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INDUCTION OF MUTANTS IN DURUM WHEAT (*TRITICUM DURUM* DESF CV. SAMRA) USING GAMMA IRRADIATION

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Abstract

A mutation breeding program was initiated in 2008 emphasizing the main constraints for sustainable production of durum wheat in Saudi Arabia. The aim of the program was to develop moderate or high yielding semi-dwarf/lodging tolerant, early maturing mutants with drought and disease tolerance from a local durum wheat cultivar (*Triticum durum* Desf. cv. Samra) which has the main defects of longer crop duration, lodging habit and low grain yield. Dry seeds of Samra were subjected to 150 and 200Gy doses of gamma irradiation and each treatment consisted of 2500 seeds. Irradiated seeds were grown as M_1 population along with parental variety as control at Almuzahmiah Research Station of Riyadh, Saudi Arabia. Decrease in germination (%) and survival rate (%) of plants was observed. A wide variation in days to flowering and plant height was found in the M_1 populations. Three seeds from each spike per plant of M_1 plants were collected, bulked dose wise and grown separately as M_2 in 2009 growing season. From these M_2, 17 desirable putative mutant plants which varied significantly with the mother were visually selected. These putative mutants were found to be semi-dwarf and early maturing in nature with other improved agronomic traits including lodging reaction and grain yield. The selected plants, when grown in progeny lines as M_3 in 2010, more or less maintained their superiority over the mother for many traits. Most of the mutant lines showed homogeneity for most of characters studied. Eleven of these 17 lines were found to be promising in respect of days to flower, plant height (for semi-dwarf) and other traits including grain yield.

Introduction

Mutation breeding is an significant breeding tool which has been used successfully in several crops for breeding agronomically important traits (Maluszynski *et al*., 1995). Therefore, mutagenesis is applied to amend few blemishes in a cultivar that has several agronomic traits preferable by farmer. In wheat breeding, Sakin *et al*., (2004, 2005) obtained superior mutant types having better agronomic values in term of yield and yield components. KuŞaksiz & Dere (2010) have succeeded to obtain certain mutant populations where their heritability and phenotypic (standard deviation values and genetic gains) were higher than those of the control populations. This doesn't mean other methods are not sufficient and generally practiced, where Jan *et al.,* (2011) reported that nitrogen fertilization increased 20% grain yield compared to control. Also, tissue culture was used in developing wheat cultivars (Mahmood *et al.*, 2012)

Gamma rays in particular, is an important physical mutagen which is well known with their effects on the plant growth and development by inducing morphological, cytological and physiological changes in cells and tissues (Thapa, 2004; Borzouei *et al*., 2010; Shah *et al.*, 2012).

The global demand and increased need of durum wheat worldwide have been lead to extensive research and development to this crop, where mutagenesis and hybridization are the most leading methods to be followed. Lodging susceptibility and straw weakness are accounted for substantially lower yields in durum wheat compared to bread wheat (Donini & Sonnino, 1998; Arain *et al*., 2001). Breeders have been trying to improve durum wheat cultivars in order to compete with the bread wheat cultivars in yielding ability (Sakin *et al*., 2004, 2005; Cagirgan, 2009; Dahot *et al*., 2010). Sakin *et al.,* (2004, 2005) succeeded to select mutant lines with increased and better agronomic value in durum wheat.

