The 19th of May

ISCP
INTERNATIONAL SYMPOSIUM ON CROP PROTECTION

Essential oil against potato late blight disease :

Chemical profiles drive biological activities

Florian MARTINI[1,2,3,4], Clément BURGEON[1,4], Jérôme MUCHEMBLED[2], Eric GONTIER[3], M. Haïssam JIJAKLI[4] and Marie-Laure FAUCONNIER[1]

[1] *Laboratory of Chemistry of Natural Molecules, Gembloux Agro-Bio Tech, Liege University, Belgium*
[2] *Laboratory of Plant Pathology and Biocontrol, ISA JUNIA, Lille, France*
[3] *Laboratory of Plant Biology and Innovation, University of Picardie Jules Verne, Amiens, France*
[4] *Laboratory of Integrated and Urban Plant Pathology, Gembloux Agro-Bio Tech, Liege University, Belgium*

1. Introduction

1.1. Agronomical context

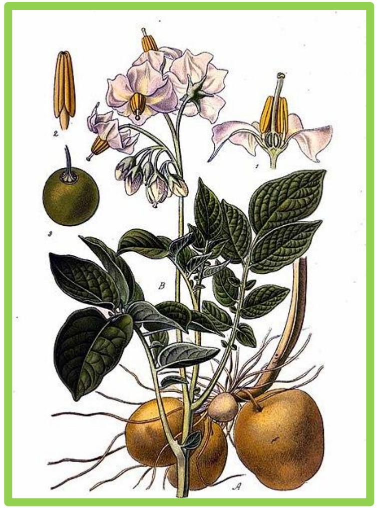
Solanum tuberosum
L., 1753

Perennial plant, *Solanaceae*

Grown as an annual crop
(almost) all around the globe

High ratio of eatable dry matter (>80%)

Tubers rich in starch and micronutrients



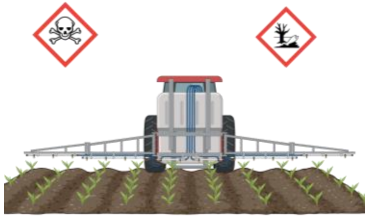
350 Mt | **200 Mha** | **17,5 t/ha**

Devaux *et al.*, 2020

1. Introduction

1.2. The agent of potato late blight

Phytophthora infestans
(Mont.) de Bary, 1876



Synthetic pesticides

10-20x **≈6B$**

Oomycota « pseudofungi »

- Cellulosic cell wall
- Plasma membrane without ergosterol
- Biflagellated spores : zoospores

➢ Disseminated through zoospores in humid conditions
➢ Sexual reproduction gives very resilient oospore

Phylogenetically closer from Algae than from Fungi

Fry *et al.*, 2015
Grünwald *et al.*, 2005
Gavino *et al.* 2000

1. Introduction

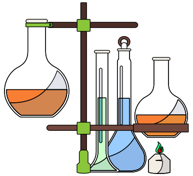
1.3. Essential oils as alternative control for late blight

Increasing resistant populations causes decreasing fungicide efficacy

Conventionnal pesticides persist in the environment causing air, water and soil pollution

Synthetic molecules are made out of unrenewable ressources

As an alternative :



Essential oils are hydrophobic complex mixtures of secondary metabolites - mainly volatile organic compounds (VOC's)- obtained from aromatic plants exclusively by certain types of extraction methods such hydrodistillation or cold expression.

Fometu *et al.*, 2019
Slavkovic *et al.*, 2023

1. Introduction

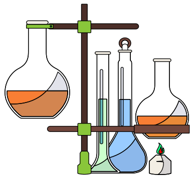
1.3. Essential oils as alternative control for late blight

Complex composition allows diverse modes of action

Easier to get biodegraded and less persistant in the environment

Extracted from plant biomass *i.e* renewable source

As an alternative :



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Fometu *et al.*, 2019
Slavkovic *et al.*, 2023

