Journal of Hazardous Materials 191 (2011) 342–348

Contents lists available at ScienceDirect

Journal of Hazardous Materials

journal homepage: www.elsevier.com/locate/jhazmat

Speciation of arsenic in rice and estimation of daily intake of different arsenic species by Brazilians through rice consumption

Bruno L. Batista, Juliana M.O. Souza, Samuel S. De Souza, Fernando Barbosa Jr.*

Laboratório de Toxicologia e Essencialidade de Metais, Faculdade de Ciências Farmacêuticas de Ribeirão Preto – USP, Avenida do Café, s/n, Monte Alegre, 14040-903 Ribeirão Preto, SP, Brazil

ARTICLE INFO

Article history:
Received 4 December 2010
Received in revised form 2 March 2011
Accepted 19 April 2011
Available online 27 April 2011

Keywords:
Arsenic
Rice
Speciation
Daily intake
Risk assessment

ABSTRACT

Rice is an important source of essential elements. However, rice may also contain toxic elements such as arsenic. Therefore, in the present study, the concentration of total arsenic and five main chemical species of arsenic (As^{3+}, As^{5+}, DMA, MMA and AsB) were evaluated in 44 different rice samples (white, parboiled white, brown, parboiled brown, parboiled organic and organic white) from different Brazilian regions using high-performance liquid chromatography hyphenated to inductively coupled plasma mass spectrometry (HPLC–ICP-MS). The mean level of total arsenic was 222.8 ng g^{-1} and the daily intake of inorganic arsenic (the most toxic form) from rice consumption was estimated as 10% of the Provisional Tolerable Daily Intake (PTDI) with a daily ingestion of 88 g of rice. Inorganic arsenic (As^{3+}, As^{5+}) and dimethylarsinic acid (DMA) are the predominant forms in all samples. The percentages of species were 38.7; 39.7; 3.7 and 17.8% for DMA, As^{3+}, MMA and As^{5+}, respectively. Moreover, rice samples harvested in the state of Rio Grande do Sul presented more fractions of inorganic arsenic than rice in Minas Gerais or Goiás, which could lead to different risks of arsenic exposure.

1. Introduction

Rice (*Oryza sativa* L.), an important component of the basic Brazilian diet, is the world's second largest cereal crop. Brazil is the most important non-Asian producer and Brazilians consume 32 kg per habitant/year [1]. Rice may accumulate considerable amounts of essential elements, but also toxic elements such as arsenic (As) [2,3].

There are considerable differences in arsenic toxicity according to its chemical form. The most toxic form of arsenic, a class 1 non-threshold carcinogen, inorganic As (arsenite – As^{3+} and arsenate – As^{5+}) is absorbed and distributed bounded to plasmatic proteins accumulating in liver and kidneys [3,4]. Monomethylarsonic acid (MMA) and dimethylarsinic acid (DMA), are metabolites of inorganic arsenic and less toxic forms. However, these organic forms have been identified as possible cancer promoters [5]. On the other hand, arsenobetaine (AsB), the main arsenic compound found in seafood, is considered virtually non-toxic [6,7]. The bioavailability of arsenic species added to these toxic differences helps the risk assessment in terms of consumption of food containing As, leading to more accurate estimates of the daily intake. Therefore, speciation is the best way of estimating risk from arsenic present in food samples, especially when we consider the bioavailability of the species and the toxicity of them as well [3,8–12].

Contamination of rice by As has several sources: pollution of paddy soils due to base and precious metal mining [13,14], irrigation of paddies with arsenic-contaminated groundwater [15,16] and the use of organoarsenical pesticides [17]. Arsenic levels in rice grain are problematic even where soil As is at background levels. Furthermore, irrigation with arsenic contaminated water can lead to a gradual increase in grain As concentration [18].

Rice, unlike other cereals, is cultivated in flooded soils, where anaerobic conditions together with excessive water lead to the mobilization of As and, consequently, an elevated accumulation in the plant [19]. Arsenite, the most toxic form of arsenic, has high water solubility and soil mobility and is thus efficiently absorbed by rice roots, reaching the grains [20].

Arsenic contamination in drinking water is a significant and internationally recognized public health concern [21]. However, high As levels found in rice may also contributes significantly to As intake in different parts of the world [22]. Since FAO recommendation for daily arsenic intake is 15 μg inorganic As/kg body weight [23], toxic effects due to cumulative As exposure through rice consumption can easily occur in some regions of the world. However, little is known about arsenic levels in Brazilian rice samples and

* Corresponding author. Tel.: +55 1636024701.
E-mail address: fbarbosa@fcfrp.usp.br (F. Barbosa Jr.).

0304-3894/$ – see front matter © 2011 Elsevier B.V. All rights reserved.
doi:10.1016/j.jhazmat.2011.04.087

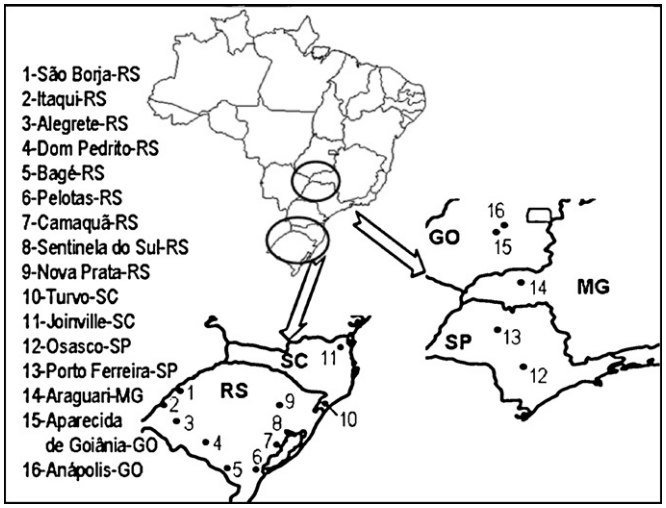

Fig. 1. A map displaying the cities where rice samples were collected.

the contribution of this staple food to the daily arsenic intake in this South American country.

Therefore, in the present study, for the first time the concentration of total arsenic and five main chemical species of arsenic (As^{3+}, As^{5+}, DMA, MMA and AsB) were evaluated in 44 different processed rice samples (white, parboiled white, brown, parboiled brown, parboiled organic and organic white) from different Brazilian regions. This study also provided estimated daily intake of arsenic and its species by Brazilians.

2. Material and methods

2.1. Reagents

High purity de-ionized water (resistivity 18.2 MΩ cm) used in all experiment was obtained using a Milli-Q water purification system (Millipore RiOs-DI™, Bedford, MA, USA). All reagents used were of analytical-reagent grade. Di-ammonium hydrogen phosphate ($(NH_4)_2HPO_4$) was purchased from Sigma (Steinheim, Germany), ammonium hydrogen phosphate ($NH_4H_2PO_4$) and ammonium hydroxide (NH_4OH) were purchased from Fluka (St. Louis, USA).

