

CHROM. 16,051

GAS CHROMATOGRAPHIC DETERMINATION OF INORGANIC ANIONS AS PENTAFLUOROBENZYL DERIVATIVES

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(Received June 13th, 1983)

SUMMARY

Simple and rapid procedures were developed for the determination of cyanide, thiocyanate, nitrite and iodide as their pentafluorobenzyl derivatives. The inorganic anion was dissolved in alkaline or acidic aqueous solution and reacted with pentafluorobenzyl bromide in acetone or ethanol. The derivative formed in the reaction solution was directly analysed by gas chromatography with flame-ionization detection. The effects of added base or acid, amount of pentafluorobenzyl bromide, reaction temperature, solvent and reaction time on the derivatization of each anion are discussed. The derivatives from the anions were identified mainly by direct gas chromatography-mass spectrometry and/or by comparison with authentic samples. The derivatives of cyanide, iodide and nitrite were pentafluorobenzyl cyanide, pentafluorobenzyl iodide and α -nitro-2,3,4,5,6-pentafluorotoluene, respectively. Thiocyanate was not converted into pentafluorobenzyl thiocyanate, but into bis(pentafluorobenzyl) sulphide.

INTRODUCTION

Much analytical work has been carried out involving the use of chemical derivatization prior to analysis. This procedure is especially used in the determination of compounds that are not accessible to direct gas chromatography (GC). Several reviews have been published on this topic¹⁻⁵. However, reports of the application of the technique to the determination of inorganic anions are relatively few⁵. We have developed several approaches for the determination of inorganic anions by GC with derivatization⁶⁻¹¹. In the method described in our previous papers^{6,7}, cyanide (CN⁻), thiocyanate (SCN⁻), bromide (Br⁻), iodide (I⁻) and sulphide (S²⁻) are converted into

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their methyl or ethyl derivatives with dimethyl or diethyl sulphate, and the resulting derivatives are determined by GC with flame-ionization detection (FID). Because of the low sensitivity of FID to methyl or ethyl derivatives of the anions, it is important to select a highly sensitive detector for their determination at low concentrations. For CN^- and SCN^- , a flame thermionic detector which is highly sensitive to nitrogen-containing compound is used⁸; for Br^- and I^- , an electron-capture detector selective to halogen-containing compounds⁹ and for S^{2-} a flame photometric detector specific for sulphur-containing compounds¹⁰ can be used. These methods are used because the alkyl moieties introduced into these derivatives are not highly sensitive to any single available detector.

We have therefore further investigated the derivatization of inorganic anions into their pentafluorobenzyl (PFB) derivatives, which give very high electron-capture detection (ECD) responses. Alkylation with pentafluorobenzyl bromide (PFB-Br) is a useful technique in GC determination with derivatization, not only because of the high ECD response but also to high volatility of PFB derivatives¹². This derivatization technique has frequently been utilized for the GC determination of organic compounds, such as fatty acids, phenols and mercaptans¹³. However, there is no report on the application of pentafluorobenylation to the GC determination of inorganic anions, except for our recent paper¹¹. In that study, we reported a highly sensitive GC method for the determination of S^{2-} which was derivatized to bis(pentafluorobenzyl) sulphide.

In this work we studied the possibility of applying pentafluorobenylation to the determination of other common anions. The anions were converted into their PFB derivatives with PFB-Br, and the resulting PFB derivatives were determined by GC with FID. The final objective was to investigate the applicability of this method to the determination of anions in various samples, especially environmental and biological matrices, by using ECD.

EXPERIMENTAL

GC conditions

A Varian 1740 gas chromatograph equipped with a dual FID system was used. The column was a coiled glass tube (1.8 m × 3 mm I.D.) packed with 5% SE-30 on Chromosorb W AW DMCS (60–80 mesh). The injection port, detector and column temperatures were varied with the anions and are shown in Table I. Nitrogen was

TABLE I
GC INJECTION PORT, DETECTOR AND COLUMN TEMPERATURES

Anion	Temperature (°C)		
	Injection port	Detector	Column
CN^-	250	270	145
SCN^-	250	270	160
NO_2^-	160	220	95
I^-	160	220	95

used as the carrier gas at a constant flow-rate of 30 ml/min for all determination. A Varian A-20 recorder was used with a chart speed of 4.2 mm/min (10 in./h).

