



# Inorganic arsenic in rice-based products for infants and young children



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## ABSTRACT

Inorganic arsenic (As<sub>i</sub>) is a chronic, non-threshold carcinogen. Rice and rice-based products can be the major source of As<sub>i</sub> for many subpopulations. Baby rice, rice cereals and rice crackers are widely used to feed infants and young children. The As<sub>i</sub> concentration in rice-based products may pose a health risk for infants and young children. As<sub>i</sub> concentration was determined in rice-based products produced in the European Union and risk assessment associated with the consumption of these products by infants and young children, and compared to an identical US FDA survey. There are currently no European Union or United States of America regulations applicable to As<sub>i</sub> in food. However, this study suggests that the samples evaluated may introduce significant concentration of As<sub>i</sub> into infants' and young children's diets. Thus, there is an urgent need for regulatory limits on As<sub>i</sub> in food, especially for baby rice-based products.

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## 1. Introduction

Arsenic (As) is ubiquitous in the environment and occurs in different forms. As<sub>i</sub> is a non-threshold human carcinogen (IARC, 2004). Other than cancer, human exposure to As has been associated to diverse health problems (WHO, 2011a), which may be exacerbated with early-life exposure (Smith et al., 2006; Vahter, 2009). The main sources of human exposure to As are water and food (WHO, 2010). As<sub>i</sub> in water is tightly regulated (Council Directive, 1998; WHO, 2011b), however, there is no European Union (EU) or United States of America (USA) standard for As<sub>i</sub> in food products despite the fact that food sources dominate exposure, especially rice and rice-based products (EFSA, 2009; US FDA, 2014). Indeed, it has been demonstrated that consumption of rice and/or rice-based products increases the occurrence of As species in human urine (Cascio, Raab, Jenkins, Felman, & Meharg, 2011; Gilbert-Diamond et al., 2011; Wei, Zhu, & Hguyen, 2014). Furthermore, a cohort study in West Bengal, India, has reported elevated genotoxic effects, as measured by micronuclei in urothelial cells, associated with the staple consumption of cooked rice with >200 µg As/kg (Banerjee et al., 2013). Recently, the JECFA proposed a maximum level of 0.2 mg/kg of As<sub>i</sub> in polished rice (JECFA, 2014). The European Food Safety Authority (EFSA) has reviewed the diet of the European Union population and has recommend that dietary exposure to As<sub>i</sub> should be reduced. Cereal and cereal-based products have been identified as contributors to daily As<sub>i</sub> exposure in the general European population and young

children (<3 years of age) have been categorised as the most exposed to As<sub>i</sub> (EFSA, 2009). The review was mainly based on data reported as total As and a number of assumptions to estimate the As<sub>i</sub> exposure were made. It was highlighted that more speciation data for different food commodities are required to support a comprehensive dietary exposure assessment and to redefine risk assessment of As<sub>i</sub>, especially to high exposure risk subpopulations. The JECFA also reviewed and stated that dietary exposure to As<sub>i</sub> should be reduced. In addition, the Provisional Tolerable Weekly Intake (PTWI) was withdrawn since cancer of the lung and urinary bladder in addition to skin and a range of adverse effects were reported at As<sub>i</sub> exposure lower than those reviewed at the time the PTWI was established (JECFA, 2010).

Rice accumulates significantly higher levels of As<sub>i</sub> from soil and water than other crops due to anaerobic paddy soil culture, which renders As<sub>i</sub> highly available for plant uptake, leading to ~10-fold higher concentrations in grain compared to aerobically grown grains such as wheat or barley (Meharg & Zhao, 2012; William et al., 2007; Xu, McGrath, Meharg, & Zhao, 2008). Levels of As<sub>i</sub> in rice are also of concern due to the fact that there is a high gut bio-availability of rice As<sub>i</sub> (Juhasz et al., 2006; Signes-Pastor, Al-Rmalli, Jenkins, Carbonell-Barrachina, & Haris Parvez, 2012). Rice-based products also have high levels of As<sub>i</sub> and show a positive correlation between rice content and their As concentration (Burlo, Ramirez-Gandolfo, Signes-Pastor, Haris, & Carbonell-Barrachina, 2012; Carbonell-Barrachina et al., 2012). Rice-based products are widely used during weaning and to feed young children due to its availability, bland taste, nutritional value and relatively low allergic potential (Da Sacco, Baldassarre, & Massotti, 2013; Meharg et al., 2008; Mennella, Ziegler, Briefel, & Novak, 2006).

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Consumption of rice and rice-based foods is particularly high for infants and young children affected by celiac disease, which is a common condition that affects 1% of children in the EU and USA, while their only viable treatment is a gluten-free diet (Da Sacco et al., 2013; Munera-Picazo, Burló, & Carbonell-Barrachina, 2014; Newton & Singer, 2012). Furthermore, the fact that infants and young children have higher food consumption rates on body weight basis than adults, *circa* 3-fold, increases their exposure to As<sub>i</sub> with respect to adults for any given item of food, which exacerbates toxicological issues for this subpopulation (EFSA, 2009; Meharg & Zhao, 2012).

Information on As speciation data of rice-based products consumed by infants and young children is needed to define risk assessment of As<sub>i</sub> for one of the most vulnerable subpopulations, and to set regulations for As<sub>i</sub> content in food to protect them. In this study, therefore, As speciation was measured in 29 commercial baby rice, 53 commercial rice cereals and 97 commercial rice crackers from the EU market, and compared to 85 commercial baby rice, 105 rice cereals and 199 rice crackers included in the US FDA survey on As<sub>i</sub> in rice and rice-based products (US FDA, 2014), and the findings put in context of exposure risks to infants and young children.

## 2. Materials and methods

Samples of baby rice ( $n=29$ ), rice cereals ( $n=53$ ) and rice crackers ( $n=97$ ) belonging to 21 different and most popular commercial brands or manufacturers were purchased from 36 food shops (15 local shops and 21 big supermarkets) in the United Kingdom (UK). Duplicate samples of the same product and brand were always purchased from different stores. The rice-based products sampled showed use by date between February 2014 and March 2016.

