

Sequential determination of nitrate and nitrite in natural waters by flow injection analysis

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Nitrate is reduced to nitrite by hydrazine sulfate and catalyzed by Cu^{2+} under alkaline conditions at an elevated temperature of 75°C . The intensely colored azo derivative which is formed upon reaction with a mixed reagent of 3-nitroaniline and N-(1-naphthyl)ethylenediamine dihydrochloride (NED) is used as basis for the flow injection analysis (FIA) simultaneous determination of nitrate and nitrite using spectrophotometric detection.

A simplex-aided optimization was used to find the best ratio of mixed reagents to be used in the single detection of these anions. Good signal enhancement, ca. 240 % (nitrate) and (400 %) nitrite were achieved in a minimum number of 15-20 experimental trials. The development of the FIA manifold for the simultaneous detection were then based from these optimized individual manifolds.

The method showed a wide linear range of 0-5.0 mg NO_3^- - N/L, 0 - 0.9 mg NO_2^- - N/L, and detection limits of 30 and 10 $\mu\text{g/L}$ nitrate-nitrogen and nitrite-nitrogen respectively, with a sampling frequency of 80 determinations per hour. Interferences due to calcium and magnesium ions were removed on-line by a cation-exchange column.

Applications of the developed FIA method to lake, river and seawater were in good agreement with standard batch methods ($r = 0.9986$) which uses copper coated cadmium granules for the reduction step and sulfanilamide-NED as diazo-coupling reagents.

Key Words: FIA, homogeneous reduction, sequential determination, nitrate and nitrite, natural waters

NITRATE IS THE FINAL OXIDATION PRODUCT OF NITROGEN compounds in seawater. This is considered the only thermodynamically stable oxidation state of nitrogen in the presence of oxygen in seawater (1). If seawater becomes depleted in oxygen as a result of microbial remineralization processes, nitrate may be used as an alternative electron acceptor instead of oxygen (2). This process called denitrification leads to a reduction of a portion of nitrate to molecular nitrogen. The reduction of sulfate to sulfides commences after the available nitrate has been used up quantitatively.

Nitrite occurs in seawater as an intermediate compound in the microbial reduction of nitrate or in the oxidation of ammonia. It may also be excreted by phytoplankton especially during periods of luxury feeding. The natural level of nitrite in seawater is usually very low but in transition zones where oxic conditions change to anoxic ones, thin layers of high nitrite concentrations may occur together with low levels of dissolved oxygen (3).

Water bodies are considered eutrophic when concentrations of nitrate and nitrite exceed 0.5 mg N/L and 0.05 mg N/L respectively (4). The existing batch methods for determining nitrate and nitrite suffer from numerous interferences, poor reproducibility and linearity in addition to being lengthy. Sophisticated and expensive chromatographic methods which allow high sampling rates are not, however, easily available.

The use of flow injection analysis (FIA) for simultaneous determination of nitrates and nitrites have earlier been reported, differing mostly, in the designs of the flow injection manifolds (5-11). In most of these FIA methods, a modified Shinn reaction (5) was used, that is, nitrate was reduced to nitrite by passing through on-line copperized cadmium column and determining the nitrite produced by the standard sulfanilamide and N-(1-naphthylethylenediamine) (NED) to form a colored azo dye which is measured at 520 nm (6-9).

Another possible approach for the determination of nitrate is the use of a homogenous mode of reduction using alkaline hydrazine sulfate catalyzed by copper ions to reduce nitrate to nitrite, followed by a diazo-coupling reaction (12-14). The use of a liquid reductant such as hydrazine sulfate eliminates the problem of decreasing reduction efficiency in solid reductants after prolonged use. This method of reduction was adapted by Madsen (15) in an FIA method for the determination of nitrate in rain water. Applications to natural waters, however were limited due to interfering calcium and magnesium ions which inhibit the reduction of nitrate. Several methods to eliminate these interferences have been reported in the batch methods. Kempers et al. (12), for example precipitated these divalent ions with sodium hydroxide and further inactivated magnesium by chelation with pyrophosphate. The formation of precipitates in a flowing system, however is not highly desirable. In a study by Mullin et al. (14), an ion-exchange column was used to remove these interferences.

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The second step in the determination of nitrate is a diazo coupling reaction used to detect nitrite. This can be achieved by either one of four composite reagents: sulphanilamide and NED, 4-nitroaniline and NED, sulphanilic acid and NED and 3-nitroaniline and NED. A closer examination of a non-flow spectrophotometric work suggests that 3-nitroaniline is an efficient diazotizing reagent. This was reported by Rathore et al. (16) and Chaube (17) who claimed the following advantages: rapid color development, excellent reproducibility, freedom from pH and temperature effects and freedom from interferences by a large group of foreign ions.

This paper reports the development of a simple, rapid and sensitive FI manifold for the sequential determination of nitrate and nitrite in natural waters. The method is based on the use of hydrazine sulfate to reduce nitrate to nitrite, catalyzed by Cu^{2+} under alkaline conditions and elevated temperatures. The method shown here has the following advantages: reduction is fast and reproducible, column regeneration and repacking problems are nonexistent and the reductant is highly stable. Interferences due to calcium and magnesium are eliminated by an on-line cation exchange column installed just before the sampling system.

The nitrite produced is determined by the diazotization coupling process using 3-nitroaniline and NED. This method proved to be very convenient for the direct determination of nitrite by simply foregoing the reduction step and thus determining these two anions simultaneously in the special FIA configuration which was developed.

