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ALCOHOLIC BEVERAGES: DISTILLATION

See **III / WHISKY: DISTILLATION**

ALDEHYDES AND KETONES: GAS CHROMATOGRAPHY

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Introduction

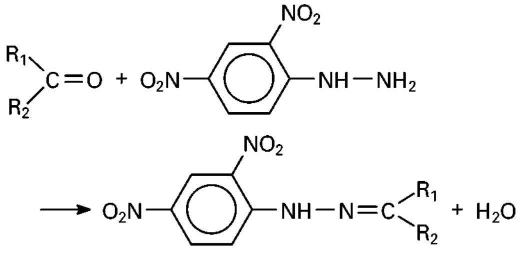
Simple aldehydes, such as formaldehyde, acetaldehyde and acrolein, are known to be hazardous air pollutants. Aldehydes are emitted from incomplete burning of various organic compounds and from various chemicals, and are formed by photochemical reaction with hydrocarbons in the atmosphere.

Volatile ketones are used as solvents in various chemical plants and laboratories and are emitted into the atmosphere. The toxicity of ketones is, in general, not as high as that of aldehydes. Carbonyl compounds are significant in environmental chemistry, i.e. in rainwater and as a photochemical oxidant.

Separation of aldehydes and ketones is very important for the determination of volatile aldehydes. Usually, analysis of aldehydes is performed by derivatization and gas chromatography (GC) or high performance liquid chromatography (HPLC). Selective and sensitive gas chromatographic methods for separation of aldehydes and ketones are described below.

2,4-Dinitrophenylhydrazone Derivatization

2,4-Dinitrophenylhydrazone (DNPH) derivatives of aldehydes and ketones have been used in gas chromatography for many years. The reaction procedure of aldehyde or ketone is as follows:



Kallio *et al.* analysed 15 carbonyl compounds (aldehydes and ketones) known to be flavour components by derivatization/GC with DNPH. The DNPHs of the carbonyl compounds were prepared by shaking 100 μL of each compound with 100 mL of a saturated solution of DNPH in aqueous 2 mol L^{-1} hydrochloric acid and allowing the mixture to stand at room temperature overnight. The precipitated DNPHs were dissolved in ethyl acetate, then analysed by GC-FID or dissolved in benzene and analysed by GC-ECD (electron-capture detector). Packed columns with silicone stationary phases were used. Relative retention times of DNPHs of aldehydes and ketones on one of these columns are listed in **Table 1**

Table 1 Relative retention times (*R*) of 2,4-dinitrophenylhydrazones of carbonyl compounds

Carbonyl compound	*2% SE-30 column 200–270°C*[a] *R*
Formaldehyde	0.52
Acetaldehyde	0.71
Propenal	0.84
Acetone	0.84
Propanal	0.86
2-Methylpropanal	0.94
2-Butanone	1.00
3-Methylbutanal	1.13
2-Butenal	1.16
Hexanal	1.42
Furfural	1.62
	1.58[b]
Heptanal	1.63
Octanal	1.83
Benzaldehyde	1.97
Nonanal	2.03

[a] Perkin-Elmer F-11 gas chromatograph equipped with a coiled glass column, 6 ft long and 1/8 in i.d.

[b] Relative retention times of secondary peaks.

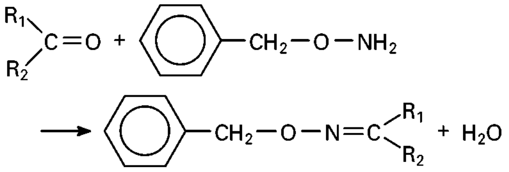
Reproduced with permission from Kallio H *et al.* (1972) Gas–liquid chromatographic analysis of 2,4-dinitrophenylhydrazones of carbonyl compounds. *Journal of Chromatography* 65: 355.

and a typical chromatogram of DNPHs of carbonyl compounds is shown in **Figure 1**. The peaks of the DNPHs of propanal, propenal and acetone are not separated under these conditions. The sensitivity was of the order of nanograms of carbonyl compounds when FID was used and five-hundred times greater with an ECD.

