

GENETIC DIVERSITY ANALYSIS IN RICE (*Oryza sativa* L.)

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Abstract

The experiment was conducted under the agro-ecological zone of Modhupur Tract (AEZ-28) to determine the genetic diversity among 39 rice genotypes, collected from Bangladesh Rice Research Institute, Lal Teer Seed Ltd. and Chittagong Hill Tracts. The genotypes were grouped into six clusters based on Mahalanobis's D^2 statistics and Canonical Variate Analysis. Cluster I consisted with the highest number of (11) genotypes from different origin, whereas cluster IV consisted with the lowest number of (3) genotypes. Cluster III consisted of BRRI dhan 28, BRRI dhan 45, Gold, Tia, Dowel and Moyna showed maximum mean yield. Maximum inter cluster distance was recorded between cluster II and cluster III. The highest inter genotypic distance was observed between Tharak dhan and BRRI dhan 45. Cluster III had the highest mean values for grain yield, harvest index, 1000-grain weight and number of effective tillers per hill and cluster V for number of primary branches per panicle and cluster VI for number of filled grains per panicle that had significant positive correlation with grain yield. Number of unfilled grains per panicle, 1000-grain weight and grain yield showed maximum contribution towards genetic divergence. The genotypes from these clusters with desirable characters may be used as potential donor for future hybridization program to develop high yielders.

Key words: Rice (*Oryza sativa* L.), genetic diversity, D^2 analysis, canonical variate analysis.

INTRODUCTION

Rice (*Oryza sativa* L.) is a self pollinated cereal crop belonging to the family Gramineae (synonym-Poaceae) under the order Cyperales. It is the staple food in Bangladesh, and grown in a wide range of environments ranging from the hilly areas like Chittagong Hill Tracts, Sylhet and Garo Hills, with little moisture, to situations where the water table is 3-4 m deep (Alim, 1982). Bangladesh has a good source of indigenous rice cultivars with two million hectares coverage, while the HYV and hybrids occupy eight million hectares and one million hectares respectively (Talukder, 2011). Knowledge of genetic diversity among existing cultivars of any crop is essential for long term success of breeding program and maximizes the exploitation of the genetic resources (Belaj *et al.*, 2002). Hybridization is one of the major tools for the improvement of a crop that needs the analysis of genetic diversity for the selection of parents (Singh, 1983). Moreover, evaluation of genetic diversity is important for the source genes of particular traits within the available germplasm (Roy and Panwar, 1993).

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Genetic diversity assessment between and within groups or clusters is very important for the proper selection of parents for the better search of heterosis (Murty and Arunachalam, 1966). Multivariate analysis with D^2 technique measures the amount of genetic diversity in a given population in respect of several characters and assesses relative contribution of different components to the total divergence (Zahan *et al.*, 2008). This study was undertaken to determine the genetic diversity in rice for the maximum utilization of the genetic resources and proper selection of donor parents.

MATERIALS AND METHODS

The experiment was conducted at the experimental field of Sher-e-Bangla Agricultural University (SAU), Dhaka 1207, Bangladesh, in irrigated condition during January to June in 2011. The experimental materials comprised of 39 rice genotypes collected from Genetic Resources and Seed (GRS) Division of Bangladesh Rice Research Institute (BRRI), Lal Teer Seed Ltd. (LTSL) and Chittagong Hill Tracts (CHT) (Table 1). The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications. Row and plant spacing were maintained 25×20 cm and two seedlings per hill were transplanted. Experimental plots were fertilized with the recommended doses of 140-20-60-20-4 kg/ha N-P-K-S-Zn (BRRI, 2011). First split of nitrogenous fertilizer and full portion of all other fertilizers were added to the main land during final ploughing. The second and third splits of nitrogenous fertilizer were applied at 4-5 tillering stage and 5-7 days before of panicle initiation, respectively.

