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Studies on physico-chemical and cooking characteristics of rice bean varieties grown in NE region of India

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Abstract Rice bean (Vigna umbellata) is grown in South and Southeast Asia, and the bean has gained importance due to its nutritional strength in terms of dietary fiber, quality protein and minerals. In current study, the nutritional and functional components, cooking and thermogravimetric properties of eleven rice bean varieties from NE India were investigated. Results revealed that the major nutrients among the varieties ranged as follows: 54.21–60.49% carbohydrates, 15.64-21.60% protein, 1.22-2.3% fat, 5.53-6.56% crude fibre, 3.34-3.8% ash; while the functional, anti-nutritional factors and mineral were present as 1189.32-1645.8 mg gallic acid equivalent (GAE)/100 g polyphenols, 205.38-432.14 mg/100 g phytic acid, 23.14-34.12 mg/100 g oxalate, 690.7-1589.5 mg/ 100 g saponins, $49.90-158.17 \mu g/100 g$ hydrocyanide, 111.51-168 calcium, 5.50-10.44 zinc, 3.72-8.37 iron. Principal component analysis revealed that varieties with higher calcium, iron and ash content had lower cooking time, swelling ratio, and cooked grain hardness. It is also revealed that varieties with higher weight loss at sixth stage in thermogravimetric graph had lower carbohydrate and higher protein content. Nagadal variety had higher fat, potassium, magnesium, calcium, sodium, iron, copper and chromium content and better cooking quality as compared to the other varieties. The study revealed that Nagadal variety was superior to other varieties with respect to

D. D. Wadikar ddwadikar@gmail.com mineral content, cooking and thermal properties and hence have better potential in the development of value added products.

Keywords Rice Bean (*Vigna umbellata*) · Cooking · Thermo-gravimetric analysis · Minerals · Anti-nutritional · Non-nutritional

Introduction

Beans play an important role in human nutrition, particularly among the low-income groups of people in developing countries like India. They are good source of dietary fiber, slow digestible starch, minerals, vitamins and bioactive phytochemicals which have capability to control or prevent various chronic and metabolic diseases especially diabetes mellitus, coronary heart disease and colon cancer (Tharanathan and Mahadevamma 2003). Rice bean (Vigna umbellata) is considered as tribal bean of South and Southeast Asia. In India, its cultivation is mainly confined to the tribal areas of Eastern and Northern India and to some extent in Orissa and Bihar where it is grown for fodder, green manure, cover crop and food. India produces 7500MT of rice bean annually for fodder and food purpose. Conventionally, rice bean is utilized to prepare vegetables (tender pod), whole dhal, deep fried snacks, soup, gruel, dessert, pastry and pickles (Wei et al. 2015).

In recent years, rice bean has gained importance, because of its nutritional strength in terms of dietary and functional fiber, quality protein, calcium and its potential health benefits. It has high quality protein with all essential amino acid in balance manner and especially rich in methionine and tryptophan. Rice beans have lower level of glycemic index, tannin, oligosaccharide and higher levels

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of potassium, calcium, iron and zinc, thiamin, niacin and riboflavin with higher calcium and zinc bio-availability. Rice bean is free from toxic and allergenic substances. It has number of bioactivities which include antioxidant, hepato-protection, anti-inflammation, anti-cancer, antifungal, anti-diabetic, anti-hypertension immunity booster, Inhibition of activities of HIV-1 and mutagenic activities (Wei et al. 2015). Besides their high nutritional value, rice bean also contain naturally occurring compounds known as anti-nutritional and or non-nutritional factor such as phytic acid, oxalate, saponin, polyphenol etc. These anti-nutritional factors interact with nutritional components such as carbohydrates, protein and mineral and reduce their nutritional value (Bressani 1993). Cooking of legumes initiates with breakdown of middle lamella which results in separation, disruption and softening of cell followed by starch gelatinization and protein denaturation. The process of starch gelatinization involves the uptake of water by the starch granules, followed by swelling during heated. Cooking is done above the gelatinization temperature for gelatinizing starch and to produces a tender edible product, to develop aroma and to improve the overall edibility of the legumes. The cooking characteristic of beans is unique feature which mainly depends on variety, grain hardness, storage condition and interaction of its constitutions. Thermo gravimetric analysis (TGA) is an analytical technique used to determine a material's thermal stability and its fraction such as weight loss percentage during heating. TGA provides a semi-quantitative understanding of thermal degradation processes occurring during thermochemical conversion under different atmospheres. With the help of derivatives of TGA, the rate of weight loss of the sample as a function of temperature and time is measured to predict thermal behaviors of the material.

However, the information on nutritional composition, anti-nutritional factors, thermal properties and cooking characteristics of rice bean of North East India is scanty. Therefore, this study has been undertaken to investigate the nutrient composition, anti-nutritional factors and thermal properties in different varieties of rice bean.

Materials and methods

Materials

Authentic samples of ten varieties of rice bean (JRC-08-7, JRC-08-8, JRC-08-10, JRC-08-12, JRC-08-15, JRC-08-16, JRC-08-32, JRC-13-11, JRC-13-13 and JRC-50) were collected from AICRP on Forage Crops and Utilization, Assam Agricultural University, Jorhat, India while variety *Nagadal* was collected from Dimapur market (Fig. 1).



Fig. 1 Photograph of eleven rice bean varieties

They were stored at 4 °C. The seeds were removed and kept at room temperature for temperature equilibrium before making any measurements. The samples were milled in vibratory micro mill (Pulverisette, M/s. Fritsch, Germany) and sieve through 250 μ m sieve.

All the chemicals and reagents used for analysis were of AR grade and procured from M/s. SD Fine Chemical Limited, M/s Merck Limited.

Methods

Proximate composition

Moisture, protein, fat, ash and crude fibre were estimated as per AOAC (1999) methods. Carbohydrates were estimated by difference method.

Polyphenols

Polyphenols were estimated as per AOAC (1984). The defatted samples (200 mg) were refluxed with acidic methanol (1 ml of concentrated HCl mixed with 99 ml methanol) for 10 min. The same extraction process was repeated for three times. The total 300 ml extracts were reduced to 90 ml using vacuum rotary flash evaporator. The contents were made up to 100 ml with acidic methanol. 0.5 ml of extract was taken and 0.5 ml of Folin–Denis reagent were added and mixed. To this, 2 ml of saturated sodium carbonate solution was added and volume made up to 5 ml with water, mixed and the absorbance was measured at 760 nm after 30 min. The results were calculated as mg gallic acid equivalent GAE/g sample and expressed as mg/100 g dry wt.

Phytic acid

Phytic acid was estimated as described by Haug and Lantzsch (1983). The sample (0.5 g) was dissolved in

30 min in boiling water bath. The contents were cooled and 4 ml of 2,2-bipyridine solution (Dissolved 10 g 2,2'-bipyridine and 10 ml thioglycollic acid in distilled water and made up to 1000 ml) was added. The intensity of colour was measured spectrophotometrically at 519 nm. The phytic acid content in the sample was calculated from standard curve made from standard phytic acid (40–200 μ g phytic acid).

