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### Volatile constituents of three *Hypericum* (Hypericaceae) species using GC-MS analysis

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#### Abstract

Essential oils (EOs) composition from three *Hypericum* (Hypericaceae) species (*H. triquetrifolium* Turra, *H. thymifolium* Banks & Sol and *H. perforatum* L.) grown wild in Syria, was investigated using Gas Chromatography-Mass Spectrometry (GC-MS) technique. Data revealed 29, 32 and 52 chemical constituents representing an average of 100, 100 and 99.97% were identified in EOs of *H. triquetrifolium*, *H. thymifolium* and *H. perforatum*, respectively. The major constituents presented were n-Hexadecanoic acid (28.58%), Octadecane (10.42%) and Tricosane (9.66%) for *H. triquetrifolium*, whereas, Isooctyl phthalate (30.39%), Tetracosane (28.18%) and Nonacosane (9.12%) were presented in *H. thymifolium*. As for *H. perforatum*, they were  $\beta$ -Selinol (18.13%), Elemol (12.77%) and  $\beta$ -Elemene (10.73%). Moreover, eleven compounds commonly presented in the three studied EOs *Hypericum* sp. of which the most presented compounds were classified in the following order: n-Hexadecanoic acid > Eugenol > Camphor > Bomeol. The current study allowed somewhat to highlight EOs composition of three *Hypericum* species in Syria for the first time.

**Key-words:** *Hypericum* species, Essential oils (EOs) composition, Gas Chromatography-Mass Spectrometry (GC-MS)

#### Introduction

*Hypericum* is a genus belongs to family Hypericaceae, includes approximately 500 species of flowering plants (herbs, shrubs and trees) (Franklin et al. 2017). It is considered as one of the 100 largest genera of flowering plants, which collectively comprise an estimated 22% of angiosperm diversity. In Syria, it has been reported the occurrence of 21 species belonged to this genus (Mouterde 1970). Many species of this genus are cultivated as ornamentals (*Hypericum* Online: <http://hypericum.myspecies.info>). Previously, Hippocrates and Paracelsus since the ancient Greeks time used this genus for treatment and healing of wounds and as mild antidepressant (Hayek 1996). Among them, *Hypericum triquetrifolium* Turra, *Hypericum thymifolium* Banks & Sol, and *Hypericum perforatum* L. species were found in Syria.

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*H. triquetrifolium* Turra, commonly called curled-leaved St. John's-wort, it is native to the Mediterranean Basin (Wikipedia). Whereas, *H. thymifolium* Banks & Sol is native to Turkey, Lebanon-Syria and Palestine (Plants of the World online). As for *H. perforatum* L. known as St. John's-wort in English; originated from most of Europe, the Azores, the Madeira Islands, the Canary Islands, north-western Africa (i.e. northern Algeria, Morocco and Tunisia), western and northern Asia (i.e. Saudi Arabia, Afghanistan, Cyprus, Iran, Iraq, Lebanon, Syria, Turkey, Armenia, Azerbaijan, Georgia, Russia, Kazakhstan, Kyrgyzstan, Tajikistan, Turkmenistan, Uzbekistan and Mongolia), China and the Indian Sub-continent (i.e. northern Pakistan and India). It is an economically important medicinal plant displayed different role in pharmacology application as anti-viral, anti-depressive and anti-cancer properties ([https://keyserver.lucidcentral.org/weeds/data/media/Html/hypericum\\_perforatum.htm](https://keyserver.lucidcentral.org/weeds/data/media/Html/hypericum_perforatum.htm)).

Different qualitative and quantitative analytical methods were widely employed in *H. perforatum* plants to identify their extracts composition; e.g. High-performance layer chromatography (HPLC), Thin-layer chromatography

(TLC), High-performance thin-layer chromatography (HPTLC) and Gas chromatography-mass spectrometry (GC-MS) techniques (Agapouda et al. 2019).

*Hypericum* species are one of the most attractive medicinal plants due to their enrichment in bioactive components mainly naphthodianthrones, flavonoids, benzophenones/xanthones and essential oils (Franklin et al. 2017). *Hypericum* genus is classified among medicinal plants and used widely in pharmaceutical sciences studies especially *H. perforatum* as nematicidal, antibacterial, antifungal and insecticidal agent worldwide, in Germany, Swiss, France, Italy, Greece, Iran, China, Lithuania, Portugal, Serbia and Uzbekistan (Crockett 2010). *Hypericum* genus is considered as a food supplement worldwide, particularly with the intention to act on the central nervous system (Agapouda et al. 2019).

