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Speciation of major arsenic species in seawater by flow injection hydride generation atomic absorption spectrometry

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Abstract Arsenic present at 1 μg L^{-1} concentrations in seawater can exist as the following species: As(III), As(V), monomethylarsenic, dimethylarsenic and unknown organic compounds. The potential of the continuous flow injection hydride generation technique coupled to atomic absorption spectrometry (AAS) was investigated for the speciation of these major arsenic species in seawater. Two different techniques were used. After hydride generation and collection in a graphite tube coated with iridium, arsenic was determined by AAS. By selecting different experimental hydride generation conditions, it was possible to determine As(III), total arsenic, hydride reactive arsenic and by difference non-hydride reactive arsenic. On the other hand, by cryogenically trapping hydride reactive species on a chromatographic phase, followed by their sequential release and AAS in a heated quartz cell, inorganic As, MMA and DMA could be determined. By combining these two techniques, an experimental protocol for the speciation of As(III), As(V), MMA, DMA and non-hydride reactive arsenic species in seawater was proposed. The method was applied to seawater sampled at a Mediterranean site and at an Atlantic coastal site. Evidence for the biotransformation of arsenic in seawater was clearly shown.

Introduction

Arsenic is present in seawater at concentration levels around 1 μg L^{-1}. Depending on biological activity, dissolved arsenic is generally distributed between various chemical forms: inorganic As(III) and As(V), monomethylated arsenic (MMA), dimethylated arsenic (DMA) and eventually non-hydride reactive organic compounds [1–2]. As arsenic is involved in biological processes, the determination of total arsenic is not sufficient to study its biogeochemical cycle and eventually its toxicity effects in seawater, therefore, its speciation is necessary. For the speciation of arsenic, a variety of methods have been developed in recent years by coupling hydride generation to different separation and detection systems. Subsequently to hydride generation, the detection has been performed by various techniques like electron capture [3], thermal neutron activation analysis [4], atomic fluorescence [5, 6], quartz or graphite furnace atomic absorption [7–12] or inductively coupled plasma mass spectrometry [13, 14]. Direct GF-AAS leads only to total arsenic and its detection limit in seawater is only about 0.3 μg L^{-1}. Consequently, for the determination and speciation of arsenic, AAS has to be coupled with preconcentration/separation systems permitting the use of larger samples. Among the different possible systems, hydride generation (HG) devices (batch or continuous flow injection systems) have been generally used. Some experimental protocols based on selective HG efficiency of arsenic species led to a partial speciation of hydride reactive species [3, 4, 8, 9, 12, 15, 16] or eventually showed the presence of non-hydride reactive species [2, 10, 17, 18]. On the other hand, different batch experimental protocols coupling HG systems with a cryogenic chromatographic trap permitted to access methylated species, but non-hydride reactive species were not determined [5, 19–21]. In this work, the potential of a commercial continuous flow injection system was examined to access a complete speciation of the major arsenic species in seawater by combining two complementary techniques. In the first step, arsenic was preconcentrated by collection of the hydride species into the furnace before AAS; this technique led to a partial speciation based on the selective HG efficiency of the arsenic species present in seawater namely the hydride reactive species, and after an oxidative UV irradiation treatment the non-hydride reactive species. A second technique was used by coupling the HG system to an electrically heated quartz furnace; in this case arsenic hydride species were trapped in a cryogenic chromatographic trap, then sequentially released by heating before AAS. Such a system permitted the separation of the different hydride species: AsH_3,

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CH_3AsH_2 and $(CH_3)_2AsH$. As an analytical application, the speciation of arsenic in seawater sampled at an Atlantic coastal site during a phytoplankton bloom and on a depth water profile at a Mediterranean site was determined.

