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Note

Separation behaviour of some isomeric organic compounds on sugars, sugar alcohols and their mixed phases by gas–liquid chromatography

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Although Kreicberger *et al.*[1] and Novrocík *et al.*[2] have reported the separation of 3- and 4-nitro-*o*-xylenes (2,3- and 3,4-dimethylnitrobenzenes) by gas–liquid chromatography (GLC), there have been few systematic separations of dimethylnitrobenzene isomers. Further, sugar alcohols were found to be effective liquid stationary phases for separating xylenol (dimethylphenol) isomers[3] and separations of nitroxylene isomers were performed on some sugar alcohols.

Fitzgerald[4], Brooks[5], Kolšek and Matičíc[6], Sassenberg and Wrabetz[7,8], Karr *et al.*[9], Jamieson[10], Mortimer and Gent[11], Janák and Komers[12], Payan[13], Bhattacharjee and Basu[14], Husain *et al.*[15] and Hrivnák and co-workers[16,17] have described separations of alkylphenol, cresol and dimethylphenol isomers by GLC. Brooks[5] separated all the dimethylphenol isomers on 2,4-xylenyl phosphate, Kolšek and Matičič[6] separated them on di-(3,3,5-trimethylcyclohexyl) phthalate, Janák and Komers[12] on salicylideneaminoguanidine and Bhattacharjee and Basu[14] on rubidium benzenesulphonate (clathrate-forming stationary phases), and Mortimer and Gent[11] separated *m*- and *p*-cresols on Bentone-34 and tolyl phosphate.

In spite of these studies, systematic studies of the separation of the dimethylphenol isomers have never been reported. 2,4,7-Trinitro-9-fluorenone[18], sugar alcohols and mixed columns of 2-nitrofluorene and dulcitol or inositol[3] have been shown to be effective liquid stationary phases. For the above reasons, the separation behaviour of nitroxylene and xylenol isomers on sugars, sugar alcohols and mixed columns has been investigated.

EXPERIMENTAL

Apparatus

A Shimadzu Model GC-5A gas chromatograph equipped with a flame-ionization detector was used.

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Chromatographic procedure

The separation column was a 2.25 m × 3 mm I.D. stainless-steel U-tube packed with acid-washed Sil-O-Cel C_{22} firebrick (60–80 mesh) support (Johns-Manville, Denver, CO, U.S.A.; purchased from Gaskuro Kogyo, Tokyo, Japan) coated with 20% (w/w) of liquid. The column and injector temperature were 140 and 270°C, respectively. Nitrogen was used as the carrier gas at 25 ml/min.

The support coated with the liquid phase was heated in an electric oven at the column temperature for 10 h in order to remove the solvent; subsequently it was kept under a stream of nitrogen for 6 h at a temperature *ca.* 20°C higher than the column temperature used.

Liquid stationary phases

Dioctyl phthalate (DOP), dulcitol (DUL), inositol (INO), sorbitol (SOR), mannitol (MAN), xylitol (XYT), glucose (GLU), mannose (MAS), sucrose (SUR), lactose (LAC), fructose (FRU), maltose (MAL), xylose (XYL), starch (soluble) (STA), Span-85 (SPA-85), diglycerol (DIL), galactose (GAL) and sorbose (SORB) of guaranteed grade from Nakarai Chemical Co. (Kyoto, Japan) were used without any purification.

2-Nitrofluorene (2-NF)[19,20], 2-nitro-9-fluorenone (2-NFO)[19–21], 2,7-dinitrofluorene (2,7-DNF)[21–23], 2,7-dinitro-9-fluorenone (2,7-DNFO)[21–23], 2,5-dinitro-9-fluorenone (2,5-DNFO)[21–23], 2,4,7-trinitro-9-fluorenone (2,4,7-TNFO)[24–26] and 2,4,5,7-tetranitro-9-fluorenone (2,4,5,7-TNFO)[25,26] were synthesized in the laboratory and their purities were confirmed by TLC and IR spectrometry.

Samples

3,5-Dimethylnitrobenzene (3,5-DMNB)[27], 2,5-dimethylnitrobenzene (2,5-DMNB)[28], 2,3-dimethylnitrobenzene (2,3-DMNB)[29], 3,4-dimethylnitrobenzene (3,4-DMNB)[30], 2,6-dimethylnitrobenzene (2,6-DMNB)[31], 2,4-dimethylnitrobenzene (2,4-DMNB)[31], 2,3-dimethylphenol (2,3-DMP)[32], 2,6-dimethylphenol (2,6-DMP)[33], 3,4-dimethylphenol (3,4-DMP)[34], 2,4-dimethylphenol (2,4-DMP)[35], 2,5-dimethylphenol (2,5-DMP)[36] and 3,5-dimethylphenol (3,5-DMP)[37] were synthesized and their purities were confirmed by GLC and IR and NMR spectrometry.