Materials

α -Bromo-2,3,4,5,6-pentafluorotoluene (PFB-Br) (Aldrich, Milwaukee, WI, U.S.A.), 1,3,5-tribromobenzene, dibenzyl and *p*-dibromobenzene (Wako, Osaka, Japan) and 5% SE-30 on Chromosorb W AW DMCS (60–80 mesh) (Gasukuro Kogyo, Tokyo, Japan) were used without further treatment. Potassium thiocyanate (E. Merck, Darmstadt, F.R.G.), potassium cyanide, sodium nitrite and sodium iodide (Wako), acetone, ethanol and other reagents were of analytical-reagent grade. Deionized, distilled water was used to prepare the aqueous solutions used. A solution of the internal standard (I.S.) was prepared by dissolving a suitable amount of each I.S. in acetone or ethanol as shown in Table II.

Procedure

A 0.9-ml volume of I.S. solution (except that for nitrite, where 0.4 ml was used) and a suitable amount of PFB-Br (see Table II) were added to 0.1 ml of an aqueous solution of each anion reference standard in a 10-ml glass-stoppered test-tube. The reaction mixture was shaken at the indicated temperature for a fixed reaction time as shown in Table II. At the end of the reaction, an aliquot of the reacted solution was subjected to GC.

RESULTS AND DISCUSSION

The pentafluorobenzoylation of several common inorganic anions was tried, except for S^{2-} , reported previously. The derivatization reaction was carried out in a system consisting of the aqueous anion solution and water-miscible organic solvent. It was found that four anions (CN^- , SCN^- , I^- and NO_2^-) give additional peaks on the chromatograms of their reaction mixtures compared with those obtained from the blank solutions excluding the anions. Therefore, these four anions were investigated further.

In order to obtain the optimum derivatization conditions for the pentafluoro-

TABLE II
DERIVATIZATION CONDITIONS

Anion	Internal standard	Solvent	Alkaline* aq. soln.	Reaction temperature	Reaction time (min)	Amount of PFB-Br (μ l)
CN^-	Tribromobenzene (0.68 mM)	Acetone	0.015 M KOH	50°C	60	5
SCN^-	Dibenzyl (0.37 mM)	Ethanol	0.15 M KOH	Room temp.	100	5
NO_2^-	Dibromobenzene (1.43 mM)	Acetone	10^{-5} M NaOH	50°C	90	1
I^-	Dibromobenzene (1.41 mM)	Acetone	0.005 M NaOH	Room temp.	60	4

* Alkaline aqueous solution used for preparation of reference standard solution.

robonylation of each anion, the effects of the concentration of base or acid, the amount of PFB-Br, solvent, reaction temperature and reaction time on the formation of the derivatives were investigated by using the peak-height ratio of the derivative to the I.S.

Effect of base or acid

As shown in Fig. 1a, for the optimum derivatization of CN^- , the concentration of the aqueous potassium hydroxide solution used should be around 0.015 M. Also as shown in Fig. 1a, the optimum concentration of potassium hydroxide is in the range 0.1–0.2 M for SCN^- . Fig. 1b demonstrates that a higher derivatization yield of I^- or NO_2^- can be obtained when the aqueous solution containing the anion covers the range from weakly acidic to weakly alkaline. In examining the effect of an alkaline solution on the derivatization of I^- , the peak from the reagent blank overlaps with that of *p*-dibromobenzene (I.S.) when the concentration of aqueous sodium hydroxide used is over 0.15 M. Therefore, *p*-nitrobromobenzene is utilized instead of *p*-dibromobenzene as the I.S. for the investigation of alkaline concentrations stronger than 0.15 M.

