Composition of the Essential Oils of *Thymus deanensis* Celak Var. *deanensis* from Different Regions of Iran

Abousaber M (Ph.D.)[1], Khanavi M (Ph.D.)[1*], Khoshchehreh M (M.D.)[2], Hadjiakhoondi A (Ph.D.)[1], Shams Ardekani MR (Ph.D.)[1], Shafiee A (Ph.D.)[3]

1- Department of Pharmacognosy and Medicinal Plants Research Center, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran
2- Evidence Based Medicine Group, the Endocrine and Metabolism Research Center, Shariati Hospital, Tehran University of Medical Sciences, Tehran, Iran
3- Department of Medicinal Chemistry, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran
*Corresponding author: Department of Pharmacognosy, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran
Tel & Fax: +98 – 21 - 66954706
Email: khanavim@tums.ac.ir

Received: 5 March 2011 **Accepted:** 4 Jan. 2012

Abstract

Background: ***Thymus deanensis*** **is a known aromatic plant which grows widely in Iran and many mediterranean regions. In previous studies composition of essential oils of many species of** ***Thymus*** **genus reported which they were rich of phenols like Thymol and Carvacrol.**

Methods: **Water distilled essential oils from aerial parts of** ***Thymus deanensis*** **var.** ***deanensis*** **from three different localities in Iran were analyzed by GC and GC/MS.**

Results: **20 compounds representing 87.8% of the region of Azerbaijan Mountains (A) oil were identified with thymol (70.8%), carvacrol (6.3%) as main constituents.**
13 compounds representing 98.6 % of Damavand, Mosha (B) oil were identified with thymol (53.6%), carvacrol (23.8%) and p-cymene (11.0 %) and then thymol (72.1%) and p-cymene (5.3%) were the major constituents of the latter oil Chaharmahal Bakhtiari (C) among the 16 compounds characterized which represented 99.6% of the oil.

Conclusions: **The composition of three samples of** ***Th. deanensis*** **oils in our research was similar to the composition of other phenol–rich Iranian species. Although the content of thymol in** ***Th. deanensis*** **is higher than many reported cultivated** ***Thymus vulgaris*****.**

Keywords: ***Thymus deanensis*** **var.** ***deanensis*****, Oil composition, Thymol, Carvacrol,** ***P*****-cymene**

[DOR: 20.1001.1.2717204.2012.11.41.4.0]

Introduction

The *Thymus* genus comprises over 300 species of which, 14 are found in Iran [1], which grow wiled in many regions and four of them are endemic [2]. Thymus is a well – known medicinal plant which is native to Southern Europe which its essential oil is manufactured commercially for use in cough drops, mouthwashes, liniments, toothpastes, detergents and perfumes. The herb is approved by Commission E in the treatment of bronchitis, whooping cough and upper respiratory inflammation.

Thyme in its crude herb form is carminative, antibiotic, anthelmintic, astringent, expectorant and antitussive. It has been used in traditional medicine to treat heat burn, gastritis, asthma, laryngitis, pertusis and bronchitis. Extracts demonstrate in vitro anti inflammatory effects on guinea pig tracheal smooth muscle tissue and the volatile oil in the herb most likely exerts spasmolytic effects on bronchial tissues in humans [3].

They are hot, energizing oils that can be used for local, respiratory and systematic infections and mental stimulants in aromatherapy [4]. In folk medicine, *Thymus* spp. are used an anthelmintic, antispasmodic, carminative, sedative, diaphoretic usually in form of an infusion, or externally in bath to cure rheumatic and skin disease [5]. Thyme oil is also carminative, expectorant and possesses antimicrobial and anthelmintic properties due to concentrated thymol and carvacrol content but it is extremely toxic. Thymus essential oil and extract is a source of aromatic terpens and terpenoids, flavonoids and phenolic acids [6].

