

The effectiveness of plant essential oils on the *in vitro* growth of postharvest phytopathogenic fungi



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Background and objectives

Plant extracts, such as essential oils (Eos) [1], which have long been used in traditional preparations, are currently adopted by the industrial manufactures for modern formulations of consumer products. They are found in perfumery and cosmetology as well as in food and pharmaceutical industries. In agriculture, it was reported that Eos could be an interesting alternative to chemical fungicides and could be used as biofungicides in postharvest biological control. The present work was therefore undertaken to study the effect of thirty species of EOs on the *in vitro* growth of *Penicillium italicum*, *Penicillium digitatum* infecting citrus (fig.1 & fig.2) and *Colletotrichum musea* infecting bananas (fig.3).



Figure 1 : Blue mold (*P. italicum*)



Figure 2 : Green mold (*P. digitatum*)



Figure 3 : Anthracnose (*C. musea*)

Material and methods

Essential oils were selected according to intrinsic (yield, phytotoxicity) and extrinsic (availability, cost, popularity) criteria [2], and were classified according to their main constituents (table 1). Their effect on the growth of three pathogens was evaluated with decreasing concentrations of application, using a microdilution method on a 96-well microplate Elisa (figure 4). Only essential oils inhibiting more than 70 % of the growth of all the fungi were retained for the following test. For each pathogen, growth was followed by recording each day the optical density (OD) at 490 nm of a solution of 200 µl containing: diluted orange juice (0,03 v/v); 10⁴ conidia/ml; essential oil (1000, 500 and 100 ppm).

The experimental design is shown in figure 4. For each pathogen, two independent experiments were performed with 8 replicates per essential oil. The mean % of inhibition of essential oils was calculated as follows:

$$[\text{OD}(\text{X}_n) - \text{OD}(\text{T}_n)] - [\text{OD}(\text{X}') - \text{OD}(\text{T}')] / [\text{OD}(\text{X}') - \text{OD}(\text{T}')] \times 100$$

Mean % inhibition =

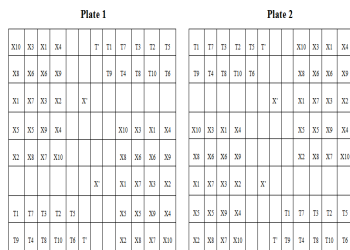
$$[\text{OD}(\text{X}_n) - \text{OD}(\text{T}_n)] - [\text{OD}(\text{X}') - \text{OD}(\text{T}')] / [\text{OD}(\text{X}') - \text{OD}(\text{T}')] \times 100$$

[OD (X_n) – OD (T_n)] = OD of the pathogen in presence of EO

[OD (X') – OD (T')] = OD of the pathogen in absence of EO

Table 1: Essential oils

Main constituents of essential oils (terpenes and aromatic compounds)	Essential oils*	Origin	Part of the plant distilled
Non-organised compounds	1	India	Leaf
	2	Brazil	Leaf
	3	Ceylon	Leaf
Alcohols	4	Basil	Flowering top
	5	Peppermint	Leaf
	6	Spearmint	Leaf
	7	Geraniol	Flowering top
	8	Geraniol	Flowering top
Aldehydes	9	Citrus	Leaf
	10	Citrus	Leaf
	11	Citrus	Leaf
Ketones	12	Ylang-ylang	Flowering top
	13	Ylang-ylang	Flowering top
	14	Ylang-ylang	Flowering top
Phenols	15	Thyme	Flowering top
	16	Thyme	Flowering top
	17	Thyme	Flowering top
Esters	18	Orange	Leaf
	19	Orange	Leaf
	20	Orange	Leaf
Ether esters	21	Ylang-ylang	Flowering top
	22	Ylang-ylang	Flowering top
	23	Ylang-ylang	Flowering top



Ye: Solution with microorganism with EO
 Te: Solution without microorganism with EO

X: Solution with microorganism without EO
 T: Solution without microorganism and EO

Figure 4 : Experimental plan

Results

- ❖ Only recorded data of the growth period 168-192 h are shown in Figure 5.
- ❖ Whatever the essential oil tested and whatever the concentration (except EO 20 tested at 500 ppm), there was a growth inhibition regardless of the species.
- ❖ In general, the three pathogens present different sensitivity levels towards the EOs. *C. musea* seems to be more sensitive than the tested *Penicillium* species.
- ❖ All pathogens appear to be more sensitive to EOs composed of alcohol, aldehyde or phenol.
- ❖ Among the 30 essential oils tested, 12 were effective against all the fungi at 1000 ppm and 3 at 100 ppm.

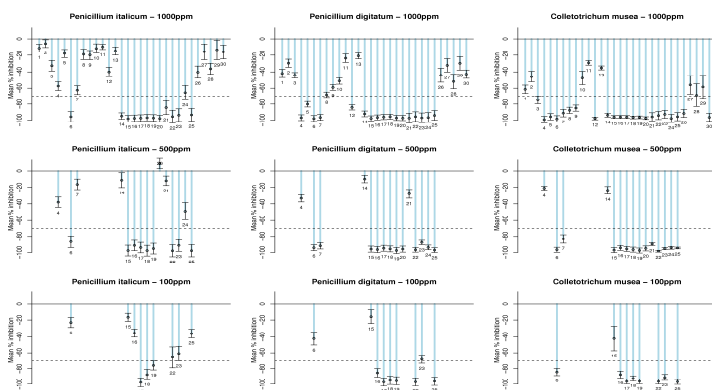


Figure 5: Mean growth inhibition (%) of essential oils against *P. italicum*, *P. digitatum* and *C. musea*

Conclusion

The present preliminary work shows that essential oils are able to partially or totally inhibit the growth of three different post-harvest fungal pathogens. This is a first encouraging study for the development of biofungicides based on essential oils as an alternative to chemical fungicides.

References

- [1] Ernest Guenther. The essential oils - vol 1: History – Origin In Plants – Production – Analysis. Read Book (2007) 452 Pages.
- [2] Pranarôm International S.A, expert en aromathérapie scientifique et médicale, Tarif export 2008.