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VOLATILE CONSTITUENTS OF *Matricaria recutita* L. FROM ESTONIA

Anne ORAV, Tiiu KAILAS, and Kaire IVASK

Institute of Chemistry, Tallinn Technical University, Akadeemia tee 15, 12618 Tallinn, Estonia; aorav@chemnet.ee

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Abstract. The volatile constituents of *Matricaria recutita* L. cultivated in Estonia were isolated by SDE and studied by GC/FID and GC/MS. Thirty-seven components were identified. The main components were bisabolol oxide A (20–33%) and B (8–12%), bisabolon oxide A (7–14%), (E)-β-farnesene (4–13%), α-bisabolol (8–14%), chamazulene (5–7%), and en-yn-dicycloether (17–22%). The content of sesquiterpenoid compounds was high, amounting to 70% of the total oil. Variations in the essential oil composition of different chamomile samples and the yields of oil during distillation are also reported.

Key words: *Matricaria recutita* L. (*M. chamomilla* L.), Compositae, chamomile, volatile oil SDE extract, GC/FID, GC/MS.

Chamomile, *Matricaria recutita* L., is a well-known medicinal plant in folk medicine cultivated all over the world. Chamomile essential oil is widely used in pharmaceutic, cosmetic, and food industries. The pharmacological effect of chamomile is mainly connected with its essential oil for its spasmolytic, antimicrobial, and disinfective properties. The biologically active substances in chamomile essential oil are α-bisabolol, bisabolol oxides, chamazulene, and en-yn-dicycloethers [1–5].

Different essential oil isolation techniques (hydrodistillation, supercritical fluid extraction, headspace analysis) and GC, GC/MS, HPLC, and TLC analysis methods have been applied for studying the volatile constituents of chamomile by several investigators. (E)-β-Farnesene, α-bisabolol, bisabolol oxide A and B, chamazulene, and en-yn-dicycloethers were found to be the main constituents in chamomile oil [2–11].

The purpose of this work was to determine the composition of volatile essential oil from different chamomile samples harvested in Estonia, to compare

the composition of the oil from Estonian chamomile with oil of foreign origin, and to study the variation in the yields and in oil composition during hydrodistillation.

EXPERIMENTAL

Commercial dried chamomile flowerheads (*flores Chamomilae*) harvested from different places of Estonia in 1997 (samples 1, 2, and 3) were supplied. The volatile fraction was isolated from the sample (~10 g) by simultaneous steam distillation and extraction (SDE) with *n*-hexane (0.5 mL) as solvent using Marcusson type micro-apparatus. The oil yield was determined by adding *n*-tetradecane (2 μL) as internal standard into the extract.

The essential oil extracts (1–5 μL) were analysed with a Chrom-5 gas chromatograph with FID on two fused silica capillary columns (50 m × 0.20 mm i.d.) with bonded stationary phases: OV-101 (film thickness 0.5 μm) and PEG 20M (film thickness 0.25 μm). As carrier gas helium with a split ratio of about 1:150, flow rate about 1.3 mL/min for OV-101 and 1.5 mL/min for PEG 20M column was applied. The oven temperature was programmed from 50 to 250 °C (OV-101) and from 70 to 250 °C (PEG 20M) at 2 °/min. The injector temperature was 160 °C. A Hewlett-Packard Model 3390A integrator was used for data processing.

The mass-spectrometric analyses were carried out on a Hitachi M-80B gas chromatograph double focussing mass-spectrometer using the OV-1 (50 m × 0.25 mm i.d.) capillary column. The temperature program was from 70 to 280 °C at 5 °/min.

The individual components were identified according to their retention indices (RI) on two columns in comparison with RI of our RI data bank or with data presented in the literature [12, 13]. The results obtained were confirmed by GC/MS.

The quantitative composition of essential oil was calculated using the peak areas without any correction for the relative response factor. The data shown are the mean values of three injections.

RESULTS AND DISCUSSION

In order to determine the reproducibility of SDE procedures three parallel distillations with Sample 2 were produced. The variation coefficients of the concentration of the components calculated on the basis of GC analysis of these extracts did not exceed 20% except for some minor components (<0.5%).

To search for optimal distillation time for the isolation of chamomile volatiles the SDE extracts from Sample 2 were analysed by GC after 0.5, 1, 2, and 3 hours of distillation. The quantitative composition of chamomile essential oil versus

distillation time is shown in Table 1 and yields of total oil and main constituents versus time in Figs. 1 and 2. It can be seen that the total yield of oil increased during 0.5 to 2 hours of distillation more than twice (from 0.8 to 2.1 mg/g) but did not change significantly from 2 to 3 hours of distillation (Fig. 1). The percentage composition of oil extracts changed insignificantly, except mono-terpenes and other more volatile compounds, whose content decreased during 3-hour distillation about four times. The concentration of chamazulene and bisabolol oxide A increased slightly as a function of time (Table 1). The yields of the main components in chamomile oil increased from 0.5 to 2 hours of distillation. Thereafter there was only a slight increase or even a decrease (Fig. 2). The results suggest that two-hour distillation is sufficient for the isolation of chamomile essential oil by the micro-SDE method.

