Determination of As(III) and As(V) in soils using sequential extraction combined with flow injection hydride generation atomic fluorescence detection

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Received 20 June 2002; received in revised form 25 October 2002; accepted 25 October 2002

Abstract

An analytical procedure for determination of As(III) and As(V) in soils using sequential extraction combined with flow injection (FI) hydride generation atomic fluorescence spectrometry (HG-AFS) was presented. The soils were sequentially extracted by water, 0.6 mol l⁻¹ KH₂PO₄ solution, 1% (v/v) HCl solution and 1% (w/v) NaOH solution. The arsenite (As(III)) in extract was analyzed by HG-AFS in the medium of 0.1 mol l⁻¹ citric acid solution, then the total arsenic in extract was determined by HG-AFS using on-line reduction of arsenate with L-cysteine. The concentration of arsenate (As(V)) was calculated by the difference. The optimum conditions of extraction and determination were studied in detail. The detection limit (3σ) for As(III) and As(V) were 0.11 and 0.07 μg l⁻¹, respectively. The relative standard deviation (R.S.D.) was 1.43% (*n* = 11) at the 10 μg l⁻¹ As level. The method was applied in the determination of As(III) and As(V) of real soils and the recoveries of As(III) and As(V) were in the range of 89.3–118 and 80.4–111%, respectively.

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Keywords: Speciation; Sequential extraction; FI-HG-AFS; Arsenic; Soils

1. Introduction

Arsenic may accumulate in soils and sediments due to the use of arsenical pesticides, fertilizers, irrigation and oxidation of volatile arsines in air, dust from the burning of fossil fuels as well as disposal of industrial, municipal and animal wastes [1]. Because the toxicity and physiological behavior of arsenic are dependent on its chemical forms, it is not sufficient to determine the total content of arsenic in order to assess the toxicity of a particular medium.

The mobility and bioavailability of metals in environmental solid samples is generally evaluated by using sequential extraction schemes (SESS) [2]. A large number of sequential extraction methods have been reported [3], but many of which are variants on the five-step sequential extraction scheme developed by Tessier et al. [4]. Under this scheme, trace metals (Cd, Co, Cu, Ni, Pb, Zn, Fe, and Mn) were classified into five fractions: exchangeable, bound to carbonates, bound to Fe–Mn oxides, bound to organic matter, and residual. The Commission of the European Communities Bureau of Reference (BCR) has proposed the production of a sediment reference material certified for its extractable heavy metal contents [5]. Recent years, different extraction schemes have also

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been developed for the speciation of arsenic in solid samples, in which the extraction media including H_2O and HCl [6], KH_2PO_4 [7], NaOH [8], $(\text{NH}_4)_2\text{C}_2\text{O}_4$, NaHCO_3 and Na_2CO_3 [9], $\text{H}_2\text{O}-\text{CH}_3\text{OH}$ [10], etc. are commonly used. However, the sequential extraction method is still few used in arsenic speciation.

Sensitive and reliable determination technique is in imperative need to be established owing to the considerably low concentration of arsenic in soil extracts. Hydride generation atomic fluorescence spectrometry (HG-AFS) is one of excellent analytical techniques compare to methods such as GF-AAS [11], ICP-AES [12], ICP-MS [13] and XRF [14], etc. for the detection of arsenic. It is characterized by high efficiency, low sample volume, low reagent consumption, improved tolerance of interference and rapid determination. Nowadays, HG-AFS coupled with different separation techniques have been widely used in arsenic speciation analysis [15–17]. Flow injection (FI) on-line separation and preconcentration can overcome the shortcomings of batch-wise operation and realize automatization easily.

The aim of this paper is to introduce a sequential extraction procedure for the arsenic in soils where the As(III) and As(V) were determined by FI-HG-AFS. The conditions of extraction and determination were studied in detail.

