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LEVELS of ARSENIC in RICE: the EFFECTS of COOKING

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SUMMARY
Total arsenic and arsenic speciation was performed on rice that had undergone various forms of cooking. Basmati, long-grain, polished (white) and wholegrain (brown), as well as parboiled rice, were investigated. The effect of rinse washing, low volume (2.5:1 water:rice) and high volume (6:1 water:rice) cooking, as well as steaming, were investigated. Rinse washing was effective at removing circa. 10% of the total and inorganic arsenic from basmati rice, but was less effective for other rice types. While steaming reduced total and inorganic arsenic rice content, it did not do so consistently across all rice types investigated. Low volume water cooking did not remove arsenic. High volume water:rice cooking did effectively remove both total and inorganic arsenic for the long-grain and basmati rice (parboiled was not investigated in high volume cooking water experiment), by 35% and 45% for total and inorganic arsenic content, respectively, compared to uncooked (raw) rice.

To reduce arsenic content of cooked rice, specifically the inorganic component, rinse washing and high volume of cooking water are effective.

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ABBREVIATIONS

As_i	Inorganic arsenic
As_o	Organic arsenic
As_T	Total arsenic
CRM	Certified Reference Material
DMA	Dimethyl Arsinic Acid
FSA	Food Standards Agency
LC-ICP-MS	Liquid Chromatography - Inductively Coupled Plasma – Mass Spectrometry
ICP-MS	Inductively Coupled Plasma – Mass Spectrometry
MMA	Monomethyl Arsonic Acid
s.e.	Standard error of mean

1. INTRODUCTION

1.1. Background

Issues surrounding arsenic in rice where reviewed for the Food Standards Agency[1]. Summarising the findings of this review:

> Rice is unusual amongst terrestrial foods, particularly for staple crops, in that that it is grown anaerobically. Under anaerobic conditions arsenic in soil is converted readily to arsenite which is mobile, leading to arsenic in rice grain being around 10-fold higher than for other crops. This occurs in soils which have no or limited anthropogenic contamination. Rice grain arsenic levels elevated further when grown in soils subject to anthropogenic contamination such as: arsenical pesticide use, base and precious mining and smelting impacted soils, and contaminated water irrigated soils.
>
> Inorganic arsenic, a class 1 non-threshold carcinogen, and dimethyl arsinic acid (DMA) constitute the dominant arsenic species present in rice, while traces of monomethyl arsonic acid (MMA) are sometimes reported, as well as a residual fraction that is either not extractable or does not elute from the chromatographic column. Inorganic arsenic can constitute up to 90% of total arsenic present in grain, but on average accounts for around 50% of total grain arsenic.

That FSA report[1] identified a need to examine if the arsenic content from rice could be reduced by the manner in which the rice was cooked. A number of previous studies had suggested that rice cooking was important to the arsenic content of the cooked grain[2-8]. A number of these studies focus on how cooking technique may reduce rice arsenic content[2,3], while others focus on how arsenic in cooking water affects arsenic content of cooked rice[4-8].

1.2. Focus of current study

A number of recent reports have shown that rinsing rice before washing and then cooking the rice in a high water:rice ratio (6:1) and not allowing the water evaporate to dryness significantly reduced the arsenic burden of the rice[2,3], with one study suggesting that the arsenic was primarily lost as inorganic arsenic, specifically arsenite[2].

Previous studies on rice cooking[2-8] had not systematically looked at: (a) differences between wholegrain (brown) or polished (white) rice or (b) commonly used cooking techniques such as low and high water:rice volume and steaming. Similarly, systematic speciation and/or mass balances are inconsistent or absent between previous studies[3-8]. Par-boiled rice also needs to be considered due to its widespread utilization.

This current study sets out the systematic determination of the effect of cooking on the concentrations of arsenic species in rice.

2. EXPERIMENAL

2.1 Samples

2.1.1. Rice samples were purchased from major UK retailers. Two varieties of Basmati one wholegrain (packed in 1 kg portions, 4 portions were mixed before use) and one polished (packed in 2 kg portions, 2 portions, mixed before use) were of Indian origin. Wholegrain long-grain (4 times 1 kg portions, mixed before use) and polished long-grain (4 times 1 kg portions mixed before use) originated according to label from more than one country. The same origin was given for the long-grain easy cook (par-boiled) rice (2 times 2 kg portion, before use). The easy cook short-grain rice (4 times 1 kg portions mixed before use) was of Italian origin.

