

## MUTAGEN-INDUCED CHANGES IN GENETIC VARIANCE AND HERITABILITY ESTIMATES FOR PEANUT SEED YIELD TRAITS

BY

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### ABSTRACT

**Locally**, only two *Arachis hypogaea* L. cultivars have been so far released for growers. Despite the unresolved controversy on the potentiality of mutation breeding, induced mutagenesis has been proven useful in widening the genetic pool of this quite important protein and oil crop among other oil crops. The main objective of this research was to estimate the induced genetic variance and broad-sense heritability for peanut seed yield component traits of mutagen-treated seeds under controlled conditions. Seeds of two cultivars, Giza 5, and Giza 6, were planted at the Egyptian Atomic Energy (EAE) Experiment Station at Anshas during June of the three consecutive years of 2002 to 2004. To induce physical and chemical mutagenesis, air-dried seeds of both cultivars were treated with both physical and chemical mutagens. The physical mutagenic effect was induced by gamma ( $\gamma$ ) rays using Cobalt 60 ( $^{60}\text{Co}$ ). Seeds were irradiated by four doses –100, 200, 300, and 400 Gray<sup>1</sup> (GY). The chemical mutagens were of two sources: ethyl methane sulphoni-ate (EMS),  $\text{C}_3\text{H}_7\text{O}_2\text{S}$  and sodium azide (SA),  $\text{NaN}_3$ . Three concentrations of 1.0, 2.0, and 3.0 mM in a 0.1 M phosphate buffer of pH 3.0 were used for each one. Mutagens showed variance heterogeneity only for cv. Giza 5. Both the within-mutagen phenotypic and genetic variances, for the SA doses, were quite heterogeneous ( $P < 0.05$ ) for all four studied seed characters – pod number plant<sup>-1</sup>, seed plant<sup>-1</sup>, pod weight plant<sup>-1</sup>, seed weight plant<sup>-1</sup>. In general, within each cultivar, broad-sense heritability estimates had a range of 0.47-0.77 for Giza 5, and of 0.56-0.79 for Giza 6 based on mutagen mean across the four characters. Mutagenwise, heritability estimates were relatively different irrespective of seed character and cultivar. Sodium azide rates resulted in a difference of eight percentage points from gamma ray doses (70% vs. 62%) and of three points from EMS doses (70% vs. 67%). Therefore, selection for these specific seed characters seems effective under SA-treated seed conditions. However, the within-mutagen heritability values were inconsistent and did not follow a definite pattern for any seed yield components. In future research, it would be more valid to select a dose range, instead, to recommend for further in-depth mutation-induced advanced research on the molecular level. This is to investigate whether these mutagen induced effects happened due to the occurrence of any real mutants and if they are new loci or allelic to existing loci. Therefore, more appropriate screening technique(s) should be suggested by breeders to identify and assay those *real* mutants on DNA level.

### INTRODUCTION

Currently in Egypt, only two ground-nut (*Arachis hypogaea* L.) cultivars have already been released by the Oil Crop Research Division, the Agricultural Research Institute<sup>2</sup>. This quite a few cultivar number released in the market, certainly, implies how this significant oil and protein crop needs,

among other main field crops, an extra effort to expand its genetic pool using various integrated plant breeding techniques.

There has still been an unresolved controversy on whether to acknowledge mutation breeding as a tool to create induced

<sup>1</sup> 1 Gray=100 rad or 1 J per Kg of the irradiated object.

<sup>2</sup> Agricultural Extension Bulletin No. 1099, 2008. Agricultural Research Center, Giza.

variation particularly in field crops. Allard (1960) severely criticized induced mutation methodology. He stated two reasons are good for the rapid loss of interest in induced mutations. First, induced mutations were almost always deleterious in their effects on the phenotype. Second, many of the genes governing characteristics in which commercial varieties required improvement occurred in one or another known stock. However, induced mutagenesis has been proven useful in widening the genetic base of many diverse crop species. This wide genetic variation, expressed as genetic variance, is urgently required for further selection, and for prediction of response to selection.

A total of 163 cultivars of annual oilseed crops have been officially approved and released for cultivation in 26 countries using induced mutation; of which 44 cultivars belong to peanut. A total of 118 cultivars has been developed as direct mutants and 45 using the induced mutants in a crossing program; in groundnut 22 cultivars out of 44 were developed after hybridization (Bhatia, *et al.*, 1999). In Egypt, however, according to them, only two sesame (*Sesamum indicum* L.) cultivars (Cairo White 8, and Sinai White 48) were released back in 1992 using gamma-ray doses.

Relying on the concept of induced mutagenesis, it is essential to obtain adequate information on the extent to which any particular mutagen is likely to cause genetic variability, and how much of this variability is inherited. Broad-sense heritability estimates are useful as initial approximations, not as definitive estimated values (Ronis *et al.*, 1985). Omoigui *et al.* (2006) argued that it is very crucial to collect fairly enough information not only about the magnitude and type of genetic variability, but also about its parallel heritability. The amount of genetic variability cannot by itself frame the success of the selection of promising genotypes, for the extent to which the desired character(s) are heritable is noteworthy. Briggs and Knowles (1967) set forth the view that if environmental variability is negligible compared to genetic variability, selection will be effective.

The five (25, 50, 100, 200, and 300 Gy) Gamma-ray doses caused a wide range of magnitude in the genetic variance compo-

nent,  $\sigma_g^2$ , estimates for per-plant of each of pod number, seed number, pod yield, and seed yield for cv Giza 5 in M2 generation (Moustafa, 1994). These varying values of  $\sigma_g^2$  were inconsistent within these characters as dose level goes up, except for both pod and seed number per plant since they were nearly linearly affected. These estimates ranged from 79 to 155 for pod number per plant, and 165 to 280 for seed number per plant.

In M1 across cvs Giza 5 and Giza 6, sodium azide doses and the above mentioned four seed yield components, the estimated variance  $\sigma_g^2$  component ranged from 13.63-76.90 (El-Shazly *et al.*, 2005); yet, the M2 range narrowed down to 21.92-55.52 compared to M1. Moreover, the M1 genetic variance estimates were consistently twice as much for Giza 6 compared to Giza 5. In M2, on the contrary, both cultivars had nearly equal genetic estimates, except for Giza 6 seed yield per plant where the estimate was twice as much.

Even within the same species, and most often the same parent cultivars that have been induced mutagenically, values of heritability were widely varied. This situation makes interpretation of the results to a great extent difficult, unless researchers state very specifically all the factors which may have affected the mutagenic process, and may aid in explanation of the outcomes. Unfortunately, most researchers do not take some/all of these factors into consideration; and if they do, they unintentionally do not report them.

Genetic parameters, in M3, were again estimated for two *Arachis hypogaea* L. genotypes (Mensah and Obadoni, 2007) when seeds were treated by 1.0, 2.0, 3.0, and 4.0 mM doses of sodium azide. For five yield parameters, the genetic variance ranged from 0.9 to 15.1. Similarly, the phenotypic variance ranged from 1.9 to 18.0. The differences between the two estimated parameters were low for both pods and seeds per plant. They inferred the low influence of environments on those two yield characters, as indicated by the high heritability values of 90% and 83%, respectively.

