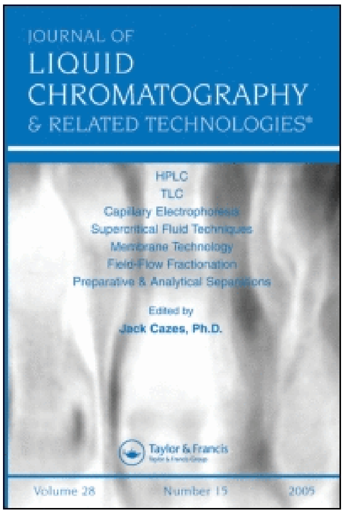

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CHROMATE AS A MOBILE PHASE FOR THE DETERMINATION OF ANIONS USING CETYLTRIMETHYLAMMONIUM-COATED COLUMNS AND INDIRECT PHOTOMETRIC DETECTION

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ABSTRACT

A suitable indirect photometry chromatographic system for the separation and detection of common inorganic and organic anions was made by dynamically coating an octadecyl-bonded silica column with cetyltrimethylammonium ions and using potassium chromate as the eluent. The effect of chromate ion concentration on the capacity factor indicates an ion-exchange mechanism. The detection limit of the anions F^-, Cl^-, NO_2^-, Br^-, and NO_3^-, corresponding to a signal-to-noise ratio of 3, were 0.29, 0.55, 0.72, 0.83 and 0.97 ppm, respectively. A linear response was observed between peak heights and injection volume of a solution with 1.19 ppm F^-, 2.22 ppm Cl^-, 2.88 ppm NO_2^-, 4.99 ppm Br^- and 3.88 ppm NO_3^-. This can further lower the detection limit of the anions by about 9 fold when a volume of 175 μL is injected. This system has been applied to the trace analysis of NO_3^- in tap water. A concentration of 1.97 ± 0.10 ppm NO_3^- was detected. This system was also found applicable for the separation of organic acids (formate, acetate, propionate and butyrate) and may also be used for the separation of a mixture of organic and inorganic anions.

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INTRODUCTION

Ion-exchange chromatography (IEC) has been widely used for the separation of ionic species. A major breakthrough in IEC was the introduction of conductivity detectors coupled with a dual-column into the system (1). This technique, called ion chromatography, is used for the determination of ions which do not absorb in the uv-vis region. Most recently, Small and Miller (2) reported the concept of indirect photometric chromatography (IPC) for the determination of these ions. This technique uses a single-column and a uv-vis detector. A light-absorbing reagent is added to the mobile phase. This reagent maintains a background signal and competes with the analyte ions for the active sites on the column. When the analyte ions elute, they are indirectly detected as negative peaks because the concentration of the light-absorbing reagent in the analyte bands decreases. The mechanism of IPC has been reported in the literature (2). The selection of light-absorbing reagent is important in determining the sensitivity and selectivity of the separation. Although several reagents such as benzoate, phthalate, salicylate, naphthalenesulfonate and other carboxylate salts have been successfully employed (2-8), the use of potassium chromate has not been described. This reagent possesses fine ion-exchange capability, good chromatographic selectivity, and large molar absorptivity (see discussion for details). In addition, chromate ion has tendency to absorb in wavelength regions above 254 nm which reduces interference from ultraviolet absorbing substances and therefore its use as an eluent is preferred over phthalate and other benzene derivatives, which are the most widely used in indirect photometric chromatography.

In indirect photometric chromatography, the capacity of the ion-exchange column is important in determining the sensitivity, speed and efficiency of separation (6). Several commercially available ion-exchange resins each with a different capacity have been reported (2-8). However, by coating a reverse-phase column with ion pairing reagents (which would not desorb in aqueous eluents) under various solvent composition and/or reagent concentrations, resin with a variety of capacities can be prepared (9-15). In addition, this approach possesses a great flexibility with regard to choice of columns, eluents, and ion-pair reagents for optimum separation.

