# Simultaneous Determination of C<sub>1</sub>-C<sub>4</sub> Carboxylic Acids and Aldehydes Using 2,4-Dinitrophenylhydrazine-Impregnated Silica Gel and High-Performance Liquid Chromatography

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A new method for the simultaneous determination of aliphatic carboxylic acids and aldehydes in air is described. In this work, carboxylic acids were allowed to react with 2,4-dinitrophenylhydrazine (DNPH) to form the corresponding carboxylic 2,4-dinitrophenylhydrazides. These derivatives have excellent thermal stability, with melting points higher than those of the corresponding hydrazones by 32-50 °C. C<sub>1</sub>-C<sub>4</sub> carboxylic acid 2,4dinitrophenylhydrazides exhibited maximum absorption wavelengths of 331-334 nm and molar absorption coefficients of  $1.4 \times 10^4$  L/mol/cm. They were completely separated by high-performance liquid chromatography (HPLC) with an RP-Amide C16 column. Cartridges packed with DNPH-coated silica particles (DNPH cartridge) were used for sampling formic acid and aldehydes. Formic acid was physically adsorbed on the silica particles as the first step of the sampling mechanism. Gradual reaction with DNPH followed. Formic acid reacted very slowly with DNPH at room temperature (20 °C), but reacted completely at 80 °C over 4 h. In field measurements, the sample air was drawn through a DNPH cartridge. After sampling, the cartridges were heated at 80 °C for 5 h and extracted with acetonitrile for HPLC analysis. Under these optimized conditions, the LOD is 0.4 ug/m<sup>3</sup> for an air sample collected for 24 h at 100 mL/min (144 L).

Carbonyl compounds, including carboxylic acids, aldehydes, and ketones, are ubiquitous pollutants throughout the troposphere, and numerous analytical methods have been reported in many papers. The first reported analysis of aliphatic carboxylic acids was carried out using a bubbler in NaOH solution followed by silica gel column chromatography titration.<sup>1</sup> Many additional methods followed. The most common collection procedures for aliphatic carboxylic acid in air involve trapping in a liquid, such as water<sup>2</sup> or aqueous solutions of NaOH<sup>3</sup>. Sample collection may

also take place on a filter impregnated with KOH <sup>4</sup> or Na<sub>2</sub>CO<sub>3</sub>;<sup>5</sup> a solid sorbent, such as a silica gel;6 or a KOH-coated C18 cartridge.7 On the basis of the large diffusion coefficient differences between gases and particles, KOH- and Na<sub>2</sub>CO<sub>3</sub>-coated denuders can be used to collect gaseous carboxylic acids.<sup>8,9</sup> The eluted solution analysis can be performed by various chromatographic methods, including ion chromatography,<sup>10</sup> ion exclusion chromatography,<sup>11</sup> high performance liquid chromatography (HPLC),<sup>12</sup> capillary electrophoresis,<sup>13</sup> and gas chromatography (GC)<sup>4</sup>. Recently, SPME-GC/MS<sup>14</sup> has been applied to the measurement of acetic and formic acids in a museum environment.

The most commonly used method to determine aldehydes and ketones exploits the reaction of an amino (NH<sub>2</sub>) group with a carbonyl (C=O) moiety to form a stable Schiff base (C=N). Derivatization is very useful in analysis by gas chromatography or high-performance liquid chromatography. Various derivatization reagents have been used for the determination of aldehydes and ketones in air, including O-(2,3,4,5,6-pentafluorobenzyl)hydroxylamine,<sup>15</sup> O-benzylhydroxylamine,<sup>16</sup> 2-diphenylacetyl-1,3-indandione-1-hydrazone,17 5-dimethylaminonaphthalene-1-sulfohydrazide (dansylhydrazine),18 N-methyl-4-hydrazino-7-nitrobenzofurazan,19

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pentafluorophenylhydrazine,<sup>20</sup> and 2,4-dinitrophenylhydrazine (DNPH).<sup>21–23</sup> Indeed, for over 70 years, aldehydes and ketones have been characterized through their reaction with DNPH to the corresponding hydrazone derivatives.<sup>24,25</sup> The derivatives are then separated using HPLC with UV detection at a wavelength near the absorption maximums of the respective hydrazones (typically 360 nm).<sup>26,27</sup> In recent years, the DNPH-HPLC method has been established as the most widely used standard procedure for the determination of aldehydes and ketones in air samples.

