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Principal component analysis of morpho-physiological traits in mutants lines of rice under submerged condition

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Abstract

Present study was undertaken to identify the principle components for the assessment of genetic diversity and extent and pattern of genetic divergence based on K-mean determination in mutant lines of rice under submerged condition. In this experiment 240 mutant lines of rice along with 3 checks (FR13A, FR13B and Labella) are evaluated in type 2 modified augmented design (MAD2) during Kharif-2017 at rice experimental area of Dr. RPCAU, Pusa, Bihar. Principle component analysis (PCA) showed that first four principle components had >1.00 Eigen value and accounted 80.24% of total variation. Rotated component matrix revealed that each principle component is separately loaded with various submergence tolerant related traits. PC1 was constituted by Tolerance score (0.41). PC2 was maximum correlated with total shoot elongation (0.59) followed by relative shoot elongation (0.48), panicle length (0.43) and plant height (0.42) while, PC₃ was maximum correlated with panicle length (0.52) followed by plant height (0.51). Therefore, intensive selection is recommended to bring about rapid improvement of submergence tolerance by selecting lines from PC₁, PC₂ and PC₃. All the 243 lines (including checks) were grouped into 16 clusters. Cluster VIII and X have higher mean value for maximum no. of traits indicating mutant lines from this trait can be used for further improvement. 3-D plot based on top three principle components indicated that Labella (200GY) entry no. 146, Labella (200 GY) entry no. 147, FR13B (350GY) entry no. 165, FR13B (200GY) entry no. 222 were found most divergent mutant lines with FR13B (200GY) entry no. 224, Labella (300GY) entry no. 102 and FR13B (200GY) entry no. 225 which can be utilized effectively in breeding programme for improvement of submergence tolerance.

Keywords: principal component analysis, genetic diversity, mutant rice, submergence tolerance

1. Introduction

Rice (Oryza sativa L.) is one of the major and staple food for more than half of the world's population. It belongs to genus Oryza, family Gramineae and sub family Oryzoidea. Approximately 90% of the world's rice is grown and consumed in Asia, whereas 50% of the population depends on rice for food (Tenorio et al., 2013). Rice provides nutrition for more people in the world than other crops, especially in developing countries (Phillips et al., 2005). The world production of rice is nearly 487.46 million tonnes being grown in an area of over 161.10 million hectares with an average productivity of 3.02 tonnes per hectare (Statista, 2017-18). In India, rice is grown over estimated area of about 44.10 million hectares with the production of 110.15 million tonnes with its productivity being 2.49 tonnes per hectare (INDIA STAT-Advance Estimate, 2017-18). It accounts for about 43% of total food grain and 55% of cereals production in country. In Bihar, the total area under rice cultivation is nearly 3.23 million hectares with production of 6.80 million tonnes and its productivity is 2.10 tonnes per hectare (Directorate of Economics and Statistics, Govt. of Bihar, 2017-18). Flooding is a serious, naturally occurring problem for rice production in the rainfed lowlands of south and south-east Asia. Rainfed lowland ecology is always prone to flash floods during rainy season. Rice is the only crop suitable for cultivation in the rainfed lowlands stress adversely affected by submergence stress. Submergence stress has been identified as the third most important constraint for higher rice productivity (Sarkar et al., 2006) as it adversely affects poor farmers living on 15 million ha of rice growing areas in the rainfed lowlands in south and south-east Asia. Bihar is the India's most flood-prone state, with 76 percent of the population, in the north Bihar living under the recurring threat of flood devastation. About 68800 Sq. Km out of total geographical area of 94160 Sq. Km is flood affected. Submergence tolerance is defined as the ability of a rice plant to survive and continue growing after being completely submerged in

