Kantoğlu Y., Çakın I., Çetintaş A. O., Göktuğ A., Laouini, M. (2025). Determining the effective mutation dose and impacts of irradiation for some vegetable species Agriculture and Forestry, 71 (2): 7-20. [*https://doi:10.17707/AgricultForest.71.2.03*](https://doi:10.17707/AgricultForest.71.2.03)

DOI: 10.17707/AgricultForest.71.2.03

Yaprak K. KANTOĞLU [1]*; Irmak ÇAKIN[1]; Aydın Ozan ÇETINTAS[1]; Aslıhan GŐKTUĞ[1]; Mouna Souihi Ep LAOUINI[2]

DETERMINING THE EFFECTIVE MUTATION DOSE AND IMPACTS OF IRRADIATION FOR SOME VEGETABLE SPECIES

SUMMARY

A comparison of winter and summer vegetable species reveals that fewer mutation breeding studies have been conducted on winter varieties. Nevertheless, the effects of global warming and climate change are expected to negatively impact the cultivation of both winter and summer vegetable species alike. Addressing this issue promptly and developing new cultivars as alternatives to existing ones in accordance with current needs is crucial. Mutation breeding methods remain significant in efforts to broaden the diminishing genetic diversity. In this research, various vegetables, including red radish, black radish, white cabbage, broccoli, cress, dill, yellow onion, purple onion, spinach, parsley, and arugula, were exposed to a cesium 137 gamma ray source. The irradiation was conducted at different dose levels (0, 50, 100, 200, 300, 400, 400, 500, 600, 700, 800, 900, 1000, 1200, 1400, 1600, and 1800 Gy), based on the specific responses of each cultivar. Following irradiation, the seed germination rate, seedling height, seedling fresh and dry weight, and seed vigor were measured, and the effective mutation dose (EMD_{50}) was determined by linear regression analysis. It was found that increasing doses had a negative effect on germination, seed strength, and fresh and dry plant weight, depending on the species. Accordingly, EMD_{50}, which is important in creating an effective mutant population, was determined as 467.62 Gy for red radish, 328.39 Gy for black radish, 354.24 Gy for broccoli, 222.25 Gy for white cabbage, 119.75 Gy for arugula, 160.68 Gy for spinach, 245.96 Gy for yellow onion, 195.55 Gy for purple onion, 132.38 Gy for dill, 1031.68 Gy for cress, and 92.77 Gy for parsley.

Keywords: Effective mutation dose, mutation breeding, winter vegetables, onion, radish, cabbage, parsley, dill, cress, broccoli

---

[1]K. Yaprak Kantoğlu (corresponding author: kadriyeyaprak.kantoglu@tenmak.gov.tr); Irmak Çakın; Aydın Ozan Çetintaş; Aslıhan Göktuğ; Türkiye Energy, Nuclear and Mineral Research Agency (TENMAK)-Nuclear Energy Research Institute TÜRKİYE

[2]Mouna Souihi Ep Laouini; National Center of Sciences and Nuclear Technology (CNSTN), Sidi Thabet-TUNISIA

Notes: The authors declare that they have no conflicts of interest. Authorship Form signed online.

Recieved:26/12/2024 *Accepted:12/05/2025*

INTRODUCTION

Throughout history, humans have influenced plant evolution through consumption habits and agricultural practices. This process began in the hunter-gatherer era and continued through the agricultural revolution as people selected and cultivated specific plant products for food. With Gregor Mendel and the process that followed him, breeding studies specific to each species have also developed and have come to the present day (Abak *et al.,* 2023). Classical breeding and biotechnological methods are effectively used in plant breeding studies in line with agricultural needs. Within the modern breeding approach, mutation breeding methods still maintain their importance to expand the shrinking gene pool and create alternative variations. Studies on mutations induced by physical mutagen applications have been actively conducted for 100 years (Ma *et al,.* 2021). The alternative variations generated through mutation in classical breeding methods have facilitated gene editing studies in biotechnological approaches to create alternative genetic resources. A chronological assessment of physical mutagen sources utilized in mutation breeding from past to present reveals that gamma rays succeeded the initial X-ray applications, followed by the adaptation of high- and low-energy accelerator technologies to mutation breeding studies (Krasaechai and Yu, 2009; Abe *et al.,* 2015). In contemporary times, the applications of cosmic radiation have expanded, and their popularity is experiencing rapid growth. Numerous countries, with China at the forefront, have developed alternative approaches by inducing mutations in plant material under space conditions within space stations (Ma *et al.,* 2021; Mohanta *et al.,* 2021). The research conducted thus far has enabled China to identify the characteristics of mutant variants encompassing over two hundred perennial and annual species, subsequently advancing them to the varietal stage (Bahanu, 2022). Consequently, mutation breeding maintains its significance in breeding studies due to the advantages it offers in conjunction with advancing technology. An examination of the International Atomic Energy Agency Mutant Database (IAEA-MVD) reveals that the number of mutant varieties developed for winter vegetable species is substantially lower compared to other summer vegetable species. While there are no registered mutant varieties in dill, arugula, broccoli, and parsley, six registered mutant varieties exist in onion and one each in cress, spinach, and radish (Micke *et al.,* 1990; MVD, 2024). In cabbage, kale-type mutant varieties predominate; however, it is acknowledged that mutant cabbage varieties exist that have not been registered in the IAEA-MVD but have been reported as developed (Wang *et al.,* 2020). Currently, mutation breeding represents a crucial tool in breeding studies for improving traits such as stunting, abiotic and biotic stress tolerance, which are characterized as the green revolution in plant breeding (Manape *et al.,* 2023). This method utilizes both physical and chemical mutagen sources and successfully overcomes genetic barriers that present challenges, particularly in breeding studies (Van Harten, 1998; Spencer-Lopez *et al.,* 2018). This approach facilitates the expansion of the existing gene pool, enabling the introduction of genotypes with diverse agricultural characteristics into agricultural production and breeding studies as genitors (Muangprom *et al.,* 2005). The COVID-19 pandemic has heightened the significance of microgreens in human nutrition and health due to their bioactive molecular content. Notable examples within this plant group include radish, cabbage, onion, parsley, broccoli, dill, amaranth, beetroot, chicory, chervil, fennel, carrot, celery, and spinach (Bhaswant *et al.,* 2023). Research on microgreens is expanding rapidly, with a focus on enhancing yield and nutrient content through improved production techniques (Samuoliené *et al.,* 2019) and breeding strategies (Lone *et al.,* 2024). Consequently, the development of microgreens, which are regarded as valuable nutritional sources, is of significant importance (Galieni *et al.,* 2020).

