

Arsenic speciation in contaminated soils

S. Garcia-Manyes^{a, b}; G. Jiménez^a; A. Padró^c; Roser Rubio^{a, *}; G. Rauret^a

^a Departament de Química Analítica, Universitat de Barcelona, Avda. Diagonal, 647, E-08028 Barcelona, Spain

^b Departament de Química Física, Universitat de Barcelona, Avda. Diagonal, 647, E-08028 Barcelona, Spain

^c Serveis Científic Tècnics, Universitat de Barcelona, Lluís Solé i Sabarís, 1–3, E-08028 Barcelona, Spain

Corresponding author. Tel.: +34-93-402-1283; fax: +3493-402-1233 E-mail address: roser.rubio@apolo.qui.ub.es (R. Rubio).

Abstract

A method for arsenic speciation in soils is developed, based on extraction with a mixture of 1 mol l⁻¹ of phosphoric acid and 0.1 mol l⁻¹ of ascorbic acid, and further measurement with the coupling liquid chromatography (LC)–ultraviolet (UV) irradiation–hydride generation (HG)–inductively coupled plasma mass spectrometry (ICP/ MS). The stability of the arsenic species in the extracts is also studied. The speciation method applied to several Spanish agricultural contaminated soils from the Aznalcollar zone shows that arsenate is the main species in all the soils analysed and that in some samples arsenite and methylated species could also be detected. The determination of the “pseudototal” arsenic in these soils, obtained by applying extraction with aqua regia (ISO Standard 11466), is also carried out. Both the speciation method and the aqua regia method are applied to several certified reference materials (CRMs) in which total arsenic content is certified. Finally, the same LC–UV–HG coupling with atomic fluorescence spectrometry (AFS) detection reveals to be a valid coupling system to perform arsenic speciation in the soils according to its fair quality parameters and easy utilisation.

1. Introduction

Arsenic can occur in agricultural soils in some regions, as a consequence of the use of arseniccontaining pesticides and herbicides [1–3]. Other contributing sources of arsenic in the soils are industrial and mine wastes. Nevertheless, the contamination of the soils due to irrigation with groundwater with high arsenic content from natural origin is widely reported since it affects large areas in the world [4–8]. In spite of the fact that arsenic in the soils and also in sediments is mainly present in inorganic forms, the organic compounds monomethylarsonate (MMA) and dimethylarsinate (DMA) may be detected [9–11]. These methylated species can originate from microorganisms-mediated oxidation–reduction reactions. Moreover, some methylated species can be demethylated to inorganic arsenic [1,3,12]. The chemical form of arsenic determines its

mobility from the soils and sediments, As(III) being the most mobile [13]; thus the knowledge of the chemical forms of arsenic can provide a good tool for the assessment of their further mobilization to the aqueous phase in equilibrium with the soils or sediments.

Arsenic speciation has acquired great importance in recent years, since the toxicity of arsenic differs dramatically with the wide range of its organic and inorganic chemical forms [14–17]. Nowadays the mechanisms that originate toxicity are not well elucidated and many efforts are going on in this subject [18–21] as well as on the development of effective therapies [22,23]. The development of analytical techniques that allow chemical speciation is therefore mandatory, since total arsenic determination cannot in many cases be an appropriate measure for assessing toxicity, environmental impact, or the effect of occupational exposure [3]. The extraction of chemical species is a crucial topic in element speciation studies in complex matrices in which the extraction system has to provide good recovery and to preserve the identity of the native species in the sample. Several extracting agents have been proposed for further measurement of the arsenic species present in the soils [24–28]. With regard to extraction conditions, microwaves are revealed as a successful technique for the extraction of elemental species [29] and is applied to extract arsenic species in biological matrices [30–32]. However, few articles [26,33] deal with the extraction of arsenic species from the sediments and soils by using low-power microwaves. Coupled systems with liquid chromatography (LC)–hydride generation (HG) and detection by atomic absorption spectrometry (AAS), atomic emission spectrometry (AES), atomic fluorescence spectrometry (AFS) or inductively coupled plasma mass spectrometry (ICP/MS) have been shown to be suitable for measuring arsenic species in the extracts obtained from natural samples even at very low concentration levels. In such couplings derivatization by generation of volatile arsines is the common step to reach good sensitivity. We present here the feasibility for extracting arsenic from the soils and sediments by using a mixture of phosphoric acid and ascorbic acid under microwaves. The stability with time of the arsenic species in the soil extracts is also evaluated by using the coupling LC–UV–HG–ICP–MS for measuring the arsenic species after extraction. The coupling LC–UV–HG–AFS is successfully applied and it is revealed as a suitable technique for arsenic speciation in the soil extracts. The quality parameters detection and quantification limits as well as precision by using this coupling are established for the soil extracts. Both couplings are applied in the present study to the soils collected in the contaminated zone of Aznalcollar (Spain) [34].

2. Materials and methods

2.1. Apparatus

A Prolabo microwave digester Model A301, 2.45 GHz, equipped with a TX32 programmer was used. The instrument can apply power settings of 20–200 W in steps of 10 W, and the microwave energy was focused into the glass vessel under atmospheric pressure. A temperature and time P/Selecta Model RAT 4000051 regulator bloc, which controls the P/Selecta Bloc Digester 12, and which allows 12 vessels to be operated at the same time, was used in the

pseudototal arsenic determination. A Perkin–Elmer ICP–AES Optima 3200 RL spectrometer provided with “cross-flow” nebulizer spectrometer was used in the measurement of total arsenic in some soil samples. The spectrometer is equipped with a 27.12 MHz, 750–1750 W work power radiofrequency source and quartz torch. Data acquisition was performed with computer software. A Karl–Fisher titrator automat 633, pump unit 681, dosimat 715 and stirrer 728 model, all of them from Metrohm, Herisaw, Switzerland have been used as an alternative technique to determine the moisture of some samples. Coupled system, LC–UV–HG–ICP/MS, was used for the determination of the arsenic species. LC–UV–HG–AFS was also applied and the quality parameters were established. The coupled systems include the following instrumentation.

