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Evaluation of Some Bread Wheat Mutants for Adult Plant Resistance to Stem Rust

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Abstract

Stem rust disease is a drastic factor to bread wheat (Triticum aestivum, L.) production in Egypt and several countries in the world. This study was carried out to evaluate twenty-two bread wheat mutant-lines in M_5 and M_6 generations as well as five check bread wheat cultivars Gemmeiza11, Sids12, Sakha 93, Sakha94 and Morocco, for high yield and an adequate level of adult plant resistance to stem rust under stress of the disease in the field. The epidemiological parameters; final rust severity (FRS%), average coefficient of infection (ACI) and area under disease progress curve (AUDPC), were used to characterize adult plant resistance to stem rust in the tested genotypes, as well as, some yield traits; grain yield, biological yield, No. of grains / spike and 1000-kernel weight were also recorded for the studied genotypes. The results showed that Sakha94 (Sk94) cultivar and the nine mutant lines, i.e. (Mut1, Mut2, Mut11, Mut25, Mut26, Mut28, Mut38, Mut59, Mut161), were resistant to stem rust. Negative correlation coefficient values existed between disease parameters and yield traits under study, showing harmful effect of stem rust on plant characteristics. Grain yield showed negative and significant correlation with ACI (r= -0.694**), AUDPC (r= -0.679**) and FRS% (r=-0.665**), while positive and significant correlation were found with biological yield ($r = 0.889^{**}$), number of grains/spike ($r = 0.411^{*}$). The best mutant lines, having an adequate levels of adult plant resistance to stem rust, combined with desirable yield traits could be introgressed into adapted Egyptian wheat cultivars; to develop durable resistance to stem rust in wheat.

Keywords: Wheat, Mutations, Puccinia graminis f. sp. tritici, Adult plant resistance (APR), Gamma radiation.

Introduction

Bread wheat (*Triticum aestivum* L.) is the most important cereal crop in Egypt and worldwide. It provides over 20% of calories consumed by the world's population (**Bushuk and Rasper, 1994**). Wheat is a main source for essential calories and protein, supplying more than 75% of protein and 65% of calories in human diet (**Mostafavi, et al., 2005**).

Stem rust, caused by Puccinia graminis f. sp. tritici is an important disease of wheat in Egypt and worldwide, as it considered the most destructive disease to the susceptible wheat cultivars. Under favorable environmental conditions, stem rust may cause yield losses up to 100 % to the susceptible wheat cultivars (Roelfs, 1985 and Leonard and Szabo, 2005). During the twentieth century, severe yield losses due to SR epidemics were reported in Europe, Asia, Australia, and the USA (Nagarajan and Joshi 1975; Roelfs 1978; Leonard and Szabo 2005). Stem rust is considered to be the most destructive disease of wheat, where the losses may reach to approximately 100% on the susceptible wheat cultivars, when conditions are favorable for the disease incidence and development (Singh et al. 2002).

Since the establishment of the Joint Food and Agriculture Organization of the United Nations/International Atomic Energy Agency (FAO/IAEA) Division of Nuclear Techniques in Food and Agriculture to assist member countries in applying radiation-induced mutation breeding technologies to improve existing and local crop varieties. Plant breeders have utilized diverse physical, chemical, and combination mutagens to produce genetic variation or diversity in various crops. More than 3362 cultivars obtained either as direct mutants or derived from their crosses have been released worldwide in more than 75 countries (Fatma Sarsu, et al., 2020, Xiong, et al., 2018 and Kong, et al., 2020). There are 2635 varieties could be generated by physical mutagens, 398 developed by chemical mutagens, and 37 produced by a mix of physical and chemical mutagens. Africa is responsible for creating 82 varieties, Asia for 2049, Australia and the Pacific for 10, Europe for 959, Latin America for 53, and North America for 209. Similarly, mutation breeding and induced mutation have produced 1602 main cereals, 501 major legumes, and 86 significant oil seed mutant crop varieties. Eighty percent of these mutants were induced using gamma rays; 274 wheat mutants and 850 rice mutants were released worldwide to face climate changes FAO/IAEA-MVD, (2018). Hence, this study was planned to evaluate and / or identify some bread wheat genotypes (22 mutant lines and 5 wheat cultivars) for high yield and an adequate and good level of adult plant resistance (APR) to stem rust disease, under stress of disease in the Egyptian field conditions.

Materials and Methods Plant materials

Twenty-two bread wheat mutant lines in M5 and M6 generations (Mut1, Mut2, Mut3, Mut11, Mut99, Mut199, Mut26, Mut28, Mut37, Mut38, Mut59, Mut64, Mut65, Mut44, Mut68, Mut25, Mut31, Mut49, Mut161, Mut166, Mut132 and Mut142) as well as five check bread wheat cultivars (Gemmeiza11, Sids12, Sakha93, Sakha94 and Morocco) were evaluated for high yield and resistance to stem rust disease. The studied 22 mutant lines were previously released as a result of exposed dry grains of the three local bread wheat cultivars (Gemmeiza11, Sids12 and Sakha 93) to different doses of gamma rays (0, 250, 300 and 350Gy) in season 2013/2014.

Field trial

Five commercial wheat cultivars, and twenty two mutant lines were evaluated against stem rust under natural field conditions for disease response, grain yield, morphological characteristic and other pathological studies. Two experiments, were carried out in 2017/2018 and 2018/2019 growing seasons which M5 and M6 generation, were planted, respectively. The tested wheat genotypes were grown at the experimental, Research Station of Moshotohor, Faculty of Agriculture, Benha University. The previous wheat genotypes were sown 15 days after sowing date (the first half of December) to expose the tested wheat plants to the suitable environment for stem rust incidence and development. The tested genotypes were sown in rows within plot, each plot consisted of 8 rows, 3m long and 30cm wide. Individual grains were spaced 10cm within row, in a randomized complete block design (RCBD), with three replicates. All recommended practices in the commercial fields *i.e.* fertilization, irrigation and other management were applied. All plots were surrounded by a spreader area of one meter in width, planted with a mixture of the two highly susceptible

wheat varieties to stem rust i.e. Morocco and Max. The spreader area of wheat plants were artificially inoculated before rust appearance during the second half of March to provide the tested plants with permanent source of stem rust urediniospores. The inoculation of all plants was carried out at booting stage, according to the methods of Tervet and Cassel (1951). Besides, the spreader plants were subjected to simultaneous injections with uredospors suspended in a distilled sterile water including uredinospores of different pathotypes of Puccinia graminis f. sp. Tritici, obtained from the Wheat Diseases Research Department, Plant Pathology Research Institute, ARC, Giza. Data of stem rust severity (%) were recorded on the adult plant stage of the tested wheat plants as adopted by Peterson et al. (1948). Data of stem rust incidence were scored, as a cultivar response (infection type) and severity (%) of infection, combined together every weak from the first rust appearance, along with the stage of growth.

