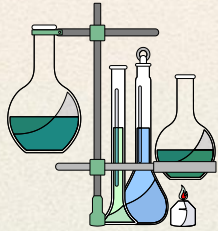


Biocontrol

Any method used to protect plants from pests of any kind
(animals, weeds, bacteria, fungi and viral diseases)
based on naturally existing mechanisms and substances



Essential oils



Essential oils (EO) are complex hydrophobic mixtures mainly composed of volatile organic compounds (VOC's) and exclusively obtained from plant biomass by some extraction methods such as steam distillation or cold pressure

Fometu *et al.*, 2019
Masango *et al.*, 2005



VOC carry many chemical functions and present diverse biological activities

Bassolé *et al.*, 2012



Plant originally use VOC to attract pollinators, fight diseases and communicate

Slavkovic *et al.*, 2019



EO have been used for decades by human in many fields, including plant protection

De Clerck *et al.*, 2021

Research context



State of the art

Many EO have shown promising properties against phytopathogens from *in vitro* to *in planta*

De Clerck *et al.*, 2020



Lack of knowledge

Molecular mechanisms of EO on *P. infestans* at the cellular scale are still not well understood

Martini *et al.*, 2023



Challenge and constraint

Due to their chemical properties, appropriate formulation is required to optimize EO activities as biofungicide

Maes *et al.*, 2019



02

Results



Experimental material



Host plant

Two varieties of
S. tuberosum
economically
important in Europe
with high sensitivity
to late blight

- Bintje
- Fontane



Pathogen

Three different
genotypes of
P. Infestans causing
massive damages in
Northern
argo-ecosystems

