

Genetic Diversity Based on SSR Markers and Morphological Analysis of faba bean (*Vicia faba* L.) in Egypt EL-Shaer¹, H.F.A. and Helal² A.A.

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Abstract

Vicia faba L. is the most vital legumes and widely cultivated in all the world. Cultivation of tolerant genetic variants is the correct way to avoid the influences of drought on crops. Allocated space adequacy of genetic difference in commercial bean cultivars. Molecular markers are benfial tools for choice among sorts. Therefore, the aim of this study was to evaluate the genetic and morphological variations to for further application in plant breeding research. In this study, five simple sequence repeats (SSR) primers were used to explain genetic and morphological diversity from 18 Egyptian faba beans cultivars were collected from different places in Egypt; 14 lines from different population in addition Giza 2, Giza 3 and Giza 716 and T.W from Sudan. The results of this study SSR markers are effective in estimating the germplasm of Egyptian beans. SSR markers results ranged from 15 bp to 202 bp with average of allele number 9.2 allele/markers. In addition, the morphological variation showed that obvious differences between the 18 genotypes in the (2017-2018) and (2018-2019) seasons also. Data results from SSR and quantitative traits showed that the best nine genotypes were in vegetative traits and tolerating conditions in Egypt Giza 2, L.168, L.171, L.175, L.180 B, L.180 C, L.219, L.223 then T.W compared with the all genotypes and the cultivated varieties.

Keywords: Vicia faba L., genetic diversity, SSR marker.

Introduction

Vicia faba L. is the most vital legumes and widely cultivated in all the world. It supports significant source of all living things including human nutritional protein diet and an excellent source for as animal feed in China and Europe, hence both are basic faba bean producers (Duc et al., 2010). The area planted with beans in the world is 2.55 million hectares and the global production is 4.3 million tons. Average productivity in Egypt is about 3 tons/hectare then Sudan (2-2.5 tons/ha), China (1.5-2 tons/ha) and Ethiopia (1-1.5)tons/ha) (FAOSTAT 2011 http://faostat.fao.org/default.aspx). Beans other than legumes are the staple food in many countries. Cultivation of tolerant genetic variants is the correct way to avoid the influences of drought on crops.

Allocated space adequacy of genetic difference in commercial bean cultivars. Molecular markers are benfial tools for choice among sorts (Tehrani *et al.*, 2009 and Hajibarat *et al.*, 2015).

Microsatellites SSRs are clusters of short tandem repeated nucleotide bases spared all over the genome. Simple sequence repeats (SSRs) markers are an excellent tool in genetic studies and marker-assisted differentiation in breeding due to their characterization of co-sites and their high allelic difference (Gupta and Varshney 2000 and Hernandez *et al.*, 2002). The classic style for developing SSR tags is an SSR enriched library architecture, cloning and sequencing this process that costs a lot of scientists (Yu *et al.*, 2009). One of the most successful methods is to use molecular markers to assess genetic diversity and to show the differences between the genetic origins of crops (Hajibarat *et al.*, 2015). Uses SSR markers in genetics research and plant breeding as they are prevalent have several advantages such as easy to score, less budget and results are consistent. Despite these few previous studies have used SSR markers as a tool to study genetic variation in beans (Rebaa *et al.*, 2017; Tufan and Erdogan 2017 and Tahir *et al.*, 2019).

In this study, we evaluated 18 faba bean genotypes from various populations for genetic variability and sorted them widely depend on the genetic background and/or origin. Five SSR primers combination were used to estimate the genetic variation in genotypes of Egyptian beans with morphological evaluation under Egyptian conditions. Therefore, this study was resulted to estimate the genetic variability in 18 faba bean genotypes of diverse origin and morphology using SSR markers and evaluate the morphology and yield related traits in dry Mediterranean environment of Egypt for identification of the faba bean genotype(s) for further application in plant breeding.

Material and methods Plant materials

Eighteen faba bean genotypes collected from Egypt population were used in this study (Table1). The present investigation was done at the Agricultural Experiments and Researches Station, Bahteem, A.R.C, Egypt, at the two seasons of (2017-2018) and (2018-2019) so as to evaluation of some faba bean genotypes.

