

See discussions, stats, and author profiles for this publication at: <https://www.researchgate.net/publication/232739359>

Spectrophotometric determination of trace amounts of phosphate in water and soil

Article in *Water Science & Technology* · October 2012

DOI: 10.2166/wst.2012.468 · Source: PubMed

CITATIONS

16

READS

28,179

6 authors, including:



Fahmida Khan

National Institute of Technology Raipur

60 PUBLICATIONS 250 CITATIONS

[SEE PROFILE](#)



N.K. Pandey

Indira Gandhi Centre for Atomic Research

92 PUBLICATIONS 304 CITATIONS

[SEE PROFILE](#)



Kamachi Mudali Mudali

Indira Gandhi Centre for Atomic Research

613 PUBLICATIONS 5,971 CITATIONS

[SEE PROFILE](#)

Some of the authors of this publication are also working on these related projects:



measurement of single ion activity coefficient using ion selective electrodes [View project](#)



Carbon Materials and Corrosion [View project](#)

Spectrophotometric determination of trace amounts of phosphate in water and soil

S. Ganesh, Fahmida Khan, M. K. Ahmed, P. Velavendan, N. K. Pandey and U. Kamachi Mudali

ABSTRACT

A simple spectrophotometric method has been developed for the determination of phosphate dissolved in soil and water. The method is based on the formation of phosphomolybdate with added ammonium molybdate followed by reduction with hydrazine in acidic medium. Orthophosphate and molybdate ions condense in acidic solution to give molybdophosphoric (phosphomolybdic) acid, which upon selective reduction (perhaps with hydrazinium sulphate) produces a blue colour, due to molybdenum blue of uncertain composition. The intensity of blue colour is proportional to the amount of phosphate. If the acidity at the time of reduction is 0.5 M in sulphuric acid and hydrazinium sulphate is the reductant, the resulting blue complex exhibits maximum absorption at 830 nm. The system obeys Lambert–Beer's law at 830 nm in the concentration range of 0.5–5 µg/mL of phosphate with a relative standard deviation (RSD) of 0.1% and correlation coefficient of 0.99. Molar absorptivity was determined to be $2.9 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}$ at 830 nm. The method is also applicable for the determination of phosphate in nuclear reprocessing plants, medical science, clinical science, agriculture, metallurgy and environmental science.

Key words | ion chromatography, molybdenum blue, phosphate, soil, spectrophotometry, water

S. Ganesh
P. Velavendan
N. K. Pandey
U. Kamachi Mudali
Reprocessing Group,
Indira Gandhi centre for atomic research,
Kalpakkam, 603102,
India

Fahmida Khan (corresponding author)
Department of Chemistry,
National Institute of Technology,
GE Road, Raipur, 492010,
India
E-mail: f.khan.chy@nitrr.ac.in

M. K. Ahmed
Nuclear Fuel Complex,
Hyderabad 500 062,
India

INTRODUCTION

Phosphorus is the most abundant element on the surface of the earth and is most commonly found as phosphate. It plays an important role in biochemical processes and is a key factor in the eutrophication of surface water (McCarty *et al.* 2003). Plants and animals depend on phosphate to perform photosynthesis and to respire. Increased phosphate concentrations are linked with increasing rates of plant growth (Yaqoop *et al.* 2003). In soils, phosphate is usually insoluble, due to it being bound to calcium, iron, and aluminum, or incorporated into organic matter – human and animal wastes or decaying matter. Usually the concentration of phosphate in the water from low fertilized soils or forest soils is only a few (µg/mL) micrograms per mL (Kempers 1975). The analytical chemistry of phosphate is very important in many fields, for example, medicine, clinical science, agriculture, metallurgy and environmental science (Motomizu *et al.* 1984).

The determination of the phosphate in soil requires the solubilization of phosphorus through the decomposition/destruction of materials in soil containing mineral and organic

phosphorus. The two most widely recognized procedures for determination of phosphate in soil are the sodium carbonate fusion method and the perchloric acid digestion method (Mendham *et al.* 2002). In recent years large quantities of phosphate have been used in beverages, detergents, fertilizer and also in sugar industries. Hence very sensitive analytical methods are required for the determination of phosphate in soil and water. Analytical methods have been proposed for the determination of phosphate at higher concentrations using titrimetry (Susic *et al.* 1961) and complexo-gravimetry (Mamadal & Kundu 2005). At ppm levels of phosphate, methods suggested include colorimetry (Krishnamurthy & Suryanarayana 1982; Williams *et al.* 1993), atomic absorption spectroscopy (Christian & Feldman 1968), flow injection analysis (Motomizu & Mitsuko 1987), high-performance liquid chromatography (HPLC) (Botker *et al.* 1994), ion chromatography (IC) (Ruiz-Calero & Galceran 2005; Nollet 2007) and spectrophotometry (Hayashi *et al.* 1960; Borissova & Mitropolitska 1979; Motomizu *et al.* 1984; Gutschik 1985; Smeller 1995; Mahadevaiah *et al.* 2007). Among such

