

AGRONOMIC AND MOLECULAR EVALUATION OF INDUCED MUTANTS IN RICE (*Oryza sativa* L.)

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ABSTRACT

The present study was conducted at the Farm of the Rice Research and Training Center, Sakha, Kafr El-Sheikh, Egypt, during 2000 to 2007 rice growing seasons. Five rice varieties, namely Giza 171, Giza 175, Giza 176, Giza 181 and GZ 1368-S-5 were the most widely grown Japonica and Indica rice varieties in Egypt, during the last period, possessed at that time many positive agronomic characteristics (e.g., wide adaptability, high yield potential, resistance to multiple diseases, and pests and good grain quality. But now, it has some drawbacks in its characters that make it ideal genotypes for identifying mutational changes traits for agronomic importance. Dry seeds of the above mentioned varieties were treated with different doses of gamma rays (100,200,300,400,and 500 Gy), at the National Center for Radiation Research and Technology, Nasr City, Cairo, Egypt, for raising M₁ generation. M₁ plants were established by transplanting in 2000 season. Seventy seven independent lines have been advanced to M₅ generation, enabling evaluation of quantitative traits by replicated trials and promising lines were selected and tested over three seasons as M₆, M₇ and M₈ generations. Morphological variations at vegetative and reproductive stages including plant type, and yield and its component characters are commonly observed in the five populations. The mutant characteristics identified, so far, consist of better resistance to lodging, blast disease, high yield potential, as well as, early maturity. The results obtained from field evaluation over three years, and, also, through PCR detection, indicated that the induced mutants were differed genetically from their parents. Therefore, these mutants could be used as a donor parents in the rice breeding program and some of them could be recommended to be new rice varieties, suitable to be grown in rice belt in Egypt, such as Giza 175- M₁₃ line.

INTRODUCTION

Rice is one of the most important crops in Egypt and its production plays a significant role in the strategy to overcome food shortage and improvement of self sufficiency for local consumption and export. Introducing new varieties of rice characterized by early heading, short stature, lodging resistance, blast resistance, and improved grain quality characters are main objectives for vertical increase of grain yield of rice. The total production increased during this period from 2.4 million tons (Ave. 1980-1989) to 4.64 million tons (Ave. 1990-2001). In 2005 season, the total rice production in Egypt reached 6.6 million tons with a national average of 10.00 tons/ha. (Badawi, 2005). This increase in rice production is mostly due to the development and releasing of new improved varieties having many desirable characters.

Most of the rice area starting from 1990 till 1995 was planted with the traditional varieties Giza171, Giza175, Giza176, Giza181 and GZ1368-S-5. These varieties have some constraints such as late maturity, lodging,

susceptibility to blast and some defects in their grain quality characters. Induced mutations have thus, played a vital role for the improvement of rice by developing in a large number of semi-dwarf, earliness, tillering ability, blast resistance, low amylose content and high yielding varieties in the world. (Soomro et al., 2006). For direct improvement of any agronomic trait, the basic requirement is the availability of adequate genetic variability. Induced mutations, with the discovery of array of radiation mutagen and improved treatments methods, offers possibility for the induction of desired changes in various attributes, which can be exploited as such or through recombination breeding (Akbar and Manzoor, 2003 and Kim, 2006).

The morphological mutations also provided genetic marker for the development of linkage maps. However, irradiation- induced mutations have not been the mainstay of gene identification tools because the mutations are not physically tagged, requiring considerable effort to isolate the gene after a phenotype has been identified. Yet, with high throughput genotyping, the efficiencies in detecting genetic polymorphism have been significantly improved. Many valuable mutant lines were induced in rice; some of them have been developed directly or indirectly into national regional new varieties. Ionizing radiation mutagenesis has been routinely used to generate genetic variability for breeding research genetic studies. More than 2200 crop varieties were released by the end of the last century using irradiation radiation mutagenesis among them 434 are rice varieties (Elayaraja et al., 2005; Wu et al., 2005 and Mohamed et al., 2006).

There are different types of ionizing radiation, namely X rays, gamma rays, protons, neutrons, alpha and beta particles. However, gamma rays are widely employed for mutation studies as they have shorter wave length and therefore, possess more energy per photon than X rays and penetrate deep into the tissue (Khin, 2006 and Zhou et al., 2006). The idea of producing artificial mutations and utilizing them for breeding cultivars plants was indicated as early as 1901 by the induction of mutations for factors which govern the heredity of quantitative characters is a promising tool for releasing new genotypes (Gomaa et al., 1995). It is an established fact that mutagen, besides causing changes in major genes, also induce mutations at loci governing the quantitative characters.