Oxalate

Oxalate was determined as per the method mentioned by Munro and Bassir (2000). 2 g of ground sample was suspended in a 250 ml volumetric flask containing 190 ml distilled water. The suspended sample was digested at 100 °C for 1 h after addition of 10 ml 6 M HCl. The sample was cooled and made up the volume to 250 ml. The digested sample was filtered and duplicate portion of 125 ml of the filtrate were measured into beaker and four drops of methyl red indicator were added, followed by the addition of concentrated NH₄OH solution (drop wise) until the solution changed from pink to yellow colour (pH 4-4.5). Each portion was then heated to 90 °C, cooled and filtered to remove the precipitate containing ferrous ion. Each of the filtrate was again heated to 90 °C and 10 ml of 5% CaCl₂ solution was added to each of the sample with stirring consistently. After cooling, the samples were left overnight. The solutions were then centrifuged at 2500 rpm for 5 min. The supernatants were decanted and the precipitates completely dissolved in 10 ml 20% H₂SO₄. The total filtrates resulting from digestion of 2 g of each of the sample was made up to 200 ml. Aliquot of 125 ml of the filtrate was heated until near boiling and then titrated against 0.05 M standardized KMnO₄ solution to a pink colour which persisted for 30 s. The oxalate content was calculated by using following formula

Ovalata	$\left(\underline{mg} \right)$	<u>Titrate value × Volume mass equivalent × Dilution factor × 2.4×100</u>
Oralate	$\sqrt{100 \mathrm{g}}$	Molar equivalent of $KMnO_4 \times weight of sample$.

25 ml of 0.2 N HCl and shaken for three hours in orbital shaker. The contents were filtered through Whatman No.1 filter paper and made up the volume of the filtrate to 50 ml with 0.2 N HCl. The 2 ml of ferric ammonium sulphate solution (0.2 g ferric ammonium sulphate dissolved in 100 ml of 0.2 N HCl) and 0.5 ml 0.2 N HCl was added into test tube containing 0.5 ml of aliquot and heated for

Saponin

The saponin was evaluated as per the procedure reported by Obdoni and Ochuko (2001). 3 g of ground sample was put into a conical flask and 30 ml of 20% aqueous ethanol were added. The dissolved sample was heated at 55 °C for 4 h with continuous stirring. The mixture was filtered and the residue re-extracted with another 30 ml 20% ethanol. The combined extracts were reduced to 15 ml over water bath at about 90 °C. The concentrate was transferred into a 250 ml separating funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated twice. 20 ml of n-butanol was added into the aqueous layer. The combined n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation the sample was dried in the oven to a constant weight; the saponin content was calculated gravimetrically.

Tannin

Tannin was analyzed using the method of Schanderi (1970). Powder sample (0.25 g) was extracted with 37.5 ml distilled water and heated the flask gently and boiled for 30 min. The sample mixer was centrifuged at 2000 rpm for 20 min and the volume of the supernatant was finally made up to 37.5 ml with distilled water in a 100 ml flask. An aliquot of 0.5 ml of the sample was treated with 1 ml of Folin–Denis reagent followed by 2 ml of sodium carbonate and allowed to stand for color development. The absorbance of the reaction mixture was measured at 700 nm in a spectrophotometer. Tannic acid used as standard. Tannin content was expressed as tannic acid equivalents (TAE) in miligram per 100 g of dry wt.

Hydrogen cyanide

The hydrogen cyanide (HCN) was determined according to AOAC (1990) method. 5 g of ground sample was dissolved in 100 ml distilled water and left to stand for 12 h. The mixture was distilled and 75 ml of the distillate was collected in 10 ml 2.5% sodium hydroxide solution. The volume of distillate was made up to 100 ml with distilled water. Three aliquots of 25 ml were obtained. 2 ml of 6 M ammonium solution and 0.5 ml of 5% potassium iodide were added to the 25 ml aliquots. This was titrated using 0.002 M silver nitrate. The readings were taken. The equation below was used to get HCN in mg. 1 ml of 0.02 M silver nitrate = 1.08 mg.

α -Amylase inhibitory activity

Amylase inhibitory activity (AIA) was carried out as per the procedure described by Katoch and Jamwal (2013) with slight modification. 5 g of ground sample was suspended in 50 ml of 0.02 M phosphate buffer (pH 6.9) and shaking for 3 h at room temperature. The suspended sample was filter through muslin cloth and the filtered solution was heated at 70 °C for 15 min. The heated content was centrifuge at 12000 g for 30 min. The supernatant was decanted and used for analysis. 0.250 ml of sample extract was added into a 15 ml test tube containing 1 ml of 0.1 M phosphate buffer (pH 7.6), 0.5 ml of 0.25 M NaCl solution and 0.250 ml α-Amylase solution. The content was mixed properly and incubated at 37 °C for 15 min. Then, 1 ml starch solution (1%) was added and mixture was incubated for 10 min. The reaction was stopped by adding 0.5 ml of dinitrosalicylic acid (DNS) reagent [Mixed the Alkaline DNS (2 g DNS dissolved in 40 ml 2 N NaOH) and 60% potassium sodium tartrate and made up the volume 200 ml] and volume was made up to 6 ml with distilled water followed by heated in boiling water bath for 30 min. A blank was set without addition of the extract and starch, and another sample blank was prepared with extract but without starch, replaced by equal quantities of buffer. The absorbance was measured at 530 nm. The reducing sugar released from starch was estimated as maltose (1 mg/ml) equivalent from the standard graph. One amylase unit was defined as the amount of enzyme that liberated microgram maltose from starch under the assay conditions (pH 6.9, 37 °C, 10 min). One unit of amylase inhibitor was defined as that which reduced the activity of the enzyme by one unit.

Trypsin inhibitory activity

Trypsin inhibitory activity (TIA) was carried out as per the procedure described by Kakade et al. (1969) with slight modification. 5 g of ground sample was suspended in 50 ml of 0.02 M phosphate buffer (pH 6.9) and shaking for 2 h at room temperature. The suspended sample was filter through muslin cloth and it was heated at 70 °C for 15 min. The heated content was cooled and centrifuged at 12000g for 30 min. The supernatant was decanted and used for analysis. 0.5 ml of sample extract was added into a 15 ml test tube containing 0.6 ml of 0.1 M phosphate buffer (pH 7.6), 0.4 ml of 0.001 M HCl solution and 0.5 ml trypsin solution (5 mg/ml). The contents were mixed properly and incubated at 37 °C for 10 min. Then, 2 ml casein solution (1%) was added and mixture was incubated at 37 °C for 20 min. The reaction was stopped by adding 5 ml of 5% trichloroacetic acid. The content was centrifuge at 10,000g for 10 min. The 0.5 ml of supernatant was mixed with reagent C [Reagent C was prepared by mixing reagent 50 ml of reagent A (2% Na₂CO₃ in 0.1 N NaoH) and 1 ml reagent B (0.5% CuSO₄ in 1% potassium sodium tartrate)] and incubated for 10 min at room temperature. Then, 0.5 ml 50% Folin-Denis reagent was added into the mixture. A blank was set without addition of the extract and casein, and another sample blank was prepared with extract but without casein,

replaced by equal quantities of phosphate buffer. The absorbance was measured at 520 nm after 30 min of incubation. The soluble protein released from casein was estimated as bovine serum albumin (BSA) (1 mg/ml) equivalent from the standard graph. One trypsin unit was defined as the amount of enzyme that liberated microgram BSA from starch under the assay conditions (pH 6.9, 37 °C, 20 min). One unit of trypsin inhibitor was defined as that which reduced the activity of the enzyme by one unit.

Mineral profile

For mineral determination, the ash was prepared as per AOAC (1999) method. Dissolved the whitish colored ash into 0.5 M HNO₃ (0.5 M HNO₃ was prepared by using MiliQ water) and filtered through Whitman No. 44 filter paper and made up the volume to 100 ml. The determination of minerals in the solution was carried out by inductively coupled plasma optical emission spectrometry (ICP-OES). The condition of ICP-OES was pump speed 15, Nebulizer flow 0.79 and Nebulizer pressure 2.52.

Thermo-gravimetric analysis

The thermo-gravimetric analysis (TGA) of rice bean varieties was performed using Thermo gravimetric analyzer (TGA Q50, TA Instruments, DE, USA). The 10 mg of sample was placed into platinum pans and heated in a furnace flushed with N₂ gas at the rate of 40 ml/min and heated from 30 to 600°C at a rate of 10 °C/min and held isothermally for 30 min.

