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Research Article
Induced Mutations in Wheat (*Triticum aestivum* L.) and Improved Grain Yield by Modifying Spike Length

A.S. Anter

Department of Field Crops Research, Biological and Agricultural Division, National Research Center, Egypt

Abstract

Background and Objective: Wheat bread is classified as the main food crop in Egypt. However, Egypt is the largest importer of wheat globally. The objectives of this study were to increase genetic variation of bread wheat and to identify among mutant lines those with high-yielding under marginal lands conditions. **Materials and Methods:** Grains of five bread wheat varieties were irradiation by three doses of gamma rays (100,200 and 300 Gy). Fifteen M_0 combination seeds along with the mother varieties arranged in randomize complete blocks design with three replicates at Agricultural Production and Research Station of National Research Centre, Egypt, during four winter seasons (2016-2019). The phenotype selection was applied to the M_3 generation based on a longer spike length, as a result, twenty-three mutant lines have been created, which considered as putative mutants. To confirm these mutant lines, five distinct mutant lines out 23 mutant lines were reevaluated based on grains yield components in M_4 generation. **Results:** Analysis of variance showed significant variation (p<0.05) among resource mutant lines for quantitative phenotypic traits in mutated generations. Compared to the mother varieties, five mutant lines presented highly outstanding quantitative phenotypic traits. All characters recorded high heritability accompanied with moderate genetic advance, therefore, the possibility of improving wheat grain yield through phenotypic selection. All characters, except plant height, evidenced to be priority traits in selection for higher grain yield due to their powerful direct association with grain yield. **Conclusion:** These mutant lines provide exciting new valuable resources to investigate and improvement of complex traits in bread wheat.

Key words: Bread wheat, new mutation resources, spike length, sustainability development, genetic diversity

Corresponding Author: A.S. Anter, Department of Field Crops Research, Biological and Agricultural Division, National Research Center, Egypt

Competing Interest: The author has declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Bread wheat (*Triticum aestivum* L.) is the primary crop in Egypt. The rising Egyptians population have resulted an increasing demand for bread wheat in the following years. Wheat grain is used as food for humans and the straw is used as fodder for animals[1]. However, Egypt is the largest wheat importer in the world due to the limited amount of cultivated land and there is little use of modern farming technology[2]. Therefore, increasing local production through improving grain yield per unit area becomes a matter of importance.

Plant breeding techniques, mutagenesis, biotechnology genetic engineering and molecular breeding have played a pivotal role in exploiting available germplasm resources to create variability and develop improved cultivars[3]. In this regard, mutagenesis shows as an easy and effective mean of inducing genetic variation. Several researchers have used mutation breeding to improve grains yield of bread wheat[4-6]. The main idea of mutation breeding is to identify individuals with a target mutation, which involves two prime steps: mutant screening and mutant confirmation[7]. Mutant screening is a selection of individuals from a huge population that meet specific selection criteria, such as height, earliness, shape, weight compared to the their parents. Wheat spike is key components for improving grain yield because it is positively correlated with grain yield components such as grains per spike and weight per spike[8,9]. Most studies have attributed the increased grain yield is due to in increases in grains per spike[10], 1000 grain weight or both[11]. In addition, the spike, non-foliar organ, can be considered as the photosynthetic organ to grain production[12,13]. Thus, improving grain yield can be achieved by indirect selection for spike length, it has the number of spikelet's, the number of fertile spikelets and the grains per spike[14].

The expected response to selection for studied traits can be estimated by some parameters like phenotypic (PCV) and genotypic (GCV) coefficient of variability, heritability and genetic advance. The GCV measures extent of genetic variability shown by studied trait but it is not possible to determine quantity of variation that is heritable. Estimation of heritability is basically for selection process as it determines the transmissibility of a studied trait in the next generation but does not give a true picture of the expected gain in the next generation. Heritability and genetic advance are two guidelines for determining the genetic control of studied traits and estimate its expected gain from selection in the next generation[15]. Correlation coefficients are prerequisites to improve any crop[16]. Knowledge of the correlation between yield and its components is essential in establishing plant selection guidelines for plant selection[15]. Therefore, the major focus in this study was on selection for a long spike length, non-foliar organ, because it can be detected easily in the field to supply quantitative character data related to grain yield. The objectives of this study were to increase genetic variation of bread wheat and to identify among mutant lines those with high- yielding under marginal lands conditions.