water for several days. Rice plants are less tolerant to submergence at the early growth stages (Adkin et al., 1990). Genetic diversity is pre-requisite for any crop improvement programme, as it helps in the development of superior recombinant (Manonmani & Fazlullah Khan., 2003). Genetic diversity represents the heritable variation within and between populations. Information on the genetic diversity and distance among the breeding lines and the correlation between genetic distances are important for determining breeding strategies, classifying the parental lines, defining heterotic groups, and predicting future hybrid performance (Acquaah G. 2012). Genetic distance estimates for population grouping can be estimated by different methods as it is crucial to understand the usable variability existing in the population panel (Nachimuthu et al. 2014). One of the approaches is to apply multivariate analysis. Principal Component Analysis (PCA) is a powerful tool in modern data analysis because this is a wellknown multivariate statistical technique which is used to identify the minimum number of components, which can explain maximum variability out of the total variability (Anderson, 1972 and Morrison, 1978) and also to rank genotypes on the basis of PC scores. Principal components are generally estimated either from correlation matrix or covariance matrix. Considering the importance of PCA this study is conducted on rice mutant lines with an objective to identification of the Morpho-physiological traits responsible for the yield differences among the rice genotypes under submerged conditions.

2. Materials and Methods

The present investigation was carried out with 240 mutant lines along with 3 untreated rice varieties as checks (FR13A, FR13B and Labella) at rice experimental area of Dr. Rajendra Prasad Central Agricultural University, Pusa, Bihar, during Kharif-2017. Field experiment was performed in type 2 modified augmented design (MAD2) for one year. The entire tank area was divided into 10 blocks. Each block accommodated 24 lines and 3 checks. The checks were randomized and replicated as per procedure described by Frank M. You et al., 2016. Each mutant lines was sown in a plot of 1m² in 5 rows with row to row distance of 20cm and plant to plant distance of 15cm. recommended dose of fertilizer was applied at the time of crop period. Three random plants were tagged from each block to record the data for yield and its related traits except for days to 50% flowering and survival percentage. Days to 50% flowering was recorded on plot basis. Submergence tolerance related traits were performed as per standard evaluation system of rice Toojinda et al., 2003 as listed below:

Survival percentage= Tota	$\frac{\text{Total number of suvived seedlings after desubmergence}}{\text{Total number of seedling before submergence}} \times 100$						
Total shoot elongation (T	SE) = after de-submergence	Height of shoot – before de-submergence					
Relative shoot elongation (RS	$SE) = \frac{Elongation growth unde}{Elongation growth under nons}$	$\frac{r \text{ submergence}}{r \text{ submerged condition}} \times 100$					

Tolerance score (TS) = Scored on the basis of survival percentage (SP).

These data were subjected to pooled analysis for genetic divergence by using statistical package WINDOSTAT version 9.2 (INDOSTAT service, Hyderabad).

3. Results and Discussion

The genetic variation present in breeding population was divided into five principal components (PCs), which explained 88.21% of total variation (Table 1). As per the criteria set by Brejda et al., 2000, the PCA with eigen values >1 and which explained at least 5% of the variation in the data were considered. The first four principle components had >1.00 eigen value and accounted to 80.24% of total variation. The first principal component (PC₁) explained 37.14% of the total variation. The second principal component (PC2) explained 20.89% variation individually and 58.03% cumulative variation. The third principal component (PC₃) explained 12.93% variation individually and 70.96% cumulatively. The fourth principal component (PC₄) explained 9.28% variation individually and 80.24% cumulatively. Rotated component matrix revealed that each principle component is separately loaded with various submergence tolerant related traits (Table 2). PC₁ was mainly correlated with Tolerance score (0.41). PC₂ was maximum correlated with total shoot elongation (0.59) followed by relative shoot elongation (0.48), panicle length (0.43) and plant height (0.42). In case of PC_4 and PC_5 the maximum correlation of 0.70 and 0.67 respectively was exhibited by days to 50 per cent flowering. Therefore, intensive selection procedures can be designed to bring about rapid improvement of above mentioned traits under submerged conditions. Similar type of work was also reported by Bisne and Sarawgi, 2008, Sanni et. al., 2012, Sarawgi et. al 2013, Kumar et. al., 2016 and Sahu et. al., 2017. It hasbeen observed that higher loading values are attributed to submergence tolerance related traits (Table 3), indicating importance of these traits in getting better recombinants for submergence tolerance. The top five mutant lines (Table 3) were ranked for each principal component (PC Score). FR13B (200GY+0.2%EMS)-entry no. 240 had highest PC

