Detection of Genetic Diversity in Egyptian Cotton (*Gossypium barbadense* L.) Varieties Using RAPD Markers and Morphological Traits

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Abstract: Two marker systems, 19 RAPD and 8 agronomic traits were used to estimate the genetic diversity in Egyptian cotton. RAPD primers produced a total of 101 amplicons, which generated 86.25% polymorphism. Number of amplification products ranged from 2 to 7 where percent genetic similarity for the studied primers ranged from 72.2% to 89.9% with an average 81.4%. PIC values of the RAPD markers ranged from 0.855 (UBC 20) to 0.909 (UBC 54) with an average of 0.896 per marker. Highly significant differences were obtained between genotypes for all traits except boll weight, lint percentage and fiber strength. PCV were higher than its corresponding GCV for number of open bolls per plant, boll weight, seed cotton and lint yields per plant. However, no great difference between PCV and GCV for the three fiber characters. Broadsense heritability estimates were ranged from 17.18% to 90.97% for boll weight and fibre strength, respectively. High genetic advance under selection was noted for lint cotton yield per plant, seed cotton yield per plant, number of open bolls per plant, fiber strength, fiber length and micronair value. However, low genetic gain obtained for boll weight and lint percentage. Number of bolls per plant showed high positive phenotypic correlation coefficients with both seed cotton and lint yields per plant. This study of the genetic diversity of Egyptian cotton varieties with RAPD markers and agronomic traits support the need to introduce new alleles into the gene pool of the Egyptian cotton breeding program.
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Key Words: Cotton (*Gossypium barbadense* L) , RAPD markers, Genetic diversity, Heritability, Genetic advance.

1. Introduction

Cotton is an economically important crop, where it is cultivated in many parts of the world (60 countries) and it is a leading textile fiber, also important source for edible oil and protein. Cotton is one of the few crops that were grown anciently in both the Old and New Worlds. Cotton and cotton products occupy a pivotal position in the world economy. In Egypt, cotton is considered one of the most important cash crops. It is important to study the genetic diversity of Egyptian present cotton cultivars, which will be used for the development of new cotton genotypes.

Knowledge of genetic diversity and relationships among breeding materials is essential to the plant breeder for improving this crop. Genetic similarity estimates among genotypes are helpful in selecting parental combinations for segregating populations so as to maintain genetic diversity in a breeding program. Crosses between genetically divergent parents are expected to have a larger genetic variance among progenies than crosses between closely related parents (**Messmer *et al*., 1993**). Many studies have suggested that cultivated cotton germplasm shows a low level of genetic diversity when evaluated by isozymes, random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), restricted fragment length polymorphism and SSRs (**Wendel *et al*., 1992; Tatineni *et al*., 1996; Pilley and Myers, 1999; Abdalla *et al*., 2001; Iqbal *et al*., 2001; Gutierrez *et al*., 2002; Lu and Myers, 2002**). Due to modern breeding, it has been suggested that genetic diversity in cotton has been increasingly narrowed. Narrow genetic diversity is problem in breeding for adaptation to biotic stresses, like diseases, and abiotic stresses, such as drought or salt tolerance. Therefore, it is necessary to investigate the genetic diversity in cotton germplasm in order to broaden the genetic variations in future cotton breeding.

Agronomic traits can be used for assessing genetic diversity but are often influenced by the environment. The use of molecular markers for evaluation of the genetic diversity is receiving much attention (**Preetha and Raveendren, 2008**). Many cotton scientists have studied genetic diversity in common cotton using different molecular markers such as RFLPs (**Meredith and Brown 1998, Wendel and Brubaker, 1993),** RAPDs **(Tatineni *et***

***al*. 1996; Rana, *et al*. 2002; Kumar *et al.*, 2003; Vafaie-Tabar *et al*. 2003),** AFLPs **(Pillay and Myers, 1999; Abdalla *et al*., 2001**) and SSRs (**Wendel *et al*., 1992; Tatineni *et al*., 1996; Pillay and Myers, 1999; Abdalla *et al*., 2001; Iqbal *et al*., 2001; Gutierrez *et al*., 2002; Lu and Myers, 2002**). RAPD is one of the Polymerase Chain Reaction (PCR) based DNA marker, defined as an assay based on the amplification of genomic DNA with single primer of arbitrary nucleotide sequence (**Williams *et al*., 1990**). RAPD can be used in studying genetic diversity, varietal identification etc. It is simple to operate, less expensive, fast, does not involve radioactive labeling and dose not require huge infrastructure to start with. For research involving cotton (*Gossypium hirsutum* L.) the most widely used molecular method has been the random amplified polymorphic DNA (RAPD) technique (**Multani and Lyon, 1995; Tatineni *et al*., 1996; Iqbal *et al*., 1997; Lu and Myers, 2002, Khan *et al.*, 2011**). The goal of this study was to: i) investigate the genetic diversity among old and newly released of commercial Egyptian cotton cultivars using RAPD markers and agronomic traits, ii) evaluate genetic relationships between commercial Egyptian cotton varieties and iii) identify RAPD markers potentially associated with lint cotton yield and fiber quality.