Saito *et al.* reported an improved GC method with DNPH for the determination of trace low molecular weight aliphatic carbonyl compounds in auto exhaust. They used a glass capillary column, 30 m × 0.27 mm i.d., coated with OV-17 and operated isothermally at 210°C. Aldehydes and ketones in the exhaust were selectively collected by passing 600 ml min^{-1} of exhaust gas through two impingers which were connected in series and which contained hydrochloric acid saturated with DNPH. The derivatives were extracted twice with chloroform in a separating funnel. After concentration by evaporation under a stream of nitrogen, anthracene was added as internal standard, and 1 μL of the solution was injected into a GC equipped with an FID. Six aliphatic aldehydes and three aliphatic ketones were analysed (**Figure 2**). The derivatives of C_3 carbonyl compounds, propionaldehyde, acetone and acrolein, were completely separated, and simultaneously determined with formaldehyde and acetaldehyde. The minimum detectable concentrations of formaldehyde, acetaldehyde and acrolein were 85, 140 and 190 ppb, respectively, for a 10 L gas sample with a relative standard deviation of less than 8%. Using this method, formaldehyde, acetaldehyde, propionaldehyde, isobutyraldehyde, acetone, acrolein, methyl ethyl ketone and methyl isopropyl ketone (or butyraldehyde) were measured in gasoline engine exhaust gas.

Benzyloxime Derivatization

Derivatization of simple aldehydes and ketones to benzyloximes was developed by Magin. The reaction proceeds as follows:



The GC used for the analysis of these derivatives was equipped with a nitrogen-selective detector and

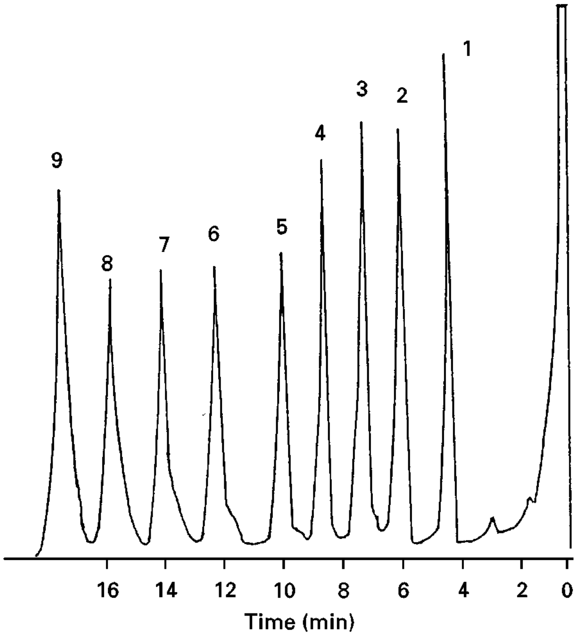


Figure 1 Chromatogram of a mixture of 2,4-dinitrophenylhydrazones of carbonyl compounds on a 2% SE-30 column. 1, Formaldehyde; 2, acetaldehyde; 3, acetone; 4, 2-butanone; 5, 2-butenal; 6, hexanal; 7, heptanal; 8, octanal; 9, nonanal. Programmed temperature analysis from 200 to 270°C (4°C min^{-1}) on a Perkin-Elmer F-11 chromatograph equipped with a hydrogen flame ionization detector. Injected sample: 1.0 μL of ethyl acetate containing about 1000 ng of each of the derivatives. Attenuation 128, range 1. (Reproduced with permission from Kallio H *et al.* (1972). Gas–liquid chromatographic analysis of 2,4-dinitrophenylhydrazones of carbonyl compounds. *Journal of Chromatography* 65: 355.)