This paper describes the results of a study of the behavior of potassium chromate as an eluent for an indirect photometric method for the separation and detection of common inorganic anions using ODS-bonded silica column coated with cetyltrimethylammonium ions. It also confirms the applicability of this system for the separation of a mixture of organic anions.

MATERIALS AND METHODS

Instrumentation

The HPLC system consisted of a Perkin Elmer Series 3 Liquid Chromatograph equipped with a variable wavelength LC-65T UV detector/oven, an LC1-100 integrator and a 20 μL (or 175 μL) Rheodyne sample injector (Model 7125, Berekely, CA). The column (5 cm x 4.6 mm i.d.) was packed with 3 micron Spherisorb ODS-bonded silica (Regis, Morton Grove, IL). A prefilter SSI. (Alltech Associates, Inc., Deerfield, IL) with a 0.2 micron filter was located between the injector and the column. The void volume of the system was calculated by using the peak of injected water. The uv-vis spectra were determined on a double beam Hitachi U-2000 spectrophotometer fitted with 1-cm cells. All pH measurements were performed with an ALTEX Model 3560 Digital pH meter and Corning combination glass electrode. The flow rate was fixed at 1.0 mL min^{-1} and a wavelength of 328 nm was selected throughout the work unless otherwise mentioned.

Chemicals and Reagents

Cetyltrimethylammonium bromide (CTAB) and potassium chromate were obtained from Aldrich Chemical Company, Inc. All other chemicals used were of analytical grade. The chemicals were used without further purification. The stock solution of 0.15 M CTAB was prepared in deionized water and filtered through a 0.45-μm membrane (Rainin Instrument Co., Inc., Woburn, MA). Stock solution of 0.10 M CrO_4^{2-} was also prepared in deionized water. Eluents were prepared by dilution of the CrO_4^{2-} stock solution and then filtered through a 0.45-μm membrane filter before using. The pH was adjusted to 7.00 with aqueous hydrochloric acid. Stock solutions of the test solutes were prepared by carefully weighing the sodium salt of the compound in a volumetric flask and then diluting it with deionized water. The working concentrations for the mixture of inorganic anions were as follows: fluoride (9.5 ppm), chloride (17.7 ppm), bromide (40.0 ppm), nitrite (23.0 ppm), nitrate (31.0 ppm). The working concentrations for the mixture of organic anions were as follows: formate (22.5 ppm), acetate (29.5 ppm), propionate (73.0 ppm), butyrate (87.0 ppm).

Preparation of the Column

The ODS-bonded silica column was coated with CTA^+ ions by passing an aqueous solution of 0.10 M CTAB at a flow rate of 0.50 mL min^{-1} through the column for 2 h, followed by washing with deionized water for 30 min. After the above dynamic modification, the column is equilibrated with potassium chromate until a steady baseline with high absorbance background is reached.

Following multiple separations the column was regenerated by washing with deionized water for 10 min at 1 mL min^{-1}, followed by pure methanol until all surfactant was removed (about 1 h), and then with deionized water until all methanol was removed (about 1 h).

RESULTS AND DISCUSSION

The absorption spectrum from 200 to 450 nm of 0.5 mM potassium chromate solution showed absorption maxima at 370.5 and 272.5 nm with molar absorptivities of 4.5×10^3 and 3.3×10^3 L cm^{-1} mol^{-1}, respectively. A detection wavelength of 328 nm ($\epsilon = 1.0 \times 10^3$ L cm^{-1} mol^{-1}) was selected throughout the work unless otherwise mentioned. This wavelength is not that of maximum sensitivity but was selected to keep the absorbance of chromate within the back-off range of the detector. However, the wavelength of maximum sensitivity can be used when working with very low concentrations of potassium chromate or when comparing the absorbance of the mobile phase in the presence and absence of the analyte (16).

