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# Bioactivity of essential oils in phytopathogenic and post-harvest fungi control

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| Keywords: | lavender, thyme, essential oils, antifungal activity |
| | |

Here there are the Reviewer(s)' Comments to Author:

Reviewer: 1

Comments to the Author
The paper reports the antifungal activity of two essential oils.
Some major and minor poisnt should be addressed.

Major points:
1. Identification of components of the essential oils. Authors used 1 column to identify the EO components.
The identification can be doubteful, namely in sesquiterpene region. In my opinion, two columns of different polarity should be used.

The reviewer is right to indicate that the greatest problem in identifying an essential oil occurs in the sesquiterpene zone, and in these cases using columns of different polarities is recommended, or better still, even a change in the temperature gradient. However as seen in Table S1, both essential oils are rich in monoterpenes, mainly oxygenated monoterpenes. The sesquiterpenic fraction is formed by hydrocarbon and oxygenated sesquiterpenes, which are very common in essential oils, and with a very characteristic mass spectrum. So it was not necessary to use columns of different polarities for these essential oils.

2. In Table S1 please add, FOR EACH COMPONENT, the method of identification (MS, co-injection, etc.). An identification based only on data bank is unacceptable.

The method of identification for each component has been added in Table S1.
The identification has not been made solely by the spectrum of the database of the equipment used, but also as indicated in material and methods using their RIs, relative to $C_8$-$C_{32}$ *n*-alkanes of each peak with RI of the literature (Adams 2007) and with mass spectra of authentic samples as well as mass spectral of the literature.

3. Authors should discuss the antifungal activity of EOs taking into account their composition.

Thank you for your comment. We have added two paragraphs about essential oil contents and antifungal activities.

In reference to the Thyme essential oil:
Between four chemotypes of *T. zygis*, one of them, chemotype thymol (23%)/*p*-cymene/γ-terpinene, showed poor antifungal activity against dermatophyte fungi and storage fungi (Gonçalves et al. 2010). However *T. zygis* here analysed (thymol 52%) showed 90-100% inhibition of fungal growth. These results corroborated previous works (Santamarina et al. 2015) that high antifungal activity is relate to a high percentage of thymol, phenolic compound recently recognized as a fungicide.

In reference to the Lavender essential oil:
The obtained results are in accordance with others authors (Erland et al., 2016) about the Lavender EO, in which a high linalool and linalyl acetate content, showed also poor antifungal activity against three important agricultural pathogens: *Botrytis cinerea*, *Mucor piriformis* and *Penicillium expansum*.

These sentences about the antifungal activity of EOs have been added to the Results and Discussion.

4. In Table S1, a SD is reported, but in Experimental no number of experiments is reported.

Thank you for your comment. A sentence has been added as a footnote in Table S1. (peak area values are the means±standard deviation of three samples).

5. Figure S1 is useless, being a repetition of the Table.

Figure S1 has been deleted as it is indeed repetitive.

6. Please, identify correcly the plants: In the paper, L.angustifolia L. (sic) and T. zygis L. (sic) are cited. The names are different in Supplementary Material (L.angustifolia Miller. (sic with point) and T. zygisBrot.

Thank you again for your comment. The scientific correct names are *Thymus zygis* Boiss. and *Lavandula angustifolia* Mill.
The names have been corrected in the manuscript and in Table S1.

Minor points
English grammar and style need revision.

The English manuscript has been reviewed by a native proofreader. I attach certificate

Reviewer: 2

Comments to the Author
Comment

This manuscript entitled "Bioactivity of essential oils in phytopathogenic and post-harvest fungi control" described the identification of biological activity and essential oil components of commercial thyme and lavender essential oils. After my careful reading of this manuscript, there were some important suggestions I have to mention.

- The manuscript had the interesting subject. It is good for publishing in this journal, but the manuscript had many grammatically mistakes especially about the time of sentences and plural and singular verbs, which some of them are highlighted in the text.
Moreover some sentences are not clear. You should review all the manuscript and improve your manuscript grammatically and change unclear sentences.

English manuscript has been reviewer by a native proofreader. I attach certificate

- I want to know that why you select these two plants and explain the innovation of your manuscript.

These two plants, Lavender and Thyme, were selected because they are characteristic species of the Mediterranean Region, to control the fungi that cause vast losses in Mediterranean crops, for sustainable agriculture. In previous studies, we found that EO

rich in phenolic compounds (eugenol) were the most active, and in the present work we wanted to test EOs rich in other phenols (thymol), and compare them with others rich in other oxygenated monoterpenes.

This sentence about the selection of these two plants and manuscript innovation has been added to the Introduction.

- In Material and Methods, authors didn't mention the voucher specimens, the condition of storage essential oils and experimental design about antifungal activity.

The authors did not mention voucher specimens because we did not collect and identify the species to obtain essential oil since we directly purchased essential oil from "Plantis Artesania Agrícola".
A sentence about the storage condition has been added to the Plant Material of the Supplementary Material (Commercial essential oils were stored in the darkness and in a refrigerator at 4°C until analysed by GC/MS or antifungal activity evaluation).

