

Genetic insights into fatty acid components of traditional Indian rice (*Oryza sativa* L.) landraces from Chhattisgarh

Parmeshwar K. Sahu, Suvendu Mondal¹, Deepak Sharma*, Richa Sao, Vikash Kumar¹ and Bikram K. Das¹

Department of Genetics and Plant Breeding, Indira Gandhi Krishi Vishwavidyalaya, Raipur 492 012; Chhattisgarh; ¹Nuclear Agriculture and Biotechnology Division, Bhabha Atomic Research Centre, Mumbai 400 085

(Received: July 2019; Revised: October 2019; Accepted: November 2019)

Abstract

Knowledge about the contents and type of fatty acids (FAs) in rice bran is beneficial, particularly from a nutritional and health standpoint. An experiment was conducted to assess the genetic variability and diversity for FA components in 215 rice landraces during kharif 2015 and kharif 2016 by following the RCB design with two replications. Palmitic (C16:0), oleic (C18:1) and linoleic (C18:2) were the major fatty acids ranging from 12.59% to 20.25%, 37.60% to 49.17% and 31.55% to 44.67%, respectively. Analysis of variance revealed significant differences in all the FA components except for linolenic acid. Results showed that environmental factors play a significant impact on the expression of FA contents. Fatty acid components showed intermediate to low genotypic and phenotypic coefficient of variation, intermediate to high heritability and low to moderate genetic advance as percent of the mean. Oleic acid content was negatively correlated with palmitic acid, stearic acid, and linoleic acid contents. Principal component analysis and cluster analysis discriminated the 215 rice landraces into five main groups with a major contribution of oleic acid, linoleic acid, and palmitic acid contents. Landraces Kadamphool, Ratanchudi and Bathrash possessed the highest amount of oleic acid/linoleic acid ratio. Information generated through this study will be functionally useful in developing rice varieties having high-quality bran oil.

Keywords: Bran oil, fatty acids, genetic variation, landraces, rice

Introduction

Rice (*Oryza sativa* L.) is the major crop for the global food and nutritional security and socio-economic development, therefore, its production potentials have to be enhanced significantly to feed the burgeoning population along with good nutritional quality traits (FAO 2017). Carbohydrate is the major component of rice, however, it has a reasonable amount of protein and oil

components mainly in the bran portion. Amount of vitamin-B complexes such as niacin, riboflavin, and thiamin has been also reported in rice bran. Attentiveness on health benefits and nutritive value and of rice is essential to increase the intake of rice in the daily diet of human beings (Fresco 2005).

Nowadays, rice bran and its oil have been getting special attention to use as a functional food due to their nutritional properties *viz.*, phenolic compounds, beneficial fatty acids, vitamins, minerals and fibers (Wilson et al. 2002). Rice bran is the outer aleuron layer of the rice kernel which can be separated during milling/ whitening of brown rice and accounted approximately 10% of the weight of rice grain (Rondanelli et al. 2011). Bran portion has ample amount of antioxidants *viz.*, tocopherols, tocotrienols, and ãoryzanol and phenolic compounds, which help to reduce LDL cholesterol, protect cardiovascular diseases and possess anticancer effects (Min et al. 2011).

Looking into the significance of bran oil, the characterization of rice landraces for FA composition may have great importance for future breeding programs. Despite abundant genetic resources available, breeders have utilized only limited rice germplasm for the improvement of fatty acid components, which cause their narrowed genetic base in improved varieties and advance breeding materials. Therefore, a study was conducted to characterize 215 rice landraces through genetic analysis based on fatty acid composition in the bran fraction. It is expected that the information obtained would help assure an effective breeding program for developing rice varieties

*Corresponding author's e-mail: deepakigkv@gmail.com

Published by the Indian Society of Genetics & Plant Breeding, A-Block, F2, First Floor, NASC Complex, IARI P.O., Pusa Campus, New Delhi 110 012; Online management by www.isgpb.org; indianjournals.com

having ample amount of desirable fatty acids.

Materials and methods

The experimental material comprises 213 traditional landraces and two improved rice varieties (Mahamaya and Rajeshwari as control) from Chhattisgarh state, India (Supplementary Table S1). All the genotypes were procured from R.H. Richharia Rice Biodiversity unit of the Indira Gandhi Krishi Vishwavidyalaya (IGKV), Raipur for research purpose. All 215 genotypes were grown at the rice research field of IGKV, Raipur for two years viz., kharif 2015 and kharif 2016 by following the randomized complete block design (RCBD) with two replications. Agronomic management practices were performed as per the standard evaluation system of IRRI, Philippines (IRRI 2002). Genotypes were harvested separately, cleaned properly, dried up to 12-14% moisture content and kept at room temperature for further utilization.

