The Effects of Different Doses of Gamma Ray and EMS on Formation of Chlorophyll Mutations in Durum Wheat (Triticum durum Desf.)

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Abstract: The main target of breeding programmes has been to develop high yielding durum wheat (*Triticum durum* Desf.) cultivars. Breeding programmes aim to exploit the existing variability and to enhance genetic variation. Genetic variation may be broadened through experimental mutagenesis. An important plant breeding problem is the selection of suitable mutagenic treatment. The aim of this study was to determine the sensitivity of durum wheat cultivars to mutagens by detecting fertility of M₁ plants and chlorophyll mutations in M₂ plants. This study was carried out in Tokat-Kazova conditions in 1996 and 1997. Gediz-75 and Sofu durum wheat cultivars were used as plant material in the frial. The gamma rays obtained from Cobalt 60 (⁶⁰Co) as physical mutagen and the EMS as chemical mutagen were used. The seeds were irradiated with 50 Gy, 100 Gy, 150 Gy and 200 Gy gamma rays or treated with 0.1 %, 0.2 %, 0.3 % and 0.4 % EMS (Ethyl Methane Sulphonate). The seeds were treated with EMS at 24 °C for 8 hours without presoaked and were washed for 6 hours after treatment. Either of treated cultivars were grown separately in the trial. Besides, the treatments of gamma rays and EMS separately were "Randomized Complete Block Design" with three replications. According to the results of this research, fertility in M₁ plants was lower with gamma ray application compared to EMS application. The spectrum and frequency of mutations were varied with treatments of mutagen and cultivars. The effect of mutagens markedly increased with high doses. The highest mutagenic efficiency was obtained from 0.4 % EMS dose in Gediz-75 cultivar and from 100 Gy gamma ray dose in Sofu cultivar.

Key Words: durum wheat, gamma rays, EMS, chlorophyll mutations, mutation frequency, mutagenic efficiency

Makarnalık Buğdayda (*Triticum durum* Desf.) Gama Işını ve EMS'nin Farklı Dozlarının Klorofil Mutasyonlarının Oluşumu Üzerine Etkileri

Özet: İslah programlarının temel hedefi verimi yüksek makarnalık buğday (*Triticum durum* Desf.) çeşitlerini geliştirmektir. İslah programları mevcut olan çeşitliliği artırmayı ve genetik varyasyonu zenginleştirmeyi amaçlar. Genetik varyasyon mutasyon yoluyla artırılabilir. Bitki islahının önemli bir sorunu uygun mutagenik muamelenin seçimidir. Bu çalışmanın amacı, makarnalık buğday çeşitlerinin mutagenlere duyarlılığını Mı bitkilerinin fertilitesi ve M₂ bitkilerinde kiorofil mutasyonlarını belirleyerek saptamaktır. Bu çalışma 1996 ve 1997 yıllarında Tokat-Kazova şartlarında yürütülmüştür. Araştırmada bitki materyali olarak Gediz-75 ve Sofu makarnalık buğday çeşitleri kullanılmıştır. Kobalt 60 (⁸⁰Co) kaynağından elde edilen gama ışını tohumları fiziksel mutagen ve EMS kimyasal mutagen olarak uygulanmıştır. EMS uygulaması ön ıslatma yapılmadan 8 saat oda sıcaklığında yapılmış ve uygulama süresi sonunda tohumlar 6 saat süreyle yıkanmıştır. Tohumlar 50 Gy, 100 Gy, 150 Gy ve 200 Gy gama ışını ile ışınlanmış ve % 0.1, % 0.2, % 0.3, % 0.4 EMS ile muamele edilmiştir. Mutagenlerin uygulandığı çeşitler ayrı ayrı denemeye alınmıştır. Ayrıca, gama ışını ve EMS uygulamaları birbirinden ayrı olarak "Tesadüf Blokları Deneme Deseni"nde 3 tekerrürlü olarak kurulmuştur. Araştırma sonuçlarına göre, Mı bitkilerinin fertilitesi EMS uygulamasıyla karşılaştırıldığı zaman gamma ışını uygulamasında daha düşük çıkmıştır. Mutasyon spektrumu ve frekansı muamelerle ve çeşitlere bağlı olarak değişmiştir. Mutagenlerin etkileri yüksek dozlarda belirgin bir şekilde artmıştır. En yüksek mutagenik verim Gediz-75 çeşidinde % 0.4 EMS dozundan, Sofu çeşidinde ise 100 Gy gama ışını dozundan elde edilmiştir.

Anahtar Kelimeler: makarnalık buğday, gama ışını, EMS, klorofil mutasyonu, mutasyon frekansı, mutagenik verim

Introduction

Durum wheat cultivars used are poor in quality, and yield is less compared to bread wheat cultivars in Turkey. Therefore, the main target of breeding programmes has been to develop high yielding durum wheat cultivars. Breeding programmes aim to exploit the existing variability and to enhance genetic variation.

