Volatile Oil Composition of Four Populations of *Satureja Montana* L. from Southern France

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Abstract

The essential oil fraction in single individuals of four populations of *Satureja montana* L. from Southern France has been analysed in order to get insight into the chemical biodiversity of this species. The main essential oil compounds found were carvacrol, p-cymene and γ-terpinene. Three populations displayed carvacrol as main compound in nearly all individuals and appeared rather uniformly.

Another population was heterogenous, about a third of the individuals were high in linalool whereas the other displayed p-cymene or carvacrol as main essential oil compound. Therefore a classification in a linalool ans a carvacrol/p-cymene chemotype is suggested.

INTRODUCTION

The Mediterranean region is rich in aromatic plants, which have been used since antiquity. Plants of the same species but from different origin may display different properties. The knowledge of this infraspecific variability is important to understand the existing uses of a plant and to propose further applications.

Savory (*Satureja montana* L., Lamiaceae) is a widespread subshrub with white flowers and small rough leaves in the rocky garrigue vegetation over lime stone underground in the northern Mediterranean region. The aromatic plant with a spicy flavour displays antimicrobial activity (Mohacsi-Farkas et al., 2001; Stahl-Biskup, 1998). The main uses are as spice and in folk medicine as a remedy for gastrointestinal disorders.

The main chemical compounds of the essential oil are aromatic monoterpenes as carvacrol, which are responsible for the characteristic taste. Although these aromatic substances represent the typical and prevailing chemotype, plants or oils rich in borneol (Lawrence, 1988) or linalool (Garnero et al., 1981) have been reported, suggesting the occurence of various chemotypes. Further active components are hydroxycinnamic acid derivatives and rosmarinic acid (Stahl-Biskup, 1998).

To get more insight in the chemical variability of this plant, we studied the essential oil patterns of single plants from four populations from southern France in order to test their homogeneity.

MATERIAL AND METHODS

Plant Material

The plant material was collected between 10th and 15th of August 2000 from 4 different locations in Southern France when the plants were flowering:
population 1: Alpes Maritimes, Utelle, 830-850 m, N 43° 55.15´; E 7° 14.66´;
population 2: Alpes de Haute Provence, Rougon, 950 m, N 43° 48.0´ ; E 4° 04.5´
population 3: Vaucluse, Mont Ventoux, 1080-1090 m, N 44° 07.91´; E 5° 17.52´
population 4: Ardèche, Plateau de Gras, 210 m, N 44° 20.65´; E 4° 35.34´.

All the populations grew in an open garrigue vegetation in the French lime stone prealps (1, 2 and 3) and west of the Rhone river on a lime stone plateau (4). 33 to 40 single flowering plants were sampled from each site. As it is necessary to analyse ontogenetically comparable plant material, only flowering branches with leaves were

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taken and dried in the shadow.

Chemical Analysis

The volatile components were extracted from 0.2 – 0.3 g of the leaves of single plants with 1.5 to 2 ml dichloromethane for 30 minutes in an ultrasonic bath at room temperature and filtered. This extract was directly used for the GC/MS analysis.

To record the pattern of volatile components a HP 6890 GC was disposable, equipped with a 5972 quadrupole mass selective detector. The separation was done on a 30 m x 0,25 mm column coated with 0,25µm HP5-MS. The analytical conditions were: carrier gas He 1.3 ml/min constant flow, injector temperature 250°C, split ratio 15:1, temperature program 2 min at 40°C, with 3°C/min up to 100°C, with 5°C/min up to 150°C, with 15°C/min up to 280°C. The compounds were identified according their mass spectra and their retention indices (Adams, 1995; McLafferty, 1989). The total ion current of each identified peak was measured and the sum of all peaks set to 100 %.

RESULTS

The variability of selected volatile compounds of the four populations tested is presented in table 1. Together they make up 85-90 % of the identified compounds. It can be seen that the main essential oil components recorded in *Satureja montana* were carvacrol, γ-terpinene and p-cymene; together they represent up to 72 % of the essential oil fraction. Further minor compounds are: α-thujene, α-pinene, camphene, ß-pinene, α-phellandrene, δ-3-carene, 1,8-cineole, trans-ß-ocimene, cis- and trans-sabinenehydrate, terpinolene, terpinene-4-ol, α-terpineole, thymol, α-copaene, ß-bourbonene, ß-caryophyllene, calarene, aromadendrene, α-humulene, α-amorphene, germacrene D, α-muurolene, γ-cadinene, δ-cadinene, spathulenol and caryophyllene oxide, each representing in most cases less than 0,5 % in the fraction.