methods, spectrophotometry involving molybdovanadate and ammonium molybdate is the most commonly used method. In the spectrophotometric method using ammonium molybdate, different reductants such as tin (II) chloride (Yathirajam & Dhamija 1979), ascorbic acid (Theodore 1986), hydrazine sulphate (Mendham *et al.* 2002; Carter & Gregorich 2008), and 1-amino-2-naphthol-4-sulfonic acid (Burton 1973) have been employed; some of these methods also involve complicated and expensive equipment and need an extraction procedure (Botker *et al.* 1994) and such techniques are usually not available in common laboratories. Therefore, a standardized, sensitive, simple spectrophotometric procedure to be used in routine process control analysis was required.

EXPERIMENTAL

Reagents

Reagents used, namely potassium di-hydrogen phosphate, ammonium molybdate and hydrazine sulphate, were of analytical reagent grade obtained from M/s Sarabhai M chemicals, Baroda, India. Water double distilled in glass was used. To start the process 14.325 mg of (100 µg/mL) potassium dihydrogen phosphate was weighed and transferred into a 100 mL standard measuring flask; it was dissolved in water and then diluted to the mark with distilled water. Then 1.7081 g of ammonium molybdate was dissolved in about 150 mL of warm water; a slightly milky solution resulted which was cooled to room temperature. It was then transferred into a 250 mL standard volumetric flask and diluted to the mark with water. Then 0.125 g of hydrazine sulphate was transferred into a clean 100 mL beaker. It was dissolved in about 50 mL of water and then the solution was transferred into a 100 mL standard measuring flask. The beaker was washed three to four times with water and washings were also transferred into the flask and the solution was diluted to the mark with water. Nitric acid (3.0 M) was prepared by suitable dilution of concentrated nitric acid (~16 M) with double distilled water.

Instrumentation

A UV-VIS Spectrophotometer UV5704SS was used for absorbance measurements. The wavelength range of this spectrophotometer is 340–900 nm. All the absorbance measurements were recorded by placing the sample tube containing the solution.

Ion chromatographic conditions

Ion chromatograph:	Metrohm modular Ion chromatography system
Analytical column:	Metrosep A Supp 5-250, 250 mm L × 4.0 mm ID
Guard column:	Metrosep A Supp 4/5
Eluent concentration:	3.2 mM Na ₂ CO ₃ + 1.0 mM NaHCO ₃
Eluent flow rate:	0.7 mL/min
Pressure:	13.5 MPa
Detector:	Suppressed conductivity
Analytical mode:	Isocratic
Injection loop:	20 µL
Temperature:	Ambient (25 °C)
Run time:	28 min
Quantitation:	Peak area

PROCEDURE

To suitable aliquots of stock standard solution, 1 mL of ammonium molybdate and 0.4 mL of hydrazine sulphate were added and the solution was made up to 10 mL with double distilled water in a standard measuring flask. The standard measuring flasks were kept in a water bath for heating for 30 min. The temperature of the water bath was set to 60 °C. While heating, a blue colour develops due to the formation of ammonium phosphomolybdate complex. After heating for 30 min the solution was cooled and its absorbance was measured at wavelength 830 nm. An experimental blank solution was used for carrying out correction for the baseline.

Water and soil samples were collected from various locations in Chhattisgarh State, India. A water sample was filtered through Whatman filter paper and collected. Aliquots of the sample were used for its phosphate analysis. Soil samples were digested, filtered and used for phosphate analysis. In the perchloric digestion method, 2 g of accurately weighed soil sample was taken in a 250 mL Erlenmeyer flask and heated in a hot plate or by using an aluminum block digester with a 250 mL digestion tube, then 20 mL of concentrated nitric acid was added into the flask and mixed well. To achieve oxidation, the organic matter in the sample was heated to approximately 130 °C. Organic matter oxidation was complete when the dark colour due to organic matter in the sample disappeared. The soil-HNO₃ mixture was allowed to cool slightly. In

the HClO_4 fume hood, 30 mL HClO_4 was added to the sample at boiling temperature $\sim 200^\circ\text{C}$ for 20 min. During that time dense white fumes appear and the insoluble solid material is left in the bottom of the flask. When necessary, a little extra HClO_4 was used to wash down any black particles that had stuck to the sides of the flask. After allowing the mixture to cool for another 10–15 min, the mixture was transferred to a 250 mL volumetric flask and made up to the mark with distilled water. Sediment was allowed to settle before taking the aliquot for analysis.

