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International Journal of Current Research Vol. 7, Issue, 01, pp.11895-11900, January, 2015 INTERNATIONAL JOURNAL OF CURRENT RESEARCH

RESEARCH ARTICLE

GRAIN QUALITY AND STARCH EVALUATION OF LOCAL VARIETIES OF RICE (*Oryza sativa* L.) GROWING IN JHARKHAND STATE, INDIA

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ARTICLE INFO	ABSTRACT					
<i>Article History:</i> Received 19 th October, 2014 Received in revised form 15 th November, 2014 Accepted 07 th December, 2014 Published online 31 st January, 2015	With the improvement of people's living standards and increasing awareness, focus is shifting towards staple food with added benefits that could be a part of healthy balanced diet. Many landraces or local varieties of rice harbour beneficial traits such as higher nutritional value and medicinal properties when compared to hybrid varieties. Efforts are required to increase public awareness about these varieties. Therefore, the present study aimed at studying physico-chemical attributes and correlation among grain quality traits of four traditionally cultivated and consumed varieties of rice					
<i>Key words:</i> <i>Oryza sativa</i> , Hulling, Milling, Physiochemical, Cooking characteristics, Amylose, Correlation.	from the state of Jharkhand, India, known for their nutraceutical properties. Most varieties are of medium size and shape. Recovery of Brown rice in selected varieties ranged from 75% to 79%. Variety Dani Goda was found to have the highest head rice recovery of 64.3%. Volume expansion ratio was observed to fall within the range of 2.11 to 2.70. Amylose content was found to range from 13 to 33% with the variety Karhani showing the maximum value. Various physiochemical and cooking properties exhibited convoluted relationship. Significant positive correlation was observed between grain weight and kernel length (r = 0.87, p< 0.05), kernel length and volume expansion (r = 0.74, p<0.05) and a significant negative correlation between volume expansion and amylose content (r = -0.65, p<0.05).					

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INTRODUCTION

Rice, truly known as grain of life is one of the major staple food that satisfies hunger of millions across the globe. Nutrient composition of rice is very rich on account of carbohydrate, protein, fibre, minerals and vitamins (Juliano, 1979). In global scenario, India and China have major share in rice production. In order to satisfy the hunger of ever increasing population and for economical reasons high vielding hybrid varieties are rapidly replacing the local varieties. If this trend continues there is an apprehension that valuable local varieties will be lost forever. The state of Jharkhand is very rich in ethnic diversity and also in plant diversities. There are large numbers of local varieties of rice which have stabilized to congenial edapho-climatic conditions of the state. These varieties carry valuable features such as medicinal properties, scent, disease resistance, early maturity, drought resistance and most importantly thrive easily without much input. Unfortunately, the cultivation of these local varieties is dwindling because of introduction of high yielding varieties and various developmental activities. It is therefore, imperative to document and conserve land races which constitute main food

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Laboratory of Plant Physiology and Biotechnology, Department of Botany, Ranchi University, Ranchi-834008, Jharkhand, India. of the ethnic communities and are under cultivation practices. The conservational practices are protection, restoration and propagation. Simultaneously, their scientific evaluation is also required for valuable attributes so that concerted effort may be taken for their acceptance amongst the masses. In the light of the above facts four local varieties, namely Dahiy, Dani Goda, Karhani and Neta have been identified in order to screen their grain quality and starch evaluation so that they may be recommended for qualitative and quantitative improvement. The variety Karhani is used in traditional medicine for treatment of Jaundice. Dahiya is known for its high energetic content. All the chosen varieties are having red kernel except for Dahiva which is having white kernel. Pigmented varieties are endowed with antioxidant properties. Free radical scavenging activity is known to inhibit etiology of number diseases as cancer, reduces plasma cholesterol level and may prevent cardiovascular disease.

Grain quality of rice plays an important role in consumer acceptability since rice is mainly consumed as whole grain especially in Asia (Seraj *et al.*, 2013). Grain quality of rice is determined by the factors: grain appearance, nutritional value, cooking and eating quality (Juliano, *et al.* 1990). Grain quality preferences vary with cultivators, millers, consumers and from place to place. Therefore its determination becomes necessary prior to any study related to rice varieties. In the present work we have evaluated rice grain quality characteristics of four varieties which are being consumed by ethnic communities of Jharkhand.