- EU-13-A2
- EU-36-A2
- EU-37-A2

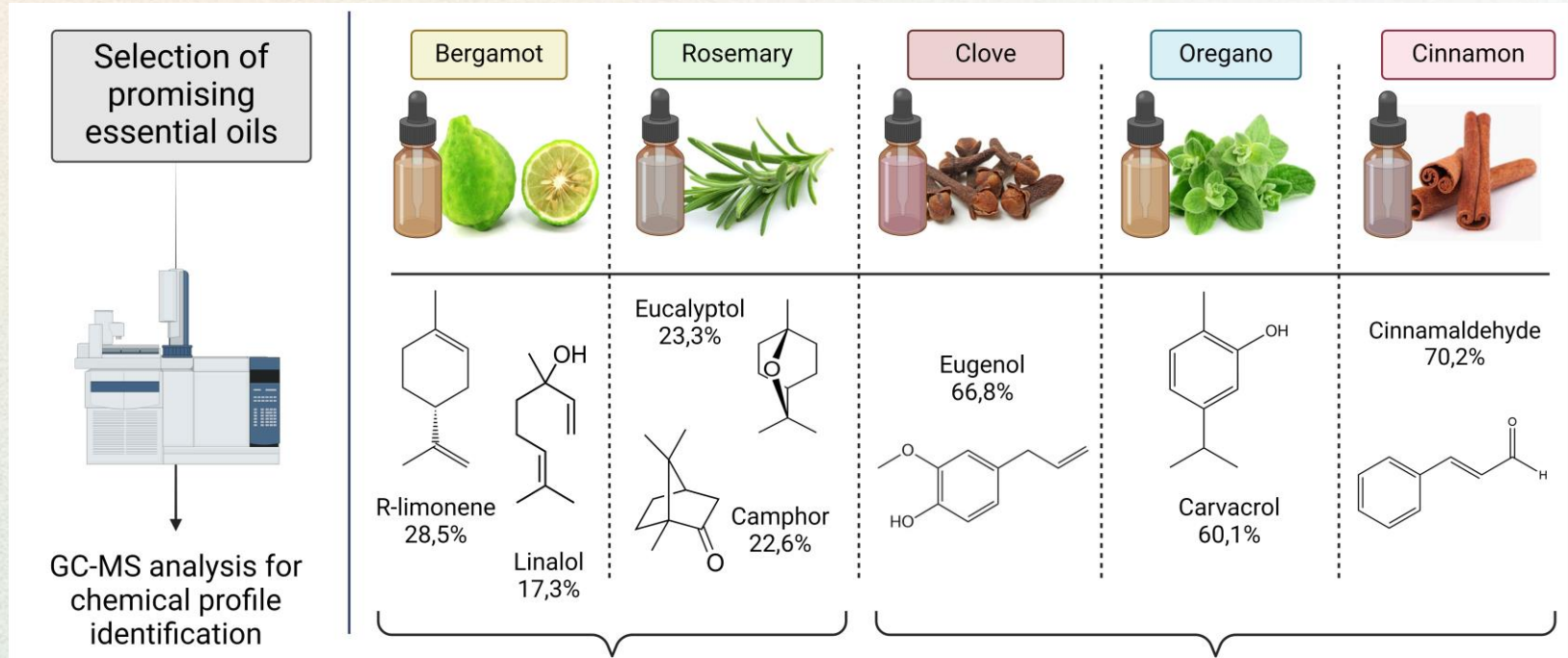


Essential oils

Five essential oils
extracted from
various sources of
biomass and with
specific chemical
composition

- Rutaceae
- Lamiaceae
- Myrtaceae
- Lauraceae

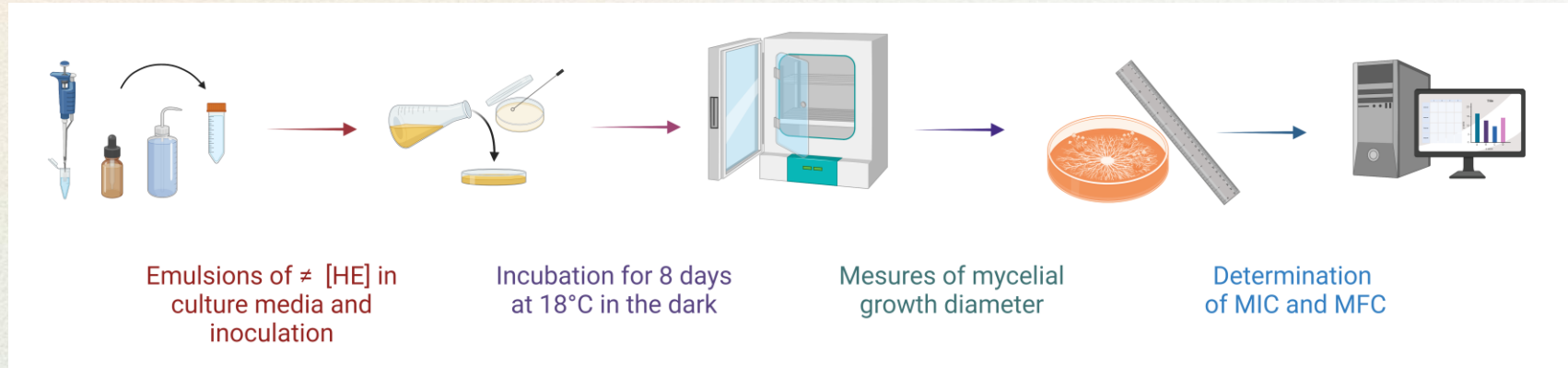
Results : essential oils chemical profile



