Deanship of Graduate Studies

Al-Quds University

Isolation and Identification of the Essential Oils of the Palestinian Thyme (*Majorana syriaca L.*) using Headspace GCMS

By

Mohammad Mahmoud Qabaja

M.Sc. Thesis

2004

Isolation and Identification of the Essential Oils of the Palestinian Thyme (*Majorana Syriaca L.*) using Headspace GCMS

By

Mohammad Mahmoud Qabaja

B.Sc. in chemistry from Al-Quds University, Palestine

A thesis Submitted in Partial Fulfillment of The requirement for the Degree of Master of Science in Applied and Industrial Technology/Graduate Studies

Al-Quds University- Jerusalem

January, 2004

Program of Postgraduate Studies in Applied & Industrial Technology
Deanship of Graduate Studies

Isolation and Identification of the Essential Oils of the Palestinian Thyme (*Majorana syriaca L.*) using Headspace GCMS

Student Name: Mohammad Mahmoud Mohammad Qabajah

Registration No: 9811371

Supervisor: Dr. Saleh Abu-Lafi
Co- Supervisor: Dr. Imad Odeh
Co- Supervisor: Dr. Hasan Dweik

Master thesis submitted and accepted, Date: 24/1/2004

The names and signatures of the examining committee members are as follows:

1- Dr. Saleh Abu-Lafi Head of Committee signature----------

2- Dr. Khaled Salem Internal Examiner signature----------

3- Dr. Mahmoud Deheidel External Examiner signature----------

Al-Quds University

2003-2004

DECLARATION

I certify that this thesis, which is submitted for the degree of master, is the result of my own research, except where otherwise acknowledged, and that this thesis (or any part of the same) has not been submitted for a higher degree to any university or institution.

Signed:  .....

Mohammad Mahmoud Mohammad Qabaja

Date: 24/1/2004

ACKNOWLEDGEMENT

I am very pleased to express my thanks to my supervisors Dr. Saleh Abu-Lafi, Dr. Imad Odeh and Dr. Hassan Dweik for their supervision, guidance and for supporting me to go forward in my study. I would also like to thank all who helped in collecting the thyme samples from different districts in Palestine. I would like to thank Mr. Amin Imam for his kind technical assistance. I would like to thank Dr. Khaled Salem for providing me with useful information related to the botanical origin of different thyme samples. Finally, I would like to thank my friend Omar Ayyad for the unlimited computer assistance.

ABSTRACT

There are over hundred varieties of thyme. Common thyme, *Majorana syriaca l.* (Zaatar in Arabic), that belongs to the mint family, Labiatae, is native to Palestine. It is one of the herbs that is cultivated widely and grows wild in the mountains of Palestine between the months April to August.

The present study investigates the use of Headspace technology coupled to Gas Chromatography Mass Spectrometry (HS-GCMS) to isolate and identify the volatile and semi-volatile components in the Palestinian dry leaves of thyme. The mode of analysis was done in the electron impact (EI) mode.

Sixty two samples growing wild and cultivated from seven populations from the West Bank territories at different harvesting times (January 2000-June 2001) were isolated by simple steam distillation and headspace followed by GCMS analysis. More than forty volatile and semi-volatile compounds were identified by the NISTL/ MS library and were confirmed from the retention times of authentic standards. Therefore, headspace developed method proved to be very fast and reliable in determining all the components present in the thyme leaves.

Monoterpenes (oxygenated and hydrocarbons) are the main constituents in thyme leaves. Only few hydrocarbons and sesquiterpenes such as caryophyllene, were also present but in smaller quantities in comparison to the oxygenated monoterpenes. The principal constituents of oxygenated monoterpenes are the phenolic compounds, thymol and its geometrical isomer, the carvacrol

The steam distillation analysis of the Palestinian thyme revealed that the yield of the oil based on air-dried weight, ranged from 1.05% (January) to 5.39% (May), a relatively high number in comparison to different countries. This result indicates that the oil production is increasing towards the end of vegetative growth. The HS-GCMS results showed that the Palestinian thyme is rich in monoterpene hydrocarbons and phenolic monoterpenes. The major constituents identified throughout all the harvesting periods were varied greatly among the thymes examined and were as

follows: α-phellandrene (1.63-8.9%), α-pinene (1.98-4.61%), β-myrecene (3.43-11.0%), m-cymene (3.21-10.6%), p-cymene (9.79-25.8%), γ-terpinene (11.96-30.8%), thymol (0.26-15.3%), carvacrol (0.44-21.7%).

In all samples, the most abundant monoterpenes were γ-terpinene and p-cymene, the biogenatic precursors (via enzymic hydroxylation) of the phenolic terpenes thymol and carvacrol. The percentages by wieght of γ-terpinene increased during February, March and April while decreased in May. The percentages of p-cymene were increased during February, March and April while decreased in May. Thymol was increased during May in cultivated samples; on the other hand carvacrol was increased during May in wild samples.

Previous investigations related the isomeric distribution of thymol and carvacrol to various factors including geographic location, irrigation and harvesting time.

Our findings, however, showed that the main factor that controls the isomeric distribution is attributed to fresh water irrigation. Thymol was the predominant constituent of cultivated thyme while carvacrol was the principal constituent of wild thyme. Therefore, this isomeric distribution ratio is used to distinguish wild from cultivated thyme. Moreover, the isomeric distribution ratio of thymol to carvacrol was found to be close using both methods the simple steam distillation and the headspace.

To My Parents, Brothers, Wife and Children

CONTENTS

LIST OF ABBREVIATIONS

HMG-CoA	3-Hydroxyl 3-Methyl Glutaryl-SoA
NADP	Nicotine Amide Diphospho Pyridine
ATP	Adonisine Tri-phosphate
MVA	Mevalonic acid
IPP	Isopentyl-pyrophosphate
DMAPP	Dimethylallyl pyrophosphate
GPP	Geranylpyrophosphate
NPP	Nerlypyrophosphate
GC	Gas chromatograph
GCMS	Gas chromatograph mass spectrometry
HS	Headspace
DHS	Dynamic headspace
SD	Steam distillation
C	Cultivated
W	Wild
DDW	Double distilled water
TIC	Total ion chromatogram
RSD	Relative standard deviation

LIST OF TABLES

Table No.

LIST OF FIGURES