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GC/MS evaluation of thyme (*Thymus vulgaris* L.) oil composition and variations during the vegetative cycle

Mohammad Hudaib [a,*], Ester Speroni [b], Anna Maria Di Pietra [a], Vanni Cavrini [a]

[a] *Dipartimento di Scienze Farmaceutiche, University of Bologna, Via Belmeloro 6, 40126 Bologna, Italy*
[b] *Dipartimento di Farmacologia, University of Bologna, Via Irnerio 48, 40126 Bologna, Italy*

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Abstract

Capillary GC/MS analysis based on polar and non-polar columns has been applied to evaluation of the volatile oils hydrodistilled from thyme (*Thymus vulgaris* L.) plants. The adopted methodology has been used to monitor seasonal variations in the composition of the oil obtained from thyme herbs harvested at different periods during the plant vegetative and life cycles. Oils from thyme plants of young (2 years) and old (5 years) cultivations have been evaluated from four and two collections, respectively, effected throughout May/December growth period. Generally, the oil was found to be rich in the active monoterpene phenols (thymol and carvacrol) and their corresponding monoterpene hydrocarbon (HC) precursors (*p*-cymene and γ-terpinene), which collectively showed synchronized patterns of variation during the different collection periods and in different seasons. The oil from old plant collected in May/June period (0.15% v/w) was characterized by significantly lower levels of monoterpene HCs (mainly γ-terpinene) and the highest levels of the oxygenated monoterpenes (linalool and borneol), monoterpene phenols (mainly thymol) and their derivatives (mainly carvacrol methyl ether), sesquiterpenes (mainly β-caryophyllene) and their oxygenated derivatives (e.g. caryophyllene oxide) in comparison with all other samples. A characteristic presence of camphor and thymodihydroquinone was also observed in the old plant oils. On the other hand, the young plant, collected in June/July just before the end of the vegetative cycle, provided the best oil yield (1.2%) with also the highest % content of the monoterpene phenols (thymol: 51.2% and carvacrol: 4%). This latter growth period can represent the best harvest time of young thyme plants in order to obtain an essential oil with better quality and quantity. © 2002 Published by Elsevier Science B.V.

Keywords: *Thymus vulgaris*; Essential oil composition; Seasonal variations; Vegetative cycle; Retention (Kovats) index; Polar and non-polar capillary column; GC/MS analysis

1. Introduction

Many phytochemical studies so far investigated the chemical composition of the essential oil from *Thymus vulgaris* L. (family: Labiatae or Lami-

* Corresponding author. Fax: + 39-051-2099-734
E-mail address: hudaib1@yahoo.com (M. Hudaib).

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aceae) from different sources and chemotypes [1–4] as well as its variation in different seasons [1] and during the plant life cycle [4–8]. Evaluations of the oil composition extracted from different parts of the plant or upon variable environmental, cultivation, and/or storage conditions have also been reported [9–11]. *T. vulgaris*, also known as common thyme, a plant native to the Mediterranean region (Spain, Italy, France, Greece, etc.), has long been used as a source of the essential oil (thyme oil) and other constituents (e.g. thymol, flavanoid, caffeic acid and labiatic acid) derived from the different parts of the plant [12]. The pharmacological properties of the plant and of its different extracts, in particular the essential oils, has been thoroughly studied and afforded the many industrial (mainly as food additive) and medical applications of the plant [13]. In addition to their numerous traditional uses, the plant (herb) and its essential oil have found diverse applications in pharmacy and medicine [12,13]. The oil was reported to have antimicrobial (bacteria and fungi) [1,14–16], carminative and expectorant [12] activities, most of which are mediated by thymol and carvacrol, as the phenolic components of the oil, with the former generally more potent. Spasmolytic [17] as well as antioxidant [18,19] activities were also reported for the alcoholic (ethanol) extract of the plant; however, these activities were found to be mediated by non-phenolic components.