Moreover, they assert that high heritability estimates were coupled with high mean

expected genetic gains (15 pods, and 10 seeds) at 10% selection pressure, as has been observed for per plant number of pod and of seeds across the two studied cultivars. They confirmed that additive gene effects played an important role in the expression of such traits. As a result, these traits could be effective in the selection of high yielding cultivar/ genotypes; yet, data show that they have only estimated the broad- and have estimated neither the narrow-sense heritability nor the genetic variance additive component,  $\sigma^2_A$ . This additive variance gene effect can be estimated in different ways (see Fehr, 1987; Singh, 1993) from results of a trial consisting of the two parents, their  $F_1$ , and  $F_2$  and the two backcrosses.

### MATERIALS AND METHODS

Seeds of two groundnut (*Arachis hypogaea* L.) cultivars, Giza 5, and Giza 6, were planted at the Egyptian Atomic Energy (EAE) Experiment Station at Anshas during June of the three consecutive years of 2002 to 2004.

To induce physical and chemical mutagenesis, prior to planting, air-dried seeds of both cultivars were treated with both physical and chemical mutagens. The physical mutagenic effect was induced in this investigation using gamma ( $\gamma$ ) rays using the Cobalt 60 ( $^{60}\text{Co}$ ) unit installed at the EAE. Seeds were irradiated by four gamma ray doses -100, 200, 300, and 400 Gray (GY). The chemical mutagens were of two sources: ethyl methane sulphonate (EMS),  $\text{C}_3\text{H}_8\text{O}_3\text{S}$ , and sodium azide (SA),  $\text{NaN}_3$ . For the chemical mutagens, three concentrations of 1.0, 2.0, and 3.0 mM in a 0.1 M phosphate buffer of pH 3.0 were used for each chemical mutagen. For each concentration, both cultivars, seeds were presoaked for 12 hr in glass bottles and kept incubated at  $25\text{ }^\circ\text{C}$ , then rinsed in tap water before planting in the field. The control (untreated) seeds were soaked in tap water for 12 h before planting.

On 2 June 2002, three seeds of each of the two cultivars were put into 30-cm hills in four 3.0 m long x 0.6 m rows. Seedlings were later thinned to one per hill. Cultivars and mutagen doses were put in three rando-

It is, therefore, apparent that mutagenesis-induced result variations considerably depend upon many influential prevailing factors as well as upon implemented statistical tools so that interpolations should be limited by these specific overall factors.

This research aims mainly at estimating the genetic variation and broad-sense heritability that may result from trying to obtain mutant-induced groundnut (*Arachis hypogaea* L.) plants in the  $M_2$  generation as a consequence of treating seeds with a range of gamma-ray, ethyl methane sulphonate and sodium azide doses.

mized complete blocks of a split plot design. Cultivars were the main plots and the mutagen doses were the subplots. Plant population and yield will be termed hereafter  $M_1$ .

At harvesting of the  $M_1$  plants, a random seed sample was collected from each treatment and saved for planting the following year to obtain the  $M_2$  population. On 20 June 2003, seeds were planted in the field to produce the  $M_2$  plants. Treatment factors were arranged in a split plot design as was done in Year 2002. At harvesting, data were recorded on a 15-guarded plants random sample selected from within each subplot. This makes a total of a 45 plants across the three replicates for each treatment combination.

In 2004, to yield the  $M_3$  plants, on 3 June 2004, bulked  $M_2$  seeds of each treatment were planted in a separate experiment in a three randomized complete blocks of a split plot design. Studied characters were recorded using a 15-plants random sample from each subplot.

Statistical analysis presented in this research depends upon data that comes from only the  $M_2$  generation. Seed yield data -- pod number  $\text{plant}^{-1}$ , seed  $\text{plant}^{-1}$ , pod weight  $\text{plant}^{-1}$ , seed weight  $\text{plant}^{-1}$  -- were analyzed considering replications and mutagens of random effect.

Both phenotypic ( $\hat{\sigma}^2_P$ ) and genotypic ( $\hat{\sigma}^2_G$ ) variances were estimated based on the individual-plants procedure derived from the procedure described by Briggs and Knowles (1967) and later by Ronis *et al* (1985). In which, the control plants of the  $M_2$  population were utilized to estimate environmental variance,  $\hat{\sigma}^2_E$ . The within-mutagenic treatment variance is considered an estimate of the phenotypic variance,  $\hat{\sigma}^2_P$ . Therefore, the genetic variance,  $\hat{\sigma}^2_G$ , is obtained by subtracting the environmental variance from the phenotypic variance. Broad-sense heritability ( $H_B$ ) was estimated as follows:

$$H = \frac{\hat{\sigma}^2_G}{\hat{\sigma}^2_G + \hat{\sigma}^2_e}$$

Genetic advance under selection ( $\hat{G}_S$ ) was calculated as follows

$$\hat{G}_S = (k)(\sqrt{\hat{\sigma}^2_P}(H))$$

where  $G_S$  is the genetic gain/advance under selection, i.e. the possible gain from selection,  $k$  is the selection differential,  $\hat{\sigma}^2_P$  is the phenotypic variance of the parent population, and  $H$  is the broad-sense heritability. The  $k$

is expressed in terms of standard deviation units, and it is inversely proportional to the selection intensity. To predict the value of  $G_S$ , the studied character should be about normally distributed (حسن، ٢٠٠٥). The  $G_S$  was estimated based on  $k$  values derived according to 20% selection intensity (see Fehr, 1987; Hallauer and Miranda, 1988) for corresponding selection differential,  $k$ , values.

The  $M_2$  data, based on individual plants, were tested for homogeneity of variances using Bartlett's test (Steel and Torrie, 1980; Damon and Harvey, 1987). Within each cultivar, both within-, and among-mutant tests have been simultaneously carried out for each of the phenotypic and the genetic variance estimates. If, for the within-mutant, the hypothesis failed to be rejected, therefore data would be interpreted based on pooled variance. If the hypothesis for the among-mutant variance homogeneity turns out to be true, in this case a pooled variance estimate would be used in the data analysis and interpretation.

## RESULTS AND DISCUSSION

### Estimated Variances

The estimated  $M_2$  phenotypic and genetic variances, for each seed character, were tested for homogeneity of variance based on Bartlett's test (Steel and Torrie, 1980; Damon and Harvey, 1987). Within-mutagen, doses showed variance heterogeneity only for cv. Giza 5 (Tables 1 & 2). Both the within-mutagen phenotypic and genetic variances, for the sodium azide mutagen doses, were quite heterogeneous ( $P < 0.05$ ) for all four studied seed characters.

It follows, therefore, that the sodium azide doses used in this research, regardless of the seed character, showed inconsistency in the estimated within-genetic variance component estimates than in those estimated component for either gamma rays or ethyl methane sulphonate doses. Hence, data should be interpreted separately by sodium azide dose.

For pod per plant and seed per plant, each of the phenotypic and genotypic variance estimates, for the 3.0 mM sodium azide dose, was a twofold value of that of the 1.0 mM (165.22 vs. 75.22 and 146.05 vs. 56.05) for pod per plant, and (373.75 vs. 211.36 and

305.28 vs. 142.89) for both estimates, respectively (Table 1). The 3.0 mM also caused increased variations ranged from two- to sixfold relative to those of the 2.0 mM (Table 1). These indicates how relatively large variation this SA dose caused in two major seed yield components. For seed weight per plant, the variation range diminishes to be as close as possible between the 3.0 mM and the 1.0 mM doses (Table 2). Still a twofold difference in variation did exist in favor of the 3.0 mM relative to the 2.0 mM dose. However, for pod weight plant, twofold phenotypic and three-fold genetic variations were obtained, but rather in favor of the little dose of 1.0 mM (Table 2).