### 2.1. Sample preparation for As speciation

All samples of rice-based products were freeze-dried, and then powdered using a Retch PM100 rotary ball-mill using a zirconium oxide lined vessel and zirconium oxide grinding balls. The powdered samples were weighed accurately to a weight of 0.1 g into 50 ml polypropylene centrifuge tubes to which 10 ml of 1% conc. Aristar nitric acid was added and allowed to sit overnight. Batches of 30 samples approximately were prepared which also include a blank and rice CRM (NIST 1568b Rice flour) which has the As species As<sub>i</sub>, dimethylarsinic acid (DMA) and monomethylarsonic acid (MMA) concentrations certified, then microwave digested in an CEM MARS 6 instrument for 30 min. at 95 °C using a 3 stage slow heating program: to 55 °C in 5 min. held for 10 min., to 75 °C in 5 min., held for 10 min. to 95 °C in 5 min., held for 30 min. A 1 ml aliquot was transferred to a 2 ml polypropylene vial and 10 µl of analytical grade hydrogen peroxide was added to convert any arsenite to arsenate to facilitate subsequent chromatographic detection.

### 2.2. Chemical analysis

To speciate As in rice-based products the diluted 1% nitric acid digested sample solutions were run on a Thermo Scientific IC5000 Ion Chromatography (IC) system, with an Thermo AS7, 2 × 250 mm column (and a Thermo AG7, 2 × 50 mm guard column) and a gradient mobile phase (A: 20 mM Ammonium Carbonate, B 200 mM Ammonium Carbonate – Starting at 100% A, changing to 100% B, in a linear gradient over 15 min.) interfaced with a Thermo ICAP Q ICP-MS that monitored  $m/z^+$  75, using He gas in collision cell mode. The resulting chromatogram was

compared with that for authentic standards; DMA, As<sub>i</sub>, MMA, tetratmethyl arsonium (TETRA) and arsenobetaine (AB). As present under each chromatographic peak was calibrated using a DMA concentration series.

### 2.3. Statistical analyses

Data were subjected to general lineal model (GLM) and the Duncan multiple range test to determine significant differences among samples. The statistical analyses were performed using IBM SPSS Statistic version 21.0.

## 3. Results and discussion

The analysis of the As speciated CRM gave excellent recovery results (mean ± SE), based on  $n=15$ , with  $105 \pm 3\%$  recovery for DMA,  $93.4 \pm 5\%$  for MMA and  $94.5 \pm 2\%$  recovery for As<sub>i</sub>. The CRM had a certified concentration of 0.182, 0.012 and 0.092 mg/kg As for DMA, MMA and As<sub>i</sub>, respectively. The limits of detection (LOD) for both DMA and As<sub>i</sub> (calculated from a DMA calibration) was 0.003 mg/kg rice d.wt.,  $n=6$ . All samples presented were above LOD for As<sub>i</sub> and only a few samples were below LOD for DMA and MMA, and in this case 1/2 LOD was used in statistical analysis of the data.

### 3.1. Baby rice

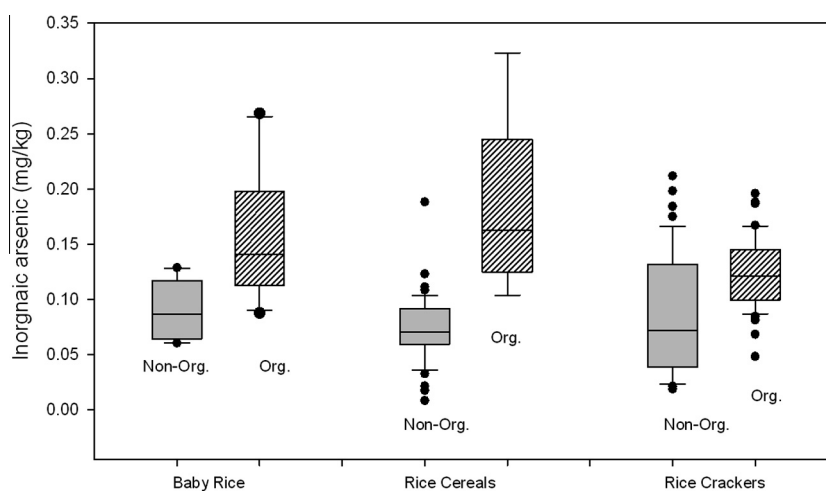
The summation of As species concentration ( $\Sigma$ As) in 29 baby rice samples belonging to 5 different commercial brands ranged from 0.063 to 0.334 mg/kg (Table 1). The As<sub>i</sub> percentage ranged from 51.4% to 84.6% of the  $\Sigma$ As species concentration. The baby rice brands M001, M003 and M005 had a significantly higher percentage of As<sub>i</sub> (71.6%, 73.4% and 79.3%, respectively) than M002 and M004 (66.3% and 53.5%, respectively;  $p < 0.001$ ). The As<sub>i</sub> concentration ranged from 0.056 to 0.268 mg/kg. This shows that 14% of the baby rice samples evaluated would be above the JECFA proposed As<sub>i</sub> maximum level for rice (0.200 mg/kg). The baby rice brand M005 had the highest As<sub>i</sub> concentration (median of 0.190 mg/kg;  $p < 0.001$ ). DMA ranged from 0.030 to 0.123 mg/kg (Table 1). The same trend of As speciation has been previously described in rice (Meharg et al., 2009; Torres-Escribano, Leal, Vélez, & Montoro, 2008). Baby rice samples labelled as produced under organic standard showed higher levels of As<sub>i</sub> than non-organic samples (Fig. 1), which was associated with the inclusion of whole grain rice. In fact, organic baby rice samples including whole grain contain a higher concentration and percentage of As<sub>i</sub> (median of 0.190 mg/kg and 79.3%) than those manufactured with milled rice (median of 0.121 mg/kg and 55.1%;  $p < 0.001$ ) (Table 2). This is in agreement with previous studies that show that brown rice contains higher proportion of As<sub>i</sub> than white rice, which is mainly concentrated at the surface of the whole grain, in the region corresponding to the pericarp and aleurone layer (Choi et al., 2014; Meharg et al., 2008). This is of particular concern as organic products are usually associated with a healthier and more nutritious option that is increasing production of organic baby food and may exacerbate As<sub>i</sub> exposure and health risk for infants and young children (Da Sacco et al., 2013).