The effect of chromate ion concentration on the retention time of F^-, Cl^-, NO_2^-, Br^-, and NO_3^- ions was examined in the range of 0.05 - 0.50 mM. The plots of log k′ against log $[CrO_4^{2-}]$ are shown in Figure 1. Increasing the chromate concentration resulted in a linear decrease in capacity factor for all five anions. Considering the competition of the solute (X^-) and chromate ions for the CTA^+ sites on the stationary phase, i.e. the ion-exchange mechanism, then:

$$(CTA^+)_2CrO_4^{2-}{}_s + 2X^-_m \rightleftharpoons 2CTA^+X^-_s + CrO_4^{2-}{}_m \qquad (1)$$

The designations s and m refer to stationary and mobile phases, respectively. The equilibrium expression in terms of concentrations of the species in the preceding equation is:

$$K = \frac{[CTA^+X^-]_s^2\ [CrO_4^{2-}]_m}{[(CTA^+)_2CrO_4^{2-}]_s\ [X^-]_m^2} \qquad (2)$$

Where K is the selectivity coefficient. An expression for capacity factor of the solute, k′, can be obtained by rearranging this equation to

$$\frac{[CTA^+X^-]_s}{[X^-]_m} = \left(\frac{K\ [(CTA^+)_2\ CrO_4^{2-}]_s}{[CrO_4^{2-}]_m}\right)^{1/2} = k' \qquad (3)$$

The logarithm of this becomes

$$\log k' = constant - \frac{1}{2} \log\ [CrO_4^{2-}] \qquad (4)$$

where the constant = 1/2 {log (K)($[CTA^+{}_2CrO_4^{2-}]_s$)}.

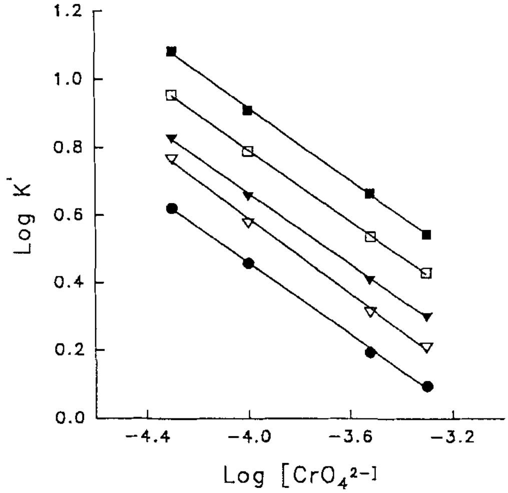

Figure 1: Effect of chromate ion concentration on the capacity factor. Conditions: eluent pH 7.0; fow rate 1.0 mL min^{-1}; detection, uv absorbance at 328 nm; injection volume, 20 μL. Line identities and solute concentrations: (●) = F^- (9.5 ppm), (▿) = Cl^- (17.7 ppm), (▾) = NO_2^- (23.0 ppm), (□) = Br^- (40.0 ppm) and (■) = NO_3^- (31.0 ppm).

Thus if the retention behavior of the anions is similar to that of ion-exchange chromatography, the slope of the plot of log k′ against log $[CrO_4^{2-}]$ should give a slope equal to 0.50. The results gave an average slope of the lines equal to 0.58 ± 0.05, which supports the ion-exchange mechanism. This is confirmed by the fact that the elution order of a mixture of these ions increases with their size (Figure 2).

The variation of capacity factor as a function of temperature was studied from 25°C to 40°C. The plot of In k′ against 1/T (van't Hoff plot) yields straight lines (Figure 3), therefore the proposed ion-exchange mechanism remains unchanged over the temperature range studied. In addition, a more significant increase was observed in the capacity factor of Br^-, NO_2^- and NO_3^- ions than that of F^- and Cl^-. This differences in slopes is related to the differences in the thermodynamic distribution behavior of these anions. In general, the overall changes in the capacity factor of the anions studied is low over the temperature