2.1.1. Separation

A Perkin–Elmer 250LC binary pump (CT, USA) with a Rheodyne model 7125 injector (CA, USA) and a 20 μ l injection loop was used. An anion exchange 250 $\times$ 4.1 mm Hamilton PRP X100 column with 10 μ m spherical poly(styrene–divinylbenzene) containing trimethyl ammonium groups as exchangers was used, with a guard column packed with the same stationary phase.

2.1.2. UV derivatization

The photoreactor system combines a Hereaus TNN 15/32 low-pressure mercury vapour lamp (λ =254 nm, external diameter 2.5 cm, 17 cm length, 15 W) and PTFE tubing (12 m length, internal diameter 0.5 mm) which constituted the photoreactor system. A computer-controlled microburette (Microbur 2031 Crison) was used to add the peroxodisulfate solution into the photoreactor.

2.1.3. Detection systems

(A) A Perkin–Elmer FIAS 400 with a gas–liquid separator equipped with a PTFE membrane was used for HG. A Perkin–Elmer Elan 6000 ICP-MS instrument was used for detecting As, and areas were calculated from custom-developed software using MATLAB language. The sample channel was connected to the outlet of the LC–UV system. The scheme of the overall coupling is reported in [30].

(B) PS. Analytical model excalibur atomic fluorescence spectrometer equipped with a As hollow cathode lamp (current intensities: primary 27.5, boost 35.0) and a Perma pure drying membrane (Perma Pure Products, Farmingdale, NJ, USA) for drying the generated hydride. Measuring wavelength was 193.7 nm. Data acquisition was performed with a microcomputer by using a homemade software (PENDRAGON 1.0). Peak heights and peak areas were measured from custom-developed software running with MATLAB language. The scheme of the overall coupling is reported in [35].

2.2. Standards and reagents

All the solutions were prepared with doubly deionized water (USF Purelab Plus, Ransbach, Baumbach, Germany) of 18.3 M Ω cm resistivity.

Standard solutions (1000 mg l⁻¹ [As]) of arsenic compounds were prepared as follows. Arsenite: As₂O₃ (Merck, Darmstadt, Germany) primary standard was dissolved in NaOH (4 g l⁻¹). Arsenate: Na₂HAsO₄·7H₂O (Carlo Erba), monomethylarsonate (CH₃)As(ONa)₂·6H₂O (Carlo Erba) and dimethylarsinate (CH₃)₂AsNaO₂·3H₂O (Fluka) were dissolved in water. All the standard solutions were standardized with respect to arsenic. These stock solutions were kept at 4 °C in darkness. More dilute solutions for the analysis were prepared daily.

Extracting reagents: Ortho-phosphoric acid (H₃PO₄, Merck Pro analysi, 85% purity) and EDTA (Merck proanalysi, 99.4–100.6%). Sodium dihydrogenphosphate anhydrous (NaH₂PO₄, Merck Suprapur) and L(+) ascorbic acid (Merck proanalysi, 99.7%) were assayed for microwave extractions.

Mobile phase: Phosphate buffer pH 6 was prepared from 100 mmol l⁻¹ of mixture of NaH₂PO₄ and Na₂HPO₄ (Merck, Suprapur). The solution, after filtering through a 0.22 μ m nylon membrane, was sonicated for 10 min.

Peroxodisulfate solution: K₂S₂O₈ (Fluka, purity $\geq$ 99.5%) at 5% prepared in sodium hydroxide (NaOH Suprapur, Merck) at 2.5% was used for photooxidation step.

Hydride-generating reagents: 10% sulfuric acid was prepared from 96% H₂SO₄ (Merck Suprapur). Sodium borohydride (NaBH₄ tablets, Fluka, purity $\geq$ 97%) at 5% in 0.2% NaOH was filtered through 0.45 μ m cellulose membrane and it was prepared daily.

Nitric acid (HNO₃, Baker, Instra-analysed, 70%) and hydrochloric acid (HCl, Baker, Instraanalysed, 36.5–38%) were used for the aqua regia digestion method.

1% potassium iodide (KI, Merck, Suprapur, minimum 99.5%) and ascorbic acid (Merck, proanalysi 99.7%) 0.2% in HCl 9% were used for prereduction in the determination of As after aqua regia leaching as well as for the determination of total arsenic in the phosphoric–ascorbic extracts, when HG–ICP/MS was used.

2.3. Certified reference materials

GBW07405 soil (412 $\pm$ 8 mg kg⁻¹ As) and GBW07311 sediment (188 $\pm$ 6 mg kg⁻¹ As) both were from NRCCRM (PR China), and BCR320 sediment (76.7 $\pm$ 4.7 mg kg⁻¹ As) was from BCR (Brussels, Belgium).

Five soil samples—2AUTr, 2DEPr, 2DEP, 1RIB2 and 2QUEh from the Aznalcóllar zone—were collected in May 1998. This area was affected in 1998 due to an accident from a mine waste reservoir. The location of sampling points, sampling and pretreatment, as well as the characterization of these soils are described elsewhere [34]. A second sampling in January 2000 was carried out from this contaminated zone, and five soil samples were selected for the study—3DEP, 3DEPr, 3RIB, 3QUE and 3QUEorg. 3QUEorg soil was sampled from the 3QUE soil, but

in a zone in which considerable amounts of manure was added. Approximately 10 kg of each of these soils was collected at 5–8 cm depth and stored in plastic containers. Samples were dried at 40 °C during 5 days, then they were gently crushed, and the particles passing through a 2 mm nylon sieve were collected and homogenized before analysis.