- In result and discussion section, you should add some relevant paragraph from previous studies about essential oil contents and antifungal activities of these two essential oils. Also, you report the chemical composition as mean ± SD, but about MGI, in spite of ten repetitions you reported without SD. Pleaseexplainwhy?

Thank you for your comment. We have added two paragraphs about essential oil contents and antifungal activities.

In reference to the Thyme essential oil:
Between four chemotypes of *T. zygis*, one of them, chemotype thymol (23%)/*p*-cymene/γ-terpinene, showed poor antifungal activity against dermatophyte fungi and storage fungi (Gonçalves et al. 2010). However *T. zygis* here analysed (thymol 52%) showed 90-100% inhibition of fungal growth. These results corroborated previous works (Santamarina et al. 2015) that high antifungal activity is relate to a high percentage of thymol, phenolic compound recently recognized as a fungicide.

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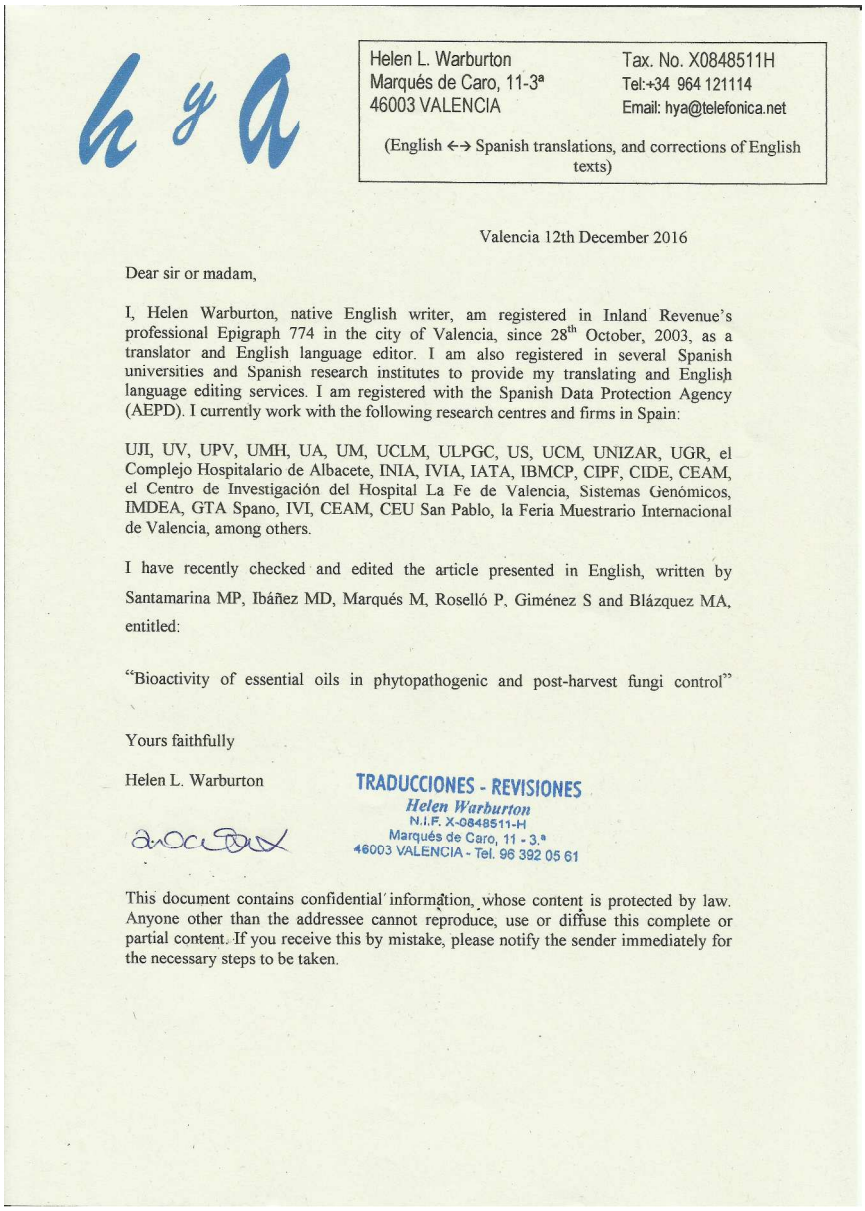
The obtained results are in accordance with others authors (Erland et al., 2016) about the Lavender EO, in which a high linalool and linalyl acetate content, showed also poor antifungal activity against three important agricultural pathogens: *Botrytis cinerea*, *Mucor piriformis* and *Penicillium expansum*.

These sentences about the antifungal activity of EOs have been added to the Results and Discussion.

In Table 2 we have added growth means and their standard deviation, MGI is a proportion, percentage of growth inhibition.