Cooking characteristics

Hardness The hardness of native, soaked and cooked grain was measured using a texture analyzer (TA.XT PlusTM, Stable Microsystems, UK). The texture analyzer was calibrated for force (2 kg) and probe height (6 mm) before commencing the tests. A cylindrical probe with 36 mm diameter was used. The load cell of 50 kg and cross head speed of 0.50 mm/s was the pre-test and post-test speeds were 1 mm/s and speed 5 mm/s, respectively. A deformation strain of 80% of the original length was applied. The hardness was measured as the peak force of the texture profile curve and corresponded to the force required to deform or crush the seed. The measured force was expressed as force per unit area (g) by normalizing the force with the cross sectional area of the probe. Twenty seeds were chosen as a representative for each experiment as variation in texture in individual grain is usually substantial. The obtained hardness results were expressed in Newton (N).

Cooking time The cooking time (CT) was determined following the method of Kaur et al. (2013). The 10 g of whole grains were soaked for 12 h. The 100 ml distilled water was brought to boil in 500 ml spout less beaker fitted with bulb condenser to prevent the water loss. The soaked grains were added into the boiled water. Boiling was continued; 4–5 boiled grains were drawn at intervals of 2 min and tested for their softness by pressing the grains between glass slides until the white core disappeared. The time from addition of grains into boiled water till achievement of the desirable softness was recorded as the cooking time.

Water uptake For determination of water uptake, the 10 g washed grains were soaked in 100 ml distilled water for 12 h followed by cooking it for minimum cooking time. The weights of cooked grains were measured after removing the surface water with tissue paper. The water uptake was calculated and express as percentage.

Solid dispersion The whole grains (10 g) were thoroughly washed with tap water to remove surface dusts and other surface contaminants. The washed grains were steep in 100 ml of double distilled water for 12 h and then cooked the steeped grains in same steep water for minimum cooking time by using 500 ml spout less beakers fitted with bulb condenser. The gruel was transferred to 250 ml beaker and then evaporated it on hot plate followed by dried completely in a hot air oven at 110 °C. The solids were subsequently weighed and solid dispersion was calculated as percentage.

Swelling ratio The volume of grains (10 g) was determined using a graduated cylinder and subsequently soaked the grains in 100 ml distilled water for 12 h followed by cooking it for minimum cooking time. The volume of cooked grains was measured after removing the surface water with tissue paper. The swelling ratio was calculated.

Degree of gelatinization (%) The degree of gelatinization (DG) was measured according to the method reported by Birch and Priestly (1973) with slight modification. The cooked rice bean was dried in oven at 58 °C and then ground and sieved through 80 mesh sieve. 1 g sample was dispersed in 50 ml distilled water containing 1 ml of 10 M KOH solution with stirring for 5 min and centrifuged at 1500 rpm for 15 min. One milliliter supernatant was pipette and added with 0.2 ml of 0.5 M HCl followed by 8 ml distilled water and 1 ml iodine solution was added. The mixture was homogenized then measuring the absorbance at 600 nm. The method was repeated using 50 ml of 10 M KOH instead of water.

 $DG (\%) = \frac{Absorbance of solution extracted with 50 ml of water containing 1 ml 10M KOH}{Absorbance of solution extracted with 50 ml of 10 M KOH} \times 100$

Statistical analysis

The results were analyzed statistically using MS-Office analysis tool-pack for ANOVA and correlation coefficient. The significance of differences was estimated by the Duncan's test. The correlation coefficient and principal components analysis were carried out by using XLSTAT 2016 software.

Result and discussion

Proximate composition

The proximate composition of rice bean varieties is shown in Table 1. JRC-08-7, JRC-08-8, JRC-08-10 and Nagadal had significantly (p < 0.05) lower (12.57-12.79%) in moisture content than the other varieties. The moisture content is an important parameter for safe storage and it depends on stage of harvesting and storage condition. JRC-08-16, JRC-13-11, JRC-13-13 and Nagadal had significantly (p < 0.05) lower amount of protein (15.64-15.95%)and higher carbohydrate (59.69-60.49%) whereas JRC-08-10, JRC-08-32 and JRC-50 had significantly (p < 0.05)higher amount of protein (20.20-21.60%) and lower carbohydrate (54.21-55.48%) as compared to other varieties. The variation in protein content may be due to genetic factor, environmental condition and soil fertility. Protein content displayed negative correlation with carbohydrate (r = -0.969, p < 0.005). The carbohydrate rich varieties

 Table 1 Proximate composition of rice bean (g/100 g)

can be utilized for development of extruded, flaked, popped products. Nagadal exhibited the highest fat $(2.30 \pm 0.15\%)$ and ash $(3.80 \pm 0.05\%)$ content while the lowest of crude fibre $(5.53 \pm 1.33\%)$ content. However, JRC-50 had the lowest $(1.22 \pm 0.10\%)$ in fat content while JRC-08-12 contained highest level of crude fibre $(6.56 \pm 1.19\%)$. The average content of moisture, carbohydrates, protein, fat, crude fibre and ash content in eleven rice bean varieties was 13.13 ± 0.34 , 57.51 ± 2.17 , 18.44 ± 2.14 . 1.54 ± 0.30 , 5.89 ± 0.37 and $3.49 \pm 0.12\%$, respectively. The average value of different chemical constituents in eleven rice bean varieties grown in NE Region showed slightly lower or comparable with earlier reported values for other varieties (Kaur et al. 2013, Rejaul et al. 2016). The proximate composition of 75 genotypes grown all over world was reviewed by Rejaul et al. (2016) and reported the moisture, carbohydrate, protein, fat, crude fibre and ash content of 9.10 ± 3.12 , 59.96 ± 2.81 , 20.78 ± 2.65 , 1.74 ± 1.42 , 5.07 ± 1.17 and $4.44 \pm 1.03\%$, respectively.

Polyphenol content

Polyphenol content in rice bean varieties ranged between 1189.32 (JCR-13-11) and 1645.80 (JCR-08-10) mg of GAE/ 100 g (Table 2). It varied significantly (p < 0.05) among the varieties. JRC-08-7, JRC-08-10, JRC-08-12, and JRC-13-13 displayed relatively more (1510.84–1645.80 mg of GAE/ 100 g) polyphenol content than other varieties. The total

Varieties	Moisture	Carbohydrate	Protein	Fat	Crude fibre	Ash
JRC-08-7	12.57 ± 0.12	57.67 ± 1.84^{b}	19.52 ± 0.04^{bcd}	1.34 ± 0.10^{a}	5.56 ± 1.12^a	3.34 ± 0.04
JRC-08-8	12.79 ± 0.10	56.18 ± 2.08^{ab}	19.59 ± 0.09^{bcd}	1.60 ± 0.11	6.42 ± 1.23	3.42 ± 0.07^{ab}
JRC-08-10	12.68 ± 0.13	55.48 ± 2.04^{ab}	20.82 ± 0.18^{e}	$1.26\pm0.09^{\rm ac}$	6.06 ± 1.10^{b}	3.63 ± 0.03
JRC-08-12	$13.19 \pm 0.10^{\rm a}$	$56.82 \pm 1.97^{\text{b}}$	18.56 ± 0.04	1.41 ± 0.10	6.56 ± 1.19	$3.46 \pm 0.09^{\rm bc}$
JRC-08-15	13.29 ± 0.11^{b}	$56.94 \pm 1.95^{\rm bc}$	$19.40 \pm 0.05^{\rm cb}$	1.30 ± 0.06^{a}	$5.64 \pm 1.21^{\rm a}$	3.43 ± 0.08^{ab}
JRC-08-16	13.25 ± 0.09^{ab}	$59.99 \pm 3.74^{\circ}$	15.86 ± 1.10^{a}	1.83 ± 0.16	$5.61 \pm 1.30^{\rm a}$	$3.46 \pm 0.04^{\rm bc}$
JRC-08-32	13.48 ± 0.08^{e}	54.86 ± 3.35^a	20.20 ± 1.01^{de}	$1.67\pm0.13^{\rm b}$	6.22 ± 1.15	3.57 ± 0.06
JRC-13-11	13.46 ± 0.22^{cde}	$60.24 \pm 2.34^{\circ}$	15.64 ± 0.38^a	$1.70\pm0.05^{\rm b}$	5.55 ± 1.18	3.41 ± 0.03^a
JRC-13-13	13.51 ± 0.09^{de}	$60.49 \pm 1.89^{\circ}$	$15.70 \pm 0.04^{\rm a}$	1.31 ± 0.13^{a}	5.56 ± 1.13^{a}	3.43 ± 0.06^a
JRC-50	$13.4 \pm 0.12^{\circ}$	54.21 ± 1.97^{a}	21.60 ± 0.07	$1.22\pm0.10^{\rm c}$	6.08 ± 1.20^{b}	$3.49\pm0.07^{\rm c}$
Nagadal	12.73 ± 0.10	$59.69\pm2.15^{\rm c}$	15.95 ± 0.38^a	2.30 ± 0.15	5.53 ± 1.33	3.8 ± 0.05