a. Disease assessment

1- Infection Type (IT):

Disease reaction was recorded based on the original scales proposed by (Roelfs et al., 1992) for stem rust in field evaluation. Five rust infection types were used for the evaluated wheat genotypes. These are; I-Immune or highly resistant, (no visible symptoms), R-resistant, (necrotic areas with or without minute uredia, MR- moderately resistant, (small uredia present surrounded by necrotic area), (MSmoderately susceptible, medium uredia with no necrosis, but with some possible distinct chlorosis), (S- susceptible, large uredia and little or no chlorosis present). Adult plant resistance response and disease severity (%) for stem rust, based on the modified Cobb's scale (Peterson et al., 1984) and the infection types by Roelfs, et al., (1992), Jin et al. (2007) and Singh et al. (2013) as shown in Table 1.

Table 1. Host response or infection type, disease severity % and Symptoms for stem rust.

Host response (Infection type)	Symptoms
Immune (I) or highly resistant	No visible symptoms or infection
Resistant (R)	Some chlorosis or necrosis and no uredia.
Moderately Resistant (MR)	Small uredia present ant surrounded by either chlorotic or necrotic
Moderately Susceptible (MS)	areas. Medium-sized uredia present and possibly surrounded by chlorotic
	areas.
Susceptible (S)	Large uredia present, generally with little or no chlorosis and no
	necrosis.

2 – Average coefficient of infection (ACI):

Average coefficient of infection, was calculated according to **Saari and Wilcoxson (1974)**, and **Pathan and Park (2007)**, by multiplying of disease severity (%) and constant values of infection type. The constant values for infection types that were used, are as follow: R=0.2, MR=0.4, M=0.6, MS=0.8 and S=1.0, as proposed by **Saari and Wilcoxson (1974)**.

3 – Rust severity (%):

Adult-plant reaction was scored as the rust severity (%) for each wheat genotype under study at the time when rust was first appeared until the early dough stage (Large, 1954). Rust severity (%) for each wheat genotype, was recorded every seven days, after the initial infection occurred, using the modified Cobb's scale (Peterson *et al.* 1948). Disease severity

(%) was calculated as the percentage of the affected tissues to the total plant tissues.

4 - Final rust severity (FRS %):

Stem rust severity percentage, was measured for the tested wheat cultivars, and mutant lines, as a percentage of stem, leaf and spike area infected or rusted, according to the modified Cobb's scale (**Peterson et al., 1948**). Final rust severity % (FRS%) was assessed as a percentage of disease rust severity% for each of the tested genotypes, when the highly susceptible (check) variety; Morocco, was severely rusted and the disease severity (%) reached its maximum or final level (**Das et al., 1993**).

5- Area under disease progress curve (AUDPC):

Area under disease progress curve (AUDPC), was estimated to compare different disease responses of the tested wheat genotypes, using the following equation, adopted by **Pandey** *et al.* (1989)

AUDPC= D [1/2 (Y1+YK) + Y2 + Y3 +Y (k-1)]

Where:

D = days between the two successive readings.

Y1 = first disease recording.

Yk = last disease recording.

Stem rust severity% data were recorded starting with the appearance of the first pustule on each of the tested wheat genotypes, and continued at 7 days intervals, between the two successive readings or scores, until the termination of the experiment.

b. Estimation of yield-related traits

At harvest stage, data were recorded on 10 individual guarded plants from the three replications for each genotype of the following traits; grains / spike (G/S), 1000-kernel weight (gm) (1000KW), grain yield (ardb/fed), and biological yield (ton/fed).

Data analysis

For comparison between the tested wheat genotypes, based on their level of stem rust resistance, the percentage of disease severity (%) was used to calculate each of the area under disease progress curve (AUDPC) and the coefficient of **Table 2.** Combined analysis of variance for stem ru infection (CI), Average coefficient of infection (ACI) by taking into account the disease severity (%) and their infection type, where; 0.0, 0.2, 0.4, 0.6, 0.8 and 1.0 represented immune or highly resistant, resistant (R), moderately resistant (MR), moderately resistant to moderately susceptible (MR-MS), moderately susceptible (MS) and susceptible (S), respectively (**Roelfs** *et al.*, **1992**).

The analysis of variance was carried out to determine the significance of the differences among the tested wheat lines (cultivars and mutant lines) for the epidemiological or disease parameters and the different agronomic traits. The least significant differences (P = 0.05), was used to compare the genotypic means. The homogeneity of error variance was done prior to combine analysis. This indicates that there are no significant differences between the two generations (years). Therefore, a combined analysis for the two generations was done according to **Snedecor and Cochran (1982).**

Correlation Analysis

Phenotypic correlation coefficient analysis was conducted to quantify the degree to which the disease level was related to different yield traits associated with yield trait. A Pearson correlation coefficient was done to establish the relationship between the different traits measured. These analyses were performed using SPSS program.

