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VARIABILITY ASSESSMENT OF SOME MAIZE (*Zea mays* L.) ELITE INBRED LINES USING MORPHOLOGICAL AND MOLECULAR METHODS

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ABSTRACT

Seed samples from the breeder's seed stocks of 11 inbred lines representing the parental inbred lines involved in the majority of the recently registered single and three-way maize crosses in Egypt were planted in South Nile Delta during 2002 season. All these inbred lines were developed and maintained by the National maize programme in the Agricultural Research Center- ARC. Since variety identification is carried out depending on the observation and recording of a range of morphological characters which can be an expensive and time consuming process. Identification and verification of maize inbred lines and other genotypes require utilizing more than one morphological attribute. Combining other biochemical and molecular fingerprinting techniques as a requirement for variety registration and national listing as well as for breeders right protection purposes and for maintaining genetic purity in the seed quality control. In this research morphological description was carried out in addition to application of the random amplified polymorphic DNA-RAPDs and simple sequence repeats or Micro-satellites- SSR as two DNA profiling techniques to investigate the variability of the 11 tested inbred lines.

Key words: ***Maize, Inbred lines, Identification, Morphology, Molecular, RAPDs, SSR, Fingerprinting.***

INTRODUCTION

Maize is the world's third leading cereal crop, after wheat and rice. It is primarily used as a major food, feed grain for livestock and for industrial products (Poehlman and Sleper 1995). In Egypt, maize comes second after wheat as per area (National Maize Research Programme,2002). There are a number of inbred lines, representing the majority of the parental material involved in the major white single and three-way crosses which cover most of the maize cultivated area. The internationally recognized descriptor of UPOV, 1994 was followed to differentiate between the tested inbred lines according to their morphological characteristics in the different growth stages as stated by Tottman (1987). However, some specific

morphological attributes may be masked due to environmental and stress conditions. This makes the development of new techniques for genetic purity determination and identification even more essential. Two DNA profiling techniques were recently used to support the morphological identification, viz. RAPDs and SSR. The random amplified polymorphic DNA (RAPDs) technique uses arbitrarily chosen oligonucleotide primers that hybridize to the genomic DNA template at two different sites, one on each stand of the complementary DNA. Under appreciate temperature alternations, a thermo-stable DNA polymerase is able to synthesize discrete DNA products that can be resolved on an agarose gel following electrophoresis. Each primer has the capability to direct consistent amplification of several unique DNA fragments in the genome. Some amplified fragments or patterns of fragments may be unique to a genotype and hence could help as a tool of the identification of genotypes.

The PCR- based analysis of simple sequence repeats (SSRs or micro-satellites) may also prove to be very useful for the identification and verification purposes. Micro-satellites are randomly repeated DNA sequences, usually with a repeat of 2-4 base pairs. In many species, multiple alleles have been shown to exist for some micro-satellites, due to variation in the copy number of this repeat unit. Micro-satellites can be analyzed by PCR using specific primers; the alleles (PCR products) can then be separated by agrose or polyacrylamide gel electrophoresis (Cooke, 1999). There are now micro-satellites available in several crop species including maize. Amplification technologies based on the polymerase chain reaction {PCR) allow the production of millions of copies of particular pieces of DNA and thus facilitating their analysis through RAPDs or SSR techniques (DeLoose and Gheysen 1995).

The objective of this work is to investigate the variability among eleven inbred lines of maize involved in the majority of white single and 3-way crosses commercially used in Egypt using morphological and molecular methods of assay.

MATERIALS AND METHODS

Seed samples from the breeder's seed stocks from: 11parental inbred lines of maize (Gmemmeiza 2,4,14,18,21,22,27and 30 and Sids 7,34 and 63) were planted in an experimental field in Toukh, Qualubia Governorate. Each variety was planted in six rows each of 5m long and 70 cm width on mid-May,2002. Readings and observations were collected on weekly intervals regularly starting from germination till the full maturity stage. The

agricultural practices were applied as recommended by the Ministry of Agriculture without using any pesticides or herbicides.