2. Experimental

2.1. Apparatus

A AF-610 atomic fluorescence spectrometer with a commercial gas–liquid separator (Beijing Raileigh Analytic Instrument Corporation) was used under the operating conditions listed in Table 1. The flow injection analytical system applied was a JTY-1B flow

Table 1
Operating conditions for atomic fluorescence spectrometer

PMT-voltage (V)	320
Lamp current (mA)	60
Argon carrier gas flow rate (ml min^{-1})	600
Atomization temperature	Low
Observation height (mm)	7

Fig. 1. Schematic diagram of the FI manifold for on-line reduction of As(V): (a) collecting sample and reduction of As(V), and (b) determining stage. P_a and P_b , peristaltic pumps; V, 8-channel rotary injection valve; T_1 and T_2 , T-tube; L, sample loop heated with water bath (100°C); R, reducing agent; S, sample; W, waste.

injection multifunction solution autohandling system (Faculty of Materials Science and Chemical Engineering, China University of Geosciences). Fig. 1 shows the manifold for on-line reduction of As(V) used in this study. The manifold program for this flow injection system is showed in Table 2. Reacting coils, sample loop and connecting tubing between them were 1 mm in inner diameter.

2.2. Reagents

Most solutions were prepared from analytical-reagent grade reagents unless stated otherwise. Distilled water was used throughout. The As(III) stock solution (1000 mg l^{-1}) was prepared by dissolving 0.6600 g As_2O_3 in 12.5 ml 1.0 mol l^{-1} NaOH

Table 2
Flow injection program used for the HG-AFS measurement

Steps	Time (s)	Pump rate (rotations min ⁻¹)		Valve position	Function
		P _a	P _b		
0	30	80	90	0	The sample and reducing agent are mixed and full of sample loop (L); pump P _b is active to keep AFS flame burning
1	60	0	90	0	Pump P _a stop; sample and reducing agent are reacted in L heated with water bath; As(V) was reduced
2	15	90	90	1	Sample is reacted with NaBH ₄ , and pushed to AFS for determination; water is pumped in P _{a2} to wash the tube

solution, neutralizing with 1.0 mol l⁻¹ HCl and diluting to 500 ml with distilled water. A 1000 mg l⁻¹ As(V) stock solution was prepared by dissolving Na₃AsO₄·12H₂O in distilled water. All the standard solutions were prepared by successive dilutions to the required concentration just before use. A 10 g l⁻¹ sodium tetrahydroborate solution was prepared by dissolving the reagent in 2 g l⁻¹ sodium hydroxide solution just before use.

One percent (w/v) L-cysteine solution, 0.5 mol l⁻¹ citric acid solution, 0.6 mol l⁻¹ KH₂PO₄ solution, 1% (w/v) NaOH solution, 1% (v/v) HCl solution and 1:1 aqua regia solution were also used in this work.

2.3. Extraction and analysis of arsenic

2.3.1. Extraction procedure

The sequential extraction procedure comprised the following four-steps.

1. One gram sample was transferred into a 10 ml centrifuge tube and 10 ml distilled water added. The tube was closed and surged for 2 h, then centrifuged (3000 rotations min⁻¹, 30 min) until the extract was limpid. The extract was decanted into a 25 ml tube, diluted to 25 ml with distilled water and kept frozen (−20 °C) until analysis.
2. Ten milliliter 0.6 mol l⁻¹ KH₂PO₄ solution was added for the second extraction (3 h). The same preparation procedure was then repeated as for the water extract.
3. Ten milliliter 1% (v/v) HCl solution was added for the third extraction (3 h). After the extract was decanted, 1.2 mol l⁻¹ NaOH solution was used to neutralize the extract, then diluted to 25 ml with

distilled water and kept frozen (−20 °C) until analysis.

4. Ten milliliter 1% (w/v) NaOH solution was then used for the fourth extraction (4 h), and 20% (v/v) HCl solution was added for neutralizing the extract. The other operations are the same for the water extract.

2.3.2. Digestion of soil sample and residue

After above extraction, the residue was transferred and dried in air. Soil sample and residue were digested for total arsenic determination.

The 0.2500 g sample was transferred into a 25 ml test tube, 10 ml 1:1 aqua regia solution was added and the solution was heated with water bath (100 °C) for 1 h. Solution was cooled and diluted to 25 ml with distilled water. Solution was allowed to stand overnight, and then total arsenic was determined by FI-HG-AFS with the given manifold program and instrument operating parameters.