FK13B (200GY+0.2%EMS)-entry no. 240 had highest PC score followed by FR13B (200GY+0.2%EMS)-entry no. 239, Labella (250GY)-entry no. 124, FR13B (200GY+0.2%EMS)entry no. 238 and Labella (350GY)-entry no. 85 in PC2 indicating that they can be used for plant height, panicle length, total shoot elongation and relative shoot elongation. Labella (350GY+0.2%EMS)-entry no. 92 showed high PC score in PC₃ followed by Labella (350GY+0.2%EMS)-entry no. 91, Labella (300GY+0.2%EMS)-entry no. 116, Labella (300GY)-entry no. 109 and Labella (200GY)-entry no. 149 for plant height and panicle length. For days to 50% flowering and leaf senescence, FR13A (250GY)-entry no. 45 showed high PC score in PC₅ followed by Labella (350GY+0.2%EMS)-entry no. 81, Labella (200GY+0.2%EMS)-entry no. 151, FR13A (250GY)-entry no. 47 and FR13B (300GY+0.2%EMS)-entry no. 196.

Cluster analysis

Cluster analysis helps in selection of suitable genotype (s) or parent to use in hybridization programme for the manipulation desirable traits. Choice of proper parent (s) plays a vital role for a successful plant breeding programme. Parents having more genetic distance believed to create higher variations by generating higher recombination frequency, which increase the genetic gain in selection. The grouping of the mutant lines was done by K-mean clustering pattern. The distribution of 240 mutant lines along with three checks into sixteen clusters and their cluster means are presented in Table 4 and 5 respectively. Cluster XVI comprised of only one mutant line forming the smallest cluster followed by cluster X which was comprised of only three mutant lines. Among other clusters the cluster III, VI comprised of twenty eight, cluster V, XIV of twenty one, cluster X of twenty, cluster VII of seventeen, cluster VIII of fourteen, cluster IV and XI of eleven, cluster I, IX and XIII of ten and the cluster XV was comprised of nine mutant lines.

The three untreated rice varieties taken as checks were FR13A, FR13B and Labella. The check FR13A was clustered in cluster VII, FR13B in cluster V and labella in cluster VIII. The clustering of checks into particular clusters shows that the present submergence tolerant varieties from same source while diverse mutant lines other than checks have the capacity to tolerate the submergence condition and can be used as recurrent parent.

The k-mean of different clusters indicated that mutant lines of cluster X possess less DFF, RSE, TS but high NTP, NFTP, SP, GYP and LS, Cluster XVI possess high PH, PL, TSE & RSE as compare to FR13B (200GY)-entry no. 224 and FR13B (200GY)-entry no. 225. It indicates that representative mutant lines can be chosen from particular diverse groups based on their cluster mean and can be involved in hybridization programme for rice improvement against submergence tolerance related traits. Based on cluster mean it was foud that Cluster VIII and X have higher mean value for maximum no. of traits indicating the possibility of obtaining transgressive segregants in early segregating and subsequent

generations. Several researchers observed the significance of high survival percentage to improve the submergence duration in lowland rice (Mackill *et. al.*, 2012; Neeraja *et. al.*, 2007; Kumar *et. al.*, 2017)

The character contribution of various clusters towards the genetic diversity by Tochers clustering method indicated that plant height and survival percentage were the major contributors towards total divergence (Table 6).

