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MAIZE (*ZEA MAYS* L.) GERMPLASM AGRO-MORPHOLOGICAL CHARACTERIZATION BASED ON DESCRIPTIVE, CLUSTER AND PRINCIPAL COMPONENT ANALYSIS

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Abstract

Broad genetic makeup is always less vulnerable to various biotic and abiotic stresses. To identify and maintain diverse genetic base, the screening of collections is carried out through different marker systems. In the current investigation a total of 153 maize genotypes including 150 accessions from China, Japan and Pakistan, and 3 check varieties were characterized for 34 agronomic and morphological traits. These traits were comprised of ten qualitative and twenty-four quantitative variables which were recorded using augmented design at Plant Genetic Resources Institute (PGRI), National Agricultural Research Center (NARC) Islamabad, Pakistan during spring-2012. Descriptive statistical analysis of the data reflected that maximum variance of 3394.8 was detected in grain yield per plant followed by 1243.3 noted in plant height. The multivariate analyses showed the maximum Euclidean distance of 13.9 was noted between 15329 and 14909, 15329 and 14959 and 24690 and 14959. Cluster analysis distributed the whole genotypes into 5 clusters indicating their broad genetic base. Principal component analysis revealed that 7 of the principal components with an Eigenvalue of more than 1 accounted for 70.56 percent of the overall variations. The identification of high level of genetic diversity during the current study could be implied for maize germplasm characterization, conservation and further improvement in maize breeding.

Key Words: Maize germplasm, Genetic divergence, Qualitative traits, Quantitative variables, Multivariate analysis.

Introduction

Maize (*Zea mays* L.) is considered as a miracle crop and one of the prominent crop species on the basis of its role in world's economy. It contributes billions of dollars worldwide to the annual revenue (Mabberley, 2008). It is leading in production (861 million tons), followed by wheat (655 million tons) (Anon., 2014) and 3rd in area under cultivation among all cereal crops. Area under maize cultivation throughout the world is 162 million hectares with average yield of 5195 kg ha^{-1} (Anon., 2012). In Pakistan maize is 3rd in production (4.63 million tons) after wheat (24.23 million tons) and rice (5.54 million tons) (Farooq, 2013). It occupies 8.98 % of the total cereal cropped area and 13.46 % of the total cereal production throughout the country, mainly grown as autumn (Kharif) crop, usually by low source farmers with marginal land (Chaudhry, 1994). In Punjab it is also grown as a spring (Rabi) crop because of the active interest of multinational companies. To address the food security problem of the outstripping population and to decrease pressure on wheat and rice availability, it is extremely vital to increase the maize production, because it is not only important for the survival of human as food and fodder for cattle but also has bright future in the form of bio-fuel, as alternative source of energy. It is encouraging that the productivity of maize has been increasing for the last five years, which was 3415 kg/ha during 2008-09 and 4268 kg/ha during 2012-13 (Farooq, 2013), however there is still enough room for further improvement in its yield and productivity. Pakistan is rich in maize local germplasm and traditional landraces. To provide broad genetic base for breeding programs it is not only pivotal to conserve and manage the present genotypes but also to screen the new indigenous as well as exotic collections. Genetic diversity is the base for crop improvement (Iqbal *et al*., 2014), which is elucidated through different marker systems such as agro-morphological, biochemical and molecular markers. Among these the agro-morphological characterization is considered as the initial step (Smith & Smith, 1989; Khan *et al*., 2014). The magnitude of genetic diversity has been calculated by researchers in other crops as well, such as in *Oryza sativa* L. (Rabbani *et al*., 2010), sesame (Akbar *et al*., 2011), maize (Iqbal *et al*., 2014), safflower (Shinwari *et al*., 2014) and in *Brassica juncea* L. (Ali *et al*., 2015). This experiment was aimed to elucidate the magnitude of genetic diversity in maize germplasm collected from diverse climates of China, Japan and Pakistan through agro-morphological traits, and to identify genotypes with broad genetic makeup for use in future breeding programs.

Materials and Methods

Experimental material and location: In the current study 153 genotypes of maize from diverged climatic conditions of the world i.e. Chinese (3), Japanese (18) and Pakistani (132) including three commercial varieties named Agaiti-2002, Sadaf and Sahiwal-2002 were characterized under field conditions during spring-2012. Seed material was obtained from gene bank of Plant Genetic Resources Institute (PGRI), National Agricultural Research Center (NARC) Islamabad, Pakistan (33° 33' N and 73° 06'E). Data were recorded on 34 agro-morphological traits (10 qualitative and 24 quantitative) using the standard descriptors formulated by IBPGR (Anon., 1991). The data recorded on 24 quantitative traits were days to field germination (DFG), days to tassel (DT), days to pollen shedding (DPS), days to silk (DS),

anthesis-silking interval (ASI), plant height (PH), number of leaves per plant (NL/P), leaf length (LL), leaf width (LW), ear height (EH), number of ears per plant (NE/P), days to ear leaf senescence (DELS), days to harvest (DH), field weight per plot (FW/Pl), ear length (EL) , ear diameter (ED), number of kernel rows (NKR), number of kernels per row (NK/R), kernel length (KL), kernel width (KW), cob diameter (CD), 1000 kernel weight (1000KW), harvest grain moisture (HGM), grain yield per plant (GY/P) and 10 qualitative traits included leaf orientation (LO), pubescence of culm (PbC), tassel type (TT), tassel size (TS), husk cover (HC), ear damage (EDa), kernel row arrangement (KRA), kernel type (KT), kernel color (KC) and cob color (CC).

