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Note

Quantitative determination of 2-amino-1,3-propanediol and its impurities by capillary gas chromatography

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2-Amino-1,3-propanediol (serinol) is an important intermediate in the synthesis of (*S*)-N,N′-bis[2-hydroxy-1-(hydroxymethyl)ethyl]-5-[(2-hydroxy-1-oxopropyl)-amino]-2,4,6-triiodo-1,3-benzenedicarboxamide (iopamidol)[1], a non-ionic water-soluble iodinated X-ray contrast medium[2]. The purity of serinol is an essential prerequisite for obtaining a contrast medium suitable for uro-angiographic and myelographic examinations, which require injection of highly concentrated solutions.

Serinol, prepared on an industrial scale according to reported methods[3–8], is often contamined by small amounts of amino alcohols as by-products that are difficult to separate from the main product prior to its quantitative determination.

The separation of amino alcohols by gas chromatography (GC) after derivatization with trifluoroacetic anhydride (TFAA) is a well know technique[9], but literature concerning serinol and related compounds is scarce. The most recent paper[10] describing a GC method using such derivatization and a packed column was found to lack the ability to separate the amino alcohol impurities from serinol.

This paper describes a GC method which, using a capillary column and stream splitting, allows the simultaneous quantitation of serinol and contaminants.

EXPERIMENTAL

Materials

All the reagents were analytical-reagent grade unless specified otherwise. Ethanolamine (ETH), 1-amino-2,3-propanediol (ISO) and 2-aminopropanol (AP) were commercially available and were used as standards without further purification. Ultrapure serinol (SER), used as a standard, was prepared and purified as described in previous work[10]. 2,3-Diaminopropanol (DAP) and 2-aminomethyl-1,3-propanediol (NME) were obtained by lithium aluminium hydride reduction[11] of the corresponding methyl esters of 2,3-diaminopropionic acid and 2-aminomethyl-3-hydroxypropionic acid, respectively.

1-Methoxy-2-aminopropanol (OME) was obtained by partial demethylation of the corresponding 1,3-dimethoxy-2-aminopropane by hydrogen bromide. A 1% (w/w) solution of methyl palmitate in chloroform was used as the internal standard in GC analyses.

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Instruments

GC analyses were performed using a Hewlett-Packard HP 5890 gas chromatograph equipped with an HP 7393A atuomatic sampler and an HP 3393A integrator. A DB 1701 fused-silica capillary column (30 m × 0.25 mm I.D.; film thickness 0.25 μm), supplied by J & W Scientific, was used under the following operation conditions: injector temperature, 200°C; flame ionization detector temperature, 220°C [hydrogen flow-rate, 33 ml/min; air flow-rate, 375 ml/min; helium flow-rate (auxiliary, 29 ml/min]; carrier gas, helium (column flow-rate, 1 ml/min; split flow-rate, 62 ml/min; purge flow-rate, 4 ml/min).

The following temperature programme was used: an initial hold of 145°C for 6.5 min followed by a ramp of 15°C/min to 10°C, held for 1 min and then a second ramp of 30°C/min to 205°C, held for 16 min.

Injections of 4-μl aliquots were performed automatically. Gas chromatography–mass spectrometry (GC–MS) was carried out on a Finnigan-MAT 8222 mass spectrometer equipped with an INCOS data system and coupled to a Varian 3400 gas chromatograph working as described above. The column was directly connected to the electron-impact source (EI) and the spectra were taken at 70 eV; filament emission, 0.5 mA; source temperature, 200°C; resolving power, 1250 (10% valley).

The molecular weight was confirmed by performing GC–MS under chemical ionization (CI) conditions using methane as reagent gas. Infrared (IR) spectra were recorded on a Perkin-Elmer Model 882 spectrophotometer. Nuclear magnetic resonance (NMR) spectra were recorded on a Bruker AC 200 FT spectometer equipped with an ASPECT 3000 computer. All the spectra were recorded in hexadeuterated dimethyl sulphoxide using tetramethylsilane as an internal standard; chemical shifts (δ) are expressed in ppm.