2.2. Rice treatment

All experiments were conducted with triplicate replication.

2.2.1. Rice rinse wash

Raw rice was first washed by placing 100 g portions of rice (packet weight) in an acid washed 800 ml beaker and then adding 600 ml of double distilled deionised (Milli-Q) water. The sample was allowed to sit for 3 minutes with routine agitation. The water was decanted and then the process repeated again with another 600 ml of water. The decanted water was then freeze dried. Dry weight determination was then made on both the raw and rinsed rice by oven drying at 80^0C until constant weight was reached. The quantity of freeze dried residue was recorded. Rinse washed rice was used in all subsequent cooking experiments.

2.2.2. Boiled rice

The quantity of packet weight used in all boiling experiments was 100 g. Double distilled deionised water was used for the cooking water. All rice, including par-boiled, was subject to 2.5:1 (low volume) water to rice (packet weight) cooking, where the water was cooked to dryness. All rice with the exception of par-boiled, were also subject to 6:1 (high volume) water:rice cooking, where the rice was cooked to eating texture. The residual water was drained off and then freeze dried. Cooked rice was then dried at 80^0C until constant weight was reached.

2.2.3. Steamed rice

Rinse washed rice (100 g packet weight) was soaked for 2 h in an acid washed 400 ml beaker with 200 mL of double distilled deionised (Milli-Q) water. On termination of soaking the water was decanted and freeze dried. The steamer was filled with 200 mL of double distilled deionised (Milli-Q) water and the soaked rice placed on arsenic and lint-free cotton-cloth. Steaming time was 2 times 15 min with stirring in between. Cooked rice was then dried at 80^0C until constant weight was reached.

2.2.4. Milling

All the oven dry samples for analysis were then milled using a coffee/spice grinder prior to analysis.

2.3. Total arsenic determination

2.3.1 Digestion

Total digestion of the samples was conducted by measuring out 0.5 g dry weight of sample into 50 ml polypropylene centrifuge tubes and adding 2.5 ml of Aristar nitric acid and 4 ml of hydrogen peroxide suprapur, followed by microwave digestion using a CEM Mars5 Microwave system. On digestion the sample was diluted to 25 ml using double distilled deionised water with Rhodium (0.02 mL 10 mg Rh/L) as internal standard.

2.3.2. CRM

CRM NIST 1568a rice powder was used throughout for the totals determination.

2.3.3 Analysis

Arsenic content was measured using an Agilent 7500c ICP-MS with hydrogen as collision/reaction gas. The ICP-MS operating conditions are given in Table A1.

2.4. Arsenic speciation

2.4.1. Extraction

Samples were extracted in 1 % Aristar nitric acid and 1% (vol/vol) hydrogen peroxide suprapur using a CEM Mars5 microwave system. The supernatant was used for determination of extractable arsenic and As-speciation. This oxidises arsenite to arsenate, improving chromatographic resolution as arsenate elutes at some distance to MMA and DMA, where arsenite elutes adjacent to MMA and DMA.

2.4.2. Analysis

Arsenic species were separated on a Hamilton PRP X 100 anion exchange column using phosphate buffer. The LC-system was a Agilent 1100 system directly coupled to the Agilent 7500c ICP-MS for arsenic determination.

2.4.3. Internal standard

Indium (0.01 mg/kg) in 1 % (v/v) nitric acid was added during the analysis via a T-piece as an internal standard.

2.4.4. CRM

CRM NIST 1568a rice powder was used throughout for speciation determinations. There is no CRM available for inorganic and organic arsenic in rice, but NIST 1568a has been used routinely in previous studies (listed in Appendix Table A2).

2.4.5. Standards

Solutions (0.1mL) containing known amounts of DMA (10 to 100 µg/kg) were subjected to LC-ICP-MS under the same conditions as the supernatants. Peak areas from these measurements were used to construct a calibration curve. Single species standards DMA, MMA and As(V) were used for identification of species by retention time.

