

# Highly Sensitive Determination Method for Total Carbonate in Water Samples by Flow Injection Analysis Coupled with Gas-Diffusion Separation

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A spectrophotometric method for the determination of total carbonate in water samples was developed. The method is based on the color change of an acid-base indicator in relation to the concentration of permeable gas substances through a membrane. By using a new portable FIA system equipped with a gas-diffusion unit, a highly sensitive and on-site determination of total carbonate in aqueous solutions was investigated. A new color-change system with 4-(2',4'-dinitrophenylazo)-1-naphthol-5-sulfonic acid (DNN5S) was developed. Absorbance changes of the reagent solution were measured at 450 nm with a light-emitting diode (LED) as a light source. A new type of gas-diffusion unit was used, and was constructed with double tubing: the inner tubing was a micro porous PTFE (polytetrafluoroethylene) tubing (1.0 mm inner diameter and 1.8 mm outer diameter, pore size 2  $\mu\text{m}$ , porosity 50%); the outer tubing was made of glass with 2.0 mm inner diameter. The optimized system conditions were as follows: the sample size was 200  $\mu\text{l}$ , the temperature of the air bath for the gas-diffusion unit was 25°C, and the length of the gas-diffusion unit was 15 cm; each flow rate was 0.3 ml  $\text{min}^{-1}$ . For measuring carbonate at low concentrations, a method for preparing water with less carbonate was proposed: the carbonate content of the water was decreased down to  $5 \times 10^{-7}$  M. The calibration graph was rectilinear from  $1 \times 10^{-6}$  M to  $10^{-3}$  M, and the detection limit (corresponding to a signal-to-noise ratio of 3) was  $1 \times 10^{-6}$  M of carbonate. The relative standard deviation (RSD) of ten measurements of  $2.3 \times 10^{-5}$  M  $\text{Na}_2\text{CO}_3$  solution was 1.9%. The total carbonate in various kinds of water (such as river, sea, rain, distilled and ultra purified) was determined.

(Received July 9, 2001; Accepted September 3, 2001)

## Introduction

Earth warming (green-house effect), abnormal weather and climate observed in recent years seem to be caused partly by the enrichment and movement of carbon dioxide into the atmosphere, and its concentration, which is now more than 0.03%. Because the carbon dioxide content in the atmosphere has gradually been increasing year-by-year, the amount of its generation has been restricted. Carbon dioxide can dissolve in environmental water, and is kept at equilibrium between the atmosphere and natural water. In order to clarify these equilibrium conditions of carbon dioxide precisely, it is necessary to develop a new sensitive and reliable determination method for total carbonate at  $10^{-5}$  M levels or less in aqueous solutions. Several methods for the determination of total carbonate (TC) in aqueous solutions have been reported. Of these, a gas-permeation detection method with a hydrophobic membrane, is the most common, and seems to be very useful. The changes in the physicochemical property of a receptor caused by permeated carbon dioxide through a hydrophobic membrane can be detected by spectrophotometry,<sup>1-4</sup> potentiometry<sup>5</sup> and conductometry.<sup>4,6</sup> Kuban and Dasgupta<sup>4</sup> reported a comparison of conductometry with spectrophotometry; they reported that a conductometric

procedure with a weak base receptor solution provided better day-to-day reproducibility, sensitivity and detection limits compared with a method with a photometric receptor consisting of a pH indicator and a carbonate-bicarbonate buffer. Aoki *et al.*<sup>6</sup> improved a conductometric method based on continuous flow reported by Carlson.<sup>11</sup> The limit of detection corresponding to a signal-to-noise ratio of three was  $1 \times 10^{-5}$  M. Motomizu *et al.* reported a gas-diffusion-FIA for carbonate<sup>1-3</sup> and ammonium.<sup>7,8</sup> However, in the methods reported so far, the sensitivity was not sufficient for determining TC at low concentrations in such samples as rain water and purified water, in which TC is less than, or around,  $10^{-5}$  M,<sup>9</sup> because the contamination of water from  $\text{CO}_2$  in air was unavoidable. In the present work, a newly designed gas-diffusion unit was used; new pH indicators used as a color-development reagent were synthesized to obtain higher sensitivity; and a method for preparing water with less carbonate was developed. The proposed method was applied for a highly sensitive determination of TC in purified water and various natural water samples.

