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Headspacing GC-MS analysis of the components of sage (*Salvia officinalis* L.) collected from Isfahan Province, Iran

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Abstract

Sage is common named (*Salvia officinalis* L.) belongs to Lamiaceae family. Use of the essential oils of *Salvia* genus in medical, culinary, food and cosmopolitic products and their biological activities depends on the chemical constitutes. Sage was used in ancient Egyptian, Greek and Roman medicines. Ancient Egyptians used it as a fertility drug. The Greeks used it to stop bleeding of wounds and to clean ulcers and sores, towards hoarseness and cough, enhancing memory functions, for gargles to treat sore mouths and throats. Sage is well known for carminative, antispasmodic, antiseptic, astringent and antihidrotic properties.The aerial parts of (*Salvia officinalis* L.) were collected from the center of Iran, Province Isfahan in 2012.The essential oil was extracted by a Clevenger approach and analyzed using GC/MS. The thirty-one compounds were identified in Sage oil. The major components were camphor (17.75%), α-thujone (13.25%), α-pinene (6%), β–thujone (5.85%) and α-humulene (5.48%). Increasing amount of camphor and α-thujone has high genetic gain through selection the best among Isfahan population.

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Introduction

The genus Salviathe largest genus (Lamiaceae: subfamily Nepetoideae, tribe Mentheae) represents a cosmopolitan assemblage of nearly 1000 species worldwide, there are 58 salvia species growing naturally in Iran, 17 species endemic (Mozaffarian, 2008).The plant is mostly diffuse in the Mediterranean Basin, in South East Africa and in Central and South America, where it is largely cultivated for culinary and medicinal purposes (Kintzios, 2000). Sage was used in ancient Egyptian, Greek and Roman medicines. Ancient Egyptians used it as a fertility drug. The Greeks used it to stop bleeding of wounds and to clean ulcers and sores, towards hoarseness and cough, enhancing memory functions, for gargles to treat sore mouths and throats. Sage is well known for carminative, antispasmodic, antiseptic, astringent and antihidrotic properties(Anonymus, 1994).ISO standard9909:1997 for the essential oil composition of common sageprescribes the following: α-thujone, 18.0-43.0%; β-thujone,3.0-8.5%; camphor, 4.5-24.5%; 1,8-cineole, 5.5-13.0%;α-humulene, 0-12%; α-pinene, 1.0-6.5%; camphene, 1.5-7.0%; limonene, 0.5-3.0%; linalool and its esters, <1%; andbornyl acetate, <2.5%. The German Drug Codex requirements differ from the above ISO and are the following: thujones (≥20.0%), camphor (14.0–37.0%), 1,8-cineole (6.0–16.0%), borneol (≤5.0%) and bornyl acetate (≤5.0%) (Teusher, 2006).It is well known that yield and yield components of plants are determined by a series of factors including plant genetic (Shafie *et al*., 2009), climate, edaphic, elevation, and topography (Rahimmalek and Goli, 2013) and also an interaction of various factors (Basu *et al.,* 2009). The aim of the present work is to headspacing GC-MS analysis of the components of Sage (*Salvia officinalis* L.) collected from Isfahan province, Iran

Materials and methods

Plant material

The aerial parts of (*Salvia officinalis* L.) were collected from the center of Iran, Province Isfahan (33°, 45_ N and 51°, 16_ E, 1998 m above sea level) in 2012.The climate of regions in Isfahan is arid and warm area (according to the Koppen climate classification) characterized by warm and dry summers.

Essential oil extraction

The fresh aerial parts of S. officinalis was dried inside for six days at room temperature (25 ± 5 °C), and the ground to fine a powder using Moulinex food processor. The essential oil was extracted from 100 g of ground tissue in 1 L of water contained in a 2 L flask and heated by heating jacket at 100 °C for 2 h in a Clevenger–type apparatus, according to producers outlined British Pharmacopoeia. The collected essential oil was dried over anhydrous sodium sulphate and stored at 4 °C until analyzed.

Identification of the oil components

Compositions of the essential oils were determined by GC–MS. The GC/MS analysis was carried out with an Agilent 5975 GC-MSD system. HP-5MS column (30 m x 0.25 mm, 0.25 μm film thickness) was used with helium as carrier gas with flow rate of 1.0 mL/min. The oven temperature was kept 20 °C at 50 °C for 4 min and programmed to 280 °C at a rate of 5 °C /min, and kept 20 °C constant at 280 °C for 5 min, at split mode. The injector temperature was at 20°C at 280 °C. Transfer 20 line temperatures 280 °C. MS were taken at 70 eV. Mass range was from *m/z* 35 to 450.Identification of the essential oil components was accomplished based on comparison of retention times with those of authentic standards and by comparison of their mass spectral fragmentation patterns (Adams, 2007).

