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GAS CHROMATOGRAPHIC CHARACTERIZATION OF FREQUENTLY OCCURRING AROMATIC COMPOUNDS IN ESSENTIAL OILS

É. LEMBERKOVICS* and G. PETRI
Institute of Pharmacognosy, Semmelweis Medical University, Üllöi út 26.III, 1085 Budapest (Hungary)

SUMMARY

Relative retention factors on stationary phases of two different polarities were determined for the gas chromatographic characterization of frequently occurring aromatic compounds in essential oils. The relative retention factors obtained with aromatic standards were in good agreement with those of the corresponding components of various essential oils examined under identical conditions.

INTRODUCTION

As part of our continuing work on the mono- and sesquiterpenes[1,2], we present here the relative retention factors of aromatic compounds frequently occurring in essential oils of plants. Our aim was to facilitate the identification of common phenylpropane derivatives in essential oils and the selection of suitable stationary phases for their analysis.

EXPERIMENTAL

We used a JEOL JGC 1100 gas chromatograph with a flame-ionization detector. The flow-rate of the carrier gas (nitrogen) was 30–40 ml/min. Column 1 was a glass spiral (3 m × 2.3 mm I.D.), coated with 3% OV-17 on 10–120-mesh Gas-Chrom Q and programmed from 60 to 230°C at 8°C/min. Its efficiency relative to linalool was 1825 plates/m with McReynolds constants of 119, 158, 162, 243 and 202[5]. Column 2 was a glass spiral (3 m × 3.4 mm I.D.) coated with 1.5% Sp-2250 + 0.95% Sp-2401 on 100–120-mesh Supelcoport and programmed from 60 to 230°C at 8°C/min. Its efficiency relative to linalool was 1776 plates/m with McReynolds constants of 129, 189, 238, 330 and 244. (The constants of the mixed phase were calculated from the data for the pure components[3].)

The concentration of the investigated solutions was 1–2 mg/ml of standard in chloroform and 10–20 mg/ml of essential oil in chloroform. Samples of 0.1–0.5 μl were injected. The temperatures of the injector and detector were both 240°C. The electrometer sensitivity range was $8 \cdot 10^{-10}$ A/mV. A JEOL IR-251 A recorder and a Digint 21 integrator (Chinoin) were used. Measurement of retention times was carried out with an accuracy of 0.1 s; relative retention factors were determined from

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3–5 parallel measurements, and the accuracy of the elution temperatures was ± 1°C.

The standard series investigated on the two types of stationary phases consisted of 15–20 components prepared from 25 aromatic compounds. Samples of anise, fennel, clove, pimenta, parsley, propolis, cinnamon, thyme and serpylli and also some Hungarian spice mixtures (salad, white meat, game, bouillon and fruit essential oils) were analysed. Identification of the component of the essential oils was carried out by the standard additions method on stationary phases of different polarities.

RESULTS AND DISCUSSION

The order of elution of aromatic compounds showed some regularities (Figs. 1 and 2).

The singly substituted aromatic compounds were eluted earlier than the doubly or multiply substituted compounds (compare the elution order of methylchavicol, safrol, myristicin, and apiol; the retention factors increase in order of the increase in molecular weight). The elution temperature of an aldehyde is always lower than that of the corresponding alcohol (see, *e.g.*, the benzaldehyde–benzyl alcohol and cinnamaldehyde–cinnamyl alcohol pairs). This effect is probably due to the lower boiling points of the aldehydes.

