## Origanum majorana Essential Oil—Chemistry and Pesticide Activity

Subjects: Chemistry, Analytical

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Origanum majorana is a medicinal and aromatic plant that belongs to the Lamiaceae family. It is cultivated in several parts of the world and, due to its splendid aroma and taste, is widely used for culinary purposes and in perfumes. The essential oil of the plant, to which is attributed its aroma, contains many secondary metabolites with valuable biological activity. One of them is the pesticide activity, which has attracted much interest. Given the necessity of replacing synthetic pesticides, essential oils are studied in an attempt to find naturally derived products.

Keywords: Origanum majorana; essential oil; natural pesticide; repellent; fumigant; insecticide

## 1. Introduction

Aromatic plants are plants that produce and exude from their different plant organs (leaves, flowers, etc.) aromatic substances, which are used for cosmetic and culinary purposes. On the other hand, according to WHO, medicinal plants are defined as those plants (wild or cultivated) that contain a mixture of active compounds, able to prevent, relieve or cure diseases or serve as lead molecules for the discovery of new drug formulations. These compounds are synthesized through common biochemical pathways shared by primary and secondary metabolism and are commonly known as secondary metabolites. Plants provide a plethora of secondary metabolites that exert significant biological activity.

Lamiaceae is a family well studied for the presence of secondary metabolites, which includes volatile and nonvolatile compounds that are present as complex mixtures. These complex mixtures provide significant biological activity, making these plants useful in the food, cosmetic and pharmaceutical industries [1][2].

O. majorana L. belongs to the large family of Lamiaceae plants, which consists of 230 genera and almost 7000 species [3]. It is a perennial aromatic, annual herb. Its synonym and accepted botanical name is *Majorana hortensis*, while the plant is commonly known as sweet marjoram. The plant is native to Greece, Cyprus and Turkey; however, it has also been cultivated in Morocco, Egypt, Tunisia, Algeria and elsewhere [4][5].

O. majorana is among the well-studied species of the Lamiaceae family. Its rich chemical profile, either referring to the essential oil fraction or the extracts of the plant, has classified O. majorana as a plant with valuable pharmacological activities [5][6][7][8][9]. In particular, the biological activity of the essential oil derived from the aerial part of the plant has been examined in various studies. Many properties have been attributed to this fraction of secondary metabolites, including antioxidant, antimicrobial, anti-inflammatory, antiacetylcholinesterase, anticancer, antidepressant and analgesic [10][11][12][13][14][15][16]. Apart from the above-mentioned biological activities, the repellent and insecticidal activity of the essential oil of the plant is of maximum importance [17][18][19]. Nowadays, in order to ensure food availability, crops are treated with synthetic pesticides, for which is intensively discussed their negative impact on human health and the environment as well [20]. Biological replacements for synthetic pesticides currently in use could be essential oils [21]. Thus, a considerable number of studies examine the biological activity of essential oils as candidate pesticides against many insect species [22] [23][24]. These naturally derived products aim to protect crops in an eco-friendly manner and at the same time not to adversely affect human health. In particular, regarding the essential oil of O. majorana, its insecticidal, larvicidal, repellent and fumigant activities have been evaluated [19][24][25][26][27][28][29][30][31].

## 2. Chemical Profile of *O. majorana* Essential Oil

Essential oils are complex mixtures, consisting of volatile, usually aromatic, colorless compounds, poorly soluble in water but highly soluble in many organic solvents such as acetone, ethanol and diethyl ether. They are products of the secretory system of the plants, obtained via different procedures, which depend on the plant part used. The most common isolation

methods are hydrodistillation and steam distillation, applied when the essential oil is obtained from the aerial parts of the plant.

Volatiles are accumulated at the glandular trichomes of the reproductive and vegetative organs of the plants that belong to the Lamiaceae family. In particular, they are more abundant in reproductive organs and young leaves [32]. Thus, in general, the most popular parts of the plants used are stems, flowers and leaves, from which essential oil is extracted mainly by steam distillation.