Data Analysis: The data recorded on quantitative traits were averaged and analyzed for simple statistical approaches i.e. mean, minimum, maximum, standard deviation and coefficient of variation to determine the extent of genetic diversity among the studied genotypes. Similarly the data recorded from qualitative traits were subjected into different classes and frequency percentage was found. Simple correlation coefficients between all pairs of quantitative traits were obtained through the way of Steele & Torrie (1980) using mean values. Through numerical taxonomic approaches the data of the whole agro-morphological characters was analyzed by applying cluster analysis and principal component analysis (PCA), according to the Sneath & Sokal (1973). For the eradication of scaling differences, the means of each trait were standardized using Z-scores before carrying out the cluster analysis and PCA. Estimates of Euclidean distance coefficients were carried out for all pairs of genotypes. The resulted matrices of Euclidean dissimilarity coefficient were applied to assess the genetic relationships between maize genotypes with a cluster analysis through complete linkage way i.e. NTSYS-pc, version 2.1 (Applied Biostatics Inc., USA). The same data matrices were utilized to carry out the PCA. Scatter plots of the first three principal components were made to determine the graphical display of the pattern of genetic diversity among the maize genotypes (Statistica, version 6.0).

Results

Descriptive statistics of the data recorded reflected high level of variation for the quantitative characters. All the traits were more or less, directly or indirectly, positively or negatively added to the yield and possessed key genetic status during the identification of productive genotypes. The basic statistical data (mean, minimum, maximum, standard deviation, coefficient of variation and variance) for every quantitative trait was calculated among all the genotypes (Table 1). Pattern of variability among the genotypes was different for various agro-morphological traits. Maximum variability was observed in grain yield per plant (2757.3) followed by plant height (1149.2), 1000 kernel weight (1052.8), ear height (301.9), leaf length (272.6) and days to harvest (116.5).

Table 1. Basic statistics of the studied genotypes based on different traits during 2012.

Trait	Mean	Minimum	Maximum	SD	CV%	Variance
DFG	12.0	9.0	19.0	1.9	15.9	3.6
DT	67.4	48.0	89.0	6.9	10.3	47.8
DPS	70.0	50.0	90.0	6.7	9.6	45.5
DS	72.6	56.0	92.0	6.6	9.0	42.9
ASI	3.3	0.0	8.0	1.5	40.8	2.2
PH	110.3	41.7	206.8	33.9	30.7	1149.2
NL/P	11.2	6.2	16.6	2.0	18.1	4.1
LL	32.9	15.6	85.0	16.5	50.2	272.6
LW	5.6	3.1	11.1	1.7	30.3	2.9
EH	48.5	13.5	98.2	17.4	35.8	301.9
NE/P	1.9	1.0	5.3	0.7	35.3	0.5
DELS	96.9	77.0	119.0	8.5	8.7	71.7
DH	115.7	93.0	135.0	10.8	9.3	116.5
FW/Pl	0.1	0.01	0.3	0.1	73.5	0.003
EL	11.2	6.0	21.3	2.9	25.9	8.5
ED	3.2	2.0	4.5	0.5	15.7	0.2
NKR	11.8	7.0	16.7	1.7	14.3	2.8
NK/R	14.6	4.3	30.7	5.9	40.2	34.5
KL	7.5	4.5	11.3	1.3	16.7	1.6
KW	6.8	4.4	11.5	1.3	19.4	1.7
CD	2.2	1.3	3.2	0.3	15.4	0.1
1000KW	219.6	145.8	338.5	32.4	27.1	1052.8
HGM	23.8	13.7	37.1	4.7	19.8	22.2
GY/P	100.1	24.0	272.0	52.5	52.5	2757.3

During the current characterization of maize germplasm through agro-morphological traits highly and positive correlation was found among most of the traits (Table 2), in which the leading one was among the traits of number of days to tassel and number of days to pollen shedding (r = 0.98**) and number of days to silking (r = 0.98**) followed by a correlation value of r = 0.95** between the traits of days to tassel and days days to silk and between days to harvest and days to ear leaf senescence (r = 0.91**). Similarly among most of other traits highly significant and significant positive association was found which shows the strong interdependency of these traits on each other. A few traits also showed highly significant and negative correlation with each other like between the traits of harvest grain moisture and 1000 kernel weight (r = -0.31**), anthesis-silking interval and days to tassel (r = -0.29**), number of kernel rows and leaf length (r = -025**) and between the traits of kernel weight and kernel length (r = -0.24**) (Table 2). Euclidean dissimilarity coefficient matrices were used to portray the relationship between maize accessions and cultivars. The maximum Euclidean genetic distance of 13.9 was noted between 15329 and 14909, 15329 and 14959 and 24690 and 14959, whereas the minimum genetic distance of 3.1 was found between 15346 and 15258, and 24670 and 24669.

