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PHENOTYPIC DIVERGENCE IN GUAR (*CYAMOPSIS TETRAGONOLOBA*) LANDRACE GENOTYPES OF PAKISTAN

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Abstract

Guar is an important commercial and export crop of Pakistan. Genetic improvement of guar regarding its yield potential has never been addressed in Pakistan. Present work was carried out to evaluate 101 guar accessions collected from diverse areas of Pakistan. Fourteen quantitative and three qualitative characters were taken under observation to estimate substantial variation and relationship among guar genotypes and identify promising accessions for future exploitation. Considerable and dissimilar level of variability was noticed for a number of agro-morphological traits on the basis of analysis of variation. High variation in germplasm was found for days to maturity, plant height, pods per plant and pod length. The correlation coefficient analysis depicted positive and significant correlation of branches per plant, clusters per plant, pods per cluster, pods per plant and pod width with seed yield plant^{-1}, while days to flower initiation, 50% flowering, days to maturity and plant height were found in significant negative correlation with seed yield plant^{-1}. Principal component analysis displayed significant variation with 30.79% of the total variation for PC1, 16.71% for PC2, PC3 for 12.66% and PC4 contributed 10.09% to the total variation. Hierarchical cluster analysis based on agro-morphological traits divided 101 guar accessions into four main groups and six sub-clusters. Clustering was found in association with similar geographic origins and also with morphological differences. On the basis of greater yield potential, seeds per pod, pods per plant and early maturity, promising genotypes have been identified for guar variety development and future breeding programs.

Introduction

Germplasm of a particular crop collected from the indigenous sources offers greater genetic variability and may furnish useful traits to widen the genetic base of crop species. The success in the improvement of crop and the development of a species requires the availability and accessibility of genetic diversity. Detection of duplicates, organization of core collection of a particular population and the selection of parents for the development of new cultivars are directly linked to the genetic variability (Jatoi *et al*., 2010; Pervaiz *et al*., 2010). Genetic diversity is one of the key factors for the improvement of many crop plants including guar. The determination of genetic diversity of germplasm will maximize the probability of transgressive segregation and increase the probability that unrelated accessions contribute with positive alleles at different loci. Among various tools available for assessing the genetic variability and relatedness among crop genetic resources, morphological characterization is the first step in the description and organization of germplasm of any crop. In the recent past efforts have been made to assess the genetic diversity and relationship among germplasm collections of radish, rice, sesame at morphological level (Siddiqui *et al*., 2007; Akbar *et al*., 2011; Jatoi *et al*., 2011), however very limited information is available on genetic analysis of guar from Pakistan.

Guar (*Cyamopsis tetragonoloba* L.) is an upright, deep rooted plant of family Leguminosae (Fabaceae), highly valued for its best yield, greater nutritional importance and drought tolerance. Guar originated in the India–Pakistan area (Purseglove, 1981) and is grown under arid/semiarid parts of the world. Pakistan is an important producer of guar in the world. During 2008-2009 guar was cultivated on an area of 155000 hectares with a gross production of 102000 tones having an average yield of 661 kg/hectare (Zahoor, 2007; Anon., 2009). In guar 27-37.2% nutritional quality protein and out of 50% endosperm, 42% mucilage or gum (guar gum) has been reported (Anderson, 1949; Whistler & Hymowitz, 1979). Guar is a versatile and multi-used legume crop, cultivated mostly for vegetable, forage, and cover crop (Arora & Pahuja, 2008) .Guar enhances soil productiveness by fixing atmospheric nitrogen for its own necessitates and also for the succeeding crop (Bewal *et al.,* 2009). Although guar is a minor crop but due to its better and finer guar gum qualities it is considered as a ready money crop for industrial gum production (Hymowitz & Matlock, 1963; Pathak *et al*., 2010) and for several pharmaceutical and nutraceutical products. India and Pakistan are the chief exporters of guar to USA. During 2008-2009, Pakistan earned about 2206400 rupees as foreign exchange by exporting 25800 tones of guar and guar based products to about 30 different countries of the world (Anon., 2009). Presently, in Pakistan three factories are processing the guar seeds to extract gum and converting into powder shape and for the most part exporting to the United States (Anon., 2011). Genetic characterization of existing gene-pools is a pre-requisite for sustainable improvement of crop species (Hodgkin, 1995), for the development of novel varieties and for successful conservation of gene-pool (Zhang *et al.,* 1999). Very little information on morphological characterization of guar is present as compared to other crops. Researchers like Dabas *et al*., (1995), Brahmi *et al*., (2004), Morris *et al*., (2004), Singh *et al*., (2005), Morris (2010) and Rai (2010) have worked out on guar but not any such work is reported in Pakistan. Despite the fact that most of the local landraces are on hand possessing considerable genetic diversity, present work attempts to explore genetic potential of guar in order to identify elite genotypes to enhance its access to international markets.

