Direct Determination Method of Nitrate Ions in Seawater by UV-Detection Ion-Chromatography with Hydrochloric Acid/Sodium Chloride Eluent

Akiharu HIOKI*, James W. McLAREN**

(May 12, 2008)

Abstract

Nitrate ions in seawater have been determined by ion-chromatography using a chloride-form ion-exchange column. Hydrochloric acid (HCl)/sodium chloride (NaCl) mixtures had an excellent effect on resolution and sensitivity. Neither dilution nor pre-treatment of seawater was needed. The limit of detection was 3.9 ng g^{-1} as NO_3^- in seawater under the eluent condition of 40 mmol L^{-1} HCl/160 mmol L^{-1} NaCl at 225 nm. Nitrite ions in seawater can be also determined in the same way without hydrochloric acid. The limit of detection was 1.2 ng g^{-1} as NO_2^- in seawater under the eluent condition of 160 mmol L^{-1} NaCl at 225 nm. The effect of a counter cation in an eluent is also discussed.

Introduction

Since the micronutrient elements (nitrogen, phosphorus and silicon) take essential roles in the marine ecosystem, it is important to monitor those elements in seawater. About 15 years ago, the National Research Council of Canada (NRC) undertook the preparation of a seawater certified reference material (CRM) for micronutrients (nitrate, phosphate and dissolved silica). In order to satisfy the NRC criterion for certification, good agreement between at least two independent methods is required. Therefore, the present study was carried out for the purpose of developing another simple and reliable method for the determination of nitrate ions at less than 1 μg g^{-1} level in seawater to complement the well-developed standard colorimetric method.1)

The strong absorption of nitrate ions in the UV region can be utilised for determination.2) In order to apply it to seawater, some pre-treatment has been required due to the interference of bromide ions and dissolved organic compounds,3) though chloride ions and sulfate ions as major constituents do not come into question. It is possible to separate nitrate ions from such kinds of ions in seawater using ion-chromatography.4)–6) One of advantages of ion-chromatography is higher selectivity compared with colorimetry. In a method in which a mixed solution of sodium chloride and sodium sulfate was used as an eluent,6) the limit of detection (LOD) for nitrate ions in low salinity seawater was reported to be 13 ng g^{-1}; on the other hand in other methods4), 5) LOD's in seawater have not been reported definitely except for LOD of nitrite ions.4) There is a report where a potassium chloride solution was used as an eluent4) and another report where a mixed solution of sodium chloride and phosphate buffer was used as an eluent for matrix elimination of 20 mg g^{-1} chloride ions.7) Though there are also some reports of ion-chromatography using a conductivity detector,8)–12) LOD's are insufficient for the accurate determination of nitrate ions in seawater at less than μg g^{-1} level, otherwise have not been reported. A possibility of LOD improvement has been suggested,9) but the approach requires pre-treatment with a silver-cation exchanger. In the present study using a chloride-form ion-exchange column, we have found the excellent effect of hydrochloric acid on the resolution for nitrate ions of chromatogram of a seawater sample. Moreover, the simultaneous determination of nitrite ions in seawater has been mentioned. The ion-chromatographic method for nitrate ions and nitrite ions was used for the production of the NRC CRM MOOS-1.13)–15) Though there have been some other reports on ion-chromatographic methods for nitrate ions in seawater recently,16)–19) LOD's in those studies are either not reported or larger than the LOD of the present study. The present study has some novelty to be reported, including the detailed investigation on the eluent conditions.

---

* Inorganic Standards Section, Inorganic Analytical Chemistry Division, MIJ

** Institute for National Measurement Standards, National Research Council of Canada

Experimental

Reagents

Water was purified by double distillation. Unless otherwise specified, all reagents used were of analytical reagent grade. Nitrate standard solutions and nitrite standard solutions were prepared from potassium nitrate (Anachemia, Montreal, Canada) and sodium nitrite (purity of 99.40 %, Fisher, Fair Lawn, USA), respectively. The seawater sample used was gathered in the Atlantic, followed by immediate filtration through 0.45 μm porosity filters; unless otherwise stated, this is referred to as seawater or a seawater sample. The seawater was different from NRC CRM MOOS-1 under current dissemination.

Instrument

The pump used for ion chromatography was an advanced gradient pump AGP-1 from Dionex (Sunnyvale, USA) equipped with four pressurized reservoirs. The flow rate was always 1.0 cm^3 min^{-1}. Sample injections were carried out through a microinjection valve of a Dionex Laboratory Chromatography Module model LCM. The volume of injection loop was *ca.* 50 μL. Unless otherwise stated, the UV detector used was a Waters (Milford, USA) model 450, equipped with a flow cell of 9.50 mm light path. The ion-exchange column used was a Dionex IonPac AS10 (4 mm I.D. and 250 mm long), which has a capacity of 170 μeq a column, and a Dionex Ion Pac AG10 (4 mm I.D. and 50 mm long) was used as a guard column. These columns are useable over the pH range from 0 to 14. For the purpose of only the comparison of UV detectors, a model 626 LC ion-chromatography system (Waters) equipped with both a manual injection valve and a model 486 absorbance detector (Waters) was also used. The light path of the flow cell of the model 486 detector was 10 mm. The laboratory temperature was (23 ± 2) ℃ in general.