## Experimental

### Apparatus

A flow-injection system for the on-site measurement of TC was assembled using a portable FIA (Asahi Technicon Co.), as used before.<sup>13</sup> The system (Fig. 1(a)) consisted of a double

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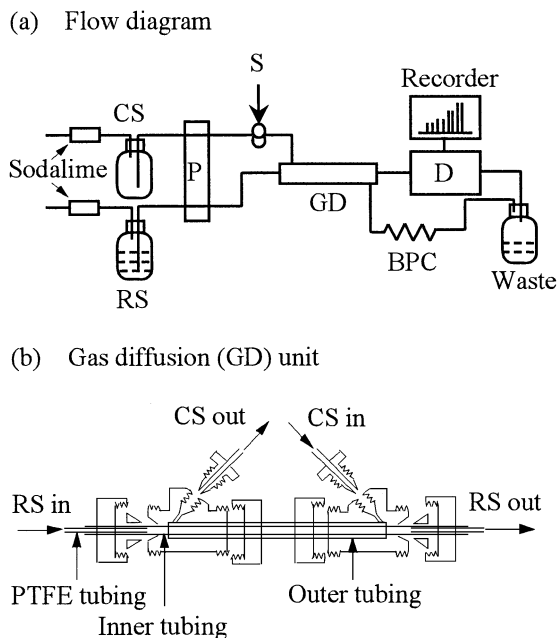


Fig. 1 Flow diagram for the total carbonate determination (a) and gas diffusion unit (b). CS, carrier solution ( $1.8 \times 10^{-3}$  M sulfuric acid); RS, reagent solution ( $2.5 \times 10^{-5}$  M DNN5S or cresol red, pH = 9.0); P, double plunger pump (each flow rate was  $0.3 \text{ ml min}^{-1}$ ); S, sample injection ( $200 \mu\text{l}$ ); GD, gas-diffusion unit (inner tube is GORE-TEX TA001: i.d., 1.0 mm; o.d., 1.8 mm; pore size, 2  $\mu\text{m}$ ; porosity, 50%; inner diameter of outer glass tube, 2 mm); BPC, back pressure coil; D, detector (450 nm).

plunger micro pump, a sample injector, a reaction coil, a detector with a flow cell and a flow-rate controller installed in the pump unit. In the detector, an LED was used as a light source, because the consumption of electricity is very low compared with a tungsten lamp. In this system, a double-plunger micro pump was used for propelling a carrier and a reagent solution; each flow rate was  $0.3 \text{ ml min}^{-1}$ . Sample solutions were injected into the carrier stream by a six-way injection valve; the sample size was  $200 \mu\text{l}$ . The gas-diffusion unit (GD unit, Fig. 1(b)) was constructed with a PTFE membrane tube (GORE-TEX TA001: 1.0 mm inner diameter and 1.8 mm outer diameter; pore size, 2  $\mu\text{m}$ ; porosity, 50%) as an inner tube; the outer tube was made of glass (2.0 mm inner diameter and 5.0 mm outer diameter) in a similar manner as before.<sup>12</sup> Two connectors made of PEEK were used to combine those two tubes. Ferrules used for fitting the tubes were reformed with a silicon material for easier connection of each part and the assembly of the GD unit. Flow-line tubes were inserted directly into the inner tube of the GD unit, which could lower the dead volume and the dispersion of a sample zone. The absorbances were measured at 450 nm using a light-emitting diode (LED) as a light source with an interference filter (450 nm); a micro flow cell (light path 10 mm, volume 8  $\mu\text{l}$ ) was also used. Signals were recorded with a strip chart recorder, or stored in a note-type personal computer.

#### Reagents

Ten kinds of reagents (Fig. 2) for color-development were synthesized by diazotization in a similar manner as in previous work.<sup>8</sup> 4-(2',4'-Dinitrophenylazo)-1-naphthol-5-sulfonic acid (DNN5S) was found to be the best of all the reagents examined.