The 3.0 mM dose of sodium azide generally caused a great variation in three of the major yield-component traits, which contribute heavily to the final seed yield. This high variation, both phenotypic and genetic, gives more likelihood of selection for higher yields in later mutation generations if high number of plant populations is involved in the selection.

However, the triple as much contribution of the 1.0 mM dose to the genetic variation of pod per plant trait as well as the equality with the 3.0 mM dose for seed weight per plant should not be overlooked. Number of pods per plant is related positively to both seed weight per pod and percentage shelling. This seed yield component trait would be of significance to be selected for if seed weight per plant genetically varies too in response to a mutagen dose as well as percentage shelling does. Therefore, selection based on just pod weight would not be beneficial to yield unless both shelling and seed weight are considered together. The 1.0 mM dose may be therefore as effective as the 3.0 mM in case if both show distinct genetic variance induction in advanced generations.

Testing the among-mutagen variance homogeneity, only per-plant pod number mean genetic variances were homogeneous ( $P > 0.05$ ) (Tables 1 & 2). This implies that the genetic variance estimates for the other three mutagen treatment groups, across the other seed characters, be analyzed and interpreted by mutagen type. There were considerable differences ( $P < 0.05$ ) between the estimated mean genetic variance components (Tables 1 & 2) within each of the three traits. Mean sodium azide estimate was again as twice as much as that of the gamma rays across the four characters.

Hence, simultaneous differences occurred within- and among-mutagen genetic variances within seed characters. The within-mutagen induced variations were of no definite trend among within any seed character. But, in case of the among-mutagen variation, there exists an increasing trend in favor of mean SA-induced genetic variance within all four seed characters. Therefore, SA shows a potential of likely induced considerable genetic variations in  $M_2$  generation. These relatively high genetic variance estimates would rather be taken into account in selection criteria for higher seed yield. Yet, this needs additional assessment in more environments, since it is too early in the  $M_2$  to decide on the effectiveness of a specific mutagen. Mutant screening techniques is crucial step in next

generations. Screening technique(s) is/are necessary to identify the genetics of any induced mutants (Swada and Palmer, 1987). Finally, large plant population should be assayed for mutants since the latter rarely occur (Allard, 1960), and as the data of Swada and Palmer (1987) has verified.

For cv Giza 6, on the other hand, all variance homogeneity tests failed to be rejected at  $P=0.05$  whether for within- (Tables 3 & 4) or among mutagen-variances. Hence, a pooled phenotypic and genetic variances across doses and mutagen type may be used for each seed character. This finding contradicted what the data of El-Shazly *et al.* (2005) postulated. In their study, averaged over three gamma-ray doses (200, 300, and 400 Gy) and over three sodium azide doses (1.0, 2.0, and 3.0 mM), estimated  $M_2$  genotypic values were substantially varied between these two mutagens for three of the four reported herein cv Giza 6 seed characters.

The comparison between both mean sodium azide genetic variance estimates for either cultivar over all four seed characters, indicated quite close values within each character, but Giza 6 seed weight per plant genetic variance was almost half as much as that of Giza 5 (105.82 vs. 207.72) (Tables 2 & 4). El-Shazly *et al.* (2005), on the contrary, found that both cultivars had also nearly equal genetic estimates, except for Giza 6 seed yield per plant where the estimate was twice as much.

Despite of similarity in both SA doses, cultivars, and variance estimation procedure, these contradictory  $M_2$  results between the two studies might be explained from two aspects. Mutagenic effects are heavily spontaneous in nature and do rely to a great extent on the nature of all the other factors that should be under control during performing seed treatment, and may influence their mutagenic action if any change happens. On the other hand, mutagen-induced effects are gene-targeted, but are greatly directed by the prevailing environmental conditions, seed source and condition during the treatment process, and dose duration. It is unlikely to

have complete control on all these varying factors so as to, expect identical results among different experiments even among those performed in the same lab. But, these factorial variations do not deny these induced mutagenic effects being of spontaneous nature. This unprompted mutagenic influence was previously manifested as Olsen *et al.* (1993) have indicated in barley. DNA sequence analysis of mutant genes revealed a range of 4-7 base substitutions in the four mutant genes, respectively. More importantly is that one mutant gene contained two adjacent base changes, whereas the remaining substitutions were scattered randomly throughout the genes. These demonstrate that azide induced mutagenesis is neither locally nor regionally targeted within the gene.

These raises a query that Singh (1993) has mentioned, which concerns the significance of reporting the exact mutagen-treated techniques and the required environmental conditions. El-Shazly *et al.* (2005) have reported neither the sample size used in estimating different variance estimates nor the environmental variance estimate values for either cultivar. Certainly, the issue also here is not the variation in the result of just one studies character, but the issue is the reverse result. Therefore, it is too soon in early generations to judge a chemical mutagen dose of being successful or not in inducing quite a genetic variation enough to be recommended in breeding program(s). In addition, since azide induced mutagenesis is neither locally nor regionally targeted within the gene (Olsen *et al.*, 1993), this prompt investigating the mode of action SA range of doses on the DNA level, as well as other mutagen mode of actions, in other field crops.

#### Broad-Sense Heritability and Genetic Gain from Selection

The  $M_2$  heritability estimates are shown in Tables 1-5 for the four seed yield component characters. In general, within each cultivar, heritability estimates had a range of 0.47-0.77 for Giza 5, and of 0.56-0.79 for Giza 6 based on mutagen mean across the four seed characters. Within Giza 5, the lower range value, 0.47, was of mean gamma ray for pod number per plant, and the upper limit,

0.77, for mean SA for seed weight per plant (Table 2). Similarly, within Giza 6, the lower limit (0.56) resulted from mean EMS for seed per plant, and the upper limit (0.79) for SA in case of number of pod per plant (Table 3).

Both cultivar mean heritability values were close and upper intermediate, 0.64 and 0.69, respectively. These suggests that either cultivar may be used as the candidate to be a base population to induce real mutants using any of the mutagen types that can be recommended for peanut crop plant. This is regardless of the variation between the two cultivars in either the more heritable seed character or the most effective mutagen type. For higher values, within each cultivar, which had resulted from different combinations of mutagen type, dose, and seed character, within Giza 5, the upper range limit, 0.77, resulted from SA in seed weight per plant (Table 2); within Giza 6, 0.79 resulted from also SA in case of number of pods per plant (Table 3). There was more upper intermediate mutagen X seed character heritability values ( $\geq 0.70$ ). In case of Giza 5, EMS X pod per plant (0.74), SA X pod per plant (0.73) (Table 2); for Giza 6, either gamma-, EMS-, or SA X pod per plant resulted in estimates of 0.78, 0.78, and 0.79, respectively, in addition to an estimate of 0.73 in case of EMS X seed weight per plant.