In solid and liquid samples, such as foods, beverages, and biological materials, 2-nitrophenylhydrazine is widely used for the derivatization of carboxylic acids, aldehydes, and ketones;<sup>28,29</sup> however, there is no report describing simultaneous analysis of aliphatic carboxylic acids, aldehydes, and ketones in an air sample. It is advantageous to measure carboxylic acid and aldehyde levels simultaneously because they are sometimes emitted from the same sources in air. In this study, we developed a new method for simultaneous measurement of  $C_1-C_4$  aliphatic carboxylic acids and aldehydes using DNPH derivatization.

#### **EXPERIMENTAL SECTION**

**Apparatus.** The HPLC system (Shimadzu, Kyoto, Japan) used included two LC-10ADvp pumps, an SIL-10ADvp autosampler, an SPD-10Avp ultraviolet absorbance detector adjusted to 350 nm, and an SPD-M10Avp photodiode array detector. The analytical column was a 250 mm long  $\times$  4.6 mm i.d. stainless steel tube packed with RP-Amide C16, 5- $\mu$ m particle size (Supelco Inc, Bellefonte, PA). Solution A of the mobile phase mixture was acetonitrile/water, 40/60 v/v, and solution B was 60/40 v/v. HPLC was carried out with 100% A for 8 min, followed by a linear gradient from 100% A to 100% B in 22 min and then held for 15 min. The flow rate of the mobile phase was 1.5 mL/min. A Discovery C18 column, 5- $\mu$ m particle size 250 mm long  $\times$  4.6 mm i.d. (Supelco Inc, Bellefonte, PA) was used as a reference, and the RP-Amide C16 column was used for most experiments.

Melting points of the DNPH derivatives were determined by a Q10 differential scanning calorimeter (DSC, TA Instruments, New Castle, DE) under nitrogen atmosphere.

The carboxylic acid DNPH derivatives were characterized by NMR spectroscopy using a JNM-ECX400 spectrometer (JEOL Ltd., Tokyo, Japan) operating at 400 MHz.

**Reagents.** The water used in HPLC and sample preparation was deionized and purified using a Milli-Q Water System equipped with a UV lamp (Millipore, Bedford, MA). Acetonitrile was HPLC grade from Wako Pure Chemical Industries Ltd. (Osaka, Japan). Formic acid 96%, acetic acid 99.8%, propionic acid 99.5%, butyric

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acid 99%, formaldehyde (37% solution in water), acetaldehyde 99.5%, propionaldehyde 97%, butyraldehyde 99.5%, and silica gel (Davisil grade 635, 60–100 mesh) were from Aldrich (Milwaukee, WI). DNPH (containing  $\sim$ 50% water) and phosphoric acid (>85%) were pure grade from Wako Pure Chemical Industries Ltd. 2,4-Dinitrophenylhydrazine hydrochloride (>85%) was from Kanto Chemical Co. (Tokyo, Japan).

**Synthesis of Carboxylic 2,4-Dinitrophenylhydrazide Derivatives.** Formic 2,4-Dinitrophenylhydrazide (Formic DNPhydrazide). Five grams of 2,4-dinitrophenylhydrazine hydrochloride was dissolved in 50 mL of formic acid. Pure water (50 mL) was added to this solution, and a light yellow precipitate formed. After 2 h, the precipitate was recovered by filtration and washed three times with 500 mL of water. The precipitate of the hydrazide derivative was recrystallized from ethanol and vacuum-dried.