MATERIALS AND METHODS

Plant material and mutagens: The basic strategy for breeding mutations is based on modifying one or more traits of distinct varieties[17]. Grains of five bread wheat varieties, Sakha 93 (P_1), Sids 13(P_2), Giza 168 (P_3), Gemmeiza 9 (P_4) and Maryout 5 (P_5) obtained of Agriculture Research Centre, Ministry of Agriculture and Land Reclamation, Egypt. The characteristic of the mother varieties are shown in Table 1.

They were irradiated by three doses of 100, 200 and 300 Gy of gamma rays (Cobalt-60) at the Egyptian Atomic Energy Authority

M_1, M_2, M_3 and M_4 generations: After the irradiation, the experiment was set up using 15 M_0 combination seeds along with the mother varieties (as control) in order to obtain M_1 plants. The generations (M_1 to M_4) was grown at the Agricultural Production and Research Station of National Research Centre, El-Nubaria, Al-Buhayrah Governorate, Egypt. The experiment was carried out in a randomized complete block design with 3 replicates during growth seasons (2016-2019). The sowing was done in November, 2016 by hand at the rate of 350 seeds m^{-2} into the 1.2 m length $\times$ 0.6 m wide, with 3 rows 0.2 m on row distance (plot size = 2.16 m^2). Sprinkler irrigation was applied. Out crossing with other varieties was avoided. Weeds were removed by hand throughout the growing seasons. The standard agricultural practice was applied. The soil characteristics presented in Table 2.

At harvest, spikes from main tillers were taken from survival seeds about ~70% as a subvitals mutation. The seeds obtained from the harvested plants in M_1 generation were sown to rise M_2 generation in 2017. Seeds collected of each combination were sown to rise M_3 generation. In 2018, each combination was grown in three replicate six-row plots, 1.2 m long and 0.60 m wide with 20 cm between rows (plot size = 4.3 m^2). At harvest, thirty spikes from each combination were selected based on Spike Length (SL),10 cm <, measured from the base of the rachis to the end of the terminal spikelet, Grains per Spike (GS), Grain Yield per Spike (GYS) and 1000 Grain Weight (TGW). Selection was delayed to M_3

Table 1: Pedigree and characterizes of mother cultivars

Variety	Pedigree	Characterizes
Sakha 93 (P_1)	Sakha 92/TR 810328 S 8871-1S-2S-1S-0S	Resistant to rusts, tolerates salinity and heat, short plant height, more tillers
Sids 13 (P_2)	KAUZ"S"//TSI//TSI/SNB"S"ICW94-0375-4AP-2AP-030AP-0APS-3AP-0APS-050AP-0AP-0SD	Resistant to rusts and water deficit, early maturing (152 d). Tolerates water salinity
Giza 168 (P_3)	MIL/BUC//Seri CM93046-8M-0Y-0M-2Y-0B	Tolerates water deficit, heat tolerant, resistant to rusts, late maturing (165-170 days), medium plant height, white grain color, thin spikes
Gemmeiza-9 (P_4)	Ald"S"/Huac"S"//CMH74A.630/5x CGM4583-5GM-1GM-0GM	Sensitive to salinity and water deficit, tall plant height, long spikes, late maturing (160 days), resistant to rusts
Maryout 5 (P_5)	Giza 162 // Bch's /4/ PI-ICW 79Su511Mr-38Mr-1Mr-0Mr	High yielding and salt tolerant

Table 2: Physical and chemical properties of the soil

	Physical properties			
Soil layer depth (cm)	0-25	25-50	50-75	75-100
Texture	Sandy	Sandy	Sandy	Sandy
Course sand (%)	48.66	55.71	37.76	37.57
Fine sand (%)	48.83	40.58	58.43	57.32
Silt+clay (%)	2.51	3.71	3.81	5.11
Bulk density (t m^{-3})	1.69	1.68	1.67	1.69
Chemical properties				
$EC_{1:5}$ (dS m^{-1})	0.44	0.53	1.00	1.56
pH (1:2.5)	8.60	8.70	9.32	9.03
Total $CaCO_3$ (%)	7.00	2.34	4.66	5.02

Table 3: Determination of mother varieties and doses used to generate mutant lines

Mutant lines	Mother variety	Dose
1	Giza 168	200
2	Gemmeiza 9	200
3	Giza 168	200
4	Giza 168	200
5	Gemmeiza 9	200
6	Sakha 93	300
7	Sakha 93	300
8	Sids 13	300
9	Sids 13	300
10	Sids 13	300
11	Gemmeiza 9	100
12	Gemmeiza 9	100
13	Gemmeiza 9	100
14	Gemmeiza 9	100
15	Sids 13	100
16	Sids 13	100
17	Sids 13	100
18	Giza 168	100
19	Giza 168	100
20	Giza 168	100
21	Sakha 93	100
22	Sakha 93	100
23	Sakha 93	100

numbered from 11-23 resulted from the 100-Gy radiation dose. Line number, mother varieties and doses are presented in Table 3.

