

Composition of Essential Oils from *Satureja darwinii* (Benth.) Briq. and *S. multiflora* (R. et P.) Briq. (Lamiaceae). Relationship Between Chemotype and Oil Yield in *Satureja* spp.

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Abstract

Air-dried aerial parts of *Satureja darwinii* (Benth.) Briq. and *Satureja multiflora* (R. et P.) Briq. (Lamiaceae) collected in Chile at full flowering stage were hydrodistilled. The yields of essential oils were 0.68% and 0.57% v/w, respectively. GC-FID and GC/MS analyses showed that the major constituents of the oils were piperitenone (57.8%) and isomenthone (83.1%) in *S. darwinii* and *S. parvifolia*, respectively. These results are compared with others reported for the genus *Satureja*.

Key Word Index

Satureja darwinii, *Satureja multiflora*, Lamiaceae, savory, essential oil composition, piperitenone, isomenthone, chemotypes.

Introduction

The genus *Satureja* (Lamiaceae) has a wide distribution which includes Europe, Asia, tropical Africa, and the Americas (1). They are mainly aromatic herbs and shrubs, some of which are used as flavoring agents and some for medicinal purposes. The composition of numerous *Satureja* oils has been reported. Among the four *Satureja* species which occur in Chile (2), and to the best of this author's knowledge only *S. gilliesii* (3) and *S. parvifolia* (4) have been studied. In this report the composition of the essential oils of the other two species, *S. darwinii* and *S. multiflora*, is reported.

Experimental

Plant material: Samples of above-ground tissue of the two *Satureja* species (three samples of ca. 200 g fr. wt each) were collected at the full flowering stage, *S. darwinii* at the Pali Aike National Park near Punta Arenas, Chile (52°04.8'S, 69°47.1'W), and *S. multiflora* by the shores of Lago Colico, near Temuco, Chile (39°04.3'S, 71°53.2'W). The material was identified by Sebastián Teillier, Universidad Central de Chile. Voucher specimens are stored at the Herbarium of Universidad de Concepción (CONC).

Oil isolation and analysis: Plant samples were air-dried, cut into small pieces, and submitted to hydrodistillation for 3 h using a modified Clevenger-type apparatus. Oils were dried over anhydrous sodium sulphate and stored in glass ampoules at 4°C until analyzed. All oils were yellowish. Each sample was processed independently.

Qualitative analyses were performed in a Hewlett-Packard 5891 gas chromatograph linked to a Hewlett-Packard 5972 mass spectrometric detector with an integrated data system (Hewlett Packard, Palo Alto, CA, USA), and quantitative analyses were performed in a Shimadzu GC-9A gas chromatograph fitted with an FID-9 detector (Shimadzu Corporation, Kyoto, Japan). The same capillary column (SPB-5, film thickness 0.25 µm, 30 m x 0.25 mm, Supelco, Deerfield IL, USA) was used in both instruments. The operating conditions were as follows: on-column injection; injector temperature, 150°C; detector temperature, 280°C; carrier gas, He; oven temperature program: 50°C for 10 min, increase to 280°C at 5°C/min, and then 280°C for 45 min. In the mass detector, ionization was by electron impact at 70 eV; scan time, 1.5 s; and acquisition mass range, 50–500 amu. The identification of compounds in the chromatographic profiles was achieved by: i) comparison of mass spectra with those in the NIST-98 library database using a reverse search technique

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Table I. Chemical composition (%) of the essential oils of *Satureja darwinii* and *S. multiflora*.

