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Chemical Compositions of the Essential Oils of Different Stages of the Growth of *Stachys inflata* Benth. from Iran

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The chemical compositions of a hydrodistilled oils of different stages of growth *Stachys inflata* Benth. Growing wild in Iran was examined by GC and GC/MS. The major components of the oils were germacrene-D (pre-f. 30.3 %, f. 12.38 % and post-f. 32.9 %), bicyclogermacrene (pre-f 3.49 % f. 11.26 %, post-f. 7.3 %), α-pinene (pre-f. 18.48 % f. 9.11 %, post-f. 2.6 %), limonene (pre-f. 16.41 % f. 11.14, post-f. 15.6 %), α-pinene (pre-f. 9.756 % f. 4.54 % post-f. 2.5 %) and spathulenol (pre-f. 3.2 %, f. 13.56 %, post-f. 7.5 %) (f - flowering).

Key Words: *Stachys inflata*, Essential oil, Geracrene-D, GC/MS analysis.

INTRODUCTION

The genus *Stachys* is one of the largest genera of the Labiatae with a worldwide distribution. About 300 *Stachys* species are reported[1] of which 34 ones are found in the flora of Iran. The Persian name of the plant is Sonboleiye Arghavani[2]. Some of the *Stachys* species are used as medicinal plants around the world[3-5]. *Stachys inflata* Benth. is one of the most distributed species in the country 6 which have been used in Iranian folk medicine in rheumatic and other inflammatory disorders[6].

The last study on the hydroalcoholic extract of aerial parts of *S. inflata* showed significant antiinflammatory activities on rats[7]. There are some reports on the phytochemical analysis of *Stachys* species, but only a very small number of these species have been studied for their essential oils[8-10].

Literature survey has shown that aerial part of *S. inflata* has been previously investigated for its essential oil[11]. In this research, the essential oils of whole plant at different stages of growth of *S. inflata* are investigated.

Essential oils of *S. inflata* yielded 0.15 % pre-flowering, 0.18 % post flowering and 0.2 % flowering (v/w) of a pale green oils with an aromatic odours. The list of compounds identified in the oils samples are presented in Table-1 with their percentage of compositions.

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EXPERIMENTAL

Stachys inflata Benth. was collected from around Borejerd in Lorestan state (South west of Iran) in march (pre-flowering), April (flowering) and June (post flowering) 2005 at an altitude of 1420 m. A voucher specimen has been deposited in the Herbarium of the College of Agriculture, Lorestan University, Khoramabad Iran.

The volatile oil of the whole plant of different stages of *S. inflata* were obtained by hydrodistillation using a Clevenger-type apparatus and dried over sodium sulfate.

The oils were analyzed by GC/MS using a gas chromatography analysis. GC analysis of the oil was conducted using a Varian CP-3800 instrument equipped with a DB-1 fused silica column (60 m × 0.25 mm i.d., film thickness 0.25 μm). Nitrogen was used as the carrier gas at the constant flow of 1.1 mL/min. The oven temperature was held at 60ºC for 1 min, then programmed to 250ºC at a rate of 4ºC/min, and then held for 10 min. The injector and detector (FID) temperatures were kept at 250 and 280ºC, respectively. GC-MS analysis was carried out on a Thermoquest-Finnigan Trace GC-MS instrument equipped with a DB-1 fused silica column (60 m × 0.25 mm i.d., film thickness (0.25 μm). The oven temperature was raised from 60 to 250ºC at a rate of 5ºC/min and then held at 250ºC for 10 min; transfer line temperature was 250ºC. In this case, the oven temperature was raised from 40 to 250ºC at a rate of 4ºC/min, then held at 250ºC for 10 min with the transfer line temperature adjusted at 250ºC. Helium was used as the carrier gas at a flow rate of 1.1 mL/min; split ratio was, 1/50. The quadrupole mass spectrometer was scanned over the 45-465 amu with an ionizing voltage of 70 eV and an ionization current of 150 μA. The constituents of the oil were identified by calculation of their retention indices under temperature-programmed conditions for *n*-alkenes (C_6-C_{24}) and the oil on DB-1 and DB-Wax columns under the same conditions. Identification of individual compounds was made by comparison of their mass spectra with those of the internal reference mass spectra library (Wiley 7.0) or with authentic compounds and confirmed by comparison of their retention indices with authentic compounds or with those of reported in the literature[12-14]. Quantitative data was obtained from FID area percentages without the use of correction factors.

