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Short communication

Analysis by gas chromatography–mass spectrometry of the volatile components of *Ageratina adenophora* Spreng., growing in the Canary Islands

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Abstract

The essential oil of *Ageratina adenophora* Spreng., growing in the Canary Islands was analyzed by GC–MS. A total of 78 volatile compounds was identified and *p*-cymene (11.6%) was the major component in the oil. The sesquiterpene fraction (44.3%) was higher than the monoterpene one (32.1%).

Keywords: *Ageratina adenophora*; Essential oils; Plant materials; Volatile organic compounds; Cymenes

1. Introduction

Ageratina adenophora Spreng., is a native species of Mexico and Costa Rica although actually it is naturalized and distributed practically all around the world (Asian, Australian, African and American continents). In fact this species is a great threat to many parts of the world, including the Canary Island's archipelago, causing problems in forest and plantation corps [1,2]. *Ageratina* L. (=*Eupatorium* L.) genus belongs to the Asteraceae family and it has been widely studied. A review of the chemical constituents of different species of this genus showed flavonids, glycosides, sesquiterpene lactones, coumarins, alkaloids and terpenes as principal chemical categories [3–5].

Although the biomass of this species has been successfully used in the cultivation of *Pleurotus sajor-caju* (Fr.) Singer [1], this species has been reported for its allelopathic effects [6]. Few studies have been done about the toxicity of this plant. A hepatotoxic compound (9-oxo-10,11-dehydroagerophorone) was identified from the crude extract of *A. adenophora*, this component promotes a chronic equine respiratory disease known as ''Numinbah horse sickness'' and liver lesions [7]. This compound had been previously described from the aerial parts of this species although its toxic effects have not been tested [8]. The authors also found β-farnesene, germacrene D, bisabolene, caryophyllene and four more cadinene derivatives were also reported [8]. Other compounds identified from the stems and leaves of this species were found to be 9-oxo-ageraphorone, 9-β-hydroxy-ageraphorone, epifriedelinol, stigmasterol, octacosanoic acid, β-daucosterol, *o*-hydroxycinnamic acid, ferulic acid, cafeic

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acid and 2-isopropenyl-5-acetyl-6-hydroxybenzofuran acetate [9].

The chemical composition of other species of this genus has also been reported. The extracts of *Eupatorium rufescens* Lund ex de Candolle with $CHCl_3$ yielded two sesquiterpene peroxides identified as zingiberene-2,6-α-endoperoxide and zingiberene-3,6-β-endoperoxide [10]. The oil of *Eupatorium stoechadosmum* Hance from Vietnam showed thymohydroquinone dimethyl ether (73.6%), selina-4,11-diene (11.0%) and β-caryophyllene (8.9%) as main constituents [11]. The volatile components of other species from Vietnam, *Eupatorium coelestinum* L., were characterized by methyl chavicol (9.7–45.2%), bornyl acetate (6.2–12.6%), camphene (0.3–11.9%), *cis*-cadin-4-en-7-ol (5.2–10.9%) as principal compounds [12]. Recently, the essential oils of two other *Eupatorium* species from Brazil have been reported. The principal compounds of *E. triplinerve* Vahl were found to be β-caryophyllene (19.7%) and 2,5-dimethoxy *p*-cymene (69.7%) while β-caryophyllene (46.8%) was the main compound in *E. paniculatum* Poepp. et Endl. [13].

In the present work, we report on the chemical composition of the essential oil from the aerial parts of *A. adenophora* Spreng. growing in the Canary Islands by gas chromatography–mass spectrometry. As far as we know this is the first report on the essential oil composition of this species.

2. Experimental

2.1. Plant material

The aerial parts of *A. adenophora* (common name: eupatorio, jediondo, jaragán, flor espuma or hierba negra) were gathered at flowering in Los Berrazales, Agaete (Canary Islands, Spain), 4 April 2000. A voucher specimen MAB-76450 has been deposited at the herbarium of the Biology Faculty in the Complutense University of Madrid, Spain.

2.2. Isolation procedure

Plant material was steam distilled in a Clevenger-type apparatus for 8 h, according to the method recommended by the Spanish Pharmacopoeia [14]. The essential oil was dried over anhydrous magnesium sulphate and stored at 4 °C in the dark. The yield was 0.76% based on dry mass of sample.

2.3. Gas chromatography (GC)

A Varian GC 3300 system was used for GC analysis, fitted with a fused methylsilicone DB-1 column (50 m×0.25 mm I.D., 0.25 μm film thickness). Oven temperature was programmed from 80 to 225 °C at 4 °C/min. Injection was performed at 280 °C in the split ratio 1:100; 0.1 μl of sample was injected. A flow of 1.5 ml/min carrier gas (N_2) was used. Flame ionization detection (FID) was performed at 300 °C.