2.4.6. Samples

The supernatants (0.1 mL) were used as they were and injected onto the column. Peak areas were used for quantification of As-species.

2.5. Quality control prerequisites

2.5.1. Total As determination

Every 10th sample was digested in duplicate and measured. Each analytical batch contained procedural blanks, spiked samples (for recovery estimate purposes) and CRM.

2.5.2. Spike recovery

Data were accepted if the spike recovery was between 80 and 120%.

2.5.3. Replicate agreement

Replicate values for a given sample must have a relative standard deviation of < 20%

2.5.4. Reference Materials

The reference material results for each batch should be within the certified range.

2.5.5. Limit of detection

The limit of detection is defined as three times the standard deviation of the signal from procedural blanks, corrected for sample weight and dilution.

3. RESULTS & DISCUSSION

3.1. QA/QC data for total analysis

The reported mean value and standard error for the CRM is 0.280 mg/kg ± 0.007 mg/kg (n=11) compared to its certified value of 0.29 mg/kg with a 95% confidence interval of ± 0.03 mg/kg, so the CRM recovery reported here is well within the 95% confidence interval. Spike recovery was 103.8 % ± 5.7 % (n=12). Limits of detection where 0.0004 mg/kg expressed on a sample weight basis.

3.2. QA/QC for speciation

Table A2 reports arsenic speciation of the rice flour CRM and compares the results of this study with those previously published in the literature as no cereal flour CRM has certified arsenic speciation reported for it. The results of this CRM analysis from the present study compare favourably with previously reported studies. Spike recoveries for arsenate and DMA are 110 % ± 6.2 % (n=5) and 103 % ± 4.3 % (n=5) respectively. Limits of detection for DMA are 0.004 mg/kg when expressed on a flour dry weight basis.

3.3. Data presentation

Summarised concentrations in raw, washed and cooked rice are presented Tables 1 and 2 respectively. A detailed breakdown of these data are provided in Table A3 for non-parboiled rice and Table A4 for parboiled rice were concentrations and percentage arsenic concentrations are also recorded for the cooking and soaking liquors. Mass balances, i.e. summation of the individual measured components with respect to the initial arsenic in raw rice, are also presented. The average mass balance for all the data ± the standard error was 100.8 ± 1.3 % (n=20).

3.4. Washing rice

There was variation in the effectiveness of rinse washing in removing total/inorganic arsenic from raw rice (Table 1 & 2). Washing removed more total arsenic for both the polished (to 87% of raw rice content) and wholegrain (to 85% of raw rice content) basmati, while for all other rice percentage arsenic remaining ranged only from 96-99% of raw rice content, including parboiled. It appears that rinse washing is more effective for basmati rice than for other types of rice, though more samples would need to be analysed to confirm this.

Virtually all the arsenic lost was inorganic while (91% on average of raw rice concentration) while negligible DMA was lost (99% on average of raw rice concentration).

3.5. Low water to rice volume cooking

3.5.1. Non-parboiled rice

Cooking rice to dryness in a 2.5:1 water:rice ratio resulted in no loss of arsenic from the cooked grain throughout for all four rice types.

3.5.2. Parboiled rice

Cooking rice to dryness in a 2.5:1 water:rice ratio resulted in no loss of arsenic from the cooked grain for both rice types.

3.6 High water to rice ratio

All rice types tested for high volume cooking (6:1 water to rice ration), that is all the non-parboiled types tested, considerably reduced both total and inorganic arsenic content. There was no reduction in organic arsenic content on high volume cooking.

Total arsenic content was reduced to 65% of raw rice content following rinsing and high volume cooking (Table 2), ranging from 55% in whole grain basmati to 72% in polished long-grain. This reduction was on average 55% for inorganic arsenic content, ranging from 51% for polished long-grain to 60% for polished basmati. Even though the rinse washing was ineffective for both types of the long-grain rice, high volume cooking water reduced inorganic arsenic contents to those of basmati rice where rinsing was more effective. This suggests that high volume cooking by itself is enough to reduce total and inorganic arsenic content, though rinse washing is normally recommended as part of the preparation of rice *per se*.