One population displayed a high variability (population 1): About a third of the individuals were high in linalool with low amounts of carvacrol and γ-terpinene and variable levels of p-cymene. The remaining individuals of this population showed variable amounts of carvacrol and p-cymene, γ-terpinene but little linalool. This population is presented in Fig. 1A.

The three remaining populations (2 to 4) looked rather uniformly, they contained mainly carvacrol, p-cymene and γ-terpinene, with carvacrol ranging from 32 to 68 % in the essential oil fraction as the main compound in nearly all individuals. An example of such a uniform population is given in Fig. 1B.

Regarding the minor compounds the three uniform populations were generally higher in myrcene, α-terpinene and ß-bisabolene but lower in limonene and borneol than the variable population mentioned before. In one population (nr. 3) about half of the tested individuals showed up to 10 % of carvacrol-methylether. Higher amounts of thymol (up to 12 %, data not shown) could be found only in one and two individuals from population 4 and 3 respectively.

DISCUSSION

In thyme the biosynthetic conversion from γ-terpinene to p-cymene and further to thymol has been demonstrated (Poulose and Croteau, 1978). Carvacrol, which is similar to thymol, may be produced by a comparable biosynthesis in *Satureja montana* from γ-terpinene via p-cymene. Therefore plants containing these three compounds may be considered as the main chemotype of *Satureja montana*. The typical essential oil of *Satureja montana* is rich in the phenolic compound carvacrol (Stahl-Biskup, 1998) and the prevailing carvacrol chemotype occurs also in Italy and the former Yugoslavia (Lawrence, 1988).

The relative amount of these three compounds in a plant or oil is influenced by various factors as plant development and climatic conditions on the growing site. It has been found that during the growing season the carvacrol content decreases whereas the p-cymene level tends to increase (Milos et al., 2001; Stanic et al., 1991; Garnero et al., 1981). During the same time interval the γ-tepinene may increase (Stanic et al., 1991) or decrease (Garnero et al., 1981). Furthermore higher temperature and less precipitation may increase the carvacrol content of *Satureja* (Piccaglia et al., 1991).

To test whether a population is homogenous, the essential oil patterns of an adequte number of individual plants have to be analysed. Homogeneity could be demonstrated in three populations (nr. 2, 3 and 4) where all individuals displayed similar oil patterns.

Another population (nr. 1) showed some individuals with linalool as main compound. These individuals can be considered as representatives of a separate linalool chemotype although carvacrol and its precursors are also present as minor compounds, indicating that the metabolic pathway producing aromatic compounds is also operating. A comparable highly variable population with individuals rich in linalool or carvacrol or p-cymene has been signalised in Spain in the province of Gerona (San Martin et al., 1973 cited in Garnero et al., 1981). These authors investigating savory from 12 spanish stations recognise a carvacrol and a linalool type with variable amount of p-cymene. Further chemotypes may occur where a borneol rich oil has also been reported from France (Lawrence, 1988) and a thymol rich one also from Spain (Lawrence, 1988). Oils with thymol and geraniol besides carvacrol have been obtained from plants from Dalmatia (Milos et al., 2001).

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Literature Cited

Adams, R.P. 1995. Identification of essential oil components by gas chromatography and mass spectroscopy. Allured, Carol Stream, IL, USA.

Garnero, J., Buil P. and Pellecuer, J. 1981. Étude de la composition chimique de l´huile essentielle de *Satureja montana* L. (labiées). Rivista Italiana EPPOS 7: 344-349.

Héjja, M., Héthelyi, É., Ferency, A.Z. and Szentgyörgyi, E. 2001. Changes of the characteristics of *Satureja hortensis* L. herb during flowering period. Internat. J. Horticultural Sci. 7: 56-60.