The choice of any combination(s) as candidates for assaying real mutants to base selection on in any mutagen breeding program depends upon which seed yield component character(s) is/are more manageable to screen mutants from. In the same time, it is important for the breeder to define which mutagen type(s) and dose(s) is/are more pronounced beneficial mutation induction potential. Both conditions do not override the degree of positive association of these yield component characters to total seed yield. Within Giza 6, all heterogeneity tests were rejected; yet estimate range was still relatively higher. The magnitude of the environmental variances (Tables 3 & 4), within Giza 6, was relatively much smaller for three out of four characters, this apparently resulted in high genetic variance estimates, and consequently led to higher estimated heritability values.

Table (1): Variance component <sup>†</sup>estimates for peanut cultivar Giza 5 per plant pod and seed number in M<sub>2</sub> generation.

| Treatment         | Seed Characters      |                      |                      |                      |                |             |                       |                      |                      |                      |                  |                |
|-------------------|----------------------|----------------------|----------------------|----------------------|----------------|-------------|-----------------------|----------------------|----------------------|----------------------|------------------|----------------|
|                   | Pod pl <sup>-1</sup> |                      |                      |                      |                |             | Seed pl <sup>-1</sup> |                      |                      |                      |                  |                |
|                   | Mean                 | $\hat{\sigma}^2_E$ † | $\hat{\sigma}^2_P$ ‡ | $\hat{\sigma}^2_G$ ¶ | H <sub>p</sub> | $\hat{G}_p$ | Mean                  | $\hat{\sigma}^2_E$ † | $\hat{\sigma}^2_P$ ‡ | $\hat{\sigma}^2_G$ ¶ | H <sub>s</sub> # | $\hat{G}_s$ †† |
| Cultivar Giza 5   |                      |                      |                      |                      |                |             |                       |                      |                      |                      |                  |                |
| Control           | 64.9                 | 19.17                | ---                  | ---                  | ---            | ---         | 111.5                 | 68.47                | ---                  | ---                  | ---              | ---            |
| <b>γ rays, Gy</b> |                      |                      |                      |                      |                |             |                       |                      |                      |                      |                  |                |
| 100               | 71.1                 | ---                  | 60.31                | 41.14                | 0.68           | 7.4         | 123.6                 | ---                  | 134.51               | 66.04                | 0.49             | 7.9            |
| 200               | 76.3                 | ---                  | 45.03                | 25.86                | 0.57           | 5.4         | 135.9                 | ---                  | 137.28               | 68.81                | 0.51             | 8.4            |
| 300               | 52.1                 | ---                  | 89.38                | 70.21                | 0.78           | 10.3        | 86.2                  | ---                  | 141.13               | 72.66                | 0.52             | 8.6            |
| 400               | 48.8                 | ---                  | 43.58                | 24.41                | 0.56           | 5.2         | 79.5                  | ---                  | 136.61               | 68.14                | 0.50             | 8.2            |
| Mean              | 62.1                 |                      | 59.58                | 40.40                | 0.65           | 7.1         | 106.3                 |                      | 137.38               | 68.91                | 0.51             | 8.3            |
| <b>EMS, mM</b>    |                      |                      |                      |                      |                |             |                       |                      |                      |                      |                  |                |
| 1.0               | 53.5                 | ---                  | 81.16                | 61.99                | 0.76           | 9.6         | 108.4                 | ---                  | 153.92               | 85.45                | 0.56             | 9.7            |
| 2.0               | 64.5                 | ---                  | 72.25                | 53.08                | 0.73           | 8.7         | 116.5                 | ---                  | 220.79               | 152.32               | 0.69             | 14.3           |
| 3.0               | 68.2                 | ---                  | 73.42                | 54.25                | 0.74           | 8.9         | 138.2                 | ---                  | 171.96               | 103.49               | 0.60             | 11.0           |
| Mean              | 62.1                 |                      | 75.6                 | 56.44                | 0.74           | 9.1         | 121.0                 |                      | 182.22               | 113.75               | 0.62             | 11.7           |
| <b>SA, mM</b>     |                      |                      |                      |                      |                |             |                       |                      |                      |                      |                  |                |
| 1.0               | 62.1                 | ---                  | 75.22                | 56.05                | 0.75           | 9.1         | 126.7                 | ---                  | 211.36               | 142.89               | 0.67             | 13.6           |
| 2.0               | 73.1                 | ---                  | 42.26                | 23.09                | 0.55           | 5.0         | 138.8                 | ---                  | 165.04               | 96.57                | 0.59             | 10.6           |
| 3.0               | 65.2                 | ---                  | 165.22               | 146.05               | 0.88           | 15.8        | 116.4                 | ---                  | 373.75               | 305.28               | 0.82             | 22.2           |
| Mean              | 66.8                 |                      | 94.23                | 75.06                | 0.73           | 10.0        | 127.3                 |                      | 250.05               | 181.58               | 0.69             | 17.7           |

Table (2): Variance component <sup>†</sup>estimates for peanut cultivar Giza 5 per plant pod and seed weight in M<sub>2</sub> generation.

| Treatment         | Seed Characters             |                      |                      |                      |                |             |                              |                      |                      |                      |                  |                |
|-------------------|-----------------------------|----------------------|----------------------|----------------------|----------------|-------------|------------------------------|----------------------|----------------------|----------------------|------------------|----------------|
|                   | Pod weight pl <sup>-1</sup> |                      |                      |                      |                |             | Seed weight pl <sup>-1</sup> |                      |                      |                      |                  |                |
|                   | Mean                        | $\hat{\sigma}^2_E$ † | $\hat{\sigma}^2_P$ ‡ | $\hat{\sigma}^2_G$ ¶ | H <sub>p</sub> | $\hat{G}_p$ | Mean                         | $\hat{\sigma}^2_E$ † | $\hat{\sigma}^2_P$ ‡ | $\hat{\sigma}^2_G$ ¶ | H <sub>s</sub> # | $\hat{G}_s$ †† |
| Cultivar Giza 5   |                             |                      |                      |                      |                |             |                              |                      |                      |                      |                  |                |
| Control           | 137.4                       | 73.63                | ---                  | ---                  | ---            | ---         | 110.0                        | 55.31                | ---                  | ---                  | ---              | ---            |
| <b>γ rays, Gy</b> |                             |                      |                      |                      |                |             |                              |                      |                      |                      |                  |                |
| 100               | 152.4                       | ---                  | 172.51               | 98.91                | 0.57           | 10.5        | 127.9                        | ---                  | 143.70               | 88.39                | 0.62             | 10.4           |
| 200               | 149.9                       | ---                  | 129.48               | 55.88                | 0.43           | 6.8         | 130.4                        | ---                  | 94.28                | 38.97                | 0.41             | 5.6            |
| 300               | 132.3                       | ---                  | 124.98               | 51.38                | 0.41           | 6.4         | 107.2                        | ---                  | 159.76               | 104.45               | 0.65             | 11.5           |
| 400               | 134.3                       | ---                  | 144.82               | 71.22                | 0.49           | 8.3         | 105.2                        | ---                  | 192.98               | 137.67               | 0.71             | 13.8           |
| Mean              | 142.2                       |                      | 142.94               | 69.35                | 0.47           | 8.0         | 117.7                        |                      | 147.68               | 92.37                | 0.60             | 10.3           |
| <b>EMS, mM</b>    |                             |                      |                      |                      |                |             |                              |                      |                      |                      |                  |                |
| 1                 | 130.2                       | ---                  | 150.24               | 76.64                | 0.51           | 8.7         | 120.0                        | ---                  | 145.57               | 90.26                | 0.62             | 10.5           |
| 2                 | 161.0                       | ---                  | 301.74               | 227.64               | 0.75           | 18.2        | 139.8                        | ---                  | 232.36               | 177.05               | 0.76             | 16.2           |
| 3                 | 173.9                       | ---                  | 180.19               | 106.59               | 0.59           | 11.1        | 159.1                        | ---                  | 161.89               | 106.58               | 0.66             | 11.8           |
| Mean              | 155.0                       |                      | 210.72               | 136.96               | 0.62           | 12.7        | 139.6                        |                      | 179.94               | 124.63               | 0.68             | 12.8           |
| <b>SA, mM</b>     |                             |                      |                      |                      |                |             |                              |                      |                      |                      |                  |                |
| 1                 | 163.1                       | ---                  | 343.51               | 269.91               | 0.79           | 20.5        | 150.0                        | ---                  | 302.83               | 247.52               | 0.82             | 20.0           |
| 2                 | 186.3                       | ---                  | 139.95               | 66.35                | 0.47           | 7.8         | 168.7                        | ---                  | 158.37               | 103.52               | 0.65             | 11.4           |
| 3                 | 189.8                       | ---                  | 141.18               | 67.58                | 0.48           | 8.0         | 155.7                        | ---                  | 327.44               | 272.13               | 0.83             | 21.0           |
| Mean              | 179.7                       |                      | 208.21               | 134.61               | 0.58           | 12.1        | 158.1                        |                      | 262.88               | 207.72               | 0.77             | 17.5           |