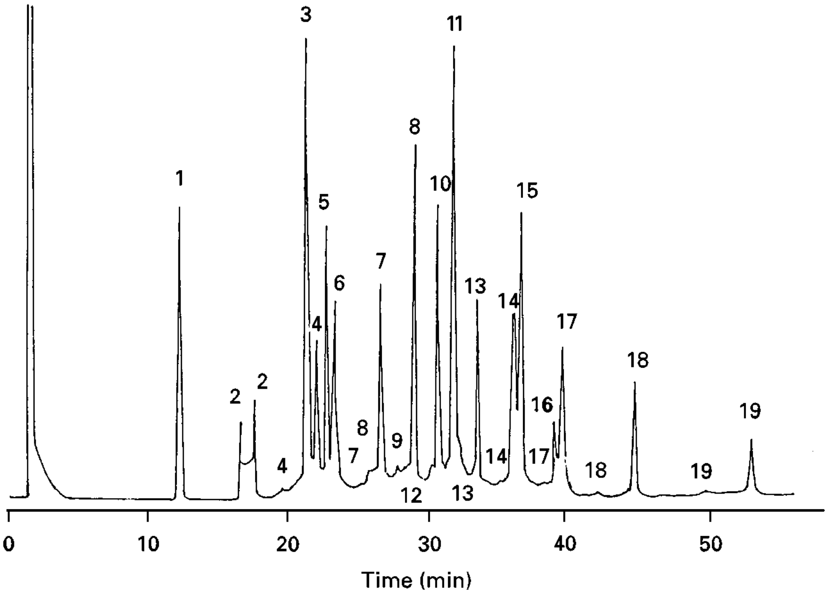


Figure 2 Chromatogram of the 2,4-dinitrophenylhydrazones of carbonyl compounds. 1, Formaldehyde; 2, acetaldehyde; 3, acetone; 4, propionaldehyde; 5, isobutyraldehyde; 6, acrolein; 7, methyl ethyl ketone; 8, butryraldehyde; 9, methyl isopropyl ketone; 10, diethyl ketone; 11, methyl *t*-butyl ketone; 12, isovaleraldehyde; 13, methyl propyl ketone; 14, methyl *s*-butyl ketone; 15, methyl isobutyl ketone; 16, valeraldehyde; 17, crotonaldehyde, 18, methyl butyl ketone; 19, capronaldehyde. (Reproduced with permission from Saito T *et al.* (1983) Determination of trace low molecular weight aliphatic carbonyl compounds in auto exhaust by gas chromatography with a glass capillary column. *Bunseki Kagaku* 32: 33.)

a glass capillary column (12 m × 0.4 mm i.d.) coated with a 0.4 μm film of free fatty acid phase (FFAP); helium was used as the carrier gas. The column temperature programme was 100–180°C at 2°C min^{-1}; the temperature was kept at 180°C to the end of the analysis. The retention times of the benzyloximes of a number of aldehydes and ketones (1–7 carbon atoms) are shown in **Table 2**. The results show almost complete separation under these conditions.

Magin also reported the application of the benzyloxime-GC analysis of aldehydes and ketones to the semi-quantitative analysis of simple monocarbonyls in cigarette smoke. The smoke was passed through a silica gel column to trap the carbonyls, followed by elution with water. About 15 mL of eluted solution was collected in a screw-capped bottle, and the benzyloximes of the carbonyls were prepared. Separation was accomplished by a temperature-programmed 12 m glass capillary FFAP column. An internal standard (hexanal) was added both as a reference for retention time determination, and as an aid in estimating the amounts of the individual carbonyls in the smoke samples. Levels of some carbonyls in the cigarette whole smoke samples are shown in **Table 3**. One problem with this method was that acetaldehyde, one of the major carbonyl compounds in cigarette smoke, could not be determined since peaks corresponding to the benzyloxime derivative of acetaldehyde appeared in the reagent blanks. Mass spectra of these peaks were identical to the spectra of the genuine acetaldehyde derivatives, but the source

