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GENETIC DIVERSITY ANALYSIS OF SOME EGYPTIAN RICE VARIETIES DIFFERING IN STORAGE ABILITY BASED ON SSR MARKERS

W.H. Elgamal[1], M.M. Kamara[2] and R.M. Soliman[2]

1. Rice Res. & Training Center, Field Crops Res. Institute, ARC.

2. Agron. Dept., Faculty of Agri., Kafrelsheikh University, Egypt.

ABSTRACT

This investigation was conducted at Sakha Agricultural Research Station during the period from 1st November 2014 to 31st October 2016 to evaluate some Egyptian rice cultivars belonging to different rice types under different storage conditions (room temperature, cooling and freezing) for 24 months with different temperature degrees. Eleven Egyptian rice cultivars with 18 SSR markers related to storage ability were used in order to determine the genetic diversity and polymorphism among the studied genotypes. Six SSR markers, namely RM1, RM5, RM71, RM251, RM161 and RM234 showed high polymorphism among the studied rice cultivars, and more-over differentiate them to different groups based on their storage ability. A total of 21 polymorphic alleles were detected and ranged between two by RM234 and five by RM71 markers. Based on the data obtained from the SSR markers, the results showed that the phylogenetic analysis divided the studied accessions into two main groups (A and B) in addition to some sub groups belong to each main group. Group A included 7 rice cultivars related to japonica rice type. On the other hand, group B included 4 cultivars divided into two sub groups (indica and indica/japonica types). The japonica cultivars were affected by storage duration more than the others, particularly for germination % and germination index under two years storage conditions. High temperature increased the negative storage effects. Indica and indica / japonica cultivars were found to be tolerant to storage more than japonica cultivars and keep more seed viability with storage time. According to the seed germination and viability after 24 months of storage, 5 Cº was considered the most appropriate storage condition for short-term storage of rice seeds.

Key words: ***Rice, Genetic diversity, Storage ability, SSR***

INTRODUCTION

Rice is life for more than half of humanity (Gnanamanickam 2009). Although rice has the second place between cereals because of the planted area, it serves as the most important food source for Asian countries mainly in south-east parts and some parts of Africa and feeding over half of the global population. (Zhou *et al* 2009). Since rice provides 21% of energy and 15% of protein for human, its quantity and quality require major attention (Gnanamanickam 2009). Rice productivity in Egypt has remarkably increased year after year according to the percentage replacement of rice area with the modern varieties to realize a maximum yield average of 10 tons /ha in season 2014 against 5.7 tons/ha for the period 1986–1998, which is considered one of the highest averages worldwide FAO STAT (2016). In Egypt, rice is rated the second among the important field crops following wheat and consider the first between the summer crops. This importance comes, for instance, because of its low cultivation costs in comparison with Synchronized crops (Elmoghazy and Elshenawy 2018). Egypt has selfsufficiency of rice and exports the rice surplus. After cultivated area limited and prices changed the storage became a very important facility for food security. Storage of rice grains is a crucial stage in the processing of paddy rice for consumption. Stored rice is preferred in some markets and whereas, in the other markets, fresh rice is preferable. Typically, rice during storage undergoes numerous changes in its physical properties and chemical composition, and these changes cause an impact on rice cooking and eating quality (Singh *et al* 2004 and Patindol *et al* 2005). The duration of the storage period depends on the objectives of the producers and marketers. Storage is also a necessity in the seed production system, rice research and genetic resources maintenance. Some properties of the rice grain are related to the duration and conditions of storage. Hull (1973) reports that under normal storage conditions grains exhibit continuous physic-chemical changes due to the physiological activities of the germ and endosperm, and these affect culinary properties and nutritive value. Simple sequence repeats (SSR) markers are the important tool for genetic variation identification of germplasm (Powell *et al* 1996 and Ma *et al* 2011). Where it has some advantages such a quickness, simplicity, rich polymorphism and stability, to present a wide range of genetic diversity (Zhou *et al* 2003, Zhang *et al* 2007, Jin *et al* 2010 and Ma *et al* 2011). Recently there is an abundance of QTLs related to rice seed storage ability to help rice breeders and whose interested in rice genetic resources. Growth and development of rice resources are mostly depending on genetic diversity among different genotypes. The objective of this work was to determine the effect of storage on rice seeds viability, grains quality and interactive effect of the two treatments (duration and temperature) on the grain quality traits of some Egyptian cultivars in addition to detecting the genetic diversity among studied genotypes using SSR markers.

MATERIALS AND METHODS

The experiment was conducted under laboratory conditions at Rice Research & Training Center (RRTC), Sakha, Egypt from 1st November 2014 to 31st October 2016 to study the effect of storage temperature, storage period and the interactions on rice storage efficiency. Also, to determine the genetic diversity among eleven rice genotypes (Table 1) using SSR markers related to storage ability based on previous studies on other genotypes. 1000-grain weight (g), germination percentage (%), germination index, hulling percentage (%), milling percentage (%) and head rice percentages (%) related storage efficiency measurements were used to evaluate the studied rice varieties.

Table 1. List of Egyptian rice genotypes, pedigree and type of studied cultivars.