Experimental

Instrumentation

A flow injection (FI) analyzer was fabricated for use in this study. This is composed of two units of a five-channel Ismatec peristaltic pumps, a six-port two-way rotary injection valve, a multiwavelength detector all housed in a 20 inches long x 21 inches wide x 10 inches high polyvinyl (PVC) box. The set-up includes accessories such as a reaction manifold, a fabricated thermostatted heater, tank of compressed air and a Hitachi recorder QD 15.

Reagents and Solutions

All reagents used were of analytical grade unless otherwise specified, prepared in distilled deionized water, mixed thoroughly and stored in appropriate reagent bottles. The standards used were dried in the oven for two hours at 100°C , cooled and stored in a dessicator. Appropriate standards for sodium nitrite and sodium nitrate (Merck) were prepared as required.

The following reagents were prepared:

Reductant	0.2 % hydrazine sulfate (Osaka, Japan) with 0.005 % copper sulfate (Merck) and 0.01% zinc sulfate (Merck)
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Mixed Color Reagent for Nitrite	0.09% 3-nitroaniline (Aldrich), 0.02% NED
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Mixed Color Reagent for Nitrate	0.35% 3-nitroaniline, 0.06% NED
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Sodium Hydroxide (Merck)	5.6%
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Procedure

Flow injection (FI) manifolds for the single component detection of nitrite and nitrate were designed. The FI manifold for nitrite, as shown in Fig. 1, was a single line system where the sample merges with a pre-mixed reagent of 3-nitroaniline and NED to form the azo dye and transported to the detector. The FI manifold for nitrate, as shown in Fig. 2, used a multiple step reaction system where the sample merges with an on-line pre-mixed sodium hydroxide-hydrazine sulfate containing copper sulfate and zinc phosphate. The sample and the reductant are mixed and heated in a reduction coil immersed in a thermostatted water bath at 75°C . The nitrite produced is merged with the pre-mixed 3-nitroaniline and NED and transported to the detector.

Both FI manifolds were optimized using a modified simplex optimization scheme. This is a computer-aided optimization procedure which operates according to the flow diagram shown in Fig. 3. Initially, four parameters are logged in. Maximum and minimum are the values which refers to the desired concentration/sample size range to be searched. Precision refers to the desired lowest value which the simplex is allowed to vary within the concentration range/sample size range defined previously.

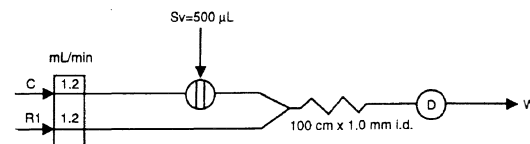


Figure 1. FI manifold for the single detection of nitrite (C-carrier, R1-mixed nitroaniline-NED)

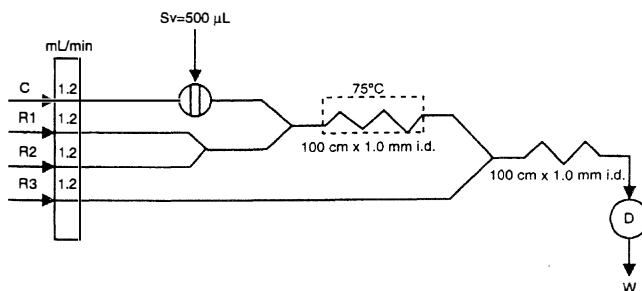


Figure 2. FI manifold for the single detection of nitrate (C-carrier, R1-hydrazine sulfate-copper sulfate-zinc sulfate, R2-NaOH, R3-mixed nitroaniline-NED)

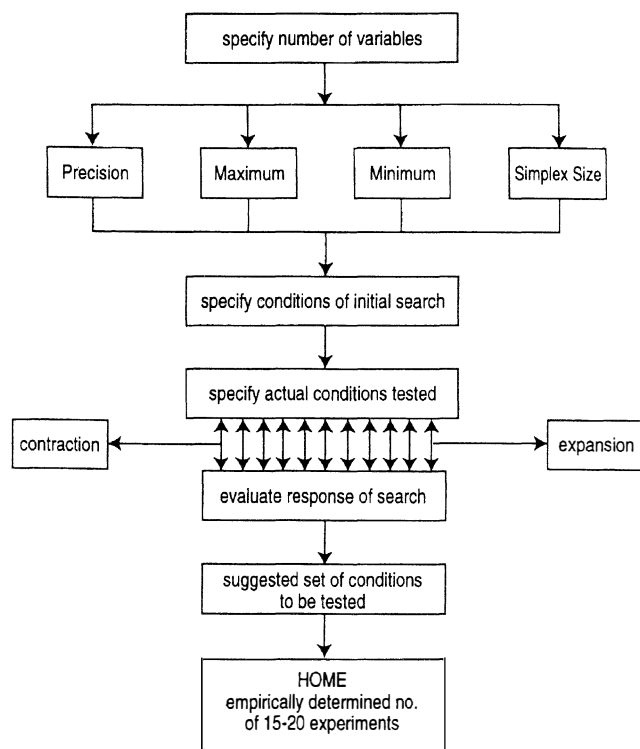


Figure 3. Flow diagram of the modified simplex program

Likewise, simplex size is defined as the desired largest value which the simplex is allowed to vary within the concentration range/sample size range defined. Initial conditions are logged in and the experiment is executed. The response (FI peak height) is inputted and evaluated by the program, after which it gives the next set of conditions to be tested. The response is again evaluated and the simplex gives another set of conditions to be tested. This process goes on until an empirically determined number of experiments have been done, and an optimum is achieved, usually after 15-20 experiments.