Genetic variation may be broadened through experimental mutagenesis. An important plant breeding problem is the selection of suitable mutagenic treatment (Wellensiek 1965). Suitable treatment doses/concentrations must be established and the relative efficiency of mutagens for chlorophyll mutations must be determined. This is established by compiling the results obtained for several varieties and analysing the M₂ chlorophyll mutation frequency on a spike progeny basis.

The frequency and type of mutation recovered can vary with dose and rate at which the mutagen is applied, the species, the genotype within a species

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(Walter et al. 1987). There are significant differences among species and genotypes within species for sensivity to mutagen treatments (Tavcar 1965, Wellensiek 1965, Siminel and Paladi 1979, Gottschalk and Wolff 1983, Yanev 1985, Tavil 1986, Walter et al. 1987). The sensitivity to mutagen depends on several genetical and external conditions (Tavcar 1965).

The differences occured in fertility of M₁ plants are a measure of sensitivity of a genotype to mutagen. With higher gamma ray and EMS doses, the fertility of M₁ plants decreased significantly compared to control in durum wheat cultivars (Taycar 1965, Yanev 1985, Çiftçi et al. 1988, Şenay 1997), Mutagens induce the reduction of the emergence rate of seedlings in M₂ generation (Peşkircioğlu 1995, Şenay 1997).

The physical and chemical mutagens induce different mutation spectra (Konzak et al. 1965, Gottschalk and Wolff 1983). Environmental conditions may markedly affect the expression of chlorophyll deficient mutants (Konzak et al. 1965). The spectrum of mutations can be quite different depending on the species, population structure, or chromosome organization (Gustafsson 1965). Nilan et al. (1965) reported that Caldecott (1955) and Mikaelsen (1958) found no differences in barley seedling mutation spectrum induced by X-rays, gamma rays, and neutrons. The largest mutation spectrum was obtained at the high doses and varied with cultivars (Paladi and Siminel 1979). Zannone (1965) observed the widest spectrum in EMS, with 12 types of mutations. Different types of chlorophyll mutations occured with different mutagens in durum wheat (Caldecott et al. 1965, D'amato 1965, Kadagidze 1979, Yanev 1980, Şenay 1997)

The mutagenic effect was evaluated on the basis of the frequency of chlorophyll mutations per M₁ plant progeny and on the basis of the ratio of M₂ mutants to the total number of M₂ seedling observed (Zannone 1965). The frequency of chlorophyll mutations were different among mutagens (Swaminathan 1965, Yanev 1980). Scarascia-Mugnoza et al. (1991) reported that the mutation frequency of the chemical mutagens were lower than those of physical mutagens. Ratio of segregating M₂ seedlings increases with increasing dose or concentration. Similar results were obtained by other researches (D'amato 1965, Swaminathan 1965, Raimkulov and Maatkarimov 1979, Şenay 1997). Also, the mutation frequency varied with cultivars (Swaminathan 1965, Minocha et al. 1979, Yanev 1985, Tavil 1986)

Konzak et al. (1965) reported that mutagens might be equal in mutagenic effectiveness, but they differ in their production of undesirable changes (such as gross chromosome aberrations, sterility and lethality). However, mutagens may differ in mutagenic efficiency. The mutagenic efficiency of the two physical mutagens (gamma rays and fast neutrons) was more or less the same (Siddiqui et al. 1991). The treatments that yielded the highest mutation frequency may not confer the highest mutagenic efficiency (Peşkircioğlu 1995, Şenay 1997).

The higher the mutation frequency, the higher the genetic variation is going to occur for selection in M₂ or M₃

generations. The aim of this study was to determine the sensitivity of durum wheat cultivars to mutagens by detecting chlorophyll mutations in M₂ plants.

Material and Methods

Sofu and Gediz-75 durum wheat cultivars (*Tnlicum durum* Desf.) were used as plant material. The gamma rays obtained from Cobalt 60 (⁶⁰Co) as physical mutagen, and the EMS (Ethyl Methane Sulphonate), which is alkylating group, as chemical mutagen were used. The seeds were irradiated with 50, 100, 150, 200 Gy gamma ray (Anonymous 1977) or treated with 0.1 %, 0.2 %, 0.3 %, 0.4 % EMS (Çiftçi et al. 1988). The cultivars were separately grown in the trial. Besides, the seeds treated with different mutagens were separately grown.