2.4. Gas chromatography–mass spectrometry (GC–MS)

Analyses were carried out in a Fison 8000 gas chromatograph fitted with a fused methylsilicone SE-30 and BP-5 column (50 m×0.25 mm I.D., 0.25 μm film thickness), coupled to a MD 800 mass detector. Column temperature was programmed from 70 to 240 °C at 4 °C/min. Injection was performed at 250 °C. Helium was used as carrier gas (0.5 ml/min). Mass spectra were recorded in the scan mode at 70 eV (35–350 U); 0.5 μl of the sample in pentane, 1 mg/ml, was injected by the splitless technique (1/100).

2.5. Qualitative and quantitative analyses

Most constituents were identified by comparison of their GC retention indices with those of authentic standards available in the author's laboratory or with GC data previously published [15–20]. Identification was confirmed when possible by comparison of their mass spectra with those stored in the MS database (National Institute of Standards and Technology, NIST, and Wiley libraries) and with mass spectra literature data [15–20]. Component relative concentrations were obtained directly from GC peak areas.

Table 1
Percentage composition of the essential oil from the aerial parts of *A. adenophora* Spreng.

Compound	*I*	%	Method
α-Pinene	926	0.2	GC^1–MS
Camphene	941	4.8	GC^1–MS
Verbenene	953	t	GC^2–MS
β-Pinene	967	0.3	GC^1–MS
3-*p*-Menthene	969	t	GC^2–MS
Dehydro-1,8-cineole	972	0.1	GC^2–MS
δ-2-Carene	991	5.0	GC^2–MS
α-Phellandrene	992	5.7	GC^1–MS
p-Cymene	1006	11.6	GC^1–MS
Limonene	1014	0.6	GC^1–MS
(*Z*)-β-Ocimene	1023	0.3	GC^1–MS
Artemisia ketone	1045	t	GC^2–MS
γ-Terpinene	1046	t	GC^1–MS
cis-Sabinene hydrate	1051	t	GC^2–MS
m-Cymenene	1065	t	GC^2–MS
Terpinolene	1068	t	GC^2–MS
Linalool	1072	0.8	GC^2–MS
1,3,8-*p*-Menthatriene	1089	t	GC^2–MS
cis-Rose oxide	1093	0.1	GC^2–MS
cis-*p*-Menth-2-en-1-ol	1107	t	GC^2–MS
cis-Limonene oxide	1120	t	GC^2–MS
trans-Sabinol	1121	t	GC^2–MS
Camphene hydrate	1132	t	GC^2–MS
Borneol	1147	0.2	GC^1–MS
m-Cymen-8-ol	1155	t	GC^1–MS
p-Mentha-1,5-dien-8-ol	1165	1.5	GC^1–MS
α-Terpineol	1180	t	GC^1–MS
Methylchavicol	1092	t	GC^2–MS
Thymyl methyl ether	1200	0.1	GC^2–MS
Carvacryl methyl ether	1217	0.1	GC^2–MS
Geraniol	1228	t	GC^2–MS
p-Cymen-7-ol	1252	t	GC^2–MS
Thymol	1255	0.2	GC^1–MS
Carvacrol	1257	t	GC^1–MS
endo-Bornyl acetate	1262	4.4	GC^1–MS
Pinocarvyl acetate	1299	0.1	GC^2–MS
trans-Carvyl acetate	1331	0.1	GC^1–MS
δ-Elemene	1335	0.3	GC^2–MS
Neryl acetate	1340	t	GC^1–MS
β-Terpenyl acetate	1342	0.2	GC^2–MS
α-Ylangene	1365	t	GC^2–MS
1,7-Di-epi-α-cedrene	1382	t	GC^2–MS
α-Cedrene	1394	0.1	GC^1–MS
α-*cis*-Bergamotene	1398	0.1	GC^2–MS
β-Cedrene	1401	t	GC^1–MS
(*E*)-Caryophyllene	1409	4.1	GC^1–MS
β-Gurjunene	1417	t	GC^2–MS
α-*trans*-Bergamotene	1420	3.1	GC^2–MS
(*Z*)-β-Farnesene	1428	t	GC^2–MS
α-neo-Clovene	1430	t	GC^2–MS
(*E*)-β-Farnesene	1431	2.1	GC^2–MS