In M_4 generation, five out of 23 mutant lines were sown as previous generations and reevaluated based on previous traits plus to Plant Height (PH), number of spikes/m^2 (NH) and Grains Yield ha^{-1} (GYH).

Statistical analysis of quantitative traits: Data of each trait were subjected to a regular analysis of variance of RCBD according to John[19]. The p-value$\leq$0.05 was considered statistically significant. $\pm$: Standard deviations. Phenotypic (PCV) and genotypic (GCV) coefficients of variation for lines were calculated according to method of Singh and Choudhary[20] as follows:

$$PCV = \frac{Vp}{X} \times 100$$

$$GCV = \frac{Vg}{X} \times 100$$

where, V_p, V_g and X are the phenotypic variance, genotypic variance and grand mean of lines, respectively for studied characters. Broad sense heritability ($h_{b2\%}$) estimated as the ratio of the genotypic variance (v_g) to the phenotypic variance (V_p) as described by Allard[21]. According to Johnson and Wichern[22], the heritability value of a trait when reaches 80%

generation due to studied traits produced a huge phenotypic effect that is recognizable on individual plant and called a macromutation. It is known that a macromutations are polygenic in nature[18]. Twenty-three mutant lines (as putative lines) were resulted as follows: lines numbered from 1-5 resulted from the 200-Gy radiation dose, lines numbered from 6-10 resulted from the 300-Gy radiation dose and lines

considered as high, moderate when it ranged from 40-80% and low when it less 40%. The expected genetic advance under selection (GA) as well as GA as percentage of the genotype mean assuming selection of the superior 5% of the genotypes were estimated in accordance with the formula illustrated by Allard [21] as follows:

$$GA = K. (\sigma^2 ph)^{1/2}. h_{b2\%}$$

$$GA \text{ (as \% of the mean lines)} = \frac{GA}{X} \times 100$$

where, k is the selection differential at 5% intensity = 2.06, σ2ph: Phenotypic standard deviation, $h_{b2\%}$: Broad sense heritability.

Deshmukh *et al.*[23] categorized genetic advance from selection as percent of mean as low (<10%), moderate (10-20%) and high (>20%). Correlation coefficients were calculated between all the studied traits according to Kashiani and Saleh[24].

RESULTS

Genetic variation among mutant lines: Twenty-three mutant lines have resulted from treatment of five wheat varieties seeds by doses of gamma rays (100,200 and 300 Gy). Analysis of variance showed a very highly significant difference ($p \leq 0.05$) among the mutant lines for all the studied traits in M_3 and M_4 generations (Table 4).

The mean square for mutant lines vs. their mother varieties was significant ($p \leq 0.05$) for all studied traits. Spike length was recorded the maximum coefficient of variation (8.3%), while 1000 grain weight recorded the least coefficient

of variation (3.7%) in M_3 generation. In M_4 generation, grains yield per spike showed the highest value of CV% (11.2), while grains yield ha^{-1} scored the lowest value (5.5%).

Quantitative traits in M_3 generation: The mean performance, grand mean and range of genotypes for the studied traits were showed in Table 5 to illustrate the differences between lines and their parents. Five mutant lines (1, 2, 3, 12 and 14) exhibited the higher mean performance for spike length (18 cm $\leq$) compared to the mother varieties. Mean performance of mutant lines for spike length ranged from 11.6 cm for line-7 to 18.5 cm for line-14 followed by line-1 (18.2 cm) while mother varieties ranged from 13.2 cm for P_3 to 17.6 cm for P_5. Grand mean of mutant lines for spike length was 15.9 cm while grand mean of mother varieties was 15.3 cm. In support of these results, Fig. 1 clearly showed that three mutant lines numbered 1 to 3 were longer spike length compare to mother varieties as shown in Fig. 2. Also, Fig. 3 showed that line -14 excelled in spike length compared to its parent and this was evidenced in Fig. 4.