Table (3): Variance component estimates for peanut cultivar Giza 6 per plant pod and seed number in M<sub>2</sub> generation.

| Treatment       | Seed Character      |                    |                    |                    |                |             |                      |                    |                    |                    |                  |                |
|-----------------|---------------------|--------------------|--------------------|--------------------|----------------|-------------|----------------------|--------------------|--------------------|--------------------|------------------|----------------|
|                 | Pod pl <sup>†</sup> |                    |                    |                    |                |             | Seed pl <sup>†</sup> |                    |                    |                    |                  |                |
|                 | Mean                | $\hat{\sigma}^2_E$ | $\hat{\sigma}^2_P$ | $\hat{\sigma}^2_G$ | H <sub>i</sub> | $\hat{G}_s$ | Mean                 | $\hat{\sigma}^2_E$ | $\hat{\sigma}^2_P$ | $\hat{\sigma}^2_G$ | H <sub>i</sub> # | $\hat{G}_s$ †† |
| Cultivar Giza 6 |                     |                    |                    |                    |                |             |                      |                    |                    |                    |                  |                |
| Control         | 65.0                | 18.25              | ---                | ---                | ---            | ---         | 84.6                 | 103.48             | ---                | ---                | ---              | ---            |
| γ rays, Gy      |                     |                    |                    |                    |                |             |                      |                    |                    |                    |                  |                |
| 100             | 67.7                | ---                | 82.17              | 63.92              | 0.77           | 9.8         | 125.1                | ---                | 260.55             | 157.07             | 0.60             | 13.6           |
| 200             | 68.3                | ---                | 83.94              | 65.69              | 0.78           | 10.0        | 118.4                | ---                | 245.93             | 142.45             | 0.58             | 12.7           |
| 300             | 61.0                | ---                | 89.54              | 71.29              | 0.79           | 10.5        | 92.9                 | ---                | 238.85             | 135.37             | 0.57             | 12.3           |
| 400             | 52.5                | ---                | 86.66              | 68.41              | 0.79           | 10.3        | 88.7                 | ---                | 227.90             | 124.42             | 0.55             | 11.6           |
| Mean            | 62.4                | ---                | 85.60              | 67.30              | 0.78           | 10.1        | 106.3                | ---                | 243.31             | 139.82             | 0.58             | 12.6           |
| EMS, mM         |                     |                    |                    |                    |                |             |                      |                    |                    |                    |                  |                |
| 1.0             | 65.1                | ---                | 84.55              | 66.30              | 0.78           | 7.2         | 108.4                | ---                | 298.68             | 195.20             | 0.65             | 15.7           |
| 2.0             | 63.2                | ---                | 87.22              | 68.97              | 0.79           | 10.3        | 114.6                | ---                | 230.10             | 126.62             | 0.55             | 11.7           |
| 3.0             | 55.3                | ---                | 80.98              | 62.73              | 0.77           | 9.7         | 95.6                 | ---                | 201.96             | 98.48              | 0.49             | 9.7            |
| Mean            | 64.5                | ---                | 84.20              | 66.00              | 0.78           | 9.1         | 106.2                | ---                | 243.58             | 140.10             | 0.56             | 12.4           |
| SA, mM          |                     |                    |                    |                    |                |             |                      |                    |                    |                    |                  |                |
| 1.0             | 69.2                | ---                | 88.20              | 69.95              | 0.79           | 10.4        | 145.4                | ---                | 286.35             | 182.87             | 0.64             | 15.2           |
| 2.0             | 58.5                | ---                | 92.63              | 74.38              | 0.80           | 10.8        | 103.3                | ---                | 261.08             | 157.60             | 0.60             | 13.6           |
| 3.0             | 60.4                | ---                | 88.75              | 70.50              | 0.79           | 10.4        | 100.8                | ---                | 202.18             | 98.70              | 0.49             | 9.7            |
| Mean            | 62.7                | ---                | 89.90              | 71.61              | 0.79           | 10.5        | 116.5                | ---                | 249.87             | 146.39             | 0.58             | 12.8           |

Table (4): Variance component estimates for peanut cultivar Giza 6 per plant pod and seed weight in M<sub>2</sub> generation.