Stock solutions (100 mL at 100 mg L^{-1} as arsenic) of the five different arsenic species were prepared and stored in amber flasks at temperatures lower than 4 °C. As^{3+} (As_2O_3, Aldrich, St. Louis, USA) was prepared by dissolving salt in 50 mL NaOH 0.18 mol L^{-1}. After salt solubilization, HCl 0.3 mol L^{-1}, which makes the medium non-oxidant and prevents the conversion of As^{3+} to As^{5+}, was added. As^{5+} ($As_2O_5{\cdot}H_2O$, Aldrich, St. Louis, USA) was prepared by dissolving oxide in 50 mL NaOH 0.18 mol L^{-1}. Then the solution was acidified with HNO_3 0.3 mol L^{-1}. DMA ($C_2H_7AsO_2$, Fluka, St. Louis, USA), MMA ($Na_2CH_3O_3{\cdot}As{\cdot}6H_2O$, Chem Service, West Chester, USA) and AsB ($C_5H_{11}AsO_2$, Fluka, St. Louis, USA) were prepared by dissolving salt in HNO_3 0.15 mol L^{-1}. Intermediary solutions at 10 mg L^{-1}, which solubilized As^{3+} in HCl 0.024 mol L^{-1} and AsB, DMA, MMA and As^{5+} in HNO_3 0.0014 mol L^{-1}, were prepared.

2.2. Rice sampling and pre-treatment

Forty-four raw rice samples produced in different regions of Brazil were purchased from markets. Samples were classified as: brown (B), parboiled brown (PB), white (W, polished), parboiled white (PW, polished), parboiled organic white (PO) and organic white (OW). Of the total samples, 10PW, 12W, 2B, 6PB, 1PO and 1OW were produced in the state of Rio Grande do Sul; 3W from the state of Goiás, 3W from the state of Minas Gerais, 2W and 1B from the state of São Paulo, 1PW and 2W from the state of Santa Catarina. Rio Grande do Sul, as the largest rice producing state in Brazil (more than 50% of total rice), had the highest participation in the sampling (32 rice samples). Fig. 1 shows the cities in Brazil where the samples were produced.

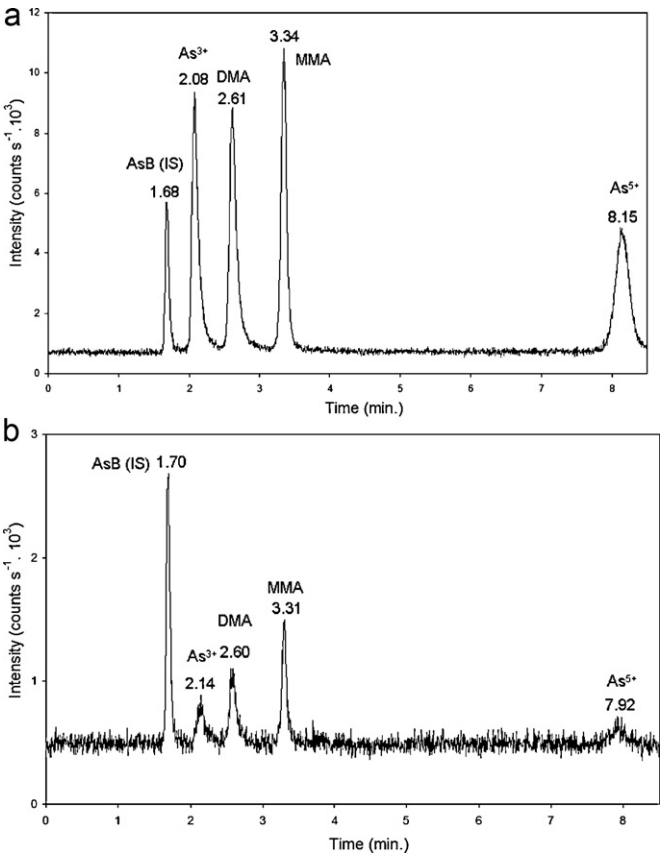

Fig. 2. Typical chromatograms obtained for (a) stock solutions of As species 10 μg L^{-1} (expressed as As) and (b) rice sample from Rio Grande do Sul. Retention time in minutes is above each peak.

After quartering, sample pre-treatment followed *Codex Alimentarius* recommendations [24]. Fifty grams of samples was individually milled, homogenized and sieved (0.25 mm) with an electric ultra-centrifugal mill (Retsch ZM200 with DR100 auto-sampler) at 6000 rpm. Then, samples were placed into clean 50 mL conic tubes (Falcon® BD) until total and chemical speciation analysis.

2.3. Determination of total arsenic in rice samples

The total amount of arsenic was determined by ICP-MS (DRC-ICP-MS ELAN DRCII, Perkin Elmer, SCIEX, Norwalk, CT, USA) in all samples to evaluate the efficiency of arsenic speciation. Results were compared with the sum of species obtained by speciation analysis. For this analysis, samples (in triplicate) were digested in closed vessels using a microwave oven decomposition system (Milestone Ethos D, Italy) according to the method proposed by Nardi et al. [25].

2.4. Rice treatment for arsenic speciation

For sample preparation we adopted a combination of two methods published by Haung et al. [26] and Sun et al. [27] in which 0.2 g of milled sample (in triplicate) was accurately weighed into a 50 mL conic tube following the addition of 10 mL of 2% (v/v) nitric

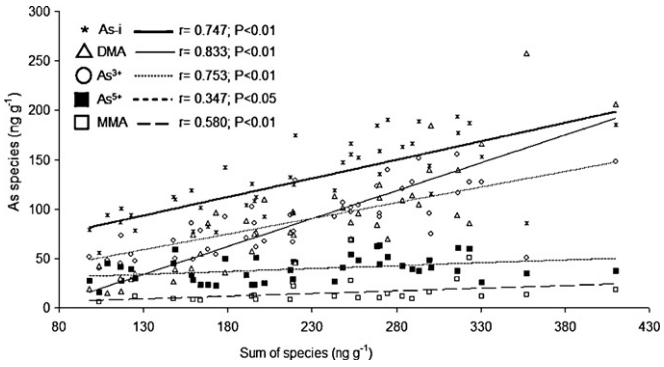

Fig. 3. Correlation between arsenic species and sum of species for 44 Brazilian rice samples.

acid. The tube was then closed with polytetrafluorethylene tape. The resulting mixture was heated in a water bath from 25 to 95 °C for 0.75 h and for 1.5 h at 95 °C. After that, samples were cooled at room temperature, filtered with a 0.20 μm cellulose filter and 380 μL was mixed with 20 μL of a solution containing 100 μg L^{-1} AsB, the internal standard (IS), at a final concentration of 5 μg L^{-1}. Then, samples were directly injected into the HPLC–ICP-MS system for chemical speciation analysis. Certified reference material (SRM 1568a) and blanks were run with each extract batch. Fig. 2 shows a common chromatogram of a standard and a sample analysis.