3.7 Steaming

Steaming did reduce total and inorganic arsenic content to 83% and 78% of the raw rice values, respectively. However, the effects were variable, ranging from 91% for wholegrain basmati to 75% for polished basmati for total arsenic. Percentage inorganic arsenic was reduced lower compared to total arsenic content, with inorganic concentrations ranging from 85% in polished long-grain to 60% in polished basmati. While steaming did reduce total and inorganic arsenic content it did not do so as effectively or as consistently as high volume cooking.

3.8 Comparison with literature

The most comparable to the present study, though more limited in cooking treatments and rice types, was a high water rice (6:1) investigation conducted by Mihucz et al.[2]. Two Hungarian and one Chinese rice types, for none of which was it recorded if the rice was wholegrain or polished, were used in that study. They found a 42-63% reduction in total arsenic in cooked rice, with the cooking liquor containing most of the removed arsenic from the rice (26-49%), while the quantity removed by rinse washing was less (8-17%). It was found that raw rice contained both arsenate and arsenite, and it was primarily arsenite that was removed from the rice on rinsing and boiling. Arsenite (As^{3+}) is uncharged at physiological pHs and hence more mobile than arsenate (As^{5+}) or DMA (As^{5+}), both of which are anionic. The DMA findings in that study[2] confirm our results. As the present report only records total inorganic arsenic, because the extraction process oxidises arsenite to arsenate (see section 2.4.1.), the observation that primarily arsenite was removed from the rice could not be confirmed.

In another comparable study, three West Bengali samples, where rice polishing state was not recorded, were rinse washed and then cooked in a large water volume[3]. Total arsenic, not speciation was determined. The rinse washing step was more exhaustive, involving 5-6 rinses until the rinse water discard was clear, the traditional Indian preparation, rather than double rinse wash step used in the experiments reported here. The rinse wash step removed 28% of the arsenic compared to raw rice. Combined rinse washing and large volume (6:1 water:rice) reduced arsenic up to 58% of raw rice content. This is compared to an average of 35% removal (maximum of 45%) or total arsenic reported in the current study (Table 2). The increased efficiency of removal for the Indian rice study of that study[3] may be due to more exhaustive rinse washing, or due to the intrinsic nature of the rice used in that study.

Other studies have been conducted on the effects of cooking rice on arsenic content, but have focused on the impact of arsenic contaminated cooking water on rice arsenic burdens[4-8]. While relevant to S.E. Asian and US scenarios where cooking and drinking water is arsenic contaminated, they are not relevant to the UK.

4. Conclusions and recommendations

It was found here that cooking rice in a large volume of water (6:1 water:rice) had the greatest effect with regards to lowering arsenic levels in cooked rice. Specifically, it preferentially reduced the inorganic arsenic content by 45% of that in the raw rice, when combined with rinse washing. It is recommended that to reduce total and inorganic arsenic content of rice that rice is rinse washed and cooked in a 6:1 water to rice ration. Exhaustive rinse washing, as practised in India, may reduce arsenic content even further when combined with large cooking water volume.

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Table 1. Summary of As_T (total) , As_i (arsenate and arsenite) and As_o (DMA and MMA) concentrations, in rice cooked in various ways. Data are the averages of 3 replicates. Numbers in italics are the standard deviation (s.e.) of the mean.

