



Genetic variation and diversity analysis of rice (*Oriza sativa* L.) based on quantitative traits for crop improvement

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ABSTRACT

The development of varieties is a continuous process and the success of the plant breeding depends upon the selection of suitable plants to be utilized. The effectiveness of selection depends basically upon the magnitude of genetic variability in the breeding material. This study was carried out at Africa Rice Center, International Institute of Tropical Agriculture (IITA) Ibadan, Nigeria. Two hundred and thirty-nine (239) lines of anther-culture derived from South Korea with an improved variety from Nigeria as check were established for their genetic variability and diversity analysis. The experiment was conducted using Alpha lattice design with four blocks each planted in 60 entries replicated tow times. The estimation of genotypic coefficient variance and phenotypic coefficient variance was found to be high (>20%) for grain yield, grain yield per plant, biomass, number of tillers, panicle weight, effective tillers, leaf area, leaf area index and number of grains per panicle. The broad sense heritability was highest for days to 50% flowering followed by plant height, 1000 grain weight, panicle length and number of tillers. The estimation of genetic advance was found to be highest for grain yield. Cluster analysis grouped the 240 accessions into four clusters (A, B, C and D), indicate wide genetic diversity among these groups. Principal component analysis showed that the first three components accounted for 64.78% of the total variation. Therefore, indicate the presence of large genetic variability, which is important as it gives wide range of selection. Among all genotypes UPN 632 and UPN 540 showed the best performance.

INTRODUCTION

Rice is one of the most important cereal crops and provides the staple food for about half of the world's population (Moosavi et al., 2015). Genetic enhancement is one of the important tools to improve the productivity. (Babu et al., 2012). Knowledge on the nature and magnitude of the genetic variation governing the inheritance of quantitative character like yield and its components is essential for effecting genetic improvement. (Rakesh et al., 2015). A critical analysis of the genetic variability parameters, namely, genotypic coefficient of variability (GCV), phenotypic coefficient of variability (PCV), heritability and genetic advance for different traits of economic importance is a major pre-requisite for any plant breeder to work with crop improvement programmes. (Kishore et al., 2015). Genetic variability for agronomic traits is the key component of breeding programs for broadening the gene pool of rice and other crops. The genetic coefficient of variation together with heritability estimate would give the best picture of the amount of advance to be expected from selection. The amount of genetic advance under selection depends mainly on the amount of genetic variability. (Immanuel et al., 2011).

Development of high yielding varieties requires the knowledge of existing genetic variability. Hence, rice breeders are interested in developing varieties with improved yield and other desirable agronomic characters. (Idris and Mohamed, 2013). Most agronomically significant characters are inherited quantitatively and are known to be affected by environmental factors. Selection based on the phenotype would be difficult for such difficult traits. In breeding programs, it is often difficult to manipulate such traits, since several inter-componential characters indirectly control them. (Immanuel et al., 2011). Genetic variability among traits is important for breeding and in selecting desirable types. The low heritability of grain characters made selection for high yielding varieties possible usually using various components traits associated with yield. (Mulugeta et al., 2012). High magnitude of variability in a population provides the opportunity for selection to evolve a variety having desirable characters. (Bornare et al., 2014). Therefore, the objectives of this study is to determine the genetic variation and diversity analysis of two hundred and twenty nine rice genotypes and an improved Nigerian variety (FARO44) were evaluated based on agronomic traits.

MATERIALS AND METHODS

The experiment was conducted at experimental field of African Rice Centre at International Institute of Tropical Agriculture (IITA), Old Oyo road, Ibadan, Oyo state, Nigeria. During 2020 cropping season. IITA is located at longitude 7°30'8"N, latitude 3°54'37"E and at elevation

243m above sea level (Ariyo et al., 2018). The experimental materials of this study consisted of two hundred and thirty-nine (239) accessions of rice anther culture derived from South Korea, of *O. sativa* L. and one improved and adapted Nigeria rice variety (FARO 44) collected from African Rice Center, IITA used as check. The experiment was established in alpha lattice design (rows x column) with four columns and planted sixty entries in each column (4x60) with two replications.