The quality control of thyme oil calls for selective and sensitive analytical methods; the European pharmacopoeia (*EP*) [20] and the literature methods are generally based on capillary gas chromatography. In the present communication we report the application of GC/MS analysis, on both polar and non-polar capillary columns, to the evaluation of the essential oils obtained from *T. vulgaris* plants (aerial parts) cultivated at the Herb Garden of Casola-Valsenio (Ravenna, Italy). The variation in the chemical composition of the oil hydrodistilled from the aerial parts at different growth stages during the plant vegetative cycle (particularly during flowering) was, as well, investigated. For better characterization of the summer–winter variations, the oil from plants that was still growing through November/December period was also hydrodistilled. Finally, the oils from young and old plant clusters were concomitantly analyzed and compared.

2. Materials and methods

2.1. Plant materials and oil distillation

The starting vegetative materials consisted of two implants (clusters) of *T. vulgaris* cultivated in the open at the Herb Garden of Casola-Valsenio (Ravenna, Italy). The materials differed principally in age: one recent (Y: young) vegetative implant of 2 years old and the other (O: old) was older with 5 years growing. No physical differences were observed for both cultivated materials except that the older plants showed particularly intense odor.

About 1 kg materials of the plants' aerial parts (tops) were collected in June, while the plant in flowering, and after flowering in July through December period (Table 1), cutting the flowered stems about 5–10 cm just below flowers and avoiding the wooden parts. The collection of the aerial parts was conducted searching the apical dominance of the plants; thus was effected, for the young plant, at the beginning of the vegetative cycle (5 June 1999: May/June period), during the vegetative cycle (3 July 1999: June/July period), before the end of the cycle (24 July 1999: July/Au-

Table 1
Plant materials (aerial parts) of *T. vulgaris* L. (young and old cultivations) collected at various times during the plant vegetative and life cycles

Collection time (year: 1999)	Young plants (2 years)	Old plants (5 years)
5 June (May/June)	+	+
3 July (June/July)	+	+
24 July (July/August)	+	N
6 December (November/December)[a]	+	N

[+]Collection was affected during the mentioned period. [N]No collection was effected in the mentioned period.

[a] Plant material collected after the vegetative cycle.