The seeds obtained from M₁ spikes in the first year were sown as a plant progeny line for the M2 generation in the second year. A control line was planted for every ten treatment lines. The number of M1 spikes obtained from 150 and 200 Gy doses were not enough and the spikes had low fertility. Therefore, the plots of these doses were taken out in the M2 generation. The rate of fertility and the percentage of M2 plants emergencing were subjected to analysis of variance (ANOVA) using Statistical Software Package. Arcsin transformation was applied to data (Düzgünes et al. 1987), Comparisons among dose means were done using the LSD (Least Significant Difference) test. Data were obtained for fertility of M1 plants according to Senay (1997), for the percentage of M2 plants emergencing according to Univer (1989), for chlorophyll mutation types according to Gustafsson (1940) and Holm (1954), and for mutation frequency and mutagenic efficiency according to Konzak et al. (1965) and Zannone (1965).

In order to estimate the fertility values, the number of kernel was divided the number of flower in spike. The percentage of M₂ plants emergencing in the field were determined per each line and calculated the averages for treatments. Chlorophyll mutations were observed on the M₂ plants of each line according to different mutation types. Mutation frequency was evaluated on a spike basis by using the formula of (total chlorophyll mutation / total M₁ spikes) x 100 and on a seedling basis by using the formula of (total chlorophyll mutation / total M₂ seedlings) x 100. The formula of (mutation frequency / % reduction) has been calculated as measure of mutagenic efficiency. Seed sterility in M₁ plants was used as % reduction.

Results and Discussion

Fertility of M₁ plants: The comparison between the plants of control and treatment plots showed significant differences in both cultivars for gamma irradiation. EMS treatments however, were not significantly different Table 1. The fertility was decreased drastically with increasing gamma ray doses. The highest fertility was observed in control, but the value decreased about 70 % in the higher doses (150 and 200 Gy), Similar result was reported by researchers (Tavcar 1965, Yanev 1985, Çifiçi et al. 1988, Şenay 1997). On the contrary, fertility of M₁ plants was

unaffected by physical mutagens, like gamma ray, X rays and neutron (Wellensiek 1965, Zannone 1965). The control plots of the cultivars and the other treatments had the high sterility. The reason for high sterility could be high temperature in flowering period.

Fertility was lower with gamma ray application compared to EMS application Figure 1. On the contrary, some researchers reported that EMS was given higher sterility than gamma rays (Zannone 1965, Hussein and Disouki 1976). This reason for this could be differences in the treatment conditions, environmental factors and cultivars or species. Drop in M1 -fertility after mutagenic treatments is a general feature in mutation breeding experiments. However, the amount of induced sterility depends on a variety of factors, among them one is the type of mutagen (Hussein and Disouki 1976). Seed sterility may be an outcome of meiotic irregularities (Datta and Biswas 1985). Most of the sterilities noted in plant species after exposure to radiations have been mainly due to chromosomal aberrations (Magri-Allegra and Zannone 1965). Radiation induced sterility has been attributed to detectable aberrations and cryptic deficiencies; while in

EMS and other chemical mutagens, sterility has been ascribed to crytic deletions and specific gene mutation (Sato and Gaul 1967).

The percentage of M₂ plants emergencing: Gamma irradiation and EMS treatments did not significantly affect the percentage of M2 plants emergencing in the both cultivars Table 2. However, gamma irradiation reduced the percentage of emergence compared to control in the both cultivars. But, the reduction was quite small (0.7% - 3.7%). The EMS treatments caused different effects according to cultivars. The highest percentage of emergence (91.6 %) was obtained from control and 0.1 % EMS dose in Gediz-75. The decreases were found at 0.2 %, 0.3 % and 0.4 % EMS doses. But, the values obtained from EMS treatments were similar or higher than control in Sofu. On the contrary, some researchers reported thad gamma irradiation and EMS treatment significantly reduced the percentage of M2 plants emergencing compared to control (Peşkircioğlu 1995, Şenay 1997). The reason for this could be differences cultivars and environmental factors.

Table 1. The fertility of M₁ plants of durum wheat cultivars after treatment with mutagens

| Treatments | Doses | | GEDIZ | -75 | | SOFU | | | | |
|------------|---------|---|-----------------|------|-------------|----------------|-----------|----------|-------------|--|
| | | Observed value | Transfo valu | 4444 | % Reduction | Observed value | Transform | ed value | % Reduction | |
| | Control | 58.50 | 49.90 | a** | | 56.73 | 48.87 | a** | | |
| | 50 Gy | 47.10 | 43.33 | a | 19.49 | 51.97 | 46.13 | ab | 8.39 | |
| Gamma | 100 Gy | 32.77 | 34.89 | b | 43.98 | 36.87 | 37.28 | b | 35.01 | |
| ray | 150 Gy | 23.20 | 28.57 | bc | 60.34 | 17.90 | 24.98 | C | 68.45 | |
| | 200 Gy | 14.73 | 22.57 | C | 74.82 | | - | | - | |
| | Average | 35.26 | | | | 40.87 | | | | |
| | LSD | 6.65 | | | | 9.54 | | | | |
| | Control | 58.20 | 49.75 | | | 56.80 | 48.93 | | | |
| | % 0.1 | 50.97 | 45.56 | | 12.42 | 55.57 | 48.20 | | 2.16 | |
| | % 0.2 | 52.00 | 46.15 | | 10.65 | 48.97 | 44.41 | | 13.79 | |
| EMS | % 0.3 | 46.07 | 42.74 | | 20.84 | 52.73 | 46.57 | | 7.17 | |
| | % 0.4 | 47.20 | 43.39 | | 18.90 | 50.00 | 45.00 | | 11.97 | |
| | Average | 011000110110111011101110110110110110110 | 50.89 | 9 | | 52.81 | | | | |
| | LSD | | NS | | | | NS | | | |