Acetic 2, 4-Dinitrophenylhydrazide (Acetic DNPhydrazide), Propionic 2, 4-Dinitrophenylhydrazide (Propionic DNPhydrazide), and Butyric 2, 4-Dinitrophenylhydrazide (Butyric DNPhydrazide). Five grams of 2, 4-dinitrophenylhydrazine hydrochloride was dissolved in 50 mL carboxylic acid with 5 mL concentrated sulfuric acid. After 24 h at room temperature, the precipitate was recovered by filtration and washed three times with 500 mL of water. The precipitates of the hydrazide derivatives were recrystallized from ethanol and vacuum-dried.

**Preparation of the DNPH Cartridge for Collection of Carboxylic Acids.** Fifty grams of silica gel was washed three times with 500 mL of water; two times with 500 mL of methanol; and last, two times with 500 mL of acetonitrile. 2,4-Dinitrophenylhydrazine hydrochloride (0.18 g) and 0.5 mL of phosphoric acid were dissolved in 150 mL of acetonitrile. This solution was added to 50 g of washed silica gel. The mixture was stirred, and then the solvent was evaporated to dryness at 40 °C under vacuum on a rotary evaporator. The DNPH-coated silica particles (500 mg) were packed into a polyethylene cartridge (50 mm long × 8.8 mm i.d.) and stored in a refrigerator (4 °C).

Generation of Standard Formic Acid Gas and Collection using DNPH Cartridge. Standard gas was generated in a dynamic system by diffusing formic acid using a gas generator (Permeater PD-1B, GASTEC Co., Kanagawa, Japan). Pure, pressure-regulated air was passed with a constant flow rate through the PD-1B gas generator including a dispersion bottle (3 mm i.d.) containing  $\sim 2$  mL of formic acid and thermostated at 30.0 °C. The concentration of generated gas was calculated from the weight loss (mg) of the dispersion bottle and the pure air volume (L) passed into the PD-1B; e.g. 5 ppm of standard formic acid gas is obtained from a flow rate of 1.35 L/min at 30 °C. One end of the DNPH cartridge was attached to the testing chamber, and the other end was connected to a sampling pump equipped with a mass flow controller (SP 208 Dual, GL Sciences Inc., Tokyo, Japan).

## **RESULTS AND DISCUSSION**

**Carboxylic DNPhydrazide Derivatives.** A DNPH cartridge saturated with formic acid vapor becomes gradually discolored and completely changes to light yellow in 6 h. The HPLC chromatogram of the eluant from this DNPH cartridge indicates complete consumption of DNPH accompanied by formation of formic 2,4-dinitrophenylhydrazide (formic DNPhydrazide). Figure 1 shows the peak area changes with time of DNPH and formic



**Figure 1.** The reaction of adsorbed formic acid and DNPH with time. ( $\lambda = 360$  nm).

DNPhydrazide at wavelength 360 nm. Acetic acid, propionic acid, and butyric acid exhibit similar behavior with longer reaction time in order of increasing carbon number.

It is postulated that carboxylic acids react with DNPH to initially form corresponding hydrazone derivatives, which then isomerize to hydrazides by keto–enol tautomerization.



2,4-dinitrophenylhydrazine

carboxylic acid



carboxylic-2,4-dinitrophenylhydrazone (enol form)



carboxylic-2,4-dinitrophenylhydrazide (keto form)

The derivatives eluted from the DNPH cartridges were identified by comparison of the HPLC retention times with authentic

Table 1. Maximum Absorption Wavelengths ( $\lambda_{Max}$ ) and Molar Absorption Coefficients ( $\epsilon$ ) of C<sub>1</sub>–C<sub>4</sub> Carboxylic Acid and Aldehyde DNPH Derivatives<sup>a</sup>