2.3.3. Determination of As(III) and total arsenic in extracts

Five milliliter above extract was transferred into 10 ml test tube and 2 ml 0.5 mol l⁻¹ citric acid solution was added. The solution was diluted to 10 ml with distilled water (pH = 3.1). The total arsenic in extract was determined by FI-HG-AFS via on-line reduction of As(V) with L-cysteine using the give manifold program as shown in Fig. 1. The reducing agent (R) was replaced by 0.1 mol l⁻¹ citric acid solution. The time of step one in operating program was changed to 5 s and the As(III) was determined. The content of As(V) was calculated by subtracting As(III) from the total As.

3. Results and discussion

3.1. Optimization of extraction procedure

Water, 0.1 mol l^{-1} KH_2PO_4 solution [7], 6 mol l^{-1} HCl solution [6] and 0.1 mol l^{-1} NaOH [8] solution has been separately used as extraction media for arsenic in solid samples. In this work, they were sequentially used to extract different forms of arsenic in soils.

The influences of extractant concentration and extraction time were studied in detail. Figs. 2 and 3 show the effects of KH_2PO_4 , HCl and NaOH concentration, respectively. The highest extraction efficiency was obtained when the KH_2PO_4 concentration was 0.6 mol l^{-1} . In contrast, the extract efficiency increased with the increasing of HCl and NaOH concentration. However, higher HCl and NaOH concentration will increase the effect of matrix, and As(III) can be converted to As(V) at higher pH value [18]. Therefore, the concentration of KH_2PO_4 , HCl and NaOH used in this work were 0.6 mol l^{-1} , 1% (v/v) and 1% (w/v), respectively.

The effect of extraction time was shown in Fig. 4. When using water as extraction media, there was no significant influence of extraction time. The effect of

extraction time by using KH_2PO_4 solution is similar to HCl solution, and the extraction efficiency was higher when extraction time was 3 h. However, the extraction efficiency increased with increasing of extraction time by using NaOH solution, and the efficiency was stable when the time exceeded 4 h. Therefore, the extraction times used in different steps were 2, 3, 3 and 4 h, respectively.

3.2. pH-selective determination of As(III)

Because the transformation of As(V) to As(III) and the ensuing generation of arsine by hydride generation is considerably slower at lower acidities, selective determination of As(III) using the FI-HG-AFS system can be achieved by controlling the reaction pH conditions and the time of the hydride generation. As a measurement for a sufficient discrimination against As(V), a contribution to the signal due to the presence of As(V) corresponding to an absorbance of $\leq 5\%$ as compared with the signal of equal As(III) concentrations was found to be appropriate [19].

For selective determination of As(III), different media including 0.1 mol l^{-1} citric acid, 1% acetic acid, 0.1 mol l^{-1} tartaric acid, 0.15 mol l^{-1} nitric acid and

Fig. 2. Effect of KH_2PO_4 concentration.

Fig. 3. Effect of HCl and NaOH concentration.

Fig. 4. Effect of the extraction time.

0.12 mol l⁻¹ hydrochloric acid were studied. When citric acid was used as medium, the flame of AFS is stable, and higher sensitivity to As(III) can be obtained. Further study showed that when concentration of citric acid was 0.1 mol l⁻¹, the influence of As(V) can be discriminated well as shown in Fig. 5. So 0.1 mol l⁻¹ citric acid was used as medium for selective determination of As(III) in this work.

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Fig. 5. Effect of the citric acid concentration.

The effect of sample flow rate on fluorescence intensity of As(III) and As(V) was studied. When the flow rate is slower than 7 ml min^{-1} , As(V) has higher fluorescence signal because of the transformation of As(V) to As(III) and the ensuing generation of arsine by hydride generation, as well as As(III) has smaller signal because little sample was introduced. But when the flow rate is faster than 7 ml min^{-1} , both As(III) and As(V) fluorescence intensities reduce with the increasing of sample flow rate. The flow rate of 13 ml min^{-1} was used for selective determination of As(III) in this work. At this flow rate, the influence of As(V) can be eliminated and As(III) has enough sensitivity as well.