Concerning with grains per spike, five mutant lines (1, 3,4,12 and 17) showed higher grains per spike (75$\leq$) compared to their parents (70.2$\geq$). Mean performance of mutant lines for grains per spike ranged from 30.0 for line-23 to 87.0 for line-12 followed by line-4 (82.0) while mother varieties ranged from 55.2 for P_1 to 70.2 for P_4. But, parents achieved the higher grand mean (66.4) compared to grand mean of mutant lines (55.3). In this study, mean performance and grand mean of parents for grains yield per spike was higher than mutant lines. But, mutant lines recorded wide range for this trait ranged from 1.2 g for line -23 to 3.6 g for line -12. Five out of twenty-three mutant lines (6,15,20,21 and 23) recorded higher mean performance for 1000 grain weight (45 g $\leq$) compared to their

Table 4: Mean squares of ANOVA for the studied traits of M_3 and M_4 generations

Trait	S.V			CV%
	Line	Error	L vs P	
M_3				
SL (cm)	7.6**	1.5	0.03**	8.3
GS	631.1**	11.9	0.04**	6.3
GYS (g)	1.3**	0.01	0.02**	5.2
TGW (g)	84.2**	1.9	0.01**	3.7
M_4				
PH (cm)	402.1**	31.2	0.02**	6.3
SL (cm)	6.9**	0.28	0.03**	7.2
GS	259.2**	26.3	0.01**	6.1
GYS (g)	1.09**	0.20	0.02**	11.2
TGW (g)	109.6**	18.3	0.04**	9.3
NH	1016.3**	132.0	0.04**	8.3
SYH (kg ha^{-1})	10244740.0**	90653.5	0.01**	5.5

S.V.: Source of variation, SL: Spike length, GS: Grains per spike, GYS: Grains yield per spike, TGW:1000 grain weight, PH: Plant height, NH: Number of spikes/m2, SYH: grains yield ha^{-1},**p<0.05, CV: Coefficient of variation, L: Lines, P: Mother varieties. Note that degrees of freedom in M_3 generation equal to 22, 44 and 1 for lines, error and lines *vs.* parents and in M_4 generation equal to 4, 16 and 1, respectively
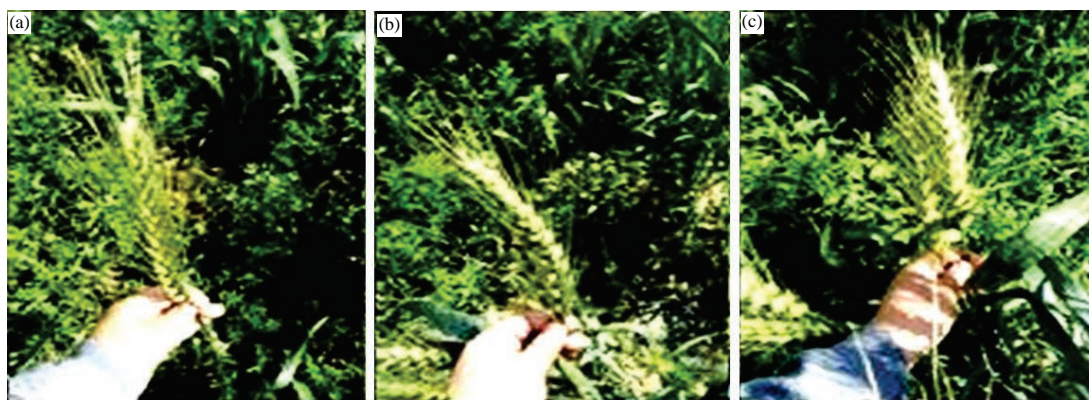
Fig.1(a-c): Spike length of mutant lines (1.2 and 3) stated from left side in the M_3 generation

Table 5: Mean performance, grand mean and range of mutant lines for studied traits in M_3 generation

Line	SL (cm)	GS	GYS (g)	TGW (g)
1	18.2±1.5	75.0±3.5	3.0±0.4	30.0±6.0
2	18.0±2.1	47.0±5.7	2.4±0.1	41. ±5.7
3	18.1±1.4	79.0±2.8	3.5±1.0	35.0±1.4
4	16.4±0.7	82.0±6.4	3.4±0.2	36.0±4.2
5	15.1±2.1	40.0±4.9	2.2±0.8	44.0±8.5
6	15.1±2.1	42.0±2.1	2.5±0.4	45.0±3.5
7	11.6±0.7	60.0±9.9	2.8±0.5	37.0±2.8
8	15.3±0.7	60.0±4.2	2.8±0.3	35.0±1.4
9	17.0±0.9	37.0±4.9	1.7±0.3	45.0±6.4
10	15.2±0.7	42.0±6.4	1.6±0.1	43.0±1.4
11	16.1±2.1	52.0±2.8	2.9±1.1	36.0±2.1
12	18.0±0.3	87.0±7.1	3.6±0.9	35.0±1.4
13	16.0±0.5	52.0±5.7	2.1±0.8	41.0±6.4
14	18.5±2.8	60.0±4.2	3.4±1.0	39.0±2.1
15	15.0±1.0	45.0±4.2	2.2±0.6	45.0±2.1
16	14.0±0.7	54.0±2.8	1.4±0.2	25.0±2.8
17	15.2±0.7	75.0±8.5	2.3±0.8	30.0±0.7
18	16.6±1.4	58.0±5.7	2.2±0.1	37.0±4.9
19	16.0±2.1	63.0±2.1	2.1±0.1	33.0±2.8
20	15.7±2.8	34.0±4.2	1.7±0.1	46.0±4.9
21	15.0±2.1	30.0±7.1	2.3±0.6	45.0±2.8
22	15.3±2.8	64.0±6.4	2.9±0.6	35.0±4.9
23	15.2±3.5	30.0±4.9	1.2±0.8	46.0±3.5
X̄	15.9	55.3	2.4	38.4
R	11.6-18.2	30.0-87.0	1.2-3.6	25.0-46.0
Parents				
P_1	14.2	55.2	3.8	43.1
P_2	17.1	66.6	3.9	42.1
P_3	13.2	60.2	4.1	41.2
P_4	14.2	70.2	4.0	44.1
P_5	17.6	80.0	3.5	41.2
	15.3	66.4	3.9	41.7

