On-line preconcentration system for flame atomic absorption spectrometry using unloaded polyurethane foam: determination of zinc in waters and biological materials

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An on-line procedure for the preconcentration and determination of zinc in waters and biological materials was developed. Zinc is preconcentrated from acidic medium (pH 3.0) as its thiocyanate complex onto a polyurethane foam mini-column placed in the loop of a four-way valve. The elution step is performed with a stream of 30% acetone in 2% HNO_3 and the zinc displaced is introduced directly into the nebuliser of a flame atomic absorption spectrometer. The system was operated under two different preconcentration times since the concentration of zinc in the two types of samples was different. For biological samples, 1 min was used and a detection limit of 3.0 $\mu g\ L^{-1}$ was achieved with a throughput of 40 samples per hour. At a concentration of 20 $\mu g\ L^{-1}$, an RSD of 2.5% was obtained. For the determination of zinc in natural water, a preconcentration time of 3 min was employed and a detection limit of 0.85 $\mu g\ L^{-1}$ was obtained. In this case, the system was slower with a throughput of 17 samples per hour. The RSD at 10 $\mu g\ L^{-1}$ was 6.0%. The continuous flow system was applied to the analysis of several biological reference materials and natural water samples.

1. Introduction

Analytical chemists frequently have to apply different previous treatments to samples that cannot be analysed in their natural state due to matrix interferences or low sensitivity of the methodology employed. Separation and preconcentration procedures have been performed in order to solve these problems. Thus, on-line preconcentration systems have proved to be the most interesting way to improve the performance of flame atomic absorption spectrometry (FAAS) in trace ion determinations, allowing high preconcentration factors with limited amounts of sample and also increasing the precision and speed of analysis. Moreover, the possibilities of contamination and losses are considerably reduced.

The process involved in continuous liquid–solid separation preconcentration always take place in two steps that involve retention and elution. The active solid phase (polyurethane foam in this study) is a permanent part of the continuous system involving mini-columns. Several flow configurations can be found in the literature employing classical ion-exchangers, chelating resins, functionalized silica and cellulose, activated alumina and charcoal.[1] With regard to zinc as an analyte, Burguera et al. in 1981[2] were the first to study its retention and elution from an ion-exchange column with chemiluminescence detection. Employing AAS, Olsen et al.[3] were the pioneers in coupling mini-column flow systems with such detection. Also, more recent papers dealing with this goal can be found in the literature, especially those of Purohit and Devi[4] and Greenway and Townshend,[5] although neither dealt with real sample analysis. Advances in this synergistic coupling (FAAS and mini-column continuous flow systems) are well documented in books by Fang[6] and Sanz-Medel.[7]

Solid-phase extraction (SPE) can be considered the most important technique for performing preconcentration in flow systems. The application of polyurethane foam (PUF) as a solid phase for analytical purposes was first reported by Bowen[8] in his pioneering research. Braun and co-workers[9–11] have reviewed this field.

Several papers have appeared in the literature employing loaded and unloaded PUF for metal sorption from aqueous medium.[12–20] Unloaded PUF has been extensively studied as an extractant for several metal cations from thiocyanate medium, with zinc being cited in some of these papers. Chow et al.[21] described cobalt retention from a medium containing 1.0 mol L^{-1} NH_4SCN under different conditions of ionic strength. They found that Ni(II) and Pb(II) were not extracted by the foam and Fe(III), Zn(II) and Cu(II) were simultaneously extracted with Co(II). Braun and Abbas[22] studied the sorption of several metal cations, including Zn(II), from different types of foam and concluded that polyether as well as polyester foams are efficient in removing metals from thiocyanate solutions. Maloney et al.[23] also reported that Fe(III), Co(II), Cd(II) and Zn(II) are extracted by polyether PUF as their thiocyanate complexes. Applications of PUF as a solid extractant in flow injection analysis (FIA) systems for the preconcentration and separation of metals have been described by our research group. Recently, de Jesus et al.[24] have performed the separation of zinc and cadmium in matrices containing high concentrations of cadmium by using PUF.

