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Arsenic(As) Removal from Rice Using Orange Peel as Bio-Sorbent

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Abstract: Rice aggregates have higher as than other plants as it is considered normal filled in overwhelmed conditions, which prompts the transcendence of the versatile as (III) or Arsenic. The objectives of this study is to determine the Arsenic concentration in commercially available brown rice and the optimal condition for the removal of arsenic by using orange peels under batch mode and to ascertain the removal efficiency of orange peels under batch mode. The rice tests were exposed to six preparation methods which included rinsing, soaking with the orange peel biosorbent, and cooking. Two different sizes of the orange peel bio-sorbent (75 μ m and 150 μ m) were additionally tried to decide the ideal size of the bio-sorbent for as removal from the rice grains. The six preparation methods could eliminate as of brown rice grains to permissible cutoff. Soaked with orange peel bio-sorbent of 75 μ m size for 24 hours, followed by washing and cooking - accomplished the most noteworthy as evacuation effectiveness (34.97±0.40%) for brown rice grains. This investigation likewise discovered that the ideal orange peel bio-sorbent size for as elimination from brown rice is 75 μ m.

Keywords: Orange Peel, Arsenic, Bio-Sorbent

1. Introduction

This Arsenic is the first group of carcinogens, according to the International Agency for Research on Cancer (IARC). Rice is a widespread staple dish eaten by people all over the world, particularly in Asia. According to WHO research, rice is the most common cause of arsenic exposure in humans. Rice is a plant that lives in an anaerobic environment. Arsenic in the soil was absorbed in anaerobic circumstances, causing arsenic levels in rice to be 10 times higher than in other plants. When rice crops are sown in soil contaminated with arsenic, such as when farmers use pesticides and irrigation water contains high arsenic levels, arsenic levels in rice grains rise. Arsenic levels in rice varied from 0.001 mg/kg to 0.42 mg/kg, according to FSA. The arsenic content of rice was determined to be 0.14 g/kg on average. The arsenic concentration of brown rice was higher than white rice in a study conducted by the Food and Drug Administration on 200 samples of rice and rice products from 2012 to 2013. The

peel of an orange is employed in our research since oranges are one of the most widely consumed citrus fruits. This fruit has an acidic flavour and is high in nutrients as well as health-promoting characteristics. Oranges are extremely low in calories and contain no saturated fats or cholesterol. The current research focuses on the use of orange peel as an agricultural and food waste bio-sorbent for removing harmful Arsenic from brown rice.

1.1 Problem Statement

In this modern era, Arsenic (As) exposure is a global public health concern affecting millions worldwide and stems from drinking water and foods containing As. Long-term exposure to As in drinking water has been associated with bladder cancer, kidney cancer, lung cancer, skin cancer and respiratory diseases. However, global is more focused on the presence of As in water as the major source of As exposure to humans is through drinking water. Arsenic content found in food is the cause of contamination of As in soil. Arsenic in soil contaminates depending on factors including geographic location, soil characteristics, microbial populations, and complicated plant-soil interactions, its concentrations and availability for plant uptake can vary greatly. Geographical areas with large concentrations of geogenic As, like Bangladesh, have high levels of water and food contamination. Due to its geochemistry and effective uptake by rice roots, As is extremely mobile in environments like rice paddies. Moreover, nearly half of the world's population eats rice, which provides 30-70 percent of their energy as well as a variety of micro and macronutrients. Brown rice contains more As compared to white rice. Brown rice is classified as a whole grain cereal since the complete outer layer has been preserved. The bran layer contains a variety of nutrients, dietary fibers, vitamins, and minerals. It is a crop that also quickly absorbs arsenic from soils. It is because the outer bran layer surrounding the endosperm of rice grains is where inorganic As is concentrated, brown rice (unmilled, unpolished rice that keeps its bran) has a higher inorganic As content than white rice.