All 153 maize genotypes were distributed into 5 main clusters, A, B, C, D and E through cluster analysis of 34 agro-morphological traits using complete linkage method at 12.6 dissimilarity coefficient value during current study (Fig.1).

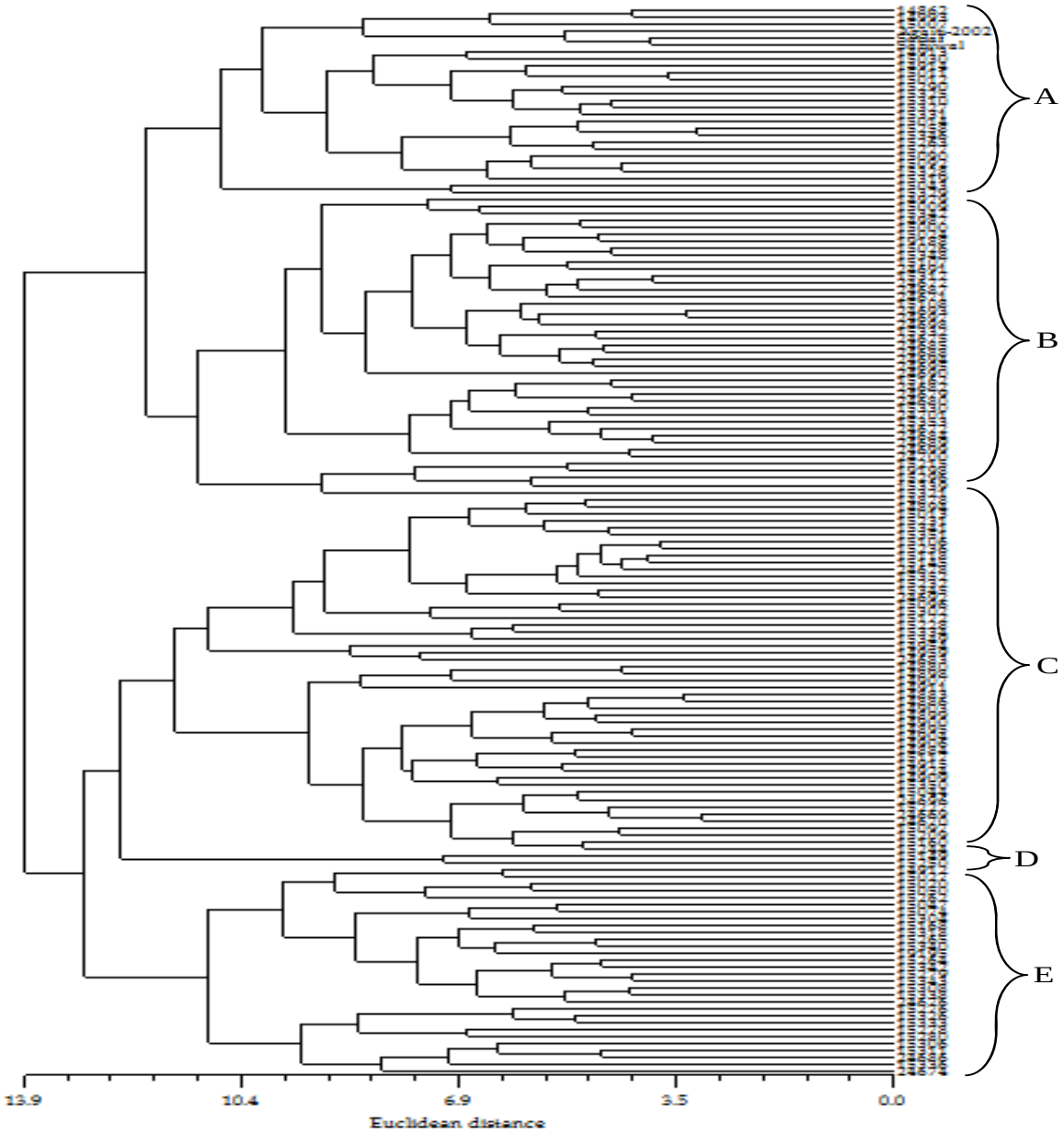

Fig.1. Dendrogram showing the genetic relationship among Chinese, Japanese and Pakistani maize genotypes based on agro-morphological traits during 2012.

Table 2. Correlation among different traits for the studied genotypes during 2012.