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(English ↔ Spanish translations, and corrections of English texts)

Valencia 12th December 2016

Dear sir or madam,

I, Helen Warburton, native English writer, am registered in Inland Revenue's professional Epigraph 774 in the city of Valencia, since 28th October, 2003, as a translator and English language editor. I am also registered in several Spanish universities and Spanish research institutes to provide my translating and English language editing services. I am registered with the Spanish Data Protection Agency (AEPD). I currently work with the following research centres and firms in Spain:

UJI, UV, UPV, UMH, UA, UM, UCLM, ULPGC, US, UCM, UNIZAR, UGR, el Complejo Hospitalario de Albacete, INIA, IVIA, IATA, IBMCP, CIPF, CIDE, CEAM, el Centro de Investigación del Hospital La Fe de Valencia, Sistemas Genómicos, IMDEA, GTA Spano, IVI, CEAM, CEU San Pablo, la Feria Muestrario Internacional de Valencia, among others.

I have recently checked and edited the article presented in English, written by Santamarina MP, Ibáñez MD, Marqués M, Roselló P, Giménez S and Blázquez MA, entitled:

"Bioactivity of essential oils in phytopathogenic and post-harvest fungi control"

Yours faithfully

Helen L. Warburton

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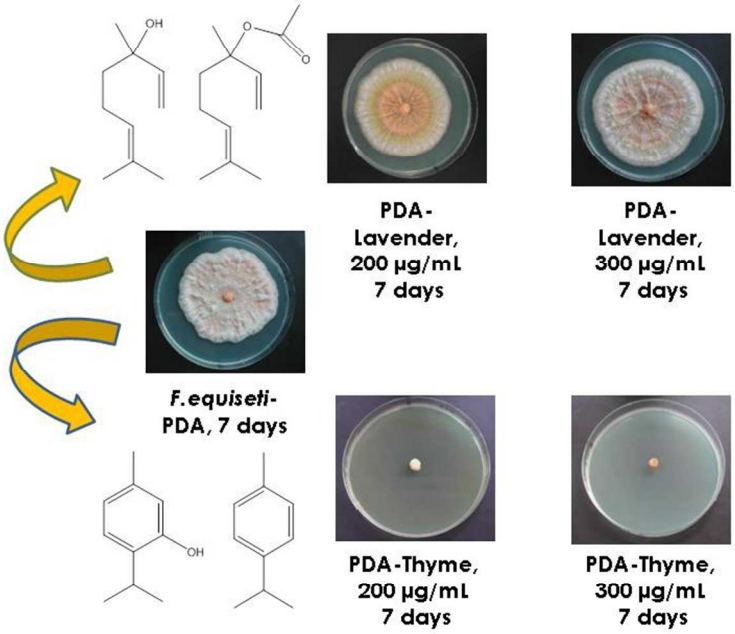


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**Presented at CIPAM 2016. Abstract number P159, page 251**

**SHORT COMMUNICATION**

# Bioactivity of essential oils in phytopathogenic and post-harvest fungi control

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# Bioactivity of essential oils in phytopathogenic and post-harvest fungi control

Commercial thyme and lavender essential oils were analysed by GC/MS. Sixty-six compounds accounting for 98.6-99.6% of total essential oil were identified. Thymol (52.14±0.21%), followed by *p*-cymene (32.24±0.16%), carvacrol (3.71±0.01%) and γ-terpinene (3.34±0.02%), were the main compounds in thyme essential oil, while large amounts of oxygenated monoterpenes linalool acetate (37.07±0.24%) and linalool (30.16±0.06%) were found in lavender one. *In vitro* antifungal activity of the essential oils was evaluated at 200 and 300 µg/mL against ten phytopathogenic and post-harvest fungi, which significantly affect agriculture. Micelial growth inhibition was calculated for each tested fungus and dose. Thyme essential oil showed satisfactory results with 90-100% growth inhibition in almost all the assayed fungi at 300 µg/mL, while Lavender essential oil showed no noteworthy inhibition data at either dose, and its growth was even enhanced. Thyme essential oil represents a natural alternative to control harvest and post-harvest fungi, and to extend the shelf-life of agriculture products.

**Keywords**: lavender; thyme; essential oils; antifungal activity.

## 1. Introduction

Fungal contamination extremely affects crops by reducing the quality and quantity of their products. Phytopathogenic fungi produce nearly 20% of losses of vast economical importance in harvests and post-harvest products (Santamarina et al. 2015). Furthermore, badly preserved foodstuff is also exposed to fungal spoilage, frequently through mycotoxins production. These relatively small molecules trigger mycotoxicosis, which involves a set of diseases and disorders that potentially affect animals and humans (Sumalan et al. 2013).

Several studies have shown that essential oils (EOs) perform antibacterial and antifungal activities (Moghaddam et al. 2015), which are simultaneously safer for both human health and the environment, and are consequently more acceptable by consumers. The Lavender (*Lavandula angustifolia* Mill.) essential oil contains principally linalool and linalyl acetate, together with moderate levels of lavandulyl acetate, terpinen-4-ol and lavandulol (Dupuy et al. 2014). The genus *Thymus* L. is widely distributed in the Iberian Peninsula, is a taxonomically complex group, and has been traditionally used as a spice or medicinal plant. The Thyme (*Thymus zygis* Boiss.) EO contains a large amount of phenolic compounds, thymol and/or carvacrol, and its biogenetic precursors, *p*-cymene and γ-terpinene. It is widely used in the the food industry for its antimicrobial activity against food-borne pathogens (Blázquez, 2014). It is also employed to prevent or delay oxidation reactions, by maintaining food quality for longer periods due to the antioxidant activity of its phenolic compounds. These two plants, Lavender and Thyme, were selected because they are characteristic species of the Mediterranean Region, to control the fungi that cause vast losses in Mediterranean crops, for sustainable agriculture. In previous studies, we found that EO rich in phenolic compounds (eugenol) were the most active, and in the present work we wanted to test EOs rich in other phenols (thymol), and compare them with others rich in other oxygenated monoterpenes.