MATERIALS AND METHODS

In the present investigation seeds of four rice varieties, Dahiya, Dani Goda, Karhani and Neta were collected from Gene Campaign, Ranchi. For physical characteristic (hullingmilling; grain shape, size and weight) procedures described by Dela Cruz and Khush, 2000 was employed.

Determination of Hulling-Milling Values

Hulling and milling values were determined from 100 gm of rice grains of each variety. Rough rice samples were dehulled and the resulting Brown rice was subjected to milling.

Values were calculated using following formulas:

Brown rice (%) =
$$\frac{\text{weight of brown rice}}{\text{weight of rough rice}} X 100$$

Hull (%) = $\frac{\text{wt. of hull}}{\text{wt. of rough rice}} X 100$
Total milled rice (%) = $\frac{\text{wt. of total milled rice}}{\text{wt. of rough rice}} X 100$
Head rice (%) = $\frac{\text{wt. of head rice}}{\text{wt. of head rice}} X 100$

Degree of milling (%) = $\frac{\text{wt. of total milled rice}}{\text{wt. of brown rice}} X100$

Determination of Physical attributes

Grain size: Ten dehusked rice kernels of each variety were arranged lengthwise and breadth wise, for cumulative measurement of length and breadth respectively in centimetres. Average length and Breadth of the rice kernels was recorded as paddy grain length and breadth respectively.

Grain shape: Based on Length to breadth ratio, the shape of the milled rice is determined as Slender (Over 3.0), Medium (2.1-3.0), Bold (1.1-2.0) and Round (1.0 or less).

The ratio of length to breadth (L/B) of dehusked and polished grain was obtained by dividing the length of each kernel by its corresponding breadth:

L/B ratio = <u>Mean length of rice kernel</u> Mean breadth of rice kernel

1000 Grain Weight (gm): In each of genotype, 100 whole kernels were counted and their weights recorded in gm and multiplied by 10 which implied 1000 grain weight.

Chemical attributes

To determine cooking quality following chemical attributes were determined.

Alkali spreading (ASV) and Gelatinization temperature (GT): Alkali spreading value was estimated according to the procedure of Biswas and Juliano, 1988. A set of six whole unmilled kernels of different rice samples without cracks were selected, placed in a petridish and soaked in 10 ml of 1.7% (0.3035 M) potassium hydroxide (KOH) solution. The samples were arranged to provide enough space between kernels to allow for spreading. The petridishes were covered and incubated for 23 hours in a 30°C oven. Starchy endosperm was rated visually based on a 7 point numerical spreading scale. Standard check varieties of high, intermediate and low gelatinization types of rice were included for every test. Gelatinization temperature is measured by determining the alkali spreading value. The GT of the rice varieties is known to vary between 50°C to 79°C and classified as low (55-69°C); intermediate (70-74 °C) and high 75-79°C (Juliano, 1979).

Gel Consistency (GC): Based on consistency of gel, rice varieties are categorized as very flaky with hard gel of 40mm or less; flaky with medium gel of 41-60 mm length and soft with gel length of 61 mm or more.100 mg kernels flour of different rice samples were collected in thin and long test tubes. 5 ml of 95% ethyl alcohol, 2-3 drop of bromophenol blue and 2 ml of 0.2 M KOH solution was added. Contents were mixed thoroughly, covered with glass marble and test tube were soaked in boiling water bath for 8 min until the height of all the sample reached 2/3 rd of the test tube. Test tubes were then removed and left to stand at room temperature for 5 min. After cooling in an ice water bath for 20 minutes, all the test tubes were laid horizontally on a graph paper and total length of gel was measured in mm (Cagampang *et al.*, 1973).

Volume Expansion ratio (VER): 10 gm rice grain were taken in 100 ml cylinder filled with 50 ml of water and the increase in volume was noted as the volume of raw rice. The entire rice sample was soaked for 30 minutes in water. Samples were cooked in boiling water for 10 minutes in glass beakers. Immediately after cooking, water was drained out first by sieve then by filter paper and increase in volume was measured by cylinder similarly as volume of raw rice was measured (Juliano, 1971).