The toxic As in rice thereby enters the body through the food chain and results in a number of diseases. According to Weber, many studies conducted are concern in removing heavy metal found in water bodies but research on removing As from rice is very limited. One of the limited studies about removal of arsenic from rice was conducted by Zhang, proofs that under carefully monitored soaking conditions, soaking can effectively remove arsenic from brown rice by lowering the amount of arsenic consumed by human. To date, As has been treated using a variety of traditional techniques, including coagulation-flocculation, precipitation, ion exchange, adsorption, ion exchange, and membrane filtration. According to Farhat, removing As by using traditional methods seems to be inefficient as compared to using bio-sorbents. Since it offers additional advantages over conventional methods. including lower operating costs, minimal sludge creation, and simpler accessibility of the bio-sorbent materials, the bio-sorption method has advanced in recent years. Accordingly, orange peel waste has demonstrated as potential bio-sorbents to remove As from groundwater. The presence of cellulose, lignin, hemicelluloses, carboxyl (-COOH), hydroxyl (-OH), and amide (-N-H) functional groups in orange peel waste serve as binding sites for metal ions and plays a significant role in the removal of metals. Therefore, orange peel waste can be used to treat As found in brown rice. According to Amin smaller bio-sorbents helps in metal adsorption. Thus, bio-sorbent particle size and soaking time are chosen as the parameters for the removal efficiency in this study.

1.2 Objectives

The objectives of this study is to determine Arsenic concentration in commercially available brown rice and the optimal condition for the removal of Arsenic by using orange peels under batch mode. Besides that, the removal efficiency of orange peels under batch mode was ascertained as well.

2. Materials and Methods

In this chapter, procedures carried out in this study will be discussed.

2.1 Materials

The materials, chemicals, reagents, and equipment needed in this study is summarized in Table 1.

Parameters	Materials/ Chemicals/Reagents	Equipment		
Arsenic	 Milli-Q water Concentrated nitric acid (HNO3) solution (65 %) Concentrated hydrochloric acid(HCl) solution 	 Drying oven Pestle and Mortar Inductively Coupled \Mass Spectrometer (ICP-MS, ELAN DRC-e, Perkin Elmer, USA) 		
Bio-sorbent particle size	-	75µm sieve150µm sieve		
Bio-sorbent dosage	-	Electronic balance		

Table 1: The materials, chemicals, reagents and equipment used in this study

2.2 Methods

The methodology of this study is illustrated in an info graphic figure. The research methodology is divided into 3 stages preparation, measurement and output. In preparation stage, detailed background study, objectives, problem statements, literature review and methodology will be determined. Then, apparatus that has been used in this study underwent preliminary cleaning which is acid wash. For Arsenic analysis, EPA Method 3050 B was chosen. In the second stage, brown rice samples and orange peel wastes has been collected and prepared under 2 batch modes, one is soaking time(1 hour and 24 hours) and another is bio-sorbent particle size(75 μ m and 150 μ m). Then all rice samples were sent for acid digestion. Acid digested samples were filtered and sent for ICP-MS analysis. After the ICP-MS analysis, results were analysed based on the objectives.



Figure 1: The framework of research methodology

2.2.1 Preparation of bio-sorbent (Orange peel powder)

The orange peels were ready as per the procedure depicted by (Khaskheli 2011). In this research, the albedo part was applied in the making of the orange peel powder as a bio-sorbent. Momentarily, orange peel waste was gathered from a restaurant. The peels were washed with deionized water (Milli-Q water) to wash out dirt particles. The peels were dried in the oven at 60°C for 6 hours. The dried peels were then grounded using pestle and mortar and afterward went through sieve shakers of various sizes (75 μ m and 150 μ m), preceding use in the adsorption tests. The grounded orange strip powder was placed in zip lock bags.



Figure 2: Orange peels were washed before oven-drying

2.2.2 Collection of rice sample

Brown rice purchased from a supermarket called Lotus's. The brand brown rice was Brand Floral which is easily available in market.

2.2.3 Screening of rice sample

Screening of raw brown rice grains done to determine the concentration of As, prior to treatment.

2.2.4 Treatments of rice sample

Treatments were modified with minor changes from (Sharafi 2019). For each treatment, brown rice samples were prepared. From section 2.2.4.1 to section .2.4.6, the details of each treatment were discussed.