	λmax (nm)	(L/mol/cm)	mp (°C)
formic DNPhydrazide	331	$1.36  imes 10^4$	185.7
acetic DNPhydrazide	333	$1.42  imes 10^4$	201.5
propionic DNPhydrazide	334	$1.44 imes10^4$	192.0
butyric DNPhydrazide	334	$1.44 imes10^4$	167.1
DNPhydrazine	351	$1.55  imes 10^4$	${\sim}200^{30}$
formaldehyde DNPhydrazone	349	$1.91  imes 10^4$	$153 - 156^{32}$
acetaldehyde DNPhydrazone	359	$2.10  imes 10^4$	$165 - 168^{31}$
propionaldehyde DNPhydrazone	360	$2.10  imes 10^4$	$152 - 155^{31}$
butyraldehyde DNPhydrazone	361	$2.18  imes 10^4$	117-1193

 $^a$  Melting points of aldehyde-2,4-DNP hydrazones are the literature values.

samples prepared by direct reaction of the carboxylic acids with DNPH (Experimental Section). Two HPLC columns with differing selectivities (C18 and RP-Amide C16) were used to confirm the retention time assignments.

Characterization. The structures of the synthesized hydrazides are consistent with the NMR data. <sup>1</sup>H NMR data of the derivatives were as follows: formic DNPhydrazide <sup>1</sup>H NMR (CD<sub>3</sub>-CN, TMS),  $\delta$  (ppm): 7.25 (d 1H, J = 9.6, a), 8.28 (dd 1H, J = 2.5, 9.4, b), 8.94 (d 1H, J = 2.8, c), 9.36 (s 1H, d), 8.49 (s 1H, e), 8.23 (s 1H, f). Acetic DNPhydrazide <sup>1</sup>H NMR (CD<sub>3</sub>CN, TMS),  $\delta$ (ppm): 7.25(d 1H, J = 9.2, a), 8.25 (dd 1H, J = 2.75, 9.6, b), 8.92 (d 1H, J = 2.8, c), 9.37 (s 1H, d), 8.49 (s 1H, e), 2.02 (s, 3H, CH<sub>3</sub>).Propionic DNPhydrazide <sup>1</sup>H NMR (CD<sub>3</sub>CN, TMS),  $\delta$  (ppm): 7.22 (d 1H, J = 9.6, a), 8.23 (dd 1H, J = 2.52, 9.39, b), 8.91 (d 1H, J =2.29, c), 9.37 (s 1H, d), 8.48 (s 1H, e), 2.30 (t, 2H, CH<sub>2</sub>), 1.13 (t, 3H, CH<sub>3</sub>). Butyric DNPhydrazide <sup>1</sup>H NMR (CD<sub>3</sub>CN, TMS),  $\delta$ (ppm): 7.21 (d 1H, J = 9.15, a), 8.24 (dd 1H, J = 2.75, 9.62, b), 8.91 (d 1H, J = 2.75, c), 9.39 (s 1H, d), 8.49 (s 1H, e), 2.27 (m, 2H, J = 7.56, CH<sub>2</sub>), 1.66 (t, 2H, J = 7.42, CH<sub>2</sub>), 0.964 (t, 3H, J = 7.327, CH<sub>3</sub>).

Carboxylic DNPhydrazide derivatives demonstrated excellent thermal stability on DSC analysis. Melting points are listed in Table 1 and are higher than those of corresponding hydrazones by 32-50 °C.

UV-visible absorption spectra of the carboxylic DNPhydrazide derivatives (20  $\mu$ mol/L acetonitrile solution) are presented in Figure 2. For reference, the absorption spectrum of DNPH is also shown.

The spectral profiles of carboxylic DNPhydrazide derivatives exhibited similar curves. Specifically, acetic, propionic, and butyric DNPhydrazide showed much the same maximum absorption wavelengths (333–334 nm) and molar absorption coefficients (1.42  $\times$  10<sup>4</sup> to 1.44  $\times$  10<sup>4</sup> L/mol/cm) (see Table 1).