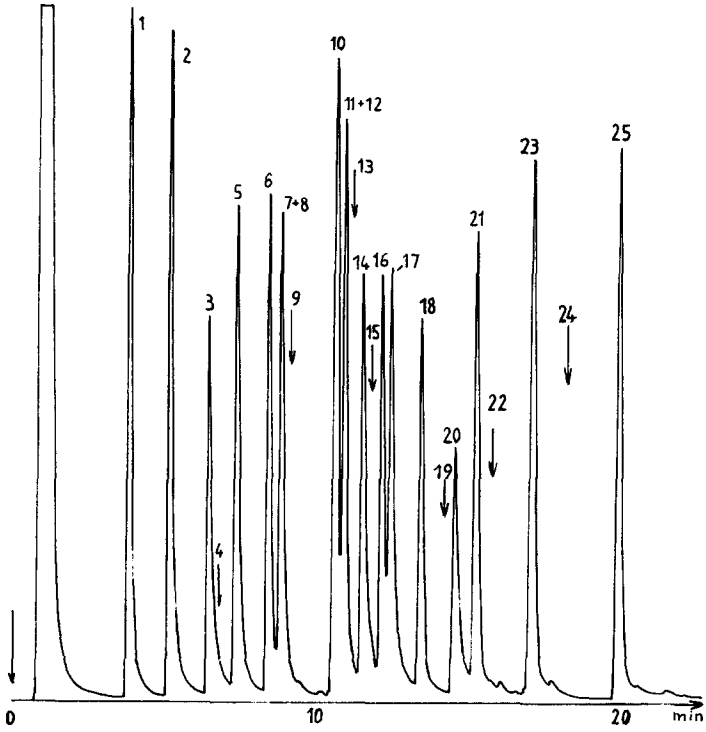

Fig. 1. Separation of aromatic compounds on a 3% OV-17 column (3 m × 2.3 mm I.D.), programmed from 60 to 230°C at 8°C/min; nitrogen flow-rate, 35 ml/min. For identification of the peaks, see Table I.

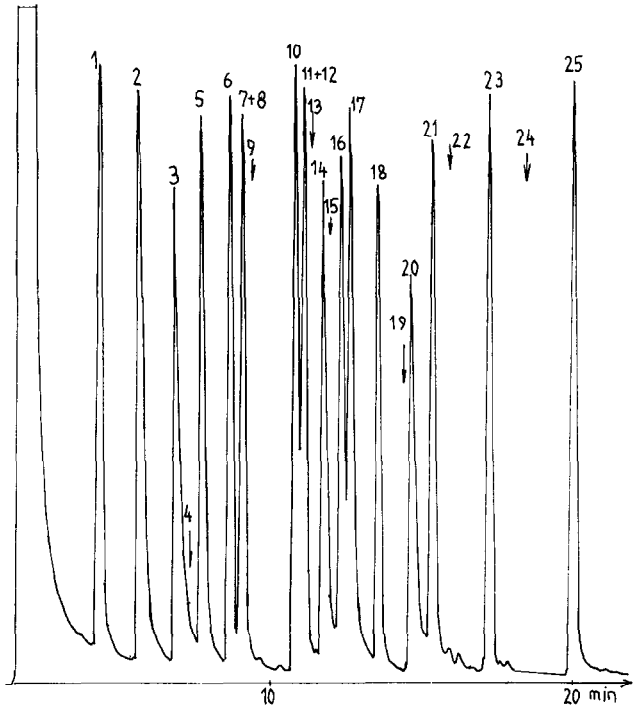

Fig. 2. Separation of aromatic compounds on a 1.5% Sp-2250–0.95% Sp-2401 column (3 m × 3.4 mm I.D.), programmed from 60 to 230°C at 8°C/min; nitrogen flow-rate, 35 ml/min. For identification of the peaks, see Table II.

The esters were eluted later than the alcohols, similarly to the behaviour of monoterpenes[1,4]. The elution order of esters depended on the number of carbon atoms in the acid components (benzyl alcohol < benzyl acetate < benzyl benzoate). When the acid component of esters was the same, the elution order was influenced by the number of carbon atoms in the alcohol components (*e.g.*, methyl benzoate < cyclohexyl benzoate < benzyl benzoate).

The difference in polarity between the two stationary phases did not materially influence the separation of aromatic compounds (Figs. 1 and 2). The mixed phase, owing to its slightly higher polarity, proved more advantageous for the separation of some monoterpene pairs[1]. However, we consider that the determination of the retention factors of aromatic compounds is also very important, because aromatic compounds and monoterpenes frequently occur together in essential oils and they have to be investigated simultaneously.

The relative retention factors of the aromatic components of the standard series were determined by reference to suitably selected aromatic compounds (Tables I and II).

To conform the reproducibility of the data, the retentions of the reference aromatic compounds were calibrated against a suitable hydrocarbon series (Tables III and IV). In order to examine the reliability and reproducibility of the relative retention factors, the standard series were compared with those of the corresponding

TABLE I

RETENTION FACTORS OF AROMATIC COMPOUNDS RELATIVE TO DIFFERENT REFERENCE AROMATIC STANDARDS ON 3% OV-17 STATIONARY PHASE

T_e = elution temperature.