2. Material & Method

2.1. Experimental material

Pathogen

Three *P. infestans* genotypes commonly found

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

Biocontrol tool

Five essential oils chosen for botanical and chemical diversity

- *Citrus bergamia*
- *Rosmarinus officinalis*
- *Syzygium aromaticum*
- *Origanum vulgare*
- *Cinnamomum verum*

Host plant

Three *S. tuberosum* varieties with ≠ sensitivity to late blight

- Bintje (2/9)
- Fontane (4/9)
- Carolus (8/9)

Martini *et al.*, 2023

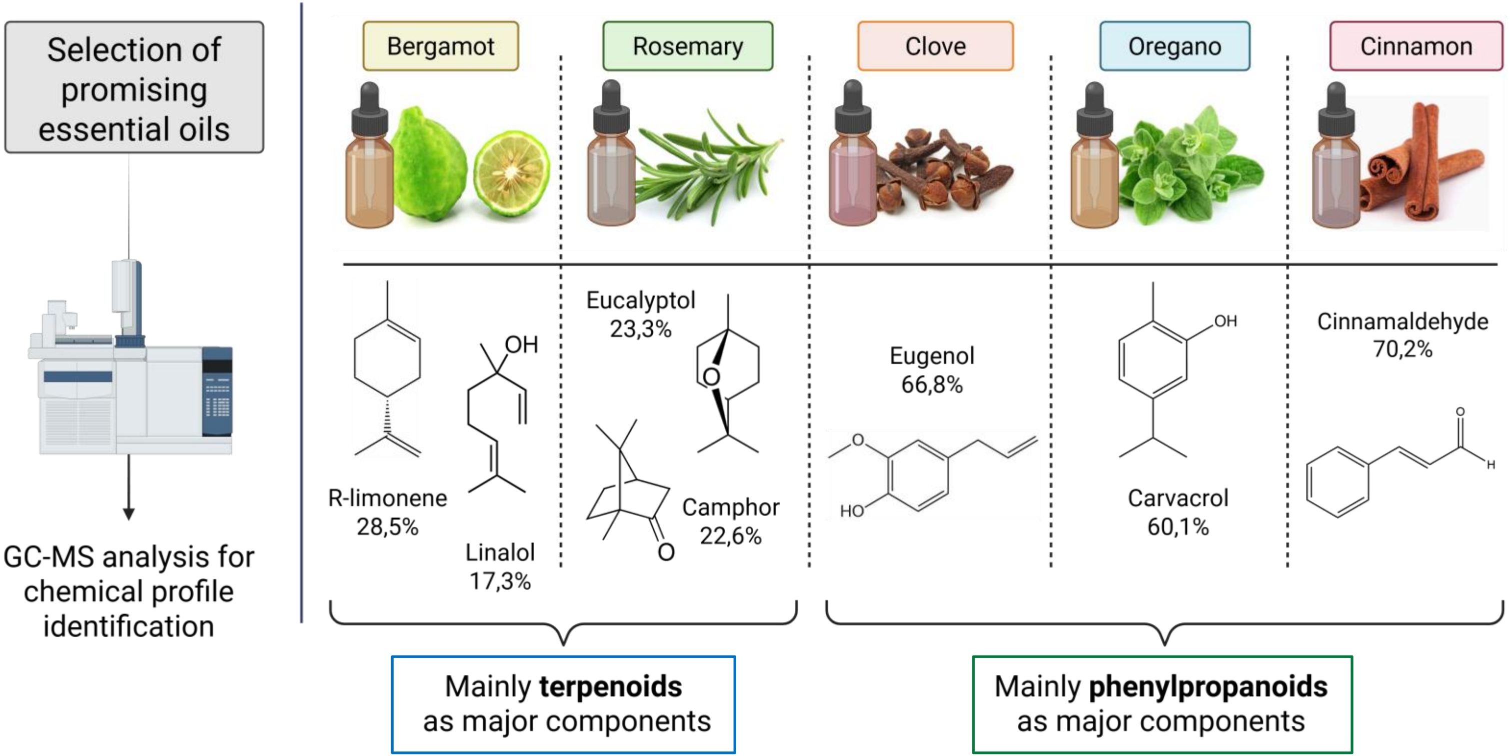
2. Material & Method

2.2. Experimental methodology

1. Essential oils chemical profil
2. Inhibition potential on mycelium growth
3. Inhibition potential on spores germination
4. Essential oils phytotoxicity tresholds
5. Most pertinent plant-pathogen couple
6. Efficacy test against late blight disease

