Speciation of Chromium Metal Ions by RP-HPLC

Varinder Kaur and **Ashok Kumar Malik***
Department of Chemistry, Punjabi University, Patiala-147 002, Punjab, India

Abstract

A high-performance liquid chromatographic method with UV detection, based on chelation with morpholine-4-carbodithioate (MDTC), has been developed for the speciation of chromium. Simultaneous preconcentration of Cr(III) and Cr(VI) in aqueous solutions was achieved by extraction of metal species in chloroform as MDTC complexes. The separation of MDTC chelates of Cr(III) and Cr(VI) was realized on a C18 column with the use of acetonitrile–water (70:30) at a flow rate of 1.0 mL/min. The Cr(III) showed a single peak in chromatogram due to $Cr(MDTC)_3$, and Cr(VI) showed two peaks due to the formation of $Cr(MDTC)_3$ and $Cr(MDTC)_2$(OMDTC). These two complexes were separated with different retention times at 5.4 and 4.3 min, respectively. The developed method can be used for rapid routine determination of chromium species with high precision and reliability. The method was validated by applying to various synthetic binary mixtures prepared by spiking Cr(III) and Cr(VI) in tap water.

Introduction

Toxicity, mobility, and bioavailability differ greatly between the two chemical species of chromium: Cr(III) and Cr(VI). The inorganic trivalent form of chromium is relatively nontoxic and is essential to maintain a glucose tolerance factor in the human body. Whereas Cr(VI) is introduced into the environment by various industries such as plastics, metal working dyes, ink, and paint industries, it is known to be carcinogenic and toxic. The toxic nature of Cr(VI) must be attributed to its high oxidation potential and relatively small size, which enables them to penetrate cell membranes. On the other hand, Cr(III) forms aqueous complexes and is unable to penetrate cell walls due to its large size (1). Measuring only the total concentration of chromium in environmental samples is crucial for the evaluation of toxicological effects, bioavailability, and monitoring. Therefore, the development of speciation techniques with high sensitivity and selectivity is an important challenge to study, because of the ambivalent nature of chromium.

Recently, selective determination of both species can be achieved successfully by using high-performance liquid chromatography (HPLC)–integrated coupled plasma–mass spectrometry (ICP-MS) (2), ion chromatography–ICP-MS (3), flame atomic absorption spectrophotometry (AAS) (4), capillary electrophoresis (5), and ICP-AES (6). ICP-MS is a promising technique used successfully with chromatographic techniques, but a major limitation generally arises from the occurrence of non-spectral as well as spectral interferences, especially when elements like C and Cl are present in the sample or eluent. It may be a severe limitation. The technique AAS is sufficiently reliable and sensitive, but it is very expensive, time consuming, and require expertise to handle flame AAS. Other techniques such as ICP-AES and CE offer limitations due to instrumental price and are very sophisticated. Recently, liquid–liquid extraction (7) and solid-phase extraction (8,9) techniques hare been used for the extraction of chromium speciation. HPLC is a technique that meets most of the analytical requirements of metal determination as it provides separation, identification, and quantification of different species down to trace levels in one procedure.