2.5. Instrumentation for arsenic speciation

A HPLC–ICP-MS system was used for chemical speciation analysis. A Perkin Elmer model L-200 LC pump, six-port injector (Rheodyne 9725) with an anion-exchange column PRP-X100 (Hamilton, Reno, NV, USA) comprised the LC system. Samples were loaded with a syringe into a 100 μL sample loop. All separations were performed at room temperature under isocratic conditions. The isocratic mobile phase, prepared daily, was 10 mM $HPO_4^{2-}/H_2PO_4^-$ (98% v/v) + methanol 2% (v/v), at pH 8.5, in accordance with Sanz et al. [28]. Analytical calibration standards for arsenic species were prepared daily over the range of 0.0–20.0 μg L^{-1} for the LC–ICP-MS method by suitable serial dilutions of the stock solution in the extractor (HNO_3 2% v/v). The flow rate was 1.0 mL min^{-1}. The effluent from the LC column was directly connected to the nebulizer with PEEK tubing (1.59 mm o.d.) and a low dead volume PEEK connector. Data evaluation was performed using Chromera® software (version 2.1.0.1631) supplied with the instrument, and quantification was based on peak high by external calibration. Typical chromatograms obtained for stock solutions of As species 10 μg L^{-1} (expressed as As) and a rice sample from Rio Grande do Sul are presented in Fig. 2.

2.6. Estimation of arsenic species daily intake

The daily intake of arsenic species depends on both the arsenic compound in rice and daily rice consumption. Estimated daily intake was calculated as: EDI = $C_{ce} \times M_{rdc}$, where EDI is the estimated compound daily intake (μg day^{-1} $person^{-1}$), C_{ce} is the mean concentration of each arsenic form in rice, weighted by Brazilian rice consumption (W, representing 77.7% of rice consumed in the country and other rice types (PW, B and PB), represented 22.3%). M_{rdc} is the mass of rice consumed daily in Brazil, based on a Brazilian national survey of rice consumption which shows consumption of polished rice (W) and other types (PW, PB and B) in the different Brazilian regions [1]. It can be pointed out that the forty-four rice samples used in the present study are produced by the biggest Brazilian producers and together represent most of the rice consumed in the whole country regardless of the region.

Estimated daily intake was compared to the Provisional Tolerable Daily Intake (PTDI) for total arsenic (JECFA/WHO guidelines) [29,30].

2.7. Data quality control

In order to guarantee the accuracy and precision of the proposed method, Rice Flour Standard Reference Material from the National Institute of Standards and Technology (NIST), SRM 1568a, was analyzed throughout the experiment, For total arsenic we found 297 ± 8 ng g^{-1} in this reference material (n = 5), which was in good agreement with the certified value (290 ± 30 ng g^{-1}). For speciation data (n = 6) we compared our results with the results of other studies in the literature for the analysis of the same SRM (Table 1).

3. Results and discussion

3.1. Total arsenic concentration in rice samples from Brazil: comparison with other countries

Total arsenic levels in the Brazilian rice samples varied from 107.9 to 427.7 ng g^{-1}, with a mean of 222.8 ng g^{-1}.

Among the different types of rice analyzed in this study B rice presented the highest levels of arsenic (mean of 348.4 ng g^{-1}) followed by PB rice (mean of 265.9 ng g^{-1}), W rice (mean of 222.9 ng g^{-1}) and PW rice (214.9 ng g^{-1}) as shown in Table 2. These values are quite close to those obtained for rice samples cultivated in other areas (Table 2), except Canada (65 ng g^{-1} for wild rice) and India (45.6 ng g^{-1} for W rice and 70 ng g^{-1} for B rice).

Rio Grande do Sul State, with the most representative number of samples (n = 32), presented mean arsenic levels of 214.8 ± 81.3 ng g^{-1} for W rice (W and OW) (Table 3) with the rice from the city of Pelotas having the highest arsenic levels. On the other hand, arsenic levels for PW rice were 223.7 ± 93.6 ng g^{-1}.

Table 1
Comparison of arsenic species determination in NIST 1568a rice flour from different studies (Arsenic certified reference value: 290 ± 30 ng g^{-1}).

Species sum (ng g^{-1})	Recovery (%)	As^{3+} (ng g^{-1})	As^{5+} (ng g^{-1})	DMA (ng g^{-1})	MMA (ng g^{-1})	Reference
272.8 ± 9.9	94.1	63.4 ± 3.5	50.3 ± 2.9	144.2 ± 4.5	14.9 ± 3.9	This study
286.4 ± 6.1	99.1 ± 2.1	129.2 ± 3.1	15.4 ± 3.8	31.5 ± 1.6	1.9 ± 0.7	[28]
281 ± 2	97.0	52 ± 1	44 ± 2	173 ± 2	12 ± 0.8	[50]
271 ± 3	93 ± 1	67 ± 5	36 ± 1	162 ± 1	5 ± 1	[14]
286.4 ± 6.2	82.3 ± 1.6	68.3 ± 3.7	20.5 ± 2.3	135.4 ± 4.1	8.1 ± 1.3	[49]
276	95.2	75	12	180	9	[51]
272	93.8	55 ± 6	41 ± 3	166 ± 6	10 ± 2	[52]
277	95.5	67 ± 4	39 ± 3	158 ± 5	13 ± 2	[53]
288.2	99.4	54.7 ± 1.4	53.7 ± 3.3	165 ± 8	14.8 ± 1.8	[54]
240 ± 40	80 ± 12	80 ± 14		160 ± 24	2	[55]
274	94	92 ± 4		174 ± 9	8 ± 2	[56]
290 ± 10	98.3	110 ± 10		180 ± 3		[27]

Table 2
Comparison of arsenic species composition in rice samples from different countries (values set as: mean (min–max)).

Samples	Rice type	AsB	{As-i}		DMA	MMA	Species sum	Total Arsenic	Reference
			As^{3+}	As^{5+}					
Brazil	W	–	78(40–156)	34(16–62)	93(39–258)	8(0–29)	212(98–357)	223(109–376)	This study
	PW	–	87(45–127)	43(24–60)	65(17–139)	10(0–51)	207(116–323)	215(108–367)	
	B	–	146(139–151)	42(37–51)	127(70–206)	11(0–18)	326(275–410)	348(271–428)	
	PB	–	104(94–129)	52(38–69)	82(46–111)	6(0–28)	244(217–270)	266(226–316)	
USA	W	–	{76(20–100)}		77(50–260)	<LOD	231(160–340)	277(170–400)	Williams et al. [55]
	W	–	92(79–101)	42(32–51)	137(141–136)	–	271(254–289)	329(308–350)	Zhu et al. [14]
	W	–	{110}		100.0	<LOD	210	280	Meharg et al. [40]
	W	–	86(49–122)	17(3–95)	155(40–302)	0.6(0–6)	257(169–382)	265(162–383)	Zavala et al. [33]
	B	–	{105(60–140)}		90(10–150)	<LOD	195(100–290)	225(110–340)	Williams et al. [55]
	B	–	{170}		140	10	320	440	Meharg et al. [40]
	B	–	131(97–168)	8.2(<5–13)	173(36–572)	1.4(0–13)	313(164–769)	331(201–710)	Zavala et al. [33]
Canada	Wi	–	{45(10–80)}		10	<LOD	50(10–90)	65(20–110)	Williams et al. [55]
India	W	–	{27(20–40)}		66	0.7	32(20–40)	46(30–50)	Williams et al. [55]
	B	–	{40}		<LOD	<LOD	40	70	Williams et al. [55]
	Re	–	{50}		10	<LOD	60	80	Williams et al. [55]
India/Bangladesh	NE	4(2–11)	240(110–341)	40(0–137)	5(1–16)	1(0–4)	290(137–479)	354(143–637)	Sanz et al. [49]
Bangladesh	NE	–	{83(10–210}		19(0–50)	–	101(20–240)	131(30–300)	Williams et al. [55]
	B	–	{280}		170	10	460	610	Meharg et al. [40]
Taiwan	W	–	{247(110–510)}		37(30–50)	32(15–60)	310(160–610)	383(190–760)	Williams et al. [55]
China	W	–	114(51–302)	40(24–84)	40(9–147)	1.3(7–13)	195(79–495)	230(19–586)	Zhu et al. [14]
	W	–	76(48–190)	6.2(0–27)	26(5–44)	1.1(0–6)	109(65–268)	114(65–274)	Liang et al. [23]
	B	–	{210}		90	10	310	360	Meharg et al. [40]
Thailand	Ja	–	{80}		30	<LOD	110	110	Williams et al. [55]
Spain	Pa	–	{80}		50	<LOD	130	170	Williams et al. [55]
Italy	B	–	{100}		50	<LOD	150	190	Williams et al. [55]
	Ri	–	{130(120–140)}		85(80–90)	<LOD	215(210–220)	220(220–220)	Williams et al. [55]

