

ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OILS OF SELECTED *Stachys* SPECIES

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Stachys species have long been used in traditional medicine to treat genital tumors, sclerosis of the spleen, inflammatory tumors, and cancerous ulcers [1]. In Turkish folk medicine, species of the *Stachys* genus are used in the same way as sage [2]. Several *Stachys* species are recognized in Iranian folk medicine [3], especially *S. inflata*, which is used to treat various inflammatory disorders [4].

In view of the growing interest in natural product applications in the food, cosmetics, and pharmaceutical industries, and having in mind the scarce previous work on the Balkan *Stachys* species, the subject of this work was to evaluate the antimicrobial activities of essential oils of selected *Stachys* species.

Stachys germanica ssp. *heldreichii* (Boiss.) Hayek, *Stachys iva* Griseb., *Stachys plumosa* Griseb., and *Stachys scardica* Griseb. essential oils were analyzed by means of GC and GC/MS. The observed antimicrobial activity was correlated with the composition.

Eighty-three identified compounds, which accounted for 96.3-99.6% of the total composition of the oils, are reported in Table 1.

The main objective of this work was to evaluate the antimicrobial activity of these oils and not to discuss the chemotaxonomic significance of the volatiles (already addressed in our previous work [5]).

The results of the bioassay are presented in Table 2 [6].

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TABLE 1. Chemical Composition of the Essential Oils of *Stachys* sp.

Compounds	RI ^a	RI ^b	<i>S. germanica</i>	<i>S. scardica</i>	<i>S. plumosa</i>	<i>S. iva</i>
α -Pinene	936	946	-	4.6	2.1	0.5
Benzaldehyde	959	1011	3.5	-	-	-
Sabinene	972	968	-	Tr.	0.7	-
β -Pinene	978	995	0.2	0.4	4.2	2.3
Limonene	1027	1054	-	0.1	-	1.2
β -Phellandrene	1028	1050	-	-	0.5	-
1,8-Cineole	1031	1050	3.0	Tr.	-	1.6
γ -Terpinene	1059	1067	-	-	0.3	-
<i>trans</i> -Linalool oxide (furanoid)	1065	1102	0.5	-	-	-
<i>cis</i> -Linalool oxide (furanoid)	1074	1089	0.5	-	-	-
Terpinolene	1085	1098	-	-	0.8	-
Linalool	1098	1130	0.7	0.2	-	-
α -Campholenal	1125	1128	-	0.1	1.1	-
<i>allo</i> -Ocimene	1128	1117	-	-	0.6	-
<i>trans</i> -Pinocarveol	1134	1143	-	-	Tr.	-
Geijerene	1139	1151	1.1	-	-	-
<i>cis</i> -Verbenol	1140	1148	-	0.2	8.2	-
Camphor	1143	1139	5.7	-	-	-
<i>trans</i> -Verbenol	1143	1162	-	0.1	1.9	-
Pinocarvone	1165	1153	0.4	0.3	9.0	-
Myrtenal	1193	1175	-	Tr.	2.8	0.1
Verbenone	1205	1183	-	-	0.6	-
Dihydroedulan II	1284	1272	Tr.	Tr.	-	-
Bornyl acetate	1285	1259	-	-	0.9	-
Bicycloelemene	1336	1318	-	-	-	4.2
δ -Elemene	1339	1321	0.2	2.6	-	-
α -Cubebene	1348	1336	0.2	2.0	-	0.1
α -Ylangene	1368	1360	-	1.4	-	-
α -Copaene	1375	1366	1.3	3.5	1.1	8.4
β -Bourbonene	1382	1374	2.3	4.6	-	-
β -Elemene	1388	1379	-	0.7	-	-
β -Cubebene	1391	1384	-	-	-	1.2
1,5-di- <i>epi</i> - β -Bourbonene	1392	1385	2.5	-	-	0.4
β -Longipinene	1398	1390	-	0.5	0.3	-
α -Gurjunene	1409	1400	-	-	-	0.6
(<i>E</i>)- β -Damascone	1412	1428	0.9	-	0.9	0.6
<i>cis</i> - α -Bergamotene	1414	1398	-	0.3	-	-
β -Caryophyllene	1418	1409	5.1	10.0	1.4	9.3
β -Copaene	1428	1418	2.0	2.3	-	-
Aromadendrene	1438	1428	1.3	0.8	-	0.2
Isogermacrene D	1439	1431	-	0.6	-	0.8
α -Humulene	1452	1440	1.2	1.2	-	1.9
(<i>E</i>)- β -Farnesene	1457	1449	3.1	-	0.8	1.6
<i>allo</i> -Aromadendrene	1458	1457	0.5	-	-	-
<i>cis</i> -Muurola-4(15),5-diene	1459	1447	-	0.6	-	-
γ -Muurolene	1477	1459	0.9	13.1	-	-
<i>ar</i> -Curcumene	1479	1464	-	-	0.3	-
Germacrene D	1480	1465	8.1	-	-	1.3
<i>cis</i> - β -Guaiane	1491	1469	-	1.2	-	-
Bicyclogermacrene	1494	1477	-	-	-	7.4
γ -Amorphene	1496	1474	-	3.1	-	-
γ -Cadinene	1513	1492	3.4	4.1	-	-
<i>cis</i> -Calamenene	1517	1494	-	2.0	-	-
δ -Cadinene	1524	1498	3.3	9.3	0.4	10.0
α -Calacorene	1540	1517	-	3.4	-	-
β -Calacorene	1561	1543	-	1.3	-	-
(<i>E</i>)-Nerolidol	1564	1556	13.5	-	-	0.1