Results and Discussion

a) Analysis of variance:

Mean squares of analysis of variance over two generations for the investigated traits are given in Table (2). Results showed highly significant mean square effects for all genotypes under study for stem rust resistance parameters estimated at adult plant stages. Thus, the obtained results referred to the high diversity in the level of stem rust resistance between the bread wheat mutant lines of the study. The interaction between years (Y) and genotypes (G); (YxG) was highly significant for all the studied disease parameters and yield traits. 6 07 1

_	a a		1.0					ã	(3.5.0)						
		genotypes	over M5 and	d M6 genera	ations.										
	Table 2.	Combined	analysis of	variance for	or stem	rust	disease	parameter	's and	yield	traits	of 27	bread	wheat	

Source of variation	d.f.		Mean Squares (MS)						
		Disease parameters				Yield traits			
		FRS%	ACI	AUDPC	G/S	1000 KW	BY	GY	
Replication	4	0.021	0.055	2.64	0.957	0.725	0.002	0.02	
Year (Y)	1	0.413*	0.001	0.66	244.5**	0.747	15.6**	23.1**	
genotypes(G)	26	41.8**	22.9**	444.8**	406.3**	307**	2.88**	34.5**	
Interaction(YxG)	26	0.45**	0.25**	8.1**	28.6**	1.24**	0.44**	0.71**	
Error	104	0.122	0.041	2.264	0.745	0.459	0.001	0.015	

* and **denote significant differences at 5% and 1% levels, respectively. FRS% = final rust severity%; ACI = average coefficient of infection; AUDPC = area under disease progress curve; G/S= Number of grains/ spike; 1000KW=1000 Kernel Weight; BY= Biological yield; GY= Grain Yield.

b) Genotype performance against stem rust disease:

1. Genotypes performance in M_5 generation: Results given in Table (3) showed that the tested wheat genotypes gave different disease reactions (infection types) and different levels of stem rust severity (%) in M5 generation (2017/2018 growing season).

However, the percentage of stem rust severity (%) reached to its maximum level (80-90%), on the check wheat variety; Morocco. In this growing season, the level of rust severity (%) was relatively higher on most of the tested wheat genotypes (table3). According to the response and the reaction of the tested wheat genotypes against stem rust, they could be divided into four main groups:

a. The first group included the highly resistant (HR) wheat genotypes where, the disease response ranged from 0 to 5R, hence this group including the mutant lines i.e. Mut1 (Tr. R), Mut2 (Tr. R), Mut11 (5R), Mut25 (5R), Mut26 (0), Mut28 (5R), Mut38 (Tr.R), Mut59 (5R), Mut161 (5R), and as well as the commercial wheat cultivar Sakha94 (5R). The ability of this variety to resist the disease might be due to it proved to contain resistance Sr gene against stem rust.

b. The second group including moderately resistant (MR), mutant lines i.e. Mut3 (20MR), Mut31 (20MR), Mut37 (10MR), Mut44 (50MR), Mut49 (10MR), Mut64 (20MR), Mut65 (10MR), Mut68 (30MR), Mut132 (10MR) and Mut166 (20MR).

c. The third group of the tested wheat genotypes were those of the moderately susceptible (MS), including one mutant line; Mut199 (40MS), and wheat cultivars Gemmeiza11 (40MS).

d. Meanwhile, the fourth group including the highly susceptible genotypes i.e. the two mutant lines Mut99 (50S) and Mut142 (40S), as well as the three wheat cultivars; Sakha93 (60S), Sids12 (80S), and the check variety; Morocco (90S) (table 3).

The pervious study of **Denbel** *et al.*, (2013) stated that the wheat variety Pavon 76 was resistant to stem rust at adult plant stage, as it has low terminal rust severity (30%, MS), under heavy stem rust epidemic. They in addition, indicated that it's resistance may be attributed to the presence of the effective Sr2 gene complex. While, the wheat varieties; Abolla, Bobicho, Galema, Hawi, Kubsa, Wetera and Sofumer, showed in general a high rust severity (40% - 50%). Therefore, these wheat varieties characterized as the susceptible wheat genotypes.

2. Genotype performance in M₆ generation:

Results obtained in Table (3) also revealed that, the tested wheat genotypes showed wide differences in their disease reactions, as they displayed different levels of stem rust incidence and severity (%). Also, the percentage of rust severity reached to 80-90% on the check wheat variety; Morocco. However, during this season severe stem rust epidemic was recorded, where most of the tested wheat genotypes were severely rusted, and showed higher percentage of rust severity (%). Based on the disease response and the level of stem rust severity (%) of the tested wheat genotypes, could be classified into four groups:

a. The first group included the resistant wheat genotypes, that displayed the lowest percentages of stem rust severity% (did not exceeded up to 5%) with infection type (R). These genotypes were, the nine mutant lines, Mut1 (Tr. R), Mut2 (Tr. R), Mut11 (5R), Mut25 (5R), Mut26 (Tr.R), Mut28 (Tr.R), Mut38 (Tr.R), Mut59 (Tr.R), Mut161 (5R) and the commercial wheat cultivar; Sakha94 (5R).

b. The second group was contained the moderately resistant (MR) group of genotypes, i.e. the nine mutant lines; Mut3 (20MR), Mut31 (10MR), Mut37 (20MR), Mut49 (10MR), Mut64 (10MR), Mut65 (10MR), Mut68 (10MR), Mut132 (20MR) and Mut166 (20MR).

c. Meanwhile, the moderately susceptible (MS) group of genotypes, including only one mutant line; Mut44 (50Ms), and an only one commercial wheat cultivar; i.e. Gemmeiza11 (50MS).

d. In contrast, the fourth group of wheat genotypes, comprised or contained those displayed the highest percentage of stem rust severity (%) (ranged from 50S to 90S). This group included the three mutant lines i.e. Mut99 (50S), Mut142 (50S) and Mut199 (60S), in addition to the three wheat cultivars; Sd12 (70S), Sk93 (50S), as well as the check variety; Morocco (90S) (table 3).

c) Characterization of adult plant resistance (APR) to stem rust in mutant wheat lines:

To gain more details on the variation of disease response and levels of adult plant resistance (APR) to stem rust, in addition to characterize more accurately this type of resistance as displayed on the tested wheat mutant lines, the three epidemiological parameters i.e. ACI, FRS (%) and AUDPC were used and recorded for each of these genotypes, during the two growing seasons i.e. 2017/2018 (M_5 generation) and 2018/19 (M_6 generation) (table 4, 5 and 6).

1 - Final rust severity (FRS %)

Final stem rust severity (%) was scored and recorded as the disease severity (%) in each of the tested wheat genotypes, when the highly susceptible check variety; Morocco, was severely rusted and the disease rate reached its highest and final level (**Das** et al., 1993).