2. Materials and Methods

Cotton materials and cultivation

Eleven Egyptian cotton varieties namely i.e., Ashmouni, Dandra, Giza 45, Giza 70, Giza 75, Giza 76, Giza 77, Giza 80, Giza 85, Giza 88 and Giza 89 were grown at the Experimental Research Station, National Research Center (NRC), Shalakan El-Kalyoubia, Egypt, during the two cotton successive growing seasons, 2006 and 2007 (Table 1). Varieties were grown in a randomized, complete-block design (RCBD) of three replicates with each plot consisting of two, 3 m long rows with 60 cm apart and 20 cm between hills. Normal agronomic practices were followed as recommended in the cotton production area.

Table 1: Names of the eleven cotton varieties and their pedigree

No	Variety name	Pedigree
1	Ashmony	MahoJumel x *G.barbadense* from south America
2	Dandara	Giza 31= Selected from Giza 3
3	Giza 45	Giza 28 x Giza 7
4	Giza 70	Giza 59A x Giza 51B
5	Giza 75	Giza 67 x Giza 69
6	Giza 76	Menoufi x Pima
7	Giza 77	Giza 70 x Giza 68
8	Giza 80	Giza 66 x Giza 73
9	Giza 85	Giza 67 x CB 58
10	Giza 88	(Giza 77 x Giza 45) B
11	Giza 89	Giza 75 x S. 6022

Evaluation of Agronomic Characters

Varieties were evaluated for a range of agronomic traits. Number of open bolls per plant, boll weight, seed cotton yield per plant and lint cotton yield per plant were determined on ten random plants per each plot. Fiber quality traits (fiber length, fineness and strength) and lint percentage were determined for each replicate.

DNA Isolation

RAPD Markers Analysis

RAPD (random amplified segments of DNA) were generated by 19 primer sets ordered from the University of British Colombia (UBC). The RAPD-PCR reactions were carried out using thermal cycler in 26 µl. RAPD reaction mixtures were 18.4 µl diH_2O, 2.5 µl 10x PCR buffer, 1.5 µl 25 mM $MgCl_2$, 0.5 µl dNTP, 1.0 µl UBC RAPD primer, 0.1 µl *Taq* DNA polymerase and 2.0 µl cotton genomic DNA.

Amplification for all UBC RAPD marker were carried out according to the following program conditions, initial denaturation at 94°C for 3 min, then denaturation for 45 cycles at 94°C for 1 min, annealing for 45 cycles at 40°C for 3 min, elongation for 45 cycle at 72° C for 1.3 min, followed by final elongation at 72°C for 5 min.

Data Collection and Diversity Analysis

Marker Polymorphism

Polymerase chain reaction (PCR) products were separated in 1.2% agarose gel using 1X TBE buffer and visualized by ultraviolet illumination after stained with ethidium bromide. To measure the

informativeness of the RAPD markers, the polymorphism information content (PIC) for each RAPD markers was calculated according to the formula: $PIC = 1 - \sum_{i=1}^{k} P_i^2$, where k is the total number of alleles detected for a locus of a marker and P the i frequency of the ith allele in the set of 11 varieties investigated.

Genetic Similarity Estimation and Cluster Analysis

Amplification profiles of the eleven cotton varieties were compared with each other and bands of DNA fragments were scored as a binary variable with (1) for presence and (0) for absence. Genetic similarity coefficients were calculated on the basis of Jacquard's coefficient (JC) using the Numerical Taxonomy Multivariate Analysis System (NTSYSpc) Version 2.1 software package. The resulting similarity coefficients were used to perform the cluster analysis by the unweighted pair group method of arithmetic mean (UPGMA). All calculations were performed using the NTSYS-pc version 2.1 software package (Biostatistics Inc., USA, **Rohlf, 1993**).

Statistical analysis

The analysis of variance for each trait was computed according to **Steel and Torrie (1980).** Phenotypic (PCV) and genotypic (GCV) coefficient of variation were calculated according to **Al-Jibouri *et al.* (1958)**. Broad sense heritability (h^2%) and expected genetic advance under selection were estimated as outlined by **Johanson *et al*. (1955).**