Abiotic and biotic factors are the main constraints in sustainable crop production in Saudi Arabia. Therefore, the newly mutation breeding program of the country emphasizing on developing moderate and /or high yielding mutants of wheat companied early maturation, drought and disease tolerance. In this regards, utilization the landraces from the local conditions in this program for breeding can be an advantage step due to the adaptation that has been developed to specific environments (Maxted *et al*., 1997). This may help in building in-house resistance to various biotic and abiotic stresses apart of changing agricultural practices and increasing yield (Maxted *et al*., 1997). Mahmood *et al*., (2011) has evaluated fifteen Pakistani wheat landraces based on Random Amplified Polymorphism DNA (RAPD) markers to be utilized further in breeding programs. On the other hand, Xiyong *et al.,* (2012) has genetically evaluated more than 200 wheat cultivars and advanced lines from Huanghuai Wheat Region of China to identify 14 agronomic traits and 7 quality traits, as well as the evolution and utilization of high molecular weight glutenin subunits (HMW-GS) and low molecular weight-glutenin subunits (LMW-GS). This concludes that local landraces outperform the best variety/cultivar in a harsh environment. As a part of this fact, farmers in Saudi Arabia grow different types of durum wheat landraces. Among them Samra cultivar that has been cultivated for a long time although it has certain defects. The main defect facing its cultivation is susceptibility to lodging which resulted in yield reduction at time of harvesting in addition to that it has a low yield.

Keeping in view all these objectives, a mutation breeding program was initiated at Atomic Energy Research Institute (AERI), King Abdulaziz City for Science and Technology (KACST) in 2008 using gamma irradiation to develop mutants having moderate/high yielding ability with short-statured/stiff-straw (lodging tolerant), early maturity and improved reaction to biotic and abiotic stresses. The present studies confirmed the importance of mutation breeding in improving the various complicated metric traits.

Materials and Methods

Dried seeds (approx. 12% moisture content) of Samra cultivar of durum wheat (*Triticum durum* Desf. cv. Samra) were subjected to 150 and 200 Gy gamma irradiation from a ^{60}Co (Cobalt-60) gamma irradiator (Gammacell Research Irradiator, Model: Excell 220, MDS, Nordion, Canada) installed at Atomic Energy Research Institute (AERI), King Abdulaziz City for Science and Technology (KACST), Riyadh, Saudi Arabia. Each treatment was comprised of 2500 wheat seeds. Irradiated seeds were grown in rows with inter and intra-row spacing of 50 and 30 cm, respectively, to raise the M_1 population. The untreated seeds (0 Gy) of mother cultivar (parental line/variety) were also planted after every five rows as control for comparison with the M_1 population. The planting was implemented in 2008 growing season in Almuzahmiah Research Station, Riyadh, Saudi Arabia. Soil is characterized by silty/clay-loam containing low organic matters. The station is lying 645 m above the sea level with 14-24°C temperature during wheat growing season (November-April) and the average rainfall is about 100 mm in wet season (December-February). Normal cultural practices including fertilization were done whenever it is necessary. Data on seed germination and surviving plants were recorded considering whole plots of M_1 population. Data on days to flower and plant height were taken from 50 randomly selected plants of each treatment representing more or less all types of morphological plants. Data is presented in Table 1. M_2 seeds were harvested as 3 seeds from each spike of each M_1 plant of both the treatments at maturity. These M_2 seeds were bulked separately for each treatment. These seeds were planted in rows with 2.5 cm spacing between plants and 25 cm among rows in year 2009. The mother variety was planted as a control after each five rows. The size of M_2 population was around 10,000 plants for the two doses 150 and 200 Gy. Selections were made in M_2 population on the basis of some specific agronomic characters viz., earliness, height, lodging reaction and yield etc. A total of 17 putative mutants (8 from 150 Gy and 9 from 200 Gy) were selected in M_2 population. Data on some agronomic traits likewise grain yield and lodging resistance were recorded from selected plants. Single spike selection was made in M_2 and the population was harvested and the seeds of each plant were bulked separately. The selected M_2 seeds were again planted separately as progeny lines in 5 m long rows in 2010 to raise M_3 population. After each 5 rows the mother cultivar was included as control. In M_3 population, observations were recorded on days to flower**,** plant height**,** spikes/plant, spike length, kernels/spike**,** 1000-grain weight**,** yield/plant and reaction to lodging from 5 randomly selected plants of each progeny lines**.** Seed coat colors were also recorded. Data of M_2 and M_3 were analyzed statistically and the mean values of different parameters were adjudged by Duncan's New Multiple Range Test (DNMRT) (Steel & Torrie, 1980). The results are summarized (Tables 1-4).