| Treatment       | Seed Characters            |                    |                    |                    |                |             |                             |                    |                    |                    |                  |                |
|-----------------|----------------------------|--------------------|--------------------|--------------------|----------------|-------------|-----------------------------|--------------------|--------------------|--------------------|------------------|----------------|
|                 | Pod weight pl <sup>†</sup> |                    |                    |                    |                |             | Seed weight pl <sup>†</sup> |                    |                    |                    |                  |                |
|                 | Mean                       | $\hat{\sigma}^2_E$ | $\hat{\sigma}^2_P$ | $\hat{\sigma}^2_G$ | H <sub>i</sub> | $\hat{G}_s$ | Mean                        | $\hat{\sigma}^2_E$ | $\hat{\sigma}^2_P$ | $\hat{\sigma}^2_G$ | H <sub>i</sub> # | $\hat{G}_s$ †† |
| Cultivar Giza 6 |                            |                    |                    |                    |                |             |                             |                    |                    |                    |                  |                |
| Control         | 142.6                      | 56.43              | ---                | ---                | ---            | ---         | 131.7                       | 29.83              | ---                | ---                | ---              | ---            |
| γ rays, Gy      |                            |                    |                    |                    |                |             |                             |                    |                    |                    |                  |                |
| 100             | 175.1                      | ---                | 204.59             | 148.16             | 0.72           | 14.2        | 151.3                       | ---                | 99.15              | 69.32              | 0.70             | 9.8            |
| 200             | 169.8                      | ---                | 211.28             | 154.85             | 0.73           | 14.8        | 149.6                       | ---                | 107.05             | 77.22              | 0.72             | 10.4           |
| 300             | 137.1                      | ---                | 163.22             | 106.79             | 0.65           | 11.6        | 114.4                       | ---                | 80.57              | 50.74              | 0.63             | 7.9            |
| 400             | 128.3                      | ---                | 150.32             | 93.89              | 0.62           | 10.6        | 98.5                        | ---                | 75.49              | 45.66              | 0.60             | 7.3            |
| Mean            | 152.6                      | ---                | 182.35             | 125.92             | 0.68           | 12.8        | 128.5                       | ---                | 90.57              | 60.74              | 0.66             | 8.9            |
| EMS, mM         |                            |                    |                    |                    |                |             |                             |                    |                    |                    |                  |                |
| 1.0             | 159.4                      | ---                | 175.02             | 118.59             | 0.67           | 12.4        | 136.4                       | ---                | 115.33             | 85.50              | 0.74             | 11.1           |
| 2.0             | 168.5                      | ---                | 165.75             | 109.32             | 0.66           | 11.9        | 141.4                       | ---                | 107.03             | 77.20              | 0.72             | 10.4           |
| 3.0             | 166.1                      | ---                | 129.83             | 73.40              | 0.56           | 8.9         | 130.0                       | ---                | 117.69             | 87.86              | 0.74             | 11.2           |
| Mean            | 164.7                      | ---                | 156.87             | 100.44             | 0.63           | 11.1        | 135.9                       | ---                | 113.35             | 83.52              | 0.73             | 10.9           |
| SA, mM          |                            |                    |                    |                    |                |             |                             |                    |                    |                    |                  |                |
| 1.0             | 179.7                      | ---                | 197.83             | 141.40             | 0.71           | 13.9        | 157.1                       | ---                | 102.12             | 72.29              | 0.71             | 10.0           |
| 2.0             | 169.1                      | ---                | 189.76             | 133.40             | 0.70           | 13.5        | 141.2                       | ---                | 139.94             | 110.11             | 0.78             | 12.9           |
| 3.0             | 160.0                      | ---                | 181.67             | 125.24             | 0.69           | 13.0        | 134.6                       | ---                | 164.90             | 135.07             | 0.81             | 14.6           |
| Mean            | 169.6                      | ---                | 189.75             | 133.35             | 0.70           | 13.5        | 112.5                       | ---                | 135.65             | 105.82             | 0.77             | 12.5           |

Heritability estimates, mutagenwise, were relatively different (Figures 1-3) irrespective of seed character and cultivar. Sodium azide rates resulted in a difference of eight percentage points from gamma ray doses (70% vs. 62%) and of three from EMS doses (70% vs. 67%) (Fig. 1). These implies that, as Figure 1 shows, the broad-sense heritability estimates, which resulted as a response to all

mutagens, especially sodium azide, averaged over all four the studied seed characters, were relatively high within the dose range employed in this research. These indicates that, in general, selection for these specific seed characters seems effective under SA-treated seed conditions.



Within mutagen (Figs. 2 & 3), heritability estimates differed to some extent. Considering both number of pod per plant and pod weight (Fig. 2), estimates for both characters were slightly higher in Giza 6 than those of Giza 5 within all mutagens; however, within both cultivars, per plant pod number had the highest heritability estimates. Since this yield component character is positively correlated to total seed yield, selection to mutants of this character using all three mutagen dose range, would seem of potential to enhance seed yield. These urges looking for both effective and efficient screening techniques to identify mutants, which would aid in selecting for this character in advanced M generations. Figure 3 shows heritability estimates for both per plant seed number and seed weight in response to mutant-induced treatments. Heritabilities for per plant seed weight, regardless of cultivar and mutagen, were marginally greater than those obtained for per plant seed number.

Heritability estimate, within Giza 6, of pod weight per plant was moderately intermediate ranged from 63%-70% (Fig. 2). This range despite being comparatively lower than that of per plant pod number; yet it can, as well, be selected for using the sodium azide dose range applied in this research. But, this character is related closely to shelling percentage since, it does not necessarily express high seed weight within pod unless the associated shelling percentage is fairly high.

By adopting the same technique of individual plants to estimate the broad-sense heritability, many studies resulted in similar trends as the one obtained in this research concerning broad-sense heritability estimates. The estimated  $H_B$ , in  $M_2$  generation of gamma-irradiated and sodium azide-treated faba bean seeds, ranged from 75 to 92% averaged over doses and cultivars considering all seed yield components (Ahmed, 2007). The estimated environmental variance was as small as one-fourth of the phenotypic variance  $\sigma_p^2$ . Consequently, this led to a quite high contribution of the genetic variance component to the heritability estimates.

Similarly, peanut induced-mutagenesis studies have been conducted by Moustafa, 1994; Kassam and Esawy, 2003; and El-Shazly *et al.*, 2005. In  $M_2$  (El-Shazly *et al.*, 2005),  $H_B$  estimates were moderate to fairly high (56-85%) for cv Giza 6 sodium azide mutants for some yield components. For cv Giza 5  $M_2$  gamma-ray mutants (Moustafa, 1994),  $H_B$  estimates spanned a very wide range of 22% - 80% across gamma ray doses and seed yield component. Kassam and Esawy (2003), in addition, obtained quite high estimates, >94%, in  $M_2$  for all yield components for both Giza 4 and Giza 5 cultivars.

In trying to interpret the broad-sense heritability estimates, Ronis *et al.* (1985) concluded that the heritability values presented were calculated from individual spaced plants; thus, they may differ from those derived from plants grown in conventional row arrangement or culture, i.e. experimental plots. Another limitation to these heritability estimates, they further added, is that the genotypic variance estimates used in the calculations are probably biased upwards since the estimates of genotype X environment interaction in this procedure, obtained by using the mean variance of the parents, are probably underestimated. This would eventually tend to bias the heritability estimates upward. Also, across 13 different character for some cowpea genotypes, Omoigui *et al.* (2006) obtained moderate-to-high genetic variance, and a wide heritability-estimate range (19%-90%). They explained that the proportion of total variation attributable to the error variance was relatively small, this in turn led to high contribution of the genetic variance to the total phenotypic resulting in high heritability estimates.

In conclusion, it would be misleading trying to quantify the precision of any particular method of heritability estimation just based on *point estimation*, i.e. an estimated mean value.

Emphasis of variance component and heritability estimation experiments has been on point estimation (Knapp, *et al.*, 1987). It is necessary to estimate the standard error of various heritability estimates, therefore,

confidence interval can be estimated at a certain probability level since this interval estimation, rather than just an estimated point, is crucial, to the statistical inference process. Knapp, *et al.*, (1985) pointed out that estimates of heritability, among which those estimated on an individual progeny basis and on progeny mean basis, are frequently interpreted without reference to precision, or misleading measures of precision are often used. He referred this to the lack of research in this area because approximate or exact confidence

intervals have been derived for only a few heritability estimators. Both experiment size and point estimate magnitude affects interval estimate width (Knapp *et al.*, 1987). Henninger, *et al.* (2000) estimated the 95%.

Confidence interval for broad-sense heritability estimates based on 19 potatoes (*Solanum tuberosum* L.) clones evaluated at six environments (three years and two locations).