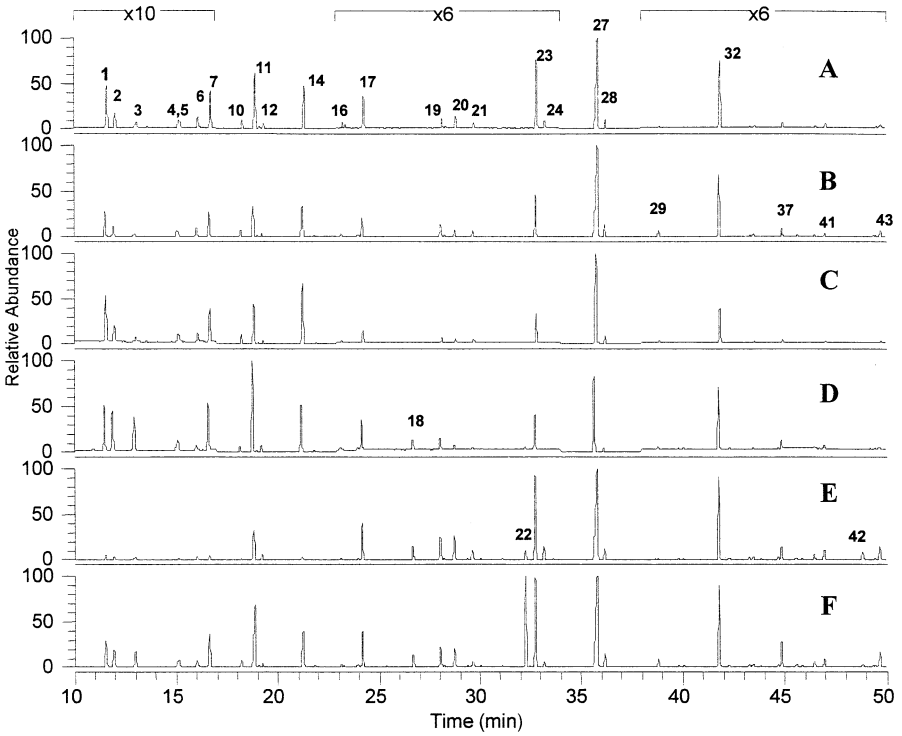

Fig. 1. Typical TIC-GC/MS chromatograms of thyme (*T. vulgaris*) oil analyzed on non-polar Rtx®-5MS capillary column showing profiles of oils obtained from young plants (aerial parts) collected on: (A) 5 June 1999; (B) 3 July 1999; (C) 24 July 1999; and (D) 6 December 1999, and from older plants collected on: (E) 5 June 1999; and (F) 3 July 1999. To ease interpretation, chromatogram-zones between retention times (min): (10–17), (23–34), and (38–50) are amplified by 10, 6, and 6 times, respectively. Peaks' numbers are according to Table 1, to which also refer for peak identity. For chromatographic conditions see experimental.

gust period) and after the cycle been ceased (6 December 1999, November/December period). On the other hand, the collection of the older thyme was only effected in the first two periods (Table 1): in 5 June and 3 July 1999; after then no collection was conducted as the plant life cycle has been ceased (no plant growth) by the end of July. Essential oil was obtained from each collected material by direct steam distillation (water and vapor method) using a medium-scale Clavenger-type apparatus.

2.2. Chromatographic analysis (GC–MS)

2.2.1. Instrumentation (GC/MS and columns)

About 0.1 μl volumes of the tested oils without further modifications were injected into a TRACE GC 2000 SERIES (ThermoQuest CE Instruments, Austin, TX, USA) gas chromatograph equipped with a split–splitless injector (split ratio; 50:1). The non-polar column was an Rtx®-5MS fused silica capillary column (30 m × 0.25 mm ID, 0.25 μm film thickness) consisting of Crossbond® (5% phenyl 95% dimethyl polysiloxane) and the polar one was a Stabilwax® w/Integra-Guard consisting of Crossbond® (Carbowax®-PEG). Helium (He) was the carrier gas at a flow rate of 1.0 ml/min. The GC was interfaced with GCQ plus (ThermoQuest, Finnigan) mass detector operated under the EI mode (70 eV) using an autotune file. The mass spectra were recorded within 40–650 (m/z), full scan mode, that revealed the total ion current (TIC) chromatograms (Fig. 1).

2.2.2. Temperature programs

2.2.2.1. Non-polar column. A linear temperature program was adapted to separate the different oil components depending on an initial temperature of 45 °C held to 10 min, then ramped by 2.5 °C/min up to 180 °C.

2.2.2.2. Polar column. The temperature of the column was initially maintained at 50 °C for 8 min, then raised at a rate of 4 °C/min to 180 °C, at which maintained for 10 min; a second ramp was then applied at 5 °C/min to 220 °C.

In both programs, the temperatures of the injector base, transfer line, and ionization source were maintained at 250, 250 and 200 °C, respectively.

2.2.3. Qualitative and quantitative analysis

The chemical identities of the separated components were determined by matching their recorded mass spectra with the data bank mass spectra (General Purpose, Terpene 'ThermoQuest' and NIST libraries) provided by the instrument software, and by comparing their retention indices (calculated on polar and non-polar columns) with literature values measured on columns with identical [21–23] polarities. Some structures were further confirmed by authentic standards analyzed under the conditions mentioned above; these included α- and β-pinene, α- and γ-terpinene, *p*-cymene, limonene, 1,8-cineole, linalool, thymol, and terpine-4-ol. A mixture of aliphatic hydrocarbons (HCs) in hexane (Sigma) was injected under the above temperature program to calculate the retention indices using the generalized equation by Van Del Dool et al. [24]. Compounds concentrations (as % content) were calculated by integrating their corresponding chromatographic peak areas (non-polar column and TIC mode), assuming a unity response by all.

