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CHEMICAL COMPOSITION OF THE ESSENTIAL OIL FROM *Stachys serotina*

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Stachys L. is one of the largest genera of the Lamiaceae of approximately 300 species, distributed mainly in the warm temperate regions of the Mediterranean and Southwest Asia, with secondary centers in North and South America and southern Africa [1–3]. The essential oil composition of the many species of this large genus has not been studied in much detail. To our knowledge, there are no published reports on the chemical composition of *Stachys serotina* (Host) Fritsch. This is an aromatic, erect, perennial plant similar to *Stachys officinalis* (L.) Trev., but distinguished clearly in very interrupted inflorescence and later flowering period [4].

In Table 1, data on essential oil chemical constituents of this species are given. Sesquiterpene hydrocarbons were the most abundant class of isolated volatiles with a predominance of β-caryophyllene (22.6%) followed by δ-cadinene (9.6%), α-humulene (7.5%), germacrene D (6.0%), and minor percentages of others. Sesquiterpene hydrocarbons were the main group of constituents of many *Stahys* species from Croatia (*S. officinalis* L. Trevis., *S. palustris* L., *S. recta* L. subsp. *recta*, *S. recta* L. subsp. *subcrenata* (Vis.) Briq., *S. salviifolia* Ten. and *S. sylvatica* L.), except *Stachys alpine* L., which was rich in oxygenated sesquiterpenes [5]. The determined percentage of β-caryophyllene in *Stachys serotina* was the highest among other *Stachys* species from Croatia [5], being most abundant in *Stachys officinalis* (14.6%). In addition, δ-cadinene was also found with greatest abundance in comparison with other *Stachys* species [5]. The determined content of α-humulene (7.5%) was close to that of *Stachys officinalis* (6.7%), while other *Stachys* species contain less (or do not contain) α-humulene. Monoterpenes were minor oil constituents of *Stachys serotina*, dominated by the monoterpene hydrocarbon α-pinene (11.1%). A high percentage of α-pinene (21.4%) was found in *Stachys sylvatica* [5]. This study revealed several differences between *Stachys serotina* and other *Stachys* species in Croatia, indicating the existence of a chemical polymorphism.

Plant Material and Isolation Procedure. The aerial parts of *S. serotina* were collected on Adriatic island Krk in July 2010. *Stachys serotina* (Host) Fritsch, Lamiaceae was authenticated by K. Hazler Pilepic. A voucher specimen is deposited in the Herbarium of the Department of Pharmaceutical Botany (FB 1302). Plant material (100 g) and water (500 mL) were placed in a Clevenger-type apparatus. The essential oils were isolated by hydrodistillation for 2 hours. The obtained essential oils were separated, dried over anhydrous sodium sulfate, and stored under argon in sealed vials at 4°C until required.

Identification of the Oil Components. The analyses of the obtained essential oil were performed by GC-FID and GC-MS.

GC-FID Analysis. Gas chromatography analyses were carried out on an Agilent Technologies (Palo Alto, CA, USA) gas chromatograph model 7890A equipped with flame ionization detector. Chromatographic separations were performed on a 30 m × 0.25 mm i.d HP-5MS capillary column (5% phenylmethylpolysiloxane, Agilent J & W GC column) with coating thickness 0.25 μm. The oven was temperature-programmed isothermal from 70°C for 2 min, then increased to 200°C at a rate of 3°C/min and held isothermal for 15 min. Helium at 1 mL/min was used as carrier gas. The injector temperature was 250°C and the detector temperature was 300°C. The injected volume was 1 μL and the split ratio was 1:50. GC-FID analysis was carried out in duplicate.

GC-MS Analysis. Analyses of volatile compounds by gas chromatography-mass spectrometry were carried out with the Agilent gas chromatograph model 7890A fitted with a mass selective detector model 5975C (Agilent Technologies, Palo Alto, CA, USA). The mass detector worked in the electron impact ionization mode at 70 eV, the mass range was *m/z* 30–300, and the ion source temperature was 280°C. Volatile compound separation was obtained using the same column and oven temperature program as previously described. GC-MS analysis was carried out in duplicate.