No.	Genotypes	Origin	Pedigree
1	Giza 2	Egypt	Individual plant selection from local variety
2	L.114	Landraces	Wadi Khairat, Aswan.
3	L.136	Landraces	19 Km. of Kalabsha, Aswan.
4	L.168	Landraces	Beni fezz, near safa, Assiut.
5	L.171	Landraces	Qena
6	L.174	Landraces	El-Abbadla, Tamia, Faiyum.
7	L.175	Landraces	14 Km. East Naji Hammadi, Qena.
8	L.180 A	Landraces	5 Km. of West meet ghamr, Dakahlia.
9	L.180 B	Landraces	Individual plant selection from 180A
10	L.180 C	Landraces	Individual plant selection from 18b
11	L.185	Landraces	5 Km. East Tanta, Gharbia.
12	L.212	Landraces	Ezbet Khalil Ibraham, Faiyum.
13	L.219	Landraces	13 Km. of West Elminia, Faiyum
14	L.220	Landraces	Ihnasya, El Madina, Beni Suef.
15	L.223	Landraces	3 Km. of Beni Mazar, Elminia.
16	Giza 716	Egypt	Giza 416×Giza 503
17	Giza 3	Egypt	Giza 1× New Accession 29
18	T.W.	Sudan	Individual plant selection in Sudan

Table (1): Origin and pedigree of the faba bean genotypes.

The names of pedigree follow the global geographic reference system.

In the current study, 14 lines from various population plus Giza 2, Giza 3 and Giza 716 from Egypt and T.W from Sudan of faba bean crop (*Vicia faba* L.) were studied and evaluated under the Egyptian

conditions. The eighteen faba bean genotypes are differed in morphological possessions and its origin and pedigree.

The experiment was coordinated in a randomized complete block design (RCBD) with four replicates. Each of them

divided into eighteen plots. The plot was three ridges, three meters long and 50 cm apart and hills (two seeds in hills) were positioned at 20 cm distance. Seeds were hand affair (dry seed in dry soil then irrigation) sown in the two sides of the ridge with 2 seeds per hill. All plots were watered by surface irrigation system. Mohaya irrigation was done after 15 day from sowing date. After that, every 15-17day intervals for all genotypes according to area conditions.

During two seasons the hoeing process and herbicide was done during two seasons, Gysabrain herbicide that was added after planting and before sowing irrigation of faba bean plants at the rate of one kg/Fed. It was added by spraying (1kg/200 liter of water/fed).

Studied attributes:

At harvest time we follower the faba bean mentioned in the International Board for Plant Genetic Resources (IBPGR 1985). Five plants were randomly selected from each plot and some measurements were taken. They were recorded to determine the following traits:

Plant height (cm), Number of branches/plant, Number of pods/plant, Number of pods/plant, Number of pods/plot, First fruiting nods/plant and Number of seeds/plants.

Statistical analysis:

Statistical analysis was performed after analysis of variance techniques (ANOVA) as described by Gomez and Gomez (1984). The mean values were compared at 5% level of significance utilize least significant variation (L.S.D) test using GENSTAT software.

DNA extraction:

Total DNA from each genotype were isolated from faba bean using DNeasy Plant Mini Kit (QIAGEN, Hilden, Germany).

PCR amplification:

Five SSR primer were used for DNA amplification (Table 2). Each PCR reaction (10 µl final volume) contained 5 ng of DNA, $1 \times PCR$ buffer (10 mM Tris-HCl pH 9.0; 50 mM KCl; 1.5 mM MgCl₂), 200 μM of each dNTP, 0.12 µM of each primer and 0.5 U of Ampli *Taq* DNA polymerase. PCR reactions were performed in an MJ PT-200 thermocycler (MJ Research Inc., Waltham, Mass.). The amplification profile consisted of an initial denaturation for 4 min at 94 °C followed by 32 cycles of 45 second at 94 °C, 30 second at the annealing temperature (40-60.5 °C, depending on primer), 30 seconds elongation at 72 °C, and a final extension step of 5 min at 72 °C. The reaction products were denatured by heating for 3 min at 94 °C with equal volume of tracking dye (98% formamide, 10 mM of EDTA, 0.25% each of bromophenol blue and xylene cyanol) then electrophoresed on 6% denaturing gels (19:1 acrylamide-bisacrylamide 7.5 M urea) in 1×TBE buffer (50 mM Tris, 50 mM boric acid, 1 mM EDTA pH 8.0) at 70 W for 2.5–6 h, depending on the fragment sizes.