Mutagen agents, including gamma rays, offered great possibilities for increasing genetic variability of quantitative traits such as yield. Mutation breeding method by using gamma rays was started in Egypt for rice in 1960 to improve some desirable characters, without disturbing the constellation of the original varieties. The present study was carried out to induce and evaluate some mutations derived promising lines that possess high yielding ability, resistant to blast, early maturity and superior in some grain quality characters. Morphological and DNA based molecular assessment of the mutant lines and their corresponding original parental lines have been conducted to assist in detection the level of genetic variation.

MATERIALS AND METHODS

Varieties developed and their drawbacks:

Giza 171: A popular short grain Japonica type variety developed from the cross Nahda and Calady 40. It was released in 1977. It is Late (160 days from sowing), tall (140 cm) susceptible to blast and it has low yielding potential.

Giza 175: The origin was breeding line (1394-10) selected from the local top cross made between IRRI varieties and the local variety, Giza 14. It was registered as the new variety in 1991. It has high yield potential and other agronomic characters. Its cooking and eating quality were less acceptable to Egyptian consumes.

Giza 176: A breeding line (2175-5-6) selected from the local top cross Calrose 76/ Giza 172//GZ 242. It was a short grain Japonica type released in 1989 and was registered in 1991 as a new variety. It has yield potential, short stature, medium growth duration (145 days) but it has low hulling and milling (%) and became susceptible to blast.

Giza 181: Introduced to Egypt from IRRI through INGER program as IR1626-203 in advanced generation. It was released in 1987 and it was characterized by medium maturity (145 days), non-lodging, high yielding, resistant to blast but was not excellent in grain and cooking qualities.

GZ 1368: It was an Indica/Japonica type bred in Egypt, medium grain, high yielding, resistance to blast, medium growth duration but also was not excellent in cooking and eating characters,

Seeds of the above mentioned five rice varieties were irradiated with different doses of gamma rays (100, 200, 300, 400 and 500 Gy, from Co⁶⁰ source at the National Center for Radiation Research and Technology, Nasr City, Cairo, Egypt.

Two hundred and fifty seeds at 14 percent moisture content were used for each treatment and the same number of seeds was kept untreated as a control for each variety. Seeds of all treatments were directly sown after radiation treatment in order to raise M₁ plants. The selection of the best mutant lines from M₁ to M₅ was carried out based on individual plants. Seedlings were transplanted to the permanent field on five June, and singly grown in hills. Three plots were lay-out for each radiation treatment. Each plot contained ten rows, five meters long with twenty cm apart between rows and hills. All plants were under keen observation from sowing till ripeness and each plant was carefully examined.

In 2001 season, M₂ plants were grown and 77 progenies were selected and harvested individually, as promising mutants. These mutants comprised eight mutants from Giza 171, twenty eight mutants from Giza 175, nine mutants from Giza 176, seven mutants from GZ 1368-S-5 and twenty five mutants from Giza 181.

In 2002 season, 77 M₃ mutants were tested with their original parents in a randomized complete block design with three replications and the best plants from each mutant were selected to raise the 77- M₄ mutants.

In 2003 season, the five parents and 77 M₄ mutants were planted in the seedbed. After thirty days, seedlings of the above populations were individually transplanted in the field in a randomized complete block design with three replications. All observations were recorded during the growing season till harvesting.

In 2004 season, the same technique was used for raising M₅ generation. Seeds of the best plants were selected and individually harvested for each mutant. The manner of planting and experimental design and collection data were similar to those followed in the previous generations. It should be noticed that all cultural practices, were followed as usually done with ordinary rice culture in each growing season and weed control were applied manually and chemically.

After harvest of M₅ plants, samples of seeds of each mutant line, as well as, the parental varieties were tested in the laboratory for hulling (%), milling (%), gel consistency, gelatinization temperature and amylose content characters. After analysis of M₅ generation, it appeared that different mutant lines were morphologically and genetically stable. From M₆ generation, those mutant lines were continuously evaluated with their respective parents in replicated yield trials at different locations and seasons from 2005 to 2007.

After analysis of M₆ generation, it appeared that different mutant lines were morphologically and genetically stable. It would be better if these mutant lines promoted to be grown under different locations as a yield trails experiments, hoping that some of these mutants lines may be surpassed the local varieties for yield and its component characters, resistant to blast as well as grain quality characters. From M₆ generation, those mutant lines were continuously evaluated in replicated yield trials at different locations and seasons from 2005 to 2007. Besides yield and its components, some agronomic characters; i.e., maturity, plant stature, photoperiod sensitivity, grain quality characters; i.e., milling recovery, amylose content and resistance to blast were evaluated. Based on the overall performance, five mutants were selected from the five varieties, as the best mutant lines, according to their desirable characters comparing with the others.