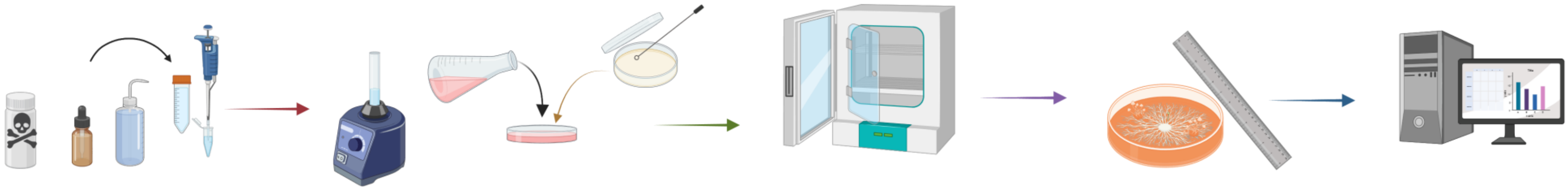
3. Results

3.1. Essential oils chemical profile



3. Results

3.2. Mycelium growth inhibition (MIC)



Emulsification of ≠ [EO] in water with 1:5 (v/v) Tween 20 and solubilization of synthetic fungicide in DMSO

Successive dilutions of EO emulsions in V8-agar culture medium

Inoculation with 0,5 cm ø mycelium plug and incubation of Petri dishes for 8 days at 18°C

Measurements of radial mycelium length

Determination of minimal inhibitory concentration (MIC) for each substance

Adapted from Soylu *et al.*, 2006

3. Results

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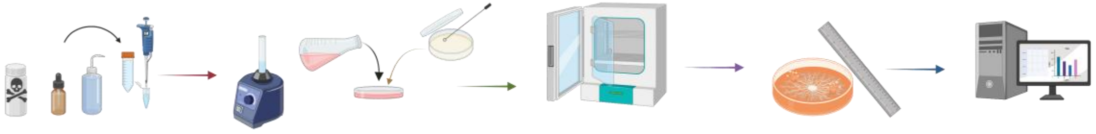
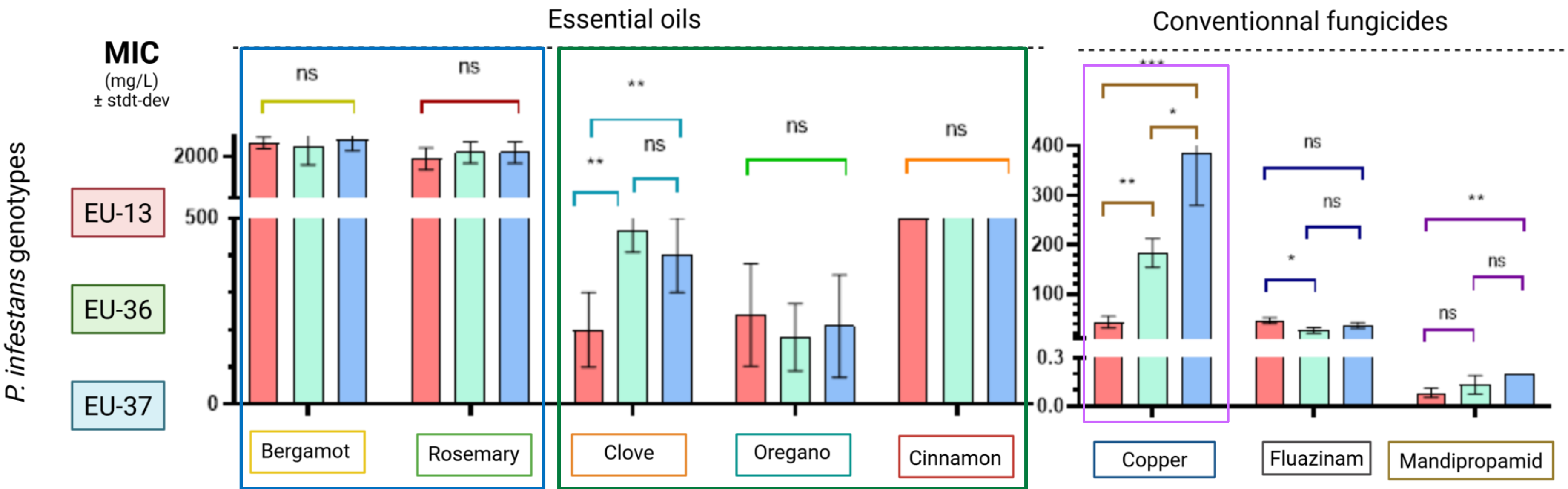
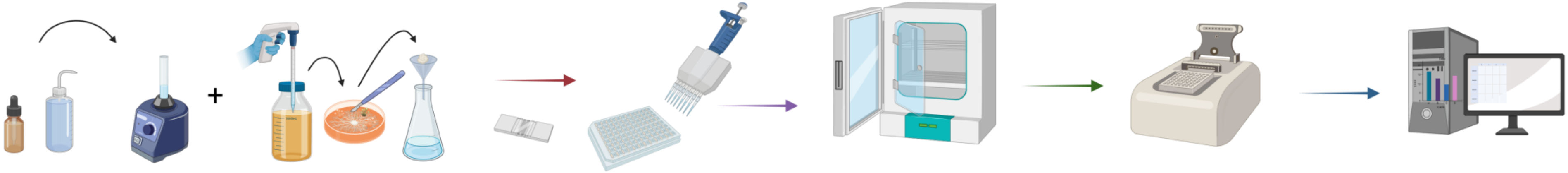