Typical constituents of the essential oils are terpenoids and more precisely monoterpenes, which are flavor compounds and sesquiterpenes, oxygenated or not. Other constituents include derivates of monoterpenes, which means compounds bearing different functional groups such as esters, acetates and alcohols [33]. Monoterpenes and sesquiterpenes are indicated by the molecular formula  $(C_5H_8)n$ , in which n = 2 in the monoterpenes case since they consist of two isoprene units. On the other hand, sesquiterpenes consist of three isoprene units; thus, n equals 3.

For *O. majorana*, characteristic volatile compounds presented in great quantities are monoterpenes hydrocarbons and oxygenated monoterpenes. Other constituents, less in quantity, are sesquiterpenes, oxygenated or not (**Table 1**). As discussed below, the oxygenated monoterpenes prevail in most cases in the *O. majorana* essential oil derived from different geographical regions, with terpinen-4-ol being the most abundant compound [34][35][36][37][38][39][40][41][42][43][44][45][46][47]

In most studies, compounds detected in abundance were terpinen-4-ol, *cis*-sabinene hydrate and *y*-terpinene, while in some cases, the essential oil is rich in carvacrol and thymol, with the concentration of terpinen-4-ol being half of carvacrol or even absent [19][47][48][49][50][51][52][53][54]. Thus, researchers have classified *O. majorana* into two main chemotypes, based on qualitative criteria. The first one is the terpinen-4-ol/cis-sabinene hydrate chemotype, and the second belongs to the carvacrol/thymol type [55][56][57]. However, according to literature data, this is not always the case, as minor exceptions exist. For example, Chaves et al. (2020) [58] studied a sample of *O. majorana* originating from Brazil, which was found rich in pulegone (57.05%). Interestingly, no terpinen-4-ol or *cis*-sabinene hydrate or carvacrol were detected [58]. Furthermore, of the four studies that were found to analyze *O. majorana* from Morocco, one of them classified the studied sample as terpinen-4-ol chemotype (however without the second major in quantity compound being *cis*-sabinene hydrate) [59]; the other study identified the compound found in abundance as 4-terpinene [26], and in the rest of the studies, linalool (32.68%), sabinene hydrate (14.08%) and *trans*-sabinene hydrate (16.0%) were the most characteristic compounds [60][61] [62]. However, remarkably, in the first two studies is the presence of terpinen-4-ol (22.30% and 13.07%, respectively).

Other studies that classified *O. majorana* to a different chemotype are those of Yang et al. (2009), Waller et al. (2016), Baj et al. (2018), Barazandeh et al. (2001) and Dantas et al. (2016)  $\frac{[63][64][65][66][67]}{[63][64][65][66][67]}$ . The first two studies  $\frac{[63][64]}{[63][64]}$  analyzed samples from India and Egypt, respectively, and found the major constituent being 1.8 cineole (50.96% and 20.9%, respectively). On the other hand, samples from Ukraine and Iran were rich in linally acetate (16.0% and 26.1%)  $\frac{[65][66]}{[65]}$ . Dantas et al. (2016)  $\frac{[67]}{[67]}$  studied a sample from Egypt. However, a different chemotype was observed, with  $\gamma$ -terpinene being the compound in abundance followed by  $\alpha$ -terpinene.

Origanum majorana grown in Greece is classified into three chemotypes. Komaitis et al. (1992)  $^{[\underline{68}]}$  determined a terpinen-4-ol chemotype. This cyclic monoterpene constitutes 37.10% of the total content of essential oil, with p-cymene and  $\alpha$ -terpineol being constituents that consist of 50% of the essential oil composition. Daferera et al. (2000)  $^{[\underline{56}]}$  also described an intermediate chemotype of thymol (14.0%) as the main compound. Carvacrol concentration reached 0.2%, while the other compounds found at higher concentrations were 3-carene (10.4%), 2-carene (7.8%), terpinen-4-ol (7.8%) and sabinene hydrate (6.0%). Finally, Giatropoulos et al. (2018)  $^{[\underline{18}]}$  identified a clear carvacrol chemotype, in which the concentration of carvacrol reached 74.8%.