3.3. On-line reduction of As(V)

The reduction agents typically applied to convert As(V) to As(III) are potassium iodide and L-cysteine. Tsalev et al. [20] pointed out some advantages of application of L-cysteine. Pre-reduction of the inorganic (i) pentavalent analytes, i-As(V) and i-Sb(V), to their trivalent oxidation states is faster and more efficient compared to other prereductants, and sensitivity can be improved at the same time. However, the application of KI demands relatively strong acidic conditions

for both the samples and the reduction medium and the high concentrations commonly used can lead to generation of considerable levels of iodine, which is especially disadvantageous if the quantification of the analyte is effected via hydride generation [19]. L-Cysteine was used for As(V) reduction in this work.

The influence of L-cysteine concentration on reduction of As(V) was studied. The results showed that 0.1% (w/v) of L-cysteine in solution was enough for reducing 50 ng ml^{-1} As(V) in solution. In order to keep the same reaction acidic conditions, the reductant used was 0.1% (w/v) L-cysteine in 0.1 mol l^{-1} citric acid solution.

Higher temperature would quicken the reduction of As(V). The time needed for reduction of 10 ng ml^{-1} As(V) solution is more than 50 min at room temperature, but is less than 60 s when immersing the sample loop into a water bath (100°C). The sample loop was immersed in boiled water in this work and the reduction time was 60 s.

The fluorescence intensity of arsenic could be improved by using of L-cysteine [21]. Two series of As(III) standard solutions with and without L-cysteine were tested shown in Fig. 6. The slope was increased by 35.1% when using of L-cysteine.

Fig. 6. Improvement of sensitivity by L-cysteine.

3.4. Interference

Interferences due to coexisting ions on the determination of $10 \mu\text{g l}^{-1}$ of As(III) were studied. The 20 mg l^{-1} Cu^{2+} , K^+ , Na^+ , Ca^{2+} , Fe^{2+} , Ba^{2+} , 10 mg l^{-1} Al^{3+} , Co^{2+} , 5 mg l^{-1} Ni^{2+} , Pb^{2+} , Cr^{3+} , Ag^+ and 5 mg l^{-1} Mn^{2+} , Fe^{3+} , Hg^{2+} , Cr^{6+} did not interfere in the FI-HG-AFS determination of As.

3.5. Analytical features of the developed procedure

The method developed for the determination of inorganic arsenic by FI-HG-AFS is based on

pH-selective determination of As(III) and on-line reduction of As(V) by L-cysteine. A regression equation (fluorescence intensity versus concentration ($\mu\text{g l}^{-1}$)), $y = 58.55x - 22.72$, with a correlation coefficient of 0.9995, was obtained for an As(III) concentration range from 10 to $50 \mu\text{g l}^{-1}$.

By using FI-HG-AFS system, a sampling frequency of 20 h^{-1} was obtained. The detection limit (3σ) of As(III) and As(V) were 0.11 and $0.07 \mu\text{g l}^{-1}$, respectively. The relative standard deviation (R.S.D.) was 1.43% ($n = 11$) at the $10 \mu\text{g l}^{-1}$ As level.

The accuracy of the method was investigated by determination the recovery percentage of spiked

Table 3
Recovery percentage obtained by sequential extraction of As in spiked soils^a (%)

Soil code	Sample 1		Sample 2		Sample 3	
	As(III)	As(V)	As(III)	As(V)	As(III)	As(V)
As added ($\mu\text{g g}^{-1}$)	0.125	0.125	0.125	0.125	0.125	0.125
Water extractable	89.6 ± 3.6	107 ± 2.1	90.8 ± 1.9	111 ± 1.3	89.3 ± 5.1	110 ± 2.2
KH_2PO_4 solution extractable	104 ± 1.2	81.6 ± 5.4	97.6 ± 4.4	85.6 ± 3.8	100 ± 1.8	83.2 ± 2.7
HCl solution extractable	102 ± 2.3	91.8 ± 2.0	106 ± 1.0	83.2 ± 2.7	111 ± 1.0	80.4 ± 3.9
NaOH solution extractable	114 ± 1.0	96.0 ± 1.8	108 ± 1.1	85.6 ± 3.1	118 ± 1.1	93.6 ± 1.6

^a Average value $\pm$ the corresponding S.D. of three independent determinations.