Table (2): Primer sequence, of faba bean SSR markers from database.

Marker Name	Marker Type	Primer Forward	Primer Reverse
FBES0025	SSR25	AATGGGATTCGTGCTTGTTC	CAAAGCATGACCCTATAAAGATTAG
FBES0026	SSR26	ACTCTAACCAAACCGCAGCA	TTGAATGGGAGATGGAGAGG
FBES0028	SSR28	CCTTACGTTGATTTCCTCCG	TTATTGGGATTCGGAGAAGG
FBES0030	SSR30	CACCTCGTCCACCTTCAAGT	ATTTGTTCGAGTTCGGTTGG
FBES0031	SSR31	ACTTCCTCCTCCTCCACCTC	TTTGCAAATCCTTTCGATGA

Data analysis

The program POPGENE version 1.31 (Yang and Yeh, 1993) calculate allele frequencies alleles per locus (N_e), observed heterozygosity (H_o) and Nei's (1973) expected heterozygosity (H_e) for each

locus. Nei's (1978) genetic identity (I) and genetic distance (D) were deliberate for all pairs of ecogeographical groups and subgroups and worth of (D) were used to conduct cluster analysis with a UPGMA algorithm and construct a dendrogram. Pairwise genetic distances among the 18 genotypes were calculated using the program MICROSAT (Minch 1997); cluster analysis was conducted with a neighbor joining algorithm using the NEIGHBOR program in PHYLIP version 3.5c (Felsenstein, 1989); and a dendrogram constructed using the program was TREEVIEW (Page, 1996). For both cluster analyses. The analysis was conducted ten times with samples in randomized order each time and the ten analyses were compared using the CONSENSE program in PHYLIP.

Results and Discussion

Molecular and agronomic estimation is used for exploration of genetic diversity between the genotypes and landraces. These the first step for advantage improvement through plant breeding programs (Farooq *et al.*, 2017).

In this study, the analysis of difference explained that the genotypes under study varies significantly in morphological and yield related traits (Table 3, 4, 5 and 6).

Plant height (cm) and number of branches/plants

Data recorded in Table (3) showed that plant height (cm) and number of

branches/plant traits as impacted by the variances between the 14 lines (2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14 and 15) comparing with 4 local varieties (1,16,17 and 18) for faba bean plants during the growing seasons of 2018 and 2019. Results indicate that there were significant differences between the18 genotypes of faba bean plants under study. The genotypes18 and 9 scored the highest plant height (121.66 and 126.66 cm) during 2018 and 2019 seasons respectively. On the other hand, the shortest plant height recorded by genotype 3 (61.66 cm) in 2018 season and genotype 12 (65.00 cm) in 2019 season as compared with other faba bean lines and local varieties under study from different population.

As for, number of branches per plants results showed that genotype 15 and 4 recorded the largest number of branches (5.33 and 5.66) during 2018 and 2019 seasons respectively as genotype 18 recorded the lowest values (2.00 and 2.66) for the same traits at the same two seasons. These results are consistent with (Rebaa *et al.*, 2017 and Elshafei *et al.*, 2019) since they found statistically significant differences between bean genotypes and all characteristics.

Table (3): plant height and Number of branches/plants as affected by variances between faba bean lines during 2018 and 2019 seasons.

faba bean	Plant hei	ght (cm)	Number of branches/Plants				
genotypes	Season 2018	Season 2019	Season 2018	Season 2019			
1	95.00	116.66	3.00	3.00			
2	75.00	106.66	4.00	3.33			
3	61.66	120.00	3.66	3.33			
4	101.66	120.00	4.66	5.66			
5	93.33	130.00	3.00	4.00			
6	100.00	98.33	4.66	3.00			
7	86.66	110.00	3.00	4.00			
8	105.00	108.33	2.66	3.00			
9	76.66	126.66	2.66	4.66			
10	90.00	98.33	2.66	33.3			
11	70.00	93.33	3.00	4.33			
12	73.33	65.00	2.66	3.00			
13	90.00	113.33	3.66	4.66			
14	73.33	91.66	3.00	3.00			
15	76.66	108.33	5.33	3.66			
16	98.33	106.66	3.33	3.00			
17	81.66	105.00	3.00	3.00			
18	121.66	125.00	2.00	2.66			
L.S.D. at 5%	24.22	15.72	1.34	1.32			