The FI manifold for the sequential determination of nitrite and nitrate are designed based on the FI manifold for the single detection of nitrite and nitrate. The sample is split after injection into two channels, one channel designed to detect nitrite alone and the other channel designed to detect the original nitrite plus nitrite produced from the reduction of nitrate by alkaline hydrazine sulfate at 75°C. Optimization of the physical variables of this manifold was done by the one-factor-at-a-time approach.

Methods for eliminating interferences from calcium and magnesium were evaluated for use in natural water bodies. Samples from Manila Bay, Marikina river and Taal Lake were collected and analyzed both by the proposed FI and batch methods.

Results and Discussion

Design and Optimization of FI Manifolds for the Single Detection of Nitrite and Nitrate

The FI manifold for the single determination of nitrite is shown in Fig. 1. This consists of two lines; one for the carrier, the other for the mixed reagent, and a mixing coil (30 cm x 0.5 mm i.d.). Simplex aided optimization (Tables 1 and 2) was used to establish the best ratio of the mixed reagent with signal enhancement of ca. 400%. The single manifold for nitrate is shown in Fig. 2. This consists of four lines, three of which merge (C, R1, R2) and heated at 75°C to effect the reduction of nitrate to nitrite. The nitrite produced merges with the mixed reagent, R3, to form the intensely coloured azo dye. Optimum concentrations for hydrazine sulfate (0.2%), sodium hydroxide (1.4 N) and copper sulfate (0.01%) were deter-

Table 1. Variables in the simplex-aided optimization of the FI manifold for nitrite determination

Variable Name	Maximum	Minimum	Precision	Simplex Size
Sample volume (μL)	500	100	50	150
NED (%)	0.14	0.05	0.01	0.03
Nitroaniline (%)	0.39	0.30	0.01	0.03

Table 2. Simplex-aided optimization of the FI manifold for nitrite determination

Experiment No.	Sample Volume (μL)	NED (%)	Nitroaniline (%)	Peak Height (mm)
1	150	0.07	0.3	1.0
2	300	0.07	0.3	2.0
3	200	0.10	0.3	1.0
4	200	0.08	0.3	1.5
5	350	0.09	0.3	1.5
6	350	0.06	0.3	2.0
7	450	0.06	0.3	2.5
8	500	0.06	0.3	3.0
9	450	0.05	0.3	5.0
10	500	0.05	0.3	5.0
11	500	0.05	0.3	5.0
12	500	0.05	0.3	5.0
13	500	0.05	0.3	5.0
14	500	0.05	0.3	5.0
15	500	0.05	0.3	5.0
16	500	0.06	0.3	3.0
17	450	0.06	0.3	2.9

*Conditions giving maximum signal are seen in bold type.

mined by a modified simplex optimization. The results of this optimization are shown in Tables 3 and 4, and showed that a signal enhancement of ca. 259 % was achieved.

Table 3. Variables in the simplex-aided optimization of the FI manifold for nitrate determination

Variable Name	Maximum	Minimum	Precision	Simplex Size
Sample Volume (μL)	500	100	50	150
Hydrazine Sulfate (%)	1.0	0.10	0.10	0.30
Copper Sulfate (%)	0.10	0.01	0.01	0.03
Sodium Hydroxide (%)	1.7	0.80	0.10	0.30

Table 4. Simplex-aided optimization of the FI manifold for nitrate determination

Experiment No.	Sample Volume (μL)	Hydrazine Sulfate (%)	NaOH (N)	CuSO_4 (%)	Peak Height (mm)
1	100	0.1	1.1	0.02	3.9
2	100	0.4	1.1	0.02	2.0
3	100	0.2	1.4	0.02	4.5
4	100	0.2	1.2	0.05	0
5	300	0.2	1.2	0.03	8.0
6	250	0.2	1.2	0.01	10.5
7	500	0.2	1.3	0.02	7.0
8	500	0.1	1.4	0.02	4.0
9	400	0.3	1.3	0.02	11.0
10	500	0.2	1.1	0.01	10.0
11	300	0.2	1.1	0.02	2.0
12	400	0.3	1.3	0.02	10.5
13	500	0.1	1.3	0.01	5.5
14	350	0.1	1.2	0.02	9.5
15	500	0.2	1.3	0.01	11.0
16	350	0.1	1.5	0.01	13.0
17	300	0.1	1.6	0.02	10.9
18	500	0.2	1.4	0.01	14.0
19	500	0.2	1.4	0.01	14.0
20	500	0.1	1.6	0.01	12.0

*Conditions giving maximum signal are seen in bold type.

Design and Optimization of the FI Manifold for the Sequential Determination of Nitrite and Nitrate

Two manifold designs combining the single manifolds for nitrate and nitrite (Figs. 4 and 5) were evaluated. In the first manifold (Fig. 4), two lines were split, (a- for the carrier line, b- for the color reagent), two were merging points (d- for the sodium hydroxide and reductant, e- detector merging point). The reaction coil length for nitrite is 30 cm x 0.5 mm i.d., while the nitrate branch utilized a 100 cm x 1.0 mm i.d. reduction coil (heated at 75°C) and another 100 cm x 1.0 mm i.d. as cooling and delay coil. A back pressure coil, 14 cm x 0.3 mm i.d. was used to regulate the flow, and a larger sample size of 600 μL was used to provide enough sample in both branches after splitting. This FI manifold design seemed feasible, however only the nitrite signals were observed, and nitrate which was expected as a second peak was not observed. This was due to a combination of inadequate tubing connectors, inappropriate flow rates, and insufficient delay time between the emergence of the nitrite and nitrate sample zones.