Table 1. Days to flower and plant height performance of M_1 populations compared to mother cultivar Samra (Untreated) in 2008.

Treatment (Gy)	No. of seeds	Days to flower			Plant height (cm)		
		Range	Mean	± SE	Range	Mean	± SE
200	2500	36-66	62.8	1.32	41-60	51.6	1.39
150	2500	42-71	65.7	1.52	33-51	46.8	1.04
0 (Untreated)	2500	49-52	50.6	0.22	49-50	49.6	0.10

Results and Discussion

M_1 generation: The results on germination of seeds, survival rate of plants at maturity derived from treated and untreated seeds are graphically depicted (Fig. 1) while days to flower and plant height are tabulated (Table 1). Seed germination and survival of plants decreased to a considerable level as compared to untreated seeds of mother cultivar Samra (Fig. 1). In case of 150 Gy, it was 72 and 67% and in case of 200 Gy it was 69 and 59%, respectively, for both the measured traits while it was 95 and 96% in the untreated (control) population. Adverse effects of gamma irradiation on germination (%) and survival (%) of plants is a common feature in most of the crops and it has been registered by many researchers in wheat (Arain *et al*., 2001; Irfaq & Nawab, 2001; Sial *et al*., 2009; Borzouei *et al.*, 2010). In case of days to flower, it was found that the mean and range values were larger than the mother cultivar. The larger range values (42-71 days for 150 Gy and 36-66 days for 200 Gy, respectively) suggested that the character have wider variations including early as well as late maturing plants in M_1 generation. In case of plant height, 150 Gy showed lower mean value and 200 Gy showed higher mean value but the ranges (33-51 cm for 150 Gy and 41-60 cm for 200 Gy) were much larger than to those of mother. This also indicated that the population has wider variation for this trait. In the present work, both early and late maturing plants were identified. Early maturing plants were reported in M_1 populations of wheat earlier by many researchers (Irfaq & Nawab, 2003; Khan *et al*., 2003; Qasim & Ahmad, 2004; Singh & Balyan, 2009). On the other hand, different flowering time wheat mutants have also been reported by others (Zhu *et al*., 1991; Díaz *et al*., 2012). Standard error (±SE) values of the treated populations were also found very high for these two traits as compared to those of the mother (Table 1). Larger standard error values with greater ranges suggested that the treated populations were heterogeneous and have larger variations for both traits due to induction of mutation. The larger variation in a population creates wider scope to a breeder for selection of desirable genotypes.

Table 2. Performance of 17 selected mutants of Samra cultivar in M_2, 2009. Mean, range and standard error (± SE) values are included.

Mutant lines/ Genotypes	Treatment (Gy)	Days to flower	Plant height (cm)	No. of spikes/ plant	Spike length (cm)	No. of seeds/ spike	1000- seed wt. (gm)	Yield/ plant (gm)	Reaction to lodging*	Seeds color
Muz-008-S-4	150	45	45	9	6.5	23	35	51	3	Gray
Muz-008-S-7	150	52	62	8	6.5	22	39	44	4	Creamy
Muz-008-S-10	150	53	50	8	6.5	23	35	46	3	Creamy
Muz-008-S-12	150	59	53	7	7.0	28	38	41	3	Creamy
Muz-008-S-13	150	57	64	9	7.2	25	38	50	5	Creamy
Muz-008-S-21	150	47	42	10	6.1	20	34	60	4	Creamy
Muz-008-S-23	150	55	40	10	6.6	28	33	60	3	Creamy
Muz-008-S-31	150	50	55	10	6.5	32	34	60	4	Creamy
Muz-008-S-6	200	53	50	8	6.4	21	35	44	5	Gray
Muz-008-S-9	200	55	45	6	7.0	26	37	36	4	Creamy
Muz-008-S-14	200	51	46	9	6.5	24	37	50	4	Creamy
Muz-008-S-15	200	49	63	9	7.5	22	35	45	5	Creamy
Muz-008-S-16	200	50	45	9	6.5	22	34	58	6	Gray
Muz-008-S-18	200	54	53	6	6.5	20	37	35	3	Gray
Muz-008-S-26	200	50	55	10	6.9	28	39	60	4	Creamy
Muz-008-S-29	200	49	55	10	7.4	33	30	62	4	Creamy
Muz-008-S-33	200	45	60	8	7.0	25	25	50	4	Creamy
Samra (Parent)	0	61	53	8	6.0	24	31	47	8	Creamy
Range	-	45-60	40-63	6.0-10.0	6.0-7.5	20-33	25-39	35-62	-	-
Mean	-	51.9	52.0	8.56	6.7	24.8	34.8	49.9	-	-
±SE	-	1.04	1.71	0.71	0.10	0.89	0.83	2.01	-	-