3. Results and discussion

3.1. General description

The unambiguous identification of the different thyme oil components was achieved using their retention indices obtained from two different (non-polar and polar) chromatographic columns in combination with their ion trap (IT) mass spectral data. The general chemical profiles of the tested oils, the identity and the percentage content of the individual components are all summarized in Table 2. Compounds are reported according to their elution order on the non-polar Rtx®-5MS column (TIC chromatograms in Fig. 1). A chemical class distribution of the oil components is also reported in Table 3. The GC/MS analyses of the oils provided the separation of 46 components, from which 45 structures were well identified using their mass spectra and retention index data [21,23], and online library. Some oil samples were further analyzed using the carbowax-based GC column. This polar column helped to improve the identification procedure of the separated components, in particular those having different elution orders in both utilized columns (Table 2), such as the couples: (α-thujene, α-pinene), (sabinene, β-pinene), (myrcene, α-phellandrene) and (*p*-cymene, limonene) as seen in Table 2. The simultaneous use of retention indices obtained from polar and non-polar columns lead, in fact, to an identification process with a high magnitude of certainty [21,22]. Quantitative measurements of the individual components were, however, obtained using the TIC chromatograms revealed from the non-polar column (Fig. 1) and after repeated analyses.

Generally, the plant oil was characterized by high percentage of the monoterpene phenols that is characteristic for the thymol chemotype growing in Italy [25]. In addition, the oil was characterized by high levels of the precursor monoterpene HCs, *p*-cymene and γ-terpinene, whose concentrations were found to vary in coincidence with the variation in their corresponding phenol products (Table 3). Compared to high level of the total monoterpene HCs, lower figures were observed for the corresponding sesquiterpenes and diterpenoids. According to Table 3 the oil also showed relatively abundant levels, though variable, of the phenolic content, in particular thymol, a marker component that plays an important role in the

Table 2
Composition of the essential oil from *T. vulgaris* L. collected in different periods during the plant's vegetative cycle

#	RI[a]	RI[b]	Compound	% Content*					
				Y1 5 June (1999)	Y2 3 July (1999)	Y3 24 July (1999)	Y4 6 December (1999)	O1 5 June (1999)	O2 3 July (1999)
1	926	1022	α-Thujene	1.28	0.96	2.06	2.90	0.15	0.80
2	932	1018	α-Pinene	0.44	0.08	0.13	2.50	0.10	0.53
3	945	1053	Camphene	0.25	0.14	0.24	2.08	0.13	0.47
4	972	1115	Sabinene	0.17	0.12	0.20	0.39	0.03	0.11
5	973	1095	β-Pinene	0.25	0.17	0.29	0.58	0.05	0.17
6	984	1283	Octen-3-ol	0.35	0.07	0.07	0.27	0.03	0.04
7	993	1156	Myrcene	1.14	0.84	1.28	2.31	0.15	0.88
8	1001	1148	α-Phellandrene	0.36	0.22	0.29	0.35	0.07	0.20
9	1010	1130	3-Carene	0.13	0.07	0.10	0.23	0.03	0.08
10	1014	1170	α-Terpinene	3.10	2.23	3.48	2.67	0.58	2.05
11	1023	1278	*p*-Cymene	21.57	11.61	14.95	32.18	12.88	21.00
12	1028	1196	Limonene	0.91	0.49	0.75	1.36	0.19	0.50
13	1030	1218	1,8-Cineol	1.90	1.15	1.39	2.48	2.40	0.90
14	1059	1230	γ-Terpinene	16.88	13.80	23.34	14.73	1.14	11.57
15	1068	1463	*E*-Sabinene hydrate	0.19	0.70	0.51	0.73	0.13	0.03
16	1089	1270	Terpinolene	0.28	0.15	0.14	0.03	0.10	0.11
17	1102	1540	Linalool	1.57	1.06	0.71	1.75	2.19	1.52
18	1144	[c]	Camphor	NF	NF	NF	0.72	0.85	0.52
19	1165	1705	Borneol	0.42	0.74	0.37	0.85	1.33	0.80
20	1177	1584	Terpin-4-ol	0.65	0.41	0.25	0.35	1.41	0.78
21	1191	1680	α-Terpineol	0.29	0.34	0.22	0.20	0.64	0.26
22	1237	[c]	Thymol methyl ether	0.06	0.03	0.17	0.16	0.55	3.84
23	1246	1965	Carvacrol methyl ether	3.36	2.44	1.63	1.98	5.02	3.68
24	1252	2180	Thymoquinone	0.37	0.03	0.10	0.01	0.84	0.27
25	1286	[c]	Isobornyl acetate	0.06	0.04	0.72	0.19	0.27	0.13
26	1289	[c]	Unk. (thymol der.)	0.11	0.25	0.14	0.12	0.25	0.20
27	1292	2152	Thymol	35.83	51.17	41.38	19.38	54.10	37.33
28	1300	2161	Carvacrol	2.62	4.00	2.47	1.43	3.55	2.96
29	1357	1945	Thymyl acetate	0.27	0.33	0.06	0.18	0.05	0.29
30	1377	1510	α-Copaene	0.02	0.02	tr	0.08	0.04	0.03
31	1386	1535	β-Bourbonene	0.03	0.02	tr	0.16	0.08	0.07
32	1416	1565	β-Caryophyllene	3.50	3.97	2.04	3.15	5.28	3.55
33	1446	[c]	Germacrene D isomer 1	0.03	tr	tr	0.14	0.10	0.06
34	1449	1634	α-Humulene	0.11	0.15	0.05	0.12	0.21	0.13
35	1457	[c]	allo-Aromadendrene	0.04	0.04	tr	0.07	0.06	0.04
36	1477	[c]	γ-Muurolene	0.06	0.04	0.02	0.20	0.18	0.10
37	1481	1675	Germacrene *D*	0.19	0.44	0.17	0.86	0.80	1.07

