Analysis of Sage Oils by GC-MS Data Bank—*Salvia officinalis L. and Salvia lavendulaefolia Vahl*

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Eastern Europe (Dalmatia, Albania, Serbia, Montenegro) is one of the main producers of *Salvia officinalis L.* These oils are yellowish or greenish-yellow possessing a warm camphoraceous, thujone-like odours and a high antimicrobial activity. They are used in the flavour industry and in perfumery. For this reason, other varieties from various countries have also been commercialized.

The chemical composition of the sage oils from various geographic origins have been studied since 1970 by a combination of chromatographic and spectroscopic techniques.[1-8] They are characterized by high α-thujone (28-40%), camphor (12-35%), and 1,8-cineole + cis-β-ocimene (9.3-13.2%) contents.[1,3-5]

The high content in 1,8-cineole (23-54%) is a characteristic of *Salvia lavandulaefolia* Vahl oils (or Spanish sage oils) which are colourless to slightly yellow and possess a camphoraceous and cineole-like odours.[2,9]

Salvia triloba L. also known as Greek sage is a third varietal form of sage oil, characterized by high contents of 1,8-cineole + limonene (30-64%) and camphor (7-17%).[5,10,11] These plants are found spontaneously growing in Greece (Crete), Sicily, Turkey, Libya and Algeria. Their oils are colourless liquids having a harsh, rosemary, spike and camphoraceous odour.

The goals of this paper are to test our SPECMA Data Bank (mass spectra and Kovats Indicies) and to detect qualitative and quantitative differences between *Salvia officinalis* L. (from Dalmatia and Albania) and *Salvia lavandulaefolia* Vahl (from Spain).

Materials and Methods

Sage oils from Albania, Dalmatia and Spain were commercially available.

Each sample was analyzed using an Intersmat IGC-15 gas chromatograph equipped with a flame ionization detector (GC-FID). A fused silica column (50 m x 0.28 mm i.d.) coated with Carbowax 20 M was used. The column oven temperature was programmed from 70°C to 220°C at a rate of 2°C/min. The injection port and detector were kept at 260°C. Helium was used as the carrier gas at a flow rate of 1 ml/mn with a split ratio of 1/20. Typically 0.04 μl (S 2.10^{-11}) of sample was injected using a conventional splitless inlet. Quantitative assessments (peak areas) were calculated from GC-FID trace using a computing integrator ICR 1B.

Qualitative analyses were recorded with a VG 70 70F mass spectrometer (MS) connected with a Perkin Elmer Sigma 3 gas chromatograph and linked on-line to the EPA/NIH data bank.

Samples were injected on a fused silica column (50 m x 0.28 mm i.d.) coated with FFAP using 1:40 split ratio. The column oven temperature was kept 8 min at 60°C and then programmed from 60 to 220°C at a rate of 2°C/min.