Table 1. List of thirty nine rice genotypes along with their sources

Accession no.	Genotypes	Source	Accession no.	Genotypes	Source
G1	Sada Kabarak	CHT	G21	BR-24 (Rahmat)	GRS, BRRI
G2	Kala kabarak	CHT	G22	BR-25 (Naya Pajam)	GRS, BRRI
G3	Kamrang dhan	CHT	G23	BR-26 (Shraboni)	GRS, BRRI
G4	Badeye dhan	CHT	G24	BRRI dhan-27	GRS, BRRI
G5	Sada gelon	CHT	G25	BRRI dhan-28	GRS, BRRI
G6	Jumma Bini (longer grain)	CHT	G26	BRRI dhan-29	GRS, BRRI
G7	Jumma Bini (shorter grain)	CHT	G27	BRRI dhan-30	GRS, BRRI
G8	Kabarak dhan	CHT	G28	BRRI dhan-31	GRS, BRRI
G9	Ameiye dhan	CHT	G29	BRRI dhan-32	GRS, BRRI
G10	Tharak dhan	CHT	G30	BRRI dhan-33	GRS, BRRI
G11	Paijam (Indian)	CHT	G31	BRRI dhan-36	GRS, BRRI
G12	Jumma Kabarak	CHT	G32	BRRI dhan-39	GRS, BRRI
G13	Gold (China)	LTSL	G33	BRRI dhan-40	GRS, BRRI
G14	Tia (China)	LTSL	G34	BRRI dhan-42	GRS, BRRI
G15	Dowel (China)	LTSL	G35	BRRI dhan-44	GRS, BRRI
G16	Moyna (China)	LTSL	G36	BRRI dhan-45	GRS, BRRI
G17	BR-3 (Biplob)	GRS, BRRI	G37	BRRI dhan-48	GRS, BRRI
G18	BR-10 (Pragati)	GRS, BRRI	G38	BRRI dhan-49	GRS, BRRI
G19	BR-11 (Mukta)	GRS, BRRI	G39	BRRI dhan-50	GRS, BRRI
G20	BR-22 (Kiran)	GRS, BRRI			

Data were collected on 12 agronomic characters *viz.* plant height at maturity (cm), number of effective tillers per hill, length of panicles (cm), days to 80% flowering (days after sowing), number of primary branches per panicle, number of secondary branches per panicle, days to maturity (days after sowing), number of unfilled grains per panicle, number of filled grains per panicle, 1000-grain weight (g), harvest index and grain yield per hill (g). Data were taken from 10 hills of each genotype. All the analyses in respect of twelve different morphological traits of 39 rice genotypes were analyzed by multivariate analyses using the GENSTAT 5-13 software program. The genotypes were grouped into clusters based on Mahalanobis's D^2 statistics and canonical variate analysis.

RESULTS AND DISCUSSION

Principal component analysis

The principal component analysis (PCA) provided eigen values and percent of variation for 12 principal component axes in 39 rice genotypes (Table 2). The results showed that only the first three principal components jointly accounted for 65.79% of the total variation among the genotypes, while these three exhibited eigen values above unity. The first two principal axes accounted for 52.76% of the total variation among the 12 component axes of the total genotypes (Table 2). Principal component analysis was also reported by Anandan *et al.* (2011) and Latif *et al.* (2011) in rice.

Table 2. Eigen values and percent of variation for 12 principal component axes in 39 rice genotypes

Component axes	Eigen values	Percent variation	Cumulative Percent variation
1	3.77	31.44	31.44
2	2.56	21.32	52.76
3	1.56	13.03	65.79
4	0.95	7.94	73.73
5	0.87	7.28	81.01
6	0.74	6.17	87.18
7	0.59	4.89	92.07
8	0.31	2.62	94.69
9	0.30	2.47	97.16
10	0.15	1.28	98.44
11	0.11	0.93	99.37
12	0.08	0.63	100.00

Construction of scattered diagram

A two dimensional scatter diagram (Z_1 - Z_2) was constructed using component 1 as X-axis and component 2 as Y-axis based on the values of principal component scores 1 and 2 obtained from the principal component analysis. The genotypes were grouped into six clusters indicating the presence of considerable genetic diversity among genotypes (Figure 1). BRR1 dhan 45 (G 36) was distantly located from Tharak dhan (G 10).