Extraction of bran, sample processing and estimation of fatty acid components

Hulling and milling process of each rice genotype was done by using a McGill No. 2 Huller & Miller (Rapsilver Supply Co. Inc., Brookshire, TX) at R.H. Richharia Rice Research Laboratory, Department of Genetics and Plant Breeding, IGKV, Raipur (C.G.), India. Bran of each sample of brown rice (decorticated rice) was collected into a small stripped polythene bag and labeled. Rice bran of each genotype was kept immediately at 4°C to avoid the harmful activities of lipase enzyme. Fatty acid estimation was done by the base-catalyzed trans-esterification method (Mondal et al. 2018) in Gas Chromatography (Shimadzu, Kyoto, Japan) at Nuclear Agriculture and Biotechnology Division (NA&BTD), Bhabha Atomic Research Centre (BARC), Mumbai, India. About 200 mg of rice bran of each genotype was taken into a 50 ml glass test tube in duplicates. One milliliter (1ml) of methanol (AR) and 1ml of 0.5M sodium methoxide (AR) were added in each tube. All the tubes were shaken thoroughly by vortex and kept at room temperature for 20 minutes. Then all the tubes were kept into a water bath at 50°C for 1 hour. After water bath treatment, tubes were kept for 10 min at room temperature for cooling. Thereafter, 2 ml of HPLC grade petroleum ether (boiling point = 60-80 °C) and 2 ml of de-ionized water was poured in each tube and vortexed for 1 min. Then tubes were kept at room temperature for 30 min to allow the phase separation. The upper petroleum ether phase was

extracted from each tube by using 1ml micro-pipette and transferred them into 1.8 ml clear GC vials. Gas chromatography (GC2010, Shimadzu, Japan) technique was used to analyze the processed samples and the final concentration of fatty acid components were obtained from GC SOLUTION software (Shimadzu, Japan) after normalization of peak areas and the recorded data were converted into a percentage value.

Statistical analysis

Proportional contents of fatty acids were transformed by ArcSine transformation as per the method suggested by Sokal and Rohlf (1995) to normalize the dataset for further analysis. Descriptive statistics, box plots, and histogram were made by using XLSTAT v18.07 (Addinsoft, NY, USA). Analysis of variance (ANOVA), genetic parameters of variability and genotypic correlation coefficients were calculated by software WINDOSTAT v9.3. Principal component analysis (PCA) based on the covariance matrix and agglomerative hierarchical cluster (AHC) analysis by following the Euclidean distance and Ward's algorithm was performed in PAST v3.15 software (Hammer et al. 2001).

Results

Gas chromatography (GC) analysis of rice bran oil revealed five fatty acid components *viz.*, oleic (C18:1), linoleic (C18:2), palmitic (C16:0), stearic (C18:0), and linolenic acid (C18:3). Analysis of variance (ANOVA) reported high significant differences for palmitic, stearic, oleic and linoleic acid during the year 2015, 2016 and over the year analysis, but they are non-significant for linolenic acid in all the conditions (Table 1). ANOVA over the year showed highly significant effects of the year for all the fatty acid components. Genotype x year interaction was significant for fatty acid components apart from for linolenic acid in rice landraces.

Genetic analysis of fatty acid contents

Since fatty acid components are very sensitive to environmental conditions therefore over the year (pooled data) analysis is more appropriate for describing the descriptive statistics, variability parameters, correlations and diversity parameters in the study. Pooled data may reduce the chances of error and combine the overall effect of environmental conditions. Palmitic acid ranged from 12.59% (Khajoor) to 20.25% (Lajini Super) with an average of 17.54%

Year	Source of variation	df		Mean sum of square					
			Palmitic acid	Stearic acid	Oleic acid	Linoleic acid	Linolenic acid		
2016	Replication	1	0.119	0.904	0.164	0.512	0.01		
	Treatment	214	2.955**	1.321*	2.77**	2.261**	0.626		
	Error	214	0.255	0.367	0.296	0.344	0.312		
	Total	429							
2017	Replication	1	1.653	1	0.67	0.738	0.119		
	Treatment	214	3.827**	1.447**	3.261**	3.074**	1.023		
	Error	214	0.414	0.415	0.444	0.596	0.189		
	Total	429							
Pooled	Replication	1	0.458	0.001	0.004	0.195	0.038		
	Year	1	425.96**	44.42**	40.89**	113.35**	75.51**		
	Variety	214	3.93**	1.53**	4.29**	4.76**	0.92		
	Year x Variety	214	3.86**	1.24*	2.75**	4.58**	0.73		
	Error B	430	0.34	0.39	0.37	0.47	0.25		
	Total	859	2.6	0.91	1.98	2.69	0.62		