Morphological Identification: The morphological identification was conducted using the UPOV (The International Union for the Protection of New Varieties of Plants) descriptors. The Decimal code for the Growth Stages of Cereals, according to Tottman (1987) was also used to standardize the growth stages of varieties during morphological description and identification.

Molecular Identification: The RAPD (Random Amplified Polymorphic DNA) and SSR (Simple Sequence Repeats, or Microsatellites as DNA profiling techniques were used to identify these inbred lines.

DNA Isolation and Sampling: Genomic DNA was isolated individually from a sample extracted from one-week old seedlings from each of the investigated inbreds. Approximately 100 mg leaf material was used to extract DNA using QIAGEN Kit. The DNA was isolated from each genotype to prepare the fingerprint which is considered specific for each genotype. Careful attention was paid to the DNA purity and concentration. Therefore, the DNA purity was first estimated using spectrophotometer A_{260}/A_{280} readings.

Polymerase Chain Reaction (PCR)

RAPD's Technique:PCR was carried out in 0.5 ml thin walled tubes in a final reaction volume of 30µl.

The reaction mix consisted of a final concentration of 1.5 mM of $MgCl_2$, 200 µM of dNTPs, 50 ng primer and 0.2 µl Taq DNA polymerase (promega) and completed by adding distilled water up to 30 µl . Five primers with arbitrary sequences were used for screening.DNA amplifications were carried out with a thermocycler (PTC-200). Forty amplication cycles were employed with each cycle consisting of preheating at 94 °C for 4 minutes and then denaturation at 94 °C for 30 seconds, annealing at 35 °C for one minute, and extension at 72 °C for 2 min, followed by a one final extension cycle for 72 °C at 10 minutes.

SSR Technique: Polymerase chain reaction (PCR) for SSR technique was performed in 0.5ml thin walled tubes in a final reaction volume of 30µl. The reaction mix containing 50ng of maize DNA, 50 ng of each primer,1.5mM of MgCl2,0.2mM of DNTps,3 µl of 10xPCR buffer, and 0.2µl Taq polymerase (RTS Taq DNA polymerase ,GIBCO-BRL). Forty amplication

cycles were employed with each cycle consisting of preheating at 94 °C for 4 minutes and then denaturation at 94 °C for 30 seconds, annealing at 60°c for Maize for one minute, and extension at 72 °C for 1.5 min, followed by a one final extension cycle for 72 °C at 10 minutes. Amplification products were separated in a 0.4 mm thick 6% polyacrylamide gel together with DNA size standard. PCR products were visualized by ethidium bromide.

RESULTS AND DISCUSSION

Variety identification and description are the key issue for variety registration and granting the breeder's rights (plant variety protection). The plant variety that should prove that it can be distinguished from other varieties which are in the public knowledge in one or more morphological, physiological, cytological, chemical or any other characteristic. Observations regarding the morphological description collected and recorded from planting till harvesting using UPOV guidelines and descriptors, are presented in Table1.

The morphological differences between the tested 11 inbred lines are very clear and can be summarized in the following points:

The inbred lines Gem.4 and 21, Sids 7 have strong anthocyanin coloration on the sheath of the first leaf at seedling stage (stage 12).

The leaves of Gem 2 appear leathery and thick in the same time while the leaves of Sids 63 have curly appearance.

The brace roots anthocyanin coloration can also be used to differentiate the inbred lines Sids 7 and Gem 22 which have strong coloration. Gems 4 and 21 have moderate degree and the other inbred lines have weak coloration.

All the inbred lines have early anthesis, except Gem 27 and Sids 7 which have early to medium, and Gem 30 which has the latest anthesis timing.

The anthocyanin coloration of glumes at the base of tassel and other glumes exist strongly in Gem 4, 18, Sids 7 and Sids 63 and more strongly in Gem 21. The other inbred lines show weak to medium coloration.