Significant operating parameters of the MS

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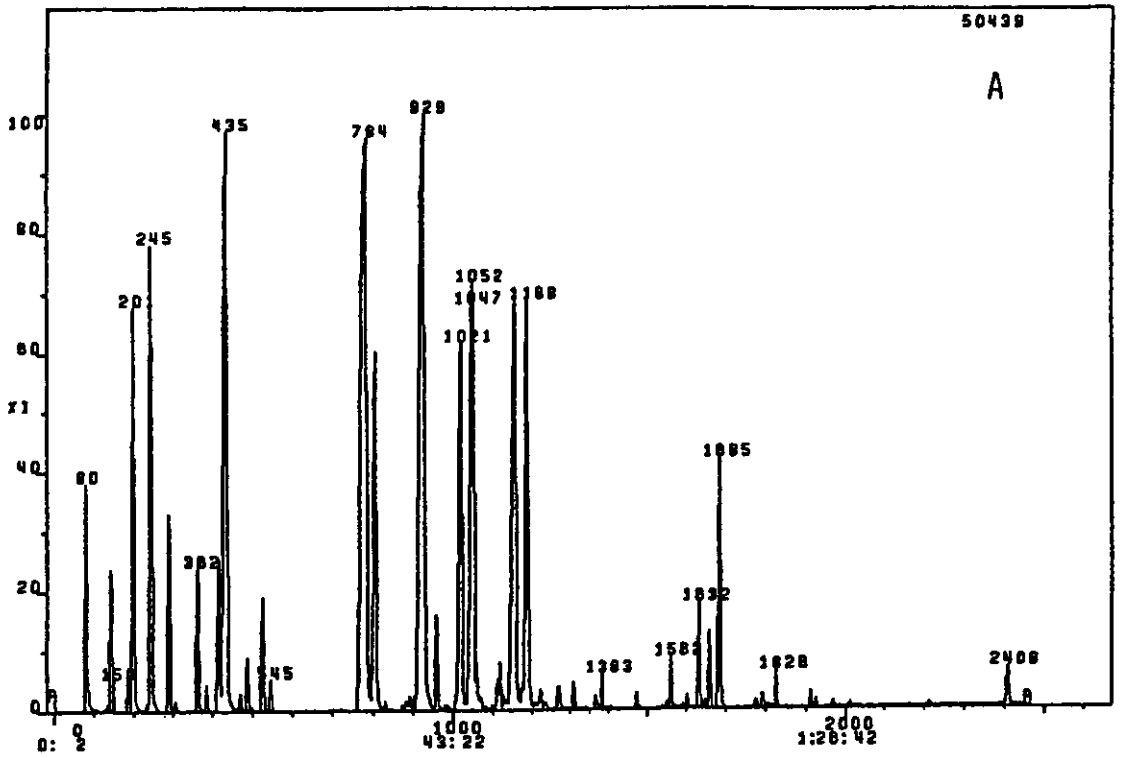
