Original Research Article

GENETIC DIVERSITY STUDIES FOR YIELD AND QUALITY TRAITS IN FOXTAIL MILLET (*Setaria italica* (L.) Beauv.)

Abstract

Field experiment was conducted using 64 genotypes of foxtail millet to assess the genetic divergence for 24 metric traits in *Kharif* 2020. The ANOVA revealed existence of highly significant variation for all the traits examined. Principal component analysis identified nine principal components with eigen value more than one and 76.59% of cumulative variance. The 2D and 3D plots indicated that genotypes SiA 60, SiA 3570, SiA 3914, SiA 3222, SiA 3629, SiA 4089, SiA 3561 and SiA 2681 were divergent for yield and quality traits. Ward's minimum variance method were grouped 64 genotypes into five clusters. The inter cluster distance between cluster I and cluster III (34.00) was maximum followed by cluster III and cluster V (31.13), therefore hybridization between the genotypes of these two clusters can result in better hybrids.

Keywords: Foxtail millet, Genetic divergence, Principal component analysis, Wards' minimum variance

Introduction:

Foxtail millet is the second-most widely cultivated species of millet after finger millet and the most important crop in East Asia (Reddy *et al*., 2006). It is an annual, self-pollinated, diploid (2n = 2x = 18), C_4 monocot belonging to the family, *Poaceae* with a genome size of ~ 515 Mb (Li and Brutnell, 2011) that offers ample scope for molecular and genetic research as a model crop to understand deeper insights into plant biology. Millets are grown for grain, which is used for human consumption and also as animal, poultry, cage birds feed and its straw is used as fodder. Millets are rich in vitamins, minerals, Sulphur-containing amino acids and phytochemicals, and hence are termed as Nutri-cereals. The foxtail millet grain is (per 100g) rich in protein (10.56 g) and iron (3.02 mg) as compared to rice (7.9 g protein and 1.8 mg Fe), Zn (2.46 mg), Mn (1.54 mg), Ca (23.94 mg) and Mg (77.65 mg) (Srilatha *et al*., 2020). It is also known as Chinese millet, Giant Setaria, German millet, Hungarian millet, Italian millet and Rala millet. Millets are nutritionally comparable or even superior to staple cereals such as rice and wheat (Gopalan *et al*., 2004).

The success of any plant breeding programme largely depends on the existence of diversity among the genotypes (Allard, 1960). In order to identify diverse genetic resources, precise information on the nature and degree of genetic diversity is required which in turn helps the plant breeders in selecting the parents for targeted hybridization (Reddy *et al*., 2015). Thus, genetic divergence studies enable us to assess the extent of genetic diversity prevalent in the genetic resources utilized for study and characterization of the genotypes can provide pivotal information for crop breeding and management of genetic resources. Clustering these genotypes is useful to identify genotypes with diverse traits, which can be useful in breeding programmes (Sapkota *et al.*, 2016).

Material and Methods

Sixty four germplasm collections of foxtail millet obtained from Nandyal, AICRP-Bangalore, ICRISAT were evaluated in 8×8 Square lattice design with three checks *viz*., Srilakshmi, Lepakshi and Krishnadevaraya during *Kharif*, 2020-21 at Agricultural college farm, Bapatla. Each genotype was raised in three rows of four meter length with a spacing of 22.5 x 10 cm between and within the rows, respectively. Standard agronomic practices and recommended fertilizer doses were applied for normal crop growth. Data recorded from the total 24 traits including five yield component characters and eight grain quality parameters. Single plant observations were recorded on five plants selected at random per genotype per replication and their means were used for the analysis. However observations on, days to 50% flowering and days to maturity were recorded on plot basis. To group the 64 genotypes along with three checks into various clusters, agglomerative hierarchical cluster analysis was followed. Principal component scores for genotypes were used as an input for clustering using Ward's minimum variance method. The tree like structure called dendrogram was constructed based on Euclidean^2 distance computed from PCA scores of genotypes. Genetic divergence analysis was done by hierarchical cluster analysis for 64 foxtail millet genotypes and these genotypes were grouped into five clusters using the Ward's minimum variance (Anderberg, 1993).