The lateral branches of tassel of Sids 34 are strongly recurved, and moderately recurved in Gem 18, 22, Sids 7 and 63. The other inbred lines have straight to slightly recurved branches.

Table 1.Morphological characteristics of maize inbred lines (according to UPOV Descriptor No. TG/2/6-1994)

No.	Characteristic and stage	Degree	Score	Inbred lines										
				Gm 2	Gm 4	Gm 14	Gm 8	Gm 21	Gm 22	Gm 27	Gm 30	S. 7	S. 34	S. 63
1	First leaf: anthocyanin coloration of sheath Stage 12(s)	Abs or v.w Weak Medium Strong Very strong	1 3 5 7 9	3	7	3	3	7	3	5	3	7	5	3
2	First leaf:shape of tip Stage 1	Pointed Pointed round Round Round spatulate Spatulate	1 2 3 4 5	1	1	1	1	1	1	1	1	1	1	1
3	Leaf:angle between blade and stem(on leaf just above upper ear) Stage 61	Very small Small Medium Large Very large	1 3 5 7 9	1	3	3	5	5	3	3	3	3	5	3
4	Leafe:attitude of blade Stage 61	Straight S. recurved Recurved Strongly recuv. Very S. recuv.	1 3 5 7 9	3	5	3	5	5	3	5	3	3	5	5
5	Stem:degree of zig-zag Stage 65	Abs. or v.slight Slight strong	1 2 3	1	1	1	1	1	1	1	3	1	1	1
6	Stem:anthocyanin coloration of brace roots Stage 65-75(S)	Abs. or v.weak Weak Medium Strong Very strong	1 3 5 7 9	1	5	1	1	5	7	1	1	7	1	1

Table 1. Cont.

No.	Characteristic and stage	Degree	Score	Inbred lines										
				Gm 2	Gm 4	Gm 14	Gm 8	Gm 21	Gm 22	Gm 27	Gm 30	S. 7	S. 34	S. 63
7	Tassel:Time of anthesis(on middle third of main axis) of 50%of plants Stage 65	Very early v.early to early early car.to medium medium medl.to late late late to v.late very late	1 3 5 7 9	3	3	3	3	3	3	4	7	4	3	3
8	Tassel:anthocyanin coloration at base of glume (in middle third main axis) Stage 65(S)	Abs.or V.weak Weak Medium Strong Very strong	1 2 3 4 5	3	7	3	7	9	1	1	1	7	3	7
9	Tassel:anthocyanin coloration of glumes excluding base(asfor 8) Stage 65(S)	Abs.or V.weak Weak Medium Strong Very strong	1 3 5 7 9	5	7	3	5	7	3	1	1	7	3	7
10	Tassel:anthocyanin coloration of anthers(as for8;on fresh anthers) Stage 65(S)	Abs.or V.weak Weak Medium Strong Very strong	1 3 5 7 9	1	1	1	1	1	1	1	1	1	1	1
11	Tassel: density of spikelets(asfor8) Stage 65	Lax Medium dense	1 2 3	5	5	5	3	5	5	5	5	5	5	5
12	Tassel:angle between main axis and lateral branches(in lower third of tassel) Stage 65	Very small Small Medium Large Very large	1 3 5 7 9	5	5	3	3	3	3	3	5	3	5	3

Table 1. Cont.