Compound	RI ^a	Concentration (%) ^b		Identification ^c
		<i>S. darwinii</i>	<i>S. multiflora</i>	
3-methylcyclohexanone	955	0.1	- ^d	RI, MS, ST
hexanoic acid	989	0.5	-	RI, MS, ST
3-octanol	995	0.7	-	RI, MS, ST
phenylacetaldehyde	1046	1.8	0.3	RI, MS, ST
6-methylheptanol	1051	0.6	-	RI, MS
trans-linalool oxide ^e	1090	0.4	-	RI, MS
cis-linalool oxide ^e	1092	0.4	-	RI, MS
linalool	1105	1.3	3.7	RI, MS, ST
trans-p-mentha-2,8-dien-1-ol	1124	0.6	-	RI, MS
menthone	1162	-	1.9	RI, MS
isomenthone	1174	-	83.1	RI, MS
borneol	1176	2.7	-	RI, MS, ST
terpinen-4-ol	1187	2.8	-	RI, MS
α -terpineol	1189	2.4	-	RI, MS, ST
neoisomenthol	1193	-	5.4	RI, MS
verbenone	1215	1.3	-	RI, MS, ST
pulegone	1249	11.4	0.7	RI, MS, ST
piperitone	1260	2.1	1.3	RI, MS
thymol	1304	0.8	-	RI, MS, ST
carvacrol	1307	4.8	-	RI, MS, ST
p-vinylguaiaicol	1323	0.9	-	RI, MS
piperitenone	1360	57.8	-	RI, MS
Oxygenated monoterpenes (%)		89.7	96.1	
Other compounds		3.7	0.3	
Total identified		93.4	96.4	
Oil yield (ml/100 g dry weight)		0.68	0.52	

^a Relative retention indices on an SPB-5 column in reference to *n*-alkanes.

^b Peak areas relative to total peak area (means of three samples).

^c MS, NIST MS library, and the literature; RI, retention index; ST, authentic standard compound.

^d Not detected.

^e furanoid form

which verifies that main peaks in the reference spectrum are present in the unknown spectrum (5); and ii) comparison of retention indexes (RI) with those reported in the literature, or with those of available standards. Quantitation was achieved by integration of peak areas in the chromatogram from the FID-fitted gas chromatograph.

Results and Discussion

Twenty-six compounds were characterized, 19 in *S. darwinii* and 7 in *S. multiflora*, representing 93.4% and 96.4% of the oils, respectively (Table I). Both oils were isolated in moderate yields (0.68% and 0.57% v/w, respectively). They contained almost exclusively oxygenated monoterpenes, mostly of the p-menthane family, the major ones being piperitenone and pulegone in *S. darwinii*, and isomenthone in *S. multiflora*.

The composition of the essential oils of more than 50 species of *Satureja* have been reported. On the basis of the data accumulated, *Satureja* oils can be assigned to one of three main chemotypes defined on the basis of the major compounds they contain, i.e., aromatic p-menthane monoterpenes, mainly carvacrol, thymol, and p-cymene (chemotype I); aliphatic p-menthane monoterpenes, mainly menthone, isomenthone, pulegone, and piperitone (chemotype II); or various mono- and sesquiterpenes (chemotype III) (Table II) (6-60). *Satureja*

atropatana (7,34), *S. mutica* (7,17,34), *S. subspicata* (29,30), *S. cuneifolia* (11,50), *S. macrantha* (6,17,56), *S. obovata* (18,57), *S. wiedemanniana* (32,60) and *S. kitaibelii* (19,55) appear as exceptions to this rule because they can be assigned to more than one chemotype. However, the first three cases involve collections performed at different times of the year, the next four cases involve collections performed at different places, and the last case is well known for being an infraspecific systematic category within the extremely polymorphous species, *S. montana* (55). As discussed below, the oil composition can be greatly affected by ontogenetic stage of the plant and its place of growth. In general, while most species studied from the Mediterranean region belong to the first chemotype, most species from South and Central America belong to the last two chemotypes. The essential oil of *S. robusta* is unique in the sense that the four major compounds found are associated to chemotypes II (menthone, 38.0%), I (thymol, 14.1%) and III (germacrene D and geraniol, 13.4% and 11.1%, respectively) (45); it thus constitutes an interesting species for the study of the regulation of terpene biosynthetic pathways.

Several reports have addressed the variability of the composition of *Satureja* oils as a function of plant origin (4,26,48,57,61-63), ontogenesis (24,57,62,64,65), plant part (24), growth environment (57,66,67), time of collection (48,57), and processing conditions (15,25,68). The yield of essential

Table II. Chemotypes of essential oils of *Satureja* species (only the latest references on each species are given).

