International Journal of Food Science and Nutrition
ISSN: 2455-4898
Impact Factor: RJIF 5.14
www.foodsciencejournal.com
Volume 3; Issue 1; January 2018; Page No. 29-33

Assessment of total arsenic in brown rice grown in the Trans Indo-Gangetic Plain (TIGP) of India

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Abstract
Rice plants uptake more arsenic as compared to other cereal crops. Arsenic levels in white rice has been studied widely in recent years however, few studies have been focused on the levels of arsenic in brown rice. Therefore, this present study has assessed the concentration of arsenic in brown rice produced in the Trans Indo-Gangetic plain (TIGP) of India, which is still an unexplored region in this context. Total arsenic in brown rice was estimated using Inductively Coupled Plasma - Mass Spectrometry (ICP-MS). Arsenic concentration in the brown rice grain varied from 14 to 1023 µg kg^{-1} (dw), with a mean value of 196 µg kg^{-1}. This value is lower than most of the published data from other countries/regions and also from the WHO recommended permissible limit in rice (1.0 mg kg^{-1}). Therefore, this study has concluded that as of now brown rice grains produced in the Trans Indo-Gangetic Plain of India are safe for human consumption.

Keywords: arsenic, brown rice, trans indo-gangetic plain (TIGP), white rice

1. Introduction

Arsenic (As) is a naturally occurring metalloid [1], which has been generally recognized as a poison [2]. Arsenic exposure causes varieties of negative health effects in humans such as hyperpigmentation, hyperkeratosis and arsenicosis [3]. Chronic exposure to As causes a risk for cardiovascular disease, hypertension and skin cancer [4]. Rice (*Oryza sativa*) is a staple food for over half of the world's population. Around 90 percent of the world's rice is produced and consumed in Asia [5]. Paddy mainly comprises of husk, bran-polish, unpolished rice (brown rice) and polished (white) rice. When paddy are dehusked, it results into brown rice with its bran layer intact, which when further milled or polished becomes 'white' or 'polished' rice by removing the bran layer[6]. In rice plants, levels of arsenic, a human carcinogen, have raised utmost concerns among public [7-10]. Rice plants are particularly susceptible to accumulate arsenic more efficiently than any other cereal crops. This is because arsenic mobility is high in flooded conditions, and when rice plants are grown under this condition with arsenic contaminated water or soil, As eventually translocate into rice plants [11]. Arsenic concentration in rice and rice-derived products vary geographically depending on various factors such as the concentration of As in soil and irrigation water, application of arsenical pesticides, processing industries and/or pollution from base and precious mining activities [12]. Arsenic residues builds up in soil when As contaminated irrigation water is continuously used in fields, because soil act as major sink to As. This increases the As concentration in rice crops grown on these fields [13-15], approximately 10-folds higher than other cereal crops [16].

In recent years, brown rice and rice bran products have gained popularity among health conscious people, as they are being recommended by nutritionist and dietitian as an excellent source of nutrition. They are increasingly being identified as "Health food" because they contains high amount of antioxidants, vitamins, minerals as well as soluble fiber [17-19]. Recent studies have shown that along with high nutrient content, brown rice and rice bran also contain higher levels of arsenic when compared with white rice [17, 18, 20, 21]. Health conscious people who generally prefer whole grain rice (brown rice) over white rice are in turn more vulnerable to As exposure. Therefore, there is a need to assess the concentration of As in Brown rice grains. Studies on concentration of As in white rice have been conducted extensively in one of the major rice growing region of India i.e., West Bengal [22-26]. However, there is little information and data available for As distribution in brown rice grown in other highest rice growing states such as Punjab, Haryana and Delhi, collectively called as Trans Indo-Gangetic Plain (TIGP) of India. Thus, the aim of the present study was therefore to assess the total As concentration of brown rice grown in the TIGP region and its comparison within the states of TIGP.