Principal coordinate analysis

Inter genotypic distance was obtained from principal coordinate analysis on auxiliary of principal component analysis for all possible combinations between pairs of genotypes (Table 3). Maximum inter genotypic distance was found between Tharak and BRR1 dhan 45 (237.41) followed by Jumma Bini (shorter grain) and BRR1 dhan 45 (224.81), Badeye and BRR1 dhan 45 (219.43), while minimum inter genotypic distance was found between Paijam (India) and BRR1 dhan 50 (1.23) followed by Kabarak and BR 25 (2.24), Kamrang and Paijam (India) (2.43). The presence of differences between the highest and the lowest inter genotypic distances indicated the presence of variability among 39 rice genotypes. Similar results also obtained by Latif *et al.* (2011) in rice.

Canonical variate analysis

The canonical variate analysis, complementary to Mahalanobis's D^2 statistics, was carried out to obtain the cluster distances (Mahalanobis's D^2 values) that indicated

the index of genetic diversity among them. There was wider genetic diversity among the genotypes of different cluster as inter cluster distances were larger than intra cluster distances (Table 4). Similar results were also suggested by Anandan *et al.* (2011) and Latif *et al.* (2011) in rice.

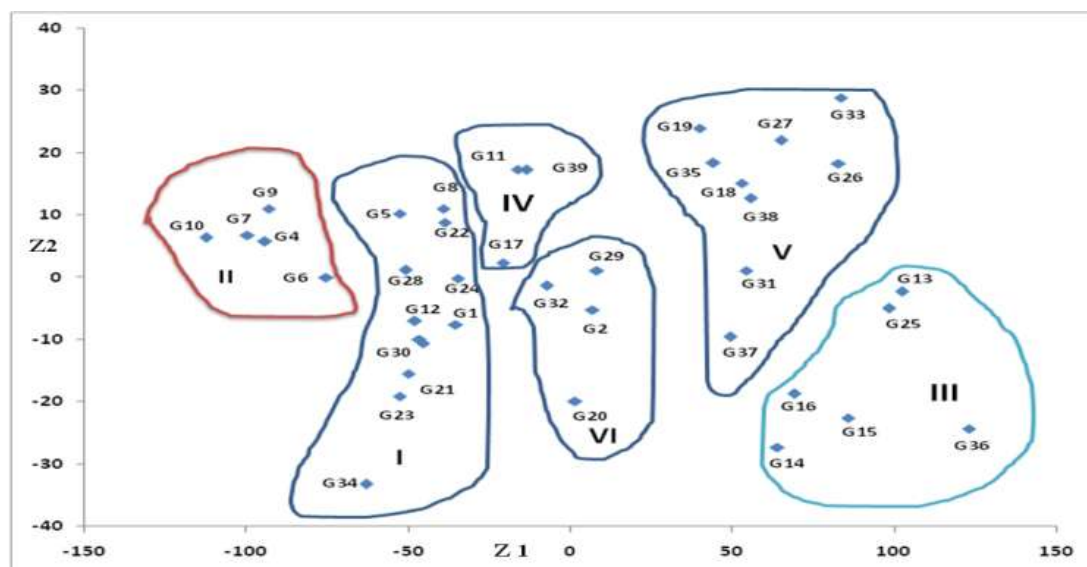


Figure 1. Scatter diagram of 39 rice genotypes based on PCA scores superimposed with clustering from D^2 analysis

Table 3. Ten highest and ten lowest inter genotypic distance among 39 rice genotypes

Highest distance				Lowest distance			
Sl. No.	Genotypes		Distance	Sl. No.	Genotype		Distance
01	Tharak	BRRRI dhan 45	237.41	01	Paijam (India)	BRRRI dhan 50	1.23
02	Jumma Bini (Shorter grain)	BRRRI dhan 45	224.81	02	Kabarak	BR 25	2.24
03	Badeye	BRRRI dhan 45	219.43	03	Kamrang	Paijam (India)	2.43
04	Ameiye	BRRRI dhan 45	218.95	04	Jumma Kabarak	BRRRI dhan 33	3.23
05	Tharak	Sada Kabarak	214.95	05	BR 10	BRRRI dhan 49	3.75
06	Tharak	BRRRI dhan 28	211.05	06	Jumma Kabarak	BRRRI dhan 50	4.25
07	Jumma Bini (Shorter grain)	Sada Kabarak	202.22	07	BR 24	BRRRI dhan 26	4.54
08	Tharak	Dowel	200.21	08	Sada Kabarak	BRRRI dhan 28	4.87
09	Jumma Bini (longer grain)	BRRRI dhan 45	199.85	09	Badeye	Jumma Bini (Shorter grain)	5.39
10	Jumma Bini (shorter grain)	BRRRI dhan 28	198.33	10	Badeye	Ameiye	5.45