^{**} indicates significance at 0.01, NS indicates not significant.

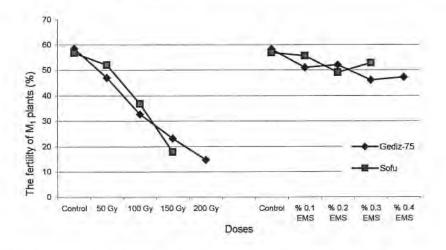


Figure 1. Percent of fertilitiy in M1 plants of durum wheat cultivars in seed treatments with gamma ray and EMS

The spectrum of chlorophyll mutation types: The spectrum of chlorophyll mutations obtained by the gamma ray application and EMS treatments were varied with the cultivar (Table 3). With the gamma irradiation, three types of chlorophyll mutations were determined in Gediz-75, while five types were observed in Sofu. Moreover, the ratios of types were different. Gustafsson (1965) and Paladi and Siminel (1979) also reported similar results. The most common chlorophyll mutation type was xhanta (% 62.4) in Gediz-75 while albino (% 26.8) and viridoalbino (%26.8) in Sofu. The albino and xantha types were also reported to be the most common by other researchers (Caldecott et al. 1965, D'amato 1965, Kadagidze 1979, Şenay 1997). However Yanev (1980) found that the highest ratio of chlorophyll mutation was striata type in durum wheat. This result could be due to different cultivar or to mutagens.

In the EMS treatment, five types of chlorophyll mutations were obtained in Gediz-75 while four types in Sofu Table 3. The highest percentage of chlorophyll mutation type was viridis in both cultivars. The distribution of other types varied with cultivar. Mutagens produced different type of chlorophyll mutations in other studies (D'amato 1965, Yanev 1980, Şenay 1997) with the durum wheat, possibly because the cultivars used were different. Besides, they were grown under different environment. Environmental conditions may markedly affect the expression of chlorophyll deficient mutants (Konzak et al. 1965). The largest mutation spectrum were obtained from 0.3 % and 0.4 % EMS doses in Gediz-75 while from 0.3 % EMS dose in Sofu, Paladi and Siminel (1979) also

reported that spectrum of mutation increased with increasing doses.

Gamma irradiation and EMS treatments affected the distributions of chlorophyll mutation types. In Table 4 and Figure 2, the frequency distributions of different types chlorophyll mutations observed in each treatment are reported. Mutation spectrum were different in the chemical and physical mutagen treatments (Konzak et al. 1965, Gottschalk and Wolff 1983). The spectrum of mutation is essentially a parameter for the index of mutation frequency (Datta and Biswas 1985). Chlorophyll development seems to be controlled by many genes located near the centromere and proximal segments of the chromosomes (Swaminathan 1965). The high incidence of different types of the chlorophyll mutations in the EMS treatments may be due to its specificity to affect certain regions of the chromosomes (Natarajan and Upadhya 1964).

Frequency of mutations: The frequency of mutations indicate the differences of cultivars and mutagen treatments Table 5. In the gamma irradiation, the frequency of mutation was 14.2 % for the M₁ spikes segregating in M₂ and 0.53% for the M₂ seedlings at 50 Gy dose in Gediz-75. These values were 23.5 % and 0.19 % at 50 Gy dose, 107.7 % and 3.95 % at 100 Gy dose in Sofu, respectively. In Sofu cultivar, mutation frequency increased with increasing gamma doses. This result was also found by other researchers (D'amato 1965, Swaminathan 1965, Raimkulov and Maatkarimov 1979, Şenay 1997).