2. Materials and Methods

2.1 Study Area

The study was conducted in total six different administrative districts of the three states Punjab, Haryana and Delhi located in the Trans Indo-Gangetic Plain of the Northern India. Purposively selected districts were Amritsar, Jalandhar and Ludhiana from Punjab; Sonipat and Panipat districts from Haryana; and North-West district from Delhi. In each district, two villages were selected randomly for assessing the arsenic concentration in brown rice. In each village, two tube well operated paddy fields were selected for collection of samples (duplicates). To collect samples, fields were selected based on two parameters: (a) where the only source of irrigation water was tubewell, to avoid arsenic contamination from any other source of irrigation water; (b) availability of the standing crops in the fields.

2.2 Sample Collection

Paddy (*Oryza sativa*) samples were collected from each selected fields of the selected area in the month of November, 2016. Five sub-samples of paddy were collected at each sampling site, from a plot (2 m^2 areas), and then, aggregated into one representative composite sample of a field for further treatment. Grains from the paddy crops were hand-picked during the harvesting stage [25, 27] and stored in airtight polyethylene bags at room temperature with proper labeling [28]. Paddy were sun-dried, and then hulled to remove rice husk and to get brown rice grain., then oven dried at 70ºC for 48 h prior to analysis.

2.3 Estimation of total arsenic

A sub-sample of homogenized brown rice grain (1-2 g) was ground and weighed (0.5 g) accurately into microwave digestion vessels. High-purity nitric acid (70 % w/w, 8 ml) was added into the vessels, which were then inserted into the rotor of the microwave. All samples were digested for 1 hour and 25 min. Digestion of the samples were done in the following manner: Firstly, temperature was increased to 100ºC in 15 min, held for 30 min. Secondly, temperature was increased from 100ºC to 190ºC in 10 min, held for a further 30 min, and then cool for 10 min. After digestion the vessels were allowed to stand at room temperature for further 15 min, and then digested samples were diluted and made up to 25 ml in plastic vials of 50 ml. Total As in the samples were analyzed using ICP-MS (Element XR, Thermo Fisher Scientific, Germany) instrument. The ICP-MS operating conditions were as follows: mass range 1 to 240 amu with automatic resolution control, sensitivity (Concentric Nebuliser) 109 counts sec^{-1} for 1 ppm, quantification Limit 1 ppt, Radio Frequency Generator 27 MHz, RF Power 2 KW, Computer controlled three Argon Mass Flow Controller for torch and nebulizer. Nebulizer for normal acid based solution: flow rate of 50 µL min^{-1} and 100 µL min^{-1} [29].

2.4 Statistical Analysis

The data statistical analysis was performed using statistical package, SPSS 20.0 and MS Excel for windows (SPSS Inc, USA). Statistical significance of differences was compared using one-way ANOVA.

3. Results and Discussion

3.1 Total As concentration in brown rice

The concentrations of Total As in brown rice grain samples from 12 villages of Trans Indo-Gangetic Plain (TIGP) of India are presented in Table 1. The total As concentration in brown rice of TIGP varied greatly, from 14 to 1023 µg kg^{-1} (dw). The range of As concentration in the present study was wider, this is due to the contribution of one sample with higher As (1023.18 µg kg^{-1}). The mean As for all 48 brown samples was 196 µg kg^{-1}, with a median value of 109 µg kg^{-1}, (dw). This mean value was lower than result from most of the other studies as presented in Table 2. Halder *et al*., 2012 [30] reported an average As concentration of brown rice of West Bengal which was 330 µg kg^{-1}. The As concentration of brown rice of the present study was 40 % lower than this data. Total As concentration in West Bengal and International brown rice published by other researchers are showed in Table 2. A comparison of our study with data published for other countries showed that As level in TIGP brown rice grain was lower than the results obtained from Bangladesh (880 and 610 µg kg^{-1}), U.S. (440 µg kg^{-1}), China (360 µg kg^{-1}) and Japan (226 µg kg^{-1}). When compared with International data, As concentration of our brown rice was 13 – 77 % lower than these data.