No	Genotypes	Pedigree	Type
1	Giza 177	Giza 171 / Yomji No.1 // Pi No.4	Japonica
2	Giza 178	Giza 175 / Milyang 49	Indica/japonica
3	Giza 179	IR 6296-12-1-2-1-1 / GZ 1368-5-5-4	Indica/japonica
4	Giza 181	IR 28 / IR 22	Indica
5	Sakha 101	Giza 176 / Milyang 79	Japonica
6	Sakha 102	GZ 4096-7-1 / Giza 177	Japonica
7	Sakha 103	Giza177 / Suweon 349	Japonica
8	Sakha 104	GZ 4096-8-1 / GZ 4100-9-1	Japonica
9	Sakha 105	GZ 5581-46-3 / GZ 4316-7-1-1	Japonica
10	Sakha 106	Giza 177 / Hexi 30	Japonica
11	Egyptian Yasmin	IR 262-43-8-1 / NAHNG SARN	Indica

Experimental Design

Each storage temperature was carried out in a separate experiment in three replicates. Every experiment of storage temperature was included eleven rice varieties under study in split-plot design; the main plot was the storage period, while the genotypes were in sub-plots. About 6 kg of paddy rice with 14 % moisture content were stored with different storage temperature degrees. The storage treatments were diverse storage periods (6, 12, 18 and 24 months from the beginning of storage) and three different degrees of storage temperature (room temperature, cooling in refrigerator on 5C^{o} ±1 and freezing on - 5C^{o} ±1). At the end of 2014 growing season, the paddy rice of eleven cultivars was harvested and dried up to 14% moisture content. All studied traits were measured as control (no storage). During 24 months the studied characters were done every six months for the three treatments of temperature degrees. All treated seeds were applied for dormancy breaking by oven-drying seeds at 50C^{o} for 48 hours according to Agrawal (1981).

Statistical Analysis

The obtained data were statistically analyzed for analysis of variance according to Gomez and Gomez (1984) by using computer statistical software of MSTAT-C. Cluster analysis, similarity and distance index were analyzed by Paleontological statistics PAST software, version 2.17 (Saitou and Nei1987). The treatment means were compared using the least significant difference (LSD) at 5%.

SSR Analysis

The fresh leaves of eleven genotypes were sampled for DNA extraction according to modified CTAB (Cetyl Try Methyl Ammonium Bromide) method (Rogers and Bendich 1988). Eaten SSR markers related to rice storage ability from previous studies were used in the polymorphism survey. PCR was performed as follow: The 10-μL PCR reaction mixture contained 1 x buffer, 0.2 μM of each primer, 50 μM of dntps, 0.5 unit of Taq polymerase (Tiangen Company, Beijing, china), and 10 ng of genomic DNA as a template. The thermal cycler was programmed for a first denaturation step of 4 min at 94°C, followed by 30 cycles, each of 94°C for 30 s, 55 for 30 s, and 72°C for 30 s and the final extension step of 5 min at 72°C. The PCR products were separated on 8.0% non-denaturing polyacrylamide gel and detected using the silver staining method. Generated DNA bands were analyzed and scored 1 for the presence- or 0 for the absence of allele. Six SSR markers (RM 1, RM5, RM71, RM251, RM161 and RM234) were highly polymorphic among the studied Genotypes were used for genotyping (Doku *et al* 2013, Hang *et al* 2015, Adeboye *et al* 2015 and Kumar *et al* 2016), these markers located on Chromosomes 1, 2, 3, 5 and 7 respectively.

Table 2 List and sequences of polymorphic primers.

No	Name	Forward sequence	Reverse sequence
1	**RM1**	**5'-GCGAAAACACAATGCAAAAA-3'**	**5'-GCGTTGGTTGGACCTGAC-3'**
2	**RM5**	**5'-TGCAACTTCTAGCTGCTCGA-3'**	**5'-GCATCCGATCTTGATGGG-3'**
3	**RM71**	**5'-CTAGAGGCGAAAACGAGATG-3'**	**5'-GGGTGGGCGAGGTAATAATG-3'**
4	**RM251**	**5'-GAATGGCAATGGCGCTAG-3'**	**5'-ATGCGGTTCAAGATTCGATC-3'**
5	**RM161**	**5'-TGCAGATGAGAAGCGGCGCCTC-3'**	**5'-TGTGTCATCAGACGGCGCTCCG-3'**
6	**RM 234**	**5'-ACAGTATCCAAGGCCCTGG-3'**	**5'-CACAGTGAGACAAAGACGGAG-3'**

RESULTS AND DISCUSSION

Analysis of variance

The mean squares due to genotypes before storage (control) were significant and highly significant for all studied traits except germination index (Table 3). Storage temperature (T), storage periods (S), and varieties mean squares were found to be highly significant for all the studied characters. The interactions of S x T, S x V, T x V and S x T x V were detected to be significant and highly significant for all studied characters except for germination index at the interactions of S x T and S x T x V (Table 4). These results indicate high variances between genotypes before storage and effective storage methods after 24 months of storage. These results agreed with previous studies by Elgamal *et al* (2018) during studies on genetic diversity of some rice cultivars. Useful breeding programs are depending on modern and adapted knowledge of genetic diversity among varieties to utilize the available genetic resources to create new genotypes, morphological markers reflect not only the genetic contribution of the genotypes but also the interaction of the genotype with the environment in which it is revealed (Thenmozhi and Rajasekaran 2013).

Table 3. Mean squares of ordinary analysis of the genotypes at control.