DNA isolation and PCR reaction:

For the molecular evaluation, DNA isolation and purification from the five parental lines as well as the five derived lines was carried out using CTAB method. The DNA was quantified using gel assay method and then PCR was performed. A total of nine pairs SSR markers were used for the screening purpose (Table 1).

The PCR was performed in 10 μ l PCR volume containing 50 ng of template DNA, five pmole of each of forward and reverse primers, 0.1mM dNTP's, 1x PCR buffer (10mM Tris, pH 8.0, 50mM KCl and 50mM ammonium sulphate), 1.8mM MgCl₂, and 0.2 units of *Taq* DNA polymerase. Initial denaturation at 94 $^{\circ}$ C for 5 minutes was followed by 35 cycles of amplification with template denaturation at 94 $^{\circ}$ C for one minute, primer annealing at 55.7 $^{\circ}$ C for one min and primer extension at 72 $^{\circ}$ C for two minutes. After the end of the 35th cycle, a final extension at 72 $^{\circ}$ C for seven

minutes was given followed by storage at 4.0 °C. The PCR products were separated using 1.5% agarose gel stained with Et Br solution (1 mg/l). The banding pattern was then scored and used to prepare the matrix. Employing the computer package DARwin software that was developed by *Perrier and Jacquemoud (2006)* was used to establish genetic relationship among the genotypes based on Unweighted Pair Group Method of Arithmetic Averages (UPGMA).

Table (1): SSR Markers Sequences used in the current study.

Marker	Chromosome	Primer sequence (F)	Primer sequence (R)
RM289	5	TTCCATGGCACACAAGCC	CTGTGCACGAACCTCCAAAG
RM223	8	GAGTGAGCTTGGGCTGAAAC	GAAGGCAAGTCTTGGCACTG
RM148	3	ATACAACATTAGGGATGAGGCTGG	TCCTTAAAGGTGGTGCAATGCGAG
RM164	5	TCTTGCCCGTCACTGCAGATATCC	GCAGCCCTAATGCTACAATTCTTC
RM212	1	CCACTTTCAGCTACTACCAG	CACCCATTTGTCTCTCATTATG
RM242	9	GGCCAACGTGTGTATGTCTC	TATATGCCAAGACGGATGGG
RM412	6	CACTTGAGAAAGTTAGTGCAGC	CCCAAACACACCCAAATAC
RM108	9	TCTCTTGC GCGCACACTGGCAC	CGTGCACCACCACCACCACCAC
RM36	3	CAACTATGCACCATTGTGCGC	GTA CTCCACAAGACCGTACC

Statistical analysis:

The analysis of variance for the randomized complete block design was done for each studied trait in the last three years, M₆, M₇ and M₈ generations under normal conditions. The combined analysis was calculated over the three years to test the interaction of the different genetic components with the three years. The homogeneity of error variance was tested as described by *Bartlett (1937)*.

The data were statistically analyzed follows *Burton (1952) and Chang et al. (1974)* and some genetic parameters ; i.e., phenotypic variance(PV), genotypic variance(GV), phenotypic coefficient of variation (PCV), genotypic coefficient of variation(GCV), heritability in broad sense(Hb) and phenotypic correlation coefficients were computed (*Johanson et al.,1955; Lush, 1940 and Burton,1951*). Means of the different mutants were compared with their respective control, using the least significant differences (L. S.D.) method.

Table (2): Rice mutants induced from local varieties by using gamma rays.

Mutant	Parent variety	Mutagenic treatment	Improved characterizes
Giza171-M ₆	Giza 171	Dried seeds, 200 GY gamma rays	High yield, resistant to blast, short stature and early maturity.
Giza 175-M ₁₃	Giza 175	Dried seeds, 300 Gy gamma rays	High yield and low amylose content
Giza 181-M ₁₁	Giza 181	Dried seeds, 300 Gy gamma rays	High yield and good grain quality characters.
Giza 176- M ₁₇	Giza 176	Dried seeds, 200 Gy gamma rays	High yield, resistant to blast, high hulling (%) and milling (%).
GZ1368 -M ₁₃	GZ 1368-S-5	Dried seeds, 100 Gy gamma rays	High yield, high milling (%) and low amylose content.

