

Original Research Article

Evaluation of Breeding Potential of Agro-morphological Important Traits in Forty Two Aman Rice Genotypes (*Oryza sativa* L.) using D² analysis

ABSTRACT: The experiment was conducted with forty two genotypes of rice at the experimental farm of Sher-e-Bangla Agricultural University, Dhaka during the period of July 2013- December 2013 to estimate the diversity among 42 Aman rice genotypes. As PCA, D² and clusters analysis the genotypes were grouped into five different clusters. Clusters IV had the maximum fourteen and cluster III had the minimum three genotypes. The highest inter-cluster distance was observed between II and III and the lowest was between IV and V. The highest and lowest intra-cluster distance was observed in III and IV respectively. Genotypes included in cluster V were important for total tillers per plant, effective tillers per plant, days to 50% flowering and thousand grains weight; cluster III were important for filled grains per panicle and unfilled grains per panicle; cluster IV were for plant height and yield per plant. Considering the diversity pattern and other agronomic performances, the genotypes Special from AL-29, AL-36, PP-4B(i), AL-17(iii)B, AL-17(iii), AL-17(ii)A, Special from-129, Special from 17(iv), AL-44(i), AL-17, Special from AL-36(D), PP-48, IR-25B, Special from AL-33, IR-25B (Tall), P-5B might be considered better parents for the future efficient hybridization programme.

KEY WORDS: Diversity analysis, Hybridization, Canonical variate analysis, Principal component analysis, Scatter diagram.

1 INTRODUCTION

Rice is a major food crop, ranking second to wheat among the most cultivated cereals in the world. It is the staple food crop of more than half of the world's population [1]. Rice provides 21% energy and 15% of per capita protein of global human [2]. The world dedicated 162.3 million hectares in 2012 for rice cultivation and the total production were about 738.1 million tons [3]. By 2030, the world must have to produce 60% more rice than it produce in 1995 to meet the demands [4].

More than 90% of the world's rice is produced and consumed in Asia [5]. Bangladesh is the fourth largest producer of rice in the world with production of 33.8 million tons in 2012 [6]. It occupies 74.77% total cropped areas and it alone constitutes 90% of the total food grain produced annually in the country [7]. Bangladesh needs 2.7% increases in rice production per year due to increasing population [8].

Rice is grown in Bangladesh under diverse ecosystem of irrigated, rain fed and deep water conditions in three distinct seasons namely Aus, Aman, and Boro [9]. Among Aus, Aman and Boro seasons, Aman occupied the highest area coverage (34% of gross cropped area) [7]. Moreover, there is lack of well developed Aman rice variety. So, we have to give more attention

for the improvement of Aman rice varieties to increase rice production in order to satisfy our population's need of food.

Genetic diversity determines the inherent potential of a cross for heterosis and frequency of desirable recombinants in advanced generations [10]. Hybridization programme involving genetically diverse parents belonging to different clusters would provide an opportunity for bringing together gene constellations of diverse nature, promising hybrid derivatives resulted probably due to complementary interaction of divergent genes in parents [11].

1.1.1 OBJECTIVE

The present study was undertaken to assess the genetic diversity among the Aman rice genotypes for identifying the genetically divergent parents to utilize them in the future hybridization programme.

2 MATERIALS AND METHODS

2.1 EXPERIMENTAL SITE AND EXPERIMENTAL PERIOD

The present research work was carried out in the experimental farm, Sher-e-Bangla Agricultural University (SAU), Dhaka during July 2013-December 2013. The location of the site is 23° 74' N latitude and 90° 35' E longitude with an elevation of 8.2 meter from sea level.

2.2 CLIMATE AND SOIL

The experimental site was situated in the sub-tropical zone. The soil of the experimental site lies in Agroecological region of "Madhupur Tract" (AEZ No. 28). Its top soil is clay loam in texture and olive gray with common fine to medium distinct dark yellowish brown mottles. The pH is 6.1 and organic carbon content is 0.82%.