3. Results and Discussion

RAPD Polymorphism

Nineteen RAPD markers were used to characterize and evaluate the genetic diversity of the studied 11 cotton varieties. A total of 101 amplification products, among which 87 were found to be polymorphic (Table 2). This resulted in 86.25 % polymorphism. The number of amplification products per locus ranged from 4 (UBC 54, UBC57, UBC65 and UBC67) to 8 (UBC64) with an average number of 5.31 bands per locus (Table 2). All the primers produced polymorphic amplification products, however, the extent of percent polymorphism varied with each primer (50-100%). The number of polymorphic band per locus ranged from 2 (UBC57, UBC 65) to 7 (UBC61 and UBC64) with an average number of 4.58 bands per locus (Table 2). **Iqbal *et al.* (1997)** also observed that 98% of the primers in their study produced polymorphic profiles. **Lu and Myers (2002)** observed a low level of DNA variation among ten varieties of G. hirsutum, as they observed only 13.5% polymorphism. **Iqbal *et al*. (1997)** found 89.1% polymorphism among 23 G. hirsutum cultivars, wherein one cultivar was of the species *G. arboreum*. **Rahman *et al.* (2002)** observed 66.2% polymorphism in 27 cotton varieties. Conflicting reports on the extent of observed polymorphism in cotton in different studies could be attributed to the nature of the genetic material under investigation. The high degree of polymorphism in our study compared to other reports, could be due to the more diverse material which belonged to different cultivated varieties of cotton. Moreover, the various cultivars within a species represented different pedigree, agronomic traits and agroclimatic conditions for cotton growth.

Gene diversity

Gene diversity for 19 RAPD loci varied from 0.855 (UBC20) to 0.909 (UBC54) with an average of 0.896 (Table 2). Data in that table show all the studied primers produced polymorphic amplification products. In cotton, **Rana and Bhat (2004)** reported low gene diversity value 0.02 to 0.34 in 20 OPA-RAPD primers, which indicates that some RAPD markers are useful for differentiating between closely related genotypes.

Genetic distances for RAPD markers

Jaccard's pair-wise similarity estimates between genotypes were calculated and have been presented in Table (3). Genetic similarities based on Jaccard's among the 11 genotypes ranged from 0.722 to 0.899, with an average 0.814. This is a high result on genetic similarity among cotton. Other genetic diversity estimates in cotton have been reported using RAPD markers to range from 1 to 8% among Australian cultivars (**Multani and Lyon, 1995**).

Genetic relationship and diversity among different cotton varieties

A dendrogram derived from UPGMA cluster analysis based on the GS coefficient matrix for the 11 cotton varieties was constructed. Basically, all varieties could be distinguished. The genetic similarity coefficient for all varieties ranged from 72.2% to 89.9% and averaged 81.4% (Table 3).

On the basis of the Jacquard's coefficient, the 11 genotypes can be classified into 3 major groups (Fig. 1); (i) group I: includes Ashmony, Dandara, Giza 88, Giza 70, Giza 76, Giza 75, Giza 89; (ii) group II: includes Giza 45, Giza 85 and (iii) group III: includes Giza 77 and Giza 80. In general, Giza 45, Giza 85, Giza77 and Giza 80were the most genetically diversified from other cultivar sources and could be important sources for new cultivar development if they differ in useful agronomic traits.

Table (2): RAPD primer name, sequence, total number of amplified bands, total number of polymorphic bands, polymorphism percentage and gene diversity

No	Primer Name	RAPD Primer sequence	Amplified bands	Polymorphic bands	Polymorphism %	Gene diversity
1	UBC 17	CCT GGG CCT C	6	6	100	0.896
2	UBC 18	GGG CCG TTT A	6	6	100	0.908
3	UBC 19	GCC CGG TTT A	7	5	71.43	0.885
4	UBC 20	TCC GGG TTT G	5	5	100	0.855
5	UBC 23	CCC GCC TTC C	6	6	100	0.894
6	UBC 25	ACA GGG CTC A	5	4	80	0.905
7	UBC 28	CCG GCC TTA A	5	5	100	0.866
8	UBC 38	CCG GGG AAA A	5	4	80	0.904
9	UBC 48	TTA ACG GGG A	5	4	80	0.896
10	UBC 53	CTC CCT GAG C	5	5	100	0.903
11	UBC 54	GTC CCA GAG C	4	3	75	0.909
12	UBC 57	TTC CCC GAG G	4	2	50	0.906
13	UBC 59	TTC CGG GTG C	5	5	100	0.900
14	UBC 60	TTG GCC GAG C	5	3	60	0.907
15	UBC 61	TTC CCC GAC C	7	7	100	0.908
16	UBC 64	GAG GGC GGG A	8	7	87.5	0.907
17	UBC 65	AGG GGC GGG A	4	2	50	0.906
18	UBC 67	GAG GGC GAG C	4	4	100	0.878
19	UBC 68	GAG CTC GCG A	5	4	80	0.899
Total			101	87	----	17.032
Average			5.31	4.58	86.25	0.896

Table (3): Genetic similarity estimates for 11 Egyptian cotton varieties based on RAPD analysis

