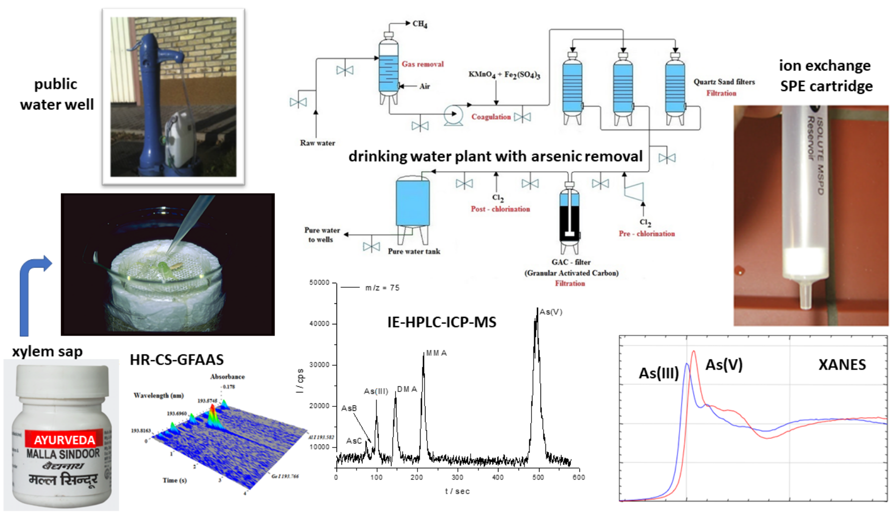
Brazilian Journal of Analytical Chemistry
2022, Volume 9, Issue 36, pp 14-51
doi: 10.30744/brjac.2179-3425.RV-119-2021

REVIEW

Regional and Global Scale Challenges for Controlling Arsenic Contamination in Agricultural Soil, Water Supplies, Foods and Ayurvedic Medicines

Victor G. Mihucz

ELTE – Eötvös Loránd University, Institute of Chemistry, H-1117 Budapest, Pázmány Péter stny 1/A, Hungary



Arsenic is naturally present at high concentration levels in aquifers adversely affecting the life of some 200 million people in a number of countries on four continents. Human exposure to As from dietary sources such as marine fish, seafood, poultry, cereals is generally much lower compared to exposure through drinking contaminated water, using contaminated water in food preparation and irrigation of crops. Arsenic toxicity depends on its four valences [As(-III), As^0, As(III) and As(V)] and chemical compounds. Thus, in seafood, As is mainly found in its less toxic organic forms. The qualitative and quantitative determinations of individual As species are crucial to understand the environmental fate and behavior of As. The aim of the present review is to give a brief overview on the main As speciation methods and to present how to control As contamination at local and global scales in several environmental (soil, waters) and biological (crops, basic and processed food) samples, as well as complementary and alternative medicinal products marketed as food supplements. In terms of chromatographic separation, emphasis is placed on separation by thin layer chromatography and solid phase extraction. Some approaches to address As contamination (e.g., stabilization in soil, provision of a safe water supply in affected communities) at global and regional scales are also presented.

Keywords: arsenate, arsenite, drinking water, organic arsenicals, speciation

Cite: Mihucz, V. G. Regional and Global Scale Challenges for Controlling Arsenic Contamination in Agricultural Soil, Water Supplies, Foods and Ayurvedic Medicines. *Braz. J. Anal. Chem.* 2022, *9* (36), pp 14-51. http://dx.doi.org/10.30744/brjac.2179-3425.RV-119-2021

Submitted 30 November 2021, Resubmitted 23 January 2022, Accepted 6 February 2022, Available online 29 March 2022.

Abbreviations used in this Review

Acronym	Meaning	Acronym	Meaning
AC	activated charcoal	K_a	acid dissociation constant
AsB	arsenobetaine	LA	laser ablation
AsC	arsenocholine	LC	linear combination
ASE	accelerated solvent extraction	LD_{50}	median lethal dose
ASHRAM	Arsenic health risk assessment and molecular epidemiology	LMWOA	low molecular weight organic acid
ATR-FTIR	attenuated total-reflection Fourier—transformed infrared spectroscopy	LOD	limit of detection
BCR	Bureau Communautaire de Référence or (European) Community Bureau of Reference	MMA(III)	monomethylarsonous acid
BMDL	benchmark dose limit	MMA(V)	monomethylarsonic acid
BW	body weight	MOE	margin of exposure
CAM	complementary and alternative medicine	MW	microwave
DMA(III)	dimethylarsinous acid	NIST	National Institute for Standards and Technology
DMA(V)	dimethylarsinic acid (cacodylic acid)	OPLC	overpressured layer chromatography
DSHEA	Dietary Supplement Health and Education Act	PEI	polyethylene imide
DW	deionized water	PTFE	polytetrafluoroethylene
EC	European Commission	Q-ICP-MS	quadrupole inductively coupled plasma mass spectrometry
EDTA	ethylene diamine tetraacetic acid	R_f	retention factor
EDXRF	energy dispersive X-ray fluorescence	RSD	relative standard deviation (%)
EXAFS	extended X-ray absorption fine structure	SAM	*S*-adenosyl methionine
FAO	Food and Agriculture Organization of the United Nations	SAX	strong anion exchanger
FIA	flow injection analysis	SPE	solid phase extraction
FRW	fresh root weight	SRM	standard reference material
FTIR	Fourier-transformed infrared spectroscopy	SR-XANES	synchrotron radiation X-ray absorption near edge spectroscopy

Continues on the next page.

Abbreviations used in this Review (Continuation)

Acronym	Meaning	Acronym	Meaning
GFAAS	graphite furnace atomic absorption spectrometry	TETRA	tetramethylarsine
GSH	reduced glutathione	TMA	trimethylarsine
HG-AAS	hydride generation atomic absorption spectrometry	TFA	trifluoroacetic acid
HPLC	high performance liquid chromatography	THGA	transversely heated graphite atomizer
HR-CS-GFAAS	high resolution continuum source graphite furnace atomic absorption spectrometry	TMAO	trimethylarsine oxide
iAs(III)	arsenite	TXRF	total-reflection X-ray fluorescence (spectrometry)
iAs(V)	(hydrogen) arsenate ions	TLC	thin layer chromatography
ICP-(SF)-MS	inductively coupled plasma (sector field) mass spectrometry	WHO	World Health Organization
IRMM	Institute for Reference Materials and Measurements	XANES	X-ray absorption near edge spectroscopy
JECFA	Joint FAO/WHO Expert Committee on Food Additives	μ-XRF	μ-X-ray fluorescence

RATIONALE OF THIS REVIEW

In the 21st century, not only did the pace of scientific development accelerate tremendously, but thanks to infocommunications, our way of life has been completely transformed. We spend nearly 90% of our time indoors, and global warming has led to the creation of energy-efficient air-conditioned buildings. There is a growing demand for healthy diet. At the same time, more and more people are turning to traditional medicine due to the lack of empathy often experienced in public health. These alternative pharmaceutical preparations are generally sold over-the-counter and considered dietary supplements.

Due to its controversial properties, arsenic (As) came early into the sight of humanity. It can occur in many chemical forms, from poisonous to harmless species. However, even small amounts of its toxic forms can have therapeutic and corroborative effects. In soil, the availability of As is mainly determined by the chemistry of iron (Fe). Arsenic phytotoxicity is closely related to the composition and pH of the soil and, last but not least, the tolerance of the plant to As. Biomethylation produces less toxic organic As compounds. The As contamination of many of the aquifers is of geological origin, adversely affecting the everyday life of some 200 million people. In Europe, the south-eastern part of Hungary is mainly affected by naturally occurring As contamination. Recently, significant efforts have been made in Hungary to comply with the 10 μg L^{-1} health limit for the concentration of As in drinking water set out in European Commission Directive 98/83/EC.[1] However, the choice of technological solution for As removal is primarily determined by the chemical form of As. The pace of development of analytical measurement techniques is extremely fast, and the development of elemental speciation analysis is also supported by the knowledge gained in many other fields of science. In addition to the coupling of separation techniques and atomic spectrometric measurement techniques, speciation procedures include a number of sample preparation procedures that can be a separate source of error. These may occur during sampling in the form of unwanted interconversion of the species, but the interference caused by sample matrix components may also be significant. In parallel with the need to improve analytical performance, it would be useful to develop cost-effective, speciation methods applicable to different matrices that could be routinely used in analytical laboratories.

The aim of the present review is to give a brief overview on main As speciation methods and to present how to address the challenges represented by As contamination at local and global scales in several environmental (e.g., soil, waters) and biological (e.g., crops, basic and processed food) samples, as well as complementary and alternative medicinal (CAM) products marketed as food supplements with special emphasis on As speciation.

ARSENIC SPECIES AND THEIR TOXICITY

Arsenic is a highly toxic metalloid that can occur in soil even at extreme concentration (e.g., 250 g kg^{-1}) due to both geogenic and anthropogenic activities.[2] Arsenic toxicity depends on its valence and chemical environment. Thus, As(III) species are more toxic than As(V) ones and inorganic As (iAs) compounds are more toxic than organic As ones. The median lethal dose (LD_{50}) values of the most common As compounds determined by oral exposition to rats have been summarized in Table I.[3]

Table I. Toxicity of arsenic species expressed as median lethal dose (LD_{50}) values*

As species	LD_{50} (mg kg^{-1})
AsH_3 (arsine)	3
As_2O_3 (arsenic trioxide)	20
Na_3AsO_3 (sodium arsenite)	60
Na_3AsO_4 (sodium arsenate)	120
$CH_3AsO(OH)_2$ [monomethylarsonic acid, MMA(V)]	700
elementary state As (As^0)	763
$(CH_3)_3As$ (trimethyl arsine, TMA)	787
$(CH_3)_4As^+$ (tetramethyl arsine, TETRA)	890
$(CH_3)_2AsO(OH)$ [dimethylarsinic acid (cacodylic acid), DMA(V)]	1600
$CH_3AsO(ONa)_2$ (disodium monomethyl arsonate)	1800
$(CH_3)_2AsO(ONa)$ (sodium cacodylate)	2600
$(CH_3)_3As^+(CH_2)_2OH$ (arsenocholine, AsC)	6500
$(CH_3)_3As^+CH_2COO^-$ (arsenobetaine, AsB)	10000
$(CH_3)_3AsO$ (trimethylarsine oxide, TMAO)	10600

*oral exposition to rats

The high toxicity of arsenite [iAs(III)] – also confirmed by the Pearson acid-base theory – can be explained by its significant affinity for the sulfur (S) atom. Thus, As can bind to the sulfhydryl group of enzymes and inhibits their function, causing cell death. The toxicity of (hydrogen)arsenate ion [iAs(V)] is due to the fact that its chemical structure is very similar to that of phosphate ion. This latter ion plays a role in the energy balance of living organisms through phosphorylation. Arsenate can be accumulated, mainly in the nails and hair. The iAs(V) taken up by plants, when replaced by phosphate ions, interfere the energy balance.

The iAs species are more toxic compared to organic derivatives such as e.g., arsenobetaine (AsB) and arsenocline (AsC). However, both dimethylarsinic acid [DMA(V)] and monomethylarsonic acid [MMA(V)] can decrease the levels of hepatic reduced glutathione (GSH) and serum alanine aminotransferase and increase those of cytochrome P450. These events were more intensive in rats at doses of 679 mg kg^{-1} MMA(V) and 387 mg kg^{-1} DMA(V) in the liver and lungs. At the same time, organ-specific DNA damage has been demonstrated.[4] Dimethylarsinous acid [DMA(III)] is a toxic intermediate of DMA(V) that can also be detected in urine in the case of As poisoning.[5] Monomethylarsonous acid [MMA(III)] is a carcinogenic and toxic human xenobiotic metabolite formed during the detoxification of GSH and MMA(V) in the liver during acute and minor As poisoning. It is then converted to DMA(V) by oxidative methylation upon reaction with *S*-adenosyl-L-methionine (SAM)[6] (Figure 1). The major metabolites of the typically non-toxic arsenic sugars, AsB and AsC, are also excreted in the urine (Figure 1). If As enters the human body in large quantities at the same time, it causes vomiting, diarrhea, increased sweating and cramps. The sodium salts of MMA(V) and DMA(V) have also been widely used as insecticides (e.g., currently still permitted in weed control for cotton cultivation).[7] Arsenic has also been used to pickle wood e.g., in the USA.[3] Among the organic As species, roxarsone should be mentioned, used mainly in poultry as a feed additive to increase weight gain and as a coccidiostat. Due to the increased concentration of iAs detected in meat from breeding animals and poultry after metabolization, the use of roxarsone was banned in the US since 2013[8] and also in the EU as early as 1999. However, it is still widely used in China and many developing countries.[9]

Among the human aspects of As toxicity, the harmful effects on health due to the chronic exposure to the consumption of As-containing drinking water in Hungary should also be mentioned. Thus, according to the results of the ASHRAM (Arsenic Health Risk Assessment and Molecular Epidemiology) research project funded by the European Commission and carried out in Hungary between 2002 and 2004, there was a positive correlation between As exposure and skin, lung, bladder and kidney cancer in several settlements located in the Hungarian Great Plain.[10] In the settlements of one the affected regions, for which the As concentration of drinking water exceeded 100 µg L^{-1}, an increase of 1.5 – 2 was observed in the number of spontaneous abortions and stillbirths compared to the control settlements.[11]

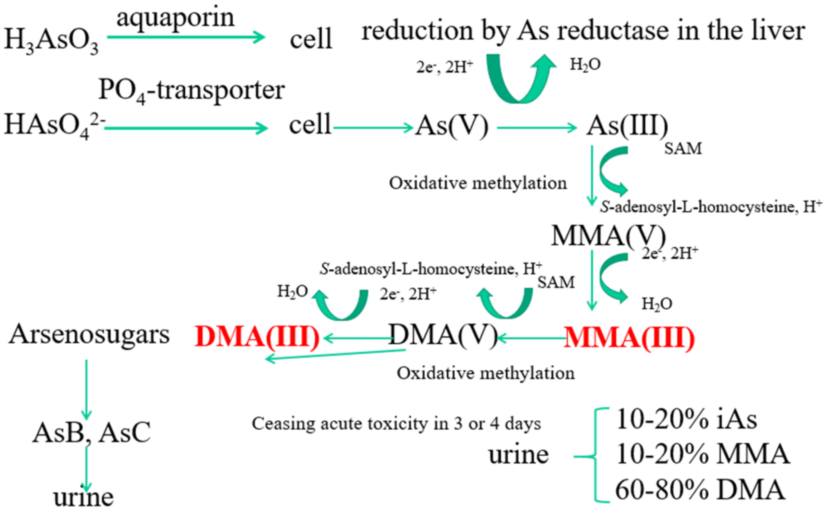


Figure 1. Mechanism of arsenic detoxification in liver for acute inorganic arsenic toxicity. Notations: iAs(III) = arsenite; iAs(V) = arsenate; AsB = arsenobetaine; AsC = arsenocholine; DMA(III) = dimethylarsinous acid acid; DMA(V) = dimethylarsinic acid; MMA(III) = monomethylarsonous acid; MMA(V) = monomethylarsonic acid; SAM = *S*-adenosyl-L-methionine.

ANALYTICAL TECHNIQUES FOR DETERMINATION OF As IN ENVIRONMENTAL AND FOOD SAMPLES AS WELL HYPHENATION POSSIBILITIES FOR As SPECIATION

Several analytical techniques have been proposed for As speciation. Analytical techniques suitable for qualitative and quantitative determination of As discussed in the present review have been compiled in Table II. It is worth mentioning that, in terms of As speciation, hydride generation atomic absorption spectrometry (HG-AAS) and synchrotron radiation X-ray absorption near edge spectroscopy (SR-XANES) allow discrimination between As(III) and As(V). Since the toxicity of As depends significantly on its chemical form, it is advisable to determine not only the total concentration of As in the samples but also that of each species. Thus, several chromatographic methods have been proposed for the separation of As species such anion/cation exchange, reversed-phase, ion pair, and size exclusion chromatography.[12] Separation of As species has traditionally been performed by high performance liquid chromatography (HPLC) using ion exchange columns. The most common species – iAs(III), iAs(V), MMA(V) and DMA(V) – are separated on an anion exchange column, while AsB and AsC are separated on a cation exchanger. In a recent review by Reid *et al.*,[12] suitability of hydrophilic interaction liquid chromatography, multiple separation mechanisms, and testing of fluorophenyl and graphene oxide stationary phases has also been discussed. Quantification of the separated species is performed using an appropriate atomic spectrometric technique connected to the chromatographic device on-line. Nowadays, inductively coupled plasma mass spectrometry (ICP-MS) is the most commonly used for this purpose.

Table II. Analytical techniques discussed in the present review suitable for As determination in environmental and food samples

Analytical technique	Sample preparation	Sample matrix	Remarks	Speciation	Ref.
Qualitative determination					
ATR-FTIR	–	food waste-derived charcoal added to As-contaminated soil	elucidation of As adsorption mechanism	n.a.	[13]
EDXRF	–	Ayurvedic ointment		– *	[14]
LA-DRC-ICP-MS	extraction	rice	monitoring of ^{13}C, ^{121}Sb and ^{75}As	off-line with PEI-TLC/OPLC	[15]
SR-XANES	–	xylem sap	LC of standard spectra	discrimination between As(III) and As(V)	[16]
Quantitative determination					
HR-CS-GFAAS	dilution with DW	thermal / well water		off-line with IE-SPE	[17,18]
HG-AAS	dry ashing	solid foods		use of pre-reducing agent (e.g., KI, L-cysteine)	[19-21]
Q-ICP-MS	dilution with DW	water	mathematical correction	[on-line with IE-HPLC & off-line with IE-SPE]	[19]
	MW-assisted acid digestion	aqueous food samples			[19]

Continues on the next page.

Table II. Analytical techniques discussed in the present review suitable for As determination in environmental and food samples (Continuation)

Analytical technique	Sample preparation	Sample matrix	Remarks	Speciation	Ref.
SF-ICP-MS	dilution with DW	water	HR when HCl is used	on-line with IE-HPLC & off-line with IE-SPE	[22]
	MW-assisted acid digestion (+H_2O_2 for organic matter)	aqueous food samples; rice; Ayurvedic formulations			[14]
	Enzymatic extraction with sonotrode	rice	for As speciation		[23]
	–	xylem sap	FIA for total As & R = 4000		[24]
TXRF	–	water		off-line with IE-SPE	[18]
	MW-assisted acid digestion	plant extracts (root, leaves)	for speciation, extraction with $NH_4H_2PO_4$ (pH=5.6)	off-line with PEI-TLC/OPLC	[25]
µ-XRF (confocal)	–	rice grains	individual item analysis	–	[26]

For acronyms see list of abbreviations; * = phase analysis.