2.4. Procedure for pseudototal arsenic determination by using aqua regia leaching

The standard ISO/CD 11466 1995 was followed for sample extraction. The soil or sediment subsample (3 g of material) was placed in the reflux vessel and it was wetted by adding 1 ml of water. Then the appropriate volume of aqua regia (28 ml per 3 g sample) was added. The cooler was connected and the soil suspension was maintained at room temperature for 16 h. The water cooler was connected and the mixture was heated at 130 °C for 2 h till the extraction was completed. Once at room temperature, the cooler was washed with 5 mol l⁻¹ of HNO₃ and the washing solution was collected into the digestion vessel. The resulting suspension was filtered through an ashless filter (approximately 8 µm) and the solid residue was washed several times with 0.5 mol l⁻¹ of HNO₃. The filtrate together with the washings was diluted up to 100 ml with 0.5 mol l⁻¹ of HNO₃. This solution was transferred to a PTFE container and stored at 4 °C until analysis. The arsenic content in the extracts was measured by ICP–AES or by HG–ICP/MS. When the latter was used, a prereduction step was carried out. For this purpose an aliquot of the aqua regia extract was taken and it was diluted 100-fold. Then 1 ml of a solution containing KI 1% and ascorbic acid 0.2% in HCl 9% was added to 9 ml of the diluted solutions, and the mixture was maintained under room temperature for 1 h before the final measurement. Matrix matching was used for calibration, and thus all three standards for the external curve were treated under the prereduction conditions described. In all the cases three independent replicates were carried out for each sample.

2.5. Procedure for the extraction of the arsenic species

100 mg of the soil or sediment and 15 ml of the extractant (1.0 mol l⁻¹ of phosphoric acid+0.1 mol l⁻¹ of ascorbic acid) previously purged with argon stream for 15 min were placed in an open reflux vessel. The latter was positioned in the cavity of the microwave digester and the mixture was maintained at 60 W for 10 min. Once the solution is at room temperature, a few millilitre of water was added, and the mixture was filtered and diluted up to 50.0 ml with water. After filtering through a 0.22 µm polysulfonic membrane, aliquots were obtained for the determination of total arsenic in the extract by using ICP/MS or by HG–ICP/MS (in this case after aqua regia digestion and the prereduction with KI was carried out as described before) and for arsenic speciation by using LC–UV–HG–ICP/MS. When the extract could not be analysed just after extraction, the aliquots were kept at 4 °C until analysis.

2.6. Measurement of the arsenic species by using LC–UV–HG–ICP/MS

20 µl of the extract was injected into the anion-exchange column of the LC system. Two phosphate buffer solutions at pH 6.0 were pumped at 1 ml min⁻¹ NaH₂PO₄– Na₂HPO₄ 5 mmol l⁻¹

(solution A) and NaH_2PO_4 – Na_2HPO_4 100 mmol l^{-1} (solution B). The gradient programme was 100% A for 2 min, decreasing to 50% A in 0.1 min and maintained for 3 min, then increasing to 100% A in 0.1 min and maintained for 7 min. The eluate reached the hydride generator system and after passing through the gas–liquid separator the volatile arsines were transferred to the ICP/MS detector with the optimal argon flow. A detailed scheme of the coupled system and experimental is reported elsewhere [30]. The arsenic species were quantified by the standard addition method in the extract.

2.7. Measurement of the arsenic species by using LC–UV–HG–AFS

Detection with AFS coupled to the system was also used. The separation conditions in this coupling were those described for the coupling with ICP/MS. The detailed scheme of the coupling and the experimental detection conditions is reported in [35]. The arsenic species were quantified by the standard addition method in the extract.

3. Results and discussion

3.1. Sample pretreatment

A study was carried out to ensure that losses of arsenic did not take place at 40 °C used for drying the soil samples. For this, subsamples of the soils were treated under room temperature (approximately 20 °C), 40 °C and 100 °C, and the total arsenic was measured in each case. The results did not indicate significant differences in the corresponding arsenic contents.

3.2. Determination of the moisture

All the results in the present study are referred to dry mass, and the moisture was determined gravimetrically after a thermal treatment at 105 °C. In spite of the fact that this is the usual method for determining moisture in the soils and in order to ascertain whether under this treatment any volatile compounds present in the soils could be lost, the Karl–Fisher method was applied to compare the results obtained with both the methods. For this purpose, two soils from the second sampling, 3DEP and 3QUE, were selected, since they presented different levels of moisture. According to the results the percentages of moisture for the soil 3DEP were 12.94% (105 °C) and 12.86% (Karl–Fisher), whereas for the soil 3QUE were 3.12% (105 °C) and 3.03% (Karl–Fisher). These results showed that only water was lost during the drying process at 105 °C.

3.3. Determination of pseudototal arsenic content

Arsenic in the aqua regia extracts was measured by ICP–AES or by HG–ICP/MS, according to the concentration values for each sample. The generation of the volatile hydrides increases sensitivity and avoids the significant interference of the chloride when As is measured directly by ICP/MS. The pseudototal arsenic content in the soils 2AUTr, 2DEPr, 2DEP, 1RIB2 and 2QUEh is reported in [34], in these soils the arsenic content ranged between 12.6 and 766 mg kg^{-1} . The results of pseudototal arsenic in the rest of the soils studied as well as in the certified reference materials (CRMs) are reported in Table 1. These results indicated that in some soils,

even after 2 years from the pollution accident, the arsenic content remained significantly high. For CRMs and in spite of the fact that aqua regia does not extract the total metal content, the obtained results are very close to those reported as certified values, except for BCR320. A lower recovery against the certified value is also reported in the literature for this material [26]. The certified arsenic content in these materials ranges between 76.7 and 412 mg kg⁻¹. This range can be considered wide enough to assess the applicability of the aqua regia method, and this leaching procedure is then a good approach for the evaluation of the arsenic content in the soils and sediments by means of an acidic-oxidative attack that avoids the use of hydrofluoric acid.