It is worth noting that medicinal plants researches increased continuously and rapidly to discover novel bioactive molecules on one hand. On the other hand, to improve plants and molecules production that serving in pharmaceutical industries (Franklin et al. 2017).

So, many researches worldwide focused on identification of the essential oils (EOs) composition using Gas Chromatography-Mass Spectrometry (GC-MS) analysis. In this regards, in Turkey (Cakir et al. 1997, Çirak et al. 2010, Ahmed et al. 2013, Özkan et al. 2009, 2013, Küçük et al. 2015, Yüce 2016), Iran (Morteza-Semnani et al. 2006; Jaimand et al. 2012; Sajjadi et al. 2015), Italy (Bertoli et al. 2003), Tunisia (Karim et al. 2007), Greece (Pavlović et al. 2006), India (Weyerstahl et al. 1995), Uzbekistan (Baser et al. 2002), Bulgaria (Vasileva et al. 2003), Serbia (Đorđević 2015), and Tajikistan (Sharopov et al. 2010) and recently in Iraq (Azeez 2017).

However, no data available regarding their EOs composition in Syria. Due to their importance in pharmacology and medicine applications, the present study was conducted on highlighting biochemical constituents of three *Hypericum* (Hypericaceae) species grown wild in Syria using GC-MS analysis.

## Material and Methods

### Plant materials

Areal parts of 10 plants were harvested and bulked as representative for each *Hypericum* sp. Sampling has been carried out during flowering stage from three wild *Hypericum* (Hypericaceae) species grown in their natural habitat from West-Southern regions in Syria. Where, *H. triquetrifolium* Turra was collected from Damascus city. Whereas, *H. thymifolium* Banks

& Sol and *H. perforatum* L. were collected from Lattakia city (Table 1).

Plant samples were shade dried for two weeks, and dry weight ranged between 100-150 g. Dried samples were milled to fine powder by special electric mill and stored separately in glass bowls until oil extraction process. One hundred grams of powder for each species were subjected to essential oil extraction.

### Essential oils (EOs) extraction

Essential oils (EOs) were extracted from areal parts during flowering stage by hydro-distillation method from *H. triquetrifolium*, *H. thymifolium* and *H. perforatum* grown in Syria as reported in many investigations (Bertoli et al. 2003, Özkan et al. 2009, Çirak et al. 2010, Sharopov et al. 2010, Küçük et al. 2015, Yüce et al. 2016) using Clevenger-type apparatus for 4 H.

### Gas Chromatography-Mass Spectrometry (GC-MS) technique

Composition of EOs from the three studied *Hypericum* species was identified by GC-MS analysis using (GC-Agilent 7986, indictor: inert-MS) apparatus equipped with a HP-5MS capillary column (30 m × 0.25 mm; coating thickness 0.25 µm). Analytical conditions were as following: injector and transfer line temperatures 250 °C for each; oven temperature programmed from 60 to 200°C at 3°C/min; carrier gas helium at 1 mL/min; triplicate injections of 0.2 mL (10% hexane solution). Identification of each chemical compound was carried out by comparing its retention index with those of authentic compounds (Formacek and Kubeczka 1982).

## Results and Discussion

Oil yield was 0.22, 0.15, and 0.31% for *H. triquetrifolium*, *H. thymifolium* and *H. perforatum*, respectively. This observation was in accordance of Özkan et al. (2009) who reported that the oil yield was 0.14% in flowering *H. thymopsis* and also with Crockett (2010) who reported that *Hypericum* species overall produced poor essential oil (generally oil yield <1%, w/w); where *H. perforatum* EOs yield was recorded to be 0.35% during the full-bloom stage.

Comparative study among three *Hypericum* (Hypericaceae) species based on their EOs chemical composition has been performed using GC-MS analysis. Essential oils (EOs) composition analyzed by GC-MS analysis was presented for *H. triquetrifolium* (Table 2), *H. thymifolium* (Table 3) and *H. perforatum* (Table 4).

Data revealed 29, 32 and 52 chemical constituents representing an average of 100, 100 and 99.97% were identified in EOs of *H. triquetrifolium* (Table 2), *H. thymifolium* (Table 3) and *H. perforatum* (Table 4), respectively.