The seedlings were raised in the wet prepared nursery bed and then transplanted to the field after 25 days under irrigated system. A single row with size of 0.2x3 m was used as plot. A single seedling was transplanted per hill at the spacing of 20x20 cm between rows and between hills. The chemical fertilizer NPK (15:15:15) was applied as a basal application of 200 kg/ha (N₂, P₂O₅ and K₂O). Urea was applied at the rate of 65 kg/ha at tillering stage and the second rate of 35 kg/ha was applied at the beginning of panicle initiative (booting) stage. The weeds were controlled twice by using selective herbicide Vespanil Plus (250 ml/ 20 liters of water) at early stage of crop development and before flowering.

Data was collected at appropriate stage of the crop development. The agronomic characters were measured by randomly selected plants from each experimental unit (row). The Standard Evaluation System (SES) by International Rice Research Institute (IRRI) for Rice reference manual IRRI, 2002 was used for all trait measurements except where stated otherwise. The data collected were plant height (cm), number of tillers per plant, days to 50% flowering, flag leaf length (cm), panicle fertility (%), productive tillers per plant, panicle length (cm), panicle weight (g), biomass (g), number of grains per panicle, 1000 grain weight (g), grain yield per plant (g) and grain yield per plot (kg/ha). Leaf area (cm²) was measured manually following the formula:

$$LA=W \times L \times K$$

Where: LA ≡ Leaf Area, W ≡ Leaf Width, L ≡ Leaf length and K ≡ Constant = 0.75

Leaf area index (m²) was calculated as described (Efisue and Dike, 2020) as follow:

$$LAI = LA / \text{area covered by the plant}$$

Where: LAI ≡ Leaf Area Index, LA ≡ Leaf Area

Genetic parameters were estimated to identify genetic variation among accessions and to determine genetic and environmental effects on various characters. These genetic parameters were calculated as described (Burton and Devane, 1953 and Johnson et al., 1955).

$$\text{Genotypic variance: } \theta_g^2 = \text{MSG-MSE}/r$$

Where MSG is the mean square of genotypes, MSE is mean square of error, and r is number of replications
Phenotypic variance:

$$\theta_p^2 = \theta_g^2 \times \theta_e^2$$

Where θ_g^2 is the genotypic variance and θ_e^2 is the mean squares of error.

Genotypic coefficient of variance (GCV) and phenotypic coefficient of variance (PCV):

$$\text{GCV (\%)} = \sqrt{\sigma_g^2} / \bar{x} \times 100$$

$$\text{PCV (\%)} = \sqrt{\sigma_p^2} / \bar{x} \times 100$$

Where $\sqrt{\sigma_g^2}$ = Genotypic variance, $\sqrt{\sigma_p^2}$ = Phenotypic variance, \bar{x} = grand mean of the trait.

GCV and PCV values were categorized as low (0-10%), moderate (10-20%) and high (20% and above).

Heritability (Broad sense) was computed as described (Amegan et al., 2020).

$$H^2 = \sigma_g^2 / \sigma_p^2 \times 100$$

Where σ_g^2 is the genotypic variance and σ_p^2 is the phenotypic variance

The Heritability was categorized as low (0-30%), moderate (30-60%) and high (60% and above) as described by Robinson et al, (1949).

Expected genetic advance (GA):

Expected genetic advance (GA) was computed according to (Allard, 1960) cited by (Saeed et al., 2018) as:

$$\text{GA} = \sigma_p \times H_b^2 \times k$$

Where, GA = Expected genetic advance, σ_p = Phenotypic standard deviation, H_b^2 = Heritability in broad sense and k = the standardize selection differential at 5% selection intensity (K = 2.063).

Principal component analysis was computed by using the principal component procedure of multivariate technique in Statistical Analysis System SAS, version 9.4 (2018) for analyzing data for all the characters. Cluster analysis was carried out to clustering the genotypes into different groups using R software version 3.9.6.

RESULTS AND DISCUSSION

Genetic Variability parameters

High variability in the initial breeding material ensures better chances of producing desired crop plant.