Variety	Ashmouni	Dandara	Giza 45	Giza 70	Giza 75	Giza 76	Giza 77	Giza 80	Giza 85	Giza 88	Giza 89
Ashmouni	-----										
Dandara	0.821	-------									
Giza 45	0.816	0.730	-----								
Giza 70	0.809	0.819	0.862	-----							
Giza 75	0.809	0.869	0.785	0.873	-----						
Giza 76	0.834	0.875	0.838	0.878	0.864	-----					
Giza 77	0.745	0.722	0.787	0.764	0.732	0.806	-------				
Giza 80	0.745	0.789	0.771	0.829	0.748	0.822	0.867	-----			
Giza 85	0.750	0.744	0.899	0.864	0.800	0.824	0.787	0.803	-----		
Giza 88	0.782	0.862	0.790	0.867	0.850	0.809	0.769	0.821	0.789	-----	
Giza 89	0.833	0.843	0.822	0.864	0.928	0.870	0.770	0.754	0.806	0.857	-----

It should be noted here that cultivar grouping here by cluster analysis depended on the polymorphic RAPD bands. Cultivars grouped together by the RAPD markers could have noticeable phenotypic differences in morphology, growth habits and agronomic traits.

The cotton genotypes mean performance over two years for agronomic traits are given in table (4). Therefore, clear variations were observed for number of open bolls per plant, boll weight, seed yield per plant and lint yield per plant and fiber quality traits (fiber length, fineness and strength) and lint percentage (Table 4). These traits were used for characterization and the observed differences among varieties indicated the possibility of using morphological markers to differentiate varieties for

germplasm collection and maintenance and for selection of suitable parents.

Table (5) shows the results of the analysis of variance for the traits studied. Highly significant differences were obtained between genotypes for all traits except boll weight lint percentage and fiber strength.

Table (4): The cotton genotype mean performance for agronomic traits of eleven cotton varieties over two years

Genotypes	No of bolls/ plant	Boll Weight (g)	Seed cotton yieled/ plant (g)	Lint cotton yield/plant (g)	Lint %	Fiber Length (mm)	Micron air	Strength
Ashmony	15.43	2.88	43.96	16.02	36.43	35.43	4.40	35.74
Dandara	15.32	2.86	41.65	14.98	35.98	34.39	4.24	35.09
Giza 45	13.55	2.86	37.33	13.31	35.86	34.26	4.22	33.04
Giza 70	13.39	2.81	36.37	12.96	35.64	33.93	4.11	32.95
Giza 75	13.15	2.77	35.26	12.47	35.54	33.41	4.01	32.87
Giza 76	12.18	2.77	34.76	11.88	35.42	31.25	3.99	31.27
Giza 77	11.81	2.72	31.09	11.10	35.37	30.97	3.90	31.13
Giza 80	11.36	2.63	30.97	11.02	35.28	30.45	3.88	29.88
Giza 85	11.26	2.60	29.59	10.44	35.28	30.37	3.85	29.50
Giza 88	10.77	2.60	29.33	10.21	34.83	30.36	3.83	27.19
Giza 89	10.52	2.48	28.11	9.92	34.30	30.03	3.72	26.57
L. S. D. 0.05	1.27	0.17	4.07	1.49	0.76	0.79	0.008	1.16
L. S. D. 0.01	1.72	0.24	5.53	2.02	1.03	1.08	0.011	1.58

Table (5): Mean square estimates of analysis of variance agronomic traits studied.

Source of variance	df	No of bolls/ plant	Boll weight	seed cotton yieled/ plant	Lint cotton yield/plant	Lint percentage	Fiber length	Micronair	Strength
Blocks	2	0.935	0.069	16.84	2.63	0.780	0.488	0.011	0.317
Genotypes	10	8.71**	0.051	80.49**	11.75**	0.964	12.26**	0.131**	26.01
Error	8	1.653	0.0315	17.057	2.281	0.594	0.652	0.006	0.833

*** and **, significant and highly significant at 0.05 and 0.01 level of probability, respectively**.

Genetic distances for agronomic characteristics

The genetic distance matrix for 11 varieties from a combination of 8 characteristics is given in Table (6) and distances ranged from 1.007 to 9.562 .

Cluster analysis

The dendrogram (Figure 2) consisted of two main groups. The first group divided into two more sub-clusters; the first sub-clusters included Ashmouni. While the second sub-clusters divided into two sub-sub-clusters, the first one included Giza 70. However, the second branches included Giza 76 and Giza 89. On the other hand, the second group divided into two more sub-clusters; the first sub-cluster divided into two sub-sub-clusters, the first one included Dandra and Giza 75. While the second one included Giza 80. On the other hand, the second branches included Giza 45 and Giza 85. The second main group includes Giza 77 and Giza 88.

The main objectives for this study were to study genetic diversity among Egyptian cotton varieties using agronomical and RAPD analyses and compare these characterization methods. Consideration of estimated genetic distance is important for comparative analysis of diversity levels (**Roldan-Ruiz *et al.*, 2001**). The overall mean of genetic similarities for agronomical data were ranged from 1.007 to 9.562 for agronomic and 0.722 to 0.899 for RAPD marker data. Therefore low genetic similarities were observed using agronomical compared to RAPD analyses. This is because agronomical characterization deals with genetic

similarities based on quantitative traits, which are influenced by the environment. According to **Poehlman (1987)**, quantitative traits that are controlled by a number of genes with small effects are tremendously influenced by environment. Genetic similarities of RAPD did not support this wider range of agronomical genetic similarities. This indicated the presence of differences between these two methods.

