

This article was downloaded by:

On: 25 January 2011

Access details: *Access Details: Free Access*

Publisher *Taylor & Francis*

Informa Ltd Registered in England and Wales Registered Number: 1072954 Registered office: Mortimer House, 37-41 Mortimer Street, London W1T 3JH, UK



Separation Science and Technology

Publication details, including instructions for authors and subscription information:

<http://www.informaworld.com/smpp/title~content=t713708471>

Merrifield Resin Supported Chelate Forming Calix[4]arene-o-vanillinthiosemicarbazone Resin Employed for the Separation, Preconcentration and Trace Determination of Cr(VI), As(III) and Tl(I) in Water Samples

Vinod K. Jain^a; Rujul A. Pandya^a; Shibu G. Pillai^a; Pranav S. Shrivastav^a; Y. K. Agrawal^b

^a Chemistry Department, School of Sciences, Gujarat University, Ahmedabad, India ^b Institute of Pharmacy and Sciences, Nirma University of Science and Technology, Ahmedabad, India

To cite this Article Jain, Vinod K. , Pandya, Rujul A. , Pillai, Shibu G. , Shrivastav, Pranav S. and Agrawal, Y. K.(2006) 'Merrifield Resin Supported Chelate Forming Calix[4]arene-o-vanillinthiosemicarbazone Resin Employed for the Separation, Preconcentration and Trace Determination of Cr(VI), As(III) and Tl(I) in Water Samples', Separation Science and Technology, 41: 1, 123 — 147

To link to this Article: DOI: 10.1080/01496390500445626

URL: <http://dx.doi.org/10.1080/01496390500445626>

PLEASE SCROLL DOWN FOR ARTICLE

Full terms and conditions of use: <http://www.informaworld.com/terms-and-conditions-of-access.pdf>

This article may be used for research, teaching and private study purposes. Any substantial or systematic reproduction, re-distribution, re-selling, loan or sub-licensing, systematic supply or distribution in any form to anyone is expressly forbidden.

The publisher does not give any warranty express or implied or make any representation that the contents will be complete or accurate or up to date. The accuracy of any instructions, formulae and drug doses should be independently verified with primary sources. The publisher shall not be liable for any loss, actions, claims, proceedings, demand or costs or damages whatsoever or howsoever caused arising directly or indirectly in connection with or arising out of the use of this material.

**Merrifield Resin Supported
Chelate Forming
Calix[4]arene-o-vanillinthiosemicarbazone
Resin Employed for the Separation,
Preconcentration and Trace Determination
of Cr(VI), As(III) and Tl(I) in Water
Samples**

**Vinod K. Jain, Rujul A. Pandya, Shibu G. Pillai, and
Pranav S. Shrivastav**

Chemistry Department, School of Sciences, Gujarat University,
Ahmedabad, India

Y. K. Agrawal

Institute of Pharmacy and Sciences, Nirma University of Science
and Technology, Ahmedabad, India

Abstract: Calix[4]arene based polymer supported chelating resin **8** has been synthesized by covalently linking calix[4]arene-o-vanillinthiosemicarbazone via its 'lower rim' to Merrifield resin and was characterized by FT-IR, ¹H-NMR and elemental analysis. Resin **8** was competently used to separate and preconcentrate toxic metal ions Cr(VI), As(III) and Tl(I) in a column prior to their determination by UV-Visible and GF-AAS with R.S.D. between 1.0–1.4%. Various physico-chemical parameters like pH, concentration of eluting agents, flow rate, total sorption capacity, metal-ligand stoichiometry, exchange kinetics, preconcentration factor, distribution coefficient, breakthrough capacity, resin stability, effect of electrolytes, and associated metal ions have been studied. Detection limits ($3\sigma_B$) of 5.83, 8.48 and $5.56 \mu\text{g L}^{-1}$, along with preconcentration factors of 117, 90 and 105 for Cr(VI), As(III) and Tl(I) respectively were achieved. Resin **8** was successfully applied to the

Received 17 August 2005, Accepted 10 October 2005

Address correspondence to Vinod K. Jain, Chemistry Department, School of Sciences, Gujarat University, Ahmedabad 380009, India. E-mail: drvkjain@hotmail.com

separation and trace determination of Cr(VI), As(III) and Tl(I) from natural water samples of Ahmedabad city and the standard environmental, biological and geological reference materials.

Keywords: Calix[4]arene, o-vanillinthiosemicarbazone, preconcentration, Merrifield resin, GF-AAS

INTRODUCTION

The concentration of toxic metal ions like Cr(VI), Cu(II), Cd(II), As(III), Pb(II), Hg(II), Tl(I) etc. has increased beyond environmentally and ecologically sustainable levels due to the increase in industrial activity in the field of chemicals, dyes, color chemicals, petrochemicals, electroplating, textiles, and pharmaceuticals sectors over the years. It has resulted in severe contamination of ground and surface water; for e.g. rivers, lakes, rivulets etc. Especially in the developing countries, water and soil degradation generated by industrial effluents has been a serious issue (1). In order to achieve balanced growth, which is sensitive towards conservation of scarce natural resources, time-to-time analysis of these metal ions in natural water samples as well as in bio fluids is necessary. In natural water and biological systems Cr(VI), As(III), and Tl(I) are found in low levels and their separation and preconcentration are obligatory ahead of their trace quantification by instrumental or chemical methods of analysis. The metabolism and toxicology of Cr(VI), As(III), and Tl(I) are of great concern as they get accumulated in body systems. Their determination in food, blood, and tissues samples is also very important in order to identify cases of human intoxication leading to untold suffering in developing and least developed countries (2–4). Extraction of these metal ions is a tedious process as they are associated with a variety of complex species present in the natural aquatic systems. Many times they are found in such a low concentration that even sophisticated instrumental techniques like FAAS and ICP-OES are not sensitive enough to detect and quantify these toxic metal ions in natural water samples and their prior separation and preconcentration is required. Polymeric chelating ion exchange resins, which act as specific, sensitive and selective complexants for metal ions have been found to be very useful. For solid phase extraction of toxic metal ions, polymeric calixarenes (5–8) can play a vital role due to their characteristics such as lower toxicity, affordability, ease of synthesis, and operation. They are more eco-friendly compared to liquid-liquid extraction method where in disposal of organic waste has been a serious environmental hazard. Various calixarene derivatives have been recently studied with Tl(I) as these cavitands have been found useful in the complexation studies (9, 10). Thrust of the ongoing research programmes is on arsenic determination at trace and ultra trace levels and its speciation (11–21).

So far, there are no reports on the use of such polymeric calixarenes for separation and preconcentration of As(III), Cr(III), and Tl(I). Therefore, we planned to introduce o-vanillinthiosemicarbazone, a versatile chelating agent for toxic metal ions (22, 23) on the calix[4]arene scaffold, held by a polymer backbone (24). In this article, synthesis of Merrifield resin supported calix[4]arene-o-vanillinthiosemicarbazone (resin **8**) and its application to the separation, preconcentration, and trace determination of Cr(VI), As(III) and Tl(I) in various natural water samples collected from Ahmedabad city and the standard environmental, biological and geological reference materials (SRMs) procured from National Institute of Standards and Technology (NIST, USA) have been described.

EXPERIMENTAL

Apparatus

Precoated silica gel plates (Merck 60 F₂₅₄) were used for TLC. FT-IR spectra were recorded on a Jasco infrared spectrophotometer as KBr pellets. Elemental analysis was done on Heraeus Carlo Erba 1108 elemental analyzer. ¹H-NMR spectra were recorded on Bruker operating at 100 MHz for proton in DMSO – d₆ with TMS as internal standard. A Systronics digital pH meter Model 335 was used for pH measurements. Flow rate in the column was adjusted with a Miclins peristaltic pump PP-10. Spectral measurements were done on a Hitachi 3210 UV-Visible spectrophotometer using 10 mm quartz cells. Atomic absorption measurements were performed on Perkin-Elmer model 420 atomic absorption spectrophotometer (AAS) pyrolytically coated HGA-76 graphite furnace (GF) for Cr(VI) and Tl(I). Total arsenic determination in the form of As(III) was done on a Chemito 201 AAS with a continuous flow hydride generator for arsine generation. Operating conditions for hydride generator were: argon flow rate-70 cm³/min; concentration of NaBH₄-1% w/v; concentration of HCl-5% v/v; sampling time-25 seconds; reaction time-25 seconds. Measurements were based on integrated absorbance.