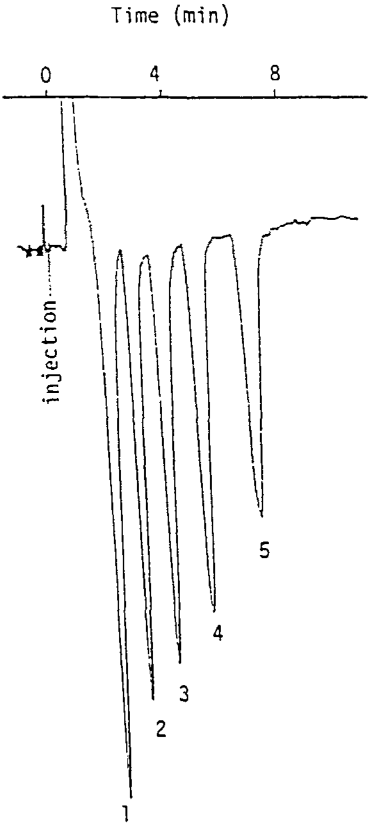

Figure 2: Separation of a mixture of inorganic anions on ODS-column coated with CTA^+. Conditions: eluent 0.5 mM potassium chromate. Other conditions and solute concentrations as for Figure 1. Peak identities: (1) = F^-, (2) = Cl^-, (3) = NO_2^-, (4) = Br^-, (5) = NO_3^-.

range studied. Therefore, experiments can be performed without careful temperature control.

In an attempt to improve the column efficiency as reported by other workers (17), the effect of 2-propanol added to the aqueous mobile phase was studied. This resulted in loss of some of the CTA^+ coated on the column as indicated by a strong positive absorbance due to the increase in concentration of

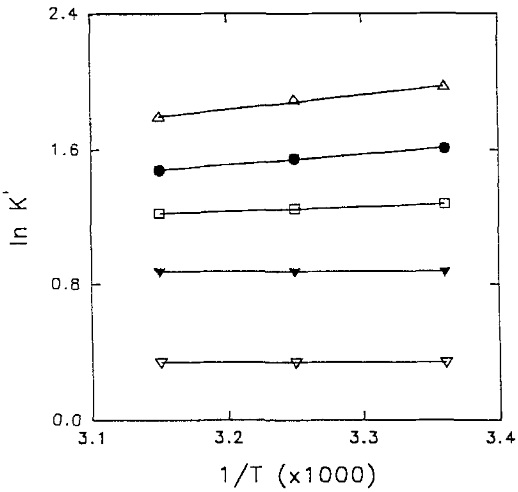

Figure 3: Effect of column temperature on the capacity factors of the investigated anions. Conditions and solute concentrations as for Figure 2. Line identities: (▿) = F^-, (▾) = Cl^-, (□) = NO_2^-, (●) = Br^- and (▵) = NO_3^-.

the chromate ion in the mobile phase. When equilibrium was reestablished, i.e. a stable baseline was obtained, a mixture of NO_2^-, Br^- and NO_3^- ions was injected. A decrease in retention time was observed (Table 1). This was expected due to the decreased ion-exchange capacity of the column. To evaluate the role of adding small amounts of the modifier to the mobile phase on the efficiency of the column, the number of theoretical plates (N) from the peaks obtained in the presence of 3% of 2-propanol were compared to those in the absence of the modifier. The values of N were calculated from the commonly used equation:

$$N = 5.54 \; (t_R / W_{0.5})^2$$

The results are shown in Table 1. Little improvement in column efficiency was obtained, therefore 2-propanol was not added to the mobile phase.

To determine the stability of the column, repeated injections of a mixture of F^-(9.5 ppm), Cl^-(17.7 ppm), NO_2^-(23.0 ppm), Br^-(40.0 ppm), and NO_3^-(31.0

TABLE 1

Variation of Capacity Factor and Column Efficiency in the Presence of 2-Propanol

	Zero% 2-Propanol		3% 2-Propanol	
Anion	K'	N	K'	N
NO_2^-	6.10	985	5.27	1024
Br^-	8.32	982	7.02	998
NO_3^-	11.41	1086	8.84	1141

0.5 mM potassium chromate at pH 7.00 and 1.0 mL min^{-1}; wavelength 328 nm.

ppm) were made on a freshly coated ODS-column with CTA^+ and a continuous flow of 0.5 mM chromate (flow rate 1 mL min^{-1}) over a period of 30 hours. No significant changes in retention times over the duration of the experiment were observed. When the column was used continuously over two weeks, a drift in the base line and a broadening in the peaks were observed. This problem was overcome by inverting the column.