MIC on mycelium growth after 8 days of incubation on solid medium treated with essential oils or conventional fungicides on Petri dishes



3. Results

3.3. Spores germination inhibition (IC_{50})



Emulsification of ≠ [EO] with Tween20 in glucose-peptone culture medium

Inoculation with a aqueous spore suspension at around 5*10[4] sporocyste/mL

Incubation of microplates for 7 days under 120 rpm at 18°C in the dark

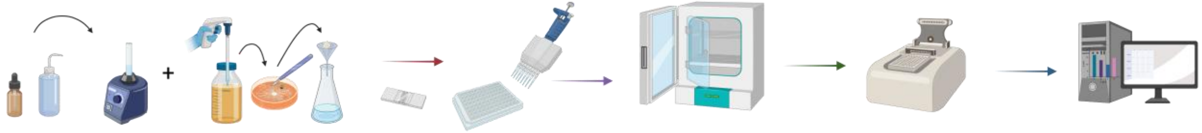
Reading of OD at 630nm to assess spores germination

Determination of IC_{50} according to a sigmoidal model

Adapted from Deweer *et al.*, 2023 and De Clerck *et al.*, 2020

3. Results

3.3. Spores germination inhibition (IC_{50})

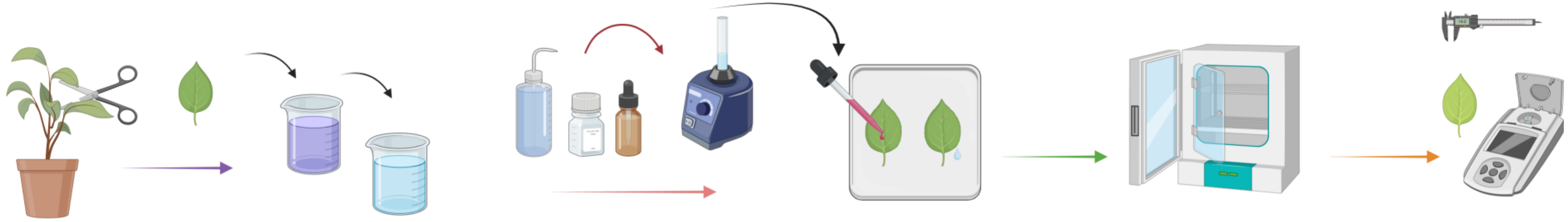


IC_{50} on zoospores germination after 7 days of incubation in liquid media treated with essential oils or conventional fungicides on microplates