RESULTS AND DISCUSSION

Separation of DMNB isomers

In the separation of DMP isomers, it was found that hydrogen bonding plays an important role. It was expected that hydrogen bonding would also be effective in separating DMNB isomers, because the nitro function of DMNB isomers will interact with the hydroxy function of the stationary phase.

The results of the separation of DMNB isomers on nitrofluorenes, nitro-9-fluorenones, sugars, sugar alcohols, polyols and mixed columns are given in Tables I–III. From Table I it can be seen that 2-NF, 2-NFO, 2,5-DNFO, 2,7-DNF, 2,7-DNFO, 2,4,7-TNFO and 2,4,5,7-TNFO resolved effectively all six isomers. In these separations, effective separations were performed at a temperature below the melting points of the liquid stationary phases[38–41].

Norman[42] reported the separation of nitrotoluene isomers by GLC on 2,4,7-

TABLE I

RELATIVE RETENTION TIMES IN THE SEPARATIONS OF DMNB ISOMERS USING NITROFLUORENES AND NITROFLUORENONES

Stationary phase	*2,6-DMNB*	*2,5-DMNB*	*2,3-DMNB*	*2,4-DMNB*	*3,5-DMNB*	*3,4-DMNB*
2-NF	1.00	1.61	1.69	2.10	2.47	3.56
2-NFO	1.00	1.73	1.91	2.21	2.83	3.49
2,5-DNFO	1.00	1.81	1.97	2.29	2.97	3.70
2,7-DNF	1.00	1.70	1.89	2.27	2.63	3.52
2,7-DNFO	1.00	1.75	1.92	2.31	2.71	2.67
2,4,7-TNFO	1.00	1.93	2.13	2.46	2.86	4.06
2,4,5,7-TNFO	1.00	1.81	2.02	2.59	2.89	4.08
2,4,7-TNFO + 2,4,5,7-TNFO	1.00	1.87	2.25	2.25	2.58	3.88

TABLE II

RELATIVE RETENTION TIMES IN THE SEPARATIONS OF DMNB ISOMERS ON MIXED COLUMNS

Stationary phase	*2,6-DMNB*	*2,5-DMNB*	*2,3-DMNB*	*2,4-DMNB*	*3,5-DMNB*	*3,4-DMNB*
2-NF + DOP	1.00	1.75	2.06	2.06	2.33	2.27
2-NF + DUL	1.00	1.91	2.27	2.27	2.59	3.95
2-NF + INO	1.00	1.79	2.16	2.16	2.16	3.53
2-NF + SOR	1.00	1.81	2.17	2.30	2.30	3.61
2-NF + MAN	1.00	1.91	2.26	2.26	2.49	4.09
2-NF + XYT	1.00	1.80	2.00	2.25	2.80	3.95
2-NFO + DUL	1.00	1.86	2.20	2.23	2.59	5.77
2-NFO + MAN	1.00	2.00	2.29	2.29	2.82	4.24
2-NFO + SOR	1.00	1.81	2.13	2.38	2.75	4.19

TNFO at 200°C, but they were resolved effectively at 140°C below its melting point[43].

From Table II, it can also be seen that the mixed column of 2-NF + XYT gave effective separations of all six isomers. Further, the mixed columns of 2-NF and sugar alcohols showed various kinds of separation modes and the mixed columns of 2-NFO and DUL or SOR separated all of the isomers. From Table III, it can be seen that sugars (mono-, di- and polysaccharides) gave effective resolutions of all

TABLE III

RELATIVE RETENTION TIMES IN THE SEPARATIONS OF DMNB ISOMERS USING SUGARS

Stationary phase	*2,6-DMNB*	*2,5-DMNB*	*2,3-DMNB*	*2,4-DMNB*	*3,5-DMNB*	*3,4-DMNB*
GLU	1.00	1.66	1.89	2.06	2.44	3.17
MAS	1.00	1.76	1.95	2.19	2.57	3.71
SUR	1.00	1.59	1.82	1.94	2.24	3.00
FRU	1.00	1.65	1.85	2.05	2.40	3.30
MAL	1.00	1.73	1.93	2.13	2.40	3.40
LAC	1.00	1.75	2.00	2.17	2.50	3.25
STA	1.00	1.59	1.73	2.05	2.35	3.15