Recommended procedure

An approximately 0.5 cm^3 portion of a seawater sample is directly introduced into the injection loop to ensure complete filling. The eluent composition of 40 mmol L^{-1} hydrochloric acid (HCl)/160 mmol L^{-1} sodium chloride (NaCl) can be chosen (*vide infra*). If necessary, there are many choices of eluent composition. Though the UV detection can be carried out at any wavelength between 210 nm and 235 nm, 225 nm was used unless otherwise specified. The chromatograms are recorded and the peak heights are calculated. Though the peak areas can be also utilised, those were not used here. A standard addition method or an addition calibration method should be adopted, because of the slight reduction of the peak height by a salt effect. Since the addition of a standard solution was carried out by weighing, the seawater density (1.023 g cm^{-3}) was used for the conversion into a volume concentration.

Results and Discussion

Selection of wavelength for detection

Figure 1 shows the UV absorption spectra of nitrate ions, chloride ions, bromide ions and seawater. Though a shorter wavelength is preferable as far as nitrate ions are concerned, the absorption of chloride and bromide ions becomes larger with decreases in wavelength. Judging from Figure 1, wavelengths near 225 nm have an advantage from the viewpoint of interferences of chloride and bromide ions. We selected the wavelength of 225 nm to examine separation performances. The dependence on wavelengths will be discussed later.

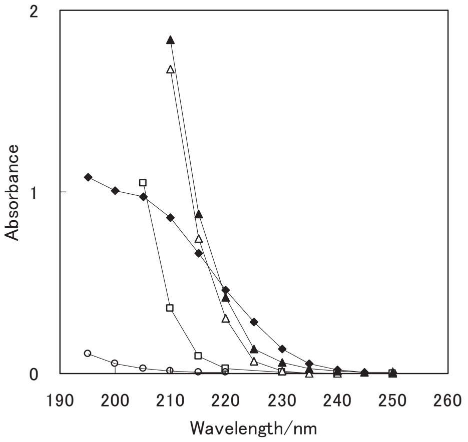

Figure 1. UV absorption spectra of nitrate ions, chloride ions, bromide ions and seawater. Reference: air. ◆: 100 μmol L^{-1} NO_3^-; □: 3.3 % NaCl; △: 0.0085 % NaBr; ▲: seawater; ○: water.

NaCl/HCl eluent

Figure 2 illustrates chromatograms of a seawater sample using various HCl/NaCl eluents. The resolution (*R*) between two adjacent peaks was defined as eq (1).

$$R = 2\,\Delta Z/(w_1 + w_2) \qquad (1)$$

where ΔZ is the difference of retention times between the

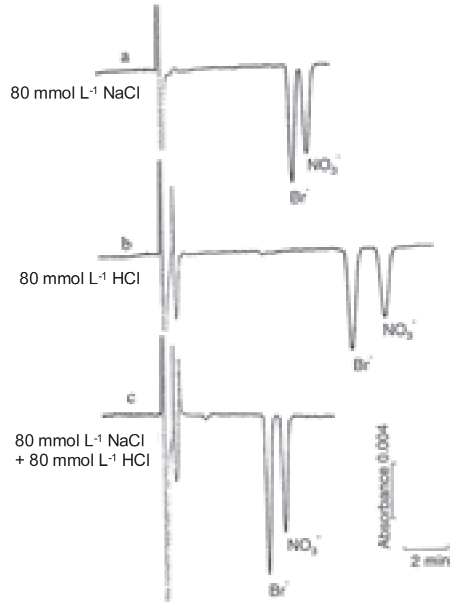

Figure 2. Chromatograms of a seawater sample. $[NO_3^-]$ = *ca.* 1 $\mu g\ g^{-1}$. Wavelength: 225 nm. (a) Eluent: 80 $mmol\ L^{-1}$ NaCl; (b) eluent: 80 $mmol\ L^{-1}$ HCl; (c) eluent: 80 $mmol\ L^{-1}$ NaCl/80 $mmol\ L^{-1}$ HCl.

two peaks, and w_1 or w_2 is each baseline peak width which is the difference of retention times between the two points where the tangents at the inflexion points of the peak across the baseline. The resolution between bromide ions and nitrate ions under the condition of an 80 $mmol\ L^{-1}$ HCl eluent was much better than that under the condition of an 80 $mmol\ L^{-1}$ NaCl eluent. Under the condition of 80 $mmol\ L^{-1}$ HCl/80 $mmol\ L^{-1}$ NaCl, both the resolution and the sensitivity were better than those under the condition of 80 $mmol\ L^{-1}$ NaCl, respectively. Thus, we examined the separation performance over a wide range of HCl/NaCl concentrations.

