IV. RESULTS AND DISCUSSION

Effect of different doses of gamma rays and sodium azide (SA) and some combinations between them on two faba bean varieties to study some growth, yield chlorophyll mutation rate, spectrum in the, M_1 generation and to induced genetic variability for select some improved type which have useful agricultural properties .

Results obtained in this investigation will be illustrated for each generation separately.

1. First mutagenic generation (M_1).

A- Analysis of variance and mean performance:

The analysis of variance for ten studied characters in the M_1 generation is presented in Table (1). Significant mean squares due to cultivars were detected for all traits except flowering date, number of branches, pods and seed yield / plant. While mutagenic treatments had highly significant differences in all characters under study except 95 % of pods maturity. Only plant height, number of pods / plant, seed yield/plant and survival plant percentage were significant in the interaction between cultivar and mutagenic treatments. These results in agreement with many authors**, Krausse (1983), Filippetti and Marzano (1984a), Filippetti and Depace (1986), Dawwam *et al.* (1986), Kumari *et* (1993), Rabie *et al*. (1996), Kumari (1996), Podlesny *et al*. (2001) and Joshi and Verma (2004).**

A.1. Varietal performance

The differences of mean values between the two faba bean cultivars (Misr 1 and Giza 429) regarding the ten studied traits are presented in Table (2), from the data obtained in this Table, it could be noticed that Giza 429 gave significantly the highest mean values for all traits except maturity date, 100 – seed weight and survival plants, while Misr1 recorded the highest mean values for the exceptional traits. The high seed yield / plant of Giza 429 cultivar could be attributed to the high number of seeds and pods / plant and number of seeds / pod.

From these results it could be concluded that, the cultivar Giza 429 produced high seed yield and early maturity date. Faba bean varieties have differences in mutagenic sensitivity; similar results were also obtained by **El-Shouny and El-Hosary (1983a), Filippetti and Depace (1986)** who reported that the minor cultivar was more sensitive to lower dose of gamma rays than major cultivar.

Mean square presented in Table (1) indicated that highly significant differences among faba bean cultivars, mutagenic treatments and there interactions were found for survival plant percentage. But it is very clear that Giza 429 was more sensitive than Misr 1 which percentage of survival plant of the two cultivars was 36.86 and 53.37 %, respectively Table (2), **Kumari (1996)** detected that faba bean genotype (VH82-1) was more sensitive to mutagenesis and there plants were less survival.

A.2. Mutagenic treatments.

Data presented in Table (3) showed the effect of mutagenic treatments (physical, chemical and their combinations) on mean performance of faba bean as combined over the two cultivars in M_1 generation. Data indicated that all faba bean characters were significantly affected by mutagenic treatments except number of days to 95 % pods maturing. number of days to 50 % flowering increased by increasing gamma rays or SA concentrations and their combination. The latest one was treatment with 80 Gy (60.0 days) while untreated populations (control) were earliest and flowered after 54.0 days (mean of the two cultivars).

With respect to plant height, it is very clear that faba bean treated by 80 Gy and its combinations with SA (0.001% or 0.002%) were significantly the shortest plants (96.8, 95.08 and 94.08 cm, respectively). On the other side, plant height of faba bean was less affected by treatments 40 Gy or SA and their combinations compared with control (107.3 cm) except 40 Gy + 0.002% SA which was 104.14 cm. Also, number of branches /plant was decreased in trend of increasing dose or concentration mutagenic, where the control was 3.35 and treatments 80 Gy + 0.002% SA and concentration of 0.002% SA had the less number of branches / plant 2.92 and 2.86, respectively.

For both number of pods and seeds / plant, the same effects were detected on faba bean plants due to applied of physical and chemical mutagenic treatments. Number of pods and seeds / plant decreased due to gamma rays effect and produced 15.97, 15.3 and 43.34, 41.47 for 40 and 80 Gy,

respectively. But the decreasing was bigger as affected by SA (14.4, 14.36 and 43.2, 43.13, for number of pods and seeds/plant treated by 0.001%, 0.002% SA, respectively). While faba bean exposed to gamma rays doses and treated by SA concentrations, had very low number of pods and seeds/plant were produced, treatments 40 Gy + .002% SA and 80 Gy + 0.002% SA produced lowest number of pods / plant (13.01) and number of seeds / plant (35.58).

The differences in number of seeds/pod and 100 – seed weight were little but significant, number of seeds/pod ranged from 3.24 for untreated faba bean to 2.76 for 80 Gy + 0.002% SA. While 100 - seed weight ranged from very heavy one (control 83.92g) to 73.75g in treatment 80 Gy + 0.001% SA, which faba bean exposed to 80 Gy and its combination produced slight little seeds 78.25, 73.75 and 78.44g for 80 Gy, 80 Gy + 0.001% SA and 80 Gy + 0.002% SA, respectively.

Faba bean seed yield /plant significantly affected by mutagenic treatments and reduced in seed yield/plant associated with increasing in doses or concentrations of mutagenic treatments or their combinations. For example treatments 40 Gy + 0.002% SA and 80 Gy + 0.002% SA produced 30.36 and 31.67g, respectively compared with control 39.15g. On the other side, treatments 40 Gy and 0.01% SA were less effected and produced 37.89 and 36.12g, respectively. These results were in agreement with **Shamsi and Sofajy (1980), Dawwam *et al.* (1986), Khan (1988), Dhole *et al*. (2003) and Maheshwari *et al*. (2003)** who subjected soybean or field bean with gamma

radiation and found in M_1 and M_2 increasing variability for most quantitative characters studied.