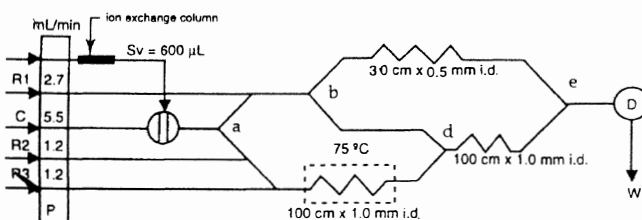


Figure 4. First design of the FI manifold for the sequential determination of nitrate and nitrite (R1-mixed reagent nitroaniline-NED, C-carrier, R2-hydrazine sulfate, R3-sodium hydroxide)

Flow injection systems allow signals to be dephased by splitting the starting flow into two channels of different length and inner diameter. In order to achieve the best possible resolution, the flow was split into two channels of 60 cm x 0.5 mm i.d. and 250 cm x 0.8 mm i.d. and an additional reaction coil (70 cm x 1.0 mm i.d.) was included.

The use of a back pressure line proved to be disadvantageous because it hindered the continuous flow in the system. Changing the back pressure line by using a wider i.d. tubing (14 cm x 1.0 mm) proved effective and heating in the reduction coil was improved. The improved FI manifold is shown in Fig. 5.

Effect of Sample Volume. The sensitivity of an FIA system can be manipulated in a number of ways. The quickest way of increasing sensitivity is by increasing the injection volume, as shown in Fig. 6. The peak height increases up to a sample volume of 800 μL . At this sample size, sample throughput is correspondingly reduced due to an increase in the analytical cycle (filling and emptying the sample loop). A compromise sample size must therefore be decided from the responses given at 600 μL .

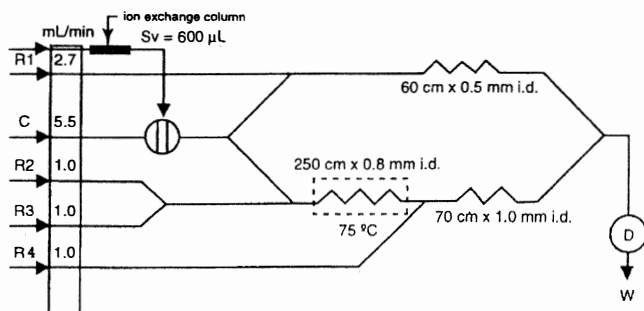


Figure 5. Improved design of the FI manifold for the sequential determination of nitrate and nitrite (R1-mixed reagent nitroaniline-NED (nitrite line), C-carrier, R2-hydrazine sulfate-copper sulfate-zinc sulfate, R3-sodium hydroxide, R4-mixed nitroaniline-NED(nitrate line))

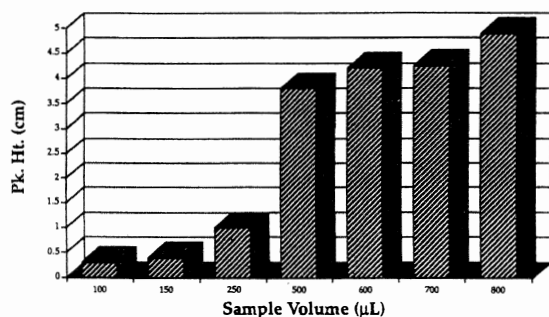


Figure 6. Effect of sample volume on the nitrate FI response

and 700 µL. At a sample volume of 600 µL, the peak height was not markedly different from that at 700 µL, but the throughput was better. Consequently, 600 µL injections were used in all measurements.

Effect of length on reduction coil and color forming coil in the nitrate line. The peak height was also highly dependent on the reduction coil length. The highest responses were obtained using a 250 cm and 320 cm coils (Figure 7). To maximize the sampling throughput and decrease the bulk of the coiled tubing in the water bath, the shorter length of reduction coil was used in subsequent measurements. Likewise, the shorter length of color forming coil (70 cm) was also used, following the observation that increasing lengths of this coil reduces the FI response considerably (Fig. 8).

Effect of length and inner diameter of mixing coil in the nitrite line. A very pronounced effect in the FI peak height was observed when the length and inner diameter of the mixing coil in the nitrite line were varied. At 0.5 mm i.d. the response was much greater compared to 0.8 mm i.d. and 1.0 mm i.d. (Fig. 9). This can be attributed to the decrease in sample zone dispersion, since contact between sample and carrier is reduced. Increasing the length of this same coil from (15 x 0.5 mm i.d.) up to (60 cm x 0.5 mm i.d.) gave a higher signal (Fig. 10). At increasing