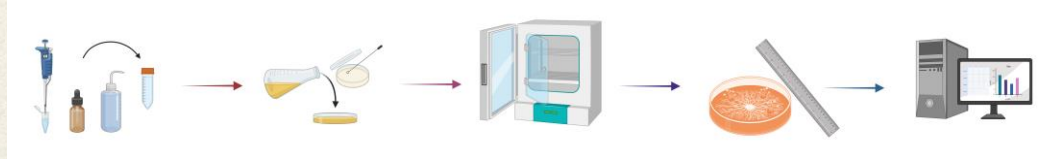
Mainly composed
of **terpenoids**

Mainly composed
of **phenylpropanoids**

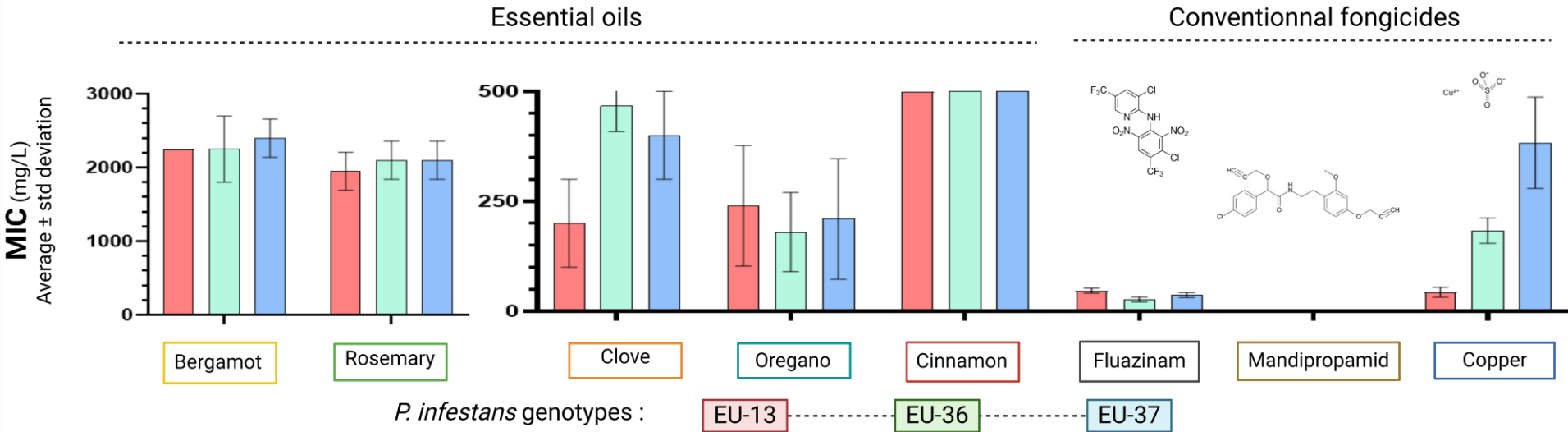
Method : mycelium growth inhibition



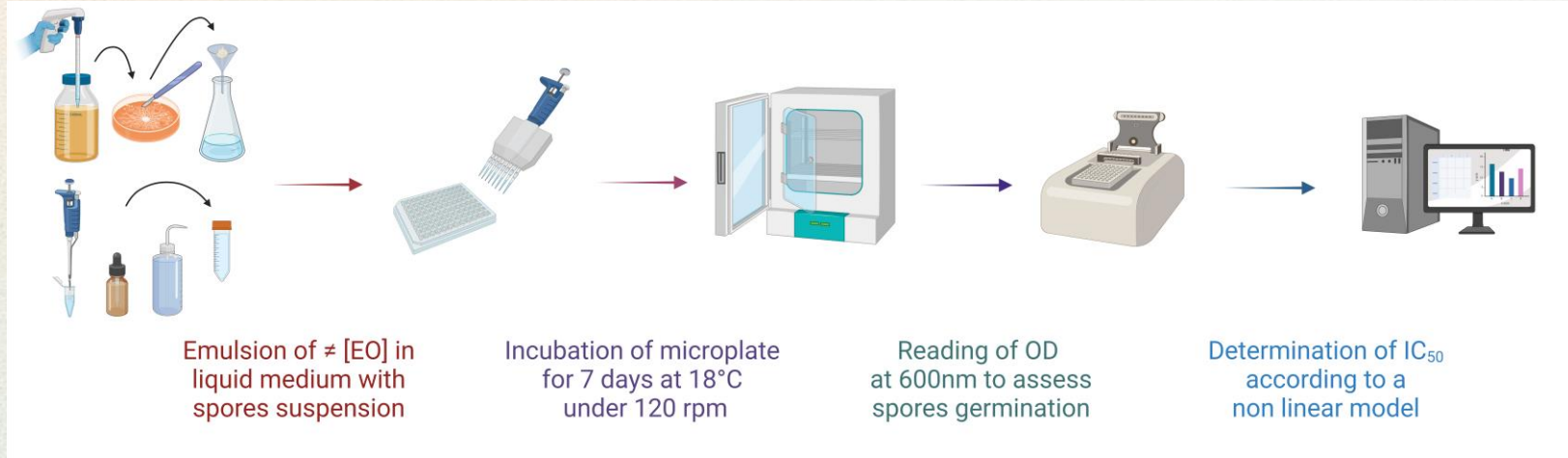
Results : mycelium growth inhibition



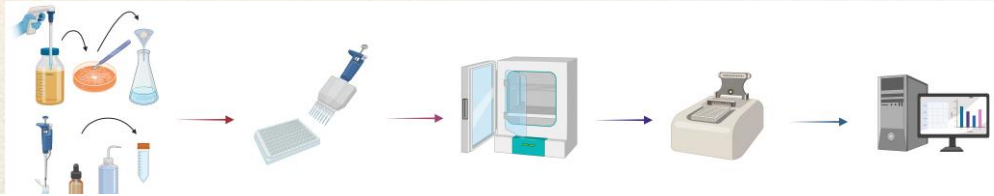
Minimum inhibitory concentrations (MIC) of chemical substances on mycelial growth of 3 *P. infestans* genotypes in Petri dish