SL: Spike length (cm), GS: Grains per spike, GYS: Grains yield per spike, TGW: 1000 grain weight, Grand mean, R: Range, L: Lines, P: Mother varieties, ± standard error, **$p<0.05$

parents (44.1g≥). Two lines, 20 and 23, recorded the highest 1000 grains weight (46 g) while P_4, the highest parent, recorded 44.1 g. Mutant lines showed wide range for 1000 grain weight ranged from 25.0 g for line-16 to 46.0 g for line-20 with grand mean 38.3 while mother varieties ranged from 41.1 g for P_2 to 44.1 g for P_4 with grand mean 41.7 g. From of M_3 generation, five mutant lines (1, 2, 3, 12 and 14) identified based studied traits and consider as putative mutants. To confirm that these mutant lines are true, they reevaluated in M_4 generation plus to plant height, number of spikes m^{-2} and grains yield ha^{-1} (Table 5).

Fig. 2: Spike length of mother variety for mutant lines 1.2 and 3 in M3 generation

Fig. 3: Spike length of mutant line-14 in the M3 generation

With regard to plant height, mean performance of mutant lines for plant height ranged from 80.1 cm for line-14 to 103.2 cm for line-3 with grand mean 88.6 cm (Table 6). While mother varieties ranged from 77.4 cm for P_4 to 105.5 cm for P_3 with grand mean 89.3 cm. The grand mean of mother varieties and mutant lines are equal. Regarding spike length, mean performance of mutant lines ranged from 15.7 cm for line-14 to 18.4 cm for line-2 with grand mean 17.0 cm. In contrast, mother varieties ranged from 12.2 cm for P_3 to 18.3 cm for P_2 with grand mean 15.5 cm. The difference between the line-2 and its parent for the spike length was evidenced in the Fig. 5.

For grains per spike, the line 2 had the highest mean performance (90.4) compared to P_4, its parent, (80.0) and other parents (85.1$\geq$). And grand mean of mutant lines (83.8) was superior to mother varieties (76.7). Regarding grains yield per spike, line 2 showed the highest mean performance (4.8 g) while P_4 recorded 3.7 g. Also, grand mean of mutant lines

(4.1g) was higher than mother varieties (3.6 g). Furthermore, line 2 was superior in 1000 grain weight (52.0 g) compared to P_4 (41.2) and grand mean of mutant lines for 1000 grain weight (45.9 g) was higher than mother varieties (42.3 g). Mutant lines ranged from 41.2 g for line-14 to 52.0 g for line-2. With respect to number of heads m^{-2} and grains yield ha^{-1}, line-2 achieved the highest mean performance (159.3 and 7441.3 kg) compared to P_4 (111.2 and 3344.5 kg) and other parents and mutant lines.

Traits selection value: The total variability was expressed in terms of the PCV and GCV. The values of PCV in all the traits were higher than the values on corresponding GCV in M_3 and M_4 generations (Table 7). In M_3 generation, the highest value of GCV (20% <) was recorded for grains per spike (25.9%) followed by grains yield per spike (16.7%). Nevertheless, the smallest GCV was for 1000 grain weight (13.5). The low