From these solutions, a suitable aliquot was taken and 1 mL of ammonium molybdate and 0.4 mL of hydrazine sulphate were added, it was made up to the mark with water in a 10 mL flask and heated for 30 min in a water bath maintained at 60°C . After blue colour developed, the solution was cooled and its absorbance measured at 830 nm as mentioned above. All solutions, namely experimental blanks, standards and unknown samples were run in duplicate.

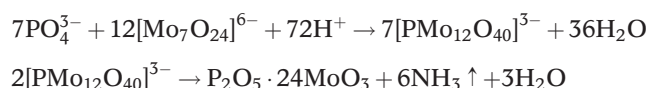
Ion chromatography

In order to compare the results obtained by spectrophotometric analysis with those of the ion chromatographic technique, experiments were conducted for the determination of phosphate present in environmental samples by the IC technique using a conductivity detector. Various phosphate standards of known concentrations ($0.1\text{--}10\text{ }\mu\text{g/mL}$) were prepared by dissolving standard potassium dihydrogen phosphate (KH_2PO_4) in Millipore water. Twenty microlitres of these solutions were injected directly into the IC column connected to the conductivity detector. A mixture of 3.2 mM sodium carbonate and 1.0 mM sodium bicarbonate solution is used as a mobile phase with a flow rate of 0.7 mL/min. A calibration graph was made for the concentration range of inorganic phosphate from 0.1 to $10\text{ }\mu\text{g/mL}$ with an RSD of 2.239% and a correlation coefficient of 0.9994. Unknown samples were filtered and $20\text{ }\mu\text{L}$ injected directly into the IC column for the determination of phosphate concentration.

RESULTS AND DISCUSSION

The developed method is based on the formation of phosphomolybdate complex due to the reaction between molybdate and phosphate, followed by its reduction with hydrazine sulphate in aqueous acidic medium. Orthophosphate and molybdate ions condense in acidic solution to

give molybdophosphoric (phosphomolybdic) acid, which upon selective reduction (perhaps with hydrazinium sulphate) produces a blue colour, due to molybdenum blue of uncertain composition. The intensity of blue colour is proportional to the amount of phosphate initially incorporated in the heteropolyacid. If the acidity at the time of reduction is 0.5 M in sulphuric acid and hydrazinium sulphate is the reductant, the resulting blue complex exhibits maximum absorption at 820–840 nm (Carter & Gregorich 2008):



Controlled heating in a water bath alone provided the uniform colour development and consistent results. Under optimized experimental conditions, with fixed concentration of molybdate and reducing agent, the colour intensity was found to be proportional to the amount of phosphate present in disodium hydrogen phosphate. The reaction conditions, as well as the various experimental parameters affecting the formation and stability of the coloured complex, were carefully investigated and optimized for quantitative determination of phosphate in various samples. The experimental variables such as concentration of ammonium molybdate and concentration of reducing agent, order of addition of the reagents and also colour stability of the complex were optimized for the effective determination of phosphate (Ganesh *et al.* 2009). Figure 1 provides the data on the optimum quantity of 0.7%

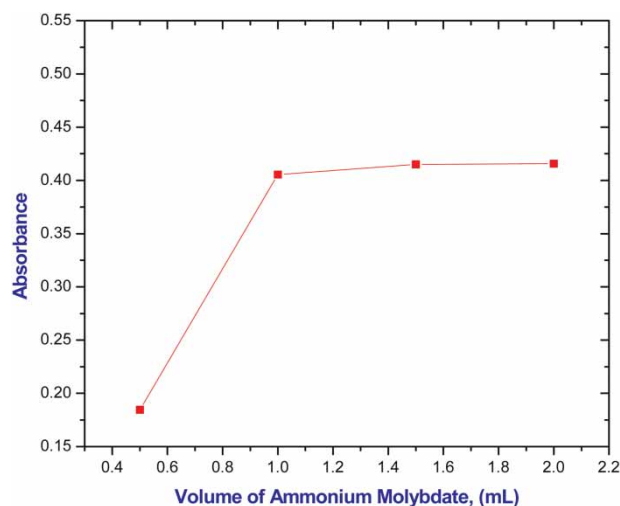


Figure 1 | Effect of ammonium molybdate.

