

# Improvement of 2,4-dinitrophenylhydrazine derivatization method for carbon isotope analysis of atmospheric acetone

Sheng Wen<sup>1</sup>, Yingxin Yu<sup>2</sup>, Songjun Guo<sup>1</sup>, Yanli Feng<sup>2</sup>, Guoying Sheng<sup>1,2</sup>, Xinming Wang<sup>1</sup>, Xinhui Bi<sup>1</sup>, Jiamo Fu<sup>1,2\*</sup> and Wanglu Jia<sup>1</sup>

<sup>1</sup>State Key Laboratory of Organic Geochemistry, Guangdong Key Laboratory of Environmental Resources Utilization and Protection, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences, Guangzhou 510640, People's Republic of China

<sup>2</sup>School of Environmental and Chemical Engineering, Shanghai University, Shanghai 200072, People's Republic of China

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Through simulation experiments of atmospheric sampling, a method via 2,4-dinitrophenylhydrazine (DNPH) derivatization was developed to measure the carbon isotopic composition of atmospheric acetone. Using acetone and a DNPH reagent of known carbon isotopic compositions, the simulation experiments were performed to show that no carbon isotope fractionation occurred during the processes: the differences between the predicted and measured data of acetone-DNPH derivatives were all less than 0.5%. The results permitted the calculation of the carbon isotopic compositions of atmospheric acetone using a mass balance equation. In this method, the atmospheric acetone was collected by a DNPH-coated silica cartridge, washed out as acetone-DNPH derivatives, and then analyzed by gas chromatography/combustion/isotope ratio mass spectrometry (GC/C/IRMS). Using this method, the first available  $\delta^{13}\text{C}$  data of atmospheric acetone are presented. Copyright © 2006 John Wiley & Sons, Ltd.

Acetone is one of the most important trace organic compounds in the tropospheric air. Its average concentration of ~0.5 parts per billion by volume (ppbv) makes it one of the most abundant organic compounds in clean air.<sup>1–4</sup> It acts as a source of HO<sub>x</sub>, organic acids, and peroxyacetyl nitrate. It may thus also influence the formation of ozone in the upper troposphere, and play an important role in global tropospheric chemistry.<sup>2,5–7</sup> There are several sources of acetone, both primary and secondary, natural and anthropogenic.<sup>1</sup> The sinks include photolysis and reaction with OH; in addition, deposition to land and oceans has been proposed as an additional sink.<sup>1,8</sup> Although many studies have been conducted in this area,<sup>1–8</sup> much is still unknown and further work is needed.

Studies of isotopic compositions of trace atmospheric species (e.g., CO<sub>2</sub>, CH<sub>4</sub>, and NMHC) have provided valuable information on the transition mechanism and source identification of these compounds,<sup>9–11</sup> but there has been no study on the carbon isotope analysis of atmospheric acetone.

In our previous study, acetone 2,4-dinitrophenylhydrazine (DNPH) derivatization in acetonitrile solution was studied,

and it was found that no  $\delta^{13}\text{C}$  fractionation occurred.<sup>12</sup> DNPH derivatization was a necessary step for the collection and measurement of atmospheric acetone in many recent studies.<sup>13–15</sup> Those results showed the possibility of carbon isotope analysis of atmospheric acetone. In the present study, the simulation experiment of atmospheric sampling was conducted. It was found that there was no  $\delta^{13}\text{C}$  fractionation during the sampling and the laboratory processes after sampling. It is possible, therefore, to obtain the carbon isotope data of atmospheric acetone by the use of DNPH derivatization sampling. By means of this method, a preliminary study has been performed to obtain the first set of carbon isotope data of atmospheric acetone.

## EXPERIMENTAL

### Chemicals and materials

All solvents employed were high-performance liquid chromatography (HPLC) grade. Water was double distilled. Acetonitrile (ACN) was purchased from Merck (Darmstadt, Germany). DNPH (99%) was purchased from Fluka (Buchs, Switzerland), and was purified by recrystallization three times from HPLC-grade ACN. DNPH of the same lot number was used for all derivatizations.

Acetone (HPLC grade) from two suppliers was used. These reagents were supplied by the Chemical Reagent

\*Correspondence to: J. Fu, School of Environmental and Chemical Engineering, Shanghai University, Shanghai 200072, People's Republic of China.