Thymol, which is the main components of many *Thymus* species is known as an antiseptic agent and is approved for diverse effects like hookworm treatment [7, 8]. Also thymol and their salts used about 0.1 - 1% in formulation of many lotions, creams and ointments. In external used about 0.1 - 1% in formulation of many lotions, creams and ointments. In external use, thymol is known as a strong antiseptic agent in toothplaste, gargle and mouthwashes [9]. The other major component of *Thymus* spp. oil is carvacrol which is used nowadays on a large scale in the food, cosmetic and mouthwashes industries. In addition, it has been shown several activities like antimicrobial, analgesic and antioxidant activities but it is toxic in high concentration [10].

There are many reports of the essential oil composition and biological activity of different *Thymus* species especially common Thyme (*Thymus vulgaris*) and wild Thyme (*Thymus serpyllum*). Hence of the use of *Thymus* species or their essential oils in the food and traditional medicine of Iran, we were interested in studying on the essential oil contents and chemical composition of all Iranian endemic species.

In this study, we reported the essential oil composition of *Thymus deanensis* Var. *deanensis* from three different regions of north, northwest and west of Iran.

Materials and Methods

Three samples of *T. deanensis* Var. *deanensis* were collected during the flowering stage in June and July 2008 from different localities in northwest, west and north of Iran, respectively.

[DOR: 20.1001.1.2717204.2012.11.41.4.0]

Voucher specimens have been deposited at the Herbarium of Research Institute of forests and rangelands, Tehran, Iran.

A: *T. deanensis*: West. Azerbaijan Mountains, June, 2008.

B: *T. deanensis*: Damavand, Moshaa, July, 2008.

C: *T. deanensis*: Chaharmahal Bakhtiary. Ardel nanman, July, 2008.

Preparation of oil

The aerial part of *T. deanensis* (100 g) was subjected to hydrodistillation using a Clevenger-type apparatus for 4h. The oils were dried over anhydrous sodium sulfate and obtained oils kept in refrigerator until doing analysis.

GC & GC _MS Analysis

Analytical gas chromatography was carried out using a Termoquest 2000 GC with capillary column DB-1 (30 m. 0.25 mm i.d., 0.25 μm film Thickness); carrier gas, He; split ratio, 1:25; and using a flame ionization detector. The column temperature was programmed at 50°C for 1 min. and then heated to 265°C at a rate of 2.5°C/min. and then kept constant at 265°C for 20 min.

GC-MS was performed on a Thermoquest 2000 with a quadruple detector, on capillary column DB-1 (GC); carrier gas, He; flow rate, 1.5 ml/min. the column was held at 50°C for 1 min. and programmed up to 265°C at rate of 2.5°C/min, then kept constant at 256°C for 20 min. The MS operated at 70 eV ionization energy. Retention indices were calculated by using retention times of *n*-alkanes that were injected after the oil at the same chromatographic conditions. Quantitative data were obtained from the electronic integration of the FID peak areas.

The components of the oils were identified by comparison of their mass spectra and retention indicates with Wiley library and those published in the literature [11 - 13].

Results

Yields of essential oils were 1% for A, 1.1% for B and 1.8% for C with yellow color and a distinct sharp odor. The chemical composition of the oils of three sample of *T. deanensis* Var. *deanensis* oil is presented in Table 1.

In sample A, 24 components were identified representing 88.6% and the main constituents of A were thymol (70.8%) and carvacrol (6.3%). In sample B, 13 components were identified representing 98.6% and the main constituents were thymol (53.6%), carvacrol (23.8%) and p-cymene (11.0%) while in sample C, 16 components representing 99.6 % and the main constituents of C were thymol (72.1%), p-cymene (5.3%), γ-terpinene (4.9%) and α-pinene (4.0%).