Experimental

Reagents. Sodium borohydride solutions were prepared daily from Acros products. Suprapur hydrochloric acid, acetic acid and sodium hydroxide from Merck were used. Potassium peroxodisulfate, tris(hydroxymethyl)-aminomethane (TRIS) pro analysi (Merck) were used. Suprapur 25% solution ammonia (Merck) was used. The solutions of As(III), MMA and DMA were obtained from sodium arsenite (Aldrich), sodium monomethylarsonate (Carlo Erba) and dimethylarsinic acid sodium salt hydrate (Acros), respectively. A 1 g L^{-1} H_3AsO_4 in 0.5 M HNO_3 standard solution (Merck) was used for As(V). Mediterranean and Atlantic seawater samples were collected with acid-cleaned Niskin samplers. The unfiltered samples were transferred to acid-cleaned polyethylene bottles and stored in a refrigerator at 4 °C until analysis.

Spectrometer. Measurements were performed with a 4100ZL Perkin-Elmer spectrometer equipped with an AS-70 autosampler. The light source used was an As Perkin-Elmer electrodeless discharge lamp working at about 480 mA. The analytical line was set at 193.7 nm with a 2 nm slit width.

Flow injection hydride generation system. The flow injection system was a Perkin-Elmer FIMS apparatus; it was used according to the schematic diagram presented in Fig. 1. Samples and hydrochloric acid solutions were continuously pumped at about 1 mL min^{-1} and a 40 g L^{-1} tetrahydroborate solution at about 2 mL min^{-1}. For As(III) determinations, a 10 g L^{-1} $NaBH_4$ solution and a 1 M Tris-HCl solution mixed on-line with the seawater sample were continuously pumped at 2 mL min^{-1}. A 2 m reaction coil before the Perkin-Elmer quartz gas/liquid separator was used.

In situ trapping. A Perkin-Elmer quartz capillary was mounted in place of the PTFE capillary tubing of the sample dispenser arm. This modification permitted injection into the hot furnace of the gaseous hydride. Perkin-Elmer end-capped tubes with integrated platforms were used. 30 µL of a 0.01 M H_2IrCl_6 solution (ABCR product, Roth- Sochiel) were deposited onto the atomizer and pretreated at 1200 °C during 30 s. The operation was repeated 3 times. Argon was used as sheath gas.

Operating conditions were as follows: the arsenic hydrides carried with the argon flow were collected into the atomizer at 300 °C. Then, the atomization took place at 2100 °C in the maximum power mode with an interrupted gas flow. A cleaning step at 2500 °C during 3 s with 250 mL min^{-1} argon was then carried out.

Cryogenic trapping on a chromatographic phase. Measurements were performed with a Perkin-Elmer electrically heated quartz cell (900 °C). The carrier gas was argon. A U-shape tube (30 cm, 0.5 mm inner diameter) filled with about 0.7 g OV-1 chromatographic phase (1%, 80–100 mesh) on Chromosorb G packing material was used. This trap, wrapped with an electrical wire (1.5 m, 30 Ω), was immersed in liquid nitrogen during 60 s (–196 °C), than electrically heated (30 V, 1 A). The arsenic hydrides eluted according to their boiling points and were detected in the heated quartz cell.

UV irradiation treatment. A Teflon tube (3 m length; 0.6 mm inner diameter) was coiled around the mercury lamp of the FIMS system. To evaluate the efficiency of this UV-irradiation system, we used a Gilson peristaltic pump out-of-line to modify the flow-rate (i.e. residence time); after UV irradiation, 4 mL of the sample were collected in a tube and As was subsequently determined by hydride generation electrothermal atomic absorption spectrometry after addition of 1 mL acetic acid. The % transformation of MMA and DMA to As(V) was calculated from the signals obtained for these species and As(V) in 20% acetic acid medium.

