Study of genetic divergence in rice (*Oryza sativa* L.) genotypes with high grain zinc using Mahalanobis’ D² analysis

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**Abstract**

The present investigation was conducted with twenty-two genotypes of rice during Kharif, 2018 in RBD with three replications in five different locations. The 22 rice genotypes were characterized based on 16 quantitative traits viz., days to first flowering, days to 50 per cent flowering, days to maturity, the total number of effective tillers per plant, plant height (cm), panicle length (cm), the number of spikelets per panicle, the number of grains per panicle, spikelet fertility percentage, grain weight per panicle (g), grain yield per plant (g), 1000-grain weight (g), grain yield per plot (kg), grain yield per hectare (kg), grain L/B Ratio, grain zinc content (ppm or mg /kg) using Mahalanobis D² statistic. D² analysis distributed the 22 genotypes into six clusters, of which cluster I was the largest with 15 genotypes. Cluster II had maximum intra-cluster values of 5.16 and the maximum inter-cluster distance was observed between the clusters III and V (40.51) followed by cluster IV and V (28.39) indicating the importance of the genotypes present in these clusters for exploiting heterosis for the desirable traits of these clusters.

**Key words**

Genetic divergence, high grain zinc, Mahalanobis D² rice.

**INTRODUCTION**

The cultivated rice plant, *Oryza sativa* L., is an annual self-pollinated short-day monocotyledous grass of family *Poaceae* (Graminae) with chromosome number 2n=2x=24. It is a warm season crop originated from South East Asia, grown extensively in humid tropical and sub tropical regions of the world. Rice is the most widely consumed staple food of more than half of the world’s population covering about 9% of the world’s arable land under its cultivation. Rice, maize and wheat account for about 49 % calories consumed by human population, where 23% is provided by rice. Therefore, rice provides almost one fourth of the calories consumed globally (Subudhi et al., 2006). Rice endosperm is an excellent source of carbohydrates and is also a good source of digestible protein but unfortunately, it is a poor source of micronutrients such as iron, zinc and vitamin A which are an essential part of balanced nutrition. The estimated number of under nourished people in the world has reached 82.1 crores (10.8 % of world’s population) and in India, 19.59 crores people (about 14.8 % of country’s population) are under nourished. Malnutrition is considered as the most common cause of zinc deficiency and its deficiency may lead to health problems including dwarfism, anorexia, weak immunity, skin lesions, diarrhea, and hypogonadism. The biofortification of rice grain by developing varieties/hybrids with high zinc levels in polished rice could be a good, cost-effective and sustainable solution for this problem.

Genetic variability exists among rice germplasm leaving a wide scope for crop improvement. Information on the genetic diversity within and among closely related
crop varieties is essential for a rational use of genetic resources. It contributes to monitoring germplasm and can also be used to predict potential genetic gains. Similarly, quantification of genetic diversity existing within and between groups of germplasm is important and the crosses between parents with maximum genetic divergence are generally the most responsive for genetic improvement (Arunachalam, 1981) for realizing higher heterosis and obtaining superior recombinants. In view of the above background, an investigation was carried out with 22 rice genotypes with high grain zinc, to study genetic divergence of sixteen yields and yield traits using Mahalanobis $D^2$ analysis.

**MATERIALS AND METHODS**

The experiment was carried out during Kharif, 2018 with 22 rice genotypes at five different locations in a Randomized Block Design (RBD) with three replications. In all the locations, a single plot consisted of three rows of 3 meters each, with 20 cm row to row and 15 cm intra-row spacing. Net plot size was 0.6 x 1 meters, i.e., 1.8 $m^2$ for all the locations. Data was recorded on 16 quantitative characters viz., days to first flowering, days to 50% flowering, days to maturity, the total number of effective tillers per plant, plant height (cm), panicle length (cm), the number of spikelets per panicle, the number of grains per panicle, spikelet fertility percentage, grain weight per panicle (g), grain yield per plant (g), 1000-grain weight (g), grain yield per plot (kg), 1000-grain weight (g), grain yield per plot (kg), grain yield per hectare (kg), grain L/B ratio, grain zinc content (ppm or mg/kg). Observations were recorded from three randomly selected competitive plants of each genotype in each replication for most of the yield and related traits whereas, days to first flowering, days to 50% flowering and days to maturity were recorded on plot basis. Grain yield per plot and grain yield per hectare were also recorded on plot basis.

