

be resolved using TMS ethers, trifluoroacetates were tried, as well as conversion of the stanols to ketones. The comparison of the R_F values of the cholestanols and cholestanones helped resolve the last doubts on the identity of the components of almost any mixture of the above-tabulated cholestanols.

Department of Chemistry, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, AP 14-740, México 14, D.F. (Mexico)

JOSEF E. HERZ
ELIZABETH GONZÁLEZ

1 J. E. HERZ, L. A. MÁRQUEZ AND E. GONZÁLEZ, *Chem. Commun.*, (1967) 1095.

2 L. F. FIESER AND M. FIESER, *Steroids*, Reinhold, New York, 1959;

C. W. SHOPPEE, *Chemistry of Steroids*, Butterworths, London, 1964.

3 J. S. MATTHEWS, *Biochim. Biophys. Acta*, 69 (1963) 163.

4 H. M. FALES AND T. LUUKKAINEN, *Anal. Chem.*, 37 (1965) 955.

Received January 2nd, 1968

J. Chromatog., 34 (1968) 251-253

CHROM 3323

Gas chromatographic separation of 4'-nitroazobenzene-4-carboxylic acid esters of alcohols

The basic reagent 4'-nitroazobenzene-4-carboxylic acid chloride was first prepared by HECKER¹ in 1955, and later on used as a reagent for hydroxyl compounds as reported by BUTENANDT^{2,3} during investigations on 'bombicol'. The separation of amines as 4'-nitroazobenzene-4-carboxamides has been described earlier⁴. In the same way the gas chromatographic separation of esters can be helpful in the identification of saturated and unsaturated aliphatic and aromatic alcohols as well as phenols.

Experimental

Apparatus. For chromatography, 1-m and 2-m stainless steel tubes (4-mm I.D.) packed with 2.5 % w/w silicone grease (E. Merck AG., Darmstadt, Germany) on 60 to 80 mesh Chromosorb G, acid washed and DMCS treated, were used in fractometer type F6 (Perkin Elmer Bodenseewerk, Überlingen, Germany). The fractometer was equipped with a flame-ionization detector and a 2.5-mV recorder (Siemens-Kompenso-graph L 288 × 288) with a paper feed of 0.5 cm/min.

Procedure. The 4'-nitroazobenzene-4-carboxylic acid esters were prepared according to HECKER¹ and purified to the melting points listed there. Previously unrecorded derivatives were crystallized to a constant melting point. Samples of 1 to 2 μ g in about 5 μ l of benzene were injected into the chromatograph.

J. Chromatog., 34 (1968) 253-256

TABLE I
RETENTION TIMES OF 4'-NITROAZOBENZENE-4-CARBOXYLIC ACID ESTERS OF ALCOHOLS

<i>Parent alcohol</i>	<i>Retention time (min)</i>
<i>2-m column</i>	
Methanol	4.0
Ethanol	4.8
1-Propanol	6.0
1-Butanol	7.6
1-Pentanol	9.0
2-Methyl-3-butanol	8.0
2,2-Dimethyl-1-propanol	7.4
1-Hexanol	12.6
1-Heptanol	16.2
3-Octanol	13.4
4-Octanol	12.8
2-Ethyl-1-hexanol	15.2
1-Nonanol	23.5
1-Dodecanol	47.8
1-Propen-2-ol (allyl alcohol)	5.8
2-Buten-1-ol	7.6
3-Hexen-1-ol	12.4
3,7-Dimethyl-2,6-octadien-1-ol (geraniol)	28.8
Lavandulol	28.2
1,2-Propanediol	8.4
2-Methyl-2,4-pentanediol	9.3
3-Hydroxy-2-butanone (acetoin)	8.3
Cyclolavandulol	29.2
Cyclopentanol	11.8
Cyclohexanol	15.6
Cycloheptanol	11.6
2,5-Dimethyl-cyclohexanol	20.0
3,4-Dimethyl-cyclohexanol	22.0
3,5-Dimethyl-cyclohexanol	17.2
Menthol	27.4
3-Thujanol	24.8
1,3,3-Trimethyl-2-norbornanol (fenchyl alcohol)	24.6 + 31.0 (two isomers)
Borneol	30.2
Phenol	16.0
<i>o</i> -Cresol	19.2
<i>m</i> -Cresol	21.0
<i>p</i> -Cresol	21.8
Benzyl alcohol	21.6
<i>o</i> -Ethyl-phenol	22.2
<i>p</i> -Ethyl-phenol	27.6
β -Phenylethyl alcohol	26.0
2,3-Xylenol	28.0
3,4-Xylenol	30.6
2,6-Xylenol	21.4
2,4-Xylenol	25.6
3,5-Xylenol	28.4
2,5-Xylenol	24.8
3,4,5-Trimethyl-phenol	42.0
2,3,5-Trimethyl-phenol	33.8
2,4,6-Trimethyl-phenol (mesitol)	28.2
Carvacrol	30.8
Thymol	28.0
Guaiacol	24.2

(continued on p. 255)

TABLE I (continued)