Data of survival plants percentage presented in Table (3) showed that chemical more affects than gamma rays and lower doses or concentrations increased survival percentage than higher doses or concentrations. In this study untreated faba bean gave 98.42% survival plants while 40Gy, 80Gy, 0.001% SA and 0.002% SA gave 42.43, 40.07, 36.53 and 33.76, respectively. Other wise their combinations were lower than gamma alone and the lowest survival (35.25 %) was found in 80 Gy + 0.002% SA. **Kumar *et al.* (1993)** found that individual application of DES on faba bean was less toxic than gamma irradiation or the combined application. **Kumari (1996)** add that plant survival decreased with increased gamma radiation dose either alone or in combination with EMS.

A.3. Interaction effects.

Table (4) showed the effect of gamma rays (40 and 80 Gy), sodium azide (0.001% and 0.002%) and some combinations on mean performance of faba bean cultivars (Misr 1 and Giza 429) in M_1 generation. Data indicated that insignificant differences among interaction of mutagenic treatments and faba bean cultivars were found in number of days to 50 % flowering, 95 % of pods maturing, branches and seeds/plant, seeds/pod and 100-seed weight. On the other side, plant height, number of pods/plant, seed yield/plant and survival plant percentage showed highly significant differences. For plant height Giza 429 (114.35 cm) was taller than Misr 1 (108.06 cm) so highest reduction in the height was recorded in Misr 1 after exposed to

80 Gy and treated to 0.002% SA or 0.001% SA (90.56 and 93.88 cm, respectively) followed by exposed Misr 1 to 80 Gy (96.19 cm) and Giza 429 exposed to 80 Gy + 0.001% SA (96.28 cm).

With respect to number of pods/plant Misr 1 and Giza 429 produced pods 16.78 and 16.08, respectively, while Misr 1 with 80Gy + 0.002% SA produced the lowest number of pods 11.71. On the other side treatments 40 Gy in Misr 1 and 0.001% SA in Giza 429 were less affected and produced 16.23 and 16.03, respectively.

Seed yield/plant (g) presented in Table (4) indicted that untreated produced highest seed yield 39.31 and 38.97 g for Misr 1 and Giza 429 , respectively followed by Misr 1 treated by 40 Gy and Giza 429 treated by 0.001 SA (38.66 and 38.19, respectively). However treatments Misr1 with treated 40 Gy + 0.001% SA and 40 Gy + 0.002% SA were the highly effected plants which produced lowest seed yield, 27.28 and 27.62g, respectively.

These results in agreement with those **Atia (1981), Voica *et al*. (1982) and Krausse (1983)** found that Sodium azide caused a greater reduction in stem length, number of pods/plant, seeds/plant, 100-seed weight and seed yield/plant than (NMU) in soybean M_1. **Ciftci *et al*. (1994)** showed that, different doses caused significant and negative effects on yield and yield components in M_1 plants.

Survival plants calculated as percentage of number of podded plants at maturing stage to number of seedling and this explain the harmful of mutagenic treatments on faba bean plants and these may be attributed to the adverse effect of mutagenic on

pollen fertility. In this study Giza 429 with treatments 0.002% SA, 80 Gy + 0.002% SA and 0.001% SA were the most effected treatments Table (4) and lost high number of plants before maturing which recorded survival plants 25.31, 27.50 and 28.00 %, respectively. These results in agreement with those, **Filippetti and Marzano (1984b), Filippetti and Depace (1986), Kumari *et al*. (1993), Ciftci *et al.* (1994), Kumari (1996) and Solanki and Sharma (2001).**

2. Second mutagenic generation (M_2).

A. Analysis of variance and mean performance:

The analysis of variance for nine studied traits following the nine mutagenic treatments (physical and chemical mutagen and same combinations) on the two faba been varieties in M_2 generations was presented in Table (5). Varietal differences in response to mutagenic treatments were found significant for all the studied traits except number of days to 50 % flowering and 95 % pod maturity and number of seeds/pod. While mutagenic treatments were highly significant for number of days to 95 % pods maturity, plant height, number of pods, number of seeds, seed yield/plant and 100 – seed weight. The interactions between cultivars and mutagenic treatments were significant for number of days to 95 % pods maturity, plant height, number of seeds/plant, 100 – seed weight and seed yield/plant. similar results were also obtained by **El-Shouny and El-Hosary (1983a), Krausse (1983), Lidanski *et al.* (1984), Krausse (1985), Filippetti and Depace (1986), Rajput (1987), Dawwam *et al*. (1988), Mehetre *et al.* (1994), Joshi and Verma (2004).**

A.1. Varietal performance

The mean performance of the two cultivars, Table (6) showed that Giza 429 cv produced the highest values for all studied characters except plant height and 100 – seed weight. It could be concluded that these variation in the results are quite expected since the tested faba bean cultivars had some differences in their genetic structure. High seed yield in Giza 429 may be attributed to increase in number of pods and seeds / plant.

A.2. Mutagenic treatments.