Speciation of arsenic using solid phase extraction cartridges

One of the pivotal points of elemental speciation is to prevent interconversion of species during sampling and analysis; therefore, it is a great advantage to be able to use an analytical technique that requires minimal and cost-effective sample preparation. Significant efforts in this direction have been seen mainly in the As speciation of water. Cost-efficient separation of iAs species can be successfully performed at the sampling site using solid phase extraction (SPE) cartridges filled with strong anion exchange (SAX) resins.[27-33] When using SAX resins it is not possible to retain iAs(III) as it is a very weak acid (Figure 2). However, by coupling two miniature columns in series and oxidation by $KMnO_4$, iAs(III) can also be retained on the microcolumn.[34] Connection in series of SPE cartridges filled with strong anion and cation exchange resins have also been shown to be robust in field studies for speciation of As characterized by samples up to 30 mL of 10 mg L^{-1} As and complex matrix materials.[33] If necessary, MMA(V) can be determined after selective elution performed with acetic acid, while DMA(V) binds upon passing the fraction not retained on the SAX cartridge through the cation exchanger one (Figure 2).[28]

Alternatively, cartridges filled with Al-containing sorbent may be used.[35] In this case, a breakthrough of the otherwise retained iAs(V) was also observed when ions competing for Al silicate sorption sites were also present in the water.[36] Other SPE cartridges for *in situ* separation of the methylated forms of iAs have also been prepared. Such sorbents include nanometer-size titanium dioxide particles immobilized on silica gel,[37] octadecyl groups,[38] yeast immobilized on controlled pore size glass,[39] PTFE turnings,[40] cetyltrimethylammonium bromide-modified alkyl silica,[41] 3-(2-aminoethylamino) propyltrimethoxysilane-modified mesoporous silica,[42] macrocyclic materials,[43] hybride resins activated with hydrated Fe oxides and silver chloride,[44] carbon nanofibers,[45] as well as nanotubes[46] and eggshell membrane.[47]

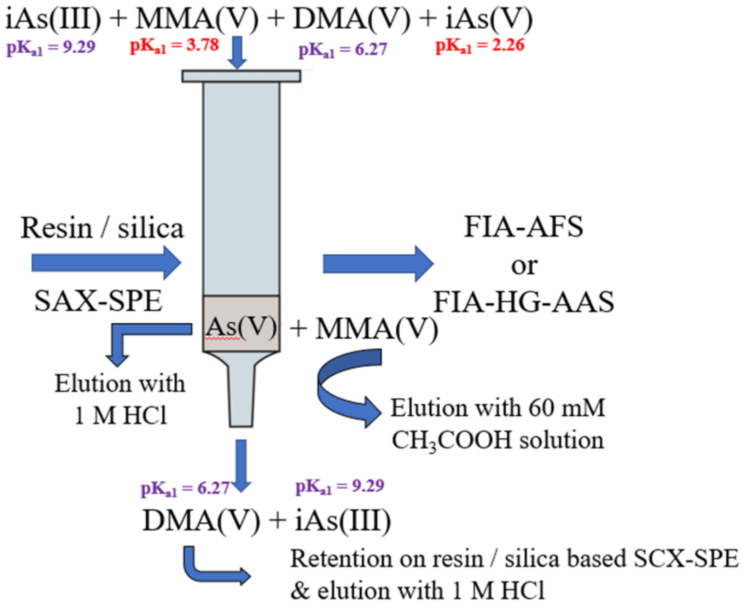


Figure 2. Diagram of a solid phase extraction process involving a cartridge filled with a strong anion exchange for *in situ* separation of arsenic species. (Figure adapted from Ref. 28.) Notations: iAs(III) = arsenite; iAs(V) = arsenate; DMA(V) = dimethylarsinic acid; MMA (V) = monomethylarsonic acid; pK_a = minus logarithm of base 10 of the corresponding acid dissociation constant.

ENVIRONMENTAL AND FOOD ANALYTICAL APPLICATIONS

Arsenic in soil

Occurrence of arsenic species in soil

Arsenic is present in many soil types at an average concentration between 0.1 and 40 mg kg^{-1}. As main constituent, it occurs in more than 200 minerals, mostly along with S. Its most important mineral is arsenopyrite (FeAsS), but realgar (α-As_4S_4) and auripigment (As_2S_3) are also noteworthy. In addition, it can occur in many sulfide minerals, mainly as a substitute for S. The concentration of As in igneous rocks is low, averaging 1.5 mg kg^{-1}. The concentration range for sedimentary rocks varies from 5 to 10 mg kg^{-1}. The concentration of As in the metamorphic rocks reflects the characteristics of the precursor igneous or sedimentary rock. Unconsolidated sediments typically contain 3-10 mg kg^{-1} of As, although this also depends on the texture and constituent minerals.[48]

In soil, the availability of As is mainly determined by the chemistry of Fe. In soils and groundwater, iAs(V) dominates. Inorganic As(V) are more likely to occur under aerobic, while iAs(III) under anaerobic conditions.[49] Inorganic As species are methylated in soils due to microbial activity (Figure 3).[48,50] Demethylation converts organic As compounds to iAs.[48] Biomethylated As(V) species account for up to 15.5–16.7% of the total As concentration of surface geothermal waters.[51] Oxyhydroxides of Fe, Al and Mn as well as clays, and mineral oxyanions (e.g., sulfates, phosphates and carbonates) in soil and sediments immobilize As.[52] Fine particles of clay minerals and (oxy)hydroxides adsorb As due to their high specific surface area.

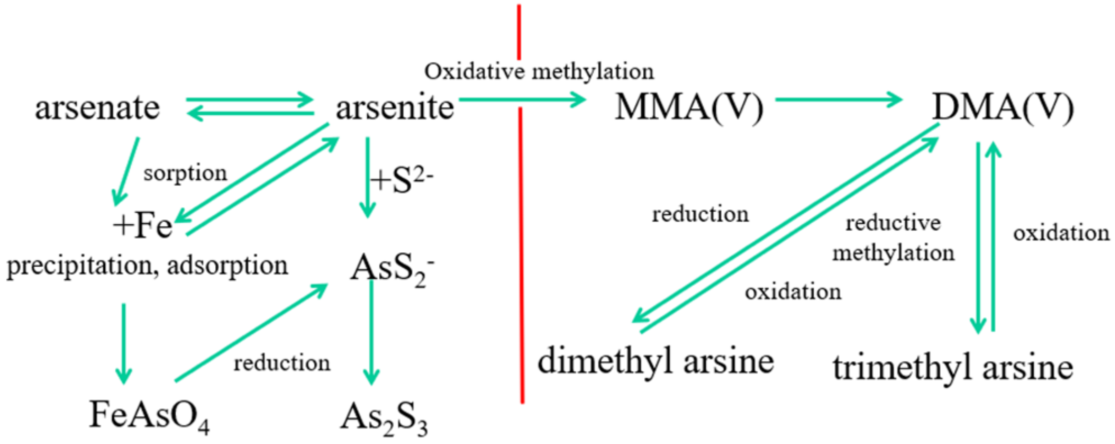


Figure 3. Simplified mechanism for the illustration of the connection of inorganic arsenic species to iron chemistry as well as bacterial methylation of in soil. Notations: $(CH_3)_2As$ = dimethylarsine; MMA(V) = monomethyl arsonic acid; DMA(V) = dimethylarsinic acid; TMA = trimethylarsine.

At regional scale, for example in Hungary, the concentration of As in soils varies between <2.5-230 mg kg^{-1}.[53] The limit value of As in Hungarian soils is 15 mg kg^{-1} dry weight (DM).[54] The European Economic Community has classified As a priority risk pollutant, i.e., As is a hazardous substance in all cases.[54] Concentrations of As in floodplain and riverbed sediments in the geochemical regions of Hungary range from 1 to 60 mg kg^{-1}.

The origin of As contamination of soils in Hungary has not been well elucidated yet (Figure 4). However, several theories have been suggested. Among the geochemical landscapes of Hungary (Figure 4), the second one is characterized by the accumulation of Ca, Mg and Sr, but As can also be found in the floodplain sediments of the Danube River. Moreover, As occurs in the southern part of the Danube-Tisza interfluve with exceptionally high values, probably due to the fact that As bound to Fe oxyhydroxides during the ice age is mobilized nowadays.[55,56] In the 3rd geochemical landscape, the dead ground originating from mining non-ferrous and precious metals (Ag, As, Au, Cd, Cu, Pb, Sb, Zn) in Transylvania (Romania) and the highlands of Slovakia, is carried by rivers to the shallow basin of the Great Plain. In this case, As is an accompanying element of non-ferrous metals.

Figure 4. Arsenic containing geochemical landscapes in Hungary. (Figure adapted from Ref. 55.)

Estimation of As concentration in agricultural soils through sequential extraction

First of all, it is important to estimate the total concentration of As in the contaminated soil. Then, it is advisable to perform sequential extraction of As on the soil. In the case of agricultural soils, As is most likely present in the form of iAs(V), so a two-step modified BCR (Bureau Communautaire de Référence) soil extraction procedure[13] is appropriate. Briefly, the water-soluble and carbonate fractions are extracted with 0.11 M acetic acid. This step is followed by extraction of the fraction bound to amorphous Fe(III)/Mn(IV) oxides with 0.5 M hydroxylamine hydrochloride solution at pH = 2. The initial samples as well as the extraction residues can be analyzed by ICP-SF-MS after MW-assisted acid digestion.[13] If appropriate soil CRMs are not available, another atomic spectrometric technique, e.g., total-reflection X-ray fluorescence (TXRF) spectrometric analysis of the same extracts can be used to check the accuracy of the results.[13] To calculate recovery, the concentration of As in the different extracts should be summed and compared to the pseudo-total concentration.[13]

Stabilization of arsenic in contaminated agricultural soils

Remediation of soils contaminated with toxic elements (e.g., As) is still a complex task of great importance nowadays, as toxic inorganic constituents cannot be easily removed from contaminated soil.[57,58] Usually *in situ* immobilization or stabilization by physical and chemical methods[59-61] have been proposed for toxic inorganic contaminants. The latter is an attractive alternative as it is a cost-effective and non-destructive process, yet suitable for remediating slightly or moderately contaminated soils. The purpose of immobilization is not only to minimize the mobility of toxic elements, but also to preserve soil structure and fertility. The most commonly used substances for stabilizing heavy metals are various minerals and organic substances [e.g., humic substances, ethylenediaminetetraacetate (EDTA), activated charcoal (AC)].[61-69]

Despite its many advantages, AC is hardly suitable for soil improvement due to its high production costs involving pyrolysis at 1000 °C for 24 h in an inert atmosphere for charring.[70] Charcoal has been used in agriculture for a long while.[71,72] However, due to its homonuclear structure, adsorption on charcoal is mainly characterized by van der Waals interaction.[73-75] The production costs of AC can be significantly reduced by choosing a large amount of available biomass waste as a feedstock. These raw materials do not always prove to be efficient enough, so activation and/or functionalization is required after charring involving mainly sulfuric or nitric acids.

However, As, the most common form of oxyanion, is not easy to stabilize in soil because plant adsorbents have a cation exchange capacity. Depending on pH of the media, iAs(III) and iAs(V) differ not only in valence but also in electrical charge significantly influencing their mobility.[76,77] Due to the carboxyl and hydroxyl functional groups contained in them, plant-derived ion exchangers are not suitable for binding negatively charged iAs(V) at soil pH. However, biomass wastes (e.g., peanut shells) are available in large quantities. The production of AC from peanut shell was carried out with concentrated sulfuric acid (H_2SO_4) for cost-effectiveness reasons. To increase the mechanical stability of the produced AC particles, composite formation with silicate-containing compounds (e.g., Florisil®) was applied.[13] Composite formation and the binding of As to AC were studied by Fourier transform infrared (FTIR) spectroscopy in attenuated total reflection (ATR) mode.[13] For example, acidic (pH = 5) sandy soil was incubated for four weeks with 15-30 mg kg^{-1} iAs(V). Freundlich isotherm proved to adequately model adsorption of As.[78] The highest adsorption rate was observed at pH=5 for Florisil®-modified AC. The slope of the line fitted for adsorption was about 30% higher at pH=5 than at pH=8 for Florisil®-AC. However, at pH 8, the rate of As adsorption decreased with Florisil®-modified AC. This suggested that the adsorption of iAs(V) is due to the formation of a covalent bond between the AC carboxyl group and one of the O atoms of iAs(V) while a water molecule is eliminated (Equation 1), as it was confirmed by the ATR-FTIR analysis:

$$\mathrm{Res-COOH+H_2AsO_4^- \rightarrow Res-COO-HAsO_3^- + H_2O} \qquad \text{Equation 1}$$

Formation of this mixed acid anhydride does not occur at alkaline pH, which was also proved by ATR-FTIR analyses.

Depending on the AC modification used, the proportion of immobilized As was approx. 30-40%, of which the soil alone immobilized about 15-20% As. The best results were obtained with Florisil®-modified AC. The amount of immobilizable As increased almost proportionally with increasing AC application dose and doubling the treatment concentration of iAs(V).[13]

Being electrically neutral over a wide pH range, iAs(III) can be more efficiently stabilized by ACs, e.g., formation of H-bonds. However, oxidation of iAs(III) → iAs(V) must also be taken into account. This cost-effective approach may be successful for acidic agricultural soils slightly contaminated with As.

Arsenic in water

Arsenic in groundwater in aquifers

The concentration of As in groundwater varies very widely (0.1-5000 μg L^{-1}).[48] This is especially problematic if the water body serves as a drinking water base. Several theories have been formulated for the large As contamination in natural water.

1) During the extraction of sulfide ores, As originating from their weathering first binds to Fe(III) (oxy) hydroxides; in parallel with the reduction of Fe(III) to Fe(II), As is released and dissolved in the groundwater in the mining areas.
2) Oxidation of As-rich pyrite or arsenopyrite results in the dissolution of As in groundwater.[79] The reductive dissolution of Fe(III) (oxy)hydroxides in groundwater under anaerobic conditions by microorganisms, e.g., may be catalyzed by *Acidithiobacillus ferrooxidans* and *Leptospirillum ferrooxidans*.[52,80]
3) A third theory that is characteristic of the Pannonian Basin in Hungary is that As is brought to the surface by thermal water that erupts from the depths.[81]

Arsenic can be accumulated in aquifers or highly reducing layers of inland/closed basins (semi-)desert areas.[48] Both of these different geological environments contain young sediments in flat, low-lying, and slow groundwater flow areas. Waters with high As concentrations (up to 5000 μg L^{-1}) adversely affect the drinking water supply of some 200 million people worldwide.[48] The As contamination of many aquifers is of geological origin. This affects the western part of the North American continent, the vast areas of Chile and Argentina in the southern hemisphere, parts of China, and the Gulf of Bengal and its associated delta region in Asia.[48] Studies in the US suggest that changes in As concentration over time cannot be clearly correlated with wet/dry seasons[82] or pre- or post-monsoon periods in the Indian subcontinent.[83] Arsenic contamination can also be attributed to a lesser extent to anthropogenic activity.[84]

Arsenic in surface and drinking water

In rivers, the As concentration is around 0.1-0.8 μg L^{-1}, which is also influenced by the flow of the rivers, lithology of the river bed and water replenishment of the area. The As concentration in lakes is close to or less than that of rivers.

For the *in situ* separation of As species from water samples taken from public wells in Hungary, the method described by Le *et al.*[28] was also suitable. For As determination in the samples, ICP-SF-MS was used. High-resolution mode (R = 10000) had to be applied only for As speciation. The sum of the As concentration in the SPE fractions was compared to the values of the total As one obtained for the corresponding original samples. Organic As species [e.g., MMA(V) or DMA(V)] could not be detected in the samples. Even a four-fold enrichment could be applied to the iAs(V)-containing fractions. After regeneration, the SPE microcolumns could be reused at least 15 more times by analyzing repeatedly carbonated mineral water samples with a salt content of 450 mg L^{-1} between pH 6 and 8 spiked to 150 μg L^{-1} iAs(V). This approach was applied to determine the ratio of iAs(III) and iAs(V) in water taken from public wells of settlements belonging to different geochemical landscapes of Hungary affected by natural As contamination (Figure 5). The total concentration of As in the samples taken from the public wells of settlements of the Csongrád plain, located east of the Tisza River and belonging to the 2nd geochemical landscape (Figure 4), reached even 210 µg L^{-1}.

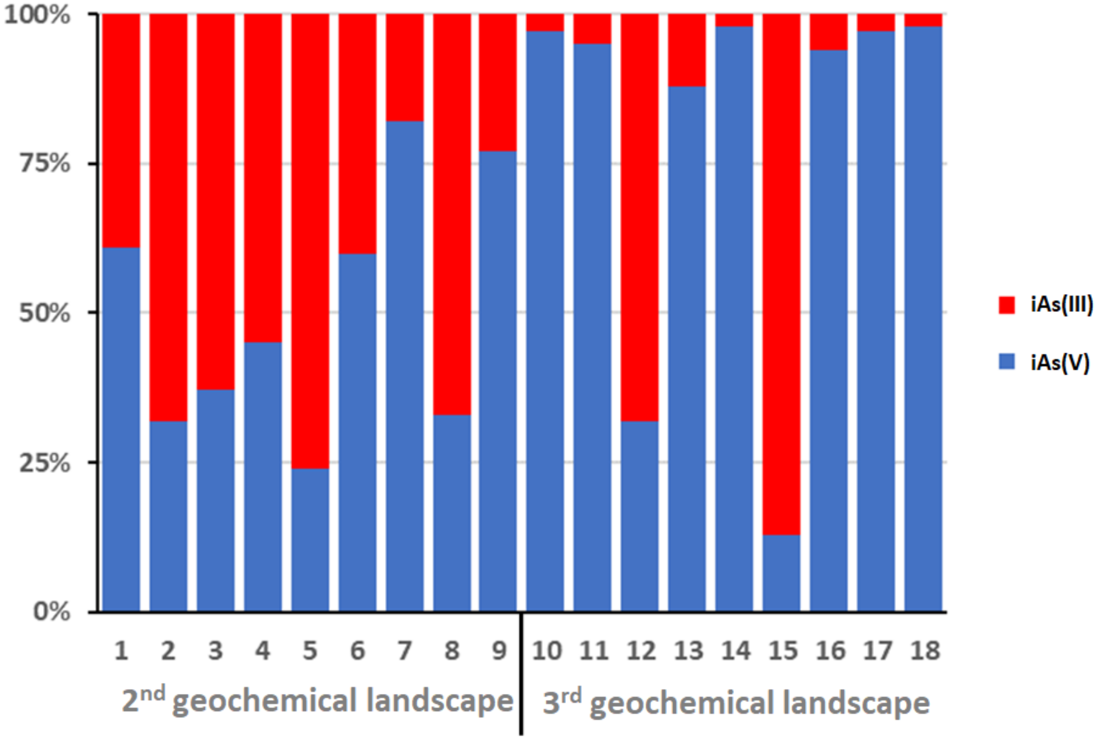


Figure 5. Arsenic distribution between arsenate [iAs(V)] (blue) and arsenite iAs(III) (red) in water samples taken from public wells in settlements located in the 2nd and 3rd geochemical landscapes affected by natural As contamination in Hungary. Grouping of data according to the relative distance of the settlements from each other. (Figure adapted from Ref. 22.)

The results obtained were in agreement with the geochemical theories on the mobility of As in soil. Thus, mobility of As deposited in flat areas from river sediments situated in the 3rd geochemical landscape, can be attributed to the iAs(V) species. The anaerobic conditions favor the formation of iAs(III) and Fe(II) in the deeper layers. In the southern part of the Danube-Tisza interfluve, located in the 2nd geochemical landscape (Figure 4), mobilization of iAs(V) adsorbed onto Fe(III) (oxo)hydroxides during the ice age could only be achieved if Fe(III) → Fe(II) reduction took place due to anaerobic conditions. The mobility of iAs(III) is due to the increased water solubility of the resulting Fe(II) compounds, and the fact that this species is electrically neutral over a wide pH range. Thus, iAs(III) is believed to be mobilized nowadays.[55,56] However, iAs(III) is again converted to iAs(V) species under aerobic conditions.

