Electronic Journal of Plant Breeding



Research Note

Variability assessment and screening of superior rice (*Oryza sativa* L.) genotypes for grain micronutrients and yield components

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Abstract

The present study was carried out to assess the genetic parameters and divergence in 38 rice genotypes for important yield and grain quality traits. Phenotypic coefficient of variation (PCV) exceeded genotypic coefficient of variation (GCV) for all the traits taken for the study, while high heritability was recorded for all the traits, except kernel length to breadth ratio and grain iron content. The trait number of grains per panicle exhibited high heritability (91.00%) and high genetic advance as a percentage of mean suggesting simple selection may be followed to improve the traits. Cluster analysis grouped the genotypes into seven clusters. The highest inter-cluster distance was observed between clusters VII and IV (537.17). Five genotypes excelled in grain yield, ten genotypes for grain zinc content and one genotype for grain iron content over the check variety Rajendra Bhagwati. Despite high variability in grain zinc and iron content. The experimental material's variability for grain nutrient status suggests their potential use in biofortification programs.

Keywords: Rice, biofortification, micronutrients, variability, genetic diversity, heritability

Rice (*Oryza sativa* L.) stands as the principal staple crop globally with Asia holding a preeminent position in production, encompassing 90% of the worldwide rice acreage (Schneider and Asch, 2020). India achieved the grain production of 112.43 MT with productivity of 2.7 t/ ha during the year 2019-20. Despite India's notable rice production, its productivity lags behind that of major rice producing countries like China (6.5 t/ha) and Indonesia (5.2 t/ha) (MoA and FW, Gol, 2019-20). Though significant

advancements have been made in rice production, there is a pressing need to enhance productivity further to address the escalating demands for food grains to feed the expanding global population. Concurrently, there is a burgeoning apprehension about ensuring nutritional security with respect to essential micronutrients, vitamins and proteins to prevent malnutrition. This concern arises due to the pervasive issue of micronutrient deficiency afflicting developing nations, exemplified by the situation

https://doi.org/10.37992/2024.1503.088

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in India (Bain et al., 2013). A diet lacking essential minerals particularly iron (Fe) and zinc (Zn) contributes to hidden hunger or micronutrient malnutrition in developing nations (Welch and Graham, 2004). According to the World Health Organization (WHO) report 2022, the prevalence of anaemia among women aged 15 to 49 remained similar in 2019 as compared to 2000. However, due to population growth, the total number of affected women increased from 492.9 million in 2000 to 570.8 million in 2019. Developing rice varieties with high yield and sufficient micronutrient content could address the challenges of both food and nutritional security challenges. In light of this, the present study aimed to investigate variability, heritability, and genetic advances in yield and yield related traits as well as grain Zn and Fe content. Additionally, the research sought to identify genotypes with high grain yield and micronutrient (Zn & Fe) content among 38 rice genotypes, exploring their potential for utilization in future rice crop improvement program for enhancing both yield and micronutrient content.

Plant material and growth conditions: The current experiments was conducted at the Research Farm of RPCAU, Pusa, Bihar during *Kharif* 2021 season. The experimental material comprising 38 rice genotypes including the check Rajendra Bhagwati obtained from the Harvest Plus program, ICRISAT, Hyderabad were planted in a Randomized Complete Block Design with two replications, adhering to standard cultural practices for puddled transplanted rice. Each entry was evaluated in experimental plots measuring 5 m².

Recording of observation: Data for 13 quantitative traits *viz.*, plant height, number of panicles per plants, panicle length, flag leaf length, number of grains per panicle, grain yield per plant, days to 50% flowering, test weight, kernel

length, kernel breadth, kernel length to breadth ratio were recorded by selecting five random plants from each plot following Standard Evaluation System, IRRI, while data on grain Iron and Zinc content was recorded through Energy Dispersive X-Ray Fluorescence (ED-XRF) on bulk sample in each replication (Chandu *et al.*,2020).

Statistical Analysis: The mean data for each trait were subjected to analysis of variance following the method outlined by Panse and Sukhatme (1978). Genotypic (GCV) and Phenotypic (PCV) coefficients of variation were calculated as per Burton (1952). Broad sense heritability was determined according to Lush (1940) and genetic advance as a percentage of mean was computed following the procedure proposed by Johnson *et al.* (1955). Genetic Divergence among 30 genotypes was assessed using D² analysis proposed by Mahalanobis (1936). The clustering of genotypes was performed utilizing the Euclidean method with Windostat version 9.2 from Indostat services software.