Figure 3-a shows the dependence of the resolution of bromide ions and nitrate ions at 225 nm on the concentrations of both HCl and NaCl. The rough outline of resolution contour in the HCl/NaCl field is depicted in Figure 3-b. Since any condition of a resolution over 1.5 which gives a complete separation[20)] is usually used for determining any analyte, from many choices we can select the conditions which are preferable from the sensitivity point of view. Figure 3-c shows a strange local maximum of the resolution near 40 $mmol\ L^{-1}$ NaCl; the reason of the local maximum appearance is not clear. Since the small concentration of HCl near zero gave relatively large influence to the resolution, an

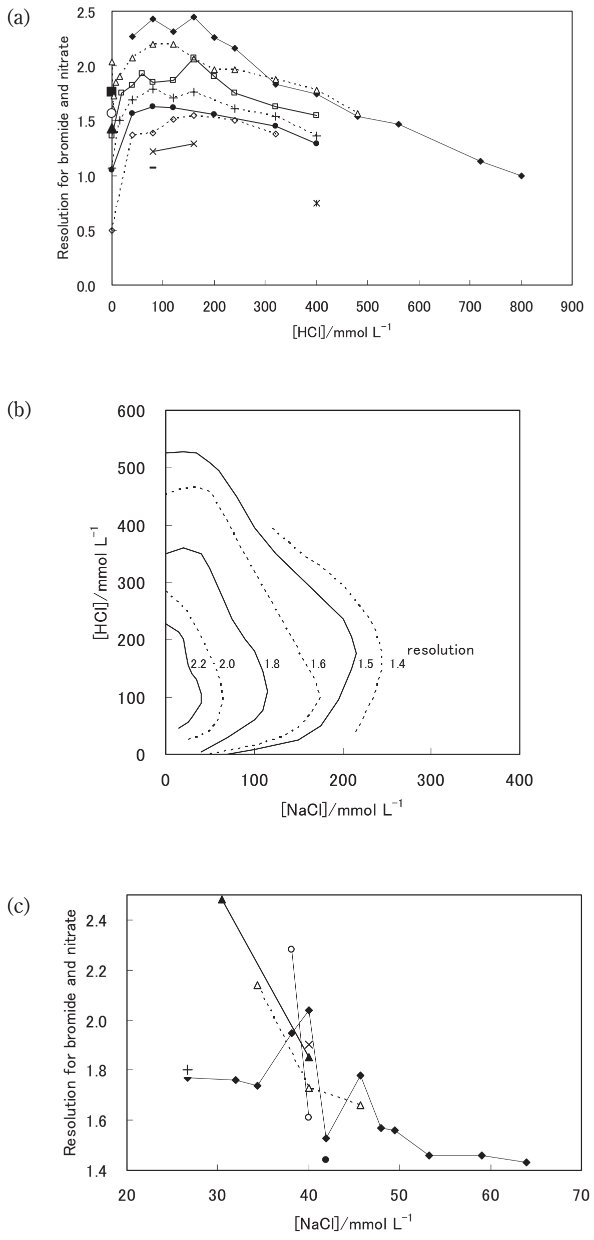

Figure 3. Dependence of resolution at 225 nm on concentrations of HCl and NaCl in eluent. $[NO_3^-]$ = *ca.* 1 $\mu g\ g^{-1}$. (a) Results obtained. Eluent: ◆, [NaCl] = 0 $mmol\ L^{-1}$; ■, 32 $mmol\ L^{-1}$; △, 40 $mmol\ L^{-1}$; ○, 48 $mmol\ L^{-1}$; ▲, 64 $mmol\ L^{-1}$; □, 80 $mmol\ L^{-1}$; +, 120 $mmol\ L^{-1}$; ●, 160 $mmol\ L^{-1}$; ◇, 200 $mmol\ L^{-1}$; ×, 280 $mmol\ L^{-1}$; −, 360 $mmol\ L^{-1}$; ⋇, 400 $mmol\ L^{-1}$. (b) Rough outline of resolution contour in [NaCl]-[HCl] field. The numbers on the lines indicate the values of resolution. (c) Local maximum of resolution near 40 $mmol\ L^{-1}$ NaCl. Eluent: ◆, [HCl] = 0 $mmol\ L^{-1}$; ○, 2 $mmol\ L^{-1}$; △, 4 $mmol\ L^{-1}$; ▲, 8 $mmol\ L^{-1}$; +, 12 $mmol\ L^{-1}$; ×, 16 $mmol\ L^{-1}$; ●, [NaOH] = 2 $mmol\ L^{-1}$.

