

EFFECT OF SOME CHEMICAL MUTAGENS ON SOME CHARACTERS OF  
THE M<sub>2</sub> AND M<sub>3</sub> PLANTS IN FIELD BEANS

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تأثير بعض المطفرات الكيماوية على بعض الصفات لنباتات الجيل الطفرى

الثانى والثالث فى القول البلدى

حسان دوام - فتحى هندوى - على الحصرى

قسم المحاصيل - كلية الزراعة ( المتوفية ) ، الزقازيق ( مشتهر )

ملخص البحث

عوملت ثلاثة أصناف من القول البلدى ( جيزة ٣ ، جيزة ٤٠٢ ، ٦١ / ٦١ / ٥٢٦ ) بأربعة تركيزات ( صفر ، ١٠٠ ، ٢٠٠ ، ٤٠٠ جزء فى المليون ) من المطفرين الكيماويين ( ايثيل ميثان سلفونات ومثيل ميثان سلفونات ) فى موسم ١٩٨٥/٨٤ وتم دراسة طفرات الكلوروفيل فى الجيل الثانى والثالث الطفرى فى موسمى ١٩٨٥/٨٦ ، ١٩٨٧/٨٦م كما درست صفات محصول البذور للنبات وعدد القرون على النبات ووزن ١٠٠ بذرة فى الجيل الثالث نتط فى موسم ١٩٨٧/٨٦ ويمكن ايجاز أهم النتائج المتحصل عليها فيما يلى :-

- ١ - زادت نسبة كل من العائلات والطرز الطفرة كنتيجة لزيادة تركيز المطفر الكيماوى ميثيل - ميثان سلفونات من صفر - ٤٠٠ جزء فى المليون فى صنفى جيزة ٣ ، ٦٦ / ٥٢٦ / ٦١ .
- ٢ - اختلفت الأصناف فى استجابتها لكل من نوع المطفر والتركيز .
- ٣ - كان تأثير المطفرين الكيماويين على نوعية الطفرات فى الجيلين الثانى والثالث متشابهة حيث تم الحصول على أعلى نسبة من طفرات الفرس يليها طفرات الزانناشم طفرات الالبيتو .
- ٤ - أدى استخدام المطفر الكيماوى ايثايل ميثان سلفونات الى زيادة عدد القرون للنبات ، ووزن ١٠٠ بذرة ومحصول البذور للنبات عن المطفر الكيماوى ميثايل ميثان سلفونات .

٥ - أدى زيادة تركيز المطفرات الكيماوية عن ١٠٠ جزء في المليون الى نقص المحصول ومكوناته .

٦ - نستخلص من هذه الدراسة أن صنف جيزة ٤٠٢ المعامل بالمطفر الكيماوى ايثايل ميثان سلفونات بتركيز ١٠٠ جزء في المليون كان أحسن المعاملات لاعطاء تباين وراثى كبير وهذا يمكن مربي النبات من الانتخاب للانتاجية العالية لمحصول الفول اللبلى .

#### ABSTRACT

Three varieties of field beans (Giza 3, Giza 402 and 61/536/66) were treated with four concentrations (0, 100, 200 and 400 ppm) of two chemical mutagens (Ethyl-methane sulphonate (EMS) and Methyl-methane sulphonate (MMS) ).

The percentage of both  $M_1$  and  $M_2$  mutated families and types were progressively increased as a result of increasing the concentration of EMS from Zero to 400 ppm in the two varieties Giza 3 and 61/536/66.

Varietal differences in response to the different mutagens i.e., EMS and MMS and their concentrations were found.

Comparing the effect of the two mutagens in both  $M_1$  and  $M_2$  generations, it was found that the most marked effect of the two mutagens were in giving viridis type (59.2% EMS and 58.8% MMS), xantha (31.6% EMS and 26.5% MMS) and albina (9.2% EMS and 14.7% MMS).

Using EMS mutagen increased the number of pods per plant, 100-seed weight and seed yield per plant than MMS mutagen did.

Increasing the concentration of chemical mutagens more than 100 ppm increased sterility percentage and subsequently decreased the yield and its components.

Giza 402 treated with EMS at 100 ppm concentration was found to be the best combination for higher productivity of field bean in this study.