Table 2. The percentage of M₂ plants emergencing of durum wheat cultivars after treatment with mutagens

| Treatments | Cultivar | Doses | The number of M ₁ spikes | The kernel of M ₁ sowing | The number of M ₂ seedlings | | ge of M ₂ plants noing (%) | % Change |
|------------|----------|---------|-------------------------------------|--|---|-------------------|--|-------------|
| | | | | | | Observed value | Transformed value | |
| | Gediz-75 | Control | 21 | 575 | 522 | 90.8 | 72.3 | |
| | | 50 Gy | 113 | 3401 | 3013 | 88.6 | 70.3 | -2.4 |
| | | 100 Gy | 50 | 1470 | 1327 | 90.3 | 71.9 | -0.7 |
| | | Average | | | | 89.9 | 3.00.00 | |
| Gamma | | LSD | ······· | | | | VS | |
| ray | Sofu | Control | 16 | 485 | 456 | 94.0 | 75,8 | |
| | | 50 Gy | 115 | 3432 | 3166 | 92.2 | 73.8 | -1.8 |
| | | 100 Gy | 13 | 392 | 354 | 90.3 | 71.9 | -3.7 |
| | | Average | | | | 92.2 | | |
| | | LSD | | - 1911 15-711111 | | 1 | VS | |
| | Gediz-75 | Control | 40 | 1203 | 1102 | 91.6 | 73.2 | |
| | | % D.1 | 94 | 2834 | 2595 | 91.6 | 73.2 | 0.0 |
| | | % 0.2 | 105 | 3174 | 2879 | 90.7 | 72.2 | -0.9 |
| | | % 0.3 | 73 | 2192 | 1975 | 90.1 | 71.7 | -1.5 |
| | | % 0.4 | 69 | 2070 | 1859 | 89.8 | 71.4 | -1.8 |
| | | Average | | | | 90.8 | | |
| EMS | | LSD | | | | 1 | NS . | |
| | Sofu | Control | 47 | 1418 | 1268 | 89.4 | 71.0 | |
| | | % 0.1 | 110 | 3307 | 2974 | 89.9 | 71.5 | - 0.5 |
| | | % 0.2 | 110 | 3304 | 3006 | 91.0 | 72.5 | - 1.6 |
| | | % 0.3 | 110 | 3310 | 2981 | 90.1 | 71.7 | = 0.7 |
| | | % 0.4 | 100 | 2997 | 2678 | 89,4 | 71,0 | 0.0 |
| | | Average | | | | 90.0 | incre- | |
| | | LSD | | | | | NS | |

Table 3. The spectrum of chlorophyll mutations in durum wheat

| Treatments | Cultivar | Doses | Albino | Xanta | Virido Albino | Viridis | Striata | Maculata | Total | % |
|------------|----------|--------|--------|-------|---------------|---------|---------|----------|-------|-----|
| | Gediz-75 | 50 Gy | 0 | 10 | 3 | 3 | 0 | 0 | 16 | 100 |
| | | 100 Gy | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | | Total | 0 | 10 | 3 | 3 | 0 | 0 | 16 | 100 |
| Gamma | | % | | 62.4 | 18.8 | 18.8 | | | | |
| ray | Sofu | 50 Gy | 5 | 9 | 6 | 4 | 0 | 3 | 27 | 66 |
| | | 100 Gy | 6 | 0 | 5 | 3 | 0 | 0 | 14 | 34 |
| | | Total | 11 | 9 | 11 | 7 | 0 | 3 | 41 | 100 |
| | | % | 26.8 | 22.0 | 26.8 | 17.1 | | 7.3 | | |
| | Gediz-75 | % 0.1 | 0 | 0 | 0 | 10 | 0 | 0 | 10 | 10 |
| | | % 0.2 | 0 | 0 | 0 | 5 | 1 | 0 | 6 | 6 |
| | | % 0.3 | 2 | 5 | 3 | 26 | 3 | 0 | 39 | 40 |
| | | % 0.4 | 7 | 0 | 11 | 20 | 5 | 0 | 43 | 44 |
| | | Total | 9 | 5 | 14 | 61 | 9 | 0 | 98 | 100 |
| EMS | | % | 9.2 | 5.1 | 14.3 | 62.2 | 9.2 | | | |
| | Sofu | % 0.1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | | % 0.2 | 1 | 6 | 0 | 4 | 0 | 0 | 11 | 33 |
| | | % 0.3 | 1 | 4 | 3 | 12 | 0 | 0 | 20 | 61 |
| | | % 0.4 | 0 | 2 | 0 | 0 | 0 | 0 | 2 | 6 |
| | | Total | 2 | 12 | 3 | 16 | 0 | 0 | 33 | 100 |
| | | % | 6.1 | 36.3 | 9.1 | 48.5 | | | | |

Table 4. The spectrum of chlorophyll mutations (values in per cent) in Triticum durum (Desf.) in different mutagenic treatments

| Treatments | Total number of mutations | Albino | Xanta | Virido albino | Viridis | Striata | Maculata |
|------------|---------------------------|--------|-------|---------------|---------|---------|----------|
| Gamma ray | 57 | 19.3 | 33.3 | 24.6 | 17.5 | 7. | 5.3 |
| EMS | 131 | 8.4 | 13.0 | 13.0 | 58.7 | 6.9 | - |