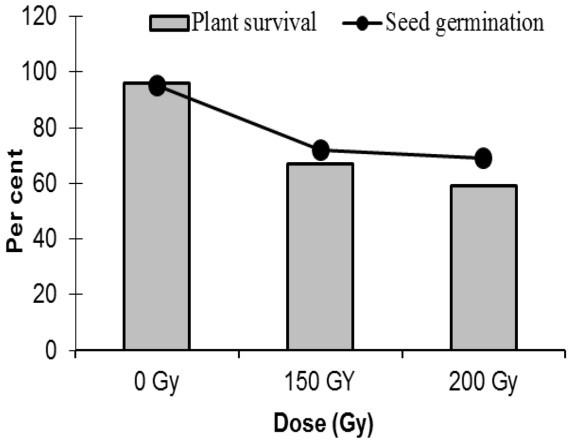

Fig. 1. Effects of different doses of gamma rays on seed germination and plant survival at maturity in Samra cultivar.

M_2 Generation: M_2 populations grown as dose-wise from separate bulk-seeds harvested in M_1, 17 desirable mutants (8 from 150 Gy and 9 from 200 Gy) were selected and their data are summarized (Table 2). It was observed that the plants varied in their performances for each trait as indicated by larger ranges and standard error values. In case of days to flower, it was found that all the mutants exhibited lower values i.e., all the mutants have become earlier maturing than the mother cultivar Samra while plant height exhibited changes in both directions, some mutants have greater height and the others have lower height than the mother. Changes in both directions i.e., increase as well as decrease in the magnitude of other characters including yield per plant were also found with the exception of spike length. Mutants showed higher or lower values for number of spikes per plant, number of seeds per spike, 1000-grain weight and yield per plant compared to mother. Spike length of the mutants was found to increase. Improvement was also noticed in the reaction to lodging and all the mutants showed higher resistance (3 to 5 scales) compared to the mother (Table 2). Four of the mutants were found to have changed seed coat color i.e., from creamy to gray. Standard error values of the agronomic character also suggested that the mutants varied significantly among themselves and also with the mother in their performances (Table 2). These plants were grown in progeny lines as M_3.

M_3 Generation: The results from analysis of variance has been summarized and tabulated (Table 3) showed that the source genotype was highly significant for all the traits indicating the existence of high variance among the entries i.e., the mutant lines performed differently with each other and also with the mother in genotypic nature for all the traits. The source sample was non-significant indicating that the progenies and the mother cultivar did not have any significant variation among themselves and these entries were mostly homogeneous in nature. The results of DMNRT on the mean values of different characters (Tables 4a & 4b) confirmed the existence of significant variation between the mutant lines and the mother cultivar for all the traits. It suggested that mean performance of the mutant progenies has significant differences within a trait. In case of days to flower and plant height, it was found that all the mutant lines were different from the mother. The observed lower values of the mutant lines for these two traits suggested that all the lines have become significantly dwarf having early maturity compared to mother. The mutant lines were about 10-20 days earlier maturing and flowered within 59 - 68 days while the mother cultivar took 78 days to flower. Mean values for plant height of the mutant lines ranged from 39.8 cm to 80.0 cm. Among the mutant lines, Muz-008-S-21-3 registered the highest plant height, which was statistically similar to the mother (84.0 cm). The most dwarf line was Muz-008-S-16-2. Singh and Balyan (2009) studied M_1 to M_3 progeny lines of wheat and isolated a number of semi-dwarf mutant lines with other improved traits.