Separation of Carboxylic DNPhydrazide Derivatives. Analytical conditions for formic, acetic, propionic, and butyric DN-Phydrazide derivatives were examined using a C18 and an RP-Amide C16 column. Figure 3 shows a chromatogram of  $C_1-C_4$ 

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**Figure 2.** UV-visible absorption spectra of carboxylic DNPhydrazide derivatives in acetonitrile solution (20  $\mu$ mol/L).



**Figure 3.** Chromatographic profile of  $C_1-C_4$  carboxylic DNPhydrazides and aldehyde DNPhydrazones on an RP-Amide C16 column (100  $\mu$ mol/L). A prime sign indicates the *Z* isomer peak; the other peaks without a prime sign are the *E* isomers.<sup>32</sup> FC, formic DNPhydrazide; AC, acetic DNPhydrazide; PC, propionic DNPhydrazide; BC, butyric DNPhydrazide; FA, formaldehyde-DNPhydrazone; AA, acetaldehyde DNPhydrazone; PA, propionaldehyde DNPhydrazone; BA, butyraldehyde DNPhydrazone.

hydrazide and hydrazone derivatives using an RP-Amide C16 column.

The reference solution was 100  $\mu$ mol/L and contained 0.1% phosphoric acid. Some small peaks, corresponding to the *Z* isomers, appeared in the chromatograms.<sup>32</sup> Aldehyde-DNPhydrazones in the standard solution (non-acid) exhibits only the *E* isomer; however, this *E* isomer can partly convert into the *Z* isomer upon addition of acid solution,<sup>33,34</sup> and the sample solution extracted from the DNPH cartridge inevitably contains phosphoric acid. The isomerization involves addition of a nucleophile (phosphate ion or water) to the protonated *E* or *Z* isomers, and an equilibrium *Z*/*E* isomer ratio was observed in 0.02–0.2% v/v phosphoric acid solutions.<sup>32</sup>

In Figure 3, the chromatographic peaks of formic, acetic, and propionic DNPhydrazide derivatives were detected before the



Figure 4. Reaction of formic acid and DNPH in the DNPH cartridge at various temperatures.

DNPH peak, and the butyric DNPhydrazide peak was detected after the DNPH peak. In the case of the C18 column, the formic DNPhydrazide peak overlapped with the acetic DNPhydrazide peak. In addition, the Z isomer of the acetaldehyde DNPhydrazone peak overlapped with the *E* isomer. The RP-Amide C16 column gave completely separated peaks. The amide column is an alkylamide reversed-phase column based on high-purity silica. The presence of the alkylamide group gives these columns an alternative selectivity compared to the simple hydrophobic selectivity of the C18 columns.<sup>35</sup> Due to the presence of both alkyl and amide functionality on the RP-Amide C16 column, two retention mechanisms are available: a mechanism based on the hydrophobic alkyl groups and a mechanism based on interaction between the polar groups of the carboxylic DNPhydrazide derivatives and the amide group. Therefore, amide columns, such as RP-Amide C16 or Bonus-RP, provide unique selectivity that leads to sufficient resolution between acetic and formic DNPhydrazide derivatives. In the chromatograms produced using the alkylamide column, C<sub>1</sub>-C<sub>4</sub> straight-chain carboxylic DNPhydrazides and aldehyde DNPhydrazones were completely separated.

The limits of detection (LOD) by HPLC, defined as three times the standard deviation of 10 replicate measurements at  $10 \ \mu g/L$ , were as follows: formic acid 1.0, acetic acid 0.7, propionic acid 1.9, butyric acid 2.4, formaldehyde 1.0, acetaldehyde 1.4, propionaldehyde 1.3, and butyraldehyde 1.9  $\mu g/L$ .