Maximum inter cluster distance was observed between clusters II and III (15.84) indicating the highest diversity among the genotypes of these clusters followed by clusters II and V (11.91), clusters I and III (11.68), and clusters III and IV (10.99), while the minimum was between clusters I and VI (3.61) indicating the lowest diversity among the genotypes of these clusters followed by clusters I and II (4.83), clusters IV and VI (5.50), and clusters V and VI (5.69). The more inter and intra cluster distances, the more variability among the genotypes between and within the cluster, respectively and vice versa. The cluster means of 12 characters of 39 rice genotypes are presented in Table 6.

There was also observed the existence of differences for almost all of the characters. Genotypes of distantly located clusters were suggested to use in hybridization programs for obtaining a wide spectrum of variation among the segregates as suggested by Yadav *et al.* (2011), Vennila *et al.* (2011) and Latif *et al.* (2011).

Table 4. Intra cluster distances (Bold) and inter cluster distances (D²) for 39 rice genotypes

Cluster	I	II	III	IV	V	VI
I	0.02	4.83	11.68	5.95	8.66	3.61
II		0.14	15.84	7.97	11.91	7.95
III			0.07	10.99	6.38	8.44
IV				0.59	6.97	5.50
V					0.03	5.69
VI						0.14

Non-hierarchical clustering

Thirty nine rice genotypes were grouped into six different clusters based on non-hierarchical clustering using co-variance matrix. Distributions of genotypes into different clusters are presented in Table 5. Cluster I contained maximum number of genotypes (11) while cluster V, cluster III, cluster II/cluster VI and cluster IV contained 9, 6, 5 and 3 genotypes, respectively (Table 5). The genotypes collected from Chittagong Hill Tracts those are mostly cultivated in Jhum (shifting cultivation) were distributed in all clusters except in cluster V as days to flowering was maximum (113.58 DAS) in this cluster (Table 6). Whereas, the genotypes collected from BIRRI were distributed in all clusters except in cluster II as plant height was maximum (114.10 cm) in this cluster (Table 6). All the genotypes collected from Lal Teer Seed Ltd. along with BIRRI dhan 28 and BIRRI dhan 45 were grouped in cluster III that were resulted maximum yield in this study (Table 6). The clustering pattern suggests that the genotypes collected from same locations (hilly area) can also be grouped into different clusters. This showed that geographic distribution was not related to genetic diversity of these materials. Similar results were also reported by Yadav *et al.* (2011) and Anandan *et al.* (2011) in rice.

Table 5. Distribution of 39 rice genotypes in different clusters

Cluster	Members	Genotypes
I	11	Sada Kabarak, Sada Gelon, Kabarak, Jumma Kabarak, BR-24, BR-25, BR-26, BIRRI dhan 27, BIRRI dhan 31, BIRRI dhan 33, BIRRI dhan 42
II	5	Badeye, Jumma Bini (longer grain), Jumma Bini (shorter grain), Ameiye, Tharak
III	6	Gold, Tia, Dowel, Moyna, BIRRI dhan 28, BIRRI dhan 45
IV	3	Paijam (Indian), BR-3, BIRRI dhan 50
V	9	BR 10, BR 11, BIRRI dhan 29, BIRRI dhan 30, BIRRI dhan 36, BIRRI dhan 40, BIRRI dhan 44, BIRRI dhan 48, BIRRI dhan 49
VI	5	Kala Kabarak, Kamrang, BR 22, BIRRI dhan 32, BIRRI dhan 39