The reagent solution contained  $2.5 \times 10^{-5}$  M DNN5S and  $3 \times$

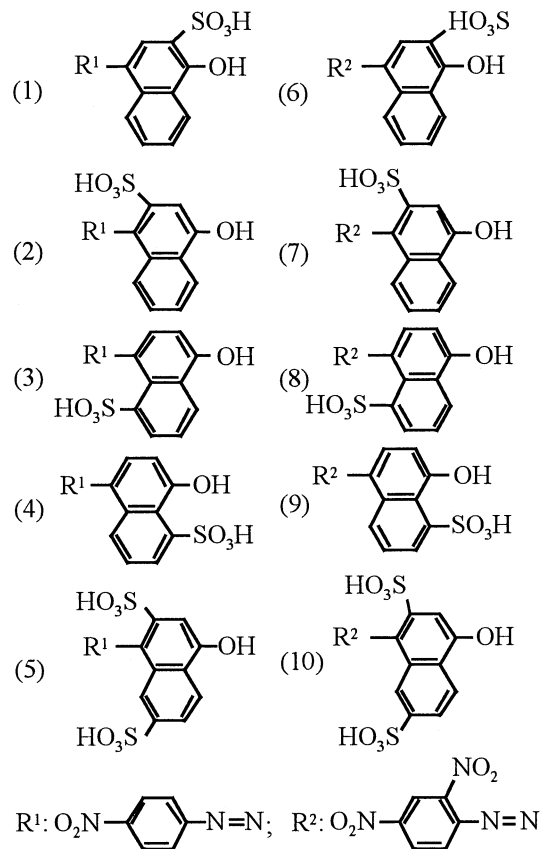


Fig. 2 Synthesized color-developing reagents.

$10^{-3}$  M sodium hydrogencarbonate; the pH of the reagent solution was adjusted to 9.0 with sodium hydroxide.

The carrier solution was  $1.8 \times 10^{-3}$  M  $\text{H}_2\text{SO}_4$ . Water with less carbonate was used for preparing the reagent and the carrier solution. For avoiding  $\text{CO}_2$  contamination from air, the solutions used in this work were all stored in an  $\text{N}_2$ -filled glass vessel with a soda-lime tube after their preparation.

## Results and Discussion

#### Selection of color-developing reagent

In previous work, cresol red was used.<sup>1,3,12</sup> For a sensitive determination, the difference in the molar absorptivity ( $\epsilon$ ) between a protonated form (HR) and a deprotonated form ( $\text{R}^-$ ) and the ratio of  $\epsilon_{\text{HR}}$  to  $\epsilon_{\text{R}^-}$  must be large. However, as shown in Table 1, the difference between  $\epsilon_{\text{HR}}$  and  $\epsilon_{\text{R}^-}$  at 450 nm and the ratio of  $\epsilon_{\text{HR}}$  to  $\epsilon_{\text{R}^-}$  are not very large. In this work, several kinds of commercially available acid-base indicators possessing  $\text{pK}_a$  at around 8.5, such as thymol blue, phenol red, *p*-xylenol blue, *m*-cresol purple, azo violet, tropeolin OO, and tropeolin O, were examined for improving the sensitivity in a carbonate determination. Of these, more sensitive reagents than cresol red could not be found. Therefore, the development of new reagents was examined. A color-developing reagent should have appropriate  $\text{pK}_a$  values of around 8.5–9.0, because the  $\text{pK}_a$  values of carbonate are 6.5 and 10.3, and its molar absorptivity of HR ( $\epsilon_{\text{HR}}$ ) must be large at 450 nm, whereas the molar absorptivity of  $\text{R}^-$  ( $\epsilon_{\text{R}^-}$ ) must be small. The acid-dissociation constants and their characteristics of absorption spectra of synthesized reagents were examined; the results are shown in

Table 1 Spectral data and acid-dissociation constants for color-developing reagents