Analytical derivatization procedure

Solutions of serinol (200 mg) in water (240 mg) spiked with various concentrations of a mixture of known impurities, each covering the range from 0.2 to 1.5% (w/w), were prepared. Each solution was derivatized in a standard screw-capped vial by adding trifluoroacetic anhydride (10 ml) cooled in a bath of solid carbon dioxide–acetone. The mixture was cautiously warmed to 40°C, then held at this temperature for 30 min; subsequently, after cooling to room temperature, methyl palmitate internal standard (100 mg) was added. Standard solutions of serinol, spiked with 0.7% of each impurity individually, were similarly derivatized and used as calibrating solutions.

Preparative derivatization procedure

To a well stirred mixture of amino alcohol (0.1 mol) and trifluoroacetic acid (0.25 mol), trifluoroacetic anhydride (0.6 mol) was added under nitrogen at 0°C. The reaction mixture was cautiously warmed to room temperature and then at 40°C for 1 h. The trifluoroacetyl derivative was recovered from the reaction mixture after evaporation of the solvent and purification either by distillation under vacuum or by crystallization from anhydrous diethyl ether. The products obtained (90–95% yield) were characterized as shown in Table I.

TABLE I

AMINO ALCOHOL TRIFLUOROACETYL DERIVATIVES PREPARED

Amino alcohol	*B.p.** (°C)	*Molecular formula***	*IR* (cm^{-1})	d_{20} ($g\ ml^{-1}$)	n_D^{20}	*^1H NMR*	*^{13}C NMR*
AP	108–110	$C_7H_7F_6NO_3$ 267.14	3308, 3101, 2992, 1788, 1707, 1553, 1461, 1405, 1386, 1354, 1164, 775, 734, 725 (neat)	1.4380	1.3640	9.4 (1H, bs, NH); 4.5 (1H, bm, CHNH); 4.3 (2H, bd, CH_2O); 1.25 (3H, bd, CH_3)	157.2 ($OCOCF_3$, J_{CCF} = 30 Hz); 156.3 ($NHCOCF_3$, J_{CCF} = 30 Hz); 115.9 ($NHCOCF_3$, J_{CF} = 289.4 Hz); 114.2 ($OCOCF_3$, J_{CF} = 289.4 Hz); 69.05 (CH_2OCO); 44.164 (CHNH); 15.13 (CH_3)
ETH	117–119	$C_6H_5F_6NO_3$ 253.12	3321, 3109, 2973, 1787, 1710, 1552, 1453, 1404, 1378, 1349, 1164, 1017, 775, 732 (neat)	1.4099	1.3730	9.65 (1H, bs, NH); 4.5 (2H, t, CH_2O); 3.5 (2H, dt, CH_2N, J = 4.2 Hz)	157.1 ($OCOCF_3$, J_{CCF} = 30 Hz); 156.8 ($NCOCF_3$, J_{CCF} = 30 Hz); 115.8 ($NCOCF_3$, J_{CF} = 289.4 Hz); 114.1 ($OCOCF_3$, J_{CF} = 289.4 Hz); 65.87 (CH_2OCO), 37.90 (CH_2NHCO)
OME	111–115	$C_8H_9F_6NO_4$ 297.16	3430, 3318, 3098, 2998, 2939, 2906, 2839, 1792, 1718, 1550, 1479, 1461, 1403, 1385, 1352, 1166, 894, 775, 735, 727 (neat)	1.3257	1.3730	9.5 (1H, bd, NHCO); 4.6 (1H, bm, CHNH); 4.5 (2H, bm, CH_2O); 3.6 (2H, bd, CH_2OCH_3); 3.3 (3H, s, CH_3O)	156.7 ($OCOCF_3$, J_{CCF} = 30 Hz); 155.4 ($NCOCF_3$, J_{CCF} = 30 Hz); 115.7 ($NHCOCF_3$, J_{CF} = 289.4 Hz); 114.1 ($OCOCF_3$, J_{CF} = 289.4 Hz); 69.69 (CH_2OCO); 66.28 (CH_2OCH_3); 58.11 (CH_3O); 48.12 (CHNH)
NME	140–141	$C_{10}H_8F_9NO_5$ 393.17	2974, 1794, 1691, 1464, 1420, 1402, 1352, 1158, 1092, 833, 774, 762, 733 (neat)	1.4465	1.3700	4.7 (5H, bm, CH_2CHCH_2); 3.1 (3H, s, CH_3N)	157.5 ($NCOCF_3$, J_{CCF} = 40 Hz); 156.0 ($OCOCF_3$, J_{CCF} = 40 Hz); 115.9 ($NCOCF_3$, J_{CF} = 289.4 Hz); 114.5 ($OCOCF_3$, J_{CF} = 289.4 Hz); 63.25 (CH_2O); 52.75 (CHN); 30.29 (NCH_3)