Table 4
Distribution of As species in soils^a (As/ng g⁻¹)

Soil code	Sample 1			Sample 2			Sample 3		
	As(III)	As(V)	Extractability (% of total As)	As(III)	As(V)	Extractability (% of total As)	As(III)	As(V)	Extractability (% of total As)
Water extractable	8.1 ± 0.5	20.0 ± 1.1	0.18	6.0 ± 0.2	16.0 ± 0.7	0.14	5.2 ± 0.1	20.4 ± 1.0	0.15
KH ₂ PO ₄ solution extractable	48.2 ± 3.5	820 ± 15	5.74	48.0 ± 2.8	935 ± 14	6.32	50.0 ± 2.5	790 ± 9.5	5.13
HCl solution extractable	12.3 ± 0.7	466 ± 6.4	3.16	16.1 ± 0.4	604 ± 8.6	3.98	18.2 ± 1.2	612 ± 8.1	3.85
NaOH solution extractable	25.1 ± 2.0	787 ± 12	5.37	30.2 ± 2.2	775 ± 10	5.17	30.0 ± 1.9	725 ± 11	4.61
Total As in residues ^b (μg g ⁻¹)	12.94 ± 0.12	85.58 ^d	12.84 ± 0.18	82.52 ^d	12.40 ± 0.20	75.79 ^d			
Total As in samples ^c (μg g ⁻¹)	15.12 ± 0.17	–	15.56 ± 0.21	–	16.36 ± 0.20	–			

^a Average of three independent analysis ± the corresponding S.D.

^b Total As was determined by FI-HG-AFS after digestion.

^c Total As was determined by FI-HG-AFS after digestion.

^d Percentage of As in residues (% of total As).

concentration of As(III) and As(V) added to extracts. Under the above conditions, the recoveries of As(III) and As(V) were in the range of 89.3–118 and 80.4–111%, respectively, as shown in Table 3.

3.6. Determination of As(III) and As(V) in soil samples

The proposed method was applied to the determination of As(III) and As(V) in three soils collected from Wuhan (Hubei province, China). The results of sequential extractability of different species are shown in Table 4. KH_2PO_4 and NaOH solutions have higher extraction efficiency compare to water and HCl solutions. However, the extractable arsenic in soils is very low. More than 75% of total arsenic can't be extracted by four kinds of extraction media used in this work. This fraction arsenic may be incorporated within the crystal structure, which is not expected to be released in solution over a reasonable time span under the conditions normally encountered in nature. Therefore, it has low mobility and is comparatively safe to human being. In soil extracts, arsenic is mainly existed as As(V) form as also shown in Table 4.

4. Conclusions

A sequential extraction method combined with FI-HG-AFS was developed for the determination of As(III) and As(V) in soils. The method is sensitive, simple and rapid for analysis of arsenic. The extractable arsenic is mainly existed as As(V) form in soils.

Sequential extraction technique is very useful in estimating the metal's mobilities and bioavailabilities. Water-extractable arsenic has the highest mobility because it can easily enter the aquatic environment and affects human health when the soil is washed by rain. KH_2PO_4 solution extractable, HCl solution extractable and NaOH solution extractable arsenic are also bioavailable arsenic forms in soil. But these four fractions of arsenic are all very little in soils. More than 75% of total arsenic can't be extracted by above four kinds of extraction media. This fraction arsenic has low mobility and therefore, is comparatively safe

to human being. The proposed sequential extraction procedure is reasonable and suitable for speciation analysis of arsenic in soils.

Acknowledgements

The authors thank the China Bureau of Geological Investigation Project (DK0003048) and Open Found from the Keylab of Environmental Sciences and Engineering, Henan Normal University for financial support.

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