Table 2 Adjusted retention times of *O*-benzyloxime derivatives

Aldehydes	*Retention time (min)*	*Ketones*	*Retention time (min)*
Formaldehyde	6.5	Acetone	12.6
Acetaldehyde	9.9	2-Butanone	14.2
Propanal	11.7	2-Pentanone	16.1
Butanal	17.6		
Pentanal	22.5	3-Pentanone	16.8
Hexanal	30.0	4-Heptanone	22.1
Heptanal	36.5		
Octanal	43.5	Cyclopentanone	34.5
		Cyclohexanone	38.4
Isobutanal	13.9	Cycloheptanone	45.0
Isopentanal	20.0		
		3-Methyl-2-butanone	14.4
Propenal	15.9	3-Methyl-2-pentanone	15.4
2-Butenal	27.9		
2-Hexenal	44.0	4-Methyl-2-pentanone	15.2
Methacrolein	17.9	5-Hexen-2-one	25.9
Benzaldehyde	67.7		

Perkin-Elmer Model 3920 gas chromatograph equipped with glass capillary column, 12 m long and 0.4 mm i.d.
Reproduced with permission from Magin DF (1979) Preparation and gas chromatographic characterization of benzyloximes and *p*-nitrobenzyloximes of short-chain (C1–C7) carbonyls. *Journal of Chromatrography* 178: 219.

Table 3 Levels of selected carbonyls in the whole smoke of some cigarettes

Carbonyl	*Cigarette A (filter)*	*Cigarette B (filter, low delivery)*	*Cigarette C (non-filter)*
Formaldehyde	31 (10–50)	10 (9–10)	21 (12–30)
Acetone	400 (325–475)	137 (130–144)	330 (310–350)
Propanal	61 (37–100)	37 (30–40)	50 (50–53)
Acrolein	23 (13–37)	3 (3–4)	22 (20–25)
Methacrolein	17 (14–38)	18 (18–19)	27 (20–32)
Butanal	20 (9–29)	13 (12–13)	18 (17–20)

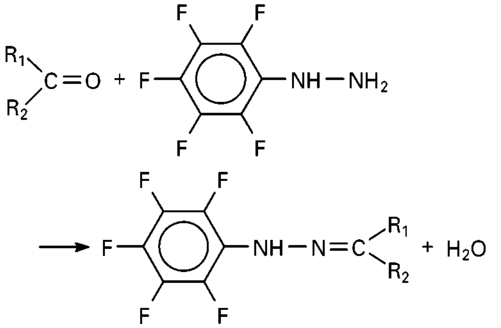
Levels are tabulated as average; values in parentheses indicate the range. The values are given in μg per cigarette.

Reproduced with permission from Magin DF (1980) Gas chromatography of simple monocarbonyls in cigarette whole smoke as the benzyloxime derivatives. *Journal of Chromatography* 202: 255.

in the blanks was unknown. Another problem was the lack of reproducibility from run to run. For this reason, the method was described as semi-quantitative.

Pentafluorophenylhydrazone Derivatization

A sensitive and selective GC analysis of lower aliphatic carbonyl compounds as their pentafluorophenylhydrazones was described by Hoshika and Muto.



The general procedure for the preparation of the pentafluorophenylhydrazone derivatives was as follows: 0.5×10^{-3} mol of lower aliphatic carbonyl compound was added, using a 100 μL microsyringe, to 1 mL methanol containing 1.01×10^{-3} mol pentafluorophenylhydrazine (PFPH). The mixture was allowed to stand overnight at room temperature. A 1 μL volume of the solution was injected into the GC. The acetone, acrolein and propionaldehyde derivatives were separated on a 30 m × 0.25 mm i.d. glass capillary column coated with polyethylene glycol 20 M at 130°C with an FID. This temperature compares favourably with the 200°C column temperature necessary with DNPH derivatives.