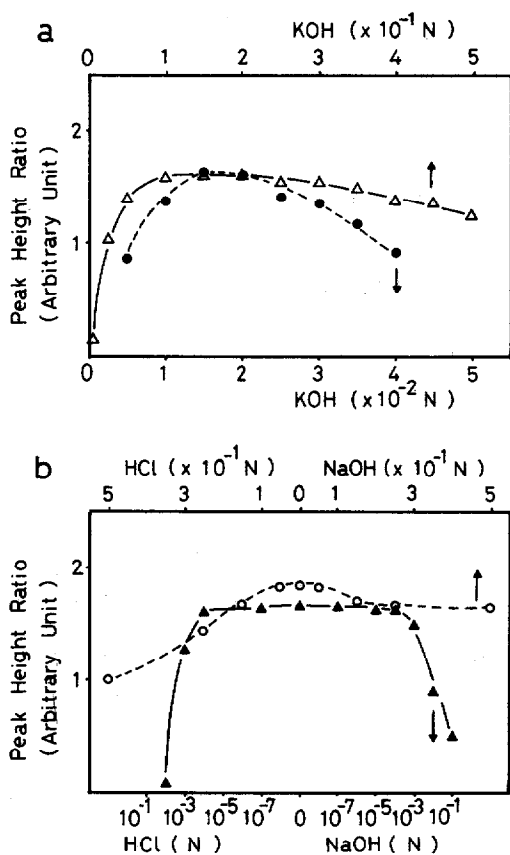


Fig. 1. Effect of base or acid on the formation of the derivatives (a) ●, CN^- ; △, SCN^- . (b) ○, I^- ; ▲, NO_2^- .

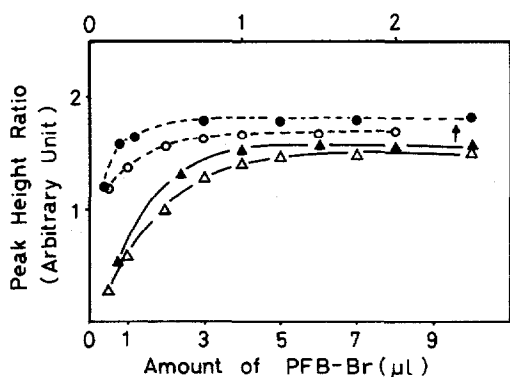


Fig. 2. Effect of amount of PFB-Br on the formation of the derivatives. ●, CN^- ; △, SCN^- ; ○, I^- ; ▲, NO_2^- .

Effect of amount of PFB-Br

To investigate the optimum amount of PFB-Br required for pentafluorobenzoylation of each anion, different levels of PFB-Br were tried for the derivatization. To achieve constant formation of the derivatives of CN^- and SCN^- , the amounts of PFB-Br required are about 3 and 5 μl , respectively (Fig. 2). Similarly, the volumes of PFB-Br required for the constant derivatization of NO_2^- and I^- are about 1 and 4 μl , respectively, also as indicated in Fig. 2.

Effects of solvent and reaction temperature

Several water-miscible organic solvents were screened for the derivatization of each anion, which were dissolved in suitable aqueous solutions of base or acid. Among the solvents acetone, acetonitrile, ethanol and methanol examined, acetone was the best choice for the derivatization of CN^- , I^- and NO_2^- , and ethanol for the derivatization of SCN^- .

In the study of the effect of reaction temperature, derivatization yields of CN^- and NO_2^- were better at 50°C than at room temperature. Owing to the relatively low boiling point of acetone, the effects of reaction temperatures higher than 50°C were not investigated. For the derivatization of I^- , the yield of the derivative obtained at 50°C was nearly identical with that obtained at room temperature. In contrast, for

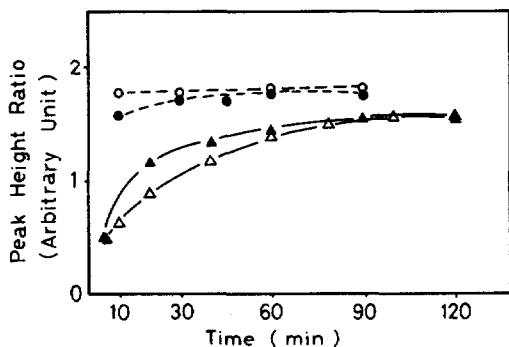


Fig. 3. Effect of reaction time on the formation of the derivatives. ●, CN^- ; △, SCN^- ; ○, I^- ; ▲, NO_2^- .

the derivatization of SCN^- , a lower reaction temperature (room temperature) is more favourable than 50°C .

