Spectroscopic Determination of Essential Elements in Unpolished Rice (*Oryza sativa* L.), Grown at RRI, Dokri

[1]G. Q. SHAR, T[2]. G. KAZI, [2]S. SAHITO, [1]S. A. ARAIN AND [1]L. A. SHAR
[1]*Department of Chemistry*
Shah Abdul Latif University Khairpur (Mirs)
Sindh, Pakistan
[2]*Centre of Excellence in Analytical Chemistry*
University of Sindh Jamshoro, Pakistan

(Received 31st January, 2004, revised 7th June, 2004)

Summary: Seven rice varieties collected from, Rice Research Institute (RRI) Dokri, Sindh in Pakistan. Agricultural soil samples was also collected to evaluate the bioavailability of eight essential elements to seven varieties of rice grown in the same agricultural plot. The agricultural soil and rice varieties were digested by wet acid digestion method. Homogeneity of the prepared materials was evaluated through the determination of eight essential elements i.e., Na, K, Mg, Ca, Fe, Mn, Zn, using flame atomic absorption spectrophotometer.

The unpolished rice varieties contained high amount of essential micronutrients such as Na, K, Mg, Ca, Fe, Mn, Zn, and Cu (616.01, 4097.15, 2626.20, 73.90, 1829.88, 50.34, 42.72 and 7.21 mg/kg) in seven different varieties of rice i.e. DR-82, DR83, DR92, Kanwal-95, Lateefi, Sadahayat and Sarshar respectively.

Introduction

Many studies have recognized the role of essential elements in human metabolism [1-3]. Macro-elements, e.g., Na, K, Mg, Ca, and micro-essential elements, e.g., Fe, Zn, Mn, are vitally important for certain biochemical systems, whereas trace and toxic elements, e.g., Cd, Cr, Co, Ni, Pb if present in relatively high amounts, adversely affect these systems [4,5]. The concentrations of essential, non-essential and trace and toxic elements in the human body are affected by a number of factors such as nutrition, sex, age, retention, chemical form of the trace elements and binding sites available to them. The intake of these elements is related to our environment; however, food is the main source. It is thus significant to assess the adequacy and safety of the diet by monitoring the concentration of essential and toxic trace elements in various food articles and integrated human diets [6-14]. Dietary inadequacies of trace elements are estimated to affect the health, mental and physical function and survival of more than two billion people worldwide. Women of child-bearing age and children are mostly at risk [15]. Inorganic nutrient metabolism traditionally has been investigated by the chemical balance technique. A controlled diet is administered over a period of time and the inorganic content of the diet and of fecal and urinary samples is then measured in order to estimate the absorption and retention of the element of interest [16]. The harmful effects of Pb and Cd have been well-documented [17-19]. Soil status and response of micronutrient to different crops are well studied [20]. Analytical results on trace elements have been reported on one sample for given species in the many of the past reports. However it would be difficult to assign to a simple value as the representative one for that species, since the variability of the concentrations seems to exist among the samples originating in different localities, depending on elemental characteristics. Trace element distributions among different parts of plants and their physiological roles are also of other interest. Cereal especially rice is staple food in most of the developing countries including Pakistan. They also constitute a major source of animal feed. Although they contribute most significantly as a source of carbohydrates and proteins, their potential contribution of minerals including trace elements is often overlooked. In the absence of recent systematic data on elements, it would be very difficult to determine the adequacy of our diet with respect to minerals, especially trace elements. Keeping in view their importance in human diet, prevalent varieties and new genotypes of rice were analysed in our laboratory for important macro- and microelements.

The present investigation was undertaken to analyse and assess the amount of various essential nutrient elements in seven rice varieties. These were

Table-1:Concentrations of Essential Elements Present In Unpolished Rice (mg/kg).

Var.	Na	K	Mg	Ca	Fe	Mn	Zn	Cu
R1	436.12	3558.93	2626.20	48.70	773.81	21.08	36.4	6.70
	±42.44	±431.66	±55.94	±2.68	±59.70	±1.43	±3.45	±0.30
R2	476.70	4097.15	2593.4	73.90	1829.88	44.09	40.7	7.21
	±42.44	±482.02	±159.57	±7.49	±94.7	±7.34	±2.58	±0.53
R3	550.19	3718.72	2317.26	55.19	1306.6	24.20	42.72	4.14
	±29.10	±464.79	±361.2	±1.03	±150.7	±4.15	±5.5	±0.25
R4	495.35	3096.41	1927.19	63.73	375.38	35.28	32.38	3.01
	±35.37	±492.37	±128.2	±7.67	±48.0	±12.9	±4.9	±0.35
R5	485.48	3247.78	1728.7	56.06	1345.1	23.92	28.35	3.15
	±11.54	±355.33	±163.62	±5.05	±149.9	±3.13	±1.1	±0.19
R6	462.44	3273.01	2136.03	71.19	1637.86	50.34	36.29	6.26
	±15.62	±124.36	±124.8	±5.0	±36.8	±6.88	±1.7	±0.73
R7	616.01	2902	2075.62	40.16	276.98	42.10	36.29	5.02
	±80.02	±156.58	±86.85	±3.26	±21.29	±1.94	±3.1	±0.25
Soil	4272.1	14073.3	39553.7	3104	5066.3	399.36	82.61	22.22
	±519.3	±412.7	±4306.8	±385.5	±475.3	±32.3	±7.1	±1.78