Table 1. Variation in the composition of the essential oil of *Matricaria recutita* L. during distillation

Component	Concentration, %[a]			
	0.5 h	1 h	2 h	3 h
6-Methyl-5-hepten-2-one	0.2	0.1	0.1	0.1
1,2,4-Trimethylbenzene	0.4	0.1	0.1	0.1
3-Octanol	0.2	0.2	0.2	< 0.05
1,8-Cineole	0.3	0.2	0.1	0.1
Artemisia ketone	0.8	0.5	0.4	0.3
Decanoic acid	2.1	1.0	1.1	1.9
(E)-β-Farnesene	4.1	3.4	3.5	3.8
γ-Muurolene + Germacrene D	0.6	0.4	0.5	0.5
α-Farnesene	0.4	0.3	0.4	0.3
Spathylenol	2.7	2.6	2.5	2.2
Bisabolol oxide B	14.2	14.3	13.4	11.8
Bisabolon oxide A	15.8	15.2	14.4	12.9
α-Bisabolol	5.6	5.8	5.9	6.2
Chamazulene	5.3	5.4	5.7	6.5
Bisabobol oxide A	31.5	34.6	34.5	37.0
En-yn-dicycloether	13.9	14.0	14.3	13.9
Total	98.1	98.1	97.1	97.6

[a] determined on OV-101 column.

The essential oils isolated with SDE during 2 hours of distillation from three samples of the commercial *flores Chamomilae* were obtained in almost the same yield of about 2 mg/g of dry weight (standard deviation 0.11). The oils showed a dark blue colour and a strong characteristic odour.

The RI of the components of three chamomile oil samples on OV-101 and PEG 20M columns, and the percentage composition of the oils determined on OV-101 are given in Table 2.

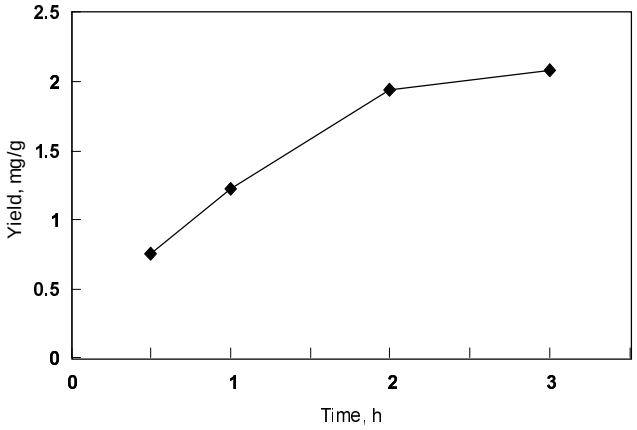

Fig. 1. Variation in the yield of the total oil of *Matricaria recutita* L. during distillation.

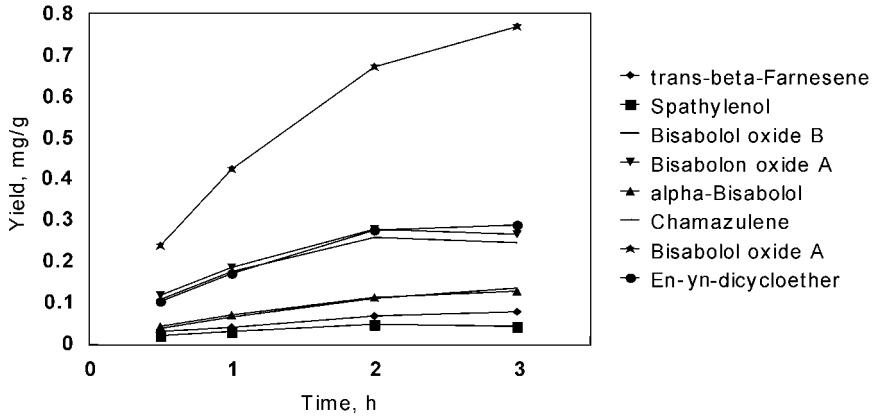

Fig. 2. Variation in the yields of the main constituents of the oil of *Matricaria recutita* L. during distillation.