2.2.4.1 Treatment 1: Rinsed 5 times + cooked

Milli-Q water was used to wash approximately 50 g of raw brown rice grains for 5 times then the samples were cooked in 100mL Milli-Q water (1:2 ratios; rice: water ratio) until the water was completely absorbed.

2.2.4.2 Treatment 2: Soaked with water and orange peel powder($75\mu m$) for 1 hour + rinsed 5 times + cooked

A beaker of 100mL Milli-Q water with particle size of 75μ m was used to soak 50 g of raw brown rice grains for 1 hour. The Milli-Q water in the beaker was discarded after soaking. Thereafter, the rice grains were rinsed 5 times with Milli-Q water. After that, the samples were cooked in 100mL Milli-Q water (1:2 ratios; rice: water ratio) until the water was completely absorbed. 2.2.4.3 Treatment 3: Soaked with water and orange peel powder ($150\mu m$) for 1 hour + rinsed 5 times + cooked

A beaker of 100mL Milli-Q water with particle size of 150μ m was used to soak 50 g of raw brown rice grains for 1 hour. The Milli-Q water in the beaker was discarded after soaking. Thereafter, the grains were rinsed 5 times with Milli-Q water. After that, the samples were cooked in 100mL Milli-Q water (1:2 ratio; rice: water ratio) until the water was completely absorbed.

2.2.4.4 Treatment 4: Soaked with water for 24 hours + rinsed 5 times + cooked

50g of raw brown rice grains was soaked in a beaker of 100mL Milli-Q water for 24 hours. The Milli-Q water in the beaker was discarded after soaking. Thereafter, the grains were rinsed 5 times with Milli-Q water. After that, the samples were cooked in 100mL Milli-Q water (1:2 ratio; rice: water ratio) until the water was completely absorbed.

2.2.4.5 Treatment 5: Soaked with water and orange peel powder (75 μ m) for 24 hours + rinsed 5 times + cooked

A beaker of 100mL Milli-Q water with particle size of 75μ m was used to soak 50 g of raw brown rice grains for 24 hours. The Milli-Q water in the beaker was discarded after soaking. Thereafter, the grains were rinsed 5 times with Milli-Q water. Then, the samples were cooked in 100mL Milli-Q water (1:2 ratio; rice: water ratio) until the water was completely absorbed.

2.2.4.6 Treatment 6: Soaked with water and orange peel powder (150 μ m) for 24 hours + rinsed 5 times + cooked

A beaker of 100mL Milli-Q water with particle size of 150µm was used to soak 50 g of raw brown rice grains for 24 hours. The Milli-Q water in the beaker was discarded after soaking. Thereafter, the grains were rinsed 5 times with Milli-Q water. After that, the samples were cooked in 100mL Milli-Q water (1:2 ratio; rice: water ratio) until the water was completely absorbed. 2.2.5 Oven-drying, grounding, and storage of rice samples

All rice samples were oven-dried at 65 °C for 72 hours and grounded using pestle and mortar. Prior to acid digestion, the ground samples were stored in acid-washed zip lock bags at room temperature.



Figure 3: Rice samples were oven dried at 65°C for 72 hours

2.3 Acid digestion of rice samples and Arsenic analysis

Acid digestion was used to remove arsenic (As) from all rice samples. Method 3050 B (Acid Digestion of Sediments, Sludges, and Soils) of the Environmental Protection Agency (EPA) was used for the acid digestion (US-EPA, 1996). Each rice sample was weighed at around 1.0g. About 10mL of 1:1 H₂O to HNO₃ (65%) was added to the rice and heated to $95^{\circ}C \pm 5^{\circ}C$ without boiling for 10 to 15 minutes. The samples were allowed to cool before being used. 5mL concentrated HNO₃ was then added. For another 30 minutes, the samples were heated. If brown fumes were produced, showing that the samples had been oxidised by HNO₃, this procedure (adding 5mL of concentrated HNO₃) was repeated until no brown fumes were produced, indicating that the reaction with HNO₃ had been completed. The solution was either allowed to evaporate to around 5mL without boiling or heated to 95°C ± 5°C for 2 hours without boiling.