Materials and Methods

Genetic evaluation of guar accessions on the basis of agro-morphological traits was performed under field conditions of Institute of Agri-Biotechnology & Genetic Resources (IABGR), National Agricultural Research Centre (NARC) Islamabad. Seed material was obtained from the gene bank of IABGR, NARC Islamabad, which were collected from a broad array of eco-geographical diverse areas of Pakistan. To prepare seed bed, pre-sowing irrigation was done under optimum moisture condition and planting was done with hand drill in augmented field design with plot size of 1×5 m^2 having 2 lines per accession during July, 2010. Length of the row was 5 m, path between beds was 2 m and row to row distance was kept as 35 cm. Two improved cultivars, BR99 and BR99-Super were repeated as checks after every 25 accessions. For healthy and vigorous plants, recommended agronomic and cultural practices were applied from the plantation to the maturity stage. Weeds eradication was done by hand once 30 days after planting. No pesticide and no fertilizer treatment were given to the crop. Harvesting of the crop was carried out when more than 70% plants pod in each accession turned brown in color.

Both quantitative and qualitative characters were brought under consideration for estimating substantial variation and relationship among guar genotypes. Fourteen quantitative characters, days to initial flowering, days to 50% flowering, days to maturity, plant height, branches per plant, clusters per plant, pods per cluster, pods per plant, pod length, pod width, pod thickness, seeds per pod, seed yield per plant and 100- seed weight while three qualitative characters, i.e., leaf texture, plant surface and seed color were taken into consideration for estimating genetic diversity (Table 1). Mean values of accessions were computed for determining analysis of variance and correlation coefficients were estimated by using the formulae of Kwon & Torrie (1964). Principal component analysis (PCA) with Eigenvalues > 1.0 and cluster analysis was also performed to assess genetic diversity among guar accessions.

Table 1. Morphological data recorded for quantitative and qualitative traits.

Trait	Scale	Description of the trait
Days to flower initiation (DFI)	Days	Number of days from seed sowing until 5% of plants have first flower in each accession
Days to 50% flowering (50%DF)	Days	Number of days from seed sowing until 50% of plants has at least one flower in each accession
Days to maturity (DM)	Days	Number of days from seed sowing until 75% of plants reaching physiological maturity
Plant height (PH)	cm	Mean height of five random plants from ground level to the apex of the main stem
Branches per plant (B/P)	No.	Total number of branches originating from main stem
Clusters per plant (C/P)	No.	Total number of clusters on a single plant. Mean of five randomly selected plant
Pods per cluster (P/C)	No.	Total number of pods in a cluster
Pods per plant (P/P)	No.	Total number of pods present on a single plant of same 5 randomly selected plants per accession
Pod length (PL)	mm	Distance from the base to the tip of five randomly selected pods
Pod width (PW)	mm	Distance across the widest point of the same pod used for length
Pod thickness (PT)	mm	Diameter of the same selected pod used for length and width
Seed yield per plant (SY/P)	g	Average seed weight of 5 randomly selected at 13% moisture content
100-Seed weight (HSW)	g	Weight of 100 random dried seeds
Leaf texture (LT)	-	1 = glabrous, 2 = pubescent, 3 = mix of glabrous & pubescent types
Plant surface (LS)	-	2 = smooth, 4 = pubescent
Seed color (SC)	-	1 = Yellow green, 2 = Dark green

