Assessment of Genetic Diversity in New Restorer Lines of Hybrid Rice

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A population panel comprising 126 newly developed restorer lines that can be utilized in hybrid rice breeding in future were evaluated for 16 agro-morphological and grain quality traits by principal component analysis for determining the pattern of genetic diversity and relationship among individuals. Agro- morphological traits included days to fifty per cent flowering, plant height, productive tillers, basal internode thickness, culm strength, flag leaf length, panicle length, number of filled grains per panicle, panicle exsertion, panicle secondary branching, spikelet fertility, 100 seed weight, single plant yield and grain quality traits were kernel length, kernel breadth and kernel LB ratio. The cumulative variance of 70.20% was explained by six principal components. Component 1 had the contribution from the traits *viz.,* productive tillers, culm strength, spikelet fertility, kernel length and panicle secondary branching which accounted 15.79% of the total variability. The remaining variability of 14.81%, 12.10%, 11.75%, 9.44% and 6.31% was consolidated in component 2, component 3, component 4, component 5 and component 6 respectively. The grain quality traits *viz.,* kernel length, kernel breadth and kernel LB ratio were loaded as major traits in PC6, PC5 and PC4 respectively. The trait single plant yield was the predominant trait in component 6. The results of present study identified the key economic traits, which accounts for maximum variation in the restorer lines under study which will be greatly beneficial to exploit in the hybrid rice breeding programmes.

Introduction

Rice (*Oryza sativa* L.) is a staple food crop that provides the primary source of calories for more than 50 per cent of the world's population. Over 90 per cent of the world's rice is produced and consumed in the Asian Region with six countries (China, India, Indonesia, Bangladesh, Vietnam and Japan) that account for 80% in the world's production and consumption (Abdullah, 2006). The United Nations estimates predict that, global population will increase 33% by 2050, from 7.2 billion today to 9.6 billion persons. To feed the ever-increasing human population, yield potential of rice should also be improved through evolution of high yielding varieties and hybrids which could be realized by

exploiting the genetically diverse breeding materials.

In this context, assessment of genetic diversity plays a definitive role to select genotypes that can be used as parents in three line hybrid breeding programme inclusive of the presence of fertility restorer genes for WA- CMS currently exploited. A set of new parental lines were developed through recombination breeding and selection from restorer x restorer crosses of TNAU and IRRI, restorer x *tropical japonica* or vice-versa from TNAU. The presence of fertility restorer genes either *Rf3* or *Rf4* or both for Wild Abortive –CMS was confirmed by molecular marker analysis. Thus the objective of the present study was to ascertain the genetic diversity through principal component analysis for agronomic and grain quality traits in the stabilized breeding lines to exploit them in hybrid rice breeding to realize the genetic gains.

Materials and Methods

In the present study, 126 restorer lines of rice were raised in augmented design with three checks *viz.,* CO (R) 50, CO 51, CO 52 during *kharif* 2018 at Department of Rice, Centre for Genetics and Plant Breeding, Tamil Nadu Agricultural University, Coimbatore. Each entry was planted at a spacing of 20 cm x 15 cm and the recommended agronomic practices were followed. Observations on 13 agronomic and yield contributing traits *viz.,* days to fifty per cent flowering, plant height (cm), productive tillers (no.), basal internode thickness (mm), culm strength (g/stem), flag leaf length (cm), panicle length (cm), number of filled grains per panicle, panicle exsertion, panicle secondary branching (no.), spikelet fertility (%), 100 seed weight (g), single plant yield (g) and three grain quality traits *viz.,* kernel length (mm), kernel breadth (mm) and L/B ratio were recorded in randomly selected five plants (SES, IRRI, 2013) except days to

50% flowering. The culm strength was measured and calculated as quoted by Hai *et al.,* (2005). Principal component analysis was performed by using STAR (Statistical Tools for Agricultural Research) software. Eigen vectors were calculated by using the following formula-

$$Percentage\ of\ variance\ explained\ for\ PC1 = \frac{Eigen\ value\ of\ PC1}{Sum\ of\ eigen\ values}$$

The cumulative percentage of contribution to variation of different traits to eigen vectors were also computed.