SER	143–146***	$C_9H_6F_9NO_5$ 379.14	3295, 3110, 2982, 2895, 1786, 1709, 1565, 1464, 1394, 1342, 1226, 1152, 989, 953, 919, 897, 882, 845, 800, 773 (neat)	1.6175***	1.3690***	9.7 (1H, bd, NH); 4.6 (5H, bm, CH_2CHCH_2)	156.86 ($NCOCF_3$, J_{CCF} = 37.4 Hz); 156.22 ($OCOCF_3$, J_{CCF} = 42.2 Hz); 115.5 ($NCOCF_3$, J_{CF} = 287.6 Hz); 114.2 ($OCOCF_3$, J_{CF} = 285.5 Hz); 65.29 (CH_2O); 47.17 (CHN)
ISO	149–150§	$C_9H_6F_9NO_5$ 379.14	3318, 3100, 2960, 1787, 1712, 1667, 1558, 1435, 1350, 1168, 799, 775, 731, 706 (neat)	–	–	9.75 (1H, bd, NH); 5.5 (1H, bs, CHO); 4.7 (2H, m, CH_2O); 3.7 (2H, bm, CH_2N)	157.35 ($OCOCF_3$, J_{CCF} = 40 Hz); 136.30 ($NHCOCF_3$, J_{CCF} = 40 Hz); 119.96 ($NCOCF_3$, J_{CF} = 289.4 Hz); 115.91 ($OCOCF_3$, J_{CF} = 289.4 Hz); 73.74 (CHO); 65.98 (CH_2O); 38.58 (CH_2NH)
DAP	142–143§§ (m.p.)	$C_9H_7F_9N_2O_4$ 378.15	3310, 3115, 1786, 1696, 1563, 1447, 1407, 1388, 1356, 1316, 1254, 1212, 1170, 1049, 976, 930, 918, 886, 850, 827, 715, 737, (KBr)	–	–	9.5 (1H, bd, NH); 4.6 (1H, m, CHN); 4.4 (2H, bd, CH_2O); 3.5 (2H, bs, CH_2N)	156.9 ($NCOCF_3$, J_{CCF} = 30 Hz); 157.2 ($OCOCF_3$, J_{CCF} = 30 Hz); 115.96 ($NCOCF_3$, J_{CF} = 289.4 Hz); 114.37 ($OCOCF_3$, J_{CF} = 289.4 Hz); 66.61 (CH_2O); 48.12 (CHNH); 39.23 (CH_2NH)

* Determined at 15 mmHg and uncorrected.
** Satisfactory microanalyses obtained: C ± 0.23, H ± 0.27, N ± 0.21, F ± 0.18%.
*** Lit.[5]: b.p. = 125°C (3 mmHg), d_{20} = 1.6175, n_D^{20} = 1.3693.
§ After distillation the product solidified (m.p. 62–63°C).
§§ From diethyl ether.

TABLE II

GC MS (EI) FRAGMENTATION OF AMINO ALCOHOL TRIFLUOROACETYL DERIVATIVES

Relative abundances (%).