Value = Mean \pm SD (n = 3). The values with the same superscript in a column not significantly different at $p \le 0.05$

Table 2 Non-r	nutritional/anti-nutritio	nal factors of rice bean						
Varieties	AIA (AIU/g)	Polyphenol (mg GAE/100 g)	Phytic acid (mg/100 g)	Oxalate (mg/ 100 g)	Saponin (mg/ 100 g)	Tannin (mg TAE/100 g)	HCN (μg/ 100 g)	TIA (TIU/g)
JRC-08-7	22.20 ± 2.21^{cd}	1510.84 ± 17^{cd}	$233.74\pm22^{\mathrm{ab}}$	$34.12 \pm 3^{\rm def}$	$1097.8 \pm 10^{\rm b}$	998.28 ± 33^{a}	$71.17\pm06^{\mathrm{a}}$	$66.48 \pm 2.33^{\rm b}$
JRC-08-8	$28.00 \pm 1.45^{\mathrm{e}}$	1271.15 ± 34	$432.14\pm57^{ m d}$	$29.80 \pm 4^{ m bcd}$	$1098.2 \pm 12^{\rm b}$	1064.11 ± 16^{b}	104.53 ± 08	$70.96\pm3.15^{\rm a}$
JRC-08-10	$23.36\pm0.95^{\rm d}$	$1645.80\pm20^{\rm e}$	$293.91 \pm 51^{\circ}$	$34.24 \pm 2^{\text{def}}$	890.20 ± 11^{a}	1007.58 ± 06^{a}	49.90 ± 04	73.90 ± 3.60^{a}
JRC-08-12	$18.48\pm2.33^{\mathrm{bc}}$	1622.40 ± 14^{e}	$379.92\pm19^{ m d}$	$30.79 \pm 5^{\rm bcde}$	785.30 ± 09	997.69 ± 10^{a}	$69.6\pm05^{\rm a}$	68.28 ± 2.70^{ab}
JRC-08-15	$20.37 \pm 2.78^{\circ}$	$1495.27 \pm 07^{\mathrm{c}}$	$423.46 \pm 43^{\mathrm{d}}$	32.97 ± 3^{de}	1753.7 ± 17	996.31 ± 10^{a}	$98.19\pm07^{ m b}$	$137.64 \pm 4.10^{\rm c}$
JRC-08-16	$18.10 \pm 1.19^{\mathrm{bc}}$	1240.93 ± 09^{a}	$279.80\pm24^{ m bc}$	$33.36\pm4^{ m def}$	896.4 ± 16^{a}	893.04 ± 11	130.44 ± 04	121.69 ± 3.22
JRC-08-32	$28.15\pm2.21^{\mathrm{e}}$	$1465.20 \pm 10^{\rm b}$	$300.85\pm15^{\rm c}$	$27.28 \pm 4^{ m abc}$	1589.5 ± 20	982.73 ± 7^{a}	158.17 ± 07	$140.34 \pm 3.89^{\circ}$
JRC-13-11	13.81 ± 3.22^{a}	1189.32 ± 07	$205.38\pm27^{\rm a}$	$30.33 \pm 6^{\mathrm{bde}}$	850.6 ± 09	$1090.06\pm20^{\mathrm{b}}$	$97.79\pm04^{ m b}$	$71.70\pm2.90^{\mathrm{a}}$
JRC-13-13	14.75 ± 4.66^{a}	$1531.67 \pm 14^{\rm d}$	214.37 ± 09^{a}	$26.14\pm4^{\mathrm{ab}}$	750.5 ± 15	976.44 ± 25^{a}	138.44 ± 05	47.37 ± 4.11
JRC-50	$15.90\pm3.25^{\mathrm{a}}$	$1450.55 \pm 13^{\rm b}$	$309.06 \pm 12^{\circ}$	$33.18\pm6^{\mathrm{de}}$	690.7 ± 08	813.57 ± 20	113.16 ± 08	$63.67 \pm 1.25^{\rm b}$
Nagadal	$18.8\pm5.66^{\mathrm{bc}}$	1229.34 ± 11^{a}	333.34 ± 10^{cd}	23.17 ± 5^{a}	875.8 ± 12	$1087.55 \pm 20^{\rm b}$	120.33 ± 09	$69.29\pm2.33^{\rm a}$
Value = Mean	\pm SD (n = 3). The v	alues with the same sup	erscript r in a column	are not significantly	different at $p \le 0.05$			
AIA amylase in	hibitory actvity, AIU	amylase inhibition unit,	GAE gallic acid equ	ivalent, HCN hydrog	en cyanide, TAE tan	nic acid equivalent, TL	4 trypsin inhibitory a	ctivity, TIU trypsin

polyphenol content determined for rice bean varieties exhibited to be similar with the values reported by Katoch (2013) and Kaur and Kapoor (1992). The difference in polyphenol content among varieties may be endorsed by the color of seed coat, seed size, growing condition, stage of seed development and storage time. The plant genetics, growing environment, state of maturity and post-harvest conditions were reported important factors responsible for polyphenols variation (Faller and Fialho 2009). Polyphenols were reported as important source of natural anti-oxidants in human diet and have ability to prevent chronic diseases (Thomasset et al. 2007). They bind with iron, zinc, calcium, charged protein and amino acid and defaces their bioavailability.

Phytic acid

The phytic acid content of rice bean varieties varied significantly (p < 0.05) and range between 205.38 mg/100 g (JCR-13-11) and 432 mg/100 g (JCR-08-8) (Table 2) which was in consistent with the range 1875–2050 mg reported by Kaur and Kapoor (1992). JRC-08-8, JRC-08-12, JRC-08-15 and *Nagadal* exhibited comparatively high (333.33–432.14 mg/100 g) phytic acid content. Phytic acids chelated with multivalent metal ions such as zinc, calcium and iron and also bind with protein and starch and thereby reducing their bioavailability (Schlemmer et al. 2009).). Phytic acid have ability to reduce the triglycerides, serum cholesterol, iron mediated oxidation (Lee et al. 2005). It also has anti-cancer, antioxidant, anti-diabetics and kidney stone decalcification properties.

Oxalate

inhibition unit

The oxalate content (23-34 mg/100 g) in ten rice bean varieties (Table 2) varied significantly (p < 0.05). The maximum oxalate content (mg/100 g) was JRC-08-10 (34.24 ± 2) while minimum for *Nagadal* (23.17 ± 5) . The varieties JRC-08-32, JRC-13-13 and Nagadal were found to be low oxalate (23.17-27.28 mg/100 g) varieties. There are hardly any reports on oxalate content of rice bean. The oxalate content of rice bean was similar to other bean like azuki bean, black gram. The oxalate content of azuki bean (Vigna angularis) was reported as 25 mg/100 g and of black gram as 30 mg/100 g (Campos-Vega et al. 2010; Chai and Liebman 2005). The higher intakes of oxalates rich food led to lowering of absorption of calcium which resulted in formation of kidney stones (Noonan and Savage 1999). Massey et al. (2001) reported that the kidney patients needs to intake of oxalate not exceeding 50-60 mg per day. However, the processing reduced the oxalate content of the beans.