Insert table 1 around here

3.4. Extraction of the arsenic species from soils

Several extractants were assayed for further determination of arsenic species in order to assess the extraction yields of arsenic. NaH₂PO₄ 0.5 mol l⁻¹, H₃PO₄ 0.6 mol l⁻¹ and EDTA 0.5 mol l⁻¹ at pH 7 were assayed independently as extracting agents on the CRMs GBW07405, GBW07311 and BCR320, and the extractions were performed under 40 W microwave power. In all cases the arsenic content in the extract was measured by using HG-ICP/MS. The values obtained were compared to those certified in order to calculate the percentage of the extraction yield of arsenic. From the results it could be observed that the extraction recoveries varied according to the type of the material. The lowest recovery was obtained by using 0.5 mol l⁻¹ of EDTA being 3% for GBW07405, 20% for GBW07311 and 35% for BCR320. The recoveries obtained by using 0.5 mol l⁻¹ of NaH₂PO₄ were 16% for GBW07405, 18% for GBW07311 and 50% for BCR320, whereas the higher recoveries were obtained by using 0.6 mol l⁻¹ of H₃PO₄, 50% for GBW07405, 70% for GBW07311 and 80% for BCR320. From these results phosphoric acid was chosen for further extraction of the arsenic species. Once the extracting agent was selected, the operational conditions were established, since the extraction system has to guarantee the inalterability of the species during the process as well as to provide reproducible results. Thus we optimized the phosphoric acid concentration, the MW power and the extraction time.

Three phosphoric acid concentrations 0.3, 0.6 and 1.0 mol l⁻¹ were assayed as extractants. The extraction time was 10 min, and the microwave powers assayed were 20 and 60 W. The arsenic content in the extracts was analysed by HG-ICP/MS. From these assays 1.0 mol l⁻¹ of phosphoric acid, 60 W and 10 min were the conditions initially adopted. This phosphoric concentration agreed with that reported in [26]. In order to prevent any As(III) oxidation that could be caused by the main soil components in the extract, the addition of a reducing agent to the extractant solution was considered. For this purpose a few preliminary experiments were carried out. In these experiments four reducing agents were assayed—sodium bromide, oxalic acid, hydroxyl ammonium chloride and ascorbic acid at several concentrations, and each of them mixed with phosphoric acid was assayed. As(III) in the extracts were analysed by LC-UV-HG-ICP/MS. From these assays ascorbic acid was revealed as the best preservative for arsenite. The recovery of As(III) without the addition of ascorbic acid reached 93.7%, whereas when ascorbic acid was added the As(III) recovery increased up to 98.9% and no peak of As(V) was observed

in the corresponding chromatogram. Thus a solution containing 1.0 mol l^{-1} of phosphoric acid and 0.1 mol l^{-1} of ascorbic acid and 60 W microwave power during 10 min were the conditions adopted for extraction.

3.5. Quality parameters

LC–UV–HG–AFS: The detection limit (DL) was calculated from the background signal in the chromatogram and considering three times its standard deviation. The concentrations at the DLs were calculated in triplicate on the corresponding standard curves which were prepared in the soil extract solutions. For quantification limit 10 times the standard deviation of the background signal was considered. The detection and quantification limits obtained, expressed as g l^{-1} in the soil extract, were as follows: 1.91 and 6.37 for As(III), 0.95 and 3.15 for DMA, 2.51 and 8.36 for MMA, and 0.93 and 3.10 for As(V).

Precision: It was calculated from the standard deviation of the peak areas from the chromatograms obtained from nine injections of the soil extract into the coupled system. For the coupling LC–UV–HG–AFS the precision values in terms of %RSD were as follows: As(III) 3.7, DMA 2.7, MMA 6.6, As(V) 2.9, for solutions containing 50 g l^{-1} of the species.

LC–UV–HG–ICP/MS: DLs and repeatability (%RSD) for As species were established in a previous work [30]. They should be taken as orientative values in order to carry out an overall comparison of the sensitivity with both coupling systems. We report here the corresponding DL and repeatability data. DLs, in the measurement solution (as g l^{-1} of As) were 0.03 for As(III), 0.10 for DMA, 0.06 for MMA and 0.12 for As(V). Repeatability, in terms of %RSD, was As(III) 0.8, DMA 3.2, MMA 4.7 and As (V) 2.3, for all the species in concentrations in the range $1\text{--}7 \text{ g l}^{-1}$ of As.

3.6. Application of the speciation procedure to CRMs and to the contaminated soils

The extraction procedure was applied to the CRMs GBW07405, GBW07311 and BCR320, in order to assess the extraction recovery in these materials and for detecting any chemical species of As. This kind of speciation studies is also reported in the literature [26–28], since there is a lack of soil and sediment CRMs in which arsenic species are certified. For this purpose aliquots of the CRMs extracts were analysed by LC–UV–HG–ICP/MS and the species were quantified by the standard addition method in the extracts. Table 2 reports the concentration of each species, the total arsenic contents in the extract (A_{sextr}) and the extraction recoveries calculated, as the percentage of the ratio of total As extracted to As certified ($A_{\text{sextr}}/A_{\text{cert}}$). The lowest recovery obtained for GBW07405 could be attributed to its higher Al and Fe content with respect to the others, elements that have high affinity for retaining arsenic. This behaviour evidences that the extraction of the arsenic species depends on the matrix composition of the materials. It can be observed that in all the materials the main species was arsenate, and small amounts of arsenite could be quantified in the three materials. As(V) and As(III) are also measured in BCR320 using other analytical methods [26,27]. The results show that the sum of the species does not match exactly with the total arsenic measured in the extract. This behaviour is also described in the