Due to the chemical polymorphism that occurs in aromatic plants, it is fundamental to know their chemical composition and to perform the isolation of specific phytopathogenic strains. So, the aims of this study were to analyse the chemical composition of the commercial thyme and lavender EOs and to determine their '*in vitro*' antifungal activity against ten harvest and post-harvest phytopathogenic fungi: *Alternaria alternata, Bipolaris spicifera, Rhizoctonia solani*, *Colletotrichum gloeosporoides*, *Curvularia hawaiiensis*, *Fusarium oxysporum* fsp. *lycopersici*, *Fusarium equiseti, Fusarium graminearum*, *Penicillium expansum* and *Penicillium italicum*.

## 2. Results and Discussion

### *2.1. Chemical composition of the commercial essential oils*

The chemical composition of the commercial Thyme and Lavender EOs was determined by a GC/MS analysis. A homologous series, listed according to Kovat's retention index, of monoterpene hydrocarbons, oxygenated monoterpenes, sesquiterpene hydrocarbons, oxygenated sesquiterpenes, and others, is offered in Table S1. Sixty-six compounds were identified, which accounted for between 98.6-99.6% of the total EO. Although oxygenated monoterpenes were the main phytochemical group in both the commercial EOs, phenolic compounds thymols (52.14±0.21%), followed by their isomer carvacrol (3.71±0.01%), were the main compounds in the *T. zygis* EO, whereas large amounts of the oxygenated monoterpenes linalool acetate (37.07±0.24%) and linalool (30.16±0.06%), were found in the *L. angustifolia* EO.

Regarding *T. zygis,* chemotypes thymol and carvacrol both showed antifungal activity, and have been described in subspecies *gracilis* and *sylvestris* (Gonçalves *et al*., 2010). In Southern Spain, seven main chemotypes, these being thymol, carvacrol, linalool, α-terpinyl acetate, thymol/*p*-cymene/γ-terpinene, 1,8-cineole/myrcene/spathulenol and 1,8-cineole/α-terpineol, with no chemotaxonomic differentiation between both subspecies (*gracilis* and *sylvestris*) for *T. zygis,* have been previously found (Pérez-Sanchez et al. 2008).

In the commercial Thyme analysed herein, chemotype thymol/*p*-cymene/γ-terpinene was found in the largest quantities of thymol (52%), followed by the identified biogenetic precursors *p*-cymene (32.24±0.16%) and γ-terpinene (3.34±0.02%). Among the other identified compounds, oxygenated monoterpenes carvacrol (3.71±0.01%), linalool (2.26±0.03%) and borneol (1.97±0.01%) reached percentages above 1%. Only two sesquiterpene hydrocarbons, β-caryophyllene (0.14%) and α-humulene (0.02%), and two oxygenated sesquiterpenes, caryophyllene oxide (0.31%), and humulene epoxide (0.02%), were detected.

Fifty compounds were identified in the *L. angustifolia* EO. Several studies have shown that its EOs are characterised by a high linalool and linalyl acetate level, moderate levels of lavandulyl acetate, terpinen-4-ol and lavandulol, and by a very low to moderate amount of 1,8-cineole and camphor (Dupuy et al. 2014). Both, the EO of *L. angustifolia* and its main compounds linalool and linalyl acetate, perform antifungal activity, and a significant difference was found among lavender, clotrimazole and the control group at higher dilutions (Adam et al. 1998). The average fungal cell count after 48 h was lower in the Lavender group compared to the isolated *C. albicans* from vaginal candidiasis.

Linalyl acetate (37.07±0.24%) and linalool (30.16±0.06%) were the main compounds found in the commercial sample used herein. Relatively large amounts of terpinen-4-ol (3.43±0.02%) and

lavandulyl acetate (3.01±0.01%), with low levels of 1,8-cineol (1.07±0.02%) and lavandulol (0.74%), and a very low level of camphor (0.23%), were found between the oxygenated monoterpenes. Moderate levels of *cis*-ocimene (4.36±0.07%) and *trans*-ocimene (2.18±0.03%), together with β-caryophyllene (4.44±0.01%) and *trans*-β-farnesene (3.00±0.03%), were observed among the 16 monoterpene hydrocarbons and the seven sesquiterpene hydrocarbons identified, respectively. Only two oxygenated sesquiterpenes were detected, caryophyllene oxide (0.30%), and *epi*-α-cadinol (0.09%). Finally, in the EO used in the biological assays, 2.48% of the total EO corresponded to low-molecular-weight aliphatic compounds in the form of alcohols, ketone ethers and esters.