Table (6): Genetic similarity matrix values for the 11 cotton varieties based on agronomic traits

	Ashmony	Dandara	Giza45	Giza70	Giza75	Giza76	Giza77	Giza80	Giza85	Giza88	Giza89
Ashmony	0.000										
Dandara	5.316	0.000									
Giza45	2.444	3.235	0.000								
Giza70	2.124	6.691	3.649	0.000							
Giza75	6.340	1.069	4.292	7.756	0.000						
Giza76	1.969	7.037	3.878	1.699	8.082	0.000					
Giza77	1.304	7.886	1.110	1.454	6.817	1.483	0.000				
Giza80	7.687	2.974	6.015	9.288	2.110	9.562	5.457	0.000			
Giza85	3.261	2.377	1.745	4.488	3.383	5.057	1.007	4.842	0.000		
Giza88	1.894	1.383	1.706	2.042	1.277	2.078	6.005	1.127	1.594	0.000	
Giza89	2.702	7.709	4.562	1.267	8.771	1.216	1.556	1.027	5.569	2.147	0.000

Phenotypic (PCV) and genotypic (GCV) coefficient of variation

Estimates phenotypic and genotypic variances (δ^2 P and δ^2G) and their coefficients of variations (PCV and GCV) are presented in Table (7). The phenotypic coefficients of variation (PCV) values were higher than its corresponding genotypic coefficients of variation (GCV) for number of open bolls per plant, boll weight, seed cotton yield per plant and lint yield per plant and lint percentage suggesting that these traits are more sensitive to the environmental conditions. However, no great difference between phenotypic and genotypic coefficient of variation for the three fiber characters indicating that these characters are less sensitive to the environmental conditions. Broad sense heritability estimates were greatly variable among the studied traits Table (7). High broad sense heritability estimates were observed for the three fiber properties i.e., fiber length (85.59%), micronaire value (87.37%) and fiber strength (90.97%). Moderate heritability values were found for number of bolls per plant (58.72%), seed cotton yield (55.35%) and lint yield (58.05%). Low broad sense heritability estimates were found for boll weight (17.178) and lint percentage (17.193).

Table (7): Estimates of variance components, genotypic (GCV) and phenotypic (PCV) coefficients of variability, broad sense heritability (h^2_b) and expected genetic advance (G.S%) for studied traits.

Trait	Grand mean	Components of variance			Genetic variability		H^2_b (%)	Genetic advance	
		σ^2g	σ^2e	σ^2ph	GCV	PCV		G.S	G.S (%)
No of bolls/plant	12.61	2.35	1.65	4.01	12.16	15.86	58.73	2.42	19.19
Boll weight	2.73	0.007	0.03	0.03	2.96	7.15	17.18	0.07	2.53
Seed cotton yield/plant	34.40	21.14	17.05	38.20	13.36	17.96	55.35	7.04	20.48
Lint yields/plant	12.21	3.16	2.28	5.43	14.55	19.09	58.05	2.78	22.84
Lint percentage	35.44	0.12	0.59	0.72	0.99	2.38	17.19	0.30	0.84
Span length	32.25	3.87	0.65	4.52	6.10	6.59	85.59	3.75	11.62
Micronair value	4.013	0.04	0.006	0.05	5.07	5.43	87.38	0.39	9.77
Fiber strength	31.38	8.39	0.83	9.23	9.23	9.67	90.97	5.69	18.13

Mohamed *et al.* (2003) showed low heritability in broad sense for boll weight (3.4%) and high value for seed cotton yield (80.4%) and lint yield (79.8%). Esmail *et al.* (1999) found high broad sense heritability estimates for number of open bolls per plant and seed cotton yield and moderate values for boll weight and lint percentage. Basbag and Gencer (2004) estimated moderate heritability values of the

seed cotton yield and plant height and low heritability estimate of the number of bolls per plant.

Esmail (2007) found high broad sense heritability for boll weight (85.1%), seed cotton yield (78.0%) and lint yield (77.2%), moderate heritability for number of open bolls per plant (69.6%) and lint percentage (61.6%).

High genetic advance under selection was noted for number of open bolls per plant, seed cotton yield, lint yield and fiber strength. However, low genetic gain obtained for boll weight and lint percentage, suggesting selection practice is limited scope for improvement of these traits Table (7). **Ahmed *et al.* (2006)** and **Esmail (2007)** found similar results. On the contrary, for traits with low heritability, the genetic gain will increase if they are selected on a family basis **Moreno-Gonzalez and Cubero (1993)**.