Trait	FGD	DT	DPS	DS	ASI	PH	NL/P	LL	LW	EH	NE/P	DELS	DH	FW/Pl	EL	ED	NKR	NK/R	KL	KW	CD	1000KW	HGM	GY/P
FGD	1.00																							
DT	0.28**	1.00																						
DPS	0.26**	0.98**	1.00																					
DS	0.23**	0.95**	0.98**	1.00																				
ASI	-0.18*	-0.29**	-0.13	0.01	1.00																			
PH	0.09	0.25**	0.25**	0.26**	0.07	1.00																		
NL/P	0.19*	0.34**	0.34**	0.35**	0.01	0.39**	1.00																	
LL	0.08	0.03	0.06	0.08	0.17*	0.16*	0.03	1.00																
LW	0.23**	0.24**	0.22**	0.21**	-0.11	0.18*	0.25**	0.42**	1.00															
EH	0.21**	0.34**	0.34**	0.35**	0.01	0.54**	0.64**	0.20*	0.30**	1.00														
NE/P	0.02	0.11	0.11	0.09	-0.08	0.01	0.07	-0.18*	0.14	0.10	1.00													
DELS	0.27**	0.79**	0.79**	0.77**	-0.19*	0.15	0.28**	-0.11	0.08	0.21**	0.08	1.00												
DH	0.25**	0.74**	0.74**	0.74**	-0.10	0.19*	0.33**	-0.17*	0.04	0.25**	0.10	0.91**	1.00											
FW/Pl	0.18*	0.31**	0.29**	0.28**	-0.12	0.24**	0.41**	0.21**	0.29**	0.41**	0.04	0.25**	0.24**	1.00										
EL	0.23**	0.36**	0.34**	0.32**	-0.16*	0.20*	0.42**	0.08	0.25**	0.40**	0.03	0.34**	0.29**	0.63**	1.00									
ED	0.17*	0.20*	0.17*	0.14	-0.20*	0.15	0.40**	0.04	0.39**	0.30**	0.13	0.12	0.19*	0.64**	0.47**	1.00								
NKR	0.12	0.15	0.10	0.07	-0.28**	0.11	0.28**	-0.25**	0.04	0.17*	0.07	0.19*	0.21**	0.35**	0.34**	0.43**	1.00							
NK/R	0.11	-0.06	-0.10	-0.10	-0.09	-0.19*	0.02	-0.03	0.18*	0.03	0.05	0.00	-0.05	0.40**	0.37**	0.39**	0.11	1.00						
KL	0.12	0.18*	0.15	0.13	-0.20*	0.11	0.22**	0.11	0.29**	0.28**	0.17*	0.08	0.07	0.40**	0.28**	0.47**	0.17*	0.37**	1.00					
KW	0.05	0.06	0.03	0.02	-0.10	0.04	0.29**	-0.06	0.04	0.13	-0.08	0.01	-0.01	0.12	0.26**	0.04	0.17*	-0.20*	-.24**	1.00				
CD	0.20*	0.15	0.12	0.10	-0.18*	0.26**	0.33**	-0.08	0.20*	0.33**	0.17*	0.11	0.19*	0.53**	0.38**	0.66**	0.49**	0.09	0.30**	0.08	1.00			
1000KW	-0.02	0.09	0.09	0.12	0.06	0.09	0.16*	-0.09	0.05	0.14	0.15	0.04	0.06	0.13	0.07	0.00	0.08	0.04	0.12	0.07	0.15	1.00		
HGM	-0.01	0.15	0.12	0.09	-0.17*	0.14	0.20*	-0.02	0.06	0.18*	0.01	0.16*	0.23**	0.14	0.06	0.18*	0.15	0.22**	0.18*	0.01	0.05	-0.31**	1.00	
GY/P	0.13	0.30**	0.27**	0.24**	-.25**	0.23**	0.23**	-0.06	0.20*	0.19*	0.20*	0.15	0.22**	0.39**	0.24**	0.23**	0.27**	0.43**	0.27**	-0.01	0.26**	0.28**	0.46**	1.00

*Significant at 0.05 (5%) probability level; **Highly significant at 0.01(1%) probability level

Table 3. Showing the share of seven PCAs in the total divergence during 2012.