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 ; 0.2]	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]

3. Results

3.4. Determination of phytotoxicity tresholds



Sampling of potato leaves from intermediate foliage stages

Successive rinsing in sterile water baths

Drop deposit of 20µL of ≠ [EO] emulsified with 1/5 (v/v) of Tween20 in water

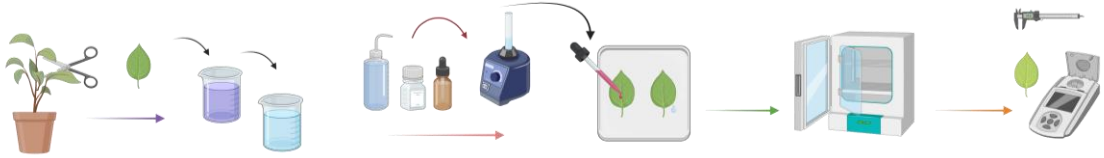
Incubation for 4 days at 18°C, 8/16h photoperiod and saturating humidity

Fluorescence measurements and necrosis observations

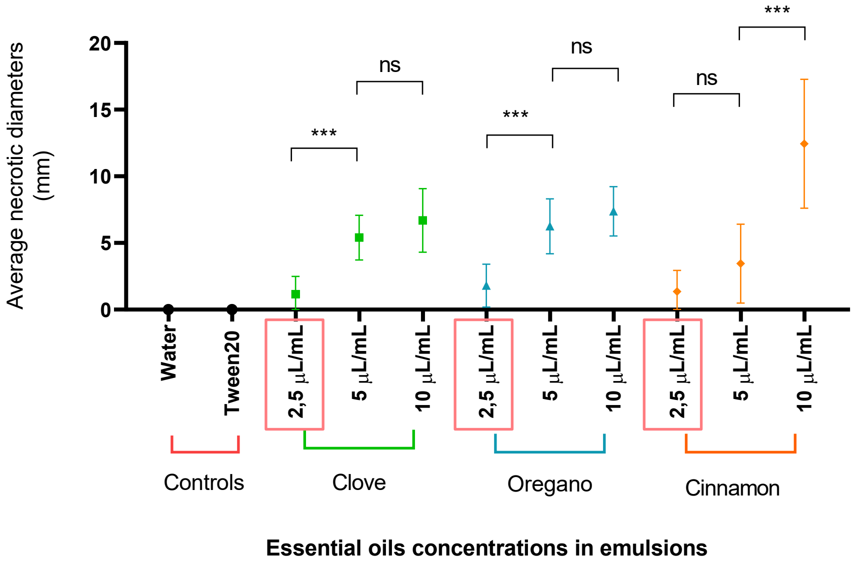
Werrie *et al.*, 2019

3. Results

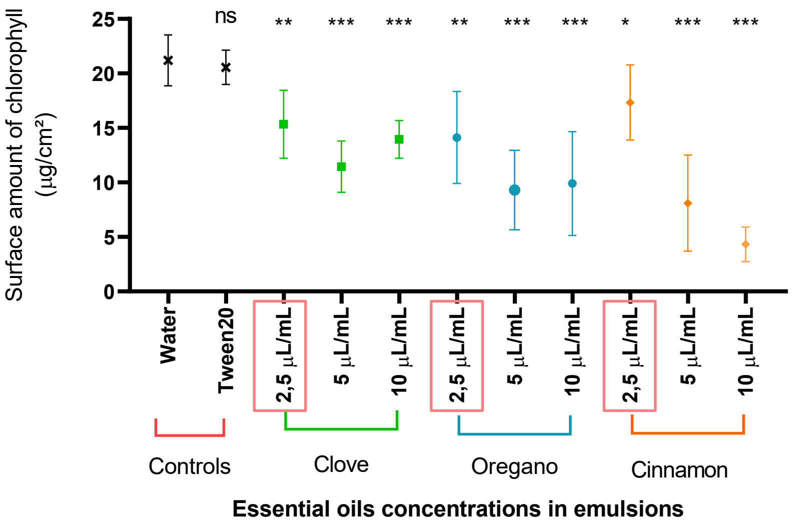
3.4. Determination of phytotoxicity tresholds