One of the most significant challenges arising from climate change is the increase in temperature and drought. Currently, research is being conducted to activate recessive genes that control tolerance to adverse factors resulting from global warming in numerous species, with a particular focus on onion (Manape *et al.,* 2023). Given that recessive genes can be rendered dominant through mutation breeding, a broad spectrum of variation is obtained both in vivo and in vitro by incorporating mutagens into breeding studies for such research. By screening this variation with relevant stress agents, it becomes feasible to select genotypes exhibiting the desired traits (Chakraborty *et al.,* 2015; Kohzuma *et al.,* 2017; Spencer-Lopez *et al.,* 2018; Wang *et al.,* 2020; Kantoğlu *et al.,* 2021; Singh *et al.,* 2021).

The primary objective of this investigation was to develop novel cultivars that demonstrate enhanced adaptation to changing climatic conditions and exhibit superior yield and quality characteristics compared to existing varieties of *Allium cepa* L. (yellow and purple onion), *Brassica oleracea* L. var. capitata (white cabbage), *Brassica oleracea* var. L. italica (broccoli), *Raphanus sativus* L. var. radicula (red radish), *Raphanus sativus* L. var. niger (black radish), *Lepidium sativum* L. (cress), *Anethum graveolens* L. (dill), *Spinacia oleracea* L. (spinach), *Petroselinum hortanse* Hoffm. (parsley), and *Eruca vesicaria* Mill. (arugula) species.

These species are significant in terms of human health and economic value, both as microgreens and for conventional consumption. This study employed mutation-breeding techniques to achieve this goal. To this end, this research aimed to determine the effective mutation dose (EMD_{50}) specific to each species and variety, which constitutes the initial phase of a well-designed mutation breeding study. Furthermore, the investigation seeks to establish EMD_{50} doses with 10% lower and upper limits through irradiation to generate a broad mutant variation for each species under examination.

MATERIAL AND METHODS

Material

Untreated (with no pest), standard red radish, black radish, white cabbage, broccoli, cress, dill, yellow onion, purple onion, spinach, parsley, and arugula seeds procured from Turkish Seed Company named Patika were utilized in the study. The seeds are from locally grown varieties.

Methods

To determine the EMD_{50} dose, the moisture content of seeds obtained from red radish, black radish, white cabbage, broccoli, cress, dill, yellow onion, purple onion, spinach, parsley, and arugula was ascertained (F.1). Subsequently, seeds were prepared for irradiation in three replicates with 30 seeds per replicate for each irradiation dose. The experimental cesium 137 (^{137}Cs) radiation source (dose rate: 821 Gy/h) at TENMAK-NÜKEN was utilized for irradiation. Seeds were irradiated to determine the EMD_{50} dose at intervals of 0, 50, 100, 200, 200, 300, 400, 500, 600, 700, 800, 800, 900, 1000, 1200, 1400, 1600, and 1800 Gy, contingent upon the irradiation dose response of the species. Following irradiation, seeds were sown in 96-well vials containing a 1:1:1 mixture (peat: burnt stable manure: garden soil) with one seed per well. The experiment was set up in a randomized plot design with three replications and 30 seeds in each replication. Seed germination and shoot development were monitored in a 250 m² cell of the greenhouse equipped with air conditioning automation under a 16/8 h photoperiod and 24±2℃ temperature. On the 35th day post-sowing, shoot length measurements were recorded (Figure 1), and wet and dry plant weight measurements and seed vigor (F.2) calculations were performed. For dry plant weight determination, plants whose fresh weight had been measured were placed in an oven at 70℃ for 24-36 hours, and dry plant weight was measured upon complete desiccation.

$$\%\ Seed\ moisture = \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} x100 \qquad \text{F.1.}$$

$$Seed\ vigor\ index = (\%) Germination\ rate\ x\ Seedling\ height\ (included\ root) \qquad \text{F.2.}$$

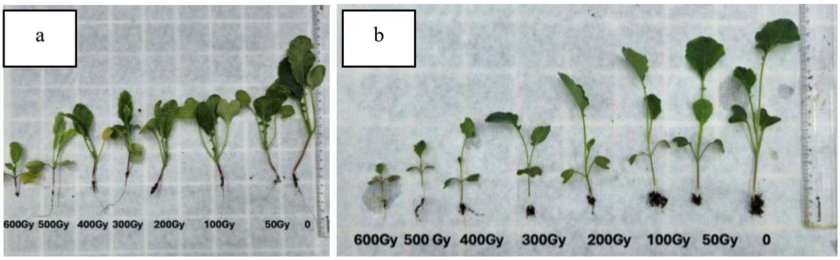

Figure 1. Shoot length measurement to determine EMD_{50} dose (broccoli (a) and red radish (b))

Calculating the EMD_{50} dose, linear regression analysis was conducted utilizing average shoot length data for all species except cress. For the cress species, linear regression analysis was performed using average fresh weight data. Linear regression analysis was conducted using the Excel package program. For other variance analysis, ANOVA under the Minitab software program and the

Duncan test were performed using the MSTATC software program to determine the statistical difference between the applications.

