CONffIDENCE: Contaminants in food and feed: Inexpensive detection for control of exposure

Determination of inorganic arsenic by MAE-SPE-HG-AAS
– a simple and inexpensive speciation alternative

Rikke V. Hedegaard and Jens J. Sloth*
National Food Institute, Technical University of Denmark
Division of Food Chemistry, Mørkhøj Bygade 19, DK-2860 Søborg, Denmark
*Corresponding author: jjsl@food.dtu.dk

Simple, inexpensive and fast methods for determination of the toxic inorganic arsenic species are called upon for the monitoring and control of food and feed samples. A simplified approach based on Microwave-Assisted Extraction (MAE) - Solid Phase Extraction (SPE) – has been developed, where inorganic arsenic is pre-separated from organoarsenic compounds by Strong Anion Extraction (SAX) SPE followed by determination of arsenic content by Hydride Generation (HG) Atomic Absorption Spectroscopy (AAS).

FIGURE 1. Principle of the simplified MAE-SPE-HG-AAS approach for selective speciation analysis of inorganic arsenic.

Introduction

The total contents of the trace elements lead, cadmium and mercury in food and feed are regulated by EU directives (EC) Nos 466/2001/EC, 78/2005/EC, 2002/32/EC and 2003/100/EC. However, for some heavy metals the chemical form (i.e. their speciation) is important in terms of food and feed safety. More than 50 different arsenic species have been found in the marine environment – including lipid-soluble arsenic compounds, however it is the ***inorganic arsenic*** **forms** that are most toxic. Since seafood is the major dietary source for arsenic exposure in the European population, marine feed and seafood are of interest.

Extraction of inorganic arsenic

Several sample extraction solvents and samples preparation approaches have been tested for extraction of inorganic arsenic in order to optimize the extraction of inorganic arsenic. These include of water, methanol, hydrochloric acid or alkaline solutions, all giving with varying results for the same reference material. Microwave assisted extraction for 20 minutes at 90 C with 0.07 M HCl/3 % H_2O_2 provided the most efficient extraction of inorganic arsenic. H_2O_2 was added to ensure quantitative conversion of arsenite As(III) to arsenate As(V) (Figure 2) and thereby facilitate the following SPE separation of inorganic As (as As(V)) from organoarsenic compounds. Importantly, no degradation/conversion of other arsenic species such as arsenobetaine (AB), which is usually the predominant species in fish, methylarsonate (MA) or dimethylarsinate (DMA) was observed under the chosen conditions.

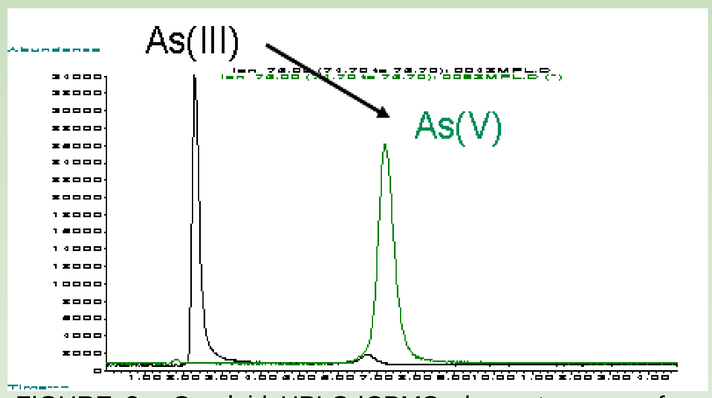

FIGURE 2. Overlaid HPLC-ICPMS chromatograms of an As(III) standard solution (10 ppb) in 0.07 M HCl / 3 % H_2O_2 before and after microwave treatment, respectively, showing the quantitative conversion of As(III) to As(V) by H_2O_2.