In September 2013 the US Food and Drug Administration (US FDA) released analytical results of As<sub>i</sub> in approximately 1100 samples of rice and rice products from the US market, which were in addition to approximately 200 samples of rice and rice products initially tested, the results of which were released in September 2012 (USFDA, 2013). These results include 85 samples of baby rice (product subcategory: infant cereals and toddler cereals). The median and range of As<sub>i</sub> in baby rice from the USA market is 0.114

**Table 1**  
Concentration (mg/kg d.wt.) of As species in rice-based products (baby rice, rice cereals and rice crackers) ordered by food category and manufacturer.

| Food category | Manufacture | n  | As <sub>i</sub> (mg/kg d.wt.)                  | DMA (mg/kg d.wt.)                 | MMA (mg/kg d.wt.)   | Σ As species (mg/kg d.wt.)         | As <sub>i</sub> (%)            |
|---------------|-------------|----|--|-----------------------------------|---------------------|------------------------------------|--------------------------------|
| Baby rice     | M001        | 4  | 0.093 <sup>bc</sup> (0.088–0.097) <sup>A</sup> | 0.037 <sup>c</sup> (0.024–0.041)  | 0.001 (0.001–0.001) | 0.131 <sup>b</sup> (0.112–0.137)   | 71.6 <sup>a</sup> (70.4–78.5)  |
|               | M002        | 7  | 0.066 <sup>c</sup> (0.056–0.092)               | 0.035 <sup>c</sup> (0.030–0.061)  | 0.001 (0.001–0.003) | 0.099 <sup>b</sup> (0.063–0.090)   | 66.3 <sup>b</sup> (59.9–69.8)  |
|               | M003        | 4  | 0.118 <sup>bc</sup> (0.062–0.129)              | 0.041 <sup>c</sup> (0.023–0.046)  | 0.001 (0.001–0.001) | 0.160 <sup>b</sup> (0.085–0.175)   | 73.4 <sup>a</sup> (73.2–74.3)  |
|               | M004        | 5  | 0.131 <sup>b</sup> (0.122–0.142)               | 0.112 <sup>a</sup> (0.107–0.120)  | 0.001 (0.001–0.003) | 0.247 <sup>a</sup> (0.230–0.263)   | 53.5 <sup>c</sup> (51.4–55.1)  |
|               | M005        | 9  | 0.190 <sup>a</sup> (0.117–0.268)               | 0.057 <sup>b</sup> (0.038–0.123)  | 0.001 (0.001–0.004) | 0.264 <sup>a</sup> (0.181–0.334)   | 79.3 <sup>a</sup> (57.2–84.6)  |
| p-Value       |             |    | <0.001   | <0.001                            | 0.342               | <0.001                             | <0.001                         |
| Rice cereals  | M006        | 5  | 0.068 <sup>c</sup> (0.066–0.123)               | 0.028 <sup>ab</sup> (0.024–0.037) | 0.001 (0.001–0.001) | 0.097 <sup>bc</sup> (0.920–0.160)  | 74.2 <sup>ab</sup> (70.6–77.8) |
|               | M007        | 3  | 0.162 <sup>b</sup> (0.104–0.167)               | 0.037 <sup>ab</sup> (0.016–0.052) | 0.001 (0.001–0.001) | 0.204 <sup>b</sup> (0.122–0.214)   | 81.5 <sup>a</sup> (75.7–85.2)  |
|               | M008        | 6  | 0.074 <sup>c</sup> (0.044–0.097)               | 0.022 <sup>b</sup> (0.005–0.035)  | 0.001 (0.001–0.001) | 0.098 <sup>c</sup> (0.049–0.132)   | 76.3 <sup>a</sup> (72.3–89.6)  |
|               | M009        | 2  | 0.234 <sup>a</sup> (0.146–0.323)               | 0.047 <sup>a</sup> (0.026–0.069)  | 0.002 (0.001–0.003) | 0.284 <sup>a</sup> (0.173–0.396)   | 83.1 <sup>a</sup> (81.7–84.5)  |
|               | M010        | 30 | 0.075 <sup>c</sup> (0.008–0.188)               | 0.045 <sup>a</sup> (0.013–0.082)  | 0.001 (0.001–0.004) | 0.120 <sup>bc</sup> (0.057–0.249)  | 62.7 <sup>b</sup> (14.2–81.6)  |
| p-Value       |             |    | <0.001   | 0.002                             | 0.422               | <0.001                             | 0.001                          |
| Rice crackers | M006        | 2  | 0.132 <sup>bc</sup> (0.128–0.136)              | 0.021 (0.021–0.022)               | 0.001 (0.001–0.001) | 0.154 <sup>bc</sup> (0.150–0.158)  | 85.7 (85.1–86.3)               |
|               | M012        | 3  | 0.140 <sup>b</sup> (0.099–0.196)               | 0.042 (0.035–0.049)               | 0.001 (0.001–0.003) | 0.175 <sup>ab</sup> (0.141–0.249)  | 78.7 (70.0–80.2)               |
|               | M001        | 6  | 0.127 <sup>b</sup> (0.110–0.162)               | 0.023 (0.014–0.029)               | 0.001 (0.001–0.001) | 0.150 <sup>bc</sup> (0.124–0.190)  | 85.2 (83.3–88.6)               |
|               | M013        | 4  | 0.099 <sup>bcd</sup> (0.032–0.160)             | 0.016 (0.006–0.047)               | 0.001 (0.001–0.001) | 0.115 <sup>bcd</sup> (0.039–0.207) | 83.2 (77.3–89.4)               |
|               | M014        | 9  | 0.125 <sup>bc</sup> (0.103–0.187)              | 0.028 (0.023–0.032)               | 0.001 (0.001–0.002) | 0.157 <sup>bc</sup> (0.131–0.220)  | 80.7 (72.1–85.0)               |
|               | M015        | 3  | 0.095 <sup>bcd</sup> (0.082–0.106)             | 0.035 (0.026–0.038)               | 0.001 (0.001–0.001) | 0.133 <sup>bcd</sup> (0.107–0.144) | 73.6 (71.6–75.9)               |
|               | M009        | 19 | 0.101 <sup>bcd</sup> (0.035–0.184)             | 0.026 (0.011–0.172)               | 0.001 (0.001–0.006) | 0.130 <sup>bc</sup> (0.046–0.328)  | 79.4 (44.8–85.1)               |
|               | M016        | 3  | 0.070 <sup>cde</sup> (0.063–0.088)             | 0.017 (0.015–0.020)               | 0.001 (0.001–0.001) | 0.085 <sup>cd</sup> (0.081–0.109)  | 81.2 (78.2–82.1)               |
|               | M017        | 4  | 0.067 <sup>bcd</sup> (0.057–0.175)             | 0.020 (0.013–0.037)               | 0.001 (0.001–0.001) | 0.088 <sup>bcd</sup> (0.076–0.215) | 78.4 (71.1–82.8)               |
|               | M005        | 25 | 0.131 <sup>bc</sup> (0.068–0.188)              | 0.026 (0.004–0.056)               | 0.001 (0.001–0.001) | 0.164 <sup>bc</sup> (0.074–0.244)  | 85.0 (73.6–95.0)               |
|               | M018        | 11 | 0.027 <sup>e</sup> (0.019–0.050)               | 0.012 (0.001–0.045)               | 0.001 (0.001–0.001) | 0.034 <sup>d</sup> (0.019–0.095)   | 71.7 (52.3–100)                |
|               | M019        | 2  | 0.205 <sup>a</sup> (0.198–0.212)               | 0.040 (0.033–0.047)               | 0.001 (0.001–0.001) | 0.245 <sup>a</sup> (0.245–0.246)   | 83.6 (80.6–86.6)               |
|               | M020        | 2  | 0.065 <sup>de</sup> (0.060–0.072)              | 0.012 (0.004–0.021)               | 0.001 (0.001–0.001) | 0.078 <sup>cd</sup> (0.064–0.092)  | 85.4 (77.3–93.6)               |
|               | M011        | 2  | 0.130 <sup>bc</sup> (0.121–0.140)              | 0.022 (0.022–0.023)               | 0.001 (0.001–0.001) | 0.152 <sup>bc</sup> (0.143–0.163)  | 85.2 (84.8–85.6)               |
|               | M021        | 2  | 0.109 <sup>bcd</sup> (0.091–0.127)             | 0.021 (0.021–0.022)               | 0.001 (0.001–0.001) | 0.130 <sup>bcd</sup> (0.113–0.148) | 83.4 (80.7–86.1)               |
| p-Value       |             |    | <0.001   | 0.230                             | 0.700               | <0.001                             | 0.104                          |