L.S.D. is the level of significance utilize least significant variation at 5%

Number of pods/Plot and Seed weight/plot

Data illustrated in Table (4) showed that the number of pods/plot and Seed weight/plot traits as affected by the variances between the14 lines from different position in Egypt (2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14 and 15) compared with the 4 local varieties (1, 16, 17 and 18) for faba bean plants during the growing seasons of 2018 and 2019. Results reveal there was significant differences between the18 genotypes of faba bean plant under study. The genotype 18 and 13 scored the highest number of pods/Plot (24.00 and 23.66) during 2018 and 2019 seasons respectively. The lowest number of pods/plots recorded by genotype 2 (4.33) in 2018 season and genotype 15 (7.00) in 2019 season as compared with other faba bean under study.

As for, seed weight/plot results showed that genotype 13 recorded the largest value (26.56 and 34.8) during 2018 and 2019 seasons respectively, compared with other genotypes. While genotype 1 and 12 recorded the lowest values (8.30 and 14.83) for the same traits at the same two seasons. Musallam et al., (2004) who mentioned significant different between environments genotypes and in generalizing morphological characteristics and genotype Х in environmental interactions to flowering days and seed vield characteristics and results are similar to those (Elshafei et al., 2019).

Table (4): Number of pods/Plot and seed weight/plot as affected by variances between faba bean lines during 2018 and 2019 seasons.

faba bean	Number of	f pods/Plot	Seed weight/plot					
genotypes	Season 2018	Season 2019	Season 2018	Season 2019				
1	10.00	16.00	8.30	26.46				
2	4.33	12.33	7.73	28.43				
3	9.00	14.33	12.20	32.96				
4	7.33	18.33	15.80	27.6				
5	8.66	9.33	12.00	18.76				
6	8.00	7.66	15.50	31.56				
7	7.00	16.00	8.63	22.46				
8	15.33	13.66	26.3	23.50				
9	10.00	10.66	16.66	25.73				
10	9.66	22.33	9.76	33.30				
11	10.00	12.66	18.83	14.96				
12	9.00	8.33	11.56	14.83				
13	17.33	23.66	26.56	34.80				
14	7.00	11.33	13.00	18.10				
15	5.66	7.00	8.93	12.76				
16	6.00	12.66	10.96	19.46				
17	7.33	14.66	18.13	17.30				
18	24.00	12.33	23.00	19.73				
L.S.D. at 5%	6.01	8.68	12.89	20.15				

L.S.D. is the level of significance utilize least significant variation at 5%

First fruiting nods/Plant and Number of seed/Plant

Results clarified in Table (5) showed that the first fruiting nods/plant and number of seed/plant traits as influenced by the variances between the14 lines (2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14 and 15) comparing with 4 local varieties (1, 16, 17 and 18) for faba bean plants during the growing seasons of 2018 and 2019. Results detected that there were significant differences between 18 genotypes of faba bean plant under study. The genotype 7, 9 and 5 scored the highest number of first fruiting nods/plant (5.33 and 4.66) during 2018 and 2019 seasons respectively. The lowest number of first fruiting nods/plant recorded by 14 (2.33) in 2018 season and genotype 1, 14 (1.66) in 2019 season as compared with other faba bean from different locations under study. As for, number of seed/plants results showed that genotype 1, 9 and 10 recorded the largest value (25.33 and 59.00) during 2018 and 2019 seasons respectively, compared with other genotypes. While genotype 2 and 12, 15 recorded the lowest values (10.00 and 16.66) for the same traits at the same two seasons. These results are like to (Elshafei *et al.*, 2019 and Rebaa *et al.*, 2017).

