GENETIC DIVERGENCE OF INDIGENOUS PUMMELO GENOTYPES

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ARTICLE INFO	ABSTRACT
	The genetic divergence was studied in 33 pummelo genotypes using D^2 statistics and principal component analysis at Regional Agricultural Research Station, BARI, Akbarpur, Moulvibazar during 2012 to 2014. The genotypes were grouped into 5 clusters and the maximum number of genotypes was included in cluster IV and V and the minimum number in cluster I. The inter cluster distance in all of the cases were higher than the intra cluster distance indicating wider genetic diversity among the accessions of different groups. The highest inter-cluster distance was observed between cluster I and II followed by cluster II and V and the lowest between III and IV. The highest intra- cluster distance was observed for the cluster II and the lowest for the cluster III. For cluster II, the highest mean values for plant height (6.13m), individual fruit weight (1141.67g), fruit length (13.03 cm) and breadth (13.15 cm), number of segments per fruit (14.41), number and weight of seeds per fruit (123.67 and 50.41g), yield per plant (50.94 kg) were observed. The first axis largely accounted for the variation among the pummelo accessions (26.16%) followed by second axis (18.75%). The first 8 axes accounted 90.56 % of the total variation. The characters individual fruit weight (g) and weight of seeds per fruit (g) showing positive value in both the vectors contributed maximum towards divergence. Considering magnitude of genetic distance, contribution of different traits toward the total divergence, magnitude of cluster means for different traits and performance the genotypes of cluster I, II, and IV may be considered as parents for future hybridization program.

INTRODUCTION

Pummelo(*Citrus maxima*) is known as one of the important commercial fruit tree under the genus *Citrus* (Verdi, 1988). It is a native plant species to tropical and subtropical regions in Asia and has been cultivated in China for over 2000 years (Corazza-Nunes *et al.*, 2002; Yong *et al.*, 2006). *C. maxima* was originated from South East Asia,which in the western regions is familiar as shaddock (Uzun and Yesiloglu, 2012). Pummelo generally produces fruits twice a year, grows on varioussoil types at the altitude of 100-400 m above sea level (Dinesh and Reddy, 2012). *C.maxima* is one of three true *Citrus* species together with *C. medica and C. reticulata* (Barrett and Rhodes,1976;Hynniewta *et al.*, 2011). Its status as true or basic species with in *Citrus* is confirmed by other researchers (Barkley *et al.*, 2006; Uzun *et al.*, 2009;Froelicher *et al.*,2011; Garcia-Lor*et al.*, 2013).Therefore, pummelo has been regarded as a parent of many citrus fruits, such as lemons, oranges and grapefruits. It is characterized for distinguished features of huge leaves borne on broadly winged petioles, very large and fragrance flowers and big fruits with a single embryo, while most of other *Citrus* species are polyembryonic (Uzun and Yesiloglu, 2012). Pummelo is one of the popular and the biggest citrus fruit of Bangladesh. In Bangladesh, it is cultivated in an area of around 7460 ha with total production of 59198 metric tons and average yield per plant is around 38.0kg (BBS, 2011). For obtaining varieties with desired traits, hybridization is a very effective tool if diversed parents with promising features are available. Pummelo is a cross pollinated crop and there is a wide variability within the species which create good opportunity to plant breeder to utilize for its improvement (Janick and Moore, 1996).

Genetic diversity is an important factor for crop improvement with desirable traits. Multivariate analysis such as D^2 cluster and factor analysis have been proved to be useful for selecting genotypes for hybridization. Mahalanobis (1949) D^2 analysis has been successfully used in measuring the diversity in several crops. An understanding of nature and magnitude of variability among the existing pummelo germplasm is a prerequisite for its improvement. Precise information on the nature and degree of genetic divergence helps the plant breeder in choosing the diverse parents for purposeful hybridization (Arunachalam, 1981; Samsuddin, 1985). Since published work on pummelo is scanty, the present study has been undertaken with 33 pummelo accession to understand the nature and magnitude of genetic divergence and the characters contributing genetic diversity by D^2 analysis.