Table 1. Year-wise Analysis of Variance (ANOVA) for fatty acid contents in rice landraces

*5% level of significance; **1% level of significance

(±1.29%). Stearic acid ranged from 1.01% (Memri Khedi) to 2.90% (Baiga Seeng) with an average 1.80% (±0.29%). A total of 25 genotypes had more than 46% oleic acid content and ranged from 37.60% (Antarved) to 49.17% (Kadamphool) with an average of 43.90% (±1.79%). Linoleic acid content ranged from 31.55% (Ratanchudi) to 44.67% (Antarved) with an average of 35.56% (±1.83%) and a total of 16 genotypes had more than 38% linoleic acid content. While linolenic acid content ranged from 0.75% (Khajoor) to 2.0% (Dadbanko) with average 1.19% (±0.18%) (Table 2).

Coefficient of variation was highest for stearic acid (16.11%) and lowest for oleic acid (4.58%). According to box plot profiling, 75% landraces have equal to or less than 18.44% palmitic acid, 1.98% stearic acid, 44.87% oleic acid, 36.53% linoleic acid, 1.28% linolenic acid content while 25% landraces have equal to or less than 16.75% palmitic acid, 1.62% stearic acid, 42.94% oleic acid, 36.53% linoleic acid, 1.28% linolenic acid content (Fig. 1). Frequency distribution on the histogram for various fatty acids showed that none of the traits followed the exact normal distribution pattern (Fig. 2). Skewness values and histogram showed that palmitic acid and oleic acid were negatively skewed whereas stearic acid, linoleic acid, and linolenic acid contents were positively skewed. All the fatty acid components were showed positive kurtosis (Table 2) and followed the leptokurtic



Fig. 1. Box plots showing the statistical parameters of fatty acid components based on pooled data



Fig. 2. Histogram showing the frequency distribution of fatty acid components based on pooled data

Fatty acid contents	Palmitic acid	Stearic acid	Oleic acid	Linoleic acid	Linolenic acid
Minimum	12.59 % (Khajoor)	1.01% (Memri Khedi)	37.60% (Antarved)	31.55% Ratanchudi	0.75% (Khajoor)
Maximum	20.25 % (Lajini Super)	2.9 % (Baiga Seeng)	49.17% (Kadamphool)	44.67% (Antarved)	2% (Dadbanko)
Mean	17.54	1.8	43.9	35.56	1.19
Standard deviation	1.29	0.29	1.79	1.83	0.18
Coefficient of variation	7.35	16.11	4.08	5.15	15.13
Skewness	-0.56	0.37	-0.21	1.43	0.83
Kurtosis	0.7	0.95	1.11	4.78	2.47
Phenotypic coefficient of variation	5.90	12.81	3.67	4.41	12.33
Genotypic coefficient of variation	5.41	9.84	3.375	4.00	9.34
Heritability(bs)	84.18	59.08	84.26	82.06	57.35
Genetic advance	2.53	1.19	2.65	2.73	0.90
Genetic advance as % of mean	10.24	15.59	6.38	7.47	14.57

Table 2. Genetic parameters of fatty acid components in rice landraces over the years

configurations. None of the traits showed mesokurtic and platykurtic configuration.

Genotypic coefficient of variations (GCV) was slightly lower than the corresponding phenotypic coefficient of variation (PCV) for oleic acid, palmitic acid, and linoleic acid content. Linolenic acid and stearic acid showed intermediate PCV with low GCV whereas Linoleic acid, oleic acid, and palmitic acid contents showed low GCV and PCV (Table 2). High heritability was expressed by oleic acid, palmitic acid, and linoleic acid content. The highest heritability exhibited for oleic acid content whereas moderate genetic advance as percent of the mean was observed for stearic acid, palmitic acid, and linolenic acid content.

Genetic correlations and diversity analysis for fatty acid components

The correlation coefficients in all possible combination of five fatty acids showed that Palmitic acid was positively correlated with linolenic acid (r = 0.21; P<0.01), while it is negatively correlated with oleic acid content (r = -0.39; P<0.01) and linoleic acid content (r = -0.29; P<0.05). Stearic acid is significant negatively correlated with oleic acid (r = -0.12; P<0.05) and linolenic acid content (r = -0.25; P<0.05). Oleic acid is significantly negatively correlated with linoleic acid (r = -0.75; P<0.01). Linoleic acid content is significant negatively correlated with linolenic acid content (r = -0.17; P<0.05) (Fig. 3).