Reagent <sup>a</sup>	pK <sub>a</sub>	$\lambda_{\max}/\text{nm}$		$\epsilon$ at $\lambda_{\max}(\text{HR})^b$		$\epsilon_{(\text{HR})}/\epsilon_{(\text{R}^-)}$ at $\lambda_{\max}(\text{HR})^c$	$\Delta\epsilon$ at $\lambda_{\max}(\text{HR})^d$	$\epsilon$ at 450 nm <sup>e</sup>		$\epsilon_{(\text{HR})}/\epsilon_{(\text{R}^-)}$ at 450 nm <sup>f</sup>	$\Delta\epsilon$ at 450 nm <sup>g</sup>
		HR	R <sup>-</sup>	HR	R <sup>-</sup>			HR	R <sup>-</sup>		
(1)	7.51	492	562	30.4	10.8	2.8	19.6	14.5	5.6	2.6	8.9
(2)	11.18	482	500	9.6	9.5	1.0	0.2	5.0	4.9	1.0	0.1
(3)	8.33	504	544	21.5	14.2	1.5	7.4	8.6	5.1	1.7	3.5
(4)	9.87	468	604	4.7	3.6	1.3	1.1	4.4	3.9	1.1	0.5
(5)	10.38	496	504	18.7	17.6	1.1	1.1	9.2	9.2	1.0	0.0
(6)	7.51	486	608	7.6	3.3	2.3	4.4	6.4	3.8	1.7	2.6
(7)	7.48	462	594	14.2	5.5	2.6	8.8	13.6	4.6	2.9	9.0
(8)	8.95	464	618	28.7	4.0	7.2	24.7	25.8	3.3	7.8	22.5
(9)	9.05	464	604	20.0	7.0	2.9	13.0	18.3	6.4	2.9	11.9
(10)	6.72	462	588	28.7	10.2	2.8	18.5	25.8	7.7	3.4	18.1
Cresol red	8.32	436	573	14.0	2.1	6.7	11.9	13.0	2.0	6.5	11.0
Thymol blue	9.34	436	597	13.7	2.2	6.4	11.5	13.1	2.4	5.6	10.7
Phenol red	8.53	432	561	23.1	2.6	8.9	20.5	21.0	4.2	5.0	16.8
<i>p</i> -Xylenol blue	9.73	438	595	16.8	3.2	5.3	13.6	16.1	3.5	4.6	12.1
<i>m</i> -Cresol purple	9.17	434	580	16.9	2.7	6.3	14.2	16.0	3.6	4.4	12.4
Azo violet	11.87	452	554	58.4	21.6	2.7	36.8	51.3	11.6	4.4	39.7
Tropeolin OO	9.43	478	519	28.0	19.8	1.4	8.2	20.8	12.4	1.7	8.4
Tropeolin	10.51	430	491	38.4	19.2	2.0	19.2	31.0	18.1	1.7	12.9

a. Reagents (1) - (10) were synthesized and correspond to those in Fig. 2. b. Molar absorptivity at each absorption maxima of HR:  $10^3 \text{ l mol}^{-1} \text{ cm}^{-1}$ . c. Ratio of molar absorptivity of HR to R<sup>-</sup> at  $\lambda_{\max}(\text{HR})$ . d. Difference in molar absorptivity at  $\lambda_{\max}(\text{HR})$  between HR and R<sup>-</sup>. e. Molar absorptivities at 450 nm (detection wavelength):  $10^3 \text{ l mol}^{-1} \text{ cm}^{-1}$ . f. Ratio of molar absorptivity of HR to R<sup>-</sup> at 450 nm. g. Difference in molar absorptivity at 450 nm between HR and R<sup>-</sup>.

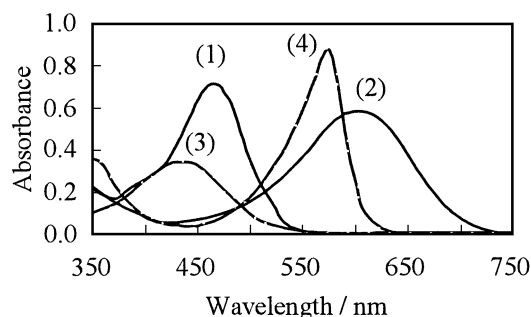


Fig. 3 Absorption spectra of DNN5S and cresol red. (1) and (2), DNN5S ( $2.5 \times 10^{-5} \text{ M}$ ); (3) and (4), cresol red ( $2.5 \times 10^{-5} \text{ M}$ ); (1) and (3), acidic form; (2) and (4), basic form.