PW, parboiled white rice; W, white rice; B, brown rice; PB, parboiled brown rice; Wi, wild; Re, red; NE, not established; Ja, jasmine; Pa, paella; Ri, risotto.

Arsenic levels for B and PB rice were 349.2 ± 111.0 and $262.3 \pm 32.7\ ng\ g^{-1}$, respectively, and the highest levels were found in rice samples produced in the cities of Nova Prata and Pelotas.

On the other hand, the states of São Paulo and Minas Gerais, presented the highest levels of arsenic in W rice (mean of $299.6 \pm 114.4\ ng\ g^{-1}$ and $256.2 \pm 78.0\ ng\ g^{-1}$, respectively) (Table 4). The state of Goiás had the lowest arsenic concentrations for W rice (mean of $195 \pm 1.4\ ng\ g^{-1}$).

In Goiás, latosol soil [31] is the most common type so rice cultivation is predominantly upland (non-irrigated). Furthermore, the

Table 3
Rice samples from Rio Grande do Sul, the largest producer in Brazil ($n = 32$).

Rice type	City	Total As ($ng\ g^{-1}$)	As^{3+} ($ng\ g^{-1}$)	As^{5+} ($ng\ g^{-1}$)	DMA ($ng\ g^{-1}$)	MMA ($ng\ g^{-1}$)	Species sum ($ng\ g^{-1}$)	Recovery (%)
W	1a	135.3 ± 3.7	47.6 ± 2.9	30.2 ± 6.9	35.3 ± 1.9	11.8 ± 3.3	124.9 ± 3.7	92.3
	2a	122.2 ± 3.6	48.6 ± 2.3	45.2 ± 6.8	14.6 ± 4.1	–	108.4 ± 4.4	88.7
	2b	270.6 ± 4.9	120.6 ± 2.1	42.4 ± 0.2	108.6 ± 4.5	11.9 ± 2.6	283.5 ± 2.4	104.8
	2c	109.3 ± 2.5	51.3 ± 3.9	27.5 ± 2.9	19.2 ± 3.1	–	98.1 ± 3.3	89.7
	3a	247.8 ± 5.2	92.1 ± 3.9	26.8 ± 0.8	113.1 ± 7.6	11.5 ± 1.5	243.5 ± 3.5	98.3
	3b	139.0 ± 5.0	78.0 ± 7.8	23.4 ± 2.6	54.7 ± 6.6	7.7 ± 1.4	163.8 ± 4.6	117.8
	4a	318.3 ± 12.3	127.4 ± 8.0	25.6 ± 0.6	165.8 ± 13.4	11.8 ± 0.7	330.6 ± 5.7	103.9
	6a	256.5 ± 7.2	66.7 ± 3.0	28.8 ± 10.1	97.6 ± 13.7	25.4 ± 0.6	218.5 ± 6.9	85.3
	6b	373.9 ± 20.4	155.7 ± 2.6	37.7 ± 7.4	93.6 ± 13.0	29.3 ± 8.9	316.3 ± 8.0	84.6
	6c	207.3 ± 10.2	66.9 ± 2.8	25.2 ± 8.1	109.4 ± 14.5	–	201.5 ± 8.5	97.2
	6d	263.1 ± 9.4	95.8 ± 1.4	48.1 ± 3.9	139.8 ± 2.8	16.2 ± 3.7	299.8 ± 2.4	113.9
	7	174.9 ± 5.4	50.1 ± 9.5	59.4 ± 8.2	39.1 ± 6.3	–	148.7 ± 8.0	85.1
OW	7	174.1 ± 16.1	85.6 ± 1.8	33.1 ± 0.7	39.9 ± 9.3	–	158.7 ± 3.9	91.2
PW	1a	119.6 ± 3.7	54.4 ± 10.6	39.1 ± 6.0	29.0 ± 4.2	–	122.5 ± 6.9	102.4
	1b	107.9 ± 2.9	44.9 ± 4.4	41.5 ± 0.5	30.0 ± 2.3	–	116.3 ± 2.4	107.8
	1c	317.1 ± 8.1	127.3 ± 5.1	39.0 ± 13.6	113.8 ± 5.7	9.1 ± 1.8	289.2 ± 6.6	91.2
	2	127.1 ± 6.7	67.8 ± 8.9	44.7 ± 2.5	26.2 ± 3.4	9.3 ± 5.3	148.0 ± 5.0	116.4
	5	366.7 ± 14.9	127.1 ± 2.2	60.2 ± 0.6	85.5 ± 8.3	50.5 ± 9.2	323.3 ± 5.1	88.2
	6a	366.6 ± 22.3	116.7 ± 8.8	60.4 ± 1.9	139.4 ± 5.0	–	316.5 ± 5.2	86.3
	6b	216.0 ± 7.2	77.5 ± 2.7	46.9 ± 3.5	73.6 ± 9.8	21.3 ± 0.7	219.3 ± 4.2	101.6
	7a	226.8 ± 4.3	61.1 ± 7.5	50.9 ± 8.9	76.3 ± 8.8	8.6 ± 1.7	196.8 ± 5.4	86.8
	7b	241.2 ± 6.1	103.6 ± 5.0	48.5 ± 3.5	95.7 ± 8.1	9.6 ± 1.7	257.4 ± 4.6	106.7
	7c	210.5 ± 7.2	102.2 ± 4.8	23.6 ± 0.3	57.0 ± 3.1	11.6 ± 1.6	194.4 ± 2.5	92.4
PO	2	161.6 ± 8.3	92.5 ± 5.4	49.9 ± 8.3	36.1 ± 0.1	–	178.5 ± 4.6	110.5
B	9	427.7 ± 16.3	148.2 ± 9.6	37.5 ± 2.3	206.2 ± 14.1	18.0 ± 1.7	410.0 ± 6.9	95.8
	7	270.7 ± 5.7	139.2 ± 4.0	51.3 ± 14.7	70.1 ± 8.2	14.2 ± 0.4	274.8 ± 6.8	101.5
PB	6a	287.7 ± 6.9	106.3 ± 7.6	40.6 ± 3.7	101.2 ± 11.3	–	248.1 ± 7.5	86.2
	6b	316.4 ± 9.0	95.5 ± 4.6	63.1 ± 6.4	111.4 ± 6.1	–	270.0 ± 5.7	85.4
	7a	240.6 ± 6.4	101.2 ± 5.6	53.9 ± 6.2	70.1 ± 0.5	27.8 ± 1.2	253.1 ± 3.4	105.2
	7b	226.3 ± 6.3	94.2 ± 3.7	38.3 ± 6.1	76.7 ± 6.7	7.9 ± 0.6	217.1 ± 4.3	95.9
	8a	255.6 ± 9.8	128.8 ± 1.8	45.3 ± 1.2	46.0 ± 9.2	–	220.1 ± 4.1	86.2
	8b	268.9 ± 5.3	97.5 ± 4.6	68.9 ± 1.8	86.5 ± 3.2	–	252.9 ± 3.2	94.1