Lawrence, B. 1988. Perfumer & Flavorist 13, Feb/March 44-50.

Milos, M., Radonic, A., Bezic, N. and Dunkic, V. 2001. Localities and seasonal variations in the chemical composition of essential oils of *Satureja montana* L. and *S. cuneifolia* Ten. Flavour and Fragrance J. 16: 157-160.

McLafferty, F.W. 1989. Registry of mass spectral data. 5th edition. John Wiley & Sons, New York.

Mohácsi-Farkas, Cs., Tulok, M. and Balogh, B. 2001. Antimicrobial activity of *Origanum vulgare* subsp. *hirtum* L. and *Satureja montana* L. extracts (essential oil and SCFE) investigated by impedimetry. World Conference on Medicinal and Aromatic Plants, Budapest, Hungary 2001, Abstract PIV/36.

Piccaglia, R., Marotti, M. and Galletti, G.C. 1991. Characterization of essential oil from a *Satureja montana* L. chemotype grown in Northern Italy. J. Essent. Oil Res. 3: 147-152.

Poulose, A.J. and Croteau, R. 1978. Biosynthesis of aromatic monoterpenes. Conversion of γ-terpinene to p-cymene and thymol in *Thymus vulgaris* L. Arch. Biochem. Biophys. 187: 307-314.

Stahl-Biskup, E. 1998. *Satureja*. In: Hagers Handbuch der Pharmazeutischen Praxis, Folgeband 3, 5th edition; Springer, Berlin; p 522-528.

Stanic, G., Petricic, J. and Blazevic, N. 1991. Gas Chromatographic investigations of essential oil of *Satureja montana* and *S. subspicata* from Yugoslavia. J. Essent. Oil Res. 3: 153-158.

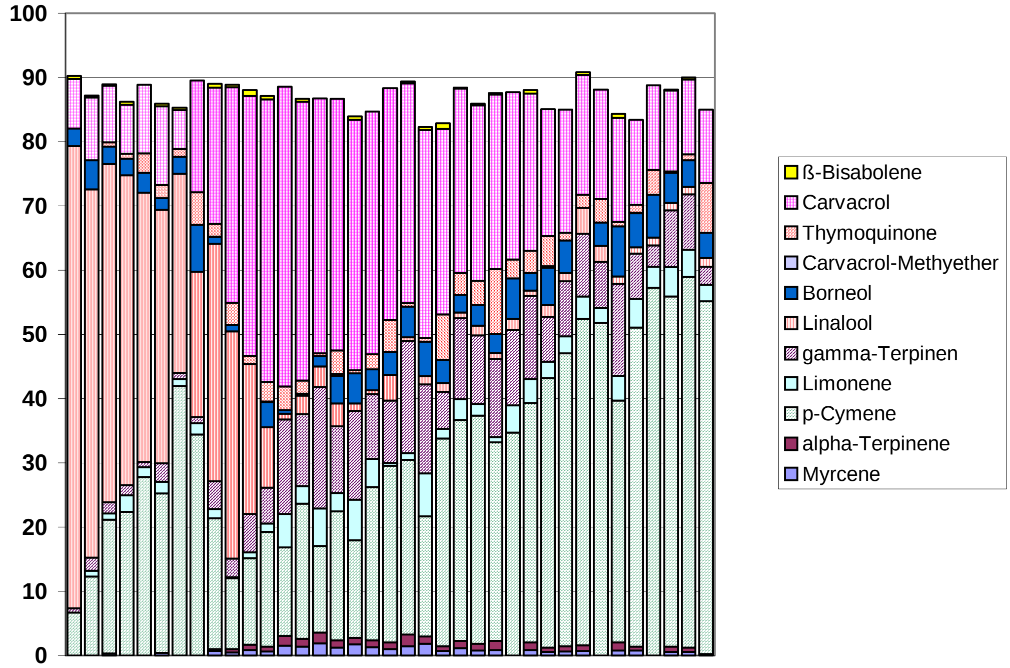
Tables

Table 1. Varialbility of selected essential oil components in four *S. montana* populations