RESULTS AND DISCUSSION

This study aimed to determine the EMD_{50} dose of commercial seeds of red radish, black radish, white cabbage, broccoli, cress, dill, yellow onion, purple onion, spinach, parsley, and arugula following irradiation with varying doses of ^{137}Cs gamma source. Additionally, the research sought to observe the effects of irradiation on seed germination and subsequent plant growth.

Prior to the irradiation process, it is essential to determine the moisture content percentage of the seed samples to be irradiated and adjust it to 7-12% if necessary. This adjustment is crucial because an increase in seed moisture content tends to elevate the production of toxic free radical substances resulting from radiation application (Marcu *et al.,* 2013). Such an increase is undesirable for accurate and effective irradiation (Van Harten, 1998). In this study, the pre-irradiation determination of seed moisture revealed a content ranging from 8.8% to 12%, depending on the species. Given that these moisture percentages fell within the accepted limits, irradiation was conducted without any additional moisture content compensation application to the seeds.

Seven days post-sowing, following irradiation, the effects of irradiation on seed germination were evaluated across all species (Table 1). The results of the germination counts indicated that irradiation had no detrimental effect on germination in cress, red radish, and broccoli, with 100% germination observed in all three species at each treatment dose.

In all species except yellow onion, the decrease in germination percentage at increasing doses was found to be statistically significant. In yellow onion, the germination rate was 96.67% higher in the 700 Gy application compared to the control.

Conversely, no germination was observed in dill after the 300 Gy application; the germination rate decreased to 0% after the 100 Gy application in parsley species, the germination rate decreased significantly after 700 Gy in arugula, and no seed germination occurred at 1000 Gy. In spinach, cabbage, and purple onion, the germination rates at 600 Gy were 73.33%, 65.00%, and 63.33%, respectively.

It is established that the dose range utilized in irradiation is higher in small-seeded species with acceptable seed moisture content (Xu *et al.,* 2019). Upon evaluation of the data obtained in this study, it was observed that there was resistance to the anticipated decrease in seed germination rates at increasing doses in all species except parsley and dill, potentially due to the negative effects of the increasing irradiation dose, contingent upon genotype response.

This can be attributed to the fact that in species with larger seeds, as a consequence of higher endosperm volume and subsequently more structural substances (water, ash, protein, carbohydrate, fat), the rate of intercellular bond breakage increases as a result of radiation application, and variations occur in the amount of dose absorbed by irradiation depending on the content of these substances. However, as a consequence of bond breakage, the formation of free radicals is likely to increase, thereby elevating the probability of adverse effects on germination and seedling development at higher radiation doses (Wang *et al.,* 2022).

Table 1. Germination values (%) of control and irradiated seeds 7 days after sowing

Dose (Gy)	**Species**							
	Spinach	**Arugula**	**Parsley**	**Dill**	**Black radish**	**Cabbage**	**Purple onion**	**Yellow onion**
0	86.67 a	96.67 a	53.33 a	100.00 a	50.00 e	83.33 b	96.67 a	90.00 d
50	86.67 a	83.33 c	53.33 a	83.33 b	76.66 a	70.00 d	93.33 b	100.00 a
100	86.67 a	90.00 b	10.00 b	83.33 b	53.33 d	70.00 d	83.33 d	93.33 c
200	50.00 c	83.33 c	0.00 c	10.00 c	63.33 b	90.00 a	90.00 c	100.00 a
300	73.33 b	66.67 d	0.00 c	10.00 c	56.67 c	76.67 c	90.00 c	100.00 a
400	73.33 b	83.33 c	0.00 c	0.00 d	43.33 f	70.00 d	90.00 c	100.00 a
500	73.33 b	66.67 d	0.00 c	0.00 d	50.00 e	66.67 e	83.33 d	96.67 b
600	73.33 b	83.33 c	0.00 c	0.00 d	45.00 f	65.00 e	63.33 e	
700	-	40.00 f	0.00 c	0.00 d	-	-	-	-
800	-	50.00 e	-	-	-	-	-	-
900	-	30.00 g	-	-	-	-	-	-
1000	-	0.00 h	-	-	-	-	-	-
Sx **LSD**	0.01291 0.04901	0.05 0.1872	0.04082 0.1543	0.08165 0.3085	0.6589 2.499	0.7106 2.698	0.3291 1.250	0.4882 1.865

Letters: a, b, c, d, e, f identify the difference between % germination values according to DUNCAN test ($p<0.01$)

As the quantity of endosperm content diminishes in smaller seeds, it is observed that the germination rate is less adversely affected by the absorbed dose and the potential formation of free radicals (Xu *et al.,* 2019). Nevertheless, the impact of radiation on plant development is more significant than its effect on germination. Given that the developmental delays occurring in plants as a consequence of radiation exposure over time are of greater importance, it is crucial to determine the effective mutation dose that influences plant development in relation to the rate of development. In this study, following irradiation in all species except cress, it was observed that the shoot development of the plants decreased compared to the control, corresponding to the effects of increasing irradiation doses at 30-35 days after seed sowing (Table 2). For cress, additional irradiation was conducted up to a dose of 1800 Gy, and plants were subsequently observed. In spinach, the shoot length value was 0.64 cm with a 500 Gy application, and according to the result of linear regression analysis, it was determined that the 50% shoot length value of the control plants was achieved at an irradiation dose of 160.67 Gy according to y = -0.0092x + 3.4782, and the EMD_{50} dose was calculated as 160.57 Gy.