Characterization of individual cluster

Intra cluster mean values of 12 different characters of 39 rice genotypes are presented in Table 6. Maximum intra cluster mean for plant height was found in cluster II (114.10) followed by cluster I (110.41) and cluster VI (106.88), whereas the lowest intra cluster mean was found in cluster IV (81.51) followed by cluster V (92.44) and cluster III (93.42). The maximum intra cluster mean for number of effective tillers per

hill was observed in cluster III (11.57) followed by cluster V (11.44) and cluster IV (10.98), whereas the lowest was found in cluster VI (10.36) followed by cluster II (10.50) and cluster I (10.64). The maximum intra cluster mean for panicle length was shown in cluster V (26.72) followed by cluster II (25.82) and cluster VI (25.32), whereas the lowest was in cluster III (23.77) followed by cluster I (25.06) and cluster IV (25.26). The maximum intra cluster mean for days to 80% flowering was observed in cluster V (113.58) followed by cluster IV (110.78) and cluster II (109.47), whereas the minimum was observed in cluster III (91.78) followed by cluster I (102.88) and cluster VI (104.60). The highest intra cluster mean for number of primary branches per panicle was observed in cluster V (11.71) followed by cluster II (10.62) and cluster VI (10.31), whereas the minimum was observed in cluster III (9.03) followed by cluster IV (9.52) and cluster I (10.18). The maximum intra cluster mean for number of secondary branches per panicle was found in cluster II (29.80) followed by cluster I (26.79) and cluster VI (25.09), whereas the minimum was in cluster III (20.24) followed by cluster IV (24.26) and cluster V (25.06). The highest intra cluster mean for number of unfilled grains per panicle was observed in cluster IV (30.82) followed by cluster II (27.31) and cluster I (22.17), whereas the minimum was in cluster III (17.39) followed by cluster VI (18.02) and cluster V (21.62). The maximum intra cluster mean was found in cluster VI (142.22) followed by cluster V (141.80) and cluster III (135.22), whereas the minimum was found in cluster IV (120.44) followed by cluster II (126.96) and cluster I (134.39). The maximum intra cluster mean for 1000-grains weight was observed in cluster III (27.69) followed by cluster VI (25.87) and cluster V (24.77), whereas the lowest was in cluster II (19.73) followed by cluster IV (22.89) and cluster I (24.40). The maximum intra cluster mean for days to maturity was shown in cluster II (143.07) followed by cluster V (142.19) and cluster IV (140.67), whereas the minimum was in cluster III (119.50) followed by cluster I (129.82) and cluster VI (130.13). The highest intra cluster mean for harvest index was observed in cluster III (0.55) followed by cluster V (0.53) and cluster IV (0.51), whereas the lowest was observed in cluster II (0.35) followed by cluster I (0.44) and cluster VI (0.46). The maximum intra cluster mean for grain yield per hill was observed in cluster III (33.27) followed by cluster V (30.17) and cluster VI (24.23), whereas the minimum was in cluster II (15.03) followed by cluster I (19.73) and cluster IV (21.52). The highest intra cluster mean for respected characters were shown in bold and the lowest intra cluster mean were shown in italic in Table 6. The cluster III had ranked first for the maximum times (4 times) followed by clusters II and V (3 times each), cluster IV and VI (once each) for maximum values, whereas cluster I had no first ranking for maximum value. However, the cluster III again had ranked in first for the maximum times (6 times) followed by cluster II (3 times), cluster IV (2 times) and cluster VI (once) for minimum values, whereas there was no first ranking of cluster I and cluster V for minimum value. Therefore, most of the important traits present in genotypes under cluster III.