Operating Conditions for AAS

	Cr(VI)	As(III)	Tl(I)
H.C. Lamp (mA)	10	10	10
Slit (nm)	0.5	0.5	0.5
Wavelength (nm)	357.9	193.7	276.8
Flame	Air-C ₂ H ₂	Air-C ₂ H ₂	Air-C ₂ H ₂
Fuel (l min ⁻¹)	2.0	2.0	2.0
Oxidant (l min ⁻¹)	8	10	8
Burner (cm)	10	10	10

Reagents

All chemicals used in this work were of analytical grade of E. Merck or Lancaster. Merrifield peptide resin [chloromethylated polystyrene divinylbenzene co-polymer or CMPDB] (1% cross linked, 1.5 mmol equivalent of Cl/g of resin, 200–400 mesh) was procured from Aldrich. Glassware were soaked in 10% HNO_3 overnight before use and cleaned repeatedly with double distilled deionized water. Solvents were purified by the method of Weissberger et al (25). All aqueous solutions were prepared with quartz distilled deionized water, which was further purified by a Millipore Milli-Q water purification system. Standard stock solutions ($2000 \mu\text{g ml}^{-1}$) of Cr(VI), As(III) and Tl(I) were prepared and standardized spectrophotometrically (26, 27). pH of solutions was maintained as described in the literature (28).

Synthesis (Fig. 1)

Synthesis of 5,11,17,23-tetratert-butyl-25,26,27,28-tetrahydroxycalix[4]arene **1** and 25,26,27,28-tetrahydroxycalix[4]arene **2** were done as per the method of Gutsche et al. [48,49]. 26,28-dimethoxy-25,27-dihydroxycalix[4]arene **3** and 11,23-dinitro-26,28-dimethoxy-25,27-dihydroxycalix[4]arene **4** were synthesized as reported by Arduini et al. [50].

Synthesis of Product 5 i.e. 11,23-diamino-26,28-dimethoxy-25,27-dihydroxycalix[4]arene

Suspension of 5 g **4** (9.2 mmol), 10 ml hydrazine hydrate and a catalytic amount of Raney nickel in 100 ml methanol was refluxed for 6 hours in a 250 ml round bottomed flask. Reaction mixture was allowed to cool to room temperature, filtered and solvent was distilled under reduced pressure using a rotary evaporator. Residue was taken in 60 ml dichloromethane, washed thrice with 30 ml water and dried over MgSO_4 . After filtration, solvent was distilled under reduced pressure using a rotary evaporator to give pure **5** in 3 g (68%) yield; mp $> 300^\circ\text{C}$ (dec.). FT-IR (KBr): 3180 cm^{-1} (O-H stretching); 1252 cm^{-1} (C-O stretching); 3435 cm^{-1} (N-H stretching). $^1\text{H-NMR}$ (DMSO-d_6): 9.7 ppm (s, 2, ArOH); 7.2 ppm (s, 10, ArH); 3.4 ppm (br d, 8, ArCH₂Ar); 3.7 ppm (s, 6, OCH₃); 5.5 ppm (s, 4, ArNH₂). Anal. calcd. for $\text{C}_{30}\text{H}_{30}\text{O}_4\text{N}_2$: C, 74.65; H, 6.27; N, 5.81. Found: C, 74.39; H, 6.08; N, 5.61.

Synthesis of Product 6 i.e. 11,23-bis(chlorodiazenyl)-26,28-dimethoxy-25,27-dihydroxycalix[4]arene

10 g amine **5** (20.7 mmol) was diazotized in 35 ml HCl (40%) at 0°C in ice-bath with constant stirring for 30 minutes using 10 g NaNO_2 (145 mmol) in 100 ml $\text{H}_2\text{O}:\text{C}_2\text{H}_5\text{OH}::1:1$ media to yield **6**, which was then reacted further in situ.

Synthesis of Product 7 i.e. 11,23-bis(o-vanillinthiosemicarbazone)-26,28-dimethoxy-25,27-dihydroxycalix[4]arene

Solution containing **6** was taken in ice-bath in a beaker with an addition funnel supported over it containing 10 g o-VTSC (44.4 mmol) [Note 1] in 40 ml NaOH (15 g, 375 mmol) in water. Addition was completed in 1 hour at 0°C with constant stirring. Brownish-red colored product **7** was solidified by

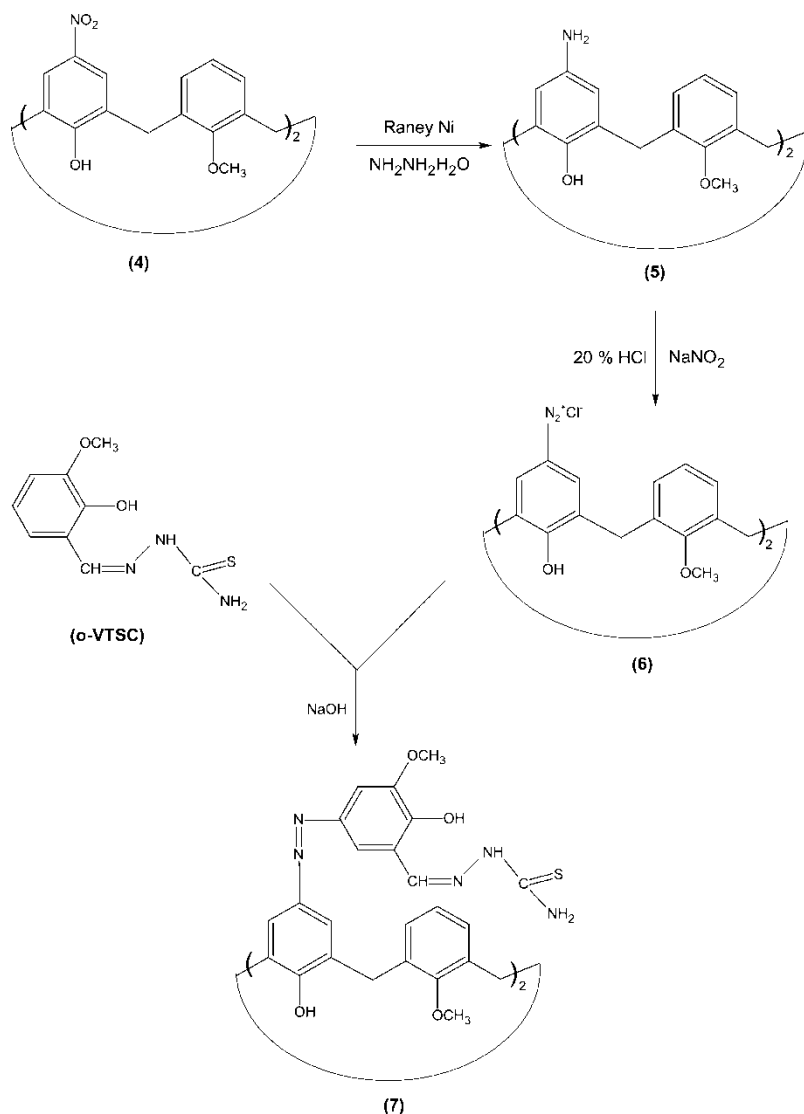
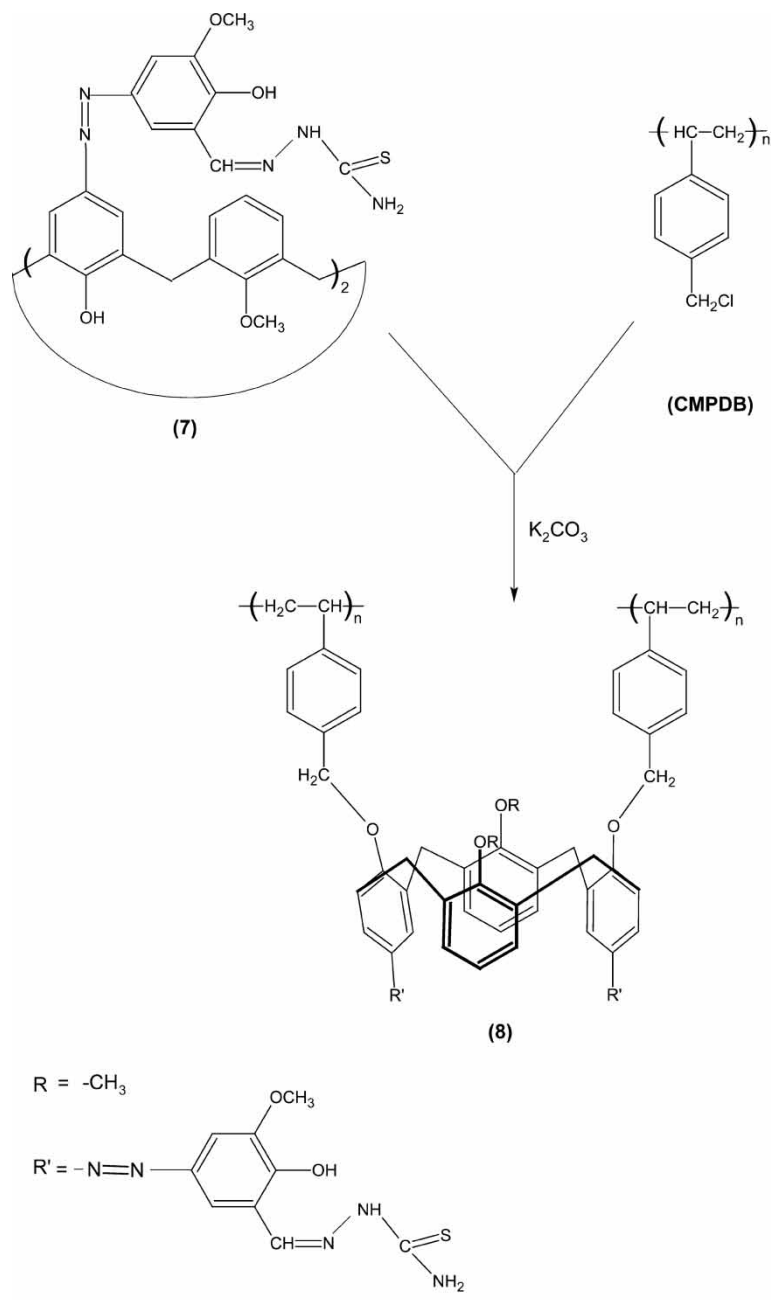