Genotypic coefficient of variation (GCV) measures the variability of any trait. The extent of the environmental influence on any trait is indicated by the magnitude of the differences between the genotypic and phenotypic coefficients of variation. Large differences reflect high environmental influence as reveal by high phenotypic influence, while small differences reveal high genetic influence (Idris and Mohamed, 2013). In this study phenotypic coefficients of variance were higher than the corresponding genotypic coefficients of variance for all the studied traits (Table 1). This indicates the presence of environmental influence to some degree in the phenotypic expression of the characters (Iftekharuddeula et al., 2001 and Idris et al., 2012) observed similar results. Likewise (Singh et al., 2019, Sameera et al., 2016, Rakesh et al., 2015 and Mulugeta et al., 2012).

The differences were observed between genotypic and phenotypic coefficients of variance for days to 50% flowering, plant height, panicle length, 1000 grain weight and fertility (Table 1). This indicates that genotypic influence had played an important role rather than phenotypic influence indicating less influence of environment on these traits. Therefore, selection on the basis of phenotype alone can be effective for the improvement of these traits (Idris and Mohamed, 2013). High differences between genotypic and phenotypic coefficients of variance were observed for biomass, grain yield, grain yield per plant, number of grains per panicle, panicle weight, and effective tillers. Therefore, indicating that phenotypic influence assert more on the existence of environment influence on these traits.

The characters studied in the present investigation exhibited high, moderate to low PCV and GCV values. The high PCV (>20%) and GCV values were recorded for grain yield, grain yield per plant, biomass, number of tillers, panicle weight, effective tillers, leaf area, leaf area index and number of grains per panicle (Table 1). The result in conformity with the reports of (Kishore et al., 2015). for grain yield; (Prasad et al., 2013) for number of tillers per plant; (Sameera et al., 2016) for number of effective tillers and number of grains per panicle; (Hasib et al., 2004) for grain yield per plant; (Bekele et al., 2013) for effective tillers per plant; (Bornare et al., 2014) for effective tillers and grain yield per plant; (Singh et al., 2019) also recorded similar observation for grain yield per plant and number of tillers. Hence, selection on the basis of these phenotypic characters in these genotypes can also be effective for improvement of grain yield. However, moderate (10-20%) genotypic and phenotypic coefficients of variance were recorded in the present study for plant height (PH), days to 50% flowering (D50%F), panicle length (PL) and 1000 grain weight (TGW) (Table 1). These results corroborate the earlier reports (Singh et al., 2019) for days to 50% flowering, panicle weight and 1000 grain weight and for days to 50% flowering (Bornane et al., 2014 and Kishore et al., 2015). In contrast, low (<10%) estimates of genotypic and phenotypic coefficients of variation were observed in fertility (FER).

Table 1: Estimates of genetic parameters, broad sense heritability, and genetic advance for yield and its components traits

TRAIL	σ_g^2	σ_p^2	GCV (%)	PCV (%)	H^2 (%)	GA
NT	6.616	10.607	30.527	38.653	62.376	442.183
PH	167.700	210.540	12.026	13.475	79.652	2702.740
D50%F	33.897	42.336	9.727	10.870	80.068	1137.393
PL	5.564	7.425	9.544	11.025	74.932	443.021
FL	28.300	61.969	17.193	25.441	45.669	779.519
ET	1.861	4.609	21.181	33.338	40.365	182.342
LA	59.137	101.779	23.884	31.333	58.103	1266.050
LAI	0.000	0.001	23.884	31.333	58.103	3.188
FER	8.481	41.910	3.192	7.096	20.237	282.829
BM	61.144	145.323	29.480	45.448	42.075	1080.359
PW	38.597	88.645	25.463	38.588	43.542	869.631
NG/P	388.986	1134.050	19.615	33.492	34.301	2474.216
TGW	8.602	11.403	10.841	12.482	75.435	556.583
GY/P	25.610	56.421	30.391	45.109	45.391	749.264
YLD	1575319.0	3038758.0	38.781	53.863	51.841	191596.882

σ_g^2 : Genotypic variance, σ_p^2 : phenotypic variance, GCV: genotypic coefficient of variance PCV: phenotypic coefficient of variance, H^2 : broad sense heritability, GA: genetic advance.