Effect of reaction time

The reaction time required for maximum formation of the derivative of each anion was examined. As shown in Fig. 3, the optimum reaction times are 60 min at 50°C for CN^- and 100 min at room temperature for SCN^- . Similarly, also as shown in Fig. 3, 90 min at 50°C are necessary for maximum derivative formation for NO_2^- , and the faster equilibrium for that of I^- can be expected from the near parallel graph of the derivatization yield for I^- against reaction time.

Analytical calibration

After the optimum derivatization conditions for each anion had been established, the quantitative application of the method to the determination of CN^- , SCN^- , NO_2^- or I^- was evaluated. Good linearity for the determination of both CN^- over a range of about 1–20 μg and SCN^- over a range of about 1–60 μg was obtained, as shown in Fig. 4a. Good linearity for quantitation of both NO_2^- and I^- over ranges of about 7–133 and 5–125 μg , respectively, was also obtained (Fig. 4b). Simultaneous

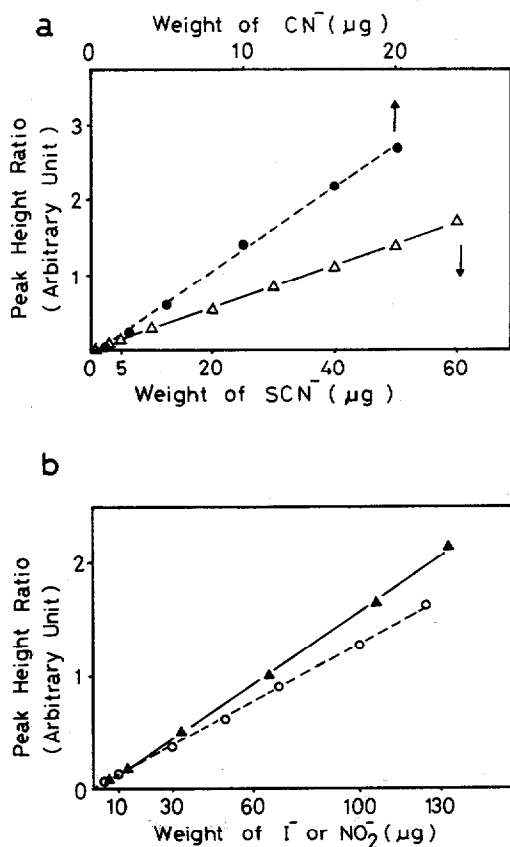


Fig. 4. Calibration graphs. (a) ●, CN^- ; △, SCN^- . (b) ○, I^- ; ▲, NO_2^- .

resolution of these four derivatives by GC is possible with a temperature programming technique.

Structure of the derivatives

To elucidate the structures of the derivatives represented by peaks 2 and 4 in Fig. 5a and b, GC-mass spectrometry (MS) was carried out using a Hitachi M-52 mass spectrometer linked to a Hitachi M-5201 gas chromatograph with a separator temperature of 260°C, an injection-source temperature of 200°C, an electron energy of 20 eV and an acceleration energy of 3.5 kV. The mass spectra obtained for the derivatives of CN^- and SCN^- exhibited parent ions of $m/e = 207$ and 394, respectively. The former represents the formation of pentafluorobenzyl cyanide from CN^- , whereas the latter precludes the formation of pentafluorobenzyl thiocyanate ($m/e = 239$) expected as the derivative of SCN^- . The pentafluorobenylation product of SCN^- was identified as bis(pentafluorobenzyl) sulphide by the complete agreement of its mass spectrum and the retention time with those of an authentic sample prepared¹¹. In addition, GC-MS using a JEOL JMS-D100 instrument was used to examine peaks 5 and 7 in Fig. 5c and d at ionization temperature 200°C, electron energy 75 eV and acceleration energy 3 kV. The mass spectra obtained exhibited fragment ions at $m/e = 181$ (equivalent to $\text{C}_6\text{F}_5\text{CH}_2^+$) and $m/e = 127$ (I^+), but no parent ion at $m/e = 308$ was detected for pentafluorobenzyl iodide. Then pentafluorobenzyl iodide was synthesized, isolated and identified by MS at an ionization source temperature of