In arugula, the average shoot length was determined to be 1.78 cm at a 700 Gy application dose, 1.29 cm at 900 Gy, and 0.00 cm at 1000 Gy. The EMD_{50} dose was calculated as 518.14 Gy (y = -0.0125x + 10.263). For *Petroselinum hortanse* Hoffm. (parsley), the shoot length decreased significantly to 0.57 cm at 100 Gy, with no shoot development observed at 400 Gy. The EMD_{50} dose was determined to be 92.77 Gy (y = -0.0144x + 3.4609). Similarly, in *Anethum graveolens* L. (dill), shoot development was not observed at doses exceeding 300 Gy, with an EMD_{50} dose of 129.17 Gy (y = -0.016x + 5.2167). In *Raphanus sativus* L. var. radicula (red radish), shoot development occurred at 600 Gy but was not observed at 700 Gy, with an EMD_{50} dose of 467.62 Gy (y = -0.0101x + 10.663). For *Raphanus sativus* L. var. niger (black radish), the EMD50 dose was determined to be 328.40 Gy (y = -0.0053x + 3.7505). Comparable to red radish, shoot development in black radish was observed up to 600 Gy; however, the plants exhibited slower development relative to red radish. In broccoli, the average shoot length was observed to be 0.00 cm in both the 200 Gy and 700 Gy treatments, while the EMD_{50} dose was determined to be 354.24 Gy (y = -0.0125x + 10.263). No significant difference was observed between the shoot lengths of plants derived from irradiated seeds and control plants in cress at irradiation levels up to 700 Gy.

Table 2. Dose effect and linear regression analysis results on shoot growth in species irradiated at different gamma ray doses

Dose (Gy)	**Species**									
	Spinach	**Arugula**	**Parsley**	**Dill**	**Red radish**	**Black radish**	**Broccoli**	**Cabbage**	**Purple onion**	**Yellow onion**
0	4	6.95	4.33	6.3	11.88	4.02	11.67	7.13	7.55	7.92
50	3.62	5.12	3.80	4.7	9.68	2.60	10.32	5.48	5.68	5.48
100	1.93	5.87	0.57	3.14	8.95	3.92	9.03	3.69	3.38	4.00
200	0.65	3.98	0.00	0.20	8.42	2.42	5.97	2.51	2.48	3.73
300	0.42	4.13	0.00	0,20	7.07	2.65	5.17	1.85	2.23	3.37
400	0.62	3.12	0.00	0.00	7.20	1.38	4.40	2.45	1.45	2.00
500	0.66	2.43	0.00	0.00	5.95	0.92	4.72	1.92	0.98	1.53
600	0.64	4.12	0.00	0.00	4.48	0.67	3.98	2.77	0.77	-
700	-	1.78	0.00	0.00	-	-	-	-	-	-
800	-	2.92	-	-	-	-	-	-	-	-
900	-	1.29	-	-	-	-	-	-	-	-
1000	-	0.00	-	-	-	-	-	-	-	-
EMD_{50} (Gy)	160.67	518.14	92.77	129.17	467.62	328.40	354.24	222.35	195.60	245.96

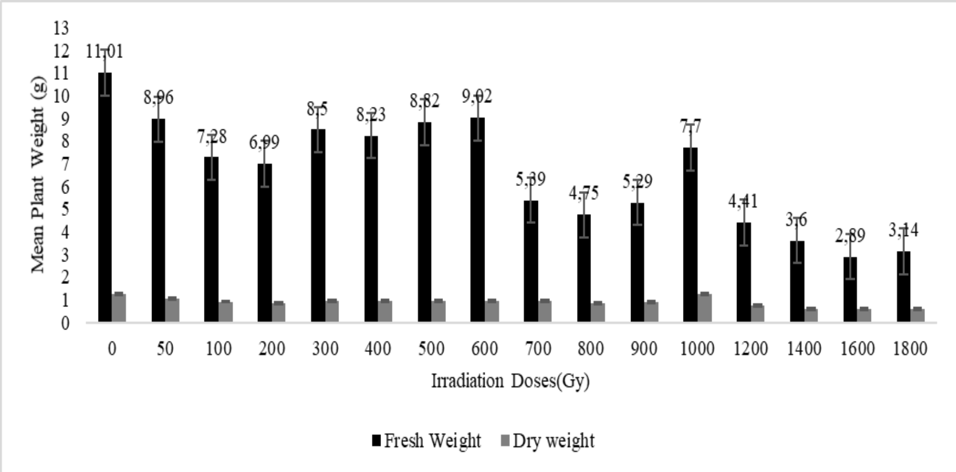

Figure 2. Effects of different radiation doses on fresh and dry plant weight of cress samples

Consequently, an additional irradiation study was conducted up to 1800 Gy, and the effects of 16 distinct radiation doses were monitored to ascertain the EMD_{50} dose. Although no statistically significant difference (at $p<0.01$) was noted in terms of shoot length, a reduction in plant growth was observed with increasing doses, as illustrated in Figure 2. The EMD_{50} dose was calculated to be 1031.68 Gy ($y = -0.0037x + 9.3222$) through linear regression analysis using fresh weight values.