No.	Characteristic and stage	Degree	Score	Inbred lines										
				Gm 2	Gm 4	Gm 14	Gm 8	Gm 21	Gm 22	Gm 27	Gm 30	S. 7	S. 34	S. 63
13	Tassel:attitude of lateral branches (as for 12) Stage 65(S)	Strainht Slightly recuv. Reacuverd Strongly recuv. v. Stro. Recuv.	1 3 5 7 9	3	3	1	5	3	5	5	3	5	7	5
14	Tassel:number of primary lateral branches Stage 65	Absent or v.few Few Medium Many Very many	1 2 3 4 5	7	3	5	5	3	5	5	7	5	7	5
15	Ear:time of silk emergency(50%of plants) Stage 65	Very early v.early to early early ear.to medium medium medi.to late late late to v.l ate very late	1 3 5 7 9	3	3	5	3	3	6	6	6	5	3	3
16	Ear: anthocyanin coloration of silks Stage 65(S)	Absent Present	1 3 5 7 9	9	9	9	9	9	9	1	1	1	9	9
17	Ear :intensity of anthocyamin coloration of silks Stage 65(S)	Very weak Weak Medium Strong Very strong	1 2 3	3	1	1	3	3	5	1	1	1	1	3
18	Leaf:anthocyanin coloration of sheath(in the middle of plant) Stage 71(S)	Abse. or v.weak Weak Medium Strong Very strong	1 3 5 7 9	3	5	3	3	7	3	5	3	5	3	3

Table 1. Cont.

No.	Characteristic and stage	Degree	Score	Inbred lines										
				Gm 2	Gm 4	Gm 14	Gm 8	Gm 21	Gm 22	Gm 27	Gm 30	S. 7	S. 34	S. 63
19	Tassel:length of main axis above lowest side branch Stage 71	Very short Short Medium Long Very long	1 3 5 7 9	3	3	3	5	5	3	7	5	5	7	5
20	Tassel: length of main axis above upper side branch Stage 71	Very short Short Medium Long Very long	1 2 3 4 5	3	3	1	1	3	3	7	5	3	5	5
21	Tassel:length of side branches (as for16) Stage 71	Very short Short Medium Long Very long	1 3 5 7 9	3	3	3	5	3	3	7	5	5	7	5
22	Plant Height: (Tassel included) Stage 75	Very short Short Medium Long Very long	1 3 5 7 9	3	7	5	5	7	3	7	7	7	7	3
23	Plant: ratio height of insertion of upper ear to plant height Stage 75	Very small Small Medium Large Very large	1 2 3	3	5	7	5	5	7	7	5	5	5	5
24	Leaf:width of blade (leaf of upper ear) Stage 75	Very narrow Narrow Medium Wide Very wide	1 3 5 7 9	3	7	3	3	7	5	5	5	3	7	7

Table 1. Cont.

No.	Characteristic and stage	Degree	Score	Inbred lines										
				Gm 2	Gm 4	Gm 14	Gm 8	Gm 21	Gm 22	Gm 27	Gm 30	S. 7	S. 34	S. 63
25	Ear:length of peduncle Stage 85	Very short Short Medium Long Very long	1 3 5 7 9	3	7	3	5	5	3	7	5	3	7	5
26	Ear :length(without husk) Stage 92	Very short Short Medium Long Very long	1 2 3 4 5	3	5	3	7	5	5	5	3	5	9	3
27	Ear: diameter in middle Stage 92	Very small Small Medium Large Very large	1 3 5 7 9	7	7	7	7	7	5	3	3	3	7	7
28	Ear shape Stage 92	Conical Con .cylindrical Cylindrical	1 3 5 7 9	1	3	2	3	3	3	3	3	3	3	2
29	Ear :number of rows of grains Stage 92	Very few Few Medium Many Very many	1 2 3	7	5	5	5	5	7	3	5	3	7	7
30	Ear: type of grain (middle third of ear) Stage 92(S)	Flint Flint like Intermediate Den tike Dent Sweet Pop	1 3 5 7 9	1	4	1	1	1	2	1	2	2	2	2

Table 1. Cont.