Starch Estimation: 0.4 gm of the sample was homogenized in hot 80% ethanol to remove sugars. The residue retained after centrifugation were washed repeatedly with hot ethanol (80%) till the washing is colourless. The residues were dried well .The extraction was done from the dried sample with the application of 5 ml water and 6.5 ml of perchloric acid (52%). The temperature was maintained at 0° C for 20 min and samples were put under centrifugation at 10,000 rpm for 8 min. The supernatant was decanted and kept for starch estimation. The extraction was repeated 2-3 times for full and final extraction. With the addition of distilled water final volume of the pooled up supernatant was made to 100ml. 0.1 ml of supernatant was pipetted out and volume was made up to 1 ml with distilled water. Similarly for reference, different aliquots of standard glucose solution were taken and volume made up to 1 ml with distilled water. 4 ml of anthrone reagent was added to each tube and heated for 8 min in water bath. Intensity of colour, green to dark green, was recorded at 630 nm (Basu et al., 2012). The glucose concentrations of the samples were determined by

using the standard graph (Fig 2 A; y = 0.003 x) and the obtained values were multiplied by a factor 0.9 to quantify the starch content.

Amylose content: Seeds of different varieties were ground to make fine powder. 100 mg of powdered samples was added with a mixture of ethanol and 1N NaOH (1ml+10 ml) and was left overnight. Subsequently, distilled water was mixed in the sample solution to make the final volume of 100 ml. An aliquot of 2.5 ml of extract was mixed with, 20 ml distilled water and 3 drops of Phenolphthalein, whereby the solution changes into pink colour. On addition of 0.1 N HCl drop by drop the pink colour disappears. To the treated sample 1 ml of iodine reagent was added and volume was made up to 50 ml by addition of distilled water and absorbance was recorded at 590 nm with reference to blank (1ml iodine reagent diluted to 50 ml with distilled water).

The amylose content of the rice seed was determined using the standard curve (Fig 2 B) derived from potato amylose (Juliano, 1971). The calculation was done using the following formula:

Amylose content = $0.D X \frac{\text{Dilution factor}}{\text{slope}}$

Since, 2.5 ml of the test solution= x mg amylose Therefore, 100 ml contains= $\frac{x}{2.5}X$ 100 amylose = % amylose

Amylopectin content:

The amylopectin content was determined by subtracting amylose content from the total starch content (Chattopadhyay *et al.*, 2008).

Statistical analysis

One-way analysis of variance (ANOVA) followed by Bonferroni test has been used for multiple comparisons, and level of statistical significance was set at $p \le 0.05$.Pearson correlation coefficients (r) were calculated by using MS Excel software.

RESULTS AND DISCUSSION

The quality of rice is considered from the view point of milling quality, grain size, shape, appearance and cooking characteristics determined to increase the acceptability to the consumer (Dela Cruz and Khush, 2000). The physicochemical characteristics include grain length (L), grain breadth (B), L/B ratio, hulling and milling percentage. The cooking qualities are amylose content, alkali spreading value, water uptake, volume expansion ratio and kernel elongation ratio (Bhonsle *et al.*, 2010). The varieties subjected for quality evaluation are native to Jharkhand and are being lost as farmers prefer growing high yielding varieties. Among the selected varieties, Karhani is the most prevalent among the tribal population as it is used to prepare a fermented drink, locally known as '*Hadia*'.

Hulling-Milling attributes

Evaluation of hulling-milling value is one of the most important physical factors to know the quality of rice in terms of head rice recovery. It is determined at two levels of processing; hulling and milling. In all varieties of rice unit mass or weight of sample was found to decrease significantly for every level of processing by removal of husk and bran layer from paddy and brown rice, respectively.

Table 1 gives the comparison of Hulling-Milling value of the chosen varieties which is important from marketing poin of view and for the profit of cultivators. Highest brown rice was recovered for Dani Goda 78.79 \pm 0.92 % and least by Dahiya 74.68 \pm 1.22. Head rice recovery ranged from 40 to 64%, in the present investigation, with maximum of 64.33 \pm 2.64% for Dani Goda and minimum of 40.3 \pm 2.16% for Neta. Head rice recovery of 65 % or more is considered a desirable trait, which in turn depends on the graintype, chalkiness, cultivation practices and drying condition (Dipti *et al.*, 2003; Bhonsle *et al.*, 2010).