faba bean	First fruiting	g nods/plant	Number of seed/Plant				
genotypes	Season 2018	Season 2019	Season 2018	Season 2019			
1	3.00	1.66	25.33	41.66			
2	4.00	2.00	10.00	35.00			
3	3.66	3.66	20.66	40.00			
4	3.33	2.33	17.33	44.33			
5	3.66	4.66	18.33	18.66			
6	3.33	2.33	21.66	17.33			
7	5.33	3.00	12.00	35.66			
8	4.00	2.33	36.00	30.66			
9	5.33	3.33	25.33	28.66			
10	4.00	3.33	24.33	59.00			
11	4.33	4.00	24.66	25.66			
12	3.33	3.00	19.33	16.66			
13	3.66	2.33	48.00	58.00			
14	2.33	1.66	18.33	28.00			
15	3.66	3.00	11.33	16.66			
16	3.66	2.66	12.33	35.33			
17	4.33	3.33	22.66	24.33			
18	3.00	3.00	54.00	43.33			
L.S.D at 5%	1.78	1.40	16.02	21.68			

Table (5): First fruiting nods/plant and Number of seed/Plant as affected by variances between faba bean lines during 2018 and 2019 seasons.

L.S.D. is the level of significance utilize least significant variation at 5%

Relationship among morphological and yield related traits under study:

The value of correlation between all quantitative traits under study were estimated as shown in Table (6). The maximum value between plant height and number of pods/plots was (0.569) in season 2018. The minimum value between number of pods/plot and number of branches/plants was (-0.501) in season 2018. There was an organization between number of pods/plot and number of seed/plants the highest value in the 2018 season which recorded 0.971. The results are similar with (Asfaw *et al.*, 2018 and Elshafei *et al.*, 2019) which appear that the field evaluations of the genotypes of Egyptian beans showed enough variation for their studied agricultural parameters.

	Trait	Season	(1)	(2)	(3)	(4)	(5)	(6)
(1)	Diant height/om	2018	1 000					
(1)	r fant neight/chi	2019	1.000					
(2)	Number of	2018	-0.165	1.000				
(2)	branches/Plant	2019	-0.118	1.000				
(3) Num	Number of node/Diet	2018	0.569	-0.501	1 000			
	Number of pous/riot	2019	0.185	0.520	1.000			
(4)	Sood weight/plat (g)	2018	0.393	-0.257	0.758	1 000		
(4)	Seed weight/plot (g)	2019	0.301	0.376	0.608	1.000		
(5)	First fruit nods/Dlant	2018	-0.234	-0.134	-0.164	-0.005	1 000	
(5)	r ii st ii uit nous/r iant	2019	0.184	0.168	-0.163	-0.260	1.000	
	Number of good/Dient	2018	0.512	-0.453	0.971	0.821	-0.173	1 000
(6)	Number of seed/Plant	2019	0.301	0.522	0.911	0.674	-0.238	1.000

Table (6): Correlation matrix between traits under study during seasons 2018 and 2019.

Molecular markers analysis and variation revealed by SSR markers:

Molecular-data-based estimation of genetic variability found among genotypes of faba bean is critical to assure the morphological diversity and use in the breeding approaches as well as in the conservation of genetic resources. Microsatellite markers are effective for assessment of polymorphism and map the diversity among genotypes (Rebaa et al., 2017; Gong et al., 2011and Tahir et al., 2019). The SSR markers as the marker were used in this study to scanner the polymorphism rate and across the genome for eighteen genotypes of faba bean from different population as shown in Figure 1.

Data resulted from SSR marker (Table 7) shows that they ranged from 156 bp to 202 bp with the average of allele number

(Na) 9.2 allele/marker. The number of alleles stretched from 3 (SSR 26) to 18 (SSR 31). Heterozygosity (Ho) was ranged from 0.0 for markers SSR 25, SSR 26 and SSR 30, 0.167 for marker SSR 28 and 0.772 for marker SSR 31 with average 0.178. While mean expected heterozygosity (He) was appeared to be 0.8196. genetic identity (I) was amidst 1.026 and 2.684 for an average of 1.9402. while the average of effective alleles per locus (Ne) was 7.096.

The analysis of variance between genotype using SSR markers (Table 7) shows the existence of variance and large differences averaged over (F) 0.804. These data agreement for (Gong *et al.*, 2011 and Tahir *et al.*, 2019). SSR is widely used in beans for preference between genotypes at the molecular level.

Table (7): Sample sizes, No. alleles, No. effective alleles, Information index, Observed and Expected Heterozygosity, and Fixation Index.