At all times, a layer of solution was kept on the bottom of the vessels. 2mL water and 3mL H_2O_2 (30%) were added to the samples after they had cooled. Warming and commencing the peroxide reaction were accomplished by returning the samples to the heat source. Throughout the digestion process, the vessels were covered with a watch glass. Excessively intense effervescence must be avoided if losses are to be avoided. After that, the vessels were heated until the effervescence ceased, and then cooled. H_2O_2 was added in 1mL aliquot with warming until the effervescence was negligible or the general appearance of the sample remained unchanged. For 2 hours, the acid-peroxide digestate was continually heated at 95°C ± 5°C without boiling. The digestate was then filtered through 0.45µm filter paper (Whatman) and the filtrates were diluted to 100mL with Milli-Q water in a volumetric flask. The samples were then sent for as analysis by Inductively Coupled Plasma Mass Spectrometry (ICP-MS).



Figure 4: Samples were sent for ICP-MS analysis

2.4 Quality Assurance and Quality Control (QA/QC)

All lab-ware was soaked overnight in 5% (v/v) nitric acid, then rinsed three times with double distilled water and dried in the oven before analysis. For each set of analyses, reagent blanks were run through the whole method. Blank samples were also prepared and subjected to the identical digestion processes as the actual samples. These blank solutions are used to correct the backdrop in the post-experiment analysis.

2.5 Equations

The removal efficiency of each treatment method was calculated by using Equation 1:

Removal efficiency (%)= $(C_i-C_f)/C_i \times 100$ % (Equation 1)

where, Ci and Cf represent the initial and final As concentration (mg/kg). The statistical analysis was carried out using SPSS 21 software (SPSS Inc., Chicago, IL, USA). To ensure that the data was normal, the Shapiro-Wilk test was used. The concentration of As in rice samples will also be compared to the Codex Alimentarius International Food Standards Schedule of the Food and Agriculture Organization of the United Nations (FAO)/World Health Organization (WHO). The FAO/WHO joint standard was chosen because it is an international food standard designed to protect consumer health and support fair trade practices.

3. Results and Discussion

This chapter will discuss about the results from experiment conducted on rice samples using orange peels.

3.1 Results

The average total As concentration in all rice samples and As removal efficiencies of the different preparation methods are summarized in Table 2.

Rice	Rice preparation methods	Average As concentration		Removal
		Before (mg/kg)	After (mg/kg)	efficiency (%)
Brown	Rinsed five times+cooked			11.75
	(Treatment 1)	0.7355 ± 0.0133	0.2425 ± 0.0125	±
				0.50
	Soaked with water and orange			10.37
	peel powder(75µm) for 1 hour + Rinsed 5 times + Cooked	0.7355 ± 0.0133	0.2463 ± 0.0227	±
	(Treatment 2)			0.90
	Soaked with water and orange peel powder(150µm) for 1 hour + Rinsed 5 times + Cooked	0.7355 ± 0.0133	0.2281 ± 0.0072	16.99
				±
	(Treatment 3)			0.51
	Soaked with water for 24			31.99
	hours+ Rinsed 5 times + Cooked	0.7355 ± 0.0133	0.2259 ± 0.0248	±
	(Treatment 4)			0.35
	Soaked with water and orange		0.1869 ± 0.0232	34.97
	hours+ Rinsed 5 times + Cooked	0.7355 ± 0.0133		±
				0.40
	(Treatment 5)			0.40

Table 2: The total arsenic concentration in all rice samples and arsenic removal efficiencies of the different rice preparation methods

Soaked with water and orange peel powder (150 µm) for 24 hours + Rinsed 5 times + Cooked	0.7355 ± 0.0133	0.1973 ± 0.0056	28.20 ±
(Treatment 6)			0.52

3.2 Arsenic removal efficiencies based on different brown rice preparation methods

Arsenic removal efficiencies based on different brown rice preparation methods ordered by increasing removal efficiency was Treatment 2 (Soaked with water and orange peel powder(75µm) for 1 hour + Rinsed 5 times + Cooked) < Treatment 1 (Rinsed five times + cooked) < Treatment 3 (Soaked with water and orange peel powder(150µm) for 1 hour + Rinsed 5 times + Cooked) < Treatment 6 (Soaked with water and orange peel powder(150µm) for 24 hours + Rinsed 5 times + Cooked) < Treatment 4 (Soaked with water for 24 hours + Rinsed 5 times + Cooked) < Treatment 5 (Soaked with water and orange peel powder(150µm) for 24 hours + Rinsed - Treatment 5 (Soaked with water and orange peel powder(150µm) for 24 hours + Rinsed + Cooked). The average total Arsenic concentration in all treated brown rice samples were successfully reduced to the maximum permitted level of inorganic As recommended by which is 0.35 mg/kg(Figure 5).