SOV	df	1000-G. weight (g)	Germination percentage (%)	Germination index	Hulling (%)	Milling (%)	Head rice (%)
Replications	2	0.033	8.656	0.001	3.291	1.608	104.429
Genotypes	10	12.771**	39.588**	0.001 ns	7.417**	8.342**	123.873*
Error	20	0.264	2.302	0.001	0.295	0.569	47.80

Table 4. Analysis of variance model for combined data of split-plot design of the separate experiments

SOV	df	1000-G. weight (g)	Germination percentage	Germination index	Hulling (%)	Milling (%)	Head rice (%)
Temperatures degrees (T)	2	14.450**	1204.98**	0.1229**	901.26**	634.09**	3,057.74**
Error (a)	6	0.309	0.698	0.0005	1.405	3.435	6.455
Storage periods (S)	3	3.922**	884.82**	0.0042**	275.31**	153.670**	106.97**
S x T	6	12.031**	1835.56**	0.0046ns	259.91**	80.65**	383.714**
Error (b)	18	0.306	2.393	0.0002	1.76	2.644	4.235
Varieties (V)	10	126.238**	173.50**	0.0175**	25.052**	68.652**	367.51**
S x V	30	373.166**	1992.5**	0.0578**	117.69**	288.21**	949.45**
T x V	20	2.910**	28.666**	0.0114**	6.892**	7.232*	90.598**
S x T x V	60	71.816**	834.526**	0.0233ns	135.74**	322.93*	764.82**
Error (c)	240	0.325	2.369	0.0005	1.260	4.366	3.770

Rice varieties performance

We can divide the studied traits into two groups; the first one for research purpose and genetic resources maintenance including 1000 grains weight, germination percentage and germination index, while the second for grain quality and consuming purpose. The evaluation of eleven varieties for the studied morphological traits before storage and within the storage period under different storage temperatures are shown in Tables (5 through 10). Regarding 1000 grains weight, the cultivars Sakha 104, Sakha 101 and Giza 181 scored the highest values at control (28.90, 28.77 and 28.67g, respectively), even at 24 months storage (27.53, 26.93 and 28.13g, respectively). In relation to storage temperature, the highest values of 1000 grains weight were observed at $5C^o$ during all storage periods for most of the studied cultivars. The highest effective storage period was 24 months, which showed the lowest 1000 grains weight values for most of the studied varieties (Table 5).

Table 5. Mean performances of studied varieties for 1000- grain weight (g) under the control and all storage conditions.

Genotyping	Control	6 months storage			12 months storage			18 months storage			24 months storage		
		R.T.	$5C^o$	$-5C^o$	R.T.	$5C^o$	$-5C^o$	R.T.	$5C^o$	$-5C^o$	R.T.	$5C^o$	$-5C^o$
Giza177	27.80	25.77	26.00	24.77	24.47	25.20	24.80	23.87	24.23	23.67	23.47	23.23	23.73
Giza178	21.37	21.70	21.77	22.03	20.80	20.80	21.27	20.60	21.40	21.13	18.77	20.07	19.80
Giza179	26.97	25.33	24.43	23.37	21.87	23.20	23.70	24.47	24.13	23.57	23.10	23.87	22.83
Giza181	28.67	28.20	28.27	27.97	27.73	28.13	27.63	26.83	28.13	27.33	26.33	28.13	27.00
Sakha101	28.77	26.00	26.07	26.80	26.40	26.40	27.87	26.40	25.30	24.53	25.37	26.93	25.27
Sakha102	27.27	25.47	25.67	26.07	25.90	26.07	26.80	23.00	25.17	24.40	22.37	24.73	24.23
Sakha103	27.80	21.73	23.73	22.33	22.13	24.60	22.73	25.93	27.13	27.43	25.50	26.17	25.77
Sakha104	28.90	22.97	22.87	22.97	23.43	24.87	23.53	25.93	27.97	27.27	24.43	27.53	26.90
Sakha105	27.33	25.50	25.70	26.77	25.43	25.63	26.87	25.20	26.80	26.50	25.13	26.80	26.40
Sakha106	27.73	26.00	25.87	27.23	26.40	26.13	27.03	25.83	26.17	26.37	25.20	26.57	26.30
E. Yasmine	27.27	22.87	22.73	23.07	23.00	22.67	23.53	22.70	23.67	23.77	22.17	23.63	23.20
LSD 5%	0.88	1.30	0.88	0.67	0.65	1.53	0.73	1.15	0.95	1.16	0.83	0.54	0.76

R.T.: room temperature condition

For germination percentage (Table 6), the main observation was the negative effects of storage duration and storage temperature on all studied varieties, the cultivars Giza 177 and Sakha 102 scored the highest germination percentage before storage (control) with the same value (96.33%), while the cultivars Giza 181 and Giza 179 showed the lowest viability with values86.67% and 87.00%, respectively.

Table 6. Mean performances of studied varieties for seed germination percentage (%) under the control and all storage conditions.

Genotyping	Control	6 months storage			12 months storage			18 months storage			24 months storage		
		R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº
Giza177	96.33	95.00	95.33	95.67	89.33	93.67	91.67	80.67	85.33	82.00	78.67	84.33	80.33
Giza178	89.33	88.33	89.33	81.67	81.00	87.33	82.00	73.00	86.67	75.33	72.67	85.33	74.67
Giza179	87.00	86.67	86.67	81.33	79.33	85.33	81.67	73.33	86.00	79.33	72.00	86.00	78.33
Giza181	86.67	85.40	85.27	85.90	83.07	84.60	84.57	80.67	84.60	82.83	84.00	84.60	83.00
Sakha101	92.33	79.00	78.33	74.00	84.33	87.67	85.33	81.00	84.67	82.33	78.33	84.67	82.00
Sakha102	96.33	91.00	90.00	77.67	87.00	89.67	88.67	80.67	86.33	83.33	78.67	85.67	81.00
Sakha103	92.33	92.33	92.67	90.67	80.67	87.67	86.67	78.33	88.33	85.67	73.00	88.00	84.00
Sakha104	94.33	90.33	91.00	83.67	85.67	90.33	88.00	80.33	89.00	86.33	78.33	89.00	84.33
Sakha105	93.00	93.33	94.00	80.33	80.00	89.33	85.67	76.33	86.67	85.33	69.67	86.33	83.67
Sakha106	94.67	92.33	92.00	83.67	85.00	89.67	87.67	79.33	89.67	87.00	77.33	89.00	82.67
E. Yasmine	92.67	92.00	90.67	81.67	79.33	88.33	84.67	74.67	87.00	85.33	76.33	86.00	82.67
LSD 5%	2.58	1.87	2.91	3.04	2.02	2.13	3.89	2.37	1.91	1.97	4.06	1.94	2.04

R.T.: room temperature condition

The highest effective storage period was 24 months and the highest effective storage temperature was room temperature, which showed the lowest germination percentage for Sakha105 with value69.67%.

