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Antimicrobial and Antioxidant Activity of *Pelargonium odoratissimum* Essential Oil Exhibits

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Results
The chemical composition of essential oils was determined by GC. Results of the analysis are shown in Table 1. There is a substantial seasonal variation in the chemical composition of the essential oils.

Conclusions
A seasonal variation is observed in the antimicrobial and antioxidant activities of *Pelargonium odoratissimum* essential oil which seems to be correlated with oil chemical composition.

As shown in Figure 2, the antioxidant activity of *Pelargonium* EO shows significant seasonal variation in a similar fashion with its antimicrobial activity; it is increased by a factor of 1.5 ± 0.3 during the months of June-July.

It is interesting to note that the similar seasonal variation of the antimicrobial and antioxidant activities seems to correlate with an increase observed in the percentage of the component geraniol of the EO during June and July (see Table 1).

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Seasonal Variation of Antimicrobial Activity of Thyme Essential Oil on *Listeria monocytogenes* and *Staphylococcus sp.*

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Introduction
Thyme essential oil (EO) is a natural organic product which can potentially be used as a food additive, as an alternative to chemical substances.

Objectives
The objective of this work was to determine and compare the chemical composition as well as the antimicrobial activity of essential oil derived from thyme collected during six consecutive months (February - July 2011).

Materials & Methods
Thyme of the carvacrol chemotype was collected in monthly intervals from a specific location of the island of Aetolia (Greece). Thyme EO was obtained by hydrodistillation and its chemical composition was determined by gas chromatography (GC). Each oil was screened for antimicrobial activity. The bacteria used were the following: *Staphylococcus epidermidis* ATCC 12228, *S. epidermidis* ATCC 14990, *S. saprophyticus* ATCC 35552, *S. hominis* ATCC 27864, *S. aureus* ATCC 12598, *S. aureus* ATCC 25923, *S. aureus* ATCC 12600, *Listeria monocytogenes* ATCC 7644, *L. monocytogenes* ATCC 15313, *Listeria monocytogenes* ATCC 19111. The antimicrobial activity was studied by determining the diameter of inhibition (disc assay) and the minimum inhibitory concentration (MIC).

Results
The chemical composition of all essential oils was determined by GC. Results of this analysis are shown in Table 1. Substances that show a considerable fluctuation include p-cymene, thymol, linalool and eucalyptol. The concentration of p-cymene decreases as the thymol concentration increases, as expected since the first substance is a precursor of the second.

Conclusions
A statistically significant seasonal variation was observed for the antimicrobial activity of thyme EO. The activity, tested in suspension (Diameter of Inhibition) was 2.5 ± 0.3 mm diameter of inhibition (DI) in MIC during the months of June and the minimum lower by a factor of 1.2 during February.

References
Eriotou, E., et al. (2011) Seasonal variation of thyme essential oil chemical composition and antimicrobial activity. *Journal of Agricultural Science*, 151, 1-6.

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Antimicrobial Activity of Thyme Essential Oil on *Staphylococcus aureus* in Cottage Cheese

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Introduction

There is an increasing demand for organic and natural products as alternatives for chemicals used in food as antimicrobials. Such products include essential oils of plant origin.

Objectives

The objective of this work was to determine the antimicrobial effect of thyme essential oil on three *Staphylococcus aureus* strains in commercial cottage cheese. To our knowledge, the effect of *S. aureus* has not been studied in milk or dairy products in the presence of thyme essential oil, and there is limited work concerning other bacteria species (1).

Materials & Methods

The essential oil tested for antimicrobial activity was obtained by hydro-distillation from local thyme and its chemical composition determined by gas chromatography (GC). The bacteria used were the following: *Staphylococcus aureus* ATCC 12602, *S. aureus* ATCC 12598 and *S. aureus* ATCC 25922. Cottage cheese used was obtained from commercial sources.

Determination of thyme oil lethal concentration: A series of sterile stomacher bags were generated for each strain, each containing 10 g of cottage cheese, 10⁷ cfu of the bacterial strain and a different concentration of thyme essential oil. Bags were homogenized, placed at 4°C for 4 hours, serially diluted and plated onto Baird Parker (BP) plates (Oxoid). The plates were incubated at 37°C and counted.

Determination of the time required for the lethal effect (Death curve): Cottage cheese had 4 weeks remaining on its shelf life. Portions of the cheese to which thyme oil at the lethal concentration was added were inoculated with a strain of *S. aureus* (10⁷ cfu/g) and kept at 4°C. Control cheese samples with no added *S. aureus* cells were also prepared as well as control samples with bacteria but no oil. 10 g samples were taken periodically from each, serially diluted and plated onto TSA plates.

Results

The results of the GC analysis of the chemical composition of thyme essential oil used are shown in Figure 1. The major components found were thymol and p-cymene (its precursor).