Saponin

The saponin content of rice bean varieties ranged between 690 mg/100 g and 1753.7 mg/100 g (Table 2). Saponin content varied among varieties because of genetics factors, locations, irrigation condition, type of soil and climatic conditions, (Campos-Vega et al. 2010). JRC-08-7, JRC-08-8, JRC-08-15 and JRC 08-32 were high (1097.8-1753.7 mg/ 100 g) in saponin content whereas JRC-50 contained the lowest content (690.7 \pm 08 mg/100 g). The average saponin content (1025.34 \pm 329 mg/100 g) in eleven rice bean varieties was lower than the earlier reported value. The average saponin content in rice bean was reported as 2285 mg/100 g for five rice bean varieties by Kaur and Kapoor (1992) while Saharan et al. (2002) reported as 2168 ± 8.4 mg/100 g. Saponin reduced the nutrient bioavailability and enzyme activity which ultimately contributed to a growth-retarding effect in animals.

Tannin

The tannin content (813.57-1090 mg TAE/100 g) of rice bean varieties (Table 2) varied significantly (p < 0.05). JRC-08-8, JRC-13-11 and Nagadal were found high (1064.11-1090.06 mg of TAE/100 g) in tannin content than other varieties whereas JRC-08-16 had the least i.e. (813.57 mg of TAE/100 g). The average tannin content $(991.58 \pm 78 \text{ mg} \text{ of TAE}/100 \text{ g})$ in eleven rice bean varieties was found to be close the earlier reported value between 1370-1550mg/100g Katoch (2013) for 16 rice bean genotypes. Verma and Mehta (1988) reported that rice bean (993 m/100 g) had higher tannin content than mung bean (612 mg/100 g). The two contrary attributes of tannin such as anti-nutritional and defence mechanisms against pathogens and pests has fascinated the plant breeder toward this compounds. Tannins form complexes with proteins, starch, minerals, digestive enzymes, vitamin B₁₂ (Leiner 1980) and reducted the nutritional value of foods. These can cause browning in foods through the action of polyphenol oxidase by darkening reactions that adversely affected the acceptability of such foods.

Hydrogen cyanide

The HCN content of rice bean varieties ranged from 49.9 to 138.44 µg/100 g (Table 2) which varied significantly (p < 0.05) among varieties. JRC-08-7, JRC-08-10, JRC-08-12 exhibited low (49.90–71.17 µg/100 g) HCN content whereas variety JRC-08-32 displayed the highest (158.17 ± 07 µg/100 g) HCN content. The average HCN content (104.71 ± 31 µg/100 g) of eleven rice bean varieties was higher than cowpea. Owolabi et al. (2012) reported HCN content as 0.7–37 µg/100 g for five varieties

of cowpea (*Vigna unguiculata*). HCN level in rice bean varieties was far below the lethal level i.e., 36 mg/100 g (Oke 1969). The pulse food was considered to be safe if cyanide content ranged from 10 to 20 mg/100 g (Manay and Sharaswamy 2008). The conventional processing such as soaking, dehulling and cooking can eliminate this anti-nutritional factor from bean.

α-Amylase inhibitory activity

The α -Amylase inhibition activity of rice bean varieties were ranged from 13.81 amylase inhibitory unit (AIU)/g (JCR-13-11) to 28.15 AIU/g (JCR-08-32). The AIA was low in JCR-13-11, JCR-13-13, JCR-50 whereas high in JCR-08-32, JCR-08-8, JCR-08-10 and JCR-08-7. The average AIA of rice bean was higher than the reported value (Katoch and Jamwal 2013). Katoch and Jamwal (2013) reported the AIA as 7.529–10.766 (IU/g) for 13 rice bean genotypes. The α -amylase inhibitor, proteineous compounds was reported to bind with active site of α amylase and reduced or inhibit their enzymatic activities which results in a decrease in starch digestibility. It is useful for diabetic and obese humans.

Trypsin inhibitory activity

Trypsin inhibitory activity varied from 47.37 to 140.34 trypsin inhibitory unit (TIU)/g with average of 84.67TIU/g. JCR-08-32, JCR-08-15, and JCR-08-16 showed higher TIA as compare to other varieties. TIA of rice bean genotypes was reported as 55.2–163.98TIU/g (Rejaul et al. 2016). Trypsin inhibitors inhibited the activities of protein digesting enzymes which reduced protein efficiency ratio. It also depressed the growth and may cause pancreatic hypertrophy.

Mineral profile

The minerals (mg/100 g) such as potassium, magnesium, calcium, sodium, zinc, iron, manganese, copper, chromium and cobalt varied in the range of 610.44–1517, 77.91–165, 111.51-168, 10.83-24.18, 3.72-8.37, 5.50-10.44, 2.04-5.00, 0.68-2.09, 0.02-0.11, and 0-0.02, respectively (Table 3). Nagadal variety revealed higher content of different minerals (mg/100 g) viz. potassium (1517.0 \pm 23) magnesium (165.0 \pm 0.09), calcium (168 \pm 2.00), sodium $(24.18 \pm 0.17),$ iron (8.37 ± 0.19) and copper (2.09 ± 0.10) as compared with other varieties. However, JRC-08-7 contained the lowest level of potassium (610.4 ± 04) , sodium (10.83 ± 0.05) , iron (3.72 ± 0.08) , manganese (2.04 ± 0.03) , and copper (0.68 ± 0.02) . There was no significant difference in calcium content among varieties except Nagadal whereas other minerals