### *2.2. Antifungal activity*

The *T. zygis* EO was more active than *L. angustifolia* against the ten harvest and post-harvest phytopathogenic fungi, and obtained MGI values that ranged from 90% to 100% in almost all the assayed fungi. At 300 µg/mL, growth was inhibited between 88.7% and 100%. *P. italicum* was the most resistant fungus. *R. solani, F. equiseti, B. spicifer*a and *C. hawaiiensis* were the most susceptible fungi with 95-100% growth inhibition at the lower assayed dose (200 µg/mL) (Table S2). The Tukey HSD intervals used to compare the means showed significant differences in fungal growth between the control and the Thyme oil. Both doses were equally effective in *R. solani, F. equiseti, B. spicifer*a and *C. hawaiiensis*.

This was due to its richness in phenolic compounds thymol and carvacrol, which have already obtained very good results against phytopathogenic fungi isolated from rice, such as *F. culmorum* and *F. verticillioides* (Roselló et al. 2015). Between four chemotypes of *T. zygis*, one of them, chemotype thymol (23%)/*p*-cymene/γ-terpinene, showed poor antifungal activity against dermatophyte fungi and storage fungi (Gonçalves et al. 2010). However *T. zygis* here analysed (thymol 52%) showed 90-100% inhibition of fungal growth. These results corroborated previous works (Santamarina et al. 2015) that high antifungal activity is relate to a high percentage of thymol, phenolic compound recently recognized as a fungicide.

In fact, thymol has been recently approved (Reg. (EU) No. 568/2013) by the European Food Safety Agency as a fungicide substance. Furthermore, it is widely used to control other food diseases caused by *Pseudomonas aeruginosa* ATCC 27853, *Salmonella enteritidis* CECT 4155, *S. typhimurium* CECT 443, *S. enterica* Enteritidis S64, *Escherichia coli* ATCC 25922, *E. coli* serovar O157:H7 CECT 4267, *Yersinia enterocolitica* serotype O:8; biotype 1 CECT 4315, *Shigella sonnei* CECT 457, *S. flexneri* serovar 2a CECT 585, *Listeria monocytogenes* ATCC 19117, *L. monocytogenes* serovar 4b CECT 935, *Staphylococcus aureus* ATCC 25923 and *S. aureus* CECT 239 (Millezi et al. 2012).

The Lavender EO displayed no antifungal activity in the fungi assayed at both tested doses: 200 and 300 µg/mL. Growth of *R. solani*, *F. equiseti*, *B. spicifera, F. oxysporum lycopersici*, *F. graminearum* or *A. alternata* was not inhibited by this EO, but it even enhanced the growth (20-50%) of certain species, in particular *Penicillium* spp. and *C. hawaiiensis*. *C. gloeosporoides* was the only fungus to obtain positive MGI values. So the Lavender EO had no significant effect on fungi inhibition. The obtained results are in accordance with others authors (Erland et al., 2016) about the Lavender EO, in which a high linalool and linalyl acetate content, showed also poor antifungal activity against three important agricultural pathogens: *Botrytis cinerea*, *Mucor piriformis* and *Penicillium expansum*.

## 3. Conclusions

The Lavender EO did not inhibit the growth of the studied fungi at the assayed doses, but even enhanced growth in some cases. The Thyme EO displayed excellent mycelial growth inhibition, of nearly 100%, in almost all the tested fungi and at both doses. Hence according to their chemical composition, their activity revealed that the Lavender EO contained oxygenated monoterpenes linalool acetate (37.07%) and linalool (30.16%) as the main compounds, whereas the main oxygenated monoterpenes in the Thyme EO were thymol (52.14%) and its isomer carvacrol (3.71%). The Thyme EO constitutes an attractive alternative to control fungal development, most of which are mycotoxigenic fungi, and could be used to extend the shelf-life of harvest and post-harvest products.

## Acknowledgements

This study has been financed by the Spanish Ministry of Economy and Competitiveness, Programme-oriented Societal Challenges 2014-2017 (reference number AGL2013-42989-R-AR).

## Supplementary Material

The experimental details are available as Supplementary Material, along with Table S1 and Table S2.

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## SUPPLEMENTARY MATERIAL

# Bioactivity of essential oils in phytopathogenic and post-harvest fungi control

Santamarina MP[1*], Ibáñez MD[2], Marqués M[1], Roselló J[1], Giménez S[1], Blázquez MA[2].

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## Abstract

Commercial thyme and lavender essential oils were analysed by GC/MS. Sixty-six compounds that accounted for 98.6-99.6% of total essential oil were identified. Thymol (52.14±0.21%), followed by *p*-cymene (32.24±0.16%), carvacrol (3.71±0.01%) and γ-terpinene (3.34±0.02%), were the main compounds in the thyme essential oil, while large amounts of oxygenated monoterpenes linalool acetate (37.07±0.24%) and linalool (30.16±0.06%) were found in the lavender one. The *in vitro* antifungal activity of the essential oils was evaluated at 200 and 300 µg/mL against ten phytopathogenic and post-harvest fungi, which significantly affect agriculture. Micelial growth inhibition was calculated for each tested fungus and dose. The Thyme essential oil showed satisfactory results with 90-100% growth inhibition in almost all the assayed fungi at 300 µg/mL, while the Lavender essential oil showed no noteworthy inhibition data at either dose, and its growth was even enhanced. The Thyme essential oil represents a natural alternative to control harvest and post-harvest fungi, and to extend the shelf-life of agriculture products.