Fig. 3. Genotypic correlation plots among the fatty acid components (pooled)

The principal component analysis (PCA) revealed two major variables, which accounted for 98% of total variation from all the fatty acids in the tested landraces. The principal component (PC1) for the first set contributed about 67.14% with eigenvalue 5.57 and PC2 for the second set contributed about 31.08% with eigenvalue 2.58. Rest of the PCs had less than 1 eigenvalue and less than 5% variability (Supplementary Table S2). The analysis detected that linoleic acid and oleic acid were the main contributory traits for PC1 whereas palmitic acid and oleic acid are the main contributing traits for PC2. Thus, it was postulated that about 98% of the total variation is represented by oleic acid, palmitic acid, and linoleic acid. Further, scattered diagram based on PCA has grouped the landraces into five groups across the PC1 and PC2 (Supplementary Fig. S1). The result of PCA was further confirmed by agglomerative hierarchical cluster analysis that divided the rice landraces into five groups in Supplementary Table S1 and Fig 4. This



Fig. 4. Scatter diagram of 215 rice landraces across the PC1 and PC2

demonstrated that the data obtained from this experiment were accurate, precise, and reliable. In AHC, cluster-V comprised of 82 genotypes (38.13%) which were the highest among all the clusters followed by Cluster-III which had 62 (28.83%) genotypes, out of 215 rice landraces. Cluster I, II and IV had 5, 38 and 28 rice genotypes, respectively (Supplementary Table S3). The Euclidean distance-based dendrogram showed that the genotypes Bansgathi, Gangachur, Nariyaljhoba, Anjaniya, and Antarved are highly diverse from the rest of the genotypes (Supplementary Fig. S1).

Discussion

Oleic acid (C18:1), linoleic acid(C18:2) and palmitic acid(C16:0) were identified as the major FAs and accounted for 77%-88% contribution of total FA components in the current study which are in accordance with the previous findings (Lugay and Juliano 1964; Yoshida et al. 2011; Yu et al. (2016). Higher oleic/linoleic (O/L) ratio determines the stability of bran oil and extends its shelf life (Holley and Hammons 1968) as well as high O/L ratio reduces the chances of cardiovascular diseases in the human

body (O'Byrne et al. 1997). The current study reported the highest oleic acid/linoleic acid ratio in landraces Kadamphool, Ratanchudi, Bathrash, Panwar, and Karigilash for the first time, which can either used as a donor to develop rice varieties for high-quality bran oil or released directly after selection. Results of analysis of variance illustrated the sufficient variation among the rice landraces for fatty acid components except for linolenic acid, indicating the huge possibility of their improvement and results are following the earlier findings in rice (Goffman 2003; Konate et al. 2016). Significant effect of year component and GxE interaction indicated that the environmental conditions and extraneous factors were not similar in two years and similar results were reported in rice (Goffman 2003), groundnut (Onemli 2012) and safflower (Gecgel et al. 2007).

Coefficients of variation studies indicated the negligible influence of extraneous factors and therefore, selection for such traits based on phenotype only could be gratifying. Nevertheless, the intermediate and low value of PCV and GCV indicated the presence narrow genetic base of FAs in rice landraces. Therefore, induced mutagenesis and hybridization could be the best method to broaden the genetic base for FAs in rice. High heritability with moderate GA for palmitic acid content has suggested the significant contribution of non-additive and additive both gene action in their inheritance. High heritability with low GA for oleic acid and linoleic acid content revealed the significant contribution of non-additive gene action. Hence, heterosis breeding may be worthwhile for the enhancement of these traits. The significant negative association of oleic acid with linoleic acid, palmitic acid and stearic acid indicated that the conversion of each fatty acid to another is dependent through various biochemical pathways. The higher negative correlation between oleic acid and linoleic acid exist due to a possible natural mutation in oleyl phosphatidylcholine desaturase gene which converts oleic acid to linoleic acid (Mondal et al. 2011) and similar results were also reported in earlier studies on rice (Ying et al. 2012) and groundnut (Onemli 2012).