Main components	<i>Satureja</i> species	References	Main components	<i>Satureja</i> species	References	
Aromatic p-menthane monoterpenes (chemotype I)	<i>aintabensis</i>	6	Aliphatic p-menthane monoterpenes (chemotype II)	<i>abyssinica</i> ssp. <i>abbysinica</i>	33	
	<i>atropatana</i>	7		<i>atropatana</i>	34	
	<i>bachtiarica</i>	8		<i>boliviana</i>	35	
	<i>boissieri</i>	9		<i>brevicalyx</i>	36	
	<i>cilicica</i>	10		<i>brownei</i>	37	
	<i>cuneifolia</i>	11		<i>douglasii</i>	38	
	<i>edmondi</i>	12		<i>fruticosa</i>	39	
	<i>hortensis</i>	13		<i>gilliesii</i>	3	
	<i>horvatii</i>	14		<i>glabella</i>	40	
	<i>horvatii</i> ssp. <i>macrophylla</i>	15		<i>grandiflora</i>	41	
	<i>icarica</i>	16		<i>mutica</i>	34	
	<i>intermedia</i>	17		<i>odora</i>	42	
	<i>intricata</i>	18		<i>paradoxa</i>	33	
	<i>kitaibelii</i> f. <i>aristata</i>	19		<i>paradoxa</i> ssp. <i>sipylea</i>	43	
	<i>khuzistanica</i>	20		<i>parvifolia</i>	35	
	<i>laxiflora</i>	21		<i>pseudosimensis</i>	44	
	<i>macrantha</i>	6,17		<i>robusta</i>	45	
	<i>mutica</i>	7,17		<i>viminea</i>	46,47	
	<i>montana</i>	22		Various monoterpenes and sesquiterpenes (chemotype III)	<i>adamovicii</i>	48
	<i>montana</i> ssp. <i>kitaibelii</i>	23			<i>alpina</i>	40
	<i>obovata</i>	18			<i>biflora</i>	44,49
<i>parmassica</i> ssp. <i>parmassica</i>	24	<i>boliviana</i>	4			
<i>pilosa</i>	25	<i>coerulea</i>	16			
<i>rechingeri</i>	26	<i>cuneifolia</i>	50			
<i>sahendica</i>	27	<i>forbesii</i>	51			
<i>spicigera</i>	28	<i>fukarekii</i>	48			
<i>spinosa</i>	29	<i>glabrata</i>	52			
<i>subspicata</i>	30	<i>innota</i>	53			
<i>subspicata</i> ssp. <i>liburnica</i>	31	<i>isophylla</i>	12			
<i>thymbra</i>	32	<i>juliana</i>	54			
<i>wiedemanniana</i>		<i>kitaibelii</i>	55			
		<i>macrantha</i>	56			
		<i>masukensis</i>	44			
		<i>obovata</i>	57			
		<i>punctata</i>	58			
		<i>salzmannii</i>	53			
		<i>spinosa</i>	59			
		<i>subspicata</i>	29			
		<i>visianii</i>	22			
		<i>wiedemanniana</i>	60			

oil has also shown considerable variability. The relationship between composition and yield of essential oils can best be assessed in studies which have used similar extraction procedures on comparable biological material. Most studies which have reported both the yield and composition of *Satureja* oils have used hydrodistillation of dry aerial plant material originally collected at the flowering stage; these studies are listed in Table III (6,7,11,12,17,20,23-26,36,40,42,43,48,56,59,60,62,63,68-82). Three yield groups may be distinguished: i) with yields higher than 1%, containing only chemotype I; ii) with intermediate yields (between 0.5% and 1%) containing chemotypes III, I and II, with the latter predominating; and iii) with low yields (< 0.5%) containing chemotypes I and III, with the latter predominating. To assess whether affiliation to a yield group

can be used for chemotype assignment, the expected and observed chemotype frequencies within each yield group were compared using the chi-squared test. The frequencies were not significantly different (chi-squared = 2.6166, $P > 0.25$, $N = 53$), showing that the affiliation of an oil to a yield group can be confidently used to assign its chemotype. It is interesting to note that this trend holds even within species. Thus, when particular collections of *S. cuneifolia*, *S. macrantha* and *S. parmassica* ssp. *parmassica* produce oils in low yields, those oils belong to chemotype III, whereas when collections of the same species produce high yield of oils, they belong to chemotype I (Table III).