Phenotypic correlation coefficients (rph) estimated among all possible pairs of traits studied are presented in Table (8). Number of bolls per plant showed highly positive significant phenotypic correlation coefficients with both seed cotton and lint yield per plant. The existence of close relationship between these traits has enabled cotton breeder to obtain more improvement in seed cotton yield through increasing the boll numbers per plant. Also, boll weight showed positive correlations coefficient with the seed cotton and lint yield per plant, as well as, lint percentage with lint cotton yield. Strong positive correlations coefficient (0.737) was found between fiber strength and fiber length. Negative association was found between fiber strength and fiber fineness. Correlation between traits can be useful in developing selection criteria, but it can also present a morass of interrelationships (**Kloth, 1998**). **Falconer (1989)** reported that if two traits are associated and one is easier to asses and select, selection pressure showed be applied to this trait to improve the other.

Table (8): Estimates of phenotypic correlation coefficients among all studied traits

Trait	No of bolls/ plant	Boll weight	Seed cotton yield/ plant	Lint cotton yield/plant	Lint percentage	Span length	Micronair	Strength
No of bolls/plant	-----	**0.052**	**0.903****	**0.890****	**0.324**	**0.234**	**-0.176**	**0.409**
Boll weight		------	**0.472***	**0.475***	**0.160**	**0.005**	**0.394**	**0.020**
Seed cotton yield/plant			------	**0.991****	**0.366**	**0.220**	**0.001**	**0.375**
Lint cotton yield/plant				------	**0.483***	**0.236**	**0.035**	**0.337**
Lint percentage					-------	**0.156**	**0.225**	**-0.133**
Span length						-------	**-0.332**	**0.737****
Micronair							-------	**-0.417**
Fiber strength								-------

*** and **, significant and highly significant at 0.05 and 0.01 level of probability, respectively.**

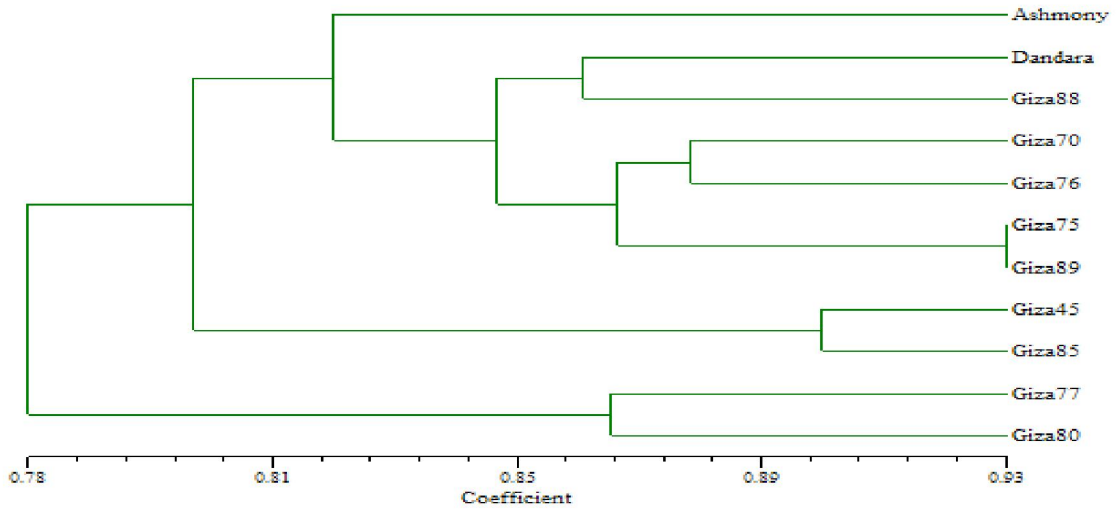

Figure (1): Dendrogram generated based on UPGMA clustering method and Jacquard's coefficient using RAPD analysis among 11 cotton varieties (Agronomic Traits)

