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Comparative Studies Revealing Arsenic Concentrations in Cooked as well as Raw Rice and Rural Bengal Cooking Method using both Arsenic Contaminated Rice and Ground Water

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ABSTRACT

The rice, specially *Oryza sativa*, is significantly known as a staple food in rural as well as urban areas of Bengal. The conventional cooking method is usually practiced to prepare cooked rice for meals and tiffins. The present study suggests that selection of both water and rice plays an important role in order to avoid Arsenic (As) exposure from food article specially rice. It is important to note that the presence of ground water with low or very low As concentration has been used for drinking purposes in several parts of rural Bengal. Additionally, several rice varieties have also been identified contaminated with As. This study also emphasizes that the uses of As free water and rice are remarkable protocol for cooking to minimize the exposure of As from both.

***Key words*:** Arsenic, Cooking method, Atomic absorption spectrophotometer, Rice

Groundwater is the main source of drinking water for about 90% of the total population (130 million) of Bangladesh and West Bengal and million of people are consuming this groundwater which is contaminated with arsenic [1-2]. The sources of arsenic contamination in this groundwater remain undiscovered. However, it is believed that long term geological changes have led to release arsenic from its core compound called arsenopyrities due to its oxidation by air reaching the underground aquifers through the tube wells conduits [3-5]. A vast portion of populations in these areas have been consuming arsenic contaminated ground water not only for drinking purposes but also for rice cultivation, particularly during summer [6]. In these countries, arable lands are under the facility of irrigation which is performed mainly with very high level of arsenic contaminated (>50-5000 ppb) underground water [7-8]. During Irrigation with arsenic contaminated groundwater, the arsenic concentration is likely to increase in top soils of paddy fields and eventually in different varieties of rice grains [9-11]. The drinking water is not only the main source of arsenic, but the intake of arsenic contaminated rice grain also greatly contributes in increasing arsenic concentration in human body as the major portion of populations consuming rice as an essential diet for their caloric intake [12-14]. The residents of these areas cook rice using both arsenic contaminated water and arsenic enriched rice grain and this cooking process eventually increases the level of arsenic concentration in their bodies. Thus, concentration of arsenic in rice grain is a raising concern in these areas [15]. There are some reports in literature on arsenic content in cooked rice [16]. However, reports on estimation of arsenic concentration in cooked rice are very rare. This work reveals the effects of traditional cooking methods on arsenic retention in cooked rice.

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MATERIALS AND METHODS

Reagents

The chemicals (HCl, NaOH, HNO_3, H_2O_2 and $HClO_4$, KI, $NaBH_4$) used in this report were of analytical grade and obtained from E. Merck, India, DM (MiliQ water) which was used in AAS method (VGA). Silver diethyldithiocarbamate (SDDC), stannous chloride, Zinc, I_2-KI, Lead acetate, Arsenous oxide used for preparation of standard solution.

Instrumentation

The spectral measurements were performed in a UV-Vis spectrophotometer (Model Shimadzu, UV-2401PC). Arsenic concentration in the experimental solution was determined by flame atomic absorption spectrophotometer (Varian AA1407) using an air-acetylene flame and hollow cathode lamp as the radiation source. Systronics digital pH meter (Model 335) was used for pH measurements.

Sampling and analytical technique

Study area

The study area is a component of the Bengal delta plain (latitude 23º/20// N-23º05/20// N; longitude 88º31// E- 88º49// E; Chakdaha block, Nadia district, 65 km north of Calcutta, West

Bengal, India, Fig 1) encompassing an integral part of the world's largest delta, the Ganga- Brahamputra deltaic alluvium. The Holocene sedimentary succession of the fluviatile deposits comprises of Fe-coated micaceous san, silt and clay with a sequence of fining upward cycles of variable thickness deposited by meandering rivers. The young gangatic flood plain is characterized by a series of meander scars of varied wavelength and amplitudes, anandoned channels and ox – bow lakes with a gradual southward slope. Other landform features are levees, back swamps in between interdistributary levees with a relief difference of a few metres. The climate is tropical, hot and humid (temperature range 16-42°C; average relative humidity >65%) with annual rainfall ranging between 1295mm and 3945mm.