In the first work describing the use of PUF in a flow system, Zn(II) was preconcentrated on a PUF mini-column from thiocyanate solution as the Zn–SCN^- complex.[25] After elution, effected with water, the metal was measured spectrophotometrically. Several biological certified reference materials were analysed with good precision and accuracy. Ferreira et al.[26] employed an FIA-PUF system to improve the selectivity in the spectrophotometric determination of nickel in alloys and silicates. Cassella et al.[27] also investigated the use of an FIA-

PUF system to enhance the selectivity of the spectrophotometry. They applied this system to the spectrophotometric determination of aluminum with MTB (Methyl Thymol Blue) in silicate samples containing large amounts of iron.

The aim of this work was to develop an analytical system capable of determining zinc at $\mu g \ L^{-1}$ levels by FAAS using zinc preconcentration, exploring the high selectivity of the FAAS technique and improving the global performance of the system. Some advantages of this new application are: very low cost of the solid phase (the PUF employed was of the same type as the domestic PUF used for cleaning purposes); very low overpressure due to the inherent physical properties of the foam (*e.g.* high resilience level) without swelling providing high sample flow rates without clogging; and very effective extraction of metals from thiocyanate solutions, removing the matrix, with very high breakthrough capacity (4.5 mg of Zn per gram of PUF).

2 Experimental

2.1 Apparatus

A Perkin-Elmer 3100 atomic absorption spectrometer (Perkin-Elmer, Norwalk, CT, USA) equipped with a zinc hollow cathode lamp was used. The instrument was connected to an RB-201 recorder (ECB-Equipamentos Científicos do Brasil, São Paulo, Brazil) and was operated according to standard conditions recommended by the manufacturer. The FIA system was constructed using an Alitea XV peristaltic pump (Seattle, WA, USA), furnished with Tygon tubes, to propel all solutions. Both preconcentration and elution steps were switched by using a Rheodyne 5041 (Cotati, CA, USA) four-way injection valve. All connections were made using fittings, unions and tees made from plastic and PEEK materials. The manifold was constructed with PTFE tubes of 0.5 mm id.

2.2 Reagents and solutions

All solutions were prepared with water obtained from a Milli-Q water purification system (Millipore, Bedford, MA, USA) and by using analytical-reagent grade reagents.

Zinc(II) standard solutions were prepared daily by appropriate dilution of a stock solution containing 1000 mg L^{-1} Zn. This solution was obtained by dissolving 2.4696 g of dried $ZnSO_4$ in 1000 mL of 0.15 mol L^{-1} HNO_3 and was standardized by complexometric titration with EDTA.[28]

A reagent solution of 1.0 mol L^{-1} potassium thiocyanate was prepared daily by dissolving 24.30 g of KSCN in water. The pH of this solution was adjusted to 3.0 ± 0.2 with 0.01 mol L^{-1} HNO_3.

Eluent solution was prepared by carefully mixing 300.0 mL of acetone with 500.0 mL of water and 20.0 mL of concentrated HNO_3, after which the volume was made up to 1000 mL.

Polyurethane foam (PUF), open cell, polyether type, was obtained as a commercial product (Vulcan of Brazil—VCON 202, 42% resilience and 10–12 cells cm^{-1}). In order to use PUF as sorbent, the foam was comminuted in a blender with Milli-Q water and washed several times with 6 mol L^{-1} HCl to ensure elimination of metallic species. A mini-column was prepared by packing 50 mg of PUF in a small plastic tube (3.0 cm × 3 mm id).