RESULTS AND DISCUSSION

Giza 171- M₆:

The rice mutant Giza 171-M₆ evolved by treating the seeds of Giza 171 with gamma rays (200 Gy). The major improvement of the Giza 171-M₆ over Giza 171 were the earliness, where this mutant has 107.00 days to heading, which is thirteen days earlier than the original variety Giza 171(120.00 days). With early maturity the mutant can be cultivated with considerably less water. The mutant, also, has reduction in plant height (95.00cm) by 40.00 cm comparing with the original variety, Giza 171(135.00 cm). It also has thirty more grains/panicle (165.00 grains/panicle) comparing with the original variety Giza 171 (135.00 grains/panicle). The mutant Giza 171-M₆ possessed high yield (35.00g/plant), good grain quality characters; i.e., high milling (72.00 %), low amylose content (19.00 %) and good resistance to blast. It could be concluded that the mutant Giza 171-M₆ possessed good characteristics, such as short growth duration, short stature, resistance to blast, high yielding ability and high milling (%) (Table 3). This mutant could easily replace the original variety due to its desirable characteristics with still higher yield potential.

Giza 175- M₁₃:

Giza 175-mutant was developed from the variety Giza 175 through gamma rays treatment (300 Gy) of dried seeds. Both yield potential and quality characteristics were significantly improved (Table 3). It has more grains/panicle (166.00 grains/panicle) than the original variety Giza 175 (155.00 grains/panicle). The yield potential is 45.00 grams/plant as compared to its parental variety 39.00 grams/plant. It has a bold grain and low amylose content (20.00%) as compared to its parent Giza 175 (26.00%). It could be concluded that Giza 175-M₁₃ are good mutant with high yield and good grain quality characters.

Giza 176- M₁₇:

Giza 176 is a japonica rice variety well known for Egyptian farmers as a breeding liner 2175-5-6 since 1991. Giza 176 was occupying a large area annually till it becomes susceptible to blast disease. Due to its susceptibility to blast disease, its long duration (145.00 days), this variety has had limited production. Through induced mutation and selection, mutant line Giza 176-M₁₇ has been developed by using 200 Gy gamma rays, (Table 3) that have improved traits in comparison to the original variety. This mutant was earlier than its original parent (105.00 days to heading), high yield potential (40.00 grams/plant), resistant to blast disease and good grain quality characters

GZ 1368 – M₁₃:

GZ 1368-S-5 is an indica / japonica rice line, resistant to salinity. The mutant GZ1368-M₁₃ was induced from this line (Table3) by exposing the seeds to 100 Gy gamma ray. This mutant has high yielding ability, and good grains quality characters.

Giza 181- M₁₁:

Is an indica type developed by exposing the seeds of a local rice variety Giza 181 to 300 Gy gamma rays. The parent variety has a long

slender grain and medium grain yield. The mutant Giza 181-M₁₁ has a medium grain and high grain yield as compared to its parent (Table 3).

It could be concluded that the results obtained by our mutation breeding program during the last seven (2000-2006) years, shows that this programme besides helping to generate an awareness of the potentiality of the mutation technique in creating valuable new variability, has resulted in some major achievements, as follows: (1) promising short culm mutants from all the studied varieties, (2) promising early maturity mutants from the late maturity varieties, (3) mutants possessing resistance to blast have been selected from the susceptible rice varieties such as Giza 171 and Giza 176, (4) amylose content in some of the studied varieties has been reduced and (5) promising high yielding mutants as compared to their parent varieties.

Phenotypic correlation coefficients:

The phenotypic correlation coefficients between grain yield and the seven agronomic characters for the three years and their combined data are presented in table (4). Obviously, grain yield was positive and highly significantly correlated with number of panicles per plant, number of grains per panicles, 100- grain weight, and milling percentage. On the other hand, negative and non- significant correlation coefficient values between grain yield and each of days to heading (-0.356) and plant height (-0.287) were obtained. This means that grain yield, might mainly depend on number of panicles per plant, number of grains per panicle, 100- grain weight for these promising mutants. The same results were obtained by *Basak and Ganguli, 1996* by using different materials. Furthermore, number of panicles per plant was highly significant correlated with each of number of grains per panicle (0.528) and 100- grain weight (0.492). Moreover, the correlation was highly significant between number of grains per panicle and 100-grain weight (0.882). Also, 100-grain weight was positive and highly significantly correlated with milling percentage (0.485)

Variation and interaction:

Results of combined analysis of the three years viz. 2005, 2006 and 2007 indicated the performance of highly significant differences between the mutant lines and their respective control, on one hand and among the lines on the other hand, for studied agronomic traits (Table 5), indicating overall wide differences among these populations and these mutant lines differ genetically .

As shown in Table (5), main effect of years was not significant for all the studied characters, indicating that there is no difference of lines performances regarding the studied characters in the three years, which may be due to the occurrence of stability and these lines did not affect significantly by environmental conditions. It could be concluded that these mutation lines could be grown in any season or year.