Primers	Range (bp)	Na	Ne	Ι	Ho	He	F
SSR25	183-191	6	5.786	1.773	0.000	0.827	1.000
SSR26	197-201	3	2.656	1.026	0.000	0.623	1.000
SSR28	172-191	10	7.448	2.137	0.167	0.866	0.807
SSR30	166-195	9	7.364	2.081	0.000	0.864	1.000
SSR31	156-202	18	12.226	2.684	0.722	0.918	0.213
Average	156-202	9.2	7.096	1.9402	0.1778	0.8196	0.804

Number of alleles (Na), Effective alleles per locus (Ne), genetic identity (I), Heterozygosity (Ho), expected heterozygosity (He), Fixation Index (F).

Estimate of correlation matrix

According to data derived from the SSR marker correlation matrix between all genotypes was estimated as presented in Table (8). The highest values of positive similarity were recorded between genotype 1 and 4 (0.487). Whereas the lowest negative similarity was recorded between genotype 1, 3, 5, 6 and genotype 17 (-0.150).

Basing on Nei and Li coefficient the phenogram generated by the UPGMA cluster analysis showed clustering of the18 genotype of faba bean (Figure 2). Investigated were divided into four groups. The first one divided into two sub-groups; first one includes genotype 3 only in sub-sub-group and genotype 7 and genotype 9 in another sub-sub-group and the second subgroup includes genotype 8 in sub-sub-group, genotype16 in sub-subgroup and genotype 10 and genotype 12 in the same sub-sub-group. The other group is divided into two subgroups; one of them includes genotype 14 only in subsub-group and genotype 1 and genotype 4 in another sub-sub-group whereas the group is further divided in two sub group the first on include genotype 2 and genotype 15 the second include genotype 17 in sub-sub-group only and genotype 11 and genotype 13 in another sub-subgroup. The third group include genotype 6 only. The fourth group include genotype 5 and genotype 18. This results agreement with (Yahia et al., 2012).



Fig. 1. Eighteen faba bean genotypes acquired with SSR marker lanes (M), 1500 bp ladder; 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18.

	Table (8): Correlation Matrix according to SSR markers																	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1	1.000																	
2	-0.150	1.000																
3	-0.150	0.233	1.000															
4	0.487	-0.135	-0.135	1.000														
5	-0.150	-0.150	-0.150	0.280	1.000													
6	-0.150	-0.150	-0.150	0.072	0.042	1.000												
7	-0.135	0.072	0.280	-0.122	-0.135	-0.135	1.000											
8	0.072	-0.135	-0.135	-0.122	-0.135	0.072	0.102	1.000										
9	0.042	-0.150	0.233	-0.135	-0.150	-0.150	0.280	0.280	1.000									
10	0.016	0.016	0.016	-0.148	-0.164	-0.164	0.046	0.241	0.016	1.000								
11	-0.135	0.072	0.072	0.102	0.072	0.072	0.102	0.102	-0.135	0.046	1.000							
12	0.042	-0.150	-0.150	-0.135	-0.150	0.233	0.072	0.072	0.042	0.375	0.072	1.000						
13	0.233	0.042	0.233	0.072	0.042	-0.150	0.072	-0.135	-0.150	-0.164	0.280	-0.150	1.000					
14	0.425	-0.150	-0.150	0.072	-0.150	0.042	-0.135	0.072	0.042	0.016	0.072	0.233	0.233	1.000				
15	0.233	0.233	0.042	0.072	-0.150	-0.150	0.072	-0.135	-0.150	-0.164	0.072	-0.150	0.233	0.233	1.000			
16	0.042	0.042	0.042	-0.135	-0.150	-0.150	-0.135	0.072	0.233	0.195	0.072	0.233	0.042	0.233	0.042	1.000		
17	-0.150	0.233	0.042	-0.135	-0.150	-0.150	0.072	0.072	-0.150	0.195	0.280	0.042	0.042	-0.150	0.042	0.042	1.000	
18	0.042	-0.150	-0.150	-0.135	0.233	-0.150	0.072	0.072	0.233	0.016	-0.135	0.042	0.042	0.042	-0.150	0.042	-0.150	1.000



Fig. 2. clustering of 18 faba bean lines based on SSR primers.