Results and discussion

Dissolved arsenic is mainly distributed in seawater between As(III), As(V), MMA, DMA and unknown organic species. Depending on the experimental conditions used, all these species do not generate the corresponding hydride species with a similar efficiency. Consequently, total arsenic concentration can be determined only if all arsenic species give a similar analytical response or if they can be transformed to species giving a similar analytical response. On the other hand, individual arsenic species can

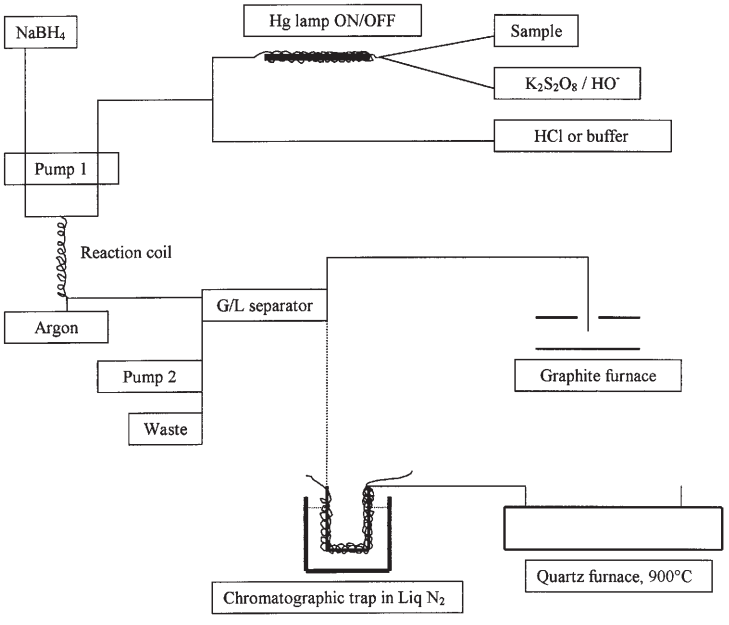

Fig. 1 Schematic diagram of the FI-HG graphite furnace and cryogenic chromatographic trap QF-AAS system

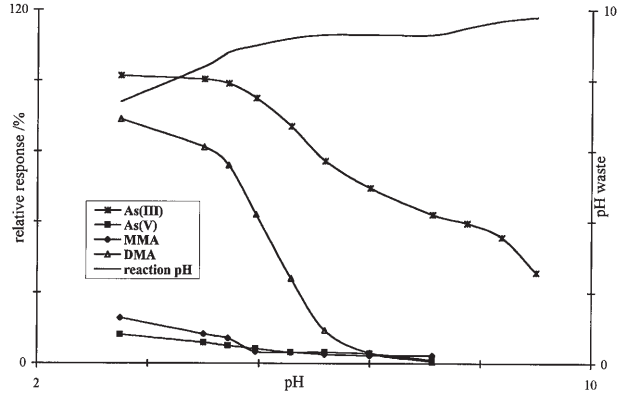

Fig. 2 Influence of the pH of a 1 M acetic acid/NH_3 buffer solution on the generation of the different arsenic hydride species ($NaBH_4$ 10 g L^{-1})

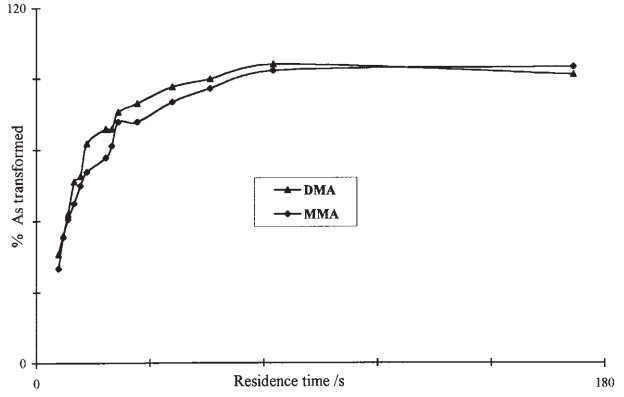

Fig. 3 Influence of the on-line UV irradiation time on the transformation of DMA and MMA to As(V) in seawater in 0.1 M $K_2S_2O_8$ / 1 M NaOH

be determined only if they are separated before their detection or if they give a typical analytical response under selective hydride generation experimental conditions.