ammonium molybdate needed for effective determination of phosphate. In this experiment, the different volumes of ammonium molybdate were added to the solution containing a constant volume of 0.4 mL hydrazine sulphate and 1.3 ppm of phosphate. From the figure, it was obvious that 1 mL of 0.7% ammonium molybdate is enough to determine the presence of phosphate. When higher volumes of ammonium molybdate were added, there was no change in absorbance. The maximum volume of hydrazine sulphate required for the determination of phosphate is presented in Figure 2. From this it was found that 0.4 mL of 0.12% hydrazine sulphate is sufficient for reduction and even if the volume of hydrazine sulphate is increased beyond 0.4 mL this has no effect on the phosphate estimation. The colour is stable with no difference in absorbance being detectable when phosphate standards were kept for up to 24 h. Figure 3 shows the calibration graph obtained at the wavelength of maximum absorption at 830 nm for standard phosphate solutions. At this wavelength the system obeys Lambert–Beer's law in the concentration range of 0.5–5.0 µg/mL of phosphate. Molar absorptivity was determined to be $2.9 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}$. Results obtained by using this methodology and instrumentation for the determination of phosphate in soil/water samples are reported in Tables 1 and 2. The results obtained are reproducible with a standard deviation of 0.1% and correlation coefficient of 0.9995. The metal ions such as Mg^{2+} , Ca^{2+} , Mn^{2+} , Co^{2+} , Ni^{2+} , Zn^{2+} , Cd^{2+} , Pb^{2+} , Hg^{2+} , Fe^{3+} , Al^{3+} , Cr^{3+} and UO_2^{2+} do not interfere with the determination of phosphate in concentrations up to 100 µg/mL. The presence of more than 500 molar amounts of As^{5+} can be tolerated by adding hydrazine

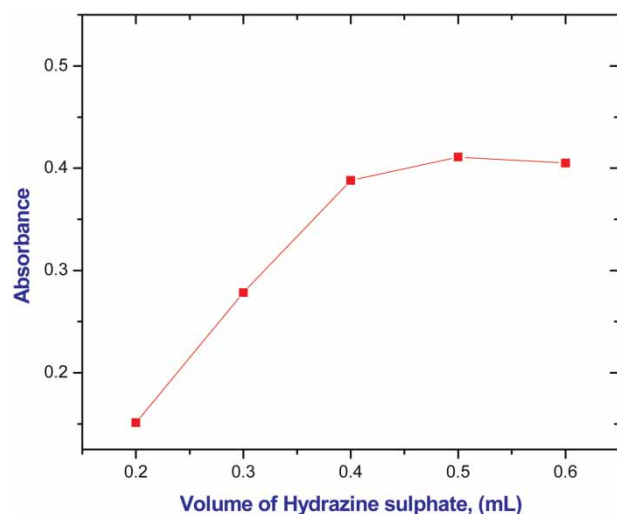


Figure 2 | Effect of hydrazine sulphate.

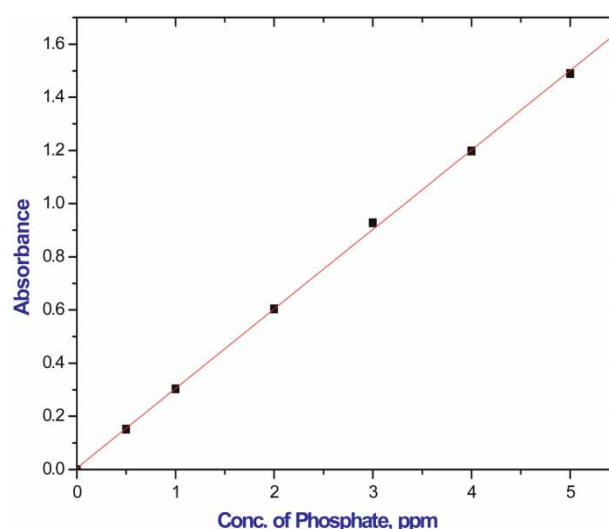


Figure 3 | Calibration graph for phosphate ion at 830 nm.