In **Table 1** is given summarized information about the collected literature data regarding the volatile profile of *O. majorana*. The most popular parts of the plant used are stems, flowers and leaves [ $^{[32]}$ ], from which essential oil is extracted mainly by steam distillation, a method adopted by the majority of researchers, as concluded from **Table 1**. Considerable variability is observed regarding the chemical composition of the plant, as well as the percentage yield of its essential oil. Terpinen-4-ol, *cis/trans*-sabinene hydrate, y-terpinene, *cis*- $\beta$ -terpineol, carvacrol and thymol are the compounds mentioned in abundance in the studied samples. Regarding the essential oil yield from the aerial parts of the plant, the % yield ranges from 0.4 to 1.85 mL/100 g of dry material, while when only leaves were used, the extent of the % yield ranges from 0.09 to 2.5 mL/100 g of dry material.

This chemical diversity of essential oil isolated from *O. majorana* samples is a product of different parameters such as the growth stage of the plant, climate variability, irrigated or arid crops, geographical area, soil salinity, storage conditions and

method of distillation [69][70]. All these variables influence the production of secondary metabolites, thus affecting both the qualitative and quantitative composition of an essential oil. In particular, limited water availability is a factor that decreases crop yield and essential oil yield, or is even responsible for altering an essential oil composition.

**Table 1.** *Origanum majorana* essential oil from different geographic regions.

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Referenc
200 g of plant material (the part used is not identified)	Hydrodistillation (clevenger apparatus)	VB-5 30 × 0.25 mm, 0.25 μm	0.8 mL/100 g dry material	4-terpinene (28.96%), y- terpinene (18.57%) and α-terpinene (12.72%), sabinene (8.02%)	Morocco	[ <u>26]</u>
1000 g of the aerial parts	Hydrodistillation (according to European Pharmacopeia 5th edition guidelines)	DB-5 30 m × 0.25 mm, 0.33 μm	0.97 mL/100 g dry material	terpinen-4-ol (34.1%), α- terpinene (19.2%), terpineol (8.9%)	South West Morocco	<u>[59]</u>
10 g of plant material	Steam distillation (Likens–Nickerson apparatus)	CP-Sil 8 30 m, 0.32 mm	-	thymol (14.0%), 3- carene (10.4%), 2- carene (7.8%), terpinen-4-ol (7.8%), sabinene hydrate (6.0%)	Greece	<u>[56]</u>
100 g of aerial part (stems, leaves and flowers)	Hydrodistillation (clevenger apparatus)	HP-5MS 30 m × 0.25 mm, 0.25 μm	1.85 mL/100 g dry material	terpinen-4-ol (23.2%), cis- sabinene hydrate (17.5%), γ- terpinene (10.5%), p- cymene (9%), α-terpineol (5.6%)	Tunisia	<u>[57]</u>
L00 g of leaves	Hydrodistillation (Quik-fit apparatus)	HP-5MS 30 m × 0.25 mm, 0.25 mm	0.09 mL/100 g dry material	terpinen-4-ol (555.1 µg/g dw), y- terpinene (192.8 mg/g dw), cis sabinene hydrate (168.8 mg/g dw)	Tunisia	<u>[34]</u>
100 g of aerial parts) three developmental stages: vegetative, flowering and post-flowering)	Hydrodistillation (clevenger apparatus)	HP-5MS 30 m × 250 m, 0.25 μM	-	terpinen-4-ol (76.94-37.15), cyclohexanol 3,3,5 trimethyl (15.99-5.41), α-terpineol (11.34-0.94); β-cymene (10.56-1.88)	Tunisia	<u>[46]</u>
-	Steam distillation	Carbowax 20M 25 m × 0.3 mm	0.20 mL/100 g dry material	terpinen-4-ol (37.10%), p- cymene (12.05%), α- terpineol (7.15%)	Greece	<u>[68]</u>