Figure 1. Reaction flowchart. (Continued)



(8) : CMPDB supported calix[4]arene-o-vanillinthiosemicarbazone

Figure 1. Continued.

evaporation of solvent. Purification was done by crystallization in iso-amyl acetate. Yield: 16 g (81%), mp > 300°C (dec.). FT-IR (KBr): 3464 & 3227 cm⁻¹ (O-H str.); 1591 cm⁻¹ (CH=N str.). ¹H-NMR (DMSO-d₆) (δ in ppm): 3.3 (s, 8, ArCH₂Ar); 3.6–3.8 (s, 12, OCH₃); 7.1–8.1 (s, 14, ArH); 10.1 (s, 4, ArOH). Anal. calcd. for C₄₈H₄₆O₈N₁₀S₂: C, 60.34; H, 4.86; N, 14.67; S, 6.72. Found: C, 59.96; H, 4.65; N, 14.76; S, 6.45.

[Note 1: Synthesis of 2-hydroxy-3-methoxybenzaldehyde thiosemicarbazone (o-VTSC) 5 g o-vanillin (33 mmol) was refluxed with 3 g thiosemicarbazide (33 mmol) in 80 ml, 95% ethanol at 75°C with constant stirring for 4 hrs. 0.5 g sodium acetate was added to maintain pH > 7. Reaction mixture was then cooled and filtered. Crude product was washed thrice with ethanol and water. Yield of pure o-VTSC was 7 g, mp 207°C.]

Synthesis of 8 by Loading of 7 on CMPDB

A mixture containing 10 g **7** (10.5 mmol), 10 g K₂CO₃ (72 mmol), and 10 g of CMPDB in 150 ml of DMF:THF::2:1 was heated at 75°C temperature for 8 hours under N₂ gas atmosphere. Thereafter polymeric beads of resin **8** were separated through a porous filter at pump. Repeated washing with DMF and THF followed by water were carried out to complete remove unreacted **7** and weight of dried resin **8** was found to be 12.34 g. Amount of **7** loaded on CMPDB was evaluated by difference of mass taken before and after coupling for dried resin and was found to be 23.4%

$$\text{Percentage of } 7 \text{ hooked on CMPDB} = \frac{W_2 - W_1}{W_1} \times 100\%$$

where, weight of dried CMPDB : (W₁)

weight of dried resin **8** : (W₂)

Elemental analysis of nitrogen and sulphur in resin **8** were found to be 2.78% and 1.26% respectively, which further confirmed amount of loading to be 23–24%. It shows that only 0.4 mmol i.e. 26.5% of total available Cl (1.5 mmol/g of CMPDB) reacted with 0.2 mmol **7**.

Procedure for Column Concentration and Determination of Cr(VI), As(III) and Tl(I)

A glass column (200 × 8.0 mm) was prepared by adding slurry of beads of same mesh size having 1 g resin **8** in CH₃OH:H₂O::1:1. Height of column section containing resin **8** (bed height) was 15 mm. It was conditioned with different buffer solutions for samples containing Cr(VI), As(III) and Tl(I) prior to their elution from column. Suitable aliquot of solution containing 0.01–3.03, 0.065–7.2, and 0.023–4.9 μg ml⁻¹ of Cr(VI), As(III) and Tl(I) respectively was passed through column after adjusting appropriate pH at an optimum flow rate. Stripping of metal ions from resin bed was carried

Table 1. Parameters optimized for sorption and desorption of Cr(VI), As(III) and Tl(I) on resin **8**

No.	Parameters	Metal Ions		
		Cr(VI)	As(III)	Tl(I)
1	pH Range	3.0–4.0	4.5–5.5	5.5–6.5
2	Flow rate (ml min ⁻¹)	1.5	1.0	1.0
3	Concentration of acid for desorption	0.1 N H ₂ SO ₄	0.5 N HCl	1 N HNO ₃ / 3 N HCl
4	Total Sorption capacity (µg g ⁻¹ of Resin)	11325	15575	34397
5	Distribution coefficient (K _d)	3360	2600	2128
6	Preconcentration factor (PF)	117	90	105
7	Breakthrough capacity (µg g ⁻¹ of resin)	2151	2647	5503
8	R.S.D. (%) ^a	1.2–1.4	1.1–1.3	1.0–1.2
9	Average recovery (%)	98	99	97–98
10	t _{1/2} for exchange (minutes)	6.5	9.0	12.5

^aTen determinations.

out by suitable mineral acids. Eluants were collected in a 25 ml volumetric flask, made to volume by double distilled water and determined spectrophotometrically as well as by GF-AAS (Table 1).

RESULTS AND DISCUSSION

Parameters Optimized for Separation and Preconcentration of Cr(VI), As(III) and Tl(I)

1. Effect of pH
- A glass column was packed with 1 g resin **8**. 100 ml solutions containing 3, 7, and 4.5 µg ml⁻¹ Cr(VI), As(III) and Tl(I) respectively were passed through the column at their optimum flow rate by varying pH. Metal sorption (%) was measured by desorbing resin **8** with a suitable eluting agent and then determined by spectrophotometry and GF-AAS. Optimum pH for Cr(VI), As(III) and Tl(I) were found to be 3.5, 5.0, and 6.0 respectively (Table 1, Fig. 2). Their selectivity in pH of sorption suggests possibility of separation of these metal ions in presence of each other in column.
2. Effect of Flow Rate
- Effect of flow rate on extent of metal ion sorbed on resin was studied in column packed with 1 g resin **8** at 30°C. Feed solutions containing 3, 7,

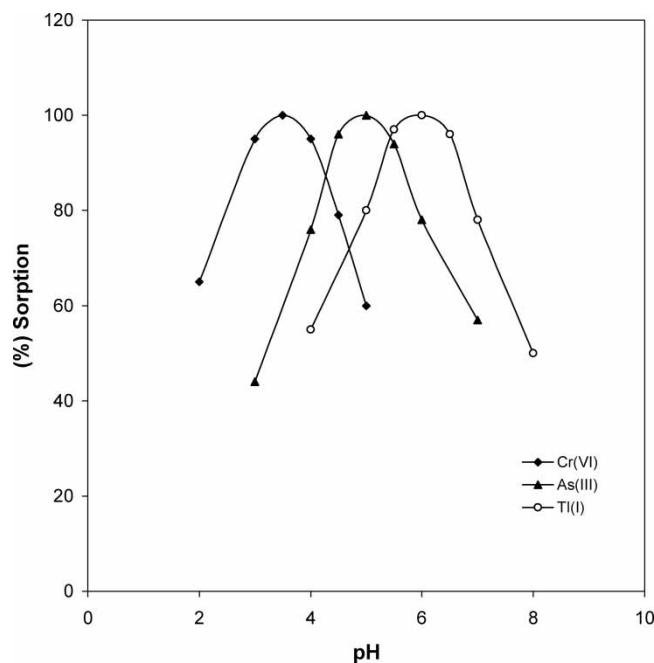


Figure 2. Effect of pH on Sorption of Cr(VI), As(III) and Tl(I) by resin **8**. Amount of resin **8** in column: 1 g; Volume of metal ion solution passed: 100 ml; Cr(VI): $3 \mu\text{g ml}^{-1}$, Elution by: 0.1 N H_2SO_4 ; As(III): $7 \mu\text{g ml}^{-1}$; Elution by: 0.5 N HCl; Tl(I): $4.5 \mu\text{g ml}^{-1}$, Elution by: 1 N HNO_3 .

and $4.5 \mu\text{g ml}^{-1}$ Cr(VI), As(III) and Tl(I) respectively were passed at different flow rates (0.5, 1.0, 1.5, 2.0 etc. ml min^{-1}) maintained by peristaltic pump. Flow rate of liquid during chromatographic separation should be such that working conditions are close to equilibrium state as flow rate depends upon particle size and temperature. Optimum flow rate may be defined as rate of flow of effluent through column at which more than 98% sorption takes place. Optimum flow rates obtained were 1.5, 1.0, and 1.0 ml min^{-1} for Cr(VI), As(III) and Tl(I) respectively. An increase in flow rate resulted in decrease in sorption due to insufficient contact time between resin phase and metal ion solution (Table 1, Fig. 3).

