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GRAIN SHAPE, PROTEIN, ZINC AND IRON CONTENT OF RICE LAND RACES IN BANGLADESH

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ABSTRACT

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More than half of the world's population relies on rice as the major daily source of calories and protein deficiency and micronutrient malnutrition are gradually increasing. Zinc and iron are the most important elements, deficiency of which is a major cause for malnutrition. The nutritional status and grain quality of rice is becoming more important. Awareness to have nutritionally enriched food and consumer acceptance may help in developing micronutrient enriched rice varieties. Thirty six rice land races along with a high yielding variety BRRI dhan48 were analyzed to know the nutritional status (protein, Zn and Fe) for their improvement. Zn ranged from 34.96 to 6.37 ppm. The highest Zn content was found in Chiritob followed by Kalanbhog, Saubail and Sakkorkhora. Jaradhan had the lowest Zn content followed by Begurbitchi, Jirakalani, Kalazira TAPL, Noyonmoni, Jirakatari, Madhimadob, Badsabhog, Bashmoti and Lalsora. Fe ranged from 11.3 to 2.07 ppm. The highest Fe content was found in Jirakalani and the lowest in Kultichikon. Protein content ranged from 7.63 to 7.02%. The highest protein content was found in Kultichikon followed by Sakkorkhora, Boishaki, Bashmoti and Noyonmoni. Chiritob and Jirakalani may be used as good source of Zn and Fe for further improvement.

***Key words**: rice, grain, quality, zinc, iron, protein*

INTRODUCTION

Rice is the most important cereal and staple food which serve as major carbohydrate for more than half of the world population. Half of the world’s population is suffering from one or more vitamin and/or mineral deficiency (World Food Program 2015). More than three billion people are affected by micronutrient malnutrition and 3.1 million children die each year out of malnutrition (Gearing 2015) and the numbers are gradually increasing (FAO 2019; Johnson *et al*. 2011). Increase in literacy percentage and awareness of diet, people tend to be more health conscious and interested to have nutritionally enriched food. The quality of rice is important for export market and consumer's preference.

Protein energy malnutrition affects 25% of children where their dietary intake is mainly on rice and staple crops have low levels of essential amino acids (Gearing 2015). The amount of PC in rice is relatively low (8.5%) as compared to other cereals like wheat (12.3%), barley (12.8%) and Millet (13.4%) and an average of PC in milled rice is about 7 and 8% in brown rice. Rice supplies about 40% of the protein to human through diet in developing countries and quality of PC in rice is high, due to rich in lysine (3.8%) (Shobha Rani *et al*. 2006). Therefore, improvement of PC in rice grain is a major target for the plant breeders and biotechnologists. So far, by classical breeding effort, very limited success has been achieved because of the complex inheritance nature and the large effect of environment on protein content (Coffman and Juliano, 1987). According to Iqbal*et al*. (2006), more than 170 million children and nourishing mothers suffered from Protein-calorie malnutrition (PCM) in developing Afro-Asian countries. In comparison with meat, plant proteins are much less expensive and nutritionally imbalanced because of their deficiency in certain essential amino acids (EAAs).