<sup>A</sup> Median (max–min); values with the same letters were not significantly different at *p*-value < 0.05 for the variable studied (Duncan's multiple range test).



**Fig. 1.** Boxplot with lines representing median, 25th and 75th percentiles, whiskers 10th and 90th percentiles and dots outliers for As<sub>i</sub> in baby rice, rice cereals and rice crackers samples. Boxplot with open bars and diagonal hashed bars show As<sub>i</sub> concentration in the rice-based products (baby rice, rice cereals and rice crackers) produced under non-organic (Non-Org.) and organic standards (Org.), respectively.

(0.039–0.254) mg/kg. As<sub>i</sub> represents a median of 65.5% of ΣAs species concentration. Our results are similar to these and no significant differences were found in the As<sub>i</sub>, DMA, ΣAs species concentration or As<sub>i</sub> percentage (Table 3). Our results are also similar to the study carried out by Meharg et al., 2008, who analysed 17 samples of baby rice bought in the city of Aberdeen, UK, and reported a median As<sub>i</sub> content of 0.110 mg/kg. However, our results show a wider range of As<sub>i</sub> (0.056–0.268 mg/kg) than this study (0.060–0.160 mg/kg).

The FAO/WHO Expert Committee on Food Additives has determined the lower limit of the benchmark dose confidence limit (BMDL) based on a 0.5% increase in the incidence of lung cancer

to be 3.0 µg/kg b.wt. per day (WHO, 2011a). The Contaminants in Food Chain Panel (CONTAM) of EFSA determined the BMDL 1% extra risk for lung, skin and bladder cancers and skin lesions to be in the range of 0.3–8 µg/kg b.wt. per day and recommended using this instead of a single reference point in the risk characterisation for As<sub>i</sub> intake (EFSA, 2009). The EFSA (2009) report stated that As<sub>i</sub> exposure from food and water ranges from 0.13 to 0.56 µg/kg b.wt. for average consumers in 19 European countries. This estimated dietary exposure is within the range of the proposed BMDL values, indicating that the risk of toxicity cannot be excluded, especially for children whose dietary exposure has been estimated to be from 2 to 3-fold (0.50–2.66 µg/kg b.wt. per day)

**Table 2**

Effect of production under organic or non-organic standards on the concentration (mg/kg d.wt.) of As species in rice-based products (baby rice, rice cereals and rice crackers).

