MULTIELEMENT ANALYSIS IN RICE GRAINS BY INSTRUMENTAL NEUTRON ACTIVATION ANALYSIS

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ABSTRACT

Over 114 countries in the world grow rice and more than 50 countries have an annual rice production of 100,000 tonnes or more. Asian farmers produce about 90% of the global total rice production. Generally, there are two most common varieties of rice; cultivated and hill rice. Nowadays a lot of agriculture land is contaminated with toxic elements owing to the use of sludge or municipal compost, pesticides, fertilizers and emissions from municipal waste incinerators, car exhausts, residues from metalliferous mines, and smelting industries. The distribution and concentration of several toxic elements in grains particularly rice has lately become a big concern. A study to determine the concentrations of some elements in a few varieties of rice in our local market using Instrumental Neutron Activation Analysis has been performed by Waste and Environmental Technology Division, Malaysian Nuclear Agency. A total of 15 elements were measured. The method was validated by analysing the Standard Reference Material SRM-1568a (Rice Flour) and SRM-1573a (Tomato Leaves) of NIST. The measured concentrations of major and minor elements were analysed in terms of the average intake of nutrient content and comparison of several toxic elements to other studied values.

ABSTRAK

Lebih 114 negara di dunia menanam padi dan lebih daripada 50 negara mempunyai pengeluaran tahunan beras sebanyak 100,000 tan atau lebih. Petani di Asia mengeluarkan sekurang-kurangnya 90% jumlah pengeluaran global beras. Lazimnya terdapat dua variati beras; beras yand di tanam di sawah dan bers bukit. Kini terdapat banyak tanah pertanian yang dicemari dengan elemen toksik disebabkan penggunaan enapcemar atau baja dari bandaran, racun serangga, baja, pelepasan dari insinerasi bahan buangan bandaran, pelepasan kenderaaan, sisa dari lombong dan industry peleburan. Taburan dan kepekatan beberapa toksik elemen dalam bijirin terutamanya beras telah mendapat perhatian *kebelakangan ini. Satu kajian telah dijalankan oleh Bahagian Teknologi Sisa dan Alam Sekitar, Agensi Nuklear Malaysia bagi menentukan kepekatan elemen dalam beberapa jenis beras yang tedapat dalam pasaran tempatan menggunakan analisis pengaktifan nutron instrumental. Sejumlah 15 elemen telah dibuat pengukuran. Pengesahan kaedah dilakukan dengan menganalisis Bahan Rujukan Piawai SRM-1568a (Rice Flour) dan SRM-1573a (Tomato Leaves) dari NIST. Kepekatan elemen major dan minor yang telah diukur dianalisis kandungan nutrisi purata pengambilan and perbandingan beberapa elemen toksik dengan kajian lalu.*

Keywords: neutron activation analysis, elements, rice

INTRODUCTION

Rice or its scientific name *Oryza sativa* is the staple food for half of the world's populations. At least 114 countries grow rice and more than 50 have an annual production of 100,000 tonnes or more (FOA, 2002). In 2000, Asian farmers produce about 91% of the global total. FOA reported in 2000, there are about 594 million tonnes of paddy produced annually over roughly 1.54 million sq km. The top producer of rice are China , Indonesia , Vietnam, Bangladesh, Myanamar, India, Thailand in order of volume.

There are a lot variety of rice. The most popular is cultivated rice. Cultivated rice is generally considered a semi-aquatic annual grass, although in the tropics it can survive as a perennial, producing new tillers from nodes after harvest. A rice plant takes from three to six months to reach maturity, depending on the variety. There is also floating paddy, dry paddy, and hill paddy which are grown on slopes and terraces in various Asian countries including China, the Philippines and East Malaysia. Rice variations show differences in nutrient contents. According to Barikmo *et al.* (2007), the differences exist for the same food due to different ecological zones.

Nowadays, more land is being contaminated with toxic elements owing to the use of sludge or municipal compost, pesticides, fertilizers and emissions from municipal waste incinerators, car exhausts, residues from metalliferous mines, and smelting industries (Gupta *et al*., 2008, Rogan *et al*., 2009). Since toxic elements in the environment can be harmful to human health through the food chain public concern about the environmental impact has grown in recent decades. Grains and cereal particularly rice, wheat, maze and barley are among the main dietary food for supplying trace elements and nutrients. The distribution and concentration of several toxic elements in those grains become highly concerned.

Therefore, the major aim of the present study was to determine the concentration of elements in variety of rice planted or marketed locally as this grain become the staple food for Malaysians. Several potentially toxic elements were also observed in this study.

METHODOLOGY

Twelve various type of rice sample were collected for this study. These rice can be categorized into three group; i) cultivated rice ii) hill rice and iii) other variety. The cultivated rice originated from Peninsular Malaysia, Sabah, Pakistan and Thailand. The hill rice mostly grew at Sabah and Sarawak highland. Other variety of rice came from Indonesia and Sarawak. Group of rice variety was presented in Table 1.