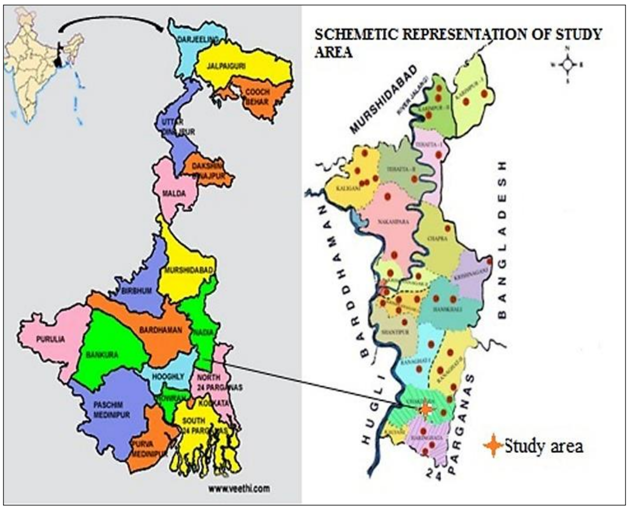

Fig 1 Schematic representation of study area

Analytical techniques

Major components

- Collection of contaminated water and raw rice samples and total As determination
- Rural experiment applied for determination of As_T (Total Arsenic)
- Sample treatment
- Sampling and classification of rice samples
- Sample digestion
- Diagrammatic representation of rural rice cooking method

Sample collection

The present study was conducted in the Village Bishnupur of Chakdaha block, Nadia District, W.B. India. We collect five different known As content water samples from house hold tube wells and five different rice samples. Water sample were collected in 1000 mL polyethylene bottle (without preservative) and another 500 mL polyethylene bottle (with preservative, conc. HNO_3). These bottles were rinsed with samples collected from water source. Lastly after collection, raw rice samples were kept in the airtight polythene zipper bags at room temperature.

Experiment applied for determination of total arsenic (As_T)

In Rural Bengal, the traditional cooking procedure was followed to cook rice samples. A desired amount of rice was washed twice with As content water, followed by boiling with that water until the rice grain became soft like properly cooked rice. Considering the above procedure, the actual experiment was carried out by a variety of rice samples with different As containing water samples. Each 5g of sample was taken in a 250g Aluminum container containing a lid and 200ml As containing water (from a specific well) was added. The whole system was then put into a clay-oven for 15-18 minutes to prepare properly cooked rice. After the scheduled time, the cooked rice and starch water samples were being separated in different containers and allowed to cool at room temperature. As soon as the samples were cooled the rice samples were collected a zipper bottle respectively. These samples were then sent to the laboratory for the detection of As content both in rice as well as in starch samples considering the conventional procedure. Calibration standards were prepared using the same HCl concentration of the samples and certified materials. The instrumental conditions used for As determination by AAS (Varian AA1407) were as follows. Reducing agent: 0.6% (m/v) $NaBH_4$ in 0.5% NaOH, 5 mL/min; HCL solution: 8 % (v/v), 10 mL/min; carrier gas: argon, 250 mL/ min flow rate; and for atomic absorption spectrometry wavelength: 185 nm; spectral band pass: 0, 5 nm; hollow cathode lamp current setting 8 mA; air/acetylene flame with a fuel flow rate of 0.8 2 /min.

The above procedure was repeated at least twice for each and every variety of rice samples containing different As content water to estimate the minimum amount of As present in starch water as in the rice sample.

Sample treatment

The rice plant samples were washed thoroughly with taped water to removed soil and other contaminates and finally ringed with de-ionized water with continuous shaking for several minutes. The samples were then dried in hot air oven at 60°C for 72 hours and store in airtight polyethylene bags at room temperature. Proper care was taken at each step to minimize any short of contamination.

Sampling and classification of rice samples

A total of 80 samples were collected, 40 from randomly selected household and 40 from randomly selected market baskets. The sample collection was followed by a detailed questionnaire survey, which includes information about the rice color, source. After collection, rice samples were stored in air tight polyethylene zipper bags at room temperature.