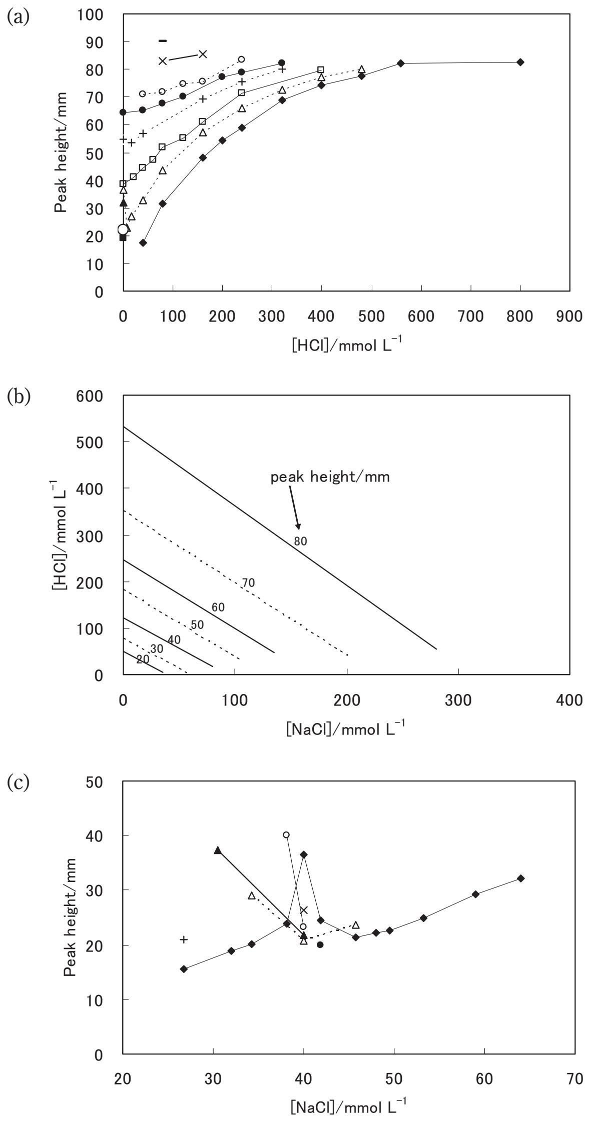

Figure 4. Dependence of sensitivity for nitrate ions at 225 nm on concentration of HCl and NaCl in eluent. The height of 100 mm corresponds to the absorbance of 0.016. [NO$_3^-$] = *ca.* 1 μg g^{-1}. (a) Results obtained. The symbols in Figure 4-a are the same as those in Figure 3-a. (b) Rough outline of sensitivity contour for nitrate ions in [NaCl]-[HCl] field. The numbers on the lines indicate the values of sensitivity. (c) Local maximum of sensitivity near 40 mmol L^{-1} NaCl. The symbols in Figure 4-c are the same as those in Figure 3-c.

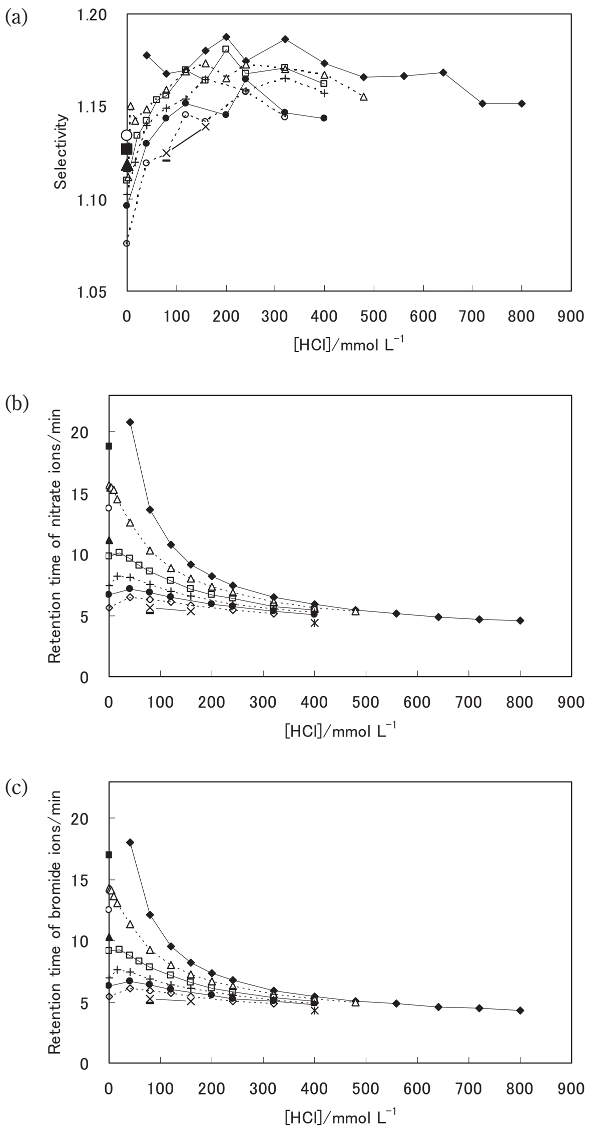

Figure 5. Dependencies of selectivity between nitrate ions and bromide ions in seawater (a), of retention time of nitrate ions in seawater (b), and of retention time of bromide ions in seawater (c) on concentrations of HCl and NaCl in eluent. [NO$_3^-$] = *ca.* 1 μg g^{-1}. The symbols in Figure 5 are the same as those in Figure 3-a.

alkaline condition intending a negative acid concentration was tried. However, NaOH instead of HCl did not give any good effect on the resolution.