Results and discussion

The analysis of variance for 24 metric traits in 64 foxtail millet genetic resources (Table 1) revealed existence of ample genetic variation in the material, an important pre-requisite that paved way for further diversity analysis. The results revealed that nine canonical roots accounted for 76.594 per cent of total divergence. PC_1 contributed maximum towards divergence (18.056 %) with eigen value of 4.334. The second, third and fourth canonical vectors contributed 12.019%, 9.374% and 8.591% to total divergence, respectively. The remaining canonical roots *viz.*, fifth, sixth, seventh, eighth and ninth accounts for 7.270 %, 6.531 %, 5.410 %, 5.003 % and 4.340 % respectively to total variability. The canonical root values, per cent of variation and cumulative variation elucidated for 64 foxtail millet genetic resources were presented in Table 2.

The PCA scores for foxtail millet genotypes in the first three principal components were computed. Principal components I, II and III were considered as three axes as X, Y and Z and squared distance of each genotype from these three axes were calculated and presented in Table 3. The analysis identified the maximum contributing variables *i.e.*, panicle length followed by number of productive tillers per plant, zinc content, protein content, harvest index, grain yield per plant, plant height, SCMR at maturity, phosphorus content and antioxidant activity in PC_1. It is important to study the variance as the relative contribution than the signs (indicative of direction) in principal component analysis.

Two dimensional (2D) and three dimensional (3D) illustrations (Fig. 1 and 2) were constructed by plotting the mean values of vectors. The 2D and 3D plots indicated that genotypes, SiA 3222, SiA 60, SiA 3570, SiA 3914, SiA 3629, SiA 4089 and SiA 2681, were divergent for yield and quality traits. Hence, hybridization between these diverse genotypes can be suggested.

Hierarchical cluster analysis (using Ward's method) for the 64 foxtail millet genotypes resulted in five clusters and the clusters are formed using $Euclidean^2$ distances (Table 4 and Fig 3). Among all the clusters, cluster I was the largest containing 29 genotypes followed by cluster II containing ten genotypes, cluster IV with nine genotypes, cluster V with nine genotypes and cluster III with seven genotypes. The estimate of intra and inter cluster distance (Table 5) revealed that the Intra cluster $Euclidean^2$ distance values are ranged from 15.09 for cluster II to 19.80 for cluster IV. The inter cluster Euclidean^2 distances varied from 17.02 (between cluster I and cluster V) to 34.00 (between cluster I and III).

Cluster means were computed for the 24 characters studied by Ward's minimum variance method are presented in Table 6. Cluster I registered high mean values for plant height (135.24 cm) followed by weight of five panicles (46.67 g), SCMR at maturity (21.10), flag leaf blade width (1.97 cm), copper content (1.92 mg/100 g) and phosphorus content (0.33 g/100 g). Cluster II reported high mean values for days to 50% flowering (51.60), panicle length (23.12 cm), number of productive tillers per plant (3.89), test weight (2.76 g), panicle exertion (16.73 cm), peduncle length (33.02 cm), iron content (4.04 mg/100 g), calcium content (25.96 mg/100 g) and grain yield per plant (15.82 g). Cluster III consist highest mean values for days to maturity (85.14) and flag leaf blade length (33.17 cm). Cluster IV reported highest mean values for panicle width (1.48 cm) and antioxidant activity (55.55 mg AAE/100 g). It also registered desired lowest mean values for days to 50 % flowering (50.14) and days to maturity (82.84). Cluster V registered highest mean values for harvest index (44.16%), SCMR at 45 days (52.12), zinc content (3.46 mg/100 g), manganese content (3.31 mg/100 g) and protein content (14.10 %)

Conclusion

The 2D and 3D plots indicated that genotypes, SiA 3222, SiA 60, SiA 3570, SiA 3914, SiA 3629, SiA 4089 and SiA 2681, were divergent for yield and quality traits. Hence, hybridization between these diverse genotypes can be suggested. But simultaneous selection for yield and quality traits may not be possible and balanced selection criteria should be followed depending on the objective.

Based on Hierarchical cluster analysis (using Ward's method) the inter cluster distance between cluster I and cluster III (34.00) was maximum followed by cluster III and cluster V (31.13), therefore hybridization between the genotypes of these two clusters can result in better hybrids.

The genotypes from the cluster II *viz.*, SiA 92, SiA 1280, SiA 3085, SiA 3353, SiA 3625, SiA 3629, SiA 3669, SiA 3642, SiA 3574 and SiA 4154 can be used for yield improvement programmes as this cluster recorded high mean values for panicle length, number of productive tillers per plant, test weight and grain panicle exertion which are yield contributing characters. Cluster I and cluster V reported high mean values for quality traits *viz.*, copper content, phosphorus content, zinc content, manganese content and protein content.