Morpholine-4-carbodithioate (MDTC) forms a water insoluble complex with metal ions when it is used as a pre-column derivatization reagent. The metal complexes precipitate as colloidal particles. Thus, the precipitates of chromium complex with MDTC have been extracted into an organic solvent such as chloroform prior to the chromatographic separation. Dithiocarbamates, such as Diehtyldithiocarbamate (10) and Ammonium pyrrolidinedithiocarbamate (11), are reported as complexing reagents for the solvent extraction and HPLC–UV determination. According to literature, Cr(III) metal ions form $Cr(DTC)_3$ [Dithiocarbamate (DTC)] complex with dithiocarbamates and Cr(VI) forms two different complexes, $Cr(DTC)_3$ and $Cr(DTC)_2$(ODTC) complex, where the Cr-O-S bond is formed during the reduction of the Cr(VI) complex by the ligand. MDTC has similar behavior of chelation and forms $Cr(MDTC)_3$ complex with Cr(III). It can be used for the speciation of Cr(III) and Cr(VI) using HPLC–UV after extraction of complexes in chloroform. It is also intended to extend the application of dithiocarbamates for chromatographic determination of Cr(III) and Cr(VI). In this method, morpholine-4-carbodithioate is used as a reagent as it is highly soluble in water, whereas commercially available ammonium pyrrolidinedithiocarbamate needs further purification. The purification of reagent lengthens the method, as it is done by the extraction of reagent in chloroform before use, as reported by Wang et al. (11). There is no need to extract when using MDTC as a reagent. This method is more precise, accurate, and simple, with very low detection limits. The reagent used in this method can be prepared by a simple method. It is more stable in acidic media and complexation with chromium is fast. The method is fast and good recoveries were found for synthetic mixtures using this method. Special attention is given to optimize the parameters, such as the effect of ligand concentration, effect of pH, effect of temperature, shaking, extraction time, effect of mobile phase composition, and diverse ion interferences.

* Author to whom correspondence should be addressed: email malik_chem2002@yahoo.co.uk.

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Experimental

Reagents

All the solvents used were of HPLC-grade and purchased from J.T. Baker Chemicals (Phillipsburg, NJ), and filtered by using Nylon 6,6-membrane filters (Rankem, India) in filtration assembly (from Perfit, India). Cr(III) nitrate nonahydrate, potassium dichromate, sodium acetate trihydrate, acetic acid glacial, and sodium hydroxide were obtained from Merck (India). All HPLC solvents were degassed with an ultrasonic bath prior to use. MDTC reagent was prepared in the lab by using the method reported by Macrotrigiano et al. (12).

Synthesis of MDTC and complex formation

MDTC was used as the complexing agent. It was prepared by the method reported by Macrotrigiano et al. (12). Morpholine was taken in dry ether in a flask, and to this, a stoichiometric amount of CS_2 was added with constant stirring and cooled in an ice-salt mixture. To this solution, a stoichiometric amount of KOH was added dropwise with constant stirring in 3–4 h. The product was filtered, washed with solvent ether, and recrystallised from isopropylalcohol. The obtained product was in the form of a white needle shaped crystals with m. pt. 175°C and was highly soluble in water. IR studies showed characteristic bands for C=S str at 1083 cm^{-1} and two bands at 1437 and 1164 cm^{-1} due to υ(C-N) and υ(C-O), respectively.

Apparatus

The Dionex HPLC unit consisted of the following components; a P 680 solvent delivery pump, an UVD 170 detector capable of detecting at four wavelengths, interfaced to a computer loaded with Chromeleon software (version 6.70), in conjunction with HP laser 1010 printer. The sample was injected into the injection valve, having volume of 20 µL. A Dionex Acclaim 120 C18 stainless steel reversed-phase column of 4.6 mm (i.d.) × 250 mm filled with C18 material with 5 µm packing (Acclaim 120) was used for separation. The temperature of column was maintained at 25°C. The peak area obtained from quantification with Chromeleon software was used to prepare the calibration graph. The IR spectra were recorded on FTIR (PerkinElmer). Elico SL-164 double beam UV-vis spectrophotometer loaded with Spectra Treatz software and interfaced to a computer in conjunction with an HP Laserjet 1010 printer was used to record the spectra. A Century CP 901 Digital pH meter was used to adjust the pH, and 1 cm quartz cuvettes were utilized for absorbance studies.