## INTRODUCTION

Through preliminary research, it was established, which is the most efficient way to obtain economic mutations. Chemical mutagens may produce mutation spectra and mutation frequencies which are different from those induced by other mutagens i.e., irradiation.

Conventional breeding methods are laborious and lengthy, requiring several generations of crossing and breeding to obtain a certain goal. Consequently attempts have been made to utilize chemical mutagens to increase the pool of variability in various characters, hoping that such a method may shorten the period required to obtain improvement in these characters.

The main objective of the present investigation being to (i) estimate the potential value of EMS and MMS chemical mutagens on the frequency and spectrum of chlorophyll mutations in the second ( $M_2$ ) and third ( $M_3$ ) generations of faba bean, (ii) to evaluate the most promising high yielding families of  $M_2$  generation for yield and some of its components in  $M_3$  generation and (iii) to determine whether the studied varieties have a differential response to the two different chemical mutagens and their concentrations.

The experiments reported herein were carried out at Experimental Farm, Faculty of Agriculture, Minufiya University in the two successive seasons 1985/1986 and 1986/1987. The details of the experimental procedures are described by (Dawwam et al., 1986). All  $M_1$  plants surviving to seed maturity were separately harvested and individually threshed. All seeds of the  $M_2$  experiment were obtained from each  $M_1$  plant and sown in one single ridge 4 m. long and 60 cm. width, with seeds spaced 10 cm. a part within ridge. During seedling stage of  $M_2$ , chlorophyll mutations in each treatment were visually identified according to Gustafsson (1940). At maturity stage, seeds of each unmutated  $M_2$  families were collected and sown

in the same way in  $M_2$  generation. At seedling stage, each  $M_1$  family was visually examined for screening new chlorophyll mutations. In addition the promising high yielding plants of each of the twenty four treatments in  $M_2$  generation were taken and threshed in order to be sown in the next generation ( $M_3$ ).

In 1986/1987 season, these seeds were arranged in split-split plot design with three replications. At seedling stage, each  $M_3$  family was visually examined for screening new chlorophyll mutations. Twenty guarded plants were taken from each sub-plot for subsequent measurements i.e., number of pods per plant, 100 seed weight and seed yield per plant. The differences among means were tested using Duncan's multiple range test (Duncan, 1955). The means followed by the same letter are not statistically different at 5% level.

## RESULTS AND DISCUSSION

### A) Chlorophyll mutation in $M_2$ and $M_3$ generations:

According to Gustafsson (1940), the types of chlorophyll mutations obtained in this study can be classified into three groups V.Z. Albina, Xantha and Viridis.

Frequencies and percentages of both mutated families and types with chlorophyll mutations which obtained after different mutagenic treatments EMS and MMS in  $M_2$  and  $M_3$ , and the total percentages of mutated families obtained in both generations of the three varieties are shown in Table (1).

The percentages of both  $M_2$  and  $M_3$  mutated families and types were progressively increased as a result of increasing the concentration of EMS from Zero to 400 ppm in the two varieties Gzia 3 and 61/536/66. Whereas, the maximum percentages of both mutated families





and type were obtained from the treatment of 200 ppm EMS and MMS applied to both Giza 402 and 61/536/66, respectively. However, increasing concentration of EMS and MMS than 100 ppm decreased the percentage of mutated families and types in the other cases. Similar results were reported in other plant species with different chemical and physical mutagens (Gustafsson, 1940; Brock, 1965 and Gupta et al., 1969). Gustafsson (1940) added that a linear relationship between radiation dose and frequency of chlorophyll mutations was observed until 15 Kr, while doses higher than 15 Kr gave lower number of chlorophyll mutations. Brock (1965) found that mutation frequency was increased by increasing the dose of X-ray but with some saturation at higher doses in Trifolium subterreneum. Gupta et al. (1969) showed that in most cases there was a linear correlation between dose rate and chlorophyll mutation frequency. These authors attributed the reduction in mutation frequency at higher mutagen doses to the saturation being happened in the mutational event. Yanaguchi (1962) considered that frequency of chlorophyll mutation did not increase in linear relationship with dose of X-ray because the high doses cause elimination of mutated genes within the killed, of these high doses, cells carrying these mutated genes, and also because of the low capacity of growing the homozygous mutants.