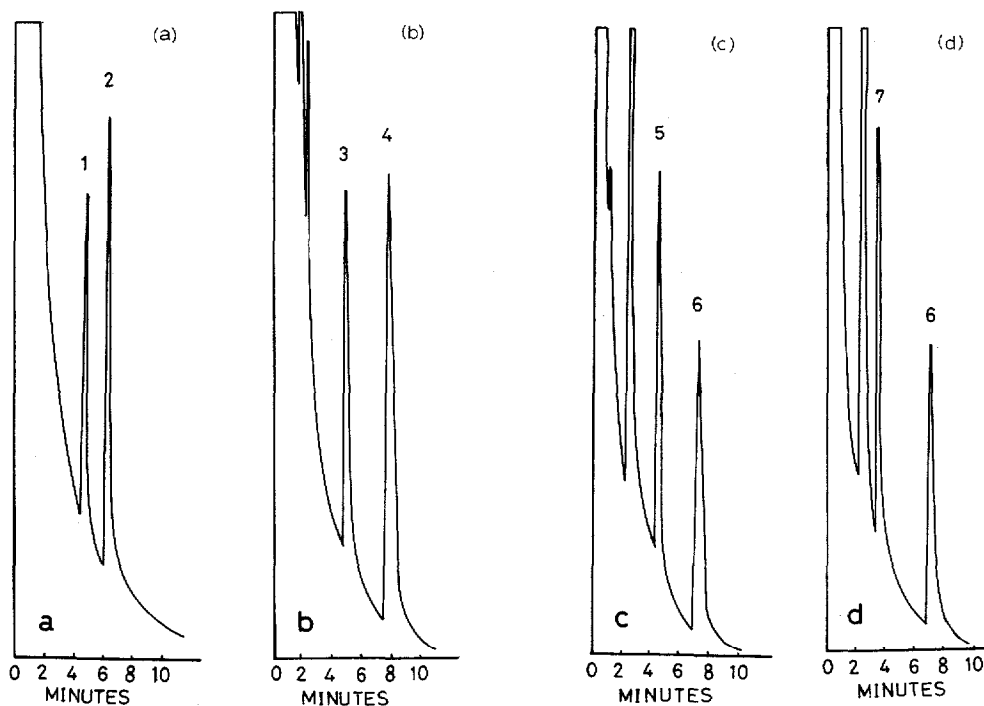


Fig. 5. Typical gas chromatograms for determination of (a) CN^- , (b) SCN^- , (c) I^- and (d) NO_2^- . Peaks: 1 = tribromobenzene (I.S.); 2 = derivative of CN^- ; 3 = dibenzyl (I.S.); 4 = derivative of SCN^- ; 5 = derivative of I^- ; 6 = dibromobenzene (I.S.); 7 = derivative of NO_2^- .

90°C, an electron energy of 12 eV and an acceleration energy of 3 kV. In this instance a parent ion peak of $m/e = 308$ was observed. The pentafluorobenylation product of I^- was identified as pentafluorobenzyl iodide by both the MS and GC experiments, based on a comparison of the retention times.

The mass spectrum obtained from peak 7 in Fig. 5d, examined under the same conditions as for the identification of the iodide derivative, exhibited as base peak at $m/e = 181$, equivalent to $M - NO_2^+$, but no molecular peak could be detected. Therefore, the nitrite derivative was also synthesized, isolated and identified by MS (Hitachi RMU-6E) at an electron energy of 70 eV. A parent ion peak at $m/e = 227$ appeared in the mass spectrum. Further analysis of the synthesized nitrite derivative was performed by IR spectrometry, the presence of IR absorption peaks at 1560 and 1364 cm^{-1} corresponding to the antisymmetric and symmetric vibrations, respectively, of the nitro group in the derivative. Therefore, based on the results of MS and IR analyses, the derivative of nitrite is identified as α -nitro-2,3,4,5,6-pentafluorotoluene.

The pentafluorobenylation of the anions is an ECD-oriented derivatization designed to enhance the sensitivity. The development of the present method for the trace determination of the anions in various matrices by GC with ECD is in progress.

ACKNOWLEDGEMENT

We are very grateful to the National Science Council, Republic of China, for financial support of part of this work (NSC 71-0204-B012-01).

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