2.3 MATERIALS

Forty two rice genotypes or lines[G1: AL-17 (iii) B, G2: AL-17 (iii), G3: AL-42, G4: From IR-25B (Yellow), G5: From IR-25B, G6: Special (Early), G7: Richer, G8: AL-35, G9: Special-130, G10: IR-25B (Dwarf), G11: AL-36(C), G12: Special from AL-36(D), G13: P-5B (i), G14: Hira, G15: AL-17 (iii) (Tall), G16: Aloron, G17: IR-25B (Tall), G18: AL-36(iii), G19: Special from AL-33, G20: Special from AL-29 (Basmati type), G21:S-1 (Basmati type), G22: S-2 (Basmati type), G23; S-5 (Basmati type), G24: AL-33(ii) (Basmati type), G25: AL-36 (Basmati type), G26: AL-42(ii) (Basmati-Yellow type), G27: AL-44(i) (Basmati-Yellow type), G28: AL-17(ii) A, G29: AL-47, G30: PP-48, G31: PP-4B(i), G32: Special from-129, G33: P-5B

(ii), G34: AL-36, G35: Special stigma color, G36: IR-25B, G37: Special from-17(iv), G38: Special from S-2, G39: Special from AL-33, G40: AL-44(i), G41: AL-17 and G42: AL-104] were used for the present study. The genetically pure and physically healthy seeds of these genotypes were obtained from the Department of Genetics and Plant Breeding (SAU), Dhaka.

2.4 LAND PREPARATION

The experimental plot was prepared by ploughing with proper tiller. The weeds and other unwanted plant materials were removed from the field during the land preparation. Proper laddering was done to bring the soil at proper tilth condition.

2.5 EXPERIMENTAL DESIGN AND LAYOUT

A Randomized Complete Block Design (RCBD) was used in the experiment with three replications. The field was divided into three blocks; the blocks were subdivided into 42 plots. Genotypes were randomly assigned into 42 plots in each block. The plot size was 2m × 1m. Block to block and plot to plot distance were 1 m and .5 m respectively.

2.6 SOWING AND TRANSPLANTING

The seeds were sown on 22 July 2013 and 1 seedling per hill was transplanted to the main plot on 24 August 2013. Plant to plant and row to row distance were maintained at 20 cm and 25 cm respectively.

2.7 FERTILIZER APPLICATION

Urea, TSP, MP, Gypsum @ 180, 55, 80, 60 kg/ha respectively were used in the experiment. Total TSP, MP and Gypsum were applied in final land preparation. Total urea was applied in three installments, at 15 days after transplanting (DAT), 35 DAT and 55 DAT respectively.

2.8 INTERCULTURAL OPERATION AND AFTER CARE

Necessary intercultural operation was taken during cropping period for proper growth and development of the plants. Weeding, during first two top dressing of urea, was done to break the soil crust, to keep the plots free from weed and to incorporate the urea fertilizer into the soil for reducing the loss of urea through denitrification. Irrigation with regular interval was given to maintain 5-7 cm water up to hard dough stage of rice.

2.9 PLANT PROTECTION MEASURES

Proper control measures were taken against rice stem borer during tillering and heading stage of rice. Furadan 5G @ 1 g per square meter were applied at active tillering stage and panicle initiation stage of rice for controlling the stem borer. Lani rate and malathion were applied to protect from rat and rice bug respectively.

2.10 IRRIGATION

2-3 cm of water level was maintained at the time of transplanting. 5 cm depth of water was given at the time of maximum tillering stage. Then field was irrigated as and when required up to physiological maturity.

2.11 HARVESTING

Harvesting was started when 80% of the plant population of each plot reached maturity.

2.12 COLLECTION OF DATA

Data were recorded on individual plant basis from 10 randomly selected plants. Observations were recorded on various plant traits i.e. days to 50% flowering, days to maturity, plant height, total tillers per plant, effective tillers per plant, panicle length, fertile grains per panicle, unfertile grains per panicle, 1000 grains weight and yield per plant.