**Keywords**: lavender; thyme; essential oils; antifungal activity.

## Experimental details

### 1. *Plant material*

Commercial samples of Lavender (*Lavandula angustifolia* Mill.) and Thyme (*Thymus zygis* Boiss.) essential oils were supplied by 'Plantis Artesanía Agrícola, S.A'. Essential oils were stored in the dark and in a refrigerator at 4°C until analysed by GC/MS or for antifungal activity evaluation.

### 2. *Fungi*

Strains of *Alternaria alternata* (AA) CECT 20923, *Bipolaris spicifera* (BS) CECT 2776, *Curvularia hawaiiensis* (CH) CECT 20934, *Fusarium equiseti* (FE) CECT 20925 and *Fusarium graminearum* (FG) CECT 20924 were isolated at the Botany Laboratory of the Department of Agroforest Ecosystems from the 'Bomba' rice caryopses collected in the 'La Albufera' Mediterranean rice-producing area (Valencia, Spain). *Rhizoctonia solani* (RS) CECT 2819, *Colletotricum gloeosporoides* (CG) CECT 20250, *Fusarium oxysporum lycopersici* (FOL) CECT 2715, *Penicillium expansum* (PE) CECT 20906 and *Penicillium italicum* (PI) CECT 2294 were supplied by Colección Española de Cultivos Tipo (CECT).

### 3. *Gas chromatography-mass spectrometry*

A gas chromatography-mass spectrometry analysis was carried out in a 5973N Agilent apparatus, equipped with a capillary column (95 dimethylpolysiloxane-5% diphenyl), HP-5MS UI (30 m long and 0.25 mm i.d., and 0.25 µm film thickness). The column temperature programme was 60°C for 5 min, with 3°C $min^{-1}$ increases to 180°C, and then 20°C $min^{-1}$ increases to 280°C. This programme was maintained for 10 min. Helium was the carrier gas used at a flow-rate of 1 mL $min^{-1}$. Split mode injection (ratio 1:30) was employed. The mass spectra that covered the *m/z* 30-500 range were taken at an ionising voltage of 70 eV. Kovat's retention index was calculated using co-chromatographed standard hydrocarbons. Individual compounds were identified by MS and their identity was confirmed by comparing their RIs, in relation to the $C_8$-$C_{32}$ *n*-alkanes, and by mass spectra with either authentic samples or the data already available in the NIST 2005 Mass Spectral Library and in the literature (Adams 2007).

### 4. *Antifungal activity study in solid media (Potato Dextrose Agar-PDA). Mycelial growth inhibition (MGI) calculations*

Essential oils were dissolved, mixed and homogenised in previously sterilised and still liquid PDA/Tween 20 (0.1%) at 200 and 300 µg/mL. Then they were distributed in 90x15 and 150x15 mm Petri dishes. Fungi were sowed in the centre of each Petri dish with 8 mm discoid explants from a 7-day culture. The experiment lasted 7 days at 25ºC in an incubator. Fungi growth was evaluated by measuring two perpendicular diameters of the colony on growth day 7, and the expansion speed was calculated. Ten repetitions were made per treatment. The control Petri dishes only had PDA/Tween 20 (0.1%) and the analysed fungus. MGI was calculated according to the following formula (Albuquerque et al. 2006):

MGI = [(CD-OD)/CD] x 100

CD: Average diameter of the colonies in the untreated dishes (without essential oil); OD: Average diameter of the colonies in the treated dishes (with essential oil).

### *5. Statistical analysis*

The fungal growth results were submitted to an analysis of variance (ANOVA). Furthermore, HSD Tukey intervals were represented to compare species and treatment averages, and also their interaction with significant values at $P<0.05$. The data analysis was performed by Statgraphics Centurion XVI.