	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Eigenvalue	6.33	3.02	1.91	1.63	1.57	1.36	1.11
Cumulative Eigenvalue	6.33	9.36	11.27	12.89	14.46	15.82	16.93
Total Variance (percent)	26.39	12.59	7.96	6.78	6.53	5.67	4.63
Cumulative Variance (percent)	26.39	38.99	46.95	53.73	60.26	65.93	70.56
Trait	**Eigenvectors**						
Days to field germination (DFG)	-0.383	0.011	-0.011	0.140	-0.104	0.188	-0.427
Day to tassel (DT)	-0.819	-0.487	0.033	0.122	-0.042	0.011	-0.107
Days to polle shedding (DPS)	-0.794	-0.536	-0.044	0.116	-0.061	-0.026	-0.025
Days to silk (DS)	-0.770	-0.550	-0.123	0.095	-0.089	-0.048	0.045
Anthesis-silk interval (ASI)	0.257	-0.114	-0.526	-0.108	-0.125	-0.168	0.509
Plant height (PH)	-0.419	0.047	-0.424	-0.321	0.328	-0.155	0.121
Number of leaves per plant(NL/P)	-0.626	0.179	-0.246	-0.392	0.087	0.024	0.083
Leaf length (LL)	-0.059	0.109	-0.725	0.383	0.106	0.182	-0.135
Leaf width (LW)	-0.404	0.277	-0.365	0.339	0.087	-0.031	-0.433
Ear height (EH)	-0.605	0.186	-0.435	-0.239	0.155	-0.076	0.071
Number of ears per plant (NE/P)	-0.184	0.073	0.219	0.031	0.039	-0.629	-0.210
Days to ear leaf senescence(DELS)	-0.714	-0.528	0.197	0.078	-0.148	0.082	0.102
Days to harvest (DH)	-0.714	-0.492	0.207	-0.008	-0.069	0.012	0.195
Field weight per plot (FW/P)	-0.648	0.469	-0.089	0.071	-0.198	0.130	0.181
Ear length (EL)	-0.637	0.312	-0.034	-0.033	-0.296	0.269	0.073
Ear diameter (ED)	-0.572	0.602	0.114	0.110	-0.051	0.030	0.098
Number of kernel rows(NKR)	-0.415	0.346	0.439	-0.339	-0.021	0.067	0.017
Number of kernels per row(NK/R)	-0.148	0.488	0.181	0.432	-0.373	0.113	0.224
Kernel length (KL)	-0.410	0.412	0.029	0.381	0.118	-0.309	0.057
Kernel width (KW)	-0.128	0.063	-0.057	-0.612	-0.103	0.442	-0.361
Cob diameter (CD)	-0.510	0.523	0.162	-0.199	-0.077	-0.133	0.023
1000 kernel weight (1000KW)	-0.145	0.039	-0.121	-0.302	-0.418	-0.572	-0.151
Harvest grain moisture (HGM)	-0.277	0.054	0.219	0.073	0.722	0.228	0.239
Grain yield per plant (GY/P)	-0.433	0.094	0.241	0.000	0.587	-0.182	-0.142

The mean and standard deviation calculated for each agro-morphological trait included in the resulted five clusters is presented in Table 5, while prominent features of the genotypes are presented in Table 4. Cluster-A comprised of 27 genotypes grouped together having peculiar attributes i.e. the highest number of days to tassel (72.52±5.04) and silking (77.22±4.96), maximum plant height (124.44±30.06), the highest number of leaves per plant (12.60±1.59), the longest leaves (46.41±22.57), and ears (14.78±3.05), the broadest ear diameter (3.59±0.49), maximum ear height (59.71±17.59) and medium grain yield per plant (92.33±40.81). Cluster-B consisted of 43 genotypes grouped together due to their similar performance for morphological traits i.e. maximum number of ears per plant (2.26±0.99), the largest number of days to ear leaf senescence (102.14±6.11) and number of days to harvest (123.30±7.46) and medium grain yield per plant (118.16±53.21). Cluster-C was having 51 genotypes converged due to distinguishing features such as the lowest number of days to tassel (60.96±4.13) and silking (66.76±3.93), the longest anthesis-silking interval (5.69±2.45), the least number of leaves per plant (9.85±2.00) and number of days to ear leaf senescence (89.37±6.05), minimum number of days to harvest (105.80±7.07), the smallest ear length (9.63±2.30), kernel length (7.16±1.32) and kernel width (6.62±1.39) and the lowest grain yield per plant (68.22±29.00). Cluster-D comprised of two accessions with prominent features like shortest anthesis-silking interval (4.00±1.41), the highest 1000 kernels weight (265.20±44.97), the broadest kernel width (8.45±2.19), minimum plant height (91.69±21.91) and ear height (31.75±16.16), the smallest leaf length (22.30±8.06), least number of ears per plant (1.33±0.47), the narrowest ear diameter (2.70±0.28) and cob diameter (2.08±0.04), the lowest number of kernel rows (11.00±0.01) and number of kernels per row (11.67±0.94). Cluster-E consisted of 30 genotypes which clustered together on the basis of agronomic features i.e. highest number of kernel rows (13.39±1.56) and number of days to field germination (12.60±2.03), the broadest cob diameter (2.46±0.27), the highest level of harvest grain moisture (26.05±6.15), maximum grain yield per plant (137.43±59.77), short anthesis-silking interval (4.33±1.54), medium plant height (105.49±24.96) and number of days to anthesis (67.73±5.71) and silking (72.03±5.50).

Table 4. Total number of genotypes per cluster, percentage and prominent features of maize germplasm separated in to five main clusters during 2012.