RESULTS AND DISCUSSION

Essential oil of *S. inflata* yielded 0.15 % pre-flowering, 0.2 % flowering and 0.18 % post-flowering (v/w) of a pale green oil with an aromatic odours. In the perflowering, flowering and post flowering respectively 35, 29, 34 compounds were identified. The list of compounds identified in the oils samples are presented in Table-1 with their percentage compositions.

TABLE-1
COMPOSITION OF THE ESSENTIAL OIL OF DIFFERENT STAGE OF GROWTH OF *Stachys inflata* Benth. FROM IRAN

Compounds	Tn	RI	1 (%)	2 (%)	3 (%)
α-Thujene	9.07	924	0.84	0.3	0.50
α-Pinene	9.28	933	18.48	2.6	12.11
Sabinene	10.10	968	2.46	0.7	1.42
β-Pinene	10.29	975	9.75	2.5	4.54
Myrcene	10.41	980	1.17	2.8	1.19
α-Phellandrene	10.89	1000	0.56	0.4	0.36
Δ-3-Carene	11.10	1008	0.63	3.0	1.40
o-Cymene	11.20	1011	0.40	0.4	–
p-Cymene	11.27	1014	0.49	1.3	0.49
Limonene	11.50	1024	16.41	15.6	13.14
cis-Ocimene	11.90	1036	0.60	0.6	–
γ-Terpinene	12.23	1050	0.24	0.8	0.44
α-Terpinolene	13.10	1081	0.74	1.6	1.93
Allo-ocimene	14.08	1117	0.18	–	–
4-Terpineol	15.40	1165	0.16	0.3	0.46
Linalyl propionate	15.69	1175	0.34	0.5	0.69
Bicycloelemene	20.00	1336	0.27	0.6	0.85
α-Copaene	21.17	1379	0.41	0.7	0.45
β-Elemene	21.40	1389	1.70	1.7	–
β-Caryophyllene	22.28	1423	0.80	1.7	0.73
Aromadendrene	23.00	1453	0.30	0.3	–
Germacrene-D	23.70	1482	30.30	32.9	15.38
Bicyclogermacrene	24.10	1497	3.49	7.3	14.26
Δ-Cadinene	24.60	1518	0.33	0.4	0.60
Aromadendrene oxide	25.70	1564	0.15	0.4	0.37
Spathulenol	25.89	1571	3.20	7.5	13.56
Caryophyllene oxide	26.10	1580	0.23	0.3	0.29
Salvial-4-(14)-en-1-one	26.20	1588	0.16	0.3	0.81
Leden oxide	26.53	1600	0.24	0.6	0.89
Tau-cadinol	27.23	1630	0.27	0.4	0.54
α-Cadinol	27.51	1644	0.55	0.9	0.99
Neoclonenoxid alcohol	28.20	1673	0.91	2.0	1.42
Hexadecane	31.50	1822	0.45	0.5	3.00
Eicosane	35.60	2021	0.80	3.0	–
Phytol	36.50	2104	2.62	3.9	1.46

Tn: Retention Time, RI: Kovats Constant, 1 pre-flowering 2: flowering
3: post- flowering

As can be seen in Table-1, the major components of the oil are germacrene-D (pre-f. 30.3 %, post-f. 32.9 % and f. 12.38 %), bicyclogermacrene (pre-f. 3.49 % post-f. 7.3 % f. 11.26 %), β-pinene (pre-f. 19.48 %, post-f. 2.6 % f 9.11 %), limonene(pre-f. 16.41 % post-f. 15.6 % f. 11.14 %), α-pinene (pre-f. 9.756 % post-f. 2.5 % f. 4.54 %) and spathulenol (pre-f. 3.2 % post-f.7.5 % f. 13.56 %). The oils of *S. inflata* consisted mainly of monoterpene hydrocarbons (pre-f. 51.55 %, f. 32.6 %, post-f. 37.49 %), oxygenated monoterpenes (pre-f. 0.16 %, f. 0.3 %, post-f. 0.46 %), sesquiterpene hydrocarbons (pre-f. 37.6 %, f. 45.6 %, post-f. 32.27 %) oxygenated sesquiterpenes (pre-f. 4.64 %, f. 10.1 %, 16.64), oxygenet diterpene (pre-f. 2.62 %, f.3.9 %, post-f. 1.46 %) and one aliphatic aldehyde (pre-f. 30.3 %, post-f. 12.38 % f. 32.9 %). Germacrene-D, the dominant compound of the essential oil of *S. inflata*, was reported as the major components of the volatile oils of *S. laxa* Boiss.[15] and *S. obliqua* L.[16].

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