Results and discussions

GC-MS analysis

Qualitative and quantitative analysis of the essential oils volatile profile are listed in Table 1.The thirty-one compounds were identified in sage oil. The major components werecamphor (17.75%), α -thujone(13.25%), α-pinene (6%), β –thujone (5.85%) and α-humulene (5.48%). Among the monoterpenes, the camphor content was highest in (*Salvia officinalis*

L.) (Table 1). The results of the bioactive components investigation in the essential oil from sage (*Salvia officinalis L.*) originating from Jordan show that this oil contains all the components that determine the chromatographic picture of the plant in accordance with the International standards (α-pinene, camphene, limonene, 1,8-cineole, α and β-thujone, camphor, lonalool, linalyl acetate, bornyl acetate and humulene.

Table 1. Chemical composition of essential oil in Sage (*Salvia officinalis* L.).

	Compound[a]	RT[b]	RI[c]	Content %
	Monoterpenes			
1.	Cis-Salvene	4.314	852	2.68
2.	Trans-Salvene	4.600	859	1.04
3.	Tricyclene	5.857	920	0.55
4.	α-Pinene	6.265	938	6.60
5.	β- Pinene	7.353	980	2.46
6.	Myrcene	7.821	991	1.49
7.	1,8-Cineole	9.143	1031	13.03
8.	γ-Terpinene	9.755	1046	3.25
9.	α- Terpinolene	10.574	1081	1.12
10.	α -Thujone	11.359	1112	13.25
11.	β-Thujone	11.987	1114	5.85
12.	3-Cyclopentene-1-acetaldehyde, 3-trimethyl	12.044	1118	0.86
13.	Camphor	12.733	1143	17.75
14.	borneol	13.114	1165	3.72
15.	Terpinene-4-ol	13.336	1177	1.14
16.	α-Terpineol	13.630	1189	0.62
17.	Borneol, acetate	16.171	1285	3.55
18.	Myrtenyl acetate	16.301	1235	0.91
19.	Carvacrol	16.622	1298	1.51
20.	Carvacryl acetate	17.888	1348	1.27
21.	2,4-Cycloheptadien-1-one imethyl-	18.334	1358	0.44
22.	Terpinolene	18.430	1379	0.38
	Sesquiterpenes			
23.	β-Caryophyllene	19.674	1418	5.07
24.	Eremophilene	20.073	1426	1.66
25.	α-Humulene	20.541	1454	5.48
26.	Aromadendrene	20.636	1464	0.64
27.	α-Guaiene	21.265	1490	0.40
28.	(+)-Leden -α	21.451	1524	1.48
29.	4(1H)-Azulenone, octahydro-1-methy	23.450	1557	0.35
30.	Viridiflorol	23.727	1590	0.98
31.	β-Selinene	33.191	1608	0.41

[a] Compounds listed in order of elution

[b] Rt (retention time)

c RI (retention index).

In the essential oil 29 components were detected, 28 of them were identified and a dominant share had α-thujone (29.9%), β-thujone (13.68%), camphor (15.74%) and 1,8-cineole (12.31%) (Amr & Dordevic, 2000). Mirza and Baher Nik (2007) reported the major constituents of the essential oil (*Salvia lachnocalyx* Hedge) were bicyclogermacrene (31.3%), α-pinene (13.2%), sabinene (11.7%) and β-pinene (10.3%). Dzumayer (1995) reported the major constituents of the essential oil (*Salvia schimperi* Benth.) were linalool (22 -32%) and linalool acetate (25 – 51%). Poor Heravi *et al.,* (2009) reported the major constituents of the essential oil (*Salvia officinalis L.*) whit HS- SPME – GC/MS method were Linalool (5.31 – 7.44%), Butyl benzoate (7.82 – 5.42%), n-Hexyl benzoate (29.17 – 40.21%) and Benzyl benzoate (42.92 – 24.14 %). It is known that genetic constitution and environmental conditions influence the yield and composition of volatile oil produced by thyme plants. Correlations between chemotype polymorphism, sexual polymorphism and the environment have been detected (Gouyon *et al.*, 1986).

Conclusion

In conclusion, the results obtained in our study indicated that the major components of oil of sage collected from Isfahan were camphor, α -thujone, α-pinene, β –thujone and α-humulene. The environmental and genetic factors had significant effects on quality and quantity of yield as well as essential oil composition of sage. Breeding the

cultivars having the higher amounts of camphor, α – thujone,...via selection the best individual among the Isfahan population is possible so leading to high genetic efficiency.

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