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Note

Gas chromatography of 2,4-dinitrophenyl derivatives of amines

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One of the major problems in the gas chromatographic analysis of amines lies in the adsorption of the compounds on the solid supports. The use of basic liquid phases or the treatment of supports with alkali often prevents tailing and provides satisfactory separations¹⁻⁵. Also, it is of great convenience to prepare amine derivatives in order to obtain symmetrical peak shapes on chromatograms^{6,7}.

In a previous paper⁸, we described a gas chromatographic method for the separation and identification of aliphatic amines in blood as their 2,4-dinitrophenyl (DNP) derivatives, and the occurrence of several primary and secondary amines in normal blood was demonstrated.

The unknown peaks simultaneously observed on these chromatograms led us to prepare more than ten DNP derivatives of amines and related compounds. This

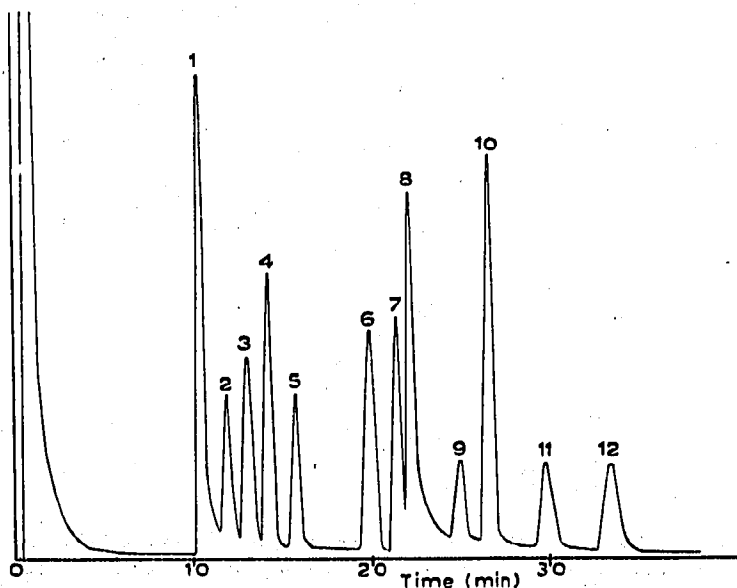


Fig. 1. Separation of a mixture of the 12 selected derivatives. (1) DNP-ammonia; (2) DNP-diethylamine; (3) DNP-isopropylamine; (4) DNP-*tert.*-butylamine; (5) DNP-*sec.*-butylamine; (6) DNP-isoamylamine; (7) DNP-*n*-amylamine; (8) TMS-DNP-ethanolamine; (9) DNP-aniline; (10) DNP-cyclohexylamine; (11) DNP-benzylamine; (12) DNP- β -phenylethylamine.

paper gives data on the melting points and the retention times of these compounds. Further, the present results, considered together with those given in the previous paper⁸, reveal that there are certain relationships between the structural formulae of the derivatives and their gas chromatographic properties.

EXPERIMENTAL

Standard DNP derivatives were prepared by the methods of Day *et al.*⁶ and Smith and Jepson⁹. The apparatus, column packing and operating conditions were described previously⁸.

TABLE I
RELATIVE RETENTION TIMES AND MELTING POINTS OF DNP DERIVATIVES

Compound	Relative retention time*	Melting point (°C)		
		Observed	Literature	Reference
Ammonia	0.73	180 -181	180	11
Methylamine	0.81	177 -180	178 -180	6
Ethylamine	0.90	112 -113	115	12
Isopropylamine	0.92	94	95	12
<i>tert.</i> -Butylamine	1.00 (14.2 min)	152 -153	154.5-155.0	6
Allylamine	1.03	73.5-75		
<i>n</i> -Propylamine	1.07	95	96.5- 97.0	6
<i>sec.</i> -Butylamine	1.10	54.5- 55.5	54.5- 55.0	6
Isobutylamine	1.16	77 - 80	79	12
<i>n</i> -Butylamine	1.29	89 - 91	89	12
Isoamylamine	1.40	92 - 93	91	12
<i>n</i> -Amylamine	1.51	81 - 82	78.0- 79.0	13
Isopropanolamine	1.54	98.5		
Ethanolamine	1.56**	88 - 89	92	12
<i>n</i> -Hexylamine	1.71	51	38.3- 39.3	14
Aniline	1.75	159 -160	153 -154	15
Cyclohexylamine	1.86	157 -159		
Benzylamine	2.09	118 -119.5		
β -Phenylethylamine	2.35	155.5-157	155	12
Dimethylamine	0.76	89 - 91	84.5- 86	6
Methylethylamine	0.83	58.5- 60.5	53 - 57	6
Diethylamine	0.84	67 - 68	67 - 68	6
Morpholine	1.28	118.5-121		
Piperidine	1.36	91 - 92	91	16
Pyrrolidine	1.53	101.5-103	102	12
Acetohexamide***	1.86	153.5-156		

* Column: 2 m \times 3 mm I.D. (glass). Packing: 10% SE-30 on siliconized Chromosorb W-AW (60-80 mesh). Rate of temperature increase: 2°/min from 190° (initial temperature) to 220°, then 3°/min to 250° (kept under the isothermal conditions when 250° was attained). Carrier gas: nitrogen at a flow-rate of 35 ml/min.

** This retention time was obtained by converting the hydroxyl group to the trimethylsilyl (TMS) ether.

*** This compound is known as an oral hypoglycaemic agent.

RESULTS AND DISCUSSION

An example of the gas chromatograms obtained is shown in Fig. 1 and the retention times of all the DNP derivatives (including the derivatives reported previously^{8,10}) relative to DNP-*tert.*-butylamine are given in Table I together with their melting points.

It is evident from Table I that the retention times of both the primary and secondary amines increase as their molecular weights increase, with a few exceptions. However, the secondary amines so far examined tend to be more rapidly eluted than primary amines, although they have higher molecular weights, because they do not have such strong hydrogen-bonding forces as primary amines. Of the isomers such as propylamines, butylamines and amylamines, those with branched chains have shorter retention times than those with straight chains. In addition, the order of emergence is not related to the melting points under the experimental conditions used.

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