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TABLE 1. Identified Constituents of the Essential Oils from *Stachys serotina*

Compound	RI	%	Compound	RI	%
(*E*)-Hex-2-enal	<900	0.1	α-Copaene	1383	0.8
α-Thujene	933	0.5	β-Bourbonene	1391	1.3
α-Pinene	943	11.1	(*E*)-β-Caryophyllene	1431	22.6
Camphene	957	0.1	β-Cubebene	1437	0.7
Verbenene	962	0.1	Alloaromadendrene	1447	0.1
Benzaldehyde	969	1.0	α-Humulene	1463	7.5
Sabinene	980	0.1	(*E*)-β-farnesene	1466	1.0
β-Pinene	984	0.4	γ-Gurjunene	1480	0.1
Oct-1-en-3-ol	986	0.7	γ-Muurolene	1486	3.8
Myrcene	995	0.7	Germacrene D	1490	6.0
Limonene	1036	0.4	β-Selinene	1493	0.4
(*Z*)-β-ocimene	1044	0.1	α-Bergamotene*	1504	3.1
Phenylacetaldehyde	1052	0.1	α-Muurolene	1508	1.2
(*E*)-β-ocimene	1055	0.1	(*E*,*E*)-α-Farnesene	1518	0.6
γ-Terpinene	1066	0.1	γ-Cadinene	1523	2.1
Linalool	1107	0.3	δ-Cadinene	1528	9.6
Nonanal	1110	0.1	Cadina-1,4-diene**	1542	0.4
α-Campholene aldehyde	1134	0.1	α-Cadinene	1547	0.5
(*Z*)-Verbenol	1153	0.2	(*E*)-Nerolidol	1575	0.3
(*E*)-Verbenol	1157	1.3	Caryophyllene oxide	1592	3.8
Terpinen-4-ol	1186	0.1	*T*-Cadinol	1655	0.8
α-Terpineol	1199	0.2	α-Cadinol	1664	2.0
Myrtenol	1208	0.1	Caryophyllenol II	1687	0.8
Verbenone	1218	0.1	Tetradecanoic acid	1873	0.1
δ-Elemene	1343	1.0	Hexadecanoic acid	1988	1.5
α-Cubebene	1357	0.4	Phytol**	2136	0.1
Eugenol	1367	0.2	Linoleic acid	2165	0.2
α-Ylangene	1378	0.3	Total identified		91.4

RI: retention indices on HP-5MS; *tentatively identified; **correct isomer not identified.

Quantitative results are mean data derived from duplicate GC-FID analyses. The individual peaks were identified by comparison of their retention indices (relative to C_9–C_{25} *n*-alkanes for the HP-5MS column) to those of authentic samples and literature [6], as well as by comparing their mass spectra with the Wiley 275 MS library (Wiley, New York, USA) and NIST98 (Gaithersburg, Germany) mass spectral database. The percentage composition of the samples was computed from the GC peak areas using the normalization method (without correction factors).

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REFERENCES

1. P. W. Ball, *Stachys* L., in: T. G. Tutin, V. H. Heywood, N. A. Burges, D. M. Moore, D. H. Valentine, S. M. Walters, and D. A. Webb (Eds.), *Flora Europaea*, Vol. **3**, Cambridge University Press, Cambridge, 1972, p. 151.
2. V. H. Heywood, *Flowering Plants of the World*, University Press, London, 1993.
3. C. King and M. Hickey, *Common Families of Flowering Plants*, Cambridge University Press, Cambridge, 1997.
4. R. Domac, *Flora of Croatia*: *Manual for Determination of Plants*, Skolska knjiga, Zagreb, 2002, p. 296.
5. V. Bilusic Vundac, H. W. Pfeifhofer, A. H. Brantner, Z. Males, and M. Plazibat, *Biochem. Syst. Ecol.*, **34**, 875 (2006).
6. A. M. El-Sayed, *The Pherobase: Database of Insect Pheromones and Semiochemicals*, http://www.pherobase.com, 2007.