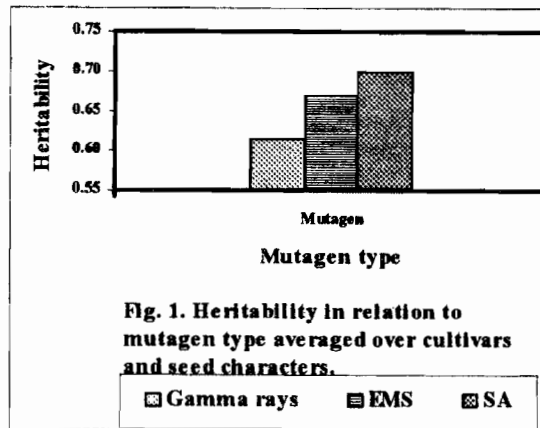


Fig. 1. Heritability in relation to mutagen type averaged over cultivars and seed characters.

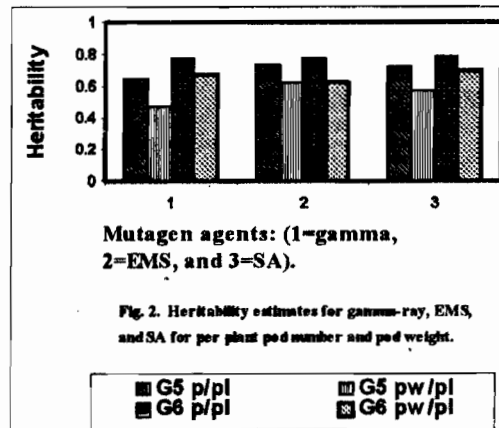


Fig. 2. Heritability estimates for gamma-ray, EMS, and SA for per plant pod number and pod weight.

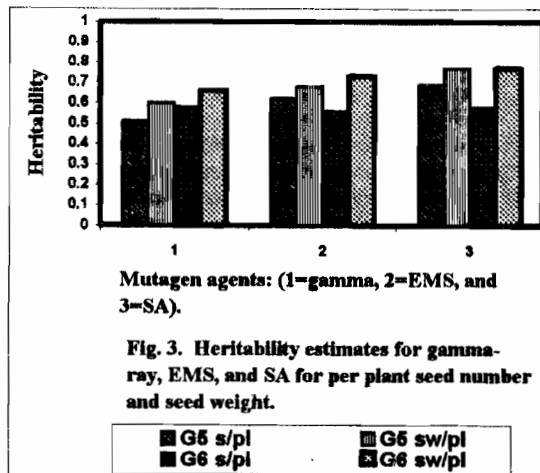


Fig. 3. Heritability estimates for gamma-ray, EMS, and SA for per plant seed number and seed weight.

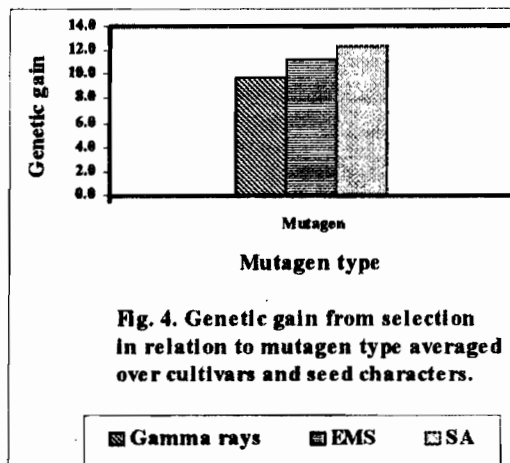


Fig. 4. Genetic gain from selection in relation to mutagen type averaged over cultivars and seed characters.

Unless genetic variance estimate can be separated from each of all possible interaction variances: genotype X location; genotype X year; and genotype X location X year, the estimated genetic variance would likely be biased up, and this would lead to expected similar bias in heritability estimate. This separation would not be possible unless evaluation be conducted in at least two locations, and during two or more years (حسن) (۲۰۰۵). This inflated heritability estimates

would probably occur if all interaction effects with genotypes are being important and neither considered nor calculated since evaluation was not carried out in more than two locations and/or more than two years (Fehr, 1987).

The estimated gain from selection,  $G_S$ , varied among mutagens within any of the four seed characters (Tables 1-5). There

was a consistent trend of a linear increase in estimated mean gain among the three mutagen types toward sodium azide within each seed character regardless of cultivar (Tables 1-4), and also when averaged over all seed characters and cultivars (Fig.4). Yet, no particular trend can be depicted among different mutagen doses within any mutagen in any of cultivar seed yield characters. In addition,

across both cultivars, difference between the same mutagen mean effect for the same seed character did differ that much. The highest difference was between SA mean effects, 17.5 vs. 12.5 g, in favor of Giza 5 seed weigh per plant, but gamma-ray mean effect caused a difference of about 5 g (12.8 vs. 8.0) for Giza 6 pod weight per plant.

Table (5): Estimates of mean, heritability and Genetic gain for all the studied traits for two Cultivars of mutagen type.

| Seed Characters | Mutagen Type   | Mean   |        | H.B  |      | G. S  |       |
|-----------------|----------------|--------|--------|------|------|-------|-------|
|                 |                | G 5    | G6     | G5   | G6   | G 5   | G6    |
| Pod PL-1        | Gamma-rays,GY  | 62.10  | 62.40  | 0.65 | 0.78 | 7.10  | 10.10 |
|                 | EMS, mM        | 62.10  | 64.50  | 0.74 | 0.78 | 9.10  | 9.10  |
|                 | SA, mM         | 66.80  | 62.70  | 0.73 | 0.79 | 10.10 | 10.50 |
| Seed PL-1       | Gamma-rays, GY | 106.30 | 106.30 | 0.51 | 0.85 | 8.30  | 12.60 |
|                 | EMS, mM        | 121.00 | 106.20 | 0.62 | 0.56 | 11.70 | 12.40 |
|                 | SA, mM         | 127.30 | 116.50 | 0.69 | 0.58 | 9.70  | 12.80 |
| Pod weight PL-1 | Gamma-rays,GY  | 142.20 | 152.60 | 0.47 | 0.68 | 8.00  | 12.80 |
|                 | EMS, mM        | 155.00 | 164.70 | 0.62 | 0.63 | 12.70 | 11.10 |
|                 | SA, mM         | 177.70 | 169.60 | 0.58 | 0.70 | 12.10 | 13.50 |
| Seed weight PL1 | Gamma-rays, GY | 117.70 | 128.50 | 0.60 | 0.66 | 10.30 | 8.90  |
|                 | EMS, mM        | 139.60 | 135.90 | 0.68 | 0.73 | 12.80 | 10.90 |
|                 | SA, mM         | 158.10 | 112.50 | 0.77 | 0.77 | 17.50 | 12.50 |

Based on the results (Tables 1-5) averaged over both mutagens and cultivars, it would be possible to change the mean pod number per plant in a few mutated generations of selection under 20% selection intensity by about 9.3 pods per plant, 11.3 seeds per plant, by 11.7 g in pod weight per plant, and by 11.7 g seed weight per plant.

Using 150 Gy gamma-ray dose (Adu-Dabaah and Sangwan, 2004), the predicted genetic gain values were high in the M<sub>3</sub> mutants. They were about 16 pods per plant, 14 g for seed index, and 12 g for seed yield per plant averaged over the tested two bambara groundnut cultivars.