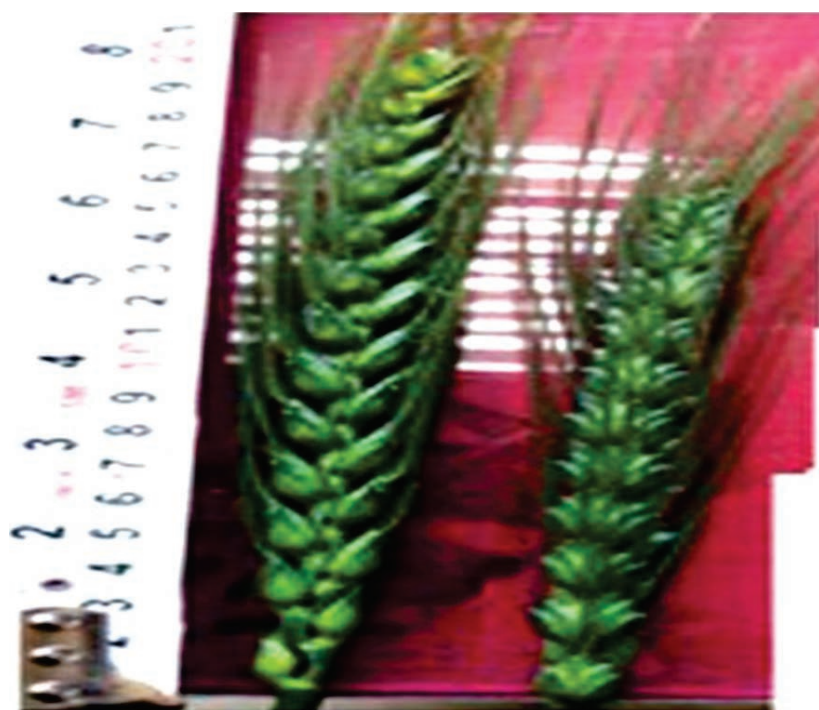Fig. 4: Spike length of the mother variety for mutant lines-14

Fig. 5: Spike length of the line-2 on the left side and his parent on the right side in M4 generation

differences between values of PCV and GCV values found for studied traits (1.1$\geq$). The highest value of heritability found for grains per spike (98.1%). In general, all traits recorded high value of heritability (80.0% $\leq$). In the same direction, all traits which recorded a high value of heritability (80%$\leq$) were associated with high genetic advance (48.2%$\geq$) as % of lines mean, except for spike length was moderate (14.7%).

In M$_4$ generation, the highest value of GCV was recorded for grain yield ha^{-1} (26.3%) followed by 1000 grain weight (11.2%) while grains per spike recorded the smallest value (10.1%). Phenotypic (PCV) and genotypic (GCV) coefficients of variability were low for all studied traits expect grain yield ha^{-1}

compared to M$_3$ generation. However, the differences between the two parameters were less (1.1$\geq$). High estimate of heritability (99.0%) accompanied with high genetic advance (53.0 %) as % of lines mean was recorded for grain yield ha^{-1}. The rest of traits recorded a high value of heritability (81.8% $\leq$) accompanied with moderate genetic advance (20.0% >) as % of lines mean. From these results, the genetic variability was presented for studied traits which provide ample opportunity for further selection.

Relationship among studied traits: In this study, data in Table 8 showed correlation coefficients among studied traits

Table 6: Mean performance, grand mean and range of mutant lines for studied traits in M_4 generation

Line	PH (cm)	SL (cm)	GS	GYS (g)	TGW (g)	NH	GYH (kg ha⁻¹)
Line							
-1.0	87.2	17.9	89.2	4.2	47.0	156.2	6548.5
-2.0	89.2	18.4	90.4	4.8	52.0	159.3	7441.3
-3.0	103.2	16.7	85.2	4.0	47.3	136.1	5484.7
-12.0	83.5	16.3	79.4	3.8	42.1	127.1	4248.6
-14.0	80.1	15.7	73.2	3.9	41.2	125.0	3769.8
X̄	88.6	17.0	83.5	4.1	45.9	139.4	5442.2
R	75.0-	15.0-	68.2-	3.0-	39.1-	120.3-	3570.2-
	115.1	19.1	100.2	6.0	55.0	172.1	7612.3
Parents							
P_1	93.3	13.2	75.2	3.6	42.1	133.2	4217.0
P_2	90.1	18.3	85.1	3.8	44.1	160.2	6012.2
P_3	105.5	12.2	70.2	3.5	41.4	129.2	3718.6
P_4	77.4	17.2	80.0	3.7	43.1	140	4827.2
P_5	87.0	18.1	73.0	3.3	41.2	111.2	3344.5
	89.3	15.5	76.7	3.6	42.3	134.8	4423.9
LSD $_{0.05}$	7.4	0.70	8.6	0.60	5.7	15.0	466.5