Contribution of characters towards divergence

The relative contributions of twelve different characters of thirty nine rice genotypes towards the total divergence are presented in Table 7. The result of the canonical variate analysis revealed that number of unfilled grains per panicle, 1000-grain weight and grain yield per hill had positive values in both the vectors, whereas number of effective tillers per hill, days to 80% flowering, number of primary branches per panicle, number of filled grains per panicle and days to maturity had negative values. The positive values in both the vectors revealed the most important whereas the negative values in both the vectors indicated the least important of these characters towards the

divergence among thirty nine rice genotypes. In the vector 1 (Z_1), the all other characters, excluding those were showing positive values in both vectors, showed negative values indicating that these characters showed less importance towards the genetic divergence. In the vector 2 (Z_2), the other important characters towards the genetic divergence were plant height, panicle length, number of secondary branches per panicle and harvest index. Therefore, importance should be given on number of unfilled grains per panicle, 1000-grain weight and grain yield per hill. Similar results were also shown by Alam *et al.* (2006) in barley.

Table 6. Intra cluster mean values (the highest with bold and the lowest with italic bold) of 12 different characters of 39 genotypes

Characters	I	II	III	IV	V	VI
Plant height (cm)	110.41	114.10	93.42	<i>81.51</i>	92.44	106.88
Effective tillers per hill (no.)	10.64	10.50	11.57	10.98	11.44	<i>10.36</i>
Panicle length (cm)	25.06	25.82	23.77	25.26	26.72	25.32
Days to 80% flowering (DAS*)	102.88	109.47	91.78	110.78	113.58	104.60
Primary branches per panicle	10.18	10.62	9.03	9.52	11.71	10.31
Secondary branches per panicle	26.79	29.80	<i>20.24</i>	24.26	25.06	25.09
Days to maturity (DAS*)	129.82	143.07	<i>119.50</i>	140.67	142.19	130.13
Unfilled grains per panicle (no.)	22.17	27.31	<i>17.39</i>	30.82	21.62	18.02
Filled grains per panicle (no.)	134.39	126.96	135.22	120.44	141.80	142.22
1000 grains weight (g)	24.40	19.73	27.69	22.89	24.77	25.87
Harvest index	0.44	0.35	0.55	0.51	0.53	0.46
Grain yield per hill (g)	19.73	15.03	33.27	21.52	30.17	24.23

*DAS=Days after sowing

Table 7. Relative contributions of the twelve characters of 39 rice genotypes to the total divergence

Characters	Vector-1	Vector-2
Plant height (cm)	-0.0297	0.1247
Tillers per hill (no.)	-0.1531	-0.0932
Days to 80% flowering	-0.0003	-0.0363
Panicle length (cm)	-0.0069	0.1609
Primary branches per panicle (no.)	-0.2114	-0.6256
Secondary branches per panicle (no.)	-0.0103	0.0717
Unfilled grains per panicle (no.)	0.0498	0.0327
Filled grains per panicle (no.)	-0.0341	-0.1004
1000-grain weight (g)	0.0363	0.1541
Days to maturity	-0.0150	-0.0185
Harvest index	-4.4883	1.3369
Grain yield per hill (g)	0.0823	0.0089

Selection of characters for future improvement

The parents for hybridization program should be selected based on the magnitude of genetic distance, contribution of different characters towards the total divergence and magnitude of cluster means for different characters performance having maximum heterosis. A higher heterosis could be produced from the crosses between genetically distant parents (Falconer, 1960). Therefore crosses between the genotypes of clusters II and III, II and V, I and III, III and IV would give high manifestation of heterosis as well as wide spectrum of genetic variation in F_2 generation. Genotypes included in cluster III

was important for number of effective tillers per hill, earliness, number of unfilled grains per panicle, 1000-grain weight, harvest index and grain yield per hill. The genotypes of cluster VI were important for number of filled grains per panicle, cluster II was important for dwarfness and number of secondary branches. The most distantly genotypes were between cluster II and cluster III. The most important cluster was cluster III (Table 6). The important characters reported by some researchers were plant height, panicle length, number of unfilled grains per panicle, 1000-grain weight (Latif *et al.*, 2011 and Sabesan *et al.*, 2009), grain yield, number of effective tillers (Baradhan and Thangavel, 2011), harvest index (Ahmed *et al.*, 2010), days to maturity (Kanwal *et al.*, 1983) and number of secondary branches per panicle (Sinha *et al.*, 1991). The most important characters contributing towards the divergence were number of unfilled grains per panicle, 1000-grain weight and grain yield per hill.

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