For *in situ* separation of As species, the SPE method described by Le *et al.*[28] was also applicable to samples taken from a geothermal well.[17] The As content of the samples was determined by high resolution continuum source graphite furnace atomic absorption spectrometry (HR-CS-GFAAS) using an Xe arc lamp as a continuous-spectrum primary radiation source equipped with a transversely heated graphite tube atomizer (THGA).[17] A linear response to As in the concentration range of 5 – 200 µg L^{-1} was obtained by external calibration. In addition, the absorbance values up to an As concentration of 500 µg L^{-1} could be properly fitted with a quadratic polynomial equation. The LOD value of the method was 0.6 µg L^{-1}. This method was characterized by adequate RSD and recovery. When the 150 µg L^{-1} of an iAs(III) fraction acidified with HNO_3 and stored for up to 30 days was analyzed by HR-CS-GFAAS, the oxidation rate of As(III) → As(V) was less than 10%. The iAs speciation could be performed with sufficient accuracy (± 5%)

up to pH 8.[17] A mass balance could be set up for the As species determined by SPE-HR-CS-GFAAS. The total As concentration was about 400 μg L^{-1}. Chloride and sulphate ions in natural waters above 100 and 50 mg L^{-1}, respectively, caused interference in the determination of As by the continuum source GFAAS method.[85] The effect of high concentration of sulphate and chloride ions generated by magmatic gases in volcanic water on the As measurements was minimized by two-fold dilution of the samples before HR-CS-GFAAS analysis. The proportion of iAs(III) in the geothermal water was about 55%. However, it was reported that the proportion of iAs(III) can be more than 70% in geothermal waters.[86] Slow cooling of the water below 66 °C did not change the proportion of As species in the samples. The SAX-SPE has been shown to be effective for As speciation in water with high salinity at about 85 °C. Due to the total concentration of As (i.e., 400 μg L^{-1}) in the water sample, almost half of which was iAs(III), the analyzed water cannot be used for recreational and/or aquaculture purposes.

For the separation and enrichment of iAs species in tap, lake and well waters, the same Dowex 1-X8 SAX-type SPE charge using electrothermal AAS was suitable for the determination of As in the fractions eluted from the cartridges.[87] Arsenic speciation of surface[88] and groundwater[89] was performed using silica gel-based SAX-SPE- HG-AAS hyphenated technique.

Arsenic removal technologies from drinking water

Arsenic has a complex water chemistry and exists in a variety of inorganic and organic forms in waters, depending on pH, salinity, acid dissociation constants (K_a) of As oxyacids, as well as redox potential of the iAs(V)/iAs(III). Chemical oxidation, co-precipitation, adsorption, ion exchange, reverse osmosis, and membrane filtration are used to remove As from (waste)water.[90-93]

From a technological point of view, As removal processes can be divided into three major groups[94]: 1) conventional technologies (coagulation, Fe-Mn removal, lime softening); 2) sorption processes (ion exchange, activated Al); 3) membrane technologies (reverse osmosis, nanofiltration, micro- or ultrafiltration). The removal efficiencies and main characteristics of the six most commonly used As removal technologies for the two iAs species present in water are summarized in Table III.[94,95]

Table III. Main features of some arsenic removal technologies

Technological solution	**Removal efficiency (%)**		**Notes**
	iAs(III)	**iAs(V)**	
Coagulation with Fe, sedimentation/filtration	60	95	1. filtration through 0.1 or 1.0 μm pore size 2. pH dependent
Coagulation with Al, sedimentation/filtration	15	80	also suitable for removal of fluoride
Lime softening	70	99	1. for hard water 2. soft water must be acidified
Ion exchange	-	>95	1. independent of pH; 2. SO_4^{2-}interference
Activated Al oxide	-	98	1. iAs(V) exchanges OH^- on the surface of Al_2O_3 2. pH dependent
Reverse osmosis	60	>95	treated water not suitable for human consumption

iAs(III) = arsenite; iAs(V) = arsenate.

As shown in Table III, each of the technologies is more efficient for the iAs(V) species, so oxidation of iAs(III) is often applied first. Oxidation by simple direct aeration is slow, but there are several chemical agents that can speed up the process such as chlorine gas, hypochlorite, ozone, $KMnO_4$, hydrogen peroxide, Mn oxides, and UV radiation.[95]

The main water treatment steps applied in the waterworks operating settlements located in the southern regions affected by natural As contamination in Hungary are summarized in Table IV.

Table IV. Steps of drinking water quality improvement technologies applied in the waterworks of the monitored settlements

Step	Water treatment step	Target compound	Notes
1	Gas removal by purging with air	CH_4	
2	Flocculation and oxidation	iAs(III), Fe(II), Mn(II)	2-4 w/w% $KMnO_4$ + 40 w/w% $Fe_2(SO_4)_3$
3	Quartz sand filtration	products of oxidation *cf.* step 2	
4	Pre-chlorination	NH_4^+	5 mg L^{-1} Cl_2
5	Granular activated carbon filtration	by-products of step 4	
6	Post-chlorination (Cl_2, ClO_2, NaOCl)	—	0.3-0.5 mg L^{-1} free Cl_2 concentration after chlorination

Water samples were also analyzed by HR-CS-GFAAS and TXRF after *in situ* separation of the As species by SPE. The total concentration of As in the raw water samples taken from the wells of the waterworks varied between 42.9 and 118 µg L^{-1}. In well waters, iAs(III) (> 80% of total As) was the predominant species in each settlement. After the addition of oxidizing and flocculant and filtration on quartz sand, the total concentration of As decreased below 10 µg L^{-1} during the applied water treatment technology. This value was typically about 5 µg L^{-1}. Regarding the proportion of species, iAs(V) was the predominant species. Arsenite was not detected in the samples taken after water treatment. Application of As speciation allowed fine tuning of the reagent use (e.g., $KMnO_4$) for the oxidation step.

In addition, application of TXRF has the advantage to provide multi-element information. Thus, changes in the concentration of other elements of interest in Directive 98/83/EC in water samples could be monitored after the addition of the oxidizing ($KMnO_4$) and flocculant [Fe(III) sulfate]. Thus, it was demonstrated that the concentrations of Mn and Fe in drinking water fell far short of the values laid down in Directive 98/83/EC. After the addition of the oxidizing agent and the flocculant, the Fe, Mn and S constituents were retained by the SPE microcolumn, but the cartridges could be regenerated with HNO_3 solution.

Moreover, dilution of contaminated water with that of adequate quality to reduce As contamination of drinking water is an accepted practice, as well. For example, the proportion of iAs(V) in the drinking water of some settlements established on the floodplain soil of the Ráckeve branch of the Danube belonging to the 2nd geochemical landscape with a total As concentration larger than 10 mg L^{-1} was found to be decreased after repeating sampling and analysis after 5 years. However, the proportion of As species was almost the same as in the samples originally taken. This result means that the reduction of the As concentration was achieved by dilution with water of adequate water quality.

Arsenic in plants and the importance of xylem sap analysis

The first studies on the accumulation of As in plants were published first in the 1970s. It was found that, the As content in plants grown on As-contaminated soils was slightly higher than in lightly contaminated soils (0.01-5 mg kg^{-1} As), but As accumulation was not observed.[13] The fern native to China (*Pteris Vittata* L.) was the first plant to be found to be capable of hyperaccumulation of As (> 20 g kg^{-1} DM). Arsenic can also be taken up in larger amounts by rice (*Oryza sativa* L.).[96] However, the As content of carrots (*Daucus carota* L.) can also reach up to 1 mg kg^{-1}.[97] Arsenic is found primarily in the roots of plants, and its concentration can be up to 75-fold higher than that determined in the shoot.[98,99] The vast majority of As is localized in the apoplastic space of the root.[100] In the rice plant grown under reducing conditions, 1% of the total As concentration (2.3 mg kg^{-1}) was transported to the shoot. In the root, 60% and 39% of the apoplastic and non-apoplastic fractions contained As, respectively.[100] In addition, the hyperaccumulative fern contained about 1/6 of the total As in the apoplast.[101]

Arsenic species found in terrestrial and aquatic plants, lichens, and fungi are divided primarily into hydrophilic and lipophilic compounds.[102] In terrestrial plants, mainly inorganic species of As, namely, iAs(V) and iAs(III), are detected, although methylated derivatives of iAs(V), DMA(V) and MMA(V), may be present in small amounts.[103]

Since iAs(V) is the most stable species in the soil under aerobic conditions, it is expected that plants will absorb it mainly in this form. Iron deposition on the root surface can significantly bind iAs(V) species.[100] In the reducing environment of root cells, iAs(V) is readily reduced to iAs(III). This reduction is thought to be an essential process in the detoxification of As, as the resulting more toxic iAs(III), being electrically neutral over a wide pH range, is transported to senescent leaves.[104] It has not been clearly demonstrated whether organic As compounds detected in plant tissues can be taken up directly from the soil. There is also no clear evidence that iAs species are methylated by microorganisms living in root symbiosis or by the plants themselves.[103]

The MMA(V) species is converted to DMA(V) in the root or xylem sap of the As hyperaccumulative fern (*Pteris Vittata* L.).[105,106] However, microorganisms may also be involved in methylation. Study of As in terrestrial plants provides a two-fold information since: (i) it may indicate the bioaccessibility of As, and (ii) As taken up by plants may enter the food chain. The phytotoxicity of As also depends on the pH, phosphate, composition (e.g., Fe, Al and organic matter content) of the soil and the tolerance of plants for As.[107] Since various As species have different toxicities, the information obtained from their identification and distribution within the plant may provide an answer to the plant metabolism of As and an assessment of the health risks posed by the introduction of As into the food chain.

The study of xylem sap playing a central role in the transport of toxic and essential elements and nutrients to plant shoots, is particularly advantageous for elemental speciation, as it requires minimal sample preparation. Xylem sap from cucumber plants (*Cucumis sativus* L.) is easily collected and a single sample preparation step consisting of filtration through 0.22 μm membrane filters should be used for chromatographic studies.[108-118] The roots of most plants should be placed in a pressurized chamber filled with an inert gas (e.g., nitrogen gas) to enhance exudation. This, together with the fact that interconversion of As species can occur relatively easily, makes it difficult to perform As speciation in xylem sap. Pickering *et al.*[114] confirmed that only a small amount of As was transported to the shoot of Indian mustard (*Brassica juncea* L. var. Czern) applying a treatment of 250 μmol L^{-1} iAs(III) or iAs(V) by XANES measurements. The iAs(V) taken up was stored by the plant both in the root and shoot mainly in the form of As(III) tris(thiolate) complexes. Kertulis *et al.*[106] have been detected mainly iAs(V) in the xylem sap of *P. vittata* L. treated with either iAs(III) or iAs(V) at concentrations up to 50 mg L^{-1}, regardless of the type of treatments used. In the case of treatments with MMA(V) or DMA(V), As was transported to the shoot in these methylated forms. However, iAs(III) has always been detected at the apex of the fern leaf, suggesting that reduction of iAs(V) occurs in the plant shoot.

Plant growth experiments conducted in nutrient solution have shown that the concentration of As(V) (2–10 μmol L^{-1}) and the iAs(V):phosphate ion concentration ratio of the hydroponics (1:100, 1:1, and 100:1) play an important role in the development cucumber plants. This phenomenon is due to the similar structure of hydrogen arsenate and hydrogen phosphate ion, taken up and transported within the plant by the same mechanism. To eliminate competition between iAs(V) and phosphate, it is advisable to set their concentration ratio in the medium to 1:1. However, the seedlings showed a severe phosphate deficiency at a phosphate ion concentration of 2 μmol L^{-1}, which could only be eliminated at a concentration of 250 μmol L^{-1}. Therefore, in plant growth experiments, it is advisable to keep the phosphate concentration at this value and to reduce it to the As concentration only at the start of As treatment.[24]

Speciation analysis of the nutrient solutions also confirms the different uptake mechanisms of the two species – electrically neutral iAs(III) and ionic iAs(V) at physiological pH.[24] As the pK_{a1} values of H_3AsO_3 and H_3AsO_4 differ significantly, i.e., 9.3 and 2.3, respectively,[105] and taking into account the pH of the hydroponics (pH 4.7–5.9), the electrical charge of these two species is significantly different. The iAs(V) uptake is similar to that of phosphate ions, with electrically neutral iAs(III) being taken up through aquaporin.

When sampling xylem sap from cucumber plants treated with As, the rate of exudation with respect to sampling time and fresh root weight (FRW) was very similar for both As treatments: 12.4 and 11.2 µL h^{-1} FRW^{-1} for iAs(V) and iAs(III) treatments, respectively.[24] Three As species – iAs(III), DMA(V) and iAs(V) – could be identified in the xylem sap. Regardless of the As treatment, the following concentration order was determined: iAs(III) > iAs(V) > DMA(V). The proportion of DMA(V) in the xylem sap did not reach 5%. The mass of the As species in the xylem sap collected for 1 h taking into account the exudation rate, was very similar: 17.65 and 16.55 ng h^{-1} for the iAs(V) and iAs(III) treatments, respectively. Arsenobetaine and AsC were not detectable in the samples by cation exchange HPLC-ICPSF-MS. The predominance of iAs(III) in xylem sap of cucumber plants grown in As containing hydroponics was consistent with literature data.[104] Zabłudowska *et al.*[119] found that As present as iAs(V) in aerobic soils was taken up by the phosphate transport system of ferns. Subsequently, a portion of the iAs(V) is reduced to the iAs(III) specific in the root by the arsenate reductase[120,121] and further translocated to the shoot *via* the xylem.[122,123] Reduction is an important process in As tolerance, as iAs(III) can react with highly reactive compounds containing SH groups,[124] such as thiol compounds, e.g. phytochelatins or GSH – and is thus immobilized by complex formation. The iAs(III) ratio was also greater than 60% in xylem sap of rice, tomatoes, wheat, cabbage, and barley. However, methylated As species are rarely detected.[114,123-126] The presence of iAs(V) in the xylem sap of plants treated with the iAs(III) species can be attributed to the partial oxidation of iAs (III) to iAs (V) in the hydroponics.

Cucumber plants also take up DMA(V) species in an unaltered way. Zhao *et al.*[127] found that not only in cucumber plants, but also in xylem sap or different tissues of common velvet grass (*Holcus lanatus*)[128] and sunflower (*Helianthus annuus*)[129] grown in iAs-containing hydroponics, methylated As species could be determined only in traces. This provided indirect evidence that the biomethylation capacity of the plants was low.

Arsenic containing xylem saps can also be studied by XANES for As speciation purposes. The normalized profile of the absorption edge of the As K shell for a xylem sap sample taken from cucumber plants (*Cucumis sativus* L.) treated with iAs(V) is shown in Figure 6. These studies also confirmed that iAs(III) is the predominant species in xylem sap regardless of the chemical form of As used for plant growth.

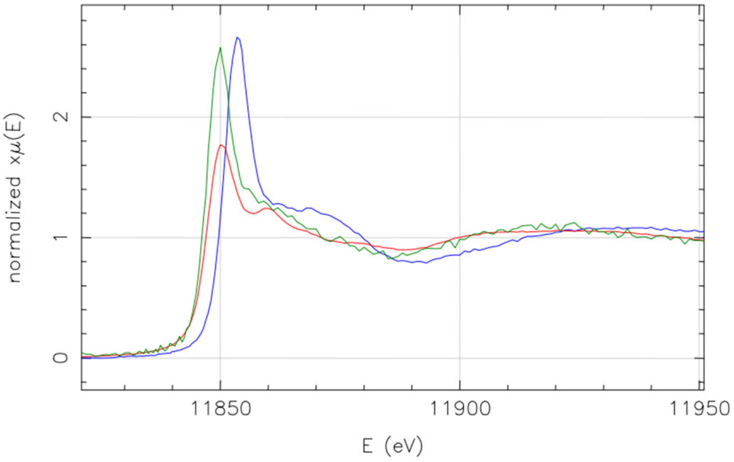


Figure 6. Spectra of a xylem sap taken from cucumber plants (*Cucumis sativus* L.) treated with iAs(V) as well as those of arsenite (red line) and arsenate (blue line) standards by near near-edge X-ray absorption spectroscopy. (unpublished figure)

The XANES spectra of nutrient solutions and xylem saps can be simulated by linear combinations (LC) of XANES spectra of iAs(III) and iAs(V) standards. Thus, LC calculations confirmed that >80% of As detected in xylem sap corresponds to the iAs(III) species regardless of the type of As treatment.[16] This is consistent with the results obtained by HPLC-ICP-MS, as the proportion of iAs(III) in those similar samples was 86%.[24]

As speciation studies on cucumber plants can be performed by applying off-line polyethylene imide thin layer chromatography – total-reflection X-ray fluorescence (PEI-TLC-TXRF) spectrometric method using acetone: acetic acid:water. The elution of the different As species was as follows: iAs(V) (R_f = 0.06) <iAs(III) (R_f = 0.38) < MMA(V) (R_f = 0.59) < DMA(V) (R_f = 0.91).[25] Due to the reproducible operating conditions ensured by overpressured layer chromatography (OPLC) such as controlled flow rate, external application of pressure and accurate flushing volume, as well as doubling of the running distance, the separation was faster and resolution was better for each 25 ng of iAs(III), iAs(V), MMA(V), and DMA(V) expressed in elemental As in a multicomponent standard solution (Figure 7).

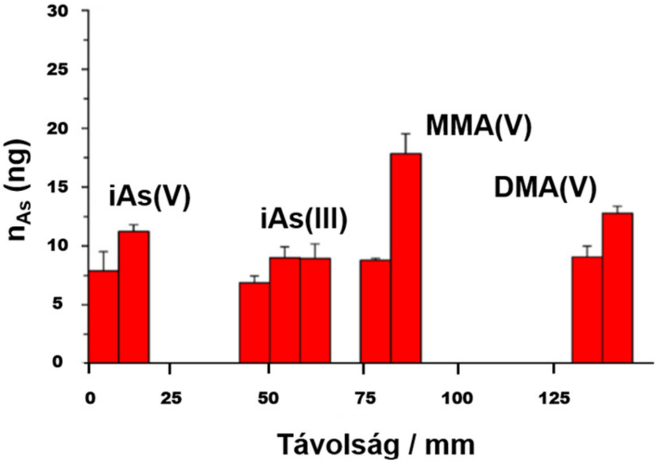


Figure 7. Off-line PEI- OPLC-TXRF chromatogram of multicomponent standard solution of arsenite [iAs(III)], arsenate [iAs(V)], monomethylarsonic acid [MMA(V)] and dimethylarsinic acid [DMA(V)] containing 25 ng of As each. [Reprinted (adapted) with permission from Ref. 25: Mihucz, V. G.; Móricz, Á. M.; Kröpfl, K.; Szikora, S.; Tatár, E.; Parra, L. M. M.; Záray, G. Development of off-line layer chromatographic and total reflection X-ray fluorescence spectrometric methods for arsenic speciation. *Spectrochim. Acta, Part B* **2006**, 61, 1124–1128. https://doi.org/10.1016/j.sab.2006.08.011 Copyright© (2022), Elsevier.]

The efficiency of As extraction from the roots of cucumber plants treated with iAs(III) species was 92%.[25] To quantify As by the developed off-line PEI-OPLC-TXRF method, the extract should have been applied in triplicate onto the plate. However, due to the high concentration of nutrient cations in the root samples and the electrostatic repulsion between positively charged PEI cellulose, this was not feasible. Only iAs(V) and iAs(III) species could be detected by applying 4 µL of the root extract. The presence of iAs(V) in the samples is due to the oxidation of iAs(III) to iAs(V) in the nutrient solution during treatment. In addition, the time-consuming extraction process could not be performed in an inert gas atmosphere.