Method : spore germination inhibition



Results : spore germination inhibition



Median inhibitory concentrations of chemical substances on spores germination of 3 *P. infestans* genotypes in microplates

IC ₅₀ (mg/L) [confidence range]	Essential oils					Conventional fungicides		
	Bergamot	Rosemary	Clove	Oregano	Cinnamon	Mandipropamid	Fluazinam	Copper
EU-13	9100 [7500 ; 13000]	9810 [7920 ; 12060]	500 [400 ; 600]	1620 [990 ; 1890]	130 [90 ; 200]	0.010 [0.03 ; 0.21]	0.35 [0.1 ; 1.3]	6 [1 ; 23]
EU-36	9900 [6300 ; 15300]	2160 [1080 ; 3330]	100 [60 ; 140]	270 [180 ; 360]	120 [50 ; 280]	0.012 [0.02 ; 0.32]	0.25 [0.05 ; 3.2]	14 [2 ; 27]
EU-37	4400 [3500 ; 5580]	5940 [4770 ; 7560]	400 [300 ; 600]	360 [180 ; 540]	80 [50 ; 130]	0.015 [0.06 ; 0.20]	0.29 [0.01 ; 6.2]	80 [30 ; 220]

Results : phytotoxicity on detached leaves



Sampling of young
leaves from potato
plants



Emulsion of essential
oils in water with
Tween20



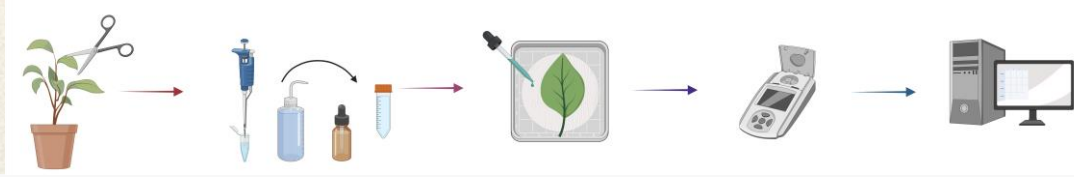
Dropping emulsion
on potato detached
leaves



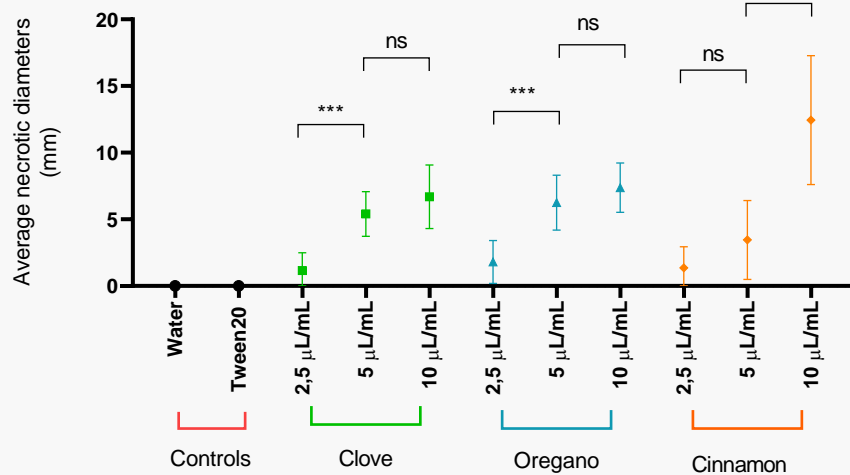
Measuring photosystem
fluorescence and
assessing phytotoxicity