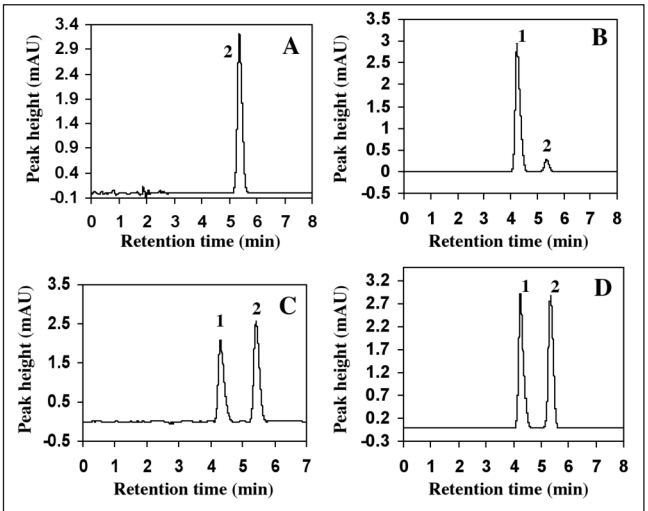

Figure 1. Chromatogram showing peaks for Cr(III) = 50 ng (A); Cr(VI) = 50 ng (B); Cr(III) and Cr(VI) = 35 ng (C) [MDTC = 1 mL of 10%, sodium acetate–acetic acid buffer (pH 4) = 1 mL, extracted with chloroform at 55°C with 20 min shaking]; Cr(VI) = 100 ng (D) [MDTC = 1 mL of 10%, sodium acetate–acetic acid buffer (pH 4) = 1 mL, extracted at 25°C with 120 min shaking]. Peak 1: $Cr(MDTC)_2(OMDTC)$ and Peak 2: $Cr(MDTC)_3$.

Procedure

Chelation, extraction, and separation of complexes

A solution containing, 0.5–250 ng of Cr(III) and Cr(VI) was transferred to a well stoppered polythene bottle and 1 mL of 10% MDTC solution (in water) was added. The pH of the solution was adjusted to 4.0 by adding 1 mL of sodium acetate–acetic acid buffer. The total volume of the solution was made equal to 25 mL. To this solution, 5 mL of chloroform was added, and the sample was placed in a water bath with temperature controlled at 55°C. The solution was shaken with wrist action for 20 min to mix the layers. After the phase separation, the organic layer was separated and placed in flask with a stopper. The solution was washed with 5 mL of triple distilled water two times. The chloroform was evaporated near to dryness in a sand bath with temperature controlled at 55°C. The dry sample was dissolved in acetonitrile and the volume was made up to 5 mL for HPLC analysis.

The solution was injected in the injection loop (with 20 µL injection volume) of HPLC and separated onto a C18 column. The temperature of the column was maintained at 25°C. The complexes were eluted with acetonitrile–water (70:30) using a flow rate of 1 mL/min at a wavelength of 320 nm. The chromatograms for Cr(III), Cr(VI), and their mixture are shown in Figure 1.

Results and Discussions

The chromium species Cr(III) and Cr(VI) were reacted with MDTC at pH 4.0 and detected by HPLC with UV detection at 320 nm. It is apparent from the chromatograms obtained from chromium species that separation is good and results are comparable to the methods reported in literature (10,11). It was observed by thin layer chromatography (TLC), UV–vis, and IR spectra that Cr(III) formed a single product, $Cr(MDTC)_3$ and Cr(VI) was reduced by reagent to give $Cr(MDTC)_2(OMDTC)$ in which oxygen is inserted in Cr-S bond. It is in agreement with the literature (11). The structures are proposed and shown (Figure 2).

TLC with methanol–petroleum ether–toluene (5:70:25) showed one spot for Cr(III)-MDTC complex at R_f value 0.143 due to the formation of $Cr(MDTC)_3$ and two spots for Cr(VI)-MDTC complexes with R_f values at 0.143 and 0.62. The IR spectrum of complex $Cr(MDTC)_2(OMDTC)$ showed absorption bands at 879.2 cm^{-1} due to S-O single bond stretching vibration, which is not observed for $Cr(MDTC)_3$ complex (13). This means two complexes are formed for Cr(VI) having the same product as the

product formed with Cr(III); that is, $Cr(MDTC)_3$. The two products are well separated by HPLC at different retention time. The second product is $Cr(MDTC)_2(OMDTC)$, as shown by FT-IR and reported in literature for other dithiocarbamates.