A faster As speciation method can be achieved by introducing the different As species separated on the PEI-TLC layer into the ICP-MS by laser ablation (LA). Even much smaller amounts of As (different As species containing 3 ng As each) can be detected with this hyphenated technique than with the PEI-TLC/OPLC-TXRF method. The R_f values calculated for the separated species also showed a good agreement with the data obtained by the PEI-TLC-TXRF method[15]. Arsenobetaine and AsC were detected together at the sampling point. The resolution calculated for each adjacent peak ranged from 1.5 to 2.5, and no OPLC separation was required. The method has significant potential for rapid identification of As-contaminated samples, which was verified on enzymatically extracted NIST SRM 1568a rice flour. Thus, up to 10 pg

MMA(V) could be distinguished from the background value. This proposed TLC-LA-ICP-MS method was later modified for silica gel-based high-efficiency TLC plates[130] for Cr speciation. It has recently been further optimized using an iAs(V)-imprinted polymer to separate iAs(III) species. The developed µ-TLC-LA-ICP-MS method has been successfully applied to mine water spiked with iAs(III) and iAs(V) species. Using the optimal conditions, the detection limit of As was 0.3 µg L^{-1}.[131]

Dietary As intake and As speciation in rice grains using simple kitchen technology operations

For the population not exposed to As-contaminated drinking water, nutrition is the most significant source of iAs intake.[132] A close to linear relationship was observed between the As concentration of foods containing more than 40% water (e.g., soups, soft drinks, soda water, beer) and that of the water used for their production.[19] Moreover, the As concentration of the samples with high water content was positively correlated with that of the water used (i.e., Pearson's correlation coefficient was 0.830-0.923).[19] However, in Bangladesh, it has been shown that a partial reduction of As contamination in drinking water was not sufficient to significantly reduce the amount of As ingested.[133] In Europe, the population consumes an average of 153 g of vegetables per day, compared to around 250 g in Asia.[134] In Chile, the total As and iAs content of cooked vegetables from an As-contaminated area was studied. During the study it was found that the concentration of As in the drinking water used for cooking varied between 41 and 572 µg L^{-1}. The results also showed that foods made a significant contribution to iAs intake, which also posed a higher health risk for those 15 years of age and younger.[135] Exposure of school children (n=55) to As-contaminated well water was assessed in several areas of Mexico. The considerable As concentration in the water showed a clear correlation with that of determined in the hair samples of children. According to the authors, As-contaminated water used for cooking may have been the main source of As exposure.[136] The use of poor-quality glass and unglazed earthenware can also cause As poisoning.[137] The concentration of As dissolved by soaking in unglazed earthenware varied from 30.9 to 800 µg L^{-1}, while the As concentration obtained in the same experiments with glazed earthenware often fell below the detection limit (<0.5 µg L^{-1}), but maximally reached only 30.6 µg L^{-1}. Valentine *et al.*[138] compared the dietary habits of the population in two As-contaminated areas (Mexico and California) where there are geographical differences in the prevalence of skin diseases. Although the population exposure to As was similar to that of drinking water, there was a difference in the diet (vitamin A consumption) for the groups studied. The issue of dietary intake has been studied in detail in Latin America.[13] The dietary exposure of the population to As has been extensively studied in different regions of France and Catalonia[140,141] as well as in Hong Kong.[142] The main route of bioavailability of As is the food chain (diet and drinking water consumption).[143,144] According to Yost *et al.,*[145] 21-40% of As ingested by terrestrial foods in North America is of inorganic origin. The average concentration of As in vegetables is around 20 µg kg^{-1}.[146]

One of the staple foods for human consumption, rice (*Oryza sativa* L.) is also often contaminated with As, at concentrations of up to 280 µg kg^{-1}.[96] Also in the United States, rice consumption is a significant source of children exposure to As.[147-149] According to the Food and Agriculture Organization (FAO), the People's Republic of China is the largest rice producer in the world. China is followed by India, Indonesia, Bangladesh and Vietnam. In many areas of these countries, groundwater is highly contaminated with As.[80] The relatively high As content of rice suggests considerable uptake and translocation of this element. According to Liang *et al.,*[150] the rice grains are first husked by grinding (and friction), i.e. the husks (bran layers) covering the grains are removed. The rice grains, stripped of their husks, are polished with a (conical) grinder and bleached with glucose or talcum. The 8-10% of the initial weight of brown rice is removed, mainly in the form of bran. As with most cereals, the distribution of the amounts of elements within the rice grain is heterogeneous. Consequently, it is worth examining the effect of milling rice grains on element content. Of the trace elements accumulating in rice, special attention is paid to the highly toxic Cd.[151-154] However, several other toxic elements have already been determined in various milled rice cultivars.[155,156] Rice plants are basically classified into two classes. We distinguish between short and long grain rice. The two most common subspecies are *indica* and *japonica*. Short-grained and cooked rice is a subspecies of *japonica*, while long-grained rice is of Indian origin. *Indica* subspecies are usually grown on water-flooded arable land, while *japonica* species can be grown on dry soil. In general, rice crops and grains grown by the flood method are studied.[152,153] However, Rascio *et al.*[154] studied Vialone Nano rice grown in the province of Verona (Italy) used to make risottos.

The rice grains must be subjected to solvent or enzymatic extraction before chromatographic separation. Methanol is often used for solvent extraction. Thus, e.g., 76% of the concentration of As in the NIST 1568a in the rice sample could be recovered by extraction with methanol-water (1:1 v/v) using ultrasonication. In the case of extraction with trifluoroacetic acid (TFA) solution at 100 °C for 6 h, the recovery in As-spiked rice samples was quantitative as opposed to methanol extraction.[155] In the latter case, the recovery varied between 31 and 72%.[155] However, a reduction in iAs (V) was observed using TFA. According to Huang *et al.,*[157] quantitative extraction could be achieved by using dilute HNO_3 solution for As speciation in rice grains.[157] By extracting As species from freeze-dried rice produced at the IRMM with water or methanol:water mixtures of different compositions (1:1, 9:1 and 1:1 then 9:1 v/v), Pizarro *et al.*[156] found that about 80% of the total concentration of As can be extracted with a 1:1 v/v mixture of methanol:water using a single extraction step. Accelerated solvent extraction (ASE) is similar to conventional solid-liquid extraction, but it is performed at higher temperatures and pressures, reducing the time required for extraction. However, depending on the type of solvent, the temperature and the pressure used, ASE causes the rice grains to swell.[155] Nevertheless, ASE has been successfully used to extract As species of marine fish[158] and seaweed.[143]

The biggest disadvantage of enzymatic extraction methods is their high time demand, which can result in the interconversion of As species.[159] To break down starch, Heitkemper *et al.* used α-amylase[155] for As determination in rice prior to extraction with methanol: water (1:1 v/v). Under these conditions, an extraction efficiency of 59% was achieved for white rice grains. However, for the NIST 1568a rice sample, this value was found to be 97%. Although the best results were obtained using TFA, it was found that iAs(V) was partially reduced to iAs(III) during extraction. In addition, Pizarro *et al.*[156,160] rejected the use of TFA for As-speciation from rice by HPLC-ICP-MS because in this case poor chromatographic resolution of the extracted As species was observed. Focused ultrasonic treatment coupled with enzymatic hydrolysis has been successfully applied to the extraction of As from rice grains for speciation purposes[23,161] as well as to other samples of biological origin.[162]

Rice is dominated by iAs(III), iAs(V), DMA(V), and MMA(V).[96,156,158-162] Arsenic speciation can be performed in rice seedlings[163] as well as in rice grains.[96,156,158-162] In raw rice grains, iAs(III) is the predominant species, accounting for about 50% of the samples.[23] The other major As species present in the samples is iAs(V), the percent of which varied between 25 and 33%.[23] At the same time, the percent of DMA(V) ranged from 8 to 10%, while that of AsC from 6 to 10%.[23] In 2015, the European Commission revised Directive 2006/1881/EC on maximum levels for iAs in food. Thus, the sum of the concentrations of iAs(III) and iAs(V) in rice used for food processing in the case of infants and young children under Regulation (EC) No 2015/1006 may not exceed 0.1 mg kg^{-1}.[164] Study of 24 high-As rice samples grown in the United States revealed that they contained mainly iAs(III) and DMA(V) species. The DMA(V) content increased proportionally with the total As content, while the concentration of iAs(III) (approximately 0.1 mg kg^{-1}) did not change significantly. From this, the authors concluded that rice can be divided into two groups according to its As content: DMA(V) and iAs rice. According to this theory, varieties grown in the United States belong to the first category, while those growing in Europe and Asia to the second one. It was suggested that this difference was due to plant genetic reasons. Although this theory cannot be completely ruled out, the type of soil composition and irrigation is more likely to be responsible for the high DMA(V) content. Furthermore, the recent use of pesticides containing DMA(V) and MMA(V) in the central and southern parts of the United States may play a role.[165,166]

It is common practice to perform As speciation of rice in ground form. This is understandable since rice flour is used for cooking in Asia and also from an analytical chemistry point of view, as the homogeneity of the sample is a key issue to obtain reliable results. In China, rice is usually cooked with small aliquots of water. In this way, rice absorbs the available water. However, this approach does not provide real information for estimating the As exposure from rice consumption for example, in the so-called Western societies, where rice is usually consumed as a side dish, so it is cooked in plenty of water and the remaining water is poured off. Thus, there is a need to perform As speciation also according to this latter kitchen technology methods. On the other hand, milk rice, risotto, etc. requires a different approach. The As speciation performed on rice cooked in As-containing water diluted with varying degrees of deionized water (DW) (1:1 to 4:1 v/v) demonstrated the ability of rice grains to absorb As during cookin.[167] Bae *et al.*[168] reached to a similar conclusion by determining the total As concentration of rice cooked with local water in Bangladeshi households.

Extraction of rice grains with room temperature and boiling DW provides valuable information on the removal of toxic elements in them. By simulating two simple kitchen technologies (washing and cooking) corresponding to one of the possible ways of preparing rice, it is possible to determine the extent to which the As content of the rice grains can be removed.[26] The minimum cooking time was chosen so that the rice grains would soften in line with the recommendations for cooking instant and long grain rice. The total As concentrations of the investigated rice samples did not exceed 171.3 ± 7.1 μg kg^{-1}, which was also in agreement with previous literature data.[96] When determining the As content of polished rice samples taken from different geographical areas of ten countries, seven-fold differences were found in the median value. Samples from Egypt (0.04 mg kg^{-1}) and India (0.07 mg kg^{-1}) had the lowest As concentration, while the United States (0.25 mg kg^{-1}) and France (0.28 mg kg^{-1}) had the highest one.[169] Since 2014, the health limit value for iAs set by WHO/FAO is 0.2 mg kg^{-1} in polished rice.[170] According to the water extraction results, 8–17% and 29–42% of As could be removed from the studied rice grains by washing room with temperature DW and cooking, respectively.[23] These results are also consistent with results reported by Bae *et al.*,[168] according to which 57-81% of As remained in varieties of rice cooked in Bangladeshi households.

For removal of elements by washing and cooking, it also plays a role if metal contamination affects the surface of the rice grains or its deeper layers.[171] Cooking may increase the diffusion rate of these ions[171]. Confocal μ-XRF analysis provides an opportunity to track the extraction of the elements and interpret the results obtained by measurements in bulk phase (e.g., ICP-MS). By confocal μ-XRF analysis, normalization of the intensity confirmed that the intensity of the X-ray fluorescent signal of most elements was higher in the near-surface part of the studied rice grains.[26] Thus, elements (e.g., As) were enriched from the surface of the rice grain to a depth of about 80 μm.[26]

In terms of As speciation in washed and cooked rice, the main extractable chemical form of As was also iAs(III), with proportions ranging from about 40 to 70% in white rice grains at room temperature and between 60 and 75% in boiling DW extracts.[23] Arsenocholine, DMA(V) and iAs(V) could also be determined in these samples. In contrast, cooked samples were generally dominated by the presence of iAs(V), suggesting that iAs(III) was partially oxidized during cooking.[23] Pal *et al.* reached to the same conclusion by performing As speciation following cooking of rice grown in As-contaminated areas of West Bengal in distilled water.[172]

Arsenic intake by breastfeeding

Exclusive breastfeeding is recommended for the first six months after birth for infants, because of providing optimal conditions for the immunological and psychological development of the newborn. However, potential inorganic contaminants in breast milk (e.g., As) are transmitted to the neonate through breastfeeding.[173] Toxic elements are absorbed to a greater extent in neonates than in adults, and bile excretion is low in the first weeks after delivery.[174] Arsenic occurrence in breast milk is mainly due to a diet rich in marine fish and seafood.[175] The As concentration of breast milk in endemic As-contaminated areas (e.g., aquifers) was found to be lower than expected.[176] According to the Joint FAO/WHO Expert Committee on Food Additives, the lower limit of the benchmark dose ($BDML_{0.5}$) with iAs-induced lung cancer as the end point of toxicity effect is 3 μg kg^{-1} BW.[177]

Twenty-four-hour pooled samples taken from the same mother[178] would be ideal for elemental analysis in breast milk. Since this is not possible, usually, samples are collected from only one mammal gland that has not been involved in breastfeeding for 2-3 hours prior to sampling.[178] The most common sample preparation procedure is MW-assisted acid digestion using a mixture of concentrated HNO_3 and hydrogen peroxide solutions. If freeze-drying is applied before analysis,[178-180] the sample demand is smaller.[180] The digested samples are nowadays analyzed by ICP-MS.[181-183]

A recent study conducted in Hungary on the determination of As in breast milk of non-smoking, exclusively breastfeeding volunteers with an average body mass index and with no occupational exposure to As in the early stages of breastfeeding, the mean concentration of As in the samples was 0.41 ± 0.20 $\mu g\ L^{-1}$. However, the median values of the As concentrations determined for all samples were 0.35 $\mu g\ L^{-1}$, respectively, indicating an almost symmetric distribution of the concentration dataset.[184] The median As concentration in the abovementioned study was approximately 45% larger than in the Hungarian mature milk samples published in 1991 and analyzed by neutron activation analysis.[185] Unlike in 1991, the results of the recent study showed that As was detectable in all samples. This is due not only to the advances in instrumental analysis, but also to the changes in the dietary habits of the last 30 years. Today, more poultry meat is consumed, because it is considered healthier, and a larger selection of seafood and marine fish is available in Hungary. Lovreglio *et al.*[186] found that less toxic AsB and AsC occur even in poultry meat. Concentration values of As in samples taken in Taiwan and Sweden were also similar to those obtained in the aforementioned recent study.[187] Higher As concentrations have been reported from Greece.[175] Outside Europe, higher values have been reported from Pakistan (0.5 $\mu g\ L^{-1}$),[188] Iran (0.85 $\mu g\ L^{-1}$),[189] Bangladesh (1 $\mu g\ L^{-1}$),[190] Japan (1.4 $\mu g\ L^{-1}$)[191] and Lebanon (2.3 $\mu g\ L^{-1}$).[192] In mature breast milk samples analyzed in Portugal, As concentration was about 5.8 $\mu g\ L^{-1}$, presumably due to regular consumption of fish and seafood, while the Pb concentration was lower.[193]

Estimation of the intake of As in breast-fed infants can be performed by the method applied by Rebelo and Caldas[194] consisting of expressing the ratio between the determined concentration and the benchmark dose limit (BMDL) value. The value thus obtained is called margin of exposure (MOE). Its estimated value is inversely proportional to the risk to public health posed by a certain toxic element.[195] In the US, this value is typically above 50 for As.[196] According to literature data, the MOE values for As in Europe are about 50 or higher.[194] No significant relationship could be found between the age of the volunteers and the As concentrations in breastmilk samples.[184] In return, the MOE values calculated for Pb proved that this element continues to pose a potential risk to infants.[184] Moreover, the Pb concentration in breast milk was significantly correlated with the age of the mothers.[184,197]

Organs playing an important role in reproduction and offspring care are protected against As, As-speciation is difficult to achieve.

Potential threat represented by As in complementary and alternative medicinal products

General considerations

According to the WHO, traditional medicine is defined as CAM. This category includes traditional Chinese medicine, the Ayurvedic or Indian medicine system, the Tamil Siddha, the Persian - Arabic Unani, and homeopathy. Of these, Ayurveda is only surpassed in popularity by traditional Chinese medicine. The main difference between these two systems is that Ayurvedic preparations can not only contain herbs but also minerals and elementary state elements (e.g., Hg, S). From an analytical chemistry point of view, Ayurvedic preparations can be classified as herbal, herbomineral and animal ones. Of the metals used to make Ayurvedic preparations, Hg is of primary importance. Among the minerals, those containing As such as realgar (α-As_4S_4) and auripigment (As_2S_3) are common constituents. Through the current numerous infocommunication channels, these products have become available worldwide. Online sales of Ayurvedic products have recently skyrocketed.[198] By performing elemental analysis of 193 Ayurvedic preparations purchased at random from the Internet, the authors of the study found that Pb, Hg or As were detectable in 21% of the products. The order of detection frequency of these toxic elements in the investigated products was: Pb> Hg> As. Heavy metal and As contamination was twice as common in herbomineral products as in other types of products. In the Netherlands, about 20% of the labels of 292 traditional herbal preparations used in Ayurveda, traditional Chinese and Tibetan medicine, indicated the presence of Hg, As or Pb. These elements could be determined by ICP-MS in 64% of the samples. In about 20% of the investigated samples (n = 59), the amount of these elements exceeded the maximum allowable intake for humans with a single recommended dose. For 26 products, the estimated weekly As intake was 0.53 and 427 mg.[199] The European Commission classifies Ayurvedic preparations only as food supplements under Directive 2002/46/EC. In the United States, where Ayurvedic preparations are subject to the requirements of the Food Supplements Act 1994 (DSHEA - Dietary Supplement Health and Education Act), there is no need to demonstrate the safe use or efficacy of these products.[198]

The large number of scientific communications on Ayurvedic research published in peer reviewed journals contain relatively less information from analytical chemistry point of view. Elemental content of Ayurvedic products has been studied by neutron activation analysis,[200] voltammetry,[201] XRF spectroscopy[199,202] or ICP-MS.[199] The advantage of XRF is that it is a non-destructive analytical technique, although not suitable for the determination of trace elements, while ICP-MS has extremely low detection limits, its acquisition and maintenance costs are much higher, and sample preparation is a much more time-consuming process. Literature data are difficult to be compared due to the wide range of Ayurvedic products. An attempt is presented hereby. Khandpur *et al.*[203] investigated eight different Ayurvedic formulations, the As concentrations in these products were ranging from 5 to 248 mg kg^{-1}. The concentration of As in a blood sample from a patient undergoing Ayurvedic treatment was 0.202 mg L^{-1}, although the normal value is generally <0.06 µg L^{-1}. Saper *et al.* studied 70 Ayurvedic formulations purchased in stores[204] and 193 online[198] in the United Stated by X-ray fluorescence spectrometry. Of the 70 samples investigated, Pb, Hg, and As were quantifiable in 14. The concentration of As ranged from 0.037 to 8.13 g kg^{-1}.[204]

In a recent study, analysis by ICP-SF-MS after MW-assisted acid digestion of six different Ayurvedic products purchased online from India and one purchased in Hungary showed that the As concentration was relatively low, i.e., 0.41 ± 0, 05 mg kg^{-1} in the plant-derived sample (containing *Centella Asiatica*).[14] The concentrations of As in the rest of the investigated samples ranged from 0.04 to 771 mg kg^{-1}. The concentration of As (106 ± 13 g kg^{-1}) was the highest in an As_2O_3-containing preparation.[14] Due to its significant As content, an Ayurvedic ointment was only worth analyzing by energy dispersive X-ray fluorescence (EDXRF). Thus, the spectra of the ointment recorded by EDXRF confirmed that As was the major component of the sample.