The mean values of all recorded attributes were compiled from all genotypes in all replications and Mahalanobis $D^2$ analysis was performed for analysis of twenty-two genotypes for all 16 characters. Tocher’s optimization method, described by Rao (1952) was used for placing genotypes into various clusters. The $D^2$ values of all the combinations were sorted in ascending order in the form of a table as outlined by Singh and Choudhary (1977). Ranking was done for each character based on their contribution towards divergence between two entries. Each of the characters were ranked for all combination of entries from rank ‘1’ (character with the highest mean difference) to rank ‘p’ (character with the lowest mean difference).

**RESULTS AND DISCUSSION**

Using the pivotal condensation method, the mean values of genotypes were transformed into standardized uncorrelated mean values. The relative contribution of different characters included in the study towards diversity is presented in **Table 1**. Days to 50% flowering contributed maximum (34.20 %) towards diversity, followed by grain yield per hectare (30.30 %), grain L/B ratio (23.38 %) and plant height (5.19 %). Thousand grain weight, days to maturity, grain yield per plot, grains per panicle and grain weight per plant showed 3.90 %, 1.30 %, 0.87 %, 0.43 %, and 0.43 % respectively. Some characters viz., days to first flowering, tillers per plant, panicle length, spikelets per panicle, spikelet fertility percentage, grain weight per panicle and grain zinc content showed no contribution towards diversity. Similar results were obtained by Beevi and Venkatesan 2015, Raghavendra et al, 2018 and Devi et al., 2019.

| Character                          | Times ranked 1st | Contribution % |
|-----------------------------------|------------------|----------------|
| Days to first flowering           | 0                | 0.00%          |
| Days to 50% Flowering             | 79               | 34.20%         |
| Days to maturity                  | 3                | 1.30%          |
| Tillers per plant                 | 0                | 0.00%          |
| Plant height (cm)                 | 12               | 5.19%          |
| Panicle length (cm)               | 0                | 0.00%          |
| Spikelets per panicle (Total)     | 0                | 0.00%          |
| Grains per panicle                | 1                | 0.43%          |
| Spikelet fertility %              | 0                | 0.00%          |
| Grain wt. per panicle(g)          | 0                | 0.00%          |
| Grain yield per plant(g)          | 1                | 0.43%          |
| 1000 grain Wt.(g)                 | 9                | 3.90%          |
| Grain yield per plot(kg)          | 2                | 0.87%          |
| Grain yield per hectare (kg)      | 70               | 30.30%         |
| Grain L/B Ratio                   | 54               | 23.38%         |
| Grain zinc content(ppm)           | 0                | 0.00%          |

Table 1. Relative contribution of 16 characters to genetic diversity in 22 genotypes of rice over five different environments

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368
Table 2. Cluster composition of 22 rice genotypes over five different environments

| Cluster | Number of genotypes | Name of the genotypes |
|---------|---------------------|------------------------|
| I       | 15                  | IR 95133:1-B-16-14-10-GBS-P1-2-2, IR 95133:1-B-16-14-10-GBS-P1-2-3, IR95133:1-B-16-14-10 GBS-P6-1-5, IR 95133:1-B-16-14-10-GBS-P5-1-3, IR95133:1-B-16-14-10-GBS-P5-2-3, IR15M1537, HUR-3022 (Local Check-2), IR-64, MTU1010, IR15M1689, IR15M1633, HURZ1, IR15M1546, DRR Dhan 48, IR99642-57-1-1-1-B |
| II      | 3                   | HURZ-3, BRRIDhan 64, BRRIDhan 72 |
| III     | 1                   | DRR Dhan 45 |
| IV      | 1                   | HUR-105 (Local Check-1) |
| V       | 1                   | Swarna |
| VI      | 1                   | Sambamashuri |