Table 1. Analysis of variance for 24 morpho-physiological and biochemical characters studied in foxtail millet (*Setaria italica* (L.) Beauv.)

S. No.	Source of variation	d.f.	DFF	PH (cm)	PL (cm)	PW (cm)	NPT	DM	TW (g)	WP (g)	H.I (%)	SCMR at 45 Days	SCMR at maturity	FLL (cm)
Mean sum of squares														
1	Replications	1	4.206	0.600	3.686	0.000	0.030	-0.104	0.001	1.472	3.930	0.635	1.238	2.702
2	Treatments (unadjusted)	63	19.454**	341.000**	34.873**	0.150**	1.362**	34.211*	0.208**	72.106**	94.376**	63.691**	34.514**	33.049**
3	Blocks within Repllicated (adj)	14	6.232	111.100	7.149	0.012	0.151	32.183	0.036	32.805	4.148	5.397	2.871	18.429
4	Intrablock Error	49	5.028	149.98	6.965	0.008	0.245	15.206	0.013	25.850	2.573	5.586	2.467	14.714
5	Total	127	12.311	239.275	20.804	0.079	0.787	26.384	0.112	49.370	48.298	34.350	18.399	24.124

* Significant at 5% and 1% levels, respectively

S. No.	Source of variation	d.f.	FLW (cm)	PE (cm)	PDL (cm)	Zn (mg/ 100g)	Cu (mg/ 100g)	Fe (mg/ 100g)	Mn (mg/ 100g)	Protein (%)	Ca (mg/ 100g)	P (g/ 100g)	AOA (% DPPH Activity)	GYPP (g)
			Mean sum of squares											
1	Replications	1	0.048	1.501	5.857	0.307	0.030	0.027	0.014	0.002	0.226	0.000	0.055	0.775
2	Treatments (unadjusted)	63	0.188**	18.745**	45.042**	0.548**	0.298**	0.774**	0.632**	7.578**	23.788**	0.006**	72.193**	21.355**
3	Blocks within Repllicated (adj)	14	0.012	0.217	7.313	0.049	0.014	0.041	0.031	0.686	1.133	0.000	7.415	4.953
4	Intrablock Error	49	0.010	0.652	3.959	0.040	0.013	0.070	0.0260	0.439	1.651	0.000	8.056	5.682
5	Total	127	0.099	9.586	24.723	0.295	0.155	0.416	0.327	4.004	12.564	0.003	39.738	13.338

* Significant at 5% and 1% levels, respectively

DFF- Days to 50 per cent flowering, PH- Plant height, PL- Panicle length, PW- Panicle width, , NPT- Number of productive tillers per plant , DM- Days to maturity, TW-Test weight, WP- Weight of five panicles, H.I- Harvest index, FLL- Flag leaf blade length, FLW- Flag leaf blade width, PE- Panicle exertion, PDL- Peduncle length, Zn- Zinc content, Cu- Copper content, Fe- Iron content, Mn- Manganese content, Ca- Calcium content, AOA- Antioxidant activity, GYPP- Grain yield per plant.

Table 2. Canonical vectors for 24 characters in 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