Results and Discussion

Analysis of variation depicted considerable level of variability among different accessions for a number of agro-morphological traits. Basic statistics (mean, coefficient of variation and variance) for quantitative characters was tabulated in Table 2. Accessions varied in several traits of economic importance and dissimilar pattern of variation among the accessions was noticed for various agro-morphological traits. The largest variation was found for days to maturity, plant height, pods per plant and pod length. Comparatively, low variation was noticed for days to flowering initiation, days to 50% flowering, branches per plant, clusters per plant, pods per cluster, pod width, pod thickness, seeds per pod, seed yield per plant and 100-seed weight. Morris (2010) findings are in accordance with the present study. Similarly, findings of Krishnan ***et al.,*** (2011) for branches per plant, pod length and seeds per pod are in strong association with the present data; however values for plant height and clusters per plant do not match with our results. Present data is less in yield, number of pods, late in 50% flowering and maturity, however possessed more branches, greater plant height but with same number of seeds per pod with respect to Punia ***et al.,*** (2009) results. Observed variability found among guar accessions can be probably attributed to the genetic differences and the environment, in which these accessions were grown.

A significant level of correlation in present study was noticed among morphological characters (Table 3). For example days to flower initiation, 50% flowering, days to maturity, pods $plant^{-1}$, pods $cluster^{-1}$, and branches $plant^{-1}$ produced positive and significant correlations with each other. However, yield $plant^{-1}$ and 100-seed weight showed negative correlation with some other traits. Branches $plant^{-1}$, clusters $plant^{-1}$, pods $cluster^{-1}$, pods $plant^{-1}$ and pod width had the significant positive contribution with seed yield $plant^{-1}$. Days to flower initiation, 50% flowering and days to maturity and plant height revealed negative and significant correlation with seed yield $plant^{-1}$. Our correlation coefficient results are in strong agreement with Rai, (2010) findings, who determined significant and positive correlation of yield with plant height, pods $cluster^{-1}$, pods $plant^{-1}$ and pod yield $plant^{-1}$.

Table 2. Descriptive statistics of morphological traits in guar genotypes.

Traits	Minimum	Maximum	Mean	SD	CV(%)	Variance
Days to flower initiation	63	69	65.5	1.5	2.3	2.2
Days to 50% flowering	64	72	68.4	1.9	2.8	3.6
Days to maturity	112	127	121.2	4.1	3.3	16.4
Plant height	50	135	97.7	14.1	14.4	197.5
Branches per plant	2.0	10.2	5.3	1.7	32.7	3.0
Clusters per plant	2.5	12.4	5.2	1.6	29.7	2.4
Pods per cluster	3.7	10.4	6.5	1.5	23.6	2.4
Pods per plant	12.0	85.0	36.8	14.5	39.4	211.3
Pod length	41.0	65.0	51.9	3.7	7.1	13.7
Pod width	5.0	7.0	5.9	0.5	7.8	0.2
Pod thickness	3.3	5.5	4.2	0.4	9.7	0.2
Seeds per pod	6.0	9.4	8.1	0.6	7.7	0.4
Seed yield per plant	1.8	15.5	5.8	2.5	43.6	6.5
100-seed weight	1.8	4.1	3.0	0.4	13.2	0.2

Table 3. Correlation coefficients among various quantitative traits.