**Collection and Derivatization of Formic Acid Using the DNPH Cartridge.** Two DNPH cartridges (primary and secondary) were connected in series, and standard formic acid gas (5 ppm) was drawn through the cartridges for 30 min. The secondary cartridge served to collect any material that may have escaped the primary cartridge. Immediately after collection, the DNPH cartridges were heated for 1–10 h in a thermostat. Later, the DNPH derivatives were removed from the adsorbent by eluting acetonitrile (5 mL) through the DNPH cartridge via a syringe to a graduated test tube over a 1-min time period. The eluate was then analyzed by HPLC. Figure 4 shows the changes in formic acid derivative content with temperature. Under all conditions

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**Figure 5.** Influence of phosphoric acid concentration on formation of hydrazide derivative.

studied, no formic acid derivative was detected in the secondary cartridge, indicating that no "breakthrough" of the primary cartridge occurred.

Formic acid was physically adsorbed onto the DNPH cartridge at a rapid rate and reacted chemically with DNPH at a slow rate. The rate of reaction was very slow at 20 °C and increased dramatically with temperature. The reaction rate at 100 °C was the most rapid of all conditions tested; however, recovery did not reach 100% (0.3 µmol) and began to decrease after 2 h. Thermal decomposition of DNPH was observed. During heating at 100 °C, the peak area of DNPH decreased linearly to 14% in 10 h, and decomposition products (1,3-dinitrobenzene and 2,4-dinitroaniline) were detected as overlapping peaks at 6.5 min on the chromatogram. It is believed that the decomposition of DNPH induced a poor recovery. During heating at 80 °C, the peak area of DNPH decreased to 76% in 10 h. However, at 20-60 °C, there were no decreases of DNPH. From these results, the standard heating condition for derivatization to hydrazide was selected as 80 °C and 5 h.

Carboxylic acids and aldehydes react with DNPH in the presence of an acidic catalyst; therefore, the effect of the amount of phosphoric acid was examined for the DNPH cartridge. DNPH-coated silica gels containing 0.1-10% v/w phosphoric acid (as 85% phosphoric acid) were prepared, and 500 mg was packed into polyethylene cartridges. All cartridges were connected to a secondary cartridge containing 1% v/w phosphoric acid, and standard formic acid gas (5 ppm) was drawn through for 30 min. Immediately after collection, DNPH cartridges were heated for 5 h at 80 °C. Figure 5 shows the recovery rates as a function of phosphoric acid concentration.

It appears that formic acid is difficult to derivatize under strong or weakly acidic conditions. However, complete adsorption of formic acid is believed to have occurred in all primary DNPH cartridges containing 0.1-10% v/w phosphoric acid since no formic acid was detected in any secondary cartridges. Therefore, the recovery rate in Figure 5 is not a reflection of the collection efficiency. The derivatization reaction to hydrazide progressed essentially to completion for the DNPH cartridges containing 0.2-1% v/w phosphoric acid. The best condition for simultaneous

Table 2. Blank Value of the DNPH Cartridge

		20 °C			80 °C		
	mean (µg)	SD (µg)	CV (%)	mean (µg)	SD (µg)	CV (%)	
acetic acid	0.0041	0.0012	29	3.1	0.095	3.0	
formic acid	0.029	0.0028	9.5	0.48	0.020	4.3	
propionic acid	0.028	0.0027	9.5	0.66	0.048	7.3	
butyric acid	nd <sup>a</sup>	nd <sup>a</sup>		nd <sup>a</sup>	nd <sup>a</sup>		
formaldehyde	0.019	0.0009	4.8	0.31	0.013	4.5	
acetaldehyde	nd <sup>a</sup>	nd <sup>a</sup>		0.077	0.0057	7.4	
prorionaldehyde	nd <sup>a</sup>	nd <sup>a</sup>		0.017	0.0032	18	
butyraldehyde	nd <sup>a</sup>	nd <sup>a</sup>		0.050	0.0042	8.2	
<sup>a</sup> Not detected.							

measurement of carboxylic acids and aldehydes is 1% v/w phosphoric acid because acidic conditions are needed for the measurement of aldehydes.