Table 1, together with those obtained for commercially available reagents. Of those, 4-(2',4'-dinitrophenylazo)-1-naphthol-5-sulfonic acid (DNN5S, the structure is shown in Fig. 2 (8)) was found to be the most suitable for the present purpose with respect to the acid-dissociation constant, the relatively large difference in the absorption maximum wavelength, the large difference at 450 nm between  $\epsilon_{\text{HR}}$  and  $\epsilon_{\text{R}^-}$  and the large ratio of  $\epsilon_{\text{HR}}$  to  $\epsilon_{\text{R}^-}$ . They were  $2.25 \times 10^4 \text{ l mol}^{-1} \text{ cm}^{-1}$  and 7.8, respectively, which were larger than those of cresol red. According to Van der Linden's consideration about the selection of a suitable indicator,<sup>10</sup> DNN5S may be the most suitable indicator for this work. The absorption spectra of DNN5S, as well as those of cresol red, are shown in Fig. 3. DNN5S has a suitable pK<sub>a</sub> value (8.95) and shows a sensitive color change with a change in its dissociated form to a protonated form at 450 nm. A comparison of the flow signals of cresol red with DNN5S showed that the sensitivity of DNN5S is twice as large as that of cresol red. The baseline stability obtained by using cresol red and DNN5S, which can also affect the improvement in the sensitivity and

Table 2 Stability and noise of baseline obtained with DNN5S and cresol red (CR)

AUF <sup>a</sup>	Baseline signal	
	DNN5S	CR
0.5	_____	_____
0.2	_____	_____
0.1	_____	_____
0.05	_____	_____
0.02	_____	_____
0.01	_____	_____

10 min  
↔

a. Absorbance unit full scale of the detector.

reproducibility of the determination, were compared with each other under the same experimental conditions (reagent concentration,  $2.5 \times 10^{-5} \text{ M}$ ; total flow rate,  $0.6 \text{ ml min}^{-1}$ ). The obtained results are shown in Table 2. When the range of the absorbance unit full scale (AUF) was changed to more than 0.1, less difference in the stability and noise of the base line between DNN5S and cresol red was found, whereas in AUF less than 0.1 DNN5S was obviously found to be better than those of cresol red. Therefore, DNN5S was selected as a color-developing reagent for the sensitive determination of carbonate. By using DNN5S, 0.02 AUF could be used for a sensitive determination of TC.

#### Preparation of water with less carbonate

Positive signals appeared when purified water samples

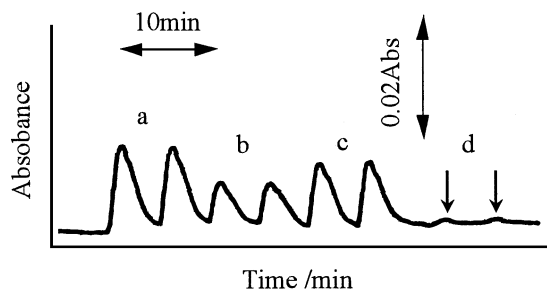


Fig. 4 Flow signals obtained for various kinds of purified water samples. (a) Water purified with the Milli Q Labo, which was agitated for 8 h in the atmosphere,  $2.2 \times 10^{-5}$  M; (b) water (Milli Q Labo) purified by bubbling with nitrogen gas for 0.5 h,  $1 \times 10^{-5}$  M; (c) fresh water purified with Milli Q Labo,  $1.7 \times 10^{-5}$  M; (d) water with less carbonate (see text),  $5 \times 10^{-7}$  M.