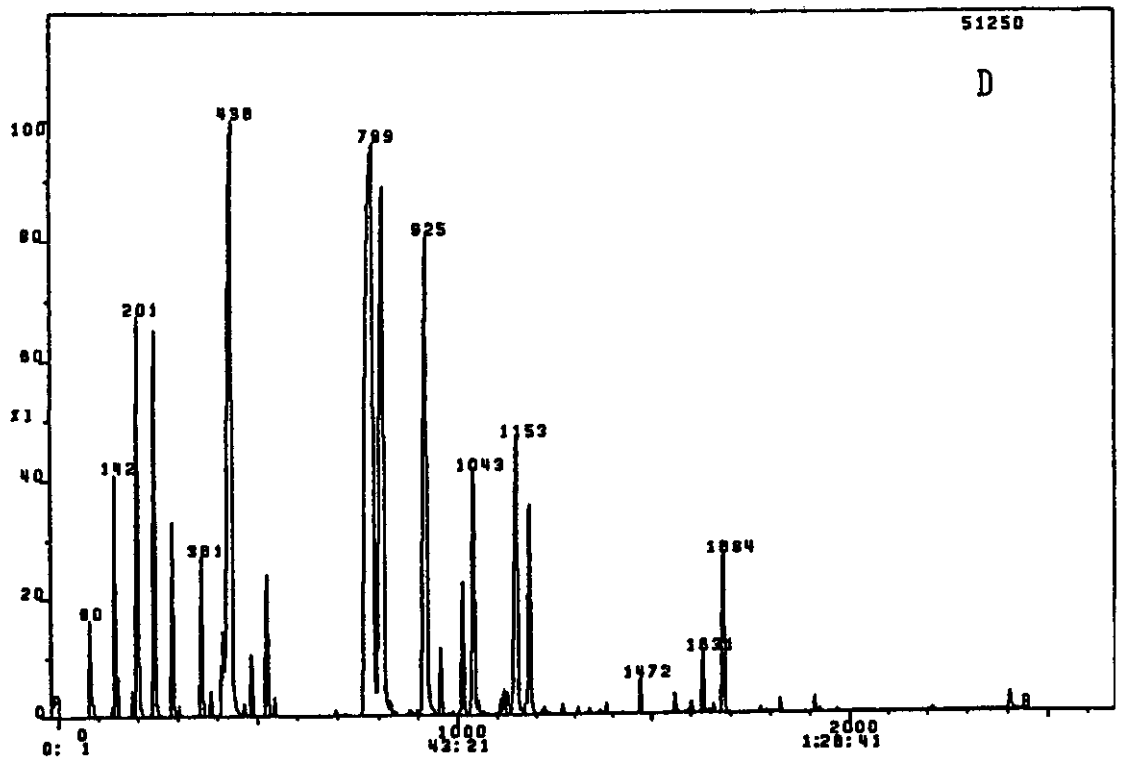
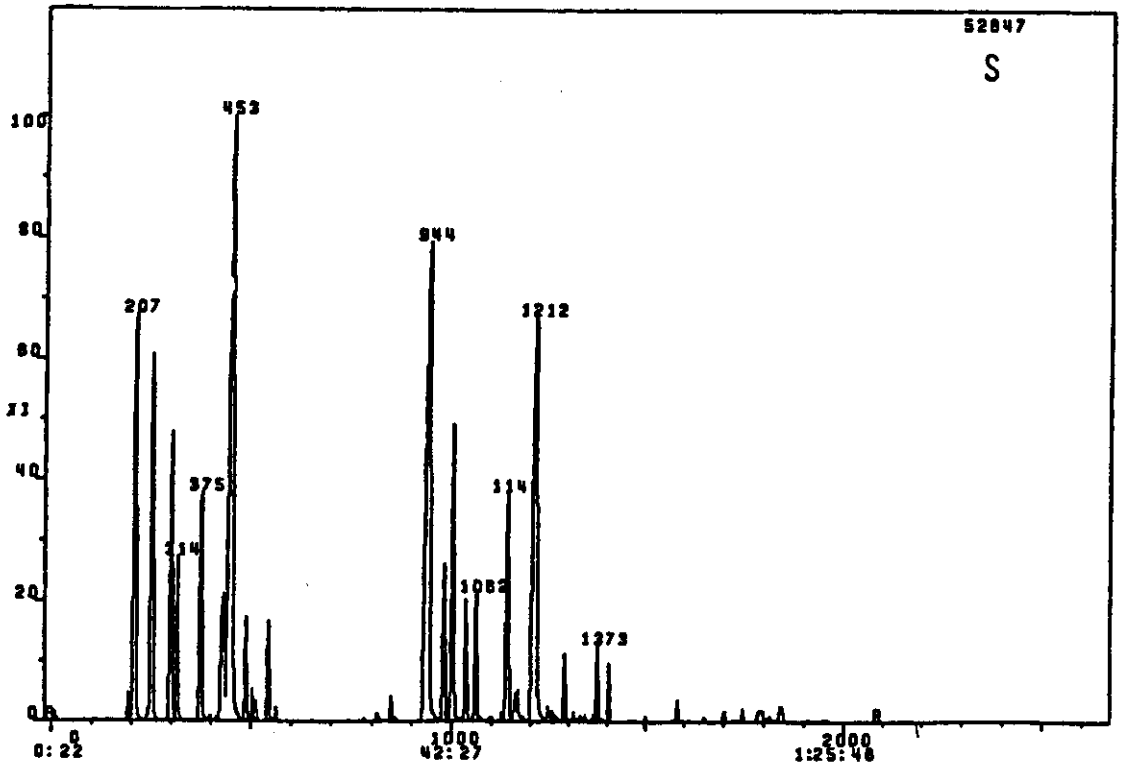
Figure 1. Total Ion Chromatograms (TIC) for Sage Oils: Albania (A), Dalmatia (D), Spain (S)