Rice type	Cooking technique	As_T (µg/kg)	s.e.	As_i (µg/kg)	s.e.	As_o (µg/kg)	s.e.
polished basmati	**raw**	162	*3*	93	*1*	18	*1*
	raw washed	141	*5*	86	*2*	19	*1*
	2.5:1 water to rice	141	*1*	90	*3*	18	*1*
	6:1 water to rice	103	*5*	56	*5*	18	*1*
	steamed	122	*8*	61	*2*	15	*2*
wholegrain basmati	**raw**	131	*8*	89	*3*	18	*1*
	raw washed	111	*3*	80	*1*	16	*3*
	2.5:1 water to rice	119	*3*	82	*1*	21	*2*
	6:1 water to rice	72	*3*	48	*2*	19	*1*
	steamed	119	*12*	76	*3*	22	*2*
polished long-grain	**raw**	229	*2*	138	*1*	58	*2*
	raw washed	222	*13*	131	*5*	59	*3*
	2.5:1 water to rice	238	*6*	144	*20*	50	*6*
	6:1 water to rice	165	*2*	70	*3*	53	*2*
	steamed	177	*4*	107	*2*	52	*1*
wholegrain long-grain	**raw**	314	*9*	183	*14*	87	*2*
	raw washed	311	*18*	157	*3*	86	*2*
	2.5:1 water to rice	324	*7*	165	*3*	109	*2*
	6:1 water to rice	219	*5*	102	*9*	87	*5*
	steamed	280	*5*	156	*24*	76	*6*
Italian parboiled	**raw**	211	*5*	157	*2*	54	*3*
	raw washed	203	*7*	149	*3*	54	*4*
	2.5:1 water to rice	211	*7*	157	*4*	54	*2*
long-grain parboiled	**raw**	186	*2*	115	*2*	56	*3*
	raw washed	180	*1*	99	*2*	57	*1*
	2.5:1 water to rice	163	*10*	86	*13*	39	*6*

Table 2. For rice cooked in various ways, summary of As_T (total) , As_i (arsenate and arsenite) and As_o (DMA and MMA), expressed as a percentage of As_T, As_i and As_o concentrations in the raw rice, respectively. Data are the averages of 3 replicates except for the overall average were the means of the individual rice types are used, i.e. n= 6 for raw washed and 2.5:1 water to rice, and n=4 for 6:1 water to rice and steamed. Numbers in italics are the standard errors (s.e.) of the mean.

Rice type	Cooking technique	As_T (%)	s.e.	As_i (%)	s.e.	As_o (%)	s.e.
polished basmati	**raw washed**	87	*3*	92	*2*	106	*3*
	2.5:1 water to rice	87	*3*	92	*2*	106	*3*
	6:1 water to rice	64	*3*	60	*6*	100	*3*
	steamed	75	*5*	66	*2*	83	*10*
wholegrain basmati	**raw washed**	85	*3*	90	*1*	89	*16*
	2.5:1 water to rice	85	*3*	90	*1*	89	*16*
	6:1 water to rice	55	*2*	54	*2*	106	*3*
	steamed	91	*9*	85	*4*	122	*13*
polished long-grain	**raw washed**	97	*6*	95	*3*	102	*5*
	2.5:1 water to rice	97	*6*	95	*3*	102	*5*
	6:1 water to rice	72	*1*	51	*2*	91	*3*
	steamed	77	*2*	78	*1*	90	*2*
wholegrain long-grain	**raw washed**	99	*6*	86	*2*	99	*2*
	2.5:1 water to rice	99	*6*	86	*2*	99	*2*
	6:1 water to rice	70	*2*	56	*5*	100	*6*
	steamed	89	*2*	85	*13*	87	*7*
Italian parboiled	**raw washed**	96	*3*	95	*2*	100	*7*
	2.5:1 water to rice	96	*3*	95	*2*	100	*7*
long-grain parboiled	**raw washed**	97	*1*	86	*2*	102	*2*
	2.5:1 water to rice	97	*1*	86	*2*	102	*2*
AVERAGE of ALL RICE TYPES	**raw washed**	**93**	***2***	**91**	***2***	**99**	***2***
	2.5:1 water to rice	**93**	***2***	**91**	***2***	**99**	***2***
	6:1 water to rice	**65**	***4***	**55**	***2***	**99**	***3***
	steamed	**83**	***4***	**78**	***5***	**96**	***9***

ANNEX

Table A1. ICP-MS setup

	Total As	**Speciation**
Instrument	Agilent 7500 c	Agilent 7500 c
ICP Power (W)	1570	1570
Nebuliser gas flow (L/min)	Optim. for maximum signal	
Lens setting	Optim. for maximum signal	
Nebuliser type	Meinhard	
Integration time (ms)	300 (75As), 100 (103 Rh, 78 Se)	500(75As), 100 (103 Rh, 77Se, 82 Se)
Reaction cell	Hydrogen	-
Reaction cell gas flow (mL/mmin)	2.5	-
Number of replicates	10	1
Scan mode Peak	Peak hopping	
LC column		Agilent 1100 Hamilton PRP x100 (150*4.5mm) + precolumn
buffer		10 mM phosphate buffer pH 6.0
Flow (mL/min)		0.8
Injected sample volume (mL)		0.1