2.3 Flow injection system

A schematic diagram of the developed flow system is depicted in Fig. 1. In this system a sample solution pumped at 6.7 mL min^{-1} merges with a 0.43 mL min^{-1} stream of 1.0 mol L^{-1} KSCN. The mixture generated percolates through the PUF mini-column where the Zn–thiocyanate complex is retained. The remaining solution goes directly to waste. After a suitable preconcentration time, the valve is switched and the
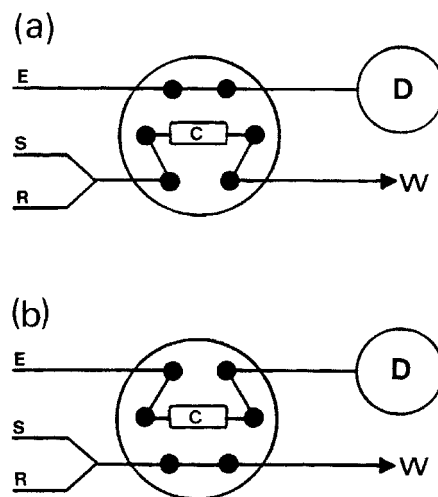
Fig. 1 Flow system manifold for zinc(II) determination after its preconcentration on a PUF mini-column. (a) Preconcentration step and (b) elution step. S = Sample, 6.7 mL min^{-1}; R = reagent, 1.0 mol L^{-1} KSCN, 0.43 mL min^{-1}; E = eluent solution, 30% acetone + 2% HNO_3 (v/v), 2.0 mL min^{-1}; C = PUF mini-column, 50 mg; D = detector, flame atomic absorption spectrometer; W = waste.

eluent solution (30% acetone + 2% HNO_3) flows at 2.0 mL min^{-1} through the mini-column, displacing Zn(II) ions to the spectrometer where absorbance signals are monitored. Peak heights were used for all calculations. After the elution step, which takes about 30 s, the mini-column is ready for a new preconcentration cycle.

2.4 Sample preparation

The flow system was used to determine zinc in several certified reference materials and water samples (sea-water and well water). The biological certified reference materials analysed were Copepoda (MA-A-1/TM), Fish Tissue (MA-B-3/TM), Fish Flesh (MA-A-2/TM) and Tuna Homogenate (IAEA-350) from the International Atomic Energy Agency (IAEA), Monaco, and Rice Flour—Unpolished Sample No. 10-a from the National Institute of Environmental Studies (NIES), Japan.

Firstly, all samples were dried overnight at $110 \pm 5 \ °C$, after which they were dissolved by the usual treatment with HNO_3 and $HClO_4$. For this purpose, different amounts of the samples (between 0.1 and 0.3 g depending on the concentration of zinc) were placed in a PTFE beaker and mixed with 10 mL of concentrated HNO_3. After standing overnight in contact with the acid as recommended by Icbinoki and Yamazaki,[29] the mixtures were heated until total dissolution of the samples had occurred and a clear pale yellow solution was obtained. Then, 4 mL of 70% $HClO_4$ were carefully added in two 2 mL portions. After evaporating to fumes of $HClO_4$, the remaining residue was cooled and further dissolved with 50 mL of water. The pH was then adjusted to 3.0 ± 0.2 and the volume was made up to 100 mL. The solution samples were stored in poly(propylene) flasks and were analysed according to the developed procedure. At least one blank solution was run for each sample to control reagent contamination.

Natural sea-water was taken from the Guanabara bay region near to a shipyard and to a sewage discharge, both located at Niterói, RJ. The well water was sampled at Itaipu balneary region near to Niterói city. For these samples, the only pre-treatment was acidification to pH 1.8, which was performed immediately after collection, in order to prevent adsorption of the Zn(II) ions on the poly(propylene) flask walls. In the laboratory, the samples were filtered and 100 mL of the filtrate were taken and the pH was adjusted to 3.0 ± 0.2. The volume was then made up to 200 mL. Also, at least one blank solution

was run for each sample in order to evaluate zinc contamination by the reagents used.

3. Results and discussion

3.1 Flow system optimization

The first procedure adopted in the development of this new methodology was the optimisation of the chemical and flow variables of the system in order to improve its performance.

In batch procedures, the pH of the sample solution was not an important parameter since the extraction was quantitative over a wide range of pH.[24] Despite this fact, the pH of the sample solution was studied since it controls the concentration of free thiocyanate ion in the medium, thereby influencing the formation of the Zn–SCN$^-$ complex. The pH was studied between 1.0 and 6.0 in the presence of 0.5% sodium citrate as masking agent for possible interferents. The highest signals were obtained for pH values up to 3.9; an abrupt decrease was observed when the pH was higher than 4.0 probably due to the high concentration of free citrate with consequent complexing of the analyte. Hence, a pH of 3.0±0.2 was chosen for all further experiments. Different results from batch procedures were observed probably due to kinetic differences between continuous and batch modes.