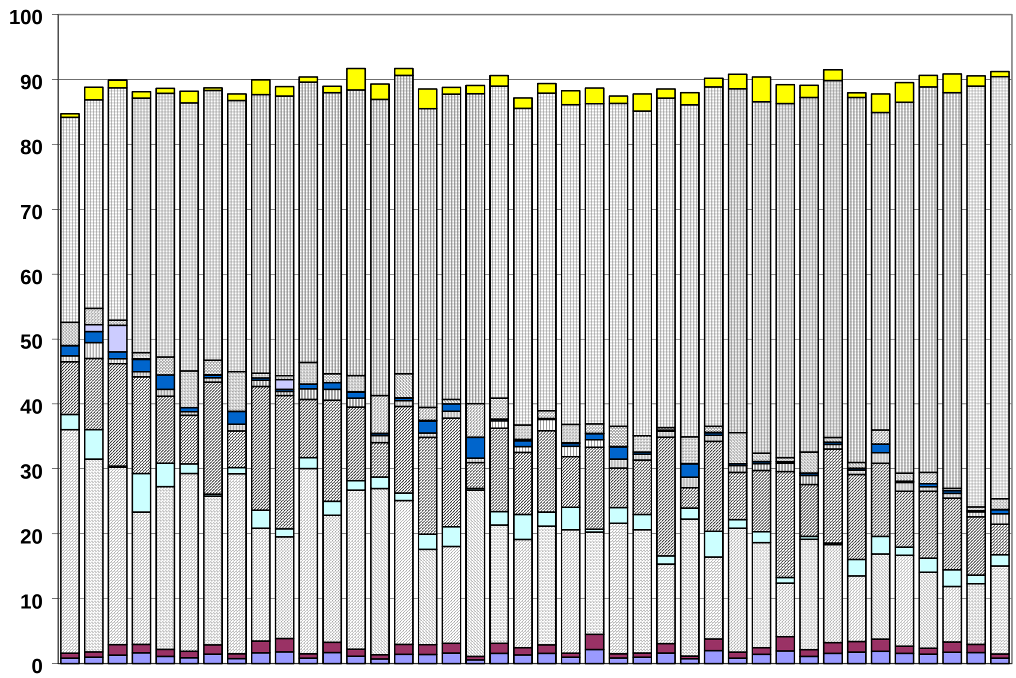
	Population 1 n = 37			Population 2 n = 40			Population 3 n = 36			Population 4 n = 33		
	min	median	max	min	median	max	min	median	max	min	median	max
Carvacrol	6,1	21,2	46,6	31,6	48,8	65,0	32,4	54,7	67,8	31,7	52,7	62,0
p-Cymene	6,7	27,8	57,7	0,0	18,3	34,4	7,0	12,6	25,1	0,0	15,9	20,8
gamma-Terpinene	0,7	8,6	19,0	3,2	11,0	20,5	4,7	12,0	24,0	4,9	12,6	22,5
Linalool	0,6	1,8	71,9	0,5	1,1	2,5	0,4	1,0	4,0	0,5	1,3	4,2
Limonene	0,0	2,6	6,7	0,2	1,7	5,9	0,0	0,4	4,6	0,1	0,4	4,4
Thymochinone	0,0	2,1	10,1	0,4	2,1	6,1	0,1	1,3	5,9	0,1	0,6	2,5
Borneol	0,0	3,5	7,8	0,1	0,5	3,2	0,3	0,7	3,2	0,4	1,0	2,4
alpha-Terpinene	0,0	0,7	1,8	0,4	1,2	2,3	0,8	1,5	2,6	0,8	1,5	2,9
ß-Bisabolene	0,0	0,3	0,9	0,4	1,6	3,8	0,3	1,8	4,0	0,5	1,9	3,8
Myrcene	0,0	0,7	1,9	0,6	1,4	2,2	0,9	1,5	2,8	0,9	1,7	2,8
Carvacrol-Methylether	0,0	0,0	0,4	0,0	0,0	4,1	0,0	1,5	9,5	0,0	0,0	0,3

Figures

Fig. 1. Composition of the volatile oil of *S. montana*. Each bar represents an individual



A: the example of a variable population (nr. 1, Utelle).



B: The example of a homogenous population (nr. 2, Rougon)