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Reference
Leaves	Hydrodistillation (clevenger apparatus)	DB-5MS 30 m × 0.25 mm × 0.25 μm	1.2 mL/100 g dry material	terpinen-4-ol (29.97%), γ- terpinene (15.40%), trans-sabinene hydrate (10.93%), α- terpinene (6.86%) and α- terpineol (6.54%)	Egypt	[ <u>13]</u>
5 g	Hydrodistillation (clevenger apparatus)	Rtx-5MS 30 m × 0.25 mm × 0.25 μm	-	terpinen-4-ol (19.7%), y- terpinene (18.4), α- terpinene (11.4%), cis- sabinene hydrate (8.6%), sabinene (7.8%)	Commercial sample Germany	[ <u>71</u> ]
Leaves	Hydrodistillation (clevenger apparatus)	DB5 30 m × 0.25 mm × 0.25 μm	-	carvacrol (57.86%), thymol (13.54%), trans- caryophyllene (11.52%), cymene (6.78%)	Iran	[ <u>48]</u>
Aerial parts	Hydrodistillation (clevenger apparatus)	DB-5 30 m × 0.25 mm, 0.25 μm	-	terpinen-4-ol (31.15%), cis- sabinene hydrate (15.76%), p- cymene (6.83%), sabinene (6.91%), trans- sabinene hydrate (3.86%), α- terpineol (3.71%)	India	[ <u>35</u> ]
500 g of leaves	Hydrodistillation (clevenger apparatus)	HP-5MS 30 m × 0.25 mm, 0.25 μm	0.6 mL/100 g dry material	cis-sabinene hydrate (30.2%), terpinen-4-ol (28.8%), γ-terpinene (7.2%), α-terpineol (6.9%), transsabinene hydrate (4.4%), linalyl acetate (3.8%), α-terpinene (3.6%)	Venezuela	[72]

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Reference
20 g of aerial part (two vegetative and two generative growth stages)	Hydrodistillation	HP-Innowax 30 m × 0.25 mm × 0.25 mm	0.04 to 0.09 mL/100 g dry material	terpinen-4-ol (29.13– 32.57%), <i>cis</i> - sabinene hydrate (19.9– 29.27%), <i>trans</i> - sabinene hydrate (3.5– 11.61%), γ- terpinene (2.11–8.20%), bornyl acetate (1.52–2.94%), linalool (1.05– 1.39%)	Tunisia	<u>[36]</u>
-	Hydrodistillation	Supelcowax 10, 60 m × 0.25 mm, 0.25 µm	0.8 mL/100 g dry material	terpinen-4-ol (30.3%), γ- terpinene (14%), linalool (12%), p-cymol (9.8%), α- pinene (5.9%), camphene (5.8%)	Hungary	<u>[37]</u>
Flowering plants	Hydrodistillation (clevenger apparatus)	Carbowax 20 M, 50 m × 0.32 mm i.d, 0.20 μm	1 mL/100 g dry material	terpinen-4-ol (38.4%), cis- sabinene hydrate (15.0%), p- cymene (7.0%), y- terpinene (6.9%).	Reunion Island	[38]
-	Hydrodistillation (clevenger apparatus)	Equity-5 60 m × 0.32 mm, 0.25 μm	0.45– 0.50 mL/100 g dry material	cis-sabinene hydrate (20.23– 46.27%), terpinen-4-ol (9.32–23.43%), y-terpinene (5.67–13.76%), α-terpinene (2.98–8.38%), sabinene (4.90–8.17%), trans-sabinene hydrate (5.01– 7.34%), α-terpineol (3.41–4.17%)	India	[49]
Leaves	Hydrodistillation (clevenger apparatus)	DB-5 (5% phenylmethylpolysiloxane) capillary column, 60 m × 0.25 mm	1.6 mL/100 g dry material	terpinen-4-ol (30.0%), y- terpinene (11.3%), trans- sabinene hydrate (10.8%)	Egypt	[25]
131 g leaves	Hydrodistillation (clevenger apparatus)	OPTIMAL-5 0.25 μm, 30 M, 0.25 mm	-	pulegone (57.05%), verbenone (16.92%), trans- menthone (8.57%)	Brasil	<u>[59]</u>
0.5 kg of aerial part	Hydrodistillation (clevenger apparatus)	HP-5 30 m × 0.25 mm, 0.25 μm	-	carvacrol (74.8%), thymol (2.7%)	Greece	[ <u>18]</u>