Table 1: Group of rice from local market

Group of rice	Variety
Cultivated Rice	Village Rice Local Rice Thailand Rice Basmathi Rice
Hill Rice	Hill Rice Bario Keladi Rice Red Bario Rice A White Bario Rice Red Bario Rice B
Other variety	Black Glutinous Rice Husk Open Rice Brown Rice

Rice sample collection and preparation

About 1 kg of rice sample was obtained from local market. Rice grains were washed three times with deionized (DI) water and then dried in a hot-air oven at 60 ± 2 ^{0}C for 4 to 6 hours. After that, the samples were ground to a homogeneous fine powder using a high speed blender in which all materials were made from titanium (Ti) to reduce contamination. The grinding step was repeated until fine powder passed completely through a No. 60 mesh sieve. Samples were weighed 0.2 to 0.3 g into polyethylene vials and heat-sealed prior to irradiate. Certified reference materials (CRM) were co-irradiated with the sample, standard and blank to ensure accuracy and precision of data.

Rice sample irradiation

The irradiations were performed in the TRIGA Mark II reactor at Malaysian Nuclear Agency. For short irradiation, all samples, standard, blank and CRM were irradiated for a period of 60 seconds, and the cooling time for 20 min for first counting, and the second counting conducted after 24 hrs. In long irradiation, each batch containing the rice samples, standard, blank and were irradiated for 6 hrs. The cooling times took 3 days for first counting, and 3 weeks for the second counting. A high resolution coaxial CANBERRA hyper-pure germanium detector (HPGe) with a resolution of 1.9 keV at 1332 keV gamma-rays line of Co-60 was used for counting. The spectral data were processed using the spectrum analysis software namely GINIE 2000. Computations of elemental concentrations were based on relative method and data were reported in dry weight (d.w.).

RESULTS AND DISCUSSIONS

Quality assessment

Certified reference material namely Tomato Leave 1573a and Rice Flour 1568a provided by National Institute of Standard and Technology were applied as quality control material of each analytical regiment implemented. Results obtained for the CRMs are presented in Table 2. The analytical result for most of the elements are in good agreement with their certified values as their errors calculated base on the average values are below 15%. Analytical results disclosed that the precision of the analysis is satisfactory where percent RSD are in the range of 1 – 15%.

Elements in rice sample

The analysis results of 15 elements in rice samples were presented in Table 3. For essential macronutrient particularly Cl, K, Mg and Na, other variety of rice such as Black Glutinous Rice, Husk Open Rice and Brown Rice showed relatively high amount of K and Mg compared to cultivated and hill rice. The cultivated rice except for Basmathi Rice showed no Mg is presented in the sample. Meanwhile, K and Mg were noted to be of higher concentration for hill rice.

We have compared the Dietary Reference Intakes (DRI) for some of the known major essential elements with that values obtained from this work based on an intake of element per day by average age of male and female (Institute of Medicine,1997). The DRI value is presented in Table 4. The potassium value of 2207mg/day was the highest value and corresponds to the concentration of obtained from Black Glutinous Rice. But this value was still below the recommended value of 3500mg/day. The concentration of Mg of most white rice samples was below the recommended dietary value (400 mg/day for male and 310 mg/day for female). The determined values of Na and Cl in the majority of the samples were lower than the DRI value.

The essential micronutrient are As, Co, Cr, Fe, Mn and Zn. The Black Glutinous Rice from Indonesia was found to have highest amount of Co, Cr, Fe and Zn compared to other variety. Below limit of detection level of Cr was recorded for Local Rice, Hill Rice, Husk Open Rice and Brown Rice. Manganese was noted to be of higher concentration in the Red Bario Rice, Black Glutinous Rice and Husk Open Rice. Very low amount of Fe was observed in Local Rice, Thailand Rice and Hill Rice. The Husk Open Rice and Brown Rice showed significantly higher As. For cultivated rice, significant amount of As was observed in Thailand Rice.

The majority of rice samples provide adequate amount of Cr, Zn and Fe for daily requirement intake. Even though several rice samples determined higher concentration of Cr than of DRI value, considerably 200 µg/day level of Cr can be consumed in daily intake. (http://www.healingwithnutrition.com/mineral.html). The concentrations of Mn in most rice samples were considerably higher than the DRI value. However, Mn has the lowest toxicity of all metals (Reilly, 1980) and an excessive amount of more than 1000 µg/g intake is required to produce any toxic effects in human.