No.	Wheat Genotypes —	Stem rust reaction (severity and infection type)						
	wheat Genotypes —	M5 generation (2017/2018)	M6 generation (2018/2019)					
	a)Mutant line							
1	Mut.1	Tr. R	Tr. R					
2	Mut.2	Tr. R	Tr. R					
3	Mut.3	20MR	20 MR					
4	Mut.11	5 R	5 R					
5	Mut.25	5 R	5 R					
6	Mut.26	0	Tr. R					
7	Mut.28	5 R	Tr. R					
8	Mut.31	20MR	10MR					
9	Mut.37	10 MR	20MR					
10	Mut.38	Tr. R	Tr. R					
11	Mut.44	50MR	50MS					
12	Mut.49	10 MR	10MR					
13	Mut.59	5 R	Tr. R					
14	Mut.64	20MR	10 MR					
15	Mut.65	10MR	10MR					
16	Mut.68	30MR	10MR					
17	Mut.99	50 S	50 S					
18	Mut.132	10MR	20MR					
19	Mut.142	40 S	50 S					
20	Mut.161	5 R	5 R					
21	Mut.166	20 MR	20MR					
22	Mut.199	40 MS	60 S					
	b)Wheat cultivars:							
1	Gemmeiza11	40MS	50 MS					
2	Sids12	80 S	70 S					
3	Sakha93	60 S	50S					
4	Sakha94	5 R	5 R					
5	Morocco (check)	90 S	90 S					

Table 3. Stem rust reaction of 27 wheat genotypes (22 mutant lines and 5 cultivar) in M5 and M6 generationsduring 2017/18 and 2018/19 growing seasons.

R: resistant, MR: moderately resistant, MS: moderately susceptible, S: susceptible.

a. FRS (%) in M5 generation:

Results presented in Table (4) indicated that the wheat mutant lines; (Mut1, Mut2, Mut11, Mut25, Mut26, Mut28, Mut38, Mut59, Mut161, Mut3, Mut31, Mut37, Mut49, Mut64, Mut65, Mut68, Mut132 and Mut166), as well as the wheat cultivar Sk94, showed in general low percentages of stem rust severity (%) (not up to 30%), during this season. Therefore, these genotypes may be characterized as slow-rusting or partial resistant (PR) wheat genotypes to stem rust. Meanwhile, wheat genotypes (4 mutant lines i.e. Mut44, Mut99, Mut142, Mut199, and one cultivar; Gemmeiza11) exhibited moderate levels of final rust severity (%) (with FRS % 30-60%).On the other hand, the three wheat cultivars (Sd12, Sk93 and Morocco) showed higher values of final rust severity %; more than 50% and the check wheat variety Morocco 90%. They, therefore characterized as the fast rusting wheat genotypes.

b. FRS (%) in M₆ generation:

Results presented in Table (4) indicated that the wheat genotypes i.e. the mutant lines (Mut1, Mut2, Mut11, Mut25, Mut26, Mut28, Mut38, Mut59, Mut161, Sk94, Mut3, Mut31, Mut37, Mut49, Mut64, Mut65, Mut68, Mut132 and Mut166) showed highly level of adult plant resistance to stem rust, as they showed low percentage of disease severity % (not up to 30%). These genotypes may be considered as the slow-rusting genotypes, or having partial resistance (PR). On the other hand, wheat genotypes (Mut44, Mut99, Mut142, Mut199, Gm11 and Sk93) exhibited moderate levels of disease severity % (30-60%). While, the wheat cultivars; (Sd12 and the check wheat variety Morocco) showed higher values of final rust severity; more than 60% and 90%. Therefore, they characterized as the fast-rusting wheat genotypes.

	Final rust sev	verity (%)
Wheat genotypes —	M ₅ generation (2017/2018)	M ₆ generation (2018/2019)
a)Mutant line		
Mut.1	4.33	3.67
Mut.2	4.33	3.00
Mut.3	20.00	16.67
Mut.11	4.33	4.33
Mut.25	5.00	6.67
Mut.26	1.00	2.00
Mut.28	5.00	3.00
Mut.31	13.33	13.33
Mut.37	16.67	20.00
Mut.38	2.00	3.00
Mut.44	56.67	50.00
Mut.49	10.00	16.67
Mut.59	4.33	3.00
Mut.64	16.67	16.67
Mut.65	13.33	13.33
Mut.68	33.33	13.33
Mut.99	43.33	53.33
Mut.132	10.00	16.67
Mut.142	43.33	46.67
Mut.161	4.33	4.33
Mut.166	13.33	13.33
Mut.199	46.67	53.33
b)Wheat cultivars:		
Gemmeiza11	43.33	53.33
Sids12	73.33	70.00
Sakha93	60.00	53.33
Sakha94	6.67	3.67
Morocco (check)	90.00	90.00
General mean	23.864	23.959
L.S.D. at 0.05	6.575	7.496
L.S.D. at 0.01	8.714	9.935
	Mut.1 Mut.2 Mut.3 Mut.3 Mut.11 Mut.25 Mut.26 Mut.28 Mut.28 Mut.31 Mut.37 Mut.38 Mut.44 Mut.37 Mut.38 Mut.44 Mut.49 Mut.59 Mut.64 Mut.65 Mut.68 Mut.99 Mut.132 Mut.163 Mut.161 Mut.166 Mut.199 b)Wheat cultivars: Gemmeiza11 Sids12 Sakha93 Sakha94 Morocco (check) General mean L.S.D. at 0.05	a)Mutant line 4.33 Mut.1 4.33 Mut.2 4.33 Mut.3 20.00 Mut.11 4.33 Mut.25 5.00 Mut.26 1.00 Mut.28 5.00 Mut.31 13.33 Mut.32 2.00 Mut.34 56.67 Mut.35 2.00 Mut.44 56.67 Mut.49 10.00 Mut.59 4.33 Mut.64 16.67 Mut.65 13.33 Mut.64 16.67 Mut.42 56.67 Mut.43 56.67 Mut.44 56.67 Mut.42 10.00 Mut.59 4.33 Mut.64 16.67 Mut.68 33.33 Mut.199 43.33 Mut.166 13.33 Mut.166 13.33 Mut.199 46.67 b)Wheat cultivars: Gemmeiza11 Genereial mean <

Table 4. Final rust severity (%) of 27 wheat genotypes (22 mutant lines and 5 cultivar) in M5 and M6 generations under field conditions during 2017/2018 and 2018/2019 growing seasons.