literature for CRM320 as well as for other CRMs [26]. This could be attributable to the fact that we are comparing the sum of results coming from individual measurements with the a unique measurement of the total arsenic in the extract, and the individual standard deviation of speciation measurements should be taken into account. Moreover MMA could be measured in GBW07405. From the chromatograms it was also observed that very small peaks, which could be attributable to arsenic compounds, eluted after As(V). Those unidentified peaks in these CRMs, all of them in a very low concentration, could correspond to compounds with high affinity for the stationary phase used for separation. A few experiments were carried out by analysing some organoarsenic compounds such as 2-nitrophenylarsonic acid, used as feed additive [36], p-arsanilic acid, o-arsanilic acid and phenylarsonic acid, used in veterinary [1], under the chromatographic system used in the present study, but none of the retention times corresponded to those of the unidentified compounds. An estimation of their concentration was made by assuming that their behaviour under photooxidation conditions was similar to the rest of the species. Fig. 1 shows an example of the chromatogram corresponding to the extract from BCR320.

Insert Table 2 around here

Insert Figure 1 around here

The speciation procedure was applied to the contaminated soils, and both couplings LC–UV–HG–ICP/MS and LC–UV–HG–AFS were applied. Table 3 reports the results obtained. Regarding extraction recoveries, expressed as the ratio of total As extracted to pseudototal As, it can be observed that the values lie between a wide range, indicating that the extraction yields depends on a great extent on the soil composition. It has been reported that arsenic adsorption is highly dependent mainly on Fe, Al and Mn contents present in the soil, as well as the pH [37–39]. In this work, the studied soils show a wide range of concentration of the three elements, which could account for significant differences in arsenic extractability. In all the chromatograms As(V) appeared as the main species, and small concentrations of As(III) were observed. The soils from the second sampling 3DEP, 3RIB, 3QUE and 3QUEorg were mainly used for assessing the extraction recoveries, for establishing the quality parameters with the coupling LC–UV–HG–AFS and for studying the stability with time of the species in the extracts. In these soils As(V) was also the main species, small amounts of As(III) were observed in all the samples and in some soils methylated species were also present. At this point it should be borne in mind that organoarsenic species are hardly ever reported in bibliography to have been detected in contaminated soils. This gives on the one hand an idea of the contamination level, and, on the other hand, on the feasibility of our two couplings to detect both inorganic and organic species in the soils without any kind of chemical or biological pretreatment. Fig. 2 shows as an example the chromatogram corresponding to a spiked soil extract used for establishing the quality parameters described below with the coupling LC–UV–HG–AFS, and Fig. 3 shows the chromatogram corresponding to the soil 3RIB obtained by using the coupling LC–UV–HG–AFS.

Insert table 3 around here

Insert figures 2 and 3 around here

From the obtained information it is worth pointing out the relationship between the organic matter content in the soil and the presence of methylated species. Thus, Figs. 4 and 5 show the chromatograms obtained from the extracts of the soils 3QUE and 3QUEorg, respectively. Fig. 4 reveals that no peaks of methylated species appeared in the first, whereas Fig. 5 shows two peaks corresponding to MMA and DMA. In these two soils the organic matter content (expressed as loss at 550 °C) is 5.74% for 3QUE soil and 8.11% for 3QUEorg soil. As it has been mentioned before, both the soils belong to the same contaminated area, but the 3QUEorg soil was collected in a zone that was amended with significant amounts of organic matter. The higher amount of organic matter in the soil 3QUEorg with respect to 3QUE may explain the presence of the methylated species in the former, probably attributed to a higher biological activity.

Insert figures 4 and 5 around here

3.7. Stability of the arsenic species in the extracts

A study on the stability of the soil extracts was carried out with the aim to guarantee the reliability of the final measurements of the arsenic species. For this purpose 3QUEorg soil was studied, due to its higher content of MMA and DMA with respect to the rest of the soils. The contents of As(III), DMA, MMA and As(V) are reported in Table 3. For the study of stability the extract from this soil was obtained according to the procedure and the extract kept at 4 °C under darkness. Under these conditions several aliquots were taken at different periods of time. The first measurement of the As species was carried out 3 days after the extraction. Another aliquot of the original extract kept at 4 °C was also measured after 6 days, and it was observed that the peak corresponding to As(III) remained practically the same as the original, whereas both DMA and MMA peaks had diminished more than 50% with respect to the former measurement. 7 days later another aliquot of the extract was analysed, and at that moment no evidence of As(III), DMA and MMA had been found. Fig. 6 shows the superposition of the corresponding chromatograms obtained along time for the soil 3QUEorg. Probably the original species As(III), DMA and MMA have been transformed into As(V), but it was actually very difficult to quantify these possible changes due to the low concentrations of the three compounds when compared with the higher concentration of As(V).

4. Conclusions and recommendations

From the results it is concluded that the extraction yields are dependent on the soil compositions, may be due to the fact that Fe, Mn and Al play an important role on arsenic adsorption in the soils. This study outlines the difficulty in the soil speciation, which mainly lies in the instability of the extract of the soil. Care must be taken so as to ensure the stability of the arsenic species in the soil extracts, and a short period of time between the extraction and the final measurement is highly recommended. The work on arsenic speciation in the soils should be carefully planned and carried out within a short period of time between sampling and measurements, in order to preserve as much as possible the integrity of the species. The quality parameters obtained by

using the coupling LC–UV–HG–AFS reveal that it is extremely adequate for arsenic speciation in the soils to detect both inorganic and organic species. However, according to the quality parameters, it is obvious that the coupling using ICP/MS as detection system is much more sensitive, reaching the ng l^{-1} range in the measurement solution. In spite of this fact, the coupling with final AFS detection, with DLs in the order of the g l^{-1} , is sensitive enough to carry out “routine” arsenic speciation experiments in the soils. No suitable CRMs of the soils and sediments are nowadays available for validating As speciation in such materials. Materials with significant differences in their composition should be desirable. The pseudototal As evaluated from the aqua regia leaching provides good recoveries for assessing the arsenic level in the soils and sediments within a wide range of concentrations.