Regarding germination index, the varieties Sakha 102 and Sakha 104 showed the best performances after germination at control condition with the same value (0.97). while under all storage treatments, the cultivars Giza 178, Giza 178 and Egyptian Yasmine scored the highest germination index with values0.97 and 0.96. The varieties Giza 181 and Sakha 106 showed the lowest value (0.82) of germination index at 24 months storage under room temperature conditions. The results of the traits which related to research purpose and genetic resources maintenance, concluded that the germination percentage was the most important traits to study storage ability and could be used as an indicator for viability after storage (Rao and Jackson 1996, Chowdhury *et al* 2014 and Mutinda *et al* 2017).

Table 7. Mean performances of Studied varieties for germination index under the control and all storage conditions.

Genotyping	Control	6 months storage			12 months storage			18 months storage			24 months storage		
		R.T.	5C°	-5C°	R.T.	5C°	-5C°	R.T.	5C°	-5C°	R.T.	5C°	-5C°
Giza177	0.95	0.94	0.93	0.95	0.95	0.94	0.95	0.95	0.94	0.95	0.95	0.94	0.95
Giza178	0.94	0.96	0.92	0.94	0.96	0.95	0.96	0.95	0.97	0.93	0.96	0.97	0.96
Giza179	0.95	0.95	0.92	0.95	0.95	0.96	0.95	0.95	0.96	0.95	0.95	0.96	0.95
Giza181	0.95	0.95	0.94	0.90	0.95	0.95	0.94	0.89	0.93	0.85	0.82	0.90	0.84
Sakha101	0.96	0.86	0.92	0.92	0.86	0.92	0.92	0.86	0.92	0.92	0.86	0.92	0.92
Sakha102	0.97	0.85	0.95	0.95	0.85	0.95	0.95	0.85	0.95	0.95	0.85	0.94	0.94
Sakha103	0.96	0.87	0.95	0.95	0.87	0.93	0.95	0.87	0.93	0.95	0.87	0.92	0.93
Sakha104	0.97	0.84	0.95	0.95	0.84	0.95	0.93	0.84	0.95	0.94	0.84	0.93	0.92
Sakha105	0.96	0.87	0.91	0.95	0.87	0.91	0.94	0.87	0.91	0.95	0.87	0.91	0.92
Sakha106	0.94	0.84	0.95	0.94	0.84	0.95	0.95	0.84	0.95	0.95	0.84	0.94	0.94
E. Yasmine	0.96	0.93	0.96	0.96	0.95	0.96	0.96	0.95	0.96	0.95	0.95	0.96	0.96
LSD 5%	0.038	0.029	0.032	0.035	0.024	0.034	0.047	0.025	0.044	0.048	0.027	0.033	0.046

R.T.: room temperature condition

In relation to grain quality traits (hulling, milling and head rice percentages) the japonica varieties showed the highest grain quality for the three quality traits at the control and during the storage periods under the different storage temperatures. On the other hand, the indica and indica / japonica varieties showed the lowest values of grain quality traits among the studied varieties (Tables 8 through 10). Sakha 105, Giza 177, Sakha 103 and Sakha 106 showed the highest hulling percentage with the values 82.11, 81.83, 81.50 and 81.34%, respectively at the control. Also, the varieties Sakha 105, Sakha 106, Sakha 102 and Sakha 101 showed the highest values during all storage periods under the storage temperature 5C°. On the other hand, the varieties Giza 178, Giza 179, Giza 181 and E. Yasmine showed the lowest values at the control (78.90, 79.61, 76.63 and 78.56%), respectively and during the most of storage periods under the room temperature and -5C° conditions (Table 8). The present data in Tables (9 and 10) showed that the milling percentage and head rice percentage follow the hulling percentage with the same trend of results. The results indicated that the indica and indica/japonica varieties showed the best performances after all storage periods up to 24 months more than the japonica varieties for research purpose, but the japonica varieties were the best for grain quality and consuming purpose.

Table 8. Mean performances of studied varieties for hulling percentage (%) under the control and all storage conditions.

Genotyping	Control	6 months storage			12 months storage			18 months storage			24 months storage		
		R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº
Giza177	81.83	78.93	80.20	76.07	75.37	78.73	75.83	72.17	76.73	76.27	74.60	77.07	75.73
Giza178	78.90	75.77	78.87	75.60	74.13	77.20	74.93	72.07	77.03	74.77	69.60	71.00	74.67
Giza179	79.61	75.27	79.63	74.67	71.93	77.50	74.33	72.03	76.17	74.10	70.00	71.57	73.43
Giza181	76.63	76.30	76.17	76.83	70.47	75.83	74.50	71.00	75.83	72.17	70.33	72.67	70.67
Sakha101	80.13	78.20	79.97	77.77	73.63	79.40	75.70	74.87	78.97	75.60	71.40	78.50	75.43
Sakha102	81.01	78.10	81.53	78.17	74.40	79.20	77.27	72.87	78.63	74.73	71.80	78.27	74.30
Sakha103	81.50	80.53	80.73	74.63	72.90	78.20	72.60	73.63	77.20	74.70	71.67	76.97	71.73
Sakha104	78.95	79.07	79.10	77.30	72.63	77.50	74.90	73.17	77.53	74.57	69.93	77.23	74.67
Sakha105	82.11	77.13	81.67	76.03	73.70	79.10	74.57	74.57	78.83	74.57	71.53	77.83	74.53
Sakha106	81.34	78.40	81.17	76.97	74.43	79.50	75.40	72.60	78.50	74.20	72.00	78.03	75.23
E. Yasmine	78.56	75.13	77.93	74.53	71.97	77.10	74.50	70.00	76.60	74.43	68.40	72.40	72.07
LSD 5%	0.92	1.41	1.12	1.34	1.83	1.14	2.34	3.81	1.03	2.07	2.22	1.75	0.94

R.T.: room temperature condition

Table 9. Mean performances of studied varieties for milling percentage (%) under the control and all storage conditions.