2 - Average coefficient of infection (ACI)

a. ACI in M₅ generation:

Results presented in Table (5) indicated that the tested genotypes (Mut1, Mut2, Mut11, Mut25, Mut26, Mut28, Mut38, Mut59, Mut161 and Sk94) showed low values of ACI. On the other hand, wheat genotypes (Mut3, Mut31, Mut37, Mut44, Mut49, Mut64, Mut65, Mut68, Mut132, Mut166, Mut99, Mut142, Mut199 and Gemmeiza11) exhibited moderate values of ACI. In contrast, wheat genotypes (Sd12, Sk93 and Morocco) exhibited high values of ACI.

b. ACI in M₆ generation:

Results presented in Table (5) indicated that the tested genotypes (Mut1, Mut2, Mut11, Mut25, Mut26, Mut28, Mut38, Mut59, Mut161 and Sk94) showed low values of ACI. On the other hand, wheat

genotypes (Mut3, Mut31, Mut37, Mut44, Mut49, Mut64, Mut65, Mut68, Mut132, Mut166, Mut142, Mut199 and Gemmeiza11) exhibited moderate values of ACI. In contrast, wheat genotypes (Mut99, Sd12, Sk93 and Yakora) exhibited high values of ACI.

3 - Area under disease progress curve (AUDPC):

Area under disease progress curve (AUDPC), as a more reliable estimator of the disease incidence and development during an epidemic, was estimated and calculated for each of the tested wheat mutant lines and cultivars. However, this disease parameter was successfully used to accurately characterized and determine the level of adult plant resistance to stem rust, expressed or displayed in the tested wheat genotypes during the two growing seasons of the study; 2017/2018 and 2018/2019 (table 6).

No.		Average coefficient of infection (ACI)					
	Wheat genotypes —	M ₅ generation (2017/2018)	M₆ generation (2018/2019)				
	a)Mutant line						
1	Mut.1	0.30	0.25				
2	Mut.2	0.30	0.20				
3	Mut.3	3.67	3.63				
4	Mut.11	0.30	0.30				
5	Mut.25	0.53	0.48				
6	Mut.26	0.03	0.10				
7	Mut.28	0.48	0.20				
8	Mut.31	2.97	2.87				
9	Mut.37	3.37	4.40				
10	Mut.38	0.07	0.20				
11	Mut.44	10.37	21.00				
12	Mut.49	2.13	3.47				
13	Mut.59	0.30	0.20				
14	Mut.64	3.30	3.37				
15	Mut.65	2.60	3.03				
16	Mut.68	6.83	2.87				
17	Mut.99	21.83	25.00				
18	Mut.132	2.47	3.43				
19	Mut.142	23.50	22.00				
20	Mut.161	0.40	0.30				
21	Mut.166	2.67	2.87				
22	Mut.199	21.67	24.33				
	b)Wheat cultivars:						
1	Gemmeiza11	20.17	22.67				
2	Sids12	50.83	43.33				
3	Sakha93	34.33	26.67				
4	Sakha94	0.70	0.32				
5	Morocco (check)	55.83	55.83				
	General mean	10.086	10.126				
	L.S.D. at 0.05	2.906	2.267				
	L.S.D. at 0.01	3.851	3.004				

 Table 5. Average coefficient of infection (ACI) of 27 wheat genotypes (22 mutant lines and 5 cultivar) in M5 and M6 generations under field conditions during 2017/2018 and 2018/2019 growing seasons.

a. AUDPC in M₅ generation:

On the basis of AUDPC values, the tested wheat genotypes could be classified into two main groups:

The first group included the wheat genotypes which showed, the relatively low AUDPC values (less than 300). Therefore, these genotypes were characterized as the adult plant resistant (APR) genotypes "slow rusters". This group comprised or contained the elite mutant lines i.e.; Mut26 (2.3), Mut38 (4.7), Mut1 (28.00), Mut2 (28.00), Mut11 (28.00), Mut59 (28.00), Mut161 (42.00), Mut28 (47.8), Mut25(54.8), Mut49 (112.00), Mut132 (123.70), Mut166 (135.3), Mut65 (147.00), Mut31 (150.5), Mut37 (170.3), Mut64 (170.3), and Mut3 (182.00), as well as the only one wheat cultivar; Sk94(70.00).

In contrast, the second group of genotypes contained wheat genotypes that revealed a considerable higher estimates of AUDPC, than the partially resistant ones (more than 300), thus they were identified as "fast- rusters" or the highly susceptible genotypes. Such genotype group was contained five mutant lines, and three wheat cultivars, as well as the check variety, Morocco; Fast-rusting genotypes were; Mut68 (344.20), Mut99 (501.7), Mut44 (515.70), Gm11 (530.80), Mut199 (565.80), Mut142 (571.7), Sk93 (840.00), Sd12 (1061.7) and Morocco (1167.00) respectively.

b. AUDPC in M₆ generation:

Similar results were obtained in this growing season (M_6 generation), to those previously found in the first growing season (M_5 generation). However, due to the obtained results, and on the basis of AUDPC values, the tested genotypes could be classified into two main groups (Table 6).

The first group contained the wheat genotypes which displayed, the relatively low AUDPC

estimates (less than 300). Therefore, they were characterized as the adult plant resistant (APR), and /or partially resistant (PR) genotypes "slow rusters". This group has been mainly contained the eighteen elite mutant lines i.e.; Mut26 (9.33), Mut2 (21.00), Mut28 (21.00), Mut38 (21.00), Mut59 (21.00), Mut1 (24.50), Mut11 (28.00), Mut161 (28.00), Mut25(44.30), Mut166 (147.00), Mut161 (28.00), Mut68 (147.00), Mut65 (158.67), Mut64 (170.33), Mut49 (173.83), Mut132 (173.83), Mut3 (182.00) and Mut37 (224.00), as well as the only one wheat cultivar; Sk94(40.83).