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Tables

Table 1. Arsenic content in the contaminated soils and in CRMs, after the aqua regia leaching, expressed as mg kg⁻¹.

Material	Arsenic content (mg kg ⁻¹)	SD (n=3)	Certified value (mg kg ⁻¹)	SD (certified)
3RIB	173 ^a	15		
3DEP	19.9 ^c	1.3		
3DEPr	15.4 ^c	1.64		
3QUEorg	712 ^a	1.9		
3QUE	769 ^a	55		
GBW07405	412 ^a	11	412	8
GBW07311	180.7 ^a	13.6	188	6
BCR320	67.3 ^c	3.2	76.7	3.4

a Measurement by ICP–AES.

b Standard deviation (n = 3).

c Measurement by HG–ICP/MS.

Table 2. As species in the CRMs GBW07405, GBW07311 and BCR320, after applying the extraction procedure proposed and measurement with LC–UV–HG–ICP/MS.

CRM	As(III)	DMA	MMA	As(V)	U1	U2	As_extr	(As_extr/As_cert) %
GBW07405	2.5 ± 0.4	n.d.	1.0 ± 0.2	275.1 ± 49.5	n.o.	18 ^a	336.6 ± 4.8	81.7
GBW07311	2.9 ± 0.5	n.d.	n.d.	157.4 ± 23.7	2 ^a	n.o.	190.8 ± 6.7	101
BCR320	7.9 ± 1.5	n.d.	n.d.	35.6 ± 7.1	1 ^a	1 ^a	73.5 ± 1.8	95.8

n.d., below the detection unit; n.o., non-observed; As_extr, total As measured in the extract; As_cert, As certified.

a Estimated value (see text).

b Standard deviation (n = 3).

Table 3. Arsenic speciation in the contaminated soils, after applying the extraction procedure proposed and measurement with LC–UV–HG–ICP/MS, expressed as mg kg⁻¹.

Sample	As(III)	DMA	MMA	As(V)	U1	U2	As_extr	(As_extr/As_aqr) %
2AUTr	0.5 ± 0.1	n.d.	n.d.	7.0 ± 1.4	1 ^a	2 ^a	n.a.	
2DEPr	1.1 ± 0.2	n.d.	n.d.	8.8 ± 1.6	1 ^a	0.5 ^a	n.a.	
2DEP	1.1 ± 0.2	n.d.	n.d.	14.7	1 ^a	0.5 ^a	n.a.	
1RIB2	2.0 ± 0.3	n.d.	n.d.	84.5 ± 13.1	n.o.	n.o.	106 ± 8.4	101
2QUEh	10.2 ± 1.4	n.d.	n.d.	394 ± 60.6	n.o.	n.o.	433 ± 19	56.5
3DEP							13.5 ± 3.9	67.8
3RIB							151 ± 6.3	87.3
3QUE							461 ± 2.6	60.0
3QUEorg	1.04	11.26	17.61	411 ^c			441 ± 2.0	61.9

n.d., below DL; n.a., not available; n.o., non-observed.

a Estimated value.

b Standard deviation (n = 3).

c Calculated by difference.

Figures

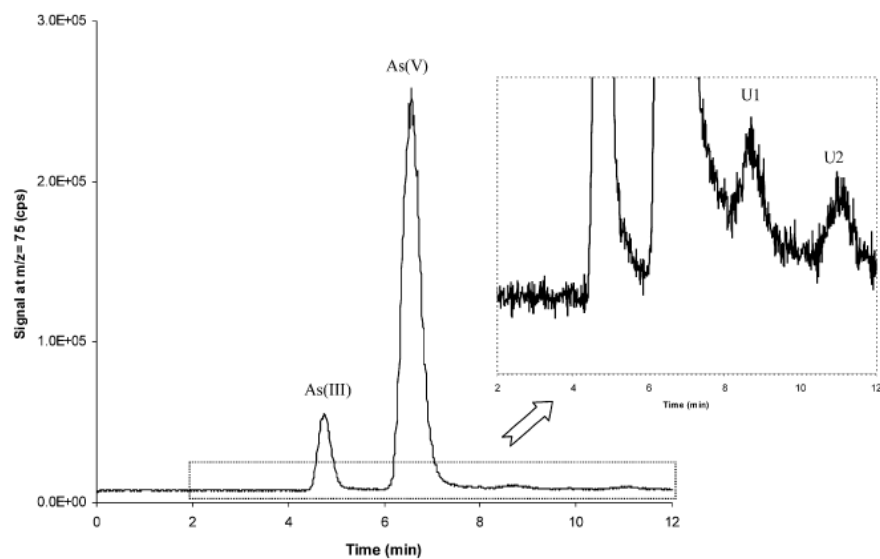


Fig. 1. Chromatogram corresponding to the BCR320 sediment (measured by LC–UV–HG–ICP/MS).