Arsenic bioaccessibility in Ayurvedic complementary and alternative medicines

Knowledge of the total concentration of a toxic element (e.g., As) is necessary but not sufficient to estimate adverse health risks, so it is important to quantify the amounts of element(s) that could be assimilated after ingestion of a certain dose. However, application of *in vivo* experiments raises ethical issues. Thus, several *in vitro* procedures have been developed to simulate the effects of the human digestive process.[205]

Data from only a few complex analytical studies on bioaccessible concentrations of toxic elements in Ayurvedic formulations have been reported.[206-208] Giacomino *et al.*[209] used a mixture of hydrochloric acid, sodium chloride and pepsin to simulate gastric juice. Pancreatin, potassium dihydrogen phosphate and sodium hydroxide solution were used to simulate duodenal fluid. However, As_2O_3 and As sulfides can hardly be dissolved in HCl due to their (thio)acid anhydride properties, so their dissolution is expected only in alkaline media. However, due to their high initial concentration, As uptake by the human body may still be significant. For samples containing As_2O_3/cinnabar red and α-As_4S_4/HgS, the estimated daily intake exceeded the WHO recommended daily dose of 3.0 µg iAs BW kg^{-1} by a factor of 11.1 to 21.4.[14]

After incubation of 17 products purchased from pharmacies and local markets in Italy and India, respectively, in suspensions of gastric and duodenal fluids, a remarkable dissolution was observed for As.[209,210]

Koch *et al.*[206] characterized the elemental content of CAMs by ICP-MS and XANES, focusing on the bioaccessibility of As and Pb, respectively, by incubation in a model suspension containing pancreatic and bile extract, respectively. Interestingly, *bhasma* formulations characterized by a strong covalent bond between As(III) and S atoms increased the bioacessibility of As. The authors explained this phenomenon by the oxidation of As(III), which was confirmed by XANES measurements. In addition, the estimated dietary intake using the bioaccessible As concentration values in several cases was greater than the daily tolerable limit set by California Proposition 65.[211] Bolan *et al.*[208] investigated bioaccessibility of As-containing rice and some Ayurvedic products of plant origin by using an exhaustive sequential extraction method proposed by Tessier *et al.*[212] as well as in simulated gastric and duodenal fluids. The concentration of bioaccessible As was highly dependent on the procedure used. However, pancreatin is one of the most preferred compounds for simulating duodenal fluid.[206,213-217] Extraction efficiencies determined by modeling bioaccessibility resulted in large differences. Therefore, it would be important to develop and adopt an appropriate extraction method.[207]

Arsenic speciation in phosphate-rich bioaccessible fractions of Ayurvedic preparations

Arsenic speciation in bioaccessible fractions is not a routine procedure. Until now, elemental speciation has been performed directly on Ayurvedic samples in solid state of matter using X-ray-based analytical techniques (e.g., XANES, extended X-ray absorption fine structure, EXAFS) due to the high toxic element concentration of the formulations.[206-208] In herbal preparations, the predominant As species were organic, while in mineral-containing Ayurvedic products, As occurs in inorganic form.

Two Ayurvedic formulations containing As in moderate concentration were studied after spiking to 150 µg kg^{-1} iAs(V) standard solution, incubation in a pH = 6.8 suspension simulating duodenal fluid and SAX-SPE separation. The highest recovery of about 80% for the iAs(V) species was obtained when the model solution did not contain phosphate ions. The presence of sodium hydroxide did not affect the recovery of iAs(V). Oxidation of iAs(III) was observed using optimal SPE conditions for two Ayurvedic (herbo)mineral preparations with high As content. Thus, 6.3% of the iAs(III) in the bioaccessible fractions were oxidized in the sample containing As_2O_3 and cinnabar red, compared to 33% in the formulation containing realgar and HgS. Spiking a simple homeopathic sample matrix containing theoretically As_2O_3 with a 1 µg L^{-1} iAs(V) standard solution, recovery of total As and iAs(V) of samples subjected to SAX-SPE were about 97% and 95%, respectively.[14] The recovery of total As and iAs(V) was approximately 90% and 80%, respectively, when the homeopathic product was spiked to 10 µg L^{-1} iAs(V) and then incubated with the duodenal modeling suspension without the addition of phosphate ions. Study of the homeopathic remedy has also shown that it was mainly phosphate ions that prevent the separation of As species with SPE and not the matrix constituents. Breakthrough results in this area with efficient As-speciation methods have yet to be published.

CONCLUSIONS

In recent decades, several As speciation methods have been developed, many of which are cost-effective as alternatives to the original on-line hyphenated techniques consisting of an HPLC separation followed by atomic spectroscopic detection. In the case of TLC, the OPLC-TXRF method provides reproducible operating conditions, resulting in faster separation and better resolution. Laser ablation for PEI-TLC plates followed by ICP-MS measurements proved to be suitable for the screening of biological samples. *In situ* separation of As species in water with SAX-SPE can be easily accomplished from matrices up to pH<9 with higher enrichment and/or dilution, followed by laboratory analysis of acid-preserved samples. The cost-efficient TXRF spectrometry allows the simultaneous determination of several other trace elements in drinking water. At the regional level (e.g., in Hungary), the exposure of the population to As has been significantly reduced since 2017 by drilling new wells and establishing new waterworks in settlements of poor water quality. To the best of my knowledge, however, As speciation has not been widely used in the development of As removal technologies. To date, there is no real breakthrough in the use of As speciation methods. Arsenic contamination is challenging since inorganic As species cannot easily be decomposed compared to purely organic contaminants. Thus, the removed As is concentrated in hazardous waste disposal, which is also costly. Another possibility is to immobilize As e.g., in the soil. In this respect, biowaste is a promising raw material after conversion to AC with concentrated H_2SO_4. In order to increase mechanical stability and adsorption, it is advisable to use a silicate-containing material after charring to stabilize As (e.g., Florisil®). Although Florisil® is a widespread chromatographic adsorbent, it is expensive, but simple charring processes can keep the cost of producing ACs low (e.g., no application of energy-demanding pyrolysis).

In the lack of a definitive solution for the removal of As from drinking water, it is important to estimate the exposure of the population in areas with extensive natural As pollution. Dietary intake is the major source for nonoccupational exposure to As. Washing and cooking rice in plenty of water is advisable in regions affected by As-contaminated water. In this way, large amounts of As can be removed. According to a recent study, significant efforts are still being made in several parts of the world (e.g., West Bengal) to develop intervention procedures to reduce As pollution of waters.[218]

The estimated intake of As from the bioaccessible fractions of Ayurvedic formulations poses a health risk. If public health employees were more empathetic to their patients, or if a significant portion of the contemporary society did not reject the results of scientific research, there would be no demand for such dubious quality drugs. A recent review paper[219] highlighted the need to (i) elucidate the mechanisms of incorporation of toxic elements into CAM formulations, (ii) to understand the biogeochemical cycles of these elements and in particular their metabolites; (iii) to develop appropriate speciation methods to study the *in vitro* bioaccessibility of toxic elements; and (iv) conducting clinical trials and developing stricter legislation. In accordance with the current practice, elemental speciation in Ayurvedic preparations containing high concentrations of toxic elements is performed directly in the solid state by X-ray-based analytical techniques (XANES, EXAFS). Although the success of X-ray measurement techniques is undeniable, more accurate information could be obtained using appropriate *in vitro* methods to study bioaccessibility, but extreme pH and complex sample/extraction matrix represent real obstacles.

Even today, several reviews deal with the estimation of health risks caused by dietary As intake[220,221] and the development of As speciation methods.[222] Regarding the reduction of As contamination in environmental compartments and food items, many questions still should be addressed in the future.

Conflicts of interest

There have not been any financial conflicts of interest for writing this manuscript.

Acknowledgements

This review has not received any funding.

REFERENCES

(1) European Commission Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption. Official Journal L 330, 05/12/1998, 0032 – 0054. https://eur-lex.europa.eu/legal-content/EN/TXT/HTML/?uri=CELEX:31998L0083&from=HU [Accessed 28 November 2021].

(2) Mikutta, C.; Mandaliev, P. N.; Mahler, N.; Kotsev, T.; Kretzschmar, R. Bioaccessibility of Arsenic in Mining-Impacted Circumneutral River Floodplain Soils. *Environ. Sci. Technol.* **2014**, *48*, 13468-13477. https://doi.org/10.1021/es502635t

(3) Prohaska, T.; Stingeder, G. Speciation of Arsenic: Arsenic and Arsenic Species in Environment and Human Nutrition. In: Cornelis, R.; Caruso, J.; Crews, H.; Heumann, K. (Eds.). *Handbook of Elemental Speciation II – Species in the Environment, Food, Medicine and Occupational Health,* Vol. 2. Wiley, Chichester, **2005**, 69-85. https://doi.org/10.1002/0470856009

(4) Brown, J. L.; Kitchin, K. T.; George, M. Dimethylarsinic acid treatment alters six different rat biochemical parameters: Relevance to arsenic carcinogenesis. *Teratog. Carcinog. Mutagen*. **1997**, *17*, 71–84. https://doi.org/10.1002/(sici)1520-6866(1997)17:2<71::aid-tcm3>3.0.co;2-b

(5) Human Metabolome Database (HMDB). Showing metabocard for Dimethylarsinous acid. http://www.hmdb.ca/metabolites/HMDB0012216 [Accessed 28 November 2021].

(6) Human Metabolome Database (HMDB). Showing metabocard for Methylarsonite. http://www.hmdb.ca/metabolites/HMDB0012259 [Accessed 28 November 2021].

(7) Odanaka, Y.; Tsuchiya, N.; Matano, O.; Goto, S. Characterization of arsenic metabolites in rice plant treated with DSMA (disodium methanearsonate). *J. Agric. Food Chem*. **1985**, *33*, 757–763. https://doi.org/10.1021/jf00064a050

(8) U.S. Food & Drug Administration (FDA). *Arsenic-based Animal Drugs*. https://www.fda.gov/animal-veterinary/product-safety-information/arsenic-based-animal-drugs [Accessed 28 November 2021].

(9) Chen, G.; Liu, H.; Zhang, W.; Li, B.; Liu, L.; Wang, G. Roxarsone exposure jeopardizes nitrogen removal and regulates bacterial community in biological sequential batch reactors. *Ecotox. Environ. Safe*. **2018**, *159*, 232-239. https://doi.org/10.1016/j.ecoenv.2018.05.012

(10) European Commission. *Arsenic health risk assessment and molecular epidemiology.* http://cordis.europa.eu/result/rcn/44352_en.html [Accessed 28 November 2021].

(11) Hamza, J. *Arsenic derogation for drinking water and related measures*. https://prezi.com/zjt6lvskaxs4/copy-of-az-ivoviz-arzen-derogacio-es-az-azzal-kapcsolatbanszukseges-intezkedesek/ [Accessed 28 November 2021].

(12) Reid, M. S.; Hoy, K. S.; Schofield, J. R. M.; Uppal, J. S.; Lin, Y.; Lu, X.; Peng, H.; Le, X. C. Arsenic speciation analysis: A review with an emphasis on chromatographic separations. *TrAC, Trends Anal. Chem*. **2020**, *123*, 115770. https://doi.org/10.1016/j.trac.2019.115770

(13) Anemana, T.; Óvári, M.; Varga, M.; Mihály, J.; Uzinger, N.; Rékási, M.; Yao, J.; Tatár, E.; Streli, C.; Záray, G.; et al. Granular activated charcoal from peanut (*Arachis hypogea*) shell as a new candidate for stabilization of arsenic in soil. *Microchem J*. **2019**, *149*, 104030. https://doi.org/10.1016/j.microc.2019.104030

(14) Kovács, D.; Veszely, Á.; Enesei, D.; Óvári, M.; Záray, G.; Mihucz, V. G. Feasibility of ion-exchange solid phase extraction inductively coupled plasma mass spectrometry for discrimination between inorganic As(III) and As(V) in phosphate-rich in vitro bioaccessible fractions of Ayurvedic formulations. *Spectrochim. Acta, Part B* **2019**, *153*, 1-9. https://doi.org/10.1016/j.sab.2019.01.002

(15) Resano, M.; García – Ruiz, E.; Mihucz, V. G.; Móricz, Á. M.; Záray, G.; Vanhaecke, F. Rapid screening method for arsenic speciation by combining thin layer chromatography and laser ablation-inductively coupled plasma-dynamic reaction cell-mass spectrometry. *J. Anal. At. Spectrom*. **2007**, *22*, 1158–1162. https://doi.org/10.1039/b704963c

(16) Meirer, F.; Pepponi, G.; Streli, C.; Wobrauschek, P.; Mihucz, V. G.; Záray, G.; Czech, V.; Broekaert, J. A. C.; Fittschen, U. E. A.; Falkenberg, G. Application of synchrotron-radiation-induced TXRF-XANES for arsenic speciation in cucumber (*Cucumis sativus* L.) xylem sap. *X-Ray Spectrom*. **2007**, *36*, 408–412. https://doi.org/10.1002/xrs.993

(17) Mihucz, V. G.; Bencs, L.; Koncz, K.; Tatár, E.; Weiszburg, T.; Záray, G. Fast arsenic speciation in water by on-site solid phase extraction and high-resolution continuum source graphite furnace atomic absorption spectrometry. *Spectrochim. Acta, Part B* **2017**, *128*, 30-35. https://doi.org/10.1016/j.sab.2016.12.010

(18) Mihucz, V. G.; Enesei, D.; Veszely, **Á.**; Bencs, L.; Pap-Balázs, T.; Óvári, M.; Streli, C.; Záray, G. A simple method for monitoring of removal of arsenic species from drinking water applying on-site separation with solid phase extraction and detection by atomic absorption and X-ray fluorescence based techniques. *Microchem. J.* **2017**, *135*, 105-113. https://doi.org/10.1016/j.microc.2017.08.006

(19) Sugár, É.; Tatár, E.; Záray, G.; Mihucz, V. G. Relationship between arsenic content of food and water applied for food processing. *Food Chem. Toxicol*. **2013**, *62*, 601–608. https://doi.org/10.1016/j.fct.2013.09.028

(20) Shraim, A.; Chiswell, B.; Olszowy, H. Speciation of arsenic by hydride generation–atomic absorption spectrometry (HG–AAS) in hydrochloric acid reaction medium. *Talanta* **1999**, *50*, 1109-1127. https://doi.org/10.1016/s0039-9140(99)00221-0

(21) Cerveira, C.; Pozebon, D.; de Moraes, D. P.; de Fraga, J. C. S. Speciation of inorganic arsenic in rice using hydride generation atomic absorption spectrometry (HG-AAS). *Anal. Methods* **2015**, *7*, 4528-4534. https://doi.org/10.1039/C5AY00563A

(22) Sugár, É.; Tatár, E.; Záray, G.; Mihucz, V. G. Field separation-based speciation analysis of inorganic arsenic in public well water in Hungary. *Microchem. J.* **2013**, *107*, 131–135. https://doi.org/10.1016/j.microc.2012.05.025

(23) Mihucz, V. G.; Tatár, E.; Virág, I.; Zang, C.; Jao, Y.; Záray, G. Arsenic removal from rice by washing and cooking with water. *Food Chem*. **2007**, *105*, 1718–1725. https://doi.org/10.1016/j.foodchem.2007.04.057

(24) Mihucz, V. G.; Tatár, E.; Virág, I.; Cseh, E.; Fodor, F.; Záray, G. Arsenic speciation in xylem sap of cucumber (*Cucumis sativus* L.). *Anal. Bioanal. Chem*. **2005**, *383*, 461–466. https://doi.org/10.1007/s00216-005-3325-y

(25) Mihucz, V. G.; Móricz, Á. M.; Kröpfl, K.; Szikora, S.; Tatár, E.; Parra, L. M. M.; Záray, G. Development of off-line layer chromatographic and total reflection X-ray fluorescence spectrometric methods for arsenic speciation. *Spectrochim. Acta, Part B* **2006**, *61*, 1124–1128. https://doi.org/10.1016/j.sab.2006.08.011

(26) Mihucz, V. G.; Silversmit, G.; Szalóki, I.; de Samber, B.; Schoonjans, T.; Tatár, E.; Vincze, L.; Virág, I.; Yao, J.; Záray, G. Removal of some elements from washed and cooked rice studied by inductively coupled plasma mass spectrometry and synchrotron based confocal micro-X-ray fluorescence. *Food Chem*. **2010**, *121*, 290–297. https://doi.org/10.1016/j.foodchem.2009.11.090

(27) Biswas, A.; Majumder, S.; Neidhardt, H.; Halder, D.; Bhowmick, S.; Mukherjee-Goswami, A.; Kundu, A.; Saha, D.; Berner, Z.; Chatterjee, D. Groundwater chemistry and redox processes: Depth dependent arsenic release mechanism. *Appl. Geochem*. **2011**, *26*, 516–525. https://doi.org/10.1016/j.apgeochem.2011.01.010

(28) Le, X. C.; Yalcin, S.; Ma, M. Speciation of Submicrogram per Liter Levels of Arsenic in Water: On-Site Species Separation Integrated with Sample Collection. *Environ. Sci. Technol*. **2000**, *34*, 2342–2347 https://doi.org/10.1021/es991203u

(29) Impellitteri, C. A. Effects of pH and competing anions on the speciation of arsenic in fixed ionic strength solutions by solid phase extraction cartridges. *Water Res*. **2004**, *38*, 1207–1214. https://doi.org/10.1016/j.watres.2003.11.023

(30) Ujević, M.; Duić, Ž.; Casiot, C.; Sipos, L.; Santo, V.; Dadić, Ž.; Halamić, J. Occurrence and geochemistry of arsenic in the groundwater of Eastern Croatia. *Appl. Geochem*. **2010**, *25*, 1017–1029. https://doi.org/10.1016/j.apgeochem.2010.04.008

(31) Issa, N. B.; Rajaković-Ognjanović, V. N.; Jovanović, B. M.; Rajaković, L. V. Determination of inorganic arsenic species in natural waters – Benefits of separation and preconcentration on ion exchange and hybrid resins. *Anal. Chim. Acta* **2010**, *673*, 185–193. https://doi.org/10.1016/j.aca.2010.05.027

(32) O’Reilly, J.; Watts, M. J.; Shaw, R. A.; Marcilla, A. L.; Ward, N. I. Arsenic contamination of natural waters in San Juan and La Pampa, Argentina. *Environ. Geochem. Health* **2010**, *32*, 491–515. https://doi.org/10.1007/s10653-010-9317-7

(33) Watts, M. J.; O’Reilly, J.; Marcilla, A. L.; Shaw, R. A.; Ward, N. I. Field based speciation of arsenic in UK and Argentinean water samples. *Environ. Geochem. Health* **2010**, *32*, 479–490. https://doi.org/10.1007/s10653-010-9321-y

(34) Jitmanee, K.; Oshima, M.; Motomizu, S. Speciation of arsenic(III) and arsenic(V) by inductively coupled plasma-atomic emission spectrometry coupled with preconcentration system. *Talanta* **2005**, *66*, 529–533. https://doi.org/10.1016/j.talanta.2004.09.024

(35) Piqué, À.; Grandia, F.; Canals, À. Processes releasing arsenic to groundwater in the Caldes de Malavella geothermal area, NE Spain. *Water Res*. **2010**, *44*, 5618–5630. https://doi.org/10.1016/j.watres.2010.07.012

(36) Meng, X.; Korfiatis, G. P.; Christodoulatos, C.; Bang, S. Treatment of arsenic in Bangladesh well water using a household co-precipitation and filtration system. *Water Res*. **2001**, *35*, 2805–2810. https://doi.org/10.1016/S0043-1354(01)00007-0

(37) Liang, P.; Liu, R. Speciation analysis of inorganic arsenic in water samples by immobilized nanometer titanium dioxide separation and graphite furnace atomic absorption spectrometric determination. *Anal. Chim. Acta* **2007**, *602*, 32–36. https://doi.org/10.1016/j.aca.2007.09.012

(38) Sperling, M.; Yin, X. F.; Welz, B. Differential determination of arsenic(III) and total arsenic using flow injection on-line separation and preconcentration for graphite furnace atomic absorption spectrometry. *Spectrochim. Acta, Part B* **1991**, *46*, 1789–1801. https://doi.org/10.1016/0584-8547(91)80206-I

(39) Koh, J.; Kwon, Y.; Pak, Y. N. Separation and sensitive determination of arsenic species (As3+/As5+) using the yeast-immobilized column and hydride generation in ICP–AES. *Microchem. J.* **2005**, *80*, 195–199. https://doi.org/10.1016/j.microc.2004.07.011

(40) Anthemidis, A. N.; Martavaltzoglou, E. K. Determination of arsenic(III) by flow injection solid phase extraction coupled with on-line hydride generation atomic absorption spectrometry using a PTFE turnings-packed micro-column. *Anal. Chim. Acta* **2006**, *573*, 413–418. https://doi.org/10.1016/j.aca.2005.12.055