In case of other traits, except spike length, some of the mutant lines exhibited higher mean values and some lower compared to mother cultivar (Tables 4a & 4b). But higher or lower magnitudes of the mean values were not fixed to any particular mutant for all characters and it varied from mutant to another. The longest spike length (7.6 cm) was produced by Muz-008-S-6-3; highest number of spikes per plant (8.4) by Muz-008-S-13-2; seeds per spike (43.4) by Muz-008-S-12-3 and Muz-008-S-9-2, and 1000-seed weight (50.0 gm) by Muz-008-S-15-4. Singh & Balyan (2009) while working with mutant generations detected some mutants producing higher number of tillers/plant and spikelets/spike, and higher 1000-grain weight compared to control in M_3 generation. Several authors (Khamkar, 1989; Jamil & Khan, 2002) have registered similar results and they found radiation effects have increased the performance of different agronomic traits in wheat. On the contrary, Muhammad *et al.,* (1985) and Irfaq & Nawab (2003) found decrease in the number of spikelets/spike and grains/spike in M_2 and M_3 populations of wheat by irradiation. Eleven of the mutant lines produced significantly higher seed yield and the remaining 6 lines produced lower than the mother cultivar. The highest seed yield (58.0 gm) was found to produce by Muz-008-S-13-2 followed by Muz-008-S-31-2 (49.2 gm) Muz-008-S-21-3 (46.4 gm). Singh & Balyan (2009) worked with M_3 lines of wheat and isolated one line which produced higher yield compared to its mother. Carvahlo (1977) also found increase in yield with low doses of gamma ray irradiation. Higher yield produced by mutants of wheat are also in agreement with the works of Jamil and Khan (2002) and Singh & Balyan (2009). However, Khamkar (1989) and Irfaq & Nawab (2009) opposed the results and they observed decrease in grain yield of wheat due to effect of radiation. However, the results of this experiment registered both increase and decrease in grain yield in the mutant generations of M_2 and M_3. Reaction to lodging was also found to be improved from susceptible (ratings of 7 in mother cultivar) to resistant or moderate resistant (ratings of 2-5 in the mutant lines). Similar trend was also noticed in M_2, where the mother cultivar showed rating of 8 in lodging standard scale while the mutant lines ratings were in between 3-6. Four of the 17 mutant lines showed changes in seed coat colour from creamy in mother cultivar to gray in the mutants, which was also observed in M_2. Changes in seed coat colour and size have been registered by Singh & Balyan in 2009. Range and standard error values, presented (Tables 4a & 4b) showed a few of the mutant lines still have some minor variations as indicated by their corresponding values for the respective characters. Such as 5 lines for days to flower, 3 for spike length, 3 for seeds per spike and seven for 1000-grain weight were found to have larger variations.

Table 4a. Performance (mean, range and standard error (±SE) values) of 17 mutant lines compared to the mother cultivar Samra in 2010.