3. Effect of Concentration of Eluting Agents

Effect of various eluting agents like HCl, HNO_3 and H_2SO_4 was studied by taking their different concentrations. 1 g resin **8** in column was conditioned at pH of maximum sorption and then fed with 100 ml solutions containing 3, 7 and $4.5 \mu\text{g ml}^{-1}$ Cr(VI), As(III) and Tl(I) respectively. Metal ions were desorbed with different mineral acids and then determined by spectrophotometry and GF-AAS (Table 1 & 2). It was observed that quantitative elution was possible with 15 ml of 0.1N

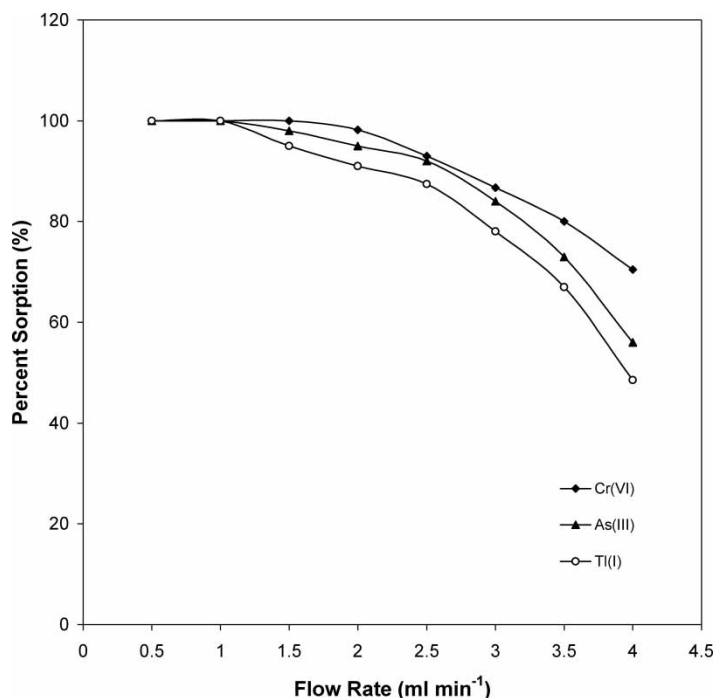


Figure 3. Effect of Flow Rate on Sorption of Cr(VI), As(III) and Tl(I) by resin 8. Amount of resin 8 in column: 1 g; Cr(VI): $3 \mu\text{g ml}^{-1}$; pH 3.5; As(III): $7 \mu\text{g ml}^{-1}$; pH 5.0; Tl(I): $4.5 \mu\text{g ml}^{-1}$; pH 6.0.

H_2SO_4 , 0.5 N HCl, and 1 N HNO_3 /3 N HCl for Cr(VI), As(III), and Tl(I) respectively.

4. Total Sorption Capacity

Total sorption capacity studies were carried out using batch techniques for Cr(VI), As(III) and Tl(I). 1 g resin 8 was equilibrated with 500 ml solution containing $100 \mu\text{g ml}^{-1}$ metal ions at their optimum pH for 2 hours at 30°C . Amount of metal ions sorbed on resin 8 was calculated from difference in metal ion concentration before and after desorption (Table 1). Total sorption capacities of resin were found to be 11325, 15575, and $34397 \mu\text{g g}^{-1}$ for Cr(VI), As(III) and Tl(I) respectively.

5. Equilibrium Distribution Coefficient K_d

Exchange equilibria are very often expressed in terms of distribution coefficient K_d . This quantity is given by ratio of equilibrium concentrations of same metal ion in resin phase and in solution. K_d of metal ions between resin 8 and aqueous phase was determined by batch experiment. 1 g resin 8 was equilibrated with 25 ml solution containing not more than 453, 623 and $1376 \mu\text{g ml}^{-1}$ Cr(VI), As(III) and Tl(I) respectively for 2 hours at 30°C . Solution was filtered to remove resin 8 and filtrate was

Table 2. Effect of concentration of eluting agents for desorption of Cr(VI), As(III) and Tl(I) from resin **8**

Conc. (N)	HCl			HNO ₃			H ₂ SO ₄		
	Cr(VI) (%)	As(III) (%)	Tl(I) (%)	Cr(VI) (%)	As(III) (%)	Tl(I) (%)	Cr(VI) (%)	As(III) (%)	Tl(I) (%)
0.01	5.5	22.8	5.0	5.3	4.3	24.5	66.6	4.8	6.0
0.1	12.5	66.5	9.7	11.5	16.3	48.8	99.8	11.1	9.0
0.5	17.1	99.7	12.0	27.7	19.3	69.0	—	16.8	13.1
1.0	21.6	—	17.9	32.5	21.6	99.9	—	22.7	18.2
1.5	29.5	—	32.3	44.6	27.5	—	—	30.5	28.5
2.0	40.7	—	63.0	50.1	36.3	—	—	38.2	39.1
3.0	49.9	—	99.8	58.1	42.8	—	—	45.9	52.3

^aAverage of ten determinations.

Experimental condition: Resin: 1 g; Volume of solution passed: 100 ml; Cr(VI): pH 3.5; 3 µg ml⁻¹; As(III): pH 5.0; 7 µg ml⁻¹; Tl(I): pH 6.0; 4.5 µg ml⁻¹.

subjected to spectrophotometric and GF-AAS determination of metal ion content. Concentration of eluted metal ion solution was also measured (Table 1). K_d for Cr(VI), As(III) and Tl(I) were found to be 3360, 2600 and 2128 respectively. K_d or efficiency of resin **8** was measured using equation,

$$K_d = \frac{\text{Amount of metal ion taken up by resin}}{\text{Amount of metal ion remaining in solution}} \times \frac{\text{Volume of solution}}{\text{Amount of resin taken}}$$

Plots of log K_d against pH (Fig. 4) were straight lines with slopes nearly equal to unity, indicating 1 : 1 stoichiometry between metal ions and resin **8** (29).

6. Exchange Kinetics

Batch experiments were carried out to determine rate of loading of Cr(VI), As(III) and Tl(I) on resin **8**. 1 g resin **8** was stirred with 100 ml solution containing 113, 156 and 344 µg ml⁻¹ Cr(VI), As(III) and Tl(I) respectively at 30° C. 5 ml of solution was withdrawn at predetermined intervals and analyzed. Concentration of metal ion in aqueous phase was determined by spectrophotometry and GF-AAS. Amount of metal ion loaded on resin phase was calculated by measuring concentration of metal ion in feed solution (Table 1, Fig. 5). Time taken for sorption of 50% of metal ions ($t_{1/2}$) was found to be 6.5, 9.0 and 12.5 minutes for Cr(VI), As(III) and Tl(I) respectively, which indicated very good accessibility of these metal ions towards chelating sites.

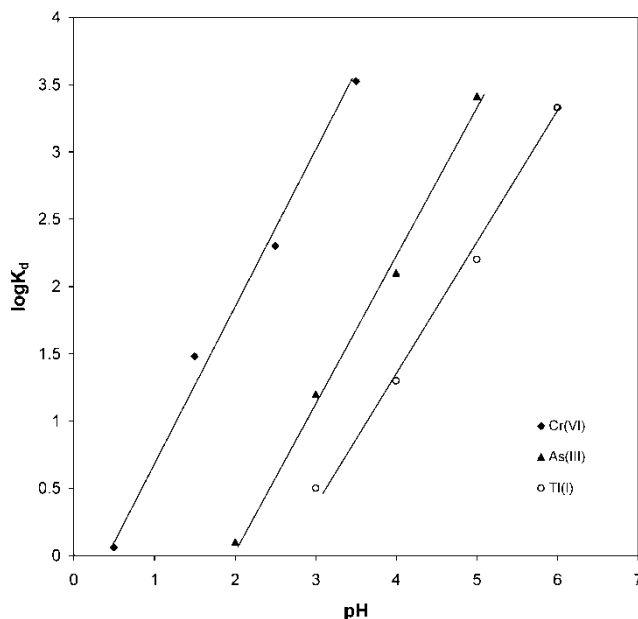


Figure 4. Plot of $\log K_d$ against pH for Sorption of Cr(VI), As(III) and Tl(I) on Resin 8. Amount of resin 8: 1 g; Volume of solution: 25 ml; Cr(VI): $453 \mu\text{g ml}^{-1}$; As(III): $623 \mu\text{g ml}^{-1}$; Tl(I): $1376 \mu\text{g ml}^{-1}$.