Figure 4-a represents the dependence of the sensitivity for nitrate ions at 225 nm on the concentrations of both HCl and NaCl. The sensitivity was defined as the peak height in the unit of mm in the full scale (250 mm) of 0.04 absorbance. The rough outline of sensitivity contour in the HCl/NaCl field is illustrated in Figure 4-b. The higher the eluent concentration was, the higher the sensitivity was. By the

comparison of Figures 3-b and 4-b, a suitable condition can be chosen, which partly depends on a possible interference from unknown substances in seawater. Figure 4-c shows a strange local maximum of the sensitivity near 40-mM NaCl similar to that shown for resolution in Figure 3-c; again, the reason is not clear. NaOH instead of HCl did not give any good effect on the sensitivity. The exact position of local maximum may depend on the salinity. If a 40 mmol L^{-1} NaCl eluent is used to determine nitrate ions, the separation performance should be preliminarily examined near the concentration of 40 mmol L^{-1}. A local maximum of the sensitivity for bromide ions was also observed near 46 mmol L^{-1} NaCl.

The selectivity (α) between two peaks was defined as eq (2).

$$\alpha = (t_2 - t_0)/(t_1 - t_0) \qquad (2)$$

where t_0 is the retention time corresponding to the sum of the void volume and the dead volume (2.8 min), and t_1 and t_2 are retention times for the two peaks ($t_1 < t_2$). The dependence of the selectivity between bromide ions and nitrate ions on the concentrations of both HCl and NaCl is indicated in Figure 5-a, which was depicted from the retention times of nitrate ions (Figure 5-b) and bromide ions (Figure 5-c). Generally speaking, the selectivity increased with increases in HCl concentration and with decreases in NaCl concentration. The slight effect of HCl on the selectivity seems to give the excellent effect on the resolution (Figure 5-a). Furthermore, the behaviour of the resolution was not monotonic as shown in Figures 3-a and 3-b; consequently the separation performance became interesting.

The repeatability of nitrate determination is summarised in Table 1. Satisfactory results were obtained under the different eluent conditions. These results also indicated that waiting times of 10 min to 18 min were sufficient for attaining to column equilibration.

Table 1. Sensitivity and RSD*

[HCl]/mmol L^{-1}	400	240	160	80	40	0	400
[NaCl]/mmol L^{-1}	80	200	120	0	160	80	80
waiting time for column equilibrium /min**	----	13.5	15	10	18	10	14.5
mean of peak height of NO_3^- /mm	----	69.7	59.8	28.2	57.9	36.8	68.1
RSD/%	----	1.6	0.4	1.3	0.9	0.6	1.4
number of measurements	----	5	5	5	5	5	5

* Indicated in the measurement order from the left to the right. $[NO_3^-]$ = *ca.* 1 μg g^{-1}.

** The waiting time after changing the previous eluent composition.

Behaviour of nitrite ions

Under the eluent condition of 200 mmol L^{-1} sodium hydroxide (NaOH), the retention times of nitrite ions and nitrate ions in seawater were *ca.* 6.5 min and *ca.* 18 min, respectively. There was no significant signal for nitrate ions in measuring a 5 μg g^{-1} nitrite standard solution. Following the eluent of 200 mmol L^{-1} NaOH, the nitrite standard solution under the eluent condition of 80 mmol L^{-1} NaCl gave a nitrite peak without any nitrate one. Under the same condition, nitrite ions were not detected in the seawater sample itself. A standard addition of nitrite ions to the seawater sample gave a good linear relationship up to at least 1 μg g^{-1} nitrite. The retention times of nitrite ions and nitrate ions were *ca.* 5.1 min and *ca.* 9.2 min, respectively. When the eluent was subsequently changed to 160 mmol L^{-1} HCl, the peak of nitrite ions in the nitrite standard solution and that in the nitrite-spiked seawater disappeared. A very small nitrate peak appeared for the former, but the nitrate peak for the latter did not increase compared with the nitrite-unspiked seawater. These phenomena were reproducible.

It seems that some kind of disproportionation of nitrite ions happened in the column. After an eluent condition including HCl, the peak of nitrite ions disappeared even under the eluent condition of 80 mmol L^{-1} NaCl. When the column was treated with a 200 mmol L^{-1} NaOH solution following the eluent condition of 80 mmol L^{-1} NaCl, the subsequent elution with 80 mmol L^{-1} NaCl gave definite nitrite peaks for the nitrite standard solution and for the nitrite-spiked seawater. Therefore, the nitrite disappearance seems to be caused by some substance from seawater accumulated on the column under an acidic eluent condition. Table 2 shows the effect of spiked nitrite ions on the peak heights of nitrate ions. The waiting time after any change of eluent composition was 22 min at least. Judging from the results, even under an eluent condition including HCl, 1 μg g^{-1} nitrite ions did not interfere the determination of 1 μg g^{-1} nitrate ions in seawater at all. Under the conditions of eluent without HCl, nitrite ions do not come into question because nitrite and nitrate ions can be detected separately. For the simultaneous determination of nitrate and nitrite ions, 80 mmol L^{-1} or 40 mmol L^{-1} NaCl can be used as an eluent. If nitrite ions alone are determined, 160 mmol L^{-1} NaCl is much more preferable.