No.	Characteristic and stage	Degree	Score	Inbred lines										
				Gm 2	Gm 4	Gm 14	Gm 8	Gm 21	Gm 22	Gm 27	Gm 30	S. 7	S. 34	S. 63
31	Ear: color of top of grain Stage 92 (S)	White	1											
		Yellowish white	3											
		Yellow	5											
		Yellow orange	7	1	1	1	1	1	1	1	1	1	1	1
		Orange	9											
		Red orange												
		Red												
		Dark red												
		Blue black												
32	Ear: color of dorsal side of grain Stage 92(S)	White	1											
		Yellowish white	2											
		Yellow	3											
		Yellow orange	4	1	1	1	1	1	1	1	1	1	1	1
		Orange	5											
		Red orange												
		Red												
		Dark red												
		Blue black												
33	Ear:Anthocyanin coloration of glumes of cob Stage 93(S)	Absent	1	1	1	1	1	1	1	1	1	1	1	1
		present	3											
			5											
			7											
			9											
34	Ear intensity of anthocyanin coloration of glumes of cob Stage 93(S)	Very weak	1											
		Weak	3	1	1	1	1	1	1	1	1	1	1	1
		Medium	5											
		Strong	7											
		Very strong	9											

The lateral branches of tassel are many in Sids 34, Gem 30 and 2; the other lines have few to medium number of lateral branches.

Most inbred lines have anthocyanin coloration on the ear silk except Gem 27, 30 and Sids 7.

The length of the main axis and side branches in Gem 27 is very clear than the others.

The shape of the main axis of the tassel is also used to identify the tested lines. The tip of the main axis of Gem 21 is binded, while the line Gem 14 has binding at the base of the main axis.

The variation in plant height is very clear among the tested lines; Gem 2 is the shortest and Gem 30 is the longest. Also, the ratio of height of the upper ear to plant hieght is varying from 0.35 in case of Gem 2 to 0.51 in Gem 27.

While the length of ear peduncle is long in Gem 4 and 27 and Sids 34, it is short in Gem 2, 14 and in Sids 7. Also the ear length is varying from very long in Sids 34 to short in Gem 2, 14 and 30 and Sids 63. There are two rudimentary small ears surrounding the main ear in Gem 30. Two molecular techniques, random amplified polymorphic DNA (RAPD's), simple sequence repeats or Microsatellites (SSR) were used to identify the tested genotypes.

The RAPD (Random Amplified Polymorphic DNA). One of the DNA profiling techniques was used to identify these inbred lines. The bands appear in figures 1 and 2 show different numbers for the investigated inbred lines and indicate differences in the molecular weights. It is concluded that identification of the maize gentypes require combining more than one morphological attribute in addition to using biochemical and molecular fingerprinting methods to give more clear-cut identification. The morphological identification according to the international schemes such as those recommended for maize can provide an efficient tool for distinguishing genotypes from each other.