7. Breakthrough Studies

Breakthrough capacities are more significant and useful than total sorption capacities in ion exchange chromatographic applications as it gives actual working capacity of resin in column. Breakthrough capacity or effective capacity may be defined as capacity at moment when adsorbed component has appeared in effluent with an increased concentration. It is one of most critical parameters when working under dynamic condition. Breakthrough studies were carried out by taking 1 g resin **8** in column having 200 mm length and 8.0 mm internal diameter with bed height of 15 mm and passing $10 \mu\text{g ml}^{-1}$ of metal ions [Cr(VI), As(III) or Tl(I)] at their optimum pH and flow rate. An aliquot of 2.0 ml eluant was collected each time and analyzed by spectrophotometry and GF-AAS for metal ion content (Fig. 6). Breakthrough capacities of resin **8** for Cr(VI), As(III) and Tl(I) were found to be 2151, 2647 and $5503 \mu\text{g g}^{-1}$ respectively (Table 1). All curves were steep at their breakthrough points at different times, indicating possibility of separating these metal ions from their mixtures.

8. Preconcentration of Cr(VI), As(III) and Tl(I)

Concentration of trace elements in natural water is too low for their direct determination. Therefore, preconcentration or enrichment step is

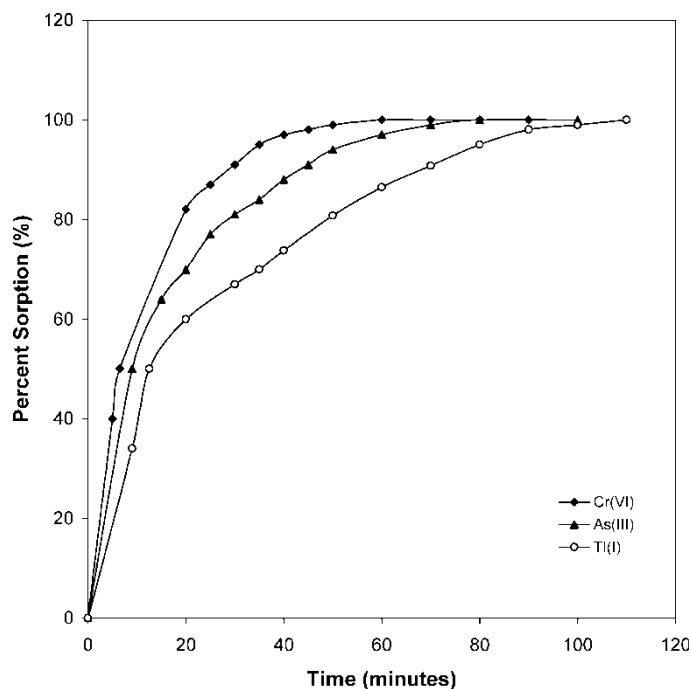


Figure 5. Exchange kinetics of Cr(VI), As(III) and Tl(I) on resin **8**. Amount of resin **8**: 1 g; Volume of feed solution: 100 ml; Cr(VI): $113 \mu\text{g ml}^{-1}$; pH 3.5; As(III): $156 \mu\text{g ml}^{-1}$; pH 5.0; Tl(I): $344 \mu\text{g ml}^{-1}$; pH 6.0.

unavoidable even with a sensitive detection method. Resin **8** was studied for column concentration of Cr(VI), As(III) and Tl(I) in terms of their pre-concentration factor (PF).

$$\text{PF} = \frac{\text{Concentration of metal in stripping solution}}{\text{Initial concentration of metal in feed solution}}$$

1000 ml solutions containing 6, 9, $6 \mu\text{g l}^{-1}$ Cr(VI), As(III) and Tl(I) at pH 3.5, 5.0, 6.0 respectively, were passed through column containing 1 g resin **8**. Preconcentrating ability of resin **8** was assessed from elution profile of metal ions by plotting concentration of effluents as a function of volume of stripping solutions: 8.5 ml, 0.1 N H_2SO_4 for Cr(VI); 11.1 ml, 0.5 N HCl for As(III); 9.5 ml, 1 N HNO_3 for Tl(I) respectively. Preconcentration factors were found to be 117, 90 and 105 for Cr(VI), As(III) and Tl(I) respectively with 98-99% recovery (Table 1 & 3, Fig. 7).

9. Resin **8** Stability Tests

Stability of resin **8** was tested to evaluate its sustainability for repeated cycles of sorption and desorption with different mineral acids. It was

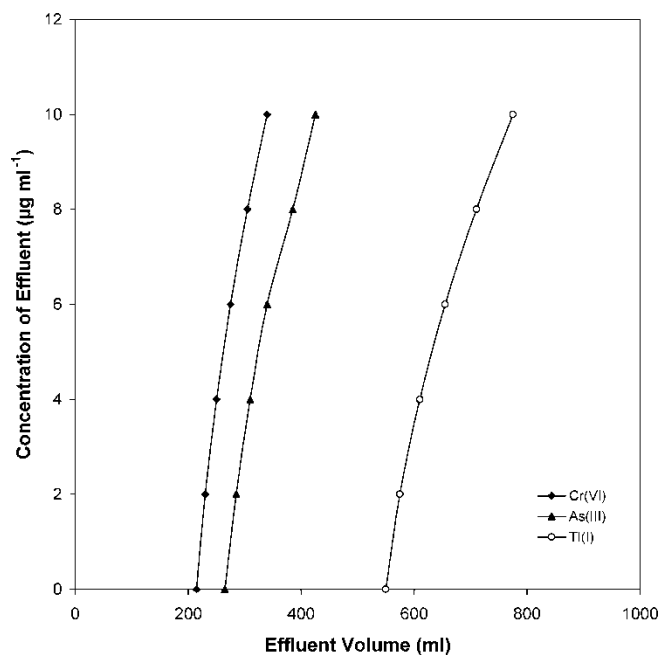


Figure 6. Breakthrough curves for Cr(VI), As(III) and Tl(I) on resin **8**. Amount of resin **8** in column: 1 g; Concentration of solution passed: $10 \mu\text{g ml}^{-1}$; Cr(VI): pH 3.5; As(III): pH 5.0; Tl(I): pH 6.0.

observed that there was no detectable change in physical and chemical properties of resin **8** in concentrated acidic solutions of 5.0 N HCl, 3.0 N HNO₃, and 3.5 N H₂SO₄. To check regenerating capacity of resin **8**, it was subjected to repeated sorption and desorption of Cr(VI), As(III), and Tl(I) at their optimum conditions in column. 1 g resin **8**

Table 3. Preconcentration factors for the Sorption of Cr(VI), As(III) and Tl(I) on resin **8**

Metal ions	Volume of solution passed (ml)	Concentration of solution passed ($\mu\text{g l}^{-1}$)	Volume of eluted solution (ml)	Recovery (%)	Preconcentration factor (PF)
Cr(VI)	1000	6	8.5	98.0	117
As(III)	1000	9	11.1	99.0	90
Tl(I)	1000	6	9.5	98.0	105

^aAverage of ten determinations.

Experimental Condition: Resin: 1 g; Cr(VI): pH 3.5; Elution by 0.1 N H₂SO₄; As(III): pH 5.0; Elution by 0.5 N HCl; Tl(I): pH 6.0; Elution by 1 N HNO₃.

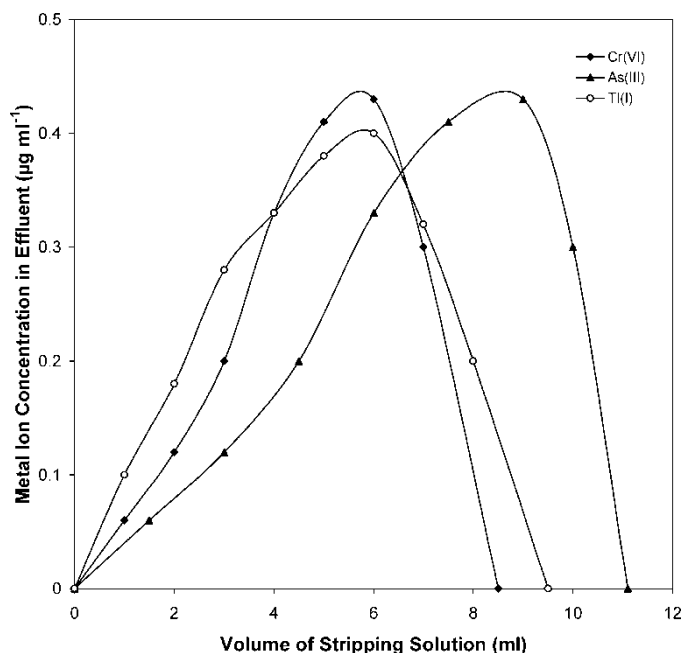


Figure 7. Elution Profile of Cr(VI), As(III) and Tl(I) on resin **8**. Amount of resin **8** in column: 1 g; Volume of solution passed: 1000 ml; Cr(VI): $6 \mu\text{g l}^{-1}$; pH 3.5; elution by 0.1 N H_2SO_4 ; As(III): $9 \mu\text{g l}^{-1}$; pH 5.0; elution by 0.5 N HCl; Tl(I): $6 \mu\text{g l}^{-1}$; pH 6.0; elution by 1 N HNO_3 .

was taken in column and 500 ml solution containing $100 \mu\text{g ml}^{-1}$ metal ions was passed through it at 30°C . Resin **8** was then stripped off metal ions with 25 ml of suitable eluant. Resin **8** showed very good stability up to 15 repeated cycles of sorption and desorption (Fig. 8), after which it showed 2–3% decrease in efficiency, which may be due to hydrolysis of resin **8** when treated with mineral acids.