Table 2. Effect of spiked nitrite ions on the peak heights (mm) of nitrate ions*

eluent	[HCl]/mmol L^{-1}	400	320	240	160	80	40	16	0
	[NaCl]/mmol L^{-1}	80	0	200	120	0	160	120	80
NO_3^- peak height	0.99 μg g^{-1} NO_2^-	*ca.*6	*ca.*5	*ca.*4	*ca.*5	3.2	2.6	3.3	3.0
	Seawater + 0.99 μg g^{-1} NO_2^-	72.6	63.2	76.2	64.1	32.2	60.9	52.1	39.9
	Seawater**	73.0	62.7	77.5	63.9	30.4	59.9	51.2	39.0

* Indicated in the measurement order from the left to the right. [NO_3^-] = *ca.* 1 μg g^{-1}.

** One percent weight of water was added to seawater in order to be consistent with the nitrite-spiked seawater.

Wavelength dependence and LOD's

The dependence of sensitivity of nitrate ions in seawater on wavelength is represented in Figure 6. With decreases in the wavelength, the sensitivity became higher, but the baseline noise also became larger. The resolution between bromide ions and nitrate ions was nearly independent of the wavelength. Moreover, Table 3 shows the LOD's for nitrate ions in seawater at 214 nm and 225 nm using the Waters model 486 UV detector which gave a much lower noise level than the Waters model 450 UV detector. LOD was defined as the concentration of nitrate ions in seawater corresponding to 3 times the noise height. The fluctuation in the time scale of tens of second in place of several seconds was adopted as the noise to calculate LOD. The shorter wavelength gave the better sensitivity but the worse LOD value. Under the condition of our instruments, 225 nm was recommended. Hu et al.[17] reported 4.0 ng g^{-1} as the LOD of nitrate ions in seawater using the sample volume of 100 μL, twice that of the present study. Ito et al. [19] recently reported a much smaller LOD (1.6 ng g^{-1}); however, their sample volume (200 μL) was four times that of the present study.

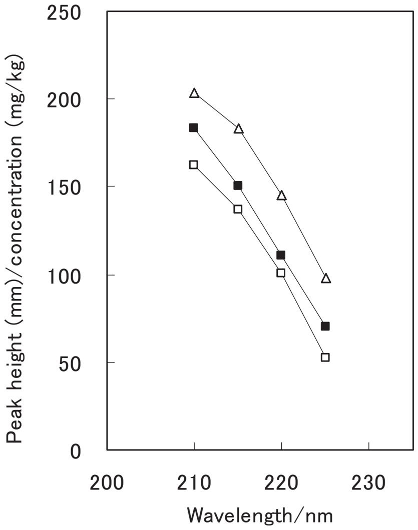

Figure 6. Dependence of sensitivity of nitrate ions and nitrite ions in seawater on wavelength. Eluent for NO_3^-: □, 240 mmol L^{-1} HCl; ■, 160 mmol L^{-1} HCl/120 mmol L^{-1} NaCl. Eluent for NO_2^-: △, 160 mmol L^{-1} NaCl. A seawater sample containing 1.060 μg g^{-1} as NO_3^- and 0.118 μg g^{-1} as NO_2^- was used.

Table 3. LOD's for nitrate determination

Wavelength (nm)	Eluent	Absorbance for NO_3^- peak height*[1]	Absorbance for noise height*[2]	LOD as NO_3^- (ng g^{-1})*[3]
214	[HCl]=240 mmol L^{-1}	0.017 75	0.000 04	5.2
214	[HCl]=40 mmol L^{-1} [NaCl]=160 mmol L^{-1}	0.020 44	0.000 05	5.7
225	[HCl]=40 mmol L^{-1} [NaCl]=160 mmol L^{-1}	0.005 93	0.000 01	3.9

*[1] 775 ng g^{-1} NO_3^- in seawater.

*[2] Peak top to valley.

*[3] Concentration of nitrate ions in seawater corresponding to 3 times the noise height.

The dependence of sensitivity of nitrite ions in seawater on wavelength is also depicted in Figure 6. The LOD's for nitrite ions in seawater at 210 nm, 215 nm, 220 nm, and 225 nm using the Waters model 486 UV detector were 11.6 ng g^{-1}, 3.5 ng g^{-1}, 1.0 ng g^{-1}, and 1.2 ng g^{-1} as NO_2^- under the eluent condition of 160 mmol L^{-1} NaCl.

Effect of counter cation in eluent

Figure 7 shows the dependence of the resolution on the kind of counter cation in an eluent at 225 nm. HCl, lithium chloride (LiCl), ammonium chloride (NH_4Cl), NaCl, potassium chloride (KCl) and magnesium chloride ($MgCl_2$) solutions were compared as unmixed eluents. Among these substances it is obvious that HCl is the most efficient eluent as far as the resolution is concerned. The separation was not as good at KCl concentrations greater than 40 mmol L^{-1}; therefore, the use of KCl as an eluent has no advantage. When a $MgCl_2$ solution was used as an eluent, the absorbance of nitrate at 225 nm was unfortunately approximately half in comparison with the other chlorides; presumably this was caused by some interaction between nitrate ion and magnesium ion. At the relatively higher concentration of chlorides the separation using LiCl was better than that using the others except for HCl. Generally speaking, the sensitivity for nitrate ions using LiCl was similar to the others except for HCl and $MgCl_2$ as illustrated in Figure 8. Thus, it is expected that the combination of LiCl and HCl may give much better separation than that of NaCl/HCl. Even under the condition of 560 mmol L^{-1} LiCl with 160 mmol L^{-1} HCl which gave high sensitivity, the resolution of 1.45 was attained in fact. Further investigation will be required regarding the use of LiCl/HCl eluent.