Table 1- Chemical composition of essential oil of aerial parts of *Thymus daenensis*

Components	RI	A	B	C
α – Pinene	929	0.2	4.6	4.0
Camphene	950	-	0.1	0.8
β-Pinene	980	0.1	0.7	0.2
Myrcene	987	0.1	-	0.1

[DOR: 20.1001.1.2717204.2012.11.41.4.0]

Table 1- Continued

Components	RI	A	B	C
α – Phellandrene	1002	-	-	2.0
ρ-Cymene	1011	1.5	11.0	5.3
α -Terpinene	1016	-	-	1.0
1,8-Cineol	1019	0.5	-	-
(E) – β-Ocimene	1042	0.6	-	-
γ- Terpinene	1057	-	3.3	4.9
Fenchone	1071	0.4	-	-
Linalool oxide*	1083	1.7	-	-
Linalool	1096	0.2	0.6	-
Limonene epoxide*	1119	0.3	-	-
Borneol	1148	0.8	-	1.5
Terpinen-4-ol	1160	0.7	-	-
Menthol	1171	1.6	-	-
(B)-Citronellol	1224	0.3	-	-
Methyl thymol	1235	-	-	1.4
Geraniol	1236	0.2	-	-
Thymol	1272	70.8	53.6	72.1
Carvacrol	1279	6.3	23.8	-
Eugenol	1326	-	-	1.0
α- Longipinene	1359	0.3	-	-
β – Caryophyllene	1432	0.5	0.3	-
Trans-9- epi-Caryophyllene	1467	-	0.1	-
β – Bisabolene	1507	0.7	0.3	1.5
γ – Cadinene	1513	-	-	2.0
(z) Nerolidol	1540	-	-	1.0
Himachalene	1544	-	-	-
Thymohydroquinone	1552	-	0.1	-
Caryophyllene alcohol	1559	-	-	-
α – Terpinyl sopantanoate	1565	-	-	-
Caryophyllene oxid	1580	-	0.1	-
Muurolol	1645	-	-	0.8
Total	--	**87.8 %**	**98.6 %**	**99.6 %**
Monoterpenes Hydrocarbons %		2.5	19.7	18.3
Monoterpenes Oxygenated %		83.8	78	75
Sesquiterpens Hydrocarbons %		1.5	0.7	3.5
Sesquiterpens Oxygenated %		0	0.2	1.8
Phenyl Propanoid		-	-	1.0

*A:T. deanensis: West. Azerbaijan Mountains, B: T. deanensis: Damavand, Moshaa, C: T.deanensis: Chaharmahal Bakhtiary. Ardel nanman,

Discussion

The oil composition of three samples were similar in containing large amounts of thymol and carvacrol together with present of p-cymene, α-pinene, γ-terpinene, β-pinene, β-bisabolene and β-caryophyllene. Linalool oxide and terpinen-4-ol were found only in sample A (1.65%, 0.66 %) respectively. Also α-terpinene and α-phellandrene were found only in sample C (1.0%, 2.0%). The oil of *T. deanensis* var. *deanensis* contained monoterpene hydrocarbons (2.5% in A, 19.7% in B and 18.3% in C samples), oxygenated monoterpenes (83.8% in A, 78% in B and 75% in C samples), sesquiterpen hydrocanrbons (1.5% in A, 0.7% in B and 3.5% in C samples)

[DOR: 20.1001.1.2717204.2012.11.41.4.0]

and oxygenated sesquiterpens (0.2% in B and 1.8% in C samples). Hence, total amount of oxygenated monoterpenes were considered as the highest fractions of the oils. In conclusion, it is evident that there are qualitative similarities between the major compound group and constituents present in these three oils. However, minor constituents are qualitatively and quantitatively different.

The composition of three sample of *T. deanensis* oils in our research was similar to the composition of other phenol–rich Iranian Thymus species [14-18], it is noticeable that content of thymol in *T. deanensis* is higher than many reported cultivated Thymus vulgaris [18].

Acknowledgments

The authors wish to thank Dr. F. Sefid-kon Research Institute of Forests and Rangelands, for the GC/MS spectra and Dr. V. Mozaffarian for his help in identifying plant materials.