2.13 STATISTICAL ANALYSIS

Multivariate analysis viz., Principal Component Analysis (PCO), Cluster Analysis (CLU) and Canonical Variate Analysis (CVA) were done by using GENSTAT programme. Genetic divergence among the genotypes was assessed by [12] generalized distance (D^2) statistic and its auxiliary analyses.

3 RESULTS AND DISCUSSION

Diversity is the function of parent selection and also heterosis. The availability of transgressive segregants in a breeding programme depends upon the divergence of parents. Thus, the accurate information on the nature and degree of diversity of the parents is the prerequisite of an effective breeding programme. Selection of parents in hybridization programme based on Mahalanobis's D^2 statistic is more reliable as requisite knowledge of parents in respect of a mass of characteristics is available prior to crossing. Reference [13] reported that the quantification of genetic diversity through biometrical procedures had made it possible to choose genetically diverse parents for a successful hybridization program. Statistical analysis

such as Mahalanobis D^2 and Canonical Variate Analysis (CVA), which quantify the differences among several quantitative traits are efficient method of evaluating genetic diversity.

3.1 PRINCIPAL COMPONENT ANALYSIS

The principal Components analysis yielded eigen values of each principal component axes of **coordination** of genotypes in which the first axes totally accounting for the variation among the genotypes, whereas four of these eigen values above unity accounted for 74.19%. The first three principal axes accounted for 62.50 % of the total variation among **the 10 characters describing in 42 rice genotypes** (Table 1). Based on principal component axis I and II, a two dimensional chart ($Z_1 - Z_2$) of the genotypes are presented in Figure 1. The scatter diagram (Figure 1) represented that apparently there were **mainly** five clusters and the genotypes were distantly located from each other.

3.2 CONSTRUCTION OF SCATTER DIAGRAM

Based on the values of principal component scores 2 and 1 obtained from the principal component analysis, a two dimensional ($Z_1 - Z_2$) scatter diagram was constructed, using component score 1 as X-axis and component score 2 as Y-axis, which is presented in Figure 1. The positions of the genotypes in the scatter diagram were random, which indicated the considerable diversity among the genotypes included in the cluster. **Some distantly located genotypes of different clusters were the genotypes number G31, G20, G25, G15, G37, G40, G36, G30, G28, G1, G32, G41, G12, G39, G33, G17.**

Table 1. Eigen values and yield percent contribution of 10 characters of 42 rice genotypes

Characters	Eigen values	Percent variation	Cumulative % of Percent variation
Plant height (cm)	3.053	30.53	30.53
Total tillers per plant	1.900	19.00	49.53
Effective tillers per plant	1.297	12.97	62.50
Days to 50% flowering	1.169	11.69	74.19
Panicle Length (cm)	0.895	8.95	83.14
Filled Grains per panicle	0.645	6.45	89.59
Unfilled Grains per panicle	0.506	5.06	94.65
Days to maturity	0.291	2.91	97.56
1000 Grains Weight (g)	0.227	2.27	99.83
Yield per plant (g)	0.016	0.17	100.00