| Food Category | Production method | Type of rice    | n  | As <sub>i</sub> (mg/kg)                       | DMA (mg/kg)         | MMA (mg/kg)         | Σ As species (mg/kg)             | As <sub>i</sub> (%)            |
|---------------|-------------------|-----------------|----|---|---------------------|---------------------|----------------------------------|--------------------------------|
| Baby rice     | Non-organic       | Milled          | 11 | 0.082 <sup>b</sup> (0.056–0.128) <sup>A</sup> | 0.038 (0.022–0.061) | 0.001 (0.001–0.003) | 0.126 <sup>c</sup> (0.084–0.175) | 67.1 <sup>ab</sup> (59.9–74.3) |
|               |                   | Whole grain     | 0  | –   | –                   | –                   | –                                | –                              |
|               | Organic           | Milled          | 9  | 0.121 <sup>b</sup> (0.087–0.141)              | 0.107 (0.024–0.120) | 0.001 (0.001–0.003) | 0.230 <sup>b</sup> (0.111–0.262) | 55.1 <sup>b</sup> (51.4–78.5)  |
|               |                   | Whole grain     | 9  | 0.190 <sup>a</sup> (0.117–0.268)              | 0.057 (0.038–0.123) | 0.001 (0.001–0.004) | 0.264 <sup>a</sup> (0.180–0.334) | 79.3 <sup>a</sup> (57.2–84.6)  |
|               |                   | <i>p</i> -Value |    | <0.001  | 0.014               | 0.302               | <0.001                           | <0.001                         |
| Rice cereals  | Non-organic       | Milled          | 48 | 0.070 <sup>b</sup> (0.008–0.188)              | 0.036 (0.005–0.081) | 0.001 (0.001–0.004) | 0.113 <sup>b</sup> (0.042–0.248) | 69.6 (14.2–89.6)               |
|               |                   | Whole grain     | 0  | –   | –                   | –                   | –                                | –                              |
|               | Organic           | Milled          | 0  | –   | –                   | –                   | –                                | –                              |
|               |                   | Whole grain     | 5  | 0.162 <sup>a</sup> (0.103–0.323)              | 0.037 (0.016–0.068) | 0.001 (0.001–0.003) | 0.204 <sup>a</sup> (0.121–0.395) | 81.6 (75.7–85.2)               |
|               |                   | <i>p</i> -Value |    | <0.001  | 0.828               | 0.442               | <0.001                           | 0.025                          |
| Rice crackers | Non-organic       | Milled          | 23 | 0.071 <sup>b</sup> (0.021–0.159)              | 0.023 (0.004–0.046) | 0.001 (0.001–0.002) | 0.095 <sup>b</sup> (0.034–0.206) | 79.8 (52.3–93.6)               |
|               |                   | Whole grain     | 22 | 0.081 <sup>b</sup> (0.018–0.211)              | 0.018 (0.001–0.048) | 0.001 (0.001–0.002) | 0.098 <sup>b</sup> (0.018–0.245) | 81.3 (66.0–100.0)              |
|               | Organic           | Milled          | 0  | –   | –                   | –                   | –                                | –                              |
|               |                   | Whole grain     | 52 | 0.121 <sup>a</sup> (0.048–0.195)              | 0.026 (0.004–0.171) | 0.001 (0.001–0.006) | 0.150 <sup>a</sup> (0.06–0.328)  | 82.5 (44.8–95.0)               |
|               |                   | <i>p</i> -Value |    | <0.001  | 0.040               | 0.576               | <0.001                           | 0.153                          |

<sup>A</sup> Median (max–min); values with the same letters were not significantly different at *p*-value < 0.05 for the variable studied (Duncan's multiple range test).

**Table 3**

Total As concentration (mg/kg) and concentration (mg/kg) of As species in rice-based products (baby rice, rice cereals and rice crackers) from USA (FDA survey). Concentration (mg/kg) of As species in rice-based products (baby rice, rice cereals and rice crackers) from EU (Present study).

| Product category | Sampling place  | n   | As <sub>i</sub> (mg/kg)          | DMA (mg/kg)                      | MMA (mg/kg)                      | Σ As species (mg/kg) | Total As (mg/kg)    | As <sub>i</sub> (%)            |
|------------------|-----------------|-----|----------------------------------|----------------------------------|----------------------------------|----------------------|---------------------|--------------------------------|
| Baby rice        | US              | 85  | 0.114 (0.039–0.254) <sup>A</sup> | 0.071 (0.015–0.204)              | 0.003a (0.001–0.012)             | 0.215 (0.550–0.341)  | 0.200 (0.060–0.373) | 65.5 (22.0–82.9)               |
|                  | EU              | 29  | 0.121 (0.056–0.268)              | 0.041 (0.023–0.123)              | 0.001 <sup>b</sup> (0.001–0.004) | 0.175 (0.084–0.334)  | –                   | 69.8 (51.4–84.6)               |
|                  | <i>p</i> -Value |     | 0.235                            | 0.085                            | 0.002                            | 0.676                | –                   | 0.028                          |
| Rice cereals     | US              | 105 | 0.091 (0.023–0.283)              | 0.042 <sup>a</sup> (0.007–0.493) | 0.001 (0.001–0.014)              | 0.135 (0.041–0.625)  | 0.132 (0.050–0.723) | 63.0 (18.8–87.7)               |
|                  | EU              | 53  | 0.075 (0.008–0.323)              | 0.037 <sup>b</sup> (0.005–0.081) | 0.001 (0.001–0.004)              | 0.119 (0.042–0.396)  | –                   | 70.6 (14.2–89.6)               |
|                  | <i>p</i> -Value |     | 0.096                            | 0.004                            | 0.590                            | 0.005                | –                   | 0.077                          |
| Rice crackers    | US              | 199 | 0.079 (0.008–0.273)              | 0.034 <sup>a</sup> (0.001–0.477) | 0.001 <sup>a</sup> (0.001–0.021) | 0.116 (0.012–0.657)  | 0.121 (0.009–1.931) | 65.1 <sup>b</sup> (18.4–97.6)  |
|                  | EU              | 97  | 0.111 (0.018–0.211)              | 0.025 <sup>b</sup> (0.001–0.172) | 0.001 <sup>b</sup> (0.001–0.006) | 0.141 (0.019–0.328)  | –                   | 80.9 <sup>a</sup> (44.8–100.0) |
|                  | <i>p</i> -Value |     | 0.011                            | <0.001                           | <0.001                           | 0.224                | –                   | <0.001                         |

<sup>A</sup> Median (max–min); values with the same letters were not significantly different at *p*-value < 0.05 for the variable studied.

that of adults due to their greater food consumption in relation to their body weight (EFSA, 2009).