1. Flow injection hydride generation in situ trapping

Determination of As(III). As shown in Fig. 2, the efficiency of HG is dependent on the nature of arsenic species and on experimental conditions, particularly on the pH of the reacting medium. It has been previously shown that As(III) could be determined alone in seawater in a mixture of As(V) and As(III) at pH about 5. However, Fig. 2 shows that dimethylarsenic can interfere with the determination of As(III) at pH about 5. Consequently, higher pH values have to be used for the determination of As(III) alone when DMA is significantly present in seawater. Using a 0.5 M TRIS-HCl buffer adjusting the reaction pH to about 8, As(III) could be determined in a mixture of As(V), DMA and MMA with a signal ratio of As(III) to As(V), MMA and DMA, 1150, 650 and 55, respectively. Therefore, these experimental conditions allow to determine As(III) alone in most of the samples in a mixture of these arsenic species. Its detection limit is about 2 ng L^{-1} for a 10 mL sample volume (3 σ, ten measurements).

Determination of hydride reactive, total and non-hydride reactive arsenic. If As(III), As(V) and methylated species are the major species present in seawater that generate hydride species, other organic species that do not generate hydride species could be present in seawater [2, 10, 17, 18]. Consequently, total arsenic can only be determined after a treatment of the seawater sample transforming all the arsenic species to hydride reactive species.

This could be achieved by using an on line UV irradiation procedure after mixing the seawater sample to an 1 M NaOH / 0.1 M persulfate solution. This alkaline medium has been shown to be more efficient than the neutral persulfate medium to decompose the organoarsenicals to inorganic arsenic. Indeed, in neutral medium, arsenic compounds, like methylated species, were not decomposed

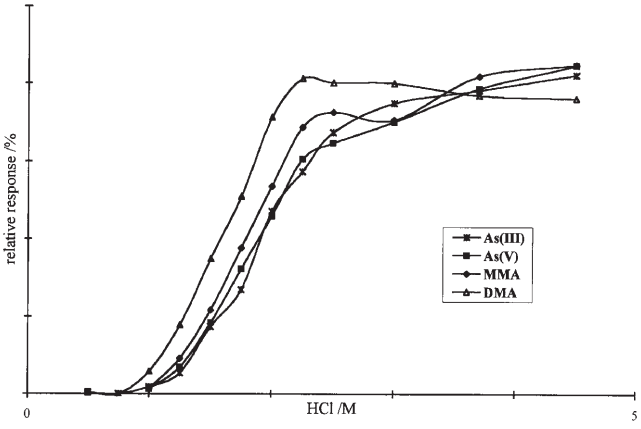

Fig. 4 Influence of the HCl concentration on the analytical response of arsenic species in seawater in the alkaline persulfate medium ($NaBH_4$ 40 g L^{-1})

and compounds, like arsenobetaine, were incompletely decomposed to a mixture of inorganic As(V) and methylated species [22]. Figure 3 confirms these observations and shows that the UV irradiation treatment used was very efficient to transform methylated species to only one inorganic species, As(V), whilst in the neutral medium no decomposition of these species was observed. Moreover, this transformation was very rapid and permitted the use of this treatment on-line coupled to continuous flow injection hydride generation. By assuming a similar decomposition process for unknown organic species in seawater like for arsenobetaine, the UV decomposition procedure in an alkaline persulfate medium leads also only to As(V) and, consequently probably to a better precision in the determination of total arsenic than in a neutral medium.

As shown in previous papers, As(III) and As(V) or methylated species could generate hydride species in seawater with similar efficiency for both high $NaBH_4$ and HCl concentrations [6, 10]. After mixing seawater with a 1 M NaOH / 0.1 M persulfate solution, hydride generation was performed at a relatively high 40 g L^{-1} $NaBH_4$ concentration. As shown in Fig. 4, hydride generation effi-

ciency was also dependent on HCl concentration and on the nature of the arsenic species. However, in the alkaline persulfate medium, hydride species were produced with a similar efficiency from As(III), As(V), MMA and DMA for a HCl concentration above 3 M. The similarity of the As(III) and As(V) curves was due to the fact that As(III) was completely oxidized to As(V) during the experimental time. Therefore, this medium appears interesting to determine total arsenic in a mixture of As(III) and As(V) because it leads only to As(V) and minimizes errors of calibration. As the analytical response was also similar for methylated species, total hydride-reactive species could be determined in seawater using these experimental conditions with an As(V) standard solution. When the seawater sample mixed with the alkaline persulfate solution was submitted to UV irradiation, all arsenic species were decomposed to only inorganic As(V). Consequently, total arsenic can be also determined by using an As(V) standard solution. The detection limits for total and hydride-reactive species are about 5 ng L^{-1} for a 2.5 mL seawater volume (3 σ, 10 measurements). The difference between these two determinations gives the non-reactive hydride arsenic concentration.