PCA detected two main components of fatty acid variability in rice landraces where in palmitic, oleic and linoleic acid played a major role in total genetic variation. This result was in agreement with earlier reports (Yoshida et al. 2011; Yu et al. 2016) however, the materials used by them had different geographic origin. PCA elucidated the high level of genetic diversity in rice landraces, which were further confirmed by agglomerative hierarchical cluster analysis. A similar pattern of PCA and AHC was also reported by Lasalita-Zapico et al. (2010) and Khatun et al. (2015). The landrace viz., Antarved, Gangachur, Nariyal Jhoba of cluster-I posses less oleic acid components as compare to the landraces Kadamphool, Panwar, Rudra, Kalajeera, which belong to cluster IV which have higher oleic acid contents. Landraces of these two clusters were genetically diverse and thus could be useful in crossbreeding towards the development of high yielding genotypes with higher oleic acid content. It can, therefore, be concluded that the identified landraces viz., Kadamphool, Ratanchudi, Bathrash, Panwar and Karigilash with good monounsaturated fatty acids and high O/L ratio as well as landraces of the diverse group have the enormous value in breeding program. Further, the inverse relationship between oleic and linoleic acid suggested that future targeted mutagenesis in olevil phosphatidylcholine desaturase or OsFAD2 which may be useful in generating higher oleic acid-containing genotypes in rice. The information, thus obtained, would be highly supportive in planning an effective rice-breeding program for developing rice varieties with a high content of desirable fatty acids for human welfare.

Authors' contribution

Conceptualization of research (SM, DS); Designing of the experiments (SM, VK, BKD); Contribution of experimental materials (DS, SM, VK, BKD); Execution of field/lab experiments and data collection (PKS, RS); Analysis of data and interpretation (PKS, SM, RS); Preparation of the manuscript (PKS, SM, RS).

Declaration

The authors declare no conflict of interest.

Acknowledgment

The research paper is the part of a Ph.D. program supported by INSPIRE fellowship awarded to senior author by the Department of Science and Technology, GOI, New Delhi. Sincere thanks to Associate Director (A), Bioscience Group and Head, NA&BTD, Bhabha Atomic Research Centre, Mumbai for their support and critical comments.

References

 FAO. 2017. Rice market monitor. 20(1): 1-38. Data accessed on 15 May 2017. Available at http:// www.fao.org/fileadmin/templates/est/COMM_ MARKETS_MONITORING/Rice/Images/RMM/ RMM_APR17_H.pdf.

- Fresco L. 2005. Rice is life. J. Food Comp. Anal., **18**(4): 249-253.
- Gecgel U., Demirci M., Esendal E. and Tasan M. 2007. Fatty acid composition of the oil from developing seeds of different varieties of safflower (*Carthamus tinctorius* L.). J. American Oil Chemists' Soc., **84**: 47-54.
- Goffman F. D., Pinson S. and Bergman C. 2003. Genetic diversity for lipid content and fatty acid proûle in rice bran. J. American Oil Chemists' Soc., 80(5): 485-490.
- Hammer O., Harper D. A. T. and Ryan P. D. 2001. PAST: Paleontological Statistics Software Package for Education and Data Analysis. Palaeontologia Elect., 4(1): 9.
- Holley K.T. and Hammons R.O. 1968. Strain and seasonal effects on peanut characteristics. Univ. Ga. Coll. Agric. Exp. Sta. Res. Bull., pp 32.
- IRRI. 2002. Standard evaluation system for rice. International Rice Research Institute, Manila. pp 50.
- Khatun M. T., Hanafi M. M., Yusop M. R., Wong M. Y., Salleh F. M. and Ferdous J. 2015. Genetic variation, heritability, and diversity analysis of upland rice (*Oryza sativa* L.) genotype based on quantitative traits. BioMed Res. Int. pp 1-7.
- Konate A. K., Zongo A., Kam H., Sanni A. and Audebert A. 2016. Genetic variability and correlation analysis of rice (*Oryza sativa* L.) inbred lines based on agromorphological traits. African J. Agril. Res., **11**(35): 3340-3346.
- Lasalita-Zapico F. C., Namocatcat J. A. and Cari-no-Turner J. L. 2010. Genetic diversity analysis of traditional upland rice cultivars in Khan, Malapatan, Sarangani Province, the Philippines using morphometric markers. Philippine J. Sci., **139**(2): 177-180.
- Lugay J. C. and Juliano B. O. 1964. Fatty acid composition of rice lipids by gas-liquid chromatography. J. American Oil Chemists' Soc., **41**(12): 273-275.
- Min B., McClung A. M. and Chen M. H. 2011. Phytochemicals and antioxidant capacities in rice brans of a different color. J. Food Sci., **76**: C117-126.
- Mondal S., Badigannavar A. M. and Dsouza S. F. 2011. Induced variability for fatty acid profile and molecular characterization of high oleate mutant in cultivated groundnut (*Arachis hypogaea* L.). Plant Breed., **130**(2): 242-247.
- Mondal S., Nazareth J., Bhad P. G. and Badigannavar A. M. 2018. Isolation of high oleate recombinants in peanut by Near Infra-Red Spectroscopy and confirmation with allele-specific polymerase chain reaction marker. J. American Oil Chemists' Soc., **95**: 113-121.