PW, parboiled white rice; W, white rice; B, brown rice; PB, parboiled brown rice; PO, parboiled organic rice (white); OW, organic white rice; a–d, different samples from the same city.

Table 4
Rice samples from other states in Brazil ($n = 12$).

Rice type	City	Total As ($ng\,g^{-1}$)	As^{3+} ($ng\,g^{-1}$)	As^{5+} ($ng\,g^{-1}$)	DMA ($ng\,g^{-1}$)	MMA ($ng\,g^{-1}$)	Species sum ($ng\,g^{-1}$)	Recovery (%)
Goiás								
W	15	193.8 ± 8.1	54.2 ± 2.9	22.2 ± 1.5	96.7 ± 3.0	–	173.2 ± 2.5	89.3
	16a	194.7 ± 7.4	58.7 ± 3.2	23.5 ± 0.4	86.0 ± 4.5	–	168.1 ± 2.7	86.4
	16b	196.6 ± 5.2	85.4 ± 5.5	22.9 ± 2.9	74.9 ± 13.9	12.8 ± 5.9	195.9 ± 7.1	99.6
Santa Catarina								
W	10	128.2 ± 3.9	39.7 ± 2.4	15.6 ± 1.3	42.6 ± 4.3	6.0 ± 1.4	103.9 ± 2.4	81.0
PW	11	118.0 ± 4.8	72.9 ± 3.4	27.5 ± 1.6	16.5 ± 0.9	–	116.9 ± 2.0	99.1
São Paulo								
W	12	320.1 ± 6.9	122.8 ± 3.5	62.1 ± 0.8	83.9 ± 2.0	–	268.8 ± 2.1	83.9
	12	375.8 ± 6.2	51.1 ± 10.0	34.7 ± 3.3	257.8 ± 4.5	13.5 ± 1.3	357.1 ± 4.8	95.0
	13	155.7 ± 5.2	48.9 ± 6.3	28.1 ± 3.7	74.0 ± 13.6	8.5 ± 3.4	159.4 ± 5.4	102.4
B	12	346.9 ± 9.2	151.1 ± 6.1	37.3 ± 1.3	105.0 ± 5.4	–	293.4 ± 4.3	84.5
Minas Gerais								
W	14a	343.9 ± 15.7	75.2 ± 12.9	40.4 ± 5.8	184.9 ± 5.9	–	300.4 ± 8.2	87.3
	14b	230.3 ± 5.6	91.9 ± 10.7	43.8 ± 7.3	124.4 ± 6.7	9.9 ± 1.6	269.9 ± 6.6	117.2
	14c	194.4 ± 3.6	70.4 ± 7.8	33.1 ± 1.7	87.7 ± 14.9	–	191.1 ± 8.1	98.3

PW, parboiled white rice; W, white rice; B, brown rice; a–c, different samples from the same city.

iron concentration in this type of soil is high, which reduces As uptake by the rice roots [20,32]. In flooded conditions such as in Rio Grande do Sul (planosol/clay soils), however, anaerobic conditions [19] and high As mobility [20] increase arsenic concentrations, as reported in Tables 3 and 4.

Williams et al. [17] reported higher levels of arsenic in rice from the south central U.S. than in rice from California. They speculated that arsenic in south central U.S. rice may have originated from pesticides in soil previously used to grow cotton. Zavala et al. [33] confirmed these results, finding especially high arsenic levels in rice from one Texas supplier. In the present study we do not have the history of the soil where the rice was grown, but an interesting finding is related to the results of arsenic levels in two organic rice samples (one PO and one OW), where pesticides were not previously used. These samples were produced in Rio Grande do Sul with mean arsenic levels of 174.1 and 161.6 $ng\,g^{-1}$ for organic white and parboiled organic rice, respectively.

3.2. Arsenic speciation in Brazilian rice

Arsenic is a ubiquitous element present in the aquatic and terrestrial environment and its chemical forms, mainly inorganic arsenic, are toxic to plants, animals and humans [4,15,33]. In roots, rice plants absorb arsenite mainly via silicic acid transporters and arsenate by competition with phosphate due to similarity. However, the methylated forms DMA and MMA are absorbed much more slowly than inorganic forms [20,34]. Xu et al. [19] showed differences between the crop type and absorption of arsenic species. DMA and inorganic arsenic species prevail in rice grains under flooded (anaerobic) and aerobic conditions, respectively [19]. These inorganic forms and other toxic elements interfere in the metabolism of plant cells interacting with sulphydryl groups, promoting replacement of phosphate from adenosine tri-phosphate [35], decreasing plant growth and crop yield [36], generating free radicals [37], promoting lipid peroxidation and reducing germination, weight and growth of roots and shoot rice [35]. Rice detoxifies arsenite by means of efflux at the roots [38] and chelation by phytochelatins, compounds containing thiol [39].

In terms of the species found in this study with Brazilian rice samples, we found four arsenic species cited above. Inorganic arsenic (As^{3+}, As^{5+}) and DMA were the predominant forms. For DMA, As^{3+}, MMA and As^{5+} the average percentages found in all samples were 38.7, 39.7, 3.7 and 17.8%, respectively.

Brown rice from São Paulo and Rio Grande do Sul presented the highest inorganic arsenic values (188.4 and 190.5 $ng\,g^{-1}$, respectively), and W rice from Santa Catarina and Goiás had the lowest levels (55.3 and 76.4 $ng\,g^{-1}$, respectively). With the lowest levels of As^{3+} and As^{5+}, W rice from Santa Catarina had results of 39.7 and 15.6 $ng\,g^{-1}$, respectively, while the highest level for As^{3+} was 155.7 $ng\,g^{-1}$ in W rice from Pelotas Rio Grande do Sul, and the highest level for As^{5+} was 68.9 $ng\,g^{-1}$ in PB rice from Sentinela do Sul, Rio Grande do Sul.