TABLE IV

RELATIVE RETENTION TIMES IN THE SEPARATIONS OF DMNB ISOMERS USING SUGAR ALCOHOLS AND POLYOLS

Stationary phase	*2,6-DMNB*	*2,5-DMNB*	*2,3-DMNB*	*2,4-DMNB*	*3,5-DMNB*	*3,4-DMNB*
XYT	1.00	1.91	2.27	2.36	2.64	3.82
MAN	1.00	1.77	1.91	2.33	2.67	3.60
SOR	1.00	1.72	2.11	2.11	2.33	3.56
DUL	1.00	1.70	2.00	2.20	2.50	3.40
INO	1.00	1.37	2.94	2.94	3.15	4.42
INO + SOR	1.00	1.86	2.29	2.29	2.29	3.86
DUL + SOR	1.00	1.90	1.90	2.57	3.15	3.93
INO + DUL	1.00	1.32	1.79	2.00	2.32	3.05
DUL + XYT	1.00	1.94	2.50	2.50	2.50	4.28
SPA-85	1.00	1.65	1.65	2.17	2.36	2.91
DIL	1.00	1.78	2.25	2.25	2.25	3.56

six isomers with almost identical relative peak separations[44]. It seems that hydrogen bonding of the hydroxy group of the stationary phase with the nitro group of the sample plays an important role.

From Table IV, in can be seen that MAN, DUL and XYT separated all of the isomers. Of the mixed columns, only INO + DUL separated all of the isomers. DIL and SPA-85 were ineffective and DUL and XYT are of specific configuration (sugar alcohols). Further, PEG 1540 (polyether) and EGA (polyester) gave effective separations[18].

Separation of DMP isomers

Table V shows that although none of the mixed columns of 2-NF and sugar alcohols or polyols gave good separations of all of the DMP isomers, the mixed columns of 2-NFO and sugar alcohols or polyols separated the isomers effectively. The mixed columns of 2-NF and DUL, INO or XYT separated all of the isomers but DUL, XYT and INO are of specific configuration (sugar alcohols); hence the carbonyl group of 2-NFO seems to be effective in these separations.

TABLE V

RELATIVE RETENTION TIMES OF DMP ISOMERS ON MIXED COLUMNS

Stationary phase	*2,6-DMP*	*2,5-DMP*	*2,4-DMP*	*2,3-DMP*	*3,5-DMP*	*3,4-DMP*
2-NF + SOR	1.00	1.73	1.73	2.26	2.52	4.91
2-NF + MAN	1.00	1.94	1.94	2.34	3.08	3.86
2-NF + DUL	1.00	1.95	2.05	2.64	3.32	4.23
2-NF + INO	1.00	1.91	2.14	2.41	3.65	4.28
2-NF + XYT	1.00	1.80	1.90	2.20	3.00	3.55
2-NFO + DUL	1.00	1.97	2.09	2.68	3.34	4.28
2-NFO + MAN	1.00	1.89	2.04	2.61	3.36	4.18
2-NFO + SOR	1.00	1.92	2.00	2.56	3.24	4.12
2-NFO + INO	1.00	1.71	1.93	2.14	2.54	3.43
2-NFO + XYT	1.00	1.97	2.09	2.67	3.36	4.33

TABLE VI

RELATIVE RETENTION TIMES OF DMP ISOMERS ON SUGAR ALCOHOLS AND POLYOLS

Stationary phase	*2,6-DMP*	*2,5-DMP*	*2,4-DMP*	*2,3-DMP*	*3,5-DMP*	*3,4-DMP*
DUL	1.00	2.13	2.38	2.63	4.13	4.62
SOR	1.00	2.05	2.19	2.74	3.69	4.62
MAN	1.00	2.20	2.53	2.95	4.37	5.00
INO	1.00	2.00	2.23	2.23	4.03	4.29
XYT	1.00	2.24	2.38	2.86	4.09	4.95
DIL	1.00	1.72	1.72	2.23	2.60	3.16
DUL + SOR	1.00	1.90	1.90	2.57	3.15	3.93
INO + DUL	1.00	2.09	2.09	2.82	3.56	4.78
INO + SOR	1.00	2.27	2.27	2.73	3.82	4.91
DUL + XYT	1.00	2.03	2.03	2.68	3.38	4.35
INO + MAN	1.00	2.05	2.15	2.45	3.60	4.15