Results and Discussion

Principle component analysis is one of the important tools of genetic diversity analysis. It is a multivariate statistical technique employed to identify the minimum number of components that explain maximum variability out of the total variability present in the breeding material analyzed. It is also used to rank the genotypes based on PC scores (Anderson, 1972 and Morrison, 1967).

Thus, the technique is very helpful for identification of plant characters that are categorized for their distinctiveness among promising genotypes. Eigen values measure the importance and contribution of each component to total variance whereas each coefficient of eigen vectors indicates the degree of contribution of every original variable with which each principal component is associated. The higher the coefficients, regardless of the sign, the more effective will be the discriminating power between accessions (Clifford and Stephenson, 1975). According to Kaiser (1960), each principal component with Eigen value greater than one could be considered to contribute at least 10% of the phenotypic variation. The higher Eigen values were considered as best representative of system attributes in principal components.

In our present study, the results of PCA explained the genetic variation among the genotypes for all agro morphological and quality characters under study. The analysis revealed six components with Eigen value more than one and explained a cumulative variation of 70.20%. The PC 1 which showed 15.79%, the highest variability among traits had an Eigen value of 2.52 (Table 1). Scree plot explained the percentage of variation associated with each principal component obtained by drawing a graph between Eigen values and principal component numbers (Figure 1). The Eigen values gradually declined. Elbow type line was shifted to straight line with minute difference with each PC value after principal component 6. Accordingly, PC2, PC3, PC4, PC5 and PC6 exhibited 14.81%, 12.10%, 11.75%, 9.44% and 6.31% variability respectively. In previous investigations on principal component analysis in rice germplasm, formation of six principal components as encountered in our present study was also reported by Ojha *et al.,* (2017), Pathak *et al.,* (2018) and Suneetha *et al.,* (2018) in 207, 98 and 95 genotypes for 17, 13 and 20 traits respectively. However with as few as 24 rice varieties, Kunusoth *et al.,* (2015) obtained seven components for 24 traits. Hasan *et al.,* (2012) performed D^2 and PCA analysis in 30 restorer and 10 maintainer lines of rice in which two principal components were derived. In 100 restorer and maintainer lines, 10 clusters were formed in cluster analysis by coefficient of parentage (Wang *et al.,* 2012) of which, the restorer lines further divided into 11 sub clusters.

The results on Eigen vectors of 16 agro-morphological traits are presented in Table 2 and Figure 2. Rotated component matrix revealed that PC1 was highly loaded with characters such as productive tillers (0.469), culm strength (-0.430), spikelet fertility (0.325), kernel length (-0.303) and panicle secondary branching (-0.259). The component

PC2 accounted for 14.81% variation which was contributed by characters *viz.,* plant height (-0.379), culm strength (-0.372), productive tillers (0.339), panicle length (-0.328) and flag leaf length (-0.302).

The traits like single plant yield (0.409), panicle length (-0.398), spikelet fertility (0.388), days to 50% flowering (-0.377), flag leaf length (-0.367), plant height (-0.344) in PC3 contributed for 12.10 % variation. The fourth principal component was loaded by major traits like kernel length breadth ratio (-0.604), kernel length (-0.398), kernel breadth (0.383), number of filled grains per panicle (-0.376), 100 seed weight (-0.293) and panicle exsertion (-0.269) that accounted for 11.75% of the variation. The variation explained by both PC3 and PC4 was almost equal.

It was observed that the characters *viz.,* kernel breadth (0.433), panicle exsertion (0.416), days to 50% flowering (-0.323), kernel length breadth ratio (-0.314), basal internode thickness (-0.287), panicle secondary branching (-0.274) contributed to the 9.4% of variation in PC5. The least variation of 6.31% accounted in PC6 were contributed by the traits kernel breadth (-0.505), 100 seed weight (0.434), days to 50% flowering (0.304), plant height (0.292), number of filled grains per panicle (0.298), kernel breadth (-0.283), basal internode thickness (0.259) and panicle secondary branching (-0.240).