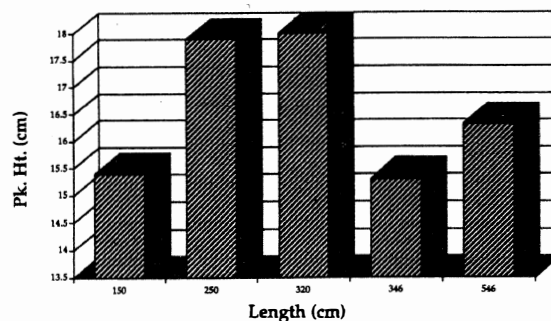


Figure 7. Effect of reduction coil length on the nitrate FI response

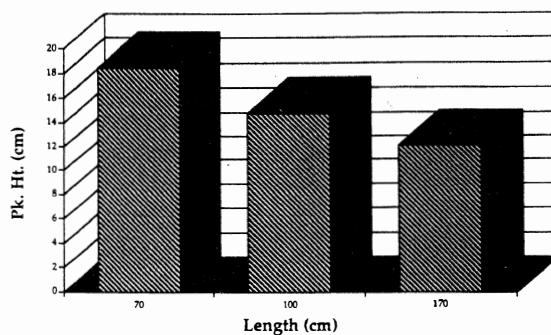


Figure 8. Effect of length of the color forming coil in the nitrate FI response

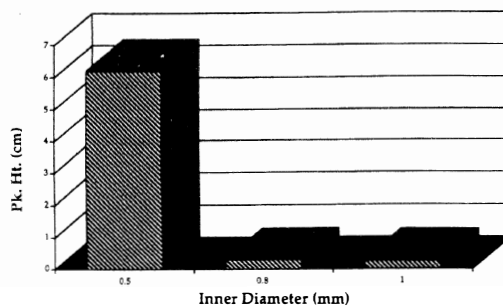


Figure 9. Effect of inner diameter on the mixing coil in the nitrite line

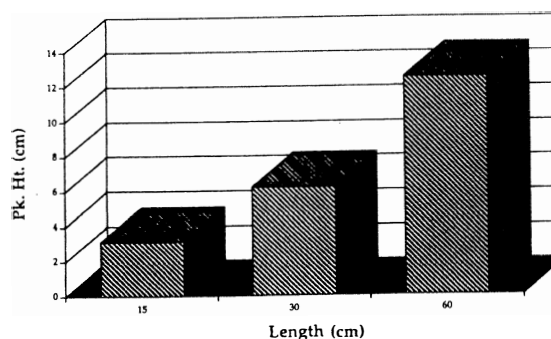


Figure 10. Effect of length of mixing coil in the nitrite line

lengths of tubing, the diazo coupling reaction is allowed to approach near equilibrium, thus the enhancement in the peak height observed.

Effect of flow rate on the carrier. A major factor which affects the shape of the FIA peak is the carrier flow rate. Fig. 11 shows the effect in the peak shape using carrier flow rates of 0.8 and 2.0 mL/min. Fig. 11a shows broad peaks while Fig. 11b consists of sharp and well defined peaks. Dispersion of the sample zone increases at higher flow rates (>2-3 mL/min) allowing a well-defined sample zone to react optimally with the merging reagents.

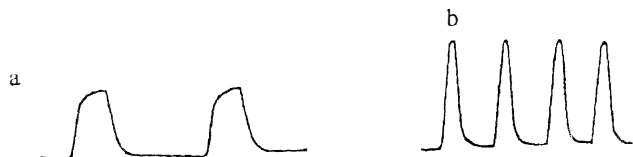


Figure 11. Effect of carrier flow rate on the peak shape of the nitrate FI response for 600 μ L sample volume a) 0.8 mL/min b) 2.0 mL/min

Effect of reagent flow rate on FI signal changes. The effect of varying flow rates in both the hydrazine and sodium hydroxide lines are shown in Figs. 12 and 13. In both cases, decreasing the flow rate to either 0.8 mL/min or 2.0 mL/min decreases the FI response. At a flow rate of 1.0 mL/min, the chemical signal was found to be maximum. For both nitrite and nitrate, signals were significant when a mixed reagent (nitroaniline-NED) flow rate of 2.0 mL/min and 0.8 mL/min were used (Figs. 14 and 15). However, considering that both reagents are scarce and expensive, this was not utilized in subsequent measurements. Instead a flow rate of 1.0 mL/min was chosen.

Effect of pH. Reduction of nitrate to nitrite by hydrazine sulfate occurs at a pH above 11. This alkaline condition was achieved by merging sodium hydroxide with hydrazine sulfate before heating. Higher concentrations of NaOH were tested up to 2.5 N. Apparently at higher concentrations of base, the wastelines are clogged with precipitates causing build-up of pressure within the FI manifold causing connections to break loose and reagent spillage.

Effect of Salinity. Water samples containing the two analytes were dissolved in different salinities from 0‰ to 35‰ to determine the effect of salinity in natural waters. This is shown in Figs. 16 and 17, where an increase in the FI signal is observed at higher salinities. This may be due to refractive index effects caused by differing ionic strength in carrier and sample. These were remedied by matching the ionic strength of the sample and the carrier.