Mean squares of the interaction between the mutant lines and years were found to be not significant for all studied characters in the three years and their combined data, if the interaction of lines was highly significant than the interaction of lines with years, and , therefore, the most superior lines could be recommended.

Table (4): Estimates of phenotypic correlation coefficients between grain yield and the other agronomic characters over three years (2005, 2006 and 2007).

Characters	Days to heading (days)	Plant height (cm)	No.of panicles/plant	No.of grains/panicle	100-grain weight (g)	Milling (%)	Amylose content (%)
Grain yield /plant(g)	-0.356	-0.287	0.975**	0.668**	0.750**	0.375	0.218
Days to heading(days)		-0.180	-0.265	-0.0127	-0.058	-0.118	0.290
Plant height(cm)			-0.372	-0.198	0.016	0.220	0.123
No.of panicles/plant				0.528**	0.492**	0.238	0.115
No.of grains/panicle					0.882**	0.318	0.210
100-grain weight(g)						0.485*	0.220
Milling (%)							0.113

Table (5): Variance of combined analysis of the three years (2005, 2006 and 2007) for the studied traits in mutation lines induced from five rice varieties.

S.O.V	Days to heading (days)	Plant height (cm)	No.of panicles/ plant	No.of grains/ panicle	100-grain weight (g)	Grain yield/ plant (g)	Milling (%)	A.C (%)
Years	9.731	18.111	1.361	1.550	0.001	0.750	0.632	0.059
Replications	0.519	0.083	4.110	2.150	0.001	2.444	0.583	0.047
Lines	686.730**	1580.770**	53.447**	66.212*	10.420*	273.370**	61.300*	52.222*
Lx Y	3.863	4.945	4.521	2.510	0.001	5.226	1.119	0.174
Error	2.021	3.626	1.825	0.990	0.001	1.990	0.484	0.102

Genetic variability:

Estimates of genetic variance (GV), coefficient of genotypic variability (GCV %), heritability (Hb) and genetic advance (GS) for the studied characters are given in Table (6).

It was evident from the relative magnitude of genetic variance that the heritable (genetic) and non-heritable (non-genetic) component of variation, were ascertained with the help of some genetic parameters, like genetic coefficient of variation, heritability estimates, and genetic advance of selection.

Table (6) show high genetic coefficient of variation for plant height, grain yield/plant and milling (%) (the range was 26.28 to 13.48%). However, moderate values (the range 8.40 to 11.59%) were obtained for days to heading, number of panicles/plant, and amylose content and low estimates (the range 1.78 to 1.96) for 100- grain weight and number of grains/panicle. These results agreed with those obtained by *Abd-Allah et al., 2002*.

Using the genetic coefficient of variation alone, however, it is impossible to estimate the magnitude of heritable variation. The heritable portion of the variation could be found out with the help of heritability estimates and genetic gain under selection. High heritability values (Table 6) had been obtained for days to heading, plant height, grain yield/plant and amylose content (the range 0.91 to 0.98). However, moderate values were

estimated for number of panicles/plant, number of grains/panicle, 100-grain weight and milling (%) (the range 0.66 to 0.87).

Johnson et al. (1955) reported that heritability estimates along with the genetic gain, were more valuable than the former alone in predicting the effect of selection. If heritability was mainly owing to non-additive gene effect, the expected gain would be low and if there was additive gene effect, a high genetic advance might be expected. In the present study, it is very interesting to note that characters having heritability estimates gave almost high values of genetic co-efficient of variation such as milling (%). These results were similar to those obtained by *Singh et al. 1996*.

Many investigations reported that genetic coefficient of variation and high heritability, were not always associated with high genetic advance for a character. But to make effective selection, high heritability should be associated with high genetic advance. In the present study, the results showed that high genetic gain was associated with relatively high heritability value, in most of the studied characters. Therefore, individual plant selection for these characters should be effective and satisfactory for successful breeding purpose.

Table(6): Estimates of genetic variance (GV), genotypic coefficient of variability (GCV), heritability (Hb) and expected genetic advance (GS) for the studied characters in induced mutation lines over three years (combined data).