High amount of Br present in Village Rice and Local Rice. Bromine concentration in cereals is dependent upon the soil conditions. Organic matter is known to accumulate Br and its enrichment in the top soil horizons is principally an effect of its precipitation with rain (Balaji *et al*., 2000). Other also reported that the presence of higher levels of Br in agricultural products is due to the application of agricultural chemicals such as methyl bromide used as a fumigant (Yamada, 1968). Cultivated rice that grows in watery soil could explain for the high level Br in those samples.

Soil pollution with potentially toxic elements resulting from rapid industrial development has caused major concerns. Excessive toxic elements in agriculture soil may restrain the growth of crops, affect their quality and safety throughout the food chain. A comparison of elements determined in this study to other reported amount of elements in rice are shown for As, Cr, Fe, Mn and Zn in Table 5.

The concentration of As, Cr and Zn were noted lower compared to those from China and Macedonia. According to Marin *et al*.(1993), food surveys had revealed that rice accumulates the highest amount of As of all cereals and mostly because of the high bioavailability of As under reduced soil conditions. Several studies showed that unpolished rice like husk open rice and brown rice contained higher amount of As than white rice due to the localization of inorganic As in the bran layer (Ren et al., 2006, Rahman et al., 2007, Meharg et al., 2008). The high amount of Cr and As in rice sample from China that came from Changshu area may be derived from some industries and pesticide use (Hang *et al*., 2009). In Kocani rice field, Macedonia, the highest concentrations of As and Zn were measured in rice grown in the most impacted soil that is close to the Zletovska River which been polluted by mining activities and acid mine drainage (Rogan *et al*., 2009). In current study, significantly high amount of Fe, Mn and Zn were observed than the rice of Pakistan. These elements have great correspondence between their content in rice grains and corresponding elements in the soil (Lin *et al*., 2009).

Table 2: Results of analysis of certified reference materials.

Elements	Measurement value	Certified or recommended value	Error (%)	RSD (%)
Tomato Leave 1573a				
Br	1114 ± 36	1300	-14.3	3.2
Cl	5800 ± 115	6600	-12.1	2.0
Co	0.65 ± 0.08	0.57 ± 0.02	14.0	12.3
Cr	1.84 ± 0.09	1.99 ± 0.06	-7.5	4.9
Cs	0.05 ± 0.007	0.053	-5.7	14.0
Fe	365 ± 16	368 ± 7	-0.9	4.5
Hf	0.14 ± 0.01	0.14	0	7.1
K	25650 ± 495	27000 ± 500	-5.0	1.9
Mg	11424 ± 913	12000	-4.8	8.0
Mn	238 ± 10	246 ± 8	-3.3	4.2
Na	124 ± 15	136 ± 4	-8.9	12.1
Rb	13.1 ± 0.8	14.89 ± 0.27	-11.9	6.1
Zn	27.7 ± 2.7	30.9 ± 0.7	-10.4	9.7
Rice Flour 1568a				
As	0.29 ± 0.04	0.29 ± 0.03	1.6	13.6
Br	7.00 ± 0.24	8.00	-12.5	3.4
Cl	273 ± 17	300	-9.0	6.3
Fe	8.5 ± 0.9	7.4 ± 0.9	14.9	10.1
K	1100 ± 110	1280 ± 80	-14.1	10.0
Mn	19.1 ± 0.2	20 ± 1.6	-4.4	1.0
Na	7.5 ± 1.1	6.6 ± 0.8	13.6	14.7
Rb	5.96 ± 0.15	6.14 ± 0.09	-3.0	2.5
Zn	18.0 ± 0.6	19.4 ± 0.5	-7.4	3.3

Table 3: Elemental concentration (mg/kg dry weight) in the rice samples.

	As	Br	Cl	Co	Cr	Cs	Fe	Hf
Village Rice	0.11±0.02	17.6±0.4	277±8	0.024±0.002	0.08±0.01	0.020±0.001	5.0±0.2	0.010±0.003
Local Rice	0.11±0.01	9.55±0.30	173±9	0.028±0.002	< 0.08	0.011±0.002	< 5.0	0.010±0.003
Thailand Rice	0.15±0.01	0.18±0.01	164±6	0.032±0.010	0.08±0.01	0.021±0.007	< 5.0	0.013±0.007
Basmathi Rice	0.08±0.01	0.52±0.01	300±5	0.029±0.002	0.11±0.02	< 0.010	8.6±0.5	0.012±0.004
Hill Rice	0.10±0.02	0.36±0.01	107±5	0.027±0.001	< 0.08	0.028±0.002	< 5.0	0.010±0.001
Bario Keladi Rice	0.13±0.01	0.82±0.01	75±1	0.026±0.002	0.09±0.01	0.020±0.003	7.7±2.5	0.010±0.005
Red Bario Rice A	0.16±0.01	0.52±0.01	147±4	0.033±0.002	0.19±0.04	0.094±0.016	16.4±2.3	0.012±0.002
White Bario Rice	0.10±0.01	0.48±0.01	178±1	0.024±0.001	0.20±0.01	0.034±0.001	8.8±4.3	0.013±0.008
Red Bario Rice B	0.12±0.01	0.71±0.02	146±2	0.035±0.001	0.08±0.04	0.066±0.009	12.3±0.6	0.012±0.003
Black Glutinous Rice	0.13±0.01	0.51±0.01	233±2	0.145±0.006	0.33±0.01	0.185±0.026	30.5±6.9	< 0.006
Husk Open Rice	0.19±0.03	0.63±0.01	131±1	0.028±0.002	< 0.08	0.053±0.011	15.6±2.7	0.010±0.004
Brown Rice	0.19±0.01	0.19±0.01	168±6	0.031±0.002	< 0.08	0.173±0.010	9.7±3.3	0.010±0.001