- O'Byrne D. J., Knauft D. A. and Shireman R. B. 1997. Low fat-monounsaturated rich diets containing high-oleic peanuts improve serum lipoprotein profiles. Lipids, **32**: 687-695.
- Onemli F. 2012. Impact of climate change on oil fatty acid composition of peanut (*Arachis hypogaea* L.) in three market classes. Chilean J. Agril. Res., **72**(4): 483-488.
- Rondanelli M., Perna S., Monteferrario F. and Opizzi A. 2011. Update on the therapeutic qualities of the rice bran in the treatment of dyslipidemia and chemo-prevention. Recent Prog. Med., **102**: 310-313.
- Sokal R. R. and Rohlf F. J. 1995. Biometry. Freeman, New York, pp. 887.
- Wilson T. A., Idreis H. M., Taylor C. M. and Nicolosi R. J. 2002. Whole fat rice bran reduces the development

of early aortic atherosclerosis in hypercholesterolemic hamsters compared with wheat bran. Nutr. Res., **22**: 1319-1332.

- XLSTAT. 2017. Data Analysis and Statistical Solution for Microsoft Excel. Addinsoft, NY, USA.
- Ying J. Z., Shan J. X., Gao J. P., Zhu M. Z., Shi M. and Lin H. X. 2012. Identiûcation of quantitative trait loci for lipid metabolism in rice seeds. Mol. Plant, 5(4): 865-875.
- Yoshida H., Tanigawa T., Kuriyama I., Yoshida N., Tomiyama Y. and Mizushima Y. 2011. Variation in the fatty acid distribution of different acyl lipids in rice (*Oryza sativa* L.) Brans. Nutrients, **3**: 505-514.
- Yu L., Li G., Li M., Xu F., Beta T. and Bao J. 2016. Genotypic variation in phenolic acids, vitamin E and fatty acids in whole grain rice. Food Chem., **197**: 776-782.



Supplementary Fig. S1. Cluster diagram (dendrogram) for 215 rice landraces based on fatty acid components based on pooled data

S. No.	Name	S. No.	Name	S. No.	Name	S. No.	Name
Gen1	Luchai	Gen55	Bhunduluchai	Gen109	OdhaDhanBanarsi	Gen163	Kalinga
Gen2	Sonagathi	Gen56	Barhasal-3	Gen110	B.D. Safri	Gen164	Bhusu
Gen3	Kherkakuchi	Gen57	Khetganga	Gen111	Lochai	Gen165	Kabeli
Gen4	Dadbanko	Gen58	Bashabhog	Gen112	GadurSela	Gen166	Gatuvan
Gen5	Danighoda	Gen59	Tulsibhog	Gen113	KareniDhan	Gen167	Baikoni
Gen6	Anjani	Gen60	Nariyalphool	Gen114	PivriLochai	Gen168	ChinniParas
Gen7	Bathrash	Gen61	Badshabhog-2	Gen115	Govardhan Kali Kamod 2	Gen169	Jalsinga
Gen8	Saraitoliha	Gen62	Lajini Super	Gen116	Loindi	Gen170	Agni Fag
Gen9	Jonyaphool	Gen63	Gangabaru	Gen117	Santio	Gen171	Lalapana
Gen10	Pratiksha	Gen64	GudkamalDhan	Gen118	Parra Dhan	Gen172	BahalBinjo
Gen11	Bhadvel	Gen65	TulsiManjari	Gen119	Godadani	Gen173	Kari Grass
Gen12	Ranikajal	Gen66	Kanakbhog	Gen120	Ramshri	Gen174	Asam Chudi-2
Gen13	Bhajna	Gen67	DRRH-3	Gen121	Danwar	Gen175	Tulsi Mala
Gen14	Ratajhinga	Gen68	Mahamaya	Gen122	ChatiyaNakhi	Gen176	Surmatiya
Gen15	Pataniyajhuli	Gen69	Rajeshwari	Gen123	BhathaMasri	Gen177	Manmohan
Gen16	Laxmibhog	Gen70	HR 14-1 Heera	Gen124	Kanchan	Gen178	Jalgundi
Gen17	Sawani	Gen71	MatkoDhan	Gen125	BaigaSeeng	Gen179	HathiPinjara
Gen18	Jhilli	Gen72	NimaliyaBanki	Gen126	Mota Safri-2	Gen180	ChurlaiBanko
Gen19	Tulsimongra	Gen73	Jonyaphool	Gen127	MohlaiBanko	Gen181	Safri-17
Gen20	DhauraMundariya	Gen74	Kadamphool	Gen128	SutaiDhan	Gen182	Rani Kajar
Gen21	Jhimipras	Gen75	Indjopa	Gen129	Haripanti	Gen183	ParwatKal
Gen22	PangudiGoindi	Gen76	Ramigauri	Gen130	Bhujnin	Gen184	Jhoomar
Gen23	Safri	Gen77	Brown Rice-1	Gen131	Manki	Gen185	Asam Chudi-3
Gen24	Barhasal	Gen78	Arokhutu	Gen132	Sadachar	Gen186	Jalkeshar
Gen25	Dubraj	Gen79	Brown Rice-2	Gen133	Mahabaikoni	Gen187	Sudama
Gen26	Agyasal	Gen80	Dubraj	Gen134	Khajoor	Gen188	Hajan
Gen27	Jauphool	Gen81	HathiPanjra	Gen135	Kumhdayin	Gen189	Ikkopatla
Gen28	Kalajeera	Gen82	Modipeera	Gen136	BhejrimaiDhan	Gen190	HarunaMasri
Gen29	Byalo	Gen83	Petgadi	Gen137	RatanChudi	Gen191	Kosawari
Gen30	Badshabhog	Gen84	Ramlaxman	Gen138	Panwar	Gen192	Bansgathi
Gen31	Chhindmauri	Gen85	Raja Banga	Gen139	Chhindmauri	Gen193	BhataNakhi
Gen32	Karhani	Gen86	Kari Gilash	Gen140	Rani Parewa	Gen194	Masri
Gen33	Sihar	Gen87	Muni Bhog	Gen141	Ramlichonch	Gen195	Ikkomota
Gen34	JhimiprasSamlayi	Gen88	SuaPankhi	Gen142	Mejhri	Gen196	Gaurimala
Gen35	Nandel	Gen89	Mala Gauri	Gen143	Bodibaja	Gen197	Ramjhilli
Gen36	Kapri	Gen90	DokraDokri	Gen144	Jela	Gen198	Sichar
Gen37	Bhusi	Gen91	NariyalJhoba	Gen145	Badshabhog Selection-1	Gen199	Rajabangla
Gen38	DhauraMundariya	Gen92	Sindursal	Gen146	Kari Alcha	Gen200	Majori
Gen39	Gangachur	Gen93	Chhindmauri	Gen147	DeshiSurmatiya	Gen201	Hardigathi
Gen40	Karhani	Gen94	Sugandha	Gen148	Anjaniya	Gen202	Ramlaxman
Gen41	Byalo	Gen95	Dandrice	Gen149	BaktiChudi	Gen203	Korma