Characters	Genetic variance (GV)	Genotypic coefficient of variability (GCV)	Heritability in broad sense (Hb)	Expected genetic advance (GS)
Days to heading(days)	77.78	8.42	0.97	0.51
Plant height(cm)	192.16	13.48	0.98	28.16
No.of panicles/plant	5.17	9.53	0.76	4.02
No.of grains/panicle	10.60	1.96	0.87	6.20
100-grain weight (g)	0.02	1.78	0.66	0.07
Grain yield/ plant(g)	28.40	13.10	0.91	10.42
Milling (%)	3.22	26.82	0.66	2.99
Amylose content (%)	5.76	11.59	0.98	4.59

Molecular diversity assessment:

Among the nine tested SSR primers used, only one primer RM223 showed monomorphic pattern, while, all others showed considerable polymorphism (Table 7). The number of alleles detected per primer ranged from one in RM223 to three alleles in RM136. The rest of primers gave two alleles. The amount of polymorphism detected reflects the existence of considerable amount of molecular diversity among the tested entries. Based on the banding pattern, similarity percentage among each pair of the tested genotype was calculated and phylogenetic tree expressing the genetic relationships, among the entries was constructed, using DARWIN software package (Figure1). The five mutants, as well as, their original parents were clustered into three main groups. The first group (A) included only the

mutation derived from the Indica/ Japonica line GZ1368-S-5 (GZ1368- M₁₃). The second group (B) subdivided further to two subgroups B1 and B2. B1 group consisted of the two very late duration parents Giza 171 and the Indica rice variety Giza 181. The other sub-grouped (B2) had Giza 175, Giza 171- M₆ and Giza 175- M₁₃ genotypes. The Third group (C) consisted of Giza 181- M₁₁, Giza 176, Giza 176- M₁₇ and GZ1368-S-5. The clustering represent very clearly that the mutations are indeed different from their original parents. Except for GZ1368-S-5 and its derived mutation, the clustering was mostly co-leaner with pedigree and/or duration. For instance Giza 171 and Giza 181, though they have different genetic background, but both are very late maturing varieties. Giza 175 and its derived mutant clustered in the same subgroup B2. Also, Giza 176 and its derived mutant were clustered in one cluster C. As the mutations were selected over many years, the genetic variability between some parents and their derived mutations as in case of GZ1368- M₁₃ and its original parent were obtained. The scenario is much less in case of Giza 181 and its derived mutant Giza 181- M₁₁ where they clustered in two neighboring subgroups B and C. The results demonstrate the efficiency of mutation breeding on both morphological and molecular levels to develop new elite lines that possess desirable characteristics and differ significantly from their original parents. However, the lack of linear relationships between molecular data and genetic background might be due to the natural divergence between mutations and their original parents by using strong ionizing irradiation that causes point mutations, as well as, serious genetic changes that was reflected in the amount of character change between the parent and its selected mutant. The other scenario of this discrepancy in the case of GZ1368-S-5 and its mutant, could be explained as low number of specific primers, were used in the current study that may not exactly reflects the genetic background.

Table (7): Description of nine DNA markers, number of alleles and gene diversity for the studied rice genotypes.

DNA markers	No. of alleles
RM289	2
RM223	1
RM148	2
RM164	2
RM212	2
RM242	2
RM412	2
RM108	2
RM136	3

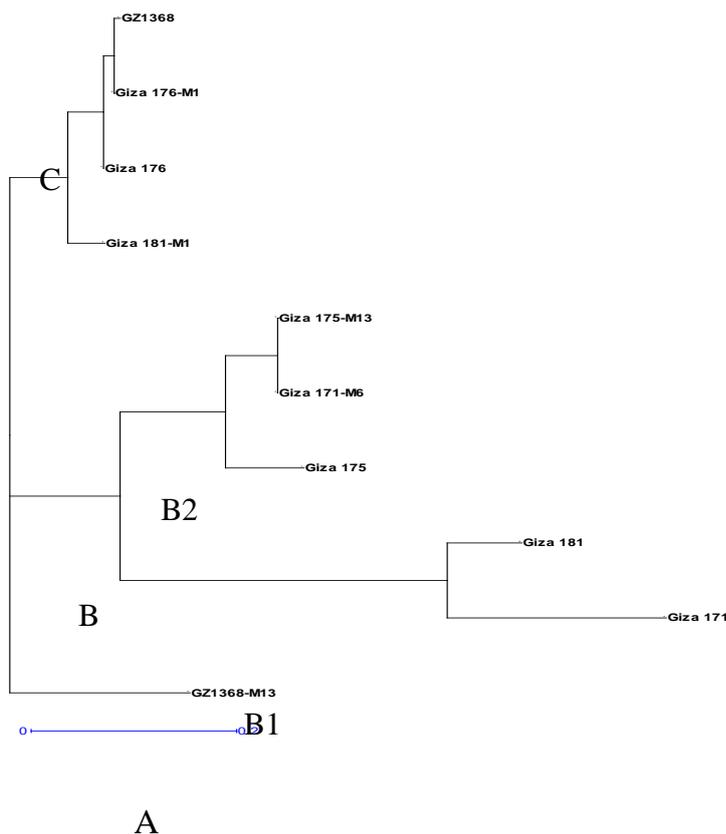


Fig. (1): UPGMA tree based on genetic distance for ten genotypes (five lines and their respective parents) with nine molecular markers.