Traits	DFI	50% DF	DM	PH	Br/P	C/P	P/C	P/P	PL	PW	PT	S/P	SY/P	100SW
DFI	1													
50% DF	0.79**	1												
DM	0.76**	0.96**	1											
PH	-0.28**	-0.28**	-0.22*	1										
Br/P	-0.06	0.01	-0.02	0.09	1									
C/P	-0.21*	-0.14	-0.13	0.30**	0.57**	1								
P/C	-0.22*	-0.22*	-0.21*	0.39**	0.17	0.35**	1							
P/P	-0.25*	-0.25*	-0.23*	0.40**	0.51**	0.76**	0.75**	1						
PL	-0.20*	-0.07	-0.11	0.09	-0.01	0.11	0.13	0.06	1					
PW	-0.13	-0.09	-0.11	-0.20*	0.11	0.02	-0.03	-0.02	0.13	1				
PT	-0.15	-0.12	-0.14	-0.1	0.25*	0.01	-0.14	-0.12	0.14	0.44**	1			
S/P	-0.12	-0.09	-0.09	-0.04	-0.13	-0.02	0.1	-0.03	0.40**	-0.06	0.15	1		
SY/P	-0.43**	-0.40**	-0.40**	0.25*	0.50**	0.59**	0.54**	0.72**	0.11	0.26**	0.16	0.06	1	
100SW	-0.40**	-0.36**	-0.37**	0.17	0.14	0.03	0.11	0.12	0.13	0.35**	0.31**	-0.12	0.41**	1

Traits = DFI (Days to flower initiation), 50%DF (Days to 50% flowering), DM (Days to maturity), PH (Plant height), B/P (Branches plant^{-1}), C/P (Clusters plant^{-1}), P/C (Pods cluster^{-1}), P/P (Pods plant^{-1}), PL (Pod length), PW (Pod width), PT (Pod thickness), S/P (Seeds pod^{-1}), SY/P (Seed yield plant^{-1}), 100-SW (100-seed weight)

Variation was also accessed using principal component analysis for 101 guar accessions by taking into account every variable at once. First four principal components accounted for a total of 70.25% variability among the accessions using quantitative and qualitative traits (Table 4). A total of 30.79 proportion of variance of the total variation was observed in the first principal component, PC2 depicted proportion of variance as 16.71%, PC3 accounted for 12.66% of the total variation and PC4 contributed 10.09% to the total variation. PC1 showed predominantly the variation in days to flower initiation, days to 50% flowering and days to maturity (Fig. 1). PC2 was mostly correlated with pod thickness, 100-seed weight and pod width. PC3 was associated with days to 50% flowering, days to maturity, branches per plant, pod width, pod thickness, and 100-seed weight, whereas 100-seed weight, pod width, plant height and branches per plant made positive contribution to PC4 but with very small magnitude.

Table 4. Principal components for 12 quantitative traits in guar.

Trait	PC1	PC2	PC3	PC4
Eigenvalue	4.31	2.34	1.77	1.41
Cumulative eigenvalue	4.31	6.65	8.42	9.84
Proportion of variance	30.79	16.71	12.66	10.09
Cumulative variance	30.79	47.50	60.16	70.25
		Eigenvectors		
Days to flower initiation	0.344	-0.307	0.163	-0.041
Days to 50% flowering	0.343	-0.342	0.244	-0.138
Days to maturity	0.337	-0.352	0.213	-0.122
Plant height	-0.228	-0.133	-0.287	0.037
Branches per plant	-0.215	-0.285	0.386	0.077
Clusters per plant	-0.298	-0.354	0.117	-0.053
Pods per cluster	-0.294	-0.260	-0.163	-0.144
Pods per plant	-0.364	-0.383	-0.022	-0.015
Pod length	-0.107	0.112	0.038	-0.631
Pod width	-0.097	0.222	0.509	0.020
Pod thickness	-0.087	0.261	0.496	-0.124
Seeds per pod	-0.037	0.125	-0.093	-0.697
Seed yield per plant	-0.401	-0.123	0.172	-0.010
100-seed weight	-0.233	0.256	0.226	0.185

Hierarchical cluster analysis based on agro-morphological trait divided 101 guar accessions into four main groups and six sub-clusters (Fig. 2; Table 5). Maximum number of accessions (38) was present in group II, followed by group IV (26), group III (25) and group I (12). Group I contained genotypes, all from Bahawalpur and group II to group V comprised of accessions of diverse geographical origin. Group 1 genotypes were characterized as very late in initial flowering, 50% flowering, late maturing and with the lowest plant height as well as yield than all other groups (Table 5). Maximum pod width, medium pod length, seeds per pod and less number of branches per plant and pods per cluster were observed in group I. Group II was the largest group comprising tall genotypes, medium in number of pods per cluster, pods per plant, late in flowering, maturing with few branches per plant, pods per plant, and also low yielding. Group III was early maturing, taller in height, more seed weight, very less number of branches and pods per cluster, numerous branches, medium height and yield. Group IV was the second largest of all groups with distinctive morphological features. Genotypes of group IV were the earliest in flowering and maturing, taller in height exhibiting maximum yield potential and 100-seed weight with maximum number of branches, clusters per plant, and pods per plant (Table 6).