Another chemical variable tested was the thiocyanate concentration, which also controls the formation of the Zn–SCN$^-$ complex. In this case, it was decided to fix the pH and to change only the concentration of the reagent, since the variation of pH could modify the concentration of free thiocyanate ions. The pH of this solution was kept at 3.0 and the concentration of SCN$^-$ was tested between 0.05 and 2.0 mol L^{-1}. The best results were observed for concentrations above 0.8 mol L^{-1}. Thus, a thiocyanate concentration of 1 mol L^{-1} was selected.

In earlier work, water was used as an eluent[24,25] to desorb Zn(II) only from the solid phase by depletion of the SCN$^-$ concentration inside the mini-column, thereby dissociating the Zn–SCN$^-$ complex. In the flow system, where detection was effected spectrophotometrically with PAR [2-(2-pyridylazo)-resorcinol], the use of organic eluents disturbed the colorimetric measurement. By using FAAS, this effect was minimized or eliminated. Hence, several solutions that could be used as eluent were tested. Nitric acid at 0.1 mol L^{-1}, 50% ethanol in 2% HNO$_3$ and 30% acetone in 2% HNO$_3$ were initially tested, and the signal obtained with water was used as a reference value. The performance of the organic eluents was much better than that observed with water and nitric acid, as can be seen in Fig. 2. Moreover, experiments were carried out with different compositions of these solutions in order to choose the best eluent solution for this system. Several concentrations of

organic solvent and acid were investigated, such as ethanol and acetone concentrations between 10 and 50% in acid solutions containing from 1 to 5% v/v HNO$_3$. For ethanol, the best combination between acid and solvent was 50% ethanol in 4% v/v HNO$_3$. However, this high concentration of ethanol caused instability of the baseline, probably due to alterations in flame transparency. For acetone, the best results were observed in solutions of 30% acetone in 2% v/v HNO$_3$ with absorbance signals of the same magnitude as that observed for the ethanol solution. Good stability of the baseline was found, in contrast to ethanol solutions. Hence, even though the signals had the same magnitude, the acetone-based eluent was chosen due to the lower noise generated and to preserve the Tygon pump tubes by using lower organic solvent concentrations.

The sample flow rate was investigated from 2.0 to 12.8 mL min^{-1} and the best response was obtained at a flow rate of 6.7 mL min^{-1}. However, the best ratio of absorbance signal : sample volume was observed at a flow rate of 3.0 mL min^{-1}. Therefore, 6.7 mL min^{-1} was chosen in order to reach a higher sample throughput and to improve sensitivity. Increasing the sample flow rate to higher values leads to a decrease of the analytical signal due to slow sorption kinetics which does not allow quantitative sorption of Zn(II) by the PUF mini-column. The thiocyanate flow rate was studied between 0.28 and 1.69 mL min^{-1} and the best analytical signals were found at 0.43 mL min^{-1}. As this flow rate maintains low sample dilution, it was chosen for further experiments. The eluent flow rate was tested from 1.69 to 5.8 mL min^{-1}. Its influence must be analysed from two points of view: (1) desorption kinetics and (2) dispersion of the sample plug. The best results were observed at 2.0 mL min^{-1} and under this condition there is a compromise between the two parameters cited above. However, by analysing the results obtained it is clear that the first parameter has little influence on the signal generated since the eluent solution provided a high desorption rate. This fact can be attested to by the short time required to complete the analytical signal. Hence, dispersion is the most important parameter in the control of the analytical signal. From 1.69 until 2.0 mL min^{-1} the signals increased because low dispersion occurred, and above 2.0 mL min^{-1} the signals decreased, probably due to the high flow rate which merely enhanced the sample dispersion and made the signals smaller. Therefore, an eluent flow rate of 2.0 mL min^{-1} was chosen.