Table 2 (*Continued*)

#	RI[a]	RI[b]	Compound	% Content*					
				Y1 5 June (1999)	Y2 3 July (1999)	Y3 24 July (1999)	Y4 6 December (1999)	O1 5 June (1999)	O2 3 July (1999)
38	1495	[c]	Valencene	0.07	0.09	0.04	0.30	0.15	0.15
39	1500	[c]	α-Muurolene	0.05	0.04	tr	0.49	0.08	0.06
40	1513	1766	γ-Cadinene	0.10	0.08	0.03	0.16	0.34	0.25
41	1523	1777	δ-Cadinene	0.23	0.18	0.07	0.32	0.66	0.37
42	1562	[c]	Thymodihydoquinone	tr	NF	NF	NF	0.54	0.14
43	1577	[c]	Spathulenol	0.07	0.07	NF	0.02	0.07	0.1
44	1581	2002	Caryophyllene oxide	0.17	0.33	0.06	0.19	0.82	0.64
45	1627	2182	γ-Eudesmol	NF	NF	NF	0.04	0.21	0.07
46	1653	[c]	α-Cadinol	0.03	0.06	NF	0.03	0.13	0.17

Y, essential oils hydrodistilled from young plants (2 years old); O, oil from old plants (5 years); NF, not found; tr, traces (<0.01%); Unk., unknown.

* Mean value of three determinations (three replicates) calculated from the non-polar (Rtx®-5MS) column-TIC chromatograms; relative standard deviation (RSD) values = 1.1–2.3%.

[a] Retention index calculated on non-polar (Rtx®-5MS) column.

[b] Retention index calculated on polar (Carbowax®-PEG) column.

[c] Compound not identified in the polar column GC/MS traces.

Table 3
Class-composition of thyme (*T. vulgaris* L.) oil at different growth stages

Class and individual components	% Content					
	Y1	Y2	Y3	Y4	O1	O2
Monoterpene HCs (mainly *p*-cymene and γ-terpinene)	46.98	31.61[b]	47.76	63.06[a]	15.79	38.54
Monoterpene alcohols	2.93	2.55	1.55[b]	3.15[a]	5.57	3.36
Monoterpene oxides (Eucalyptol)	1.90	1.15[b]	1.39	2.48[a]	2.40	0.90
Monoterpene ketones (Camphor)	NF	NF	NF	0.72[a]	0.85	0.52
Monoterpene phenols						
Thymol	35.83	51.17[a]	41.38	19.38[b]	54.1	37.33
Carvacrol	2.62	4.00[a]	2.47	1.43[b]	3.55	2.96
Monoterpene phenol derivatives (thymol and carvacrol ethers, thymoquinone)	4.17[a]	3.08	2.1[b]	2.45	6.71	8.28
Thymodihydroquinone (TDHQ)	tr	NF	NF	NF	0.54	0.14
Sesquiterpene HCs	4.41	5.07	2.42[b]	6.05[a]	7.98	5.88
Oxygenated sesquiterpenes	0.27	0.51	0.06[b]	0.28[a]	1.32	1.02

Y and O: see Table 1; NF, not found; tr, traces; HCs, hydrocarbons.
[a] Maximum recorded level.
[b] Minimum recorded level.

chemotaxonomy and the overall biological activity of the plant or its oil [4,12,26].