SL: Spike length (cm), GS: Grains per spike, GYS: Grains yield per spike, TGW: 1000 grain weight, PH: Plant height, NH: Number of spikes m⁻², GYH: Grains yield ha⁻¹, X̄ Overall mean, R: Range, LSD$_{0.05}$: Least significant difference

Table 7: Estimates of genetic parameters of studied traits in M_3 and M_4 generations

	M_3				M_4					
	SL	GS	GYS	TGW	PH	SL	GS	GYS	TGW	GYH
PCV	11.1	26.2	17.1	13.8	11.0	12.0	10.6	11.6	12.1	26.6
GCV	10.0	25.9	16.7	13.5	10.7	11.0	10.1	10.5	11.2	26.3
PCV-GCV	1.1	0.3	0.4	0.3	0.3	1.0	0.5	1.1	0.9	0.3
$h_{b2\%}$	80.0	98.1	97.5	97.5	92.0	94.0	89.0	81.0	81.8	99.0
GA%	14.7	91.8	97.9	48.2	18.1	13.0	15.6	19.0	16.6	53.0

SL: Spike length, GS: Grains per spike, GYS: Grains yield per spike, TGW: 1000 grain weight, PH: Plant height, NH: Number of spikes m⁻². GYH: Grain yield ha⁻¹, PCV: Phenotypic coefficient of variation, GCV: Genetic coefficient of variation; h$_{b2\%}$: Heritability in broad sense; GA% genetic advance as % of lines mean

Table 8: Correlation coefficients between quantitative traits in M_4 generation

	SL	GS	GYS	TGW	NH	GYH
PH	0.133	0.34	0.248	0.145	0.139	0.325
SL	1	0.840**	0.559	0.849**	0.02	0.901**
GS		1	0.612	0.477	0.471	0.875**
GYS			1	0.147	0.147	0.801**
TGW				1	0.134	0.820**
NH					1	0.830**
						1

SL: Spike length, GS: Grains per spike, GYS: Grains yield per spike, TGW: 1000 grain weight, PH: Plant height, NH: Number of spikes m⁻², GYH: Grain yield ha⁻¹, **p<0.05

illustrated that grain yield ha⁻¹ recorded a positive significant correlation with spike length (0.901) grains per spike (0.875), grains yield per spike (0.801),1000 grain weight (0.820) and number of spikes/m² (0.830). Spike length recorded a positive significant correlation with grains per spike (0.840) and 1000 grain weight (0.849).

DISCUSSION

In this study, gamma irradiation was used as a strategy for a broad genetic base within bread wheat to improve the efficiency of the breeding programs because the human selection contributed to a long time in the loss of important

allelic variance[25]. Data in the Table 4 showed that presence variation among mutant lines in M_3 and M_4 generations, indicating that mutant lines did not perform uniformly across different gamma rays. Traits showed that the CV% was less than 20%, indicating that experimental data were not dispersed and traits were not severely affected environment[5]. Often the mutations are recessive and of low frequency. Thus, the major focus in this study was on phenotypic selection for a long spike length because it can be detected easily in the field to supply quantitative character data related to grain yield. In the M_3 generation, some putative mutant lines (1, 2, 3, 12 and 14) were identified by a longer spike length (18 cm ≤) compared to their parents. To confirm these

mutant lines are true, they were screened for the spike length in the M_4 generation and found that line 2 recorded the longest spike length (18.4 cm) compared to their parents (18.1 cm) [5,6]. Wheat spike comprises spikelets, each with several florets. Consequently, an increase in the spike length can be selected concurrently with increased grains per spike[26]. In this study, plant height trait was estimated in M_4 generation which was equal with mother varieties. In the current study, five mutant lines (1,3,4,12 and 17) recorded higher grains per spike (75.0$\leq$) compared to their mother varieties (70.2<). Mutant line-12 showed the highest grains per spike (87.0) in M_3 generation. While in M_4 generation the P_5 superior to mutant lines for grains per spike[5,26]. This finding showed that the increase in the spike length of line 2 had a positive effect on the grain per spike compared to other mutant lines. For grains yield of each spike, mean performance of parents was higher than mutant line in M_3 generation, but in M_4 generation, the situation changed in desired direction. Because, the mutant line 2 achieved the highest mean performance of grains yield per spike compared to mother varieties and other lines[6,8]. Regards to 1000 grain weight, six mutant lines (6, 9, 15, 20, 21 and 23) were heavier 1000 grain weight than mother varieties in M_3 generation[6,8,27]. However, these lines had a short spike length (16 cm >) and low grains per spike (45>). In M_4 generation, mutant line-2 recorded the highest 1000 grain weight (52.0 g) compared to mother varieties. Grain yield improvement has been significantly connected with increased 1000 grain weight[28]. In contrast, Xiao *et al.*[29] found that grain yield negatively correlated with 1000 grain weight. For both number of spikes m^{-2} and grain yield ha^{-1}, mean performance of mutant lines and mother varieties are equal for number of spikes m^{-2} in M_4 generation. Mean performance of mutant line 2 for grains yield ha^{-1} was higher than mother varieties and other mutant lines[4-6,27]. The reason for superior mutant lines vs. mother varieties for spike length, grains per spike, grains yield per spike, 1000 grain weight and grain yield ha^{-1} may be due to generate of new mutated alleles responsible for increasing these traits. Hence, specific merit of mutation induction is the possibility of winning unselected genetic variation, improvement of one or a few characters of an excellent cultivar is to be modified[30]. Therefore, the maximize grain yield in bread wheat obtained by manipulating and predicting spike morphology traits such as spike length, grains per spike, grain yield per spike and 1000 grain weight. It noted that grand mean of mutant lines for grains per spike, grains yield per spike and 1000 grain weight in M_3 generation were lower