Genotyping	Control	6 months storage			12 months storage			18 months storage			24 months storage		
		R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº
Giza177	69.33	66.43	68.83	67.53	66.87	69.10	65.17	65.30	68.47	65.40	64.07	67.93	65.23
Giza178	66.15	62.77	68.20	65.30	64.50	67.27	61.30	62.63	67.37	61.30	62.57	63.43	60.97
Giza179	67.60	63.07	67.23	69.63	65.20	68.00	62.00	61.83	65.13	62.10	61.43	62.27	60.87
Giza181	65.00	64.13	64.47	63.67	62.33	63.67	62.33	59.67	62.33	60.67	58.00	59.33	56.67
Sakha101	68.27	67.20	68.43	65.10	65.73	68.53	64.43	62.63	67.73	64.93	63.57	66.57	64.70
Sakha102	70.28	61.10	70.00	66.47	65.77	69.23	64.27	64.27	68.70	63.60	62.20	67.83	63.13
Sakha103	69.10	63.93	66.50	63.30	64.77	68.33	64.23	63.43	68.13	63.90	62.20	66.53	63.90
Sakha104	67.66	68.73	69.10	69.07	64.10	67.43	63.90	62.93	67.00	63.33	61.57	65.83	63.10
Sakha105	70.60	65.40	70.43	65.30	64.00	66.87	61.10	63.40	66.33	61.50	61.47	66.00	62.80
Sakha106	69.81	66.40	70.20	66.87	64.53	68.23	63.83	61.10	67.77	63.93	66.70	66.80	63.03
E. Yasmine	69.11	63.53	68.33	62.97	64.93	67.77	62.33	61.33	67.30	62.40	60.90	63.33	62.33
LSD 5%	1.28	1.69	0.89	1.91	2.65	1.42	3.36	3.81	1.79	2.80	9.75	2.30	1.98

R.T.: room temperature condition

Table 10. Mean performances of studied varieties for head rice percentage (%) under the control and all storage conditions.

Genotypes	Control	6 months storage			12 months storage			18 months storage			24 months storage		
		R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº	R.T.	5Cº	-5Cº
Giza177	64.73	62.27	63.63	58.40	56.97	61.77	56.73	53.57	63.17	53.90	56.03	62.17	56.23
Giza178	60.90	51.47	63.60	49.07	57.53	63.17	55.93	48.77	61.03	45.10	47.33	60.27	48.23
Giza179	56.63	44.80	62.43	50.43	49.07	60.73	48.60	46.30	60.47	44.27	48.07	60.10	52.07
Giza181	52.67	53.33	53.83	52.17	52.33	52.83	51.17	50.33	51.83	50.33	47.00	51.83	47.67
Sakha101	65.8	58.97	65.03	58.03	57.53	64.40	62.63	58.83	62.97	57.80	57.70	62.07	58.27
Sakha102	63.9	55.93	59.83	51.30	52.67	62.83	50.93	55.60	60.57	53.20	57.90	59.70	58.63
Sakha103	64.43	46.50	62.90	45.37	57.53	63.40	54.77	57.50	62.73	55.63	47.87	61.13	47.83
Sakha104	65.27	63.87	64.47	63.33	62.83	64.77	61.93	60.43	62.13	55.10	56.53	61.47	56.73
Sakha105	64.23	55.97	63.07	52.30	49.73	61.20	52.53	59.67	61.90	54.37	50.43	60.87	52.93
Sakha106	65.33	58.27	66.40	57.07	58.87	65.13	58.43	55.73	63.60	57.00	56.43	62.93	57.20
E. Yasmine	63.63	55.53	60.80	49.53	51.23	61.53	51.10	46.20	61.97	46.33	47.57	55.60	48.50
LSD 5%	11.78	4.67	2.54	2.70	1.95	2.47	4.57	3.21	2.25	4.05	3.59	1.32	4.30

R.T.: room temperature condition

Japonica group varieties need more special storage conditions like cold conditions during long-term storage. Chang (1991), Rao and Jackson (1996) and Ellis (2011) reported that japonica cultivars possessed intrinsically poorer storage characteristics than the indica cultivars and more sensitive to high temperature store conditions. Saida Naik and Chetti (2017) revealed that among the storage conditions, cold storage (4 ± 1 ºC) recorded higher score of sensory evaluation of rice over room temperature (25 ± 2º C), irrespective of the storage containers throughout the storage period of 18 months. Kandil et *al* (2012) showed that rice cultivars (Sakha 102, Sakha 104, Sakha 105, Sakha 106, Giza 178, Giza 181, Giza 182, Egyptian Yasmin, Egyptian Hybrid 1 and Egyptian Hybrid 2) significantly varied in means of final germination percentage, germination rate and germination index. Bhardwaj and Sharma (2015) indicated that impacts of the storage period of paddy rice under different environments have a profound effect on storage in terms of decreased bulk density and germination percentage.