In the studies of Sathish *et al.,* (2017) and Gour *et al.,* (2017), the number of productive tillers was loaded in PC1 as realized in our analysis. So also, similar type of loading in PC1 for kernel length was reported by Mengli *et.al* (2017). The traits *viz.,* basal internode thickness and leaf length loaded in PC5 (9.4% variability) and PC3 (12.1% variability) respectively in the present investigation was realized by Kunusoth *et al.,* (2015) in PC1explaining 18.32% of the variability.

Table.1 Eigen value and contribution of variability in 126 rice restorer lines

S.No.	Principal components (PCs)	1	2	3	4	5	6
1.	Eigen value	2.526	2.370	1.935	1.880	1.510	1.009
2.	Variability (%)	15.79	14.81	12.10	11.75	9.44	6.31
3.	Cumulative%	15.79	30.60	42.70	54.45	63.89	70.20

Table.2 Eigen vectors of 16 agro-morphological traits in rice restorer lines

S.No.	Traits	Eigen vectors					
1.	Days to 50% flowering	-0.087	0.065	-0.377	0.194	-0.324	0.305
2.	Plant height	0.149	-0.380	-0.345	-0.049	0.001	0.293
3.	Productive tillers	0.470	0.340	-0.065	-0.016	0.122	-0.027
4.	Basal internode thickness	-0.209	-0.029	0.021	-0.214	0.288	0.259
5.	Flag leaf length	0.090	-0.303	-0.367	-0.067	-0.035	-0.274
6.	Panicle length	0.218	-0.329	-0.399	-0.111	0.126	-0.174
7.	Number of filled grains per panicle	0.226	0.099	0.004	-0.376	-0.169	0.298
8.	Panicle exsertion	0.177	-0.039	-0.209	-0.269	0.416	-0.034
9.	Panicle secondary branching	-0.260	-0.009	-0.076	0.202	-0.274	-0.240
10.	Culm strength	-0.434	-0.372	-0.043	-0.006	-0.147	0.107
11.	Kernel length	-0.304	-0.024	0.073	-0.399	0.074	-0.505
12.	Kernel breadth	-0.148	-0.260	0.158	0.383	0.433	-0.283
13.	Length breadth ratio	-0.095	-0.260	-0.090	-0.604	-0.314	-0.098
14.	Spikelet fertility	0.326	-0.263	0.389	-0.119	-0.167	-0.025
15.	100 seed weight	-0.189	-0.169	0.150	-0.294	0.186	0.434
16.	Single plant yield	0.216	-0.327	0.410	-0.113	-0.124	0.125