than M_4 generation. The ranges gained in the M_3 generation were higher than M_4 generation for studied traits (Table 5 and 6). Due to the variability dive gradually from M_2 to M_4 generations[17]. This study identified line 2 as a promising line. Because, it achieved the highest level of performance for studied traits. It was cleared that the use of the spike length character had a positive effect for identifying the distinct mutant among a large number of plants, especially in early generations. The sum variability was showed in terms of the PCV and GCV. In this context, the values of PCV were higher than their corresponding GCV for all studied traits in two mutated generations, which environmental impact was comparatively low on the expression of studied traits. In this study, a narrow gap was found between the PCV and GCV values of studied traits in two mutated generations, indicating that the environmental factors had lower influence on the expression of these characters than the genetic factors. Grains yield ha^{-1} recorded a high value of PCV and GCV, suggesting that this trait is under the influence of genetic control. This indicated that this trait could be improved through hybridization and selection for better yield performance. Heritability and genetic advance give generic behavior of traits which increases the efficiency of selection. High heritability (80% <) was coupled with high genetic (20% <) advance as percent of mean lines observed for studied traits except for spike length was moderate (14.7%) in M_3 generation. In M_4 generation, grains yield ha^{-1} recorded high heritability coupled with high genetic advance. This indicated that mutation take place in more than one locus[17]. This indicated that the additive gene action controlled in grains yield ha^{-1} and can easily be selected through phenotypic selection[5]. All traits, except plant height, recorded a positive and significant correlation with grain yield ha^{-1} indicates that the important role of these traits in determining grain yield, consequently, these traits have priority in selection for higher grain yield. Spike length was positively correlated with grains per spike and grains weight[14,31]. This indicated that spike length has a great role in improving grain yield ha^{-1} of bread wheat in sandy soil. This finding suggests that crossing between proper mutant lines possessing longer spike length (18 cm$\leq$), higher grains per spike (80<) and heavier 1000 grain weight (46 g $\leq$) and crossing with commercial cultivars would be given positive results. Another use of these lines is further evaluation under biotic and abiotic stresses with evaluation in many locations for many years. Consequently, broaden genetic variation of bread wheat in Egypt by add a novel genes and give a chance for grain yield improvement. The introduction

of a new adaptive allele in the diverse genetic backgrounds may assist to promote grain yield of old or newly developed varieties of bread wheat to balance supply and demand in Egypt.

CONCLUSION

Five varieties of bread wheat were irradiated by three doses of gamma rays to increase genetic variability and to improve grain yield. Twenty -three mutant lines were generated based on spike length in M_3 generation. Five out 23 mutant lines were reevaluated based on grain yield components. Mutant lines showed significant variation in the studied traits that were established through phenotypic and genotype coefficients of variation. Several mutant lines were superior to their mother varieties for most studied characters in M_3 and M_4 generations. High values of heritability associated with moderate genetic gain observed in all studied characters. This indicated these characters were controlled by additive gene effects and selection may be effective for these characters to grain yield improve. All traits, except plant height, recorded a positive and significant correlation with grain yield ha^{-1} indicates that the important role of these traits in determining grain yield. These mutant lines provide exciting new valuable resources to investigate and improvement of complex traits in bread wheat.

SIGNIFICANCE STATEMENT

This study discovered the direct selection for spike length has a positive effect of grain yield of bread wheat that can be beneficial for wheat breeders. This study will help the wheat breeder to use new valuable resources to investigate and improvement of complex traits to uncover the critical areas of grain yield improvement that many researchers were not able to explore.

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