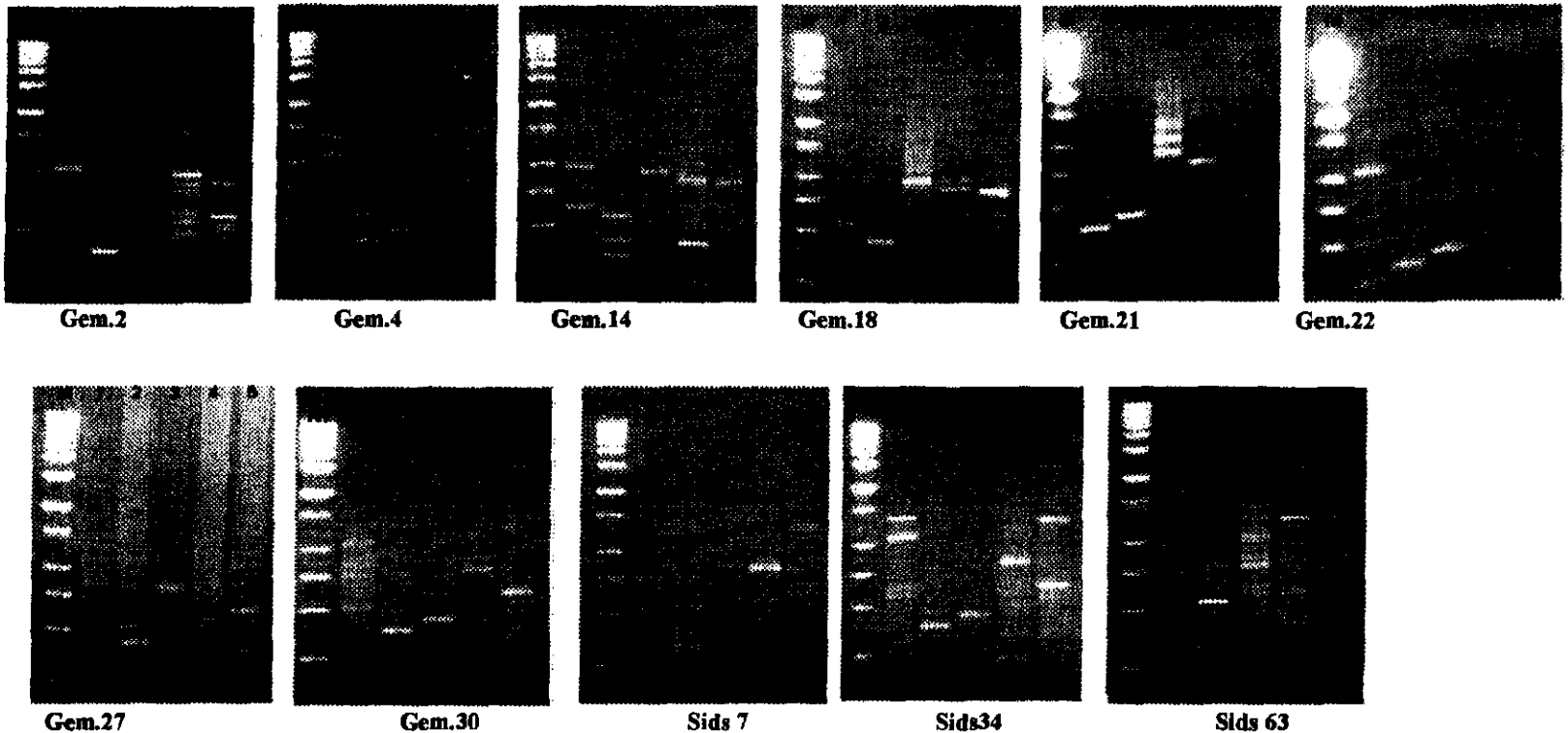


Figure 1: Gels of 11 maize inbred lines with polymorphic bands scored as potentially diagnostic using 5 primers (RAPD's Method).

Table 2: Alignment of sequences and No. of bands of 11 maize inbred lines

Line	Primers				
	1	2	3	4	5
Gem.2	3142.9	2228.8	2695.5	3283.9	3611.3
	2816.4	1639.5		2900	2900
	2041.6	1223.7		2637	2656.4
	1738.2	836.6		2415.5	2363.1
	1580.6			1911.6	2133.2
	1437.3			1675.8	1816.2
				1479.9	1615.7
				1395.8	1375.6
				1197.1	1065
				1088.6	867.8
				824.5	
Gem. 4	4745	3094	1633	4919	3315
	4229	2170		3672	
	1860	1402		3299	
	1159	1159		2531	
				2270	
				1916	
				648	
Gem.14	3005.2	2880.8	4125.8	2820.6	
	2332.1	2471.6	2880.8	2511.1	
	1928.3	2165.9	2022.1	2307.6	
	1663.2	1744.1	1645.7	1887.9	
	1480.7	1585.9	3673.2	1672	
	1277.1	1411.9	3218.8	1520.3	
	920.4	1325.2	2989.3		
		1217.8	2805.7		
		1137	2647.3		
		1017.6	2484.7		
			1897.9		
			1689.7		
			1577.6		
			1283.7		
			1050.4		
			828.2		
Gem. 18	1038.2	462.4	2428.8	3329	2330.9
	616.7	233	1821.2	2863	1747.9
	361.3		1442.7	2260.6	1275.2
	179.6			1897.7	1067.1
				1223.8	822.4
				856.6	678.8
				747.1	
				414.4	
				332.8	
				211.7	