Results : phytotoxicity on detached leaves

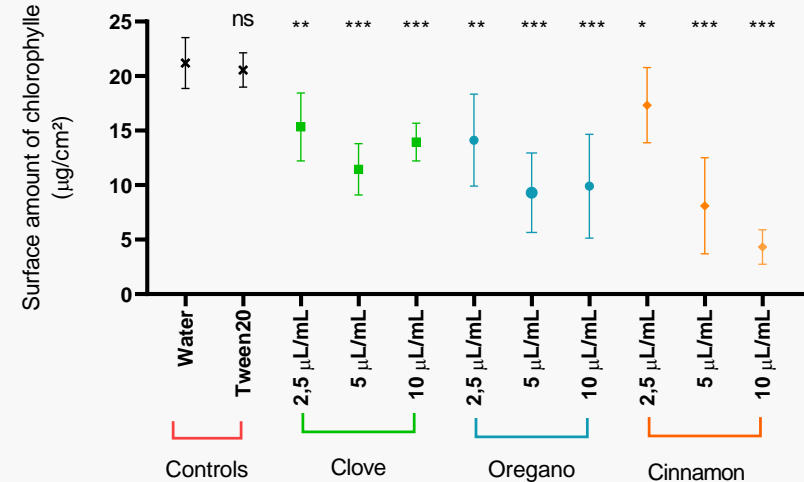


Necrotic diameters on potato detached leaves after drop treatments of various concentrations of essential oils emulsions



Essential oils concentrations in emulsions

Surface amount of chlorophyll on potato detached leaves after drop treatments of various concentrations of essential oils emulsions



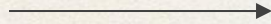
Essential oils concentrations in emulsions

03 Conclusion and Perspectives

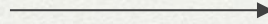


Conclusion : From *in vitro* to *in vivo*

1

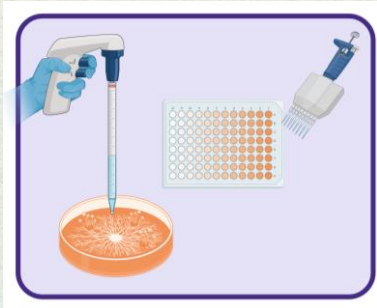


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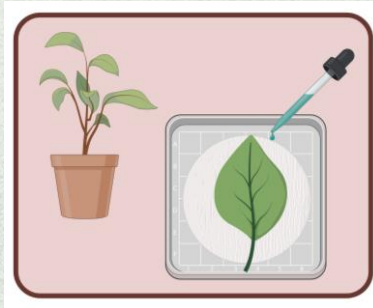


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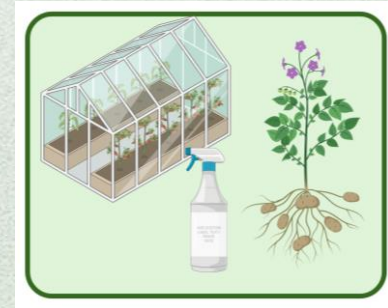
In vitro tests showed promising anti-oomycete properties of specific EO's



Ex vivo assays gave phytotoxicity thresholds on potato detached leaves



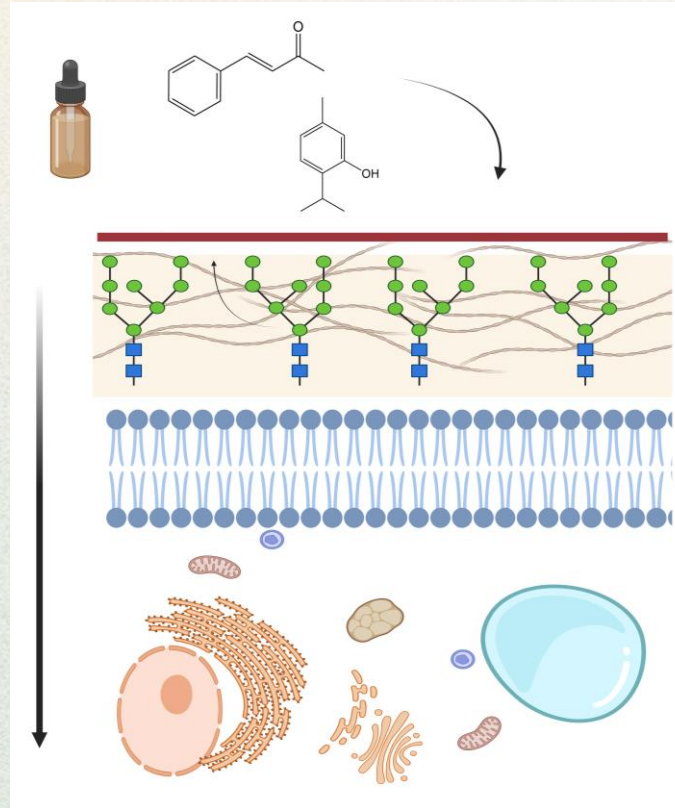
In planta experiments must be carried out to confirm EO protective properties against late blight



Perspectives : Cellular site of action

Successive extractions with different solvents on treated *P. infestans* biomass followed by GC-MS analysis should help determine :

- in which cellular compartment
- how long EO are retained.