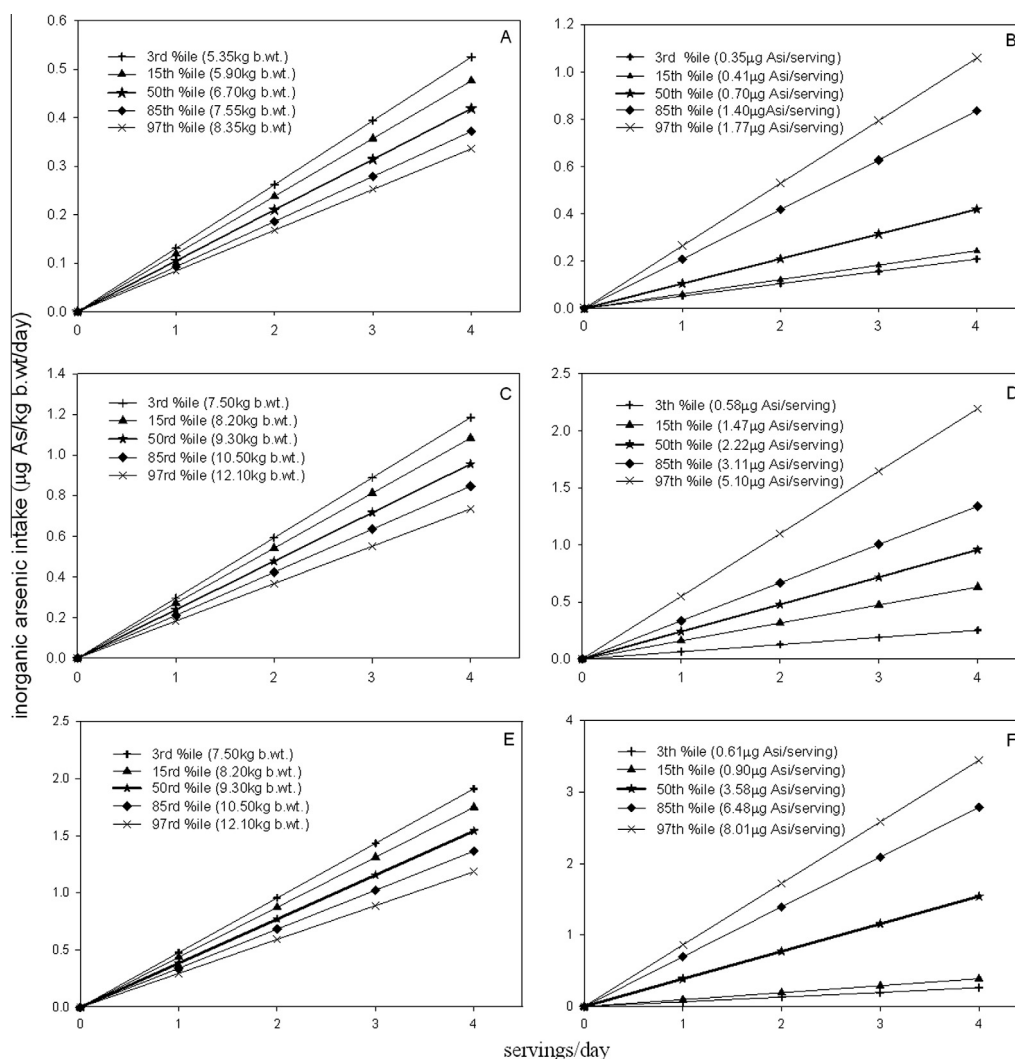
The plots A and B, included in Fig. 2, show the As<sub>i</sub> exposure for infants (4 months of age) consuming baby rice. The median of 0.70 µg As<sub>i</sub>/serving (Table 4) and the 3<sup>th</sup> (5.35 kg), 15<sup>th</sup> (5.90 kg), 50<sup>th</sup> (6.70 kg), 85<sup>th</sup> (7.55 kg) and 97<sup>th</sup> (8.35 kg) percentiles of body weight (WHO, 2014) have been used for calculations in the plot A. The median of 6.70 kg b.wt. and the 3<sup>th</sup> (0.35 µg As<sub>i</sub>/serving), 15<sup>th</sup> (0.41 µg As<sub>i</sub>/serving), 50<sup>th</sup> (0.70 µg As<sub>i</sub>/serving) 85<sup>th</sup> (1.40 µg As<sub>i</sub>/serving) and 97<sup>th</sup> (1.77 µg As<sub>i</sub>/serving) percentiles of As<sub>i</sub> concentration per serving have been used for calculations in the plot B. When the median body weight and the median As<sub>i</sub> concentration per serving are used for calculations, 3 serving lead to an exposure of 0.31 µg As<sub>i</sub>/kg b.wt. per day. This could be 1.67 to 3.37-fold higher when 4 servings are consumed and the median As<sub>i</sub> concentration per serving with the 3<sup>th</sup> percentile of body weight, or the median body weight with the 97<sup>th</sup> percentile of As<sub>i</sub> concentration per serving is used for calculations, respectively. These values are within the BMDL<sub>01</sub> range identified by EFSA (2009), which highlights that the risk cannot be excluded for infants consuming baby rice.

### 3.2. Rice cereals

The ΣAs species concentration in 53 rice cereal samples belonging to 6 different commercial brands ranged from 0.042 to 0.396 mg/kg (Table 1). The As<sub>i</sub> percentage ranged from 14.2% to 89.6% of the ΣAs species concentration. The rice cereal brands M006, M007, M008, M009 and M011 had a significantly higher

percentage of As<sub>i</sub> (median of 74.2%, 81.5%, 76.6%, 83.1% and 76.6%, respectively) than the M010 (median of 62.7%; *p* < 0.001). The As<sub>i</sub> concentration ranged from 0.008 to 0.323 mg/kg and showed a good correlation with the rice content, which means that most of the As<sub>i</sub> is coming from the rice. Due to As<sub>i</sub> concentration level 2% of the rice cereal samples evaluated would be above the JECFA proposed maximum level. In addition, the rice cereal brand M009 had a higher As<sub>i</sub> concentration (median of 0.234 mg/kg; *p* < 0.001) than the other rice cereals samples analysed, which could reach up to 1.6 times the JECFA maximum level. The major organic As species for each baby rice was DMA, ranging from 0.005 to 0.082 mg/kg. Rice cereal samples labelled as produced under organic standards showed higher levels of As<sub>i</sub> (median of 0.162 mg/kg) than non-organic samples (median of 0.070 mg/kg; *p* < 0.001) (Fig. 1). As pointed out in the section on baby rice this is associated with the common inclusion of whole grain rice to produce organic products (Table 2).