In the observations conducted on cabbage, plant growth was observed at doses up to 500 Gy; however, no shoot development occurred in 600 and 700 Gy applications, and the EMD_{50} dose was determined to be 222.35 Gy ($y = -0.0091x + 5.5884$). For purple onion, shoot development decreased significantly from 100 Gy, with a substantial plant regression observed from 500 Gy, and the EMD_{50} dose was calculated as 195.60 Gy ($y = -0.0098x + 5.6914$). In yellow onion, development decelerated from 400 Gy application; nevertheless, plant development was marginally superior at increasing doses compared to the yellow onion variety, and shoot development was not achieved in 600-700 Gy applications. The EMD_{50} dose for yellow onion was determined to be 245.96 Gy ($y = -0.0085x + 6.0507$). When comparing the findings obtained with the results of other studies conducted thus far, in the mutation breeding study performed on garlic, increasing doses exhibited a negative effect on shoot development, and 4.55 Gy gamma ray dose was established as EMD_{50} for the T6 garlic clone, resulting in a substantial variability following irradiation with this dose (Taner *et al.,* 2004; Beşirli *et al.,* 2022). Comparable results were reported for pepper (Tepe *et al.,* 2004; Kantoglu *et al.,* 2014), lettuce (Sarıçam *et al.,* 2017), watermelon (Velkov *et al.,* 2016; Kantoğlu, 2022), purple carrot (Yarar *et al,.* 2021), cucumber and okra (Jaipo *et al.,* 2019), zucchini (Kurtar *et al.,* 2017), and Chinese carnation (Çakın *et al*., 2024). Furthermore, it has been reported that

EMD_{50} doses of different cultivars vary within the same species (Sarıçam *et al.,* 2017; Sarıçam Kökpınar *et al.,* 2024). Consequently, this study reaffirms the necessity of determining the EMD_{50} dose according to genotype.

Upon examination of mutation breeding studies conducted on winter vegetable species by determining the EMD_{50} dose, Sing *et al.,* (2011) report the development of mutant lines exhibiting high yield and quality, extended shelf life, suitability for mechanical harvesting, and increased seed yield from the mutant population generated with the EMD_{50} dose in studies utilizing chemical and physical mutagens for onion. In a subsequent study focused on onion, molecular investigations conducted by Manape *et al.,* (2023) revealed the acquisition of purple leaf spot-resistant lines at an EMD_{50} dose of 50 mM ethyl methane sulfonate (EMS) application (Chakraborty *et al.,* 2015).

Furthermore, it was observed that bright leaf formation was facilitated by a recessive gene that activates the resistance mechanism against biotic and abiotic stress factors. Oxalate and nitrate accumulation and abiotic stress tolerance in spinach are two significant issues that researchers have investigated. In a study, it was determined that an EMD_{50} dose of 50 mM for 6 hours resulted in low oxalate accumulation in a mutant line (Murakami *et al.,* 2009). In another mutation breeding study on Chinese cabbage, a 16-hour application of 0.04% EMS was identified as the EMD_{50} dose, and it was observed that the rate of yellow leaf formation was higher in the mutant population created with this dose, which influenced glucosinate activity (Sun *et al.,* 2022). In a subsequent mutation breeding study conducted by Wang *et al.,* (2020) on cabbage, it was determined that the recessive gene (Bra019346), which maintains leaf color green for an extended period, became dominant in a mutant line.

When evaluating the data from studies conducted on different species and varieties, it is evident that EMD_{50} doses range from 160-280 Gy for onion, 800-1600 Gy for turnip, and 300-500 Gy for spinach (Spencer-Lopez *et al.,* 2018). Similarly, although the data obtained for spinach and onion in this study are nearly consistent with these results, variations due to genotype response are an anticipated outcome (Ribera *et al.,* 2020).

Another significant effect of irradiation is on seed vigor. It is essential to determine seed vigor by evaluating the combined effect of doses on germination and plant growth rate. When the data obtained were evaluated in this context, it was determined that plant growth capacity was negatively affected on seed vigor index, particularly due to the effect of increasing doses (Table 3).

Although the seed vigor appeared to be higher than the other species because 100% germination was achieved at each dose in only cress species, the expected decrease in seed vigor due to the effect of increasing dose, as a result of the linear decrease in plant weight due to increasing doses, was clearly observed with 900 Gy application. This finding aligns with the results of the research conducted by Urva *et al.,* (2017).