Figure 5: The average As removal efficiencies of the different treatments of brown rice

The mean total As content of brown rice samples after undergone treatments ordered by decreasing total As content was Treatment 2(0.2463 mg/kg) (Soaked with water and orange peel powder(75 μ m) for 1 hour + Rinsed 5 times + Cooked) < Treatment 1(0.2425 mg/kg) (Rinsed five times + cooked) < Treatment 3(0.2281 mg/kg) (Soaked with water and orange peel powder(150 μ m) for 1 hour + Rinsed 5 times + Cooked) < Treatment 6(0.1973 mg/kg) (Soaked with water and orange peel powder(150 μ m) for 24 hours + Rinsed 5 times + Cooked) < Treatment 4(0.2259 mg/kg) (Soaked with water for 24 hours + Rinsed 5 times + Cooked) < Treatment 5(0.1869 mg/kg) (Soaked with water and orange peel powder(150 μ m) for 24 hours + Rinsed + Cooked). The average total Arsenic concentration in all treated brown rice samples were successfully reduced to the maximum permitted level of inorganic As recommended by which is 0.35 mg/kg(Figure 6).



Figure 6: The average total As concentration in the brown rice samples based on the different treatments compared to the permissible limits stipulated by FAO/WHO Codex Alimentarius Commission.

3.3 Effect of the application of bio-sorbent of varying sizes for the Arsenic removal in brown rice

Varying sizes of bio-sorbent has effected the results of Arsenic removal in brown rice. Treatment 5 (Soaked for 24 hours with the bio-sorbent size of 75 μ m, followed by rinsing and cooking) removed the greatest amount of total As content (34.97%) in brown rice samples. When comparing the preparation methods that involved soaking with the bio-sorbent (orange peel powder), it can be seen that the total As concentration in brown rice sample that were soaked with orange peel powder of 75 μ m particulate size (Treatment 6) was lower than those soaked with the orange peel powder of 150 μ m particulate size. The average As concentration of samples that underwent Treatment 2 and Treatment 3 were 0.2463 \pm 0.0227 mg/kg and 0.2281 \pm 0.0072 mg/kg respectively, whereas samples that underwent Treatment 5 and Treatment 6 were 0.2259 \pm 0.0248 mg/kg and 0.1973 \pm 0.0056 mg/kg, respectively. A post-hoc test validated that the removal efficiency of Treatment 5 is statistically significantly higher than Treatment 6. This finding justified that bio-sorbent with a smaller particulate size (75 μ m) has a higher As adsorption capacity than the larger particulate size (150 μ m).

Hence, for this study, the optimum size of bio-sorbent for As removal from brown rice was 75 μ m, removing up to 34.97% of total As in brown rice. Previous studies on As removal have also proven similar results. (Sumalatha 2017) investigated the effect of biosorption of As from aqueous solutions using peels of Citrus limonium. Results of their research demonstrated that the percentage of As biosorption decreased with increasing particle size from 0.112 to 0.206 mm. Additionally, based on an adsorption study done by (Amin 2017), the As removal efficiency of orange peels shows a decreasing trend when the size of the bio-sorbent is increased from 45 μ m to 251 μ m. This is because bio-sorbents with smaller size provides a larger surface area for the metal ions to adsorb onto the bio-sorbent. In other words, decreasing bio- sorbent size contributes to increasing metal adsorption (Amin 2017). For these reasons, it is recommended for future studies to explore the effect of bio-sorbent that are smaller than 75 μ m in size to obtain a higher As removal efficiency for brown rice grains.