Cluster	Total Genotypes	Percentage	Prominent Features
A	27	17.66	Highest number of days to tassel and silking, maximum plant height and ear height, largest number of leaves, longest leaves, broadest ear diameter, longest ears and medium grain yield.
B	43	28.10	Maximum number of ears per plant, number of days to ear leaf senescence and number of days to harvest, high grain yield per plant.
C	51	33.34	Lowest number of days to tassel and silking, longest anthesis-silking interval, least number of leaves, number of days to ear leaf senescence and days to harvest, shortest ear length, kernel length and kernel width, lowest grain yield per plant.
D	02	1.30	Shortest anthesis-silking interval, highest 1000 kernels weight, broadest kernel width, smallest leaf length, number of ears per plant, number of kernel rows, number of kernels per row, narrowest ear diameter and cob diameter, minimum ear height, plant height and leaf length.
E	30	19.60	Maximum number of kernel rows and number of days to field germination, largest cob diameter, highest harvest grain moisture and grain yield per plant, short anthesis-silking interval, medium plant height, number of days to tassel and silking.

Table 5. Standard deviation and mean of genotypes included in different clusters during 2012.

Trait	Cluster-A	Cluster-B	Cluster-C	Cluster-D	Cluster-E
DFG	12.41±1.76	12.02±2.01	11.25±1.66	12.50±0.71	12.60±2.03
DT	72.52±5.04	71.53±5.43	60.96±4.13	66.50±2.12	67.73±5.71
DPS	74.93±4.84	74.44±5.17	63.90±4.19	68.00±2.83	69.90±5.52
DS	77.22±4.96	76.95±5.06	66.76±3.93	70.50±3.54	72.03±5.50
ASI	4.74±1.79	5.47±2.04	5.69±2.45	4.00±1.41	4.33±1.54
PH	124.44±30.06	120.88±38.72	97.46±31.52	91.69±21.91	105.49±24.96
NL/P	12.60±1.59	11.41±1.54	9.85±2.00	10.00±1.41	11.77±1.80
LL	46.41±22.57	27.29±11.73	34.15±16.72	22.30±8.06	27.29±5.37
LW	7.03±1.95	5.49±1.36	4.84±1.13	5.22±2.76	5.71±1.87
EH	59.71±17.59	51.76±13.50	39.40±17.51	31.75±16.16	50.52±14.18
NE/P	1.72±0.35	2.26±0.99	1.76±0.44	1.33±0.47	1.96±0.51
DELS	100.78±7.57	102.14±6.11	89.37±6.05	94.00±1.41	98.97±6.94
DH	119.37±9.04	123.30±7.46	105.80±7.07	108.50±3.54	118.93±9.23
FW/Pl	0.13±0.05	0.05±0.03	0.047±0.03	0.03±0.01	0.09±0.05
EL	14.78±3.05	10.43±1.93	9.63±2.30	10.17±0.71	11.92±1.94
ED	3.59±0.49	3.02±0.36	2.94±0.41	2.70±0.28	3.53±0.43
NKR	12.25±1.89	11.36±1.09	11.07±1.41	11.00±0.01	13.39±1.56
NK/R	17.28±5.99	11.88±4.78	14.08±5.60	11.67±0.94	17.16±5.99
KL	8.33±1.12	7.314±1.04	7.16±1.32	7.25±1.77	7.85±1.23
KW	7.03±1.28	6.78±1.08	6.62±1.39	8.45±2.19	6.93±1.47
CD	2.40±0.38	2.16±0.31	2.09±0.30	2.08±0.04	2.46±0.27
1000KW	229.53±32.80	221.92±33.35	215.76±31.02	265.20±44.97	210.68±29.37
HGM	22.52±2.96	25.29±4.60	22.01±3.64	19.50±2.12	26.05±6.15
GY/P	92.33±40.81	118.16±53.21	68.22±29.00	69.50±6.36	137.43±59.77