		Column Used for the GC		Chemical		
Plant Material	Extraction Method	Analysis	% Yield	Composition	Region	Reference
-	-	DB-1MS 30 m × 0.25 mm, 0.25 μm	-	terpinen-4-ol (22.96%), linalool (15.32%), y- terpinene (12.92%), p- cymene (6.37%)	Commercial sample, Korea	[ <u>23</u> ]
-	-	VF-5MS 30 m × 0.25 mm, 0.25 μm	-	terpinen-4-ol (33.8%), terpinolene (16.5%), linalool (14.7%), α- terpinene (6.8%)	Commercial sample	[73]
20 g dried leaves	Hydrodistillation (clevenger apparatus)	-	12.70 μL·g <sup>−1</sup>	terpinen-4-ol (23.83%) <i>cis</i> -β- terpineol (21.63%),	-	[7 <u>4</u> ]
-	-	DB-5MS 30 m × 0.25 mm, 0.25 μm	-	1,8-cineole (50.96%), linalool (24.04%), limonene (6.38%)	India	[ <u>63]</u>
Two samples from different regions were analyzed	Hydrodistillation	-	6.5–7.7 mL/100 g dry material	carvacrol (78.27– 79.46%), p- cymene (4.31– 4.68%), y- terpinene (3.72–4.84%)	Turkey	<u>[55]</u>
100 g of fresh plant material	Steam distillation	SE-54 50 m × 0.32 mm		linalool (32.68%), terpinen-4-ol (22.30%), p- cymene (8.07%)	Morocco	<u>[60]</u>
80 g of aerial parts	Hydrodistillation (clevenger apparatus)	HP-5MS 30 m × 0.25 mm, 0.25 μm	17.2 g/kg	terpinen-4-ol (20.9%), linalool (15.7%), linalyl acetate (13.9%), limonene (13.4%), α- terpineol (8.57%)	Pakistan	[ <u>39]</u>
1 kg of dried aerial parts	Hydrodistillation	DB-5 30 m × 0.25 mm, 0.33 μm	0.4 mL/100 g dry material	terpinen-4-ol (29.6%), δ-2- carene (20.1%), camphene (13.4%), α- pinene (7.9%)	Italy	[11]
100 g dried aerial parts	Hydrodistillation (clevenger apparatus)	Cp WAX 52 CB 50 m × 0.32 mm, 1.2 μm	-	carvacrol (52.5%), linalool (45.4%),	Turkey	[ <u>50]</u>