A procedure for the GC determination of low molecular weight carbonyl compounds in aqueous solution as their pentafluorophenylhydrazones and its application to such compounds produced by photolysis of α-amino acids were reported by Kobayashi *et al.* (**Figure** 3). The calibration graphs for formaldehyde, acetaldehyde and isobutyraldehyde showed good linearity in the range 10–40 μg. Solutions of DL-alanine, DL-valine, DL-leucine or DL-isoleucine in phosphate buffer containing mercury (II) chloride were irradiated in a quartz vessel with a 20 W blacklight lamp (300–400 nm) as a light source for 300 h and the mixtures were measured by this method. Two peaks corresponding to formaldehyde and acetaldehyde were observed for valine.

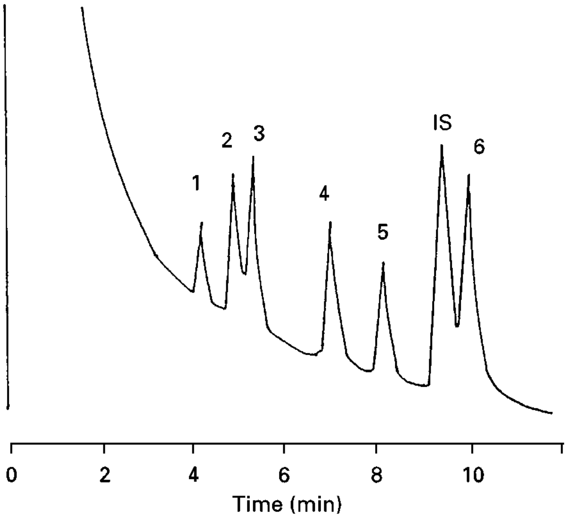
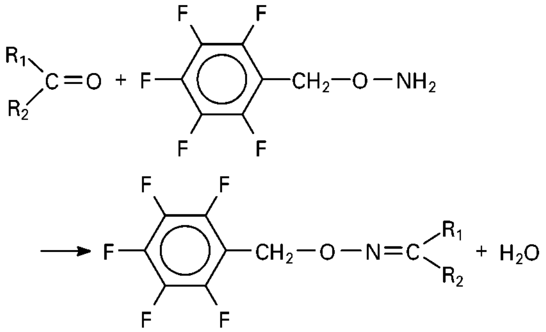


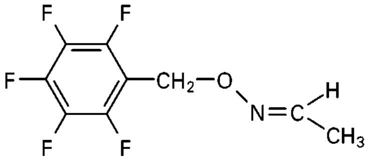
Figure 3 Chromatogram of some carbonyl compounds as their pentafluorophenylhydrazones. Conditions: 3% XE-60, 2.0 m glass column, temperature-programmed from 105 to 130°C at 2°C min^{-1}, FID. 1, Formaldehyde; 2, acetaldehyde; 3, acetone; 4, isobutyraldehyde; 5, diethyl ketone; IS, *p*-xylylene dichloride; 6, methyl isobutyl ketone. (Reproduced with permission from Kobayashi K *et al.* (1979) Gas chromatographic determination of low-molecular-weight carbonyl compounds in aqueous solution as their pentafluorophenylhydrazones. *Journal of Chromatography* 176:118.)

Pentafluorobenzyloxime Derivatization

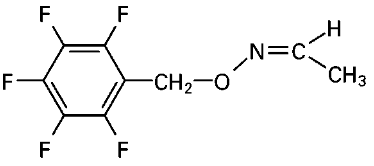
Pentafluorobenzyloxyamine (PFBOA) was synthesized by Nambara *et al.* as a new derivatizing agent for GC of ketones using electron-capture detection. Kobayashi *et al.* reported a GC analysis of low molecular weight carbonyl compounds in aqueous solution as their O-pentafluorobenzyloximes. The reaction procedure of aldehyde or ketone is as follows:



The retention times of 13 O-pentafluorobenzyloxime (O-PFBO) derivatives of carbonyl compounds relative to the internal standard obtained on XE-60 and other columns are shown in **Table 4**. When some carbonyl compounds react with PFBOA, two peaks are obtained, corresponding to syn- and anti-isomers formed by the condensation reaction with PFBOA:



syn



anti

Good linearity of response was obtained for formaldehyde, acetaldehyde, isobutyraldehyde and diethyl ketone in the range 1–50 μg in 0.5 mL of aqueous solution. The utility of PFBOA was compared with that of PFPH. The formation of the O-PFBO derivatives of aldehydes was easily achieved with a much lower concentration of PFBOA than that required for PFPH. The derivatization of aldehydes with PFBOA is complete in 20 min at room temperature, but the reaction with ketones proceeds slowly (within 24 h). The PFBO derivatives are much more volatile than the corresponding pentafluorophenylhydrazones and therefore the separation can be carried out at lower temperatures (70–100°C). It was shown that the PFBO derivatives are stable in ethyl acetate at room temperature for several days.

Table 4 Relative retention times of the *O*-pentafluorebenzyloximes of carbonyl compounds

Parent compound	*Stationary phase and column temperature (each column: 2 m × 3 mm i.d.)*			
	3% XE-60 (90°C)	*3% XF-1105 (100°C)*	*3% SE-30 (80°C)*	*2% OV-17 (70°C)*
PFBOA	0.62	0.69	0.88	0.65
HCHO	0.19	0.27	0.29	0.19
CH_3CHO	0.32, 0.34[a]	0.48, 0.51[a]	0.57, 0.59[a]	0.43
C_2H_5CHO	0.47, 0.51[a]	0.80	1.05	0.72
n-C_3H_7CHO	0.80	1.41	1.83, 1.92[a]	1.30, 1.38[a]
iso-C_3H_7CHO	0.57	1.01	1.32	0.88
n-C_4H_9CHO	2.72	1.91, 1.99[a]	2.55, 2.72[a]	1.76, 1.94[a]
CH_3COCH_3	0.40	0.70	0.88	0.65
$CH_3COC_2H_5$	0.61	1.13	1.50	1.04
CH_3CO-iso-C_3H_7	0.73	1.48	2.08	1.24, 1.43[a]
CH_3CO-iso-C_4H_9	1.16	2.36	3.38	2.08
$C_2H_5COC_2H_5$	0.85	1.74	2.43	1.61
C_2H_5CO-*n*-C_3H_7	1.27	2.70	3.95	2.61
$CH_2ClC_6H_4CH_2Cl$[b]	1.00	1.00	1.00	1.00

[a] Double peaks.

[b] Internal standard (α,α′-dichloro-*p*-xylene). PFBOA, Pentafluorobenzyloxyamine. Shimadzu Model GC-4APF gas chromatograph equipped with an FID.

Reproduced with permission from Kobayashi K *et al.* (1980) Gas chromatographic determination of low-molecular-weight carbonyl compounds in aqueous solution as their *O*-(2,3,4,5,6-pentafluorobenzyl) oximes. *Journal of Chromatography* 187: 413.