Line	Primers				
	1	2	3	4	5
Gem.21	2347.9	1684.6	5086.1	4574.1	4051.6
	1747.1	1669.4	4404.1	3727.7	3671.7
	1270.8	1413	3930.8	2902.9	2513.6
	1092.1		3277.1	2571.4	1899
			2859.2	2002.4	1682.1
				850.4	
Gem.22	1488.5	944.6	440.9	1108.3	1222.8
	738.7	584.9			766.5
	510.9	371.2			629.7
	430.1				
	259.9				
Gem.27	3172.2	4365.8	2157.9	3412.4	3025.8
	2348.1	4106.4	1012	2408.4	1728.4
	1140.1	2139.8	759.4	2000	945.2
	689.2	1536	348.6	1878.7	
		1074.1		1570.4	
		592		1048.8	
				655.1	
Gem. 30	2462.8	1548.6	764.3	2361.1	3073
	2102.2	1015.9		1932.6	2336.4
	1932.6	652.6		1632.6	1649.9
	1739.2	234.7		687.9	1165.2
	1484.9			593.5	984.4
	1093.8			406.1	702.5
	904.6				581.1
	780.6				496.1
	659.5				
	485.8				
	401.8				

Table 2 - Continued

Line	primers				
	1	2	3	4	5
Sids 7	2056	5415	3769	3131	3769
	864	3682	2530	2428	3033
		2056		1801	2428
		1938		844	1844
		1671		481	
		1460			
		1025			
		851			
		693			
Sids 34	4658	5104	879	4956	4623
	3776	1967		4258	3486
	1296	1755		3583	1950
	1197	1443		3000	1361
	1073	1184		1967	1184
	947	932		1418	720
	500	697		1237	513
	349	409		1073	398
		239		574	
				239	
Sids 63	4341	4353	7040	7217	6102
	3944	4116	6458	6814	4710
	3024	3627	5262	5696	4161
	2413	3124	4675	4908	4013
	2152	2837	4450	4687	3737
	1879	2631	4205	4450	3558
	1500	2239	4052	4192	2957
		1912	3833	3917	2533
		1656	3640	3695	1739
		1519	3477	3384	
		1453	3265	3111	
		1231	2674	2815	
		1027	2533	2631	
			2381	2468	
			2207	2250	
			2033	2076	
			1771	1838	
			1500	1656	
				1422	

Gem. 2
P1 GTAGACCCGT
P2 CCTTGACGCA
P3 TTTGCCCGGA
P4 AGGGAACGAG
P5 CCACAGCAGT

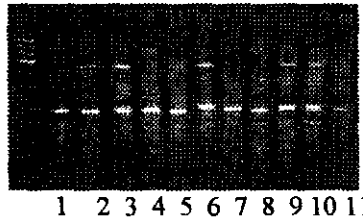
Gem. 27
P1 GTAGACCCGT
P2 TCCGCTCTGG
P3 AGGGAACGAG
P4 CCACAGCAGT
P5 GGACCCTTAC

Gem. 4
P1 GTAGACCCGT
P2 CCTTGACGCA
P3 TCCGCTCTGG
P4 AGGGAACGAG
P5 CCACAGCAGT

Gem 30
P1 GTAGACCCGT
P2 CCTTGACGCA
P3 TCCGCTCTGG
P4 AGGGAACGAG
P5 CCACAGCAGT

Gem.14	P1 GTAGACCCGT	**Sids 7**	P1 GTAGACCCGT
	P2 CCTTGACGCA		P2 CCTTGACGCA
	P3 TTTGCCCGGA		P3 TTTGCCCGGA
	P4 AGGGAACGAG		P4 AGGGAACGAG
	P5 CCACAGCAGT		P5 CCACAGCAGT
Gem. 18	P1 CCTTGACGCA	**Sids 34**	P1 GTAGACCCGT
	P2 TCCGCTCTGG		P2 CCTTGACGCA
	P3 TTTGCCCGGA		P3 TCCGCTCTGG
	P4 AGGGAACGAG		P4 AGGGAACGAG
	P5 CCACAGCAGT		P5 CCACAGCAGT
Gem. 21	P1 CCTTGACGCA	**Sids 63**	P1 GTAGACCCGT
	P2 TCCGCTCTGG		P2 CCTTGACGCA
	P3 TTTGCCCGGA		P3 AGGGAACGAG
	P4 AGGGAACGAG		P4 CCACAGCAGT
	P5 CCACAGCAGT		P5 GGTCCCTTAC
Gem 22	P1 GTAGACCCGT		
	P2 CCTTGACGCA		
	P3 TCCGCTCTGG		
	P4 AGGGAACGAG		
	P5 CCACAGCAGT		