Mean performances of mutagenic treatments are presented in Table (7). All mutagenic treatments affects significantly on all traits except number of days from sowing to 50 % flowering, number of branches and seeds/pod. The number of days from sowing to 95 % pods maturity insignificantly increased by increasing gamma – rays from 40 Gy to 80 Gy. The numbers of days to 95 % pods maturity were 148.01 and 148.76 in the same order. Also, the effect of SA showed the same trend. In addition, the combination of gamma – ray and SA gave insignificantly effects compared with gamma – ray or SA when used alone. Generally, the SA gave the earliest than other treatments.

For plant height, the effect of 40 Gy, 40 Gy + 0.001% SA, 40 Gy + 0.002% SA and 80 Gy gave significantly increased plant height compared with control. However other mutagenic treatments expressed significantly decreased highest of plant. The treatment 80 Gy + 0.001% SA gave the lowest value of plant height, however, 40 Gy + 0.001% SA gave the highest

ones. The depressing effect of high mutagenic doses plant height may by attributed to the active disturbances some enzymes involved in the synthetic of growth. Similar decrease in plant height was previously obtained by **Muhammad and Bashir (2003), Metwally *et al*. (2004), Joshi and Verma (2004) El-Gazar *et al*. (2004) and Wani and Samiullah *et al.* (2004)**. The highest mean values for number of pods, number of seeds and seed yield/plant were recorded with treated by 40 Gy + 0.001% SA followed by 40 Gy and then by 40 Gy + 0.002% SA. On the other hand, the treatments of 80 Gy + 0.002% and 0.001% SA gave the lowest ones for these traits Table (7). These results were agreements with the previously results obtained by **Filippetti and Depace (1986), Goranova and Aleksieva (1986) Joshi and Verma (2004) and Samiullah *et al.* (2004).**

The effect of treatments of 0.001% SA and 80 Gy 0.001% SA expressed significantly heavier 100 – seed weight. However, the treatments 40 Gy + 0.001% SA recorded the lowest one. Generally, the reduction in seed yield and some of its components with increasing gamma rays (40 Gy to 80 Gy) or combination between gamma rays + SA may be due to the reduction in fertility which may by attributed to chromosomal aberrations or attributed to physiological damage. It may be also attributed to the inhibiting effect of gamma – rays on cell division and consequently on the DNA replication. However, the low concentrations of SA may effect the activity of certain enzymes involved in the synthesis of endogenous hormones, but the mechanism involved is still not clear. Similar results were obtained by **Filippetti and Depace (1986), Goranova and**

Aleksieva (1986) Joshi and Verma (2004) and Samiullah *et al.* (2004).

High number of varieties of different crops that were developed by mutagenesis has been released approved for cultivation. The polygenic characters *Viz* high seed yield early maturity, plant type, quality characters, seed quality, abiotic stress and biotic resistance have been improved by mutagenesis. These findings are enough in the crop improvement.

A.3. Interaction effects.

The interactions between mutagenic treatments and faba bean cultivars in M_2 generation are presented in Table (8). Insignificant interaction between mutagenic treatments and faba bean cultivars were found in 50 % flowering, number of branches/plant, pods/plant and seeds/pod, while significant interaction was showing in 95 % of pods maturing, plant height, number of seeds/plant, 100 – seed weight and seed yield/plant. For 95 % of pods maturing untreated plants were later than mutagenic treatments, while later treatment was Giza 429 (151.20 days) and earliest one was Misr 1 treated by 0.001% SA (144.96 days) and 80 Gy + 0.002% SA (146.09 days).

Mean performance of plant height (cm) detected that the tallest plants were found in cultivar Misr 1 exposed to 40 Gy + 0.001% SA and 40 Gy only with mean values 132.60 and 131.65 cm, respectively. On the other side shortest plants were showed in Misr 1 with treated by 80 Gy + 0.002% SA (101.50 cm) and Giza 429 treated by 0.002% SA (101.35 cm).

Number of seeds/plant showed significant interaction among cultivars and mutagenic treatments. The highest numbers

of seeds (30.82, 30.47 and 29.22) were found in treatments Giza 429 with 40 Gy + 0.01 SA, Misr 1 with 40 Gy and Misr 1 with 40 Gy + 0.001% SA, respectively. While the lowest numbers (16.11, 16.51 and 16.63) were found in cultivar Misr 1 with treatments 80 Gy + 0.002% SA, only 0.002% and 80 Gy + 0.001% SA, respectively.

With respect to 100-seed weight, the heaver seeds (82.22 and 81.43 g) were detected in cultivar Misr 1 (control) and with 40 Gy, respectively. On the other side, the little weight of seed (100 – seed weight is 72.43 and 72.85g) was found in Giza 429 as control (untreated) or when exposed to 40 Gy Gamma rays, respectively.

High seed yield / plant produced by treatments faba bean by Misr 1 with 40 Gy and Giza 429 with 40 Gy + 0.001% SA recorded 23.68 and 23.55 g /plant, respectively. While the lowest seed yield/plant (12.36, 13.25, 13.25, 13.36 g) were found in cultivar Misr 1 with treatments 80 Gy + 0.002% SA, control, 80 Gy + 0.001% SA and 0.001 SA%, respectively. The same tread was obtained by many author in different crops, in faba bean **Atia (1981)** reported that application of IAA and SA tended to increase faba bean plant height. **Lidanski *et al.* (1984), Dawwam *et al.* (1986) and Dawwam *et al* (1988)** in soybean **Rajput (1987)** found that mean values of the yield components increased in M_2 as compared to controls. In Mung bean, **Khan (1988)** reported that mean values of Number of pods, 100-seed weight and seed yield/plant increased in the M_2 as compared to the control. While **Joshi and Verma (2004)** showed remarkable increase in the mean pod length and 100-seed weight (in M_2 and

M_3 faba bean generations) with slight reduction in plant height, number of seeds and pods/plant.