(Figure 1. continued)



were: ionization voltage: 70 eV, ionization current: 100 μA and source temperature 220°C. Data acquisition and reprocessing were performed by a VG 2035 system.

Results and Discussion

Figure 1 shows the reconstructed chromatograms of Albanian, Dalmatian and Spanish sage oils.

The chromatograms were similar for both Albanian and Dalmatian oils, although quantitative differences appear in some peaks. Spanish oil is quite different both quantitatively and qualitatively.

One may note here that the chromatograms of sage oil reported by Masada[12] as a Spanish oil is actually a *Salvia officinalis* L. oil, identified by its high content of α-thujone and camphor.

Compounds identified in the present investigation are listed in Table I. New identified compounds are indicated by an asterisk. The scan numbers, the Kovats Indices on the FFAP column calculated as described previously,[13] the corresponding literature data,[14] the molecular weight and the main fragments in EI mode and classified in decreasing order of intensity, and their occurrence in studied sage oils are also given. Among seventy-two separated compounds sixty were identified. Besides the products reported by Lawrence et al.[1] we found twelve new compounds in *Salvia officinalis* L: β-caryophyllene, isoborneol, α-terpineol, neryl acetate, neral, geranyl isobutyrate, geranyl propionate, a α-humulene iosmer, eugenol, thymol, carvacrol and the humulene epoxide II. On the other hand, cis-alloocimene, α,p-dimethylstyrene, α-cubebene, α-gurjunene, cis and trans-α-bergamotene were not found.

The composition of the monoterpenic hydrocarbons in the three examined sage oils are given in Table II. The two main notable features are the higher content in sabinene and cis-β-ocimene in the Spanish sage oil when compared to the two others which are less rich in hydrocarbons.

In Table III a comparative study of the chemical composition of the three various sage oils studied is given. The chemical composition of the Dalmatian oil is very similar to that reported by Ivanic et al. for various regions of Yugoslavia, except for the camphor content which was confused with β-thujone.[4] The thujone content in these oils (51%) is higher than that observed in the Albanian oil (27%). With regard to the Spanish oil, it is characterized by a high content of monoterpenic compounds (27.6%), 1,8-cineole (21%), camphor (22.3%), linalyl acetate (3.9%), borneol (13%), probably an aromatic ester (3.7%) and geranyl esters (1%).

Table I. GC-MS Qualitative Analysis of Albanian (A), Dalmatian (D) and Spanish (S) Sage Oils