Otherwise, eleven compounds commonly presented in the three studied EOs *Hypericum* sp. (Figure. 1).

From data presented in Figure. 1, *H. triquetrifolium* exhibited the highest content compared to the other *Hypericum* sp. Where, these compounds classified in the following order: n-Hexadecanoic acid > Eugenol > Camphor > Borneol. In this regards, n-Hexadecanoic acid was recorded to be 28.58, 2.21 and 6.46% for *H. triquetrifolium*, *H. thymifolium* and *H. perforatum*, respectively. Whereas, its content was 28.0 % in *H. aviculareifolium* oil (Küçük et al. 2015). While, it was presented in the percentage of 2.7%, 17.7%, 9.2% and 23.2% in *H. uniglandulosum*, *H. scabroides*, *H. kotschyanum* and *H. salsugineum*, respectively (Özkan et al. 2013).

It has been demonstrated that n-hexadecanoic acid exhibited different biological properties e.g. as anti-inflammatory, antioxidant, hypocholesterolemic and antibacterial (Aparna et al. 2012, Abubakar and Majinda 2016, Adeoye-Isijola et al. 2018), nematicide, pesticide, lubricant and immunostimulant activities (Adeoye-Isijola et al. 2018).

Nonacosane was 9.12% in *H. triquetrifolium*; whereas, Özkan et al. (2013) reported its percentage of 3.2%, 4.4%, 11.1% and 42.7% in *H. uniglandulosum*, *H. scabroides*, *H. kotschyanum* and *H. salsugineum*, respectively. Whereas,  $\beta$ -Selinene was recorded to be 2.07 and 5.98% for *H. thymifolium* and *H. perforatum*, respectively. Whereas, Yüce (2016) reported a higher value of 19.4% in *H. perforatum*.

EOs composition for each studied *Hypericum* species has been separately discussed. In this regards for *H. triquetrifolium*, the present study revealed that the major identified constituents in their EOs were n-Hexadecanoic acid (28.58%), Octadecane (10.42%), Tricosane (9.66%), Eugenol (6.98%) and Z-7-Hexadecenal (5.41%). The other remaining compounds were weakly presented or in scarce amounts.

Bertoli et al. (2003) reported that the main oil constituents were n-nonane (8%, 15%),  $\beta$ -pinene (8%, 4%),  $\alpha$ -pinene (13%, 10%), myrcene (16%, 5%),  $\beta$ -caryophyllene (5%, 11%), germacrene-D (10%, 13%), sabinene (13%, 3%) and caryophyllene oxide (5%, 12%) in the leaves and flowers *H.*

*triquet trifolium* oils, respectively. Whereas, Karim et al. (2007) reported the major constituents in its oil were Alpha-humulene, cis-calamenene, delta-cadinene, bicyclogermacrene, eremophilene, betacaryophyllene and (E)-gamma-bisabolene in *H. triquet trifolium* oils. Indeed, Jaimand et al. (2012) reported that n-tetradecane (21.3%),  $\alpha$ -himachalene (14.2%) and  $\alpha$ -pinene (10.7%) on flowers, and that  $\alpha$ -himachalen (27%), n-tetradecane (25.7%) and n-pentadecane (7.0%) were presented on leaves were presented in *H. triquet trifolium* oil. Moreover, Sajjadi et al. (2015) reported that Germacrene-D (21.7%),  $\beta$ -caryophyllene (18.3%),  $\delta$ -cadinene (6.4%), trans- $\beta$ -farnesene (4.3%),  $\alpha$ -humulene (3.8%),  $\beta$ -selinene (3.7%),  $\gamma$ -cadinene (3.3%) and trans-phytol (3.2%) were mainly constituents identified in its oil. Recently, Azzez (2017) identified 33 constituents of which Hexenal (E) (12.63%), Octane, 2,3,3-trimethyl (11.36%), Pentadecane, 7-methyl- (9.7%), Undecane (6.15%) and  $\alpha$ -Pinene (5.75%) were mainly found in *H. triquet trifolium* oils leaves.

Whereas, for *H. thymifolium*, the present study revealed that their major identified constituents were Isooctyl phthalate (30.39%), Tetracosane (28.18%), Nonacosane (9.12%), Heneicosane (5.81%), Caryophyllene Oxide (3.34%), 1-Tetradecanol (2.75%),  $\beta$ -Caryophyllene (2.37%),  $\alpha$ -Selinene (2.12%) and  $\beta$ -Selinene (2.07%). Whereas, other compounds presented in scarce amounts e.g. Camphor (0.6%), Borneol (0.53%) and Carvacrol (0.22%).