Optimization of extraction condition

Effect of ligand concentration

The effect of ligand concentration on complex formation was studied. The ligand concentration was varied from 1–5%. Incomplete extraction was observed at 1% and 1.5%. The results indicated complete extraction of metal at 2% and greater concentrations. It was observed constant up to 4% of reagent. For further studies, the concentration of the ligand was kept 2%.

Effect of pH

The effect of pH was observed by varying the pH of the solution from 2 to 6. Maximum complex formation was observed in pH 2.5 to 5. The pH was adjusted by using sodium acetate (0.02 M)–acetic acid buffer of pH 4, as the complex is more stable at this pH. The buffer concentration of 1% was sufficient to control the pH of solution.

Effect of shaking and temperature on extraction of metal complexes

Effect of shaking time and temperature was observed on Cr(III)–MDTC and Cr(VI)–MDTC complexes. It was observed that both metal complexes could be extracted in chloroform after shaking the sample up to 20 min. The effect was studied by varying the extraction time (Figure 3). At 30°C, the complex was recovered up to 70% in ~ 2 h for both species. As the temperature was increased to 55°C, both complexes were recovered up to 95% in ~ 20 min of shaking. Increase in temperature improved the extraction efficiency and reduced extraction time, but the complex formation in case of Cr(VI) is greatly affected by extraction time and temperature. In case of Cr(VI), two complexes are formed in different ratios at different temperature and shaking time. It was optimized by varying temperature and extraction time. The ratio was determined by repeating the same experiment five times. The ratio varies from 9.3–9.6, and the average was calculated. The ratio of $Cr(MDTC)_2(OMDTC)$–$Cr(MDTC)_3$ for Cr(VI) was found to be 9.4 after shaking the sample for 20 min at 55°C. At lower temperatures (25°C and 30°C) the ratios were found to be very low (Figure 4). We concluded that the maximum $Cr(MDTC)_2(OMDTC)$ complex was recovered at 20 min extraction with 95% recovery. The peak areas were measured and recoveries were calculated for both metal complexes at different concentrations.

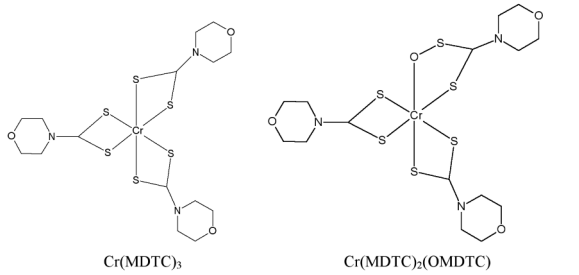

Figure 2. The structures of $Cr(MDTC)_3$ and $Cr(MDTC)_2(OMDTC)$.

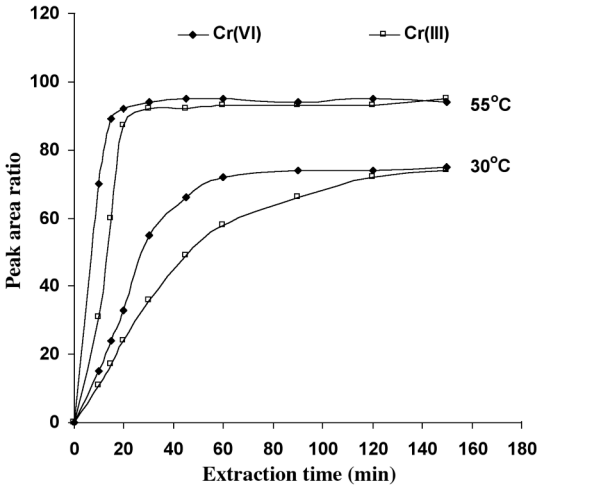

Figure 3. Recovery versus the extraction time: Cr(III)–MDTC and Cr(VI)–MDTC at pH 4 and 55°C and 30°C.