RI and GC/MS data allowed identification of 37 constituents representing over 95% of the total oil. Most of the identified compounds in Estonian chamomile oil have been reported previously; however, 1,2,4-trimethylbenzene and decanoic acid were not mentioned in the literature [1–11]. En-yn-dicycloether was identified only by literature data on nonpolar columns [2, 5, 7, 10, 11] because the peak of this compound was very weak on the polar PEG 20M column.

The results obtained show that monoterpenes and oxygenated monoterpenoid compounds contribute very little (<1%) to the chamomile oil composition. Sesquiterpenes (5–16%) are mainly represented by (E)-β-farnesene (4–13%). Oxygenated sesquiterpenes are the most characteristic chamomile essential oil components as confirmed by a number of authors [1–11], and in these samples they represent 50–70% of the total oil, among which bisabolol oxide A accounts

Table 2. Identification data and percentage composition of the essential oils of *Matricaria recutita* L.

Component	RI		Concentration, %[a]		
	OV-101	PEG 20M	Sample 1	Sample 2	Sample 3
α-Pinene	926	1029	tr.	tr.	tr.
Sabinene	964	1124	tr.	0.1	0.1
6-Methyl-5-hepten-2-one[b]	967	1338	0.1	0.1	0.1
Myrcene	981	1158	tr.	0.1	0.1
1,2,4-Trimethylbenzene[b]	984	1289	tr.	0.1	0.1
3-Octanol	987	1395	0.1	0.2	0.2
p-Cymene	1010	1270	tr.	tr.	0.1
1,8-Cineole[b]	1019	1211	0.1	0.1	0.1
Limonene	1020	1204			
(E)-β-Ocimene	1038	1247	tr.	0.1	0.1
Artemisia ketone[b]	1044	1348	0.2	0.4	0.4
γ-Terpinene	1047	1243	tr.	0.1	0.1
Terpinolene	1073	1281	tr.	0.1	0.1
Terpinen-4-ol	1161	1596	tr.	0.1	tr.
α-Terpineol	1172	1688	tr.	0.1	tr.
Carvone[b]	1215	1725	tr.	0.1	0.1
Not identified	1255		0.1	0.1	0.1
γ-Elemene	1344		0.3	0.1	0.3
Decanoic acid[b]	1371	2270	2.1	1.1	1.7
β-Caryophyllene	1411	1587	0.1	0.1	0.1
(E)-β-Farnesene[b]	1445	1662	12.6	4.3	8.3
Germacrene D	1471	1700	1.7	0.6	0.5
γ-Muurolene[b]	1474	1693			
α-Farnesene[b]	1485	1742	0.9	0.4	0.3
β-Bisabolene	1494	1737	0.7	tr.	0.1
γ-Cadinene	1500	1749	0.1	tr.	tr.
σ-Cadinene	1509	1749	0.1	tr.	tr.
(E)-Nerolidol	1546	2035	0.1	0.1	0.1
Spathylenol[b]	1558	2100	3.6	2.3	3.2
Caryophyllene oxide	1562	1968	tr.	0.2	0.1
Not identified	1594		0.4	0.5	0.5
Caryophyllenol	1611	2121	0.2	0.3	0.3
T-Cadinol	1620	2156	0.2	0.2	0.2
Bisabolol oxide B[b]	1636	2115	7.9	12.4	12.3
Bisabolon oxide A[b]	1657	2158	9.2	13.6	6.7
α-Bisabolol	1670	2200	7.8	5.2	2.9
Chamazulene[b]	1702	2370	5.6	5.3	7.2
Bisabolol oxide A[b]	1730	2400	20.2	33.1	32.9
En-yn-dicycloether	1830		21.7	17.1	17.6
Not identified	1900		0.9	0.6	0.5
Total			97.0	99.3	97.5
Yield, mg/g			2.09	1.96	1.88

tr., traces (<0.05%); [a] determined on OV-101; [b] identification by GC/MS.

for 20–33%, bisabolon oxide A for 7–14%, bisabolol oxide B for 8–12%, and α-bisabolol for 3–8%. Chamazulene represents 5–7% and en-yn-dicycloether contributes 17–22% of the total area of the GC peaks.

Comparison of the composition of the studied chamomile samples showed that Sample 1 contained more (E)-β-farnesene, α-bisabolol, and en-yn-dicycloether than the other two samples, which had much more bisabolol oxides. The concentration of chamazulene in all samples remained between 5 and 7% and was in agreement with published data (6–7%) [2, 5, 8–11].

The composition of Estonian chamomile essential oil resembled that of chemotype oils rich in bisabolol oxides described by Grgesina et al. [5], Vuorela et al. [8], and Reverchon & Senatore [10]. The amount of en-yn-dicycloether in chamomile oil SDE extracts from Estonia harvested in 1997 was somewhat higher (17–22%) than in samples studied in 1981 (10–16%) [2] and also in extracts from Italy (13%) [10], Hungary (12%) [9], and Croatia (8%) [8].