obtained by a deionization/distillation method or by treating it with Milli Q Labo (Millipore) were injected into the carrier shown in Fig. 1(a) as a blank. This indicates that the purified water prepared in the usual ways contained small amounts of carbonate. To eliminate such carbonate dissolved in purified water and to prepare water with less carbonate, the purification of water was examined. Deionized-distilled water was treated with a purification apparatus (Milli Q Labo) for 6 h, and transferred into a glass bottle equipped with a soda-lime tube. Nitrogen gas was then introduced in this water from an  $N_2$  cylinder and bubbled for 1.5 h in a water bath at  $50^\circ\text{C}$  under ultrasonic radiation. After being cooled to room temperature, it was used as water with less carbonate. This water was used for preparing the carrier solution. During a measurement of the total carbonate, soda-lime tubes were equipped to the carrier and the reagent bottles to avoid the contamination of carbonate from air. In Fig. 4, peak profiles of the water with less carbonate, as well as other purified water samples, are shown. The amount of carbon dioxide dissolved in the water with less carbonate was found to be  $5 \times 10^{-7}$  M, which indicates the difference in the concentration between this water and the carrier acidified with sulfuric acid. The carbonate content in the water with less carbonate is sufficiently low for the present purpose of a sensitive determination of carbonate at the  $10^{-6}$  M level. By using this water, the absorption behavior of carbon dioxide in the atmosphere was examined under the agitation of water. As shown in Fig. 5, carbon dioxide in the atmosphere gradually dissolved into the water with less carbonate, and was equilibrated slowly with the total carbonate in the water; the equilibrated concentration of carbon dioxide in the water was  $2.2 \times 10^{-5}$  M at  $25^\circ\text{C}$ .

#### Optimization of FIA conditions for a sensitivity improvement

Sample solutions were injected into a carrier stream (CS) containing sulfuric acid, where dissolved carbonate was converted to gaseous carbon dioxide; then, generated carbon dioxide was passed through a PTFE membrane tube, and mixed with the reagent stream (RS), which contained DNN5S and buffer components. The resulting pH change in RS caused an absorbance change of the reagent (DNN5S). The absorbance was measured at 450 nm with an LED as a light source. The optimal concentration of sulfuric acid in CS and pH of RS were the same as in a previous study.<sup>2</sup>

The effect of the flow rate on the peak profiles was examined by varying it from 0.2 to  $1.0 \text{ ml min}^{-1}$ . As shown in Fig. 6, the height of the signals increased along with a decrease in the flow

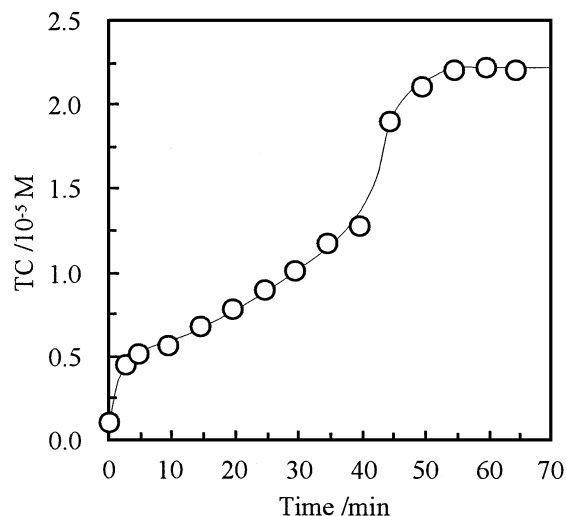


Fig. 5 Absorption equilibrium of  $\text{CO}_2$  between water with less carbonate and air. The equilibrated concentration of carbonate was  $2.2 \times 10^{-5}$  M at  $25^\circ\text{C}$ .

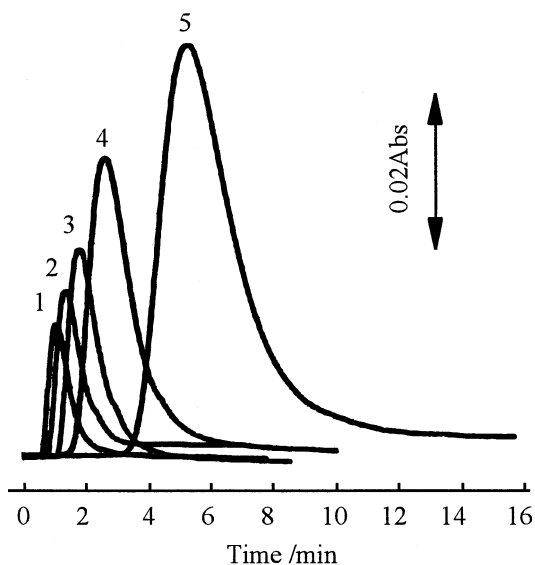


Fig. 6 Effect of the flow rate on the peak profiles. Each flow rate ( $\text{ml min}^{-1}$ ): (1) 0.5; (2) 0.4; (3) 0.3; (4) 0.2; (5) 0.1; sample,  $8.0 \times 10^{-5}$  M  $\text{NaHCO}_3$ ; sample size,  $100 \mu\text{l}$ ; GD unit length, 15 cm.