Supplementary Table S1. List of rice genotypes used for the investigation

(<i>iii</i>)	Parmeshwar K. Sahu et al.					[Vol. 79, No. 4	
Gen42	Bhusu	Gen96	BansveeraDhan	Gen150	JhilliSafri	Gen204	Dowana
Gen43	Safri-2	Gen97	Jhingapuchhi	Gen151	Nanded	Gen205	Luchai- 2
Gen44	Sanchorma	Gen98	BeedelaDhan	Gen152	TuriyaKhudig	Gen206	Maidubraj
Gen45	SatraSafri	Gen99	Sonagathi-2	Gen153	Antarved	Gen207	Bhaisapuchhi
Gen46	Barhani	Gen100	PhalodDhan	Gen154	Rang Chudi	Gen208	Pancho
Gen47	Kanakbans	Gen101	LalmaDhan	Gen155	MotaChudi	Gen209	Bhata Masri-2
Gen48	Jhimipras-2	Gen102	BaiganiDhan	Gen156	B.D. Safri-2	Gen210	Gomti
Gen49	Dhaniyaphool	Gen103	MaranDhan	Gen157	KharikhaKuchi	Gen211	Sonkharcha
Gen50	Lalbarhasal	Gen104	LoktiMusi	Gen158	MemriKhedi	Gen212	Katrani-4
Gen51	Alsenga	Gen105	AsamChudi	Gen159	Ankapalli	Gen213	Katrani-7
Gen52	Barhasal-2	Gen106	Jana Dhan	Gen160	Samarlengda	Gen214	Katrani-9
Gen53	RuchiDhan	Gen107	RelaDhan	Gen161	Mayath	Gen215	KondhaKoya
Gen54	Rudra	Gen108	Jhunuprash	Gen162	MotaSafri		

Note: All the collected landraces/ varieties are conserved in IGKV, Raipur (C.G.), India

Supplementary Table S2. Eigen value, variability % and eigen vectors based on principal component analysis