Figure 2. Gels of 11 maize inbred lines with polymorphic bands scored as potentially diagnostic using SSR (Microsatellite Technique).



1 2 3 4 5 6 7 8 9 10 11

1=Gem 2 , 2= Gem 4, 3=Gem 14, 4=Gem18, 4=Gem21, 6=Gem22, 7=Gem17, 8=Gem30, 9= Sids 7, 10=Sids 34, 11=Sids63.

Table 3. Alignment of sequences and no .of bands of 9 rice varieties - ssr.

P	N	Inbred lines										
		1	2	3	4	5	6	7	8	9	10	11
	1	184.56	479.46	47.46	650.43	479.46	492.34	512.32	189.52	498.91	492.34	479.46
	2	134.25	187.02	187.02	485.86	182.12	199.84	192.04	141.56	199.84	194.61	189.52
	3	108.59	134.25	132.48	189.52	134.25	170.44	168.2	108.59	157.02	179.73	134.25
	4	75.91	110.04	102.98	163.79	107.16	136.04	136.04	76.92	143.45	139.7	105.75
	5		72.95	72.95	130.74	74.91	107.06	105.75		110.04	111.51	76.92
	6				105.75		78.99	75.91		78.99	81.11	
	7				72.95							

Phi 049 GTNTGGCCATACCGTACTGCTTCT

PHI 050 TCCAGTTCTTCCGAAACGAAAGGG

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تقدير التباين فى بعض السلالات النقية المستخدمة كأباء فى الذرة الشامية باستخدام الطرق المورفولوجية والجزيئية

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زرعت عينات من تقاوى المربى من ١١ سلالة مرباه داخليا تمثل اباء الهجن الفردية والثلاثية من الذرة الشامية والتى تم تسجيلها مؤخرا فى مصر فى حقل تجريبى فى جنوب الدلتا لموسم ٢٠٠٢. وقد تم استنباط هذه السلالات ويتم المحافظة على نقاوتها الوراثية من خلال برنامج الذرة الشامية فى مركز البحوث الزراعية . وحيث أن تعريف الاصناف النباتية الذي يتم اعتمادا على

الملاحظة وتسجيل سلوك مجموعة من الصفات المورفولوجية التى تأخذ الكثير من الوقت والجهد. كما ان تعـريف سـلالات الـذرة الشـامية وغيرها من التراكيب الوراثية يستلزم استغلال اكثر من صفة مورفولوجـية . الا أن الصـفات البيوكيمائية والجزيئية وغير ذلك من طرق البصمة الوراثية تستخدم بغرض تسجيل هذه التراكيب وحماية حق المربي بالاضافة للحفاظ على النقاوة الوراثية ولغرض مراقبة جودة التقاوى . وقد تم فى هذا البحث بالاضافة الى التوصيف المورفولوجى استخدام طريقتين من طرق البصـمة الوراثـية هما طريقة التكبير العشوائى لقطع من الحامض النووى RAPDs وطريقة تكرارات التـتابعات البسيطة SSR لتوصيف التباينات والفروق بين هذه الاحدى عشر سلالة من الذرة الشامية وذلـك بتطبـيق طريقة متكاملة تجمع بين استخدام نماذج التوصيف المورفولوجي وغيرها من الطرق الكيميائية والجزيئية حتي يتم تمييز كل تركيب وراثي تمييزا قاطعا .