10. Effect of Electrolytes and Associated Cations

Limit of tolerance of anions and cations on the sorption of Cr(VI), As(III) and Tl(I) is defined as that concentration which causes an error of 2–3% in the recovery of these metal ions. Effect of commonly associated cations in natural water samples such as Fe(II), Fe(III), Co(II), Ni(II), Mn(II), alkali [Li(I), Na(I), K(I), Rb(I), Cs(I)], and alkaline earth [Mg(II), Ca(II), Sr(II), and Ba(II)] had been studied under optimum conditions of metal ion sorption. Mn(II) did not affect sorption of Cr(VI), As(III) and Tl(I) up to 4 mg, however, for other cations tolerance limits were much higher (Table 4). Effect of anions and their limit of tolerance on sorption of Cr(VI), As(III) and Tl(I) by resin **8** was studied by taking different concentrations of some electrolytes like NaCl, NaF, NaBr, NaNO_2 , NaNO_3 , Na_2SO_4 , Na_3PO_4 , and CH_3COONa . Results (Table 5) showed that

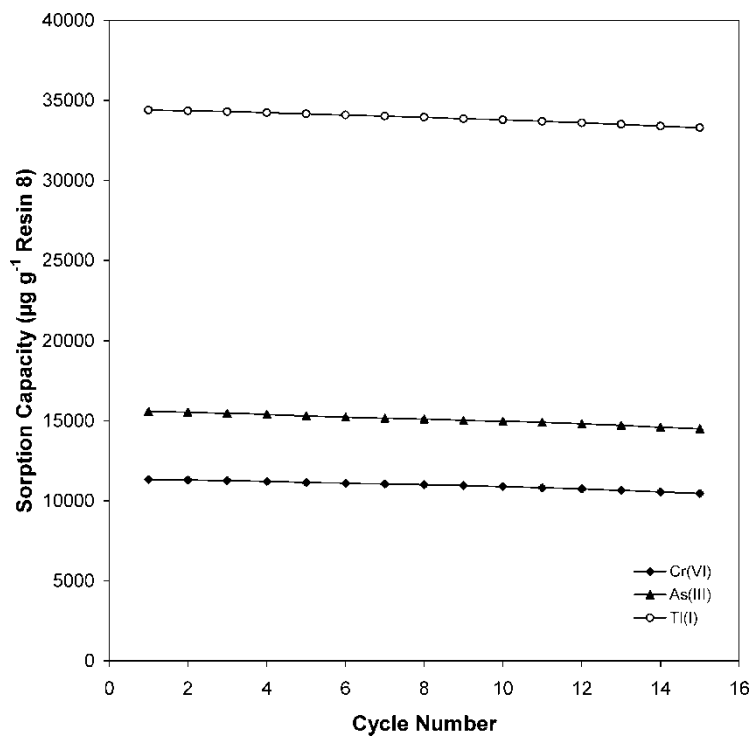


Figure 8. Evaluation of stability of resin 8 for Cr(VI), AS(III) and Tl(I) by Sorption and elution. Amount of resin **8** in column: 1 g; Volume of feed solution: 500 ml; Concentration of feed solution: 100 µg ml⁻¹; Cr(VI): pH 3.5; elution by 0.1 N H₂SO₄; As(III): pH 5.0; elution by 0.5 N HCl; Tl(I): pH 6.0; elution by 1 N HNO₃.

Table 4. Effect of commonly associated cations in water samples on the Sorption of Cr(VI), As(III) and Tl(I) on resin 8

Metal ions (100 µg each)	Interfering ions (mg)						Alkaline earth metal ions
	Fe(II)	Fe(III)	Co(II)	Ni(II)	Mn(II)	Alkali metal ions	
Cr(VI)	16	21	17	20	4	DNI	DNI
As(III)	14	17	16	15	4	DNI	DNI
Tl(I)	7	15	11	14	5	DNI	DNI

^aAverage of ten determinations. DNI: Do not interfere.
Experimental Condition: Resin: 1 g; Volume of solution passed: 100 ml; Cr(VI): pH 3.5; As(III): pH 5.0; Tl(I): pH 6.0.

Table 5. Tolerance limits of electrolytes on the Sorption of Cr(VI), As(III) and Tl(I) on resin 8

Metal ions (2.5 $\mu\text{g ml}^{-1}$)	Concentration of electrolytes (mol l^{-1})							
	NaF	NaCl	NaBr	NaNO ₂	NaNO ₃	Na ₂ SO ₄	Na ₃ PO ₄	CH ₃ COONa
Cr(VI)	1.5	2.3	2.5	3.0	2.5	1.3	0.2	2.5
As(III)	2.0	3.0	2.0	2.1	2.8	1.2	0.3	2.3
Tl(I)	1.2	2.0	1.4	2.2	3.0	1.7	0.4	2.1

^aAverage of ten determinations.

Experimental condition: Resin: 1 g; Volume of solution passed: 100 ml; Cr(VI): pH 3.5; As(III): pH 5.0; Tl(I): pH 6.0.

except Na₃PO₄, all other electrolytes did not interfere between 1.0-3.0 M concentration range, which further augments potential application of resin 8 for analysis of real samples.

Separation of Cr(VI), As(III) and Tl(I)

Resin 8 was found useful for separating chromium, arsenic and thallium from mixture due to favourable kinetics and different breakthrough capacities in column. Resin 8 was tested for simultaneous separation of their ternary mixture. Hence, their mixture (each 100 μg in 25 ml buffer solution) was passed through column at optimum pH and flow rate. Column effluents were analyzed for metal ions by spectrophotometry and GF-AAS. In separation of their ternary mixture containing 100 μg each of Cr(VI), As(III), and Tl(I) was passed through column maintained at pH 3.0 at flow rate of 1.0 ml min^{-1} . At this pH, most of As(III) and Tl(I) came out with effluent while Cr(VI) was retained in column. Cr(VI) was eluted with 0.1 N H₂SO₄. Small amounts of As(III) and Tl(I) ($\leq 2\%$) which were sorbed by resin 8 were then separated on the basis of selective elution with different eluting agents. As(III) was eluted first with 0.5 N HCl followed by Tl(I) with 1 N HNO₃. Effluent containing As(III) and Tl(I) was then passed through a fresh column maintained at pH 5.5. Both metal ions were retained in column and As(III) was desorbed first with 0.5 N HCl followed by Tl(I) with 1 N HNO₃. Thus, quantitative separation was achieved as seen in separation pattern (Fig. 9).

Analytical Performance (Calibration Graph, Detection Limit and Precision)

Standard stock solutions of these elements were used after appropriate dilution for preparation of calibration standards for Cr(VI), As(III), and Tl(I). Precision of proposed method was examined based on optimum

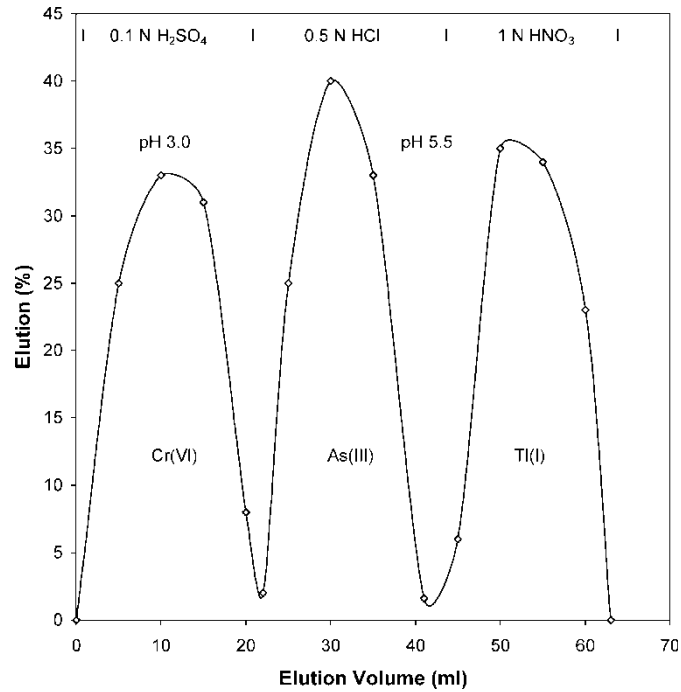


Figure 9. Separation of Cr(VI), As(III) and Tl(I) on Resin 8. Amount of resin 8: 1 g; Column maintained first at pH 3.0 and then at pH 5.5; Cr(VI): 100 µg in 25 ml buffer; As(III): 100 µg in 25 ml buffer; Tl(I): 100 µg in 25 ml buffer.

condition for preconcentration in column and expressed as R.S.D.; 1.3% for Cr(VI), 1.2% for As(III), and 1.1% for Tl(I). Limit of detection (LOD) was defined as that analyte concentration giving a signal equal to three times standard deviation of blank signal (42). Limit of quantification (LOQ) was then set as ten times standard deviation of blank signal (Table 6).