The estimates of gain from selection need to be carefully interpreted since many factors may influence these values. Any bias in heritability estimates –due to any of/all reasons that have been previously discussed— would accordingly lead to similar upward bias in the estimated values of gain from selection as well. Ronis *et al.* (1985) argued these were also heritability values in the broad-sense and are useful as first approximation, not as definitive values. Assuming that effective mutant screening procedure(s) have already been set up; hence, screening for induced mutant by narrowing down selection intensity to 10% or 5% (i.e. selection differential of k = 1.75 at 10% or = 2.06 at k= 5%) in few generations would greatly change expected gain.

### CONCLUSIONS

Egypt lags behind many developing countries which have seriously listed the issue of enhancing their oil crop genetic pool among their national agronomic advancement plans

especially those on the mutagenic level. It is, therefore, urgent to locally incorporate a more advanced-level mutagen-based research plan starts at the recent DNA-level research results.

In induced mutagenesis research area, the stage of investigating just the mutagen-induced effects on different crop yields and chemical traits whether from the genetic or the character location perspectives should be pursued by more advanced analytical stages. These subsequent stages should be based on valid estimates of heritability using more advanced statistical models. In addition, they should possess the capability of offering more tangible evidence of the potentials of induced mutagenesis as a concrete plant breeding tool to share effectively in developing new released cultivars superior to the already released ones in one or more character. The

success of these advanced stages of research projects will certainly depends upon the appropriateness of identifying real induced mutants resulted from a range of effective mutagen doses rather than selecting just a single dose based on insufficient data. Additionally, effective and efficient screening techniques, at the molecular levels, should be employed on the part of induced-mutagenesis plant breeder to identify and assess the resultant few real mutants. This is for the purpose of trying to understand the mode(s) of action of the effective mutagen dose *via* sequencing gene(s) encodes certain characters in these mutants.

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التغيرات المستحدثة باستخدام المطفرات في تقديرات التباين الوراثي ودرجة التوارث لصفات محصول بذور الفول السوداني

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\*\* قسم بحوث النبات، هيئة الطاقة الذرية، أنشاص، مصر

في الوقت الحالي يفتقر نبات الفول السوداني للقاعدة الوراثية العريضة في مصر حيث يوجد حالياً صنفان فقط منه متوفران للمزارع المصري. ولما كان استنباط أصنافاً جديدة يعتمد اعتماداً كبيراً على وجود تلك القاعدة الوراثية العريضة كخطوة أساسية وضرورية للمربي للانطلاق منها، لذا يلزم تضاعف كافة طرق تربية النبات الممكنة والتي تناسب طبيعة نمو هذا النبات، من الناحية الوراثية، كنبات ذاتي التلقيح لتوسيع تلك القاعدة حتى يتسنى للمربي استنباط سلالات مبشرة تتميز بتفوقها في المحصول أو/و مقاومتها للأمراض.

من تلك الطرق استخدام الطفرات المستحدثة صناعياً جراً معالجة البذور بالمطفرات الفيزيائية والكيميائية. حيث تعد أشعة جاما و سلفات ايثيل الميثان وأزاييد الصوديوم من أكثر المطفرات شيوعاً في الاستخدام في مجال تربية النبات باستخدام المطفرات. وتختلف الجرعات المؤثرة من كل مطفر حسب نوع النبات المحصولي تحت الدراسة. وتم تنفيذ هذا البحث أساساً إلى تقدير كل من التباين الظاهري و الوراثي، معامل التوريث بالمعنى الواسع، وكذلك التقدم الوراثي الناتج عن الانتخاب لأهم صفات المحصول لبذور صنفين تجاريين من الفول السوداني -جيزة ٥، وجيزة ٦- التي عولمت بجرعات مختلفة من أشعة جاما ومادتا سلفات ايثيل الميثان وأزاييد الصوديوم بهدف استحداث تغيرات طفورية ومن ثم محاولة تقييمها في الجيل الانعزالي الثاني. حيث تعرضت بذور صنف الفول السوداني لأربعة جرعات من أشعة جاما (كوبالت ٦٠) - ١٠٠، ٢٠٠، ٣٠٠، ٤٠٠ جراي- بالإضافة إلى ثلاث جرعات من كل من سلفات ايثيل الميثان و أزاييد الصوديوم -١، ٢، ٣ مللي مول- عند درجة حوضة ٣٠، في محلول فوسفاتي ٠،١ مول. وقد أجريت التجارب الحقلية في محطة بحوث تجارب هيئة الطاقة المصرية بأنشاص في الأعوام ٢٠٠٢، ٢٠٠٣، ٢٠٠٤.

أوضحت النتائج في الجيل الثاني إحداث تباينات ظاهرية ووراثية غير متجانسة بين المطفرات الثلاثة المختلفة فقط بالنسبة للصفة جيزة ٥. ففي الصنف جيزة ٥، أدت المعالجة بجرعات آزاييد الصوديوم إلى إحداث اختلافات معنوية فيما بينها في التباينات الظاهرية والوراثية في صفات المحصول تحت الدراسة وهي عدد القرون، عدد البذور، وزن القرون، وأخيرا وزن البذور للنبات. عموما، كان مدي قيم درجة التوارث علي المستوي العريض يتراوح ما بين ٠,٤٧-٠,٧٧ لجيزة ٥، وما بين ٠,٥٦-٠,٧٩ لجيزة ٦ ذلك بناءا علي المتوسط الحسابي عبر المطفرات الثلاثة وعبر صفات المحصول. حيث أحدث آزاييد الصوديوم ٨% تفوقا عن أشعة جاما (٧٠% مقابل ٦٢%)، و آخر قدره ٣% عن سلفات ايثيل الميثان. أما الاختلافات في قيم درجة التوارث بين جرعات أي من المطفرات الثلاثة فلم تسلك اتجاها محددًا لأي من صفات المحصول المدروسة علي عكس نتائج أبحاث أخرى شملت تقريبا نفس جرعات المطفرة.

وبرغم وجود جدل كبير حول اعتبار المطفرات كوسيلة فعالة من وسائل تربية النبات لأحداث توسيع للقاعدة الوراثية لنبات ما، وكذلك وجود تساؤلات كثيرة حول فعاليتها في هذا المجال، إلا أنها تستخدم علي نطاق واسع في كثير من بلدان العالم خاصة دول العالم الثالث. في تلك الدول ينصب غالبا الاهتمام البحثي علي مجال استخدام تلك المطفرات لتحسين إنتاجية المحاصيل من خلال الاهتمام بما تحدته تلك المطفرات الفيزيائية منها أو الكيميائية من تغيرات -أي كانت- يسفر عنها تحسن في صفات المحصول المختلفة. أما في الدول المتقدمة، فهناك خطوات تخطت بها مجال الأبحاث تلك المرحلة إلى دراسات علي مستوي الحمض النووي لمحاولة فهم ما تحدته تلك المطفرات من تغيرات في الجينات المسؤولة عن صفة تهم المربي في بعض تلك الطفرات المستحدثة. يتحتم إذن في الأبحاث المحلية المستقبلية تعدي مرحلة البحث في التغيرات سواء أكانت في قيمة المتوسط الحسابي للمتغير التابع أم في قيمة المتغير الوراثي التي تحدتها أي من المطفرات إلى البحث في كون تلك التغيرات نشأت نتيجة طفرات حقيقية، وهل هي موقع جيني جديد أم مجرد أليلات لجينات هي بالفعل كانت موجودة مسبقا