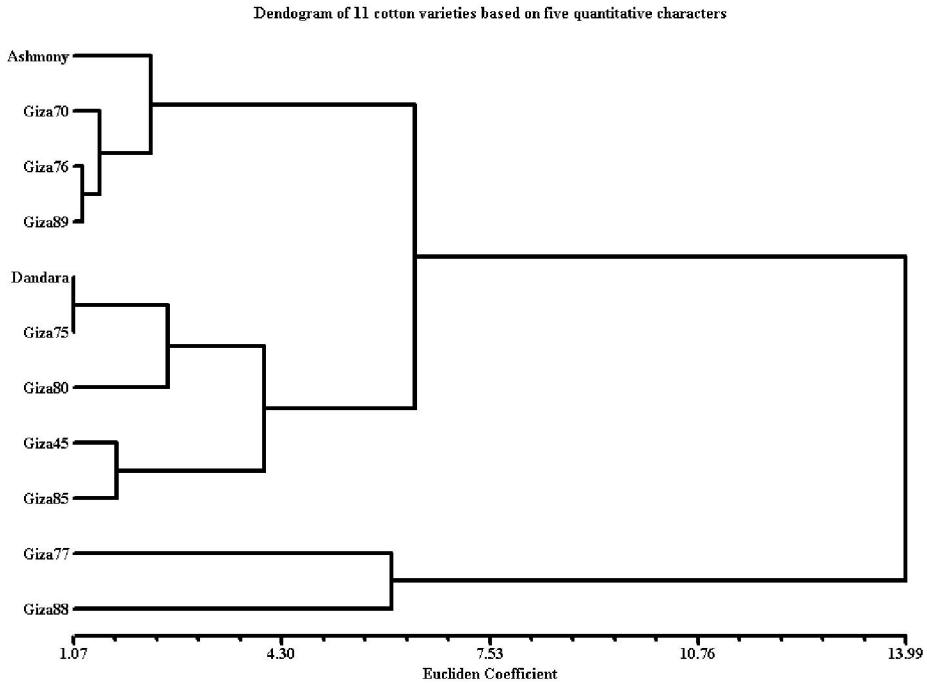

Figure (2): Agronomical characterisation of 11 varieties using UPGMA clustering method and Eucliden similarity coefficient

It could be concluded that RAPD markers were found to reveal sufficient genetic diversity and a high level of polymorphism. Low levels of correlation existed between agronomical and RAPD based genetic similarities in the current study. RAPD analysis reflected the true expression of genotypes, while agronomical analysis encompassed the expression of genotype, environment and their interactions.

Agronomical characteristics are not consistent and few, whereas RAPD analysis appeared to provide more accurate estimates and utility of genetic diversity measurements. All methods have advantages and disadvantages for practical applications under different circumstances. Consequently, both methods should continue rendering valuable services to farmers, breeders and genetic resource curators. The overall findings from this study indicated that RAPD analysis and to a certain extent qualitative traits and quantitative traits, sufficiently detected genetic diversity to differentiate Egyptian cotton varieties. Genetically distinct varieties were identified that could be potentially important sources of germplasm for cotton improvement. Although all methods did not provide exactly the same description of relationships between varieties, they existed some consistency in discriminating varieties which were closely related and ones which were distantly related. RAPDs analysis are more efficient and provide exciting insights (**Lu and Myers, 2002; Kumar *et al.*, 2003, Khan *et al.*, 2011**). Application of DNA markers could accelerate the process of finding markers related to specific agronomical traits of interest, such as disease and pest tolerance (**Spielmeyer *et al.*, 1998**). Gossypium barbadense has limited genetic diversity, therefore RAPD analysis may offer a powerful tool for analyzing the inheritance and relationships of important traits in cotton breeding. Therefore, future research should be focused on comparing the two methods in terms of feasibility, efficiency and accuracy by involving more tests over different environmental trials and years (for agronomic characteristics). Molecular analysis using more primer combinations and different molecular markers, along with costs and benefits, should be included.

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References

1. Abdalla AM, Reddy OUK, El-Zik KM, Pepper AE. (2001). Genetic diversity and relationship of diploid and tetraploid cottons revealed using AFLP. Theor. Appl. Genet., 102: 222-229.