Ion	*m/z (%)*						
	AP	*ETH*	*OME**	*NME*	*SER*	*ISO*	*DAP*
M	267(<1)	253(<1)	297(<1)	393(4)	379(<1)	379(<1)	378(<1)
M + 1**	268(<1)	254(1)	298(1)	394(<1)	380(1)	380(1)	379(1)
$M-CH_3$	253(<1)	–	–	–	–	–	–
M−F	–	–	278(<1)	374(5)	360(6)	380(2)	359<1)
$M-CF_3$	198(5)	184(20)	228(8)	324(9)	310(2)	310(1)	309(3)
$M-OCH_3$	–	–	266(1)	–	–	–	–
$M-CH_3OH$	–	–	265(<1)	–	–	–	–
$H-COCF_3$	–	156(1)	–	296(1)	282(<1)	282(<1)	281(<1)
$M-CO_2CF_3$	154(16)	140(50)	184(34)	280(32)	266(68)	266(24)	265(41)
$M-CO_2HCF_3$	153(5)	139(83)	183(5)	279(21)	265(36)	265(58)	264(33)
$M-CH_2OCOCF_3$	140(100)	126(100)	170(41)	266(99)	252(83)	252(2)	251(13)
$C_2H_4NHCOCF_3$	140(100)	–	140(7)	140(9)	140(8)	140(65)	140(50)
$CH_2NHCOCF_3$	126(7)	126(2)	126(3)	126(2)	126(30)	126(100)	126(59)
CO_2CF_3	113(8)	113(3)	113(2)	113(1)	113(18)	–	113(4)
$M-CO_2CF_3-CO_2HCF_3$	–	–	–	166(28)	152(79)	152(45)	151(17)
$M-CH_2OCOCF_3-CH_2HCF_3$	–	–	–	152(100)	138(100)	138(8)	137(100)
$M-CH_2OCOCF_3-CF_3$	–	–	–	210(3)	106(48)	196(9)	195(14)
$M-CO_2HCF_3-COCF_3$	–	–	–	182(1)	168(9)	168(9)	167(3)

* Base peak *m/z* 45.

** Confirmed by CI.

RESULTS

The separation achieved by GC analysis of serinol containing other amino alcohols after derivatization is shown in Fig. 1. Material corresponding to each peak, analysed in conjunction with the mass spectrometer, showed, in addition to a molecular ion, the typical fragmentation pattern of N,O-trifluoroacetyl derivatives[9]. The same fragmentations were observed by GC–MS (EI) analysis of the trifluoroacetyl derivatives obtained preparatively. Table II gives the results.

The quantitation of serinol and amino alcohol contaminants was subsequently examined. The precision of the method was characterized by the standard deviation (S.D.) obtained from five replicate analyses and the accuracy was characterized as the percentage error (Δ%) in the analyses of six samples (A F) with different predetermined compositions (Table III).

CONCLUSION

The procedure described represents a simple, accurate and precise method that allows the quantitation of serinol and amino alcohol contaminants. There is no *a priori* reason why the method should not also be valid outside the composition range described. Further, to our knowledge, except with serinol, none of the trifluoroacetyl derivatives, which have been fully characterized here, have been reported previously.