In conclusion, *Satureja* oils may be assigned to three different chemotypes depending on the nature of their main

Table III. Yield and composition of *Satureja* oils obtained by hydrodistillation of dry aerial plant material originally collected at the flowering stage.

Species	Chemotype	Main components (%)	Reference	Yield (%)
<i>rechingeri</i>	I	carvacrol (86.6)	25	4.2
<i>spicigera</i>	I	thymol (35.1), p-cymene (22.1), γ -terpinene (13.7)	69	3.8
<i>khuzistanica</i>	I	p-cymene (39.6), carvacrol (29.6), γ -terpinene (18.9)	70	3.0
<i>hortensis</i>	I	carvacrol (36.2), γ -terpinene (30.9), thymol (11.5)	63	2.5*
<i>cuneifolia</i>	I	thymol (65.5), p-cymene (9.8), carvacrol (7.2)	11	2.5
<i>thymbra</i>	I	thymol (41.0), γ -terpinene (22.2), p-cymene (11.8)	24	2.4
<i>mutica</i>	I	carvacrol (30.9), thymol (26.5), γ -terpinene (14.9), p-cymene (10.3)	17	2.3
<i>sahendica</i>	I	p-cymene (30.2), thymol (29.6), γ -terpinene (27.7)	26	2.3
<i>cuneifolia</i>	I	thymol (43.6), carvacrol (31.2), p-cymene (11.5)	71	2.2
<i>thymbra</i>	I	thymol (17.2), γ -terpinene (12.5), carvacrol (29.2), p-cymene (10.9)	72	2.2
<i>pilosa</i> var. <i>pilosa</i>	I	thymol (46.1), p-cymene (12.7), γ -terpinene (8.7)	23	2.1
<i>boissieri</i>	I	carvacrol (40.8), γ -terpinene (26.4), p-cymene (14.5)	73	2.1
<i>aintabensis</i>	I	p-cymene (59.0), thymol (17.5)	6	2.0
<i>cuneifolia</i>	I	carvacrol (46.4), p-cymene (15.8), γ -terpinene (13.0)	74	1.9
<i>subspicata</i>	I	carvacrol (16.8), α -pinene (13.6), p-cymene (10.8), thymol methyl ester (8.8)	75	1.8
<i>cuneifolia</i>	I	carvacrol (59.3), thymol (15.7), p-cymene (9.7)	76	1.7
<i>macrantha</i>	I	p-cymene (25.8), limonene (16.3), thymol (8.1)	17	1.5
<i>intermedia</i>	I	thymol (32.3), γ -terpinene (29.3), p-cymene (14.7)	17	1.5
<i>pamassica</i> ssp. <i>sipylea</i>	I	carvacrol (42.9), p-cymene (20.1)	43	1.5
<i>pamassica</i> ssp. <i>pamassica</i>	I	thymol (20.3), carvacrol (34.6), γ -terpinene (16.7), p-cymene (6.9)	24	1.4
<i>montana</i>	I	carvacrol (29.8), p-cymene (14.6), thymol (7.7)	62	1.3*
<i>khuzistanica</i>	I	carvacrol (80.6)	20	1.2
<i>spicigera</i>	I	thymol (25.8), p-cymene (32.0), carvacrol (5.5)	77	1.2*
<i>hortensis</i>	I	thymol (40.5), γ -terpinene (18.6), carvacrol (14.0)	78	1.1
<i>icarica</i>	I	carvacrol (53.4), p-cymene (14.8)	79	1.1*
<i>parvifolia</i>	I	carvacrol (34.0), carvacryl acetate (14.7), p-cymene (14.0), γ -terpinene (11.3)	42	1.1
<i>pilosa</i>	I	carvacrol (53.5), p-cymene (17.4)	79	1.1
<i>glabella</i>	II	isomenthone (39.9), pulegone (33.3),	40	1.1*