E-mail: fujm@gig.ac.cn; fujm@staff.shu.edu.cn

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Factory of Guangzhou (China) (M1) and the Xinjian Chemistry Factory of Guangzhou (China) (M2), respectively. They were distilled before use.

### Simulation experiment for atmospheric sampling

The experiments were performed using a home-made system, consisting of a 100 L Teflon sample bag (SKC Inc., Eighty Four, PA, USA), connected to a sampling pump (Thomas, Sheboygan, WI, USA). Before every experiment, the bag was flushed at least three times with high-purity N<sub>2</sub> (99.99%). Then acetone was injected into the bag, heated to aid evaporation, and then kept for about 30 min to reach equilibrium. The gas (containing acetone) was then drawn out and through a sampling cartridge (DNPH-coated) using the sampling pump; the flow rate was about 2 L/min. After all the gas in the bag had been drawn out (for acetone concentration of low level, several bags of gases were needed to get sufficient analyte (200 ng per injection) for gas chromatography/combustion/isotope ratio mass spectrometry (GC/C/IRMS) analysis), the cartridge was removed and the acetone-DNPH derivatives were washed out by 1 mL ACN, and then about 20 µL saturated ammonia solution was added for neutralization. The solution was blown to dryness by a gentle flow of N<sub>2</sub> before analysis. For GC/C/IRMS analysis, 20–100 µL CHCl<sub>3</sub> was added to make solutions.

### Atmospheric acetone sampling

The sampling method has been described in detail in previous studies.<sup>13,15</sup> The sampling medium was a Sep-Pak silica cartridge (Waters, Millipore Corp., Milford, MA, USA). Each cartridge was rinsed by attaching a syringe filled with 10 mL of ACN and then coated slowly by passing 7 mL of the freshly made coating solution, which contained 60 mL DNPH-ACN-saturated solution and 4 mL concentrated orthophosphoric acid in 500 mL ACN, through the cartridge by gravity. When there was no more solution flowing out of the cartridge, the cartridge was dried with a gentle flow of nitrogen for 15 min, and then wrapped in aluminum foil, sealed in a Teflon bag, and stored in a refrigerator at 4°C until use. Three blank cartridges from each cartridge batch were analyzed and the results were below the EPA blank criteria (acetone concentration <0.1 µg/cartridge, mostly less than 10% of the concentration for the analyte from an air sample when the acetone was collected as its DNPH derivative).

Samples were collected by drawing air with a sampling pump (Thomas) through the cartridge. A potassium iodide denuder was connected upstream of the cartridge to prevent ozone interference.<sup>16</sup> After sampling, each cartridge was wrapped in aluminum foil, resealed in a Teflon bag, transported back to the laboratory and stored in the refrigerator before being analyzed. For each sampling program one laboratory blank and one field blank were also included. The recovery test was performed to obtain a recovery efficiency of more than 95% for acetone in general.

A batch of samples was taken at the foot of Prince Hill, a small hill about 90 km from Guangzhou City, on 10–11 July, 2004. The sampling site was in a forested agricultural area with low population density. The sampling flow rate was 2 L/min, and the sampling duration was 3 h.

Another batch of samples was taken at the top of an office building, about 10 m high, in Guangzhou Institute of Geochemistry, Chinese Academy of Sciences, Guangzhou, Guangdong Province, China. The sampling period was from 5–7 July, 2005. The sampling site was about 300 m away from the expressway and railway. The sampling flow rate was 1 L/min, and the sampling duration was 12 h.

After sampling, the sampled cartridge was eluted with 1 mL of ACN and about 20 µL saturated ammonia solution was added for neutralization. The solution was then blown to dryness by a gentle flow of nitrogen before analysis. For GC/C/IRMS analysis, 10–100 µL CHCl<sub>3</sub> was added to make solutions.