Table 5. Means of different quantitative traits for different clusters of 101 guar genotypes.

Trait	Group-1	Group-2	Group-3	Group-4
Days to flower initiation	67.5	66.1	65.1	64.2
Days to 50% flowering	70.3	69.2	67.9	66.7
Days to maturity	124.8	122.8	120.8	117.5
Plant height	79.9	98.6	104.1	98.5
Branches per plant	5.0	4.8	5.1	6.3
Clusters per plant	4.3	4.9	5.1	6.2
Pods per cluster	5.5	6.3	7.0	6.9
Pods per plant	24.2	33.0	40.7	44.6
Pod length	52.2	51.3	51.7	52.8
Pod width	6.3	5.7	5.7	6.2
Pod thickness	4.3	4.1	4.0	4.5
Seeds per pod	8.0	8.2	8.0	8.2
Seed yield per plant	4.1	4.6	5.6	8.6
100-seed weight	3.0	2.8	3.1	3.3

Table 6. Characteristics of Pakistani guar genotypes in sub-clusters.

Main group	Sub-cluster	No. of lines	%age of lines	Prominent traits/Main features
I	1	12	12%	Maximum pod width, very late in flower initial, late maturing, lowest yield potential and medium in seeds/pod
II	1	24	24%	Tall stature, medium in pods per cluster, pods per plant, late in flowering and maturing, lower yield and 100-seed weight
	2	14	14%	Very late in initial flowering, maturing, less in height, minimum pod width and 100 seed weight among all guar genotypes
III	1	11	11%	Early maturing, tall in height, greater seed weight, medium pod length, very less number of branches and pods per cluster
	2	14	14%	More number of branches, numerous pods per plant, minimum pod thickness and less number of seeds per pod
IV	1	5	5%	Tallest in height, highest yielding, maximum number of branches and pods/plant, medium pod length, width and 100-seed weight
	2	21	21%	Earliest in initial flowering, maximum yield potential and 100-seed weight, medium in plant height, pod length but with maximum pod thickness