TABLE 1 (continued)

Compounds	RI ^a	RI ^b	<i>S. germanica</i>	<i>S. scardica</i>	<i>S. plumosa</i>	<i>S. iva</i>
γ -Calacorene	1571	1553	-	0.7	-	-
Spathulenol	1577	1566	-	-	-	8.1
β -Copaen-4 α -ol	1579	1583	-	-	-	4.1
Caryophyllene oxide	1581	1567	13.4	8.2	6.5	5.4
4(14)-Salvialen-1-one	1589	1578	Tr.	3.0	-	-
1- <i>epi</i> -Cubenol	1625	1608	-	1.3	-	-
τ -Cadinol	1635	1640	2.8	3.3	0.3	3.4
τ -Muurolol	1641	1626	2.4	2.9	0.2	2.0
α -Cadinol	1656	1653	3.3	3.7	-	-
Valeranone	1672	1668	4.8	-	0.2	8.7
β -Bisabolol	1673	1674	-	0.7	-	-
α -Bisabolol	1682	1699	-	-	0.2	0.1
Bisabol-1-one	1713	1754	-	0.2	-	-
Isopimara-8,15-diene	1908	1906	-	-	0.8	-
(<i>Z</i>)-Nuciferyl propionate	1910	1901	0.2	-	-	-
(<i>Z</i>)-Nuciferyl isobutyrate	1935	1945	5.5	-	-	14.0
Manool oxide	2007	1998	-	-	0.6	-
(<i>Z</i>)-Nuciferyl isovalerate	2025	2058	0.8	-	-	-
(<i>Z</i>)-Nuciferyl 2-methylbutyrate	2030	2074	0.3	-	-	-
Kaurene	2036	2006	-	-	1.5	-
Manool	2055	2087	-	-	1.6	-
<i>ar</i> -Abietatriene	2058	2039	-	-	45.5	-
(<i>E</i>)-Nuciferyl isovalerate	2075	2091	0.2	-	-	-
(<i>E</i>)-Nuciferyl 2-methylbutyrate	2081	2101	0.2	-	-	-
<i>n</i> -Tricosane	2300	2301	0.2	-	-	-
<i>n</i> -Pentacosane	2500	2500	Tr.	-	-	-
Total			99.5	98.6	96.3	99.6
Yield (% w/w)			0.024	0.025	0.037	0.035
Grouped components						
Terpenoids						
Monoterpene hydrocarbons			1.3	5.1	9.2	2.8
Oxygenated monoterpenes			11.7	0.9	23.6	2.9
Sesquiterpene hydrocarbons			35.4	69.3	4.3	48.0
Oxygenated sesquiterpenes			40.2	23.3	8.3	31.9
Diterpenoids			-	-	50.0	-
Terpene esters			7.2	-	0.9	14.0
Others			5.7	-	0.9	0.6

Components listed in order of elution from an HP-5MS column.