S. No.	Parameter	PC_1	PC_2	PC_3	PC_4	PC_5	PC_6	PC_7	PC_8	PC_9
1	Eigenvalue (Root)	4.334	2.884	2.250	2.062	1.745	1.567	1.299	1.201	1.042
2	Variability (%)	18.056	12.019	9.374	8.591	7.270	6.531	5.410	5.003	4.340
3	Cumulative variability %	18.056	30.075	39.449	48.040	55.310	61.841	67.252	72.255	76.594
S. No.	**Character**									
1	Days to 50% flowering	0.271	0.125	0.125	0.115	0.042	0.222	0.348	0.077	0.172
2	Plant height (cm)	0.147	0.124	0.220	-0.068	0.049	0.352	-0.338	0.053	-0.414
3	Panicle length (cm)	0.353	0.122	0.155	-0.194	-0.032	-0.276	0.123	-0.002	-0.004
4	Panicle width (cm)	-0.029	-0.144	0.437	0.091	-0.270	-0.042	-0.166	0.311	-0.089
5	Number of productive tillers per plant	0.278	0.353	-0.055	0.077	-0.001	0.017	0.068	-0.073	-0.312
6	Days to maturity	0.297	0.185	0.181	0.085	0.099	0.106	0.255	0.124	-0.088
7	Test weight (g)	-0.134	0.075	-0.086	0.271	-0.384	0.395	0.205	0.007	0.058
8	Weight of five panicles (g)	-0.067	0.125	0.364	-0.113	-0.260	0.179	-0.029	0.394	0.090
9	Harvest index (%)	0.231	-0.196	-0.173	-0.094	-0.218	0.135	-0.104	0.324	0.397
10	SCMR at 45 days	-0.150	0.051	0.330	0.019	0.427	0.145	-0.117	0.101	0.227
11	SCMR at maturity	0.133	0.242	-0.331	0.056	0.003	0.192	-0.139	0.150	0.217
12	Flag leaf blade length (cm)	0.283	0.021	0.038	-0.127	-0.162	-0.191	0.287	-0.060	0.189
13	Flag leaf blade width (cm)	0.303	-0.216	0.071	0.177	-0.007	0.199	0.030	-0.213	0.108
14	Panicle exertion (cm)	-0.093	0.224	0.211	-0.242	-0.202	-0.049	-0.315	-0.324	0.241
15	Peduncle length (cm)	0.072	0.312	-0.147	-0.181	-0.326	0.129	-0.225	-0.316	0.130
16	Zinc content (mg/100g)	0.270	-0.258	0.076	0.094	-0.085	-0.066	-0.266	-0.180	-0.154
17	Copper content (mg/100g)	-0.081	-0.018	0.190	0.407	-0.340	-0.349	0.001	-0.079	0.040
18	Iron content (mg/100g)	-0.159	0.318	-0.110	-0.162	0.116	-0.162	-0.091	0.288	0.173
19	Manganese content (mg/100g)	0.290	-0.100	-0.054	0.113	-0.046	-0.286	-0.292	0.187	0.155
20	Protein content (%)	0.258	-0.272	-0.120	-0.076	0.247	0.045	-0.199	0.113	0.087
21	Calcium content (mg/100g)	-0.122	0.195	-0.090	0.454	0.094	-0.239	0.005	0.139	0.082
22	Phosphorus content (g/100g)	0.079	0.139	-0.201	0.441	0.003	0.104	-0.338	0.064	-0.090
23	Antioxidant activity (% DPPH Activity)	0.022	0.034	-0.276	-0.241	-0.236	-0.169	0.049	0.366	-0.437
24	Grain yield per plant (g)	0.192	0.387	0.167	0.084	0.175	-0.211	-0.107	-0.023	0.060

Table 3. Mean values of canonical vectors for 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

S. No.	Genotype	PCA I	PCA II	PCA III
		X Vector	Y Vector	Z Vector
1	**SiA 60**	52.676	19.569	7.882
2	**SiA 78**	50.069	15.929	12.490
3	**SiA 92**	47.948	11.779	10.097
4	**SiA 3613**	40.713	14.465	9.221
5	**SiA 326**	45.214	13.662	18.764
6	**SiA 723**	47.675	18.124	5.946
7	**SiA 3563**	45.898	12.073	7.607
8	**SiA 1280**	46.813	20.726	-1.136
9	**SiA 1331**	45.307	14.384	9.673
10	**SiA 3639**	45.856	12.348	11.225
11	**SiA 3569**	41.608	17.655	8.215
12	**SiA 3553**	47.021	17.461	5.298
13	**SiA 2681**	42.505	10.466	7.694
14	**SiA 2851**	45.162	12.211	15.370
15	**SiA 3085**	50.022	17.460	5.785
16	**SiA 3088**	42.071	20.480	6.616
17	**SiA 3640**	36.182	15.160	10.771
18	**SiA 3159**	41.596	19.197	12.209
19	**SiA 3549**	42.567	14.406	9.977
20	**SiA 3552**	42.942	17.394	9.881
21	**SiA 3577**	46.759	14.944	6.388
22	**SiA 3326**	46.251	12.762	4.992
23	**SiA 3330**	44.873	12.865	7.388
24	**SiA 3353**	43.043	17.254	17.075
25	**SiA 3556**	38.284	18.728	12.277
26	**SiA 3390**	43.748	13.971	7.596
27	**SiA 3396**	42.164	15.916	6.320
28	**SiA 3561**	40.960	23.488	13.709
29	**SiA 3597**	42.612	13.735	11.715
30	**SiA 3422**	40.136	17.120	10.370
31	**SiA 3625**	44.177	18.532	6.666
32	**SiA 3570**	57.055	12.272	7.604
33	**SiA 3630**	57.152	12.263	7.502
34	**SiA 3588**	42.923	17.592	9.766