3-D Plot diagram was constructed on the first three principle components (Fig. 1). Researchers use 3-D plot in principle component analysis to visually assess which components explain most of the variability in the data. In 3-D diagram Labella (200GY)- entry no. 146, Labella (200 GY)-entry no. 147, FR13B (350GY)- entry no. 165, FR13B (200GY)-entry no. 222 were plotted at distant end whereas, FR13B (200GY)entry no. 224, was plotted at another end of 3-D indicating their effectiveness in breeding programme for improvement of submergence tolerance trait as diverse parents. Based on 3-D diagram genotypes Labella (200GY)-entry no. 146, Labella (200 GY)-entry no. 147, FR13B (350GY)- entry no. 165, (350GY+0.2%EMS)-entry no. 176, FR13B FR13B (250GY+0.2%EMS)- entry no. 213 and FR13B (200GY)entry no. 225 also exhibited good divergence with Labella (300GY)-entry no. 102 and FR13B (200GY)-entry no. 225 therefore, they can also be used in hybridization programmes for improvement of submergence tolerance traits.

Table 1: Eigen values and variability explained by each principal components (PCs)

	PC1	PC2	PC3	PC4	PC5
Eigene Value (Root)	4.09	2.30	1.42	1.02	0.88
% Var. Exp.	37.14	20.89	12.93	9.28	7.98
Cum. Var. Exp.	37.14	58.03	70.96	80.24	88.21

Table 2: Factors loadings (Eigen vectors) for the different morph-physiological characters for the principal components

S. No.	Traits	PC1	PC2	PC3	PC4	PC5
1	Days to 50% flowering (Days)	0.05	0.16	0.15	0.70	0.67
2	Plant height (cm)	-0.09	0.42	0.51	-0.04	-0.17
3	Panicle length(cm)	-0.07	0.43	0.52	-0.08	-0.11
4	No.of tillers per plant	-0.45	-0.03	-0.02	0.07	-0.07
5	No. of fertile tillers per plant	-0.45	-0.01	0.01	0.05	-0.09
6	Survival percentage (%)	-0.44	-0.11	0.00	0.05	0.07
7	Total shoot elongation(cm)	-0.07	0.59	-0.35	0.01	-0.07
8	Relative shoot elongation (cm)	-0.04	0.48	-0.56	0.05	-0.01
9	Tolerance score	0.41	0.10	0.03	-0.05	-0.09
10	Grain yield per plant (g.)	-0.44	-0.06	-0.04	0.06	-0.03
11	Leaf senescence (SPAD Value)	-0.12	0.10	0.02	-0.69	0.69

Table 3: Top five mutant lines contributing to principal component 2, 3 & 5

	PC2	PC3	PC5
	(PH, PL, TSE, RSE)	(PH, PL)	(DFF, LS)
Ι	FR13B (200GY+0.2%EMS)-entry no. 240	Labella (350GY+0.2%EMS)-entry no. 92	FR13A (250GY)-entry no. 45
II	FR13B (200GY+0.2%EMS)-entry no. 239	Labella (350GY+0.2%EMS)-entry no. 91	Labella (350GY)-entry no. 81
III	Labella (250GY)-entry no. 124	Labella (300GY+0.2%EMS)-entry no. 116	Labella (200GY+0.2%EMS)-entry no. 151
IV	FR13B (200GY+0.2%EMS)-entry no. 238	Labella (300GY)-entry no. 109	FR13A (250GY)-entry no. 47
V	Labella (350GY)-entry no. 85	Labella (200GY)-entry no. 149	FR13B (300GY+0.2%EMS)-entry no. 196

