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Research Article
Essential Oil Constituents of Summer Savory Plants Propagated and Adapted under Egyptian Climate

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Abstract
The essential oils obtained by water distillation from aerial parts of summer savory (*Satureja hortensis* L.) yielded 1.2% v/w on a dry weight. Eight constituents representing 99.7% of the *S. hortensis* essential oils were identified. The major constituents of *S. hortensis* essential oils were γ-terpinene (46.4%), carvacrol (40.2%) and α-thujene (8.8%). The obtained constituents from *S. hortensis* essential oil under egyptian conditions grouped into three classes which are Monoterpene Hydrocarbons (MH), Oxygenated Monoterpenes (OM) and Sesquiterpene Hydrocarbons (SH). It is evident that the MH reached its highest concentrations (57.8%) followed by OM (41.3%) in essential oil compared with the minor chemical class (SH) (0.6%).

Key words: *Satureja hortensis* L., essential oil, monoterpene hydrocarbons, oxygenated monoterpenes, sesquiterpene hydrocarbons

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The use of natural products with therapeutic properties is an ancient as human civilization and for a long time, plant products were the main sources of drugs (De Pasquale, 1984). World Health Organization (WHO) estimated that 80% of the populations of developing countries rely on traditional medicines, mostly plant drugs for their primary health care needs. Also modern pharmacopoeia still contains at least 25% drugs derived from plants and many others which are synthetic analogues built on prototype compounds isolated from plants (Bodeker *et al*., 1997). Summer savory or *Satureja hortensis* L., belongs to Lamiaceae family. It is a widely distributed, annual plant, cultivated in many parts of the world. It is one of the most important of many classified *Satureja* species (Silic, 1979). *Satureja hortensis* is an aromatic herb, spice and natural food preservative. It used in traditional folk medicine in the treatments of cardiovascular diseases, thrombosis (Yazdanparast and Shahriyary, 2008), muscle pain, stomach and intestinal disorders (Hajhashemi *et al*., 2000) and an anti inflammation agent in the treatment of rhino sinusitis (Uslu *et al*., 2003). Extracts and essential oils of this plant species have antioxidant (Gulluce *et al*., 2003), antibacterial (Gulluce *et al*., 2003; Sahin *et al*., 2003) and antifungal activities (Gulluce *et al*., 2003; Boyraz and Ozcan, 2006). Chemical composition essential oil of *S. hortensis* has a no studies under the conditions of Egypt, so, the present investigation was carried out for the propagation and adaptation to the environmental conditions of Egypt as well as study the essential oil composition of *S. hortensis* plants imported from Enza Zaden Worldwide Company, Netherlands, as a new natural source of essential oil in Egypt.

MATERIALS AND METHODS

Plant material: *Satureja hortensis* seeds were introduced from Enza Zaden Worldwide Company, Netherlands and were cultivated in Egypt for the propagation and adaptation to the environmental conditions of the country as a new source of natural products. Seeds were sown in the open field during the first week of April, 2014.

Harvesting: After 60 days from sowing date, the plants were harvested by cutting the plants 5 cm above the soil surface. Total fresh and dry weights of the herbs (gram per plant) were recorded.

Isolation of the essential oil: Air-drying of plant material was performed in a shady place at room temperature for 10 days. The dried aerial parts [100 g from each replicate (four replicates)] were cuts and subjected to hydro-distillation for 3 h using a clevenger-type apparatus (Clevenger, 1928).

The essential oil content was calculated as a relative percentage (v/w). The resulting essential oil was dried over anhydrous sodium sulfate and stored at 4°C.

Essential oil yield analysis: The qualitative analysis of the essential oil compounds was performed on a Gas Chromatograph (GC) coupled to a Mass Spectrometer (MS) (GC-MS; Shimadzu QP5000), operating at an MS ionization voltage of 70 eV. Model: GC-MS- QP 2010 SE, Column flow up to 4 mL min^{-1} and variety of column selection, analytical method transfer available based on GCMS-QP5000 series, nables direct sample injection (DI) and easy expandability without Any changes to the GC, ecology mode for lower running cost in laboratory; power consumed in analysis standby mode reduced 40%.

The chromatography was equipped with a fused silica capillary column DB-5 (J and W Scientific; 30 m × 0.25 mm × 0.25 μm) and helium was used as the carrier gas. The following chromatography conditions were used: injector at 240°C, detector at 230°C, gas flow 1.0 mL min^{-1}, split 1/20, initial column temperature of 60-240°C at a rate of 3°C/per minute and a 1 μL injection of solution (1 mg of essential oil and 1 mL of ethyl acetate).

The compounds were identified using the comparative analysis of the acquired mass spectra with those stored in the GC-MS database of the system (Nist 62.Lib), the literature (McLafferty and Stauffer, 1989) and retention indices (Adams, 2007), which were obtained from the injection of a mixture of n-alcanes (C_9H_2O-$C_{25}H_{52}$, Sigma Aldrich, 99%) employing a column temperature program as follows: 60-240°C at a rate of 3°C min^{-1}. Separation and quantification (normalization area method) of the substances were carried out by GC with the same type column as used for GC/MS and same conditions.