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Reference
100 g of dried aerial part	<ul> <li>Microwave- assisted extraction (MWE)</li> <li>Hydrodistillation (HD)</li> <li>Steam distillation (SD)</li> </ul>	TR-5 MS 30 m × 0.32 mm, 0.25 μm	(HD) 0.73 mL/100 g dry material (MWE) 0.80 mL/100 g dry material (SD) 0.66 mL/100 g dry material	terpinen-4-ol MWE: 22.28%, HD: 28.49%, SD: 26.72% trans-sabinene hydrate MWE: 13.05%, HD: 11.69%, SD: 3.04% y-terpinene MWE: 13.20%, HD: 7.87%, SD: 13.72% \alpha-terpinene MWE: 9.07%, HD 3.89%, SD: 9.46%	Egypt	[47]
300 g of plant material	Hydrodistillation (clevenger apparatus)	-	1.7 mL/100 g dry material	terpin-4-ol (27.32%), γ- terpinene (15.67%), α- terpinene (11.08%) α- terpineol (6.90%), sabinene (5.53%)	Tunisia	[ <u>51</u> ]
Aerial parts	Extraction with organic solvent	ZB-5MS (Phenomenex), 30 m × 0.25 mm, 0.25 μm		trans-sabinene hydrate (16.0%), sabinene (14.1%), cissabinene hydrate (11.8%), yterpinene (10.2%), $\alpha$ terpinyl acetate (10.0%), $\alpha$ terpinene (8.9%)	Yemen	[ <u>62]</u>
-	-	DB-5 30 × 0.25 × 2.5 mm	-	terpinen-4-ol (20.55%), terpinene (13.13%), trans-terpineol (12.67%), 2- carene (7.67%), sabinene (6.96%)	-	[ <u>40]</u>
-	-	ZB-5 MS 30 m, 0.25 mm, 0.25 μm	-	linalyl acetate (16.0%), linalool (14.7%), $\alpha$ -terpineol (13.8%), limonene (11.5%)	Commercial sample produced in Ukraine	[65]
-	Hydrodistillation (clevenger apparatus)	HP-5MS 30 m× 0.25 mm, 0.25 mm	-	terpinen-4-ol (32.69%), γ- terpinene (12.88%), trans-sabinene hydrate (8.47%), α- terpinene (7.98%), sabinene (6.21%)	-	<u>[15]</u>

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Reference
200 g of aerial part	Hydrodistillation (Dean–Stark apparatus)	VB5 30 m × 0.25 mm 0.25 μm	1.06 mL/100 g dry material	Sabinene hydrate (14.08%), α- terpineol (13.95%), (-)- terpinen-4-ol (13.07%), (+)- sabinene (5.67%)	Morocco	[ <u>61]</u>
-	-	HP-5 30 m× 0.32 mm× 0.25 mm	-	1,8-cineole (20.9%), terpinen-4-ol (20.4%), p- cymene (7.0%), sabinene (6.7%)	Commercial sample Egypt	[ <u>64</u> ]
Dried leaves	Hydrodistillation (clevenger apparatus)	DB-5 30 m × 0.25 mm 0.25 mm	1.20 mL/100 g dry material	terpinen-4-ol (30.41%), γ- terpinene (13.94%), cis- sabinene hydrate (9.64%), α- terpinene (7.70%)	Egypt	[ <u>52]</u>
-	-	Restek 30 m × 0.32 mm, 0.50 μm	-	terpinen-4-ol (21.3%), trans- sabinene hydrate (15.5%), γ- terpinene (14.0%) and α- terpinene (8.9%)	Commercial product Albania	<u>[53]</u>
Aerial parts of plant material collected in different regions	Hydrodistillation (clevenger apparatus)	FSC 60 m × 0.25 mm, 0.25 μm	-	terpinen-4-ol (8–14%), linalyl acetate (7–10%), <i>trans</i> - sabinene hydrate (6–7%)	Turkey	[ <u>41</u> ]
100 g of air- dried aerial parts	Hydrodistillation (Dean–Stark apparatus)	HP-101 25 m × 0.32 mm	1.40 mL/100 g dry material	terpinen-4-ol (32.8%), y- terpinene (9.9%), <i>cis</i> - sabinene hydrate (8.6%)	Tunisia	<u>[54]</u>
Dried leaves	Hydrodistillation (clevenger apparatus)	TR-5MS 30 m × 0.25 mm, 0.25 μm	2.5 mL/100 g dry material	terpinen-4-ol (33.0%), caryophyllene oxide (11.9%), p-cymene (6.8%), \(\alpha\)- terpineol (6.7%) spathulenol (6.0%)	Commercial sample China	[ <u>42</u> ]