Mutant lines/ Genotypes	Gy	Days to flower			Plant height (cm)			No. of spikes per plant			Spike length (cm)		
		Range	Mean	± SE	Range	Mean	± SE	Range	Mean	±SE	Range	Mean	± SE
Muz-008-S-4-3	150	66-70	68.0 b	0.87	60-69	65.2 d-e	1.46	4.0-6.0	5.0 de	0.32	6.5-7.4	7.1 a-c	0.17
Muz-008-S-7-3	150	60-65	62.0 c-e	1.04	48-52	50.0 i	0.63	6.0-8.0	7.0 b	0.32	5.0-6.0	5.5 d	0.15
Muz-008-S-10-1	150	57-67	61.8 c-e	1.83	57-63	59.6 gh	1.08	6.0-7.0	6.4 bc	0.24	5.0-6.5	5.9 cd	0.29
Muz-008-S-12-3	150	59-70	65.6 b-d	2.16	62-69	64.6 de	1.33	4.0-6.0	5.0 de	0.32	5.0-8.0	6.4 a-d	0.49
Muz-008-S-13-2	150	60-68	65.0 b-d	1.20	74-81	78.0 bc	1.30	8.0-9.0	8.4 a	0.24	6.0-7.5	6.9 a-c	0.24
Muz-008-S-21-3	150	60-65	62.6 b-e	0.87	76-83	80.0 ab	1.22	6.0-8.0	7.2 ab	0.37	7.0-8.0	7.4 ab	0.19
Muz-008-S-23-3	150	59-63	61.8 c-e	0.80	46-52	48.8 ij	1.01	6.0-8.0	6.6 bc	0.40	6.0-7.0	6.5 a-d	0.19
Muz-008-S-31-2	150	61-65	62.8 b-e	0.66	72-78	75.0 c	1.00	7.0-9.0	7.6 ab	0.40	5.0-6.5	6.0 cd	0.27
Muz-008-S-6-3	200	58-70	64.6 b-e	2.23	52-58	55.6 h	1.17	5.0-7.0	6.2 b-d	0.37	6.9-8.0	7.6 a	0.21
Muz-008-S-9-2	200	59-66	62.6 b-e	1.17	40-46	42.6 j	1.08	4.0-6.0	5.0 de	0.32	4.5-7.0	5.9 cd	0.43
Muz-008-S-14-4	200	58-66	63.2 b-e	1.78	41-48	45.0 j	1.22	4.0-6.0	5.0 de	0.45	5.0-7.0	6.3 b-d	0.37
Muz-008-S-15-4	200	58-62	60.8 c-e	0.80	66-71	68.8 d	0.86	4.0-5.0	4.4 e	0.25	5.0-6.5	5.8 cd	0.26
Muz-008-S-16-2	200	58-70	66.6 bc	2.27	39-41	39.8 k	0.37	6.0-8.0	7.0 b	0.32	6.0-7.0	6.6 a-d	0.19
Muz-008-S-18-2	200	64-66	64.6 b-e	0.34	60-64	62.4 e-g	0.81	4.0-6.0	4.8 de	0.37	5.0-8.0	6.8 a-c	0.52
Muz-008-S-26-2	200	58-62	60.0 de	0.63	58-63	60.2 fg	1.02	5.0-7.0	5.4 c-e	0.25	6.0-7.5	6.9 a-c	0.24
Muz-008-S-29-2	200	60-60	60.0 de	0.00	72-80	75.6 bc	1.50	6.0-7.0	6.8 b	0.20	6.0-7.5	6.7 a-d	0.24
Muz-008-S-33-2	200	57-60	59.0 e	0.55	63-67	64.8 de	0.66	5.0-7.0	6.2 b-d	0.37	6.0-7.0	6.4 b-d	0.26
Samra (Mother)	0	78-81	79.2 a	0.58	82-86	84.0 a	0.79	5.0-6.0	5.4 c-e	0.24	6.5-7.0	6.7 a-d	0.12

*Mean values with same letters in a column do not differ at 1% level of significance

Table 4b. Performance (Mean, range and standard error (±SE) values) of 17 mutant lines compared to the mother cultivar Samra in 2010.