	K	Mg	Mn	Na	Rb	Sb	Zn
Village Rice	585±48	< 150	6.1±0.4	23.4±2.0	1.8±0.1	< 0.003	9.2±0.6
Local Rice	560±30	< 150	6.3±0.8	3.9±0.3	2.4±0.1	< 0.003	11.0±0.5
Thailand Rice	427±4	< 150	5.8±0.4	3.4±0.1	3.8±0.1	0.150±0.005	10.1±0.1
Basmathi Rice	530±28	274±17	5.9±0.2	6.0±0.3	0.7±0.1	0.127±0.006	11.1±0.2
Hill Rice	532±18	248±15	8.1±0.5	3.7±0.8	2.3±0.1	0.091±0.004	9.9±0.4
Bario Keladi Rice	556±7	586±96	10.2±0.9	2.7±0.1	9.4±0.3	0.007±0.001	13.1±0.2
Red Bario Rice A	974±29	1025±31	25.9±0.2	5.4±0.4	18.2±2.3	0.147±0.006	14.7±0.4
White Bario Rice	231±32	241±15	7.4±0.4	2.7±0.1	3.3±0.3	0.083±0.031	11.4±0.2
Red Bario Rice B	869±23	511±17	12.8±1.2	4.1±0.1	9.2±0.2	0.010±0.001	14.7±0.2
Black Glutinous Rice	2207±107	1072±110	25.6±1.6	79.2±1.1	16.7±0.4	0.093±0.007	18.1±0.3
Husk Open Rice	1874±24	1375±152	25.5±1.3	7.1±1.0	7.8±0.1	0.010±0.003	12.7±0.3
Brown Rice	1576±124	1094±202	16.1±0.3	4.6±0.2	11.3±0.8	0.010±0.001	16.1±2.7

Table 4: Dietary Reference Intake: Recommended intake for elements per day of individual in average age of 19 to 30.

Elements	**Male**	**Female**
Chromium (μg)	35	25
Chloride (mg)	2300	2300
Iron (mg)	8	18
Magnesium (mg)	400	310
Manganese (mg)	2.3	1.8
Potassium (mg)	4700	4700
Sodium (mg)	1500	1500
Zinc (mg)	11	8

Source: (Institute of Medicine,1997)

Table 5: A comparison study of elements concentration

	This study			**China** (Hang et al., 2009)			**Pakistan** (Sarfraz et al., 2007)			**Macedonia** (Rogan et al., 2009)		
	Mean mg/kg	**Min mg/kg**	**Max mg/kg**	**Mean mg/kg**	**Min mg/kg**	**Max mg/kg**	**Mean mg/kg**	**Min mg/kg**	**Max mg/kg**	**Mean mg/kg**	**Min mg/kg**	**Max mg/kg**
As	0.13	0.08	0.19	0.199	0	0.587	nd	nd	nd	0.28	0.15	0.53
Cr	0.14	<0.08	0.20	0.29	0.03	0.74	nd	nd	nd	nd	nd	nd
Fe	12.72	<5	30.49	nd	nd	nd	5.26	5.14	5.30	nd	nd	nd
Mn	12.96	5.74	25.88	nd	nd	nd	1.50	1.48	1.53	nd	nd	nd
Zn	12.67	9.17	18.1	19.1	9.10	28.3	2.74	2.50	2.98	28	13	67

CONCLUSIONS

Rice variety that has been collected into the same group showed similar elemental distribution for several elements. However, each variety demonstrate specific elemental characteristic. Soil and climatic differences could directly affect the elemental content of plant origin. On the other hand bioavailability of elements depends on several other factors such as chemical state, fibre content, the presence of complexing agents accompanying the intake and the extent to which other interacting elements are present or absent from the diet. Elemental toxicity in man arising from excess intake of food is not frequently reported unless there is an occurrence of industrial or environmental contamination. Analysis of food items not only provides data on nutritional surveillance programmes, but also on the contamination brought in by several indiscriminating activities.

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