Physical attributes

The length and breadth of paddy and kernels are depicted in Table 2. Based on the length: breadth ratio of kernels all the selected rice varieties fall in medium sized grain except for Karhani which is short grain size category. For shape all are under medium category.

Varieties	Wt. of rough rice (gm)	Brown rice % [#]	Hull% [#]	Total milled rice % [#]	Head rice % (HRR) [#]	Degree of milling % #
Dahiya	100	74.68±1.22 ^a	26.75±2.93 ^a	67.99± 1.28 °	58.9±1.68 ^a	93.43±2.06 ª
Dani Goda	100	78.79± 0.92 ^a	22.46±2.58 a	71.72± 1.77 ^a	64.33±2.64 ^a	90.23±1.78 ^a
Karhani	100	74.93± 2.73 ^a	24.77±1.38 a	72.09 ± 1.84^{a}	$44.06 \pm .76$ ^b	95.02 ± 2.76^{a}
Neta	100	75.12± 2.83 ^a	24.91±2.55 a	69.72 ± 2.50^{a}	40.3 ± 2.16^{b}	91.78± 1.21 ^a

[#]Means with different letters within a column are significantly different ($p \le 0.05$).

Values are means of triplicate measurements \pm standard deviations.

Varieties	Husked Grain [#] (cm)		Dehusked Grain [#] (cm)		Milled Head Rice Grain [#] (cm)		Size	Shape	1000grain weight [#] (gm)
	L	W	L	W	L	W		-	
Dahiya	0.79 ± 0.03^{a}	0.23 ± 0.03^{a}	0.60 ± 0.02^{a}	0.23 ± 0.01^{a}	0.54 ± 0.02^{a}	0.20 ± 0.04^{a}	Medium	Medium	18.630±0.051 ^a
Dani Goda	0.76 ± 0.03^{a}	0.27±0.02 ^a	0.53 ± 0.04^{a}	0.22 ± 0.03^{a}	0.57 ± 0.05^{a}	0.24 ± 0.02^{a}	Medium	Medium	18.838±0.114 ^a
Karhani	0.70 ± 0.03 ^b	0.27±0.05 ^a	0.55 ± 0.03^{a}	0.24 ± 0.03^{a}	0.48 ± 0.01 ^b	0.21 ± 0.02^{a}	Short	Medium	15.359±0.044 ^b
Neta	0.82 ± 0.03^{a}	0.33±0.03 ^a	0.63 ± 0.02^{a}	0.27 ± 0.03^{a}	0.60 ± 0.02^{a}	0.22 ± 0.02^{a}	Medium	Medium	20.784±0.094 °

Means with different letters within a column are significantly different ($p \le 0.05$).

[#]Values are means of triplicate measurements ± standard deviations.

The physical appearance varies with respect to its shape and size and is important from consumers point of view. The size and shape of rice grain is determined by length:breadth ratio (Dela Cruz and Khush, 2000).Highest 1000 grain weight was recorded for Neta as 20.784 ± 0.094 gm while lowest was for Karhani as 15.359 ± 0.044 gm. Both Dani Goda and Neta showed nearly 18 gm as 1000 grain weight. Preferences for grain size and shape vary from one group of consumers to another. Some ethnic groups prefer short bold grains, while medium and long slender grains are prized by others (Shobha Rani *et al.*, 2006). Information about the size and density of the grain is also given by grain weight. The density of different rice grains effect the cooking quality. So grain weight should be uniform because it determines the quality of grains.

Chemical attributes and cooking characteristics

Table 3 depicts the analyzed values of various chemical and cooking characteristics. Volume expansion ratio of the undertaken local rice varieties ranged from 2 to 2.5. The maximum VER was found for Neta (2.56 ± 0.015) followed by Dahiya (2.37 ± 0.015), Dani Goda (2.20 ± 0.037) and Karhani (2.11 ± 0.015).