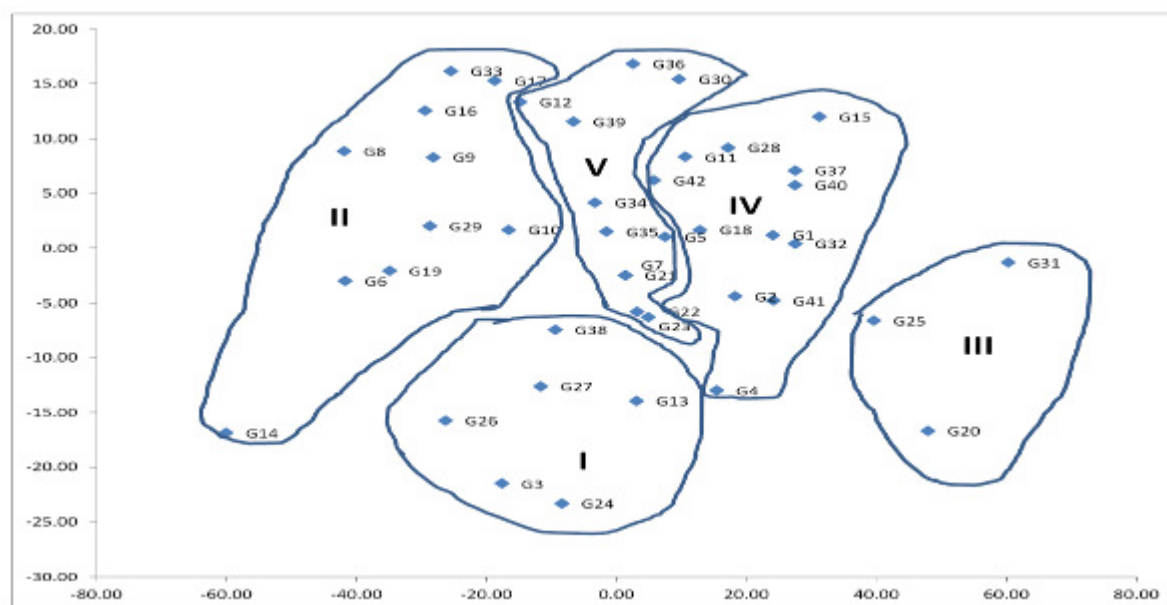


Fig. 1. Scatter Diagram of Forty Two Rice Genotypes based on their Principal Component Scores

3.3 NON-HIERARCHICAL CLUSTERING

By using these inter-genotypic distances intra-cluster genotypic distances were calculated (Table 4) as suggested by [14]. On the basis of D^2 values, the 42 genotypes were grouped into five highly divergent clusters (Table 2). The clusters divergence was proved by the high inter-cluster and low intra clusters D^2 values. Cluster IV was the largest and consisted fourteen genotypes followed by cluster II with ten genotypes, clusters V, I and III had 9, 6 and 3 genotypes respectively. The grouping pattern did not show any relationship between genetic divergence and geographical diversity which has been a point of debate in the past. A perusal of the Table 2 clearly showed the genotypes usually did not cluster according to geographical distributions. One of the possible reasons may be the fact that it is very difficult to establish the actual location of origin of a genotype. The free and frequent exchange of genetic material among the crop improvement programmes in the country makes it difficult to maintain the real identify of the genotypes. Moreover, breeding progenies incorporate genes from varied sources, thus losing the basic geographical identity of the genotype. The absence of relationship between genetic diversity and geographical distance indicates that forces others than geographical origin, such as exchange of genetic stocks, genetic drift, spontaneous variation, natural and artificial selection are responsible for genetic diversity. It may also be possible that causes for clustering pattern were much influenced by environment and (genotype x environment) interaction resulting in differential gene expression. Another possibility may be that estimates might not have been sufficient to account for the variability caused some other traits of physiological or biochemical nature which might have important in depicting the total genetic diversity in the population.

The cluster mean of 42 genotypes (Table 3) showed that the mean value of clusters varied in magnitude for all the ten characters. Genotypes in cluster I showed maximum performance for

panicle length (27.14). Cluster II showed maximum performance for days to maturity (134.13). Cluster III recorded highest mean performance for filled grain per panicle (197.11) and unfilled grains per panicle (31.70). Cluster IV showed maximum performance for plant height (108.15) and yield per plant (41.99). Cluster V showed maximum performance for total tillers per plant (22.90), effective tillers per plant (21.83), days to 50% flowering (103.11) and thousand grains weight (24.93).