Var. = Varieties
R1= DR-82, R2= DR-83, R3= DR-92, R4= Kanwal-95
R5= Lateefi, R6= Sadahayat, R7= Sarshar

collected from rice research institute (RRI) Dokri as a representative sample.

Results and Discussion

The results of essential nutrients of seven rice varieties are reported in the Table 1. Highest amount in mg/kg of sodium was detected (616.01±80.02) in the Sarshar variety and lowest amount (436.12± 54.68) was spotted in samples of the DR-82. Level of the sodium in DR-83 and Sadahayat is close to one an other. Similarly the concentration of sodium in Kanwal-95 and Lateefi are with the margin of 10mg/kg, which shows similar characteristics of the absorption of sodium in these varieties. Potassium is one of the principal elements, which carries the highest concentration as compared to all nutrients in Table 1. Among the seven varieties, the maximum concentration (4097.15±482.02mg/kg) of potassium was occupied by the DR-83 variety on the same plot where all these varieties were grown. In case of minimum concentration, the Sarshar shows its bottom level i.e.2902.99±156.58 of the potassium. The uptakes of potassium in Lateefi and Sadahayat varieties are with a little difference. However, there is a much and more difference in the uptake of potassium in rest of rice varieties. It may be due to its genetic characters.

High magnesium level (2626.20±55.94) was detected in DR-82 and low (1728.71±163.62) in Lateefi and the value of other five that lie between both of these varieties. The content of the magnesium in DR-83 is very close to the DR-82 variety, which is important for providing the maximum quantity of the magnesium. Maximum concentration of calcium (73.90±7.49) was found in DR-83, where as the low quantity (40.16±3.26) was determined in the Sarshar variety. There is no significant difference of the content of the calcium in DR-83 and Sadahayat.

Most of the microelements are very important and their requirement is as much essential as that of macro elements, but the concentration of these are sufficiently less as compared to the macro- nutrients. Among these micro- nutrients iron, manganese, zinc and copper are the most important ones. In this regard DR-83 was on top of the iron content (1829.88±94.72mg/kg) and Sarshar was at the bottom (276.98±21.29 mg/kg). Rest of the varieties has a lot of difference in the absorption of iron from its soil, which shows that physiological role has its own importance. The highest level (50.34±6.88 mg/kg) of manganese was present in Sadahayat and the lowest amount was observed in DR-82 i.e. (21.08± 1.43mg/kg), whereas DR-92 and Lateefi varieties possess insignificant difference in the content of manganese. The highest amount of Zn was observed in DR-92 (42.72±5.48) and the lowest amount of Zn was found in Latefi (28.35±1.121mg/kg), grown at the same plot. Three cultivars of this location i.e. DR-82, Sadahayat and Sarshar have same concentration of zinc.

The level of copper is lower as compared to all macro and micro- nutrients in all mentioned varieties. The value of copper exhibits no significant difference in DR-82 and Sadahayat. Similarly, no significant difference was observed in Kanwal-95

Table-2:Instrumental Conditions for the AAS Measurement of Na, K, Ca, Mg, Fe, Zn, Mn and Cu

Elements	Wave length (nm)	Slit width (nm)	Lamp current (mA)	Fuel flow (acetylene) (L/min)	Flow rate (Air) (L/min)	Burner height (mm)	Oxidant (Air) kg/cm^2	Fuel (Acetylene) kg/cm^2	Signal out put
Na	590	0.4	9.5	2.21	9.4	7.5	1.60	0.25	100%
K	766.8	2.6	9.5	2.3	9.4	7.5	1.60	0.3	100%
Ca	422.2	2.6	7.3	2.6	9.4	12.5	1.60	0.4	100%
Mg	285.5	2.6	7.0	2.0	9.4	7.5	1.60	0.2	100%
Fe	248.3	0.2	9.5	2.30	9.4	7.5	1.60	0.3	100%
Zn	214.0	1.3	9.5	2.0	9.4	7.5	1.60	0.2	100%
Mn	279.8	0.4	9.5	2.0	9.4	7.5	1.60	0.2	100%
Cu	325.0	1.3	9.5	2.0	9.4	7.5	1.60	0.2	100%

and Lateefi, but most significant difference was seen in DR-83 and Lateefi.