RI^a: Experimental retention indices on the HP-5MS column; RI^b: Experimental retention indices on the SPB 1 column.

-: not detected; Tr.: trace amount (<0.1%).

TABLE 2. Antimicrobial Activity^a of the *Stachys* sp. Oils and Corresponding Pure Constituents

Microorganism	<i>E. coli</i>		<i>S. enteritidis</i>		<i>P. aeruginosa</i>		<i>K. pneumoniae</i>		<i>S. aureus</i>		<i>C. albicans</i>		<i>A. niger</i>	
	Sample (1:10 dilution)													
Sample	C ^b	S ^c	C	S	C	S	C	S	C	S	C	S	C	S
<i>S. plumosa</i>	17	-	-	18	-	20	-	13	-	19	-	16	-	21
<i>S. germanica</i>	20	-	14	16	-	18	-	14	-	-	20	22	-	-
<i>S. scardica</i>	18	30	18	32	15	-	-	14	17	31	14	17	14	18
<i>S. iva</i>	16	18	16	20	17	21	-	16	14	18	-	19	-	18

TABLE 2. (continued)

Microorganism	<i>E. coli</i>	<i>S. enteritidis</i>	<i>P. aeruginosa</i>	<i>K. pneumoniae</i>	<i>S. aureus</i>	<i>C. albicans</i>	<i>A. niger</i>							
Standard (1:10 dilution)														
β -Caryophyllene	16	-	30	-	14	18	-	-	14	-	-	-	-	
Caryophyllene oxide	8	26	25	-	20	24	21	-	20	-	-	21	21	-
Camphor	38	-	28	32	18	31	21	29	29	-	22	-	29	-
1,8-Cineole	-	18	-	20	-	17	-	-	-	17	-	18	-	18
α -Pinene	-	-	-	-	-	-	-	-	-	-	-	-	-	-
β -Pinene	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Linalool	37	-	34	-	33	-	27	-	24	-	40	-	34	-
<i>cis/trans</i> -Linalool oxide (furanoid)	25	-	25	-	-	18	-	16	-	17	-	21	19	-
Bornyl acetate	-	36	-	35	-	22	-	-	-	30	-	34	-	-
Antibiotics														
Cefquinome	29	32	24	28	26	28	18	20	23	26	14	17	16	-
Tetracycline	30	32	26	28	26	28	27	28	25	28	22	26	22	28
Unipen	-	15	-	17	-	18	-	14	-	15	-	15	-	17
Nystatin	Nt.		Nt.		Nt.		Nt.		Nt.		18	-	17	-

^aAntimicrobial activities represented as the inhibition zones, in mm, including the disk diameter, 6 mm. Values for static (S) zones represent the extra millimetres around the cidal (C) zone (or the sole disk if no cidal activity) in which the growth of microorganisms was inhibited but in which the microorganisms were not killed;

^bBacteri- and fungicidal zones;

^cBacteri- and fungistatic zones;

-: no activity observed, Nt.: not tested.

REFERENCES

1. H. D. Skaltsa, D. M. Lazari, I. B. Chinou, and A. E. Loukis, *Planta Med.*, **65**, 255 (1999).
2. E. Sezik and A. Bascaran, *J. Fac. Pharm.*, **21**, 98 (1985).
3. A. Zargari, *Medicinal Plants*, Vol. **4**, Teheran University Publications, Teheran, 1990.
4. N. Maleki, A. Garjani, H. Nazemiyah, N. Nilfouroushan, A. T. Eftekhar Sadat, Z. Allameh, and N. Hasannia, *J. Ethnopharmacol.*, **75**, 213 (2001).
5. N. Radulovic, J. Lazarevic, N. Ristic, and R. Palic, *Biochem. Syst. Ecol.*, **35**, 196 (2007).
6. NCCLS (National Committee for Clinical Laboratory Standards). *Performance Standards for Antimicrobial Disk Susceptibility Test*, 6th ed., Approved standard: P. A. Wayne, M100-S9 (1997).