Necrotic diameters on potato detached leaves after 4 days with drop-application of different concentrations of essential oils emulsions

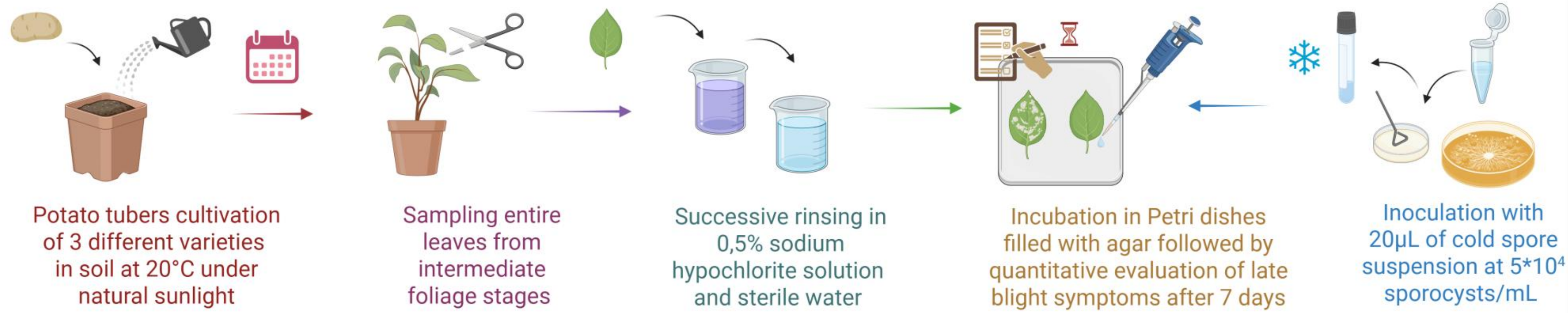


Surface amount of chlorophyll in potato detached leaves after 4 days with drop-application of different concentrations of essential oils emulsions