**Table S1. Identified compounds in commercial thyme and lavander essential oils**

| COMPOUND | RI | RI Ref | Peak Area (%) Mean ±SD Thyme | Peak Area (%) Mean ±SD Lavander | Identification Method |
|---|---|---|---|---|---|
| ***Monoterpene hydrocarbons*** | | | **37.31±0.19** | **9.07±0.16** | |
| α-thujene | 930 | 930 | 0.02±0.00 | 0.09±0.00 | MS, RI |
| α-pinene | 937 | 939 | 0.45±0.01 | 0.16±0.01 | MS, RI |
| camphene | 951 | 954 | 0.03±0.00 | 0.12±0.00 | MS, RI |
| *trans*-pinane | 971 | 975 | 0.01±0.00 | - | MS, RI |
| sabinene | 975 | 975 | - | 0.04±0.00 | MS, RI |
| β-pinene | 978 | 979 | - | 0.03±0.00 | MS, RI |
| 3-*p*-menthene | 985 | 987 | 0.02±0.00 | - | MS, RI |
| myrcene | 992 | 990 | 0.63±0.00 | 0.43±0.00 | MS, RI |
| α-phellandrene | 1004 | 1002 | - | 0.04±0.00 | MS, RI |
| δ-3-carene | 1010 | 1011 | - | 0.17±0.00 | MS, RI |
| α-terpinene | 1018 | 1017 | 0.02±0.00 | - | MS, RI |
| *p*-cymene | 1024 | 1024 | 32.24±0.16 | 0.05±0.00 | MS, RI |
| *o*-cymene | 1026 | 1026 | - | 0.19±0.01 | MS, RI |
| β-phellandrene | 1031 | 1029 | - | 0.74±0.02 | MS, RI |
| limonene | 1032 | 1029 | 0.51±0.05 | - | MS, RI |
| *cis*-ocimene | 1040 | 1037 | - | 4.36±0.07 | MS, RI |
| *trans*-ocimene | 1053 | 1050 | - | 2.18±0.03 | MS, RI |
| γ-terpinene | 1061 | 1059 | 3.34±0.02 | 0.13±0.00 | MS, RI |
| terpinolene | 1088 | 1088 | 0.01±0.00 | 0.15±0.00 | MS, RI |
| *p*-cymenene | 1089 | 1091 | 0.03±0.00 | - | MS, RI |
| *allo*-ocimene | 1132 | 1132 | - | 0.16±0.01 | MS, RI |
| ***Oxygenated monoterpenes*** | | | **61.79±0.19** | **77.94±0.15** | |
| 1,8-cineole | 1033 | 1031 | 0.83±0.01 | 1.07±0.02 | MS, RI |
| *cis*-sabinene hydrate | 1069 | 1070 | - | 0.06±0.00 | MS, RI |
| *cis*-linalool oxide | 1074 | 1072 | 0.02±0.00 | 0.11±0.00 | MS, RI |
| *trans*-linalool oxide | 1086 | 1086 | 0.01±0.00 | - | MS, RI |
| 6,7-epoxymyrcene | 1094 | 1092 | 0.01±0.01 | - | MS, RI |
| linalool | 1101 | 1096 | 2.26±0.03 | 30.16±0.06 | MS, RI, ST |
| α-fenchol | 1112 | 1116 | 0.01±0.00 | - | MS, RI |
| *cis*-*p*-menth-2-en-1-ol | 1123 | 1121 | - | 0.04±0.00 | MS, RI |
| camphor | 1144 | 1146 | - | 0.23±0.02 | MS, RI |
| isoborneol | 1156 | 1160 | 0.43±0.03 | - | MS, RI |
| borneol | 1166 | 1169 | 1.97±0.01 | 0.57±0.01 | MS, RI |
| lavandulol | 1171 | 1169 | - | 0.74±0.00 | MS, RI |
| terpinen-4-ol | 1177 | 1177 | 0.02±0.00 | 3.43±0.02 | MS, RI |
| isocitral | 1179 | 1180 | 0.08±0.01 | - | MS, RI |
| isomenthol | 1180 | 1182 | 0.02±0.01 | - | MS, RI |
| cryptone | 1184 | 1185 | - | 0.28±0.02 | MS, RI |
| α-terpineol | 1189 | 1188 | 0.22±0.02 | 0.82±0.00 | MS, RI |
| γ-terpineol | 1201 | 1199 | 0.04±0.00 | - | MS, RI |
| nerol | 1231 | 1229 | - | 0.11±0.00 | MS, RI |
| carvacrol methyl ether | 1245 | 1244 | 0.02±0.00 | - | MS, RI |
| linalyl acetate | 1260 | 1257 | - | 37.07±0.24 | MS, RI, ST |
| lavandulyl acetate | 1293 | 1290 | - | 3.01±0.01 | MS, RI |
| thymol | 1295 | 1290 | 52.14±0.21 | - | MS, RI, ST |
| carvacrol | 1305 | 1299 | 3.71±0.01 | - | MS, RI, ST |
| neryl acetate | 1363 | 1361 | - | 0.23±0.00 | MS, RI |
| ***Sesquiterpene hydrocarbons*** | | | **0.15±0.00** | **8.39±0.04** | |
| β-bourbonene | 1385 | 1388 | - | 0.04±0.00 | MS, RI |
| β-caryophyllene | 1417 | 1419 | 0.14±0.00 | 4.44±0.01 | MS, RI |
| α-*trans*-bergamotene | 1434 | 1434 | - | 0.15±0.00 | MS, RI |
| α-humulene | 1451 | 1454 | 0.02±0.00 | 0.11±0.00 | MS, RI |
| *trans*-β-farnesene | 1457 | 1456 | - | 3.00±0.03 | MS, RI |