Hilan and Sfeir (2001) identified 12 chemical constituents using Gas Chromatography tool, of which Limonene (16.7%), Geranyl acetate (15.4%), Terpineol (7.6%) and Geraniol (2.5%) were a major compounds presented in *H. thymifolium* oil. Whereas, the remaining compounds presented in scarce amounts e.g. Camphor (0.2%), Borneol (0.3%) and Carvacrol (0.02%).

As for *H. perforatum*, they were  $\beta$ -Selinol (18.13%), Elemol (12.77%) and  $\beta$ -Elemene (10.73%), n-Hexadecanoic acid (6.46%),  $\beta$ -Selinene (5.98%), Valencene (4.59%), 1S,Cis-Calamenene (3.82%), Aromadendren epoxide -(I) (3.16%), Germacrene d (2.88%), Delta-Cadinene (2.53%), Isoledene (2.42%) and Spathulenol (1.47%) and other components.

Previously, Cakir et al. (1997) reported the presence of  $\alpha$ -pinene (61.7%), 3-carene (7.5%),  $\beta$ -caryophyllene (5.5%), myrcene (3.6%), cadalene (3.2%) and other components in *H. perforatum* oil. Whereas, Baser et al. (2002) reported the occurrence of  $\beta$ -caryophyllene (11.7%), caryophellene oxide (6.3%), spathulenol (6.0%),  $\alpha$ -pinene (5.0%) as a

main constituents in *H. perforatum* oil. Indeed, Vasileva et al. (2003) reported the presence of nonacosane (12%),  $\beta$ -epi-bicyclosesquiphellandren (10%) and bexadecanioc acid, bis(2-2thyl-hexyl)ester (10%) in *H. perforatum* oil.

Whereas, Pavlović et al. (2006) reported the presence of  $\alpha$ -pinene (21.0%) and 2-methyl octane (12.6%) as a major components in *H. perforatum* oil. Moreover, Çirak et al. (2010) reported the presence of  $\beta$ -caryophyllene (4.08–5.93%),  $\gamma$ -muurolene (5.00–9.56%),  $\beta$ -selinene (5.08–19.63%),  $\alpha$ -selinene (4.12–10.42%), d-cadinene (3.02–4.94%), spathulenol (2.34–5.14%), and caryophyllene oxide (6.01–12.18%) as a major compounds in *H. perforatum* oil. Indeed, Sharopov et al. (2010) identified 66 compounds of which germacrene D (13.7%),  $\alpha$ -pinene (5.1%), (E)-caryophyllene (4.7%), n-dodecanol (4.5%), caryophyllene oxide (4.2%), bicyclogermacrene (3.8%), and spathulenol (3.4%) were the major constituents in *H. perforatum* oil. Whereas, Jaimand et al. (2012) reported the presence of E- $\beta$ -farnesene (14.7%), n-hexadecanal (9.1%) and E-nerolidol (7.8%) as major compounds in *H. perforatum* flowers. Moreover, Đorđević (2015) reported germacrene D (18.6%), (E)-caryophyllene (11.2%), 2-methyloctane (9.5%),  $\alpha$ -pinene (6.5%), bicyclogermacrene (5.0%) and (E)- $\beta$ -ocimene (4.6%) as a main compounds in *H. perforatum* oil. While, Yüce (2016) reported the occurrence of forty components in *H. perforatum* oil of which  $\beta$ -selinene (19.4%), bicyclogermacrene (15.3%), 2 tetradecene (8.2%) and  $\alpha$ -amorphene (8.1%) were the major components.

GC/MS analysis has been also employed to investigate EOs composition in other *Hypericum* species. In this regards, Baser et al. (2002) reported that  $\alpha$ -pinene (11.2%), spathulenol (7.2%), p-cymene – (6.1%), acetophenone (4.8%) and carvacrol (4.7%) were the major constituents in *H. scabrum* L. EOs. Indeed, Schwob et al. (2002) reported that ar-curcumene (40%) and g-cadinene (15%) were mainly found in *H. coris* EOs. Whereas, Demirci et al. (2005) reported that  $\beta$ -selinene (15%) and ar-curcumene (8%) were major compounds found in *H. patulum* EOs. Whereas, Morteza-Semnani et al. (2006) reported 85 components in *H. scabrum* L. EOs, of which  $\alpha$ -pinene (45.3%), n-nonane (5.6%) and thymol (5.3%) were found as a main constituents. While, Özkan et al. (2009) reported the presence of 72 compounds of which spathulenol (10.8%), d-cadinene (7.1%), germacrene D (6.1%), g-muurolene (5.9%), 2,3,6-trimethylbenzaldehyde (5%) and g-cadinene (4.4%) were mainly found in *H.*