Principal component analysis based on 24 various quantitative agro-morphological traits determined seven PCs with Eigen value greater than 1 (Table 3, Fig. 2). Collectively the contribution of these PCs was 70.56% in the overall variability among the genotypes. The contribution of PC1 was found to have 26.39% in the total divergence of the studied population, in which the major contributing traits were anthesis-silking interval (0.257), while the contribution of number of days to tassel (-0.819), number of days to pollen shedding (-0.794), number of days to silk (-0.770), number of days to ear leaf senescence (-0.714), number of days to harvest (-0.714), field weight per plant (-0.648), ear length (-0.637), number of leaves per plant (-0.626), ear diameter (-0.572) and cob diameter (-0.510) was negative to add to this component. The second principal component explained 12.59 percent of the total variations found in agro-morphological traits which portrayed the variations in ear diameter (0.602), cob diameter (0.523), number of kernels per row (0.488), field weight per plot (0.469), kernel length (0.412), number of kernel rows (0.346), ear length (0.312), leaf width (0.277), ear height (0.186) and number of leaves per plant (0.179). The traits grain yield per plant, number of ear per plant, kernel width, harvest grain moisture, plant height and 1000 kernel weight also loaded positively but with low intensity. In contrary number of days to silk (-0.550), number of days to pollen shedding (-0.536), number of days to ear leaf senescence (-0.528), number of days to harvest (-0.492) and number of days to tassel (-0.487) were having negative weights. The third principal component illustrated 7.96 percent of the total variations found quantitative agro-morphological characters in maize which determined the pattern of variation in number of kernel rows (0.439), grain yield per plant (0.241), harvest grain moisture (0.219), number of ears per plant (0.219), number of days to harvest (0.207), number of days to ear leaf senescence (0.197), number of kernels per row (0.181), cob diameter (0.162) and ear diameter (0.114). The traits number of days to tassel and kernel length also added positively to this component but with low magnitude. On the other hand leaf length (-0.725), anthesis-silking interval (-0.526), ear height (0.435), plant height (-0.424) and leaf width (-0.365) contributed negatively to this component. PC4 comprised of 6.78 percent mainly enlightened the variability in number of kernels per row (0.432) was positive load and kernel width (-0.612) was negative load on PC4, PC5 accounted for 6.53 percent of the total genetic divergence mainly depicted the variations in harvest grain moisture (0.722) was positive load and 1000KW (-0.418) was negative load on PC5, PC6 accounted for 5.67 percent of the total variations mainly illuminated the kernel width (0.442) was positive load and number of ears per plant (-0.629) was negative load on PC6 and PC7 comprised of 4.63 percent mainly enlightened the variability in anthesis-silking interval (0.509) was positive load and leaf width (-0.433) was negative load on PC7.

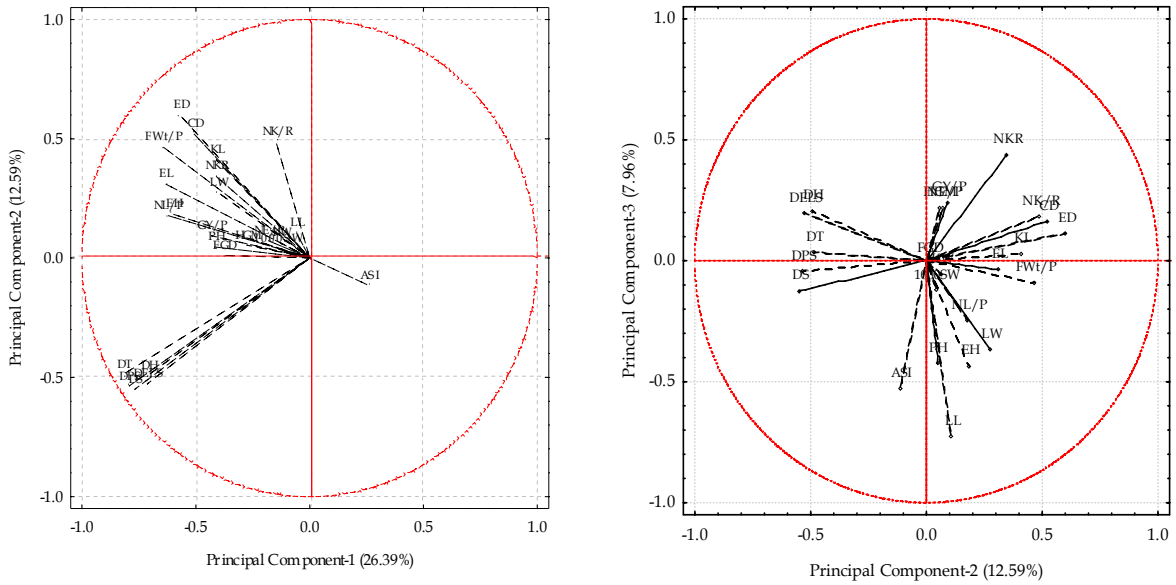

Fig.2. The contribution of the traits in first three PCs.

Discussion

The exploitation of genetic diversity for crops improvement is vital which depends upon the study of all parameters related to the exploration of genetic diversity (Iannetta *et al*., 2007). To conserve the genetic diversity of crops is one of the mile stones to be achieved especially due to post green revolution consequences. It is obvious that the elucidation of genetic diversity is extremely necessary for the effective maintenance, evaluation and utilization of germplasm which is the only source to be exploited for the development of new varieties during breeding programs (Baranger *et al*., 2004). Breeders inserted desirable

