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Chemical Composition of Essential Oils from some *Salvia* species

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The essential oils from *Salvia aurea* L., *Salvia ianthina* Otto et A. Dietr., *Salvia iodantha* Fernald and *Salvia cinnabarina* M. Martens et Galeotti were analysed by means of GC/MS. A total of 89 components were identified and the variation of composition during the year of the first three species was studied. The antimicrobial activity of the essential oil from *Salvia aurea* L. was also tested. © **1998 John Wiley & Sons, Ltd.**

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INTRODUCTION

Several *Salvia* species are widely used in folk medicine (Penso, 1983) and some species are listed in the modern Pharmacopoeias (Zepernick *et al.*, 1984). In folk medicine *S. aurea* is used in Africa as a decoction or infusion for the treatment of coughs, colds and female ailments (Watt and Breyer-Brandwijk, 1962). In a previous work a preliminary investigation of the essential oil from *Salvia aurea* L. was reported (Serrato *et al.*, 1997).

We now report the composition of the essential oils from *Salvia aurea* L., *Salvia iodantha* Fernald, *Salvia ianthina* Otto et A. Dietr. and *Salvia cinnabarina* M. Martens et Galeotti and, for the first three species, qualitative and quantitative variations throughout some periods of the year. The antimicrobial activity of the essential oil from *Salvia aurea* L. was also tested.

MATERIALS AND METHODS

Plant materials. Each of the *Salvia* species was obtained from the collection of *Salvia* species established and available in the 'Hanbury Botanical Gardens' of La Mortola (Ventimiglia, Italy) and identified by Dr P.G. Campodonico. Voucher specimens were deposited in the herbarium there.

Extraction of the essential oils. Fresh aerial parts of non-flowering plants were subjected to steam distillation under reduced pressure to produce oils in the following yields [% w/w (*year/month of collection*)]: *S. aurea* [0.25% (*96/03*), 0.39% (*96/07*), 0.20% (*96/11*)], *S. iodantha* [0.02% (*96/07*), 0.007% (*96/10*)], *S. ianthina* [0.005% (*96/07*), 0.006% (*96/10*)], *S. cinnabarina* [0.052% (*96/11*)].

Analysis of the essential oils. Essential oils were analysed using a Hewlett-Packard 5890 Series II GC-MS apparatus equipped with a capillary column HP-Innowax (crosslinked polyethylene glycol, 30 m long, 0.25 mm internal diameter, 0.25 μm film thickness). The carrier gas was helium with a flow rate of 1 mL/min and the injection temperature was 250 °C. The column temperature programme was 60 °C for 8 min^{-1}, followed by an increase of 3 °C per min to 180 °C, then this temperature for 5 min, followed by an increase of 40 °C per min to 250 °C. Identification of the components was achieved by comparison of their mass spectra with WILEY-NBS Library (Wiley275) and of retention indices with authentic samples. Retention indices were experimentally determined in the same conditions of analyses, by use of standard mixture of n-alkanes (Sigma Chem. Comp.). They were calculated by linear interpolation between retention times of compound and alkanes, notwithstanding the isothermal periods of analysis, and are quite acceptable only for internal comparisons within the actual set of chromatograms (Sun *et al.*, 1993). These values are reported in Table la to three significant figures.

The quantitation of each component was performed by integration of total ion chromatograms (autointegration method).

Antimicrobial assay. The pure (10 μL, 8.4 μg) and diluted oil [dimethylsulphoxide (DMSO) solution] were tested. Standard laboratory strains of bacteria were selected as test organisms for the study. These included both Gram-positive (*Bacillus cereus var. micoides* 213

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Table 1a. Components[a] of the essential oil of *Salvia spp.*