Particulars	PC1	PC2	PC3	PC4	PC5		
Eigenvalue	5.577	2.582	0.101	0.042	0.003		
Variability (%)	67.148	31.087	1.216	0.509	0.040		
Cumulative variability (%)	67.148	98.235	99.451	99.960	100.000		
Eigen vectors							
Palmitic acid	-0.041	-0.798	-0.315	-0.306	0.412		
Stearic acid	-0.001	-0.027	0.870	-0.271	0.412		
Oleic acid	-0.685	0.460	-0.268	-0.270	0.418		
Linoleic acid	0.727	0.389	-0.269	-0.270	0.418		
Linolenic acid	-0.001	-0.027	-0.007	0.829	0.558		

Supplementary Table S3. Grouping of rice landraces in different clusters

Cluster	Total genotypes	Name of the rice genotypes
1	5	Bansgathi, Gangachur, Nariyal Jhopa, Anjaniya, Antarved
2	38	Modipeera, Badshabhog, Chhindmauri, Dokra Dokri, Jhoomar, Sindursal, Lajini Supar, Nimlayi Banko, Danwar, Sonkharcha, Saraitoliha, Barhasal-3, Bhata Mahsuri-3, Danighoda, Turiya Khudig, Dadbanko, Pancho, Kanakbans, Brownrice-2, Jhimipras Samlayi, Sanchorma, Ramigauri, Byalo, Gudkamal Dhan, Alsenga, Karhani, Bhujnin, Sihar, Dhaura Mundariya, Nariyalphool, Bhatamasri, Bhusu, Munibhog, Masri, Ruchi Dhan, Parwatkal, Bhusi, Manki
3	62	Motachudi, Majori, Haruna Masri, Bhaisa Puchhi, Badshahbhog, Jalsinga, Byalo, Safri-2, Ankapalli, Rajeshwari, Anjani, Loktimusi, Nandel, Brown irce-1, Bhusu-1, Gaurimala, Loindi, Kabeli, Mahamaya, Sua Pankhi, Kalinga, Mota Safri, Odha Dhan Banarsi, Ratajhinga, Chhindmauri, Memrikhedi, Satra Safri, Matkodhan, Chatiyanakhi, Sudama, Junupras, Ramlaxman, Hathi Panjra, B.D. Safri-2, Mohlayi Banko, Beedela Dhan, Sonagathi-2, Khairka Kuchhi, Maidubraj, Ghodadani, Ramshri, Marandhan, Phalod Dhan, Kanchan, Churlayi Banko, Raja Banga, Haripanti, Kosawari, Kumhadayin, Mala Gauri, Parra Dhan, Sanwali, Pivri Luchai, DRRH-3, Reladhan, Kareni Dhan, Sadachar, Bhajan, Baigani Dhan, Sutai Dhan, Hathi Panjara
4	28	Gathuvan, Asamchudi-3, Gadursela, Katarni-9, Gangabaru, Tulsimala, Korma, Kapri, Tulsibhog, Karigrass, Rudra, Jalkeshar, Govardhan Kali Kamod, Kondha Koya, Dubraj, Rajabangla, Lalma Dhan, Khajoor, Kadamphool, Panwar, Tulsimongra, Jauphool, Bansveera, Badshahbhog sel-1, Badshahbhog-2, Tulsimanjari, Laxmibhog, Kalajeera
5	82	Chhindmauri, Jhimipras-2, Khetganga, Ramlaxman, Surmatiya, Bhunduluchai, Lalbarhasal, Arokhutu, Kanakbhog, Mota Safri-2, Indjhopa, Bhaktichudi, Barhani, HR-14-1Heera, Hardigathi, Barhasal, Ramkajal, Agyasal, Dhaniyaphool, Agnifag, Ramjhilli, Luchai-2, Barhasal-2, Baiga seeng, Nanded, Jhingapuchhi, Janadhan, Mejhari, Jhimiprash, Santio, Chiniparas, Karhani, Bhejrimai Dhan, Petgadi, Gomti, Rangchudi, Bhadvel, Ranikajar, Samarlengda, Bahal Binjo, Jonyaphool, Baikoni, Manmohan, Katarni-7, Ikkopotla, Lalapana, Bhatanakhi, Katarni-4, Pataniyajhuli, Safri, Sichar, Jhela, Pratiksha, Jonyaphool, Asamchudi, Deshi Surmatiya, Dowana, Mayath, Mahabaikoni, Rani Parewa, Bodibaja, Karialcha, Lochai, Safri-17, Jalgundi, Dubraj, Pangudi Goindi, Dandrash, Bathrash, Ratanchudi, Sonagathi, Karigilash, Sugandha, Luchai, Ikkomota, Jhilli, B.D. Safri, Asam Chudi-2, Kherkakuchi, Dhaura Mundariya, Bhajna, Jhilli Safri
Тс	otal 215	