B: Chlorophyll mutation:

During the season 2005 / 2006, all observations on M_2 plants were taken during growth period from germination period to complete ripening. All plants were screened to chlorophyll mutation and data of all traits i.e. flowering and maturity dates, plant height, number of branches/plant, number of seeds/plant, number of seeds/pod, 100 – seeds weight and seed yield/plant were determined separately in each treatments to determine the genetic variance, heritability and genetic advance.

Frequency of chlorophyll mutated forms in M_2 was determined in each treatment by three methods:

1- Percentage of M_2 mutated plants to total M_2 plants,

2- Percentage of M_2 mutated families to total M_2 families,

3- Percentage of M_2 mutation types to total M_2 families,

In this study, variations of chlorophyll pigments were not found in the first mutagenic generation. Chlorophyll mutations were appeared and were screened. In the second generation, most of them were lethal in the seedling stage, while some of them continued their growth and survived till ripening stage.

Chlorophyll mutations obtained in this investigation were classified according to **Gustafsson (1940)** to: albina (A), xantha (X) and viridis (V).

Data on chlorophyll mutations induced in M_2 after different mutagenic treatments in separate varieties and over them are presented in Tables (9, 10, 11 and 12). Frequency of

mutated plants, mutation families and mutation types after different mutagenic treatments was estimated.

B1. Percentages of mutated plants in M_2 generations:

Percentages of mutated plants obtained in the M_2 as affected by mutagenic treatments over two varieties are tabulated in Tables (9 and 10).

B1.1. Mutagenic treatments:

B1.1.1. Gamma rays

The percentages of total M_2 mutated plants increased by increasing radiation dose from 40 Gy to 80 Gy. These percentages were 0.0, 2.01 and 2.23 % for control, 40 Gy and 80 Gy, respectively Table (9).

B1.1.2. Sodium azide (SA) treatments:

The percentage of SA, was found that the percentages of mutated plants increased by increasing concentrations of SA from 0.001 to 0.002.

These percentages were 2.17 and 2.98% for 0.001 and 0.002 SA concentrations, respectively.

B1.1.3. Combination effects.

From Table (9) it is shown that combination of gamma – rays (40 Gy and 80 Gy) and each of SA (0.001% and 0.002% SA concentrations) gave higher percentage of M_2 mutated plants with chlorophyll deficiencies than the same dose of gamma – ray or SA when it used alone. These percentages were 2.24, 2.22, 3.06 and 3.26 in the case of 40 Gy + 0.001% SA, 40 Gy + 0.002% SA, 80 Gy +0.001% SA and 80 Gy + 0.002% SA, respectively.

B1.2. Varieties

Means of the two faba bean varieties for all traits in M_2 are presented in Table (10). It is clear that the two varieties clearly differed than each other in their reaction over all mutagenic treatments. Thus, it is shown from this Table that the variety Giza 429 gave less percentages of mutated plants with chlorophyll mutation in the M_2 generation than the second variety Misr 1 over all treatments.

B1.3. Interaction effects

From Table (10), it is clear that the two varieties clearly differed than each other in their reaction to different mutagenic treatments. Thus it is shown from this Table (10) that variety Giza 429 gave less percentage of mutated plants with chlorophyll mutation in the M_2 than Misr 1 after seed treatments with all physical, chemical mutagenic and combinations between them except 40 Gy + 0.001% SA and 0.002% SA treatments whereas the Giza 429 gave the highest percentages of mutated plants.

B2. Percentages of mutated families in M_2:

Percentages of mutated families in M_2 as affected by mutagenic treatments, varieties and interaction between them are tabulated in Tables (11 and 12).

B2.1. Mutagenic treatments:

B2.1.1. Gamma rays:

The percentages of M_2 mutated families increased by increasing radiation dose from 0.0 to 80 Gy. These percentages

were 0.00, 23.93 and 28.99 % for controls, 40 Gy and 80 Gy, respectively.

B2.1. 2. Sodium azide (SA) treatments:

The percentages of M_2 mutated families increased by increasing SA % concentrations from 0.001 % to 0.002 % SA . Percentages of mutated families were 0.002, 26.8 % and 33.33 % for control, 0.001 and 0.002 % SA, respectively.

B2.1. 3. Combination effects:

From Table (11) it is shown that combination of gamma – rays (40 Gy) and each of sodium azide 0.001% and 0.002 % gave higher percentages of M_2 mutated families with chlorophyll deficiencies than the same of gamma – rays when it was used alone but did not surpass the treatment of SA in the same concentration when it was used alone. These percentages were 23.93, 26.80 and 33.33 % in the case of 40 Gy and (0.001% and 0.002 % SA), separately and 25.58 and 29.13 % after combination between gamma – rays 40 Gy and SA (0.001% or 0.002 %) concentration, respectively. On the other hand, when Gamma – rays dose 80 Gy was used in combination with SA in concentration of 0.001 % or 0.002 % , the frequency of the induced mutated families with chlorophyll deficiencies were slightly les than the percentage obtained after treatment only with gamma – rays or SA in the same dose when used alone. This combination effect may by attributed to the cumulative effect of radiation and chemical mutagenic, especially if they were different in their mode of action. **Mohan Rao (1972)** found that the additively of the two mutagens (X – ray + DES post – treatment on barley) in combination treatments with regard to M_2

– chlorophyll mutant frequency indicates that the two mutagens acted independently in mutation induction probably by different mechanisms.