Brown rice from Rio Grande do Sul and W rice from São Paulo presented the highest average values of organic arsenic (154.3 and 271.3 $ng\,g^{-1}$, respectively), and PW and W rice from Santa Catarina had the lowest levels (16.5 and 48.6 $ng\,g^{-1}$, respectively). W rice from Santa Catarina showed the lowest level of DMA, at 16.5 $ng\,g^{-1}$, while the lowest detectable level of MMA was 6.0 $ng\,g^{-1}$, also in Santa Catarina W rice. The highest level for DMA for rice produced in Brazil was 206.2 $ng\,g^{-1}$ in B rice from Nova Prata and for MMA it was 50.5 $ng\,g^{-1}$ in PW rice from Bagé, both in Rio Grande do Sul.

Table 5
Estimated daily intake of arsenic species and potential health risk due to consumption of rice (B, PB, W, PW, OW and PO) for Brazilians of different regions, based on the POF/IBGE [1].

Estimated daily intake							
Region	North	Northeast	Southeast	South	Center-West	Brazil (Mean intake) (% of the PTDI[A])	PTDI[A]
Arsenic species ingestion (μg/day)[B]							
As^{3+}	7.85	7.85	7.61	6.24	10.76	7.72 (5.2)	150 (Total As)
As^{5+}	3.54	4.15	3.44	2.77	4.82	3.45 (2.3)	
DMA	7.71	7.42	7.53	5.97	10.46	7.49 (5.0)	
MMA	0.75	0.69	0.73	0.14	1.00	0.71 (0.5)	
Total As	21.33	20.94	20.77	16.74	29.10	20.85 (13.9)	

B-(considering 70 kg body weight person). A-PTDI = Provisional tolerable daily intake for toxic elements, based on PTWI/7, (WHO guidelines) [29,30].

A comparison between arsenic species levels found in Brazilian rice and in rice samples produced in other countries is shown in Table 2. Interestingly, AsB was only found in rice from India and Bangladesh.

Recently, Meharg et al. [40] demonstrated that B rice had a higher concentration of As-i than W rice, which can be confirmed when we compare the As-i average concentration in B and W rice from the USA or Brazil (Table 2).

4. Fractions of inorganic arsenic and DMA in Brazilian rice samples

According to Zavala et al. [33], rice may be divided into two types, depending on the form of arsenic in the grain: inorganic arsenic-type and DMA-type. They reported that as arsenic levels rise, rice contains more methylated arsenic (DMA) but Meharg et al. [3] noted that the amount of DMA is dependent on the rice cultivar.

As shown in Fig. 3, Brazilian rice samples did not tend toward the inorganic or DMA type since there was a high correlation between DMA and As^{3+} and the sum of arsenic species (Fig. 3). However, it seems that rice samples from Rio Grande do Sul contained more fractions of inorganic arsenic than those from Minas Gerais or Goiás regions (Tables 3 and 4).

5. Estimation of the daily intake of arsenic species from rice consumption and exposure risk assessment

Arsenic toxicity depends on its chemical form. For inorganic arsenic, the lethal dose for 50% of rats varied from 15 to 293 mg As kg^{-1} body weight [41]. For humans, 70–80 mg of arsenic trioxide ingestion was reported as fatal [42]. As^{3+} has a high affinity for the sulphydryl groups of biomolecule such as glutathione (GSH), lipoic acid and the cysteinyl residues of many enzymes [43].

The oral route is the most common for inorganic arsenic exposure. However, in Brazil, a staple food such as rice is an important source of exposure. Average rice consumption in Brazil is 86.5 g day^{-1} $person^{-1}$, of which 77.7% is polished rice (W rice) and 22.3% other rice types (PW, B, PB, PO or OW) [1]. Brazilians' total daily intake of arsenic represents approximately 14% of PTDI (Table 5).

Estimated daily intake of As^{3+}, As^{5+}, MMA and DMA through rice consumption was 7.72 μg; 3.45 μg; 0.71 μg and 7.49 μg, respectively and the variations in the five regions of Brazil is shown in Table 5. These values was estimated based on the mean daily ingestion of rice by Brazilians and represent a considerable percentage of the PTDI for arsenic where inorganic arsenic (the most toxic form) is in some Brazilian rice the predominant form. However, it can be pointed out that a consumption of more than 350 g of rice per day is very common by Brazilian males. Then, the intake of inorganic arsenic from rice must easily reach the PTDI for arsenic for many Brazilians. Torres-Escribano et al. [44] found average values ranging from 1.2 to 44.7% of PTDI for inorganic arsenic ingestion through rice consumption in Spain and Asia, respectively. Liang et al. [22] found PTDI of 45.1 μg for inorganic arsenic and highlighted that in China, inorganic arsenic consumption through rice is 25 times higher than through potable water.

Considering that water is a source of arsenic exposure, it is important to note that the bioavailability of metal and its main species in the body undergo changes that may be significant and result in further damage to human health.

Brazilian laws prescribe limits to the amount of arsenic found in drinking water. In terms of arsenic content in drinking water, resolutions of the Brazilian Environmental Agency [45] and Brazilian Health Ministry [46] establish limits of 50 μg L^{-1} and 10 μg L^{-1}, respectively. The latter limit cited is the one suggested by the World Health Organization [47]. Knowing that a person consumes an average of 2 L of water per day, we can predict an average daily intake of 20 μg to be the maximum intake through water contaminated with arsenic at the highest level allowed by WHO. The total levels of arsenic accumulated throughout a Brazilian's life through consumption of drinking water and rice can be estimated by the following equation:

$$\left[\frac{\text{water}}{\text{rice As level}}\right] \times [\text{average lifetime}\,(73.1\ \text{year})] \times [\text{ingestion rate}\,(365\ \text{days year}^{-1})] \times \left[\frac{\text{water}\,(2\ \text{L})}{\text{rice daily consumption}\,(86.5\ \text{g})}\right]$$

Then, hypothetically, the ingestion of arsenic via drinking water (considering 10 μg L^{-1} in water) and rice for the lifetime of a Brazilian is 533.6 mg and 556.3 mg, respectively. Some works showed the importance of the study of bioavailability of arsenic species *in vivo* and *in vitro*. Animal model (swine) was used for the study of bioavailability in vegetables [12] and rice [11] containing arsenic; cell culture and *in vitro* gastrointestinal digestion was used for simulate the absorption of arsenic from rice samples [10]. These studies showed that inorganic As, specially As^{5+}, could be available for intestinal absorption than organic forms. For swine, the bioavailability achieved almost 90% for inorganic species, 16.7% for MMA and 33.3% for DMA through oral exposition. Furthermore, arsenic bioavailability is modified according to the food type and the cooking [44,48]. Differences in arsenic absorption were observed in swine fed with rice, lettuce, mung beans, chard and radish [11,12]. On the other hand, rice cooked with arsenic contaminated water showed more bioavailability of inorganic arsenic than rice cooked with non-contaminated water [10]. Therefore, future studies are necessary to better understand the real risks of the intake of high levels of inorganic arsenic from rice, to reduce uncertainties in estimating exposure and legislation on food safety must consider the risks.