Table 3 Mine	sral (mg/100 g) pro	file of rice bean varie	eties							
Varieties	K^{766}	Mg^{279}	Ca ³⁹⁶	Na^{588}	Zn^{206}	Fe ²⁵⁹	Mn ²⁵⁷	Cu ³²⁷	Cr^{367}	C0 ²²⁸
JRC-08-7	610.4 ± 04	$155.6\pm0.66^{\rm ab}$	$133.3 \pm 3.21^{\rm b}$	10.83 ± 0.05	$8.45\pm0.23^{\mathrm{c}}$	3.72 ± 0.08	2.04 ± 0.03	0.68 ± 0.02	0.02^{a}	ND
JRC-08-8	743.5 ± 04	77.9 ± 0.58	125.6 ± 4.22^{ab}	$19.48\pm0.16^{\mathrm{bc}}$	$7.57\pm0.24^{ m b}$	$6.57\pm0.05^{\rm c}$	$3.62\pm0.02^{ m b}$	1.28 ± 0.01	0.03	0.01
JRC-08-10	$1065.6\pm09^{\rm cd}$	$159.9\pm0.30^{\rm ab}$	$129.7 \pm 5.32^{\mathrm{b}}$	21.01 ± 0.25	10.44 ± 0.28	$7.36\pm0.11^{ m d}$	4.45 ± 0.10	1.62 ± 0.01	0.02^{a}	0.02^{a}
JRC-08-12	$1028.1\pm09^{\mathrm{ab}}$	$161.9\pm0.55^{\mathrm{b}}$	$131.1 \pm 3.44^{\rm b}$	12.61 ± 0.31	$6.34\pm0.12^{\rm a}$	$5.58\pm0.02^{\rm a}$	$3.65\pm0.06^{\mathrm{b}}$	1.03 ± 0.02	0.02^{a}	0.02^{a}
JRC-08-15	$1006.9\pm07^{\mathrm{a}}$	$160.3\pm0.58^{\rm ab}$	$131.4\pm1.36^{\rm b}$	17.96 ± 0.21	9.98 ± 0.30	$6.23\pm0.13^{ m b}$	$3.37\pm0.04^{\mathrm{a}}$	$1.19\pm0.02^{\mathrm{b}}$	0.02^{a}	0.02
JRC-08-16	1145.1 ± 13	$158.5\pm0.45^{\rm ab}$	$130.7\pm5.33^{\mathrm{b}}$	21.51 ± 0.04	$7.28\pm0.20^{\mathrm{b}}$	$6.20\pm0.18^{\rm b}$	3.17 ± 0.06	1.44 ± 0.05	0.02^{a}	0.02^{a}
JRC-08-32	$1187.0\pm08^{\rm e}$	$157.9\pm0.18^{\rm a}$	$131.3\pm3.33^{\mathrm{b}}$	$19.17\pm0.36^{\mathrm{ab}}$	$6.24\pm0.30^{\rm a}$	$6.34\pm0.01^{ m bc}$	$3.40\pm0.12^{\mathrm{a}}$	$1.13\pm0.02^{\rm a}$	0.04^{b}	0.02^{a}
JRC-13-11	1072.0 ± 13^{d}	$143.3 \pm 0.37^{\rm a}$	$111.5\pm2.25^{\rm a}$	$19.54\pm0.31^{ m c}$	$6.54\pm0.04^{\rm a}$	5.55 ± 11^{a}	2.86 ± 0.04	1.33 ± 0.01	0.02^{a}	0.02^{a}
JRC-13-13	$1169.2 \pm 01^{\mathrm{e}}$	$159.7\pm0.21^{\rm ab}$	131.8 ± 1.26^{b}	$18.16\pm0.40^{\mathrm{a}}$	5.50 ± 0.07	5.26 ± 0.19	2.70 ± 0.06	$1.21\pm0.03^{\mathrm{b}}$	0.04^{b}	0.02^{a}
JRC-50	$1047.9 \pm 17^{\rm bc}$	$160.2\pm0.79^{\rm ab}$	129.7 ± 1.19^{b}	19.02 ± 0.24^{a}	$8.24\pm0.18^{\rm c}$	7.30 ± 0.23^{d}	5.00 ± 0.08	$1.11\pm0.02^{\mathrm{a}}$	0.04^{b}	0.02^{a}
Nagadal	1517.0 ± 23	$165.0\pm0.09^{\mathrm{b}}$	168.0 ± 2.00	24.18 ± 0.17	$7.49 \pm 0.06^{\mathrm{b}}$	8.37 ± 0.19	4.08 ± 0.10	2.09 ± 0.10	0.11	ND
Value = Mea	$n \pm SD (n = 3). Tt$	ie values with the sam	te superscript in a col	lumn not significantly	y different at $p \leq 0$.	05. Minerals supers	cript indicates wav	elength of analysis	, ND: Not e	letected



Fig. 2 Mineral ratio of rice bean varieties (a, b)

varied significantly (p < 0.05) among varieties. The variation among varieties may be due to genetic and environmental factors, agronomics practice and soil composition. The average potassium (1053 \pm 223.7 mg/100 g), magnesium $(150.9 \pm 23.68 \text{ mg}/100 \text{ g}),$ calcium $(132.0 \pm 12.69 \text{ mg}/100 \text{ g})$, sodium $(18.50 \pm 3.62 \text{ mg}/100 \text{ g})$ 100 g), iron $(6.22 \pm 1.18 \text{ mg}/100 \text{ g}),$ manganese $(3.49 \pm 0.78 \text{ mg}/100 \text{ g})$, copper $(1.28 \pm 0.34 \text{ mg}/100 \text{ g})$ zinc $(7.64 \pm 1.45 \text{ mg/100 g})$ content among eleven varieties was found within range of reported earlier (Rejaul et al. 2016).

The important mineral ratio is shown in Fig. 2a, b. Na/K ratio in the body is of great concern for prevention of high blood pressure, Na/K ratio was less than recommended. Hence, in the present study, all the varieties of rice bean would probably help to reduce high blood pressure due to low Na/K (0.01–0.03). In addition, high K/Na ratio in plants parts is important indicator for stress tolerance (Wenxue et al. 2003). Diets low in potassium and high in sodium may partially or directly linked with chronic illnesses, including hypertension, stroke, kidney stones, osteoporosis etc. (Afolabi et al. 2015). The healthy ratio of Ca:Mg in diet of humans should be 2:1. The ratio of Ca:Mg in this study was lower than the recommended values. Lower Ca:Mg ratios increased the risk for heart diseases. In most humans, healthy blood pressure depends upon a



Fig. 3 a Thermo-gravimetric profile of rice bean varieties, **b** percentage of weight loss at different temperature zone

balance of both Na:K and Mg:Ca ratios at both cellular and whole body levels which, in turn, require adequate, long-term intakes of nutritional magnesium (Afolabi et al. 2015). The Na/Mg, Ca/K and Fe/Cu ratio among rice bean varieties ranged between 0.07–0.25, 0.1–0.22, and 4.00–5.61 respectively. Each of these ratio have nutritional significance in humane diet.

Thermo-gravimetric analysis

The TGA was done to evaluate the behavior and influence of constituents of rice bean such protein, carbohydrate and minerals during processing. The TGA curve of all varieties (Fig. 3a) exhibited uniform trend; however it can be grouped into six different thermal events/stages (Fig. 3b). Each of this stage has significant correlation with chemical composition (Table 4). In the first stage of heating (0–200 °C), all the varieties showed 10.32–12.60% weight loss which may be attributed to free water evaporation and volatile compounds. In first stage, JRC-08-12 showed minimum weight loss whereas JRC-08-8 showed maximum. In second stage (200–270 °C), the weight loss of all analyzed varieties was ranged from 5.83 to 6.92%. This loss may be attributed to loss of bound water, disintegration

	Moisture	CHO	Protein	Fat	Ash	Polyphenol	Phytic acid	Oxalate	Tannin	HCN	Potassium	Magnesium	Sodium	Zinc	Copper
WL1	-0.435													0.514	
WL2	0.695						-0.498			0.453	0.486				
WL3				0.448	0.466		-0.544				0.681	0.459	0.592		0.503
WL4		0.507	-0.601					-0.740		0.741	0.450			-0.856	
WL5		0.591	-0.672	0.471				-0.553	0.621		0.493				0.483
WL6		-0.627	0.728	-0.565				0.549	-0.565		-0.591		-0.449		-0.553
DG	-0.512			0.768	0.476	-0.560			0.553						0.671
DG de	gree of gelati	nization, WI	LI weight lo	58 in first st fth stage (4	tage (0–20 50–530 °C	0 °C), <i>WL2</i> we	ight loss in seco	ond stage (2)	00–270 °C) °C) <i>CHO</i>), <i>WL3</i> we	ight loss in thi	ird stage (270–3 trogen evanide	50 °C), WL	4 weight los	s in forth