Iron and zinc micronutrients are the most important elements, deficiency of which is a major cause for malnutrition. More than half of the world population is suffering from bioavailable nutrient deficiencies particularly in developing countries (Seshadri 1997; Shahzad *et al.* 2014). The main reason of these deficiency occurred due to consumption of polished cereal based food crops as rice, wheat and maize (Pfeiffer and McClafferty, 2007). Modern high yielding rice varieties are poor sources of essential micronutrients like Fe and Zn (Zimmerman and Hurrel, 2002). On an average, polished rice has 2 mg kg^{-1}, while the recommended dietary intake of Fe for humans is 10–15 mg kg^{-1}. Therefore, globally more than 3 billion people were affected by Fe deficiency, particularly in developing countries (Graham *et al*. 1999; Welch and Graham, 2004). Pregnancy maternal mortality by anemia leads to 1.15 lakh deaths per year, resulting in 3.4 million disability-adjusted life-years (DALYs), has been recognized to Fe deficiency (Stoltzfus *et al*. 2004). Hence, improvement of Fe content in rice grain is necessary, which is a major challenge to the plant breeders. In plants, Zn plays a significant role in the biosyntheses and turnovers of proteins, nucleic acids, carbohydrates and lipids, with functional aspects as integral cofactor for more than 300 enzymes, coordinating ion in the DNA-binding domains of transcription factors and equally important as Fe and vitamin A (Marschner 1995). Males within the age bracket of 15–74 years require approximately 12–15 mg of Zn daily, while females within 15–74 years of age group need about 68 mg of Zn (Sandstead 1985). Generally, the content of Zn in polished rice is an average of only 12 mg kg^{-1}, whereas the recommended dietary intake of Zn for humans is 12–15 mg kg^{-1} (FAO 2001). About 17.3% of the global population is under risk of Zn deficiency and in some regions of the world, it is as high as 30% due to dietary inadequacy (Wessells and Brown, 2012). Therefore, to enhance the concentration of these micronutrients in rice grain could be possible as signified the presence of vast genetic potential of various rice germplasm by adapting appropriate genetic approaches. However, major attention to date has been paid on identification and development of genetically engineered rice grains with increased bioavailable contents of Fe and/or Zn. Recently, Indian Institute of Rice Research, Hyderabad has developed a genotype (IET 23832) that possesses high Zn (19.50 ppm). As the brown rice has higher amount of Fe and Zn, more than 70% of micronutrients are lost during polishing (Sellappan *et al*. 2009) as they are located on the outer layer of the kernel. Martinez *et al*. (2010) found 10–11 ppm Fe and 20–25 ppm Zn in brown rice, while 2–3 ppm Fe and 16–17 ppm Zn was observed in milled rice.

Grain size and shape (length-width ratio) is a very stable varietal property that can be used to measure the varietal purity of a sample. The wide diversity of plant genetic resources provides opportunity for identifying micronutrient-rich genotypes for direct use or for genetic enhancement of staple crops using breeding strategies. The nutritional status and grain quality of rice is becoming more important. Considering consumer preference and market price, farmers aim is for the production of fine rice. In Bangladesh, cultivation of high yielding rice varieties has been increased and information of land races particularly nutritional status is becoming unknown. So, this study was undertaken to know the shape of grains and protein, Zn and Fe content of land races and their further improvement.

MATERIALS AND METHODS

Rice sample preparation

Thirty seven rice genotypes (36 land races and a high yielding variety BRRI dhan48) were collected from Bangladesh Rice Research Institute and different parts of Bangladesh. The research was conducted in Crop Physiology Division, Bangladesh Institute of Nuclear Agriculture, Mymensingh and Department of Agricultural Chemistry, Bangladesh Agricultural University, Mymensingh during 2018 to 2020. The samples were manually cleaned to remove cracks kernels and the husk of the paddy was removed to get rice. Rice grains were grinded for analyzing.

Determination of kernel length and breadth

Ten randomly selected whole kernels of rice in three sets were taken and length and breadth of each grain was measured by using a slide calipers. The average value for each observation was considered as final reading. The length and breadth of rice kernel were expressed in millimeter (mm).

Determination of kernel length/breadth (L/B) ratio

The L/B ratio was calculated by dividing the average length by the average breadth of kernel. L/B ratio= Average length of the rice (mm)/ average breadth of the rice (mm). The scores are recorded for brown rice to evaluate the traits as genetic characteristics avoiding the effect of milling on size and shape. The rice grains were classified by standard evaluation system (SES) for rice (IRRI 1996), provides the following scales: For size extra long (>7.50), long (6.61-7.50), medium/ intermediate (5.51-6.60), and short (< 5.50) mm.

Estimation of protein

Micro-Kjeldahl method was used for the estimation of total nitrogen in rice grain. Then total nitrogen was multiplied by conversion factor to obtained protein content.

Digestion: Powdered rice samples (0.2 g) taken in a 75 ml Kjeldahl flask and 5ml of concentrated H_2SO_4, 1 gm of digestion mixture was added. The flask was placed on digestion chamber and boiled until the mixture content becomes clear. The flask was cooled and the digested sample was diluted with distilled water.