Acknowledgements

Authors are grateful to Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for financial support and fellowships.

References

[1] Consumer Expenditure Survey – POF 2002–2003; Brazilian Institute of Geography and Statistics (IBGE), Brazilian Government, http://www.ibge.gov.br/english/presidencia/noticias/noticia_impressao.php?id_noticia=278, 2003 (accessed November 2010).
[2] P.N. Williams, A. Villada, C. Deacon, A. Raab, J. Figuerola, A.J. Green, J. Feldmann, A.A. Meharg, Greatly enhanced As shoot assimilation in rice leads to elevated grain levels compared to wheat and barley, Environ. Sci. Technol. 41 (2007) 6854–6859.
[3] A.A. Meharg, P.N. Williams, E. Adomako, Y.Y. Lawgali, D. Deacon, A. Villada, R.C.J. Cambell, G. Sun, Y.G. Zhu, J. Feldmann, A. Raab, F.-J. Zhao, R. Islam, S. Hossain, J. Yanai, Geographical variation in total and inorganic As content of polished (white) rice, Environ. Sci. Technol. 43 (2009) 1612–1617.
[4] Toxicological Profile for Arsenic, Agency for Toxic Substances and Disease Registry (ATSDR), Public Health Service, Atlanta, 2007.
[5] J. Brown, K. Kitchin, Dimethylarsinic acid treatment alters six different rat biochemical parameters: relevance to arsenic carcinogenesis, Teratog. Carcinog. Mutagen. 17 (1997) 71–84.
[6] M. Vahter, E. Marafante, L. Dencker, Metabolism of arsenobetaine in mice, rats and rabbits, Sci. Total Environ. 30 (1983) 197–211.
[7] E. Marafante, M. Vahter, L. Dencker, Metabolism of arsenocholine in mice, rats and rabbits, Sci. Total Environ. 34 (1984) 223–240.
[8] W.R. Cullen, K.J. Reimer, Arsenic speciation in the environment, Chem. Rev. Am. Chem. Soc. 89 (1989) 713–764.