Dahiya, Karhani and Neta with GC value of less than 40 mm comes under hard GC while Dani Goda with a value greater than 61mm falls in soft GC category. Grains with hard GC have fluffy texture and become hard upon cooling. Soft GC indicates that rice grain remains soft when cooled after cooking. It has been observed that Waxy rice varieties rich in amylopectin, have a sticky texture and remains firm when cooked. Dahiya(13%), Dani Goda(17%) and Neta(16%) have low amylose content while Karhani has high amylose content of 33%. Amylose as compared to amylopectin are digested slowly as such high amylose rice have low glycemic index. The variety Karhani could be further investigated in this aspect and for its suitability for diabetics.

Correlation among various physiochemical attributes

The correlation coefficients for various physiochemical attributes analyzed are depicted in Table 4. Correlation analysis always helps the consumers to select better rice varieties for their consumption and use (Seraj *et al.*, 2013).

Table 3. Chemical attributes and cooking characteristics of Selected varieties

Variety	ASV	GT	GC (mm)	Volume Expansion Ratio (VER)	Starch % [#]	Amylose % #	Amylopectin% [#]
Dahiya	3 (low)	75-79 (high)	22 (very flaky)	2.37 ± 0.015 ^a	42.2±3.76 ^a	13.06 ^a (low)	29.13 ± 1.650^{a}
Dani Goda	4 (interme diate)	70-74 (intermediate)	88 (soft)	2.20 ± 0.037 ^b	49.7 ± 2.99^{a}	17.03 ^a (low)	32.66 ±1.105 ^b
Karhani	5 (interme diate)	70-74 (intermediate)	30 (very flaky)	2.11 ± 0.015^{b}	44.79± 3.32 ^b	33.07 ^b (high)	10.72±1.379 ^b
Neta	4 (interme diate)	70-74 (intermediate)	35 (very flaky)	$2.56 \pm 0.015^{\circ}$	62.03±3.21 ^a	16.45° (low)	45.58 ±1.239 °

Means with different letters within a column are significantly different ($p \le 0.05$).

[#] Values are means of triplicate measurements ± standard deviations.

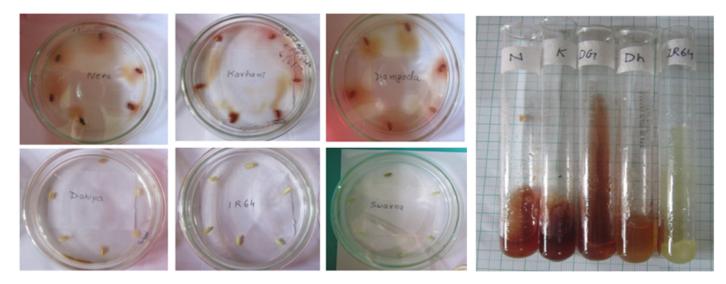


Fig. 1. ASV of selected local varieties and reference varieties, GC of selected rice varieties

The cooking and eating characteristics of rice is the basis of choice for the consumers and is mainly affected by starch content and properties. The proportions of two types of starch-amylose and amylopectin, ASV, GT and GC are the major characteristics to be determined for quality evaluation. Dani Goda, Karhani and Neta showed intermediate GT as evidenced by ASV of 4, 5 and 4 respectively. Dahiya with ASV of 3 has high GT (Fig 1)

Grain weight was found to have a highly significant and positive correlation with Length of the Kernel (r = 0.87, p<0.05). Kernel Length was also found to have a positive correlation with Volume expansion (r = 0.74, p<0.05). Chauhan *et al.* (1995) also reported a positive correlation between these paramers. However, no relation between volume expansion with grain size and shape was observed.