DFF= Days to 50% flowering PH= Plant height

PL= Panicle length TSE= Total shoot elongation

RSE= Relative shoot elongation LS= Leaf senescence

Group	No. of mutants	Mutant lines in cluster					
K	within cluster						
I	10	FR13A (350GY+0.2% EMS)-entry no. 9, FR13A (350GY)-entry no. 15, FR13A (300GY)-entry no. 28, FR13A (300GY+0.2% EMS)-entry no. 32, FR13A (300GY+0.2% EMS)-entry no. 37, FR13A (300GY+0.2% EMS)-entry no. 38, FR13A (250GY+0.2% EMS)-entry no. 60, FR13A (200GY)-entry no. 64, FR13B (350GY)-entry no. 166, FR13B (250GY+0.2% EMS)-entry no. 220.					
П	29	 FR13A (350GY)-entry no.11, FR13A (350GY)-entry no.12, FR13A (350GY)-entry no.14,FR13A (300GY)-entry no.30, FR13A (300GY+0.2% EMS)-entry no.34, FR13A (300GY+0.2% EMS)-entry no.39, FR13A (250GY+0.2%EMS)-entry no.55, FR13A (200GY)-entry no.63, FR13A (200GY+0.2%EMS)-entry no.78, FR13A (200GY+0.2%EMS)-entry no.80, Labella (350GY+0.2%EMS)-entry no.92, Labella (300GY+0.2%EMS)-entry no.112, FR13B (350GY)-entry no.167, FR13B (350GY)-entry no.169, FR13B (350GY+0.2%EMS)-entry no.178, FR13B (350GY+0.2%EMS)-entry no.180, FR13B (300GY)-entry no.182, FR13B (300GY)-entry no.185, FR13B (300GY+0.2%EMS)-entry no.191, FR13B (300GY+0.2%EMS)-entry no.193, FR13B (300GY+0.2%EMS)-entry no.197, FR13B (300GY+0.2%EMS)-entry no.198, FR13B (300GY+0.2%EMS)-entry no.200, FR13B (250GY)-entry no.205, FR13B (250GY)-entry no.209, FR13B (250GY+0.2%EMS)-entry no.216, FR13B (250GY+0.2%EMS)-entry no.217, FR13B (250GY+0.2%EMS)-entry no.219, FR13B (200GY)-entry no.227. 					
ш	28	 FR13A (300GY)-entry no.22, FR13A (300GY)-entry no.25, FR13A (250GY)-entry no.42, FR13A (250GY)-entry no.44, FR13A (250GY)-entry no.45, FR13A (250GY)-entry no.47, FR13A (200GY)-entry no.70, Labella (350GY)-entry no.90, Labella (350GY+0.2%EMS)-entry no.91, Labella (300GY)-entry no.109, Labella (250GY+0.2%EMS)-entry no.137, Labella (250GY+0.2%EMS)-entry no.138, Labella (250GY+0.2%EMS)-entry no.137, Labella (250GY+0.2%EMS)-entry no.138, Labella (250GY+0.2%EMS)-entry no.140, Labella (200GY)-entry no.142, Labella (200GY)-entry no.144, Labella (200GY+0.2%EMS)-entry no.153, Labella (200GY+0.2%EMS)-entry no.157, Labella (200GY+0.2%EMS)-entry no.159, Labella (200GY+0.2%EMS)-entry no.160, FR13B (350GY+0.2%EMS)-entry no.173, FR13B (350GY+0.2%EMS)-entry no.177, FR13B (300GY)-entry no.181, FR13B (300GY)-entry no.194, FR13B (250GY)-entry no.201, FR13B (250GY)-entry no.206, FR13B (250GY)-entry no.210, FR13B (200GY)-entry no.223. 					
IV	11	FR13A (350GY+0.2%EMS)-entry no.3, FR13A (350GY+0.2%EMS)-entry no.6, FR13A (350GY)-entry no.13, FR13A (350GY)-entry no.16, FR13A (350GY)-entry no.18, FR13A (350GY)-entry no.19, FR13A (350GY)-entry no.20, FR13A (300GY)-entry no.21, FR13A (300GY+0.2%EMS)-entry no.40, FR13A (250GY+0.2%EMS)-entry no.54, FR13A (200GY)-entry no.67.					
v	21	 FR13A (350GY+0.2% EMS)-entry no.4, FR13A (350GY+0.2% EMS)-entry no.10, FR13A (350GY)-entry no.17, FR13A (300GY)-entry no.29, FR13A (250GY)-entry no.46, FR13A (250GY)-entry no.50, FR13A (250GY+0.2% EMS)-entry no.51, FR13A (250GY+0.2% EMS)-entry no.52, FR13A (250GY+0.2% EMS)-entry no.58, FR13A (200GY+0.2% EMS)-entry no.121, Labella (250GY+0.2% EMS)-entry no.123, Labella (250GY)-entry no.130, Labella (250GY+0.2% EMS)-entry no.132, Labella (250GY+0.2% EMS)-entry no.135, Labella (250GY+0.2% EMS)-entry no.130, Labella (200GY)-entry no.150, FR13B (300GY)-entry no.138, FR13B (250GY)-entry no.208, FR13B (200GY+0.2% EMS)-entry no.232, FR13A c-entry no.242. 					