MATERIALS AND METHODS

The experiment was conducted at the pummelo orchard and laboratory of the the Regional Agricultural Research Station, Bangladesh Agricultural Research Institute, Akbarpur, Moulvibazar during February 2012 to October 2014.Thirty three pummelo genotypes were included in the present study and data were recorded from three different plants of an accession where each plant was considered as a replication. Individual plants were fertilized with cowdung (20 kg), urea (500 g), TSP (500 g), MP (500 g), gypsum (200 g) , Zinc (4 g), boron (4 g) in two equal installments one at the onset and other at the end of rainy season. (FRG,2012). Irrigation, weeding and other crop management practices were followed as recommended by Ullah *et al.* (2006) to have a good healthy plant. Data on plant height (m), canopy spreading (m), number of flowers per cluster, number of fruits per plant, individual fruit weight (g), fruit length (cm) and breadth (cm), number of segments per fruit, number and weight (g) of seeds per fruit, edible percentage, % Brix (TSS) and yield per plant(kg)were recorded.

Genetic diversity was studied following Malanobsis's (1949) generalized distance (D^2) extended by Rao (1952). Clustering of genotypes was done according to Tocher's Method (Rao, 1952) and principal component analysis was done according to Rao (1964). All the statistical analysis were carried out using GENSTAT-5 computer software. Average intracluster distance was calculated by the following formula as suggested by Singh and Chaudhury (1985).

$$\text{Average intracluster } D^2 = \frac{\sum D^2 ?}{?}$$

Where, $\Sigma D^2 i$ = Sum of distances between all possible combination (n) of the varieties/lines included in a cluster and n = All possible combinations

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RESULTS AND DISCUSSION

In this study the 33 accessions of pummelo were grouped into five clusters based on D^2 values (Table 1). The distribution pattern indicate that the maximum number (9 each) of genotypes were included in cluster I V and V followed by cluster II and III. The minimum numbers of genotypes were included cluster I. Rahman and Al Munsur(2009) grouped 40 genotypes of lime into 6 clusters.

Table 1. Distribution of 33 pummelo accessions in five clusters with location

Cluster	Numbers	Accessions	Percentage
I	3	CM-Akb-141, CM-Akb-171, CM-Akb-178	9.09
II	6	CM-Akb-144, CM-Akb-147, CM-Akb-148, CM-Akb-158, CM-Akb-159, CM-Akb-166	18.18
III	6	CM-Akb-145, CM-Akb-150, CM-Akb-156, CM-Akb-162, CM-Akb-164, CM-Akb-165	18.18
I V	9	CM-Akb-134, CM-Akb-139, CM-Akb-146, CM-Akb-151, CM-Akb-153, CM-Akb-160, CM-Akb-161, CM-Akb-163, CM-Akb-184	27.27
V	9	CM-Akb-136, CM-Akb-137, CM-Akb-138, CM-Akb-170, CM-Akb-172, CM-Akb-173, CM-Akb-176, CM-Akb-177, CM-Akb-182	27.27

A two dimensional scatter plotting diagram (Z_1-Z_2) constructed using component score 1 on X axis and component score 2 on Y axis exhibited that the genotypes were fallen into five clusters (Figure 1).
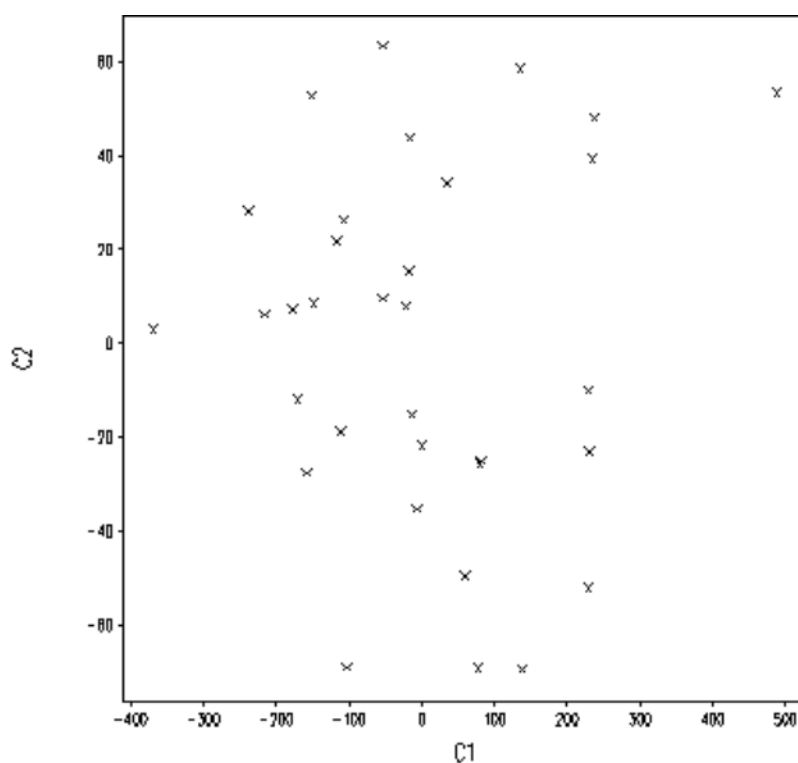
Figure 1. Scattered distribution of 33 pummelo genotypes based on the principal component score