### Analytical procedure

Two GC/C/IRMS systems were used in our study for compound-specific carbon isotope analysis. Analyses of acetone-DNPH derivatives were performed using an HP 6890 gas chromatograph (Agilent, Wilmington, DE, USA) combined with a GV IsoPrime mass spectrometer. The GC system was equipped with a DB-5 column (30 m × 0.32 mm × 0.25 µm). For all the measurements, the injector was set at 230°C in splitless mode, and the following oven temperature program was used: 60°C at the start, 20°C/min to 150°C, 3°C/min to 230°C, and then 20°C/min to 290°C. Helium was used as the carrier gas at a flow rate of 1.5 mL/min. The combustion interface was operated at 850°C. The mass spectrometer provided real-time measurements of the <sup>13</sup>C/<sup>12</sup>C ratios for each CO<sub>2</sub> peak. An external CO<sub>2</sub> reference gas (δ<sup>13</sup>C = -27.45‰) was used to obtain highly accurate isotopic compositions. The reproducibility and accuracy of carbon isotope analyses were evaluated routinely every day using ten laboratory isotopic standards (C<sub>12</sub>, C<sub>14</sub>, C<sub>16</sub>, C<sub>18</sub>, C<sub>20</sub>, C<sub>22</sub>, C<sub>25</sub>, C<sub>28</sub>, C<sub>30</sub>, C<sub>32</sub> n-alkanes supplied by Indiana University), and the procedure was the same as that used in a previous study.<sup>12</sup> The synthesized acetone-DNPH standard solutions, used as the laboratory isotopic standards, were prepared by a method similar to that described by Wen *et al.*,<sup>13</sup> and measured over 30 times by the same GC/C/IRMS system to obtain the δ<sup>13</sup>C result of -27.21‰ (with one standard deviation of less than 0.5‰). However, for the δ<sup>13</sup>C analysis of underivatized acetone, another GC/C/IRMS system was used as described previously,<sup>12</sup> the analytical conditions used in the elemental analyzer/isotope ratio mass spectrometry (EA/IRMS) were also the same as before.<sup>12</sup>

All <sup>13</sup>C/<sup>12</sup>C ratios are expressed in conventional delta (δ) notation, which is the per mil (‰) deviation from the standard Pee Dee Belemnite (PDB). The reproducibility and accuracy of these IRMS systems were shown to be good.<sup>12,13</sup>

### Measurement of δ<sup>13</sup>C values of DNPH, acetone and its derivative

The δ<sup>13</sup>C values of the DNPH reagent were determined using the EA/IRMS system; for acetone and its acetone-DNPH derivatives, the δ<sup>13</sup>C values were determined using the two GC/C/IRMS instruments, described above. The detailed descriptions were reported in a previous study.<sup>12</sup>

**Table 1.** Stable carbon isotopic compositions of predicted and measured acetone-DNPH derivatives in simulation experiment of atmospheric sampling

Supplier	Acetone conc. ( $\mu\text{g m}^{-3}$ )	$\delta^{13}\text{C}$			$\Delta^e$
		Underivatized acetone <sup>a,b</sup>	Measured acetone-DNPH derivatives <sup>a-c</sup>	Predicted acetone-DNPH derivatives <sup>a,c,d</sup>	
M1	80		$-27.10 \pm 0.05$		$-0.42$
	161	$-29.07 \pm 0.10$	$-27.21 \pm 0.08$	$-27.52$	$-0.31$
	4480		$-27.23 \pm 0.20$		$-0.29$
M2	20		$-25.62 \pm 0.15$		$-0.14$
	40	$-23.86 \pm 0.31$	$-25.68 \pm 0.26$	$-25.76$	$-0.08$
	4480		$-25.39 \pm 0.33$		$-0.37$
	7552		$-25.68 \pm 0.06$		$-0.08$

<sup>a</sup> Stable carbon isotopic compositions reported in per mil relative to PDB ( $\pm 1$  standard deviation for measured data).

<sup>b</sup>  $\delta^{13}\text{C}$  determined by triplicate analyses of GC/C/IRMS.

<sup>c</sup>  $\delta^{13}\text{C}_{\text{DNPH}} = -26.72 \pm 0.24\text{‰}$ , from eight analyses determined by EA/IRMS.

<sup>d</sup> Based on the mass balance relationship (Eqn. (1)) of  $\delta^{13}\text{C}$  of acetone determined by GC/C/IRMS and  $\delta^{13}\text{C}$  of DNPH determined by EA/IRMS.

<sup>e</sup> Predicted  $\delta^{13}\text{C}$ –measured  $\delta^{13}\text{C}$ .