2. Flow injection hydride generation cryogenic trapping chromatography

Determination of MMA and DMA. Coupled to hydride generation, cryogenic trapping on a chromatographic phase permits the preconcentration of the different arsenic hydrides and, by heating the chromatographic trap the sequential release of the arsenic hydrides into a detector. In this work, the FIMS Perkin-Elmer flow injection system coupled to a Perkin-Elmer electrically heated quartz cell was used instead of the graphite furnace, the Perkin-Elmer 4100ZL spectrometer used as detector (Fig. 1). Arsenic hydrides were trapped on a cold chromatographic OV-1 column (–196 °C), then released by heating. The retention time, the separation and the magnitude of the analytical signals of AsH_3, $MMAH_2$ and DMAH were highly dependent on the mass of Chromosorb, the cooling time, the carrier gas flow and the heating power. Because the data acquisition system of the Perkin-Elmer 4100ZL had a maximum read time of 50 s, all arsenic hydrides had to be released and separated during this period. This could be achieved with 0.7 g Chromosorb, one minute cooling time and at 30 V, 1 A heating and a carrier gas flow fixed to about 100 mL min^{-1}. In Fig. 5, the chromatogram obtained for a mixture of As(V), MMA and DMA using these experimental conditions is presented. Peak heights could be directly determined with the commercial Perkin-Elmer software by clicking on the maximum of each peak. However, a slight drift of the baseline was generally observed during the 50 s measurement time inducing important errors at low concentration levels. Therefore, the Perkin-Elmer data were transformed to ASCII files into a PC Microsoft Windows 95 software permitting a base-line correction for each different species. Peak area measurements, less subject to experimental condition variations, were used. The reproducibility of peak area measurements for all arsenic species was about 2.5%. The detection limit obtained for MMA and DMA was about 1.5 ng L^{-1} for a 2.5 mL seawater volume (3 σ, 10 measurements). The chromatograms obtained, by using our protocol, for a surface and a bottom seawater samples collected in Mediterranean Sea in December 1997 at Lat. 36.04.518N, Long. 01.35.991W, are presented in Fig. 6.

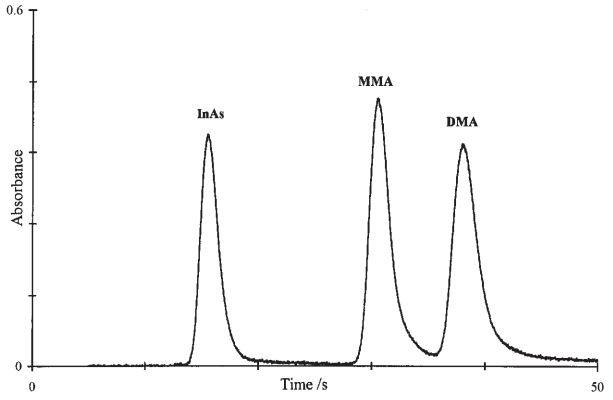

Fig. 5 Typical chromatogram of hydride arsenic species in seawater. 5 ng of inorganic As, MMA and DMA. Ar flow ~ 100 mL min^{-1}, 30 V heating voltage, 1 A