of simple sugar etc. In this stage, the gravimetric reduction (weight loss) was positively correlated with levels of moisture (r = 0.68), potassium (r = 0.49) content while negative correlation with phytic acid (r = -0.50) content. The weight loss among varieties in third stage (270-350 °C) was ranged between 35.53 (JRC-08-8) and 42.38% (JRC-13-11). This loss may be attributed to decomposition of starch, oligosaccharide. Khan et al. (2015) reported 60% carbon loss at 220.85-407.7 °C to cellulose and hemicelluloses. The weight loss in this third stage showed positive correlation with levels of fat (r = 0.44, p < 0.05), ash (r = 0.46, p < 0.05), potassium (r = 0.68, p < 0.05), sodium (r = 0.59, p < 0.05) and copper (r = 0.50, p < 0.05) whereas negative correlation with phytic acid content (r = -0.54, p < 0.05). The weight loss in fourth stage (350-450 °C) was ranged from 10.64 (JRC-08-10) to 14.39% (JRC-13-13). The loss in this stage may be contributed by complex carbohydrates including lignin. Yang and Wu (2009) reported that the cellulose and hemicelluloses were predominantly degraded below 400 °C while lignin materials above 400 °C. In the present report, weight loss in 4th stage was positively correlated (0.50-0.74) with carbohydrate (r = 0.507,p < 0.05) and HCN (r = 0.741, p < 0.005) contents and negative with protein(r = -0.601, p < 0.05), oxalate(r = 0.741, p < 0.005) and zinc contents(r = -0.85, p < 0.005) ranged from 13.36% (JRC-50) to 25.57% (Nagadal) which may be due to delayed decomposition of nitrogenous compound such as protein associated complexes. The weight loss in this stage showed positive correlation with carbohydrate (r = 0.59, p < 0.05), fat (r = 0.67, p < 0.05), tannin (r = 0.62, p < 0.05), potassium (r = 0.49, p < 0.05) and negative with protein (r = -67, p < 0.05) and oxalate (r = -0.55, p < 0.05). The weight loss of sixth stage (530-590 °C) may be due decomposition of complex nitrogenous compound. The weight loss in sixth stage showed positive correlation with protein (r = 0.728, p < 0.005), oxalate (r = 0.549,p < 0.05) and and negative correlation with carbohydrate (r = -627, p < 0.05), fat(r = -0.565, p < 0.05), tannin(r = -0.565, p < 0.05), potassium (r = -0.591, p < 0.05)and copper (r = -0.553, p < 0.05). The maximum weight loss was observed in third stages (35.43-42.38%) whereas minimum weight loss (0.54-5.93%) in sixth stage except varieties JRC-50 and JRC-08-7. The degradation of carbohydrates, fat start at second stage and ended in fifth stage. The maximum decomposition of carbohydrates and proteins occurred at fifth and sixth stage, respectively.

Cooking characteristics

The Cooking Characteristics of eleven rice bean varieties is shown in Table 5.

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Varieties	Grain hardness ((N)		Cooking characteristic	ss cooking			
	Native	Soaked	Cooked	Cooking time (min)	Water uptake (%)	Solid dispersion (%)	Swelling ratio	Degree of gelatinization ($\%$
JRC-08-7	128.61 ± 2.16	$70.24 \pm 1.18^{\mathrm{ab}}$	25.11 ± 2.16^{bc}	32 ± 3^{ab}	124.0 ± 11^{cd}	$26.8 \pm 5.5^{\circ}$	1.11 ± 0.32^{a}	50.32 ± 3.65^{b}
JRC-08-8	114.88 ± 2.45	$53.46\pm1.86^{\rm c}$	$24.03 \pm 1.57^{\mathrm{ab}}$	$30 \pm 2^{\rm c}$	$115.0\pm15^{\mathrm{ab}}$	$25.0\pm 8.5^{ m b}$	$0.996\pm0.55^{\rm a}$	$52.66 \pm 2.25^{\rm b}$
JRC-08-10	66.51 ± 1.77	$43.56\pm2.55^{\rm d}$	$23.54\pm2.06^{\rm a}$	$29\pm3^{\mathrm{a}}$	$118.8 \pm 13^{\mathrm{bc}}$	$27.7 \pm 7.4^{\rm cd}$	$1.09\pm0.33^{\mathrm{a}}$	$51.65 \pm 1.65^{\mathrm{b}}$
JRC-08-12	72.5 ± 1.96	$47.97 \pm 1.96^{\mathrm{d}}$	$23.45\pm1.57^{\rm a}$	$29\pm3^{\mathrm{a}}$	$108.5\pm10^{\mathrm{a}}$	28.5 ± 6.6^{a}	$0.988 \pm 0.33^{\rm a}$	$52.11\pm3.00^{\mathrm{b}}$
JRC-08-15	94.86 ± 2.26	$53.66\pm1.76^{\rm c}$	$26.19 \pm 1.47^{\text{cdef}}$	$39 \pm 5^{\rm c}$	114.2 ± 13^{a}	$24.5 \pm 3.2^{\mathrm{b}}$	$0.997\pm0.45^{\mathrm{a}}$	49.32 ± 2.21^{a}
JRC-08-16	87.21 ± 1.86	$58.47 \pm 2.65^{\circ}$	25.11 ± 2.26^{bcd}	$40 \pm 2^{\rm d}$	$126.5 \pm 12^{ m cd}$	$29.0\pm 8.5^{\mathrm{ad}}$	$1.222\pm0.65^{\rm a}$	$52.32 \pm 1.78^{\rm b}$
JRC-08-32	101.04 ± 2.40	$67.98 \pm 2.35^{\rm b}$	$27.76 \pm 1.37^{\mathrm{f}}$	$38 \pm 1^{\rm c}$	$131.0 \pm 11^{\rm d}$	$26.0 \pm 7.7^{\circ}$	$1.320\pm0.32^{\rm a}$	49.11 ± 1.95^{a}
JRC-13-11	77.6 ± 1.76	$60.33 \pm 1.76^{\rm bc}$	$26.00 \pm 1.67^{\mathrm{cde}}$	$37 \pm 2^{\rm c}$	$125.0\pm09^{ m cd}$	$25.5 \pm 4.5^{\mathrm{bc}}$	$1.022\pm0.22^{\mathrm{a}}$	$52.49 \pm 1.55^{\mathrm{b}}$
JRC-13-13	130.08 ± 2.55	$63.27 \pm 1.86^{\rm bc}$	$27.96 \pm 1.76^{\rm f}$	$35 \pm 3^{\rm c}$	$108.7\pm10^{\mathrm{ab}}$	$25.3 \pm 5.5^{\mathrm{b}}$	$0.998 \pm 0.23^{\rm a}$	48.65 ± 2.65^{a}
JRC-50	137.73 ± 3.24	$75.14\pm2.35^{\mathrm{a}}$	$26.78 \pm 1.27^{\mathrm{ef}}$	$41 \pm 3^{\rm d}$	132.0 ± 15^{d}	$27.5\pm8.0^{\circ}$	$1.350\pm0.45^{\mathrm{a}}$	49.22 ± 2.40^{a}
Nagadal	75.05 ± 2.31	52.88 ± 1.86^{cd}	21.58 ± 1.57	30 ± 5^{a}	127.5 ± 17^{cd}	$28.5\pm6.5^{ m d}$	1.115 ± 0.70^{a}	55.05 ± 1.66
Value – Me	(1) = 30 (n - 3)	The values with the	e same sunerscrint ir	a column not significat	ntly different at $n < 0$	0.65		

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 Table 5
 Cooking characteristics of rice bean varieties

Hardness

The hardness of native rice bean grains ranged from 66.51 N (JCR-08-10) to 137.73 N (JCR-50) and varied significantly (p < 0.05) among the varieties. JRC-08-7, JRC-08-8, JRC-08-32, JRC-13-13, and JRC-50 observed native hardness of more than 100N. The average value of native hardness (98.73 N) was exhibited to be lesser than mung bean but more than but cowpea (Henshaw 2008, Kaur et al. 2015). The native hardness was directly related to grain structure, packing and compactness of seed constituents such carbohydrates, protein and other phytochemicals with minerals. It was one of the important physical parameter for size reduction or milling, soaking and cooking. The hardness of soaked and cooked rice bean grains ranged from 43.56 N (JCR-08-10) to 75.14 N (JCR-50) and 21.58 N (Nagadal) to 27.96 N (JCR-13-13), respectively. The results of cooked and soaked hardness were agreed with earlier reported results. (Singh et al. 2010, Kaur et al. 2013, Wani et al. 2013). The soaking and cooking treatment reduced the native hardness by 22.25-53.46 and 64.60-80.6%, respectively which concurred with earlier reported results (Kaur et al. 2013).