Peak N.[b]	Compound[c]	RI[d]	*S. aurea*			*S. iodantha*		*S. ianthina*		*S. cinnabarina*
			96/03	*96/07*	*96/11*	*96/07*	*96/10*	*96/07*	*96/10*	*96/11*
1	α-Pinene	1.02	7.2	5.6	6.9	0.5	0.2	6.1	0.3	0.3
2	Camphene	1.07	8.2	7.0	8.0	0.2	–	5.6	–	0.9
3	β-Pinene	1.11	1.4	1.0	1.3	0.8	0.2	–	–	0.4
4	Sabinene	1.12	–	–	–	0.2	–	–	–	–
5	3-Carene	1.15	15.0	13.2	15.0	–	–	3.3	–	–
6	α-Phellandrene + Myrcene	1.16	1.5	1.3	1.7	–	–	–	–	–
7	α-Terpinene	1.18	0.3	0.3	0.3	–	–	–	–	–
8	Sylvestrene	1.20	–	2.7	–	–	–	–	–	–
9	Limonene + Sylvestrene	1.20	5.6	2.4	5.3	–	–	–	–	–
10	Limonene	1.20	–	–	–	0.1	–	–	–	–
11	β-Phellandrene	1.21	2.4	1.8	2.3	–	–	–	–	–
12	1,8-Cineole	1.21	–	–	–	–	0.4	–	–	–
13	(Z)-β-Ocimene	1.24	–	–	–	0.2	–	–	–	–
14	Unidentified (MI 136)	1.24	0.2	0.2	0.2	–	–	–	–	–
15	γ-Terpinene	1.24	0.4	0.4	0.3	–	–	–	–	–
16	(E)-β-Ocimene	1.25	–	–	–	3.3	0.8	–	2.9	–
17	*p*-Cymene	1.27	1.1	1.2	1.0	–	–	–	–	–
18	α-Terpinolene	1.28	0.5	0.4	0.5	–	–	–	–	–
19	Unidentified (MI 136)	1.28	0.3	0.3	0.3	–	–	–	–	–
20	(Z)-3-Hexen-1-yl acetate	1.32	–	–	–	0.2	–	–	1.2	1.0
21	3-Octyl acetate	1.34	–	–	–	–	–	–	–	0.8
22	1-Octen-3-yl acetate	1.38	–	–	–	–	–	–	–	2.6
23	(*Z*)-3-Hexenol	1.39	–	–	–	–	0.3	–	1.7	0.7
24	3-Octanol	1.40	–	–	–	0.1	1.0	–	–	2.2
25	Unidentified (MI 204)	1.45	–	–	–	0.3	0.4	–	–	–
26	1-Octen-3-ol	1.46	–	–	–	0.3	8.2	–	0.8	1.9
27	(*Z*)-3-Hexenyl butanoate	1.46	–	–	–	–	–	–	0.9	–
28	δ-Elemene	1.46	–	–	–	0.3	–	–	–	–
29	*trans*-Sabinene hydrate	1.47	0.3	–	0.3	–	–	–	–	0.8
30	Cyclosativene	1.47	–	–	–	–	–	–	–	0.7
31	Bicycloelemene, stereoisomer	1.48	–	–	–	3.8	3.9	–	–	1.7
32	α-Copaene	1.48	–	0.4	–	0.2	0.3	2.2	1.9	2.9
33	Camphor	1.50	29.1	28.4	30.6	0.4	0.2	1.0	–	0.4