| | | | | | |
|---|---|---|---|---|---|
| germacrene D | 1483 | 1485 | - | 0.49±0,00 | MS, RI |
| γ-cadinene | 1510 | 1513 | - | 0.15±0.01 | MS, RI |
| ***Oxygenated sesquiterpenes*** | | | **0.33±0.00** | **0.39±0.04** | |
| caryophyllene oxide | 1578 | 1583 | 0.31±0.00 | 0.30±0.01 | MS, RI |
| humulene epoxide II | 1603 | 1608 | 0.02±0.00 | - | MS, RI |
| *epi*-α-cadinol | 1636 | 1640 | - | 0.09±0.00 | MS, RI |
| ***Others*** | | | - | **2.48±0.03** | |
| 1-methoxy-hexane | 826 | | - | 0.07±0.00 | MS, RI |
| hexanol | 867 | 870 | - | 0.04±0.00 | MS, RI |
| 1-octen-3-ol | 981 | 979 | - | 0.20±0.00 | MS, RI |
| 3-octanone | 988 | 983 | - | 0.74±0.03 | MS, RI |
| 3-octanol | 996 | 991 | - | 0.14±0.00 | MS, RI |
| hexyl acetate | 1016 | 1009 | - | 0.37±0.00 | MS, RI |
| octenyl acetate | 1117 | | - | 0.78±0.00 | MS, RI |
| hexyl isobutanoate | 1153 | 1151 | - | 0.06±0.00 | MS, RI |
| hexyl butanoate | 1193 | 1192 | - | 0.31±0.00 | MS, RI |
| hexyl hexanoate | 1383 | 1383 | - | 0.06±0.00 | MS, RI |
| **TOTAL IDENTIFIED** | | | **99.59±0.02** | **98.58±0.02** | |

Compounds listed in order of elution in the HP-5MS column. RI: retention index relative to $C_8$-$C_{32}$ n-alkanes on the HP-5MS column. Peak area values are means ± standard deviation of three samples. Identified compound by MS (mass spectra) RI (Kovat's index, Adams 2007) and ST (authentic sample).

**Table S2. (a) Mean growth and standard deviation values for each fungus grown on PDA, thyme and lavender essential oils. (b). Mycelial Growth Inhibition (MGI) percentage for each fungus grown on PDA, thyme and lavender essential oils.**

| a. Mean growth and standard deviation values for each fungus grown on PDA, thyme and lavender essential oils | | | | | |
|---|---|---|---|---|---|
| Species | Control | Concentration (µg/mL) | | | |
| | | 200 | | 300 | |
| | | Thyme | Lavander | Thyme | Lavander |
| CG | 68.70 ± 1.64 | 30.50 ± 2.67 | 61.40 ± 0.84 | 0.00 ± 0.00 | 58.90 ± 2.33 |
| FG | 112.60 ± 11.52 | 26.6 ± 3.20 | 114.70 ± 4.50 | 0.00 ± 0.00 | 107.10 ± 6.37 |
| FOL | 59.60 ± 2.76 | 12.38 ± 1.69 | 61.70 ± 1.34 | 4.30 ± 1.50 | 57.70 ± 2.11 |
| FE | 74.40 ± 4.84 | 1.30 ± 0.82 | 83.40 ± 3.10 | 0.00 ± 0.00 | 85.40 ± 5.58 |
| AA | 55.20 ± 4.73 | 11.67 ± 1.37 | 60.40 ± 2.50 | 0.50 ± 0.85 | 58.40 ± 2.46 |
| CH | 23.20 ± 4.42 | 1.25 ± 1.39 | 30.50 ± 2.27 | 0.00 ± 0.00 | 32.5 ± 7.17 |
| BS | 81.80 ± 0.92 | 0.10 ± 0.32 | 77.00 ± 1.83 | 0.00 ± 0.00 | 80.70 ± 4.72 |
| RS | 116.60 ± 5.19 | 0.00 ± 0.00 | 120.50 ± 1.58 | 0.00 ± 0.00 | 120.6 ± 0.84 |
| PE | 26.70 ± 6.04 | 11.17 ± 1.17 | 33.9 ± 1.66 | 2.10 ± 1.10 | 32.30 ± 2.67 |
| PI | 21.30 ± 4.16 | 11.33 ± 1.21 | 31.20 ± 2.04 | 2.40 ± 0.70 | 32.40 ± 2.12 |

| b. Mycelial Growth Inhibition (MGI) percentage for each fungus grown on PDA, thyme and lavender essential oils | | | | |
|---|---|---|---|---|
| Species | Concentration (µg/mL) | | | |
| | 200 | | 300 | |
| | Thyme | Lavander | Thyme | Lavander |
| CG | 55.60 | 10.60 | 100 | 14.30 |
| FG | 76.40 | -1.90 | 100 | 4.90 |
| FOL | 78.80 | -3.50 | 92.80 | 3.20 |
| FE | 98.30 | -12.10 | 100 | -14.80 |
| AA | 78.30 | -9.40 | 99.50 | -5.80 |
| CH | 94.60 | -31.50 | 100 | -40.10 |
| BS | 100 | -2.00 | 100 | -4.40 |
| RS | 100 | 6.90 | 100 | -3.50 |
| PE | 58.20 | -27.00 | 92.10 | -21.00 |
| PI | 46.80 | -46.50 | 88.70 | -52.10 |

CG: *Colletotrichum gloeosporoides*, FG: *Fusarium graminearum*, FOL: *Fusarium oxysporum* fsp. *lycopersici*, FE: *Fusarium equiseti*, AA: *Alternaria alternata*, CH: *Curvularia hawaiiensis*, BS: *Bipolaris spicifera*, RS: *Rhizoctonia solani*, PE: *Penicillium expansum*, PI: *Penicillium italicum*.