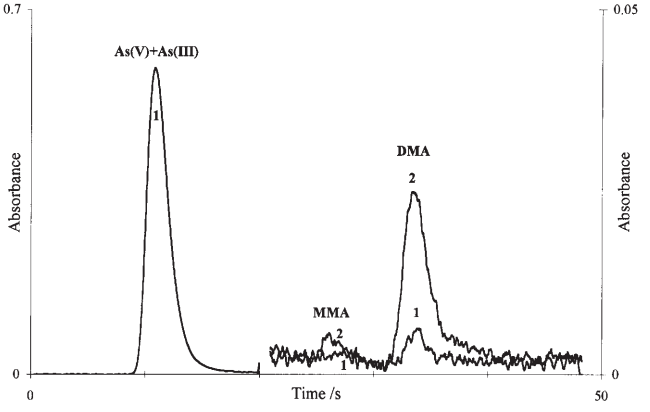

Fig. 6 Typical chromatograms of Mediterranean seawater samples (2.5 mL): (1) bottom seawater; (2) surface seawater (~ 100 mL min^{-1} argon, 30 V heating voltage, 1 A)

We could not determine MMA and DMA in certified seawater samples because we observed an interference effect using our experimental conditions. This effect was attributed to the presence of HNO_3 (~ 2.5×10^{-2} M) in the CASS-2 and SLEW-1 samples. Indeed, as observed in Fig. 7, the analytical response obtained for DMA dramatically decreased with increasing concentrations of HNO_3. The analytical responses of AsH_3 and MMA were not subject to similar interference effects. This interference could lead to significant errors in the determination of DMA and, consequently in the speciation of arsenic in seawater. Therefore, it appears that the use of HNO_3 as a preservative is not adequate when using this technique; this medium is further not recommended for the preser-

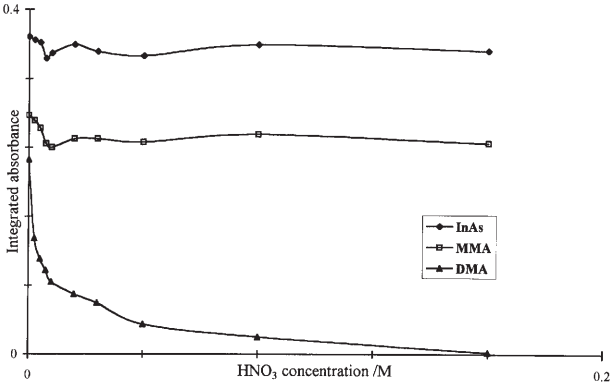

Fig. 7 Interference of nitric acid on the signal of inorganic As, MMA and DMA in seawater

vation of species due to the oxidation of As(III) to As(V) [23]. It can be noted that the cold trapping technique can be used for the determination of As(III) by performing hydride generation at a pH about 7. This technique can be also eventually used, in the presence of high HCl and $NaBH_4$ concentrations, for the determination of total arsenic with a preliminary alkaline persulfate UV irradiation treatment to transform all arsenic species to inorganic As(V). However, the determination of non-hydride reactive arsenic species is less precise using this protocol than the in situ collection GF-AAS technique, because non-hydride reactive arsenic concentration is obtained by the difference between total arsenic and the sum of three concentrations [As(III)+As(V)]+[MMA]+[DMA] and, consequently the sum of the corresponding errors. Moreover, continuous flow injection hydride generation in situ collection GF-AAS measurements are entirely automated and, consequently more reproducible.

3. Speciation protocol

From this study, a protocol based on four experiments can be proposed for the speciation of arsenic in seawater.

Three experiments based on continuous flow injection hydride generation, in situ collection, followed by GF-AAS

- HG in 10 g L^{-1} $NaBH_4$ and TRIS-HCl, pH~7 → [As(III)]
- seawater mixed with alkaline persulfate HG in 40 g L^{-1} $NaBH_4$ and ~ 3 M HCl → [As(V)+As(III)+DMA+MMA]
- UV irradiation in alkaline persulfate HG in 40 g L^{-1} $NaBH_4$ and ~ 3 M HCl → [As(V)+As(III)+DMA+MMA+NHR-As]

One experiment based on continuous flow injection hydride generation, cryogenic chromatographic trapping, followed by GF-AAS

- HG in 40 g L^{-1} $NaBH_4$ and ~ 2 M HCl → [As(III)+As(V)], [MMA], [DMA]

From these four experiments, the different individual arsenic species: As(V), As(III), MMA, DMA and non-hydride reactive arsenic species can be generally determined in seawater.

