Composition of commercial Cape chamomile oil (*Eriocephalus punctulatus / Eriocephalus tenuifolius*)

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ABSTRACT

Cape chamomile oil obtained from the herb *Eriocephalus punctulatus* DC (Asteraceae) was analysed by means of GC and GC-MS. It was proven to consist of about 50 aliphatic esters together amounting to more than 50% of the oil. 2-Methylbutyl 2-methylpropanoate, 2-methylbutyl 2-methylbutanoate, 2-methylpropyl 2-methylpropanoate, and 7-methyl-2octyl acetate were the main components. In the terpenoid portion (about 37%) linalyl acetate and a-pinene are the major compounds. In the higher boiling fraction the artedouglasia oxides A-D, several davanones and laciniata furanones, oxygenated sesquiterpenes previously found in *Artemisia* species, could be detected in amounts of 5.9%, 0.4% and 1.4%, respectively. Commercial samples of different years showed almost identical compositions.

KEY WORDS

Eriocephalus punctulatus, Asteraceae, essential oil, commercial Cape chamomile oil, 2-methylbutyl 2-methylpropanoate, artedouglasia oxide, davanone, laciniata furanone

Introduction

Eriocephalus punctulatus DC (Asteraceae), "Cape chamomile", is an endemic plant growing on the north-east slopes of the Drakensberge in the Province Freestate (South Africa). It is a white flowering small shrub with fleshy, gland dotted leaves. Although in folk medicine several species of *Eriocephalus* are traditionally used as diaphoretics and diuretics[1] there have been few publications concerning special uses of *E. punctulatus*; local people appreciate its benefit in the treatment of stomach diseases or for the fumigation of huts[2].

Twenty years ago a practically feasible method of vegetative propagation was developed and a few hundred genetically identical plants have been produced for plantings in the Amatola mountains, Ciskei.[3] Nowadays commercial Cape chamomile oil is produced from cultivars in the Cape Province (South Africa). Due to its pleasant odour the oil has been used as a fragrance in cosmetics and toiletries; it is increasingly employed in aromatherapy.

The blue colour of the commercial Cape chamomile oil is striking and one associates it automatically with the European chamomile oil from *Matricaria recutita* L. (Asteraceae). In both oils the blue colour is caused by azulene derivatives in the oils which are formed by a decomposition of proazulenes during steam distillation. Aside from the blue colour these two oils have nothing in common.

To this date only few components have been identified in Cape chamomile oil. 2-Methylbutyl isobutyrate, 2-methylpropyl isobutyrate, *p*-cymene, a-pinene, 2-methylbutyl

isovalerate, 3-methylbutyl isobutyrate were reported to be the main components besides eight minor components.[6, 7] In Table I the previously published components are labelled with asterisks.

Table 1. Constituents of commercial Cape chamomile oil (*Eriocephalus punctulatus* DC)

Compound name	**RI**	**Percentage**
2-Methyl-1-butanol *	801	0.4
2-Methylpropyl acetate	810	t
3-Methyl-2-butyl acetate	850	t
Propyl 2-methylpropanoate	866	t
2-Methylpropyl propanoate	873	t
3-Methylbutyl acetate	878	t
2-Methylbutyl acetate	879	0.1
2-Methylpropyl 2-methylpropanoate *	909	5.3
Ethyl 3-methyl-2-butenoate	911	t
Tricyclene	917	t
α-Thujene	922	t
2-Methylpropyl methacrylate	926	0.1
α-Pinene *	932	1.9
Camphene	942	0.7
Verbenene	945	t
3-Methylbutyl propanoate	952	t
2-Methylbutyl propanoate	955	0.2
Sabinene	963	0.1
β-Pinene *	966	0.3
(*E*,*Z*)-1,2-Diethylidenecyclopentane	969	t
Dehydroxylinalool oxide A	977	0.5
β-Myrcene	979	0.1
2-Methylpropyl 2-methylbutanoate *	986	1.4
2-Methylpropyl 3-methylbutanoate *	989	1.2
3-Methylbutyl 2-methylpropanoate *	995	2.6
2-Methylbutyl 2-methylpropanoate *	1002	21.2
α-Terpinene	1005	0.4
p-Cymene *	1007	2.0
1,8-Cineole	1013	0.2
Limonene *	1015	0.7
3-Methylbutyl methacrylate	1019	t
2-Methylbutyl methacrylate	1021	0.2
2-Heptyl acetate	1022	0.1
cis-Arbusculone	1026	t