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Reference
200 g dried flowers 200 g dried leaves	Hydrodistillation (clevenger apparatus)	Supelcowax 10 30 m × 0.32 mm, 0.5 pm	12.8 mL/100 g dry material (flowers) 8% ml/100 g dry material (leaves)	Leaves: cissabinene hydrate (33.3%), terpinen-4-ol (21.6%), y-terpinene (8.3%), \(\alpha\)-terpineol (7.3%), \(transsabinene\)-table hydrate (4.7%) Flowers: \(cissabinene\)-terpineol (16.6%), \(\alpha\)-terpinene (10.6%) Stems: terpinen-4-ol (19%), \(\alpha\)-terpineol (14.25%), y-terpinene (11.1%), \(cissabinene\)-terpinene (11.1%), \(cissabinene\)-terpinene hydrate (7.4%)	Cyprus	[69]
Flowers	Steam distillation	DB-1 60 m × 0.25 mm, 0.25 pm	0.3 mL/100 g dry material	linalyl acetate (26.1%), sabinene (12%), y- terpinene (8.8%), cis- sabinene hydrate (8.7%)	Iran	[ <u>66]</u>
-	-	-	-	-	Egypt	[ <u>43</u> ]
-	-	DB-1 30 m × 0.25 mm, 0.25 μm	-	terpinen-4-ol (20.8%), y- Terpinene (14.1%), cis- sabinene hydrate (10.8%) sabinene (9.3%), α- terpinene (9.2%)	Commercial sample UK	[44]
300 g of aerial parts	Hydrodistillation (clevenger apparatus)	-	1.72 mL/100 g dry material	terpinen-4-ol (26.7%), γ- terpinene (16.96%), p- menthenol (11.85%), α- terpinen (9.22%), α- terpineol (5.76%), p-cymene (5.27%)	Tunisia	[ <u>45]</u>
Dried leaves	Hydrodistillation (clevenger apparatus)	Durabond-DB5 30 m × 0.25 mm × 0.25 μm	-	y-terpinene (25.73%), α- terpinene (17.35%), terpinen-4-ol (17.24%), sabinene (10.8%), β- phellandrene	Egypt	[ <u>67]</u>

Plant Material	Extraction Method	Column Used for the GC Analysis	% Yield	Chemical Composition	Region	Reference
200 g aerial part	(a) Microwave- assisted hydrodistillation (b) Hydrodistillation	HP-5 ms capillary 30 m × 0.25 mm, 0.25 μm	5 mL/100 g of dry material	(a) carvacrol (41.3%), linalool (12.2%), terpinen-4-ol (6.6%), linalyl acetate (6.8%), y-terpinene (5.1%) (b) carvacrol (39.1%), linalool (7.2%), terpinen-4-ol (10.1%), linalyl acetate y-terpinene (6.8%), (3.2%)	Greece	(7 <u>0</u> )
-	Hydrodistillation (clevenger apparatus)	-	0.2 mL/100 g of dry material	carvacrol (43.7%), thymol (18.3%), γ- terpinene (14.1%), ο- cymene (8.1%), α- terpinene (2.0%)	Greece	[ <u>75</u> ]

The concentration of *cis*- and *trans*-sabinene hydrate ranges from 0.95% to 46.27%. This difference can be explained by taking into account the influence of abiotic components on the production of essential oils. As reported in the study of Novak et al. (2002) [71], increased temperature resulted in increased production of sabinene hydrate. In addition, apart from the effect of temperature, a longer period of sunlight had a positive influence on the production of *cis*- and *trans*-isomers, while the opposite was observed regarding the terpinene content [76]. Furthermore, row planting arrangement seems to be important. Single-row planting yielded essential oils richer in sabinene than binate rows. This effect was explained by the fact that single rows receive more sunlight [77]. On the other hand, the cyclic monoterpenes  $\alpha$ - and  $\gamma$ -terpinene are frequently stated as components of the essential oil. Terpinen-4-ol,  $\alpha$ -terpinene and  $\gamma$ -terpinene are typical products derived from a rearrangement reaction that follows the distillation process due to elevated temperature [78][79].

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