NT=Number of tillers; PH=Plant height; D50%F=Days to 50% flowering; PL=Panicle length; FL=Flag leaf; ET=Effective tillers; LA=Leaf area; LAI=Leaf area index; FER=Fertility; BM=Biomass; PW=Panicle weight; NG/P=Number of grains per panicle; TGW=1000grain weight; GY/P=Grain yield per plant; YLD= yield.

Heritability

The heritability estimates act as predictive instrument in expressing the reliability of phenotypic value. Therefore, good knowledge of transmission of a particular trait assists the plant breeders in predicting the behavior of succeeding generations because high heritability of a trait simplifies the selection procedure (Khaliq et al., 2009). In this study, heritability in broad sense was calculated for all characters under study and is presented in (table 1). Heritability was classified as low (<30%), medium (30-60%) and high (>60%). High heritability was recorded for days to 50% flowering (80.07), plant height (79.65), 1000 grain weight (75.44), panicle length (74.93) and number of tillers (62.38). These results are in conformity with the reports of (Idris et al., 2013) for 1000 grain weight and plant height; (Bisne et al., 2009) for number of tillers; (Dhanwani et al., 2013) for days to 50% flowering. Similar results were also quoted by (Rakesh et al., 2015), (Subbaiah et al., 2011) and Ananadarao et al., 2011). Low heritability value was recorded for fertility (20.24). High heritability

values indicate that the characters under study are less influenced by environment in their expression, therefore, the selection may base on the phenotypic expression of these characters in the individual plant by adopting simple selection methods. High heritability indicates the scope of genetic improvement of these characters through selection. The low heritability recorded for fertility indicate that direct selection for this trait will be ineffective.

Genetic advance

The genetic advance is a useful indicator of the progress that can be expected as result of exercising selection on the pertinent population. Genetic advance I (Table 1) was highest for grain yield . This result is in conformity with the findings of (Kishore et al., 2015, Dhanwani et al., 2013 and Bornare et al., 2014). This suggest that this character is predominantly controlled by additive gene action. Hence genetic improvement through selection for this trait may be effective (Singh et al., 2019). The lowest genetic advance was noticed for leaf area index (3.188).

The information on genetic variation, heritability and genetic advance helps to predict the genetic gain that could be obtained in later generations, if selection is made for improving the particular trait under study. Selection for the traits having high heritability coupled with high genetic advance is likely to accumulate more additive genes leading to further improvement of their performance (Panse and Suhatme, 1957) cited by (Rakesh et al., 2015). High heritability along with high genetic advance was noticed for traits days to 50% flowering and plant height (Table 1) this was also reported by (Seyoum *et al.*, 2012 and Rakesh et al., 2015). Characters which showed high heritability coupled with moderate or low genetic advance such as 1000 grain weight, panicle length and number of tillers can be improved by intermating superior genotypes of segregating population developed from combination breeding (Samadhia, 2005), (Babu et al., 2012).

Cluster Analysis of Rice Genotypes Evaluated

Cluster analysis of some agronomic traits and yield was conducted and dendrogram was constructed using rice genotypes values presented in the (Figure 1). Four major groups (A, B, C, and D) were observed among 240 rice genotypes including an improved variety (FARO44). Cluster analysis provide a useful means for estimating morphological diversity within and between genotypes evaluated. Its useful tool to detect the potential of breeding value. Cluster A consisted of 73 genotypes, Cluster B had least genotypes (20) of the group with two sub groups; B1 contain three genotypes (UPN_643, UPN_611 and UPN_639) and B2 comprise 17 genotypes. Cluster C consisted 35 genotypes. Cluster D recorded 112 genotypes which is the biggest and the check (FARO44) was clustered in this group (Figure 1) and FARO 44 contains fortified iron (Ikuli et al., 2017) that showed more genetic relatedness among the genotypes as compared to any other cluster. Cluster analysis showed the genetic variation among 240 genotypes indicating wide diversity of genetic material. Hence, crossing between two groups could give good result and cluster also assist breeders in making good choice in selection of parents in crop improvement. However, breeders need to evaluate genotypes accurately in each grouping before use in rice breeding programme (Maji et al., 2012). For the selection of parents, genetic diversity is one of the important decisive factors that will enhance crop improvement (Tuhina-khatun et al., 2015).