## RESULTS AND DISCUSSION

### $\delta^{13}\text{C}$ analysis of acetone and derivatives

The reproducibility of the carbon isotope analysis was evaluated for two batches of acetone (with different  $\delta^{13}\text{C}$  values); their  $\delta^{13}\text{C}$  values and those of the corresponding DNPH derivatives are given in Table 1. The analytical errors (standard deviation) obtained for three GC/C/IRMS analyses of acetone from the same supplier ranged from 0.10‰ to 0.31‰, and, for their DNPH derivatives, the analytical errors were from 0.05‰ to 0.33‰. The accuracy of these results was well within the instrument specifications. The reproducibility was also excellent compared with that of previous studies.<sup>10,11,17</sup>

The reproducibility of the derivatization process was also evaluated (Table 1). For acetone from the same supplier, the differences in the  $\delta^{13}\text{C}$  values of its derivatives were all within the instrument specification, even with the large variation in acetone concentrations (from tens to thousands of  $\mu\text{g m}^{-3}$ ) used to perform several derivatizations. The small differences obtained showed the good reproducibility of the derivatization processes.

### Isotopic fractionation during sampling

Theoretically, the acetone-DNPH derivatives should exhibit  $\delta^{13}\text{C}$  compositions that reflect the relative contributions of carbon from each component and their respective  $\delta^{13}\text{C}$  values. If there is no isotopic fractionation during the acetone derivatization reaction, the generalized stoichiometric mass balance equation for acetone and its DNPH derivatives may be written as:

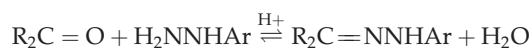
$$\delta^{13}\text{C}_{\text{derivatives}} = f_{\text{Acetone}}\delta^{13}\text{C}_{\text{Acetone}} + f_{\text{DNPH}}\delta^{13}\text{C}_{\text{DNPH}} \quad (1)$$

where  $f_{\text{Acetone}}$  and  $f_{\text{DNPH}}$  are the fractions of carbon in the acetone-DNPH derivatives due to the underivatized acetone and DNPH reagent, respectively. Here,  $f_{\text{Acetone}}$  has the value of 1/3 for the derivatization of acetone. It can be used to estimate the carbon isotope composition of the original acetone ( $\delta^{13}\text{C}_{\text{Acetone}}$ ) from the known isotopic composition of DNPH ( $\delta^{13}\text{C}_{\text{DNPH}}$ ) and the measured isotopic composition of acetone-DNPH derivatives ( $\delta^{13}\text{C}_{\text{derivatives}}$ ).

Table 1 compares the  $\delta^{13}\text{C}$  values of the DNPH derivatives predicted by Eqn. (1) with those measured by GC/C/IRMS. The predicted and measured values agreed to within the precision limits of the GC/C/IRMS measurements (ranging from  $-0.08\text{‰}$  to  $-0.42\text{‰}$ ), and it should be noted that the calculated isotopic difference between the predicted and measured values is minor.

From the data above, the conclusion could be drawn that no  $\delta^{13}\text{C}$  fractionation occurs during the atmospheric sampling process via DNPH derivatization. The detailed theory has been described elsewhere.<sup>12,13,18,19</sup>

In the present study, the acetone reacts with DNPH as follows:



where  $\text{R} = \text{CH}_3$  and  $\text{Ar} = 2,4\text{-dinitrophenyl}$ . It can be seen that acetone is the only carbon-containing compound that is altered in the rate-determining step. Both in the simulation experiment and in the environment sampling, acetone reacted quantitatively with excess DNPH. Thus, it is predicted that only minor carbon isotopic fractionation should occur during these processes, and this is confirmed by the results of our present studies. The computation of the original, underivatized atmospheric  $\delta^{13}\text{C}$  values of acetone using the mass balance equation (Eqn. (1)) is then possible.

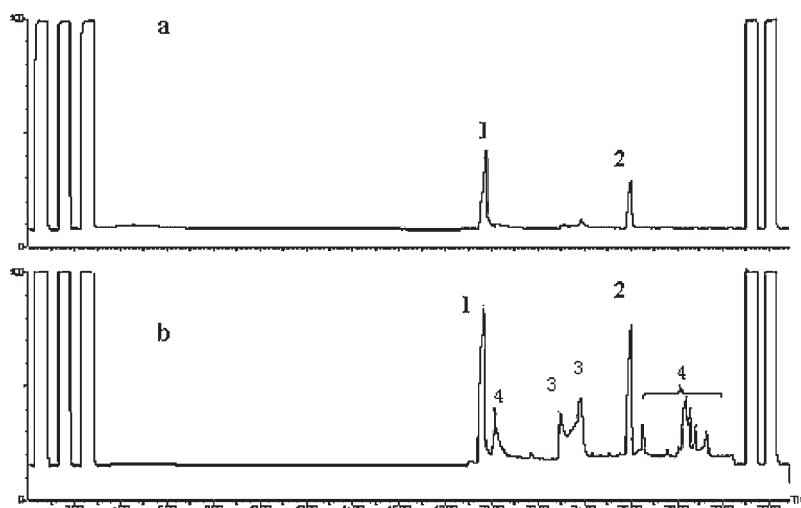
It should be pointed out that the analytical error of the calculated data for acetone (usually expressed as the standard deviation, S) could be calculated by the following equation (derived from Eqn. (1)):