The US FDA dataset on As<sub>i</sub> in rice and rice products from the US market includes 105 rice cereals (product subcategory: hot/ready-to-eat cereals). The median and range of As<sub>i</sub> in rice cereals from the USA market is 0.091 (0.023–0.283) mg/kg. As<sub>i</sub> represents a median of 63.0% of ΣAs species concentration. Our results are similar to those and no significant differences were found in the As<sub>i</sub> concentration or percentage (Table 3). However, rice cereals from the USA market had higher levels of DMA (median of 0.042 mg/kg) and ΣAs species concentration (median of 0.135 mg/kg) than rice cereals from the EU market (0.037 mg/kg; *p* = 0.004 and 0.119 mg/kg; *p* = 0.005, respectively) (Table 3). This supports the fact that the geographical origin affects the As concentration and speciation in



**Fig. 2.** Estimation of  $As_i$  exposure for infants and young children from baby rice, rice cereals and rice crackers. (The A and B plots show the  $As_i$  exposure of infants consuming baby rice. The median of  $0.70 \mu g As_i$ /serving and the 3<sup>rd</sup>, 15<sup>th</sup>, 50<sup>th</sup>, 85<sup>th</sup> and 97<sup>th</sup> percentiles of body weight have been used for calculations in plot A. The median of  $6.70 \text{ kg b.wt.}$  and the 3<sup>rd</sup>, 15<sup>th</sup>, 50<sup>th</sup>, 85<sup>th</sup> and 97<sup>th</sup> percentiles of  $\mu g As_i$ /serving have been used for calculations in plot B. The C, D, E and F plots show the  $As_i$  exposure of young children consuming rice cereals (C and D) and rice crackers (E and F). The median of  $2.22 \mu g As_i$ /serving, the median of  $3.58 \mu g As_i$ /serving and the 3<sup>rd</sup>, 15<sup>th</sup>, 50<sup>th</sup>, 85<sup>th</sup> and 97<sup>th</sup> percentile of body weight have been used for calculations in the plots C and E, respectively. The median of  $9.30 \text{ kg b.wt.}$  and the 3<sup>rd</sup>, 15<sup>th</sup>, 50<sup>th</sup>, 85<sup>th</sup> and 97<sup>th</sup> percentile of  $\mu g As_i$ /serving have been used for calculations in plot D and F).

**Table 4**

Estimation of the  $As_i$  intake ( $\mu g$ /serving) from baby rice, rice cereals and rice crackers.

| Food category | Sampling place | n   | Moisture (%)  | $As_i$ (mg/kg w.wt.) | Recommended unit (g w.wt.) | Recommended serving (g w.wt.) | $As_i$ intake per unit (mg) | $As_i$ intake per serving (mg) |
|---------------|----------------|-----|---------------|----------------------|----------------------------|-------------------------------|-----------------------------|--------------------------------|
| Baby rice     | US             | 85  | n.a           | 0.114 (0.039–0.254)  | n.a                        | 15.0 (15.0–15.0)              | n.a                         | 1.7 (0.6–3.8)                  |
|               | EU             | 29  | 2.3 (0.8–5.0) | 0.117 (0.056–0.262)  | n.a                        | 6.0 (6.0–12.0)                | n.a                         | 0.7 (0.3–2.2)                  |
| Rice cereals  | US             | 105 | n.a           | 0.091 (0.023–0.283)  | n.a                        | 30.0 (15.0–55.0)              | n.a                         | 2.6 (0.8–11.0)                 |
|               | EU             | 53  | 1.1 (0.3–7.6) | 0.075 (0.008–0.313)  | n.a                        | 30.0 (22.0–35.0)              | n.a                         | 2.2 (0.2–9.3)                  |
| Rice crackers | US             | 199 | n.a           | 0.079 (0.008–0.273)  | n.a                        | 30.0 (7.0–30.0)               | n.a                         | 2.4 (0.2–8.2)                  |
|               | EU             | 97  | 3.1 (0.3–5.8) | 0.108 (0.018–0.201)  | 1.6 (1.4–19.2)             | 30.0 (8.0–50.0)               | 0.3 (0.0–2.0)               | 3.5 (0.5–9.1)                  |

rice. Meharg et al. (2009) reported that the relationship between  $As_i$  content versus total As content differs among countries, with Bangladesh and India having the steepest slope in linear regression, and the US having the shallowest slope due to higher levels of DMA.

The plots C and D, included in Fig. 2, show the  $As_i$  exposure for young children (12 months of age) consuming rice cereals. The

median of  $2.22 \mu g As_i$ /serving (Table 4) and the 3<sup>rd</sup> (7.50 kg), 15<sup>th</sup> (8.20 kg), 50<sup>th</sup> (9.30 kg), 85<sup>th</sup> (10.50 kg) and 97<sup>th</sup> (12.10 kg) percentiles of body weight (WHO, 2014) have been used for calculations in the plot C. The median of  $9.30 \text{ kg b.wt.}$  and the 3<sup>rd</sup> ( $0.58 \mu g As_i$ /serving), 15<sup>th</sup> ( $1.47 \mu g As_i$ /serving), 50<sup>th</sup> ( $2.22 \mu g As_i$ /serving) 85<sup>th</sup> ( $3.11 \mu g As_i$ /serving) and 97<sup>th</sup> ( $5.10 \mu g As_i$ /serving) percentiles of  $As_i$  concentration per serving have been used for calculations in

the plot D. When the median body weight and the median  $As_i$  concentration per serving are used for calculations, 2 serving lead to an exposure of 0.47  $\mu g As_i/kg$  b.wt. per day. This could be 2.48 to 4.58-fold higher when 4 servings are consumed and the median  $As_i$  concentration per serving with the 3<sup>th</sup> percentile of body weight, or the median body weight with the 97<sup>th</sup> percentile of  $As_i$  concentration per serving is used for calculations, respectively. These values are within the BMDL<sub>01</sub> range identified by EFSA (2009), which highlights that the risk cannot be excluded for infants consuming rice cereals.