rate, because the generated carbon dioxide needed much time to diffuse through the membrane and into the reagent solution. However, the decrease in the flow rate resulted in an increase in the residence time. Considering the sensitivity and sampling time, each flow rate was fixed to  $0.3 \text{ ml min}^{-1}$  as a compromise.

The effect of the sample size on the peak profiles was examined by varying from 20 to  $700 \mu\text{l}$ . With an increase in the sample size, the peak height increased, and reached its maximum at  $200 \mu\text{l}$ , above which the peak broadening was remarkable. When the sample size was larger than  $500 \mu\text{l}$ , the peaks split into two peaks. This is because the middle region of the sample zone could not mix effectively with the carrier solution. The sample size adopted in the proposed system was determined to be  $200 \mu\text{l}$ .

The effect of the length of the reaction coil from sample injection to GD was examined. The peak height was almost the

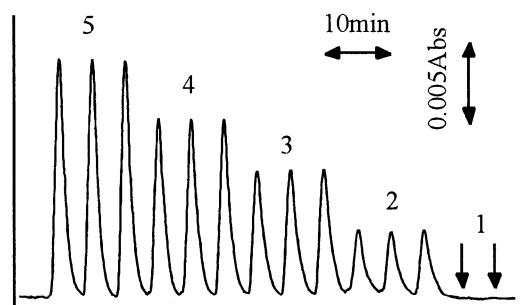


Fig. 7 Flow signals for the determination of the total carbonate (TC) at low concentrations. Sample ( $\text{NaHCO}_3/10^{-5}$  M): (1) blank (water with less carbonate); (2) 2.0; (3) 4.0; (4) 6.0; (5) 8.0; sample size, 200  $\mu\text{l}$ ; GD unit length, 15 cm.

same as a length of 5 – 100 cm, which means that the generation of permeable carbon dioxide occurred immediately. In the proposed system, a 5 cm reaction coil was sufficient.

This meant that the efficiency of the diffusion of the gaseous substance through the PTFE membrane was improved so as to be higher than in the previous system.<sup>3</sup> To improve the permeable efficiency, a back-pressure coil was connected just after the GD unit. The length of the back-pressure coil (0.25 mm i.d.) was examined from 0 to 50 cm; the peak height was increased by up to 1.5 times. Therefore, a length of 50 cm was adopted. The length of the PTFE membrane tube was tested by varying it from 5 cm to 60 cm. The peak height obtained with 60 cm was 2-times higher than that with 5 cm. However, the peaks became wider and the occurrence of small air bubbles became remarkable with an increase in the length. From the viewpoint of convenience and a stable background, the length of the PTFE membrane tube was fixed at 15 cm. For maintaining the hydrophobicity and the permeability of the membrane tube, the carrier and the reagent solution were removed from the GD unit after the experiment was finished, and air was introduced before and after the GD unit.<sup>12</sup> By substituting air for aqueous solutions, the hydrophobicity of the PTFE membrane was maintained for several months.

#### Improvement of gas-diffusion unit

The outer tube of the GD unit was changed from PTFE to a rigid glass tube (2.0 mm inner diameter and 5.0 mm outer diameter). By using a glass tube, the aqueous streams became stable, and furthermore the assembly became easier than that in the previous one.<sup>1-3</sup> Two tee connectors were changed for Y connectors made of PEEK for lowering the dead volume in the connectors. Ferrules made of silicon material were used for easier and more reliable connections; the flow-in and flow-out tubes for the reagent stream could be inserted directly into the inner tube of the PTFE membrane, and fastened tightly. Two 6-way valves were equipped in the carrier and the reagent line for removing the solutions and substituting air for them in the GD unit.