34	β-Bourbonene	1.51	–	–	–	0.3	0.4	1.8	2.5	0.3
35	α-Gurjunene	1.52	0.2	0.2	0.1	0.2	0.1	–	–	–
36	Unidentified (MI 204)	1.53	–	–	–	–	–	–	0.4	–
37	Unidentified (MI 204)	1.53	0.1	0.1	0.1	–	–	–	–	–
38	α-Zingiberene	1.53	1.4	1.3	1.3	–	–	–	–	–
39	*cis*-Sabinene hydrate	1.55	0.3	0.1	0.3	–	–	–	–	0.5
40	Linalool	1.56	–	–	–	0.8	1.5	–	0.6	32.8
41	2-Isopropyl-5-methyl-9-methylene[4.4.0]dec-1-ene (*tentative*)	1.56	–	–	–	–	–	2.0	2.2	–
42	Unidentified (MI 204)	1.56	–	–	–	0.4	0.3	–	–	–
43	α-Bergamotene, stereoisomer	1.56	1.3	1.5	1.3	–	–	–	–	–
44	Methyl β-cyclogeranate	1.57	–	–	–	–	–	–	–	0.6
45	Elemene, isomer	1.57	–	–	–	–	–	1.8	1.7	–
46	Bornyl acetate	1.58	–	–	–	2.9	3.4	–	–	0.6
47	β-Cubebene	1.58	–	–	–	–	–	1.2	1.2	–
48	α-Bergamotene, stereoisomer	1.58	0.9	1.0	0.8	–	–	–	–	–
49	β-Elemene	1.58	–	–	–	–	–	30.1	27.0	–
50	*trans*-Caryophyllene	1.59	1.7	3.9	1.6	43.7	35.0	–	–	1.3
51	Aromadendrene, stereoisomer	1.59	0.2	0.3	0.2	–	–	–	–	0.4
52	4-Terpineol	1.60	1.3	1.4	1.3	–	0.3	–	–	0.9
53	β-Cyclocitral	1.61	–	–	–	–	–	–	–	1.0
54	Unidentified (MI 204)	1.63	–	–	–	–	–	0.5	0.7	–
55	Alloaromadendrene	1.63	–	–	–	0.3	0.9	–	–	–
56	Benzeneacetaldehyde	1.64	–	–	–	0.3	–	–	–	–
57	Unidentified (MI 204)	1.65	–	–	–	–	–	0.4	0.6	–
58	α-Humulene	1.66	0.9	2.2	0.8	8.5	6.9	0.7	1.4	0.4
59	Unknown	1.67	–	–	–	0.2	–	–	–	–
60	β-Farnesene, E or Z isomer	1.67	1.4	1.7	1.4	–	–	–	–	–
61	β-Farnesene, Z or E isomer	1.68	0.2	0.2	0.2	–	–	–	–	–
62	Salicylaldehyde	1.68	–	–	–	–	0.3	–	–	–
63	Unidentified (MI 204)	1.68	0.1	–	–	0.2	–	–	–	–
64	γ-Curcumene	1.69	1.1	1.2	1.0	–	–	–	–	–
65	Germacrene-D	1.70	–	–	–	3.0	2.2	36.1	36.1	0.9
66	α-Terpineol	1.70	–	–	–	–	–	–	–	2.7