Peak[a]	Scan[b]	KIP[c]	Lit.[d]	m/z(EI)[e]	Compounds (identification)[f]	Occurrence
1	80	600		57, 43, 41, 56	Hexane	A, D
2	84	610		56, 41,67	Methyl cyclopentane ?	A, D
3	142	990		81, 68, 41, 67, 109	(Z)-2-methyl-3-methylene hept-5-en ?	A, D
4	150	996		41, 81, 67,109	Isomer of peak 3	A, D
5	187	1021	1036	93, 121, 136	α-Thujene	A, D, S
6	200	1030	1039	93, 92, 91, 77	α-Pinene	A, D, S
7	240	1056	1083	93, 121, 79, 107	Camphene	A, D, S
8	290	1089	1124	93, 69, 41	Beta-Pinene	A, D, S
9	304	1099	1130	93, 77, 91	Sabinene	A, D, S
10	360	1136	1156	41, 93,69	Myrcene	A, D, S
11	385	1152	-	121, 93, 136	α-Terpinene	A, D, S
12	400	1154	-	109, 43, 79, 124	Unidentified	S
13	416	1173	1206	68, 93, 67	Limonene	A, D, S
14	430	1182	1228	43, 81, 71	1,8-Cineole	A, D, S
15	470	1209	1228	93, 77, 79, 91	(Z)-Beta-Ocimene	A, D, S
16	485	1219	1251	93, 77, 91, 136, 121	γ-Terpinene	A, D, S
17	500	1229	1250	93, 77, 91	(E)-Beta-Ocimene	S
18	523	1244	1272	119,134,91	ρ-Cymene	A, D, S
19	545	1259	1287	121, 93, 136	Terpinolene	A, D, S
20	770	1408	-	81, 110, 68, 41	α-Thujone	A, D, S
21	806	1432	-	110, 81, 68, 95	Beta-Thujone	A, D
22	810	1435	1420	57	1-Octen-3-ol	A
23	830	1448	-	43, 71, 81, 93	(E)-Sabinene hydrate ?	D, S
24	886	1486	1480	82, 67, 57	(Z)-3-Hexenyl isovalerate	A
25	893	1490	1520	119, 105, 161	α-Copaene	A
26	903	1497	-	85, 57, 41, 55, 82	(E)-3-Hexenyl isovalerate ?	A, D, S
27	918	1507	1518	95, 81, 41, 108, 69	Camphor	A, D, S
28	960	1535	1506	71, 93, 41, 55, 43, 69	Linalool	A, D, S
29	980	1548	1538	93, 43, 80, 69	Linalyl acetate	S
30	988	1553		93, 79, 107	Unidentified	D
31	1010	1568	-	93, 69, 41, 133	Isocaryophyllene	A
32	1016	1572	1585*	95, 43, 93, 121, 136	Bornyl acetate	A, D, S
33	1040	1588	1628	71, 111	Terpinen-4-ol(+ Beta-caryophyllene)	A, S
34	1048	1593	1616	69, 93, 41, 133	Beta-Caryophyllene	D
35	1060	1601	-	41, 61, 93	Aromadendrene	A
36	1113	1637	-	41, 91, 93, 161	Allo-aromadendrene	A
37	1120	1641	1651	91, 43, 92	Sabinyl acetate ?	A, D, S
38	1128	1647	1683	93, 43, 41, 55, 95	?	D
39	1138	1653	1660	95, 41, 60, 43	Isoborneol	A
40	1140	1655	-	59, 81, 43, 93	δ-Terpineol	S
41	1150	1661	1682	93, 80, 121	(Z)-α-Humulene	D, S
42	1160	1669	-	93, 80, 121	(E)-α-Humulene	A
43	1175	1678	1685*	161, 93, 105	γ-Muurolene*	A, D
44	1180	1681	1661	59, 93, 121, 136	α-Terpineol	D
45	1185	1684	-	95, 59	Borneol (+α-terpineol)	A, S
46	1187	1686	1698	95	Borneol	D
47	1200	1695	1687	121, 93, 136, 43	α-Terpinyl acetate	S
48	1203	1696	1698	91, 43, 92, 93, 121	Linalyl isovalerate*	S
49	1225	1711	1730	41, 43, 69, 93, 105	δ-Selinene + α-muurolene ?	A
50	1228	1715	1699	69, 41, 43, 68, 93	Neryl acetate	S

(Continued)

(Table I. continued)

Peak[a]	Scan[b]	KIP[c]	Lit.[d]	m/z(EI)[e]	Compounds (identification)[f]	Occurrence
51	1232	1716	1730	69, 41	Geranial	S
52	1240	1721	1715	93, 121, 41, 82	Germacrene B + carvone ?	S
53	1268	1740	1735	69, 41, 43, 68, 93	Geranyl acetate*	S
54	1278	1746	1766	161, 134, 119, 204	δ-Cadinene	D
55	1290	1754	-	119, 132, 105, 41	α-Curcumene	S
56	1316	1772	-	79, 81, 108	Mixture ?	A, S
57	1320	1774	1757	69, 41	Nerol	S
58	1340	1788	1795	69, 43, 41	Geranyl isobutyrate*	S
59	1353	1796	1799	69, 57, 41	Geranyl propionate	S
60	1360	1801	1790	109, 84, 41, 55	Carveol I*	A
61	1380	1814	-	43, 135	ρ-Cymen-8-ol	A, D
62	1385	1817	1797	69, 41	Geraniol	S
63	1473	1876		43, 97, 85, 55	A Ketone	A, D
64	1560	1934		41, 43, 79, 93, 69	Caryophyllene oxide	A, D, S
65	1602	1962		93, 80, 121, 41, 43	Humulene derivative*	A, D
66	1630	1980		43, 109, 41, 67, 138	Humulene epoxide II*	A, D
67	1660	2000		153, 43, 168	Unidentified	A, D
68	1684	2016		43, 109, 69, 85, 55, 161	Humulenol or caryophyllenol ?	A, D
69	1775	2077	2103	164, 149	Eugenol*	A, D, S
70	1793	2089	2100	135, 150	Thymol*	A, D, S
71	1825	2110	2160	135, 150	Carvacrol	A, D, S
72	2408			137, 81, 97, 69 41, 55	Unidentified	A, D