#### Calibration graph

The calibration graphs were linear over a wide dynamic range from  $10^{-6}$  M to  $10^{-3}$  M. An example of the calibration graph of the total carbonate (TC) at the  $10^{-5}$  M level is shown in Fig. 7, where TC in the water with less carbonate could not be detected. The relative standard deviation (RSD) of 10 measurements of the distilled water (average concentration: 2.3

Table 3 Determination of the total carbonate in water samples

Sample	Found/ $10^{-4}$ M
River water A	5.8
River water B	4.2
Seawater A	20.0
Seawater B	14.0
Tap water	4.6
Rain water	0.20
Ultrapure water <sup>a</sup>	0.23
Ultrapure water <sup>b</sup>	0.17
Ultrapure water <sup>c</sup>	0.22
Water with less carbonate <sup>d</sup>	~0.005

a. Deionized and distilled water.

b. Fresh water purified with Milli Q Labo.

c. Water purified with Milli Q Labo, and equilibrated with carbon dioxide in air.

d. See text.

$\times 10^{-5}$  M) was 1.9%. The limit of detection (LOD) corresponding to a signal-to-noise ratio of three was  $1 \times 10^{-6}$  M of carbonate, which was two-times the concentration of the water with less carbonate. The sampling rate was about 20 samples  $\text{h}^{-1}$ .

#### Application to the determination of carbonate in water samples

The proposed method was applied to the determination of carbonate in water samples, such as natural, rain, tap and purified water samples. The results are shown in Table 3. Because the proposed method is highly sensitive, the total carbonate concentration in the water with less carbonate, which was purified by the deionization/distillation method, with a Milli Q Labo and with  $\text{N}_2$ -bubbling under ultrasonic radiation, could be determined to be about  $5 \times 10^{-7}$  M.

By using the established system, the total carbonate in environmental water samples, such as rain water, river and sea water, was found to be easily determined on site.

## Conclusion

An FIA system with a gas-diffusion apparatus was improved for the sensitive and reproducible determination of carbonate in water samples, and a new color-developing reaction with DNN5S (4-(2',4'-dinitrophenylazo)-1-naphthol-5-sulfonic acid) was proposed. For an accurate determination, the method for preparing water with less carbonate was investigated according to simple procedures. By the proposed sensitive system, a highly sensitive method with an LOD of  $1 \times 10^{-6}$  M was developed.

## Acknowledgements

This work was supported by Grant-in-Aid No. 10558084 from the Ministry of Education, Science, Sports and Culture, Japan.

## References

1. S. Motomizu, K. Toei, T. Kuwaki, and M. Oshima, *Anal. Chem.*, **1987**, 592, 930.
2. T. Kuwaki, K. Toei, M. Akiba, M. Oshima, and S. Motomizu, *Bunseki Kagaku*, **1987**, 36, T132.



3. M. Sanada, M. Oshima, and S. Motomizu, *Bunseki Kagaku*, **1993**, *42*, T123.
  4. V. Kuban and P. K. Dasgupta, *Talanta*, **1993**, *40*, 831.
  5. P. Linares, M. D. Lague de Castro, and M. Valcarcel, *Anal. Chim. Acta*, **1985**, *225*, 443.
  6. T. Aoki, Y. Fujimura, Y. Oka, and K. Fujie, *Anal. Chim. Acta*, **1993**, *284*, 167.
  7. T. Kuwaki, M. Akiba, M. Oshima, and S. Motomizu, *Bunseki Kagaku*, **1987**, *36*, T81.
  8. T. Takayanagi, H. Tanaka, and S. Motomizu, *Anal. Sci.*, **1997**, *13*, 11.
  9. W. Stumm and J. J. Morgan, "Aquatic Chemistry", 2nd ed., **1981**, Wiley, New York, 149.
  10. W. E. Van der Linden, *Anal. Chim. Acta*, **1983**, *155*, 273.
  11. R. M. Carlson, *Anal. Chem.*, **1978**, *50*, 1528.
  12. K. Higuchi, A. Inoue, T. Tsuboi, and S. Motomizu, *Bunseki Kagaku*, **1999**, *48*, 253.
  13. L. Ma, M. Oshima, T. Takayanagi, and S. Motomizu, *J. Flow Injection Anal.*, **2000**, *17*, 188.
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