Parameters resulted from all SSR markers and yield related traits under study:

The assessment of genetic diversity morphological characteristics and is challenging as it can be influenced by environmental factors. Hence the increased use of molecular markers to characterize the differences between and within crop groups at the DNA level (Hoxha et al., 2004). Data came from SSR and quantitative traits are shown in Table (9) was used to study the correlation among genotypes. The maximum similarity was 1.00 between genotype 1, 2, 4, 5, 7, 15 and 16 while the minimum value was 0.92 between genotype 3, 5, 10 and 12. These results indicate that the best one genotypes are 7 and 14 compared with all genotype.

The Principal Component Analysis (PCA) from all SSR markers and morphological data was performed as shown in Figure 3. The results divided the 18 genotypes of faba bean in to two major group. The first major group include two subgroups one of which contains only the genotype 10 only. On the other hand, the other subgroup divided into two subsubgroup, the first include genotypes 6 and genotype 12, the two genotype from Faiyum Govenorate, the second sub-subgroup divided into two sub-subgroup one of them genotype 13 only and the other include genotypes 8 and 18. The second major group divided into two groups one of them include genotype 3 only (from Aswan Governorate), the other group divided into two subgroups, the first of them is divided into two sub-subgroup genotypes 2 (from Aswan Governorate) only in one group as the other is divided into two sub-subsubgroup with genotypes 1 and genotype 14, the other is divided into two sub-subsub-subgroup with genotype 16 in one and genotypes 4 and genotype 7 in the other sub-sub-sub-subgroup. The second is divided into two sub-subgroup genotypes with genotypes 5 and genotype 15 in the one sub-subgroup while the other subsubgroup is divided into two sub-subsubgroups with genotypes 9 only in one and genotypes 11 and genotype 9 in the same sub-sub-subgroup.

EL-Shaer, H.F.A. and Helal, A.A.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1	1.00																	
2	0.99	1.00																
3	0.98	0.99	1.00															
4	1.00	0.99	0.97	1.00														
5	0.98	0.98	0.97	0.98	1.00													
6	0.98	0.97	0.94	0.98	0.98	1.00												
7	1.00	1.00	0.98	1.00	0.99	0.98	1.00											
8	0.98	0.96	0.94	0.98	0.97	0.99	0.98	1.00										
9	0.98	0.99	0.99	0.98	0.99	0.97	0.98	0.97	1.00									
10	0.96	0.96	0.94	0.97	0.92	0.93	0.96	0.94	0.93	1.00								
11	0.99	0.98	0.97	0.99	0.99	0.98	0.99	0.99	0.99	0.95	1.00							
12	0.98	0.96	0.92	0.98	0.97	0.99	0.97	0.99	0.95	0.94	0.98	1.00						
13	0.98	0.96	0.96	0.97	0.94	0.94	0.96	0.97	0.96	0.96	0.98	0.95	1.00					
14	1.00	0.99	0.98	1.00	0.99	0.98	1.00	0.99	0.99	0.96	1.00	0.98	0.97	1.00				
15	0.98	0.98	0.96	0.98	1.00	0.97	0.99	0.97	0.99	0.92	0.98	0.96	0.93	0.99	1.00			
16	0.99	0.99	0.96	1.00	0.98	0.98	1.00	0.98	0.97	0.95	0.98	0.98	0.95	0.99	0.98	1.00		
17	0.99	0.98	0.97	0.99	0.99	0.98	0.99	0.99	0.99	0.94	1.00	0.98	0.97	1.00	0.99	0.99	1.00	
18	0.98	0.95	0.94	0.97	0.96	0.97	0.96	0.99	0.96	0.94	0.98	0.99	0.98	0.98	0.95	0.97	0.98	1.00

Table (9): Correlation matrix generated from all data SSR markers and yield related traits under study.

140



Fig. 3. Dendrogram resulted from all data SSR markers and yield related traits under study.

Conclusion

There was significant diversity and variation among the examined faba bean genotypes for morphological and yield linked traits. Great genetic variability in the faba bean genotypes was characterized by higher level of polymorphism with SSR markers. Based on the field performance and genetic diversity information. Nine genotypes had the highest distinction that can be selected to use in breeding and hybridization strategies.

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