EP in its 4th edition put quality standards for the thyme herb and the oil used as drug in two separate monographs. These standards dealt mainly with the % yield (v/w) of the oil obtained from the herb drug as well as the % content (w/w) of the volatile phenols (expressed as thymol) in the herb. For the thyme oil, allowable percentage ranges (chromatographic profile) of the principal components of the oil are also reported. Worth noting that in the oil monograph a type GC chromatogram is usually provided for information, whereas the chromatographic profile (representative and characteristic components with their tolerance ranges) represents always a standard to be met.

Our tested oils (*young plants*) were found in substantial agreement with these standards (Tables 3 and 4); in particular, those related to the content of thymol (36–55% by *EP*, 35.83–51.17% recorded) and/or the entire phenolic fraction. However, relatively higher and lower levels were observed for γ-terpinene and linalool, respectively (Table 4). Therefore, since most of the oil activities are attributed mainly to the phenolic components, as mentioned above, the oil could serve as a therapeutic equivalent according to official limits and quality standards.

3.2. Compositional variations during vegetative cycle

The data presented in Tables 2 and 3 show the oil composition to vary in a regular manner during the different collection periods (see also Fig.

Table 4
Percent contents of principal thyme oil components compared to official *EP* (4th edition) chromatographic profile

Component	Percentage range by *EP* (% content)	Average % content in the tested oil (Y1–Y3)*
β-Myrcene	1.0–3.0	1.09
γ-Terpinene	5.0–10.0	18.01
p-Cymene	15.0–28.0	16.04
Linalool	4.0–6.5	1.11
Terpin-4-ol	0.2–2.5	0.44
Thymol	36.0–55.0	42.75
Carvacrol	1.0–4.0	3.03

* Oil samples from young plants collected within May/August period.

Table 5
Percentage yields of thyme oil from young and old implants of *T. vulgaris* during the vegetative cycle of the plant

Collection time and period (year: 1999)	% Yield (v/w fresh aerial parts)[a]	
	Young plants (age: 2 years)	Old plants (age: 5 years)
5 June (May/June)	0.52	0.15
3 July (June/July)	0.50	0.15
24 July (July/August)	1.20	–
6 December (November/December)[b]	0.08	–

[a] The limit stated by the *EP* (4th edition) is not less than 1.2% (v/w) of essential oil calculated with reference to the anhydrous drug.
[b] Plants collected after the vegetative cycle been ceased.

1); variations that are mainly attributed to certain individual components and/or compound classes of the oil. This was valid through the correlation between the individual monoterpene phenols (thymol and carvacrol) and their corresponding precursors (*p*-cymene and γ-terpinene) in one side, and between the total HCs and phenol fractions on another side (Table 3). The biosynthetic relationships between these components, reported so far by other researchers [4,26], explains clearly their regular and correspondent variation within the plant life cycle. As expected the young plant was found to have the highest % yield of the oil ($\sim$1.20%, Table 5) during July/August (collection in 24/7/1999) just before the end of the vegetative cycle. In the same collection period (Y3), the highest monoterpene HCs level was observed, while the plant was found to be richest in phenols during June/July period (Y2, Table 3) while in full bloom. These observations were found, moreover, to be in good agreement with those reported by previous investigations carried out on thyme [5–8]. The older plants, on the other hand, provided comparable oil yield during the both collection periods ($\sim$0.15% v/w), with the highest level of phenols and monoterpene HCs in May/June (O1) and June/July (O2), respectively (Tables 2, 3 and 5).