$$S_{\text{Acetone}}^2 = 1/f_{\text{Acetone}}^2 \times S_{\text{derivatives}}^2 + (f_{\text{DNPH}}/f_{\text{Acetone}})^2 \times S_{\text{DNPH}}^2 \quad (2)$$

where  $f_{\text{Acetone}}$  and  $f_{\text{DNPH}}$  have the same meaning as in Eqn. (1), and  $S_{\text{derivatives}}$  and  $S_{\text{DNPH}}$  are the standard deviation (S) of the measured carbon isotope data of the acetone-DNPH derivative and DNPH, respectively.

### Measurements of atmospheric samples

The acetone-DNPH derivatives in the samples collected at both the sampling sites (described above) were analyzed by



**Figure 1.** Typical GC/C/IRMS chromatograms of acetone-DNPH derivatives from (a) standard sample and (b) atmospheric sample collected at the top of the office building in Guangzhou. 1: formaldehyde-DNPH derivative; 2: acetone-DNPH derivative; 3: acetaldehyde-DNPH (E- and Z-isomers); 4: unknown compounds.

**Table 2.** Stable carbon isotope composition of acetone in samples collected from environmental atmosphere

Sampling site	Sampling time	Acetone conc. ( $\mu\text{g m}^{-3}$ ) <sup>f</sup>	$\delta^{13}\text{C}$	
			Measured acetone derivatives <sup>a-c</sup>	Calculated underivatized acetone <sup>a,c-e</sup>
S1	9 am–9 pm, 5th	10	$-26.21 \pm 0.31$	$-25.19 \pm 1.05$
	9 am–9 pm, 6th	14	$-26.49 \pm 0.36$	$-26.03 \pm 1.18$
	9 pm 6th–9 am, 7th	6	$-26.51 \pm 0.23$	$-26.09 \pm 0.84$
S2	0 am–3 am, 11th	6	$-28.09 \pm 0.12$	$-30.83 \pm 0.60$
	3 am–6 am, 11th	4	$-28.33 \pm 0.28$	$-31.55 \pm 0.97$
	6 am–9 am, 11th	3	$-27.87 \pm 0.09$	$-30.17 \pm 0.55$

S1: At the top of the office building. S2: At the foot of Prince Hill.

<sup>a</sup>Stable carbon isotope compositions reported in per mil relative to PDB ( $\pm 1$  standard deviation).

<sup>b</sup> $\delta^{13}\text{C}$  determined by triplicate analyses of GC/C/IRMS.

<sup>c</sup> $\delta^{13}\text{C}_{\text{DNPH}} = -26.72 \pm 0.24\text{‰}$ , from eight analyses determined by EA/IRMS.

<sup>d</sup>Based on the mass balance relationship (Eqn. (1)) of  $\delta^{13}\text{C}$  of acetone determined by GC/C/IRMS and  $\delta^{13}\text{C}$  of DNPH determined by EA/IRMS.

<sup>e</sup>Standard deviation was calculated according to Eqn. (2).

<sup>f</sup>The acetone concentration was determined by HPLC as described in Refs. 13 and 15.

GC/C/IRMS, and the target compound was well separated from the other components (Fig. 1).  $^{13}\text{C}$  data of the target compounds are shown in Table 2. It can be seen that there was almost no variation in the  $^{13}\text{C}$  data of atmospheric acetone collected at the same sampling site, which might reflect the structural stability of the acetone in air. [Note that Atkinson reported that acetone had a much longer life time than other volatile organic compounds (VOCs) in the atmosphere even under the effect of  $\cdot\text{OH}$ ,  $\text{O}_3$  and photolysis.<sup>20</sup>] The  $^{13}\text{C}$  data showed large differences between the two sampling sites, which might reflect the different source of the atmospheric acetone (urban and rural). For samples collected at the office building, the acetone might mainly come from vehicle emission and reagent evaporation from factories,<sup>21</sup> whereas, for the samples collected in the forest, the emission of biomass was the main contributor.<sup>20</sup> Carbon isotope

analysis might thus be very useful for the identification of the source of atmospheric acetone, and compound-specific carbon isotope analysis might be a powerful tool in this field.