Table 1: Arsenic levels (µg kg^{-1}, dw) in brown rice grain collected from 12 villages of Trans Indo-Gangetic Plain (TIGP) of India

Village	District	State	n	Mean	Median	SD
Khasa	Amritsar	Amritsar	4	74	73	43
Atari	Amritsar	Amritsar	4	53	52	12
Bidhipur	Jalandhar	Amritsar	4	115	114	78
Paragpur	Jalandhar	Amritsar	4	472	465	76
Khanna	Ludhiana	Amritsar	4	74	73	41
Doraha	Ludhiana	Amritsar	4	115	115	83
Raipur	Sonipat	Haryana	4	468	369	391
Joshi Jat	Sonipat	Haryana	4	348	309	248
Patti Kalyana	Panipat	Haryana	4	64	55	29
Bhodwal Majri	Panipat	Haryana	4	404	382	67
Naya Bansh	North-West Delhi	Delhi	4	44	43	26
Khera Khurd	North-West Delhi	Delhi	4	119	120	68

Studies have shown that arsenic is mostly localized at the surface of the brown rice, mainly in the pericarp and aleurone layer of the grain [21]. Meharg *et al*., 2008 [21] reported that arsenic, is present in higher proportion in brown rice as compared to white rice. The present study showed that brown rice from TIGP contained much lower As concentration. In order to better understand the contaminated status of As in brown rice, we compared our data with the WHO recommended permissible limit of 1.0 mg kg^{-1} for As in rice. Our present study resulted that almost all of the collected samples (97%) from TIGP region contained total As lower than WHO recommended permissible limit (1.0 mg kg^{-1} for As). Only 2% brown rice samples from the collected samples contained total As above WHO limit. Thus, it can be stated that as of now brown rice of TIGP region is safe for human consumption. However, As content was present in all of the 48 samples of brown rice of TIGP region, and therefore, brown rice contributes as a potential source of arsenic intake among people who consider brown rice as "Health food" [17,18].

Table 2: Total As concentration in West Bengal and International brown rice published by other researchers

Regions	No. of samples	Total As concentrations (μg kg^{-1})	Reference
Bangladesh	3	800 ± 100	Rahman *et al.*, 2007 [31]
Bangladesh	1	610	Meharg *et al.*, 2008 [21]
China	1	360	Meharg *et al.*, 2008 [21]
U.S.	1	440	Meharg *et al.*, 2008 [21]
China	282	92	Fu *et al.*, 2011 [32]
Australia	12	170	Fransisca *et al.*, 2015 [33]
Australia	12	100	Fransisca *et al.*, 2015 [33]
West Bengal	157	330	Halder *et al.*, 2012 [30]
Japan	199	160	MAFF, 2006 [34]
Japan	10	226	Naito *et al.*, 2015 [35]

3.2 Comparison of as content in brown rice of TIGP within the states

Mean As content in brown rice grain from 12 villages of Trans Indo-Gangetic Plain (TIGP) of India (error bars are SD) are shown in the Figure 1. The total As concentration in brown rice of Punjab ranged from 33.226 to 559.41 μg kg^{-1} (dw), with mean As concentration of 151.036 ± 158.37 μg kg^{-1} (dw). The highest and lowest total As content in brown rice grain samples of Punjab was present in Paragpur and Atari village, with mean As concentration of 472 ± 76 and 53 ± 12 μg kg^{-1} (dw) respectively. The total As concentration in brown rice of Haryana ranged from 38.853 to 1023.18 μg kg^{-1} (dw), with mean As concentration of 321.57 ± 263.66 μg kg^{-1} (dw). The highest and lowest total As content in brown rice grain samples of Haryana was present in Raipur and Patti Kalyana village, with mean As concentration of 468 ±391 and 64 ± 29 μg kg^{-1} (dw) respectively. In Delhi, Total As in brown rice grain samples ranged from 14.104 to 184.64 μg kg^{-1} (dw). Highest As content was found in Khera Khurd, while lowest As content was found in Naya Bansh, with mean value of 119 ± 68 and 44 ± 26 μg kg^{-1} (dw) respectively. The average concentration of As in brown rice samples was found in the order of Paragpur > Raipur> Bhodwal Majri > Joshi Jat > Khera Khurd > Bidhipur-Doraha > Khasa – Khanna > Patti Kalyana > Atari > Naya Bansh.