Principal Components Analysis

The result of principal component analysis explained the genetic diversity of the rice accessions for all traits under study. The PCA with eigenvalues >1 and which explained 20% of the variation were considered. Out of fifteen only three principal components exhibited more than one eigenvalue, and showed 64.78% of total cumulative variability among the traits studied (Table 2). It also revealed that the first principal component accounted for 37.17% of total variance and traits such as plant height (0.32), panicle weight (0.32), panicle length (0.31), biomass (0.31) and grain yield per plant (0.30) are the most positively contributed. The second component accounted for 20.39% of the total variance with most positively contribution variables such as effective tillers (0.37), number of tillers per plant (0.31) and grain yield (0.30). The third principal component accounted for 7.22% and the variables contributing most positively were number of tillers (0.55), biomass (0.39) and effective tillers (0.32). These results conformed presence of strong differences among genotypes. Through the PCA, the number of characters, which are responsible for the observed variation within a group could be identified. The principal component with higher eigenvalues and variables which had high loading were considered as best representative of system attributes (Pachauri et al., 2017), which may be considered in the utilization of these traits for crop improvement. These result in agreement with the findings of (Ayenew et al., 2020) and (Worede et al., 2014) explained 73.5% of the total variability and (Singh et al., 2013) explained 62.72% of total variation in rice using the first three principal components.

Agronomic parameters projection in the PCA plots showed the phenotypic variation among the populations this indicated how dispersed along each principal component (Figure 2). The genotypes UPN540 and UPN632 on the top right part of the plot showed the highest performance in either axis of principal component 1 and 2. While the genotypes UPN643 is the lowest performance in either axis of the principal component 1 (Figure 2). The first three principal components accounted for 64% of the total variation, which indicated a very strong correlation among the characters being studied. Accordingly, in the first PCA, the plant height, panicle length, biomass, panicle weight, and grain yield per plant were important in separating the genotypes due to their high loadings. Similarly, (Tuhina-khatun et al., 2015 and Worede et al., 2014) explained 61.2% of the total variability using the first and second principal components.

Table 2: Principal Component Analysis among Agronomic Traits

Variables	PC1	PC2	PC3
<i>Eigenvalue</i>	5.5759	3.0579	1.0831
Difference	2.5181	1.9747	0.1382
Proportion (%)	0.3717	0.2039	0.0722
Cumulative (%)	0.3717	0.5756	0.6478
<i>Eigen Vectors</i>			
NT	-0.051	0.312	0.553
PH	0.323	-0.133	-0.001
D50%F	0.288	0.050	0.070
PL	0.315	-0.150	-0.057
FL	0.218	-0.336	0.006
ET	0.143	0.373	0.319
LA	0.274	-0.345	0.133
LAI	0.274	-0.345	0.133
FER	0.206	0.216	-0.403
BM	0.313	-0.008	0.393
PW	0.323	0.224	0.048
NG/P	0.257	0.186	-0.471
TGW	0.121	-0.260	0.038
GY/P	0.300	0.284	-0.050
YLD	0.280	0.301	-0.061

PC1: First principal component, PC2: Second principal component, PC3: Third principal component. NT=Number of tillers; PH=Plant height; D50%F=Days to 50% flowering; PL=Panicle length; FL=Flag leaf; ET=Effective tillers; LA=Leaf area; LAI=Leaf area index;