Table 1a. Continued

Peak N.[b]	Compound[c]	RI[d]	*S. aurea*			*S. iodantha*		*S. ianthina*		*S. cinnabarina*
			96/03	*96/07*	*96/11*	*96/07*	*96/10*	*96/07*	*96/10*	*96/11*
67	Borneol	1.70	0.6	0.5	0.6	0.8	1.1	–	–	5.5
68	α-Muurolene	1.72	–	0.2	–	–	0.1	–	1.8	–
69	β-Bisabolene	1.72	0.3	0.4	0.3	–	–	–	–	–
70	Bicyclogermacrene	1.72	–	–	–	8.1	8.5	1.1	–	4.9
71	Unidentified (MI 204)	1.73	–	–	–	0.4	–	–	–	–
72	α-Cedrene	1.74	5.6	5.6	5.3	0.4	–	–	–	–
73	δ-Cadinene	1.75	1.0	1.8	0.9	–	–	3.4	2.2	1.2
74	*E,E*-α-Farnesene	1.75	–	–	–	6.9	3.5	–	–	–
75	Methyl salicylate	1.77	–	–	–	–	1.7	–	0.4	1.1
76	α-Curcumene	1.77	5.1	7.1	4.9	0.4	–	–	–	–
77	Unidentified (MI 222)	1.81	–	–	–	0.1	0.3	–	–	–
78	Thymol acetate	1.85	–	–	0.1	–	–	–	–	–
79	Benzyl alcohol	1.88	–	–	–	0.2	2.0	–	0.4	–
80	Unknown	1.90	–	–	–	–	–	–	–	1.8
81	Phenethyl alcohol	1.92	–	–	–	–	0.3	–	–	–
82	Shyobunol	1.93	0.9	0.3	0.9	–	–	–	–	0.7
83	Carotol (*tentative*)	1.94	–	–	–	–	–	–	0.6	–
84	*Z*-Jasmone	1.94	–	–	–	0.4	0.4	–	–	1.1
85	Unidentified (MI 220)	1.96	–	–	–	0.2	0.4	–	–	–
86	Caryophyllene oxide	1.97	0.3	0.5	0.3	0.9	2.0	–	0.5	–
87	Unidentified (MI 222)	1.99	0.3	0.2	0.4	–	–	–	–	–
88	Unidentified (MI 220)	2.01	–	–	–	–	–	–	–	2.9
89	Ledol	2.03	–	–	–	–	1.2	–	–	–
90	Humulene oxide	2.03	0.2	0.3	0.2	0.1	0.3	–	–	–
91	1–*endo*-Bourbonanol	2.05	–	–	–	4.4	1.4	1.7	0.9	1.0
92	Unidentified (MI 222)	2.06	–	0.2	0.2	–	–	–	–	–
93	Globulol	2.07	–	–	–	0.6	0.2	–	–	–
94	Unidentified (MI 220)	2.08	–	–	–	–	–	–	–	0.5
95	Elemol	2.08	–	–	–	–	–	–	–	7.4
96	Guaiol	2.09	–	–	–	–	–	–	0.4	–
97	Unidentified (MI 222)	2.10	0.3	0.5	0.4	–	–	–	–	–
98	Spathulenol	2.12	–	–	–	0.4	1.6	–	–	3.4
99	Z-3-Hexenyl benzoate	2.12	–	–	–	0.2	–	–	–	1.1
100	Unknown	2.13	–	–	–	–	–	–	0.4	–
101	Unknown	2.14	–	–	–	0.2	0.3	–	–	0.4
102	Unknown	2.15	–	–	–	–	–	–	0.5	–
103	8–*epi*-β-Bisabolol (*tentative*)	2.15	0.2	0.2	0.2	–	–	–	–	–
104	γ-Eudesmol	2.17	–	–	–	–	–	–	–	0.6
105	Torreyol	2.18	–	–	–	–	–	–	–	0.7
106	Eugenol	2.18	–	–	–	–	1.3	–	–	–
107	T-Muurolol	2.19	–	–	–	0.2	0.6	–	0.7	–
108	Unknown	2.21	0.1	0.1	0.1	–	–	–	–	–
109	α-Eudesmol	2.21	–	–	–	–	–	–	–	1.3
110	β-Eudesmol	2.22	–	–	–	–	–	–	–	2.1
111	α-Cadinol	2.23	–	–	–	0.8	–	0.9	0.9	0.5
112	Unknown	2.26	–	–	–	–	–	–	–	1.1
113	Unidentified (MI 222)	–	0.3	–	0.4	–	–	–	–	–
114	Caryophylla-4(12),8(13)-dien-5β-ol	–	–	–	–	–	0.3	–	–	–

Table 1b. Cumulative composition[a] of the essential oil of *Salvia spp.*

Components	*S. aurea*			*S. iodantha*		*S. ianthina*		*S. cinnabarina*
	96/03	*96/07*	*96/11*	*96/07*	*96/10*	*96/07*	*96/10*	*96/11*
Monoterpene hydrocarbons	44.1	37.8	43.1	5.3	1.6	15.0	3.2	1.6
Oxygenated monoterpenes	31.6	30.4	33.2	4.9	6.5	1.0	0.6	45.8
Sesquiterpene hydrocarbons	21.5	29.1	20.2	77.4	62.4	81.3	79.7	14.7
Oxygenated sesquiterpenes	2.5	2.2	3.0	7.7	8.3	2.6	4.0	22.9
Others	0.1	0.1	0.1	2.1	15.8	–	6.3	14.0
Linear alkanes C_{19}-C_{23}	–	–	–	0.2	2.5	–	3.2	–

[a] % composition
[b] In order of elution
[c] The compounds, for which correct characterization of isomer was not possible, are specified by words 'isomer' and 'stereoisomer'. The term 'unidentified' was reserved to compounds with surely attributable molecular ion (MI).
[d] R.I. × 10^3; see Analysis of essential oils in Materials and methods.