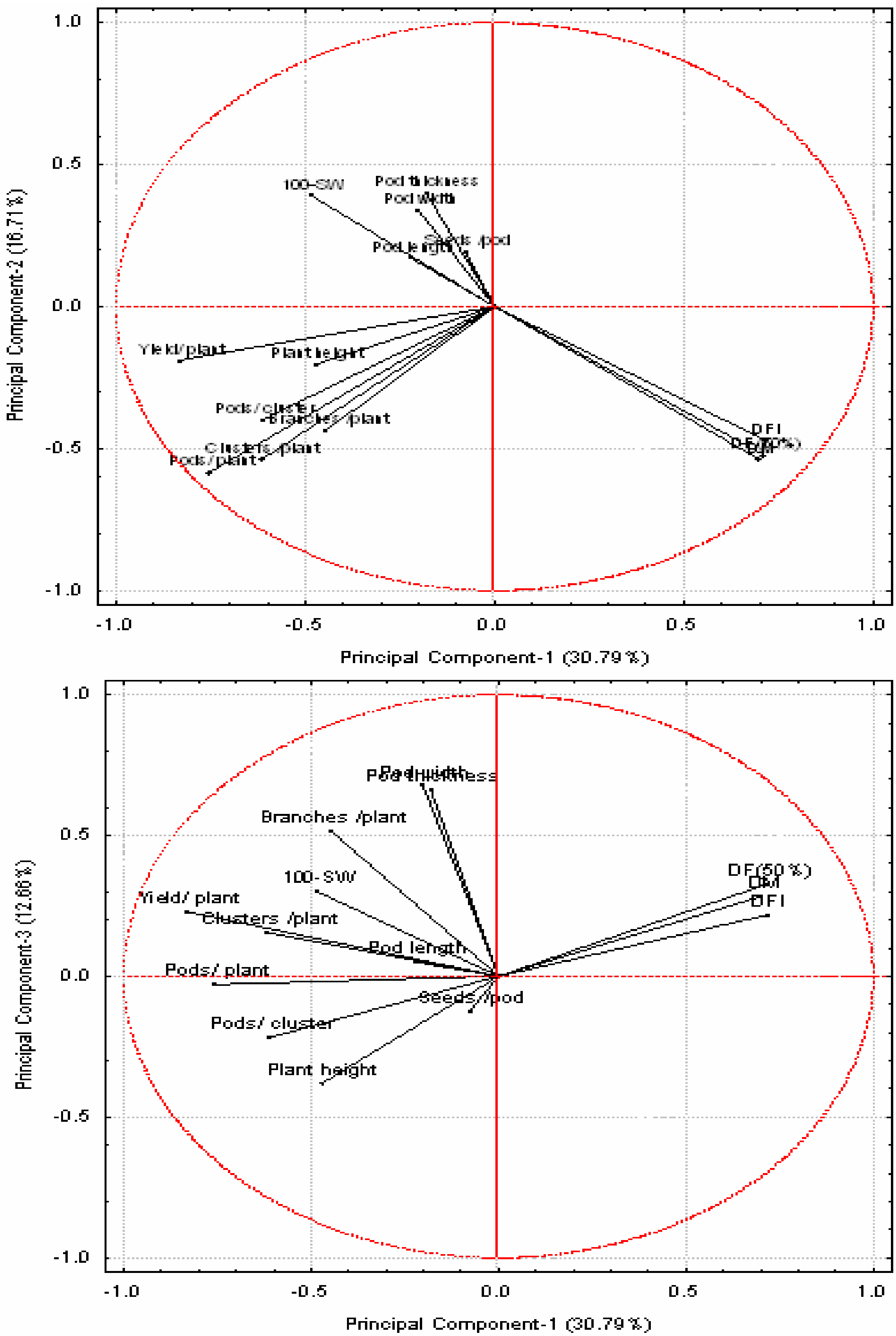

Fig. 1. Contribution of morphological characters in first three principal components.