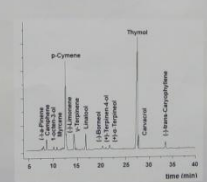


Figure 1: GC chromatogram of thyme oil.

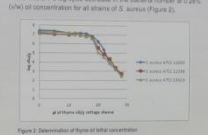


Figure 2: Determination of thyme oil concentration. There was a 4-5 log-cycle decrease in the bacteria number at 0.25% (v/v) oil concentration for all strains of *S. aureus* (Figure 2).

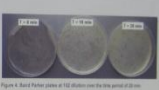


Figure 4: Baird Parker plates at 100 dilution on the 25th period of 20 days.

Conclusions

The results showed that thyme essential oil can be used as a food additive aiming at the growth control of *S. aureus* in food products, provided that it is incorporated safely in the respective compositions. Moreover, determination of the mechanism of action of thyme essential oil will potentially strengthen its practical application in food products.

References

1. Baga, V. K., Baga, K. H. and King, S. C. 2012. Control of *Salmonella* in foods by using essential oils. *Antonie van Leeuwenhoek* 102: 722.

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The Antimicrobial Activity of Thyme Essential Oil on *Salmonella* sp. in Milk is Influenced by Lipid Content

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Introduction

There is no doubt that more effective antimicrobials will be useful in the safe extension of food shelf life as well as meeting consumer changing demands (1). Thyme essential oil (EO), a potent antimicrobial, shows decreased antimicrobial activity when added to food products indicating an interaction of the oil with some food components(2). Therefore, if such a product is to be used as a food antimicrobial, its interactions with different food components should be evaluated.

Objectives

The objective of this work was to determine the effect of lipid content of source milk on thyme essential oil antimicrobial activity.

Materials & Methods

Thyme essential oil was obtained by hydrodistillation of local thyme and its chemical composition was determined by gas chromatography (GC). Lipid content of commercial pasteurized milk was controlled by addition of pasteurized milk cream. Six *Salmonella* species were used as test microorganisms: *S. enterica* ATCC 13314, *S. subenterica* ATCC 9759, *S. typhimurium* ATCC 14028, *S. enteritidis* ATCC 13076, *S. saintpaulus* ATCC 12032 and *S. dublinus* ATCC 19619.

The essential oil antimicrobial activity was assessed by determining its lethal concentration. To achieve this, a series of sterile test tubes were generated for each strain, each containing 10 ml of milk, 10⁷ cfu of the bacterial strain and a different concentration of thyme essential oil. Test tubes were stored at 4°C for 4 hours, serially diluted and plated onto Salmonella-Shigella agar (SSS) plates (Oxoid). The plates were incubated at 37°C and counted. Experiments were performed in triplicate. Statistical analysis was done by using the t-test (5% significance level or less).

Results

Figure 1 and Table 1 show the results obtained by GC.

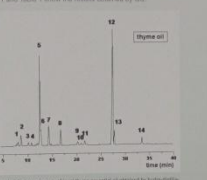


Figure 1: GC chromatogram of local thyme essential oil obtained by hydro-distillation.

Peak	Retention Time (min)	Chemical Compound	Area	Chemical Compound	Area	
1	7.84	Thymol	10000	12	Thymol	10000
2	8.90	p-Cymene	10000	13	Thymol	10000
3	10.02	Carvadiol	10000	14	Thymol	10000
4	11.15	Carvadiol	10000	15	Thymol	10000
5	12.28	Thymol	10000	16	Thymol	10000
6	13.41	Thymol	10000	17	Thymol	10000
7	14.54	Thymol	10000	18	Thymol	10000
8	15.67	Thymol	10000	19	Thymol	10000
9	16.80	Thymol	10000	20	Thymol	10000
10	17.93	Thymol	10000	21	Thymol	10000
11	19.06	Thymol	10000	22	Thymol	10000

Figure 2: Chemical composition of thyme essential oil.

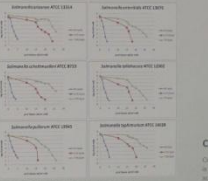


Figure 3: Determination of thyme oil concentration for six different *Salmonella* species.




Figure 4: Six S.S.S. plates at 100 dilution on the 25th period of 20 days.

Conclusions

One of the most diverse and interesting areas of plant essential oils is to explore their natural pathogenesis against *Salmonella* species. In addition to the suppression of food, one should determine the necessary concentration of the essential oil in the specific food product. Our research showed that the antimicrobial activity of thyme essential oil on *Salmonella* sp. decreases significantly as the lipid content of the food under investigation increases.

References

1. Hock, R. A. and G. Patel. 2000. Enhancement in *Salmonella* strains by plant essential oils and animal antimicrobials. *Food Microbiol.* 22: 273.

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