Speciation of arsenic in fish products by LC-ICP/MS

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Introduction

While many elements are considered essential to human health, others can be toxic. However, because the intake, accumulation, transport, storage and interaction of these different metals and metalloids in nature is strongly influenced by their specific elemental form, complete characterization of the element is essential when assessing its benefits and/or risks. This is why, elemental speciation, which typically involves the coupling of a separation technique to a specific detector, represents the technique of choice to determine the different forms of an element.

Arsenic speciation is of particular interest in biological, food and environmental specimens. It is currently considered that the total amount of arsenic ingested by humans depends on the amount of seafood included in the diet [1]. However, the high arsenic content detected in seafood products, of the order of micrograms per gram, is not subjected to legislative control in most countries.

Furthermore, when arsenic content is found in levels close or higher than the admissible values, it is of interest to identify which of the toxic or nontoxic form of arsenic is present. This is particularly true for seafood products, given that arsenobetaine is commonly the major arsenic form present [2-3].

Following our first experience with speciation of arsenic in hair [4-5], the purpose of this study was to validate our method for prawn paste by LC-ICP/MS in order to identify and quantify carcinogen inorganic forms, arsenite As(III), arsenate As(IV), monomethylarsonic acid (MMAA) and dimethylarsinic acid (DMAA) and relatively nontoxic organic arsenobetaine (AsB), respectively.

Analytical aspects

Material

Deionized water produced by a Direct-Q3 from Millipore (Molsheim, France) was used for the preparation of reagents and standards.

Suprapur nitric acid (65%) and analytical grade Triton X-100 were purchased from Elvetec (Heillecourt, France).

As(III), As(IV), MMAA, DMAA and AsB were purchased from Fluka (Saint Quentin Fallavier, France). Standards were prepared by dissolving appropriate amounts of commercially available salts in water.

Hamilton cations (PRP-X200, 250 x 4.1 mm, 10µm) and anions (PRP-X100, 250 x 4.1 mm, 10µm) exchange columns were purchased from Chromoptic (Courtaboeuf, France).

Analyses were achieved on a TSP chromtographic system coupled to a Thermo Elemental X7 Series ICP/MS.

Prawn paste specimens were provided by an industry processing seafood products.

Methods

First, total arsenic concentration was measured using ICP/MS after mineralization of prawn paste (1.5 g) in 10 ml concentrated nitric acid (65%) for 90 min at 75°C. After dilution (1/40) in 1% nitric acid and 0.01% Triton X100, acquisition of the specific m/z value (75As) for arsenic was performed by ICP/MS. Quantification, based on external calibration, was done using Rhodium (113Rh) as internal standard.

Speciation of arsenic was achieved using HPLC-ICP/MS after two successive extraction of prawn paste (1.5 g) with 10 ml water for 20 min in an ultrasonic bath. In contrast with hair specimen, an additional step was necessary for fish products to isolate high amounts of arsenobetaine from other arsenic species.

After filtration of the water extract, purification of inorganic species and quantification of AsB were achieved on a cations exchange PRP-X200 column (250 x 4.1 mm, 10µm) using a mobile phase with a pH gradient ranging from 1.6 to 9.0 in 10 min.

Inorganic species were isolated in the fraction collected between 1.5 and 2.5 min while AsB was identified and quantified at 3 min.

In a second step, separation of As(III), As(IV), MMAA and DMAA was obtained on an anions exchange PRP-X100 column (250 x 4.1 mm, 10µm) . The 12.5 mM phosphate buffer 3% MeOH (pH 8.5) mobile phase flow rate was of 1.5 ml/min.

Arsenic species were identified based on their specific retention times and the specific m/z value (75As) for arsenic using ICP/MS. Quantification was done using an external calibration. Under these conditions, the principal inorganic species, arsenite, arsenate, DMA and MMA were separated in 10 minutes with a good resolution (Fig. 1).

Results and discussion

Detector linearity was checked for each compound between 0.5 and 20 µg/L, with coefficients of correlation always higher than 0.9999 (n = 5).

Relative extraction recovery of the speciation method was 85% using the total arsenic determination by mineralization as reference. The limits of quantification were fixed from 0.6 µg/L for arsenite to 1.6 µg/L for arsenate.

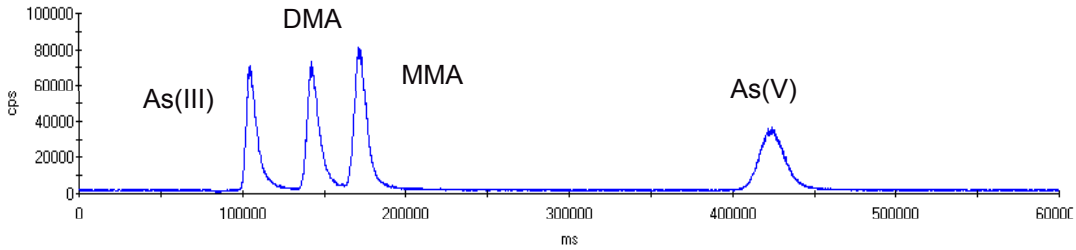

Fig. 1. Typical chromatogram of the principal inorganic species separated in 10 minutes (final concentration of 10µg/L).

The analysis of prawn paste samples revealed the presence of the inorganic form AsB in all the specimens with concentrations ranging from 27.24 to 61.47 µg/g. These results were in accordance with those previously published by Suner and coll. (2) who reported concentrations ranging from 0.3 and 104.1 µg/g, and confirmed that Asb represents the major part (72 and 99%) of the arsenic species content in seafood products.

Toxic As(III) concentrations of ranged from 0.01 to 0.22 µg/g (0.01 to 0.35%), from 0.03 to 0.09 µg/g (0.03 to 0.52%) for As(V), and from not detected to 0.05 µg/g (ND to 0.06%) for the metabolites MMAA and DMAA, in prawn paste samples, respectively. Suner and coll. (2) reported that the metabolites did not appear in all their analyzed specimens (concentrations : ND to 0.5 µg/g).

Conclusion

The proposed method for the speciation of arsenical species, adapted from our previous procedure dedicated to hair specimens, enables the identification and quantification of arsenite, arsenate, monomethylarsonic acid, dimethylarsinic acid and arsenobetaine in prawn paste specimens by LC-ICP/MS.

The analysis of prawn paste specimens revealed that only less than 1 % of the highly toxic mineral forms As(III) and As(V) were present.

References

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