Comparing the ability of different mutagenic treatments on the basis of frequency of chlorophyll mutations obtained in both generations (Table 1), it can be shown that EMS gave the highest mutation rate than MMS did.

Varietal differences in response to the different mutagens and their concentrations were found (Table 1). Varietal differences in sensitivity and mutability has been indicated early by many authors (Gichener et al., 1968; Ghafoor, 1974 and El-Hosary, 1977).

To compare the effect of both mutagens on the three studied varieties concerning the spectrum of chlorophyll mutations, the frequency of each type obtained in all treatments in both generations ( $M_1$  and  $M_2$ ) will be taken together (Table 2).

No varietal differences in response to the two different mutagens i.e., EMS and MMS could be detected as regards to the spectrum of chlorophyll mutations (see Table 2). These results are in agreement with the previous results obtained by El-Shouny and El-Hosary (1983) in field bean and disagreed with those obtained by Marki and Bianu (1970) in flax.

Comparing the effect of the two mutagens in both  $M_1$  and  $M_2$  generations it was found that the most marked effect of the two mutagens were in giving viridis type (59.2% EMS and 58.8% MMS), xantha (31.6% EMS and 26.5% MMS) and albina (9.2% EMS and 14.7% MMS) (see Table 3). Similar pattern was previously found by L'undquist and Wettstein (1962) in barley and El-Hosary (1977) in field bean.

## B) Yield and some of its components in the $M_1$ generation:

### 1- Varietal effect:

Comparing the average of the three tested varieties, it was found that they significantly differed in number of pods per plant and 100-seed weight. Giza 402 and 61/536/66 had the highest values of 100-seed weight and number of pods/plant, respectively. Though, the three examined varieties did not significantly differ in seed yield/plant, and that may be due to the fact that, both number of pods/plant and seed index are not the only components of the yield in faba bean (see Table 4).



Table (2) : Effect of varieties on spectrum of chlorophyll mutation induced in  $M_2$  and  $M_3$  generations .

Trait Variety	Total E.F	Total M.F	Mutation spectrum in both generations						M.T.
			Albina		Xantha		Viridis		
			No.	%	No.	%	No.	%	
Giza 3	188	28	4	12.5	9	28.13	19	59.38	32
Giza 402	182	29	3	7.69	12	30.77	24	61.54	39
61/536/66	189	37	7	17.50	10	25.00	23	57.50	40

Table (3) : Effect of mutagenic type on spectrum of chlorophyll mutation in  $M_2$  and  $M_3$  generations .

Trait Mutagen	Total E.F	Total M.F	Mutation spectrum in both generations						M.T.
			Albina		Xantha		Viridis		
			No.	%	No.	%	No.	%	
RMS	301	62	7	9.2	24	31.6	45	59.2	76
MMS	258	32	5	14.3	10	28.6	20	57.1	35

E.F. = Examined families .

M.F. = Mutated families .

M.T. = Mutated types .



## 2- Effect of mutagen type:

The data in Table (5) show that using EMS increased the number of pods per plant, 100-seed weight and seed yield/plant more than MMS did. This results could be due to that the effectiveness and efficiency of ethylating agents (EMS) were higher than methylating agents (MMS) in increasing the variability. This may be related to the role of different alkali groups that passed by each mutagen.

## 3- Effect of the concentration of chemical mutagens:

The data in Table (6) indicate in general that, the concentration of 100 ppm gave the highest values of the three studied traits in comparison with the other two concentrations. However, the concentration of 100 ppm exhibited less seed yield/plant and seed index than the control but, the decrease in seed yield/plant was not significant. So, it could be suggested that increasing mutagen concentration increased the sterility percentage and chromosomal aberration in the  $M_1$ -generation and consequently decreased the three examined characters.

## 4- Interaction effect:

### a) Interaction of variety with chemical mutagen:

Varietal differences in response to the chemical mutagens were found to be significant in only 100-seed weight (see Table 7). Giza 402 variety exhibited the highest seed index when treated with EMS mutagen.

### b) Interaction of variety with mutagen concentration:

Varietal differences in sensitivity to the mutagenic concentration were found to be only in 100-seed weight (Table 8). Giza 402 when treated with 100 ppm produced the heavy seed index.