No.	*Aromatic compound*	*Retention factor relative to selected standards*				
		Phenyl vinyl ether (T_e = 92°C)	*Anisyl vinyl ether (T_e = 132°C)*	*Anethole (T_e = 148°C)*	*Methylisoeugenol (T_e = 185°C)*	*Benzyl benzoate (T_e = 228°C)*
1	Phenyl vinyl ether	1.000	0.385	0.307	0.209	0.153
2	Benzaldehyde	1.452	0.559	0.445	0.304	0.223
3	Benzyl alcohol	1.839	0.708	0.564	0.385	0.282
4	Phenylacetaldehyde	1.935	0.745	0.594	0.405	0.297
5	Methyl benzoate	2.145	0.826	0.658	0.449	0.329
6	Guaiacol vinyl ether	2.484	0.956	0.762	0.520	0.381
7	Anisyl vinyl ether	2.597	1.000	0.797	0.544	0.398
8	Benzyl acetate	2.597	1.000	0.797	0.544	0.398
9	Methylchavicol	2.677	1.031	0.822	0.561	0.411
10	Thymol	3.193	1.230	0.980	0.669	0.490
11	Carvacrol	3.258	1.255	1.000	0.682	0.500
12	Anethole	3.258	1.255	1.000	0.682	0.500
13	Safrol	3.355	1.292	1.030	0.703	0.515
14	Anisaldehyde	3.516	1.354	1.079	0.736	0.540
15	Cinnamaldehyde	3.548	1.366	1.089	0.743	0.544
16	Cinnamyl alcohol	3.693	1.422	1.134	0.774	0.567
17	Eugenol	3.742	1.441	1.148	0.784	0.574
18	Methyleugenol	4.112	1.584	1.262	0.861	0.631
19	Isoeugenol	4.419	1.702	1.356	0.926	0.678
20	Vanillin	4.516	1.739	1.386	0.946	0.693
21	Methylisoeugenol	4.774	1.838	1.465	1.000	0.733
22	Myristicin	5.000	1.925	1.535	1.047	0.767
23	Cyclohexyl benzoate	5.419	2.087	1.663	1.135	0.832
24	Apiol	5.968	2.298	1.832	1.250	0.916
25	Benzyl benzoate	6.516	2.509	2.000	1.365	1.000

TABLE II

RETENTION FACTORS OF AROMATIC COMPOUNDS RELATIVE TO DIFFERENT REFERENCE AROMATIC STANDARDS ON 1.5% Sp-2250–0.95% Sp-2401 STATIONARY PHASE

No.	*Aromatic compound*	*Retention factor relative to selected standards*				
		Phenyl vinyl ether (T_e = 98°C)	*Anisyl vinyl ether* (T_e = 140°C)	*Anethole* (T_e = 156°C)	*Methylisoeugenol* (T_e = 190°C)	*Benzyl benzoate* (T_e = 230°C)
1	Phenylvinyl ether	1.000	0.390	0.311	0.217	0.167
2	Benzaldehyde	1.437	0.561	0.447	0.313	0.241
3	Benzyl alcohol	1.828	0.713	0.568	0.398	0.306
4	Phenylacetaldehyde	1.937	0.756	0.602	0.422	0.325
5	Methyl benzoate	2.094	0.817	0.650	0.456	0.351
6	Guaiacol vinyl ether	2.406	0.939	0.747	0.524	0.403
7	Anisyl vinyl ether	2.562	1.000	0.796	0.558	0.429
8	Benzyl acetate	2.562	1.000	0.796	0.558	0.429
9	Methylchavicol	2.625	1.024	0.810	0.571	0.440
10	Thymol	3.125	1.219	0.970	0.680	0.524
11	Carvacrol	3.219	1.256	1.000	0.701	0.539
12	Anethole	3.219	1.256	1.000	0.701	0.539
13	Safrol	3.312	1.293	1.029	0.721	0.555
14	Anisaldehyde	3.437	1.341	1.068	0.748	0.576
15	Cinnamaldehyde	3.515	1.372	1.090	0.765	0.589
16	Cinnamyl alcohol	3.609	1.408	1.121	0.786	0.605
17	Eugenol	3.734	1.457	1.160	0.813	0.626
18	Methyleugenol	4.031	1.573	1.254	0.877	0.675
19	Isoeugenol	4.281	1.671	1.330	0.932	0.717
20	Vanillin	4.406	1.719	1.369	0.959	0.738
21	Methylisoeugenol	4.594	1.793	1.430	1.000	0.770
22	Myristicin	4.781	1.866	1.485	1.041	0.801
23	Cyclohexyl benzoate	5.219	2.036	1.621	1.136	0.874
24	Apiol	5.641	2.201	1.752	1.228	0.945
25	Benzyl benzoate	5.969	2.329	1.854	1.299	1.000