Sample digestion

About 0.5g of the rice sample was taken into clean dry digestion tubes and 5 mL of conc. HNO_3 was added to it. The mixture was allowed to stand overnight under fume hood. In the following day, the digestion tubes were placed on heating block and heated at 60°C for two hours and rises the temperature at 100°C for two hours. The tubes were then allowed to cool at room temperature. About 2 mL of Conc. $HClO_4$ was added to the rice samples. Then the tubes were heated at 150°C for about 3-4 hours. The heating was stopped when the dense white fume of $HClO_4$ was emitted. Then H_2O_2 was added into the digestion tube till evaporation was stopped and colorless. Then the solution was heated till the volume became 1/3. The solution was poured in the 10mL volumetric flask.

Sample analysis

The total arsenic concentration of rice was analyzed by atomic absorption spectroscopy till the dense white fume of $HClO_4$ was emitted. Then H_2O_2 was added in the digestion tube till evaporation was stopped and colourless. Then the solution was heated till the volume became 1/3. The solution was poured into 10 mL volumetric flask. The Spectrophotometric determination of As was performed, where the standard solution was passed first time and got the standard curve. The arsenic concentration of colourless rice sample solution was measured with respect to the standard curve. The above cooking method is shown by the following diagram (Fig 2).

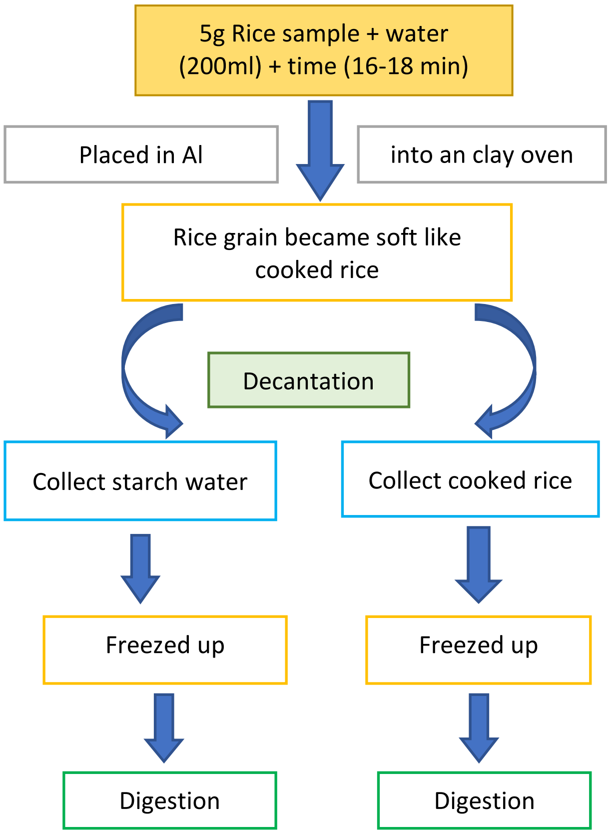

Fig 2 Diagrammatic representation of cooking method and its digestion

RESULTS AND DISCUSSION

Validation of total as in raw and cooked rice

Total As (As_T) contents of raw and cooked rice samples were shown in Table no. 3 to 8 of supporting information respectively of supporting information respectively. The total As content in raw rice sample varied from 11.08 to 4.82 ppb with a mean value 6.86 ppb which was quite comparable (Table 1).

Table 1 Raw rice samples and total As content

Name of the raw rice sample	Total As (ppb)
Miniket (amon)	11.08
Miniket (boro)	10.15
Swarno	9.95
Ranjit	7.30
Nagra	4.82

The range of (11.08-4.82) total As concentrations exceeded the global normal distribution range of 0.08-0.02 ppb. The total As concentrations varied with different types of rice samples such as Minikit (Amon), Minikit (Boro), Swarno, Ranjit, Nagar. It was found that Minikit (Amon) contained the highest As concentrations whereas Nagar (short-grain) red color was identified with lowest As concentration. The raw rice was cooked with 5 different concentrations of ground water samples collecting from 5 different wells such as Well A, Well B, Well C, Well D, Well E (Table 2).

Table 2 Raw tube well water samples and total As content

Name of the different tube well	Total As (ppb)
Well A	73.6
Well B	176
Well C	262
Well D	223
Well E	137

The raw rice and ground water were preceded by traditional cooking method. The cooked rice and starch water were collected for As determination (Table 3-7).