2-Methylpropyl angelate	1030	1.6
(*E*)-β-Ocimene	1033	t
2-Methylbutyl butanoate	1037	0.1
trans-Arbusculone	1039	t
γ-Terpinene	1043	1.0
trans-Sabinene hydrate / 7-Methyl-2-octanol**	1046	0.7
Butyl angelate	1055	0.8
2-Nonanone / *p*-Cymenene **	1066	0.1
Butyl tiglate	1068	0.1
Terpinolene *	1072	0.3
cis-Sabinene hydrate	1075	0.1
Linalool	1077	0.5
3-Methylbutyl 2-methylbutanoate	1082	1.3
2-Methylbutyl 2-methylbutanoate	1087	5.6
2-Methylbutyl 3-methylbutanoate *	1089	1.1
Hexyl 2-methylpropanoate	1092	t
2,2,3-Trimethyl-3-cyclopentene-1-acetaldehyde	1097	t
cis-*p*-Menth-2-en-1-ol	1099	0.2
Camphor	1111	1.4
trans-Pinocarveol / *trans*-*p*-Menth-2-en-1-ol **	1116	0.1
2-Octyl acetate	1121	0.4
Menthone	1125	0.1
3-Methylbutyl angelate	1128	0.3
2-Methylbutyl angelate	1134	3.5
Borneol	1143	1.4
Terpinen-4-ol *	1156	1.7
Pentyl angelate	1162	1.3
α-Terpineol *	1166	0.2
Pentyl tiglate	1174	0.2
7-Methyl-2-octyl acetate	1187	4.5
cis-Carveol	1200	0.0
Nerol	1207	0.1
Pulegone	1209	0.1
2-Nonyl acetate	1221	0.6
Geraniol	1234	t
Linalyl acetate	1241	4.4
Bornyl acetate	1266	0.3
Thymol	1267	t
Diosphenol	1273	t
Carvacrol	1277	t

trans-Sabinene hydrate acetate	1281	t
(E,E)-2,4-Decadienal	1288	t
Myrtenyl acetate	1304	t
Eugenol	1327	t
α-Terpinyl acetate	1332	t
Neryl acetate	1344	0.6
α-Longipinene	1347	0.2
Geranyl acetate	1362	0.5
2-Phenylethyl 2-methylpropanoate	1368	0.5
Longicyclene	1370	0.3
α-Copaene	1374	1.4
Modhephene	1376	t
α-Isocomene	1383	0.1
β-Elemene / α-Funebrene** / Lyratyl acetate **	1387	0.4
Longifolene	1401	0.2
Unknown MW=220 (A) / 2-Methylbutyl benzoate **	1409	0.1
β-Caryophyllene	1414	0.6
β-Copaene	1424	t
α-Humulene	1447	0.1
Unknown MW=206 (B) / *allo*-Aromadendrene **	1452	0.4
Unknown MW=202 (C) / 3-Methylbutyl phenylacetate **	1460	0.3
2-Methylbutyl phenylacetate	1464	0.7
Unknown MW=234 (D)	1469	0.1
α-Selinene	1478	0.1
Bicyclogermacrene	1489	0.2
α-Muurolene	1493	0.1
Laciniata furanone G	1498	0.2
Artedouglasia oxide C	1500	1.7
Laciniata furanone F	1503	0.4
Artedouglasia oxide A	1511	2.2
Laciniata furanone E	1514	0.3
1,4-Dimethylazulene	1517	0.3
Laciniata furanone H	1522	0.5
α-Calacorene	1526	t
Artedouglasia oxide D	1532	0.7
Isodavanone	1537	0.2
Davanone B	1542	0.2
Artedouglasia oxide B	1553	1.3
Davanone D	1557	t