Whereas, the second group contained wheat genotypes which revealed a considerable high estimates of AUDPC estimated rather than partially resistant ones (more than 300). Thus they were identified as "fast- rusters" or the highly susceptible genotypes. However, this genotype group found to be contained four mutant lines, and three wheat cultivars, as well as the check variety Morocco; they were; Mut142 (495.83), Mut199 (525.00), Mut44 (560.00), Gm11 (571.67), Sk93 (618.33), Mut99 (682.50), Sd12 (910.00) and Morocco (1178.33) respectively (Table 6). The previous study of **Macharia and Wanyera (2012)**, found that the wheat line; 102091 showed low value of AUDPC, while the wheat cultivars; Chozi and Duma showed high estimates of AUDPC during an epidemic stem rust in Kenya.

Table 6. Area under disease progress cu	rve (AUDPC) of 27 wheat genotyp	pes (22 mutant lines and 5 cultivar) in
M5 and M6 generations under fi	ield conditions during 2017/2018 a	and 2018/2019 growing seasons.

No.	1171 4	Area under disease progress curve (AUDPC)				
	Wheat genotypes —	M ₅ generation (2017/2018)	M ₆ generation (2018/2019)			
	a)Mutant line					
1	Mut.1	28.0	24.50			
2	Mut.2	28.0	21.00			
3	Mut.3	182.0	185.50			
4	Mut.11	28.0	28.00			
5	Mut.25	54.8	44.33			
6	Mut.26	2.3	9.33			
7	Mut.28	47.8	21.00			
8	Mut.31	150.5	147.00			
9	Mut.37	170.3	224.00			
10	Mut.38	4.7	21.00			
11	Mut.44	515.7	560.00			
12	Mut.49	112.0	173.83			
13	Mut.59	28.0	21.00			
14	Mut.64	170.3	170.33			
15	Mut.65	147.0	158.67			
16	Mut.68	344.2	147.00			
17	Mut.99	501.7	682.50			
18	Mut.132	123.7	173.83			
19	Mut.142	571.7	495.83			
20	Mut.161	42.0	28.00			
21	Mut.166	135.3	147.00			
22	Mut.199	565.8	525.00			
	b)Wheat					
	cultivars:					
1	Gemmeiza11	530.8	571.67			
2	Sids12	1061.7	910.00			
3	Sakha93	840.0	618.33			
4	Sakha94	70.0	40.83			
5	Morocco (check)	1167	1178.33			
	General mean	282.402	271.401			
	L.S.D. at 0.05	81.11	59.49			
	L.S.D. at 0.01	107.50	78.847			

d) Mean performance of mutant wheat genotypes:

1- Thousand Kernel Weight (TKW)

Mean performance for mutant genotypes for Thousand Kernel Weight is presented in Table (7). Results show that significant mean performance difference for thousand kernel weight were found among tested wheat genotypes in M5 and M6 generations. Thousand Kernel Weight measure was the best indicator to estimate the effect of stem rust on grain yield of bread wheat. Thousand kernel weights is the integral parameter of overall yield and differ from genotype to another.

Results presented in Table (7) recorded that thousand kernel weight ranged from 38.10 to 62.13gm and from 37.90 to 62.40gm for susceptible (Mut.44) to resistant (Mut.2) genotypes in M5 and M6 generations, respectively. The stem rust reduced the TKW of wheat genotypes by shriveling of wheat kernels on susceptible genotypes.

In the present field experiment, thousand kernel weights showed significant variation among the wheat genotypes. The disease pressure was greater at flowering and milk stage of the crop development and resulted significant impact on grain feeling process of few susceptible varieties. Infection of wheat stem by wheat stem rust affects the transport of assimilates to the developing kernel and results in shriveled kernel (Everts, et al., 2001).

2 - Number of grains/ spike (G/S)

Results presented in Table (7) showed that number of grains - spike ranged from 60.02 (Mut.44) to 108.62 (Mut.132) for susceptible to resistant genotypes in M5 generation and from 69.00 to 102.00 for susceptible (Sakha 93) to resistant (Mut.132) genotypes in M6 generation. The stem rust reduced the G/S of wheat genotypes by shriveling of wheat grains on susceptible genotypes.

Table 7. Mean performance of the 27 bread wheat genotypes (22 mutant lines and 5 cultivar) for number of grains/ spike and1000- kernel weight in M5 and M6 generations during 2017/18 and 2018/19 growing seasons.

No.	sons.	Number of a	grains/ spike	1000 kernel	weight (gm)
	Wheat genotypes	M ₅ generation (2017/2018)	M ₆ generation (2018/2019)	M ₅ generation (2017/2018)	M ₆ generation (2018/2019)
	a)Mutant line				
1	Mut.1	81.67	84.00	59.30	59.60
2	Mut.2	79.22	83.00	62.13	62.40
3	Mut.3	71.97	85.33	55.77	56.03
4	Mut.11	87.83	94.00	59.43	60.20
5	Mut.25	76.63	80.00	40.17	39.90
6	Mut.26	75.02	80.33	44.07	44.47
7	Mut.28	86.52	89.00	42.37	42.50
8	Mut.31	77.00	81.67	42.17	42.27
9	Mut.37	81.33	80.33	39.10	39.17
10	Mut.38	89.33	83.00	44.90	45.10
11	Mut.44	60.02	72.00	38.10	37.90
12	Mut.49	81.65	88.67	40.57	41.00
13	Mut.59	81.67	83.00	43.73	44.47
14	Mut.64	75.45	76.33	39.67	40.90
15	Mut.65	79.67	81.67	39.67	40.03
16	Mut.68	72.00	75.67	45.00	45.90
17	Mut.99	86.67	85.00	52.90	52.33
18	Mut.132	108.62	102.00	52.67	53.63
19	Mut.142	80.00	81.33	52.47	52.43
20	Mut.161	72.00	71.67	41.83	42.63
21	Mut.166	83.23	84.00	40.30	40.50
22	Mut.199	82.33	85.33	51.83	51.50
	b)Wheat cultivars:				
1	Gemmeiza11	74.13	78.00	52.80	54.43
2	Sids12	95.67	94.67	48.90	48.33
3	Sakha93	70.00	69.00	44.30	41.73
4	Sakha94	73.43	77.67	43.67	44.43
5	Morocco (check)	68.00	71.00	39.00	38.67
	General mean	79.679	82.136	46.647	46.758
	L.S.D. at 0.05	1.354	1.472	1.346	0.9912
	L.S.D. at 0.01	1.794	1.950	1.784	1.313

3 - Grain Yield

Yield is the most desirable and important parameter of crop plant. Grain yield vary from one genotype to another genotype due to genetic variations of crop plant. Grain yield depends on genetic potential of a genotype against biotic and abiotic stresses and overall performance of individual plant.