In Haryana, significantly higher total As content in brown rice was found when compared with Punjab (p=0.015) and Delhi (p=0.020) brown rice. The difference in the concentration of As in brown rice between different states could be because As concentration varies geographically. Arsenic content in rice plants depends on various factors such as concentration of arsenic in irrigation water and soil, and other farm management activities like fertilization practices, crop rotation, and herbicidal/insecticidal uses [17].

According to Mani *et al.*, 2004 [36] and Lalwani *et al.*, 2004 [37], higher As content has been reported in some industrial area of Haryana and Delhi, which can be one of the source of arsenic in brown rice. In Punjab 40% tubewell water samples contain As greater than the WHO permissible limit (10 μg As L^{-1}). This suggested that irrigation with As contaminated water over a longer period of time may have detrimental effects on soil as well as on crops [38].

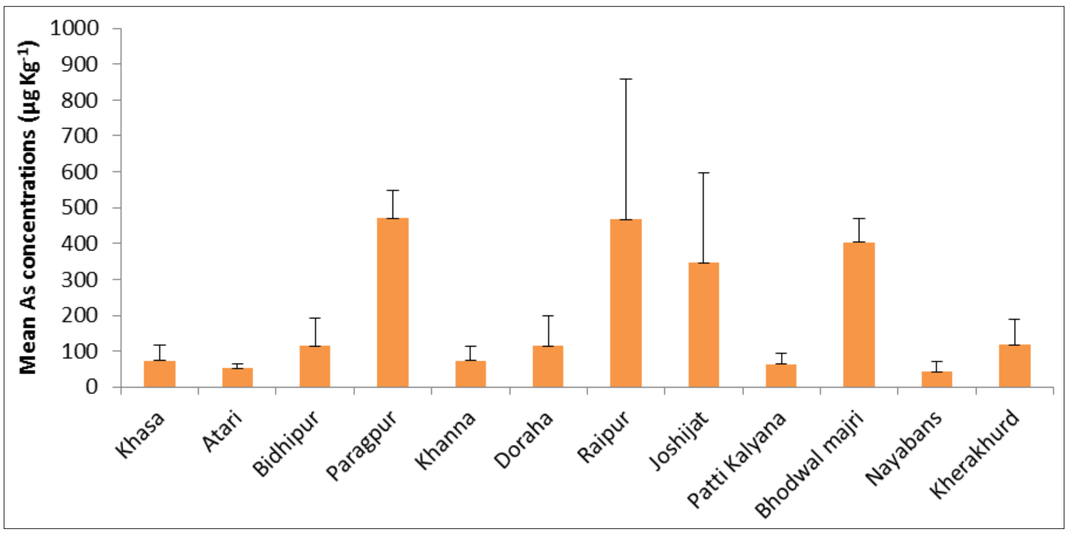

Fig 1: Mean As content in brown rice grain from 12 villages of Trans Indo-Gangetic Plain (TIGP) of India (error bars are SD)

4. Conclusion

The present study reveals the Total As concentration in the brown rice grain sample of TIGP region, India. Arsenic concentration was lower in brown rice grain when compared with most the published data and WHO recommended permissible value. This suggests that they are safe for consumption, unless brown rice makes up a large proportion in the diet. Though the As concentration was lower in all of the brown rice grain samples, but it is still one of the hidden source of As exposure in Humans, as any amount of arsenic is likely to be matter of concern to the public. Therefore, present study suggests the regular monitoring of arsenic content in brown rice.

5. Acknowledgement

The authors are cordially acknowledging the Sophisticated Analytical Instrument Facility (SAIF) Lab, IIT Bombay for giving the permission to use their ICP-MS Lab and Chemical Analysis Lab. The first author is also thankful to the University Grants Commission (UGC), for awarding the

UGC-NET/JRF fellowship for the research work.

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