Test for Stability of the Reduction Reagent. The reductant mixture was shown to be stable for a month even at room temperature. This was tested by monitoring the re-

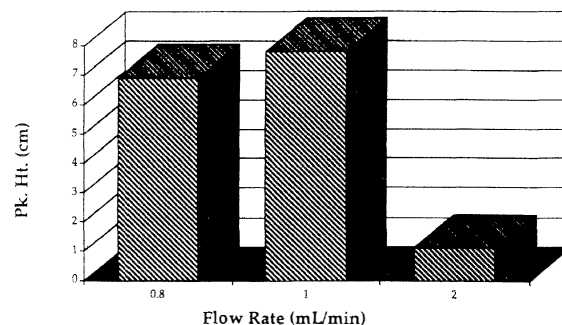


Figure 12. Effect of flow rate in the hydrazine reagent line

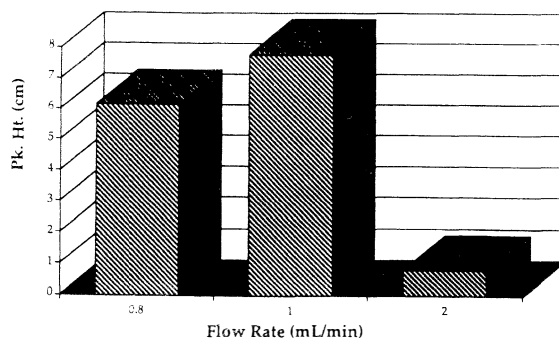


Figure 13. Effect of flow rate in the sodium hydroxide reagent line

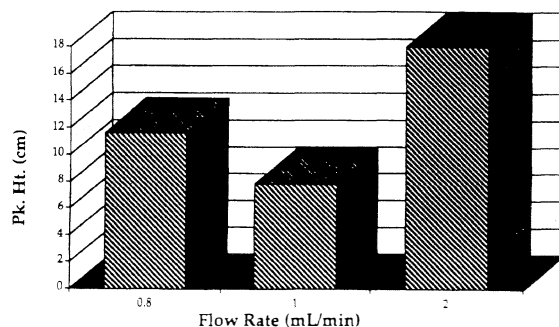


Figure 14. Effect of flow rate in the mixed reagent line for nitrate

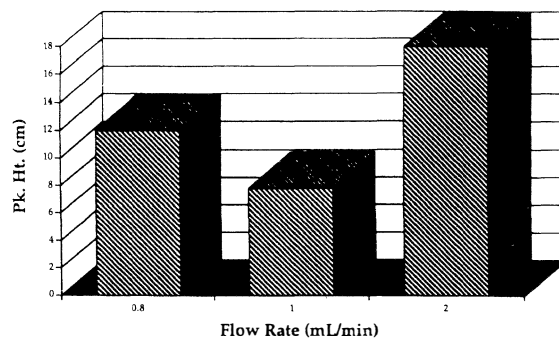


Figure 15. Effect of flow rate in the mixed reagent line for nitrite

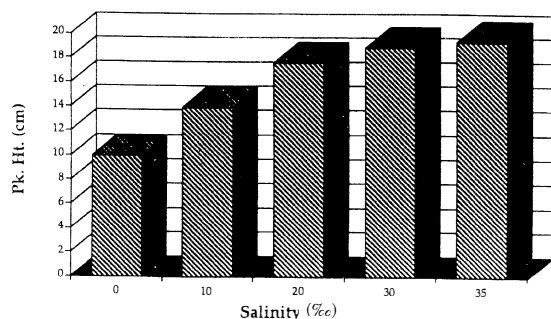


Figure 16. Effect of increasing salinities on the nitrite FI response

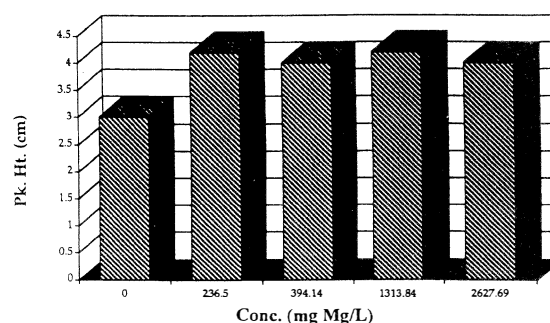


Figure 18. Effect of increasing concentrations of magnesium on the nitrate FI peak height

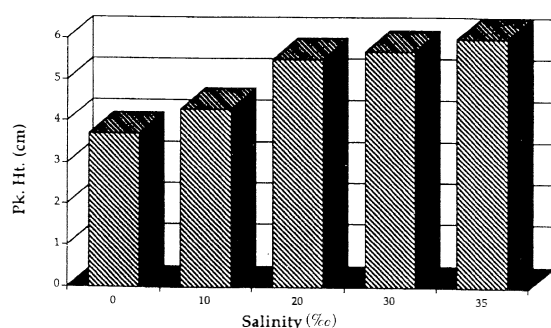


Figure 17. Effect of increasing salinities on the nitrate FI response

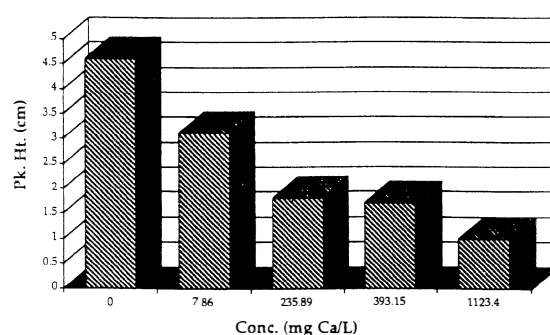


Figure 19. Effect of increasing concentrations of calcium on the nitrate FI peak height

producibility of the FI signal for a given concentration of nitrate (1.0 mg N/L) over a period of one month.

Interference Study

The presence of calcium and magnesium ions in the water sample inhibits the reduction of nitrate to nitrite by hydrazine sulfate. A series of tests, at concentration levels normally found in seawater, ca. 1326 mg Mg^{2+} /L and 400 mg Ca^{2+} /L (18) were performed and the results are shown in Figs. 18 and 19. Increasing levels of magnesium enhance the FI signal, ca. 40% at the normal levels, at ca 1100 mg Mg^{2+} /L. The effect of increasing levels of calcium was to suppress the responses, by as much as 62% compared with those obtained for Ca^{2+} standards.