VI	28	 41 FR13A (250GY)-entry no. 41, FR13A (200GY+0.2%EMS)-entry no. 73, Labella (350GY)-entry no. 82, Labella (350GY+0.2%EMS)-entry no. 95, Labella (350GY+0.2%EMS)-entry no. 97, Labella (350GY+0.2%EMS)-entry no. 100, Labella (300GY)-entry no. 103, Labella (300GY)-entry no. 104, Labella (300GY+0.2%EMS)-entry no. 113, Labella (300GY+0.2%EMS)-entry no. 115, Labella (300GY+0.2%EMS)-entry no. 118, Labella (250GY)-entry no. 115, Labella (250GY)-entry no. 127, Labella (250GY)-entry no. 128, Labella (250GY)-entry no. 126, Labella (250GY)-entry no. 127, Labella (250GY)-entry no. 128, Labella (250GY)-entry no. 136, Labella (200GY)-entry no. 141, Labella (200GY)-entry no. 143, Labella (200GY)-entry no. 149, Labella (200GY+0.2%EMS)-entry no. 151, Labella (200GY+0.2%EMS)-entry no. 152, FR13B (350GY+0.2%EMS)-entry no. 172, FR13B (300GY+0.2%EMS)-entry no. 199, FR13B (250GY)-entry no. 203, FR13B (250GY+0.2%EMS)-entry no. 212, FR13B (200GY)-entry no. 234. 					
VII	17	 FR13A (350GY+0.2%EMS)-entry no.7, Labella (350GY)-entry no. 83, Labella (350GY+0.2%EMS)-entry no. 93, Labella (350GY+0.2%EMS)-entry no. 96, Labella (300GY)-entry no. 107, Labella (300GY+0.2%EMS)-entry no. 114, FR13B (350GY)-entry no. 168, FR13B (300GY)-entry no. 184, FR13B (300GY)-entry no. 186, FR13B (300GY)-entry no. 187, FR13B (300GY)-entry no. 188, FR13B (250GY)-entry no. 202, FR13B (200GY)-entry no. 231, FR13B (200GY+0.2%EMS)-entry no. 231, FR13B (200GY+0.2%EMS)-entry no. 237, FR13B (200GY+0.2%EMS)-entry no. 238, Labella c-entry no.241. 					
VIII	14	FR13A (300GY)-entry no. 27, FR13A (200GY+0.2%EMS)-entry no. 74, Labella (350GY+0.2%EMS)-entry no. 94, Labella (350GY+0.2%EMS)-entry no. 98, Labella (300GY)-entry no. 108, Labella (300GY+0.2%EMS)-entry no. 119, Labella (300GY+0.2%EMS)-entry no. 120, Labella (250GY+0.2%EMS)-entry no. 134, FR13B (350GY)-entry no. 161, FR13B (350GY+0.2%EMS)-entry no. 176, FR13B (250GY+0.2%EMS)-entry no. 213, FR13B (200GY)-entry no. 222, FR13B (200GY+0.2%EMS)-entry no. 235, FR13B c-entry no.243.					
IX	10	 FR13A (350GY+0.2% EMS)-entry no.2, FR13A (350GY+0.2% EMS)-entry no.5, FR13A (300GY)-entry no.26, FR13A (300GY+0.2% EMS)-entry no.33, FR13A (200GY)-entry no.61, FR13A (200GY)-entry no.62, FR13A (200GY)-entry no.65, FR13A (200GY)-entry no.66, FR13A (200GY)-entry no.68, FR13A (200GY)-entry no.69. 					
X	3	FR13A (350GY+0.2% EMS)-entry no.1, Labella (200GY)-entry no.146, Labella (200GY)-entry no.147.					
XI	11	FR 13A (250GY)-entry no. 48, Labella (350GY)-entry no. 84, Labella (300GY)-entry no. 101, Labella (300GY+0.2%EMS)-entry no. 116, Labella (300GY+0.2%EMS)-entry no. 117, Labella (250GY)-entry no. 124, Labella (200GY)-entry no. 145, FR13B (350GY)-entry no. 165, FR13B (350GY+0.2%EMS)-entry no. 175, FR13B (200GY)-entry no. 230, FR13B (200GY+0.2%EMS)-entry no. 236.					