Table 3. Contd…

S. No.	Genotype	PCA I	PCA II	PCA III
		X Vector	Y Vector	Z Vector
35	**SiA 3629**	40.252	27.558	13.584
36	**SiA 3669**	48.986	18.458	7.853
37	**SiA 3619**	40.713	19.345	8.189
38	**SiA 3566**	45.951	10.474	11.280
39	**SiA 3770**	36.801	21.392	15.154
40	**SiA 3855**	43.115	15.636	8.913
41	**SiA 3872**	46.160	10.646	9.625
42	**SiA 3222**	28.227	16.718	4.955
43	**SiA 3891**	38.526	16.024	9.053
44	**SiA 3642**	50.247	13.626	10.397
45	**SiA 3914**	50.797	8.868	7.528
46	**SiA 3915**	39.387	20.178	8.760
47	**SiA 3928**	47.525	19.156	4.135
48	**SiA 3934**	43.606	14.610	8.791
49	**SiA 4032**	39.736	20.261	11.973
50	**SiA 3554**	40.440	17.658	11.612
51	**SiA 3574**	44.204	22.672	9.274
52	**SiA 4089**	44.851	7.076	15.434
53	**SiA 3596**	46.392	19.000	11.336
54	**SiA 4134**	41.714	17.668	6.766
55	**SiA 4145**	46.551	21.934	13.798
56	**SiA 3284**	42.707	13.739	13.411
57	**SiA 3643**	50.718	15.677	6.527
58	**SiA 3562**	43.462	14.962	12.984
59	**SiA 3568**	41.038	18.802	9.996
60	**SiA 3912**	40.534	18.861	10.012
61	**SiA 4154**	44.271	22.939	9.994
62	**Lepakshi**	48.140	17.719	8.557
63	**Krishnadevaraya**	47.902	24.419	11.952
64	**Srilakshmi**	45.763	23.720	9.025

Table 4. Clustering pattern estimated by Ward's minimum variance method in 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

Cluster. No	No. of genotypes	Name of genotype(s)	Source
I	29	SiA 60, SiA 78, SiA 326, SiA 2851, SiA 3088, SiA 3159, SiA 3552, SiA 3326, SiA 3330, SiA 3556, SiA 3390, SiA 3396, SiA 3561, SiA 3588, SiA 3619, SiA 3566, SiA 3770, SiA 3891, SiA 4032, SiA 3554, SiA 3596, SiA 4145, SiA 3284, SiA 3643, SiA 3562, SiA 3568, SiA 3912, Krishnadevaraya, Srilakshmi	Nandyal, AICRP-Bangalore, ICRISAT
II	10	SiA 92, SiA 1280, SiA 3085, SiA 3353, SiA 3625, SiA 3629, SiA 3669, SiA 3642, SiA 3574, SiA 4154	Nandyal, ICRISAT, AICRP-Bangalore
III	7	SiA 3613, SiA 3639, SiA 2681, SiA 3640, SiA 3597, SiA 3855, SiA 3915	AICRP-Bangalore, Nandyal
IV	9	SiA 723, SiA 1331, SiA 3569, SiA 3422, SiA 3222, SiA 3934, SiA 4089, SiA 4134, Lepakshi	Nandyal, AICRP-Bangalore, ICRISAT
V	9	SiA 3563, SiA 3553, SiA 3549, SiA 3577, SiA 3570, SiA 3630, SiA 3872, SiA 3914, SiA 3928	AICRP-Bangalore, ICRISAT

Table 5. Average intra and inter-cluster Eucledian^2 values among the five clusters in 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

Cluster No.	I	II	III	IV	V
I	16.30	20.41	34.00	25.21	17.02
II		15.09	19.45	17.16	20.42
III			16.74	17.39	31.13
IV				19.80	24.64
V					18.76

Table 6. Cluster means of five clusters estimated by Ward's minimum variance method in 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