Intra and inter cluster distances are presented in Table 2. The inter-cluster distances were higher than the average intra-cluster distances, which indicated wide genetic diversity among the pummelo accessions of different groups than those of same cluster. Rahman and Munsur (2009) found same result in case of lime. The highest inter cluster distance was observed between cluster I and II (16.638) and followed by cluster II and V (13.374) and the lowest between III and IV (4.482) (Table 2). The highest intra- cluster distance was observed for the cluster II (1.766) and the lowest for the cluster III (1.121). The highest values for intercluster distance indicated that the accessions belonging to cluster I was far away from those of cluster II. The minimum intercluster divergence was observed between III and IV indicating that the genotype of these clusters was genetically closer. Hybridization among the genotypes drawn from widely divergent clusters with high yield potential would likely to manifest maximum heterotic combinations as well as new recombination with desired traits. Similar findings were observed in lemon, sweet gourd and jackfruit (Ismail 2008; Rahman *et al.* 2006 and Saifullah*et al.* 1999).

Table 2. Average intra (bold) and inter-cluster distances for 33 pummelo accessions

Clusters	I	II	III	I V	V
I	**1.3**				
II	16.638	**1.766**			
III	12.11	5.51	**1.121**		
I V	8.093	8.894	4.482	**1.359**	
V	4.624	13.374	8.952	4.909	**1.513**

Table 3. Cluster mean values for yield and yield contributing characters of 33 pummelo accessions

Characters	Cluster means				
	I	II	III	IV	V
Plant height (m)	5.30	6.13	5.71	5.27	5.15
Canopy spreading at north-south direction (m)	5.97	6.15	5.95	6.24	5.23
Canopy spreading at east-west direction (m)	5.33	5.30	5.45	6.0	6.38
Number of flowers per cluster	5.88	4.75	5.16	5.55	5.64
Number of fruits per plant	41.0	43.17	39.83	43.77	33.34
Individual fruit weight (g)	593.33	1141.67	965.0	850.92	730.56
Fruit length (cm)	11.37	13.03	12.05	11.75	12.07
Fruit breadth (cm)	11.0	13.15	12.65	12.12	11.31
Number of segments per fruit	13.33	14.41	14.33	14.34	13.17
Number of seeds per fruit	102.33	123.67	77.67	111.53	97.11
Weight of seeds per fruit (g)	35.0	50.41	29.5	44.61	37.22
Edible percentage	53.54	49.37	52.73	49.53	48.12
% Brix (TSS)	8.33	9.17	9.50	9.37	8.97
Yield per plant (kg.)	24.68	50.94	38.49	37.14	24.18

Cluster mean values of 14 different characters are shown in Table 3. Difference in cluster means existed for almost all the characters studied. The highest mean value for number of flowers per cluster (5.88),edible part of fruit (53.54%) were observed in cluster I. For cluster II, the highest mean values for plant height

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(6.13m), individual fruit weight (1141.67g), fruit length (13.03 cm) and breadth (13.15 cm), number of segments per fruit (14.41), number and weight of seeds per fruit (123.67 and 50.41g), yield per plant (50.94 kg) were recorded. It was revealed that parental lines fallen in this cluster having the genetic potentiality to contribute better for yield maximization of improved pummelo varieties. In case of cluster III, only the character % Brix (TSS) showed the highest mean value (9.50).Cluster IV showed highest mean values for canopy spreading (6.24 m) for north-south direction, and number of fruits per plant (43.77).Only canopy spreading (6.38 m) for east-west direction was highest mean value in Cluster V.