Table 1. Continued

Compound	*I*	%	Method
α-Humulene	1435	t	GC^1–MS
n.i. 1	1444	1.2	—
Geranyl *n*-propanoate	1451	0.8	GC^2–MS
γ-Curcumene	1460	5.0	GC^2–MS
Germacrene D	1464	3.0	GC^1–MS
n.i. 2	1468	4.1	–
α-Zingiberene	1473	0.6	GC^2–MS
Bicyclogermacrene	1483	1.4	GC^2–MS
β-Bisabolene	1488	3.2	GC^2–MS
(*Z*)-γ-Bisabolene	1494	t	GC^2–MS
β-Sesquiphellandrene	1503	2.2	GC^2–MS
β-Vetivenene	1507	0.6	GC^2–MS
6,11-oxido-Acor-4-ene	1511	0.1	GC^2–MS
Cadina-1,4-diene	1516	0.1	GC^2–MS
1,1,4,8-tetramethyl-*cis*,*cis*,*cis*-4,7,10-cycloundecatriene	1519	0.5	GC^2–MS
n.i. 3	1525	0.8	–
(*Z*)-Nerolidol	1535	0.3	GC^1–MS
Geranyl isovalerate	1542	0.8	GC^2–MS
Geranyl *n*-butyrate	1549	0.3	GC^2–MS
Spathulenol	1557	0.4	GC^1–MS
ar-Turmerol	1560	t	GC^2–MS
Caryophyllene oxide	1563	0.5	GC^1–MS
Globulol	1567	0.2	GC^2–MS
Viridiflorol	1575	0.4	GC^2–MS
10-epi-γ-Eudesmol	1596	0.8	GC^2–MS
γ-Eudesmol	1602	1.2	GC^2–MS
epi-α-Cadinol	1612	7.4	GC^2–MS
epi-α-Muurolol	1615	t	GC^2–MS
n.i. 4	1629	1.2	–
n.i. 5	1644	0.9	–
epi-α-Bisabolol	1655	4.7	GC^2–MS
n.i. 6	1666	3.4	–
n.i. 7	1692	0.8	–

t, trace (<0.1%); *I*, retention index according to *n*-alkanes on DB-1 column; MS, mass spectrum; GC^1, retention index according to authentic standards; GC^2, Kováts index according to literature; n.i., not identified; n.i. 1: $I=1444$, *m*/*z* (rel.int.): 204[M^+](10), 148(100), 133(40), 43(23), 105(22), 119(21), 189(15), 175(13), 93(12), 91(10); n.i. 2: $I=1468$, 204[M^+](5), 69(100), 93(85), 133(52), 120(33), 79(25), 109(23), 41(19), 55(13), 121(10), 147(9), 189(8); n.i. 3: $I=1525$, 202[M^+](3), 146(100), 131(71), 187(42), 105(40), 145(35), 91(30), 133(29), 159 (28), 202(28), 147(25), 93(22), 79(17); n.i. 4: $I=1629$, 236[M^+](6), 234(100), 137(98), 119(56), 109(48), 219(35), 91 (30), 136(30), 150(30), 206(25), 83(22), 43(20); n.i. 5: $I=1644$, 218[M^+](5), 119(100), 175(85), 161(60), 105(60), 218(57), 91(53), 69(48), 107(47), 147(47), 133(41), 93(93), 43(33); n.i. 6: $I=1666$, 218[M^+](3), 218(100), 161(98), 203(78), 133(73), 105(46), 147(37), 175(30), 91(29), 119(28), 185(20), 79(16), 41(15), 220(8); n.i. 7: $I=1692$, 218[M^+](35), 143(100), 105(98), 91(92), 185(90), 109(80), 119(75), 145(73), 79(70), 129(65), 93(68), 157(66), 171(64), 200(45), 55(27), 41(23), 67(20).

3. Results and discussion

The identified compounds of the oil from the aerial parts of *A. adenophora* Spreng., their retention indices and percentage composition are given in Table 1 where the components are listed in order of their elution on the DB-1 column.

The major compound of the essential oil studied was found to be *p*-cymene (11.6%). Other representative components of the oil were identified as epi-α-cadinol (7.4%), α-phellandrene (5.7%), δ-2-carene (5.5%), γ-curcumene (5.0%), camphene (4.8%), epi-α-bisabolol (4.7%), endo-bornyl acetate (4.4%), (*E*)-caryophillene (4.1%), β-bisabolene

(3.2%), α-*trans*-bergamotene (3.1%), germacrene D (3.0%), β-sesquiphellandrene (2.2%) and (*E*)-β-farnesene (2.1%). The complete list of compounds identified appears in Table 1. Although some compounds could not be identified, their mass spectra fragmentation are given in Table 1.

The total amount of sesquiterpene fraction (44.3%) was higher than the monoterpene one (33.1%). In comparison with other oils of this genus previously reported [11–13], only the oil of *E. coelesticum* L. showed monoterpene as the dominant fraction although all the compounds of this oil could not be identified. According to our results and those mentioned above, the oils of *Ageratina* (=*Eupatorium*) are richer in sesquiterpenes than in monoterpene compounds. We have not detected the compounds previously described for this species, possibly because we could not extract them by steam distillation.

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