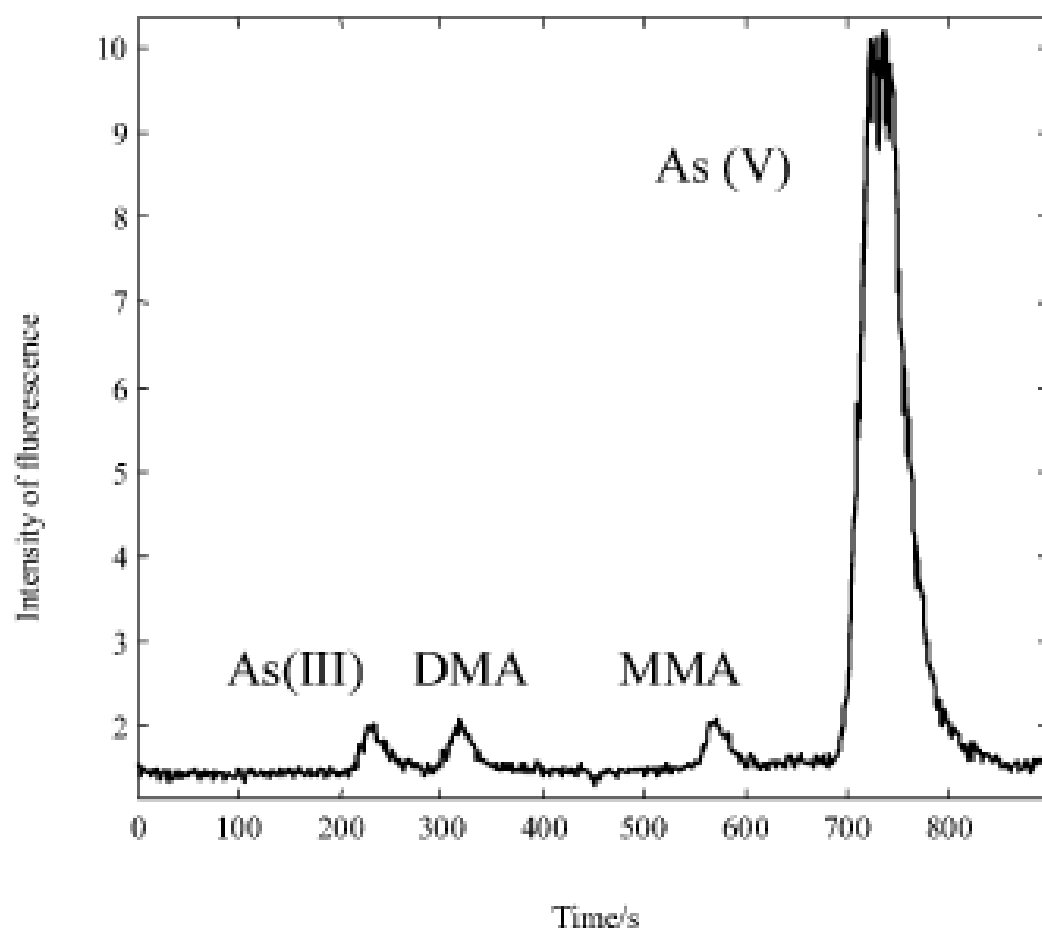


Fig. 2. Chromatogram corresponding to one of the spiked soil extract used to determine the quality parameters measured by LC–UV–HG–AFS.

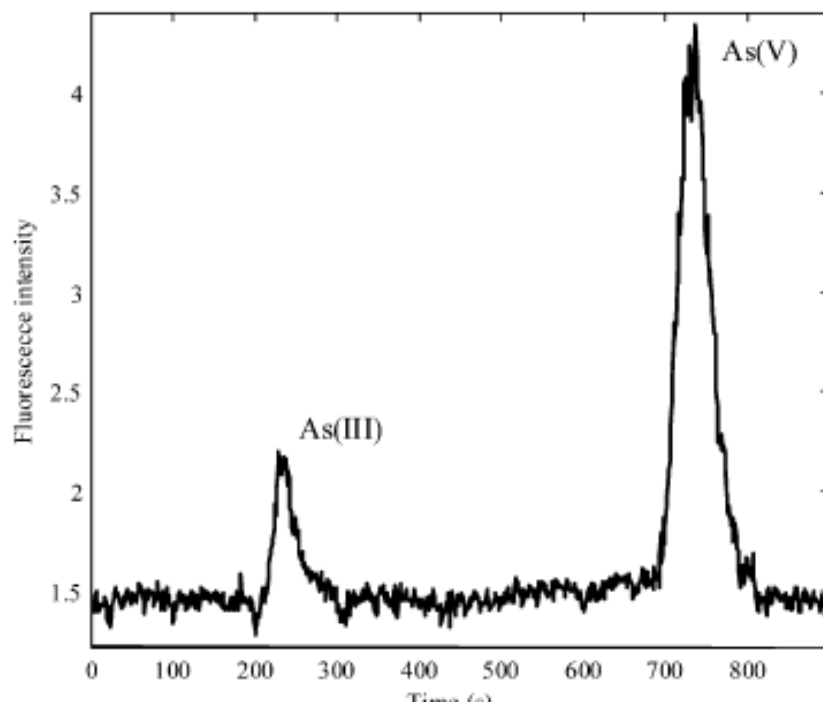


Fig. 3. Chromatogram corresponding to the 3RIB soil (measured by LC–UV–HG–AFS).

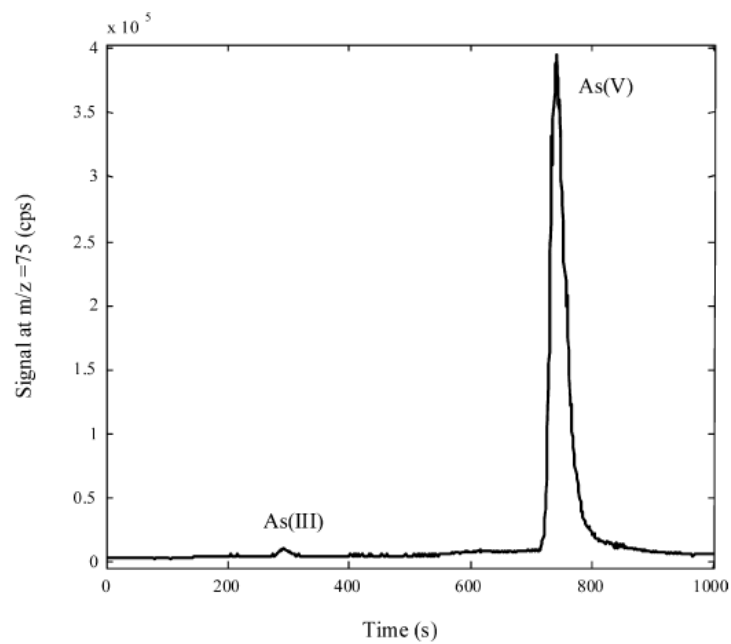


Fig. 4. Chromatogram corresponding to the 3QUE soil (measured by LC–UV–HG–AFS).