SSR polymorphism

Eighteen SSR markers were used for investigating the polymorphism among the common eleven Egyptian rice cultivars based on their abilities of storage for 24 months. Six SSR markers showed highly polymorphic pattern among the studied genotypes, the electro-photogram for the amplified DNA fragments for these markers are shown in Figures (1through 6). A total of 21 polymorphic alleles were detected and ranged between two by RM234 and five by RM5 markers. The RM1 marker amplified two different alleles for the studied rice accessions (Figure 1), the molecular weight of these alleles ranged between 80 bp and 120 bp, while the RM 5 marker amplified five different alleles, (Figure 2), the molecular weight of these alleles ranged between 110 bp and 200 bp. The RM71 marker amplified four different alleles with molecular weight ranged between 150 bp and 230bp (Figure 3). The RM251 marker amplified four different alleles with molecular weight ranged between 140 bp and 200 bp (Figure 4). The RM161 marker amplified two different alleles with molecular weight ranged between 80 bp and 110 bp (Figure 5). The RM234 marker amplified two different alleles with molecular weight ranged between 180 bp and 220 bp (Figure 6).

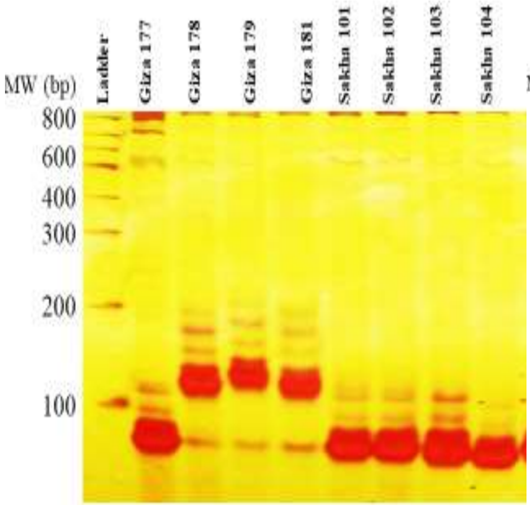


Fig. 1. Genetic polymorphism among the seven genotypes using RM 1.

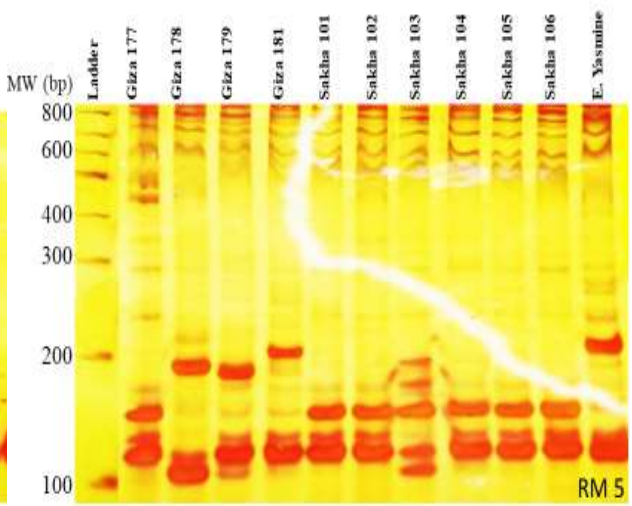
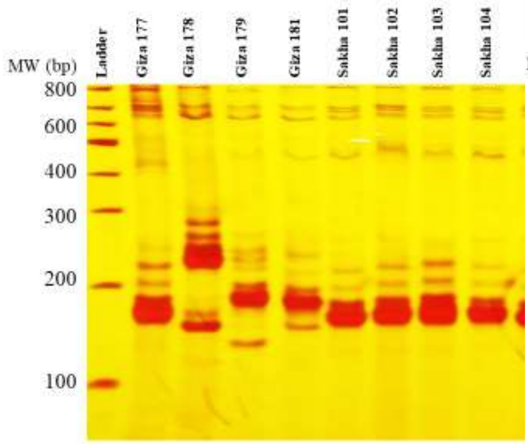


Fig. 2. Genetic polymorphism among the seven genotypes using RM 5.



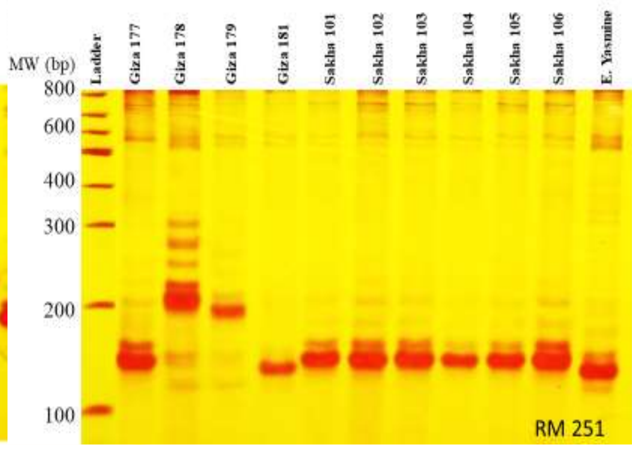
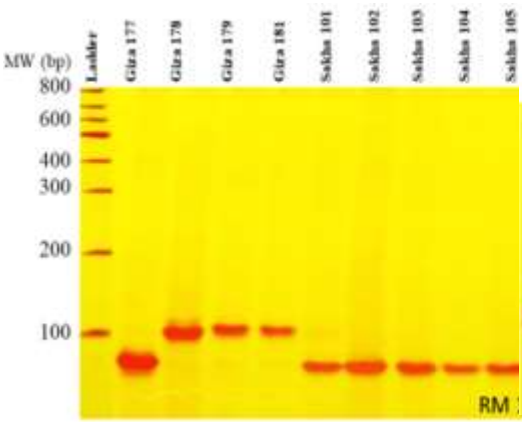


Fig. 3. Genetic polymorphism among the seven genotypes using RM 71.