Table 1 | Typical results of phosphate in soil

S. No	Absorbance	Conc. of phosphate (µg/mL)
S-1	0.892	2.942
S-2	0.928	3.061
S-3	0.609	2.001
S-4	0.525	1.732
S-5	0.198	0.637
S-6	0.325	1.072
S-7	0.389	1.283
S-8	0.274	0.904
S-9	0.468	1.544
S-10	0.555	1.831

Soil samples were collected from various sites located in Chhattisgarh state, India.

Table 2 | Typical results of phosphate in water

S. No	Absorbance	Conc. of phosphate (µg/mL)
W1	0.411	1.356
W2	0.292	0.963
W3	0.310	1.022
W4	0.642	2.118
W5	0.788	2.600
W6	0.175	0.577
W7	0.243	0.802
W8	0.364	1.201
W9	0.511	1.685
W10	0.812	2.678

Water samples were collected from various sites located in Chhattisgarh state, India.

sulphate solution (Ohashi *et al.* 1981). The accuracy of the present method was checked by determining phosphate in various samples by both the present method and by an independent technique, namely, IC. A typical chromatogram and a calibration curve are shown in Figures 4 and 5. The results obtained for phosphate by the proposed method and by the IC method agreed well within the limits of experimental error and are reported in Table 3.

CONCLUSION

A modified spectrophotometric method for the determination of phosphate in soil is proposed. It is simple, fast and accurate. On the basis of recent investigations, this method shows potential for application to the determination

Table 3 | Comparison of two different analytical techniques

S. No.	Concentration of phosphate ($\mu\text{g/mL}$)	
	Present method	Ion-chromatographic technique
1	3.588	3.609
2	1.080	1.107
3	4.111	4.230
4	2.218	2.275
5	0.803	0.717
6	0.140	0.132

of phosphate in sediments, soil, plants and ores as well as in biological materials such as urine and blood serum.

REFERENCES

- Borissova, R. & Mitropolitska, E. 1979 Spectrophotometric determination of phosphate ions with the system cerium (III)-Arsenazo III. *Talanta* **26**, 543–547.
- Botker, H. E., Kimose, H. H., Helligso, P. & Nielsen, Y. T. 1994 Analytical evaluation of high energy phosphate determination by high performance liquid chromatography in myocardial tissue. *J. Mol. Cell. Cardiol.* **26**, 41–48.
- Burton, J. D. 1973 Problems in the analysis of phosphorous compounds. *Water Res.* **7**, 291–307.
- Carter, M. R. & Gregorich, E. G. 2008 *Soil Sampling and Methods of Analysis*, 2nd edition. CRC Press, Taylor & Francis Group, London, p. 265.
- Christian, G. D. & Feldman, F. J. 1968 Determination of nonmetals by atomic absorption spectrophotometry. *Anal. Chem. Acta* **40**, 173–179.
- Ganesh, S., Velavendan, P., Nair, B. S., Selvi, T., Ahmed, M. K. & Geetha, R. 2009 IGC Technical Report-302. Indira Gandhi Centre for Atomic Research Internal Technical Report, Kalpakkam, India.
- Gutschik, V. P. 1985 Occurrence of consistent nM levels of phosphate in doubly demineralized and demineralized-distilled water. *Talanta* **32**, 93–94.
- Hayashi, K., Dazuka, T. & Ueno, K. 1960 Spectrophotometric determination of phosphate using lanthanum chloranilate. *Talanta* **4**, 244–249.
- Kempers, A. J. 1975 Determination of submicroquantities of plant-available phosphate in aqueous soil extracts. *Plant Soil* **42**, 423–427.
- Krishnamurthy, K. & Suryanarayana, A. V. 1982 New spot test for phosphate. *Fresenius Z. Anal. Chem.* **312**, 546–546.
- Mahadevaiah, M. S., Yokendra, K., Mansour, S., Abdul, G., Suresha, M. S., Sathish, M. A. & Nagendrappa, G. 2007 A simple spectrophotometric determination of phosphate in sugarcane juices, water and detergent samples. *E. J. Chem.* **4**, 467–473.

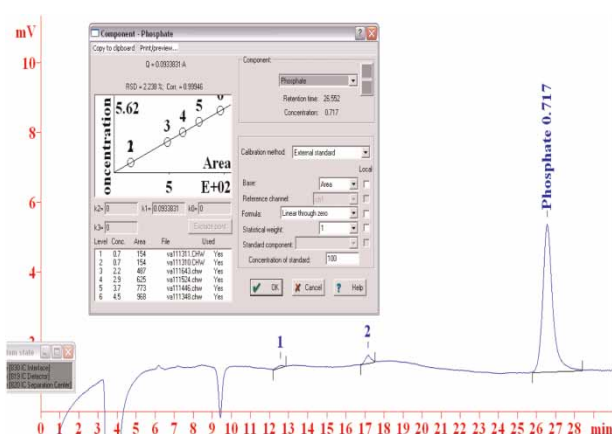


Figure 4 | Typical ion chromatogram for phosphate ion.