REFERENCES

Abd Allah, A. A.; A. B. El-Abd and A.A. El- Hissewy (2002). Effect of gamma rays on some root characters and grain yield in M₂ generation of different rice varieties (*Oryza sativa* L.), Six Arab Conference on the Peaceful uses of Atomic Energy, Cairo, Egypt, 14- 18 Dec. vol. IV, 193-200.

Akbar, A. and B.Manzoor (2003). Radio sensitivity studies in basmati rice. Pak.J.Bot, 35(2):197-207.

Badawi, A.T. (2005). Sustainability of rice productivity in Egypt. Egypt J.Agric.Res. 83(5A).

- Basak, A. K. and P. K. Ganguli (1996). Variability and correlation studies on yield and yield components in induced plant type mutants of rice. *Indicant Agriculturist*, 40 (3) 171-181.
- Bartlett, M.S. (1937). Properties of sufficiency and statistical tests. *Proc.Roy.Soc.London, Series A*, 160: 268-282.
- Burton, G .W. (1951). Quantitative inheritance in Millet. *Agron. J.* 43, 409-417.
- Burton, G.W. (1952). Quantitative inheritance in grasses.*Proc.6 Int. Grassid Conger.1:277-*
- Chang, T.T.; G.C. Loresto and O.T.Aum (1974). Screening rice germplasm for drought resistance. *SABRAO. J.* 6(1): 9-16.
- Elayaraja, K.; M. Prakash; K. Saravanan; S. B. Kumar and J. Ganesan (2005). Studies on variability, heritability and genetic advance for certain quantitative characters in mutant population of rice (*Oryza sativa* L.) crop Research-Hisar. (1): 134-137.
- Gomaa, M. E.; A. A. El-Hissewy; A. B.Khattab and A. A. Abd-Allah (1995). Improvement of yield and some related characters of rice (*Oryza sativa* L.), BY Irradiation. *Menofiya Journal of Agricultural of Research*, Vol. 20, No. 2: 395-408.
- Johanson, H.W.; H.F. Robinson and R.E. Comstock (1955). Estimates of genetic and environmental variability in soybean.*Agron. J.*47:314-318.
- Khin, T. (2006). Rice mutation breeding for varietals improvement in Myanmar. *Plant mutation reports* 1, (1); 34-36.
- Lush, J.L. (1940). Intra-sire correlation and regression of off-spring on dams as a method of estimating heritability of characters. *Proc Amer Soc Animal Production* 33, 293-301.
- Mohamed, O.; B. N. Mohd; 1. Alias; S. Azlan; H. Abdul-Rahim; M. Z. Abdullah; O. Othman; K. Hadzim; A. Saad; H. I. Habibuddin and F. Golam (2006). Development of Improved Rice varieties through use of induced mutation in Malaysia. *Plant mutation Report.* 1, (1):27-33.
- Perrier, X. and J.P.Jacquemoud (2006). Genetic diversity of cultivated tropical plants. *Enfield, Science Publishers. Montpellier*: pp 43 – 76.
- Soomro, B.A.; A.M. Naqvi; M.H. Bughio; H.R. and M.S.Bughio (2006). Sustainable enhancement of rice production through the use of mutation breeding. *Plant mutation report*, Vol.1: 13-17.
- Singh, S.; R. P. S. Dhaka and S.S. Malik (1996). Heritability and genetic Advance in irradiated population of rice (*Oryza sativa* L.), *Crop Research* 11 (2): 184-188.
- Wu, J.L.; C.Wu; C.Lei; M.Baraoidan; A.Bordeos; M.R.S.Madamba; M.Ramos; R.Mauleon; A. Portugal; V.J.Ulat; R.Bruskiewich (2005). Chemical and irradiation induced mutants of indica rice IR64 for forward and reverse genetics. *Plant Molecular Biology* 59:85-97.
- Zhu, X.D.; H.Q.Chen and J.X.Shan (2006). Nuclear techniques for rice improvement and mutant induction in China National Rice Research Institute. *Plant mutation Report*, Vol.1: 25-28.