Table 6. Regression analysis for the calibration graph, linear range, detection limit and precision

Metal Ions	Linear range (µg ml ⁻¹)	Regression equation	Correlation coefficient	LOD (µg l ⁻¹)	LOQ (µg l ⁻¹)	R.S.D. ^a (%)
Cr(VI)	0.010–3.03	A = C × 0.599 – 0.0011	1.0010	5.83	19.43	1.3
As(III)	0.065–7.20	A = C × 0.201 – 0.0076	0.9979	8.48	28.26	1.2
Tl(I)	0.023–4.90	A = C × 0.3469 – 0.0006	1.0110	5.56	18.56	1.1

^aTen determinations. A = Absorbance, C = Concentration (µg ml⁻¹).

Table 7. Determination of Cr(VI), As(III) and Tl(I) in natural water samples on resin 8

Sample	Method	Cr(VI)		As(III)		Tl(I)	
		Amount ($\mu\text{g l}^{-1}$)	R.S.D. ^a (%)	Amount ($\mu\text{g l}^{-1}$)	R.S.D. ^a (%)	Amount ($\mu\text{g l}^{-1}$)	R.S.D. ^a (%)
Sabarmati River, Near Thermal Power Station, Ahmedabad	Present method	42	1.0	39	1.1	25	1.1
	Standard addition	43	1.1	38	1.2	24	1.3
Ground Water, University Area, Ahmedabad	Present method	20	1.4	29	1.0	19	1.3
	Standard addition	19	1.2	30	1.2	20	1.1
Ground Water, Vatva Industrial Zone, Ahmedabad	Present method	94	1.2	67	1.0	40	1.2
	Standard addition	96	1.3	69	1.1	39	1.2

^aTen determinations.

Experimental condition: Resin: 1 g; Sample volume: 1000 ml.

Table 8. Determination of Cr(VI), As(III) and Tl(I) in Standard reference materials (SRM) of NIST on resin **8**

Sample	Metal ion concentration ^a found by GF-AAS		
	Cr(VI) ($\mu\text{g g}^{-1}$)	As(III) ($\mu\text{g g}^{-1}$)	Tl(I) ($\mu\text{g g}^{-1}$)
SRM 361 (Low alloy steel)	6937.5 (6940)	168.9 (170)	—
SRM 2711 (Montana soil)	47.2 (47)	104.1(105)	2.39 (2.47)
SRM 1573a (Tomato leaves)	1.99 (1.99)	0.111 (0.112)	—
SRM 1577b (Bovine lever)	—	0.049 (0.047)	—

(Certified Values in the Brackets)
^aThree determinations.
Experimental condition: Resin: 1 g; Sample: 1 g.

Application to Analysis of Water Samples

Validity of system was established by subjecting resin **8** to natural water samples and SRMs procured from NIST. Results obtained for water samples by proposed method were compared with that of standard addition method. Natural water samples and aqueous samples of reference standards were prepared as per methods described in experimental section and then passed through column as explained in the general procedure for column concentration. SRMs were decomposed in mixture of acids as described in literature (30). Amount of metal ions present in water samples and their R.S.D. values are given in Table 7 and method validation with respect to NIST standards has been given in Table 8. Higher efficiency of separation curves with lower R.S.D., suggests utility of resin **8** for various analytical applications in determination of Cr(VI), As(III) and Tl(I).

CONCLUSION

Polymer supported calixarenes have established themselves with prospective ionophoric properties that can be used for varied analytical applications. Resin **8** appears capable for trace enrichment of metal ions as noticeable from column studies. Separation curves exhibit efficiency of resin **8** for real-time separation of Cr(VI), As(III) and Tl(I) in presence of each other with minimum interference from alkali and alkaline earth metal ions. Resin **8** shows high separating and preconcentration ability for these metal ions compared to other preconcentration methods (31–45) reported in literature as summarized in Table 9. Resin **8** can be successfully regenerated for 15

Table 9. Summary of preconcentration methods reported for arsenic, chromium, and thallium

Metal ions	Preconcentration procedure	Analytical technique	Preconcentration factor	Detection limit	Application	References
As(III) & As(V)	Cloud Point Extraction (Triton X-114)	ET-AAS	52.5	0.01 ppm	Tap water and biological samples	(31)
As(III) & As(V)	Anion Exchange Resin	ICP-AES	—	0.1 ppm	Fresh water samples	(32)
As(III) & As(V)	Co-precipitation	ICP-AES	70	0.02 ppm	Water samples	(33)
As(V)	Solid Adsorbent	Potentiometric flow analysis	—	30.6 ppm	Ground water samples	(34)
As(III)	PTFE knotted reactor	ET-AAS	44	0.008 ppm	Sea water and Reference material (NASS-4)	(35)
Cr(III) & Cr(VI)	On-line flow injection	Derivative F-AAS	—	0.244 & 0.235 ppm	Environmental water samples	(36)
Cr(VI)	Dowex-1 Oxalate form	ET-AAS	25	0.027 ppb	Potable water samples	(37)
Cr(III) & Cr(VI)	On-line flow injection	ICP-OES	70	0.029 ppb	Parental solutions	(38)
Cr(III) & Cr(VI)	Zirconium(IV) Phosphate Immobilized on Silica gel	F-AAS	20.8 & 24.9	1.9 & 2.3 ppm	Water samples	(39)
Cr(III)	Saccharomyces Carlsbergensis immobilized on amberlite XAD-4	F-AAS	—	7.4 ppb	Water samples	(40)

(continued)

Table 9. Continued

Metal ions	Preconcentration procedure	Analytical technique	Preconcentration factor	Detection limit	Application	References
Cr(III)	Bentonite modified with trioctylamine	F-AAS	—	1.7 ppm	Waste water samples	(41)
Cr(III)	Agrobacterium tumefaciens immobilized on amberlite XAD-4	F-AAS	25	3.6 ppb	Water samples, alloys, infant foods and standard reference materials	(42)
Cr	Nanometer-sized alumina	ICP-AES	5	—	Environmental reference materials	(43)
Tl(III)	Polyurethane foam	Platform-GF-AAS	—	0.05 ppm	Urine	(44)
Tl	Chelating resin	ICP-MS	20	—	Sea water samples	(45)
Cr(III), As(III) & Tl(I)	Merrifield resin supported calix[4]arene derivative	GF-AAS	90,117 & 105	8.48, 5.83 & 5.56 ppb	Natural water samples & environmental, biological & geological reference materials	Present method

cycles of operation in column. Results obtained from analysis of natural water samples and SRMs prove reliability of proposed method for its application to a range of samples for their determination by GF-AAS.

ACKNOWLEDGEMENT

Authors are thankful to GUJCOST, Gujarat for financial assistance. Authors also acknowledge CDRI, Lucknow for instrumental facilities and INFLIBNET, Ahmedabad for e-journals.

REFERENCES

1. Clayton, G.D. and Clayton, F.E. (1994) *Patty's Industrial Hygiene and Toxicology*. Wiley-Interscience: New York; Vol. 2C.
2. Patai, S. (1994) *The Chemistry of Organic Arsenic, Antimony and Bismuth Compounds*; John Wiley & Sons: New York.
3. Nriagu, J.O. and Nieboer, E. (1988) *Chromium in the Natural and Human Environments*; John Wiley & Sons: New York.
4. Browning, E. (1969) *Toxicity of Industrial Metals*, 2nd Ed.; Appleton-Century-Crofts: New York, 317.
5. Yilmaz, M. and Cheremisinoff, N.P. (1997) Solution state metal complexes of calixarenes and polymeric calixarenes. In *Handbook of Engineering Polymeric Materials*; Marcel Dekker Inc.: New York.
6. Gutsche, C.D. and Stoddart, J.F. (1998) *Calixarenes Revisited, Monographs in Supramolecular Chemistry*. Royal Society of Chemistry: Cambridge.
7. Asfari, Z., Bohmer, V., Harrowfield, J., and Vicens, J. (2001) *Calixarenes 2001*; Kluwer Academic Press: Dordrecht.
8. Mandolini, L. and Ungaro, R. (2000) *Calixarenes in Action*; Imperial College Press: London.
9. Mohammed-Ziegler, I. (2003) Selective complex formation of some chromogenic calix[4]arene derivatives detected by fourier transforms infrared spectroscopy 2. Solid experiments. *Spectrochim. Acta, Part A: Mole. Biomol. Spectroscopy*, 59A: 3239.
10. Matthews, S.E., Rees, N.H., Felix, V., Drew, M.G.B., and Beer, P.D. (2003) Thallium-cations complexation by calix[4]tubes: ^{205}Tl NMR and X-ray Evidence. *Inorg. Chem.*, 42: 729.
11. Vela, N.P., Heitkemper, D.T., and Stewart, K.R. (2001) Arsenic extraction and speciation in carrots using accelerated solvent extraction, liquid chromatography and plasma mass spectrometry. *Analyst*, 126: 1011.
12. Lenoble, V., Deluchat, V., Serpaud, B., and Bollinger, J. (2003) Arsenite oxidation and arsenate determination by the molybdene blue method. *Talanta*, 61: 267.
13. Vaisanen, A., Suontamo, R., and Rintala, J. (2002) Control of matrix interferences by the multiple linear regression model in the determination of arsenic, antimony and tin in lead pellets by inductively coupled plasma emission spectrometry. *J. Anal. At. Spectrom.*, 17: 274.
14. Heitkemper, D.T., Vela, N.P., Stewart, K.R., and Westphal, C.S. (2001) Determination of total and speciated arsenic in rice by ion chromatography and inductively mass spectroscopy. *J. Anal. At. Spectrom.*, 16: 299.