Effect of mobile phase composition

It is important to choose a mobile phase to optimizing the separation of the chromium species. Selection of the mobile phase depends upon resolution of peaks as well as signal response and separation time. The results obtained showed that complexes are more strongly retained with increasing amounts of water. But increase in concentration of acetonitrile has greater influence on the retention time and peak shape. The ratio of water and acetonitrile was varied. The results showed best resolution at 70:30 (acetonitrile–water) at a flow rate of 1 mL/min. So, this composition was used throughout the experiment for obtaining good separation. The peaks were obtained at a retention time of 4.3 and 5.4 min for $Cr(MDTC)_2(OMDTC)$ and $Cr(MDTC)_3$, respectively.

Characteristics of chromium determination

The calibration curve was prepared at different concentrations by measuring peak areas (Figure 5). The ratio of both products

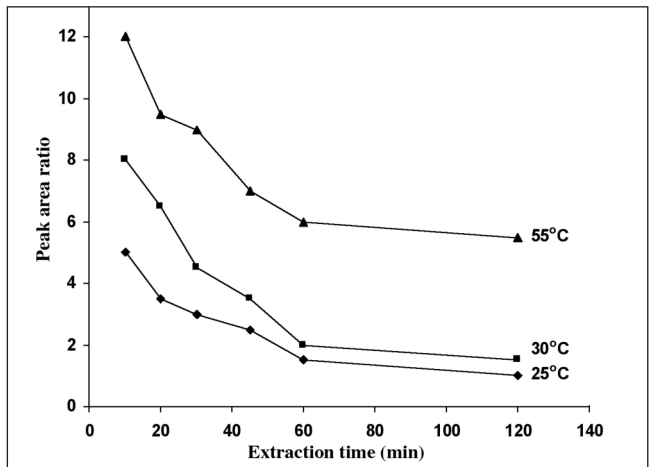

Figure 4. Peak area ratio versus extraction time, Cr(VI) + MDTC, extracted into $CHCl_3$ at 55°C, 30°C, and 25°C.

generated from Cr(VI) and Cr(III) showed a linear relation over a range of 0.1–50 ng/mL, with regression coefficient of 0.9990 and 0.9991. In the unknown samples, concentrations of Cr(VI) can be determined by measuring the peak areas obtained due to $Cr(MDTC)_2(OMDTC)$ and calculating the concentration from regression equation obtained from the calibration graph. The concentration of Cr(III) can be calculated by subtracting the value corresponding to $Cr(MDTC)_3$ obtained from Cr(VI). The interference due to $Cr(MDTC)_3$ obtained from Cr(VI) was calculated from the relationship obtained from the ratio of $Cr(MDTC)_2(OMDTC)/Cr(MDTC)_3$. The regression equations obtained are given in Table I. The relative detection limits calculated with respect to the S/N=2 criterion are 0.005 and 0.007 µg/L for Cr(III) and Cr(VI), respectively. These detection limits are better when compared to methods reported in literature (Table II).

Table I. Different Parameters for $Cr(MDTC)_3$ and $Cr(MDTC)_2(OMDTC)$ Complexes Obtained from Cr(III) and Cr(VI)

Parameters	$Cr(MDTC)_3$ in Cr(III)	$Cr(MDTC)_2(OMDTC)$ in Cr(VI)
Regression equation	0.094x + 0.015	0.064x + 0.047
Correlation coefficient (r^2)	0.9991	0.9990
RSD (%)	4.9	4.3
LOD, S/N =3 (ng/mL)	0.005	0.007
Retention time (min)	5.4	4.3
RSD for variation in time	1.3	1.1