### 3.3. Rice crackers

The  $\Sigma As$  species concentration in 97 rice cracker samples belonging to 15 different commercial brands ranged from 0.019 to 0.246 mg/kg (Table 1). The  $As_i$  percentage ranged from 71.1% to 100% of the  $\Sigma As$  species concentration. The  $As_i$  concentration ranged from 0.019 to 0.212 mg/kg and showed a good correlation with the rice content, which means that most of the  $As_i$  is coming from the rice. Due to the  $As_i$  concentration level 1% of the rice crackers samples evaluated would be above the JECFA proposed maximum level. The rice cracker brand M019 had the highest  $As_i$  concentration (median of 0.205 mg/kg;  $p < 0.001$ ). The major organic As species for each rice cracker was DMA, ranging from 0.001 to 0.172 mg/kg. Rice crackers labelled as produced under organic standards showed higher levels of  $As_i$  (median of 0.121 mg/kg) than non-organic samples elaborated with milled rice (median of 0.071 mg/kg;  $p < 0.001$ ) (Table 2 and Fig. 1).

The US FDA dataset on  $As_i$  in rice and rice products from the USA market includes 199 samples of rice crackers (product subcategory: rice cakes and savoury rice snacks). The median and range of  $As_i$  in rice crackers from the US market is 0.079 (0.008–0.273) mg/kg.  $As_i$  content represents a median of 65.1% of  $\Sigma As$  species concentration. Our results are similar to those and no significant differences were found in the  $As_i$  or  $\Sigma As$  species concentration (Table 3). However, rice crackers from the US market had higher levels of DMA (median of 0.034 mg/kg) than rice crackers from the EU market (median of 0.025 mg/kg;  $p < 0.001$ ). Thus the  $As_i$  percentage in rice crackers from the EU market (median of 80.9%) was higher than the  $As_i$  percentage in rice crackers from the USA market (median of 65.1%;  $p < 0.001$ ) (Table 3). This is in agreement with the geographical variation in As concentration in rice, which might be associated with breeding/cultivar selection and agronomic practices (Meharg & Zhao, 2012; Meharg et al., 2009). Rice processing is another factor that may affect the As concentration in the final product. Extensive rinsing of uncooked grain followed by cooking rice in a large excess of water and discarding the water on cessation of cooking may reduce the  $As_i$  content of food by up to 30% when  $As_i$  free water is used (Raab, Baskaran, Feldman, & Meharg, 2008; Sengupta et al., 2006; Signes, Mitra, Burlo, & Carbonell-Barrachina, 2008).

The plots E and F, included in Fig. 2, show the  $As_i$  exposure for young children (12 months of age) consuming rice crackers. The median of 3.58  $\mu g As_i/serving$  (Table 4) and the 3<sup>th</sup> (7.50 kg), 15<sup>th</sup> (8.20 kg), 50<sup>th</sup> (9.30 kg), 85<sup>th</sup> (10.50 kg) and 97<sup>th</sup> (12.10 kg) percentiles of body weight (WHO, 2014) have been used for calculations in the plot E. The median of 9.30 kg b.wt. and the 3<sup>th</sup> (0.61  $\mu g As_i/serving$ ), 15<sup>th</sup> (0.90  $\mu g As_i/serving$ ), 50<sup>th</sup> (3.58  $\mu g As_i/serving$ ) 85<sup>th</sup> (6.48  $\mu g As_i/serving$ ) and 97<sup>th</sup> (8.01  $\mu g As_i/serving$ ) percentiles of  $As_i$  concentration per serving have been used for calculations in the plot F. When the median body weight and the median  $As_i$  concentration per serving are used for calculations, 1 serving lead to an exposure of 0.38  $\mu g As_i/kg$  b.wt per day. This could be 4.95 to 8.93-fold higher when 4 servings are consumed and the median  $As_i$  concentration per serving with the 3<sup>th</sup> percentile of body weight, or the median body weight with the 97<sup>th</sup> percentile of  $As_i$  concen-

tration per serving is used for calculations, respectively. These values are within the BMDL<sub>01</sub> range identified by EFSA (2009), which highlights that the risk cannot be excluded for infants consuming rice crackers.

## 4. Conclusions

Baby rice, rice cereals and crackers are widely used during weaning and to feed young children due to its availability, bland taste, nutritional value and relatively low allergic potential. They are particularly used to feed infants and young children affected by celiac disease whose only viable treatment is a gluten-free diet. The commercial brand affected the  $As_i$  concentration in the baby rice, rice cereals and rice crackers evaluated, which may be associated to the amount of rice, rice origin and manufacturing process. In fact, rice-based products including whole grain rice showed the highest  $As_i$  concentrations and they were usually labelled as produced under organic standards. The  $As_i$  concentration found in several samples was higher than the JECFA maximum level. The  $As_i$  exposure for infants consuming baby rice and young children consuming rice cereals and rice crackers are close or within the range of BMDL<sub>01</sub> values identified by EFSA (2009) and therefore the risk cannot be excluded. In addition, this could be worse when these products represent a major part of the diet and  $As_i$  contribution is accumulated. Despite this scenario there are currently no EU or USA regulations applicable to  $As_i$  in food to protect the most vulnerable subpopulation. Thus, we conclude that there is an urgent need for regulatory limits on  $As_i$  in food, especially in rice and rice-based products.

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