3. Results

3.5. Plant-pathogen compatibility



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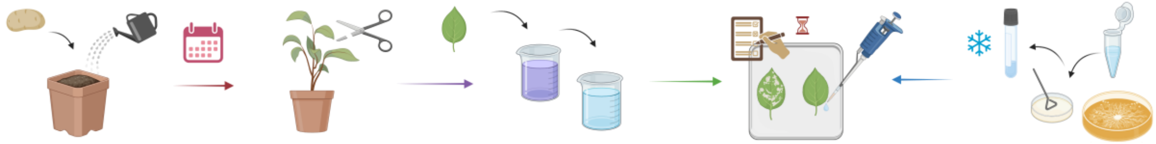
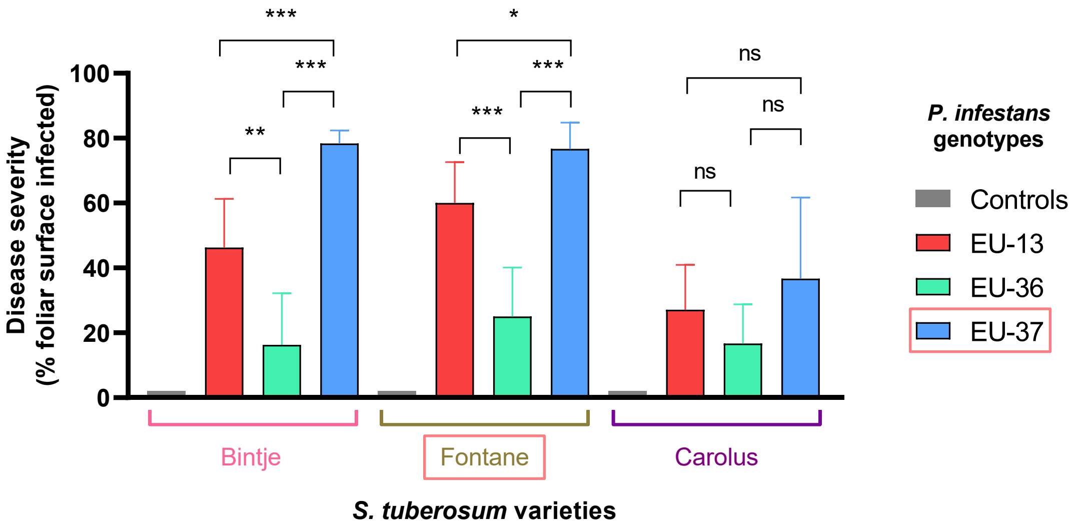
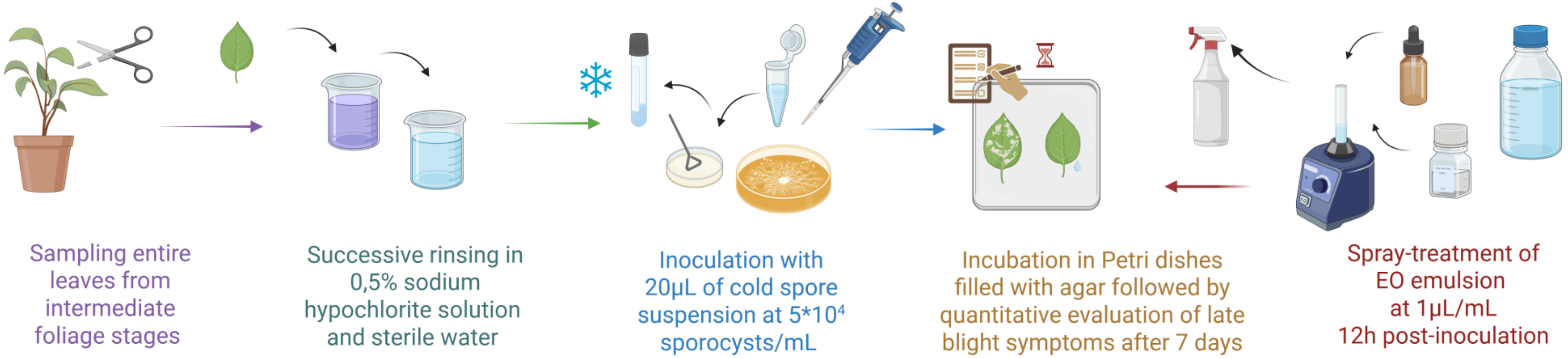


Severity of late blight early symptoms caused by three different genotypes of *P. infestans* on detached leaves of three varieties of potato after 7 days of sporal infection



3. Results

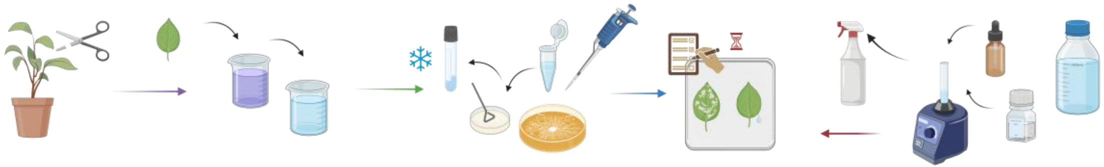
3.6. Essential oils protective effects against late blight