The sensitivity of the method was investigated by preparing various calibration plots for 20 μL injections of the anions using 0.5 mM CrO_4^{2-} as the mobile phase. Plots of the peak height against concentration injected were linear over the range up to 14.4 ppm F^-, 17.7 ppm Cl^-, 23.0 ppm NO_2^-, 40.0 ppm Br^- and 31.0 ppm NO_3^- studied (Figure 4). Reproducible results can be obtained in low ppm ranges (Table 2). The detection limits of the anions F^-, Cl^-, NO_2^-, Br^- and NO_3^-, corresponding to a signal-to-noise ratio of 3, were 0.29, 0.55, 0.72, 0.83 and 0.97 ppm, respectively. These detection limits are comparable with

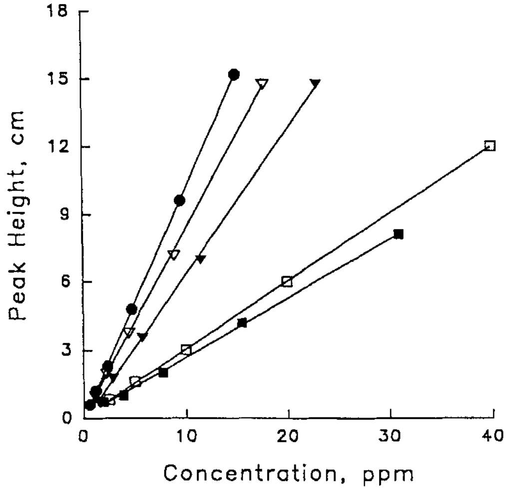

Figure 4: Calibration curves for anion determination with CTA$^+$-coated column. Conditions as for Figure 2. Line identities: (■) = NO_3^-, (□) = Br^-, (▼) = NO_2^-, (▽) = Cl^- and (●) = F^-.

TABLE 2

Repeatability of Anion Determinations

Anion	Conc.[a], ppm	RSD[a*] %	RSD[b*] %
F^-	9.5	3	
	1.2	5	0.6
Cl^-	17.7	0.4	
	2.2	6	1.1
NO_2^-	23.0	1.1	
	2.9	7	0.9
Br^-	40.0	3	
	5.0	5	1.1
NO_3^-	31.0	2	
	3.9	3	0.8

* Relative standard deviation of 6 samples
a 20 μL injected volume
b 175 μL injected volume

Chromatographic conditions as in Table 1

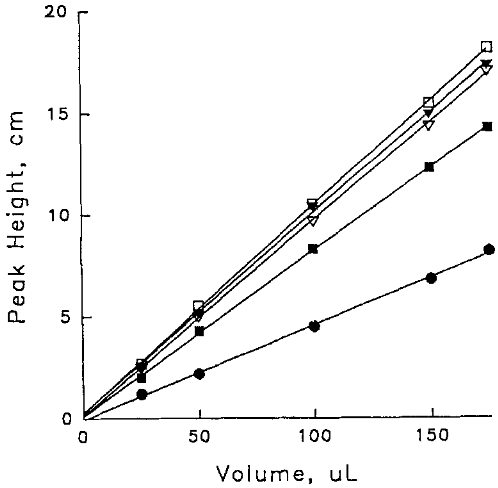

Figure 5: Relationship of detector response to injection volume. Conditions as for Figure 2. Line identities and solute concentrations: (●) = F^- (1.19 ppm), (■) = NO_3^- (3.88 ppm), (▿) = Cl^- (2.22 ppm), (▾) = NO_2^- (2.88 ppm) and (□) = Br^- (4.99 ppm).

those reported using conductivity (18-22) or indirect refractive index methods (23).