4. Application

The variation of the arsenic speciation in Atlantic seawater samples collected near Brest in April-May 1999 during a phytoplankton bloom is presented in Fig. 8. The variation of the chlorophyll concentration measured during this period is also presented. As clearly shown, before the phytoplankton bloom practically all arsenic is occurring as inorganic As(V). Then, during the phytoplankton bloom all inorganic As(V) was converted to inorganic As(III) and to about 20% DMA; MMA representing only

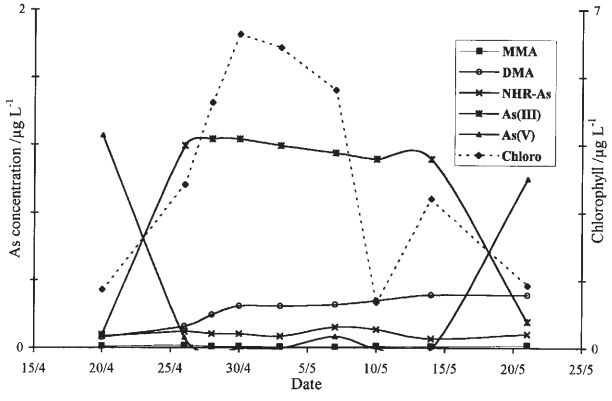

Fig. 8 Distribution of major arsenic species at a coastal Atlantic station observed during a phytoplankton bloom

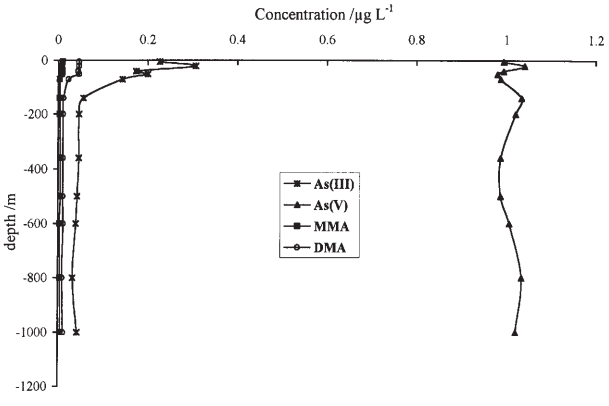

Fig. 9 Depth distribution of major arsenic species at a Mediterranean station

about 3% whilst non-hydride reactive species were relatively constant at about 10% of total arsenic. After the phytoplankton bloom, all As(III) was rapidly reoxidized to inorganic As(V). It could be noted that the DMA and MMA concentrations did not decrease after the phytoplankton bloom showing their high stability in seawater.

As an another example, the speciation of arsenic in a Mediterranean depth profile was determined. The variation of arsenic speciation in the water column confirmed the upper observations (Fig. 9). Indeed, in the surface seawater < 200 m with a higher biological activity, biotransformation of arsenic was evidenced by the presence of a significant proportion of As(III) (30%) and the simultaneous presence of DMA (5%). MMA at the 1 ng L^{-1} concentration level and non-hydride reactive arsenic species were not detected in these seawater samples. All these observations are in good agreement with laboratory studies showing that As(V) could be taken up by phytoplankton and subsequently released as As(III), MMA and DMA [24–28].

Conclusion

By combining the analytical responses obtained by selective flow injection hydride generation, collection in a graphite furnace or cryogenically trapping on a chromatographic column followed by AAS, an experimental protocol for the speciation of the major arsenic species, As(V), As(III), MMA, DMA and non-hydride reactive arsenic at the ng L^{-1} concentration level, in seawater was proposed. Using this analytical protocol, the important modifications of the distribution of arsenic species in seawater observed during a phytoplankton bloom were shown. The results confirmed clearly the evidence of the in situ biotransformation of arsenic and particularly the importance of speciation modifications in the presence of biological activity.

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