Table II. Comparison with Reported Methods

S. No.	Matrix	Technique used	LOD [Cr(III), Cr(VI)]*	Ref.
1	Waste water	IC–ICP-MS	Below 0.2	(3)
2	Soil/plant samples	HPLC–DRC-ICP-MS	0.05	(16)
3	Drinking water	HPLC–FAAS	3.7, 2.0	(17)
4	Wastewater	HPLC–UV	2.2, 4.5	(18)
5	Airborne matter	HPLC–ICPMS	160	(19)
6	Water	CPE-HPLC	3.4, 5.2	(20)
7	Natural waters	HPLC–ICPMS	0.5	(21)
8	Water sample	HPLC–UV	400, 1000	(22)
8	Spiked samples	HPLC–DAD	7000	(23)
9	Surface water	HPLC–ICPMS	0.13	(24)
10	Water	HPLC–ICPMS	0.050, 0.12	(25)
11	Drinking water	HPLC–UV	0.005, 0.007	DM†

* ng/mL; † DM = developed method.

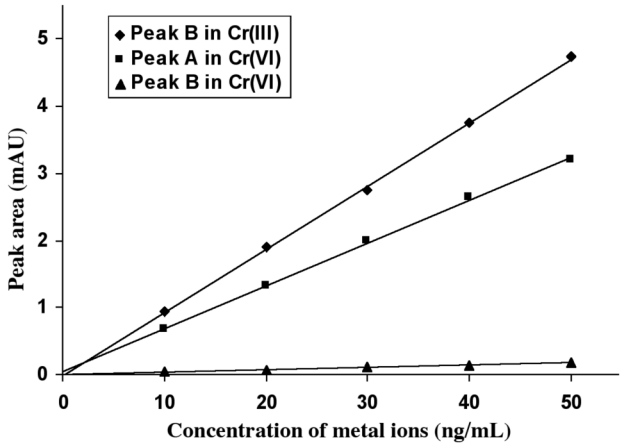

Figure 5. Peak area versus the concentration of Cr, MDTC = 1 mL of 10%, sodium acetate–acetic acid buffer (pH 4) = 1 mL, extracted with chloroform at 55°C with 20 min shaking [Peak A is $Cr(MDTC)_2(OMDTC)$ and Peak B is $Cr(MDTC)_3$ complex].

Effect of diverse ions

The effect of various ions was studied. Different amounts of some alkali metal salts and metal ions were added individually to aliquots containing 50 ng of Cr(III) and Cr(VI). The interferences due to all metals can be avoided by pre-extraction in chloroform at room temperature. The complex of chromium is not formed at room temperature, so Cr(III) or Cr(VI) remain in the aqueous phase, which can be analysed by the general procedure. The tolerance limit of different ions is given in Table III.

Applications

The validity of developed method was checked by the determination of Cr(III) and Cr(VI) in synthetic mixtures. The solutions were prepared by adding different amounts of Cr(III) and Cr(VI) to tap water. The results obtained were satisfactory with recoveries more than 90%. The relative error was calculated for each sample (Table IV).

Conclusions

The method of speciation of chromium has enabled accurate and reproducible measurements of Cr(III) and Cr(VI) in aqueous

Table III. Effect of Diverse Ions on 50 ng of Cr(III) and Cr(VI)

Anions added	Tolerance limit (ng)	Cation added*	Tolerance limit (ng)
Bromide	30	Copper	500
Iodide	50	Lead	600
Chloride	50	Zinc	1000
Fluoride	30	Bismuth	150
Acetate	60	Cadmium	600
Oxalate	18	Manganese	600
Citrate	110	Indium	400
Phosphate	35	Mercury	100
Thiocyanate	65	Silver	500
Sulphate	60	Cobalt	200
Tartrate	10	Nickel	600
Cyanide	30	Uranium	200
Nitrate	50	Arsenic	600
EDTA	0.1	Gold	200

* These ions can be pre-extracted into chloroform at pH 2–4.5 and Cr(III) and Cr(VI) remain in the aqueous solution.