(41) Xiong, C. M.; He, M.; Hu, B. On-line separation and preconcentration of inorganic arsenic and selenium species in natural water samples with CTAB-modified alkyl silica microcolumn and determination by inductively coupled plasma-optical emission spectrometry. *Talanta* **2008**, *76*, 772–779. https://doi.org/10.1016/j.talanta.2008.04.031

(42) Chen, D.; Huang, C.; He, M.; Hu, B. Separation and preconcentration of inorganic arsenic species in natural water samples with 3-(2-aminoethylamino) propyltrimethoxysilane modified ordered mesoporous silica micro-column and their determination by inductively coupled plasma optical emission spectrometry. *J. Hazard. Mater*., **2009**, *164*, 1146–1151. https://doi.org/10.1016/j.jhazmat.2008.09.022

(43) Rahman, I. M. M.; Begum, Z. A.; Furusho, Y.; Mizutani, S.; Maki, T.; Hasegawa, H. Selective Separation of Tri- and Pentavalent Arsenic in Aqueous Matrix with a Macrocycle-Immobilized Solid-Phase Extraction System. *Water Air Soil Pollut*. **2013**, *224*, 1526-1537. https://doi.org/10.1007/s11270-013-1526-0

(44) Issa, N. B.; Rajaković-Ognjanović, V. N.; Marinković, A. D.; Rajaković, L. V. Separation and determination of arsenic species in water by selective exchange and hybrid resins. *Anal. Chim. Acta* **2011**, *706*, 191–198. https://doi.org/10.1016/j.aca.2011.08.015

(45) Chen, S. Z.; Zhan, X. L.; Lu, D. B.; Liu, C.; Zhu, L. Speciation analysis of inorganic arsenic in natural water by carbon nanofibers separation and inductively coupled plasma mass spectrometry determination. *Anal. Chim. Acta* **2009**, *634*, 192-196. https://doi.org/10.1016/j.aca.2008.12.018

(46) López-García, I.; Rivas, R. E.; Hernández-Córdoba, M. Use of carbon nanotubes and electrothermal atomic absorption spectrometry for the speciation of very low amounts of arsenic and antimony in waters. *Talanta* **2011**, *86*, 52-57. https://doi.org/10.1016/j.talanta.2011.07.105

(47) Zhang, Y.; Wang, W.; Li, L.; Huang, Y.; Cao, J. Eggshell membrane-based solid-phase extraction combined with hydride generation atomic fluorescence spectrometry for trace arsenic(V) in environmental water samples. *Talanta* **2010**, *80*, 1907–1912. https://doi.org/10.1016/j.talanta.2009.10.042

(48) Smedley, P. L.; Kinniburgh, D. G. A review of the source, behaviour and distribution of arsenic in natural waters. *Appl. Geochem*. **2002**, *17*, 517–568. https://doi.org/10.1016/S0883-2927(02)00018-5

(49) Abedin, M. J.; Feldmann, J.; Meharg, A. A. Uptake Kinetics of Arsenic Species in Rice Plants. *Plant Physiol*. **2002**, *128*, 1120–1128. https://doi.org/10.1104/pp.010733

(50) Takamatsu, T.; Aoki, H.; Yoshida, T. Determination of arsenate, arsenite, monomethylarsonate, and dimethylarsinate in soil polluted with arsenic. *Soil Sci*. **1982**, *133* (4), 239–246.

(51) Farnfield, H. R.; Marcilla, A. L.; Ward, N. I. Arsenic speciation and trace element analysis of the volcanic río Agrio and the geothermal waters of Copahue, Argentina. *Sci. Total Environ*. **2012**, *433*, 371–378. https://doi.org/10.1016/j.scitotenv.2012.05.098

(52) Wang, S.; Mulligan, C. N. Occurrence of arsenic contamination in Canada: Sources, behavior and distribution. *Sci. Total Environ*. **2006**, *366*, 701–721. https://doi.org/10.1016/j.scitotenv.2005.09.005

(53) Varsányi, I.; Matray, J. M.; Kovács, L. O. Hydrogeochemistry in two adjacent areas in the Pannonian Basin (Southeast-Hungary). *Chem. Geol*. **1999**, *156*, 25-39. https://doi.org/10.1016/S0009-2541(98)00178-8

(54) KvVM-EüM-FVM common order about the standard limits and measurement of contamination for the protection of underground water and geological medium. (In Hungarian). *Magyar Közlöny* **2009**, *51*, 14398-14414.

(55) Ódor, L.; Horváth, I.; Fügedi, U. J. Low-density geochemical mapping in Hungary. *Geochem. Explor*. **1997**, *60*, 55–66. https://doi.org/10.1016/S0375-6742(97)00025-3

(56) Szocs, T.; Horváth, I.; Bartha, A.; Bertalan, É.; Ballók, M.; Horváth, É. Speciation studies in understanding high As content in ground water. *Mineral. Mag*. **2008**, *72*, 107-111. https://doi.org/10.1180/minmag.2008.072.1.507

(57) Bezuglova, O. S.; Gorbov, S. N.; Tischenko, S. A.; Shimko, A. E. Use of brown coal as a detoxifier of soils contaminated with heavy metals. *J. Geochem. Explor*. **2018**, *184*, 232–238. https://doi.org/10.1016/j.gexplo.2016.11.004

(58) Pukalchik, M.; Panova, M.; Karpukhin, M.; Yakimenko, O.; Kydralieva, K.; Terekhova, V. J. Using humic products as amendments to restore Zn and Pb polluted soil: a case study using rapid screening phytotest endpoint. *Soils Sediments* **2018**, *18*, 750–761. https://doi.org/10.1007/s11368-017-1841-y

(59) Zhang, W. H.; Tong, L. Z.; Yuan, Y.; Liu, Z. Y.; Huang, H.; Tan, F. F.; Qiu, R. L. Influence of soil washing with a chelator on subsequent chemical immobilization of heavy metals in a contaminated soil. *J. Hazard. Mater*. **2010**, *178*, 578-587. https://doi.org/10.1016/j.jhazmat.2010.01.124

(60) Bolan, N.; Kunhikrishnan, A.; Thangarajan, R.; Kumpiene, J.; Park, J.; Makino, T.; Kirkham, M. B.; Scheckel, K. Remediation of heavy metal(loid)s contaminated soils – To mobilize or to immobilize? *J. Hazard. Mater*. **2014**, *266*, 141-166. https://doi.org/10.1016/j.jhazmat.2013.12.018

(61) Tsang, D. C. W.; Yip, A. C. K. Comparing chemical-enhanced washing and waste-based stabilisation approach for soil remediation. *J. Soils Sediments* **2014**, *14*, 936-947. https://doi.org/10.1007/s11368-013-0831-y

(62) Lee, S. H.; Lee, J. S.; Choi, Y. J.; Kim, J. G. In situ stabilization of cadmium-, lead-, and zinc-contaminated soil using various amendments. *Chemosphere* **2009**, *77*, 1069-1075. https://doi.org/10.1016/j.chemosphere.2009.08.056

(63) Komarek, M.; Vanek, A.; Ettler, V. Chemical stabilization of metals and arsenic in contaminated soils using oxides – A review. *Environ. Pollut*. **2013**, *172*, 9-22. https://doi.org/10.1016/j.envpol.2012.07.045

(64) Tsang, D. C. W.; Olds, W. E.; Weber, P. A.; Yip, A. C. K. Soil stabilisation using AMD sludge, compost and lignite: TCLP leachability and continuous acid leaching. *Chemosphere* **2013**, *93*, 2839-2847. https://doi.org/10.1016/j.chemosphere.2013.09.097

(65) Michalková, Z.; Komarek, M.; Sillerova, H.; Puppa, L. D.; Joussein, E.; Bordas, F.; Vanek, A.; Vanek, O.; Ettler, V. Evaluating the potential of three Fe- and Mn-(nano)oxides for the stabilization of Cd, Cu and Pb in contaminated soils. *J. Environ. Manage*. **2014**, *146*, 226-234. https://doi.org/10.1016/j.jenvman.2014.08.004

(66) Ahmad, M.; Rajapaksha, A. U.; Lim, J. E.; Zhang, M.; Bolan, N.; Mohan, D.; Vithanage, M.; Lee, S. S.; Ok, Y. S. Biochar as a sorbent for contaminant management in soil and water: A review. *Chemosphere* **2014**, *99*, 19-33. https://doi.org/10.1016/j.chemosphere.2013.10.071

(67) Bian, R.; Joseph, S.; Cui, L.; Pan, G.; Li, L.; Liu, X.; Zhang, A.; Rutlidge, H.; Wong, S.; Chia, C.; et al. A three-year experiment confirms continuous immobilization of cadmium and lead in contaminated paddy field with biochar amendment. *J. Hazard. Mater*. **2014**, *272*, 121-128. https://doi.org/10.1016/j.jhazmat.2014.03.017

(68) Tsang, D. C. W.; Yip, A. C. K.; Olds, W. E.; Weber, P. A. Arsenic and copper stabilisation in a contaminated soil by coal fly ash and green waste compost. *Environ. Sci. Pollut. Res*. **2014**, *21*, 10194-10204. https://doi.org/10.1007/s11356-014-3032-3

(69) Rajapaksha, A. U.; Ahmad, M.; Vithanage, M.; Kim, K. R.; Chang, J. Y.; Lee, S. S.; Ok, Y. S. The role of biochar, natural iron oxides, and nanomaterials as soil amendments for immobilizing metals in shooting range soil. *Environ. Geochem. Health* **2015**, *37*, 1-12. https://doi.org/10.1007/s10653-015-9694-z

(70) Mohan, D.; Singh, K. P.; Singh, V. K. Trivalent chromium removal from wastewater using low cost activated carbon derived from agricultural waste material and activated carbon fabric cloth. *J. Hazard. Mater*. **2006**, *135*, 280-295. https://doi.org/10.1016/j.jhazmat.2005.11.075

(71) Marris, E. Black is the new green. *Nature* **2006**, *442*, 624-626. https://doi.org/10.1038/442624a

(72) Glaser, B.; Birk, J. State of the scientific knowledge on properties and genesis of Anthropogenic Dark Earths in Central Amazonia (terra preta de Índio). *J. Geochim. Cosmochim. Acta*, **2012**, *82*, 39-51. https://doi.org/10.1016/j.gca.2010.11.029

(73) Yin, C. Y.; Aroua, M. K.; Daud, W. M. A. W. Review of modifications of activated carbon for enhancing contaminant uptakes from aqueous solutions. *Sep. Purif. Technol*. **2007**, *52*, 403-415. https://doi.org/10.1016/j.seppur.2006.06.009

(74) Lin, S. H.; Juang, R. S. Adsorption of phenol and its derivatives from water using synthetic resins and low-cost natural adsorbents: A review. *J. Environ. Manage*. **2009**, *90*, 1336-1349. https://doi.org/10.1016/j.jenvman.2008.09.003

(75) Dias, J. M.; Alvim-Ferraz, M. C.; Almeida, M. F.; Rivera-Utrillo, J.; Sánchez-Polo, M. Waste materials for activated carbon preparation and its use in aqueous-phase treatment: A review. *J. Environ. Manage*. **2007**, *85*, 833-846. https://doi.org/10.1016/j.jenvman.2007.07.031

(76) Samsuri, A. W.; Sadegh-Zadeh, F.; Seh-Bardan, B. J. Adsorption of As(III) and As(V) by Fe coated biochars and biochars produced from empty fruit bunch and rice husk. *J. Environ. Chem. Eng*. **2013**, *1*, 981-988. https://doi.org/10.1016/j.jece.2013.08.009

(77) Tavares, D.; Lopes, C.; Coelho, J.; Sánchez, M.; Garcia, A.; Duarte, A.; Otero, M.; Pereira, E. Removal of Arsenic from Aqueous Solutions by Sorption onto Sewage Sludge-Based Sorbent. *Water Air Soil Pollut*. **2012**, *223*, 2311-2321. https://doi.org/10.1007/s11270-011-1025-0

(78) Toor, M.; Jin, B. Adsorption characteristics, isotherm, kinetics, and diffusion of modified natural bentonite for removing diazo dye. *Chem. Eng. J*. **2012**, *187*, 79–88. https://doi.org/10.1016/j.cej.2012.01.089

(79) Kim, M. J.; Nriagu, J.; Haack, S. Arsenic species and chemistry in groundwater of southeast Michigan. *Environ. Pollut*. **2002**, *120*, 379–390. https://doi.org/10.1016/S0269-7491(02)00114-8

(80) Varsányi, I.; Kovács, L. Ó. Arsenic, iron and organic matter in sediments and groundwater in the Pannonian Basin, Hungary. *Appl. Geochem*. **2006**, *21*, 949-963. https://doi.org/10.1016/j.apgeochem.2006.03.006

(81) Rowland, H. A. L.; Omoregie, E. O.; Millot, R.; Jimenez, C.; Mertens, J.; Baciu, C.; Hug, S. J.; Berg, M. Geochemistry and arsenic behaviour in groundwater resources of the Pannonian Basin (Hungary and Romania). *Appl. Geochem*. **2011**, *26*, 1–17. https://doi.org/10.1016/j.apgeochem.2010.10.006

(82) Thundiyil, J. G.; Yuan, Y.; Smith, A. H.; Steinmaus, C. Seasonal variation of arsenic concentration in wells in Nevada. *Environ. Res*. **2007**, *104*, 367–373. https://doi.org/10.1016/j.envres.2007.02.007

(83) Kumar, P.; Kumar, M.; Ramanathan, A. L.; Tsujimura, M. Tracing the factors responsible for arsenic enrichment in groundwater of the middle Gangetic Plain, India: a source identification perspective *Environ. Geochem. Health* **2010**, *32*, 129–146. https://doi.org/10.1007/s10653-009-9270-5

(84) Nordstrom, D. K. Worldwide Occurrences of Arsenic in Ground Water. *Science* **2002**, *296*, 2143-2144. https://doi.org/10.1126/science.1072375

(85) Ajtony, Z.; Laczai, N.; Szoboszlai, N.; Bencs, L. Quantitation of Toxic Elements in Various Water Samples by Multi-element Graphite Furnace Atomic Absorption Spectrometry. *At. Spectrosc*. **2014**, *35*, 33-42.

(86) Lord, G.; Kim, N.; Ward, N. I. Arsenic speciation of geothermal waters in New Zealand. *J. Environ. Monit*. **2012**, *14*, 3192-3201. https://doi.org/10.1039/C2EM30486D

(87) Smichowski, P.; Valiente, L.; Ledesma, A. Simple Method for the Selective Determination of As(III) and As(V) by ETAAS After Separation With Anion Exchange Mini-column. *At. Spectrosc*. **2002**, *23*, 92-97.

(88) Yalçin, S.; Le, X. C. Speciation of arsenic using solid phase extraction cartridges. *J Environ Monit*. **2001**, *3*, 81-85. https://doi.org/10.1039/B007598L

(89) Sigrist, M.; Albertengo, A.; Beldoménico, H.; Tudino, M. Determination of As(III) and total inorganic As in water samples using an on-line solid phase extraction and flow injection hydride generation atomic absorption spectrometry. *J. Hazard. Mater*. **2011**, *188*, 311–318. https://doi.org/10.1016/j.jhazmat.2011.01.126

(90) Baranik, A.; Gagor, A.; Queralt, I.; Marguí, E.; Sitko, R.; Zawisza, B. Determination and speciation of ultratrace arsenic and chromium species using aluminium oxide supported on graphene oxide. *Talanta* **2018**, *185*, 264-275. https://doi.org/10.1016/j.talanta.2018.03.090

(91) Agrafioti, E.; Kalderis, D.; Diamadopoulos, E. Arsenic and chromium removal from water using biochars derived from rice husk, organic solid wastes and sewage sludge. *J. Environ. Manag*. **2014**, *133*, 309-314. https://doi.org/10.1016/j.jenvman.2013.12.007

(92) Mohan, D.; Pittman Jr., C. U. Arsenic removal from water/wastewater using adsorbents – A critical review. *J. Hazard. Mater*. **2007**, *142*, 1-53. https://doi.org/10.1016/j.jhazmat.2007.01.006

(93) Zhang, M.; Gao, B.; Varnoosfaderani, S.; Hebard, A.; Yao, Y.; Inyang, M. Preparation and characterization of a novel magnetic biochar for arsenic removal. *Bioresour. Technol*. **2013**, *130*, 457-462. https://doi.org/10.1016/j.biortech.2012.11.132

(94) Dombay, G. "Az arzéneltávolítás korszerű technológiai lehetőségei." *Vízmű Panoráma* **2002**, *10*, 38-40. (in Hungarian)

(95) Litter, M. I.; Morgada, M. E.; Bundschuh, J. Possible treatments for arsenic removal in Latin American waters for human consumption. *Environ. Pollut*. **2010**, *158*, 1105-1118. https://doi.org/10.1016/j.envpol.2010.01.028

(96) D'Amato, M.; Forte, G., Caroli, S. Identification and Quantification of Major Species of Arsenic in Rice. *J. AOAC Int*. **2004**, *87*, 238–243. https://doi.org/10.1093/jaoac/87.1.238

(97) Bohari, Y.; Lobos, G.; Pinochet, H., Pannier, F.; Astruc, A.; Potin-Gautier, M. Speciation of arsenic in plants by HPLC-HG-AFS: extraction optimisation on CRM materials and application to cultivated samples. *J. Environ. Monitor*. **2002**, *4*, 596–602. https://doi.org/10.1039/B203988P

(98) Rofkar, J. R.; Dwyer, D. F. Effects of Light Regime, Temperature, and Plant Age on Uptake of Arsenic by Spartina Pectinata and *Carex Stricta*. *Int. J. Phytoremediat*. **2011**, *13*, 528-537. https://doi.org/10.1080/15226514.2010.495151

(99) Rahman, M. A.; Kadohashi, K., Maki, T.; Hasegawa, H. Transport of DMAA and MMAA into rice (*Oryza sativa* L.) roots. *Environ. Exp. Bot*. **2011**, *72*, 41-46. https://doi.org/10.1016/j.envexpbot.2010.02.004

(100) Bravin, M. N.; Travassac, F.; Le Floch, M.; Hinsinger, P.; Garnier, J. M. Oxygen input controls the spatial and temporal dynamics of arsenic at the surface of a flooded paddy soil and in the rhizosphere of lowland rice (*Oryza sativa* L.): a microcosm study. *Plant Soil* **2008**, *312*, 207-218. https://doi.org/10.1007/s11104-007-9532-x

(101) Chen, T.; Yan, X.; Liao, X., Xiao, X.; Huang, Z.; Xie, H., Zhai, L. Subcellular distribution and compartmentalization of arsenic in *Pteris vittata* L. *Chin. Sci. Bull*. **2005**, *50*, 2843-2849. https://doi.org/10.1360/982005-943

(102) Dembitsky, V. M.; Rezanka, T. Natural occurrence of arseno compounds in plants, lichens, fungi, algal species, and microorganisms. *Plant. Sci*. **2003**, *165*, 1177–1192. https://doi.org/10.1016/j.plantsci.2003.08.007

(103) Meharg, A. A.; Hartley-Whitaker, J. Arsenic uptake and metabolism in arsenic resistant and nonresistant plant species. *New Phytol*. **2002**, *154*, 29–43. https://doi.org/10.1046/j.1469-8137.2002.00363.x

(104) Zhang, W.; Cai, Y.; Tu, C.; Ma, L. Q. Arsenic speciation and distribution in an arsenic hyperaccumulating plant. *Sci. Total Environ*. **2002**, *300*, 167–177. https://doi.org/10.1016/s0048-9697(02)00165-1

(105) Chen, R. X.; Smith, B. W.; Winefordner, J. D.; Tu, M. S.; Kertulis, G.; Ma, L. Q. Arsenic speciation in Chinese brake fern by ion-pair high-performance liquid chromatography–inductively coupled plasma mass spectroscopy. *Anal. Chim. Acta* **2004**, *504*, 199–207. https://doi.org/10.1016/j.aca.2003.10.042