References

1. Rechinger KH, Hedge IC. Flora Iranica. Vol. 150. Akademisch Druck-u.Verlag sanstalt, Graz, *Austria* 1982; 532 - 51.

2. Mozaffarian V. A dictionary of Iranian Plant Names.Vol.196.: Farhang Moaser Publishers. Tehran, Iran. 1996, p: 547.

3. Van Den Broucke CO, Lemli JA. Spasmolytic activity of the flavonoids from Thymus vulgaris. *Pharm. Weekbl Sci*. 1983 Feb 25; 5 (1): 9 - 14.

4. Sefidkon F. Kalvandi R. Atri M. Barazandeh M.M. Essential oil variability of Thymus eriocalyx (Ronniger) Jalas. *Flavour and Fragrance J*. 2005 Sep 20; 5: 521 – 4.

5. Rustaiyan A, Masoudi S, Monfared A, Kamalinejad M, Lajevardi T, Sedaghat S, Yari M. Volatile constituents of three Thymus species grown wild in Iran. *Planta Med*. 2000 Mar; 66 (2): 197 - 8.

6. Stahl-Biskup E. & Sáez F. Thyme – the Genus Thymus. Taylor & Francis. London. 2002, 75.

7. Sefidkon F, Askari F and Mirmostafa SA. The essential oil of *Thymus carnosus* Boiss. from Iran. Journal of Essential Oil Research 2001; 13: 192 – 3.

8. Evans WC. Trease and Evans' Pharmacognosy, Saunders: London, 1998, pp: 171, 263.

9. Zargari A. Pharmaceutical Plants. 4th ed. Tehran University Publications. Tehran, Iran. 1990, pp: 28 - 48.

10. Monzote L, Stamberg W, Staniek K, Gille L. Toxic effects of carvacrol, caryophyllene oxide, and ascaridole from essential oil of Chenopodium ambrosioides on mitochondria. *Toxicol. Appl. Pharmacol*. 2009 Nov 1; 240 (3): 337-47. Epub 2009 Aug 8.

11. Ryhage R. Sydow E. V. Mass spectrometry of terpenes. I. Monoterpene hydrocarbons. *Acta Chem. Scand*. 1963; 17: 2025 - 35.

12. Sydow EV. Mass spectrometry of terpenes, II. Monoterpene alcohols. *Acta Chem. Scand.* 1963; 17: 2504 - 12.

13. Davies NW. Gas chromatographic retention indices of monoterpenes and sesquiterpenes on methyl silic one and carbowax 20 M phases. *J. Chromatogr*. 1990;

[DOR: 20.1001.1.2717204.2012.11.41.4.0]

313: 1 – 24.

14. Kalvandi, R. Sefidkon, F. Atri, M. Mirza, M. Analysis of the essential oil of Thymus eriocalyx from Iran. *Flavour and Fragr. J*. 2004; 19 (4): 341 - 3.

15. Miri R, Ramezani M, Javidnia K and Ahmadi L. Composition of the volatile oil of Thymus transcaspicus Klokov from Iran. *Flavour Fragr. J*. 2002; 17: 245 - 6.

16. Rasooli I, Mirmostafa SA. Bacterial susceptibility to and chemical composition of essential oils from Thymus kotschyanus and Thymus persicus. *J. Agric. Food Chem*. 2003; 51 (8): 2200 - 5.

17. Rasooli I, Mirmostafa SA. Antibacterial properties of Thymus pubescens and Thymus serpyllum essential oils. *Fitoterapia* 2002; 73: 244 - 50.

18. Hajimehdipoor H, Shekarchi M, Khanavi M, Adib N, Amri M. A validated high performance liquid chromatography method for the analysis of thymol and carvacrol in *Thymus vulgaris* L. volatile oil. *Pharmacognosy Mag*. 2010; 6 (23): 154 - 8.

[DOR: 20.1001.1.2717204.2012.11.41.4.0]