These results are in agreement with **Arnason *et al*. (1963)** who showed that when EMS was applied before ethyleneimine, more than additive mutation frequencies were produced, combination of nucleotids with Co^{60} gamma – radiation or ethylmethan sulphate gave higher than additive frequencies. **Debelyi and Ptashenchuk (1973)** found that, the number of chlorophyll mutation induced in M_2 after treatment of *Vicia Sativa* (L gov 31 – 292) with gamma – rays and various chemical mutagens, alone or in combination, were higher in combined treatments.

The highest mutation rates were from combined treatments with 40 Gy of Gamma rays and (0.001 and 0.002 % SA) solutions of various chemical mutagens. Similar results were obtained by **El-Shouny and El-Hosary (1983 b), Lidanski *et al*. (1984), Filippetti and Depace (1986), Dawwam *et al*. (1988), Bhatnagar *et al*. (1992), Vandana (1992 a), Yasin (1996), Solanki and Sharma (2001 a), Solanki and Sharma (2001 b) and Dhole *et al.* (2003).**

B2.2. Varieties:

The data presented in Table (12) show significant differences between genotypes, it is clear that the two faba bean varieties clearly differed in their reaction over all mutagenic treatments, it is apparent that the variety Misr 1 is more mutable (29.43) than the variety Giza 429 (25.63 %). Varietal difference in sensitivity and mutability has been indicated early by many

authors **Shamsi and Sofajy (1980)** found that the French cv. was more sensitive than the Egyptian one to high doses. **El-Shouny and El-Hosary (1983 b)** observed varietals differences in spectrum of chlorophyll mutations between the two varieties Giza 1 and Giza 2. Similar results were reported by Yasin **(1996), Kumari (1996), Hajduch et al. (1999) and Muhammad and Bashir (2003).**

B2.3. Interaction effects:

From Table (12) it is clear that the effect of interaction between varieties and mutagenic treatments. It is shown from this Table (12) that the variety Giza 429 gave the highest mutated families 42.11 % when treated with 0.002 % SA, fallowed by variety Misr 1 when treated with 80 Gy (33.33 %). Generally, the variety Misr 1 gave the higher percentages of families with chlorophyll mutation in the M_2 than the second one (Giza 429).

C. Percentages of mutated types in M_2 generation:

C.1.Mutagenic treatments:

When mutation frequency was calculated as the percentage of mutation types obtained in each treatment, it was found that it took the same trend as it was found when it was calculated as the percentages of mutated families Table (11). For all mutagenic treatments, the percentages of mutation types were increased than that of mutated families in M_2 Table (11). The percentages of mutation types were 0.00, 31.02, 35.66, 38.66, 38.83, 40.58, 42.47, 44.23, 39.18 and 45.98 % for control, 40 Gy , 40 Gy + 0.001 % SA , 40 Gy + 0.002 % SA , 80 Gy , 80 GY

+ 0.001 % SA , 80 Gy + 0.002 % SA , 0.001 % SA and 0.002 % SA, respectively .

The frequency of chlorophyll mutation recoverable, in a mutation induction program, may be taken as a good indicates for the frequency of mutations with later expression and a good indication of the effectiveness of a given mutagen, **Smith (1950), Mesken and Van der Veen (1968), Bhatnagar *et al.* (1992), Vandana (1992 a), Yasin (1996), Solanki and Sharma (2001 a), Solanki and Sharma (2001 b), Dhole *et al.* (2003).** For comparison between different mutagenic treatments we shall consider the dose of Gamma – rays and concentrations of SA which cause nearly the same effects in M_1 – plant and the total percentage of chlorophyll mutations obtained. A relationship between increase in Gamma – rays dose or SA concentrations and mutation frequency was detected. Similar results were reported in various other plant species with different chemical and physical mutagens **(Gupta, *et al* 1969**, **Lidanski *et al*. 1984 and Tickoo and Chandra 1999**).

Added that a linear relationship between radiation dose and frequency of chlorophyll mutations was observed until 15 Kr , while doses higher than this gave lower yield of chlorophyll mutations. EMS – chlorophyll mutation frequency had increased by an exponent greater than 1, even in treatments giving extremely high M_1 – plant lethality. **Brock (1965)** found that mutation frequency increased linearly with increasing the dose of X – rays or thermal neutrons with same saturation at higher doses in *Trifolium subterreneum*. **Gupta *et al* (1969)** showed that in

most cases there is a linear correlation between dose rate and chlorophyll mutation frequency.

These authors attributed the reduction in mutation frequency at higher mutagen doses to saturation in mutational event. An alternative explanation is suggested by **Gaul (1957) and Hildering (1962)** who attributed this reduction in mutation frequency to statistical bases , as in low M_1 – fertility with higher mutagen doses, many M_2 families contain a low number of plant progenies, with the result that mutations in the M_1 generation had a smaller probability of being represented by mutants in M_2. **Yanaguchi (1962)** considers that frequency of chlorophyll mutations do not increase in linear relationship with dose 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.