(106) Kertulis, G. M.; Ma, L. Q.; MacDonald, G. E.; Chen, R.; Winefordner, J. D.; Cai, Y. Arsenic speciation and transport in *Pteris vittata* L. and the effects on phosphorus in the xylem sap. *Environ. Exp. Bot*. **2005**, *54*, 239-247. https://doi.org/10.1016/j.envexpbot.2004.09.001

(107) Cullen, W. R.; Reimer, K. J. Arsenic speciation in the environment. *Chem. Rev*. **1989**, *89*, 713–764. https://doi.org/10.1021/cr00094a002

(108) Unterbrunner, R.; Puschenreiter, M.; Sommer, P.; Wieshammer, G.; Tlustoš, P.; Zupan, M.; Wenzel, W. W. Heavy metal accumulation in trees growing on contaminated sites in Central Europe. *Environ. Pollut*. **2007**, *148*, 107–114. https://doi.org/10.1016/j.envpol.2006.10.035

(109) Mihucz, V. G.; Tatár, E.; Varga, A.; Záray, G.; Cseh, E. Application of total-reflection X-ray fluorescence spectrometry and high-performance liquid chromatography for the chemical characterization of xylem saps of nickel contaminated cucumber plants. *Spectrochim. Acta Part B* **2001**, *56*, 2235–2246. https://doi.org/10.1016/S0584-8547(01)00345-7

(110) Gasparics, T.; Mihucz, V. G.; Tatár, E.; Záray, G. Hyphenated technique for investigation of nickel complexation by citric acid in xylem sap of cucumber plants. *Microchem. J*. **2002**, *73*, 89–98. https://doi.org/10.1016/S0026-265X(02)00055-3

(111) Tatár, E.; Mihucz, V. G.; Varga, A.; Záray, G.; Cseh, E. Effect of lead, nickel and vanadium contamination on organic acid transport in xylem sap of cucumber. *J. Inorg. Biochem*. **1999**, *75*, 219–223. https://doi.org/10.1016/S0162-0134(99)00091-4

(112) Mihucz, V. G.; Tatár, E.; Kmethy, B.; Záray, G.; Cseh, E. Investigation of the transported heavy metal ions in xylem sap of cucumber plants by size exclusion chromatography and atomic absorption spectrometry. *J. Inorg. Biochem*. **2000**, *81*, 81-87. https://doi.org/10.1016/S0162-0134(00)00091-X

(113) Tatár, E.; Mihucz, V. G.; Kmethy, B.; Záray, G.; Fodor, F. Determination of organic acids and their role in nickel transport within cucumber plants. *Microchem. J*. **2000**, *67*, 73–81. https://doi.org/10.1016/S0026-265X(00)00101-6

(114) Pickering, I. J.; Prince, R. C.; George, M. J.; Smith, R. D.; George, G. N.; Salt, D. E. Reduction and Coordination of Arsenic in Indian Mustard. *Plant Physiol*. **2000**, *122*, 1171–1177. https://doi.org/10.1104/pp.122.4.1171

(115) Salgueiro, M. J.; Zubillaga, M. B.; Lysionek, A. E.; Caro, R. A.; Weill, R.; Boccio, J. R. The role of zinc in the growth and development of children. *Nutrition* **2002**, *18*, 510–519. https://doi.org/10.1016/S0899-9007(01)00812-7

(116) Untoro, J.; Karyadi, E.; Wibowo, L.; Erhardt, J.; Gross, R. Multiple Micronutrient Supplements Improve Micronutrient Status and Anemia but not Growth and Morbidity of Indonesian Infants: A Randomized, Double-Blind, Placebo-Controlled Trial. *The Journal of Nutrition* **2005**, *135*, 639S–645S. https://doi.org/10.1093/jn/135.3.639S

(117) Chan, S. S. L.; Ferguson, E. L.; Bailey, K.; Fahmida, U.; Harper, T. B.; Gibson, R. S. The concentrations of iron, calcium, zinc and phytate in cereals and legumes habitually consumed by infants living in East Lombok, Indonesia. *J. Food Compos. Anal.* **2007**, *20*, 609–617. https://doi.org/10.1016/j.jfca.2007.03.003

(118) Liang, J.; Han, B. Z.; Nout, M. J. R.; Hamer, R. J. Effects of soaking, germination and fermentation on phytic acid, total and in vitro soluble zinc in brown rice. *Food Chem*. **2008**, *110*, 821–828. https://doi.org/10.1016/j.foodchem.2008.02.064

(119) Zabłudowska, E.; Kowalska, J.; Jedynak, Ł.; Wojas, S.; Skłodowska, A.; Antosiewicz, D. M. Search for a plant for phytoremediation – What can we learn from field and hydroponic studies? *Chemosphere* **2009**, *77*, 301-307. https://doi.org/10.1016/j.chemosphere.2009.07.064

(120) Duan, G. L.; Zhou, Y.; Tong, Y. P.; Mukhopadhyay, R.; Rosen, B. P.; Zhu, Y. G. A CDC25 homologue from rice functions as an arsenate reductase. *New Phytol*. **2007**, *174*, 311–321. https://doi.org/10.1111/j.1469-8137.2007.02009.x

(121) Su, Y. H.; McGrath, S. P.; Zhu, Y. G.; Zhao, F. J. Highly efficient xylem transport of arsenite in the arsenic hyperaccumulator *Pteris vittata*. *New Phytol*. **2008**, *180*, 434–441. https://doi.org/10.1111/j.1469-8137.2008.02584.x

(122) Ellis, D. R.; Gumaelius, L.; Indriolo, E.; Pickering, I. J.; Banks, J. A.; Salt, D. E. A Novel Arsenate Reductase from the Arsenic Hyperaccumulating Fern *Pteris vittata*. *Plant Physiol*. **2006**, *141*, 1544–1554. https://doi.org/10.1104/pp.106.084079

(123) Xu, X. Y.; McGrath, S. P.; Zhao, F. J. Rapid reduction of arsenate in the medium mediated by plant roots. *New Phytol*. **2007**, *176*, 590–599. https://doi.org/10.1111/j.1469-8137.2007.02195.x

(124) Gonzaga, M. I. S.; Santos, J. A. G.; Ma, L. Q. Arsenic phytoextraction and hyperaccumulation by fern species. *Sci. Agric*. **2006**, *63*, 90–101. https://doi.org/10.1590/S0103-90162006000100015

(125) Ma, J. F.; Yamaji, N.; Mitani, N.; Xu, X. Y.; Su, Y. H.; McGrath, S. P.; Zhao, F. J. Transporters of arsenite in rice and their role in arsenic accumulation in rice grain. *Proc. Natl. Acad. Sci. U.S.A.* **2008**, *105*, 9931–9935. https://doi.org/10.1073/pnas.0802361105

(126) Su, Y. H.; McGrath, S. P.; Zhao, F. J. Rice is more efficient in arsenite uptake and translocation than wheat and barley. *Plant Soil* **2010**, *328*, 27–34. https://doi.org/10.1007/s11104-009-0074-2

(127) Zhao, F. J.; McGrath, S. P.; Meharg, A. A. Arsenic as a Food Chain Contaminant: Mechanisms of Plant Uptake and Metabolism and Mitigation Strategies. *Annu. Rev. Plant Biol*. **2010**, *61*, 535-559. https://doi.org/10.1146/annurev-arplant-042809-112152

(128) Quaghebeur, M.; Rengel, Z. The Distribution of Arsenate and Arsenite in Shoots and Roots of *Holcus lanatus* is Influenced by Arsenic Tolerance and Arsenate and Phosphate Supply. *Plant Physiol*. **2003**, *132*, 1600–1609. https://doi.org/10.1104/pp.103.021741

(129) Raab, A.; Williams, P. N.; Meharg, A.; Feldmann, J. Uptake and translocation of inorganic and methylated arsenic species by plants. *Environ. Chem*. **2007**, *4*, 197–203. https://doi.org/10.1071/EN06079

(130) LaFleur, J. P.; Salin, E. D. Speciation of Chromium by High-Performance Thin-Layer Chromatography with Direct Determination by Laser Ablation Inductively Coupled Plasma Mass Spectrometry. *Anal. Chem*. **2008**, *80*, 6821–6823. https://doi.org/10.1021/ac8010582

(131) Meharg, A. A.; Williams, P. N.; Adomako, E. E.; Lawgali, Y. Y.; Deacon, C.; Villada, A.; Cambell, R. C. J.; Sun, G.; Zhu, Y. G.; Feldmann, J.; et al. Geographical Variation in Total and Inorganic Arsenic Content of Polished (White) Rice. *Environ. Sci. Technol*. **2009**, *43*, 1612–1617. https://doi.org/10.1021/es802612a

(132) Yost, L. J.; Tao, S. H.; Egan, S. K.; Barraj, L. M.; Smith, K. M.; Tsuji, J. S.; Lowney, Y. W.; Schoof, R. A.; Rachman, N. J. Estimation of Dietary Intake of Inorganic Arsenic in U.S. Children. *Hum. Ecol. Risk Assess*. **2004**, *10*, 473–483. https://doi.org/10.1080/10807030490452151

(133) Ohno, K.; Yanase, T.; Matsuo, Y.; Kimura, T.; Rahman, M. H.; Magara, Y.; Matsui, Y. Arsenic intake via water and food by a population living in an arsenic-affected area of Bangladesh. *Sci. Total Environ*. **2007**, *381*, 68–76. https://doi.org/10.1016/j.scitotenv.2007.03.019

(134) Rahman, M. A.; Hasegawa, H. High levels of inorganic arsenic in rice in areas where arsenic-contaminated water is used for irrigation and cooking. *Sci. Total Environ*. **2011**, *409*, 4645–4655. https://doi.org/10.1016/j.scitotenv.2011.07.068

(135) Díaz, O. P.; Leyton, I.; Muñoz, O.; Nuñez, N.; Devesa, V.; Suner, M. A.; Velez, D.; Montoro, R. Contribution of Water, Bread, and Vegetables (Raw and Cooked) to Dietary Intake of Inorganic Arsenic in a Rural Village of Northern Chile. *J. Agric. Food Chem*. **2004**, *52*, 1773–1779. https://doi.org/10.1021/jf035168t

(136) Monroy-Torres, R.; Macias, A. E.; Gallaga-Solorzano, J. C.; Santiago-Garcia, E. J.; Hernandez, I. Arsenic in Mexican Children Exposed to Contaminated Well Water. *Ecol. Food Nutr*. **2009**, *48*, 59–75. https://doi.org/10.1080/03670240802575519

(137) Henden, E.; Cataloglu, R.; Aksuner, N. Determination of arsenic leaching from glazed and non-glazed Turkish traditional earthenware. *Sci. Total Environ*. **2011**, *409*, 2993–2996. https://doi.org/10.1016/j.scitotenv.2011.04.027

(138) Valentine, J. L.; Cebrian, M. E.; Garcia-Vargas, G. G.; Faraji, B.; Kuo, J.; Gibb, H. J.; Lachenbruch, P. A. Daily Selenium Intake Estimates for Residents of Arsenic-Endemic Areas. *Environ. Res*. **1994**, *64*, 1–9. https://doi.org/10.1006/enrs.1994.1001

(139) Bundschuh, J.; Nath, B.; Bhattacharya, P.; Liu, C. W.; Armienta, M. A.; Moreno López, M. V.; López, D. L.; Jean, J. S.; Cornejo, L.; Lauer Macedo, L. F.; et al. Arsenic in the human food chain: the Latin American perspective. *Sci. Tot. Environ*. **2012**, *429*, 92–106. https://doi.org/10.1016/j.scitotenv.2011.09.069

(140) Arnich, N.; Sirot, V.; Riviere, G.; Jean, J.; Noel, L.; Guerin, T.; Leblan, J. C. Dietary exposure to trace elements and health risk assessment in the 2nd French Total Diet Study. *Food Chem. Toxicol*. **2012**, *50*, 2432–2449. https://doi.org/10.1016/j.fct.2012.04.016

(141) Martorell, I.; Perelló, G.; Martí-Cid, R.; Llobet, J. M.; Castell, V.; Domingo, J. L. Human Exposure to Arsenic, Cadmium, Mercury, and Lead from Foods in Catalonia, Spain: Temporal Trend. *Biol. Trace Elem. Res*. **2011**, *142*, 309–322. https://doi.org/10.1007/s12011-010-8787-x

(142) Wong, W. W. K.; Chung, S. W. C.; Chan, B. T. P.; Ho, Y. Y.; Xiao, Y. Dietary exposure to inorganic arsenic of the Hong Kong population: Results of the first Hong Kong Total Diet Study. *Food Chem. Toxicol*. **2013**, *51*, 379–385. https://doi.org/10.1016/j.fct.2012.10.010

(143) Gallagher, P. A.; Wei, X. Y.; Shoemaker, J. A.; Brockhoff, C. A.; Creed, J. T. Detection of arsenosugars from kelp extracts via IC-electrospray ionization-MS-MS and IC membrane hydride generation ICP-MS. *J. Anal. At. Spectrom*. **1999**, *14*, 1829–1834. https://doi.org/10.1039/A906249A

(144) Tao, S. S. H.; Bolger, P. M. Dietary arsenic intakes in the United States: FDA Total Diet Study, September 1991 – December 1996. *Food Addit. Contam*. **1999**, *16*, 465–472. https://doi.org/10.1080/026520399283759

(145) Yost, L. J.; Schoof, R. A.; Aucoin, R. Intake of Inorganic Arsenic in the North American Diet. *Hum. Ecol. Risk Assess*. **1998**, *4*, 137–152. https://doi.org/10.1080/10807039891284244

(146) Schoof, R. A.; Yost, L. J.; Eickhoff, J.; Crecelius, E. A.; Cragin, D. W.; Meacher, D. M.; Menzel, D. B. A Market Basket Survey of Inorganic Arsenic in Food. *Food Chem. Toxicol*. **1999**, *37*, 839–846. https://doi.org/10.1016/S0278-6915(99)00073-3

(147) Davis, M. A.; Mackenzie, T. A.; Cottingham, K. L.; Gilbert-Diamond, D.; Punshon, T.; Karagas, M. R. Rice Consumption and Urinary Arsenic Concentrations in U.S. Children. *Environ. Health Perspect*. **2012**, *12*, 1418–1424. https://doi.org/10.1289/ehp.1205014

(148) Meacher, D. M.; Menzel, D. B.; Dillencourt, M. D.; Bic, L. F.; Schoof, R. A.; Yost, L. J.; Eickhoff, J. C.; Farr, C. H. Estimation of Multimedia Inorganic Arsenic Intake in the U.S. Population. *Hum. Ecol. Risk Assess*. **2002**, *8*, 1697–1721. https://doi.org/10.1080/20028091057565

(149) Meliker, J. R.; Franzblau, A.; Slotnick, M. J.; Nriagu, J. O. Major contributors to inorganic arsenic intake in southeastern Michigan. *Int. J. Hyg. Environ. Health* **2006**, *209*, 399–411. https://doi.org/10.1016/j.ijheh.2006.03.006

(150) Liang, J; Li, Z.; Tsuji, K.; Nakano, K.; Nout, M. J. R.; Hamer, R. J. Milling characteristics and distribution of phytic acid and zinc in long-, medium- and short-grain rice. *J. Cereal Sci*. **2008**, *48*, 83–91. https://doi.org/10.1016/j.jcs.2007.08.003

(151) Grant, C. A.; Clarke, J. M.; Duguid, S.; Chaney, R. L. Selection and breeding of plant cultivars to minimize cadmium accumulation. *Sci. Total Environ*. **2008**, *390*, 301–310. https://doi.org/10.1016/j.scitotenv.2007.10.038

(152) Liu, H.; Probst, A.; Liao, B. Metal contamination of soils and crops affected by the Chenzhou lead/zinc mine spill (Hunan, China). *Sci. Total Environ*. **2005**, *339*, 153–166. https://doi.org/10.1016/j.scitotenv.2004.07.030

(153) Liu, H. J.; Zhang, J. L.; Zhang, F. S. Role of iron plaque in Cd uptake by and translocation within rice (*Oryza sativa* L.) seedlings grown in solution culture. *Environ. Exp. Bot*. **2007**, *59*, 314–320. https://doi.org/10.1016/j.envexpbot.2006.04.001

(154) Rascio, N.; Vecchia, F. D.; La Rocca, N.; Barbato, R.; Pagliano, C.; Raviolo, M.; Gonnelli, C.; Gabbrielli, R. *Environ. Exp. Bot*., **2008**, *62*, 267–278. https://doi.org/10.1016/j.envexpbot.2007.09.002

(155) Heitkemper, D. T.; Vela, N. P.; Stewart, K. R.; Westphal, C. S. Determination of total and speciated arsenic in rice by ion chromatography and inductively coupled plasma mass spectrometry. *J. Anal. At. Spectrom*. **2001**, *16*, 299–306. https://doi.org/10.1039/B007241I

(156) Pizarro, I.; Gómez, M.; Palacios, M. A.; Cámara, C. Evaluation of stability of arsenic species in rice. *Anal. Bioanal. Chem*. **2003**, *376*, 102–109. https://doi.org/10.1007/s00216-003-1870-9

(157) Huang, J.-H.; Ilgen, G.; Fecher, P. Quantitative chemical extraction for arsenic speciation in rice grains. *J. Anal. At. Spectrom*. **2010**, *25*, 800-802. https://doi.org/10.1039/C002306J

(158) McKiernan, J. W.; Creed, J. T.; Brockhoff, C. A.; Caruso, J. A.; Lorenzana, R. M. A comparison of automated and traditional methods for the extraction of arsenicals from fish. *J. Anal. At. Spectrom.* **1999**, *14*, 607–613. https://doi.org/10.1039/A808824A

(159) Caruso, J. A.; Heitkemper, D. T.; B'Hymer, C. An evaluation of extraction techniques for arsenic species from freeze-dried apple samples. *Analyst* **2001**, *126*, 136–140. https://doi.org/10.1039/B009825F

(160) Pizarro, I.; Gómez, M.; Cámara, C.; Palacios, M. A. Arsenic speciation in environmental and biological samples: Extraction and stability studies. *Anal. Chim. Acta* **2003**, *495*, 85–98. https://doi.org/10.1016/j.aca.2003.08.009

(161) Sanz, E.; Muñoz-Olivas, R.; Cámara, C. A rapid and novel alternative to conventional sample treatment for arsenic speciation in rice using enzymatic ultrasonic probe. *Anal. Chim. Acta* **2005**, *535*, 227–235. https://doi.org/10.1016/j.aca.2004.12.021

(162) Sanz, E.; Muñoz-Olivas, R.; Cámara, C. Evaluation of a focused sonication probe for arsenic speciation in environmental and biological samples. *J. Chromatogr. A* **2005**, *1097*, 1–8. https://doi.org/10.1016/j.chroma.2005.08.012

(163) Abedin, M. J.; Cresser, M. S.; Meharg, A. A.; Feldmann, J.; Cotter-Howells, J. Arsenic Accumulation and Metabolism in Rice (*Oryza sativa* L.). *Environ. Sci. Technol*. **2002**, *36*, 962–968. https://doi.org/10.1021/es0101678

(164) European Commission Regulation (EU) 2015/1006 of 25 June 2015 amending Regulation (EC) No 1881/2006 as regards maximum levels of inorganic arsenic in foodstuffs. https://eur-lex.europa.eu/legal-content/EN/TXT/HTML/?uri=CELEX:32015R1006&from=EN [Accessed 28 November 2021].