Table IV. Determination of Cr(III) and Cr(VI) in Synthetic Binary Mixtures Spiked in Tap Water

	Taken (µg/L)		Found (ng/mL) (Recovery %)		Relative error (%)	
S. No.	Cr(III)	Cr(VI)	Cr(III)	Cr(VI)	Cr(III)	Cr(VI)
1	25	–	24 (96.0)	–	–0.04	–
2	50	–	49 (98.0)	–	–0.02	–
3	–	30	–	29 (96.6)	–	–0.03
4	–	60	–	59 (98.3)	–	–0.01
5	85	20	80 (94.1)	19 (96.0)	–0.05	–0.05
6	35	45	35 (100)	44 (97.7)	0.0	–0.02
7	25	50	24 (96.0)	48 (96.0)	–0.04	–0.04
8	65	35	63 (96.9)	33 (94.2)	–0.03	–0.05
9	15	80	14 (93.3)	77 (96.2)	–0.06	–0.03

solutions. This method showed a reduction in analysis time as complete elution was obtained in 6 min, and a run of 8 min is sufficient to analyse one sample. Thus, a batch of 10 samples requires only 80 min for analysis. The use of morpholine-4-carbodithioate as a reagent presents an alternate method and extends the use of dithiocarbamates for chromium speciation. It eliminates the need of purification, which is time consuming and is required for commercially available reagents. The method showed improved detection limits of 0.005 and 0.007 µg/L for Cr(III) and Cr(VI), respectively, over reported methods (Table IV). The method is applied for the speciation of chromium to synthetic mixtures. The results are found to be satisfactory. Therefore, the proposed method can be used for the speciation of chromium in unknown environmental samples.