Parent alcohol	Retention time (min)
<i>2-m column</i>	
2-Allyl-phenol	25.7
1,2,3,4-Tetrahydro-2-naphthol	62.0
1-Naphthol	60.4
2-Naphthol	67.6
2-Methoxy- <i>p</i> -cresol (creosol)	29.2
<i>o</i> -Methoxy-benzyl alcohol	31.0
<i>m</i> -Methoxy-benzyl alcohol	31.0
4-Ethyl-guaiacol	36.4
4-Vinyl-guaiacol	39.2
5-Vinyl-guaiacol	43.6
2-Methoxy-4-propenyl-phenol (isoeugenol)	60.6
4-Allyl-2-methoxy-phenol (eugenol)	46.4
2,6-Dimethoxy-phenol	36.6
4-(2,6,6-Trimethyl-2-cyclohexen-1-yl)-butan-2-ol	46.4
4-(2,6,6-Trimethyl-1-cyclohexen-1-yl)-butan-2-ol	∞
<i>p</i> -Hydroxy-benzaldehyde	38.6
Vanillin	27.0
Isovanillin	58.2
5-Allylvanillin	66.0
2-(<i>N</i> -ethyl-anilino)-ethanol	44.6
Furfuryl alcohol	11.4
5-(Hydroxymethyl)-furfurol	28.8
3-Pyridinol	16.8
4-Pyridinol	19.4
<i>1-m column</i>	
1-Heptanol	4.2
3-Octanol	4.4
4-Octanol	4.0
2-Ethyl-1-hexanol	4.6
1-Nonanol	7.6
1-Undecanol	12.0
1-Dodecanol	14.6
1-Octadecanol	56.8
3-Hexen-1-ol	3.8
Nerol	9.0
1,3,3-Trimethyl-2-norbornanol (fenchyl alcohol)	9.4
<i>o</i> -Cresol	5.8
3,5-Xylenol	8.0
3,4,5-Trimethyl-phenol	12.0
Santalol	28.6
4-Methyl-1,2,3,4-tetrahydro-1-naphthol	19.8
1-Naphthol	18.6
2-Naphthol	20.4
<i>o</i> -Methoxy-benzyl alcohol	9.6
4-Ethyl-guaiacol	11.0
Isovanillin	16.7
Vanillylacetone	24.6
Furfuryl alcohol	3.8
3,4-(Methylenedioxy)-phenol (sesamol)	11.6
3-Pyridinol	4.8

A slight peak distortion caused by some bleeding of silicone grease from the column was overcome by using a relatively high hydrogen flow rate to the flame-ionization detector (30 ml/min).

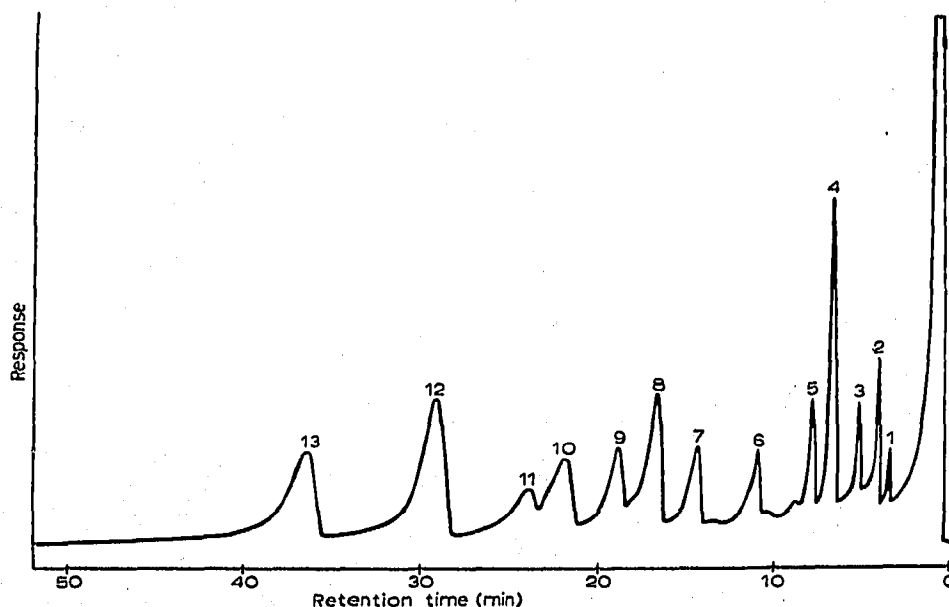


Fig. 1. Gas chromatographic separation of 4'-nitroazobenzene-4-carboxylic acid esters. Sample size: 1 to 2 μ g of each derivative in 5 μ l benzene. Constant column temperature: 270°; injection temperature: 300°; carrier gas: helium; flow rate: 30 ml/min. 1 = Methanol; 2 = ethanol; 3 = *n*-propanol; 4 = *n*-butanol; 5 = *n*-pentanol; 6 = *n*-hexanol; 7 = phenol; 8 = *o*-cresol; 9 = *p*-cresol; 10 = 2,3-xyleneol; 11 = 3,4-xyleneol; 12 = 2,3,5-trimethyl-phenol; 13 = 3,4,5-trimethyl-phenol.

Results

Fig. 1 shows the gas chromatographic separation of a mixture of 4'-nitroazobenzene-4-carboxylic acid esters. Table I lists the retention times of all derivatives tested on the 2-m and 1-m columns under the conditions indicated in the legend of Fig. 1. The retention time of the reagent, 4'-nitroazobenzene-4-carboxylic acid chloride, is about that of the solvent, thus excluding any interference with the derivatives.

Zentrallaboratorium H. F. & Ph. F. Reemtsma, 2 Hamburg 50,
Luruper Chaussee 145 (Germany)

GEORG NEURATH
WERNER LÜTTICH

1 E. HECKER, *Chem. Ber.*, 88 (1955) 1666.

2 A. BUTENANDT AND H. REMBOLD, *Z. Physiol. Chem.*, 308 (1957) 284.

3 A. BUTENANDT, D. STAMM AND E. HECKER, *Chem. Ber.*, 94 (1961) 1931.

4 G. NEURATH AND W. LÜTTICH, Paper presented at the Gas Chromatographic Symposium, Rome 1967; *J. Chromatog.*, (1968) in press.

Received November 10th, 1967