Table (4) : Mean values of seed yield and some yield components for M<sub>3</sub>-generation of three field bean varieties .

Trait Variety	No. of Pods/plant	100-seed weight (gm)	Seed yield / plant (gm)
Giza 3	20.09 b	65.62 b	35.76 a
Giza 402	20.12 b	67.34 a	34.43 a
61/536/66	21.99 a	63.99 c	34.64 a

Table (5) : Effect of chemical mutagens on seed yield and some yield components of M<sub>3</sub>-generation of field bean .

Trait Mutagen	No. of Pods/plant	100-seed weight (gm)	Seed yield / plant (gm) .
EMS	22.02 a	66.91 a	38.23 a
MMS	19.44 a	64.39 b	31.65 b

Table (6) : Effect of mutagenic concentrations on seed yield and some yield components of M<sub>3</sub>- generation of field bean .

Trait Con- centration (ppm)	No. of Pods/plant	100-seed weight (gm)	Seed yield / plant (gm) .
0	22.82 a	67.99 a	39.14 a
100	22.96 a	65.88 b	37.04 a
200	18.32 b	65.06 b	31.26 b
400	18.84 b	63.67 c	32.34 b



Table (7) : Effect of the variety X mutagen interaction on seed yield and some yield components of M<sub>3</sub>-generation of field bean .

Mutagen Variety	No. of pods/plant		100-seed weight (gm)		Seed yield/plant (gm)	
	EMS	MMS	EMS	MMS	EMS	MMS
Giza 3	21.35 a	18.83 a	65.58 b	65.67 b	37.65 a	33.87 a
Giza 402	21.49 a	18.75 a	68.68 a	66.00 b	38.90 a	29.96 a
6L/536/66	23.23 a	20.75 a	66.48 b	61.50 c	38.14 a	31.13 a

Table (8) : Effect of the variety X mutagenic concentration interaction on seed yield and some yield components of M<sub>3</sub>-generation of field bean .

Concent- ration (ppm)	No. of pods/plant				100-seed weight (gm)				Seed yield/plant (gm)			
	0	100	200	400	0	100	200	400	0	100	200	400
Giza 3	17.50 <sup>a</sup>	21.77 <sup>a</sup>	22.00 <sup>a</sup>	19.09 <sup>a</sup>	64.33 <sup>de</sup>	65.22 <sup>c-e</sup>	66.99 <sup>bc</sup>	65.95 <sup>b-d</sup>	29.83 <sup>a</sup>	39.56 <sup>a</sup>	39.37 <sup>a</sup>	34.28 <sup>a</sup>
Giza 402	18.67 <sup>a</sup>	22.74 <sup>a</sup>	23.34 <sup>a</sup>	15.75 <sup>a</sup>	65.33 <sup>c-e</sup>	71.22 <sup>a</sup>	67.82 <sup>bc</sup>	65.00 <sup>c-e</sup>	34.50 <sup>a</sup>	40.13 <sup>a</sup>	35.59 <sup>a</sup>	27.51 <sup>a</sup>
6L/536/66	20.33 <sup>a</sup>	23.97 <sup>a</sup>	23.53 <sup>a</sup>	20.13 <sup>a</sup>	61.33 <sup>f</sup>	67.55 <sup>b</sup>	63.18 <sup>ef</sup>	64.22 <sup>de</sup>	32.65 <sup>a</sup>	37.71 <sup>a</sup>	36.17 <sup>a</sup>	31.99 <sup>a</sup>

c) Interaction of mutagenic type with the concentration:

Significant interaction of the mutagenic type and their concentrations was found for number of pods/plant, 100-seed weight and seed yield/plant (Table 9). The 100 ppm of EMS produced the highest mean values of the three investigated traits, revealing that this concentration produced high genetic variability in these three traits. However, the concentration of 400 ppm of MMS exhibited the lowest ones.

d) Effect of interaction between variety, type of mutagen and concentration:

From the analysis of variance, it was found that the interaction of variety with mutagenic type and concentration was significant in the three studied traits of the M<sub>1</sub> generation (Table 10). Giza 402 variety gave the highest values of the three examined traits when treated with 100 ppm of EMS. However, the lowest values of both number of pods/plant and seed yield/plant were given by Giza 402 when treated with 400 ppm of MMS mutagen. The variety 61/536/66 exhibited the lowest value of 100-seed weight when treated with 200 or 400 ppm of MMS mutagen. Similar results were obtained by Dawwam et al. (1986).