Results in M5 generation (2017/18) presented in Table (8) showed that the maximum grain yield of 19.71, 17.85, 17.71 and 17.70 (ardb/ fed) was obtained from Mut.11, Mut.31, Mut.68 and Mut.59, respectively. Whereas the Mut.11 and Mut.59 were resistant and the Mut.31 and Mut.68 were moderately resistant, which had better effect in reducing stem rust epidemics, increased grain yield and yield components, and showed higher economic benefits over other wheat genotypes.

Results in the M6 generation (2018/19) presented in Table (8) recorded that the maximum grain yield of 20.08, 19.56, 18.92 and 18.66 (ardb/ fed) was obtained from Mut.11, Mut.59, Mut.31 and Mut.2, respectively. The Mut.11, Mut.59 and Mut.2 were resistant for stem rust and Mut.31was moderately resistant. These mutant wheat genotypes had better effect in reducing stem rust epidemics, increased grain yield and yield components, and showed higher economic benefits over other wheat genotypes. From the result of research finding it is possible to reduce yield loss due to stem rust using relatively resistant mutant wheat genotypes Mut.11, and Mut.59 in both M5 and M6 generations. The result of research finding was similar with the finding of (Mebrate, et al., 2008) who reported that host resistance is the most economical and safest method for controlling the disease. He also reported that the highest grain yield of 3816.7, 3959, 3159, 3434, and 3624kg/ha-1 were obtained from the moderately susceptible varieties, i.e. Tate, Mangudo, Ude, Odda and Assasa, respectively. The highest grain yield was 3418, 3183 and 2995kg/ha-1 were obtained from susceptible varieties of Mukye, Mesebo, and Toltu, respectively. Majority of varieties showed low grain yield which represents that susceptibility to biological stress may be the cause of lower yield in these varieties. Similarly Singh, et al., (2008) reported that stem rust reduced yield irrespective of the type and level of resistance possessed by the wheat varieties. The effect of stem rust infection on grain yield losses of wheat genotypes possibly in consequence of the effect on the photosynthetic area of the top three leaves especially flag leaf, which shares with its sheath by about 75% in determining the grain weight.

4 - Biological Yield (ton/fed)

Mean performance values of the tested mutant bread wheat genotypes in M5 generation (2018/19) for biological yield shown in Table (8) indicates that biological yield (ton/fed) ranged from 4.60 to 7.44 (ton/fed) from susceptible (Morocco) to resistant (Mut.11) genotypes, respectively. The stem rust reduced the biological yield of wheat genotypes by shriveling of wheat biological yield on susceptible genotypes.

Mean performance of the 27mutant wheat genotypes in M6 generation (2018/19) detected for biological yield is presented in Table (8). Results showed that the biological yield (ton/fed) ranged from 5.10 to 8.60 (ton/fed) from susceptible (Morocco) to resistant (Sakha94) genotypes, respectively. The stem rust reduced the biological yield of wheat genotypes by shriveling of wheat biological yield on susceptible genotypes.

e) Phenotypic correlation coefficient studies:

Phenotypic correlation coefficient among three stem rust parameters and four yield traits over M5 and M6 generations was presented in Table (9).

Correlation coefficients has been mainly defined the level of relationship between two variables. It is valuable in plant breeding since it can show a foretelling association that can be exploited in practice, and it overs evidence about the relationship between several preferred traits. It overs a core concept of the association among various yieldcontributing traits, which is beneficial for plant breeders in choosing varieties having desired attributes or possessing desirable traits (**Ghafoor, et al., 2013 and Liu, 2016**).

Results in Table (9) indicated in general that grain yield (GY) and biological yield (BY) displayed negative and highly significant correlations with each of average coefficient of infection (ACI), area under disease progress curve (AUDPC), and final rust severity % (FRS%). Which, gave values of the relatively high significant negative values of correlation coefficient between grain yield and each of; ACI ($r = -0.694^{**}$), AUDPC ($r = -0.679^{**}$) and FRS % (r =-0.665**), respectively, and with biological yield were; ACI (r = -0.724**), AUDPC $(r = -0.606^{**})$ and FRS% $(r = -0.692^{**})$, respectively. On the other hand, highly significant positive correlation between grain yield and biological yield (r= 0.889**) and significant with grains/spike (r= 0.411*). Positive and highly significant correlation was observed between ACI with and AUDPC (r = 0.983^{**}), and FRS (r = 0.995^{**}) which means that genotypes high for ACI, AUDPC and FRS. Positive and highly significant correlation was found between FRS with AUDPC (0.984^{**}) .

Table 8. Mean performance of the 27 bread wheat genotypes (22 mutant lines and 5 cultivar) for grain yield and
biological yield in M5 and M6 generations during 2017/18 and 2018/19 growing seasons.