Table A2. Performance of CRM speciation compared to previous studies. As_i refers to inorganic arsenic (arsenate and arsenite). As_o refers to inorganic arsenic (DMA and MMA). Numbers in italics are the standard error of the mean from the current study. Column recovery is the sum of species expressed as a percentage of total arsenic determined in that solution.

Extraction	**As_o (µg/kg)**	**As_i (µg/kg)**	**Σ of species (µg/kg)**	**Extraction efficiency (%)**	**Column recovery (%)**	**Reference**
2M TFA	180	87	267	95	96	4
Enzymatic digest, pepsin and pancreatin	159	101	260	*	*	4
2M TFA	182	92	274	112	84	9
2M TFA	162	80	240	*	*	10
Methanol:Water with sonication	180	109	288	99	*	11
Enzymatic hydrolysis, α-amylase	171	106	277	*	*	12
Ultrasonic & enzy. hydrol., protease & α-amylase	143	88	231	99	81	13
1 M H_3PO_4 with sonication	164	102	267	*	*	14
1% HNO_3	**185**	**99**	**284**	**104**	**98**	**this study**
	3	***2***	***4***	***1***	***1***	

Table A3. Arsenic speciation and total arsenic for rice cooked in various ways. Concentration data for rice is expressed per gram of rice analysed, while rinse wash and cooking liquor are expressed per gram of raw rice. As_T refers to total arsenic. As_i refers to inorganic arsenic (arsenate and arsenite). As_o refers to organic arsenic (DMA and MMA). "Extr. efficiency" refers to extraction efficiency, "recov." for recovery. Reported means are for the average of 3 replicates, with the exception of steaming cooking liquor were all 3 replicates were cooked together to give one liquor. Numbers in italics are the standard deviation of the mean. Mass balances were obtained by summing the rice with rinse wash and cooking liquor. Note all rice was rinse washed with the exception of raw rice. Percentage arsenic speciation is presented as percentage of sum of species determined chromatographically. Column recovery is the sum of species expressed as a percentage of total arsenic determined in extraction solution. Rinse wash and cooking liquors were freeze dried and extraction efficiency is expressed as a percentage extraction from that freeze dried material.

Part a. Polished basmati.

cooking treatment	rice or wash – cooking liquor	moisture loss on drying (%)	As_T (ng/g)	As_T, % of raw (%)	extr. effic. As_T (%)	column recov. (%)	As_i (ng/g)	As_o (ng/g)	As_i (%)	As_o (%)	mass balance (%)
raw	**rice**	10.5	162	-	71	97	93	18	84	16	-
		0.3	*6*	-	*5*	*2*	*2*	*1*	*1*	*1*	-
rinsed rice	**rice**	11.9	141	87	77	97	86	19	82	18	**98**
		0.2	*8*	*5*	*6*	*2*	*3*	*1*	*1*	*1*	***6***
	rinse wash	-	18	11	84	115	-	-	90	10	-
		-	*1*	*1*	*13*	*10*	-	-	*1*	*1*	-
2.5:1 water to rice	**rice**	12.2	141	87	72	96	90	18	92	18	**98**
		0.1	*2*	*1*	*1*	*4*	*5*	*1*	*1*	*1*	***2***
6:1 water to rice	**rice**	16.0	103	64	71	100	56	18	75	24	**98**
		0.6	*8*	*5*	*9*	*3*	*9*	*1*	*2*	*2*	***6***
	cooking water	-	38	23	89	91	-	-	95	5	-
		-	*2*	*1*	*3*	*9*	-	-	*2*	*2*	-
steaming	**rice**	15.6	122	75	54	106	61	15	80	20	**103**
		0.4	*14*	*8*	*3*	*7*	*4*	*3*	*2*	*2*	***3***
	soaking water	-	17	10	105	98	-	-	93	1	-
		-	*1*	*1*	*8*	*2*	-	-	*1*	*1*	-
	cooking liquor	-	1	0.6	98	99	-	-	85	15	-