Table 2

Test bacteria	Samples[a]			Reference antibiotic		
	neat	1/10	1/100	Lincomycin	Gentamycin	Amikacin
Bacillus cereus	9	–	–	22	nt[b]	nt
Bacillus subtilis	9	–	–	20	nt	nt
Sarcina subflava	9	–	–	38	nt	nt
Enterococcus faecalis	8	–	–	11	nt	nt
Staphylococcus aureus	9	–	–	23	nt	nt
Escherichia coli ATCC 25922	7	–	–	nt	22	nt
Escherichia coli ATCC 35218	–	–	–	nt	19	nt
Proteus mirabilis	–	–	–	nt	22	nt
Yersinia enterocolitica	–	–	–	nt	26	nt
Aeromonas hydrophila	8	–	–	nt	25	nt
Salmonella typhi Ty_2	–	–	–	nt	25	nt
Salmonella paratyphi	–	–	–	nt	17	nt
Pseudomonas aeruginosa	–	–	–	nt	nt	25

[a] Mean of three replicates: diameter of inhibition zone (mm)
[b] Not tested.

P.C.I., *Bacillus subtilis* ATCC 6633, *Sarcina subflava* ATCC 7468, *Enterococcus faecalis* ATCC 29212, *Staphylococcus aureus* ATCC 25923) and Gram-negative (*E. coli* ATCC 25922, *E. coli* ATCC35218, *Proteus mirabilis* ATCC 12453, *Yersinia enterocolitica* ATCC 9610, *Aeromonas hydrophila* ATCC 35654, *Salmonella typhi Ty2* ATCC19430, *Salmonella paratyphi* ATCC 12176, *Pseudomonas aeruginosa* ATCC 27853) bacteria. The *in vitro* paper-disc diffusion method (Bauer *et al.*, 1966) was used for the antibacterial assay: 25 mL of Agar Mueller-Hinton with a standardized inoculum of the test microrganism (standard 0.5 McFarland) were placed into Petri plates (10 cm diameter). Then 10 μL of diluted solutions of the essential oil was added to antibiotic sterile assay paper-discs Whatman no. 1 (0.6 cm of diameter) and placed on agar plates. Plates were examined for zones of inhibition after 24 h of incubation at 37°C. Each test was carried out in triplicate; a blank containing DMSO showed no inhibition in preliminary studies. Controls were performed with lincomycin and gentamycin for Gram-positive and Gram-negative bacteria respectively and amikacin for *Pseudomonas aeruginosa*.

RESULTS AND DISCUSSION

The composition of each oil can be seen in Tables 1a and 1b. The following observations are possible.

S. aurea: the data reported here are in substantial agreement with those of a previous paper (Serrato *et al.*, 1997), considering the following: (a) different type of GC column; (b) incomplete analysis reported there; (c) different period of collecting. However, a small amount of oxygenated sesquiterpenes in the present work was found. Variations throughout the year are not particularly important. A ratio of around 2:1 between camphor and 3-carene percentages was constantly observed.

S. iodantha, *S. ianthina* and *S. cinnabarina*: in these species, the production of essential oil is scarce and parallels the poor occurrence of monoterpene hydrocarbons. High (or much higher [*S. cinnabarina*]) content of non-terpenoid compounds, especially in the autumn, was observed. *S. iodantha* and *S. ianthina*: the composition of essential oil is strongly characterized by some specific sesquiterpene hydrocarbons (*trans*-caryophyllene in *S. iodantha*, *β*-elemene and germacrene-D in *S. ianthina*). *S. cinnabarina* is characterized by a high amount of various oxygenated monoterpenes (particularly linalool) and oxygenated sesquiterpenes.

The antimicrobial activity of the essential oil from *S. aurea*, which showed a higher yield, was also tested. The results of the antimicrobial assay carried out on 13 bacteria strains are given in Table 2. The table shows that the oil does not possess significant activity against all the tested Gram-positive and Gram-negative microorganisms.

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