[9] V. Foà, A. Colombi, M. Maroni, M. Buratti, G. Calzaferri, The speciation of the chemical forms of arsenic in the biological monitoring of exposure to inorganic arsenic, Sci. Total Environ. 34 (1984) 241–259.
[10] J.M. Laparra, D. Vélez, R. Barberá, R. Farré, R. Montoro, Bioavailability of inorganic arsenic in cooked rice: practical aspects for human health risk assessments, J. Agric. Food Chem. 53 (2005) 8829–8833.
[11] A.L. Juhasz, E. Smith, J. Weber, M. Rees, A. Rofe, T. Kuchel, L. Sansom, R. Naidu, *In vivo* assessment of arsenic bioavailability in rice and its significance for human health risk assessment, Environ. Health Perspect. 114 (2006) 1826–1831.
[12] A.L. Juhasz, E. Smith, J. Weber, M. Rees, A. Rofe, T. Kuchel, L. Sansom, R. Naidu, Application of an in vivo swine model for the determination of arsenic bioavailability in contaminated vegetables, Chemosphere 71 (2008) 1963–1969.
[13] X. Liao, T. Chen, H. Xie, Y. Liu, Soil As contamination and its risk assessment in areas near the industrial districts of Chenzhou city, Southern China, Environ. Int. 31 (2005) 791–798.
[14] Y.G. Zhu, G.X. Sun, M. Lei, M. Teng, Y.X. Liu, N.C. Chen, L.H. Wang, A.M. Carey, C. Deacon, A. Raab, A.A. Meharg, P.N. Williams, High percentage inorganic arsenic content of mining impacted and non-impacted Chinese rice, Environ. Sci. Technol. 42 (2008) 5008–5013.
[15] A.A. Meharg, M. Rahman, Arsenic contamination of Bangladesh paddy field soils: implications for rice contribution to As consumption, Environ. Sci. Technol. 37 (2003) 229–234.
[16] P.N. Williams, M.R. Islam, E.E. Adomako, A. Raab, S.A. Hossain, J.Y. Zhu, J. Feldmann, A.A. Meharg, Increase in rice grain As for regions of Bangladesh irrigating paddies with elevated As in groundwaters, Environ. Sci. Technol. 40 (2006) 4903–4908.
[17] P.N. Williams, A. Raab, J. Feldmann, A.A. Meharg, 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. 39 (2007) 5531–5540.
[18] Y. Lu, E.E. Adomako, A.R.M. Solaiman, M.R. Islam, C. Deacon, P.N. Williams, G.K.M. Rahman, A.A. Meharg, Baseline soil variation is a major factor in As accumulation in Bengal delta paddy rice, Environ. Sci. Technol. 43 (2009) 1724–1729.
[19] X.Y. Xu, S.P. McGrath, A.A. Meharg, F.J. Zhao, Growing rice aerobically markedly decreases As accumulation, Environ. Sci. Technol. 42 (2008) 5574–5579.
[20] J.F. Ma, N. Yamaji, N. Mitani, X.Y. Xu, Y.H. Su, S.P. McGraph, F.J. Zhao, Transporters of arsenite in rice and their role in As accumulation in rice grain, Proc. Natl. Acad. Sci. U.S.A. 105 (2008) 9931–9935.
[21] Arsenic, drinking-water and health risks substitution in arsenic mitigation: a Discussion Paper; World Health Organization (WHO), 2003.
[22] F. Liang, Y. Li, G. Zhang, M. Tan, J. Lin, W. Liu, Y. Li, W. Lu, Total and speciated arsenic levels in rice from China, Food Addit. Contam. 27 (2010) 810–816.
[23] Joint FAO/WHO Expert Committee on Food Additives (JECFA), Evaluation of Certain Food Additives and Contaminants. Fifty-third Report of the Joint FAO/WHO Expert Committee on Food Additives. WHO Technical Report Series 896, WHO, Geneva, Switzerland, 2000.
[24] General Guidelines on Sampling, Codex Alimentarius Commission CAC-GL-50CAC, 2004.
[25] E.P. Nardi, F.S. Evangelista, L. Tormen, T.D. Saint'Pierre, A.J. Curtius, S.S. Souza, F. Barbosa, The use of inductively coupled plasma mass spectrometry (ICP-MS) for the determination of toxic and essential elements in different types of food samples, Food Chem. 112 (2009) 727–732.
[26] J.-H. Huang, G. Ilgen, P. Fecher, Quantitative chemical extraction for arsenic speciation in rice grains, J. Anal. At. Spectrom. (2010), doi:10.1039/c002306j.
[27] G.X. Sun, P.N. Williams, Y.G. Zhu, C. Deacon, A.M. Carey, A. Raab, J. Feldmann, A.A. Meharg, Survey of arsenic and its speciation in rice products such as breakfast cereals, rice crackers and Japanese rice condiments, Environ. Int. 35 (2009) 473–475.
[28] E. Sanz, R. Muñoz-Olivas, C. Cámara, Evaluation of a focused sonication probe for arsenic speciation in environmental and biological samples, J. Chromatogr. 1097 (2005) 1–8.
[29] Evaluation of certain food additives and contaminants; Fifty-third Report of the Joint FAO/WHO Expert Committee on Food Additives (JECFA); WHO technical report series 896: Geneva, 2000.
[30] Safety evaluation of certain food additives and contaminants; Sixty-first Meeting of the Joint FAO/WHO Expert Committee on Food Additives (JECFA); WHO Food Additives Series No. 52: Geneva, 2004.
[31] A.C.M. Igreja, L.P. Yokoyama, M.B. Rocha, J. Almeida, Culturas do arroz nos estados de Goiás e Tocantins: aspectos sócio-ecnômicos e tecnológicos nos sistemas de cultivo, Agric. São Paulo 42 (1995) 65–93.
[32] H.Y. Liu, A. Pribst, B.H. Liao, Metal contamination of soils and crops affected by Chenzhou lead/zinc mine spill (Hunan, China), Sci. Total Environ. 339 (2005) 153–166.
[33] Y.J. Zavala, R. Gerads, H. Gurleyuk, J.M. Duxbury, Arsenic in rice: II. Arsenic speciation in USA grain and implications for human health, Environ. Sci. Technol. 42 (2008) 3861–3866.
[34] M.J. Abedin, M.S. Cresser, A.A. Meharg, J. Feldmann, J. Cotter-Howells, Arsenic accumulation and metabolism in rice (*Oryza sativa* L.), Environ. Sci. Technol. 36 (2002) 962–968.
[35] M. Shri, S. Kumar, D. Chakrabarty, P.K. Trivedi, S. Mallick, P. Misra, D. Shukla, S. Mishra, S. Srivastava, R.D. Tripathi, R. Tuli, Effect of arsenic on growth, oxidative stress and antioxidant system in rice seedlings, Ecotoxicol. Environ. Saf. 72 (2009) 1102–1110.
[36] A.A. Carbonell-Barrachina, F.B. Carbonell, J.M. Beneyto, Arsenic uptake, distribution, and accumulation in tomato plants: effect of arsenite on plant growth and yield, J. Plant Nutr. 18 (1995) 1237–1250.
[37] F. Zhang, W. Shi, Z. Jin, Z. Shen, Response of antioxidative enzymes in cucumber chloroplasts to cadmium toxicity, J. Plant Nutr. 26 (2002) 1779–1788.
[38] X.Y. Xu, S.P. McGrath, F.J. Zhao, Rapid reduction of arsenate in the medium mediated by plant roots, New Phytol. 176 (2007) 590–599.
[39] A. Raab, H. Schat, A.A. Meharg, J. Feldmann, Uptake, translocation and transformation of arsenate and arsenite in sunflower (*Helianthus annuus*): formation of arsenic–phytochelatin complexes during exposure to high arsenic concentrations, New Phytol. 168 (2005) 551–558.
[40] A.A. Meharg, E. Lombi, P.N. Williams, K.G. Scheckel, J. Feldmann, A. Raab, Y. Zhu, R. Islam, Speciation and localization of arsenic in white and brown rice grains, Environ. Sci. Technol. 42 (2008) 1051–1057.
[41] A.K. Done, A.J. Peart, Acute toxicities of arsenical herbicides, Clin. Toxicol. 4 (1971) 343–355.
[42] B.L. Vallee, D.D. Ulmer, W.E. Wacker, Arsenic toxicology and biochemistry, AMA Arch. Ind. Health 21 (1960) 132–151.
[43] H.V. Aposhian, M.M. Aposhian, Arsenic toxicology: five questions, Chem. Res. Toxicol. 19 (2006) 1–15.
[44] S. Torres-Escribano, M. Leal, D. Vélez, R. Montoro, Total and inorganic arsenic concentrations in rice sold in Spain, effect of cooking, and risk assessments, Environ. Sci. Technol. 42 (2008) 3867–3872.
[45] Brazilian National Council of Environment, Resolução n° 20 do Conselho Nacional do Meio Ambiente (CONAMA). Public law 07/18/1986; http://www.mma.gov.br/port/conama/res/res86/res2086.html (accessed November 2010).
[46] Control of water quality. Brazilian Ministry of Health. Public law 1.469 29/12/2000; http://portal.saude.gov.br/portal/arquivos/pdf/portaria_1469.pdf (accessed November 2010).
[47] Guidelines for Drinking-water Quality: Incorporating the First and Second Addenda to Third Edition; World Health Organization (WHO): Geneva, 2008.
[48] P.N. Williams, S. Islam, R. Islam, M. Jahiruddin, E. Adomako, R.M. Soliaman, G.K.M.M. Rahman, Y. Lu, C. Deacon, Y.-G. Zhu, A.A. Meharg, Arsenic limits trace mineral nutrition (selenium, zinc, and nickel) in Bangladesh rice grain, Environ. Sci. Technol. 43 (2009) 8430–8436.
[49] E. Sanz, R. Muñoz-Olivas, C. Cámara, M.K. Sengupta, S. Ahamed, Arsenic speciation in rice, straw, soil, hair, and nails samples from the arsenic affected areas of middle and lower Ganga plain, J. Environ. Sci. Health Part A 42 (2007) 1695–1705.
[50] T. Narukawa, K. Inagaki, T. Kuroiwa, K. Chiba, The extraction and speciation of arsenic in rice flour by HPLC–ICP-MS, Talanta 77 (2008) 427–432.
[51] I. Pizarro, M. Gómez, C. Cámara, M.A. Palacios, Arsenic speciation in environmental and biological samples – extraction and stability studies, Anal. Chim. Acta 493 (2003) 85–98.
[52] J.L.G. Mar, L.H. Reyes, G.M.M. Rahman, H.M.S. Kingston, Simultaneous extraction of arsenic and selenium species from rice products by microwave-assisted enzymatic extraction and analysis by ion chromatography-inductively coupled plasma-mass spectrometry, J. Agric. Food Chem. 57 (2009) 3005–3013.
[53] U. Kohlmeyer, E. Jantzen, J. Kuballa, S. Jakubik, Benefits of high-resolution IC–ICP-MS for the routine analysis of inorganic and organic arsenic species in food products of marine and terrestrial origin, Anal. Bioanal. Chem. 377 (2003) 6–13.
[54] M. D'Amato, G. Forte, S. Caroli, Identification and quantification of major species of arsenic in rice, J. AOAC 87 (2004) 238–243.
[55] P.N. Williams, A.H. Price, A. Raab, S.A. Hossain, J. Feldmann, A.A. Meharg, Variation in arsenic speciation and concentration in paddy rice related to dietary exposure, Environ. Sci. Technol. 39 (2005) 5531–5540.
[56] D.T. Heitkemper, N.P. Vela, K.R. Stewart, C.S. Westphal, Determination of total and speciated arsenic in rice by ion chromatography and inductively coupled plasma mass spectrometry, J. Anal. At. Spectrom. 16 (2001) 299–306.