In this investigation reduction in mutation frequency by increasing doses was not noticeable, perhaps because all doses used in this study did not reach that high level which can follow the above mentioned explanations.

C.2. Varieties:

Data shown in Table (12) indicated that the two faba bean varieties (Misr 1 and Giza 429) clearly differed from each other in their reaction for all mutagenic treatments, it is apparent that the variety Misr 1 is more mutable types (44.95 %) than the variety Giza 429 (34.52 %) .

C.3. Interaction effects:

From Table (12), it is clear that the effect of interaction between varieties and mutagenic treatments. Thus it is shown from this Table (11) the treatment 0.002 % SA gave increased mutation types in each varieties Miser 1 or Giza 429. Generally, Misr 1 gave a higher percentages of mutated types percentages than Giza 429. These percentages were 37.5 and 28.57 , 40.63 and 30.77 , 45.1 and 32.70 , 47.62 and 37.5 , 51.52 and 35.0 , 50.0 and 40.0 , 41.51 and 36.36 and 46.94 and 44.74 % for Misr 1 and Giza 429 when treated with 40 Gy , 40 Gy + 0.001 % SA , 40 Gy + 0.002 % SA , 80 Gy , 80 Gy + 0.001 % SA , 80 Gy + 0.002 % SA , 0.001 % SA and 0.002 % SA , respectively .

D: Spectrum of chlorophyll mutations

Tables (9, 10, 11 and 12) show the effect of mutagenic treatments, varieties and interactions on the spectrum of induced chlorophyll mutation in the M_2. According to **Gustafsson (1940)**, types of chlorophyll mutations observed in this study were albina (A), zantha (X), and viridis (V).

D1. Effect of mutagenic treatments on spectrum of chlorophyll mutations in M_2.

D1.1. Mutagenic treatments:

D1.1.1. Gamma rays:

From Table (9) it is clear that the spectrum of chlorophyll mutation plants was affected by Gamma – rays doses. This, at the lowest dose 40 Gy the albina type was 0.86 % while at the high dose 80 Gy was 0.91 %. Also, xantha and viridis types were increased by increasing dose of Gamma – rays from 40 Gy to 80 Gy. The percentages of different types of plant mutations at

different doses of Gamma – rays were 0.86, 0.62 and 0.53 % at 40 Gy, 0.91, 0.70 and 0.63 at 80 Gy for albina, xantha and viridis, respectively.

D1.1.2. Sodium azide (SA):

From Table (9), it is clear that the plant mutation spectrum was clearly affected by different concentrations of SA. Percentages of different types differed according to the concentration of SA. The plant mutation types were increased by increasing SA. The percentages of different types of plant mutation types were 0.88, 0.68 % and 0.60 at 0.001 % SA and 1.40, 0.88 and 0.70 % at 0.002 % SA for

albina, xantha and viridis , respectively.

D1.1.3. Combination effects:

Concerning the effect of combination of different mutagens, the three types of chlorophyll plant mutation were presented in Table (9). Although it was found that the albina type was the dominant and followed by xantha and than by viridis. Also, all types of chlorophyll plant mutations were increased with combination treatments than the effect of Gamma – ray or SA alone.

D1.2. Varieties:

From Table (10) it is clear that the two varieties slightly differed for mutation plant types. These percentages were 1.03, 0.75 and 0.67 of Misr-1 and 1.04 %, 0.72 and 0.61 % of Giza 429 for albina, xantha and viridis types, respectively.

D1.3. Interaction effects:

From Table (10), it is clear that the effect of interaction between varieties and mutagenic treatments. Thus, it is shown from this Table (10) that the variety Misr-1 gave the highest percentages of mutation spectrum plants of albina , xantha and viridis when treated with 80 Gy + 0.002 % SA . On the other hand the variety Giza 429 with treated by 40 Gy or 0.002 % SA gave the lowest for the three types of chlorophyll mutation plants.

D2. Effect of mutagenic treatments on spectrum of chlorophyll mutation families

D2.1. Mutagenic treatments:

D2.1.1. Gamma rays

From Table (11) it is clear that the spectrum of chlorophyll mutation families was affected by Gamma – rays doses. The three types of mutation were increased by increasing Gamma – rays doses from 40 Gy to 80 Gy. The percentages of mutated chlorophyll families at different doses of Gamma – ray were 12.82, 10.26 and 8.55 % at 40 Gy, 15.94, 14.49 and 10.14 % with 80 Gy for albina, xantha and viridis, respectively.

D2.1.2. Sodium azide:

From Table (11), it is clear that the mutation spectrum from families was clearly affected by the two concentrations of SA. The mutation types were increased by increasing SA from 0.001 to 0.002 %. Also, the albina type was the highest followed xantha and viridis. The percentages of different of different types (families) were 16.49, 12.37 and 10.31 % at 0.001 % SA, 19.54,

14.94 and 11.49 % at 0.002 % SA for albina, xantha and viridis, respectively.