From the previous results, it could be concluded that the EMS mutagen was found to be more effective in inducing mutations in field bean. That may enable the breeder to select desirable genotypes which could be used as new varieties or in faba bean breeding programmes to improve the crop.

REFERENCES

- Brock, R.D. (1965). Response of Trifolium subterraneum to X-rays and thermal neutrons. Rad. Boty. 5: 543-555.



Table (9) : Effect of mutagen X concentration interaction on seed yield and some yield components of M<sub>3</sub>-generation of field bean .

Concentration (ppm)	No. of pods/plant			100-seed weight (gm)			Seed yield/plant (gm)					
	0	100	200	400	0	100	200	400	0	100	200	400
Mutagen												
EMS	18.83	25.39	23.35	20.53	63.66	68.54	68.54	68.43	32.34	43.44	40.86	36.29
MMS	18.83	20.26	22.56	16.11	63.66	67.44	63.33	63.11	32.34	34.83	33.22	26.22

Table (10) : Effect of the variety X mutagen X concentration interaction on seed yield and some yield components of M<sub>3</sub>-generation of field bean .

Concentration	No. of pods/plant			100-seed weight (gm)			Seed yield/plant (gm)					
	0	100	200	400	0	100	200	400	0	100	200	400
Concentration												
Variety												
Giza 3	17.50	21.90	25.00	21.00	64.33	62.77	67.97	67.33	29.83	38.97	46.74	37.05
Giza 402	18.67	17.67	25.67	13.50	65.33	70.00	64.33	64.67	34.50	32.83	32.33	20.17
61/536/66	20.33	21.50	23.00	18.17	61.33	64.67	59.67	60.33	32.65	31.50	33.33	27.00

- Dawwam, H.A.; A.A. El-Hosary and S.M. Abdel-Aal (1986). Effect of some chemical mutagens on yield and its components of the  $M_1$  plants in some varieties of field beans (Vicia faba, L.). Minufiya J. Agric. Res., 11: 63-80.
- Duncans, D.B. (1955). Multiple range and Multiple F. tests. Biometrics, 11: 1-42.
- El-Hosary, A.A. (1977). Effect of some chemical and physical mutagens on Vicia faba, M.Sc. Thesis, Fac. of Agric., Ain-Shams Univ., Egypt.
- El-Shouny, K.A. and A.A. El-Hosary (1983). Effect of some chemical and physical mutagens on Vicia faba, L. IV. Spectrum of chlorophyll mutations in  $M_2$  and  $M_3$  generations. First Conf. of Agron. 85-94 (Egypt).
- Ghafoor, A. (1974). The effect of ethylmethane sulfate treatment on floret sterility and chlorophyll mutation rate in barley. Rod. Bot., 14: 349-352.
- Gichener, T.; L. Ehrenberg and C.A. Wachtmeister (1968). The mutagenic activity of B-hydroxyethyle methone sulfonats, B-methoxy ethylemethan sulfonate, and diethyl 1,3-propanedi-sulfonate. Hereditas Lund. 252-262.
- Gupta, M.; M.P. Singh, and C.S. Kalla (1969). Frequency and spectrum of induced chlorophyll mutation in Triticum dicoccum. Wheat Inform. Serv., Kyoto No. 29: 4-6.
- Gustafsson, A. (1940). The mutation system of the chlorophyll apparatus. Sundo. Univ., Arakr. Avd., 36, PP. 1-40.
- L'undquist, U. and D. Von Wettstein (1962). Induction of mutants in barley by ionizing radiations and chemical mutagens. Hereditas, 48: 242-362.
- Marki, A. and M. Bianu (1970). Gamma rays and EMS induced mutations of flax (Linum usitatissimum, L.) Genetika: 24-28.
- Yanaguchi, H. (1962). The chimaeric formation in an  $X_1$  panical after irradiation of dormant rice seeds. Faculty of Agriculture, Univ. of Tokyo, Japan. Rod. Bot., 2: 71-77.