No.		Grain Yield	l (Ardb/ fed)	Biological Yield (ton/fed)			
	Wheat Genotypes	M ₅ generation (2017/2018)	M ₆ generation (2018/2019)	M ₅ generation (2017/2018)	M ₆ generatio (2018/2019)		
	a)Mutant line						
1	Mut.1	16.93	17.09	6.89	7.31		
2	Mut.2	17.59	18.66	7.21	7.68		
3	Mut.3	14.64	15.99	6.10	7.29		
4	Mut.11	19.71	20.08	7.44	7.79		
5	Mut.25	16.69	17.65	7.36	7.78		
6	Mut.26	16.02	17.34	6.43	7.52		
7	Mut.28	16.93	17.89	6.63	7.25		
8	Mut.31	17.85	18.92	7.31	7.78		
9	Mut.37	14.58	15.80	6.20	7.17		
10	Mut.38	16.27	16.78	6.70	7.10		
11	Mut.44	11.61	11.86	5.27	5.77		
12	Mut.49	17.31	18.34	7.05	7.79		
13	Mut.59	17.70	19.56	6.86	7.53		
14	Mut.64	16.53	16.54	6.99	7.27		
15	Mut.65	16.76	17.29	6.74	7.76		
16	Mut.68	17.71	18.51	6.87	7.43		
17	Mut.99	17.46	18.09	7.04	7.23		
18	Mut.132	17.16	17.60	6.60	6.78		
19	Mut.142	16.46	17.08	6.51	6.86		
20	Mut.161	15.93	16.35	6.89	7.03		
21	Mut.166	16.37	16.71	6.66	6.82		
22	Mut.199	16.19	16.08	6.53	6.76		
	b)Wheat cultivars:						
1	Gemmeiza11	13.89	13.87	6.57	6.83		
2	Sids12	11.59	11.88	5.03	5.21		
3	Sakha93	11.28	14.52	5.32	7.73		
4	Sakha94	15.63	15.90	6.61	8.60		
5	Morocco (check)	8.84	9.57	4.60	5.10		
	General mean	15.763	16.516	6.533	7.154		
	L.S.D. at 0.05	0.2136	0.1795	0.0518	0.518		
	L.S.D. at 0.01	0.2839	0.2373	0.0812	0.0564		
	enotypic correlation coe ead wheat genotypes (22						
Variable		AUDPC FF		TKW	BY		
ACI	1						
AUDPC	0.983**	1					

FRS	0.995**	0.984**	1				
G/S	-0.116	-0.161	-0.138	1			
TKW	-0.01	-0.058	0.010	0.411*	1		
BY	- 0.724**	-0.606**	-0.692**	0.131	0.182	1	
GY	-0.694**	- 0.679**	-0.665**	0.411*	0.297	0.889**	1

* and **denote significant differences at 5% and 1% levels, respectively. ACI = average coefficient of infection; AUDPC = area under disease progress curve; FRS% = final rust severity%; G/S= Number of grains/ spike; TKW=1000 Kernel Weight; BY= Biological yield; GY= Grain yield.

Conclusion

Mutation breeding considered to be one of the most superior methods or the best techniques, which have been employed to enhance, and / or increase genetic variation within wheat genotypes. This breeding method is necessary to create and obtained the possible elite or advanced wheat mutant lines having an adequate levels of adult plant resistance (APR) to rust fungi, combined with the desirable yield traits. Results obtained during this study revealed, in conclusion, that the nine wheat mutant lines i.e. Mut1, Mut2, Mut11, Mut25, Mut26, Mut28, Mut38, Mut59 and Mut161 having an adequate levels of adult plant resistance to stem rust, combined with some desirable vield traits or characters. These advanced mutant lines could be successfully used as the new and good sources of resistance to rust pathogens, especially stem rust, as well as they desirable yield traits. Also, the best mutant lines, could be introgressed into adapted Egyptian wheat cultivars; to develop durable resistance to stem rust in wheat. Furthermore, mutation induction should be facilitate the useful as of the best mutant lines both in stem rust disease resistance and drought tolerance, as well as high yielding, for further screening in the national breeding programs in Egypt.

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تقييم بعض طفرات قمح الخبز لمقاومة النباتات البالغة لصدأ الساق

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يعتبر مرض صدأ الساق في القمح من أهم الأمراض التي تهدد محصول القمح في مصر والعديد من دول زراعة القمح الأخري في العالم. تم تقييم اثنين وعشرين سلالة طفرية في الجيل الخامس والجيل السادس بالاضافة الي خمسة أصناف تجارية من قمح الخبز وهم جميزة 11 وسدس 12 وسخا93 وسخا94 وموروكو من أجل انتاجية عالية للمحصول ومقاومة جيدة لمرض صدأ الساق تحت الضغط المرضي في الحقل. تم استخدام ثلاثة مقاييس مرضية هامة وهي: شدة الإصابة النهائية للمرض (%FRS) ومتوسط معامل الإصابة (ACI) والمساحة الواقعة تحت منحني الإصابة المرضي (AUDPC) لتوصيف الانماط الجينية لمقاومة مرض صدأ الساق. كما تم تسجيل الصفات المحصولية للتراكيب الوراثية منحني الإصابة المرضي (AUDPC) لتوصيف الانماط الجينية لمقاومة مرض صدأ الساق. كما تم تسجيل الصفات المحصولية للتراكيب الوراثية منحني الإصابة. أظهر الصنف سخا94 وتسعة سلالات طفرية من قمح الخبز وهي السلالات الطفرية (, KM13, Mut2, Mut1, Mut2, Mut1, Mut2, الطفرية (, وهي الملالات الطفرية (, وهي المدلالات الطفرية (, ومي المداراثية, Mut3, Mut2, Mut1, Mut2, Mut3, Mut38, Mut59 محصول الحبوب والمحصول النيولوجي حيث قد اعطت قيما قاومة مبولة وجيدة لصدأ الساق حيث انها قد تميزت بإنخفاض في قيم كلا من محصول الحبوب والمحصول البيولوجي حيث قد اعطت قيما كانت علي المعنوية بين المقاييس الوبائية للمرض والصفات المحصولية وهي (*ACI, FRS, AUDPC محصول البيولوجي حيث قد اعطت قيما كانت علي المعنوية بين المقاييس الوبائية للمرض والصفات المحصولية وهي محصول الحبوب والمحصول البيولوجي حيث قد اعطت قيما كانت علي التوالي G/s (r= 0.619), وقد انتمح من النتائج السابقة انه يمكن أدخال محصول الحبوب والمحصول البيولوجي حيث قد اعطت قيما كانت علي التوالي G/s (r= 0.419), وقد التمنح من النتائج السابقة انه يمكن أدخال محصول الحبوب والمحصول البيولوجي حيث قد اعطت قيما كانت علي التوالي G/s (r= 0.619), ووقد التمحصولية وهي (, الحبوب والمحصول البوج ارتباط اليجابيا مع (*9898) و C/s ((الحاصة) و مراه (), وراه (), وراه (), وراه الموان والحفان في أدخال (الانماط الجينية أو السلالات الطافرة الماق وذات الصفات المحصولية الجيدة في برامج التربية حيث يتم تهجينها مع الأصناف الانماط الجينية أو السلالات الطافرة الماق ومي نفس الوقت تكون عالية في المي .