● Hyphae surface

● Cell wall

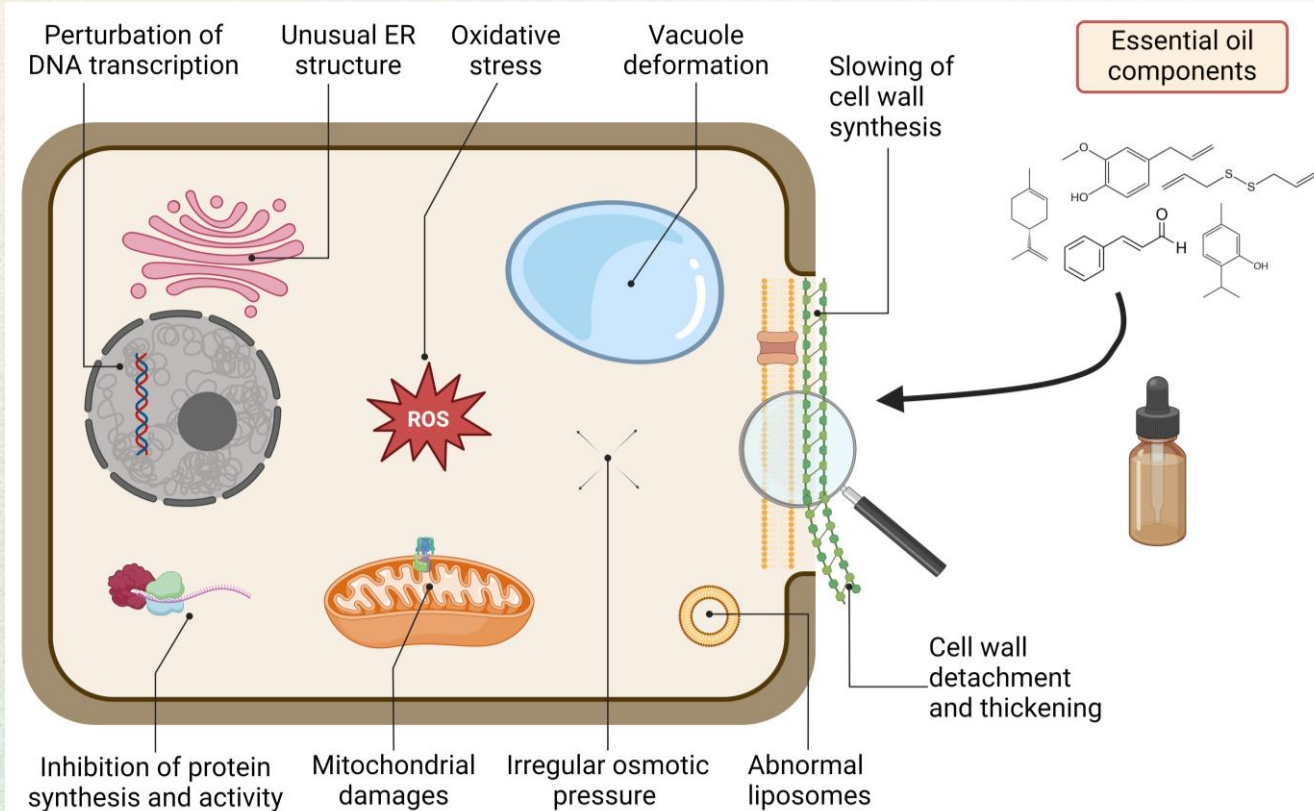
● Plasma Membrane

● Cytoplasm

Perspectives : Mechanisms of action

Many mechanisms have been described on how EO's perturb cellular activities.

Our hypothesis :
→ Focus on plasma membrane



Thanks for listening!

I'd be happy to answer any questions!

florian.martini@uliege.be

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