Table 5. The frequency of mutation of gamma ray and EMS

| Treatments | Cultivar | Doses | M ₁ spikes no. | M ₂ seedling no. | Segregating spikes no. | Total chlorophyll mutation | Mutation M ₁ spike | frequency M ₂ seedling |
|------------|----------|---------|---------------------------|------------------------------|------------------------|----------------------------|----------------------------------|--------------------------------------|
| | Gediz-75 | 50 Gy | 113 | 3013 | 5 | 16 | 14.2 | 0,53 |
| | | 100 Gy | 50 | 1327 | | | | |
| Gamma | | Average | | | | | 14.2 | 0.53 |
| ray | Sofu | 50 Gy | 115 | 3166 | 6 | 27 | 23.5 | 0.19 |
| | | 100 Gy | 13 | 354 | 3 | 14 | 107.7 | 3.95 |
| | | Average | | | | | 65.6 | 2.07 |
| | Gediz-75 | % 0.1 | 94 | 2595 | 4 | 10 | 10.6 | 0.39 |
| | | % 0.2 | 105 | 2879 | 4 | 6 | 5.7 | 0.21 |
| | | % 0.3 | 73 | 1975 | 11 | 39 | 53.4 | 1.97 |
| | | % 0.4 | 69 | 1859 | 7 | 43 | 62.3 | 2.31 |
| EMS | | Average | | manus aparticus Sandariomics | | | 33.0 | 1.22 |
| | Sofu | % 0.1 | 110 | 2974 | | | | |
| | | % 0.2 | 110 | 3006 | 3 | 11 | 10.0 | 0.37 |
| | | % 0.3 | 110 | 2981 | 9 | 20 | 18.2 | 0.67 |
| | | % 0.4 | 100 | 2678 | 1 | 2 | 2.0 | 0.07 |
| | | Average | | | | | 10.1 | 0.37 |

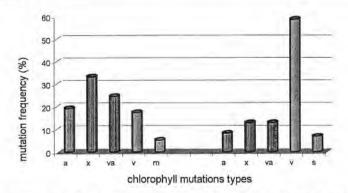


Figure 2. The spectrum of chlorophyll mutations (values in per cent) in *Triticum durum* (Desf.) in different mutagenic treatments (a= albino; x= xantha; va= virido albino; v= viridis; s= striata; m= maculata)

With EMS treatment, mutation frequency gone up with increasing doses. The highest mutation frequency was obtained from 0.4 % EMS dose in Gediz-75, but from 0.3 % in Sofu. These findings are in agreement with previous studies (D'amato 1965, Swaminathan 1965, Raimkulov and Maatkarimov 1979, Senay 1997). The response of durum wheat cultivars to EMS treatments were different. The effects of mutagens varied with cultivars (Swaminathan 1965, Minocha et al. 1979, Yanev 1985, Tavil 1986). M₁ seed sterillty has demostrated parallel relationship with mutation frequency mostly Table 1. Donini et al. (1974) also reported that the highest mutation frequencies are reached in those M₁ plants showing the highest reduction in spike fertility.

The frequency of mutations with EMS treatments were higher than those of gamma irradiation with Gediz-75 cultivar. These results confirm the findings of Gaul (1962). However, some researchers reported much higher segregating M₁ spikes with physical mutagens (D'amato 1965, Yanev 1980, Scarascia-Mugnozza et al. 1991). Similarly, gamma irradiation resulted in a higher mutation frequency than EMS treatment in Sofu cultivar. The cultivars reacted in different ways to mutagen.

Mutagenic efficiency: The mutagenic efficiency indicates differences of cultivars to gamma irradiation Table 6. The mutagenic efficiency was found as 0.73 in Gediz-75 and 2.80 in Sofu at 50 Gy dose. The mutagenic efficiency varied with the rate of mutation frequency and sterility at the same dose for both cultivars. Although the ratios of mutation frequency obtained from 50 Gy and 100 Gy doses were quite different, mutagenic efficiency values of doses were similar in Sofu cultivar. As a result, the highest mutagenic efficiency can not be achieved from treatments, which give the highest mutation frequency. Senay (1997) also reported similar results

The mutagenic efficiency was high at high EMS doses. The highest values were obtained from high doses (0.3 % and 0.4 % EMS) that give the highest mutation frequency in Gediz-75. Similarly, the highest mutagenic efficiency was obtained from 0.3 % EMS doses in Sofu cultivar because the rate of mutation frequency was higher and sterility was lower in this dose. However some researchers achieved the highest mutagenic efficiency in lower doses (Peşkircioğlu 1995, Şenay 1997).