التقييم المحصولي والجزئي لسلاسل طفرية مستحدثة من الأرز
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أجريت هذه الدراسة فى مزرعة مركز البحوث و التدريب فى الأرز -سحا- كفر الشيخ - مصر خلال المواسم الزراعية من ٢٠٠٠ وحتى ٢٠٠٧ . أستخدم فى هذه الدراسة خمسة أصناف أرز مصرية بعضها يتبع الطراز اليابانى والبعض يتبع الطراز الهندى وهى : جيزة ١٧٧، جيزة ١٧٥ ،جيزة ١٧٦ ،جيزة ١٨١ والسلاسل جى زد ١٣٦٨ . كانت تلك الأصناف تمتلك صفات محصولية ممتازة فى الفترة السابقة، مثل الإنتاجية العالية من محصول الحبوب ، المقاومة للأمراض والحشرات و صفات جوده ممتازة، ولكن حاليا تفتقد تلك الأصناف إلى بعض الصفات مثل المقاومة لمرض اللفحة كما فى بعض الأصناف مثل جيزة ١٧١ وجيزة ١٧٦ ، و بعض صفات جودة الحبوب كما فى جيزة ١٧٥ والسلاسل جى زد ١٣٦٨ بالإضافة الى أن تلك الأصناف متأخرة النضج . ولقد تم معاملة بذور هذه الأصناف بجرعات مختلفة من أشعة جاما وهى ٤٠٠، ٣٠٠، ٢٠٠، ١٠٠، ٥٠٠ جى راد للحصول على نباتات الجيل الأول الإشعاعى عام ٢٠٠٠ وتم إنتخاب ٧٧ سلالة طفرية من نباتات الجيل الخامس الإشعاعى من الأصناف الخمسة ، حيث تم تقييم تلك السلالات بعد ذلك ، لمدة ثلاثة أجيال ، وهى الجيل السادس والجيل السابع والجيل الثامن الإشعاعى (٢٠٠٥-٢٠٠٦-٢٠٠٧) فى ثلاثة مكررات، باستخدام تصميم القطاعات الكاملة العشوائية.

تم إنتخاب خمسة سلالات طفرية من الأصناف الخمسة، تمتلك صفات خضرية و صفات محصولية و صفات جودة ممتازة، وتعتبر تلك السلالات سلالات مبشرة لتفوقها على الأباء (الأصناف) الناتجة منها ، حيث أن تلك السلالات مقاومة للرقاد، مقاومة لمرض اللفحة ، مبكرة النضج بالإضافة الى الإنتاجية العالية من محصول الحبوب .

أوضحت النتائج أن هناك تباينات وراثية بين السلالات الطفرية المستحدثة و الأباء الناتجة منها، و لقد تم تأكيد تلك الاختلافات الوراثة الموجودة بين الأباء (الأصناف) والسلالات الطفرية، عن طريق تكنيك الـ P C R واستخدام الدلائل الجزيئية، و بذلك فان تلك السلالات تعتبر طفرات حقيقية ، ويمكن الاستفادة من بعضها كأباء معطية فى برنامج تربية الأرز لتحسين بعض الصفات الهامة بالتهجين مع الأصناف الأخرى ، وبعضها يمكن التوصية به كصنف جديد من الأرز، يمتلك صفات متميزة مثل صفة التبيكر و صفة المقاومة لمرض اللفحة والإنتاجية العالية من محصول الحبوب مثل السلالة الطفرية (Giza 175-M₁₃) جيزة ١٧٥-ام ١٣ .

Table (3): Performance of the promising mutants selected in M5 generation and evaluated in three successive generations (M6, M7 and M8) of the studied traits.

Designation	DH (days)	Plant height (cm)	No. of panicles/plant	No. of grains/panicle	Grain yield/plant (g)	100-grain weight (g)	Milling (%)	Blast reaction	A.C (%)
Giza 171-M ₆	107.00	95.00	28.00	165.00	35.00	2.60	72.00	R	19.00
Giza 171	12.00	135.00	22.00	135.00	32.00	2.50	72.00	S	19.50
Giza 175-M ₁₃	100.00	102.00	28.00	166.00	45.00	2.52	70.00	R	20.00
Giza 175	105.00	95.00	24.00	155.00	39.00	2.45	69.00	R	26.00
Giza176-M ₁₇	105.00	100.00	29.00	187.00	40.00	2.62	70.00	R	19.00
Giza 176	110.00	102.00	25.00	145.00	36.00	2.56	68.00	S	19.00
Giza181-M ₁₁	99.00	100.00	28.00	165.00	38.00	2.55	70.00	R	20.00
Giza 181	100.00	95.00	20.00	150.00	36.00	2.50	69.00	R	19.50
GZ1368 -M ₁₃	109.00	98.00	23.00	156.00	43.00	2.58	71.00	R	19.00
GZ1368-S-5-4	101.00	98.00	21.00	128.00	35.00	2.50	69.00	R	24.00
LSD 0.05	3.89	4.32	2.50	7.60	3.40	0.03	0.08	-	1.50