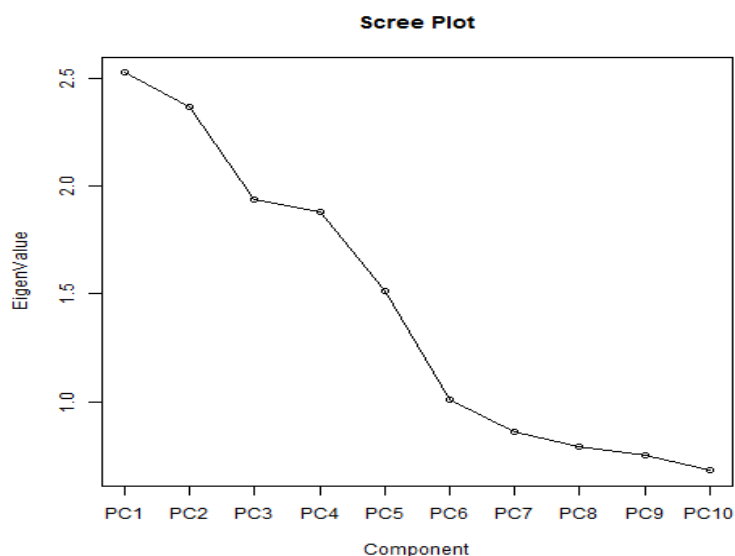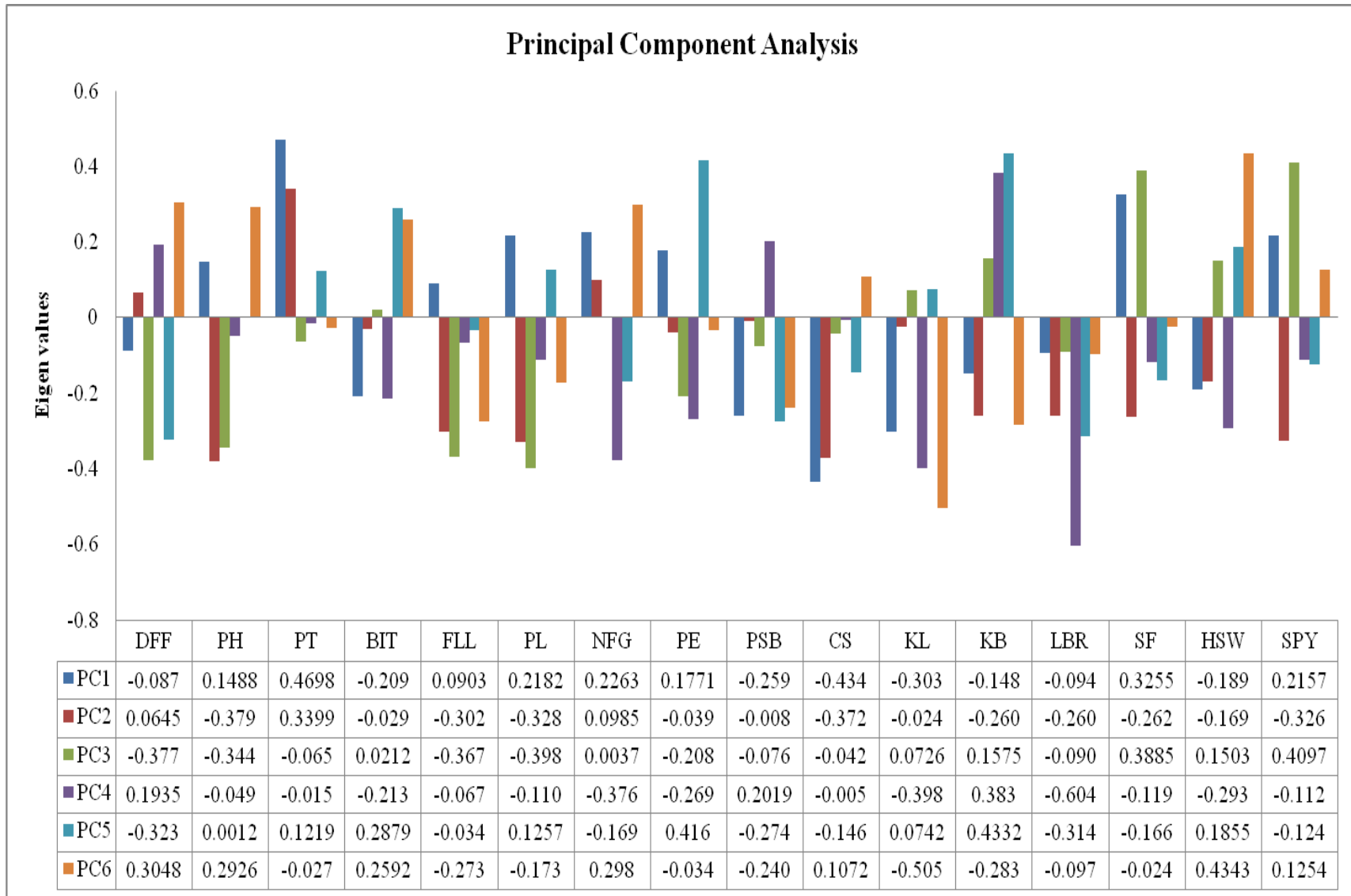**Fig.1** Scree plot showing Eigen values and percentage of cumulative variability