Three methods were tested to suppress these interferences. The first method involved masking of calcium and magnesium with hydroxyquinoline. This showed promising results, but the hydroxyquinoline is highly insoluble in water, the effective working concentration was difficult to reproduce everytime a new stock was prepared.

A cation exchange column for removal of Ca^{2+} and Mg^{2+} was tested by installing it in the sample line. Initially, a column size of 191.6 mm x 2 mm i.d. was used, which proved to be effective at concentrations of up to 24 mg/L of the interferents in lake and river water. For marine waters, the high ionic strengths of the water samples

caused immediate saturation of the ion exchange sites. A bigger ion-exchange column was prepared (320 mm x 3 mm) and this was found effective for saline waters. In both cases, regeneration of the column was found necessary, and was achieved by passing 1N HCl through the sample line followed by rinsing with distilled water.

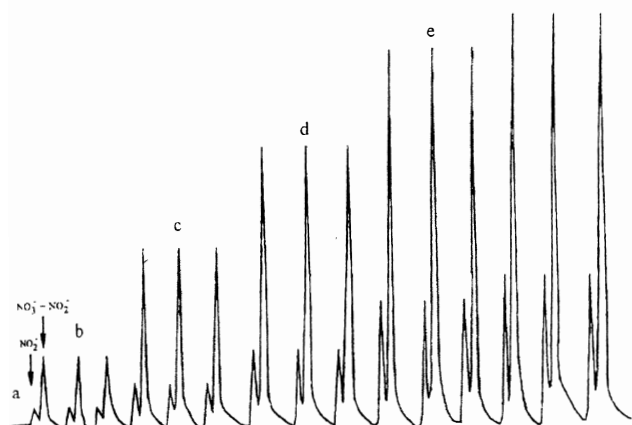
An alternative method used to eliminate these interferences was by sample pretreatment using ammonium chloride-ethylenediaminetetraacetic acid (EDTA). This also acts as a masking agent for calcium and magnesium.

A statistical analysis of the proposed method was performed and results of this are shown in Table 5. A typical recorder output for the mixed standards using a range of 0-5.0 mg N/L is shown in Fig. 20. A single injection gives two peaks, the first one, which is detected at 5 s, corresponds to nitrite. The second peak, detected after 30 s, corresponds to total nitrite and nitrate. The difference between the two peaks therefore is a measure of the levels of nitrate in natural waters.

The limit of detection is 0.03 mg NO_3^- -N/L and 0.01 mg NO_2^- -N/L with a linear range of 0- 5.0 mg NO_3^- -N/L and 0 - 0.9 mg NO_3^- - N/L. A sampling throughput of 80 determinations per hour is achievable and with the use of an ion exchange column, to remove calcium and magnesium interferences, the method is applicable to lake, river and seawater samples.

Table 5. Results of the statistical analysis of the proposed FI manifold for the sequential determination of nitrate and nitrite

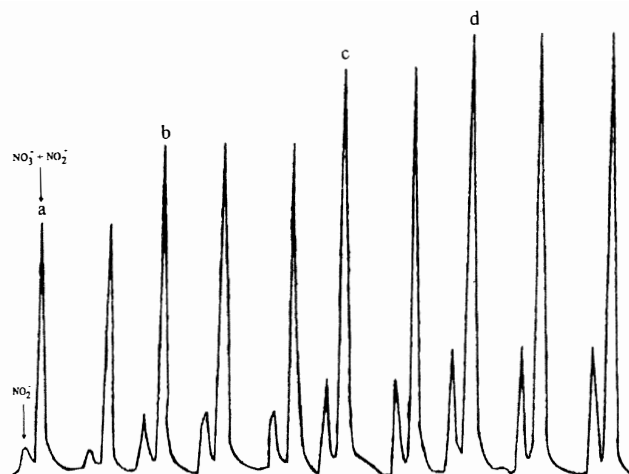
Parameter	Nitrite	Nitrate
rsd for n = 7 (%)	0.76	0.88
limit of detection (mg N/L)	0.01	0.03
linear range (mg N/L)	0 - 0.90	0 - 5.00
calibration curve	$y = 1.4575 + 58.5781x$	$y = -1.6 + 27.34x$
correlation coefficient	0.9933	0.9942

**Figure 20.** Typical recorder output for mixed nitrate and nitrite standards (0-0.5 mg NO_3^- - N/L and 0-0.9 mg NO_2^- - N/L)

- a) 0 mg NO_2^- - N/L + 0 mg NO_3^- - N/L
 b) 0.10 mg NO_2^- - N/L + 1.00 mg NO_3^- - N/L
 c) 0.30 mg NO_2^- - N/L + 2.00 mg NO_3^- - N/L
 d) 0.50 mg NO_2^- - N/L + 3.00 mg NO_3^- - N/L
 e) 0.70 mg NO_2^- - N/L + 4.00 mg NO_3^- - N/L
 f) 0.90 mg NO_2^- - N/L + 5.00 mg NO_3^- - N/L

Application to Natural Waters

Samples of river, lake and seawater were collected in February 1994. The samples were filtered on site through 0.45 μm Millipore filters, placed in prewashed nalgene or polyethylene bottles and stored in ice until analyzed, usually within 24 hours. Analyses of water samples were done for three stations of Manila Bay, two stations for Marikina River and four stations for Taal Lake (Table 6). Figure 21 shows the typical recorder out-

**Figure 21.** Typical recorder output for the simultaneous determination of nitrite and nitrate in Marikina water sample (MR-2) where

- Cs1 = 1000 mg NO_2^- - N/L;
 Cs2 = 1000 mg NO_3^- - N/L
 a) MR - 2 only
 b) MR - 2 + 15 μL mixed Cs1 and Cs2
 c) MR - 2 + 30 μL mixed Cs1 and Cs2
 d) MR - 2 + 45 μL mixed Cs1 and Cs2

put for the determination of nitrite and nitrate in Marikina water sample.