According to our previous studies,[25] the mini-columns were constructed with 50 mg of PUF packed in mini-tubes (3 cm × 3 mm id). Under the optimized experimental conditions, very low overpressure was observed, in contrast to other sorbents employed in FIA systems.

The PUF mini-column retention capacity was evaluated by a simple experiment in which a defined volume (50 mL) of several solutions containing different amounts of zinc was pumped
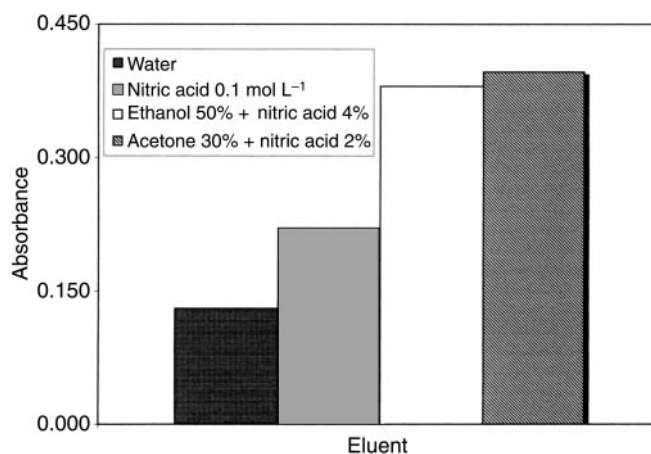

Fig. 2 Signals obtained with different eluent solutions for Zn^{II} preconcentration solutions containing 100 ng mL^{-1} during 1 min.

Table 1 Retention capacity of the PUF mini-column (50.6 mg) for Zn(II) under the optimized conditions; 50 mL of sample solution containing Zn(II)

Zn(II) solution/µg mL^{-1}	Zn retained/µg	Zn retained (%)
1	43.8	89.8
5	225	90.0
10	345	69.0
20	540	54.1
50	800	32.0
100	1485	29.7
200	2780	28.0
400	4388	21.9
600	4536	15.1

through the mini-column. Zinc was determined in the collected effluent and the amount retained was calculated by difference. Table 1 shows the behaviour of the system when these different solutions are pumped into the mini-column in terms of amount and per cent. of Zn(II) retained. From these data, the maximum retention capacity can be estimated as 4.5 mg of Zn per gram of PUF. However, as can also be seen in Table 1, in solutions with concentrations higher than 5 µg mL^{-1} of Zn(II), the Zn(II) retained decreases abruptly, decreasing the process efficiency.

3.2 Interference study

AAS has an inherent characteristic that is its selectivity. However, when working with PUF mini-columns, the presence of metal cations can affect the zinc signal by competition of their thiocyanate complexes for the PUF active sites. Hence, several possible interferent species, especially those which form thiocyanate complexes, were tested in order to verify the selectivity of the procedure: Fe(III), Cu(II), Co(II), Mn(II) and Cd(II). However, by using a mixed solution containing thiocyanate and 0.5% sodium citrate as masking agent, all interferents could be suppressed. Higher concentrations of sodium citrate were not used because they could affect the zinc signal by complexation with the metal.[26] Al(III) and Ni(II) were not tested because the rate of formation of their thiocyanate complexes is negligible. Several zinc:interferent ratios were tested such as 1:50, 1:100, 1:200 and 1:500 and no interferences were observed for any ratio for any of the species studied. Interference was considered to have occurred when a signal difference greater than 10% was observed.

3.3 Analytical features

The flow system was operated employing two different preconcentration times due to differences in the concentration of zinc in the two types of samples analysed. For the determination of zinc in the biological samples, a 1 min preconcentration time (6.7 mL of sample) was used. Under this condition, the system shows linearity for concentrations of Zn(II) between 20 and 100 ng mL^{-1}, which can be represented by the equation: $A = 0.0045$ [Zn(II) (ng mL^{-1})] + 0.003, $r = 0.999$. The detection limit, calculated as three times the standard deviation, was 3.0 ng mL^{-1}. The RSD was calculated by taking ten measurements of a 20 ng mL^{-1} solution and was always better than 2.5%. The quantification limit found was

10 ng mL^{-1}. The throughput achieved under the optimized experimental conditions was 40 samples per hour.