On the basis of these results, the following optimized conditions for derivatization were identified: DNPH cartridge with 1% v/w phosphoric acid, 80 °C reaction temperature, and 5 h reaction time.

**Blank Test and LOD.** Seven DNPH cartridges were kept for 5 h in a thermostat at 80 or 20 °C and analyzed by the abovementioned procedure. The blank values of  $C_1-C_4$  carboxylic acids and aldehydes are shown in Table 2.

The blank values in the case of heating at 80 °C were higher than at 20 °C, especially for carboxylic acids. It is believed that the unreacted background compounds in the DNPH cartridge formed DNPH derivatives on heating. The LOD for measurement of formic acid, defined as three times the standard deviation of the blank (3 $\sigma$ ), is 0.06  $\mu$ g per cartridge. When the collection of air sample occurs for 24 h at 100 mL/min (144 L), the LOD is 0.4  $\mu$ g/m<sup>3</sup>.

**Measurement of Indoor Air.** Indoor air in an apartment house was passed through the DNPH cartridges for 24 h at a flow rate of 50 mL/min. After sampling, the cartridges were maintained for 5 h in a thermostat at 80 or 20 °C and then analyzed by the above-mentioned procedure. The chromatograms in Figure 6 illustrate measurements made under the two different temperature conditions.

It was confirmed that carboxylic acids and aldehydes in sample air were completely collected in the primary DNPH cartridge because the amounts of compounds detected in secondary cartridges were equivalent to background levels. The concentrations of aldehydes including formaldehyde, acetaldehyde, and propionaldehyde following heating at 80 °C were the same as those at 20 °C. Formic acid collected with the DNPH cartridge was detected following heating at 80 °C.

Some unknown peaks were detected in Figure 6. In samples without heat treatment, peaks of "unknown 1" were often detected in the sample solution immediately after collection of the air sample. The peak corresponding to "unknown 2" was detected in the sample solution from the DNPH cartridges stored for longer time periods. This suggests that unknown 1 was converted to unknown 2. The retention time for unknown 1 fits well to 2,4-dinitrophenyl azide, a known interfering agent of the DNPH method resulting from the reaction of the reagent with nitrogen dioxide.<sup>36</sup> This compound is known to be thermolabile. It decom-



**Figure 6.** Chromatographic profiles of DNPH derivatives from air sample measured with a heat treatment of 80 and 20 °C. FC, formic DNPhydrazide; AC, acetic DNPhydrazide; PC, propionic DNPhydrazide; FA, formaldehyde DNPhydrazone; AA, acetaldehyde DNPhydrazone; PA, propionaldehyde DNPhydrazone; BA, butyraldehyde DNPhydrazone; AN, acetone; DB, 1,3-dinitrobenzene; DA, 2,4-dinitroaniline.

poses under thermal stress to nitrobenzofurazan-3-oxide (unknown **2**), which is more stable.<sup>37,38</sup>

## CONCLUSIONS

In this study, it was found that  $C_1-C_4$  carboxylic acids reacted with DNPH to form the corresponding hydrazide derivatives. These compounds are thermally stable and exhibit maximum absorption wavelengths near the aldehyde DNPhydrazone derivatives.  $C_1-C_4$  carboxylic DNPhydrazides and aldehyde DNPhydrazones were completely separated and detected by HPLC using a RP-Amide C16 column and 350-nm detection. These properties permit the simultaneous measurement of carboxylic acids and aldehydes in air using a DNPH cartridge containing DNPH-coated silica gel. Heat treatment of the collected DNPH cartridge was necessary in order to measure carboxylic acid levels because the reaction times of derivatization are very slow. In the case of formic acid, 5 h reaction time at 80 °C is needed after collection.

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