15. Akl, M.A.A., Kenawy, I.M.M., and Lasheen, R.R. (2004) Organically modified silica gel and flame atomic absorption spectrometry: employment for separation and preconcentration of nine trace metals for their determination in natural aqueous system. *Microchem. J.*, In Press.
16. Wangkarn, S. and Pergantis, S.A. (2000) High-speed separation of Arsenic compounds using narrow-bore high performance liquid chromatography on-line with inductively coupled plasma mass spectrometry. *J. Anal. At. Spectrom.*, 15: 627.
17. Caruso, J.A., Heitkemper, D.T., and Hymer, C.B. (2001) An evaluation of extraction techniques for arsenic species from freeze-dried apple samples. *Analyst*, 126: 136.
18. Gallagher, P.A., Schwegel, C.A., Wei, X., and Creed, J.T.J. (2001) *Environ. Monitoring*, 3: 371.
19. Shraim, A., Chiswell, B., and Olszowy, H. (2000) Use of perchloric acid as a reaction medium for speciation of arsenic by hydride generation atomic absorption spectrometry. *Analyst*, 125: 949.
20. Munoz, O., Velez, D., Montoro, R., Arroyo, A., and Zamorano, M. (2000) Determination of inorganic arsenic [As(III) + As(V)] in water samples by microwave assisted distillation and hydride generation atomic absorption spectrometry. *J. Anal. At. Spectrom.*, 15: 711.
21. Yan, X., Yin, X., He, X., and Jiang, Y. (2002) Flow injection on-line sorption preconcentration coupled with hydride generation atomic fluorescence spectrometry for determination of ultratrace amounts of arsenic(III) and arsenic(V) in natural water samples. *Anal. Chem.*, 74: 2162.
22. Garg, B.S. and Jain, V.K. (1988) Analytical application of thiosemicarbazone and semicarbazone. *Microchem. J.*, 38: 144.
23. Jain, V.K., Sait, S.S., Shrivastav, P., and Agrawal, Y.K. (1997) Application of chelate forming resin amberlite XAD-2-o-vanillinthiosemicarbazone to the separation and preconcentration of Cu(II), Zn(II) and Pb(II). *Talanta*, 45: 397.
24. Jain, V.K., Pandya, R.A., Pillai, S.G., Shrivastav, P.S., and Agrawal, Y.K. (2004) Application of a chelate forming calix[4]arene-o-vanillinthiosemicarbazone resin to the separation, preconcentration and Trace determination of Cu(II), Cd(II) and Pb(II) in natural water samples. *Microchim. Acta*, 147: 253.
25. Weissberger, A., Proshauer, E.S., Riddick, J.A., and Toops, E.E., Jr. (1955) *Techniques of Organic Chemistry*; Interscience: New York; Vol. VII.
26. Marczenko, Z. (1976) *Spectrophotometric Determination of Elements*; Ellis-Horwood Ltd.: Chichester.
27. Jeffery, G.H., Bassett, J., Mendham, J., and Denny, R.C. (1989) *Vogel's Textbook of Chemical Analysis; 5th Ed.*; Wiley: New York.
28. Dean, J.A. (1999) *Lange's Handbook of Chemistry; 15th Ed.*; McGraw-Hill: New York.
29. Gidwani, M.S., Menon, S.K., and Agrawal, Y.K. (2002) Chelating Polycalixarene for the chromatographic separation of Ga(III), In(III) and Tl(III). *React. Funct. Polym.*, 53: 143.
30. Lajunen, L.H.J. (1992) *Spectrochemical Analysis by Atomic Absorption and Emission*; Royal Society of Chemistry: Cambridge, 9.
31. Shemirani, F., Baghdadi, M., and Ramezani, M. (2005) Preconcentration and determination of ultra trace amounts of arsenic(III) and arsenic(V) in tap water and total arsenic in biological samples by cloud point extraction and electrothermal atomic absorption spectrometry. *Talanta*, 65 (4): 882.

32. Jitmanee, K., Oshima, M., and Motomizu, S. (2005) Speciation of arsenic (III) and arsenic (V) by inductively coupled plasma-atomic emission spectrometry coupled with preconcentration system. *Talanta*, 66 (3): 529.
33. Kim, C-G. and Pak, Y. (2004) Continuous co precipitation preconcentration-hydride generation for arsenic in inductively coupled plasma atomic emission spectrometry. *J. Korean. Chem. Soc.*, 48 (8): 585.
34. Rodriguez, J.A., Barrado, E., Vega, M., and Lima, J.L.F.C. (2005) Speciation of inorganic arsenic in waters by potentiometric flow analysis with on-line preconcentration. *Electro analysis*, 17 (5–6): 504.
35. Herbello-Hermelo, P., Barciela-Alonso, M.C., Bermejo-Barrera, A., and Bermejo-Barrera, P. (2005) Flow on-line sorption preconcentration in a knotted reactor coupled with electrothermal atomic absorption spectrometry for selective as(III) determination in sea-water samples. *J. Anal. At. Spectrom.*, 20 (7): 662.
36. Sun, H.-W., Kang, W.-J., Ha, J., Liang, S.-X., and Shen, S.-G. (2004) Determination of Cr(III) and Cr(VI) in environmental waters by derivative flame atomic absorption spectrometry using flow injection on-line Preconcentration with double-micro column adsorption. *J. Iranian Chem. Soc.*, 1 (1): 40.
37. Sahayam, A.C., Venkateswarlu, G., and Chaurasia, S.C. (2005) Determination of Cr(VI) in potable water samples after selective preconcentration on oxalate form of dowex-1 and electrothermal atomic absorption spectrometric determination. *Anal. Chim. Acta*, 537 (1–2): 267.
38. Gil, R.A., Cerutti, S., Gasquez, J.A., Olsina, R.A., and Martinez, L.D. (2005) On-line preconcentration and determination of chromium in parental solutions by inductively coupled plasma optical emission spectrometry. *Spectrochim. Acta, Part B: At. Spectrom.*, 60B (4): 531.
39. Maltez, H.F. and Carasek, E. (2005) Chromium speciation and preconcentration using Zirconium(IV)phosphate chemically immobilized onto silica gel surface using a flow system and F-AAS. *Talanta*, 65 (2): 537.
40. Baytak, S. and Tuerker, A.R. (2005) Determination of Iron(III), Cobalt(II) and Chromium(III) in various water samples by flame atomic absorption spectrometry after preconcentration by saccharomyces carlsbergensis immobilized on Amberlite XAD-4. *Microchim. Acta*, 149 (1–2): 109.
41. Aktas, Y.K. and Ibar, H. (2005) Determination of chromium, copper, manganese, nickel and zinc by flame atomic absorption spectrometry after separation of bentonite modified with trioctylamine. *J. Ind. Chem. Soc.*, 82 (2): 134.
42. Baytak, S. and Tuerker, A.R. (2005) The use of agrobacterium tumefaciens immobilized on amberlite XAD-4 as a new biosorbent for the column preconcentration of Iron(III), Cobalt(II), manganese(II) and chromium(III). *Talanta*, 65 (4): 938.
43. Yin, J., Jiang, Z., Chang, G., and Hu, B. (2005) Simultaneous on-line preconcentration and determination of trace metals in environmental samples by flow injection combined with inductively coupled plasma mass spectrometry using a nanometer-sized alumina packed micro-column. *Anal. Chim. Acta*, 540 (2): 333.
44. Meng, Y., Zhang, K., and Ye, B. (2004) Platform-GFAAS determination of trace thallium in urine after preconcentration by polyurethane foam. *Zhongguo Weisheng Jianyan Zazhi*, 14 (4): 424.
45. Zhu, Y., Itoh, A., and Haraguchi, H. (2005) Multielement determination of trace metals in seawater by ICP-MS using a chelating resin-packed minicolumn for Preconcentration. *Bull. Chem. Soc. Jpn.*, 78 (1): 107.