Character	Cluster number				
	I	II	III	IV	V
Days to 50% flowering	50.14	**51.60**	50.79	48.61	51.44
Plant height (cm)	**135.24**	117.62	103.13	112.45	129.85
Panicle length (cm)	21.09	**23.12**	19.24	19.46	22.49
Panicle width (cm)	1.43	1.35	1.30	**1.48**	1.30
Number of productive tillers per plant	3.84	**3.89**	3.15	3.68	3.50
Days to maturity	82.84	84.72	**85.14**	80.72	84.00
Test weight (g)	2.70	**2.76**	2.67	2.46	2.58
Weight of five panicles (g)	**46.67**	43.77	41.33	44.94	43.16
Harvest index (%)	29.39	33.57	32.00	32.21	**44.16**
SCMR at 45 days	47.79	43.03	46.01	44.56	**52.12**
SCMR at maturity	**21.10**	20.74	15.98	20.34	20.76
Flag leaf blade length (cm)	32.79	32.69	**33.17**	27.61	32.36
Flag leaf blade width (cm)	**1.97**	1.90	1.75	1.82	1.93
Panicle exertion (cm)	14.98	**16.73**	12.55	13.67	13.65
Peduncle length (cm)	29.15	**33.02**	24.40	25.93	27.37
Zinc content (mg/100g)	3.37	3.42	3.12	3.13	**3.46**
Copper content (mg/100g)	**1.92**	1.88	1.82	1.86	1.56
Iron content (mg/100g)	3.59	**4.04**	3.89	3.63	3.38
Manganese content (mg/100g)	2.84	3.05	2.80	2.91	**3.31**
Protein content (%)	13.63	12.97	13.47	13.13	**14.10**
Calcium content (mg/100g)	24.03	**25.96**	23.91	23.72	24.31
Phosphorus content (g/100g)	**0.33**	0.30	0.28	0.32	**0.33**
Antioxidant activity (mg AAE/100g)	48.83	44.10	44.80	**55.55**	48.74
Grain yield per plant(g)	14.19	**15.82**	11.97	14.49	13.96