XII	20	 FR 13A (250GY+0.2% EMS)-entry no. 53, FR 13A (200GY+0.2%EMS)-entry no. 77, FR 13A (200GY+0.2%EMS)-entry no. 79, Labella (350GY)-entry no. 81, Labella (350GY)-entry no. 87, Labella (350GY)-entry no. 89, Labella (300GY)-entry no. 105, Labella (300GY+0.2%EMS)-entry no. 111, Labella (250GY+0.2%EMS)-entry no. 133, Labella (200GY+0.2%EMS)-entry no. 155, FR13B (350GY)-entry no. 162, FR13B (350GY)-entry no. 164, FR13B (350GY)-entry no. 170, FR13B (350GY+0.2%EMS)-entry no. 179, FR13B (300GY)-entry no. 183, FR13B (300GY+0.2%EMS)-entry no. 196, FR13B (250GY+0.2%EMS)-entry no. 214, FR13B (250GY+0.2%EMS)-entry no. 218, FR13B (200GY)-entry no. 226, FR13B (200GY)-entry no. 229.
XIII	10	FR13A (300GY+0.2%EMS)-entry no. 35, FR13A (200GY+0.2%EMS)-entry no.72, Labella (350GY)-entry no. 85, Labella (350GY)-entry no. 88, Labella (350GY+0.2%EMS)-entry no. 99, Labella (250GY)-entry no. 125, Labella (200GY+0.2%EMS)-entry no. 156, FR13B (300GY)-entry no. 190, FR13B (300GY+0.2%EMS)-entry no. 195, FR13B (200GY+0.2%EMS)-entry no. 239.
XIV	21	 FR13A (350GY+0.2%EMS)-entry no. 8, FR13A (300GY)-entry no. 23, FR13A (300GY)-entry no. 24, FR13A (250GY)-entry no. 43, FR13A (250GY)-entry no. 49, FR13A (250GY+0.2% EMS)-entry no. 56, FR13A (250GY+0.2% EMS)-entry no. 57, FR 13A (250GY+0.2%EMS)-entry no. 59, FR13A (200GY+0.2%EMS)-entry no. 71, FR13A (200GY+0.2%EMS)-entry no. 76, Labella (300GY)-entry no. 106, Labella (250GY)-entry no. 129, Labella (200GY)-entry no. 148, Labella (200GY+0.2%EMS)-entry no. 154, FR13B (350GY)-entry no. 163, FR13B (350GY+0.2%EMS)-entry no. 171, FR13B (300GY+0.2%EMS)-entry no. 192, FR13B (250GY)-entry no. 204, FR13B (250GY)-entry no. 207, FR13B (250GY+0.2%EMS)-entry no. 211, FR13B (250GY+0.2%EMS)-entry no. 215.
XV	9	FR13A (300GY+0.2%EMS)-entry no. 31, FR13A (300GY+0.2%EMS)-entry no. 36, Labella (350GY)-entry no. 86, Labella (300GY)-entry no. 102, Labella (300GY)-entry no. 110, Labella (200GY+0.2%EMS)-entry no. 158, FR13B (350GY+0.2%EMS)-entry no. 174, FR13B (200GY)-entry no. 224, FR13B (200GY)-entry no. 225.
XVI	1	FR13B (200GY+0.2%EMS)-entry no. 240