The analysis of variance (ANOVA) presented in Table 1 indicated significant mean squares of treatment for all traits, implying noteworthy differences among suggests the suitability of the genotypes and it experimental material for further genetic investigations. The ANOVA results gain significance due to the diverse nature of the experimental material comprising rice genotypes obtained from various rice research centres across the country characterized by distinct lineages and adaptation to diverse agro-ecological conditions. This finding aligns with earlier reports in rice, where disparate genotypes exhibited significant variations (Das and Borthakur, 1974; Dhanwani et al., 2013; Nachimuthu et al., 2014; Abebe et al., 2017; Parihar et al., 2017). The observed variations underscore the genetic diversity

Table 1. Analysis of variance (ANOVA) for 13 quantitative traits in rice evaluated during Kharif- 2021
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Traits	Mean Squares						
	Replication (df = 1)	Treatment (df = 37)	Error (df = 37)				
Plant height (cm)	29.239	496.194**	47.258				
Days to 50% flowering	21.053	234.721**	20.215				
Number of panicles/ plant	1.914	6.630**	1.297				
Panicle length (cm)	0.144	10.302**	1.657				
Flag leaf length (cm)	3.513	52.156**	6.466				
Number of grains/panicle	862.249	6139.212**	279.727				
Test weight (g.)	3.522	25.465**	1.286				
Kernel length (mm)	0.065	0.520**	0.116				
Kernel breadth (mm)	0.038	0.065**	0.013				
Kernel length to breadth ratio	0.029	0.225**	0.064				
Grain Iron content (ppm)	0.064	3.455**	0.910				
Grain Zinc content (ppm)	2.434	28.064**	1.964				
Grain yield per plant (g.)	0.441	23.580**	2.398				

**- Significant at α=0.01

inherent in the experimental material, affirming its potential for comprehensive genetic exploration.

The analysis of coefficients of variation presented in Table 2 elucidates that the calculated values for genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) exhibit minimal disparities among all the investigated traits, suggesting a limited influence of environmental factors on phenotype. Specifically, GCV and PCV values for kernel length (6.78 and 8.5%), kernel breadth (7.53 and 9.3%) and panicle length (8.1 and 9.53%) were below 10%, aligning with earlier findings of Singh et al. (2020). Traits such as kernel length to breadth ratio (9.03 and 12.11%), days to 50% flowering (11.59 and 12.63%), plant height (13.39 and 14.74%), flag leaf length (14.57 and 16.5%), grain iron content (12.84 and 16.82%), test weight (16.03 and 16.86%), grain zinc content (15.89 and 17.05%), grain yield per plant (17.58 and 19.47%) and number of panicles per plant (16.67 and 20.32%) exhibited moderate GCV and PCV values ranging from 10 to 20%. Notably, similar observations regarding panicle length, test weight and days to 50% flowering have been documented by Khan et al. (2009), Raza et al. (2019), Kashinath et al. (2024). Conversely, number of grains per panicle displayed high GCV and PCV values (>20%), akin with the findings of Bekele et al. (2013). The minimal disparity between PCV and GCV across all traits in the present investigation suggests that the phenotypic variation observed was predominantly due to genetic factors rather than environmental influences.

The outcomes derived from the assessments of heritability and genetic advance as a percentage of the mean (GAM) presented in **Table 2** reveal that traits such as plant height (83.00 and 25.08%), days to 50% flowering (84.00 and 21.90%), number of panicles per plant (67.00 and 28.17%), flag leaf length (78.00 and 26.49%), number of grains per panicle (91.00 and 46.93%), test weight (90.00 and 31.40%), grain zinc content (87.00 and 30.53%), grain iron content (58.00 and 20.20%), and grain yield per plant (82.00 and 32.70%) exhibit high heritability (>60%) and substantial GAM. These findings suggest that these traits hold potential for direct selection to influence their modification in the desired direction, ultimately leading to an enhancement in grain yield (Agrawal, 2003; Girma et al., 2018 and Vallala et al., 2024). Conversely, other traits, including panicle length (72.00 and 14.19%), kernel length to breadth ratio (56 and 13.87%), kernel breadth (66.00 and 12.57%) and kernel length (64.00 and 11.15%), demonstrate high heritability but low GAM. It may be attributed to the influence of environmental factors on trait expression. Consequently, simple selection strategies may not yield significant improvements in these traits (Mall et al., 2005; Khan et al., 2009; Brar et al., 2011; Kumar et al., 2020; Vanlalrinngama et al., 2023).