a) In elution order on FFAP column.
b) Scan numbers as recorded on the VG 70-70F spectrometer.
c) Kovats Indices on polar column calculated as described previously (13).
d) Kovats Indices as reported by Jennings and Shibamoto (14).
e) Main fragments in decreasing intensity order.
f) Identification was achieved using either the SPECMA data bank or our own manual files. When identification was not confirmed by visual comparison with the mass spectrum references, this is indicated by a question mark.

Table II. Chemical Composition of the Monoterpenic Hydrocarbons of Sage Oils from Albania, Dalmatia and Spain.

Compounds	% (relative to the fraction) Albania	Dalmatia	Spain
α-Thujene	1.41	1.10	0.80
α-Pinene	28.30	36.80	30.60
Camphene	40.80	29.50	20.30
Beta-Pinene	9.00	9.00	14.00
Sabinene	0.80	0.90	5.40
Myrcene	4.54	6.70	10.00
α-Terpinene	0.70	1.30	trace
Limonene	12.20	11.60	13.10
cis-Beta-Ocimene	0.30	0.20	3.60
γ-Terpinene	0.90	1.70	1.00
trans-Beta-Ocimene	0.15	0.10	0.70
Terpinolene	0.90	1.10	0.50

Table III. Comparative Chemical Composition of Albanian, Dalmatian and Spanish Sage Oils

Compounds	% Albanian	Dalmatian	Spanish
Monoterpenic compounds	12.00	10.82	27.60
ρ-Cymene	0.80	1.12	0.97
1,8-Cineole	8.20	12.00	21.10
α-Thujone	23.40	37.15	1.30
Beta-Thujone	3.45	14.20	trace
Camphor	22.45	12.30	22.30
Linalool	0.51	0.40	1.70
Linalyl acetate	trace	trace	3.90
Bornyl acetate	3.50	0.86	0.97
Beta-Caryophyllene(+ terpinen-4-ol)	6.50	2.20	0.80
Aromatic ester	-	-	3.70
α-Humulene	6.90	3.87	0.24
Borneol (+ α-terpineol + terpinyl acetate)	5.30	1.87	13.10
Geranyl acetate	-	-	0.33
Geranyl propionate	-	-	0.42
ρ-Cymen-8-ol	0.13	0.05	0.10
Geraniol	-	-	0.37
Humulene oxides	0.80	0.48	0.20
$C_{15}H_{26}O$	1.46	1.08	-
Miscellaneous	4.60	1.60	1.90

Note: These analyses were carried out on an Intersmat IGC 120 FL apparatus (Carbowax 20 M, 50 m x 0.2 i.d. column) coupled with an Intersmat ICR 1B computer integrator.

Conclusion

This study allowed us to test our data bank and to demonstrate its usefulness in the analysis of various aroma and essential oils. Not only the well-known fundamental differences in the composition of sage essential oils related to their origin have been observed but, in addition, twelve new minor compounds have been identified. They may play an important olfactive influence. However, it would be necessary to obtain samples from a very wide survey of large numbers of Salvia populations and to treat them by statistical methods in order to demonstrate this hypothesis.

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