Table 2. Distribution of 42 rice genotypes in different clusters

Cluster no.	No. of Genotypes	No. of populations	Name of genotypes
I	3, 13, 24, 26, 27, 38	6	AL-42, P-5B (i), AL-33(ii), AL-42(ii), AL-44(i), Special from S-2
II	6, 8, 9, 10, 14, 16, 17, 19, 29, 33	10	Special (Early), AL-35, Special-130, IR-25B (Dwarf), Hira, Aloron, IR-25B (Tall), Specil from AL-33, AL-47, P-5B (ii)
III	20, 25, 31	3	Special from AL-29, AL-36, PP-4B(i)
IV	1, 2, 4, 5, 11, 15, 18, 22, 28, 32, 37, 40, 41, 42	14	AL-17(iii)B, AL-17(iii), From IR-25B (Yellow), From IR-25B, AL-36 (C), AL-17(iii) (Tall), AL-36(iii), S-2, AL-17(ii) A, Special from-129, Special from-17(iv), AL-44 (i), AL-17, AL-104
V	7, 12, 21, 23, 30, 34, 35, 36, 39	9	Richer, Special from AL-36(D), S-1, S-5, PP-48, AL-36, Special stigma color, IR-25B, Specil from AL-33

Table 3. Cluster mean values of 10 different characters of 42 genotypes

Characters	I	II	III	IV	V
Plant height (cm)	99.67	100.37	102.37	108.15	97.56
Total tillers per plant	16.46	22.12	19.52	19.20	22.90
Effective tillers per plant	15.07	20.10	18.74	17.77	21.83
Days to 50% flowering	98.50	102.57	100.67	101.12	103.11
Panicle length (cm)	27.14	24.42	27.03	26.49	25.42
Filled grains per panicle	137.57	115.76	197.11	165.63	148.28
Unfilled grains per panicle	20.73	16.41	31.70	21.31	14.54
Days to maturity	130.56	134.13	134.11	133.19	134.00
Thousand grains weight (g)	24.24	23.71	23.62	24.55	24.93
Yield per plant (g)	23.02	40.13	35.58	41.99	41.38

3.4 CANONICAL VARIATE ANALYSIS (CVA)

Canonical Variate Analysis was performed to compute the inter-cluster Mahalanobis's values. Statistical distances represent the index of genetic diversity among the clusters. The divergence within the cluster (intra-cluster distance) indicates the divergence among falling in the same cluster. On the other hand, inter cluster divergence suggest the distance (divergence)

between the genotypes of different clusters. The intra and inter clusters D^2 values among 42 genotypes presented in Table 4 revealed that cluster IV showed minimum intra cluster D^2 value (0.01) distance followed by cluster II (0.105), whereas, maximum intra cluster D^2 value (0.81) was shown by cluster III followed by cluster I (0.10) indicated that **genotypes included in this cluster are very diverse** and was due to both natural and artificial selection forces among the genotypes. Minimum inter cluster D^2 value was observed between the clusters IV and V (3.82) indicated close relationship among the genotypes included in these clusters. Maximum inter-clusters D^2 value was observed between the clusters II and III (10.94) indicated that the genotypes belonging to these groups were genetically most divergent and the genotypes included in these clusters can be used as a parent in hybridization programme to get higher heterotic hybrids from the segregant population [15]. Several authors also reported profound diversity in the germplasm of rice by assessing genetic divergence on the basis of quantitative traits following Mahalanobis D^2 statistics [11], [10] and [16]. Average inter and intra-cluster distance revealed that, in general inter-cluster distance were much higher than those of intra-cluster distances, suggesting homogenous and heterogeneous nature of the germplasm lines within and between the clusters, respectively. These results are in accordance with the findings of [11].

Results obtained from different multivariate techniques from which it may be concluded that all the techniques gave more or less similar results and one technique supplemented and confirmed the results of another one.