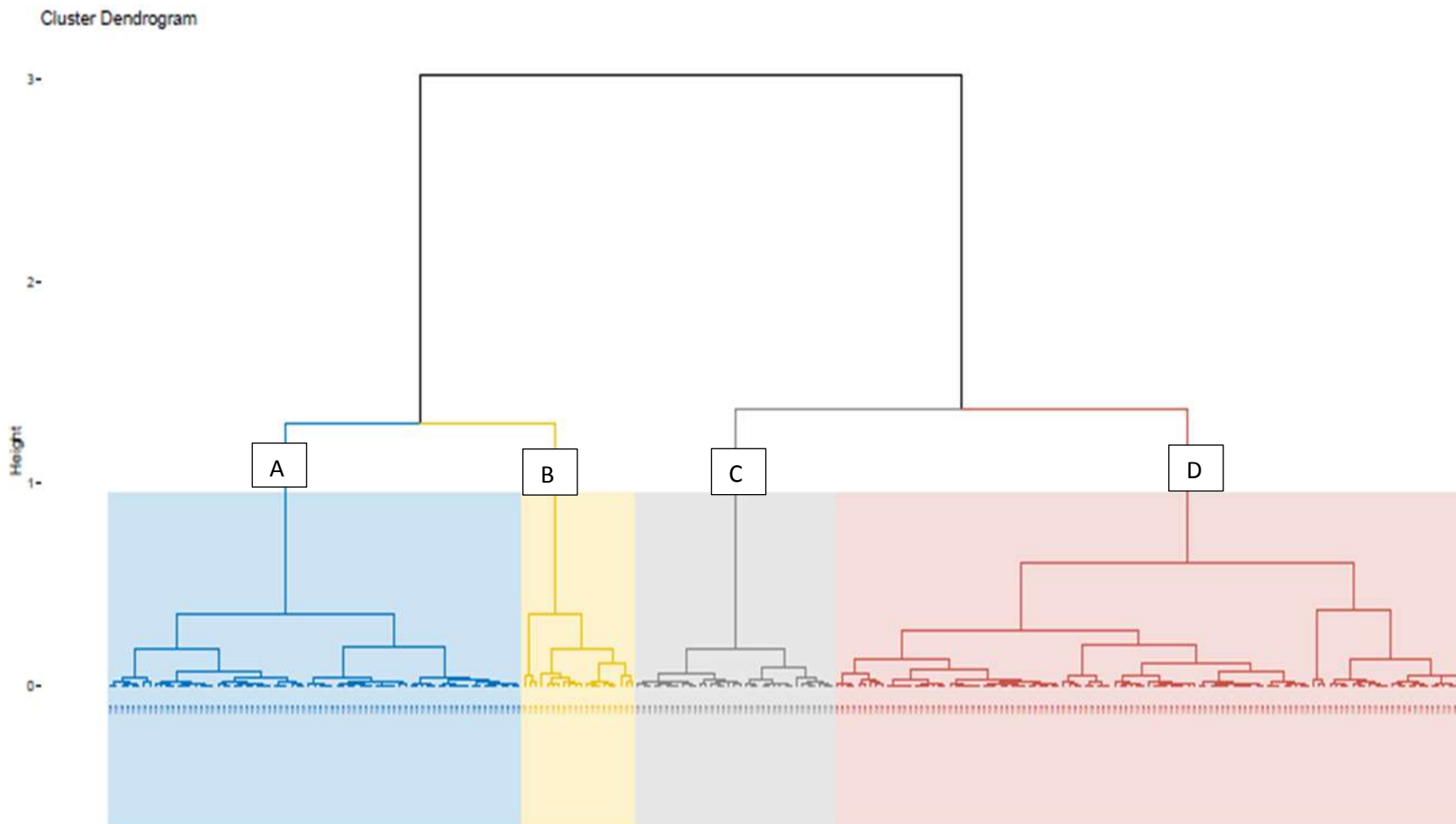


Figure 1: Cluster dendrogram representing distribution of 240 rice genotypes based on quantitative traits