Distillation: 25 ml of diluted digested samples was taken and 25 ml of 40% NaOH was poured into the flask slowly holding the flask about 45· angle and connected to the distillation set. The distillate was collected in a conical flask containing 10ml of 2% Boric acid solution and 2-3 drops of mixed indicator.

Titration: Total distillate was titrated with 0.1N HCL and titration value was recorded.

Percentage of N was calculated by the following × formula:

% of nitrogen= (Ts-Tb) × normality of acid × 0.014 × 100/weight of samples (g),

Where Ts= Titre value of the sample, Tb= Titre value of the blank
0.014= Milli equivalent weight of nitrogen
% protein= % of nitrogen × C.F.
C.F.= Conversion factor (5.5 for plant sample)

Sample preparation and determination of Zn and Fe

Collected samples were dried in an oven at 65°C for 24 hours and ground by a grinding machine after cooling. The prepared samples were then kept into plastic bottles until extract preparation. The plant extract was prepared by wet oxidation method using di-acid mixture following Singh *et al*. 1999. Exactly, 1.0 g of finely ground plant material was taken into a 250 mL conical flask and 10 mL of di-acid mixture (HNO_3 : $HClO_4$= 2:1) was added to it. Then, it was placed on the electric hot plate for heating at 180-200°C until white fumes were evolved and subsequently cooled at room temperature. The digest was washed with distilled water repeatedly and filtered into a 100 mL volumetric flask through filter paper (Whatman No. 42) and the volume was made up to the mark with distilled water. Amaranth extracts were preserved separately in plastic bottles for subsequent chemical analysis.

The concentrations of Fe and Zn ions in the extracts were analyzed by atomic absorption spectrophotometer (AAS) (Model: SHIMADZU AA-7000) at the wavelengths of 248.3 and 213.9 nm, respectively as described by APHA 2012.

RESULTS AND DISCUSSION

The highest Zn content was found in Chiritob (34.96 ppm) followed by Kalanbhog (34.83 ppm), Saubail (30.11 ppm) and Sakkorkhora (28.38 ppm) (Table 1). Jaradhan had the lowest Zn content (6.37 ppm) followed by Begurbitchi (7.23 ppm), Jirakalani (8.14 ppm), Kalazira TAPL (9.00 ppm), Noyonmoni (9.14 ppm), Jirakatari (10.18 ppm), Madhimadob (11.55 ppm), Badsabhog (11.67 ppm), Bashmoti (12.52 ppm) and Lalsora (13.09 ppm). The highest Fe content was found in Jirakalani (11.3 ppm) and the lowest in Kultichikon (2.07 ppm). The results partially agree with Martinez *et al*. (2010) who found 10–11 ppm Fe and 20–25 ppm Zn in brown rice, while 2–3 ppm Fe and 16–17 ppm Zn was observed in milled rice. The results also partially agree with Maganti *et al*. (2019) who found Fe concentration varied from 6.9 to 22.3 mg/kg, whereas Zn concentration ranged from 14.5 to 35.3 mg/kg in unpolished, brown rice. The highest protein content was found in Kultichikon (7.63%), followed by Sakkorkhora (7.26%), Boishaki (7.20 ppm), Bashmoti (7.07 ppm) and Noyonmoni (7.02 ppm). The results are in conformity of Shobha Rani *et al.* (2006) and Mahender *et al*. (2016).