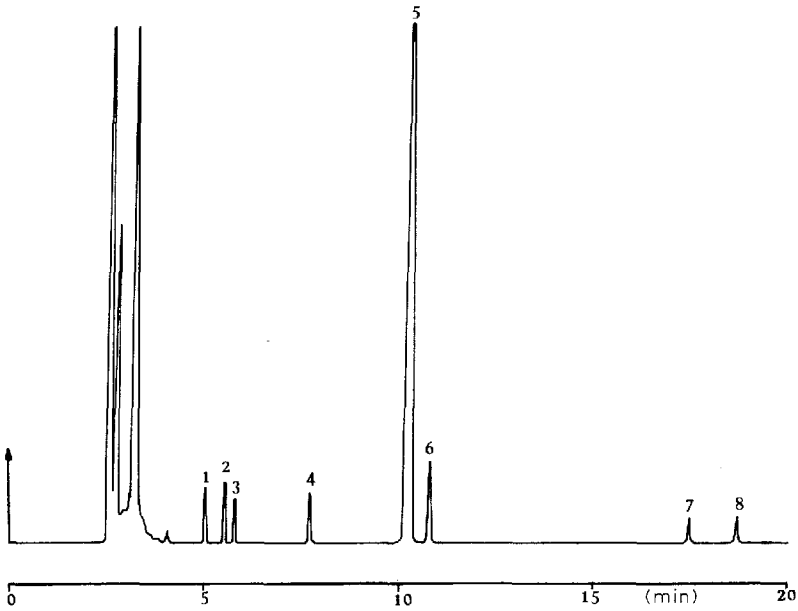

Fig. 1. Gas chromatogram of a sample of serinol and its impurities after derivatization. Peaks and retention times: 1 = 2-aminopropanol (5.003 min); 2 = 2-ethanolamine (5.535 min); 3 = 1-methoxy-2-aminopropanol (5.821 min); 4 = 2-aminomethyl-1,3-propanediol (7.776 min); 5 = serinol (10.404 min); 6 = 1-amino-2,3-propanediol (10.861 min); 7 = 2,3-diaminopropanol (17.486 min); 8 = methyl palmitate (18.789 min).

TABLE III

ACCURACY AND PRECISION OF ANALYTICAL PROCEDURE

Amino alcohol	*Parameter*	*Sample*					
		A	*B*	*C*	*D*	*E*	*F*
AP	Content (mg)	1.155	1.991	1.991	3.270	0.209	0.852
	Found ± S.D. (mg)	1.221 ± 0.004	1.465 ± 0.010	2.128 ± 0.005	3.438 ± 0.022	0.022 ± 0.002	0.912 ± 0.007
	Δ%	5.74	4.78	6.88	5.14	5.26	7.04
ETH	Content (mg)	2.207	3.071	0.249	0.687	1.216	1.565
	Found ± S.D. (mg)	2.301 ± 0.016	3.259 ± 0.026	0.271 ± 0.001	0.723 ± 0.010	1.314 ± 0.047	1.676 ± 0.012
	Δ%	4.26	6.12	8.84	5.24	8.06	7.00
OME	Content (mg)	1.577	2.031	3.003	0.282	0.615	1.017
	Found ± S.D. (mg)	1.668 ± 0.006	2.147 ± 0.016	3.158 ± 0.023	0.307 ± 0.004	0.672 ± 0.002	1.082 ± 0.014
	Δ%	5.77	5.71	5.16	8.87	9.27	6.29
NME	Content (mg)	3.263	0.255	0.641	1.115	1.531	2.233
	Found ± S.D. (mg)	3.488 ± 0.011	0.278 ± 0.003	0.695 ± 0.014	1.194 ± 0.008	1.641 ± 0.052	2.412 ± 0.0215
	Δ%	6.90	9.02	8.91	7.09	7.18	8.02
SER	Content (mg)	200.40	201.40	225.70	214.60	202.10	197.50
	Found ± S.D. (mg)	201.90 ± 0.004	203.73 ± 0.008	216.10 ± 0.010	216.10 ± 0.012	203.41 ± 0.002	198.64 ± 0.006
	Δ%	0.75	0.66	0.79	0.70	0.65	0.58
ISO	Content (mg)	0.255	0.711	1.134	1.670	2.342	3.454
	Found ± S.D. (mg)	0.268 ± 0.009	0.767 ± 0.003	1.189 ± 0.020	1.770 ± 0.022	2.484 ± 0.032	3.636 ± 0.018
	Δ%	5.10	5.06	4.89	5.99	6.06	5.27
DAP	Content (mg)	0.691	0.988	1.426	2.259	3.312	0.310
	Found ± S.D. (mg)	0.727 ± 0.008	1.047 ± 0.039	1.497 ± 0.022	2.365 ± 0.031	3.462 ± 0.013	0.327 ± 0.015
	Δ%	5.21	5.97	4.98	4.47	4.53	5.48

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