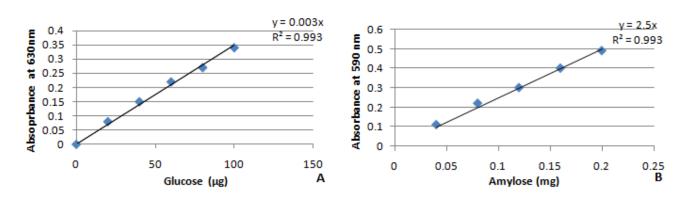


Fig. 2. (A): Standard glucose curve for estimation of starch; (B): Standard curve for amylose

Table 4. Correlations among various physiochemical characteristic

	L	W	L/W	HRR	1000 Gr. Wt.	V ER	ASV	GT	GC	Starch%	Amylose%	Amylo pectin%
L	1											
W	0.54	1										
L/W	0.17	-0.7	1									
HRR	0.18	0.27	-0.1	1								
1000 Gr.Wt.	0.87**	0.23	0.43	0.05	1							
VER	0.74*	0.05	0.52	-0.28	0.89**	1						
ASV	-0.45	0.13	-0.56*	-0.52	-0.59*	-0.53	1					
GT	-0.05	-0.40	0.48	0.40	0.07	0.17	-0.82**	1				
GC	0.31	0.55	-0.37	0.57*	0.17	-0.26	0.10	-0.48*	1			
Starch%	0.73*	0.51	-0.05	-0.43	0.68*	0.68*	0.11	-0.53	0.15	1		
Amylose%	0.60*	0.08	-0.6*	-0.42	-0.81**	-0.65*	0.87**	-0.48	-0.16	-0.15	1	
Amylopectin%	0.88**	0.28	0.37	0.01	0.99**	0.88**	-0.52	-0.02	0.20	0.75*	-0.76*	1

*p <0.05; **p<0.001

Similar results have al been reported by (Sivasubramanian et al., 1973); Sood and Siddiq, 1986. Length was found to be significantly and positively correlated with Starch (r = 0.73, p<0.05) and amylopectin (r = 0.88, p<0.05) whereas negatively with amylose content (r = -0.63, p< 0.05). The negative correlation between amylose content and length of kernel has also been reported by Vanaja and Babu, (2003). A negative correlation was also found between shape and size of the grain defined by length: breadth ratio and amylose content. This is in agreement with the findings of Vanaja and Babu, 2003 that slender grains have lesser amylose content. They have also reported significant negative correlation between Width and Length/Width ratio. A similar result has been obtained in this study with r = -0.73 at p<0.05 between the above two parameters. No correlation could be established between Length:Breadth ratio and ASV similar to that reported by Vanaja and Babu, (2003).

HRR could not be related to any of the chosen physiochemical characteristics. (Hussain *et al.*, 1987 and Chauhan *et al.*, 1995) Have also reported no relationship of Hulling-Milling recovery with shape of grain, ASV and amylose content. 1000 grain weight was found to have positive correlation with VER (r = 0.89, p<0.001), Starch (r = 0.68, p<0.05) and amylopectin (r = 0.88, p<0.001) while a negative correlation with amylose content (r = -0.81, p<0.001). VER depends upon the composition of starch content of rice grains. It had significant positive correlation with starch (r = 0.69, p<0.05) and amylopectin content (r = 0.88, p<0.001). Similar to the findings of (Zaman *et al.*, 1985 and Chauhan *et al.*, 1995) VER

and amylose content were found to be negatively correlated (r = -0.65, p<0.05). This is in contrary to various other researchers (Sood *et al.*, 1986; Hussain *et.al.*, 1987 and Yadav *et. al.*, 2007) who reported a positive correlation indicating more water absorption by high amylose rice.

The positive significant correlation of ASV with amylose content (r = 0.88, p<0.001) and negative significant correlation with GT (r = -0.82, p<0.001) obtained signifies that GT decreases with increase in amylose content. The findings are in accordance with Tomar and Nanda, 1982.

Conclusion

The selected rice varieties cover wide range of characteristics. With respect to Hulling and Milling values Dani Goda was found to show a desirable head rice recovery.

For physical attributes Dahiya, Dani Goda and Neta were found to be of medium shape and size with low amylase content whereas Karhani, a short grain variety has high amylose content. It is necessary to conserve these varieties for their diverse physical and chemical characteristics. These traditional varieties could serve as parental donors for further breeding programmes.

The variety DaniGoda shows and Karhani could be further investigated and subjected to improvement programmes. Increasing public awareness and concerted efforts for restoration of these traditional varieties is the need of the hour.

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