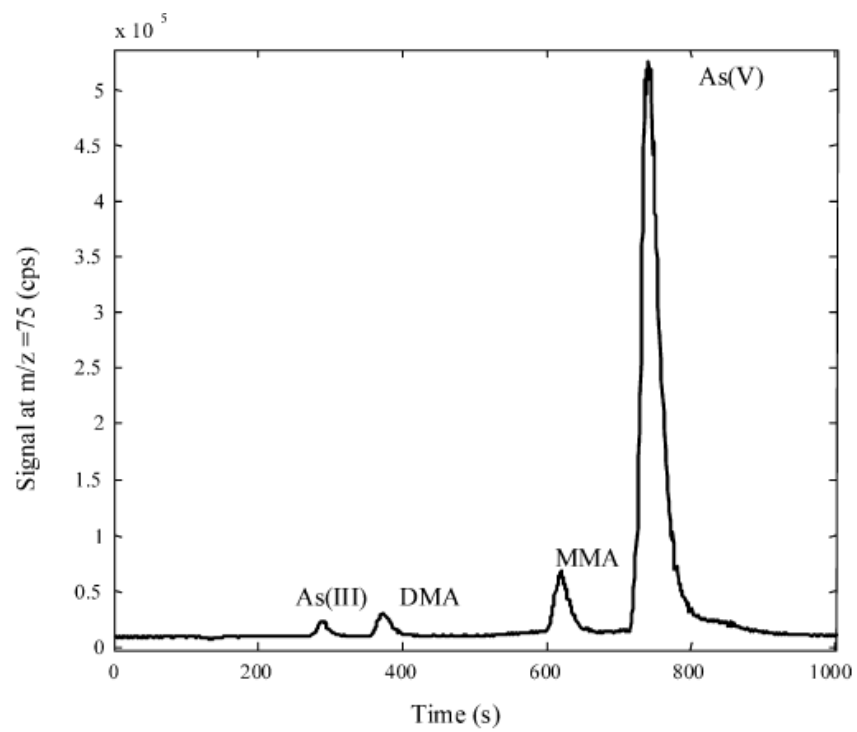


Fig. 5. Chromatogram corresponding to the 3QUE org soil (measured by LC–UV–HG–ICP/MS).

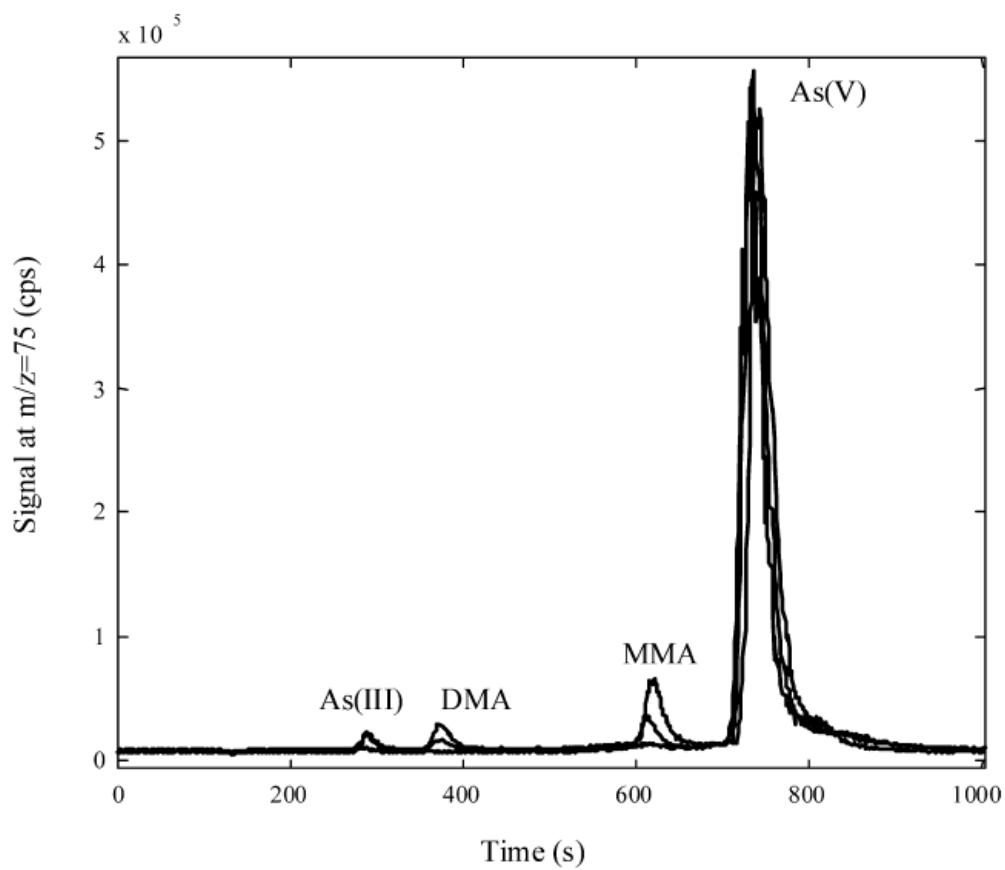


Fig. 6. Sequence of chromatograms corresponding to the soil 3QUEorg, obtained from the measurement of the extract by LC–UV–HG–ICP/MS along the period described in the text. The instability of As(III), DMA and MMA, and their final transformation into As(V) can be observed.