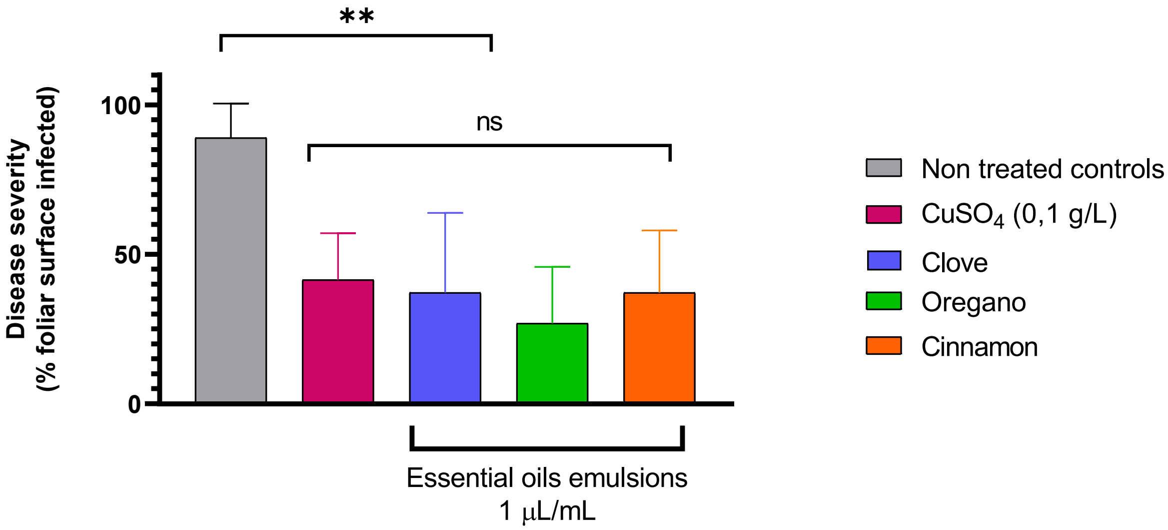
Adapted from Najdabbasi *et al.*, 2020

3. Results

3.6. Essential oils protective effects against late blight



Severity of late blight early symptoms 7 days after sporal infection by EU-37 genotypes of *P. infestans* on Fontane detached leaves treated 12 hours post-inoculation with essential oils emulsions

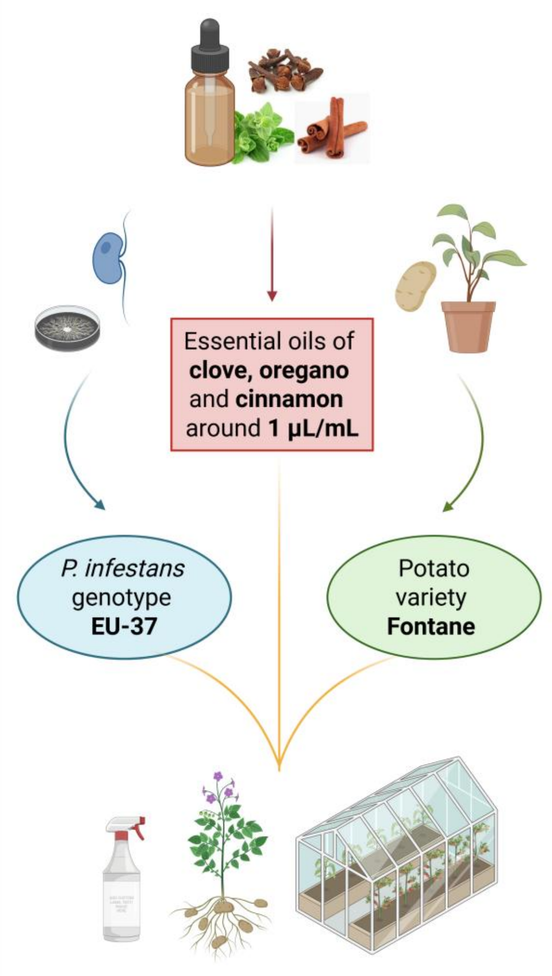


4. Conclusion & Perspectives

Essential oil potential activity against late blight disease must be confirmed *in planta* on a longer kinetic and semi-controlled conditions

Appropriate formulation is recquired in order for EO to fully express their inhibitory potential in agronomical conditions

Modes of action at the cellular scale are still to be further explored to comprehend what the real targets are both on the pathogen, and on the induction of plant resistance



Maes *et al.*, 2019
Martini *et al.*, 2023
Wang *et al.* 2019

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Thank you for your attention!

I’d be happy to answer questions.