Since the functional group of the ion-exchange resin used in the IonPac AS10 column is alkanol quaternary ammonium, it is possible that there is some electrostatic interaction between the alcoholic oxygen and a cation; especially protonation for HCl. In general, the magnitude of the resolution in Figure 7 is in the order of the atomic masses (formula mass for ammonium ion) for the monovalent cations.

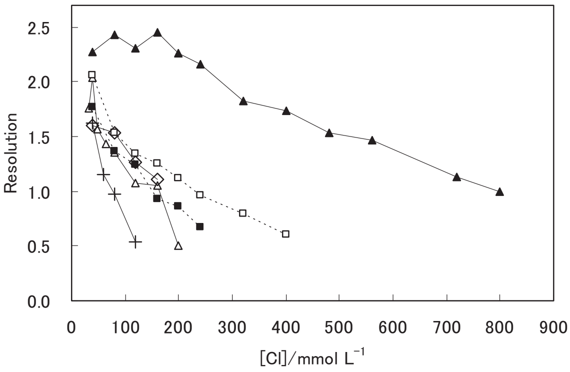

Figure 7. Dependence of resolution at 225 nm on chloride concentration and on the kind of counter cation in eluent. ▲: HCl; □: LiCl; △: NaCl; +: KCl; ■: NH_4Cl; ◇: $MgCl_2$.

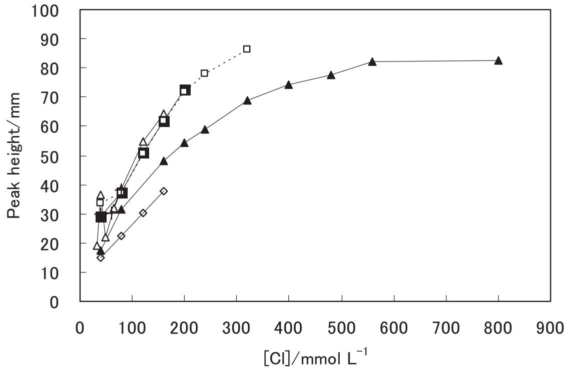

Figure 8. Dependence of sensitivity for nitrate ions at 225 nm on chloride concentration and on the kind of counter cation in eluent. The symbols in Figure 8 are the same as those in Figure 7.

Seawater analysis

According to the recommended procedure, CSK standard solutions (Wako Pure Chemical Industries, Osaka, Japan) containing 0.0 μmol L^{-1}, 10.0 μmol L^{-1} or 20.0 μmol L^{-1} nitrate ions in 3.05 % NaCl solution were analysed. The results obtained for the three solutions were less than LOD, 10.2 μmol L^{-1} and 20.8 μmol L^{-1}, respectively. These indicated a good agreement with the guaranteed values by Sagami Chemical Research Center.

A candidate seawater CRM for micronutrients was also analysed with a standard addition method. A so-called addition calibration method which is applicable to a series of samples having the same matrix was also used. The coefficients of correlation of calibration curves for standard solutions up to 37 μmol L^{-1} nitrate ions and for nitrate-spiked seawater samples up to 50 μmol L^{-1} nitrate ions were 1.00000 and 0.99995, respectively, under the eluent condition of 40 mmol L^{-1} HCl with 160 mmol L^{-1} NaCl. The same coefficients of correlation were 0.99987 and 0.99977, respectively, under the eluent condition of 240 mmol L^{-1} HCl.

The coefficient of correlation of calibration curve for nitrite-spiked seawater samples up to 2.6 μmol L^{-1} nitrite ions and that for nitrite-spiked seawater samples up to 4.3 μmol L^{-1} nitrite ions were 0.99983 under the eluent condition of 160 mmol L^{-1} NaCl and 0.99993 under the eluent condition of 80 mmol L^{-1} NaCl, respectively.

Three seawater reference solutions from IFREMER

(L'Institut Francais de Recherche pour l'Exploitation de la Mer, Brest, France) were determined under the eluent condition of 240 mmol L^{-1} HCl for nitrate ions. The obtained concentrations of nitrate ions were 14.29 μmol L^{-1}, 1.69 μmol L^{-1}, and 0.86 μmol L^{-1}. The expanded uncertainty (coverage factor k = 2) for those values was 0.03 μmol L^{-1} judging from the linearity of standard addition. No certified value for the first value had been reported. The last two values agreed well with the certified values (1.74 μmol L^{-1} and 0.90 μmol L^{-1}, respectively).

The present study was carried out at NRC during 1994-1995 and originally presented at the annual meeting of the Japan Society for Analytical Chemistry in 1996.[21)] Though it has not been published so far, many people have recently shown their interests in the ion-chromatographic method for nitrate ions and nitrite ions used for the production of the NRC CRM MOOS-1;[13)–15)] therefore, we decided to publish it.