Cooking time, water uptake, solid dispersion and swelling ratio

CT with prior soaking (12 h) significantly (p < 0.05) varied among the varieties. CT varied from 29 to 41 min, JCR-50 displayed the highest and JCR-08-10, JCR-08-12 displayed the lowest CT. Similar findings have been reported earlier in other varieties of rice bean (Kaur et al. 2013). A positive correlation (0.496) of CT and native hardness was observed. The water uptake of varieties ranged from 108.5% for JCR-08-12 to 132.0% for JCR-50. The water uptake was an important cooking parameter that increased the yield of cooked grain. Water uptake of grain depended upon the seed coat structure, grain composition and compactness of the seed cells. The solid dispersion of analyzed varieties ranged from 24.5% (JCR-08-15) to 28.5% (JCR-08-12, Nagadal). The water uptake and solid dispersion of studied varieties exhibited more while CT lesser than the earlier reported values (Hira et al. 1988; Saikia et al. 1999). These variations were due to the soaking treatment. CT water uptake and solid dispersion of rice bean without soaking was reported as 50-57 min, 94-117% and 4.2-12%, respectively. The swelling ratio of rice bean varieties ranged between 0.996 and 1.350. The higher value of swelling ratio was displayed by JCR-50, JCR-08-32, JCR-08-16, JCR-13-11, JCR-08-7 and Nagadal. For optimum cooking quality, the variety should have maximum water uptake and swelling ratio and minimum solid dispersion, and cooked grain hardness.

Degree of gelatinization

DG of rice bean varieties during cooking is given in Table 5. The degree of starch gelatinization ranged from 48.65 to 55.33%. The highest (55.33%) gelatinization was observed for Nagadal. DG was attributed to starch content and their starch properties, including amylose/amylopectin content and gelatinization enthalpy. The DG of rice bean varieties was lower than the mung bean varieties (62.99%). Arntfield et al. (1997) observed 29% starch gelatinized when lentil was pre-tempered to 25% moisture and increased to 68% when moisture was increased to 33%. DG depended on seed composition, cooking and hydration and thermal properties. The varieties with high polyphenol content showed less DG. These varieties may be having thick seed coat which decreased the water absorption capacity during 12 h of soaking and resulted in reduced DG. It was observed that DG exhibited (Table 4) negative correlation with polyphenol (-0.56), and grain hardness (-0.64). However, DG showed positive correlation with tannin (0.55), copper (0.68) and fat (0.77). The tannin content decreased during soaking due to leaching in soaked water. The leaching of tannin facilitated water to enter into seed through seed coat which increased the swelling ratio. The swelled seed having more water content which bound with starch may have led to higher degree of gelatinization during cooking.

In addition, it was witnessed that the weight loss graph of fifth stage (450–530 °C) followed reverse trend with that of sixth stage (530–590 °C) as seen Fig. 3b. The weight loss graph of fifth stage (450–530 °C) followed similar trend with DG values (Table 5) except for JRC-08-15 and JRC-08-32 varieties. In JRC-08-15, the DG is low due to presence high polyphenol content whereas JRC-08-32 showed high DG due to presence of high fat and carbohydrate content. Based on the relation between the DG and the weight loss of fifth and sixth stage, the TGA curve can be used for prediction of DG. The *Nagadal* showed highest weight loss at fifth stage (450–530 °C) and lowest weight loss at six stage (550–590 °C) which also showing highest DG.

Principal component analysis

Principal component analysis (PCA) was carried out to observe the relationships between the chemical, thermogravimetric and cooking parameters of rice bean varieties (Fig. 4). The PC-1, PC-2, PC-3 and PC-4 represented for 68.9% of cumulative variance. PC-1 asserted for % variance with protein, fat, ash, oxalate, potassium, calcium, sodium, copper, native hardness, DG, WL3 (weight loss in third stage), WL5 (Weight loss in fifth stage), WL6 (Weight loss in sixth stage) being the chief contributor. PC-



Fig. 4 Loading plot of first two components from PCA analysis for chemical, thermo-gravimetric and cooking parameters of rice bean varieties. *HCN* hydrogen cyanide, *TIA* trypsin inhibition activity, *AIA* amylase inhibition activity, *C.Fibre* crude fibre, *K* potassium, *Na* sodium, *Ca* calcium, *Fe* iron, *Cu* Copper, *Mn* Manganese, *Zn* Zinc, *N.Hard* native hardness, *S.Hard* soaked hardness, *C.Hard* cooked

2 accounted for 18.6% variance with moisture, phytic acid, HCN, Zinc, soaked hardness, cooked hardness, cooking time, WL2 (weight loss in second stage) and WL4 (weight loss in forth stage) being the major contributor. PC-3 accounted for 14.7% variance with carbohydrate, tannin, iron, manganese, water uptake, swelling ratio being the key player. PC-4 accounted for 9.9% variance with AIA, TIA, saponin and solid dispersion was major player. Polyphenol, protein, oxalate, zinc, phytic acid and WL6 were situated on the same side indicating their positive relationships with each other. Oxalate, protein, polyphenol were located toward opposite side of carbohydrate which indicated their negative relationship. The variety with lower carbohydrate content had higher oxalate, protein and polyphenol. The cooking time, swelling ratio, soaked hardness, cooked hardness and native hardness were situated on same side which indicated positive relationship between whereas these parameters were also located toward opposite side of calcium, iron, DG, ash, solid dispersion indicated negative correlation. The native hardness was positioned in an angle of ninety degree with protein and carbohydrate which showed no relation with them. WL2, WL3, WL4 were having positive relationship with carbohydrate and negative relationship with protein and WL6 (Fig. 4).

Conclusion

The 11 varieties of rice bean from NE region of India had the average content of carbohydrates, protein, fat, crude fibre, ash, polyphenols, tannin, calcium and iron content of

hardness, *CT* cooking time, *WU* water uptake, *SD* solid dispersion, *SR* swelling ratio, *DG* degree of gelatinization, *WL1* weight loss in first stage (0–200 °C), *WL2* weight loss in second stage (200–270 °C), *WL3* weight loss in third stage (270–350 °C), *WL4* weight loss in fourth stage (350–450 °C), *WL5* weight loss in fifth stage (450–530 °C), *WL6* weight loss in six stage (530–590 °C)

13.13 \pm 0.34%, 57.51 \pm 2.17%, 18.44 \pm 2.14%, 1.54 \pm 0.30%, $5.89 \pm 0.37\%$, $3.49 \pm 0.12\%$, 1422.95 ± 155 mg GAE/100 g, 991.58 \pm 77.59 mg TAE/100 g, 132.22 \pm 5.58 mg/100 g and 6.23 ± 1.54 mg/100 g, respectively. JRC-08-7 had the lowest ash, potassium, sodium, iron and copper content whereas JRC-08-10 had the highest content of polyphenols, oxalate, and zinc. JRC-50 had the highest protein, manganese and the lowest of carbohydrates, fat, saponins and tannin content. However, Nagadal had highest content in fat, ash, HCN, potassium, magnesium, calcium, sodium, iron, copper and chromium. JRC-08-10 had the lowest cooking time whereas JRC-50 had the highest cooking time and cooking hardness, water uptake, swelling ratio as well. Nagadal showed the highest weight loss at fifth stage (450-530 °C) and the lowest weight loss at six stage (550-590 °C) which also showed highest DG. Principle component analysis revealed that varieties with higher calcium, iron, ash had lower cooking time, swelling ratio, and cooked hardness. Nagadal was found to be the best in term of DG, mineral content, cooking and thermal properties. Hence, among the varieties evaluated, Nagadal had higher carbohydrates content and has potential to develop of value added products through extrusion, flaking and frying technologies.

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