A linear response between peak heights and injection volume for the anions (1.19 ppm F^-, 2.22 ppm Cl^-, 2.88 ppm NO_2^-, 4.99 ppm Br^- and 3.88 ppm NO_3^-) was observed up to an injection volume of 175 μL studied (Figure 5). In addition, reproducibility significantly increased when 175 μL was injected in comparison to 20 μL for the same analyte concentration (Table 2). Therefore, the injection of 175 μL lowers the detection limit by about nine fold compared to a volume of 20 μL. Provided that volumes larger than 175 μL do not destabilize the baseline and the large solvent peak does not interfere with early eluting peaks, then this approach of improving the detection limit can be extended to even larger injection volumes.

This system was used to analyze NO_3^- in tap water. A mobile phase of 0.5 mM $CrO4^{2-}$ at pH 7.02 was used and a sample of 175 μL was injected. A

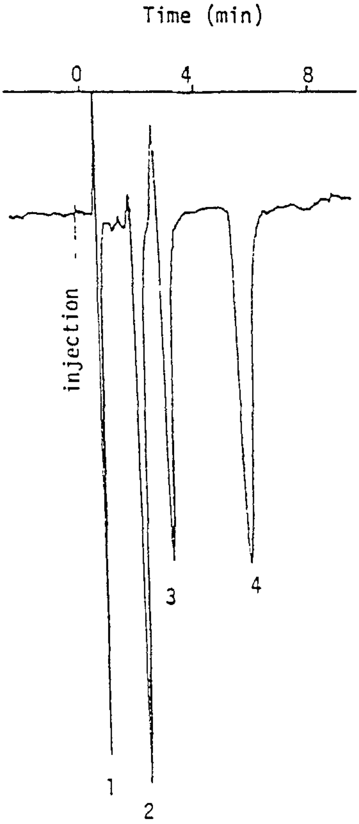

Figure 6: Separation of a mixture of organic anions on ODS-column coated with CTA+. Conditions as for Figure 2. Peak identities and solute concentrations: (1) = formate (22.5 ppm), (2) = acetate (29.5 ppm), (3) = propionate (73.0 ppm), (4) = butyrate (87.0 ppm).

concentration of 1.97 ± 0.10 ppm NO_3^- was detected based on the calibration-curve method using peak height.

The ODS-bonded column dynamically coated with CTA^+ and loaded with CrO_4^{2-} ions was tested for its ability to separate a mixture of organic anions (formate, acetate, propionate and butyrate). Figure 6 illustrates that a complete separation of the four species was obtained in less than 6 minutes. In addition,