When the mean data of trait for all the genotypes were subjected to D² analysis, it became apparent that the experimental materials exhibited significant diversity. Utilizing Euclidean methods, genotypes were organized into seven clusters as detailed in Table 3. The distribution of genotypes across clusters varied, with Cluster VII containing the highest number of genotypes (13) followed by Clusters I and VI (6 genotypes each), Cluster III (5 genotypes) and Cluster IV with the lowest number of genotypes (2). Cluster means for all studied traits are furnished in Table 4. Cluster V demonstrated superior mean performance for the traits, number of panicles per plant, panicle length, test weight, kernel length and grain yield per plant. The Cluster VII exhibited optimal mean performance for traits such as kernel length to breadth ratio, grain Fe and Zn content, suggesting potential suitability of genotypes from this cluster as parent for yield and grain Fe and Zn content improvement in rice. Table 5 provides insights into the intra cluster and inter cluster distances among all seven clusters derived from

Table 2. Genetic parameters for 13 quantitative traits in rice evaluated during Kharif- 2021

Characters	Mean	Ran	ge	Coefficient of	variation (%)	Heritability	GA as % of
		Min	Мах	Genotypic	Phenotypic	(%)	Mean
Plant height (cm)	111.87	84.5	146.57	13.39	14.74	83.00	25.08
Days to 50% flowering	89.37	72.00	120.00	11.59	12.63	84.00	21.90
Number of panicles per plant	9.80	6.67	12.92	16.67	20.32	67.00	28.17
Panicle length (cm)	25.67	21.17	29.17	8.10	9.53	72.00	14.19
Flag leaf length (cm)	32.82	22.50	41.59	14.57	16.50	78.00	26.49
Number of grains per panicle	227.00	134.50	370.00	23.85	24.96	91.00	46.93
Test weight (g.)	21.69	12.33	28.95	16.03	16.86	90.00	31.40
Kernel length (mm)	6.63	5.55	7.65	6.78	8.50	64.00	11.15
Kernel breadth (mm)	2.13	1.75	2.65	7.53	9.30	66.00	12.57
Kernel length to breadth ratio	3.14	2.56	3.96	9.03	12.11	56.00	13.87
Grain Iron content (ppm)	8.78	6.25	11.80	12.84	16.82	58.00	20.20
Grain Zinc content (ppm)	22.73	15.15	32.75	15.89	17.05	87.00	30.53
Grain yield per plant (g.)	18.52	11.68	26.97	17.58	19.47	82.00	32.70

Table 3. Clustering pattern of 38 genotypes based on Euclidean distance of 13 quantitative tra	aits in rice
evaluated during Kharif- 2021	

Cluster No.	No. of genotypes within cluster	Genotypes in cluster
I	6	R-RGY-IS-110, NVSR 522, DRR Dhan 45, CR 2819-1-5-3-2B-12-1, R-RGY-RH-16, JDP 2520-2-4-1
П	3	Samba Mahsuri, R-RH2-M1-93, R-RHZ-IR-140
Ш	5	R-RHZ-MB-119, R-RHP-IK-120, IR 64, BPT 3144, R-RHZ-IH-82
IV	3	MTU 1356, MTU 1357, BPT 3137
V	2	R-RHZ-SK-128, CR 2818-1-11-1-B-1-1-2-B-1
VI	6	CR Dhan 310, CGZR-2, CSR HZR 17-41, Rajendra Bhagwati, R-RGY-MH-113, DRR Dhan 48
VII	13	R-RHZ-IR-131, MI 156, R-RHZ-IR-142, CR Dhan 311, DRR Dhan 49, R-RHZ-IB-80, Zinco rice, R-RHZ-IR-132, CGZR-1, CSR HZR 17-8, CSR HZR 17-42, MI 127, R-RHZ-SM-14

Table 4. Cluster Mean of 13 quantitative traits in rice evaluated during Kharif- 2021

Traits Cluster	PH (cm)	DFF	NPP	PL (cm)	FLL (cm)	NGP	1000-GW (g.)	KL (mm)	KB (mm)	L/ B	FeC (ppm)	ZnC (ppm)	YPP (g.)
Cluster I	124.99	95.92	10.83	25.46	33.79	211.44	21.58	6.60	2.19	3.03	7.78	19.42	17.46
Cluster II	93.83	98.33	10.78	26.18	35.94	250.81	22.87	6.60	2.30	2.89	7.25	18.75	24.60
Cluster III	106.37	87.60	8.70	23.97	33.70	266.97	19.86	6.38	2.05	3.13	8.46	22.55	18.44
Cluster IV	103.57	109.83	9.33	26.67	32.31	327.78	15.68	5.78	2.07	2.81	8.73	19.47	17.39
Cluster V	99.00	93.50	12.21	28.58	33.84	267.58	28.80	7.17	2.18	3.30	8.13	22.18	26.60
Cluster VI	123.16	84.58	9.88	26.54	37.35	223.80	23.73	6.85	2.28	3.05	9.27	22.01	18.64
Cluster VII	110.77	81.81	9.22	25.22	29.17	185.29	21.52	6.77	2.03	3.35	9.61	26.42	16.59