In this connection many authors pointed out that, mutation of albina type are mostly point (gave) mutations, which are not connected (related) with chromosomal aberrations **(Gustafsson 1938), Edith *et al.* (1964), Butany and Singh (1965)**, while, viridis and xantha are macromutations which are connected chromosomal aberrations (**Gustafsson, *et al.* 1946)**. Also, **Heiner *et al.* (1960)**, after treatment of barley seed with Gamma – rays and DES they analyzed chlorophyll mutations and chromosome aberrations, and did not find any relationship between them. They concluded that occurrence of chlorophyll mutations is a different matter from those of chromosomal aberrations, **Bhaskaran and Swaminathan. (1963),** in barley did not find any relationship between the degree of M_1 fertility and frequency of any type of M_2 chlorophyll mutations. On the other hand, **Matsumura *et al.* (1963),** reported a linear relationship between frequency of all types of chlorophyll mutations and chromosomal aberrations and radiation dose, and therefore. They considered chlorophyll mutations to be in relation with chromosomal aberration.

The results obtained here are in general agreement with those previously obtained by **Bhatnagar *et al.* (1992), Vandana (1992 a), Yasin (1996), Solanki and Sharma (2001 a), Solanki and Sharma (2001 b), Dhole *et al.* (2003).**

D2.1.3. Effect of combination:

Concerning the effect of combination between Gamma – rays doses and SA concentrations, the three types of chlorophyll

mutated families are presented in Table (11). Although it was found that, the albina type was the highest percentage followed by xantha and viridis. It is clear that combinations of Gamma – rays 40 Gy and each of 0.001 or 0.002 % SA gave albina type more than other types, and it was increased than the same concentration in treatments with Gamma – rays (40 Gy), but less than SA (0.001 or 0.002 %) separately . Also, the effect of combination of gamma – ray 80 Gy with SA (0.001 or 0.002 %) gave albina types more than the same dose of gamma – ray 80 Gy when used separately. While, it gave the same percentages when compared with SA (0.001 or 0.002 %) alone.

Also, the same trend was obtained for xantha and viridis types. Where, the effect of combination between gamma – ray (40 Gy or 80 Gy) and each of concentrations of SA (0.001 and 0.002 %) gave more percentages of xantha or viridis than the same concentration treatments with Gamma – ray and SA when used separately.

D2.2. Varieties:

From Table (12), it is clear that the two varieties clearly differed than each other in their reaction to different mutagenic treatments. Thus it is shown from this Table (12) that the varieties Misr 1 gave more percentages of the three types of chlorophyll mutation in the M_2 (albina , xantha and viridis) than the second variety Giza 429. These percentages were 17.72, 15.02 and 11.41 % in Misr 1 and 14.21, 10.91 and 9.39 % in Giza 429 for albina, xantha and viridis, respectively.

D2.3. Interaction effects:

From Table (12) it is clear that the effect of interaction between varieties and mutagenic treatments. Thus, it is shown from this Table (12) that the variety Misr 1 with exposed to 80 Gy and treated of each 0.001 % SA, 0.002 % SA and Giza 429 treated by 0.002 SA alone gave the highest percentages of albina type. These percentages were 21.73, 22.73 and 21.05 % in the same order. However, the variety Giza 429 when treated with 40 Gy gave the lowest one (11.68). While, variety Misr 1 when treated with 80 Gy or 80 Gy + 0.001 % SA gave the highest percentages of xantha type (19.05 and 18.18 %). However, variety Giza 429 when treated with 40 Gy , 40 Gy +0.001 % SA and 40 GY +0.002 % SA had the lowest ones (9.09 , 9.23 and 9.62 %) . For viridis type, the highest percentage was detected from Misr 1 when treated with 80 GY + 0.002% SA (13.64 %). While, Giza 429 when treated with 40 Gy gave the lowest one (7.79 %).

E: Estimates of genetic components

The response to mutagens, as measured by the magnitude and the nature of the induced variability varied from character to character and between the two varieties. The estimates of phenotypic and genetic variability, heritability, genetic advance (ΔG) and ΔG % for each mutagenic treatment were important for improved characters.

Flowering and maturity

Mean performance, variance, heritability, genetic advance (ΔG) and ΔG % for number of days from sowing to 50 % flowering, 95 % pods maturity, and plant height in both varieties

are presented in Table (13). The phenotypic variance associated with the three traits were found to be increased by increasing gamma – ray (from 40 Gy to 80 Gy), SA (from 0.001% to 0.002% SA) and combination between Gamma – rays and SA. Also, these variances were more than the untreated (control) population and consequently increase in the genotypic variation Table (13).

High heritability valves were detected for the three traits (Number of days to 50% flowering 95 % pods maturity and plant height at different mutagenic treatments in both varieties.

Flowering date:

The genetic advance under selection of the percentages (Δg %) were found to be moderate in magnitude for flowering date at the different mutagenic treatments in both varieties varied from 21.55 (Misr 1 when treated with 80 Gy + 0.002% SA) to 9.32 (Giza 429 when treated with 0.001% SA).

Maturing date:

The genetic parameters of number of days to 95 % pods maturity for the two varieties were found to be progressively increased by different mutagenic treatments and combination treatments. It could also be noticed that the most pronounced estimates of the heritability and genetic advance under selection were found by 80 Gy + 0.002% SA in both varieties. These pronounced values of both heritability and genetic advance may give possibilities of improving this trait during the successive generations. Generally high heritability values and low genetic gain under selection were found in this trait. The results obtained here are in general agreement with those previously obtained by

Khan (1988), Vadana and Dubey (1992), Mehetre *et al.* (1994), and Samiullah *et al.* (2004).