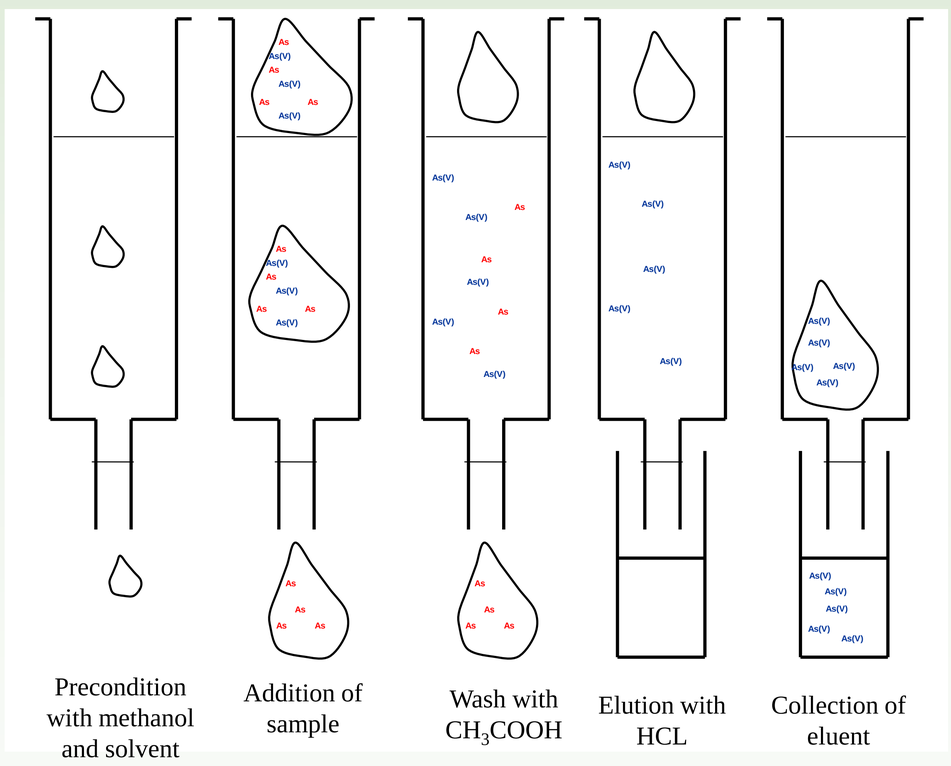

FIGURE 3. Separation of inorganic arsenic (as As(V)) from organoarsenic forms (As) by SAX SPE. The columns are preconditioned with methanol and afterwards with the sample solvent sample before loading the sample. The organoarsenic compounds are coeluted with CH_3COOH prior to the elution of inorganic arsenic with HCl .

Selective separation of inorganic arsenic

Following extraction of the sample the separation of inorganic arsenic in the form of As(V) (pKa ~ 2.3/6.7/11.6) from organoarsenic compounds is done by the use of a silica-based Strong Anionic Extraction (SAX) SPE column (from Phenomenex or ThermoFisher). Standards of As(V) were quantitatively retained on the SAX SPE column at pH 6. However, at pH 6 other organicarsenic compounds commonly found in marine products such as MA (pKa ~3.6/8.2), DMA (pKa ~ 1.3/6.3)) and AB (pKa 2.2) were also retained on the SAX SPE column, with approximately 80, 52 and 10 %, respectively. It was possible to selectively elute MA, DMA and AB with CH_3COOH prior to elution of inorganic arsenic with 1M HCl without loss of As(V) and thereby enable separation of As(V) from the organoarsenic compounds by SPE. The principle of the SPE method is shown in Figure 3.

Detection by HG-AAS

An ICE 3300 Atomic Absorption Spectrometer from Thermo Scientific (Picture 1) is used for the determination of the arsenic content in the eluent from the SPE separation. The sample introduction system to the AAS consist of an 250 Auto sampler (CETAC Technologies, Omaha, NE, USA) and a VP100 Continuous Flow Vapour Generator. The samples are prereduced with potassium iodide and ascorbic acid reducing As(V) to As(III) prior to analysis, due to the better hydride generating capacity of As(III). The 0.5 % $NaBH_4$ in 0.5 % NaOH and 6 M HCl reagents were used in the continuous flow hydride generation system. In samples such as fishmeal and the CRM TORT-2 (*lobster hepatopancreas*) recoveries of inorganic arsenic 90-100 % were found compared to analysis by a HPLC-ICPMS reference method.

PICTURE 1. ICE 3300 AAS from Thermo Scientific with a VP100 for hydride generation system and an electrically heated cell.

The research leading to these results has received funding from the European Community's Seventh Framework Programme (*FP7/2007-2013)* under *grant agreement* n 211326.