*thymopsis* EOs. Whereas, Ahmed et al. (2013) reported that  $\alpha$ -pinene was presented in *H. kotschyanum* and *H. thymopsis*; whereas,  $\alpha$ -pinene, baeckeol, limonene and spathulenol were presented in *H. thymopsis* as major components in their EOs. Indeed, Küçük et al. (2015) reported that Hexadecanoic acid (28.0 %), lauric acid (11.3%), myristic acid (9.7%) and caryophyllene oxide (8.7 %) were found as a main components in *H. avicularifolium* EOs. Moreover, Yüce (2016) reported 41 components in *H. Lanuginosum* EOs of which spathulenol (17.3%), caryophyllene oxide (13.1%),  $\alpha$ -pinene (11.7%) and undecane (6.2%) were found as a main constituents. Özkan and Mat (2013) reported the utility of *H. perforatum* in burns, wounds, haemorrhoids, diarrhoea and ulcers treatment in Turkish traditional medicine. Moreover, it displayed wide range spectrum in pharmacology as anti-depressant, anti-inflammatory, anti-microbial, antiviral, anti-nociceptive and wound healing. This species displayed also a potential role in pharmacology studies as anticancer, anti-tumors and anti-AIDS, etc. (Özkan et al. 2009).

It has demonstrated that n-hexadecanoic acid, nonacosane and tricosane exhibited a potential biological role as antioxidant, hypocholesterolemic, anti-inflammatory and antibacterial (Mihailović et al. 2011, Sermakkani et al. 2012, Yugeswari et al. 2012). Whereas, Eugenol exhibited antifungal (Schmidt et al. 2013), anti-allergenic, antiseptic and anaesthetic (Sarkic and Stappen 2018) properties. While, Camphor has anti-inflammatory and analgesic activities (Ghori et al. 2016, Sikka and Bartolome 2018)

The monoterpenes are natural products belonging to the chemical group of terpenes and the main constituents of essential oils. They are found in many bioactive essential oils and medicinal plants [12]. Considering that the monoterpenes are common in many plant species and are used in cosmetic and pharmaceutical preparations, as well as in the food industry. Overall, *Hypericum* spp. EOs composition investigated in the current study was comparable with that those reported by other researches in other countries. These differences could be attributed to the fact that the composition of essential oils notably varied according to geographical distribution, climatic conditions, and other factors (Sanli and Karadogan 2017) within the same species. Of which, geographical distribution and phonological stage were the main factors affect EOs composition in *Hypericum* spp. (Bagci and Bekci 2010), *Thymus algeriensis* and other lamiaceae species (Tangpao et

al. 2018). Thereby, observed differences in the *Hypericum* spp. EOs composition in our case study, significantly related to the fact that the studied samples belonged to three different *Hypericum* spp., and collected from different regions differ in their altitude and annual rainfall.

In conclusion, GC-MS technique was employed to determine EOs composition from three *Hypericum* species (*H. triquetrifolium*, *H. thymifolium* and *H. perforatum*) grown in Syria. GC-MS analysis revealed 29, 32 and 52 chemical constituents representing an average of 100, 100 and 99.97% were identified in EOs of *H. triquetrifolium*, *H. thymifolium* and *H. perforatum*, respectively. Moreover, the major constituents recorded were varied according to the each studied *Hypericum* species. In this regards, for *H. triquetrifolium* they were n-Hexadecanoic acid (28.58%), Octadecane (10.42%) and

Tricosane (9.66%). Whereas, Isooctyl phthalate (30.39%), Tetracosane (28.18%) and Nonacosane (9.12%) were presented in *H. thymifolium*. While, for *H. perforatum*, they were  $\beta$ -Selinol (18.13%), Elemol (12.77%) and  $\beta$ -Elemene (10.73%). Otherwise, data revealed eleven compounds commonly occurred in the three studied EOs *Hypericum* sp. Thereby, future performance research in EOs *Hypericum* sp. is requested due to their high effectiveness as a cheap, natural and bioactive agent in pharmacology studies.