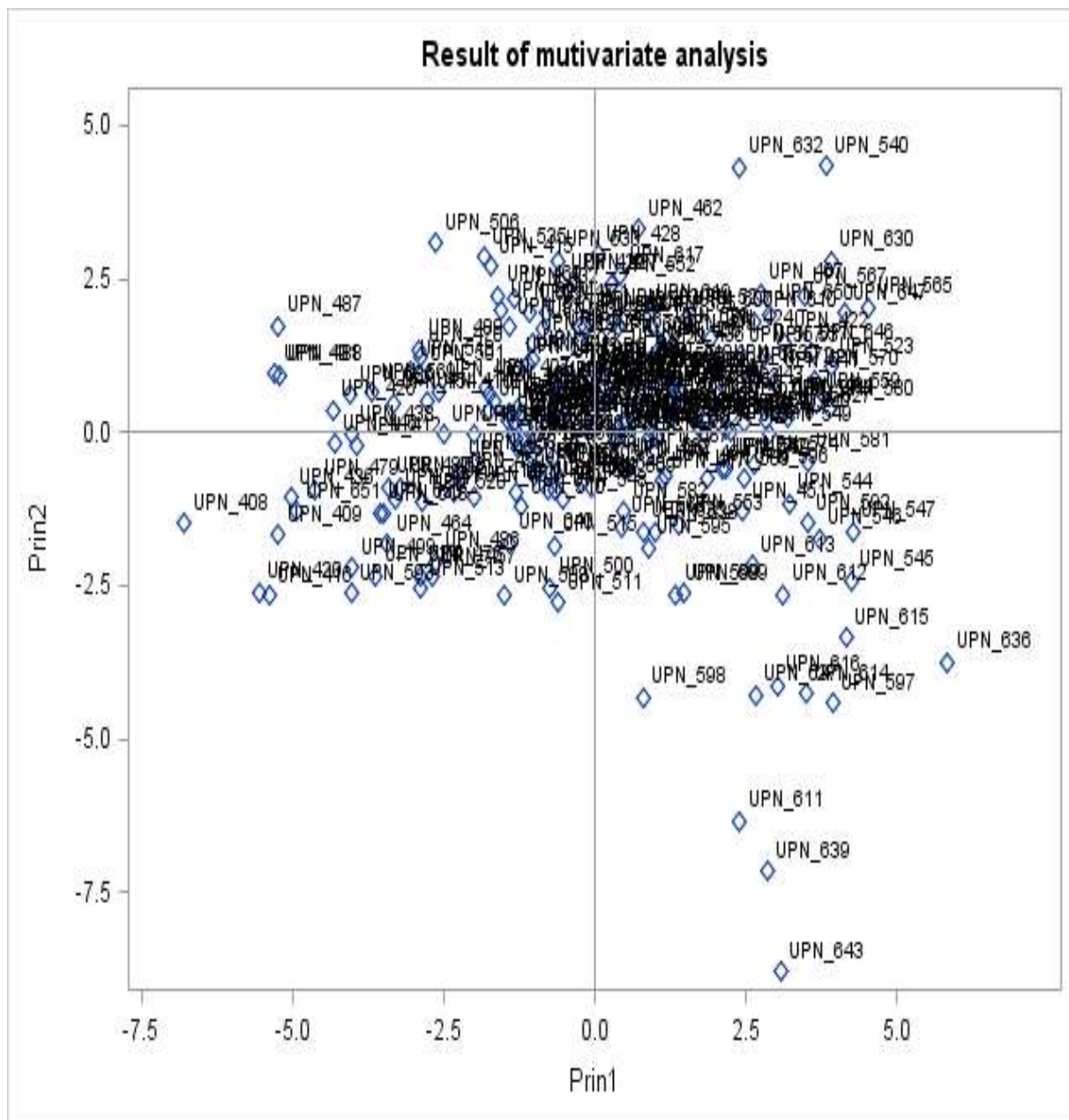


Figure 2: distributions of genotypes across the principal component 1&2 axis

CONCLUSION

Results of variability indicated that there is adequate genetic variability among the genotypes studied. Characters such as grain yield per plant, biomass and number of tillers showed high genotypic and phenotypic coefficient variance whereas days to 50% flowering, plant height, panicle length and 1000 grain weight showed high heritability likewise number of grains per panicle, biomass, leaf area, days to 50% flowering and plant height showed high genetic advance suggesting a scope for improvement of grain yield through selection. Dendrogram cluster analysis based on quantitative traits classified 240 rice genotypes including an improved variety (FARO44) into four groups (A, B, C and D) which

detected the divergences among genotypes. The principal component analysis revealed that the first three components (PC1, PC2 and PC3) accounted for 64.78% of the total cumulative variability among the traits studied confirmed the presence of ample genetic diversity for use in improvement.

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