Table 3. Effect of different radiation doses on seed vigore index

Dose (Gy)	**Species**										
	Spinach	**Arugula**	**Parsley**	**Dill**	**Cress**	**Red radish**	**Black radish**	**Broccoli**	**Cabbage**	**Purple onion**	**Yellow onion**
0	346.68 ±0.57	671.86 ±0.82	230.92 ±0.58	1058.00 ±0.58	1101.00 ±1.29	1188.00 ±5.80	201.00 ±0.82	1167.00 ±5.80	594.14 ±0.82	729.86 ±5.77	712.80 ±0.80
50	313.75 ±0.58	426.65 ±0.01	202.65 ±0.58	1100.00 ±0.57	896.00 ±0.58	968.00 ±5.80	199.32 ±0.58	1032.00 ±0.57	383.60 ±0.82	530.11 ±0.58	548.00 ±0.80
100	167.27 ±0.57	528.30 ±0.58	5.70 ±0.63	1065.00 ±0.58	728.00 ±0.82	895.00 ±0.82	209.05 ±0.58	903.00 ±0.58	258.30 ±0.82	281.66 ±0.58	373.32 ±0.80
200	32.50 ±0.58	331.65 ±0.82	0.00 ±0.00	1100.00 ±0.58	699.00 ±0.58	842.00 ±5.77	153.2 ±0.58	597.00 ±0.58	225.90 ±0.58	223.20 ±0.58	373.00 ±0.80
300	30.80 ±0.57	275.35 ±0.58	0.00 ±0.00	1000.00 ±0.58	850.00 ±5.80	707.00 ±5.80	150.18 ±0.58	517.00 ±0.57	141.84 ±0.82	200.70 ±0.58	337.00 ±0.80
400	45.46 ±0.57	259.99 ±0.58	0.00 ±0.00	985.00 ±0.58	823.00 ±5.80	720.00 ±5.80	59.80 ±0.58	440.00 ±0.57	171.50 ±0.82	130.50 ±0.58	200.00 ±05.8
500	48.40 ±0.58	162.01 ±0.58	0.00 ±0.00	1056.00 ±0.57	882.00 ±0.58	595.00 ±5.63	46.00 ±0.57	472.00 ±0.58	128.01 ±0.82	81.66 ±0.57	147.91 ±0.57
600	46.93± 0.57	343.32 ±0.58	0.00 ±0.00	756.00 ±0.57	992.00 ±5.80	448.00 ±1.29	30.15 ±0.57	398.00 ±0.57	180.51 ±0.82	48.76 ±0.57	-
700	-	71.20 ±0.82	0.00 ±0.00	865.00 ±0.57	539.00 ±0.58	-	-	-	-	-	-
800	-	146.00 ±1.29	-		475.00 ±5.81	-	-	-	-	-	-
900	-	38.70 ±0.82	-		529.00 ±1.29	-	-	-	-	-	-
1000	-	0.00 ±0.00	-		77.00 ±5.80	-	-	-	-	-	-
1200	-	-	-		44.10 ±5.83	-	-	-	-	-	-
1400	-	-	-		36.00 ±0.82	-	-	-	-	-	-
1600	-	-	-		28.9 ±5.80	-	-	-	-	-	-
1800	-	-	-		31.4 ±0.08	-	-	-	-	-	-

The positive effects of low-dose ionizing radiation on seed germination rate, young embryo stimulation, plant development, and the quantity of internal matter are well established in the scientific literature (Osborne and Bacon, 1960; Sjödin, 1962; Marcu *et al*., 2013; Urva *et al*., 2017; Jaipo *et al.,* 2019). While numerous researchers have reported that low doses of ionizing radiation stimulate ribonucleic acid (RNA) and protein activation in seeds and enhance the priming effect, ongoing investigations aim to determine the efficacy of various ionizing radiation sources (Abdel-Haly *et al.,* 2008; Wang *et al.,* 2022). Upon evaluation of the research findings, it was observed that the germination rates of yellow onion and black radish increased with low-dose application compared to the control, reaching 76.66% and 100%, respectively. This observation aligns with the results of previous studies in the field.

CONCLUSIONS

Mutation breeding is a method that has been utilized to obtain different genotypes within existing variability. Currently, this method maintains its significance in research on the development of new varieties with drought tolerance, which has become increasingly important due to the climate crisis. The Atoms4Food initiative of the IAEA is likely to further enhance the prominence of this technique in the future. Mutation breeding is crucial for creating genetic resources for both classical breeding studies and research conducted using biotechnological methods, particularly for winter vegetable species and microgreens, which are important for human nutrition and health. Determining the EMD_{50} dose according to the genotype under study is one of the most critical aspects in establishing the main mutant population, which serves as the starting source for a successful mutation breeding study. This research aimed to emphasize this point and determine the red effective mutant population used in the study. Additionally, it sought to establish species-specific doses for radish, black radish, white cabbage, broccoli, cress, dill, yellow onion, purple onion, spinach, parsley, and arugula varieties, as well as to elucidate the differences in dose absorption and dose effect between species. The study results indicated that cress exhibited the highest survival and germination capacity under ionizing radiation treatment. For this species, a preliminary study established an application dose of 1800 Gy, and the EMD_{50} dose could be determined through irradiation across a broader dose range. Conversely, yellow onion demonstrated no positive viability outcomes in applications exceeding 500 Gy. This observation underscores the critical nature of determining and applying the EMD_{50} dose to generate a breeding population with a high mutation frequency. In contemporary practice, biotechnological approaches have gained prominence alongside classical breeding methods. While mutation remains a breeding tool, researchers also consider it a significant resource, particularly for gene editing studies. Consequently, the development of effective mutant populations continues to be of importance for future research, and its contribution to the enrichment of the gene pool remains valuable. In addition to these, it is imperative to utilize irradiation technology for agricultural production in conjunction with breeding studies. Seed irradiation applications employing the low dose effect to enhance germination rates and quality characteristics (malt, antioxidant content, etc.) that are crucial for the product are implemented on a limited scale. Acknowledging this positive effect of irradiation, it would be advantageous to expand research efforts to incorporate radiation technology into the production process, thereby maximizing its potential benefits.

ACKNOWLEDGEMENTS

The research was supported by the Turkish Energy, Nuclear and Mineral Research Agency under project A2.H1.P14.