In the analysis of water samples, a longer preconcentration time must be used since the Zn(II) was below the concentration range used. A preconcentration time of 3 min (20.1 mL of sample) was sufficient to reach a measurable analytical signal. Calibration graphs were constructed from 10 to 50 ng mL^{-1}, the characteristic equation being $A = 0.0085$ [Zn(II) (ng mL^{-1})] + 0.002, $r = 0.999$. The detection limit, calculated as described above, was 0.85 ng mL^{-1}. The RSD, assessed by ten measurements of a 10 ng mL^{-1} solution, was 6.0%. In this case, the quantification limit was 5 ng mL^{-1} and the throughput was 17 samples per hour.

Preconcentration factors were calculated by comparing the calibration graphs constructed with the flow system with that obtained by direct aspiration of Zn(II) solutions. For a preconcentration time of 1 min, the preconcentration factor was 8 and for 3 min it was 15.

3.4 Applications

The developed flow system showed good performance in the preconcentration and determination of zinc in different matrices such as biological materials and natural waters.

Table 2 shows the results obtained in the analysis of biological reference materials and compares them with the certified values. In Table 3, the results obtained in the analysis of natural saline and non-saline waters and the recoveries found are presented. The results are expressed as the mean of three determinations and their confidence limits. As can be seen, good agreement was observed between the certified values and those obtained by the FIA method, and good recoveries were obtained from spiked natural waters.

4. Conclusions

An increase in the speed of the analytical process and a reduced sample manipulation were possible by coupling an on-line preconcentration procedure to a flame atomic absorption spectrometer.

PUF can be considered an effective extractant for zinc from thiocyanate solutions under flow conditions. Under the described conditions, it is possible to retain large amounts of Zn(II), *viz.*, 4.5 mg per gram of PUF. The PUF provides a useful means of concentrating zinc from different matrices by the continuous mode. Also, very low overpressure is observed inside the mini-column, in contrast to other sorbents frequently used as solid phases in flow systems. Other advantages in the

Table 3 Results obtained for the analysis of natural water samples, expressed as mean and standard deviation of three independent determinations, and recoveries after spiking with 20 µg L^{-1} of Zn(II)

Sample	Obtained value/µg L^{-1}	Recovery (%)
Sea-water[a]	63.0 ± 0.5	106
Sea-water[b]	40.2 ± 0.6	94
Underground water	30.0 ± 0.6	96

[a]Sample collected near to the sewage discharge of a Domestic Waste Treatment Plant. [b]Sample collected near to a shipyard.

Table 2 Results obtained for the analysis of biological reference materials. Results in µg g^{-1} dry mass

Reference sample	Obtained value	Certified value
Tuna Homogenate, IAEA, 350	16.9 ± 0.8	17.4 ± 0.8
Fish Tissue, IAEA, MA-B-3/TM	118.1 ± 6.9	109.2 ± 2.8
Rice Flour, Unpolished, High Cd Level, No. 10c, NIES	22.8 ± 1.5	23.1 ± 0.8
Copepoda, IAEA, MA-A-1/TM	156.6 ± 6.7	158 ± 2
Fish Flesh, IAEA, MA-A-2/TM	33.1 ± 2.4	33 ± 1

use of PUF as sorbent are the simplicity and the very low cost of constructing the flow system.

The main objective of this work was to improve both the selectivity and analytical throughput of the system developed previously[25] as well as to enhance sensitivity. The selectivity was excellent, making it possible to detect zinc in the presence of concentrations of interferents 500 times higher. The analytical throughput was improved, reaching a frequency of 40 samples per hour, but the sensitivity was virtually the same when determining zinc at the same concentration levels as those determined by FIA-spectrophotometry.[25] The detection limits achieved were satisfactory for the samples studied, and can be improved by using more sensitive detectors such as ICP-AES or ICP-MS employing a similar manifold to that used in this work.