Table 5: Mean characteristics (K-Mean) on various submergence tolerance related traits for each cluster in mutant lines of rice

	DFF	PH	PL	NTP	NFTP	SP	TSE	RSE	TS	GYP	LS
1 Cluster	123.73	99.67	20.27	5.54	3.23	48.43	15.40	16.02	8.19	22.03	40.04
2 Cluster	121.20	116.08	22.22	4.42	2.76	47.59	20.50	17.90	8.16	21.70	38.89
3 Cluster	123.47	119.68	22.80	6.80	3.99	64.44	23.55	19.89	7.00	22.64	40.23
4 Cluster	114.00	97.17	20.26	6.67	4.31	64.03	15.17	15.36	6.94	22.61	37.79
5 Cluster	121.43	116.18	21.33	9.30	6.88	76.04	22.06	19.06	6.04	23.57	40.54
6 Cluster	123.44	126.00	23.71	8.69	5.69	70.11	29.65	23.77	6.73	23.10	40.88
7 Cluster	124.15	132.39	24.28	6.35	4.10	60.16	33.46	25.85	7.01	22.68	40.64
8 Cluster	119.43	124.01	23.73	11.09	8.53	80.77	26.88	22.17	5.06	24.03	41.08
9 Cluster	116.67	105.61	19.66	9.32	5.34	79.70	16.46	15.90	5.15	23.78	41.01
10 Cluster	106.86	118.94	22.90	13.90	10.65	89.81	17.11	14.47	5.01	26.15	41.97
11 Cluster	126.18	134.54	25.13	9.87	7.44	78.40	33.60	25.60	5.40	23.82	40.47
12 Cluster	127.63	120.95	22.69	5.07	3.14	54.20	28.38	23.92	7.41	22.17	39.83
13 Cluster	125.77	126.54	23.09	5.09	2.90	38.90	33.66	27.13	9.01	21.71	39.77
14 Cluster	118.03	113.23	21.70	6.14	3.71	64.08	19.29	17.21	6.98	22.53	39.45
15 Cluster	125.56	117.84	22.27	4.77	2.78	35.70	26.57	23.68	9.04	21.78	37.93
16 Cluster	120.63	137.97	26.33	6.01	4.03	62.03	60.81	43.22	7.04	22.30	39.18

 Table 6: Contribution percentage of eleven morpho-physiological traits towards genetic divergence in mutant lines of rice under submergence condition

S. No.	Source	Abbreviation	Contribution (%)	Times Ranked 1st		
1	Days to 50% flowering (Days)	DFF	22.05	6482.00		
2	Plant height (cm)	PH	30.90	9086.00		
3	Panicle length (cm)	PL	0.03	9.00		
4	No. of tillers per plant	NTP	0.07	22.00		
5	No. of fertile tillers per plant	NFTP	0.03	9.00		
6	Survival percentage (%)	SP	38.26	11249.00		
7	Total shoot elongation (cm)	TSE	7.43	2185.00		
8	Relative shoot elongation (cm)	RSE	0.88	260.00		
9	Tolerance score	TS	0.01	0.00		
10	Grain yield per plant (g)	GYP	0.01	0.00		
11	Leaf senescence (SPAD Value)	LS	0.34	101.00		



Fig 1: Distribution of 184 rice land races between first, second and third principle components (3-D components (3-D Plot diagram)

4. Conclusion

Based on above discussion, PCA analysis revealed the possibility for improvement of rice mutant lines for improvement of submergence tolerance by manipulation of various traits *viz.*, TS, TSE, RSE, PL and PH. Labella (200GY)- entry no. 146, Labella (200 GY)-entry no. 147, FR13B (350GY)- entry no. 165, FR13B (200GY)-entry no. 222 were found most divergent mutant lines with FR13B (200GY)-entry no. 224, Labella (300GY)-entry no. 102 and FR13B (200GY)-entry no. 225 which can be utilized effectively in breeding programme for improvement of submergence tolerance. Suitable mutant lines selected from cluster VIII and cluster X; the two diverse groups based on their cluster mean may also be involves in breeding programmes for rice improvement against submergence tolerance.

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