Table 4. Intra (Bold) and inter cluster distances (D^2) for 42 rice genotypes

Cluster	I	II	III	IV	V
I	0.10	4.66	8.54	5.47	4.15
II		0.03	10.94	6.61	4.56
III			0.81	4.80	7.37
IV				0.01	3.82
V					0.04

The clustering pattern of the genotypes revealed that varieties or lines originating from the same places did not form a single cluster because of direct selection pressure. This indicated that geographic diversity was not related to genetic diversity that might be due to continuous exchange of genetic materials among the countries of the world. Same results have been reported by [17] and [18] in brown mustard; [19] in sunflower; [20] in groundnut and soybean. It had

been observed that geographic diversity was not always related to genetic diversity and therefore, it was not adequate as an index of genetic diversity. Reference [21] studied that genetic drift and selection in different environment could cause greater diversity than geographic distance. Furthermore, there was a free exchange of seed material among different region, as a consequence, the characters constellation that might be associated with particular region in nature, lose their individually under human interference, and however, in some cases effect of geographic origin influenced clustering that was why geographic distribution was not the sole criterion of genetic diversity.

The free clustering of the genotypes suggested dependence upon the directional selection pressure applied for realizing maximum yield in different regions; the nicely evolved homeostatic devices would favor constancy of the associated characters would thus indiscriminate clustering. This would be suggested that it was not necessary to choose diverse parents for diverse geographic regions for hybridization.

3.5 CONTRIBUTION OF CHARACTERS TOWARDS DIVERGENCE OF THE CULTIVARS

The character contributing maximum to the divergence were given greater emphasis for deciding on the cluster for the purpose of further selection and choice of parents for hybridization [22]. The PCA revealed that in vector I (Z1) the important characters responsible for genetic divergence in the major axis of differentiation were plant height, panicle length, filled grains per panicle, unfilled grains per panicle, days to maturity and yield per plant (Table 5). In vector II (Z2) that was the second axis of differentiation, plant height, total tillers per plant, effective tillers per plant, days to 50% flowering, days to maturity and yield per plant were important. The role of plant height, days to maturity and yield per plant in both the vectors were positive across two axes indicating the important components of genetic divergence in those materials.

3.6 COMPARISON OF DIFFERENT MULTIVARIATE TECHNIQUES

The clustering pattern of D^2 analysis through non-hierarchical clustering had taken care of simultaneous variation in all the characters under study. However, the distribution of genotypes in different clusters of the D^2 analysis had followed more or less similar trend of the Z1 and Z2 vector of the principal component analysis. The D^2 and principal component analysis was found to be alternative methods in giving the information regarding the clustering pattern of genotypes. However, the principal component analysis provided the information regarding the contribution of characters towards divergence of rice.

Table 5. Relative contributions of the ten characters of 42 rice genotypes to the total divergence

Characters	Vector-1	Vector-2
Plant height (cm)	0.09867	0.02898
Total tillers per plant	-0.01777	0.20832
Effective tillers per plant	-0.00486	0.21611
Days to 50% flowering	-0.02063	0.11111
Panicle length (cm)	0.03660	-0.02108
Filled grains per panicle	0.98511	-0.02257
Unfilled grains per panicle	0.10797	-0.37279
Days to maturity	0.00299	0.10897
Thousand grains weight (g)	-0.00053	-0.02131
Yield per plant (g)	0.07778	0.86282

3.7 SELECTION OF CULTIVARS FOR FUTURE HYBRIDIZATION

Genotypically distant parents were able to produce higher heterosis [23], [24], [25], [26], [27], [28], [29]. Beside this, reference [27] reported in groundnut that the higher heterosis for yield and its components could be obtained from the crosses between the intermediate divergent parents than extreme ones. Reference [29] also reported the same in chick pea that medium divergent genotypes showed higher heterosis in crosses for different yield contributing characters. Reference [30] reported in triticale that very high or very low parental divergent failed result in heterosis.

Considering this idea and other agronomic performances, the genotypes Special from AL-29, AL-36, PP-4B(i) from cluster III; AL-17(iii)B, AL-17(iii), AL-17(ii)A, Special from-129, Special from 17(iv), AL-44(i) and AL-17 from cluster IV; Special from AL-36(D), PP-48, IR-25B, Special from AL-33 from cluster V; IR-25B (Tall) and P-5B(ii) from cluster II might be considered better parents for efficient hybridization programme.

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