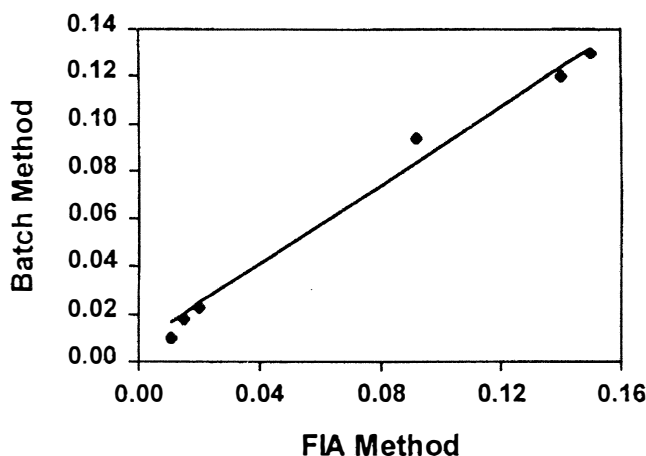
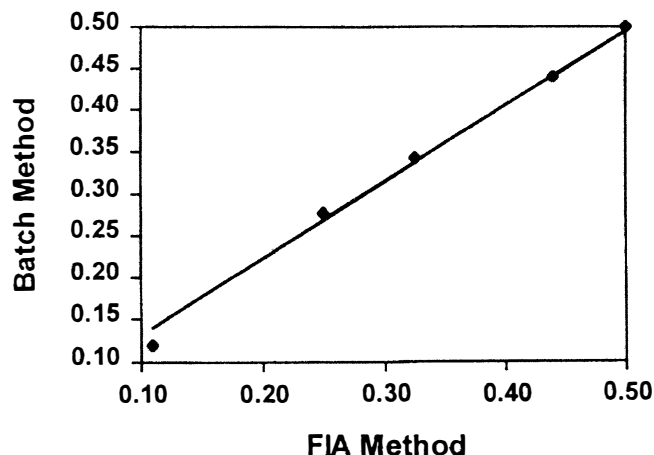
Results obtained using this FI method were compared with the batch method using copper coated granules for reduction and sulfanilamide and NED as diazo coupling reagents. A good correlation ($r = 0.9981$ for nitrate and 0.9947 for nitrite) was obtained as shown in Fig. 22 for nitrate and Fig. 23 for nitrite.

Conclusion

Hydrazine sulfate is comparable with copper coated cadmium granules as reductant in the determination of nitrate as nitrite followed by detection as an azo derivative. This homogeneously catalyzed reduction of nitrate is rapid and reproducible, and is more convenient for laboratory and monitoring purposes in terms of ease in preparation and the high stability of the reductant even at room temperatures. In the developed FIA configuration, the direct determination of nitrite is possible without going through the reduction process and thus provides an even more powerful method

Table 6. Nitrate and nitrite analyses in natural waters by FIA and Batch methods

Sampling Site	Sample Code	$\text{NO}_3^- + \text{NO}_2^-$ (mg/L)	$\text{NO}_3^- + \text{NO}_2^-$ (mg/L)	NO_2^- (mg/L)	NO_2^- (mg/L)
		FIA Method	Batch Method	FIA Method	Batch Method
Manila Bay (Stn1)	MB-1D	0.12 ± 0.03	0.13 ± 0.02	0.011 ± 0.002	0.010 ± 0.002
Manila Bay (Stn 2)	MB-5D	0.34 ± 0.05	0.36 ± 0.04	0.015 ± 0.002	0.018 ± 0.002
Manila Bay (Stn 3)	MB-10D	0.27 ± 0.05	0.30 ± 0.04	0.020 ± 0.001	0.023 ± 0.001
Manila Bay (Stn 4)	MB-0D	0.10 ± 0.02	0.14 ± 0.03	0.092 ± 0.003	0.094 ± 0.002
Marikina River (Stn 1)	MR-1 S	0.65 ± 0.10	0.63 ± 0.09	0.150 ± 0.002	0.130 ± 0.001
Marikina River (Stn 2)	MR-2 S	0.58 ± 0.11	0.56 ± 0.10	0.14 ± 0.003	0.12 ± 0.002
Taal Lake (Stn 1)	TL-1 SS	<0.03	<0.03	<0.01	<0.01
Taal Lake (Stn 2)	TL-2 SR	<0.03	<0.03	<0.01	<0.01
Taal Lake (Stn 3)	TL-3 SM	<0.03	<0.03	<0.01	<0.01
Taal Lake (Stn 4)	TL-4 SL	<0.03	<0.03	<0.01	<0.01


Figure 22. Correlation graph for the analysis of nitrate by Batch and FIA Methods

Figure 23. Correlation graph for the analysis of nitrite by Batch and FIA Methods

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