Fig. 4. Genetic polymorphism among the seven genotypes using RM 251.



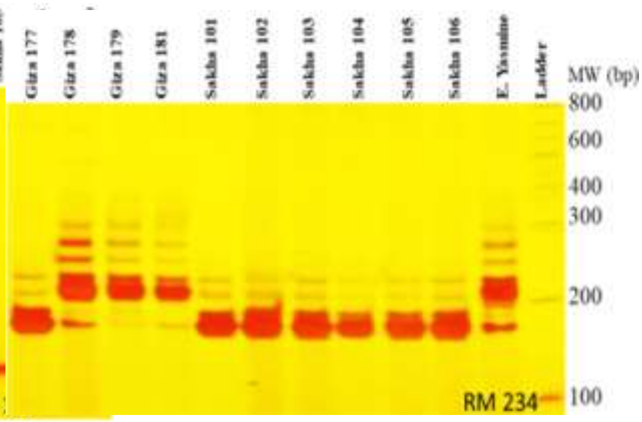


Fig. 5. Genetic polymorphism among the seven genotypes using RM 161.

Fig. 6. Genetic polymorphism among the seven genotypes using RM 234.

Cluster analysis

To determine the strong degree of the relation of the genotypes and present in a simple way, we use the cluster analysis depending on the data out of all SSR polymorphic markers (Figure 7). The cluster analysis showed that the phylogenetic divided the studied accessions into two main groups (A and B) in addition to some sub groups belong to each main group. Group A included seven rice varieties all of them belong to Japonica type, sub groups appeared but without significant distances. On the other hand, the group B included four rice varieties were divided into two sub groups, two varieties per each. The first subgroup included indica types varieties; Giza 181 and Egyptian Yasmine. The second subgroup included the two Indica / Japonica varieties Giza 178 and Giza 179. Japonica and indica varieties were obviously separated into clusters, the genetic diversity was greater for indica than japonica varieties, as reported in previous studies by Oka and Morishima (1982), Junjian *et al* (2002) and Lin *et al* (2012).

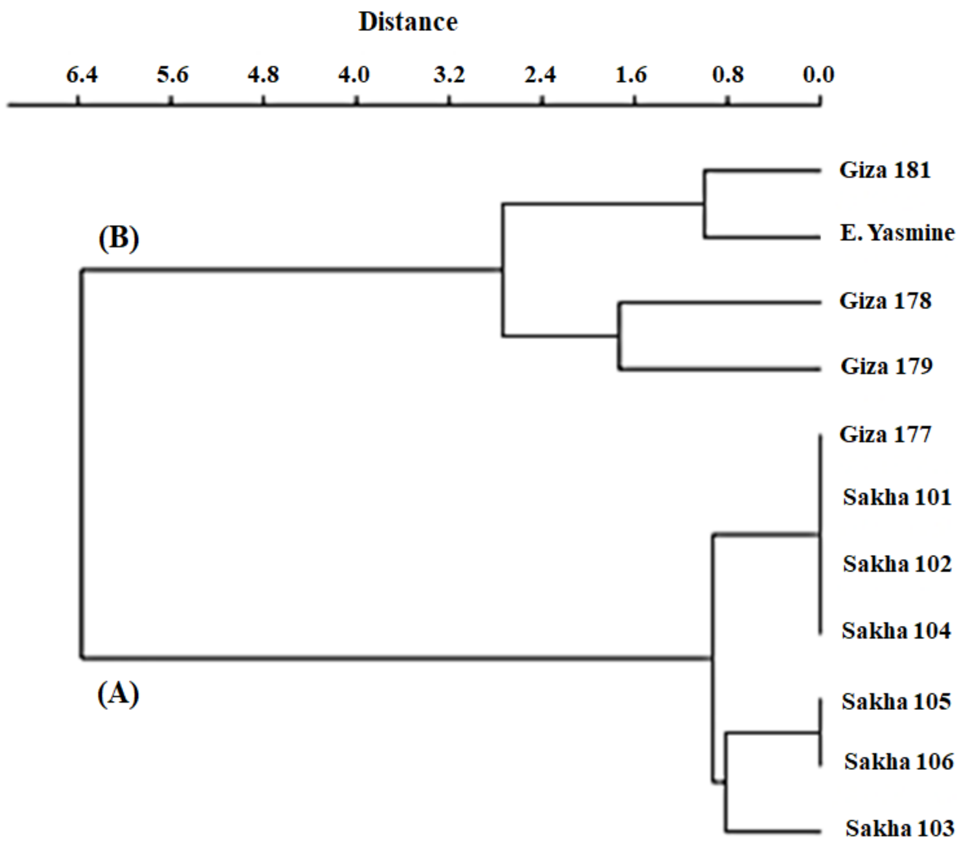


Fig. 7. Dendrogram of eleven rice accessions based on genotyping data using hierarchical cluster analysis (wards method).