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REFERENCES

1. Koppel, T., Arak, E. & Türi, E. Taimse päritoluga ravimite antimikroobse toime uurimine I. *Eesti Rohuteadlane*, 1993, **3**, 107–109.
2. Arak, E. H. Results of essential oil analysis of pineapple food and wild chamomile by gas chromatographic method. In *Abstract Book of II Congress of Estonian Pharmacists*. Tallinn, 1981, 79–80.
3. Arak, E. H., Pehk, T. T., Mäeorg, U. J., Vahar, V. E. & Altsaar, T. M. Isolation of spathylenol from essential oil of wild chamomile and its identification. In *Abstract Book of II Congress of Estonian Pharmacists*. Tallinn, 1981, 84–87 (in Russian).
4. Brunke, E.-J., Hammerschmidt, E.-J. & Schmaus, G. Headspace analysis of selected European medicinal plants. In *Proceedings of the 12th International Congress of Flavours, Fragrances and Essential Oils, Vienna, Austria, Oct. 4.–8. 1992* (Woidlich, H. & Buchbauer, G., eds.). Fachzeitschriftenverlags, Vienna, 1992, 105–124.
5. Grgesina, D., Mandic, M. L., Karuza, L., Klapec, T. & Bočkinac, D. Chemical composition of different parts of *Matricaria chamomilla*. *Prehrambeno-tehnol. biotehnol. rev*., 1995, **33**, 111–113.
6. Pekic, B., Zekovic, Z. & Tolic, A. Study of the extraction of chamomile flowers with supercritical carbon dioxide. *J. Serb. Chem. Soc*., 1995, **60**, 439–443.
7. Zekovic, Z., Pekic, B., Lepojevic, Z. & Petrovic, L. Chromatography in our investigations of chamomile (*Matricaria chamomilla* L.). *Chromatographia*, 1994, **39**, 587–590.
8. Vuorela, H., Holm, Y. & Hiltunen, R. Application of headspace gas chromatography in essential oil analysis. Part VIII. *Flavour Fragr. J*., 1989, **4**, 113–116.
9. Vuorela, H., Holm, Y., Hiltunen, R., Tarvela, T. & Laitinen, A. Extraction of the volatile oil in chamomile flowerheads using supercritical carbon dioxide. *Flavour Fragr. J*., 1990, **5**, 81–84.

10. Reverchon, E. & Senatore, F. Supercritical carbon dioxide extraction of chamomile essential oil and its analysis by gas chromatography–mass spectrometry. *J. Agric. Food Chem*., 1994, **42**, 154–158.
11. Simandi, B., Kery, A., Lemberkovics, E., Oszagyan, M., Ronyai, E., Mathe, I., Fekete, J. & Hethelyi, E. Supercritical fluid extraction of medicinal plants. In *Process Technology Proceedings, 12. High Pressure Chemical Engineering. Proceedings of the 3rd International Symposium of High Pressure Chemical Engineering, Zürich, Switzerland, October 7–9, 1996* (von Rohr, Ph. R. & Trepp, Ch., eds.). Elsevier, Amsterdam, 1996, 357–362.
12. Davies, N. W. Gas chromatographic retention indices of monoterpenes and sesquiterpenes on methyl silicone and Carbowax 20M phases. *J. Chromatogr*., 1990, **503**, 1–24.
13. Venskutonis, P. R., Dapkevicius, A. & Baranauskiene, M. Flavour composition of some lemon-like aroma herbs from Lithuania. In *Food Flavors: Generation, Analysis and Process Influence* (Charalambous, G., ed.). Elsevier Science B.V., 1995, 833–847.

EESTIS KULTIVEERITUD *Matricaria recutita* L. EETERLIKU ÕLI KOOSTIS

Anne ORAV, Tiiu KAILAS ja Kaire IVASK

Eestis kultiveeritud teekummeli (*Matricaria recutita* L.) eeterlik õli eraldati samaaegse destillatsiooni ja ekstraktsiooni mikromeetodil ning analüüsiti seda kapillaargaasikromatograafia ja kromatomassispektromeetria abil. Õlis identifitseeriti 37 komponenti, neist suurima sisaldusega olid bisabolooloksiid A (20–33%) ja B (8–12%), bisaboloonoksiid A (7–14%), (E)-β-farneseen (4–13%), α-bisabolool (8–14%), hamasuleen (5–7%) ja en-üün-ditsükloeeter (17–22%). Uuriti ka teekummeli õli koostise ja saagise sõltuvust destillatsiooni ajast.