## CONCLUSIONS

Reduplicate  $\delta^{13}\text{C}$  analysis proved that simulation experiments for atmospheric acetone sampling via DNPH derivatization were reproducible, and that no carbon isotope fractionation occurred. This permits the computation of the original, underivatized  $\delta^{13}\text{C}$  values of acetone through a mass balance equation. Using this method, the preliminary study of carbon isotope analysis of atmospheric acetone was performed and we were able to acquire the first carbon isotope data for atmospheric acetone.

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## REFERENCES

- Singh HB, O'Hara D, Herlth D, Sachse W, Blake DR, Bradshaw JD, Kanakidou M, Crutzen PJ. *J. Geophys. Res.* 1994; **99**: 1805.
- Singh HB, Kanakidou M, Crutzen PJ, Jacob DJ. *Nature* 1995; **378**: 50.
- Singh HB, Chen Y, Tabazadeh A, Fukui Y, Bey I, Yantosca R, Jacob D, Arnold F, Wohlfrom K, Atlas E, Flocke F, Blake D, Blake N, Heikes B, Snow J, Talbot R, Gregory G, Sachse G, Vay S, Kondo Y. *J. Geophys. Res.* 2000; **105**: 3795.
- Singh HB, Chen Y, Staudt A, Jacob D, Blake D, Heikes B, Snow J. *Nature* 2001; **410**: 1078.
- McKeen SA, Gierczak T, Burkholder JB, Wennberg PO, Hanisco TF, Keim ER, Gao RS, Liu SC, Ravishankara AR, Fahey DW. *Geophys. Res. Lett.* 1997; **24**: 3177.
- Wennberg PO, Hanisco TF, Jaeglé L, Jacob D, Hintsä E, Lanzendorf EL, Anderson JG, Gao RS, Keim ER, Donnelly S, Del Negro L, Fahey DW, McKeen SA, Salawitch RJ, Webster CR, May RD, Herman R, Proffitt MH, Atlas E, Schauffler S, Flocke FT, McElroy E, Bu P. *Science* 1998; **279**: 49.
- Collins WJ, Stevenson DS, Johnson CE, Derwent RG. *J. Geophys. Res.* 1999; **104**: 26927.
- Chatfield RB, Gardner EO, Calvert JG. *J. Geophys. Res.* 1987; **92**: 4208.
- Conny JM, Currie LA. *Atmos. Environ.* 1996; **30**: 621.
- Rudolph J, Czuba E, Norman AL, Huang L, Ernst D. *Atmos. Environ.* 2002; **36**: 1173.
- Saito T, Tsunogai U, Kawamura K, Nakatsuka T, Yoshida N. *J. Geophys. Res.* 2002; **107** (D4): doi: 10.1029/2000JD000127.
- Wen S, Feng Y, Wang X, Sheng G, Fu J, Bi X. *Rapid Commun. Mass Spectrom.* 2004; **18**: 2669.
- Wen S, Feng Y, Yu Y, Bi X, Wang X, Sheng G, Fu J, Peng P. *Environ. Sci. Technol.* 2005; **39**: 6202.
- Ho KF, Lee SC, Louie PKK, Zou SC. *Atmos. Environ.* 2002; **36**: 1259.
- Feng Y, Wen S, Wang X, Sheng G, He Q, Fu J. *Atmos. Environ.* 2004; **38**: 103.
- Sirju A, Shepson PB. *Environ. Sci. Technol.* 1995; **29**: 384.
- Abrajano TA Jr, Murphy DE Jr, Fang J, Comet PA, Brooks JM. *Org. Geochem.* 1994; **21**: 611.
- Rieley G. *Analyst* 1994; **119**: 915.
- Melander L, Saunders WH. *Reaction Rates of Isotopic Molecules*. John Wiley: New York, 1980.
- Atkinson R. *Atmos. Environ.* 2000; **34**: 2063.
- Feng Y, Wen S, Chen Y, Wang X, Lü H, Bi X, Sheng G, Fu J. *Atmos. Environ.* 2005; **39**: 1789.