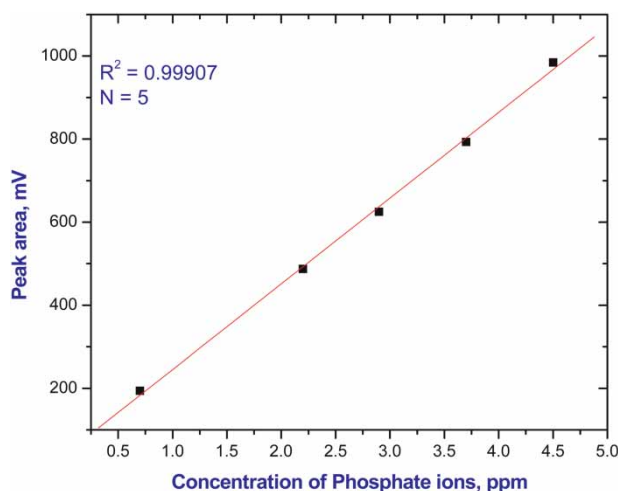


Figure 5 | Calibration graph for phosphate ion by the ion chromatographic (IC) technique.

- Mamadal, S. & Kundu, D. 2005 *J. Indian Chem. Soc.* **82**, 1030–1031.
- McCarty, C., McCarty, P. & Parkin, G. F. 2003 *Chemistry for Environmental Engineering and Science*, 5th edition. Tata McGraw-Hill Publishing Company Ltd., Noida, p. 677.
- Mendham, J., Denney, R. C., Barnes, J. D. & Thomas, M. 2002 *Vogel's Text book of Quantitative Chemical Analysis*, 6th edition. Longmans, London, pp. 473 and 672.
- Motomizu, S. & Mitsuko, O. 1987 Spectrophotometric determination of phosphorus as orthophosphate based on solvent extraction of the ion associate of molybdophosphate with malachite green using flow injection. *Analyst* **112**, 295–300.
- Motomizu, S., Wakimoto, T. & Kyoji, T. 1984 Solvent extraction-spectrophotometric determination of phosphate with molybdate and malachite green in river water and sea-water. *Talanta* **31**, 235–240.
- Nollet, M. L. N. 2007 *Handbook of Water Analysis*, 2nd edition. CRC Press, Taylor and Francis Group, London, 284 pp.
- Ohashi, K., Enomoto, T. & Yamamoto, K. 1981 Dual-wavelength spectrophotometric determination of trace amounts of phosphate in the presence of large amounts of silicate using molybdenum blue. *Bull. Chem. Soc. Jpn.* **54**, 1889–1890.
- Ruiz-Calero, V. & Galceran, M. T. 2005 Ion chromatographic separations of phosphorus species: a review. *Talanta* **66**, 376–410.
- Smeller, J. M. 1995 Comparison of sample preparation methods for the spectrophotometric determination of phosphorus in soil and coal fly ash. *Analyst* **120**, 207–210.
- Susic, S. K., Njengovan, V. N. & Solaja, B. Z. 1961 Über die Verwendung von Hexamethylentetramin zur gravimetrischen Trennung dreiwertiger Kationen von den zweiwertigen. *Z. Anal. Chem.* **183**, 412–416.
- Theodore, G. T. 1986 Determination of aqueous phosphate by ascorbic acid reduction of phosphomolybdic acid. *Anal. Chem.* **58**, 223–229.
- Williams, K. E., Haswell, S. J., David, A. B. & Preston, G. 1993 Determination of total phosphate in waste waters by on-line microwave digestion incorporating colorimetric detection. *Analyst* **118**, 245–248.
- Yaqoop, M., Nabi, A. & Paul, J. W. 2003 Determination of nanomolar concentrations of phosphate in fresh waters using flow injection with luminal chemiluminescence detection. *Anal. Chimica Acta* **510**, 213–218.
- Yathirajam, V. & Dhamija, S. 1979 Extractive separation and spectrophotometric determination of tungsten as ferrocyanide. *Talanta* **24**, 317–321.

First received 24 December 2011; accepted in revised form 27 June 2012