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**Table 1: Description sites of *Hypericum* sp. Collection**

<i>Hypericum</i> spp.	Original site	Sampling date	Altitude (m)	Annual rainfall (mm)
<i>Hypericum triquetrifolium</i> Turra	Damascus	June	970	240
<i>Hypericum thymifolium</i> Banks & Sol	Latakia	May	90	800
<i>Hypericum perforatum</i> L.	Latakia	May	420	850

**Table 2: GS-MS analysis of *H. triquetrifolium* EOs**

Peak Number	Retention Time	Area%	Compound name
1	12.867	0.59	1.8-Cineole
2	15.708	1.91	Trans-Linalool oxide
3	16.775	1.63	Linalool oxide Cis
4	17.808	1.90	Linalool
5	20.383	4.83	Camphor
6	22.150	4.11	Borneol
7	22.600	1.78	Terpineol-4
8	23.617	0.94	$\alpha$ -Terpineol
9	24.225	0.42	Verbenone
10	29.983	1.45	Carvacrol
11	32.050	6.98	Eugenol
12	34.825	0.41	Trans-Caryophyllene
13	37.492	1.05	$\alpha$ -Amorphene

14	39.242	0.84	$\gamma$ -Muurolen
15	39.492	0.83	Delta-Cadinene
16	39.625	0.45	Eugenyl Acetate
17	52.742	1.05	Hexahydrofarnesyl acetone
18	54.867	0.62	Nonadecane
19	58.125	<b>28.58</b>	<b>n-Hexadecanoic acid</b>
20	58.575	0.82	Hexadecane
21	63.242	<b>10.42</b>	<b>Octadecane</b>
22	63.808	2.99	Phytol
23	65.942	2.88	Ambrettolide
24	66.417	5.41	Z-7-Hexadecenal
25	67.950	2.24	Octadecanoic acid
26	69.467	1.83	Tetracosane
27	74.392	1.65	2,6,9,12,16-Pentamethylheptadeca-2,6,11,15-tetraene-9-carboxylic acid
28	78.317	<b>9.66</b>	<b>Tricosane</b>
29	90.717	1.73	Heptacosane
Total		100	

Table 3: GS-MS analysis of *H. thymifolium*EOs

Peak Number	Retention Time	Area%	Compound name
1	12.792	0.21	1,8-Cineole
2	15.625	0.10	Linalool Oxide Trans
3	16.717	0.06	Linalool Oxide CIS
4	17.750	0.29	Linalool
5	20.333	0.60	Camphor
6	22.083	0.53	Borneol
7	22.550	0.21	Terpineol -4
8	23.583	0.10	$\alpha$ -Terpineol
9	29.983	0.22	Carvacrol
10	31.492	0.25	$\alpha$ -Terpineol acetate
11	32.033	0.35	Eugenol
12	33.492	0.29	$\beta$ - Elemene
13	34.600	0.29	(+)-Beta Funebrene
14	34.842	2.37	$\beta$ - Caryophyllene
15	35.008	0.11	Cedrene
16	36.575	1.21	$\beta$ -Famesene
17	37.850	1.28	Cyclododecane

18	38.100	2.07	β.-Selinene
19	38.442	2.12	α-Selinene
20	42.300	3.34	Caryopyllene Oxide
21	45.000	1.59	1-Dodecanol
22	46.517	2.75	1-Tetradecanol
23	52.767	0.39	Hexahydrofarnesyl acetone
24	54.325	0.49	1-Pentadecanol
25	57.250	<b>30.39</b>	<b>Isooctyl phthalate</b>
26	57.742	2.21	n-Hexadecanoic acid
27	59.575	5.81	Heneicosane
28	60.117	<b>9.12</b>	<b>Nonacosane</b>
29	60.908	<b>28.18</b>	<b>Tetracosane</b>
30	63.242	0.43	Nonadecane
31	63.817	1.40	Phytol
32	78.358	1.24	Octadecane
Total		100	