genes into the genome of plants and removed the unwanted ones to get the favorable breeds (Narain, 2000). This kind of estimation of genetic diversity and resemblance is extremely pivotal during genetically characterizing the genotypes to identify the cultivars, eliminate the duplicated genotypes present in collected germplasm and more commonly to select the genotypes with desired character(s) for the purpose of hybridization. Descriptive statistics of the data reflected high level of variation for the quantitative characters i.e. days to anthesis, days to silk, plant height, number and length of leaves, ear height, kernel per ear, kernel length and width, 1000 kernel weight and seed yield. High variability for agronomic traits in our germplasm was also supported by findings of previous studies (Dijak *et al*., 1999; Beyene *et al*., 2005; Ihsan *et al*., 2005; Miguel *et al*., 2008; Ranatunga *et al*., 2009; Shrestha, 2013). Correlation plays pivotal role in the selection of right traits for breeding purposes. The correlation between various traits is because of the presence of linked genes. Environment-gene interaction is considered in the outcome of this association between any two correlated traits. Sometimes the role of environment for both the traits is direct and synchronized in the same direction, while in other cases is in opposite or different directions (Yucel *et al*., 2009). Among the total 276 combinations of agro-morphological characters, measurements of some functionally related characters presented significant correlation with each other like traits related to flowering (i.e. number of days to tassel, number of days to silk, number of days to pollen shedding, anthesis-silking interval), morphology of the leaf (such as number of leaves, leaf length, leaf width), traits related to yield (such as number of kernel rows, number of kernels per row, kernel length, kernel width, 1000 kernel weight, grain yield per plant). Our findings were in complete agreement with the results of Bolanos & Edmeades (1996); Banziger *et al*., (2002); Betran *et al*., (2003) and Cortes *et al.,* (2007), who found strong correlation between different agronomic traits in maize. Aliu *et al*., (2013) found highly significant positive correlation of grain yield with cob diameter and 1000 kernels weight and significant positive correlation with plant height which was confirmed by our studies. It is clear from the cluster and principal component analysis that the maize germplasm investigated has tremendous genetic diversity which may be of a great potential source for the future maize breeding strategies. Different quantitative traits preferably number of kernel rows, number of kernels per row, 1000 kernels weight, ear length, ear height, ear diameter, number of ears per plant, number of days to tassel and silking and anthesis-silking interval, in the combination of few or more can be useful for breeding programs. The dendrogram grouped all genotypes on the basis of their similarity into five clusters, A, B, C, D and E. The crosses made among the genotypes from cluster A and E will be more fruitful which agrees with the findings of Sharma *et al*., (2013). Distribution of genotypes into multiple clusters was tantamount to presence of genetic divergence among them. Therefore, on the basis of agro-morphological performance the germplasm from Pakistan and Japan were divided into four different clusters denoting broad genetic base. Likewise, Chinese accessions were found in two groups. Based on these facts it is obvious that the distribution of the studied accessions is independent of their geographical background. So these two complementary analyses portrayed that there is no geographical relationship among the studied genotypes because genotypes collected from same place were distributed into different clusters, which is supported by the findings of Gupta *et al*., (1991) during studying the genetic diversity of mustard, who found that the genetic diversity present in mustard germplasm was not because of their geographical background. Amurrio *et al*., (1995), reported that geographical origin of the pea landraces had no role in their clustering into groups. Similarly, PCA grouped together the maize accessions with similar morphology but not because of their geographical background. PCAs displayed that the total genetic diversity was clearly distributed throughout the agro-morphological traits. It is important to point out that Pakistan possesses a variety of environmental and ecological conditions which effectively contribute to the genetic diversity of different organisms. Along with other crops, the genetic diversity of maize is also greatly benefited from this. The genetic diversity in an area has a direct relationship with the ecological and environmental conditions instead of affected by the geographical distribution (Li & Rutger, 2000). The overall distribution of the studied germplasm on the basis of agro-morphological traits, it can be concluded that landraces of maize in Pakistan possess significant amount of genetic variability, which have the potential to be used for the development of productive varieties. It is needed to cash this tremendous level of genetic variation by developing varieties with early maturity, as some of the accessions tested, were having shorter life cycles than check varieties. The accessions collected from the areas of harsh environmental conditions such as drought, poor soils, more radiations and possess the adaptation traits like short stature, early flowering and narrow leaves, provide the base and opportunity to develop drought resistant varieties for those parts of the country that receive little rain. During the present investigation various agro-morphological traits displayed significant level of variation which can be used directly for the development of high yielding cultivars. The selection process promotes the development of cultivars on one hand but on the other hand results in the decrease of the genetic diversity of the gene pool, which is immensely necessary to conserve and utilize in future (Rabbani *et al*., 1998). The little fluctuation noted between the results from previous studies may be because of the difference in the genetic makeup of experimental material (Iqbal *et al*., 2014) and change in the environmental conditions.

Conclusion

It can be inferred from the present investigation that high level of genetic diversity was present in agro-morphological traits like grain yield per plant, plant height, 1000 kernels weight etc in the tested germplasm. Similarly best performing promising genotypes with more genetic diversity were identified. The identification of high level of genetic diversity during the current study could be implied for maize germplasm characterization, conservation and further improvement in maize breeding in future.

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