Aldehydes in air are important as pollutants and photochemical products because they are irritants to the skin, eyes and nasopharyngeal membranes. Trace amounts of formaldehyde in air was determined as the PFBO derivative using GC-ECD by Nishikawa *et al.* Sub-ppb levels of formaldehyde in air could be determined in this way. Although the FID or ECD can be used with PFBO derivatives, the FID has poor sensitivity and in some cases the peaks of the derivatives suffer interference from co-existing materials with both detectors. In the case of analysis of samples with a complex matrix such as exhaust gas or emission gas, the thermionic detector (TID) is more effective. The TID is very sensitive and selective to nitrogen compounds such as oximes and can therefore be used with advantage for the analysis of aldehydes as PFBOA derivatives in automobile exhaust and emission gas. Gas (2–30 L) was collected by bubbling the exhaust through two impingers connected in series. Each impinger contained 10 mL of the absorption solution (300 mg L^{-1} PFBOA$\cdot$HCl in ethanol). The sample gas was drawn at a rate of 0.5 L min^{-1}. After sampling, the absorbed solution was made up to 20 mL with ethanol and was allowed to stand for 80 min at room temperature. A 10 mL portion of the solution and 20 mL of distilled water were mixed well and the mixture was passed through a Sep-Pak C_{18} cartridge. The cartridge was eluted with 1.5 mL hexane and the eluate was analysed with an SE-52 fused silica column (25 m × 0.25 mm i.d.) at 130°C with an FTD. The relative retention times for aldehyde and ketone derivatives are shown in **Table 5**. Except for formaldehyde, acetone and diethyl ketone, each derivative has syn- and anti-isomers and the double peaks appear for the isomers. The peaks of the formaldehyde, acetaldehyde, propionaldehyde and butyraldehyde derivatives were completely separated from those of the ketones. Although the first peak of propionaldehyde was not separated from the first peak of acrolein, the second peak of propionaldehyde was separated from the second peak of acrolein. Therefore, it can be judged whether the first peak of propionaldehyde contained the peak of acrolein or not. The calibration graphs obtained show good linearity over the range 0–5 µg for formaldehyde, 0–4 µg for acetaldehyde, 0–15 µg for propionaldehyde and 0–13 µg for butyraldehyde in 1 mL of the absorption solution. The determination limits ranged from 14 ppb (v/v) for formaldehyde to 67 ppb for propionaldehyde when a 30 L gas sample was used.

Table 5 Relative retention times of *O*-pentafluorobenzyloxime derivatives

Parent compound	*Relative retention time*
Formaldehyde	1.00
Acetaldehyde	1.30, 1.34[a]
Propionaldehyde	1.75, 1.79[a]
Butyraldehyde	2.52, 2.58[a]
Acrolein	1.76, 1.88[a]
Crotonaldehyde	–[b]
Acetone	1.63
Methyl ethyl ketone	2.20, 2.24[a]
Methyl isopropyl ketone	2.69, 2.75[a]
Diethyl ketone	3.00
Methyl propyl ketone	3.03, 3.14[a]

GC conditions: SE-52 capillary column (25 m × 0.25 mm i.d.), temperature 130°C.

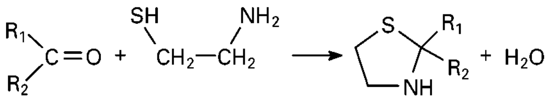
[a] Double peaks; [b] no peaks appeared.

Reproduced with permission from Nishikawa H *et al.* (1987) Determination of aldehydes in exhaust gas and thermal degradation emission using volatile derivatization and capillary GC with flame thermionic detector. *Bunseki Kagaku* 36: 381.

This method is very selective, without interference from ketones, and is well-suited to analyse simple aldehydes in various exhaust gas samples. The PFBO method was applied to analysis of carbonyl compounds in clothes, river water, seawater, tap water, indoor air samples and environmental air samples.

Thiazolidine Derivatization

Hayashi *et al.* developed a method to determine formaldehyde and methyl glyoxal in foods and beverages. This method is known as the cysteamine method. Volatile aliphatic aldehydes and ketones react with 2-aminoethanethiol (cysteamine) to form thiazolidine compound as follows:



Only one derivative is formed from each aldehyde or ketone; the reaction proceeds rapidly under mild conditions and the derivatives can be readily separated on a capillary column. The derivatives can be detected selectively with a nitrogen–phosphorus detector (NPD). Carbonyl compounds in the headspace gases above heated pork fat were analysed as thiazolidine derivatives by Yasuhara and Shibamoto (**Figure 4**). A GC with a 30 m × 0.25 mm i.d. DB-Wax fused silica capillary column and an NPD was used for the qualitative and quantitative analysis of the carbonyl compounds in the samples. The column temperature was held at 50°C for 2 min and then programmed to 190°C at 3°C min^{-1}. Nine aldehydes and four ketones were analysed as thiazolidine derivatives in traps containing aqueous cysteamine or