PH- Plant height, DFF- Days to 50% flowering, NPP- Number of panicles per plant, PL- Panicle length, FLL- Flag leaf length, NGP-Number of grains per panicle, 1000GW- Test weight, KL- Kernel length, KB- Kernel breadth, L/B- Kernel length to breadth ratio, FeC-Grain iron content, ZnC- Grain zinc content, YPP- Yield per plant

Table 5. Inter- and Intra-Cluster distance (Euclidean²) of clusters formed by 38 genotypes evaluated during *Kharif*- 2021

Clusters	I	II	Ш	IV	V	VI	VII
I	59.54	136.35	121.70	216.13	264.41	116.89	264.09
II		87.26	155.75	190.45	197.24	205.40	399.91
III			88.00	197.62	223.74	127.50	202.16
IV				107.85	467.26	363.48	537.17
V					66.17	171.97	278.70
VI						66.14	155.18
VII							100.73

this investigation. The highest intra-cluster distance was observed within the Cluster IV (107.85) comprising three genotypes followed by Cluster VII (100.73) with 13 genotypes, then Cluster III (88.00) with five genotypes and Cluster II (87.26) with three genotypes. Inter-cluster distances are further delineated in descending order, highlighting the maximum Euclidean² distance between Cluster IV and VII (537.17) followed by Cluster IV and VI (467.26), Cluster II and VII (399.91), Cluster IV and VI (363.48), Cluster V and VII (264.41), Cluster II and

IV (264.41), Cluster I and VII (264.09), Cluster III and V (223.74), Cluster I and IV (216.13), whereas the minimum distance recorded between Cluster I and VI (216.13). Crosses among the genotypes selected from these cluster pairs are expected to yield diverse segregants, enabling effective selection as the clustering pattern reflects genetic diversity. Utilizing parents from diverse clusters facilitates the accumulation of favourable alleles within a single genetic background. Similar findings in rice were reported earlier by Anjali *et al.* (2014), Rayala *et al.* (2016),

Table 6. Identification of g	genotypes superior	over check variety f	for grain micronutrients	content and grain vield
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Traits	Number of genotypes	f Genotypes
Grain yield (R. Bhagwati- 18.98 g.)	5	R-RHZ-SK-128 (26.97 g.), CR 2818-1-11-B-1-1-2-B-1 (26.22 g.), Samba Mahsuri (26.04 g.), R-RH2-M1-93 (24.62 g.), R-RHZ-IR-140 (23.14 g.)
Grain Iron content (R. Bhagwati- 9.55 ppm)	1	R-RGY-MH-113 (11.8 ppm)
Grain Zinc content (R. Bhagwati- 22.20 ppm)	10	R-RHZ-SM-14 (32.75 ppm), Zinco rice (31.375 ppm), CGZR-1 (28.3 75 ppm), R-RHZ-IR-132 (27.5 ppm), CSR HZR 17-8 (26.85 ppm), R-RHZ-IH-82 (26 ppm), CSR HZR 17-42 (25.95 ppm), R-RHZ-IB-80 (25.35 ppm), DRR Dhan 49 (25.25 ppm), CR Dhan 311 (25.15 ppm)

CD @5%

Ashok *et al.* (2017), Behera *et al.* (2018), Tripathy (2020) and Singh *et al.* (2020). In breeding for specific traits, parents should be selected from diverse clusters based on inter-cluster distance and appreciable differences in cluster mean values for the specified traits. Moreover, while selecting parents for trait improvement, the importance of yield should not be underestimated.

Based on the mean value it was observed that five genotypes (R-RHZ-SK-128, CR 2818-1-11-1-B-1-1-2-B-1, Samba Mahsuri, R-RH2-M1-93, and R-RHZ-IR-140) excelled in yielding ability. In addition, the genotype (R-RGY-MH-113) was superior for grain iron content and 10 genotypes (R-RHZ-SM-14, Zinco rice, CGZR-1, R-RHZ-IR-132, CSR HZR 17-8, R-RHZ-IH-82, CSR HZR 17-42, R-RHZ-IB-80, and DRR Dhan 49) outperformed the check variety Rajendra Bhagwati for grain Zinc content (**Table 6**). However, no genotype exhibited superiority over the check variety for all crucial traits essential for food and nutritional security.

In conclusion, the genotypes used in this experiment display considerable genetic divergence, indicating their strong potential for utilization in biofortification programs. This diversity could be instrumental in enhancing the nutritional quality of crops, thereby contributing to improved food security and public health.

ACKNOWLEDGEMENT

The authors greatly acknowledge AICRIP coordinating centre, IIRR, Hyderabad and HarvestPlus, Hyderabad for providing the seed material and conducting micronutrient analysis for conducting this research.

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