Fig. 1. Intra & inter cluster distances between genotypes by Tocher Method

Table 3. Inter-cluster and Intra-cluster (diagonal) $D^2$ values of 22 rice genotypes over five different environments

| Clusters     | Cluster 1 | Cluster 2 | Cluster 3 | Cluster 4 | Cluster 5 | Cluster 6 |
|--------------|-----------|-----------|-----------|-----------|-----------|-----------|
| Cluster 1    | 3.49      | 10.67     | 7.89      | 12.82     | 27.75     | 20.65     |
| Cluster 2    | 5.16      | 21.46     | 9.48      | 22.62     | 12.5      | 17.45     |
| Cluster 3    | 0         | 5.12      | 21.75     | 12.5      | 17.45     | 10.87     |
| Cluster 4    | 0         | 0         | 27.75     | 17.45     | 10.87     | 7.65      |
| Cluster 5    | 0         | 0         | 0         | 9.48      | 7.65      | 0         |
| Cluster 6    | 0         | 0         | 0         | 0         | 0         | 0         |
The minimum inter-cluster distance was recorded between cluster III and IV (5.12), followed by cluster V and VI (7.65), and cluster I and III (7.89) suggesting closeness and similarities among the genotypes in these clusters for most of the traits. The maximum inter-cluster distance was observed between cluster III and V (40.51) followed by cluster IV and V (28.39), cluster I and V (27.75) and cluster II and IV (22.62) which indicated the existence of high genetic diversity among genotypes in these clusters, and therefore crosses between the genotypes of these clusters could yield desirable transgressive segregants. Similar explanation was given by Ranjith et al., 2018, Amegan et al., 2020 and Sudeepthi et al., 2020.

Cluster means of all the characters is presented in Table 4. The cluster mean values varied in all the clusters for all the 16 quantitative traits studied. Cluster III recorded the highest mean values for most number of traits viz., plant height (120.74), Grains per panicle (164.45), spikelet fertility % (87.47), grain yield per plant (37.33) and grain zinc content (24.22). Further, the clusters II, IV and VI recorded the highest mean values for 3 traits each. The results indicated the existence of high genetic diversity among genotypes in these clusters, therefore crosses between the genotypes of these clusters could yield desirable transgressive segregants. Similar explanation was given by Muthuramu and Sakthivel 2017 and Sudeepthi et al., 2020.

Based on D² values, twenty two genotypes were grouped into six clusters using Tocher method (Singh and Choudhary, 1977). Clusters with their genotypes are presented in Table 2. Cluster I had fifteen genotypes, Cluster II had three genotypes, whereas Cluster III, IV, V, and VI had single genotype in each cluster.

Range of an average intra-cluster D² values was 0 to 5.16 (Table 3). Cluster II had a maximum intra-cluster value (5.16), while cluster I showed a minimum intra-cluster distance (3.49). Cluster III, IV, V, and VI had zero intra-cluster distance, as they were having single genotype each. High intra-cluster distance in Cluster II suggests wide genetic diversity among the genotypes in this cluster.

The minimum inter-cluster distance was recorded between cluster III and IV (5.12), followed by cluster V and VI (7.65), and cluster I and III (7.89) suggesting closeness and similarities among the genotypes in these clusters for most of the traits. The maximum inter-cluster distance was observed between cluster III and V (40.51) followed by cluster IV and V (28.39), cluster I and V (27.75) and cluster II and IV (22.62) which indicated the existence of high genetic diversity among genotypes in these clusters, and therefore crosses between the genotypes of these clusters could yield desirable transgressive segregants. Similar explanation was given by Ranjith et al., 2018, Amegan et al., 2020 and Sudeepthi et al., 2020.

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Mahalanobis D² statistic has been used by many researchers for multivariate analysis such as degree of divergence studies in crop germplasm collections. Kandhola and Panwar (1999), Singh et al. (2006), Kuchanur et al. (2009), Vennela et al. (2017), Bhati et al. (2015) and Behera et al. (2018) has reported the success of this model for studying genetic divergence in rice genotypes.
The genotypes belonging to cluster III and V showed a maximum divergence followed by cluster IV and V, therefore, they can be used for making crosses. Considering per se yield and yield contributing characters along with divergence, genotypes, viz., DRR Dhan 45, DRR Dhan 48, HUR-105 (Local check1) and Swarna can be best utilized in breeding programmes.

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Fig. 2. Dendrogram showing relationship among 22 rice (*Oryza sativa* L.) genotypes in six clusters based on Mahalanobis’ $D^2$ values
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