Qualitative and quantitative analyses: Identifications were made by library searches combining MS and retention data of authentic compounds by comparison of their GC Retention Indices (RI) with those of standards available in our laboratories. The retention indices were determined in relation to a homologous series of n-alkanes under the same operating conditions. Further identification was made by comparison of their mass spectra on both columns with those stored in NIST 98 and Wiley 5 Libraries or with mass spectra from literature.

Component relative concentrations were calculated based on GC peak areas without using correction factors.

RESULTS

The essential oils obtained by water distillation from aerial parts of *S. hortensis* yielded 1.2% v/w on a dry weight. Eight constituents representing 100% of the *S. hortensis* essential oils were identified. The major constituents of *S. hortensis* essential oils were γ-terpinene (46.4%), carvacrol (40.2%) and α-thujene (8.8%). Table 1 represents the obtained constituents from *S. hortensis* essential oil under egyptian conditions. The identified constituents were grouped into three classes which are Monoterpene Hydrocarbons (MH), Oxygenated Monoterpenes (OM) and Sesquiterpene Hydrocarbons (SH). From the same table it is evident that the MH reached its highest concentrations (57.8%), followed by OM (41.3%) in essential oil compared with the minor chemical class (SH) (0.6%). The MH included the constituents of α-thujene, β-thujene, ρ-cymene, γ-terpinene and 1, 3, 8-ρ-menthatriene. The OM included the constituents of carvacrol and carvacrol acetate. The SH Included the constituent of β-caryophyllene. These results indicate that *S. hortensis* grown in Egypt belongs to the γ-terpinene-carvacrol chemo-type. These constituents represent 86.6% (area percent) of the total *S. hortensis* essential oil.

DISCUSSION

The previous investigations showed a significant effect of different locations on essential oil extracted from *S. hortensis* essential oil (Ibrahim *et al*., 2014). The essential oil of *S. hortensis* collected in Serbia yielded 2.1%. Thirty six components (86.1%) were identified as constituents of this essential oil by combined GC/FID and GC/MS analyses. The major components were carvacrol (67.00%), γ-terpinene (15.3%) and ρ-cymene (6.73%). In a smaller percent, α-terpinene (1.29%), β-caryophyllene (1.90%) and β-bisabolene (1.01%) were identified as constituents of the oil.

Table 1: *Satureja hortensis* L. essential oil constituents

Constituents (%)	RI	Peak area (%)	Class	Identified method
α-thujene	923	8.8	MH	RI, MS
β-thujene	980	0.5	MH	RI, MS
ρ-cymene	1027	1.7	MH	RI, MS
γ -terpinene	1062	46.4	MH	RI, MS
1, 3, 8-ρ-menthatriene	1108	0.4	MH	RI, MS
Carvacrol	1299	40.2	OM	RI, MS
Carvacrol acetate	1371	1.1	OM	RI, MS
β -caryophyllene	1428	0.6	SH	RI, MS
MH = Monoterpene hydrocarbons		57.8		
OM = Oxygenated monoterpenes		41.3		
SH = Sesquiterpene hydrocarbons		0.6		
Total identified		99.7		

RI: Confirmed by comparison with retention indices on DB5 column (Adams, 2007)

The monoterpene prevalence in the oil (82.3%) was evident, while the most abundant were oxygenated monoterpenes (69.1%).

In addition, sesquiterpene hydrocarbons (3.2%) and oxygenated sesquiterpenes (0.5%) were isolated (Mihajilov-Krstev *et al*., 2009). Thymol, ρ-cymene, γ-terpinene and carvacrol were the main components of *Satureja hortensis* L. grown in Iran (Mahboubi and Kazempour, 2011). According to Gulluce *et al*. (2003), the main constituents of Croatian *S. hortensis* essential oil were thymol (29.0%), carvacrol (26.5%), γ-terpinene (22.6%) and ρ-cymene (9.3%). Baser *et al*. (2004) pointed out that the plants cultivated on the territory of the former Yugoslavia had the highest yield of essential oil (2.7%), while the plant material cultivated in Italy had the lowest yield of oil (0.6%). In addition to the high content of the essential oil, *S. hortensis* cultivated on the territory of the former Yugoslavia contained a relatively high level of carvacrol (44.0%). Beside carvacrol, the essential oil also contains γ-terpinene from 6.0% (South American oil), to 60.3% (oil from material of Lawrence), ρ-cymene from 4.5% (Lawrence oil) to 35.8% (Russian oil) and thymol from 8.6% (Russian oil) to 18.0% (South American oil).

The same authors, according to their analysis of 20 samples from different localities in Turkey, concluded that cultivated forms of *Satureja hortensis* L. contained carvacrol as the dominant component of oil (42.0-63.0%), while in the oil of wild growing forms thymol dominated as the main component (29.0-43.0%) (Baser *et al*., 2004). It may be concluded that the changes in essential oil of *Satureja hortensis* L. cultivated in Egypt compared with the essential oil of *Satureja hortensis* L. cultivated in other countries may be due to its effect of locations on enzyme activity and metabolism of essential oil production and its constituents (Burbott and Loomis, 1969).

CONCLUSION

It may be concluded that the essential oils obtained by water distillation from aerial parts of *S. hortensis* yielded 1.2% v/w. The major constituents of *S. hortensis* essential oils grow in Egypt were γ-terpinene, carvacrol and α-thujene. The MH reached its highest concentrations, followed by OM in essential oil compared with the minor chemical class (SH).

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