The results of principal component analysis (Table 4) revealed that the first axis largely accounted for the variation among the pummelo accessions (26.16%) followed by second axis (18.75%). The first 8 axes accounted 90.56 % of the total variation among 14 characters of describing 33 pummelo genotypes.The rest six characters contributed remaining 9.44% of total variation. In a study of diversity of acid lime, Ranpise and Desai (2003) found that fruits per tree, yield per plant, juice volume and juice percentage were major contributing traits towards divergence. The character with maximum contribution to the divergence should be given more emphasis for selection in breeding (Jagadev *et al.*, 1991).

Table 4. Latent root (Eigen value) and percent of variation in respect of fourteen characters in 33 pummelo accessions

Plant characters	Eigen value	Percent of Variance	Cumulative Percentage
Plant height (m)	3.6622	26.16	26.16
Canopy spreading at north-south direction (m)	2.6244	18.75	44.91
Canopy spreading at east-west direction (m)	2.1726	15.52	60.43
Number of flowers per cluster	1.2808	9.15	69.58
Number of fruits per plant	1.08	7.71	77.29
Individual fruit weight (g)	0.7334	5.24	82.53
Fruit length (cm)	0.6289	4.49	87.02
Fruit breadth (cm)	0.4958	3.54	90.56
Number of segments per fruit	0.4531	3.24	93.8
Number of seeds per fruit	0.3416	2.44	96.24
Weight of seeds per fruit (g)	0.3197	2.28	98.52
Edible percentage	0.1331	0.95	99.47
% Brix (TSS)	0.0660	0.47	99.94
Yield per plant (kg.)	0.0083	0.06	100

Contributions of different characters responsible for genetic divergence are presented in Table 5. The canonical variate analysis (CVA) revealed that invector I (Z_1), the important characters responsible for genetic divergence in the major axis of differentiation number of segments per fruit, number of flowers per cluster, number of fruits per plant, edible percentage, fruit breadth (cm),individual fruit weight (g),weight of seeds per fruit(g). In vector II (Z_2), fruit length (cm), canopy spreading at east-west direction (m), weight of seeds per fruit (g),plant height (m), yield per plant (kg.),individual fruit weight (g), number of seeds per fruit had positive impact towards divergence. The characters individual fruit weight (g) and weight of seeds per fruit (g) showing positive value in both the vectors contributed maximum towards divergence. So, the divergence in the present materials due to these two traits will offer a good scope for improvement of pummelo varieties through selection of parents.

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Table 5. Latent vectors for fourteen characters of 33 pummelo genotypes

Characters	Vector-I (Z_1)	Vector-II (Z_2)
Plant height (m)	0.0110	0.0214
Canopy spreading at north-south direction (m)	-0.0098	-0.0355
Canopy spreading at east-west direction (m)	-0.0460	0.0712
Number of flowers per cluster	0.3353	-0.2807
Number of fruits per plant	0.2379	-0.0444
Individual fruit weight (g)	0.0427	0.0031
Fruit length (cm)	-0.0090	0.3826
Fruit breadth (cm)	0.0536	-0.6583
Number of segments per fruit	0.3362	-0.3755
Number of seeds per fruit	-0.0148	0.0003
Weight of seeds per fruit (g)	0.0208	0.0670
Edible percentage	0.0613	-0.0709
% Brix (TSS)	-0.4291	-0.0778
Yield per plant (kg.)	-0.2619	0.0041

CONCLUSION

Crosses involving parents belonging to most diverse genotypes are expected to exhibit maximum heterosis and create wide variability in genetic architecture. Considering magnitude of genetic distance, contribution of different traits toward the total divergence, magnitude of cluster means for different traits and performance the genotypes of cluster I, II, and IV may be considered as parents for future hybridization program.

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