Plant height:

The different genetic parameters which illustrated in Table (13) were found to be progressively increased as the Gamma – rays, SA and

combinations doses increased, for instance, the phenotypic variation which resulted from the treatment Miser 1 with 0.002% SA was found to be increased twelve times more than the untreated (control) population of the Misr 1 cv. High heritability estimates were found to be associated with low to moderate genetic advance as percentage of the mean (Δg %) in most mutagenic treatments. Therefore, selection for plant height could be effective and Satisfactory during the successive generations. The same conclusion was reported by **Voica *et al.* (1982), Vandana and Dubey (1992), Mehetre *et al.* (1994), Joshi and Verma (2004), El-Gazar *et al.* (2004).**

Number of branches / plant

The different genetic parameters which presented in Table (14). The phenotypic variation was found to be increased by increasing doses of mutagenic treatments especially 80 Gy + 0.002% SA with each variety Misr 1 or Giza 429 up to seven times more than the untreated (control) population and consequently increase in the genotypic variation. High heritability valves were detected for number of branches/plant at the different mutagenic treatment in both varieties and it ranged from 0.75 to 0.87. Also, genetic advance under selection (Δg %) were found to be high in magnitude at the different mutagenic

and varied from 51.23 to 88.73. It is of interest to mention that both heritability and genetic gain were detected to be progressively increased as mutagenic doses increased. In addition high heritability valves were found to be associated with high genetic advance as percentage of the mean (Δg %) in all mutagenic treatments.

Similar results were previously obtained by **Vandana and Dubey (1992), Mehetre *et al*. (1994), Samiullh *et al*. (2004).**

Number of pods and seeds/plant

The phenotypic variation was found to be increased by increasing mutagenic doses alone or combinations between physical and chemical mutagens especially 80 Gy + 0.002% SA when treated Misr 1 up to 13 times for number of pods and nine times in number of seeds/ plant more than the untreated (control) population and consequently increase in the genotypic variation Table (14). High heritability valves were detected for number of pods and seeds / plant at the different mutagenic treatments in both varieties, Also, genetic advance under selection (Ag %) were found to be high in magnitude at the different mutagenic doses and varied from 52.27 to 137.8 of number of pods / plant and 54.09 to 134.3 of number of seeds / plant. It is of interest to mention that both heritability and genetic gain were detected to be progressively increased as mutagenic doses (physical or chemical) and combination between them. Also, high heritability valves were found to be associated with high genetic advance as percentage of mean (Ag %) in all mutagenic treatments. Moreover, is no great differences between the

phenotypic and genotypic variance could be detected which would indicate the possibility of improving both characters. Therefore, it could be concluded that selection for number of pods and seeds / plant would be effective and satisfactory in the successive generations. Similar results were previously obtained by **Khan (1988), Filippetti and Depace (1986), Vandana and Dubey (1992), El-Gazar *et al.* (2004),** for number of pods / plant, **Singh *et al.* (2001), Samiullh *et al*. (2004), Mehetre *et al*. (1994),** and **Dawwam *et al*. (1988)**, **Joshi and Verma (2004),** for number of seeds / plant.

Number of seeds/pod

The different mutagenic treatments were found to be progressively increased the phenotypic variation and consequently the genotypic variation, for instance, the treatments 80 Gy + 0.001% SA and 80 Gy + 0.002% SA with Giza 429 increased the phenotypic variation up to five times more than the control (untreated) population Table (15). High heritability valves and moderate genetic gain estimates were detected for all mutagenic treatments in both varieties. As it is well known, heritability estimates along with the genetic gain were found to be more valuable than the former alone in predicting the effectiveness of selection. Therefore, selection for number of seeds/pod would be effective and satisfactory during the successive generations in the two varieties. Similar results were previously obtained by **Khan (1983), Filippetti and Depace (1986), Vandana and Dubey (1992), Singh *et al.* (2001), Metwally *et al*. (2004).**

100 – seed weight

The different genetic parameters were found to be increased by increasing Gamma – rays doses or SA concentrations and combination between them (Table 15). The phenotypic variances were found to be increased about four or five times more than the untreated (control) populations of both varieties. High heritability values were found to be associated with moderate genetic advance as percentage of the mean (Δg %) in all mutagenic treatments used in both varieties. Therefore selection for this trait could be effective and satisfactory during the successive generation. The results obtained here are in general agreement with those previously obtained by **Khan (1983), Khan (1988), Dawwam *et al*. (1988), Vandana and Dubey (1992), Mehetre *et al*. (1994) and Singh *et al.* (2001).**

Seed yield/plant

The different genetic parameters presented in Table (15) were found to be increased by increasing Gamma – rays or SA doses and combination between them. For instance, the phenotypic variations were found to be increased seven to four times more than the untreated populations of both varieties. SA it has been previously mentioned, heritability estimates along with genetic gain were found to be more valuable than the former alone in predicting the effectiveness of selection. In this study, high heritability values were found to be associated with high genetic advance as percentage of the mean (Δg %). The results obtained here are in general agreement with those previously obtained by **Khan (1988), Vadana and Dubey (1992), Mehetre *et al*. (1994), Singh *et al.* (2001), El-Gazar *et al.* (2004) and Samiullah *et al.* (2004).**