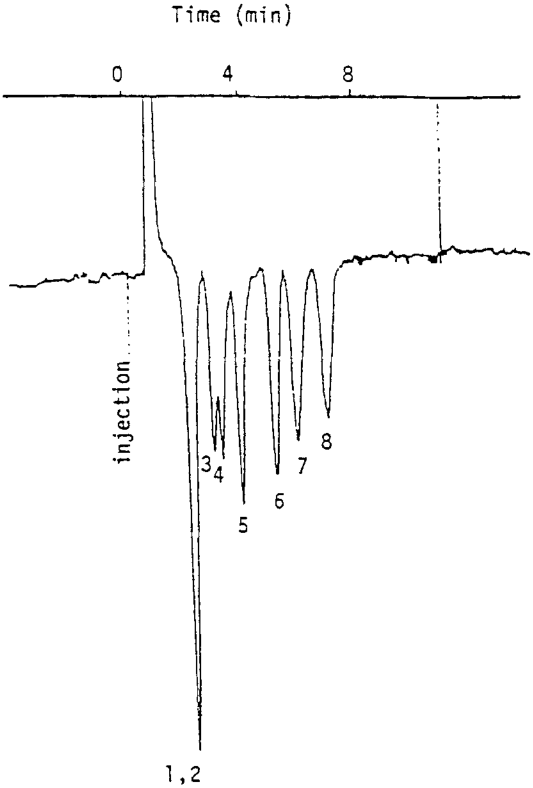

Figure 7: Separation of a mixture of inorganic and organic anions on ODS-column coated with CTA$^+$. Conditions as for Figure 2. Peak identities and concentrations: (1) = fluoride (9.5 ppm), (2) = acetate (29.5 ppm), (3) = chloride (17.7 ppm), (4) = propionate (73.0 ppm), (5) = nitrite (23.0 ppm), (6) = bromide (40.0 ppm), (7) = butyrate (87.0 ppm), (8) = nitrate (31.0 ppm).

the elution order as expected based on the size of these ions. When a mixture of the inorganic anions, fluoride, chloride, bromide, nitrite, and nitrate, and the organic anions acetate, propionate, and butyrate was injected, a complete overlap of fluoride and acetate, a partial overlap of chloride and propionate, and a complete separation of the other anions was obtained (Figure 7). Further studies to improve this separation and to include other organic anions are underway.

CONCLUSIONS

Coating an octadecyl-bonded silica column with cetyltrimethylammonium ions and using an aqueous solution of potassium chromate as the mobile phase provides a method that is comparable to that of other HPLC methods reported for the separation and determination of inorganic anions in low ppm concentration levels. In addition, this system is applicable for the separation of organic anions and may be used for the separation of mixtures of organic and inorganic anions This approach to detect trace anions may be attractive because ODS-columns are readily available and used by most high-performance liquid chromatographers.

REFERENCES

1. H. Small, T.S. Stevens and W.C. Bauman, Anal. Chem., 47, 1801(1975).
2. H. Small and T.E. Miller, Anal. Chem., 54, 462(1982).
3. S. A. Malik and N. D. Danielson, Anal. Chem., 63, 699(1991).
4. S. A. Malik and N. D. Danielson, J. Chromatogr., 452, 101(1991).
5. S. Motomizu, M. Oshima and T. Hironaka, Analyst, 116, 695(1991).
6. P. J. Naish, Analyst, 109, 809(1984).
7. H. Sato, Anal. Chim. Acta, 206, 281(1988).
8. Y. Yokoyama and H. Sato, J. Chromatogr. Sci., 26, 561(1988).
9. Y. Michigami, Y. Yamamoto and K. Ueda, Analyst, 114, 1201(1989).
10. P. Janos, K. Stulik and V. Pacakova, Talanta, 39, 29(1992).
11. T. Takeuchi and E. S. Yeung, J. Chromatogr., 370, 83(1986).
12. M. Deacon and M. R. Smyth, Analyst, 116, 897(1991).

13. T. Takeuchi, E. Suzuki and D. Ishii, J. Chromatogr., 447, 221(1988).

14. K. Ito, Y. Ariyoshi, F. Tanabiki and H. Sunahara, Anal. Chem., 63, 273(1991).

15. E. Papp, J. Chromatogr., 402, 211(1987).

16. A. L. Heckenberg and P. R. Haddad, J. Chromatogr., 299, 301(1984).

17. K. McCormic and B. L. Karger, Anal. Chem., 52, 2249(1980).

18. A. E. Buchholz, C. I. Verplough and J. L. Smith, J Chromatogr. Sci., 20, 499(1982).

19. T. Okada and T. Kuwamoto, Anal. Chem., 55, 1001(1983).

20. J. A. Glatz and J. E. Girard, J. Chromatogr. Sci., 20, 266(1982).

21. H. Mackie, S. J. Speciale, L. J. Throop and T. Yang, J. Chroamtogr., 242, 177(1982).

22. R. A. Cochrane and D. E. Hillman, J. Chromatogr., 241, 392(1982).

23. P. R. Haddad and A. L. Heckenberg, J. Chromatogr., 252, 177(1982).

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