The fifty soil samples collected form the agricultural plot of the Rice Research Institute, Dokri possesse 4272.1±519.29, 14073.3±412.7, 39553.7±4306.8, 3104.0±385.5, 5066.3±475.3, 399.36±32.25, 82.61±7.07, 22.22±1.78 mg/kg of sodium, potassium, magnesium, calcium, iron, manganese, zinc and copper respectively.

Experimental

Sample Collection

Samples of paddy (DR-82, DR-83, DR-92, Kanwal-95, Lateefi, Sadahayat, and Sarshar) varieties were collected from Rice Research Institute, RRI, Dokri as a representative sample.

Samples of paddy were brought to the Nuclear Institute of Agricultural Tandojam for dehusk, where these were dehusked by dehusker machine into brown rice form. These varieties were packed into polythene bags and brought to the laboratory of the Centre of Excellence in Analytical Chemistry, University of Sindh Jamshoro, Pakistan where they were washed with distilled/deionized water and finely oven dried at 110^{0}C to constant weight.

The fifty samples of clay loam soil was collected from experimental field of Rice Research Institute (RRI) Dokri, (Sindh) Pakistan where all these seven rice varieties were grown. The soil was air-dried and sieved (<0.5mm).

Contamination Control

All glassware were soaked in chromic acid for 24 hours and washed with water oven dried and stored in dust and fume free atmosphere without touching their insides.

Pre-treated filter papers were used to filter digested rice samples. For this purpose they were soaked in 0.1 M (approximate) disodium salt of EDTA and washed with water to remove suspected metal impurities. The process was continued until washings were free from traces of sodium.

Procedure

All seven rice varieties samples were dried at 105°C in oven. Replicate 2g of each rice variety were weighed in to 100ml conical flask.

Five rice varieties were dried at 105°C in oven. Replicate 2g sample of each rice variety was weighed in to 100 ml conical flasks and treated with 5 ml of nitric acid. 5ml of nitric acid was also added to empty conical flask serving as a blank. The flasks were covered with watch glasses, and their contents were heated to reflux gently on an electric plate. After refluxing for one hour, 5 ml of nitric acid and , 2 ml of 35% hydrogen peroxide was added to the contents of flasks, and the gentle reflux was continued for another hour [21,22]. The watch glasses were removed from the flasks, and heating was continued until the volumes of their contents were reduced to 2-3 ml. The contents of flask were cooled, diluted with high purity water, and filtered through whatman # 42 paper in to 25 ml volumetric flasks. The contents of the flasks were brought to volume with high purity water and examined by atomic absorption spectrometry for their sodium, potassium, magnesium, calcium, iron, manganese, zinc, and copper levels.

Instrumentation

A Hitachi Model 180-50 atomic absorption spectrophotometer equipped with standard burner and air-acetylene flame was used for the determination of elements such as, sodium, potassium, calcium, magnesium, iron, zinc, manganese and copper. The

Table-3:Statistical Data for Standards of Elements

Elements	Concentration range ppm (x)	Absorbance (y)	Statistical calculation y = m x + c		
			m	c	r^2
Na	0 - 0.25	0 - 0.084	0.3344	0.0001	0.9996
K	0 - 1	0 - 0.207	0.2069	0.0006	0.9999
Ca	0 - 5	0 - 0.256	0.0508	0.0024	0.9993
Mg	0 - 1	0 - 0.885	0.8856	0.0017	0.9999
Fe	0-1	0 - 0.096	0.0976	-0.0016	0.9989
Zn	0 - 0.5	0 - 0.138	0.2761	-0.0004	0.9999
Mn	0 – 1	0 – 0.196	0.1962	0.0005	0.999
Cu	0 – 1	0 –0.086	0.0862	0.0004	0.9989

hollow-cathode lamps (made by Mtiorika company) of all above elements were operated at lamps current 9.5, 9.5, 7.3, 7.0, 9.5, 9.5, 7.0 and 7.0 mA respectively. The flow-rate for fuel 2.30 L min^{-1} and air 9.40 L min^{-1} was used respectively to obtain a clear yellow flame (reducing condition). The spectrophotometer out put was connected to a Hitachi recorder 056 with a range of 5mV. The signals measured were the heights of the absorbance. All instrumental parameters are given in Table 2.

Reagents and Calibration

The supra pure nitric acid (65% w/v) and hydrogen peroxide (35% w/v) reagents (Merck), high-purity water (electrical resistivity $\rangle$ 10mΩ cm) was produced with a Milli-Q system Millipore, MA, USA).