<i>boliviana</i>	II	isomenthone (29.7), menthone (24.2)	36	1.0
<i>edmondi</i>	I	p-cymene (61.1), γ -terpinene (9.6)	12	1.0
<i>brevicalix</i>	II	menthone (37.5), isomenthone (25.2)	36	1.0
<i>hortensis</i>	I	γ -terpinene (38.5), carvacrol (47.0)	68	1.0*
<i>visianii</i>	III	linalool (68.6), thymol (5.6)	48	0.80
<i>odora</i>	II	pulegone (61.5), isomenthone (5.8), menthone (3.4)	42	0.70
<i>darwinii</i>	II	piperitenone (57.8), pulegone (11.4)	this work	0.68
<i>khuzistanica</i>	I	carvacrol (93.9)	20	0.60
<i>multiflora</i>	II	isomenthone (83.1), neoisomenthol (5.4)	this work	0.57
<i>horvatii</i>	I	p-cymene (27.6), thymol (27.4), carvacrol (17.8)	80	0.53
<i>pamassica</i> ssp. <i>pamassica</i>	III	β -caryophyllene (20.9), carvacrol (20.4), spathulenol (17.2), p-cymene (13.0)	81	0.44
<i>cuneifolia</i>	III	α -pinene (10.5), limonene (8.5), carvacrol (7.7), p-cymene (6.4), borneol (6.4), β -cubebene (5.8), β -caryophyllene (6.0)	62	0.43*
<i>boissieri</i>	I	carvacrol (70.1), γ -terpinene (6.8), p-cymene (6.3)	82	0.30
<i>spicigera</i>	I	thymol (37.3), p-cymene (14.6), γ -terpinene (14.5), carvacrol (9.2)	56	0.30
<i>isophylla</i>	III	α -eudesmol (11.3), camphor (7.1), β -caryophyllene (6.1), γ -eudesmol (5.8), geranial (5.5)	12	0.29
<i>fukarekii</i>	III	α -phellandrene (17.3), <i>iso</i> -longifolene aldehyde (13.2), limonene (12.6)	48	0.25
<i>adamovicii</i>	III	α -phellandrene (13.0), <i>iso</i> -longifolene aldehyde (13.3), limonene (8.1)	48	0.20
<i>macrantha</i>	III	spathulenol (14.0), vanillin (13.4), p-cymene (12.3), caryophyllene oxide (7.2)	56	0.20
<i>mutica</i>	I	thymol (62.6), p-cymene (9.4), carvacrol (6.6)	7	0.20
<i>subspicata</i> subsp. <i>subspicata</i>	I	carvacrol (58.1), terpinen-4-ol (18.5)	80	0.19
<i>montana</i> ssp. <i>kitaibelii</i>	III	limonene (15.7), p-cymene (13.1), germacrene D (8.1)	23	0.19
<i>spinosa</i>	III	linalool (47.4), germacrene D (4.2), bicyclogermacrene (4.8)	59	0.12
<i>atropatana</i>	I	thymol (62.1), p-cymene (6.1)	7	0.10
<i>wiedemanniana</i>	III	caryophyllene oxide (8.5), borneol (8.3), germacrene D (8.1), limonene (7.7), spathulenol (7.7), β -caryophyllene (6.5), β -bisabolene (6.0)	60	0.06
<i>alpina</i>	III	germacrene D (33.2), α -muurolene (6.1), β -caryophyllene (4.4), β -bourbonene (4.1)	40	<0.01

* Mean of values reported.

constituents. The yield of oil is correlated with the nature of the chemotype. The oils of the two *Satureja* species described in this paper conform to these general trends.

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