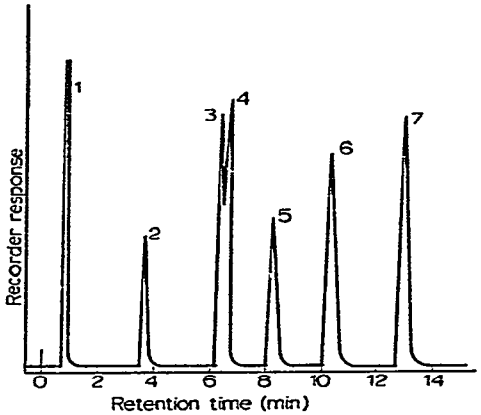
Table VI indicates that all of the sugar alcohols except INO separated all of the isomers. DIL did not give effective separations. INO is a different polyol from DUL and the others used, and DIL is a tetrahydric polyol, but not with vicinal tetrahydroxy groups. Further, the mixed columns of sugar alcohols except for INO and MAN did not give useful separations. Table VII shows that SUC, FRU, MAS, GLU, GAL, MAL, LAC and SORB separated all of the isomers effectively.

Although STA is a polyol, it could not separate 2,3-DMP from 2,4-DMP. The mixed columns of two kinds of sugars separated all of the isomers effectively. Although the retention times of DMP isomers on sugars or sugar alcohols are too short, an improvement was obtained on the mixed columns of 2-NF and INO or DUL and of 2-NFO and DUL or MAN. The improved chromatograms obtained on the mixed liquid stationary phases (2-NFO and DUL or MAN) are shown in Figs. 1 and 2.

TABLE VII

RELATIVE RETENTION TIMES OF DMP ISOMERS ON SUGARS AND POLYOLS

Stationary phase	*2,6-DMP*	*2,5-DMP*	*2,4-DMP*	*2,3-DMP*	*3,5-DMP*	*3,4-DMP*
SUC	1.00	2.13	2.31	2.69	4.00	4.62
FRU	1.00	1.63	1.73	1.97	2.63	2.97
MANS	1.00	1.96	2.24	2.50	2.74	4.33
GAL	1.00	1.94	2.18	2.47	3.65	4.12
GLU	1.00	1.79	2.05	2.32	3.36	3.79
MAL	1.00	1.80	2.00	2.33	3.20	3.66
LAC	1.00	2.13	2.44	2.63	4.00	4.38
SORB	1.00	1.62	1.85	1.92	2.31	3.31
STA	1.00	1.86	2.04	2.04	3.51	3.89
GLU + GAL	1.00	1.68	1.84	2.11	3.11	3.63
MAL + FRU	1.00	2.00	2.25	2.50	3.50	4.00
FRU + GAL	1.00	1.65	2.25	2.50	3.50	4.00
MAL + GLU	1.00	2.13	2.50	2.63	3.00	4.00
GAL + MAS	1.00	1.94	2.12	2.41	3.47	3.88
SPA-85	1.00	1.65	1.65	2.17	2.36	2.91



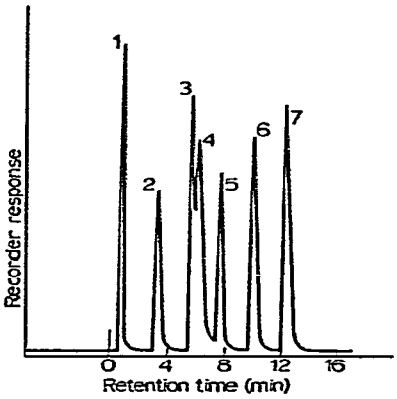


Fig. 1. Separation of DMP isomers on 2-NFO + DUL. Peaks: 1 = solvent; 2 = 2,6-DMP; 3 = 2,5-DMP; 4 = 2,4-DMP; 5 = 2,3-DMP; 6 = 3,5-DMP; 7 = 3,4-DMP.

Fig. 2. Separation of DMP isomers on 2-NFO + MAN. Peaks: 1 = solvent; 2 = 2,6-DMP; 3 = 2,5-DMP; 4 = 2,4-DMP; 5 = 2,3-DMP; 6 = 3,5-DMP; 7 = 3,4-DMP.

In the separation of DMP isomers on the mixed columns of 2-NF and INO and of 2-NF (or 2-NFO) and sugar alcohols, the properties of the component stationary phases are additive. The functional group combined with sugars or sugar alcohols for the improvement of retention is generally preferable, if it can separate the isomers individually.