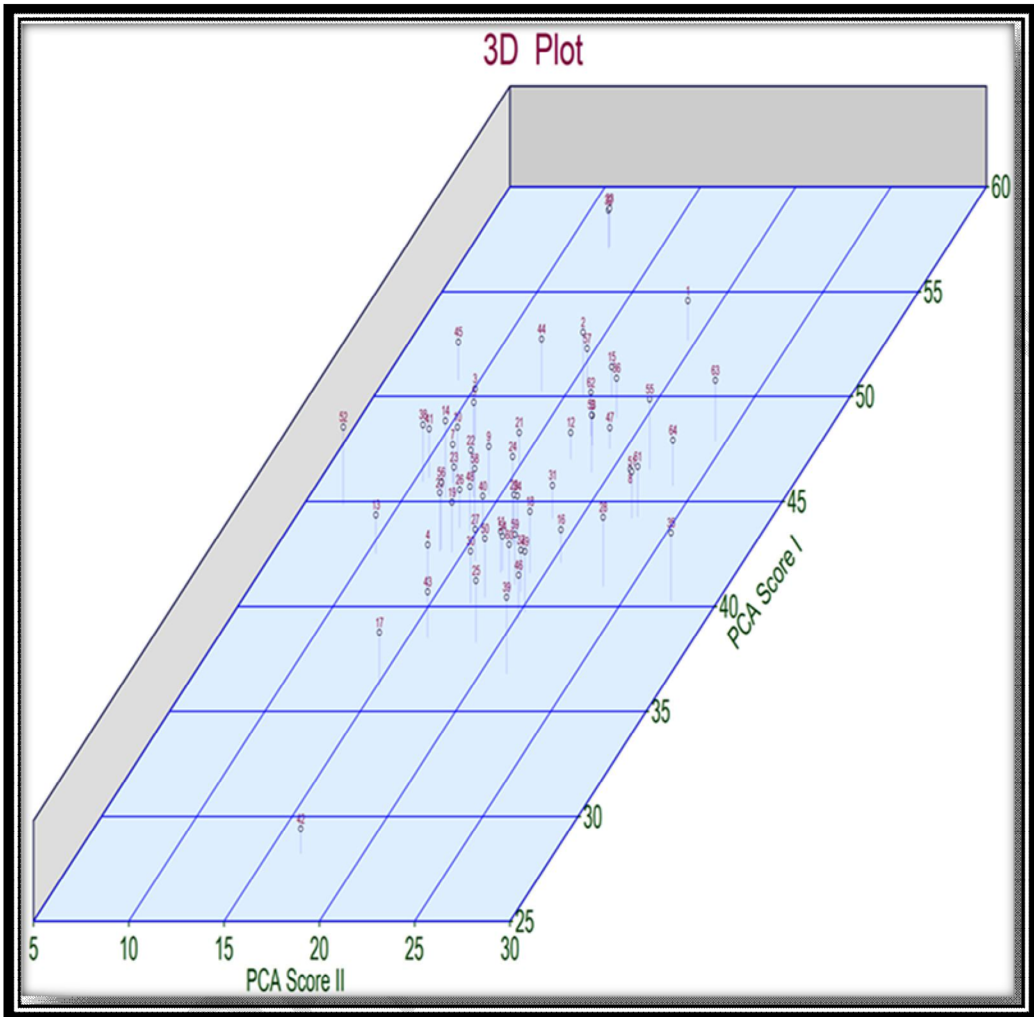

Fig. 1. Three dimensional (3D) graph showing relative positions based on PCA scores in 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

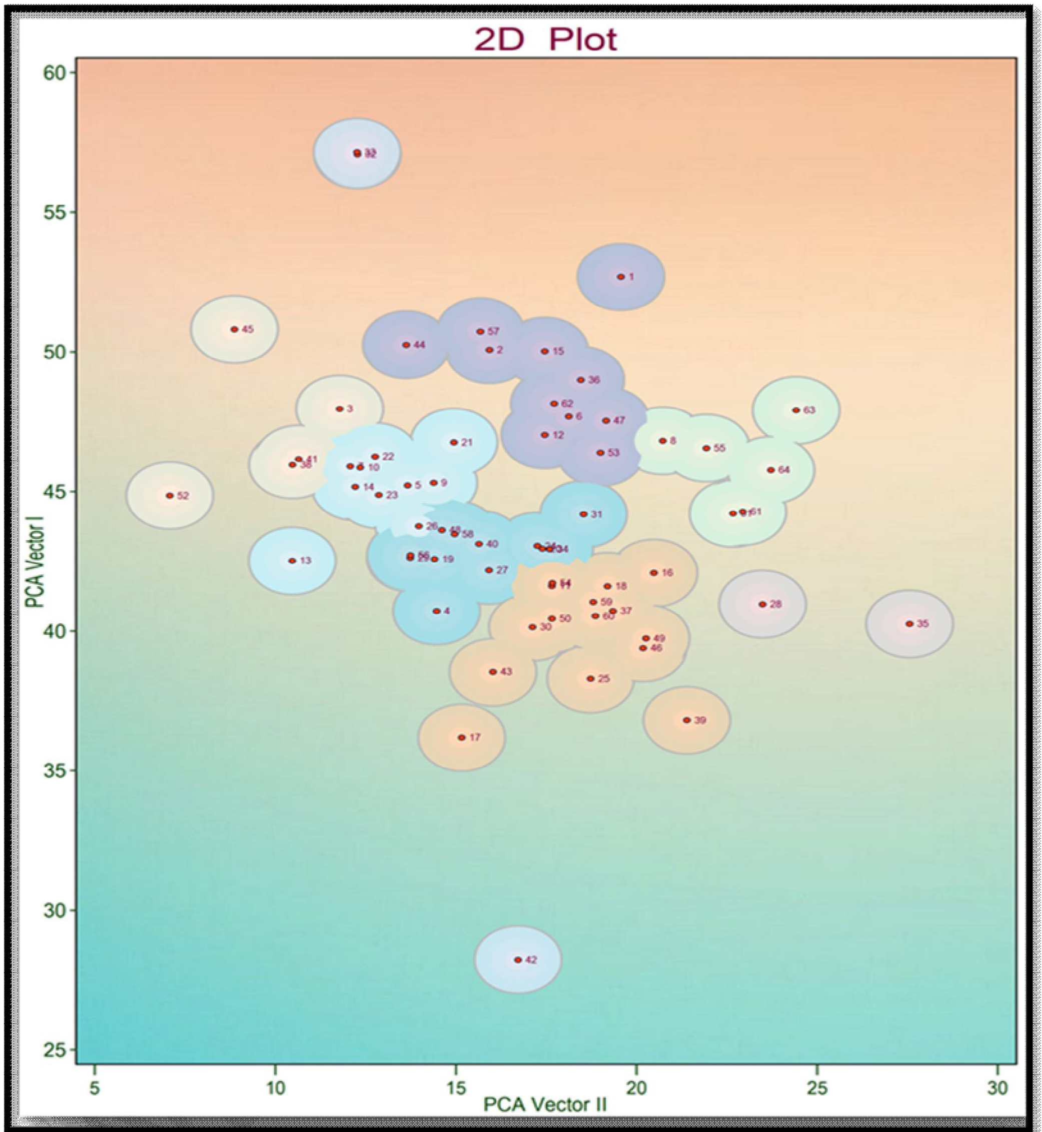

Fig. 2. **Two dimensional (2D) graph based on PCA scores showing relative positions of 64 genotypes in foxtail millet (*Setaria italica* (L.) Beauv.)**