The results obtained for the reference materials analysed were in good agreement with the certified values. Fresh and saline water samples can be analysed accurately by this methodology.

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References

1 M. Valcárcel and M. D. Luque de Castro, *Non-chromatographic Continuous Separation Techniques*, Royal Society of Chemistry, Cambridge, 1991.
2 J. L. Burguera, M. Burguera and A. Townshend, *Anal. Chim. Acta*, 1981, **127**, 199.
3 S. Olsen, L. C. R. Pessenda, J. Ruzicka and E. H. Hansen, *Analyst*, 1983, **108**, 905.
4 R. Purohit and S. Devi, *Analyst*, 1991, **116**, 825.
5 G. M. Greenway and A. Townshend, *Anal. Proc.*, 1991, **30**, 438.
6 Z. Fang, *Flow Injection Separation and Preconcentration*, VCH, Weinheim, 1993.
7 A. Sanz-Medel *Flow Injection with Atomic Spectrometric Detectors*, Elsevier, Amsterdam, 1999.
8 H. J. M. Bowen, *J. Chem. Soc.*, 1970, 1082.
9 T. Braun and A. B. Farag, *Talanta*, 1975, **22**, 699.
10 T. Braun and A. B. Farag, *Anal. Chim. Acta*, 1978, **99**, 1.
11 T. Braun, J. D. Navratil and A. B. Farag, *Polyurethane Foam Sorbents in Separation Science*, CRC Press, Boca Raton, FL, 1985.
12 T. Braun and A. B. Farag, *Anal. Chim. Acta*, 1974, **73**, 301.
13 D. Wildhagen and V. Krivan, *Anal. Chim. Acta*, 1993, **274**, 257.
14 S. L. C. Ferreira, V. A. Lemos, A. C. S. Costa, D. S. de Jesus and M. S. Carvalho, *J. Braz. Chem. Soc.*, 1998, **9**, 151.
15 J. J. Oren, K. M. Gough and H. D. Gesser, *Can. J. Chem.*, 1979, **57**, 2032.
16 M. S. Carvalho, I. C. S. Fraga, K. C. M. Neto and E. Q. S. Filho, *Talanta*, 1996, **43**, 1675.
17 M. S. Carvalho, M. L. F. Domingues, J. L. Mantovano and E. Q. S. Filho, *Spectrochim. Acta, Part B*, 1998, **53**, 1945.
18 T. Braun and M. N. Abbas, *Anal. Chim. Acta*, 1980, **119**, 113.
19 T. Braun and A. B. Farag, *Anal. Chim. Acta*, 1974, **69**, 85.
20 D. W. Lee and M. Halmann, *Anal. Chem.*, 1976, **48**, 2214.
21 A. Chow, G. T. Yamashita and R. F. Hamon, *Talanta*, 1981, **28**, 437.
22 T. Braun and M. N. Abbas, *Anal. Chim. Acta*, 1982, **134**, 321.
23 M. P. Maloney, G. J. Moody and J. D. R. Thomas, *Analyst*, 1980, **105**, 1087.
24 D. S. de Jesus, M. S. Carvalho, A. C. S. Costa and S. L. C. Ferreira, *Talanta*, 1998, **46**, 1525.
25 D. S. de Jesus, R. J. Cassella, S. L. C. Ferreira, A. C. S. Costa, M. S. Carvalho and R. E. Santelli, *Anal. Chim. Acta*, 1998, **366**, 263.
26 S. L. C. Ferreira, D. S. de Jesus, R. J. Cassella, A. C. S. Costa, M. S. Carvalho and R. E. Santelli, *Anal. Chim. Acta*, 1999, **378**, 287.
27 R. J. Cassella, R. E. Santelli, A. G. Branco, V. A. Lemos, S. L. C. Ferreira and M. S. Carvalho, *Analyst*, 1999, **124**, 805.
28 H. A. Flaschka, *EDTA Titrations—An Introduction to Theory and Practice*, Pergamon Press, Oxford, 2nd edn., 1964.
29 S. Icbinoki and M. Yamazaki, *Anal. Chem.*, 1985, **57**, 2219.

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