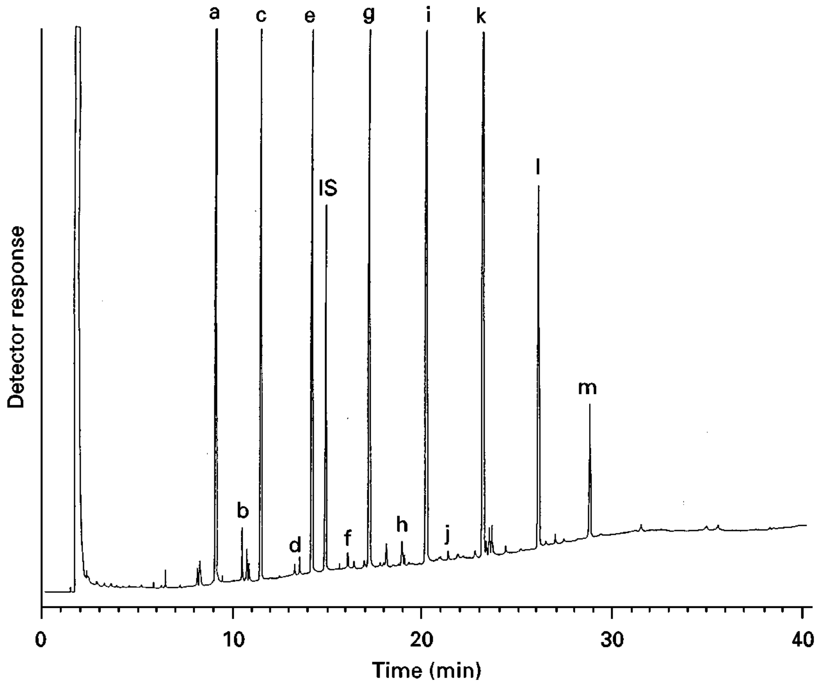


Figure 4 Chromatogram of the thiazolidine derivatives of carbonyl compounds produced from heated pork fat. a, Acetaldehyde; b, formaldehyde; c, propionaldehyde; d, 2-pentanone; e, butyraldehyde; f, 2-hexanone; g, valeraldehyde; h, 2-heptanone; i, hexylaldehyde; j, 2-octanone; k, heptaldehyde; l, octaldehyde; m, nonylaldehyde; IS, N-methyl acetamide. (Reproduced with permission from Yasuhara A (1991) Analysis of lower aldehydes in air. *Journal of Environmental Analytical Chemistry* 1: 253.)

aqueous sodium bisulfite. The major compounds produced from the samples were pentanal, hexanal and heptanal. Generally, aqueous cysteamine was more efficient at trapping carbonyl compounds than was aqueous sodium bisulfite but formaldehyde and acetaldehyde were trapped better by sodium bisulfite. Acrolein and malonaldehyde were analysed as 1-methyl-2-pyrazoline and 1-methyl-pyrazole, respectively, from the trap containing methylhydrazine. Good separation of volatile aldehydes and ketones was obtained by this method.

The cysteamine derivatization method has been applied to the determination of trace amounts of carbonyl compounds in automobile exhaust, air samples, the headspace of heated food oils, foods and beverages.

Conclusion

The DNPH-GC and PFBO-GC methods are now widely used for the derivatization and gas chromatographic analysis of aldehydes and ketones. The PFBO-GC and cysteamine-GC methods will become more common. These reactions proceed under mild conditions and the derivatives are well separated with a capillary column at comparatively low temperatures. The GC-MS measurement of these derivatives will be utilized for analysis of trace amounts of aldehydes and ketones in a variety of samples.

See also: **II/Chromatography: Gas:** Derivatization; Detectors: Selective.

Further Reading

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