TABLE III

RETENTION FACTORS OF REFERENCE AROMATIC STANDARDS RELATIVE TO THE *n*-ALKANE SERIES (C_9–C_{20}) ON 3% OV-17 STATIONARY PHASE

n-Alkane reference	*Phenyl vinyl ether*	*Anisyl vinyl ether*	*Anethole*	*Methyl-isoeugenol*	*Benzyl benzoate*
C_9H_{20}	1.409	3.659	4.591	6.727	9.182
$C_{10}H_{22}$	0.838	2.176	2.730	4.000	5.459
$C_{11}H_{24}$	0.574	1.491	1.870	2.741	3.741
$C_{12}H_{26}$	0.430	1.118	1.403	2.055	2.805
$C_{13}H_{28}$	0.344	0.894	1.122	1.644	2.244
$C_{14}H_{30}$	0.284	0.738	0.927	1.358	1.853
$C_{15}H_{32}$	0.248	0.644	0.808	1.184	1.616
$C_{16}H_{34}$	0.221	0.575	0.721	1.057	1.443
$C_{18}H_{38}$	0.194	0.503	0.631	0.925	1.262
$C_{19}H_{40}$	0.176	0.457	0.574	0.841	1.147
$C_{20}H_{42}$	0.161	0.419	0.526	0.771	1.052

aromatic components of several different essential oils (Table V). The retention data obtained showed reasonable agreement (standard deviation < 0.015).

CONCLUSION

The retention factors of the aromatic compounds obtained facilitate the qualitative investigation of essential oils, provide an aid for the selection of a suitable stationary phase for the separation of known aromatic compounds and afford a starting point for the analysis of essential oils of unknown origin.

TABLE IV

RETENTION FACTORS OF REFERENCE AROMATIC STANDARDS RELATIVE TO THE *n*-ALKANE SERIES (C_9–C_{20}) ON 1.5% Sp-2250–0.95% Sp-2401 STATIONARY PHASE

n-Alkane reference	*Phenyl vinyl ether*	*Anisyl vinyl ether*	*Anethole*	*Methyl-isoeugenol*	*Benzyl benzoate*
C_9H_{20}	1.333	3.416	4.292	6.125	7.958
$C_{10}H_{22}$	0.800	2.050	2.575	3.675	4.775
$C_{11}H_{24}$	0.547	1.402	1.761	2.513	3.265
$C_{12}H_{26}$	0.415	1.065	1.338	1.909	2.480
$C_{13}H_{28}$	0.333	0.854	1.073	1.531	1.990
$C_{14}H_{30}$	0.282	0.722	0.907	1.295	1.683
$C_{15}H_{32}$	0.245	0.628	0.789	1.126	1.464
$C_{16}H_{34}$	0.227	0.581	0.730	1.042	1.355
$C_{18}H_{38}$	0.199	0.509	0.640	0.913	1.186
$C_{19}H_{40}$	0.183	0.468	0.588	0.840	1.091
$C_{20}H_{42}$	0.169	0.434	0.545	0.778	1.010

TABLE V

AROMATIC COMPOUNDS IDENTIFIED (+) IN DIFFERENT ESSENTIAL OILS

No.	*Aromatic compound*	*Anise*	*Fennel*	*Thyme, Serpylli*	*Clove, Pimenta*	*Parsley*	*Propolis*	*Cinnamon*	*Spices*
1	Phenyl vinyl ether						+		
2	Benzaldehyde							+	
3	Benzyl alcohol						+		
4	Phenylacetaldehyde								
5	Methyl benzoate								
6	Guaiacol vinyl ether						+		
7	Anisyl vinyl ether						+		
8	Benzyl acetate						+		
9	Methylchavicol		+						+
10	Thymol			+					+
11	Carvacrol			+					
12	Anethole	+	+					+	+
13	Safrole							+	
14	Anisaldehyde	+	+						
15	Cinnamaldehyde						+	+	+
16	Cinnamyl alcohol						+		
17	Eugenol				+				+
18	Methyleugenol				+			+	+
19	Isoeugenol				+				
20	Vanillin						+		
21	Methylisoeugenol						+		
22	Myristicin					+			+
23	Cyclohexyl benzoate						+		
24	Apiol					+			+
25	Benzyl benzoate						+		

REFERENCES

1 É. Lemberkovics, *J. Chromatogr.*, 286 (1984) 293.
2 É. Lemberkovics and G. Verzár-Petri, *J. Chromatogr.*, 318 (1985) 125.
3 W. O. McReynolds, *J. Chromatogr. Sci.*, 8 (1970) 685.
4 Analytical Methods Commitee, *Analyst (London)*, 106 (1981) 448.