Mutant lines/ Genotypes	Gy	Seeds per spike (no)			1000-grain weight (gm)			Yield per plant (gm)			Reaction to lodging*	Seed color
		Range	Mean	± SE	Range	Mean	± SE	Range	Mean	± SE		
Muz-008-S-4-3	150	20-25	22.6 e-g	0.81	32-41	36.6 d-f	1.72	29-36	30.6 g	1.36	3	Gray
Muz-008-S-7-3	150	22-26	23.6 d-g	0.81	29-36	33.6 f-h	1.50	43-49	45.6 b-d	1.08	4	Creamy
Muz-008-S-10-1	150	33-39	36.0 b	1.10	35-38	36.0 d-f	0.63	37-42	40.0 e	0.94	4	Creamy
Muz-008-S-12-3	150	41-47	43.4 a	1.03	27-34	30.8 gh	1.28	28-34	29.8 gh	1.11	5	Creamy
Muz-008-S-13-2	150	18-22	20.0 fg	0.71	34-38	35.6 d-f	0.67	55-61	58.0 a	1.00	4	Creamy
Muz-008-S-21-3	150	10-17	12.8 h	1.20	37-41	38.8 de	0.66	44-50	46.4 bc	0.95	5	Creamy
Muz-008-S-23-3	150	35-44	39.6 ab	1.81	27-35	30.0 h	1.41	39-43	41.2 de	0.73	3	Creamy
Muz-008-S-31-2	150	15-23	19.8 fg	1.39	43-48	46.0 ab	0.95	46-53	49.2 b	1.24	3	Creamy
Muz-008-S-6-3	200	23-29	24.8 c-f	1.07	32-36	34.6 e-g	0.67	37-43	39.6 e	1.21	2	Gray
Muz-008-S-9-2	200	40-48	43.4 a	1.33	34-36	34.8 e-g	0.49	28-31	29.6 gh	0.51	2	Creamy
Muz-008-S-14-4	200	18-21	19.6 g	0.51	31-34	33.0 f-h	0.55	27-35	30.6 g	1.33	4	Creamy
Muz-008-S-15-4	200	26-30	27.8 cd	0.73	48-53	50.0 a	0.95	24-29	25.6 h	0.93	5	Creamy
Muz-008-S-16-2	200	33-40	37.0 b	1.22	26-33	30.0 h	1.14	44-48	45.6 b-d	0.81	3	Gray
Muz-008-S-18-2	200	30-42	37.8 b	2.11	39-48	43.6 bc	1.57	25-33	28.6 gh	1.33	5	Gray
Muz-008-S-26-2	200	27-33	29.4 c	1.12	36-44	39.6 cd	1.44	37-44	41.0 e	1.18	5	Creamy
Muz-008-S-29-2	200	25-30	26.6 c-e	0.93	33-38	34.8 e-g	0.92	41-46	44.0 c-e	0.95	3	Creamy
Muz-008-S-33-2	200	35-43	39.2 ab	1.69	36-43	39.8 cd	1.28	37-44	40.0 e	1.14	3	Creamy
Samra (Mother)	0	36-39	37.6 b	0.51	35-37	36.2 d-f	0.44	32-38	35.0 f	1.14	7	Creamy

Mean values with same letters in a column do not differ significantly at 1% level
*9 Standard Scale of Rating

Table 3. Results analysis of variance (only mean values are included).

Source of variation	Df	Mean squares						
		Days to flower	Plant height	Spikes per plant	Spike length	Seeds per spike	1000-grain weight	Yield per plant
Sample	4	5.73	5.36	0.16	0.41	2.00	1.43	1.79
Genotype	17	101.42***	864.71***	6.36***	1.62***	431.61***	145.39***	370.83***
Error	68	8.72	6.42	0.56	0.42	7.41	6.21	6.03

*** Significant at 0.1% level

More selections in M_4 will improve traits of the respective mutant lines. However, selection was done with respect to and emphasizing on days to flower, plant height and seed yield per plant. The mutant lines, which were semi-dwarf and early maturing having other improved characters with considerable high yield compared to mother cultivar were selected. Thus, 11 mutant lines out of 17 were selected for evaluation in the next generation as M_4 population for future breeding program.

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