2. Ahmed HM, Kandhro MM, Laghari S, Abro S. (2006). Heritability and genetic advance as selection indicators for improvement in cotton (*Gossypium hirsutum* L). J. Biol. Sci., 6 (1): 96-99.
3. Al-Jibouri HA, Miller PA, Robinson HF. (1958). Genotypic and environmental variances and co-variances in upland cotton cross of interspecific origin. Agron. J., 50: 633-636.
4. Basbag S, Gencer O. (2004). Investigations on the heritability of seed cotton yield, yield components and technological characters in cotton (*Gossypium hirsutum* L). Pakistan J. of Biol. Sci., 7 (8): 1390-1393.
5. Esmail R. M. (2007). Early generation testing for vegetative and yield characters in thirty Egyptian cotton populations. Bull. N.R.C. Egypt, 32 (4): 445- 457.
6. Esmail RM, Hendawy FA, Rady MS, Abdel-Hamid AM. (1999). Genetic studies on yield and yield components in one inter-and two intra-specific crosses of cotton. Egypt. J. Agron., 21: 37-51.
7. Falconer DS. (1989). Introduction to Quantitative Genetics. Longman Group, New York.
8. Gutierrez OA, Basu S, Saha S, Jenkins JN, Shoemaker DB, Cheatham CL, McCarty JC. (2002). Genetic distance among selected cotton genotypes and its relationship with F_2 performance. Crop Sci., 42: 1841-1847.
9. Iqbal MJ, Aziz N, Saeed NA, Zafar Y, Malik KA. (1997). Genetic diversity evaluation of some elite cotton varieties by RAPD analysis. Theor. Appl. Genet., 94: 139-144.
10. Iqbal MJ, Reddy OUK, Ez-Zak KM, Pepper AE. (2001). A genetic bottleneck in the evolution under domestication of upland cotton. (*Gossypium hirsutum* L.) examined using fingerprinting. Theor. Appl. Genet., 103: 547-554.
11. Johanson HW, Robinson HF, Comstock RE. (1955). Estimation of genetic and environmental variability in soybeans. Agron. J., 47: 314-318.
12. Khan AI, Khan IA, Awan FS, Sadaqat HA, Bahadur S. (2011). Estimation of genetic distance based on RAPDs between 11 cotton accessions varying in heat tolerance. Genetics and Molecular Research, 10 (1): 96-101
13. Kloth RH. (1998). Analysis of Commonality for Traits of Cotton Fiber. J. Cotton Sci., 2:17-22.
14. Kumar P, Singh K, Vikal Y, Randhawa LS, Chahal GS. (2003). Genetic diversity studies in elite cotton germplasm lines using RAPD markers and morphological characters. Indian J. Genet., 63: 5-10.
15. Lu HJ, GO Myers. 2002. Genetic relationships and discrimination of ten influential upland cotton varieties using RAPD markers. Theor. Appl. Genet. 105: 325-331.
16. Meredith WR, Jr. Brown JS. (1998). Heterosis and combining ability of cotton originating from different regions of the United States. J. Cotton Sci., 2:77-84.
17. Messmer MM, Melchinger AE, Herrmann RG, Boppenmaier J. (1993). Relationship among early European maize inbreds: II. Comparison of pedigree and RFLP data. Crop Sci., 33: 944-950.
18. Mohamed SAS, El-Adly HH, Eissa AEM. (2003). Evaluation of some Egyptian cotton genotypes under different environments. Egypt J. Agric. Res., 81 (4) :1797-1816.
19. Moreno-Gonzalez J, Cubero JI. (1993). Selection strategies and choice of breeding methods. In: Plant Breeding Principles and Prospects. Eds. Hayward. M. D., Bosemark, N. O. and I. Romagosa. Chapman & Hall, New York.
20. Multani, DS, Lyon BR. (1995). Genetic fingerprinting of Australian cotton cultivars with RAPD markers. Genome, 38: 1005-1008.
21. Pillay AA, Myers GO. (1999). Genetic diversity in cotton assessed by variation in ribosomal RNA genes and AFLP markers. Crop Sci., 39: 1881-1886.
22. Poehlman JM. (1987). Breeding field crops. (second edition), 724pp. AVI Publishing Company Ltd Westport, Connecticut, USA.
23. Preetha S, Raveendren TS. (2008). Molecular marker technology in cotton. Biotechnology and Molecular Biology Review, 3 (2): 032-045.
24. Rahman M, Hussain D, Zafar Y. (2002). Estimation of divergence among elite cotton cultivars-genotypes by DNA fingerprinting technology. Crop Sci., 42: 2137-2144.
25. Rana MK, Bhat KV. (2002). Genetic diversity analysis in Indian diploid cotton (*Gossypium* spp.) using RAPD markers. Indian J. Genet., 62: 11-14.
26. Rohlf FJ. (1993). NTSYS-pc. Numerical taxonomical and multivariate analysis system. Exeter Software, Setauket, New York, USA.
27. Roldan-Ruiz I, van Eeuwijk FA, Gilliland TJ, Dubreuil P, Dillmann C, Lallemand J, De Loose M, Baril CP. (2001). A comparative study of molecular and morphological methods of describing relationships between perennial ryegrass (*Lolium perenne* L.) varieties. Theor. Appl. Genet., 103: 1138-1150.
28. Spielmeyer W, Green AG, Bittisnich D, Mendham N, Lagudah ES. (1998). Identification of quantitative trait loci contributing to Fusarium wilt resistance on AFLP linkage map of flax. Theor. Appl. Genet., 97: 633-641.
29. Steel RGD, Torrie JH. (1985). Principles and Procedures of Statistics. Mc Graw-Hill. New York.
30. Tatineni V, Cantrell RG, Davis DD. (1996). Genetic diversity in elite cotton germplasm determined by morphological characteristics and RAPDs. Crop Sci., 36: 186-192.
31. Vafaie-Tabar M, Chandrashekaran S, Singh SP, Rana MK. (2003). Evaluation of genetic diversity in Indian tetraploid and diploid cotton (*Gossypium* spp.) by morphological characteristics and RAPDs. Indian J. Genet., 63: 230-234.
32. Wendel JF, Brubaker CL, Percival AE. (1992). Genetic diversity in *Gossypium hirsutum* and the origin of upland cotton. American J. Botany, 79: 1291-1310.
33. Wendel JF, Brubaker CL. (1993). The gene pool of *G hirsutum*. In: D.J. Heber and D.A. Richter (Eds.), Proceedings Belt wide cotton conference, 1556pp. National Cotton Council, Memphis, TN.
34. Williams J, Kubelik A, Liviak JL, Rafalski JA, Tingey SV. (1990). DNA polymorphism amplified by random primers are useful as genetic markers. Nucleic acid Res., 18: 6531-6535.