Table 6. Mutagenic efficiencies of mutagens

| Treatments | Cultivar | Doses | Sterility (M ₁) | Mutation frequency M ₁ spike | Mutagenic efficiency |
|------------|----------|--------|--------------------------------|---|-------------------------|
| Gamma | Gediz-75 | 50 Gy | 19.49 | 14.2 | 0.73 |
| Ray | | 100 Gy | 43.98 | | |
| 4.5 | Sofu | 50 Gy | 8.39 | 23.5 | 2.80 |
| | | 100 Gy | 35.01 | 107.7 | 3.08 |
| | Gediz-75 | % D.1 | 12.42 | 10.6 | 0.85 |
| | | % 0.2 | 10.65 | 5.7 | 0.54 |
| | | % D.3 | 20.84 | 53.4 | 2,56 |
| EMS | | % 0.4 | 18.90 | 62.3 | 3,30 |
| | Salu | % D.1 | 2.16 | | |
| | | % D.2 | 13.79 | 10.0 | 0.73 |
| | | %03 | 7.17 | 18.2 | 2.54 |
| | | % D.4 | 11.97 | 2.0 | 0.17 |

Different results were found for mutagenic efficiency with the treatments of gamma ray and EMS in durum wheat cultivars. The effects of mutagens varied with cultivars. Similar effects were reported by Konzak et al. (1965). Besides, Siddiqui et al. (1991) determined that the mutagenic efficiency of the two physical mutagens (gamma rays and fast neutrons) was nearly the same.

Conclusion

Comparing to EMS, the sterility in M₁ was higher with gamma ray treatment. The percentage of M₂ plants emergencing was unaffected by mutagen treatments. The spectrum and frequency of mutations were varied with treatments of mutagen and cultivars. The effect of mutagens markedly increased with high doses. The highest mutagenic efficiency was obtained from 0.4 % EMS dose in Gediz-75 cultivar and from 100 Gy gamma ray dose in Sofu cultivar.

References

- Anonymous, 1977. Manual on Mutation Breeding, Technical Report Series No. 119, IAEA, Vienna, p. 44-45.
- Caldecott, R.S., D.T. North, Kao Fa-ten, V.S. Hiatt and N.A. Tuleen, 1985. Forward mutations in avena and triticum polyploid series. Supplement to Radiation Botany, 5, 753-760.
- Çiftçi, C.Y., G. Akbay ve S. Ünver, 1988. Kunduru-1149 (Triticum durum L..) makarnalık buğday çeşidine uygularını farklı EMS (ethyl methane sulphonate) dozlarının Mı bitkilerinin bazı özellikleri üzerine etkileri-II, Ankara Üniv. Ziraat Fak. Yıllığı, 39 (1- 2) 349- 360.
- D'amato, F, 1965. Chimera formation in mutagen-treated seeds and diplontic selection. Supplement to Radiation Botany, 5, 303-316.
- Datta, A.K. and A.K. Biswas, 1985. Induced mutagenesis in Nigella sativa L.. Cytologia, 50, 545-562.
- Donini, B., M. Devreux and G.T. Scarascia-Mugnozza, 1974. Genetic effects of gametophyte irradiation in durum wheat. Proc. FAO/IAEA/Eucarpia Meeting, Bari, 1972, IAEA, Vienna, 127-138.
- Düzgüneş, O., T. Kesici, O. Kavuncu ve F. Gürbüz, 1987. Araştırma ve Deneme Metodları (İstalistiki Metodlar II), Ankara Üniv. Ziraat Fak, Yayınları: 1021, Ders Kitabı: 295, Ankara.
- Gaul, H. 1962. Ungewohnlich hohe mutations raten bei gefste nach anwendiung von aethylemethanesulphonat unda rontgenstrahlen. Naturwissensch, 49, 431.
- Gottschalk, W. and G. Wolff, 1983. Induced Mutations In Plant Breeding. Monographs On Theoretical and Applied Genetics 7, Berlin Heldelberg New York Tokyo, 10-14.
- Gustafsson, A. 1940. The mutation system of the chlorophyll aparatus. Lunds Univeritets Araskift, n.f. Avd.2, 36 (11) 1-40.
- Gustafsson, A. 1965. Characteristics and rates of high-productive mutants in diploid barley. Supplement to Radiation Botany, 5, 323-337.
- Holm, G. 1954. Chlorophyll mutations in barley. Acta Agric. Scand, 4, 457-471.