Spathulenol	1561	1.1
Caryophyllene oxide	1566	0.8
β-Copaen-4-α-ol	1572	0.5
Viridiflorol	1578	0.3
Unknown MW=250 (E)	1586	0.2
Unknown MW=222 (F)	1589	0.1
Unknown MW=218 (G)	1598	0.1
Unknown MW=222 (H)	1608	0.6
Caryophylla-3(15),7(14)-dien-6-ol	1614	0.9
Chamazulene	1700	0.2
		94.4

*previously mentioned / ** co-eluted / t = < 0.05%

Mass spectra: EI-Mode, 70eV, (m/z)

(A) 220(28), 177(64), 149(100), 135(47), 107(37), 93(47), 91(37), 43(66)
(B) 206(28), 96(100), 82(32), 81(67), 68(88), 67(47), 55(32), 41(34)
(C) 202(8), 159(75), 115(27), 97(31), 71(39), 70(26), 43(200), 41(20)
(D) 234(13), 161(100), 109(83), 105(71), 93(69), 91(64), 43(63), 41(60)
(E) 250(3), 177(70), 138(68), 124(93), 96(74), 81(84), 43(100), 41(69)
(F) 222(5), 109(67), 105(51), 95(58), 93(64), 81(51), 43(100), 41(68)
(G) 218(8), 105(66), 95(72), 93(100), 91(88), 67(70), 43(73), 41(88)
(H) 222(6), 204(55), 189(32), 161(100), 121(77), 105(59), 95(46), 43(98)

Results and Discussion

GC analysis of 5 commercial samples produced from harvests in 1997- 2001 revealed the Cape chamomile oil to consist of about 220 compounds (Figure 1). 104 of them were elucidated by GC-MS analysis and by comparison of retention indices. If available co-chromatography of authentic substances was performed. The results are listed in Table I which shows the data of sample 2 produced in 1999. Besides 123 identified components, 8 additional components with percentages above 0.1% are listed whose identity remained unknown. Their ms data are given below Table 1 (A)-(H). The components listed in Table 1 cover 92-94% of each oil sample. The remaining portion of 6-8% is distributed over about 100 peaks below 0.1% each.

Aliphatic esters are the dominant group in the oils amounting to more than 50% with 2 -methylbutyl 2-methylpropanoate (21.2%) being the main component. Further important esters are 2-methylbutyl 2-methylbutanoate (5.6 %), previously described as 2-methylbutyl isovalerate3 , 6, 7, 2-methylpropyl 2-methylpropanoate (5.3%), 7-methyl-2-octyl acetate (4.5%), 3-methylbutyl 2-methylpropanoate (2,6%). Furthermore 28 minor aliphatic esters were detected among them 5 angelates and 2 tiglates (in total 7.8 %).

The terpenoid portion consisted of 27 oxygenated monoterpenes (together 11.2%) and 15 monoterpene hydrocarbons (together 7.7%) with linalyl acetate and a-pinene, respectively, as dominant compounds. Furthermore 16 sesquiterpene hydrocarbons

(together 4.5%) and 16 oxygenated sesquiterpenes (in total 11.3%) were detected. The presence of the artedouglasia oxides (A-D, together 5.9%), davanones (0.4%) and laciniata furanones (1.4%) reminds one of the sesquiterpene fractions of oils which have been isolated from *Artemisia* species[10, 11].

In Table II the 25 most abundant components (equal to or larger than 0.1%) of the oils of the five investigated samples are listed including a calculation of the percentage ranges and the mean percentages. With regard to the peak pattern as well as to the peak percentages, the table reflects a good correspondence of the commercial Cape chamomile oils produced in the years 1997-2001.

Table 2. Percentage composition of 5 commercial Cape chamomile oils (25 most abundant components)