Part b. Wholegrain basmati.

cooking treatment	rice or wash – cooking liquor	moisture loss on drying (%)	As_T (ng/g)	As_T, % of raw (%)	extr. effic. As_T (%)	column recov. (%)	As_i (ng/g)	As_o (ng/g)	As_i (%)	As_o (%)	mass balance (%)
raw	**rice**	10.2	131	-	83	99	89	18	83	17	**-**
		0.2	*13*	-	*6*	*2*	*5*	*1*	*1*	*1*	-
rinsed rice	**rice**	10.5	111	85	96	92	80	16	83	17	**88**
		0.2	*6*	*5*	*14*	*11*	*1*	*5*	*4*	*4*	***5***
	rinse wash	-	8	6	94	101	-	-	67	33	-
		-	*8*	*6*	*20*	*10*	-	-	*40*	*40*	-
2.5:1 water to rice	**rice**	11.3	119	91	86	101	82	21	80	20	**97**
		0.2	*5*	*4*	*9*	*3*	*2*	*4*	*2*	*2*	***5***
6:1 water to rice	**rice**	15.5	72	55	96	99	48	19	71	29	**96**
		0.7	*5*	*4*	*10*	*2*	*3*	*1*	*2*	*2*	***3***
	cooking water	-	45	34	91	98	-	-	93	7	-
		-	*4*	*3*	*7*	*6*	-	-	*2*	*2*	-
steaming	**rice**	15.3	119	91	75	111	76	22	78	22	**100**
		0.8	*21*	*16*	*7*	*3*	*6*	*4*	*2*	*2*	***19***
	soaking water	-	4	3	91	99	-	-	67	33	-
		-	*1*	*1*	*21*	*8*	-	-	*23*	*23*	-
	cooking liquor	-	0.3	0.2	114	108	-	-	88	12	-

Part c. Polished long-grain.

cooking treatment	rice or wash – cooking liquor	moisture loss on drying (%)	As_T (ng/g)	As_T, % of raw (%)	extr. effic. As_T (%)	column recov. (%)	As_i (ng/g)	As_o (ng/g)	As_i (%)	As_o (%)	mass balance (%)
raw	**rice**	10.9	229	-	79	109	138	58	71	29	-
		0.1	*3*	-	*2*	*1*	*1*	*3*	*1*	*1*	-
rinsed rice	**rice**	12.1	222	97	84	103	131	59	69	31	**103**
		0.1	*23*	*10*	*5*	*6*	*8*	*5*	*1*	*1*	***13***
	rinse wash	-	13	6	95	96	-	-	88	12	-
		-	*2*	*1*	*27*	*7*	-	-	*3*	*3*	-
2.5:1 water to rice	**rice**	12.9	238	104	78	104	144	50	73	27	**110**
		0.1	*10*	*4*	*7*	*10*	*35*	*10*	*8*	*8*	***5***
6:1 water to rice	**rice**	17.0	165	72	68	111	70	53	57	43	**113**
		1	*4*	*2*	*5*	*7*	*5*	*3*	*2*	*2*	***1***
	cooking water	-	82	36	83	95	-	-	92	8	-
		-	*5*	*2*	*1*	*2*	-	-	*1*	*1*	-
steaming	**rice**	17.7	177	77	77	116	107	52	67	33	**99**
		0.2	7	3	2	1	3	2	1	1	**4**
	soaking water	-	31	13	80	101	-	-	85	15	-
		-	*2*	*1*	*6*	*1*	-	-	*1*	*1*	-
	cooking liquor	-	7	3	22	102	-	-	95	5	-

Part d. Wholegrain long-grain.