(165) Williams, P. N.; Raab, A.; Feldmann, J.; Meharg, A. A. Market Basket Survey Shows Elevated Levels of As in South Central U.S. Processed Rice Compared to California: Consequences for Human Dietary Exposure. *Environ. Sci. Technol*. **2007**, *41*, 2178–2183. https://doi.org/10.1021/es061489k

(166) Zavala, Y. J.; Duxbury, J. M. Arsenic in Rice: I. Estimating Normal Levels of Total Arsenic in Rice Grain. *Environ. Sci. Technol*. **2008**, *42*, 3856–3860. https://doi.org/10.1021/es702747y

(167) Ackerman, A. H.; Creed, P. A.; Parks, A. N.; Fricke, M. W.; Schwegel, C. A.; Creed, J. T.; Heitkemper, D. T.; Vela, N. P. Comparison of a Chemical and Enzymatic Extraction of Arsenic from Rice and an Assessment of the Arsenic Absorption from Contaminated Water by Cooked Rice. *Environ. Sci. Technol*. **2005**, *39*, 5241–5246. https://doi.org/10.1021/es048150n

(168) Bae, M.; Watanabe, C.; Inaoka, T.; Sekiyama, M.; Sudo, N.; Bokul, M. H.; Ohtsuka, R. Arsenic in cooked rice in Bangladesh. *Lancet* **2002**, *360*, 1839–1840. https://doi.org/10.1016/S0140-6736(02)11738-7

(169) Kisomi, A. S.; Alizadeh, T.; Shakeri, A.; Nouri, A.; Farsadrooh, M.; Najafi, S.; Pashaki, A. Application of μ-TLC for speciation of inorganic arsenic by laser ablation inductively coupled plasma mass spectrometry. *Microchem. J*. **2020**, 105443. https://doi.org/10.1016/j.microc.2020.105443

(170) The Food and Agriculture Organization (FAO) Codex Alimentarius Commission - Geneva 14-18 July 2014. http://www.fao.org/news/story/en/item/238558/icode/SCOOP [Accessed 28 November 2021].

(171) Sharafi, K.; Yunesian, M.; Mahvi, A. H.; Pirsaheb, M.; Nazmara, S.; Nodehi, R. N. Advantages and disadvantages of different pre-cooking and cooking methods in removal of essential and toxic metals from various rice types- human health risk assessment in Tehran households, Iran. *Ecotoxicol. Environ. Saf.* **2019**, *175*, 128-137. https://doi.org/10.1016/j.ecoenv.2019.03.056

(172) Pal, A.; Chowdhury, U. K.; Mondal, D.; Das, B.; Nayak, B.; Ghosh, A.; Maity, S.; Chakraborti, D. Arsenic Burden from Cooked Rice in the Populations of Arsenic Affected and Nonaffected Areas and Kolkata City in West-Bengal, India. *Environ. Sci. Technol*. **2009**, *43*, 3349–3355. https://doi.org/10.1021/es803414j

(173) Isaac, C. P.; Sivakumar, A.; Kumar, C. R. Lead Levels in Breast Milk, Blood Plasma and Intelligence Quotient: A Health Hazard for Women and Infants. *Bull. Environ. Contam. Toxicol*. **2012**, *88*, 145-149. https://doi.org/10.1007/s00128-011-0475-9

(174) Oskarsson, A.; Hallén, I. P.; Sundberg, J.; Grawé, K. P. Risk assessment in relation to neonatal metal exposure. *Analyst* **1998**, 123, 19-23. https://doi.org/10.1039/A705136K

(175) Miklavčič, A.; Casetta, A.; Tratnik, J. S.; Mazej, D.; Krsnik, M.; Mariuz, M.; Sofianou, K.; Spiric, Z.; Barbone, F.; Horvat, M. Mercury, arsenic and selenium exposure levels in relation to fish consumption in the Mediterranean area. *Environ. Res*. **2013**, *120*, 7-17. https://doi.org/10.1016/j.envres.2012.08.010

(176) Concha, G.; Vogler, G.; Lezcano, D.; Nermell, B.; Vahter, M. Exposure to Inorganic Arsenic Metabolites during Early Human Development. *Toxicol. Sci*. **1998**, *44*, 185-190. https://doi.org/10.1006/toxs.1998.2486

(177) Joint FAO/WHO Expert Committee on Food Additives. Seventy-second meeting, Rome, 16–25 February 2010. http://www.who.int/foodsafety/chem/summary72_rev.pdf [Accessed 28 November 2021].

(178) Ballard, O.; Morrow, A. L. Human milk composition: nutrients and bioactive factors. *Pediatr. Clin. North Am*. **2013**, *60*, 49-74. https://doi.org/10.1016/j.pcl.2012.10.002

(179) Leotsinidis, M.; Alexopoulos, A.; Kostopoulou-Farri, E. Toxic and essential trace elements in human milk from Greek lactating women: Association with dietary habits and other factors. *Chemosphere* **2005**, *61*, 238-247. https://doi.org/10.1016/j.chemosphere.2005.01.084

(180) Letinić, J. G.; Sarić, M. M.; Piasek, M.; Jurasović, J.; Varnaic, V. M.; Grgec, A. S.; Orct, T. Use of human milk in the assessment of toxic metal exposure and essential element status in breastfeeding women and their infants in coastal Croatia. *J. Trace Elem. Med. Biol*. **2016**, *38*, 117-125. https://doi.org/10.1016/j.jtemb.2016.08.002

(181) Örün, E.; Yalçın, S. S.; Aykut, O.; Orhan, G.; Morgil, G. K.; Yurdakök, K.; Uzun, R. Breast milk lead and cadmium levels from suburban areas of Ankara. *Sci. Total Environ*. **2011**, *409*, 2467-2472. https://doi.org/10.1016/j.scitotenv.2011.02.035

(182) Coni, E.; Bocca, B.; Galoppi, B.; Alimonti, A.; Caroli, S. Identification of chemical species of some trace and minor elements in mature breast milk. *Microchem. J*. **2000**, *67*, 187-194. https://doi.org/10.1016/S0026-265X(00)00116-8

(183) Martino, F. A. R.; Sánchez, M. L. F.; Sanz-Medel, A. The potential of double focusing-ICP-MS for studying elemental distribution patterns in whole milk, skimmed milk and milk whey of different milks. *Anal. Chim. Acta* **2001**, *442*, 191-200. https://doi.org/10.1016/S0003-2670(01)01170-9

(184) Ecsedi-Angyal, M.; Tatár, E.; Óvári, M.; Kurin-Csörgei, K.; Záray, G.; Mihucz, V. G. Determination of low-level arsenic, lead, cadmium and mercury concentration in breast milk of Hungarian women. *Int. J. Environ. Anal. Chem*. **2020**, *100*, 549-566. https://doi.org/10.1080/03067319.2019.1637429

(185) Parr, R. M.; DeMaeyer, E. M.; Iyengar, V. G.; Byrne, A. R.; Kirkbright, G. F.; Schöch, G.; Ninistö, L.; Pineda, O.; Vis, H. L.; Hofvander, I.; Omololu, A. Minor and trace elements in human milk from Guatemala, Hungary, Nigeria, Philippines, Sweden, and Zaire. *Biol. Trace Elem. Res*. **1991**, *29*, 51-75. https://doi.org/10.1007/BF03032674

(186) Lovreglio, P.; D'Errico, M. N.; Gilberti, M. E.; Drago, I.; Basso, A.; Apostoli, P.; Soleo, L. The influence of diet on intra and inter-individual variability of urinary excretion of arsenic species in Italian healthy individuals. *Chemosphere* **2012**, *86*, 898-905. https://doi.org/10.1016/j.chemosphere.2011.10.050

(187) Chao, H. H.; Guo, C. H.; Huang, C. B.; Chen, P. C.; Li, H. C.; Hsiung, D. Y.; Chou, Y. K. Arsenic, Cadmium, Lead, and Aluminium Concentrations in Human Milk at Early Stages of Lactation. *Pediatr. Neonatol*. **2014**, *55*, 127-134. https://doi.org/10.1016/j.pedneo.2013.08.005

(188) Khan, S.; Ismail, A.; Gong, Y. Y.; Akhtar, S.; Hussain, M. Concentration of Aflatoxin M1 and selected heavy metals in mother milk samples from Pakistan. *Food Control* **2018**, 91, 344-348. https://doi.org/10.1016/j.foodcont.2018.04.015

(189) Samiee, F.; Vahidinia, A.; Javad, M. T.; Leili, M. Exposure to heavy metals released to the environment through breastfeeding: A probabilistic risk estimation. *Sci. Total Environ*. **2019**, *650*, 3075-3083. https://doi.org/10.1016/j.scitotenv.2018.10.059

(190) Kippler, M.; Hossain, M. B.; Lindh, C.; Moore, S. E.; Kabir, I.; Vahter, M.; Broberg, K. Early life low-level cadmium exposure is positively associated with increased oxidative stress. *Environ. Res*. **2012**, *112*, 164-170. https://doi.org/10.1016/j.envres.2011.11.012

(191) Sakamoto, M.; Chan, H. M.; Domingo, J. L.; Kubota, M.; Murata, K. Changes in body burden of mercury, lead, arsenic, cadmium and selenium in infants during early lactation in comparison with placental transfer. *Ecotoxicol. Environ. Saf*. **2012**, *84*, 179-184. https://doi.org/10.1016/j.ecoenv.2012.07.014

(192) Bassil, M.; Daou, F.; Hassan, H.; Yamani, O.; Kharma, J. A.; Attieh, Z.; Elaridi, J. Lead, cadmium and arsenic in human milk and their socio-demographic and lifestyle determinants in Lebanon. *Chemosphere* **2018**, *191*, 911-921. https://doi.org/10.1016/j.chemosphere.2017.10.111

(193) Almeida, A. A.; Lopes, C. M. P. V.; Silva, A. M. S.; Barrado, E. Trace elements in human milk: Correlation with blood levels, inter-element correlations and changes in concentration during the first month of lactation. *J. Trace Elem. Med. Biol*. **2008**, *22*, 196-205. https://doi.org/10.1016/j.jtemb.2008.03.007

(194) Rebelo, F. M.; Caldas, E. D. Arsenic, lead, mercury and cadmium: Toxicity, levels in breast milk and the risks for breastfed infants. *Environ. Res*. **2016**, *151*, 671-688. https://doi.org/10.1016/j.envres.2016.08.027

(195) Rebelo, F. M.; Cunha, L. R. D.; Andrade, P. D.; Costa Jr., W. A. D.; Bastos, W. R.; Caldas, E. D. Mercury in breast milk from women in the Federal District, Brazil and dietary risk assessment for breastfed infants. *J. Trace Elem. Med. Biol*. **2017**, *44*, 99-103. https://doi.org/10.1016/j.jtemb.2017.06.009

(196) European Food Safety Authority (EFSA). Opinion of the Scientific Committee on a request from EFSA related to A Harmonised Approach for Risk Assessment of Substances Which are both Genotoxic and Carcinogenic. Published: 27 October 2005. Adopted: 18 October 2005. https://doi.org/10.2903/j.efsa.2005.282

(197) Mieczan, A. W. Cadmium, Lead, Copper and Zinc in Breast Milk in Poland. *Biol. Trace Elem. Res*. **2014**, *157*, 36-44. https://doi.org/10.1007/s12011-013-9870-x

(198) Saper, R. B.; Phillips, R. S.; Sehgal, A.; Khouri, N.; Davis, R. B.; Paquin, J.; Thuppil, V.; Kales, S. N. Lead, Mercury, and Arsenic in US- and Indian-Manufactured Ayurvedic Medicines Sold via the Internet. *JAMA* **2008**, *300*, 915–923. https://doi.org/10.1001/jama.300.8.915

(199) Martena, M. J.; Van Der Wielen, J. C. A.; Rietjens, I. M. C. M.; Klerx, W. N. M.; de Groot, H. N. D.; Konings, E. J. M. Monitoring of mercury, arsenic, and lead in traditional Asian herbal preparations on the Dutch market and estimation of associated risks. *Food Addit. Contam. Part A* **2010**, *27*, 190-205. https://doi.org/10.1080/02652030903207235

(200) Kumar, A.; Nair, A. G. C.; Reddy, A. V. R.; Garg, A. N. Availability of essential elements in bhasmas: Analysis of Ayurvedic metallic preparations by INAA. *J. Radioanal. Nucl. Chem*. **2006**, *270*, 173–180. https://doi.org/10.1007/s10967-006-0326-z

(201) Sharma, S.; Dhingra, P.; Pandey, R. S. Trace determination of Pb, Cu, Cd and Zn in Ayurvedic, drug, "Mahayograj Guggulu" via polarographic technique. *J. Indian Chem. Soc*. **2008**, *85*, 962–965.

(202) Mahawatte, P.; Dissanayaka, K. R.; Hewamanna, R. Elemental concentrations of some Ayurvedic drugs using energy dispersive XRF. *J. Radioanal. Nucl. Chem*. **2006**, *270*, 657–660. https://doi.org/10.1007/s10967-006-0444-7

(203) Khandpur, S.; Malhotra, A. K.; Bhatia, V.; Gupta, S.; Sharma, V. K.; Mishra, R.; Arora, N. K. Chronic arsenic toxicity from Ayurvedic medicines. *Int. J. Dermatol.* **2008**, *47*, 618–621. https://doi.org/10.1111/j.1365-4632.2008.03475.x

(204) Saper, R. B.; Kales, S. N.; Paquin, J.; Burns, M. J.; Eisenberg, D. M.; Davis, R. B.; Phillips, R. S. Heavy Metal Content of Ayurvedic Herbal Medicine Products. *JAMA* **2004**, *292*, 2868–2873. https://doi.org/10.1001/jama.292.23.2868

(205) Giacomino, A.; Abollino, O.; Malandrino, M.; Karthik, M.; Murugesan, V. Determination and assessment of the contents of essential and potentially toxic elements in Ayurvedic medicine formulations by inductively coupled plasma-optical emission spectrometry. *Microchem. J*. **2011**, *99*, 2–6. https://doi.org/10.1016/j.microc.2011.01.002

(206) Oomen, A. G.; Hack, A.; Minekus, M.; Zeijdner, E.; Cornelis, C.; Schoeters, G.; Verstraete, W.; Van de Wiele, T.; Wragg, J.; Rompelberg, C. J. M. et al. Comparison of Five In Vitro Digestion Models To Study the Bioaccessibility of Soil Contaminants. *Environ. Sci. Technol*. **2002**, *36*, 3326–3334. https://doi.org/10.1021/es010204v

(207) Koch, I.; Moriarty, M.; House, K.; Sui, J.; Cullen, W. R.; Saper, R. B.; Reimer, K. J. Bioaccessibility of lead and arsenic in traditional Indian medicines. *Sci. Total Environ*. **2011**, *409*, 4545–4552. https://doi.org/10.1016/j.scitotenv.2011.07.059

(208) Koch, I.; Moriarty, M.; Sui, J.; Rutter, A.; Saper, R. B.; Reimer, K. J. Bioaccessibility of mercury in selected Ayurvedic medicines. *Sci. Total Environ*. **2013**, *454-455*, 9–15. https://doi.org/10.1016/j.scitotenv.2013.02.089

(209) Bolan, S.; Kunhikrishnan, A.; Chowdhury, S.; Seshadri, B.; Naidu, R.; Ok, Y. S. Comparative analysis of speciation and bioaccessibility of arsenic in rice grains and complementary medicines. *Chemosphere* **2017**, *182*, 433–440. https://doi.org/10.1016/j.chemosphere.2017.04.126

(210) Giacomino, A.; Abollino, O.; Casanova, C.; La Gioia, C.; Magi, E.; Malandrino, M. Determination of the total and bioaccessible contents of essential and potentially toxic elements in ayurvedic formulations purchased from different commercial channels. *Microchem. J.* **2015**, *120*, 6–17. https://doi.org/10.1016/j.microc.2014.12.005

(211) California Office of Environmental Health Hazard Assessment (OEHHA). Proposition 65 in Plain Language. https://oehha.ca.gov/proposition-65/general-info/proposition-65-plain-language [Accessed 28 November 2021].

(212) Tessier, A.; Campbell, P. G. C.; Bisson, M. Sequential extraction procedure for the speciation of particulate trace metals. *Anal. Chem*. **1979**, *51*, 844–851. https://doi.org/10.1021/ac50043a017

(213) Koch, I.; McPherson, K.; Smith, P.; Easton, L.; Doe, K. G.; Reimer, K. J. Arsenic bioaccessibility and speciation in clams and seaweed from a contaminated marine environment. *Mar. Pollut. Bull*. **2007**, *54*, 586–594. https://doi.org/10.1016/j.marpolbul.2006.12.004

(214) Vinas, P.; López-Garcia, I.; Merano, B.; Cempillo, N.; Hernández-Córdoba, M. Speciation of arsenic in baby foods and the raw fish ingredients using liquid chromatography-hydride generation-atomic absorption spectrometry. *Chromatographia* **2003**, *57*, 611–616. https://doi.org/10.1007/BF02491737

(215) Moreda-Pineiro, A.; Moreda-Pineiro, J.; Herbello-Hermelo, P.; Bermejo-Barrera, P.; Moniategui-Lorenzo, S.; López-Mahía, P.; Prada-Rodríguez, D. Application of fast ultrasound water-bath assisted enzymatic hydrolysis – High performance liquid chromatography–inductively coupled plasma-mass spectrometry procedures for arsenic speciation in seafood materials. *J. Chromatogr. A* **2011**, *1218*, 6970–6980. https://doi.org/10.1016/j.chroma.2011.07.101

(216) Almela, C.; Laparra, J. M.; Vélez, D.; Barberá, R.; Farré, R.; Montoro, R. Arsenosugars in Raw and Cooked Edible Seaweed: Characterization and Bioaccessibility. *J. Agric. Food Chem*. **2005**, *53*, 7344–7351. https://doi.org/10.1021/jf050503u

(217) Pardo-Martínez, M.; Vinas, P.; Fisher, A.; Hill, S. J. Comparison of enzymatic extraction procedures for use with directly coupled high performance liquid chromatography-inductively coupled plasma mass spectrometry for the speciation of arsenic in baby foods. *Anal. Chim. Acta* **2001**, *441*, 29–36. https://doi.org/10.1016/S0003-2670(01)01070-4

(218) Bhowmick, S.; Pramanik, S.; Singh, P.; Mondal, P.; Chatterjee, D.; Nriagu, J. Arsenic in groundwater of West Bengal, India: A review of human health risks and assessment of possible intervention options. *Sci. Total Environ*. **2018**, *612*, 148-169. https://doi.org/10.1016/j.scitotenv.2017.08.216

(219) Bolan, S.; Kunhikrishnan, A.; Seshadri, B.; Choppala, G.; Naidu, R.; Bolan, N. S.; Ok, Y. S.; Zhang, M.; Li, C.-G.; Li, F.; et al. Sources, distribution, bioavailability, toxicity, and risk assessment of heavy metal(loid)s in complementary medicines. *Environ. Int.* **2017**, *108*, 103–118. https://doi.org/10.1016/j.envint.2017.08.005

(220) Cubadda, F.; Jackson, B. P.; Cottingham, K. L.; Van Horne, Y. O.; Kurzius-Spencer, M. Human exposure to dietary inorganic arsenic and other arsenic species: State of knowledge, gaps and uncertainties. *Sci. Total Environ.* **2017**, *579*, 1228-1239. https://doi.org/10.1016/j.scitotenv.2016.11.108

(221) Arslan, B.; Djamgoz, M. B. A.; Akün, E. Arsenic: A Review on Exposure Pathways, Accumulation, Mobility and Transmission into the Human Food Chain. In: Gunther, F. A.; de Voogt, P. (Eds.) *Reviews of Environmental Contamination and Toxicology*, Vol. 243 of Springer Reviews of Environmental Contamination and Toxicology (Continuation of Residue Reviews). Springer, Cham, **2016**, 27-51. https://doi.org/10.1007/398_2016_18

(222) Ardini, F.; Dan, G.; Grotti, M. Arsenic speciation analysis of environmental samples. *J. Anal. At. Spectrom*. **2020**, *35*, 215-237. https://doi.org/10.1039/C9JA00333A