- Hussein, H.A.S. and I.A.M. Disouki, 1976. Mutation breeding experiments in *Phaseolus vulgaris* (L.): I. EMS and gammaray induced seed coat colour mutant. Z. Pflanzenzüchtg. Journal of Plant Breeding, 190-199.
- Kadagidze, M.G. 1979. Experimental mutagenesis in breeding the georgian durum wheat shaupka. Plant Breeding Abstracts, 45 (10) 731.
- Konzak, C.F., R.A. Nilan, J. Wagner and J. Foster, 1965. Efficient chemical mutagenesis. Radiat. Bot. 5 (suppl.), 49-70.
- Magri-Allegra, G. and L. Zannone, 1965. Effects of chemical and physical mutagens on forage vetch. II. Comparison of chromosome aberration produced by ethyl methane sulphonate, ethyleneimine and X-rays. In The Use of Induced Mutations in Plant Breeding, Radiat. Bot. 5 (suppl.), 215-226.
- Minocha, J.L., R.G. Saini and J.S. Sidhu, 1979. Mutations induced by ethylmethanesulfonate in three wheat cultivars. Plant Breeding Abstracts, 49 (1) 14.
- Natarajan, A.T. and M.D. Upadhya, 1964. Localized chromosome breakage by EMS and HA in Vicia faba. Chromosoma 15, 156-169.
- Nilan, R.A., C.F. Konzak, J. Wagner and R.R. Legault, 1965. Effectiveness and efficiency of radiations for inducing genetic and cytogenetic changes. Supplement to Radiation Botany, 5, 71-89.
- Paladi, N.I. and V.D. Siminel, 1979. A study of the second generation of irradiated wheat plants. Plant Breeding Abstracts, 49 (10) 730.
- Peşkircioğlu, H. 1995. Arpa (Hordeum vulgare L.)'ya Birlikte Uygulanan EMS (Ethyl Methane Sulphonate) ve Gama Işınlarının M₁ ve M₂ Bitkilerinin Bazı Özellikleri Üzerine Etkileri. A. Ü. Fen Bilimleri Enstitüsü, Doktora Tezi, 93.s.
- Raimkulov, K.R. and A. Maatkarimov, 1979. Variability of spring wheat after treatment with chemical mutagens. Plant Breeding Abstracts, 49 (9) 648.
- Sato, M. and H. Gaul, 1967. Effect of ethyl methane sulphonate on the fertility of barley. Radiat. Bot., 7, 7-15.
- Scarascia-Mugnozza, G.T., F. D'amato, S. Avanzi, D. Bagnara, M.L. Belli, A. Bozzini, A. Brunori, T. Cervigni, M. Devreux, B. Donini, B. Giorgi, G. Martini, L.M. Monti, E. Moschini, C. Mosconi, G. Porreca and L. Rossi, 1991. Mutation breeding programme for durum wheat (*Triticum turgidum ssp. durum* Desf.) improvement in Italy. IAEA, 1, 95-109.

- Siddiqui, K.A., G. Mustafa, M.A. Arain and K.A. Jafri, 1991. Realities and possibilities of improving cereal crops through mutation breeding. IAEA, 1, 173-185.
- Siminel, V.D. and N.I. Paladi, 1979. Variability of M₂ winter wheat varieties following the irradiation of grains with different dose of Rays. Plant Breeding Abstracts, 49 (10) 729.
- Swaminathan, M.S. 1965. A Comparison of mutation induction in diploids and polyploids. Supplement to Radiation Botany, 5, 619-641.
- Şenay, A. 1997. Makarnalık Buğday'da (*Triticum durum* Desf.) Gama Işını ve EMS'nin Farklı Dozlarının Ayrı Ayrı ve Birlikte Uygulamasının M₁ ve M₂ Bitkilerindeki Etkileri. A. Ü. Fen Bilimleri Enstitüsü, Doktora Tezi,103 s.
- Tavil, M.V. 1986. Mutability of varieties of bread and durum wheat. Tsitologiya-i. Genetika, 20 (2) 130-134.
- Tavcar, A. 1965. Gamma-ray irradiation of seeds of wheat barley and inbreds of maize and the formation of some useful point mutations. Supplement to Radiation Botany, 5, 159-174.
- Ünver, S. 1989. Arpada Uygulanan EMS (Ethyl Methane Sulphonate) Dozları, Yıkama Suyu ve Sıcaklık Süresinin M₁ ve M₂ Bitki Özelliklerine Etkileri. Ankara Üniv. Fen Bilimleri Enstitüsü, Doktora Tezi, 132 s., Ankara.
- Walter, R.F., L.F. Elinor and J.J. Holly, 1987. Mutation Breeding Principles of Cultivar Development, Theory and Technique. Macmillan Publishing Company A Dision of Macmillan, Inc, 287-303, New York.
- Wellensiek, S.J. 1965. Comparison of the effects of EMS, neutrons gamma and X- rays on peas. Supplement to Radiation Botany, 5, 227-235.
- Yanev, S.H. 1980. Use of experimental mutagenesis in breeding durum wheat, Rastenieu"dni-Nauki, 22 (12) 9-14.
- Yanev, S.H. 1985. Biological and genetic effect of some phsical and chemical mutagens on new durum wheat cultivars. Genetics and Breeding, 18 (5) 426-434, Sofia.
- Zannone, L. 1965. Effect of mutagenic agents in Vicia sativa L. comparison between effects of ethyl methane sulphonate, ethylene imine and X-rays on induction of chlorophyll mutations. Supplement to Radiation Botany, 5, 205-213.