3.4 Effect of using varying soaking time for the Arsenic removal in brown rice

In term of soaking time of brown rice samples, 1 hour and 24 hours were measured. Treatment 5 (0.1869 mg/kg) and Treatment 6 (0.1973 mg/kg) that were soaked with orange peel powder for 24 hours have lesser As concentrations (p < 0.05) compared to Treatment 2 (0.2463 mg/kg) and Treatment 3 (0.2281 mg/kg) that were soaked with orange peel powder for 1 hour. According to (Zhang 2020), under carefully monitored parameters, soaking can effectively remove As from brown rice, lowering the amount of As consumed by brown rice consumers. In addition, it also mentioned that soaking duration helped in efficiently removing As from brown rice grains and the rate of maximum reduction was around 40 %. Total concentration of As can be significantly removed (p < 0.05) as the soaking duration increases. The soaking, germination, and fermentation processes can

soften grain texture and improve the solubility of minerals in foods, as well as reduces As concentration.

3.5 Effect of rinsing and cooking on the Arsenic removal from brown rice

In the context of rinsing processes, Treatment 1 which involved only rinsing, was effective in removing total As from brown rice. Treatment 1 was the preparation method that achieved As removal efficiency of (11.75%). The average removal efficiency of Treatment 1 was statistically higher than Treatment 2. This result is in oppose to the findings by (Gray 2015) as no As reduction was detected in the rinsed brown rice grains of their study. On the contrary, previous studies have demonstrated positive As removal through washing brown rice samples. For instance, (Liu 2018) experimented the effect of washing on brown rice grains of japonica and indica cultivar and noticed significant reductions in total As levels for all rice grains. (Naito 2015) also discovered that washing brown rice samples for 3 times with deionized water contributes to a reduction of 71-83 % of the total As content.

In terms of the influence of cooking, when evaluating between raw (Treatment 1: 0.2425 ± 0.0125 mg/kg) and cooked (Treatment 2: 0.2463 ± 0.0227 mg/kg) brown rice samples that underwent rinsing, the average As concentration in brown rice grains that were rinsed before cooking, were higher than the raw rinsed brown rice grains (p<0.05). Moreover, the average removal efficiency of Treatment 1 (only rinsing) was statistically significantly higher than the brown rice samples that underwent Treatment 2 (rinsing + cooking) (p<0.05). Treatment 1 removed up to 11.75 %, while Treatment 2 removed a total percentage of 10.37%. Thus, carrying out an additional cooking process after the rinsing step did not further enhance the removal of total As in brown rice. This result is similar to the findings reported by (Naito 2015). In their study, the cooked brown rice grains were prepared under typical Japanese conditions. The brown rice grains were firstly washed and cooked in low-volume (rice:water 1:1.4–2.0) until complete dryness. Comparison of total As concentrations between cooked washed rice and raw washed rice revealed that these levels were nearly equal. In other words, subsequent cooking after rinsing did not significantly reduce the As levels in brown rice samples.

4. Conclusion

In this study, the total As concentration of local Malaysian commercially available brown rice $(0.7223 \pm 0.0144 \text{ mg/kg})$ exceeded the FAO/WHO Codex Alimentarius Commission permissible limits (permissible limit for brown rice: 0.35 mg/kg). As brown rice exceeded the maximum levels recommended by FAO/WHO, it can be perceived that there is a potential human health risk through the long-term consumption of local Malaysian commercially available rice. The application of the orange peel bio-sorbent had successfully reduced the total As levels of brown rice samples to below stipulated permissible limits. Treatment 5, which involved soaking with the bio-sorbent of 75µm in size for 24 hours, followed by rinsing and cooking, achieved the highest removal efficiency for brown rice samples. Treatment 5 removed up to 34.97% of total As from brown rice grains. The removal efficiency of Treatment 4 was also higher than the treatment method Treatment 6 that incorporated a larger bio-sorbent size (150µm). Hence, it is justified that the optimum size of the orange peel bio-sorbent for removal of As from brown rice grains is 75µm. The results of brown rice samples also prove the potential of orange peels as a bio-sorbent in reducing total As in brown rice grains, thereby safeguarding human health. In conclusion, both the objectives of this study is achieved.

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