Acknowledgement.

The authors are indebted to Dr. Alain Aminot, IFREMER, for his supply of seawater reference solutions of IFREMER.

References

1) J. P. Riley and G. Skirrow, eds.: *Chemical Oceanography* (Academic Press, London, 1975) 2nd ed., Vol. 3, pp. 415-419.
2) p. 416 in reference 1.
3) J. Mertens and D. L. Massart: Determination of Nitrate Ion in Marine Biotopes with High Nitrate Content by UV Spectrophotometry, *Bull. Soc. Chim. Belg.* **80** (1971) 151-158.
4) S. Rokushika, K. Kihara, P. F. Subosa and W. Leng: Ion Chromatography of Nitrite, Bromide and Nitrate Ions in Brine Samples Using a Chloride-form Anion-exchange Resin Column, *J. Chromatogr.* **514** (1990) 355-361.
5) J. Zou, S. Motomizu and H. Fukutomi: Reversed-phase Ion-interaction Chromatography of Inorganic Anions with Tetraalkylammonium Ions and Divalent Organic Anions Using Indirect Photometric Detection, *Analyst* **116** (1991) 1399-1405.
6) U. Haldna and I. Yakovleva: Nitrate Determination in Sea Water by On-column Matrix Elimination, *Eesti Tead. Akad. Toim., Keem.* **43** (1994) 83-85.
7) Marheni, P. R. Haddad and A. R. McTaggart: On-column Matrix Elimination of High Levels of Chloride and Sulfate in Non-suppressed Ion Chromatography, *J. Chromatogr.* **546** (1991) 221-228.
8) H. Itoh and Y. Shinbori: Determination of Anions in Sea Water by Ion Chromatography, *Bunseki Kagaku* **29** (1980) 239-242.
9) S. Y. Tyree, Jr., and M. A. O. Bynum: A Method for Nitrate and Phosphate in Saline Water, *Limnol. Oceanogr.* **29** (1984) 1337-1339.
10) P. Pecevich: Ion Chromatograph Provides Rapid Analysis of Air and Water, *Pollut. Eng.* **17** (1985) 53-55.
11) L. Chen and K. Liu: Ion Chromatographic Determination of Bromide, Sulfite, Fluoride, Chloride, Nitrate and Sulfate, *Sepu* **2** (1985) 207-208; *Chem. Abstr.* **104** (1986) 95115.
12) A. A. Ivanov, I. N. Voloshchik and O. A. Shpigun: Removal of Anion Interferences in Ion Chromatography, *Zh. Anal. Khim.* **44** (1989) 699-702; *J. Anal. Chem. USSR* **44** (1989) 569-572.
13) http://inms-ienm.nrc-cnrc.gc.ca/calserv/crm_files_e/MOOS-1_e.pdf
14) A. Hioki, J. W. H. Lam and J. W. McLaren: Online Determination of Dissolved Silica in Seawater by Ion Exclusion Chromatography in Combination with Inductively Coupled Plasma Mass Spectrometry, *Anal. Chem.* **69** (1997) 21-24.
15) L. Yang, R. E. Sturgeon and J. W. H. Lam: On-line Determination of Dissolved Phosphate in Seawater by Ion-exclusion Chromatography Inductively Coupled Plasma Mass Spectrometry, *J. Anal. At. Spectrom.* **16** (2001) 1302-1306.
16) E. Nakamura, J. Inoue and H. Namiki: Determination of Total Nitrogen in Seawater by Ion Chromatography Using a High-capacity Anion Exchange Column and a UV Detector, *Bunseki Kagaku* **45** (1996) 711-715.
17) W. Hu, K. Hasebe, Ming-Yu Ding and K. Tanaka: Direct Determination of Nitrite Traces in High Ionic-strength Samples by Electrostatic Ion Chromatography Using Diluted Acid Solutions as Eluent, *Fresenius J. Anal. Chem.* **371** (2001) 1109-1112.
18) G. Hashida, Y. Yokoyama and H. Sato: Determination of Total Nitrogen in Seawater by Spectrophotometric/PLS Regression Method in UV Region and Ion Chromatographic Method after Oxidative Digestion with Potassium Peroxodisulfate in Alkaline Medium, *Bunseki Kagaku* **53** (2004) 1455-1461.

19) K. Ito, Y. Takayama, N. Makabe, R. Mitsui and T. Hirokawa: Ion Chromatography for Determination of Nitrite and Nitrate in Seawater Using Monolithic ODS Columns, *J. Chromatogr. A* **1083** (2005) 63-67.

20) D. A. Skoog: *Principles of Instrumental Analysis* (Saunders College Publishing, Philadelphia, 1985) 3rd ed., p. 741.

21) Partly presented in A. Hioki and J. W. McLaren: Direct Determination Method of Nitrate and Nitrite Ions in Seawater by UV-detection Ion Chromatography using Hydrochloric Acid/Sodium Chloride Eluents, Proc. of 45th Annual Meeting of the Japan Society for Analytical Chemistry (Japan Society for Analytical Chemistry, Sendai, 1996) 1E11 (Abstract).