Table 3 The change of As_T in rural rice (Nagar) cooking method

Rice sample name (Nagra)		Tube well name (Well A)		Total As_T (ppb) (Rice + Water)		Total change of mass (change of As_T) after cooking. (ppb)
Types of rice	As_T (ppb)	Types of water	As_T (ppb)	Types of rice + Types of water	As_T (ppb)	= (78.42-59.52) =18.9
Raw rice	4.28	Raw water	73.6	Raw rice + Raw water	78.42	
Cooked rice	8.92	Starch water	50.6	Cooked rice + starch water	59.52	

Table 4 The change of As_T in rural rice (Ranjit) cooking method

Rice sample name (Ranjit)		Tube well name (Well B)		Total As_T (ppb) (Rice + Water)		Total change of mass (change of As_T) after cooking. (ppb)
Types of rice	As_T (ppb)	Types of water	As_T (ppb)	Types of rice + Types of water	As_T (ppb)	= (183.30-178.75) = 4.55
Raw rice	7.30	Raw water	176	Raw rice + Raw water	183.30	
Cooked rice	10.95	Starch water	167.8	Cooked rice + starch water	178.75	

Table 5 The change of As_T in rural rice (Mniket - Boro) cooking method

Rice sample name (Mniket - Boro)		Tube well name (Well C)		Total As_T (ppb) (Rice + Water)		Total change of mass (change of As_T) after cooking. (ppb)
Types of rice	As_T (ppb)	Types of water	As_T (ppb)	Types of rice + Types of water	As_T (ppb)	= (272.15 -259.35) =12.8
Raw rice	10.15	Raw water	262	Raw rice + Raw water	272.15	
Cooked rice	13.35	Starch water	246	Cooked rice + starch water	259.35	

Table 6 The change of As_T in rural rice (Miniket-Amon) cooking method

Rice sample name (Miniket-Amon)		Tube well name (Well D)		Total As_T (ppb) (Rice + Water)		Total change of mass (change of As_T) after cooking. (ppb)
Types of rice	As_T (ppb)	Types of water	As_T (ppb)	Types of rice + Types of water	As_T (ppb)	
Raw rice	11.08	Raw water	223	Raw rice + Raw water	234.08	= (234.08-217) =17.08
Cooked rice	32	Starch water	185	Cooked rice + starch water	217	

Table 7 The change of As_T in rural rice (Swarno) cooking method

Rice sample name (Swarno)		Tube well name (Well E)		Total As_T (ppb) (Rice + Water)		Total change of mass (change of As_T) after cooking. (ppb)
Types of rice	As_T (ppb)	Types of water	As_T (ppb)	Types of rice + Types of water	As_T (ppb)	
Raw rice	9.95	Raw water	137	Raw rice + Raw water	146.95	= (234.08-217) =17.08
Cooked rice	11	Starch water	122	Cooked rice + starch water	133	

The concentration of total As in cooked rice sample was envisioned to vary from 32-8.92ppb. In each case it was found that the concentration in the cooked rice increased depending on the concentration of As in ground water [17].

The result showed that the mean As concentration of total As in the raw rice and indicated that the cooking method and use of water, notably as contaminated water may have an influence on cooking rice. These were clearly reflected on each sample where both ground water total As concentration and raw rice total As concentration were found different [18-19].

The total As in starch water also varied with a mean value 154.28. It was also found that the concentration of total As in starch water was relatively low with respect to cooked rice. This was noticed in each sample and varied with respect to raw. The results indicated that the total As in cooked rice largely depended on initial total As concentration of raw rice and total As concentration of raw water [20]. The conventional cooking method (raw rice-water-boiling in container-starch water-cooked rice) may not have much impact on the increase of As concentration in cooked rice. Alternatively, the choice of rice and the selection of water contaminated with As are important issues to understand the level of As in cooked rice [21].

CONCLUSION

As the cultivation of rice requires huge volume of water, long term use of arsenic contaminated ground water for irrigation may results in the increase of arsenic concentration in the agricultural soil and eventually accumulation in rice plant. In the rice plant, the highest accumulation of arsenic was noticed in the root and lowest in the grain. The selection of rice and choice of water are principle issues to understand the enrichment of As level in cooked rice. This work significantly provides a new approach about the effect of cooking method and rice type on arsenic concentration in cooked rice and the estimation of arsenic dietary intake in a rural village in West Bengal, India.

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