REFERENCES

Abak, K., Balkaya, A., Ellialtıoğlu, Ş. Ş. & Düzyaman. E. (2023). Sebze Islahı Cilt VI: Leguminosae (Baklagiller) Compositae (Bileşik Çiçekliler). Gece Kitaplığı, Ankara. p 5-6. (in Turkish)

Abe, T., Kazama, Y., & Hirano, T. (2015). Ion beam breeding and gene discovery for function analyses using mutants. Nucl Phys News. (2015) 25:30–4. doi: 10.1080/10619127.2015.1104130

Abdel-Hady, M. S., Okasha, E., M., Soliman, S. S. A. & Talaat, M. (2008). Effect of gamma radiation and gibberellic acid on germination and alkaloid production in *Atropa belladonna* L. Australian Journal of Basic and Applied Sciences 2(3): 401-405.

Bahanu, A., N. (2022). Space breeding: Space inspired mutation breeding for crop imorovement. Grain 92-102.

Beşirli G., Yarali Karakan F., Sönmez I, Çetin B. E., Erol Ü. H., Kantoğlu Y. T., & Kunter B. (2022). Characterization of mutant garlic genotypes based on volatile sulfur compounds and mineral content. J. Elem., 27(3): 489-506. DOI: 10.5601/jelem.2022.27.2.2276

Bhaswant, M., Shanmugam, D. K., Miyazawa, T., Abe, C. & Miyazawa T. (2023). Microgreens-A comprehensive review of bioactive molecules and health benefits. Molecules 15, 28(2):867. https://doi.org/ 10.3390/molecules28020867

Çakın, I., Kantoğlu, K. Y., Kunter, B. & Göktuğ, A. (2024). Çin karanfili (*Dianthus chinensis* L.) için mutasyon ıslahı çalışmasına yönelik etkili mutasyon dozun belirlenmesi. Düzce Üniversitesi Orman Fakültesi Ormancılık Dergisi 20(Special issue), 44-54. https://doi.org/10.58816/duzceod.1537332 (in Turkish).

Chakraborty, M., Quddus, T., Rahman, S., Azad, M. A. K., Hossain, M. R., Hoque, A., Saha, M. K. & Haque, M. A. (2015). Molecular characterization of selected mutant lines of onion (*Allium cepa* L.) against purple leaf blotch disease using SSR markers. American Journal of Experimental Agriculture 8(4): 261-267.

Galieni, A., Falcinelli, B., Stagnari, F., Datti, A. & Benincasa, P. (2020). Sprouts and microgreens: Trends, opportunities, and horizons for novel research. Agronomy 10: 1424.

Jaipo, N., Kosiwikul, M., Panpuang, N. & Prakrajang, K. (2019). Low dose gamma radiation effects on seed germination and seedling growth of cucumber and okra. Journal of Physics: Conference Series 1380, 012106 doi:10.1088/1742-6596/1380/1/012106

Kantoğlu, K. Y., Tepe, A., Kunter, B., Fırat, A. F. & Peşkircioğlu, H. (2014). Vegetable Crops Breeding by Induced Mutation and A Practical Case Study of *Capsicum annuum* L. In: N.B. Tomlekova, M.I. Kozgar and M.R. Wani, (eds) Mutagenesis: exploring genetic diversity in crops. Wageningen Publisher, Holland. 41-56.

Kantoğlu, K. Y., Kuşvuran, Ş., & Ellialtıoğlu, Ş. Ş. (2021) Doku kültürü yöntemlerinin stres çalışmalarında kullanımı. In: Sebzelerde Stres Toleransı ve Islah Stratejileri, Ellialtıoğlu, Ş. Ş., Daşkan, H. Y., & Kuşvuran, Ş. (Eds). Gece Kitaplığı, Ankara, pp 708–741. ISBN: 978-625-8449-01-3 (in Turkish)

Kantoğlu, K. Y. (2022). Effects of Gamma Irradiation on seed germination in watermelon (*Citrullus lanatus* (Thunb.) Matsum & Nakai) and determination of effective mutation dose. Alatarım 21(1):10-17. ISSN: 1304-2653

Krasaechai, A., & Yu, L. D. (2009). Low- energy ion beam modification of horticultural plants for induction of mutation. J. Surface and Coatings Technol.203: 2525-2530.

Kohzuma, K., Chiba, M., Nagano, S., Anai, T., Ueda, M. U., Oguchi, R., Shirai, K., Hanada, K., Hikosaka, K. & Fujii, N. (2017). Mutant selection in the self-incompatible plant radish (*Raphanus sativus* L. *var. sativus*) using two-step TILLING. Breed Sci 67(3):268-276. doi: 10.1270/jsbbs.16200

Kurtar, E. S., Balkaya, A. & Kandemir, D. (2017). Determination of semi-lethal (LD50) doses for mutation breeding of Turkish winter squash (*Cucurbita maxima* Duch.) and pumpkin (*Cucurbita moschata* Duch.). Fresenius Environ Bull 26:3209-3216.

Lone, J. K., Pandey, R. & Gayacharan. (2024). Microgreens on the rise: Expanding our horizons from farm to fork. Heliyon 10(4): 25870. https://doi.org/10.1016/j.heliyon.2024.e25870

Ma, L., Kong, F., Sun, K., Wang, T. & Guo, T. (2021). From classical radiation to modern radiation: Past, present, and future of radiation mutation breeding. Frontiers in Public Health 9: 768071. https://doi.org/10.3389/fpubh.2021.768071

Manape, T. K., Soumia, P. S., Khade, Y. P., Satheesh, V., & Anandhan, S. (2023). A glossy mutant in onion (*Allium cepa* L.) shows decreased expression of wax biosynthesis genes. Frontiers in Plant Science 14, 1245308. https://doi.org/10.3389/fpls.2023.1245308

Marcu, D., Damian, G., Cosma, C. & Cristea, V. (2013). Gamma radiation effects on seed germination, growth and pigment content, and ESR study of induced free radicals in maize (*Zea mays*). J Biol Phys 39(4):625-34. https://doi.org/10.1007/s10867-013-9322-z

Micke, A., Donini, B. & Maluszynski, M. (1990). Induced mutations for crop improvement. Mutation Breeding Review 7, 1-41.