References

1. A.M. Standeven and K.E. Wetterhahn. Chromium(VI) toxicity: Uptake, reduction, and DNA damage. *J. Amer. Col. Toxicol.* **8:** 1275–83 (1989).
2. F. Laborda, M.P. Gorriz, E. Bolea, and J.R. Castillo. Mobilization and speciation of chromium in compost: A methodological approach. *Sci. Total Environ.* **373:** 383–90 (2007).
3. Z. Chen, M. Megharaj, and R. Naidu. Speciation of chromium in wastewater using ion chromatography inductively coupled plasma mass spectrometry. *Talanta* **72:** 394–400 (2007).
4. A.N. Anthemidis and S.V. Koussoroplis. Determination of chromium(VI) and lead in water samples by on-line sorption preconcentration coupled with flame atomic absorption spectrometry using a PCTFE-beads packed column. *Talanta* **71:** 1728–33 (2007).
5. F.P. Capote and M.D.L. Castro. Speciation of chromium by in-capillary derivatization and electrophoretically mediated microanalysis. *J. Chromatogr. A* **1113:** 244–50 (2006).
6. T. Sumida, T. Ikenoue, K. Hamada, A. Sabarudin, M. Oshima, and S. Motomizu. On-line preconcentration using dual mini-columns for the speciation of chromium(III) and chromium(VI) and its application to water samples as studied by inductively coupled plasma-atomic emission spectrometry. *Talanta.* **68:** 388–93 (2005).
7. A. Beni, R. Karosi, and J. Posta. Speciation of hexavalent chromium in waters by liquid–liquid extraction and GFAAS determination. *Michrochem. J.* **85:** 103–108 (2007).
8. V. N. Bulut, C. Duran, M. Tufekci, L. Elci, and M. Sotlak. Speciation of Cr(III) and Cr(VI) after column solid phase extraction on Amberlite XAD-2010. *J. Haz. Mat.* **143:** 112–17 (2007).
9. I. Narin, Y. Surme, M. Soylak, and M. Dogan. Speciation of Cr(III) and Cr(VI) in environmental samples by solid phase extraction on Ambersorb 563 resin. *J. Haz. Mat.* **136:** 579–84 (2006).
10. T. Tande, J. E. Paettersen, and T. Torgrimsen. Simultaneous determination of Cr(III) and Cr(VI) in water by reversed phase HPLC, after chelating with sodium diethyldithiocarbamate. *Chromatographia* **13:** 607–10 (1980).
11. J.S. Wang and K. Chiu. Simultaneous extraction of Cr(III) and Cr(VI) with a dithiocarbamate reagent followed by HPLC separation for chromium speciation. *Anal. Sci.* **20:** 841–46 (2004).
12. G. Macrotrigiano, G.C. Pellacani, C. Preti, and G. Tosi. *Bull. Chem. Soc., Jpn.* **48:** 1018–20 (1975).
13. G. Aravamudan, D.H. Brown, and D. Venkappayya. Some metal complexes of morpholine-4-carbodithioate. *J. Chem. Soc. A* 2744–47 (1971)
14. S. Pramanik, S. Dey, and P. Chattopadhyay. A new chelating resin containing azophenolcarboxylate functionality: synthesis, characterization and application to chromium speciation in wastewater. *Anal. Chim. Acta* **58:** 469–76 (2007).
15. A. Beni, R. Karosi, and J. Posta. Speciation of hexavalent chromium in waters by liquid–liquid extraction and GFAAS determination. *Microchem. J.* **85:** 103–108 (2007).
16. C.Y. Kuo, S.J. Jiang, and A.C. Sahayam. Speciation of chromium and vanadium in environmental samples using HPLC-DRC-ICP-MS. *J. Analyt. Atom. Spect.* **22:** 636–41 (2007).
17. D.Y. Sarica, A.R. Turker, and E. Erol. On-line speciation and determination of Cr(III) and Cr(VI) in drinking and waste water samples by reversed-phase liquid chromatography coupled with atomic absorption spectrometry. *J. Sep. Sci.* **29:** 1600–1606 (2006).
18. M.A. Hossain, M. Kumita, Y. Michigami, T.S.A. Islam, and S. Mori. Rapid speciation analysis of Cr(VI) and Cr(III) by reversed-phase with UV detection. *J. Chromatogr. Sci.* **43:** 98–103 (2005).
19. M. Takaya. Novel characterization method for airborne particulate matter in workplace-chemical speciation method for heavy metals and real time size classified analysis. *Bunseki Kagaku* **53:** 1193–94 (2004).
20. A.N. Tang, D.Q. Jiang, Y. Jiang, S.W. Wang, and X.P. Yan. Cloud point extraction for high-performance liquid chromatographic speciation of Cr(III) and Cr(VI) in aqueous solutions. *J. Chromatogr. A* **1036:** 183–88 (2004).
21. F. Seby, S. Charles, M. Gagean, H. Garraud, and O.F.X. Donard. Chromium speciation by hyphenation of high-performance liquid chromatography to inductively coupled plasma-mass spectrometry - Study of the influence of interfering ions. *J. Anal. Atom. Spect.* **18:** 1386–90 (2003).
22. I. Ali and H.Y. Aboul-Enein. Speciation of arsenic and chromium metal ions by reversed phase high performance liquid chromatography. *Chemosphere* **48:** 275–78 (2002).
23. S. Cathum, C.E. Brown, and W. Wong. Determination of Cr3+, CrO42-, and Cr2O72- in environmental matrixes by high-performance liquid chromatography with diode-array detection (HPLC-DAD). *Anal. Bioanal. Chem.* **373:** 103–10 (2002).
24. Y. Martinez-Bravo, A.F. Roig-Navarro, F.J. Lopez, and F. Hernandez. Multielemental determination of arsenic, selenium and chromium(VI) species in water by high-performance liquid chromatography-inductively coupled plasma mass spectrometry. *J. Chromatogr. A* **926:** 265–74 (2001).
25. F. Vanhaecke, S. Saverwyns, G.D. Wannemacker, L. Moens, and R. Dams. Comparison of the application of higher mass resolution and cool plasma conditions to avoid spectral interferences in Cr(III)/Cr(VI) speciation by means of high-performance liquid chromatography—inductively coupled plasma mass spectrometry. *Anal. Chim. Acta* **419:** 55–64 (2000).

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