Similarity and distance index

Similarity and distance index are indicators of the similarity degree and genetic distance between genotypes, its values ranged from zero distance in the case of complete similarity and unity in case of non-similarity. The present results in Table 11 based on the combined data across all studied traits. The highest distances between studied cultivars before storage (below diagonal) were 0.659, 0.654 and 0.614 which scored between Giza 181-Sakha 106, Giza 178-Sakha 106 and Giza 181-Sakha 105, respectively, while the lowest distances before storage were scored between Sakha 101- Sakha 103, Sakha 105- Sakha 106 and Sakha 102- Sakha 103 with values 0.137, 0.152 and 0.161, respectively. After 24 months storage (above diagonal), the highest distances between studied cultivars were scored between Giza 178-Sakha 106, Giza 178-Sakha 101 and Giza 181-Giza 177 with values 0.702, 0.628 and 0.603, respectively. While, the lowest distances were between Sakha 104- Sakha 102, Sakha 105- Sakha 103 and Giza 178-Giza 179 with values 0.093, 0.131 and 0.179, respectively. Most of the present results explain the strength of genetic relationship within rice groups types indica, japonica or indica/japonica, which hadn't been affected by storage conditions.

Table 11. Similarity and distance index among studied cultivars based on the combined data across all studied traits.

Genotypes	Giza 177	Giza 178	Giza 179	Giza 181	Sakha 101	Sakha 102	Sakha 103	Sakha 104	Sakha 105	Sakha 106	E. Yasmine
Giza177	0	0.403	0.362	0.603	0.314	0.331	0.425	0.371	0.424	0.555	0.467
Giza178	0.588	0	0.179	0.578	0.628	0.442	0.308	0.498	0.369	0.702	0.355
Giza179	0.502	0.275	0	0.399	0.487	0.340	0.291	0.331	0.241	0.598	0.254
Giza181	0.585	0.392	0.264	0	0.419	0.376	0.270	0.283	0.346	0.445	0.456
Sakha101	0.185	0.473	0.432	0.486	0	0.224	0.338	0.214	0.394	0.309	0.513
Sakha102	0.196	0.471	0.392	0.602	0.252	0	0.272	0.093	0.318	0.303	0.315
Sakha103	0.207	0.453	0.319	0.505	0.137	0.161	0	0.237	0.131	0.425	0.379
Sakha104	0.206	0.469	0.495	0.513	0.161	0.185	0.205	0	0.262	0.267	0.350
Sakha105	0.237	0.597	0.463	0.614	0.244	0.231	0.169	0.353	0	0.369	0.295
Sakha106	0.244	0.654	0.520	0.659	0.289	0.258	0.203	0.314	0.152	0	0.428
E. Yasmine	0.400	0.268	0.269	0.322	0.282	0.279	0.262	0.253	0.405	0.462	0

Data obtained after 24 months storage (above diagonal)

Data obtained from Control (below diagonal)

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تحليل التنوع الوراثي لبعض اصناف الأرز المصرية المختلفة في القدرة التخزينيه معتمدا علي المعلمات الجزيئية SSR

وليد حسن الجمل [١]، محمد محمد قمرة [٢] و رمضان محمد سليمان [٢]

١. قسم بحوث الأرز – معهد بحوث المحاصيل الحقليه – مركز البحوث الزراعيه

٢. قسم المحاصيل – كلية الزراعة – جامعة كفرالشيخ

أجريت هذه الدراسة بمحطة بحوث سخا الزراعية – قسم بحوث الارز خلال الفتره من نوفمبر ٢٠١٤ إلي أكتوبر ٢٠١٦ بهدف تقييم بعض أصناف الأرز المصرية المنزرعه و التي تنتمي الي طرز مختلفة من الأرز لقدرتها علي تحمل التخزين تحت ظروف تخزين مختلفة (تبريد و تجميد و درجة حرارة الغرفه) لمدة أربعة و عشرون شهرا. بالإضافة إلى تقدير التنوع الوراثي بين هذه التراكيب الوراثية تحت الدراسة باستخدام المعلمات الجزيئية SSR المتعلقة بالقدرة علي التخزين. تم استخدام أحد عشر من أصناف الأرز المصرية المنزرغه في هذه الدراسة. تم استخدام ١٨ معلم جزيئي SSR المتعلقة بقدرة التخزين لتحديد التنوع الوراثي وتعدد الأشكال بين التراكيب الوراثية المدروسة. أظهرت ستة من المعلمات الجزيئيه SSR وهي RM1 و RM5 و RM71 و RM 251 و RM161 و RM234 تباينًا كبيرًا بين أصناف الأرز المدروسة وأكثر من تمييزها إلى مجموعات مختلفة بناءً على قدرتها التخزينية. تم اكتشاف اجمالي ٢١ من الأليلات متعددة الأشكال وتراوحت أعدادها بين اثنين بواسطة RM 234 وخمسة بواسطة المعلم الجزيئي RM 71. فيما يتعلق بالتحليل العنقودي للبيانات التي تم الحصول عليها من المعلمات الجزيئية SSR ، أظهرت النتائج أن التحليل العنقودي قسّم الأصناف المدروسة إلى مجموعتين رئيسيتين (أ و ب) بالإضافة إلى بعض المجموعات الفرعية تنتمي إلى كل مجموعة رئيسية. المجموعة (أ) ضمت سبعة أصناف من الأرز كلهم ينتمون إلى مجموعة الأرز ياباني الطراز. من ناحية أخرى ، تضمنت المجموعة (ب) أربعة أصناف مقسمة إلى مجموعتين فرعيتين (أنواع الهندي و الهندي/ياباني). تأثرت الأصناف اليابانيه بمدة التخزين أكثر من الأنواع الأخرى خاصة عند نسبة الإنبات٪ ودليل الإنبات بعد سنتين من التخزين، درجة الحرارة العالية أدت الي زيادة آثار التخزين السلبية. الأصناف الهنديه و الهنديه/يابانية ذات قدره أكبر على التخزين أكثر من الأصناف اليابانية وحافظت على المزيد من حيوية البذور مع طوال فترة التخزين. درجة الحراره خمسه هي أفضل درجة حرارة لتخزين البذور لفترة قصيرة (المجموعه النشطة).

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