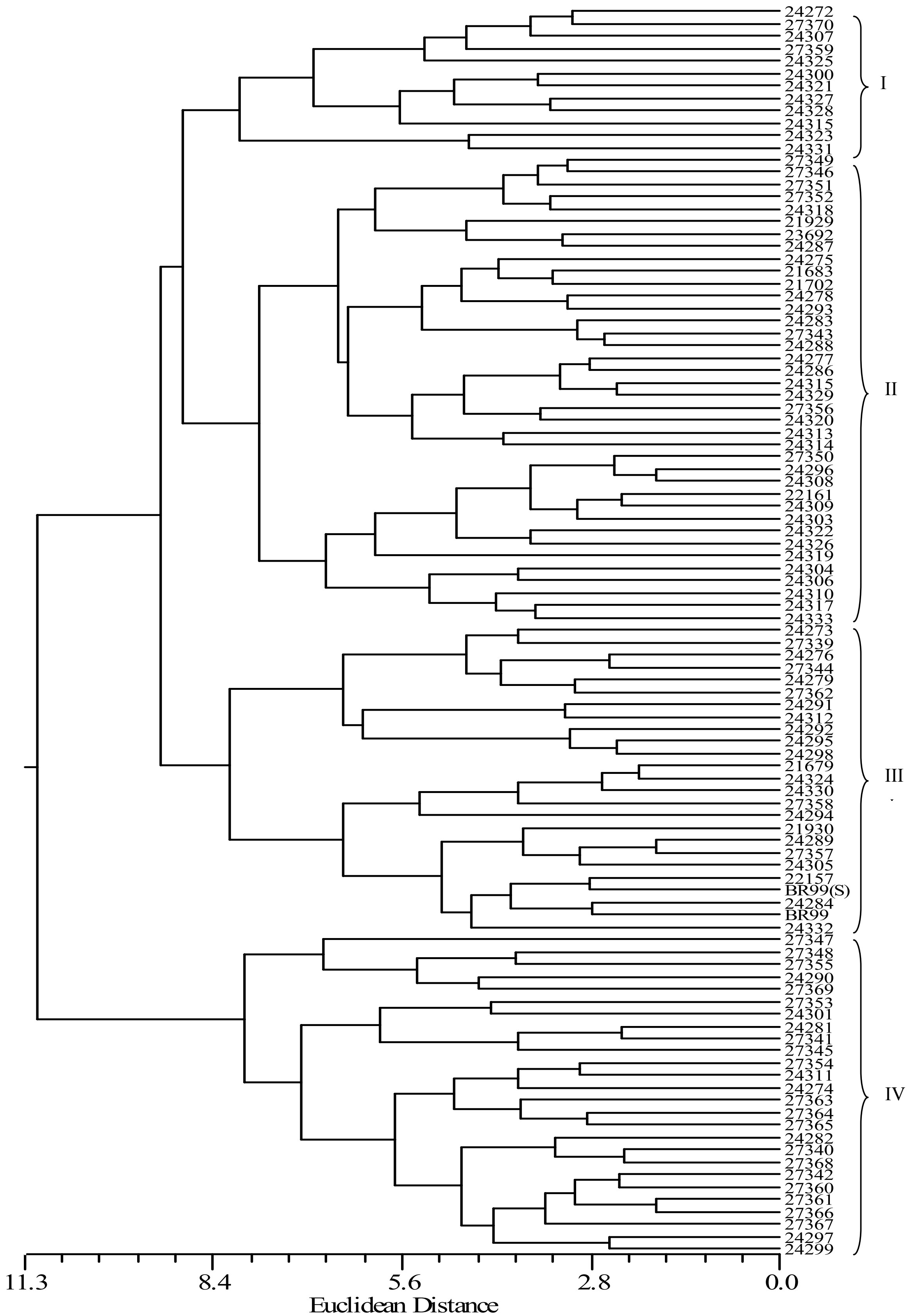

Fig. 2. Dendrogram showing the genetic relationship of Pakistani guar genotypes.

Divergence studies of morphological characters of guar using cluster analysis have been conducted by researchers Punia *et al*., (2009), Morris (2010), and Rai (2010). Conclusions of the work done by all these researchers agree to the present outcome that this method can clear complex relationships between populations of diverse origins in a more simplified way. By hierarchical cluster analysis, present study revealed that some of the accessions collected from various geographical regions were grouped into the same cluster, while some other accessions fell into different clusters. Present findings for group I are in accordance with the results of Morris (2010) that these clusters are likely to define accession groups with similar geographic origins. However for rest of the groups it can be generalized that grouping on cluster basis is not always associated with the geographical distribution instead accessions are grouped on the basis of their morphological differences. Our findings also come to agreement with the findings of Brahmi *et al*., (2004), who declared that cluster groups were not associated to the geographical origin of guar accessions.

According to the results of current work an extensive range of genetic diversity has been explored in guar germplasm of Pakistan. Important agro-morphological traits like greater yield potential, seeds per pod, pods per plant and early maturity, etc. served as a criterion to select promising guar genotypes (Table 7). For the improvement of cultivated guar, it is dire need to use diverse collections with more variability for the purpose of variety development and for use in future breeding programs of Pakistan.

Table 7. Elite guar genotypes identified on the basis of important agro-morphological traits for future use.

Trait of interest	Range	Accessions identified
Days of maturity	< 115	24281, 27341, 27345, 24292, 27363, 24298
Pods per plant	≥ 70	27358, 24290, 27369, 27347, 27355
Seeds per pod	≥ 9	27339, 24301, 27351, 27349, 24281, 27341, 27355, 27362, 24310, 24323, 24333
Seed yield per plant	≥ 11g	24290, 27348, 27369, 24301, 27355, 27341
100-seed weight	≥ 3.6g	24291, 27345, 24307, 27364, 27363, 27368, 27353, 24312

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