Compound Name	Ri	**Sample**					Mean
		A	B	C	D	E	
2-Methylpropyl 2-methylpropanoate	909	12.4	5.3	11.8	11.7	10.8	10.1
α-Pinene	932	1.2	1.9	0.8	1.3	2.3	1.7
2-Methylpropyl 2-methylbutanoate	986	1.7	1.4	1.5	1.6	1.5	1.5
2-Methylpropyl 3-methylbutanoate	989	1.2	1.2	1.9	1.5	0.6	1.1
3-Methylbutyl 2-methylpropanoate	995	2.5	2.6	2.2	2.7	2.2	2.5
2-Methylbutyl 2-methylpropanoate	1002	23.0	21.2	19.9	23.1	20.0	21.5
p-Cymene	1007	1.8	2.0	1.7	1.2	1.1	1.3
2-Methylpropyl angelate	1030	3.0	1.6	1.8	1.9	1.5	1.9
γ-Terpinene	1043	1.0	1.0	0.7	1.0	1.5	1.2
3-Methylbutyl 2-methylbutanoate	1082	1.2	1.3	2.9	1.0	1.4	1.6
2-Methylbutyl 2-methylbutanoate	1087	4.6	5.6	3.4	4.4	4.3	4.4
2-Methylbutyl 3-methylbutanoate	1089	0.8	1.1	0.8	0.9	0.7	0.9
Camphor	1111	0.7	1.4	1.6	1.4	2.2	1.6
2-Methylbutyl angelate	1134	3.1	3.5	3.9	3.2	2.4	3.0
Borneol	1143	1.0	1.4	2.6	1.5	2.4	1.9
Terpinen-4-ol	1156	2.3	1.7	2.7	1.9	2.6	2.2
Pentyl angelate	1162	1.0	1.3	1.0	1.1	1.0	1.1
7-Methyl-2-octyl acetate	1187	3.8	4.5	4.6	4.1	3.2	3.8
Linalyl acetate	1241	5.7	4.4	2.6	4.2	3.2	3.9
α-Copaene	1374	1.0	1.4	0.6	0.9	1.2	1.1
Artedouglasia oxide C	1500	1.2	1.7	1.4	1.2	1.6	1.4
Artedouglasia oxide A	1511	1.6	2.2	1.8	1.5	2.2	1.8
Artedouglasia oxide B	1553	0.9	1.3	0.9	0.8	1.2	1.0
Spathulenol	1561	1.0	1.1	0.8	0.8	0.9	0.9
Caryophylla-3(15),7(14)-dien-6-ol	1614	0.5	0.9	0.4	0.5	0.5	0.6
Total		78.2	72.9	74.1	75.4	72.5	74.0

It was mentioned above that 1,4-dimethylazulene (0.3%) and chamazulene (0.2%) were proven to be artefacts formed during steam distillation. In spite of their low percentages in the oils they are responsible for the typical blue colour of the oils. Aside from colour no further chemical similarity with German chamomile oil could be found. The latter consists mainly of sesquiterpenes of the bisabolane type and other sesquiterpenoids. There is a closer correspondence to Roman chamomile oil, which is known to consist mainly of propionates, butyrates, isobutyrates, angelates, tiglates, and further aliphatic esters.

Experimental

Commercial samples of Cape chamomile oil were provided by Grassroots Natural Products, Gouda 6821, South Africa. Sample 1997(A), sample 1999 "Berger" (B), sample June 1999 (C), sample 2000 (D), sample 2001 (E). For GC and GC-MS the oils were dissolved in n-hexane (5.0%).

Gas Chromatography

A Hewlett-Packard 5890 Series II equipped with a FID (250°C) and a Phenomenex column ZB-1, 30 m x 0.25 mm, 0.25 µm film thickness was applied. The injection (1 µL) was carried out with a Hewlett-Packard 7673 automatic-sampler using the split mode (ratio 1:10) at 250°C. The carrier gas N2 was pressure controlled with a flow of 1 mL/min (45°C). The temperature program was 3°C/min from 45°C to 230°C. The relative amounts of individual compounds are based on the peak area obtained without FID response factor correction. Retention indices of the components were determined relative to *n*-alkanes.

Gas chromatography-mass spectrometry

A Hewlett-Packard 5890 Series II equipped with a Chrompack column CP-Sil 5, 30 m x 0.25 mm, 0 .25 µm film thickness, carrier gas He, flow 0.9 mL/min., temperature program: 5°C/min from 45°C to 220°C, injector (split 1:10): 220°C, coupled with a Hewlett-Packard MSD 5970 B detector; injection volume 1 µL. Mass spectra were recorded in the scan mode (solvent cut) at 70 eV over the mass range 39-310 Da.

Component Identification

The constituents of the essential oil were identified by matching their 70eV EI mass spectra and retention indices with reference libraries .

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