The variation in the oil within the different periods can be generally discussed through the correlation between the precursor monoterpene HCs (*p*-cymene and γ-terpinene, Fig. 2) and the product phenol terpenes (thymol and carvacrol, Fig. 2), which were reported to occur at the end of the biosynthetic pathway as according to Vernert et al. [26]. More specifically, this relation is more observable between *p*-cymene and thymol as strongly manifested by their corresponding % content figures in all of the tested oil samples (Table 2). Consequently, according to Stahl-Biskup [4] and by treating *p*-cymene as a dependent component, a thymol chemotype should be assigned to the plant under study, a typical chemotype of *T. vulgaris* growing in Italy [25,27]. Moreover, the oils showed contrasting highest-content figures for both components (*p*-cymene and thymol) between summer and winter months (Y2 and Y4, Tables 2 and 3). A phenomenon, that according to Weiss and Flück [5], could also assign a summer and a winter chemotypes for the thyme under study.

Switching in between the young and the old plant oils, some interesting observations could be highlighted. The first was concerning the monoterpene ketone (camphor), a compound not frequently reported in the oils from *T. vulgaris* [4]. Camphor was only detected in the oils from the

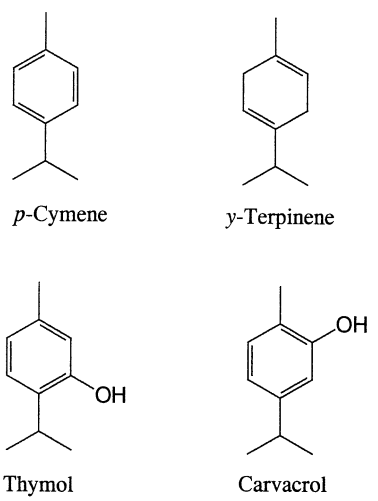

Fig. 2. Structures of the monoterpene HC precursors (*p*-cymene and γ-terpinene) and their biosynthetic products: monoterpene phenols (thymol and carvacrol), quality marker constituents of thyme oil.

old plants as well as the young one collected late in December, when the vegetative cycle of the plant has ceased (Y4, Tables 2 and 3), and was absolutely absent from the young plant oils obtained within May/August period (Y1–Y3, Tables 2 and 3). A delayed activation of the biosynthetic pathway for this ketone could be a reasonable explanation for the mentioned phenomenon [26]. The second observation was exhibited by the distinctive low level of the monoterpene HC γ-terpinene in the oil from the old plant collected in May/June (1.14%, O1, Table 2) compared to that of the other old plant oil (11.57%, O2). This remarkably reduced level of γ-terpinene was not associated with an equivalent variation in the level of the other major monoterpene HC (*p*-cymene) in the same oil sample (relative level of *p*-cymene/γ-terpinene (C/T) = 11.3 in O1 versus 1.81 in O2), whereas a comparable pattern of variation was always characteristic for these two precursors in all of the other samples (Y1–Y4, C/T: 0.6–2.2).

To be mentioned, moreover, that in the same oil sample (O1) the lowest levels of the monoterpene HCs (compounds # 1–12, Table 2 and Fig. 1) and the highest levels of the oxygenated monoterpenes (compounds # 18–21, Table 2), monoterpene phenols (mainly thymol) and their derivatives (mainly carvacrol methyl ether), sesquiterpenes (mainly β-caryophyllene) and their oxygenated derivatives (e.g. caryophyllene oxide) were observed in comparison with all other samples. A particular occurrence of thymodihydroquinone (TDHQ) could be also observed as this thymol derivative was detected almost only in the old plant oils and in the highest concentration in the oil of the first collection (O1).

4. Conclusions

Throughout the entire vegetative phase, the young *T. vulgaris* plant provided a marked higher essential oil yield relative to the older cultivation, with the highest oil content (1.2%) observed in July/August collection period. Regarding the oil composition, the young plant oils were found to be the richest in phenols in June/July (3/7/99) with the minimum levels recorded in November/December (6/12/99), while the contrary figures were observed for monoterpene HCs in both collection periods, respectively. Both component classes (monoterpene HCs and phenols) varied also in the same manner (maximum and minimum) in the older plant samples during the vegetative cycle. For the other minor components (monoterpene alcohols, oxides and ketones and sesquiterpene HCs) a higher content was generally observed in the November/December collection (Y4, Table 2). The study, therefore, emphasized the importance of choosing the appropriate collection (harvest) period of thyme herbs in order to achieve the highest quality and quantity of the essential oil, whose activity is known to be essentially correlated with the content of phenol components.

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