Calibration was obtained with external standards. The standards solutions were prepared by diluting a 1000mg/L multi element solution (ICP Multi element standard iv, Merck, Darmstadt, FRG) with the same acid mixture used for sample dissolution. Glassware were cleaned by soaking with the contact over night in a 10% (w/v) nitric acid solution and then rinsed with deionized water.

Solutions were aspirated into atomic absorption spectrophotometer and absorbance measurements were made for each element using optimum instrumental conditions for flame atomization mode.

Reference standards were also run in parallel for inter calibration of our own standards. Elemental concentration were computed on an IBM compatible PC using Excel software program.

The statistical calculations for standards are given in Table 3.

Conclusions

It was concluded that highest concentration of sodium was found in Sarshar; potassium, calcium, iron and copper was detected in DR-83 cultivar; magnesium and zinc was detected in DR-82 and DR-92 respectively. Similarly highest content of manganese was observed in Sadahayat variety. The lowest content of sodium and manganese was found in DR-82 cultivar. The lowest level of magnesium and zinc was observed in Lateefi variety. In the same way the lowest contents of potassium, calcium and iron were determined in Sarshar; and copper in Kanwal-95 variety.

AcknowledgementS

Ghulam Qadir Shar is thankful to Syed Iqrar Hussain Shah and Muhammad Aslam Nushad (Seed Certification Officers), Seed Certification & Registration Department for their co-operation in sampling of the paddy rice from RRI Dokri, District Larkana. Thanks are also due to my parent institute Shah Abdul Latif University, Khairpur, Sindh, Pakistan for financial support and grant of study leave.

References

1. B.L.O'Dell, R.A. Sunde (Eds), "Handbook of Nutritionally Essential Mineral Elements", Marcel Deller, Inc., NewYork, (1997).
2. N.R. Maturu, IAEA Technical Report, Series No. 197, Vienna, (1980).
3. A.S. Prasad, "Trace Elements in Human Health and Diseases" Vols.**1** and **2**, Academic Press, New York, USA., (1976).
4. S.J. Khurshid, I.H. Qureshi, *Nucleus*, **21**, 3 (1984).
5. H.R.Roberts, "Food Safety" John Wiley & Sons, NewYork, Chap. 3 (1981).
6. Y. Kagawa, supervised for the "4th compilation Standard Tables of Food Composition in Japan"

Kagawa Nutrition College, Publ. Dept., Tokyo (1998).

7. Y.Suzuki, S.Tanushi (Eds), "Table of Trace Element Content in Japanese Foodstuffs". Daiichi Shuppan Co. Ltd., Tokyo, (1993).
8. G.F. Clemente, *J. Radioanal. Chem.*, **32**, 25 (1976).
9. D.L. Samudralwar, A.N. Garg, Frescenius *J. Anal. Chem.*, **348**, 433 (1994).
10. V. Singh, A.N. Garg, *J. Radioanal. Nucl. Chem.*, **217**, 139 (1997).
11. V. Singh, A.N. Garg, *Private Communication*, (1998).
12. I.H. Qureshi, J.H. Zaidi, M. Arif, A. Mannan, I. Fatima, *Intern. J. Environ. Anal. Chem.*, **44**, 265 (1991).
13. I.H. Qureshi, A. Mannan, J.H. Zaidi, M. Arif, N. Khalid, *Intern. J. Environ. Anal. Chem.*, **38**, 565 (1990).
14. J.H. Zaidi, I.H. Qureshi, M. Arif, A. Mannan, I. Fatima, *Intern. J. Environ. Anal. Chem.*, **60**, 15 (1995).
15. R.S. Gibson, F. Yeudall, C. Hotz and E.L. Ferguson, *New Zealand Dielectric Association Conference Proceedings*, 72 (1998).
16. M. Kamao, N. Tsugawa, K. Nakagawa, Y. Kawamoto, K. Fukui, K. Takamatsu, G. Kuwata, M. Imai, and T.J. Okano, *Nutr. Sci. Vitaminol*, **46**, 34 (2000).
17. C. Reilly, "Metal contamination of food" Ist Edition, Chap.5 and 6, (Applied Sci. Publishers, London, 1980).
18. E.J. Underwood, "Trace elements in human and animal nutrition" 4th Edition, Chap. 9 and 17, (Academic Press Inc. New York, (1977).
19. S.J. Khurshid and I.H. Qureshi, The *Nucleus*, **21**, 3 (1984).
20. S. Yashida and A. Tanaka, *Soil Sci. Plant Nutr.*, **15**, 75 (1969).
21. A.F. Ward, L.F. Marciello, C.Carrara and V.J. Luciano, *Spectrosc. Letters*, **13**, 803 (1980).
22. A.Abu-Samara, J.R. Morris and S.R.Koityohann, *Anal. Chem.*, **47**(8), 1475 (1975).