Scree Plot

Fig.2 Eigen vectors of 16 agro-morphological traits in rice restorer lines

	DFF	PH	PT	BIT	FLL	PL	NFG	PE	PSB	CS	KL	KB	LBR	SF	HSW	SPY
■PC1	-0.087	0.1488	0.4698	-0.209	0.0903	0.2182	0.2263	0.1771	-0.259	-0.434	-0.303	-0.148	-0.094	0.3255	-0.189	0.2157
■PC2	0.0645	-0.379	0.3399	-0.029	-0.302	-0.328	0.0985	-0.039	-0.008	-0.372	-0.024	-0.260	-0.260	-0.262	-0.169	-0.326
■PC3	-0.377	-0.344	-0.065	0.0212	-0.367	-0.398	0.0037	-0.208	-0.076	-0.042	0.0726	0.1575	-0.090	0.3885	0.1503	0.4097
■PC4	0.1935	-0.049	-0.015	-0.213	-0.067	-0.110	-0.376	-0.269	0.2019	-0.005	-0.398	0.383	-0.604	-0.119	-0.293	-0.112
■PC5	-0.323	0.0012	0.1219	0.2879	-0.034	0.1257	-0.169	0.416	-0.274	-0.146	0.0742	0.4332	-0.314	-0.166	0.1855	-0.124
■PC6	0.3048	0.2926	-0.027	0.2592	-0.273	-0.173	0.298	-0.034	-0.240	0.1072	-0.505	-0.283	-0.097	-0.024	0.4343	0.1254

Gana *et al.,* (2013) and Nachimuthu *et al.,* (2014) also published that leaf length was loaded in PC1 explaining 19.1% and 28.46% variability respectively.

For the trait 100 grain weight, Gana *et al.,* (2013), Mengli *et al.,* (2017) and Suneetha *et al.,* (2018) reported its loading in PC1 unlike our study in which it was loaded in PC6. Also, kernel breadth that fell in PC5 was reported in PC1 by Sathish *et al.,* (2017).

In contrast to the equal loading of days to 50% flowering in PC 5 and 6 in our studies, it occupied the first principal component in the publications by Sanni *et al.,* (2012), Nachimuthu *et al.,* (2014), Mahendran *et al.,* (2015), Pachauri *et al.,* (2017) and Suneetha *et al.,* (2018).

For the traits plant height and panicle length that branched off in PC2 and PC 3 was reported by Nachimuthu *et al.,* (2014) and Pachauri *et al.,* (2017) in first component.

Many workers reported number of filled grains in first component unlike fourth component in this investigation.

The ultimate trait single plant yield loaded in PC3 was detected in first component in the experiments of Gour *et al.,* (2017) and Pachauri *et al.,* (2017).

The total variation of 70.20% observed in the present study is in accordance with the reports of Sanni *et al.,* (2012), Gana *et al.,* (2013), Ojha *et al.,* (2017), Pachauri *et al.,* (2017) and Suneetha *et al.,* (2018) where the variation ranged from 68.86 to 72.48%.

In exceptional cases, high total variation of 92.75% and 80.56% were realized by Gour *et al.,* (2017) and Nachimuthu *et al.,* (2014) in their PCA analysis with 33 and 12 traits respectively in 83 and 192 germplasm lines.

In this study, phenotypic traits could be identified which are responsible for the observed genotypic variation present within each component besides traits with maximum variability thus highlighting adequate variability among the restorer lines. These lines with fertility restorer genes for WA- CMS could be utilized to strike at heterotic hybrid combinations in future to realize the yield gains.

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