cooking treatment	rice or wash – cooking liquor	moisture loss on drying (%)	As_T (ng/g)	As_T, % of raw (%)	extr. effic. As_T (%)	column recov. (%)	As_i (ng/g)	As_o (ng/g)	As_i (%)	As_o (%)	mass balance (%)
raw	**rice**	11.2	314	-	84	104	183	87	68	32	**-**
		0.1	*16*	-	*12*	*7*	*25*	*3*	*2*	*2*	-
rinsed rice	**rice**	12.1	311	99	74	107	157	86	65	35	**104**
		0.2	*31*	*10*	*8*	*1*	*5*	*3*	*1*	*1*	***11***
	rinse wash	-	15	5	105	106	-	-	92	8	**-**
		-	*5*	*2*	*3*	*18*	-	-	*4*	*4*	-
2.5:1 water to rice	**rice**	12.9	324	103	78	108	165	109	60	40	**108**
		0.2	*12*	*4*	*3*	*3*	*6*	*3*	*1*	*1*	***5***
6:1 water to rice	**rice**	17.7	219	70	91	95	102	87	54	46	**104**
		0.7	*9*	*3*	*2*	*7*	*16*	*9*	*1*	*1*	***3***
	cooking water	-	93	30	79	104	-	-	86	14	**-**
		-	11	4	7	3	-	-	2	2	-
steaming	**rice**	18.0	280	89	78	107	156	76	66	34	**102**
		0.5	9	3	5	18	42	10	8	8	**4**
	soaking water	-	24	8	96	97	-	-	74	26	**-**
		-	7	2	7	4	-	-	7	7	-
	cooking liquor	-	1	0.3	84	106	-	-	83	17	**-**

Table A4. Arsenic speciation and total arsenic for parboiled rice cooked in a water to rice ratio of 2.5:1. Concentration data for rice is expressed per gram of rice analysed, while rinse wash and cooking liquor are expressed per gram of raw rice.As_T refers to total arsenic. As_i refers to inorganic arsenic (arsenate and arsenite). As_o refers to organic arsenic (DMA and MMA). "Extr. efficiency" refers to extraction efficiency, "recov." for recovery. Reported means are for the average of 3 replicates. Numbers in italics are the standard deviation of the mean. Mass balances were obtained by summing the rice with rinse wash and cooking liquor. Note all rice was rinse washed with the exception of raw rice. Percentage arsenic speciation is presented as percentage of sum of species determined chromatographically. Column recovery is the sum of species expressed as a percentage of total arsenic determined in that solution. Column recovery is the sum of species expressed as a percentage of total arsenic determined in extraction solution. Rinse wash and cooking liquors were freeze dried and extraction efficiency is expressed as a percentage extraction from that freeze dried material.

rice type	cooking treatment	rice or wash – cooking liquor	moisture loss on drying (%)	As_T (ng/g)	As_T, % of raw (%)	extr. effic. As_T (%)	column recov. (%)	As_i (ng/g)	As_o (ng/g)	As_i (%)	As_o (%)	mass balance (%)
Italian	**raw**	**rice**	11.4	211	-	86	104	157	54	75	25	**-**
			0.1	*9*	-	*7*	*4*	*3*	*6*	*2*	*2*	-
	rinsed rice	**rice**	11.3	203	96	89	104	149	54	73	27	**100**
			0.1	*12*	*6*	*11*	*5*	*5*	*7*	*2*	*2*	***5***
		rinse wash	-	17	8	93	115	-	-	81	19	**-**
			-	*1*	*0.5*	*14*	*12*	-	-	*2*	*2*	-
	2.5:1 water to rice	**rice**	12.1	211	100	92	107	157	54	75	25	**97**
			0.5	*12*	*6*	*5*	*1*	*7*	*4*	*1*	*1*	***9***
long-grain	**raw**	**rice**	10.7	186	-	90	103	115	56	67	33	**-**
			0.4	*3*	-	*6*	*5*	*3*	*6*	*2*	*2*	-
	rinsed rice	**rice**	10.8	180	97	91	96	99	57	63	37	**104**
			0.1	*2*	*1*	*3*	*5*	*4*	*2*	*1*	*1*	***1***
		rinse wash	-	13	7	73	105	-	-	69	31	**-**
			-	*2*	*1*	*18*	*5*	-	-	*1*	*1*	-
	2.5:1 water to rice	**rice**	12.9	163	88	73	105	86	39	69	31	**95**
			0.2	*18*	*10*	*14*	*4*	*22*	*10*	*1*	*1*	***12***