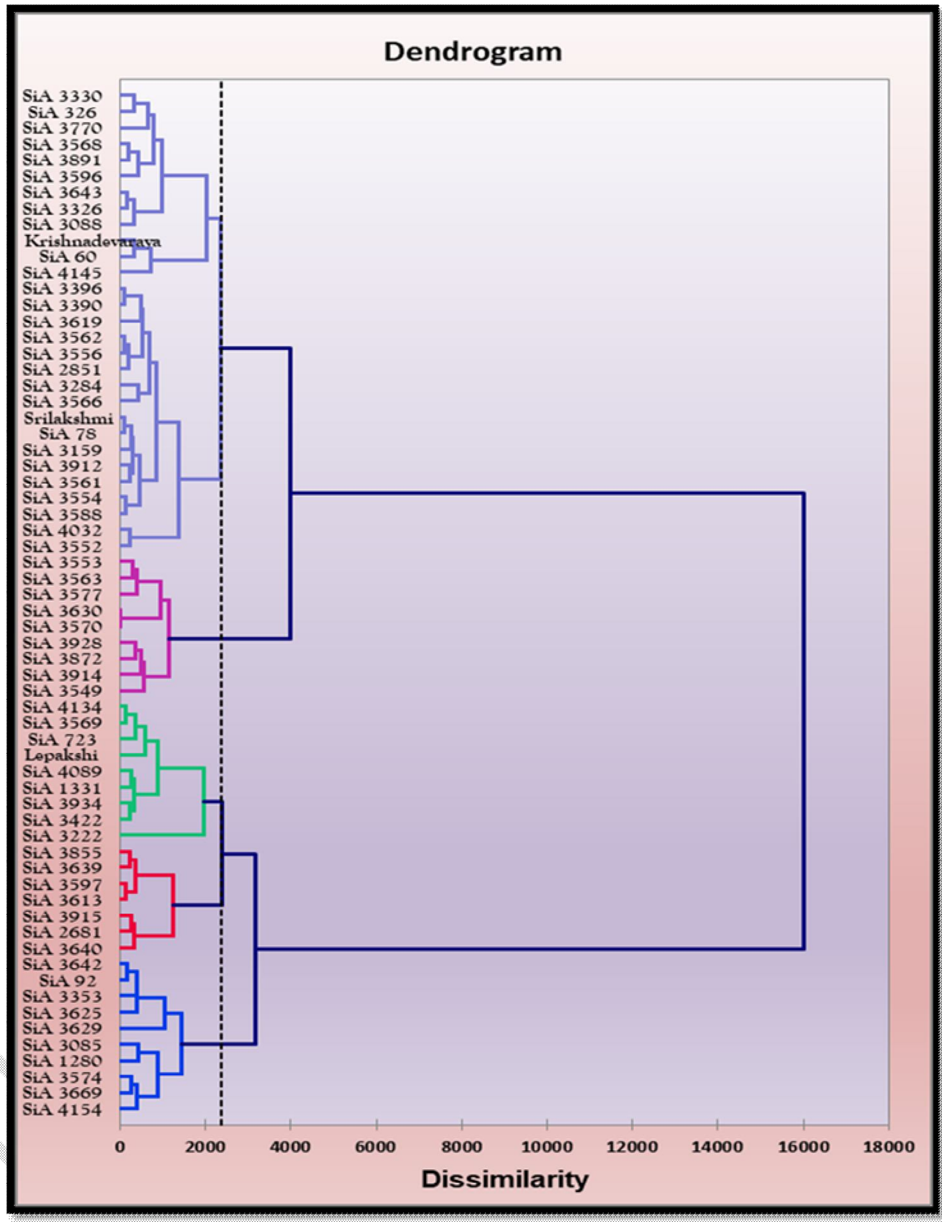

Fig. 3. Dendrogram showing relationship in five clusters based on Eucledian^2 distance in 64 genotypes of foxtail millet (*Setaria italica* (L.) Beauv.)

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