Table 1. Grain shape, Zn, Fe and protein content of rice land races

Genotypes	**Kernel length (mm)**	**Kernel breadth (mm)**	**Length /Breadth ratio**	**Zn content (ppm)**	**Fe content (ppm)**	**Protein** (%)
Doiargura	4.30f-k	1.75jk	2.45b-h	27.75b	3.90lmn	6.94bcd
Boishaki	4.04k	1.75jk	2.30e-j	14.88e-j	7.79de	7.20abc
Tulsimala	4.25g-k	1.90fghij	2.23e-j	16.66d-h	4.85i-l	6.30b-g
Kalajira-TAPL	4.53c-k	1.70k	2.67a-f	9.00i-l	5.80f-k	6.73b-e
Bashmoti	5.40ab	1.78hijk	3.05a	12.52f-l	9.59c	7.07a-d
Kataribhog	4.43d-k	1.89f-k	2.35d-j	14.72e-j	5.42h-k	6.02c-h
Jirakatari	4.64c-k	2.03b-f	2.29e-j	10.18g-l	5.00h-l	6.07c-h
Lalsora 1	4.80b-j	2.16ab	2.22e-j	14.87e-j	6.16f-i	5.26e-h
Bowigiaki	4.06jk	1.97b-i	2.06hij	14.50e-k	5.46g-k	5.18fgh
Lalsora 2	4.09ijk	1.98b-g	2.06hij	13.09f-l	7.04ef	5.93c-h
Madhimadob	4.06jk	1.90f-j	2.14g-j	11.55f-l	4.50k-n	6.29b-g
Jaradhan	4.16h-k	2.13abc	1.95ij	6.373l	3.50mno	5.59d-h
Chinikarai	4.52c-k	2.10a-e	2.16g-j	14.55e-k	5.40h-k	4.86gh
Deshi kaluni	4.32e-k	2.25a	1.92j	17.59d-g	5.60g-k	5.65d-h
Jirabhog	4.40d-k	2.13abc	2.06hij	15.69e-i	3.42no	6.61b-f
Bashmoti sufaid	5.20abc	1.86f-jk	2.80a-d	16.98d-g	3.19nop	5.21fgh
Tulsimala 2	4.67c-k	1.86f-k	2.49b-h	26.24bc	5.95f-j	6.31b-g
Kalaribhog	4.19h-k	1.90e-j	2.20f-j	34.83a	5.94f-j	5.92c-h
Lunia	4.07jk	1.81g-k	2.25e-j	17.73d-g	10.8b	5.17fgh
Bashmoti 71	4.13ijk	2.11a-d	1.96ij	16.84d-g	4.05lmn	5.10gh
Begunbitchi	4.51c-k	1.87f-k	2.41c-i	7.233kl	3.53mno	4.62h
Badsabhog	5.01a-f	2.11a-d	2.38c-j	11.67f-l	3.26nop	5.09gh
Ranisalut	4.61c-k	1.98b-h	2.34d-j	23.57bcd	3.80lmn	6.69b-e
Baoibhog	4.43d-k	2.00b-g	2.22e-j	15.72e-i	2.23op	5.05gh
Sakkorkhani	4.95a-g	1.88f-k	2.62a-g	28.38ab	3.33nop	5.68d-h
Kultichikon	5.45ab	1.92d-j	2.84abc	20.81cde	2.07p	7.63ab
Sakkorkhora	4.79b-k	1.83g-k	2.62a-g	17.99def	5.36h-k	7.26abc
Jirakalani	4.47c-k	2.05b-f	2.17g-j	8.14jkl	11.3a	6.94bcd
Saubail	5.10a-d	1.96c-i	2.60a-g	30.11ab	6.36fgh	5.14fgh
Noyonmoni	4.83b-i	1.73jk	2.79a-d	9.14h-l	4.78j-m	7.02a-d
Tilokkachori	4.83b-i	2.05b-f	2.35d-j	12.99f-l	7.04ef	4.99gh
Bashful	4.97a-g	1.75jk	2.84abc	12.40f-l	8.79cd	5.20fgh
Kamianasaru	4.90a-h	1.82g-k	2.69a-e	27.36bc	7.70de	4.63h
Chiniatob	5.00a-f	1.73jk	2.89ab	34.96a	9.08c	5.69d-h
Gahinda	5.07a-e	1.95c-i	2.59a-g	14.15e-k	6.78efg	5.87c-h
BRRI dhan48	5.58a	2.27a	2.49b-h	10.84f-l	14.03a	8.37gh

Common letter(s) in a column do not differ significantly at 5% level as per DMRT

CONCLUSION

Zn content of rice land races ranged from 34.96 to 6.37 ppm, Fe content from 11.3 to 2.07 ppm and protein content from 7.63 to 7.02%. The highest Zn content was found in Chiritob and the highest Fe content in Jirakalani. So, Chiritob and Jirakalani may be used as good source of Zn and Fe for further improvement.

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