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REFERENCES

1 D. Kreicberger, S. Velfere and O. Neilands, *Latv. PSR Zinat. Akad. Vestis, Kim. Ser.*, (1970) 170.
2 J. Novrocík, K. Komárek and J. Poskočil, *J. Chromatogr.*, 124 (1976) 73.
3 A. Ono, *J. Chromatogr.*, 193 (1980) 300.
4 J. S. Fitzgerald, *Aust. J. Appl. Sci.*, 10 (1959) 169.
5 V. T. Brooks, *Chem. Ind. (London)*, (1959) 1317.
6 J. Kolšek and M. Matičič, *J. Chromatogr.*, 12 (1963) 305.
7 W. Sassenberg and K. Wrabetz, *Z. Anal. Chem.*, 184 (1961) 423.
8 K. Wrabetz and W. Sassenberg, *Z. Anal. Chem.*, 179 (1961) 333.
9 C. Karr, Jr., P. M. Brown, P. A. Estep and G. L. Humphrey, *Anal. Chem.*, 30 (1958) 1413.
10 G. R. Jamieson, *J. Chromatogr.*, 3 (1960) 494.
11 J. V. Mortimer and P. L. Gent, *Anal. Chem.*, 36 (1964) 754.
12 J. Janák and R. Komers, *Z. Anal. Chem.*, 164 (1958) 69.
13 P. S. Payan, *Chem. Ind. (London)*, (1960) 1090.
14 A. Bhattacharjee and A. N. Basu, *J. Chromatogr.*, 71 (1972) 534.
15 S. Husain, P. Kunzelmann and H. Schildknecht, *J. Chromatogr.*, 137 (1977) 53.
16 J. Hrivnák and J. Macák, *Anal. Chem.*, 43 (1971) 1039.
17 J. Macák, P. Buryan and J. Hrivnák, *J. Chromatogr.*, 89 (1974) 309.
18 A. Ono, *J. Chromatogr.*, 180 (1979) 170.
19 W. E. Kuhn, *Org. Syn.*, Collect. Vol. 2 (1943) 447.
20 E. A. Calderon, *An. Asoc. Quim. Argent.*, 33 (1945) 35.

21 E. H. Hunters, E. B. Hershberg and I. S. Cliff, *J. Amer. Chem. Soc.*, 53 (1931) 2720.
22 S. V. Anantakrishnan and E. D. Hughes, *J. Chem. Soc.*, (1935) 1607.
23 C. W. Bennett, W. G. Jewsbury and J. P. Dupuis, *J. Amer. Chem. Soc.*, 68 (1946) 2489.
24 M. Orchin and E. O. Woolfolk, *J. Amer. Chem. Soc.*, 68 (1946) 1727.
25 M. Orchin, L. Reggel and E. O. Woolfolk, *J. Amer. Chem. Soc.*, 69 (1947) 1225.
26 E. O. Woolfolk and M. Orchin, *Org. Syn.*, Collect. Vol. 3 (1955) 837.
27 W. A. Noyes, *J. Amer. Chem. Soc.*, 20 (1898) 789.
28 H. R. Snyder and F. J. Pilgrim, *J. Amer. Chem. Soc.*, 70 (1948) 3787.
29 O. H. Emerson and L. I. Smith, *J. Amer. Chem. Soc.*, 62 (1940) 141.
30 S. F. Birch, R. A. Dean, F. A. Filder and R. A. Laury, *J. Amer. Chem. Soc.*, 71 (1949) 1362.
31 E. Grevigk, *Chem. Ber.*, 17 (1884) 2429.
32 W. F. Short, H. Stromberg and A. E. Wiles, *J. Chem. Soc.*, (1936) 319.
33 R. B. Carlin and H. P. Landerl, *J. Amer. Chem. Soc.*, 72 (1950) 2762.
34 O. Jacobsen, *Chem. Ber.*, 17 (1884) 159.
35 W. R. Hodgkinson and L. Linpach, *J. Chem. Soc.*, 77 (1900) 65.
36 E. Noelting, O. N. Witt and S. Forel, *Chem. Ber.*, 18 (1885) 2664.
37 A. Toel, *Chem. Ber.*, 18 (1885) 362.
38 P. Urone, J. E. Smith and J. R. Katnik, *Anal. Chem.*, 34 (1962) 476.
39 D. Nogare, *Anal. Chem.*, 37 (1965) 1456.
40 R. R. Claeys and H. Freund, *J. Gas Chromatogr.*, 6 (1968) 421.
41 G. Dahlman, H. J. K. Köser and H. H. Oelert, *J. Chromatogr.*, 171 (1979) 398.
42 R. O. C. Norman, *Proc. Chem. Soc., London*, (1958) 151.
43 A. Ono, unpublished results.
44 W. L. Jones and R. Kieselbach, *Anal. Chem.*, 30 (1958) 1590.