Table 4: GS-MS analysis of *H. perforatum*EOs

Peak Number	Retention Time	Area%	Compound name
1	9.042	0.22	β-Pinene
2	12.617	0.20	D-Limonene
3	12.850	0.14	1,8-Cineol
4	15.667	0.06	Trans-Linalool oxide
5	16.758	0.05	Linalool Oxide CIS
6	17.800	0.28	Linalool
7	20.142	0.03	Trans-Pinocarveol
8	20.400	0.77	Camphor
9	22.150	0.57	Borneol L
10	22.608	0.25	Terpineol-4
11	23.333	0.09	Propanoic acid, 2-methyl-, hexyl ester
12	23.608	0.21	α-Terpineol
13	24.192	0.04	Verbenone
14	25.150	0.04	Isobomylformate
15	26.125	0.23	Cuminal
16	26.542	0.08	Linalylanthranilate
17	28.250	0.02	Bornyl acetate
18	28.417	0.03	Neryl Acetate

19	28.575	0.12	Trans-Anethole
20	30.067	0.64	Carvacol
21	31.375	0.06	$\alpha$ -Cubebene
22	31.558	0.54	$\alpha$ -Terpineol acetate
23	32.167	0.85	Eugenol
24	32.783	0.34	$\alpha$ -Copaene
25	33.175	0.78	(-)- $\beta$ -Elemene
26	33.775	<b>10.73</b>	<b><math>\beta</math>-Elemene</b>
27	34.958	1.55	$\beta$ -Caryophyllene
28	35.392	0.18	$\beta$ -Cubebene
29	36.633	1.25	$\alpha$ -Humulene
30	36.817	1.08	Alloaromadendrene
31	37.667	0.95	$\alpha$ -amorphene
32	37.925	2.88	Germacrene d
33	38.367	5.98	$\beta$ -Selinene
34	38.700	4.59	Valencene
35	38.792	2.42	Isoledene
36	39.667	2.53	Delta-Cadinene
37	39.892	3.82	1S,Cis-Calamenene
38	40.608	0.60	$\alpha$ -Calacorene
39	41.417	<b>12.77</b>	<b>Elemol</b>
40	42.100	0.84	3-Hexen-1-ol, benzoate, (Z)-
41	42.408	0.96	Caryophyllene Oxide
42	43.642	1.21	Humulene Oxide
43	43.750	0.59	Cubenol
44	44.050	0.59	Juniper camphor
45	44.150	0.65	10-Epi- $\gamma$ -eudes mol
46	44.867	6.62	$\gamma$ -Eudes mol
47	46.058	<b>18.13</b>	<b><math>\beta</math>-Selinol</b>
48	47.892	1.47	Spathulenol
49	48.600	3.16	Aromadendren epoxide -(I)
50	58.367	6.46	n-Hexadecanoic acid
51	66.658	0.99	9,12,15-Octadecatrien-1-ol, (Z,Z,Z)-
52	74.542	0.33	5. $\alpha$ -Androstan-3. $\beta$ -ol, 4,4-dim
Total		99.97	

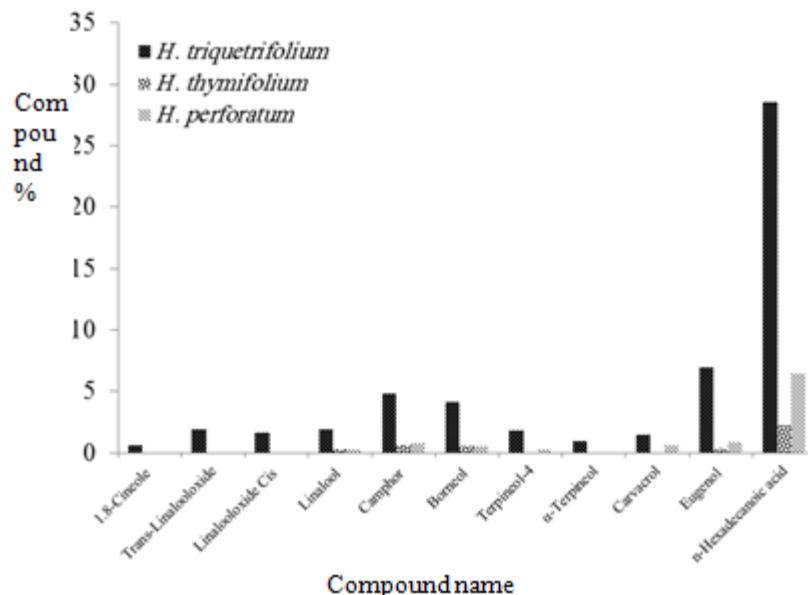


Figure 1: Common compounds (%) presented in the three studied *Hypericum* EO species

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