Mohanta, T. K., Mishra, A. K., Mohanta, Y. K. & Al-Harrasi, A. (2021). Space Breeding: The next-generation crops. Front Plant Sci Sec Plant Breeding 12: 771985. https://doi.org/10.3389/fpls.2021.771985

Muangprom, A., Thomas, S. G., Sun, T. P. & Osborn, T. C. (2005). A novel dwarfing mutation in a green revolution gene from *Brassica rapa*. Plant Physiol 137(3):931-8. doi: 10.1104/pp.104.057646

Murakami, K., Edamoto, M., Hata, N., Itami, Y. & Masuda, M. 2009. Low-oxalate spinach mutant induced by chemical mutagenesis. J Japan Soc Hort Sci 78 (2): 180–184.

MVD. (2024). https://nucleus.iaea.org/sites/mvd/SitePages/Search.aspx (acsess date: 30 October 2024)

Osborne, T. S. & Bacon, C. A. (1960). Radiosensitivity of seeds: I. Reduction or stimulation of seedling growth as a function of gamma-ray dose. Radiation Research 13(5): 686-690. https://doi.org/10.2307/3571031

Ribera, A., Bai, Y., Wolters, A. A., van Treuren, R. & Kik, C. (2020). A review on the genetic resources, domestication and breeding history of spinach (*Spinacia oleracea* L.). Euphytica 216, 48. https://doi.org/10.1007/s10681-020-02585-y

Samuoliené, G., Brazaityte, A., Viršile, A., Miliauskiene, J., Vaštakaite-Kairiene, V. & Duchovskis, P. (2019). Nutrient levels in *Brassicaceae* microgreens increase under tailored light-emitting diode spectra. Front Plant Sci 10, 1475.

Sarıçam Kökpınar, Ş., Kantoglu, K. Y., & Ellialtıoğlu, Ş. Ş. (2024). Marulda (*Lactuca sativa* var. *longifolia*) mutasyon ıslahı yönteminin morfolojik etkileri. Bahçe 53(Special issue 1), 364-373. https://doi.org/10.53471/bahce.1509609

Sarıçam, Ş., Kantoğlu, K. Y. & Ellialtioğlu, Ş. Ş. (2017). Determination of Effective Mutagen Dose for Lettuce (*Lactuca sativa* var. *longifolia* cv. Cervantes) Seeds. Eurasian Journal of Agricultural Research, 1(2): 108-114.

Singh, H., Khar, A. & Verma, P. (2021). Induced mutagenesis for genetic improvement of Allium genetic resources: a comprehensive review. Genet Resour Crop Evol 68:2669–2690. https://doi.org/10.1007/s10722-021-01210-8

Sjödin, J. (1962). Some observations in X_1 and X_2 of *Vicia faba* L., after treatment with different mutagens. Hereditas 48,565-586.

Spencer-Lopes, M. M., Jankuloski, L., Mukhtar Ali Ghanim, A., Matijevic, M., & Kodym, A. (2018). Physical Mutagenesis. In: Manual on Mutation Breeding Third Edition Spencer-Lopes, M. M. Forster, B. P. & Jankuloski, L. (Eds). Food and Agriculture Organization of the United Nations. Rome, Italy. 301pp.

Sun, X., Li, X., Lu, Y., Wang, S., Zhang, X., Zhang, K., Su, X., Liu, M., Feng, D., Luo, S., Gu, A., Fu, Y., Chen, X., Xuan, S., Wang, Y., Xu, D., Chen, S., Ma, W., Shen, S., Cheng, F. & Zhao, J. (2022). Construction of a high-density mutant population of Chinese cabbage facilitates the genetic dissection of agronomic traits. Mol Plant 15, 913–924.

Taner, Y., Beşirli, G., Kunter, B. & Yanmaz, R. (2004). Determining effective radiation mutagen dose for garlic (*Allium sativum* L.). Bahçe 33 (1-2): 95-99. (in Turkish).

Tepe, A., Fırat, A. F., Taner, K. Y., Kunter, B., Peşkircioğlu, H. & Ekiz, H. (2003). Sera Demre 8 biber çeşidinde mutasyon ıslahına yönelik olarak etkili mutasyon dozunun belirlenmesi. IV. Ulusal Bahçe Bitkileri Kongresi. 8-12 Eylül 2003, Antalya-Türkiye, 365-366 (in Turkish).

Urva, H. S., Jamil, Y., Ul Haq, Z., Mujahid, T., Khan, A. U., Iqbal, M. & Abbas, M. (2017). Low power continuous wave-laser seed irradiation effect on *Moringa oleifera* germination, seedling growth and biochemical attributes, Journal of Photochemistry and Photobiology B: Biology 170, 314-323, https://doi.org/10.1016/j.jphotobiol.2017.04.001

Van Harten, A. M. (1998). Mutation Breeding Theory and Practical Applications. Cambridge University Press, London. 353.

Velkov, N., Tomlekova, N. B. & Sarsu, F. (2016). Sensitivity of watermelon variety Bojura to mutant agents ^{60}Co and EMS. J BioSci Biotech 5(1): 105-110.

Wang, J., Zhang, Y., Zhou, L., Yang, F., Li, J., Du, Y., Liu, R., Li, W. & Yu, L. (2022). Ionizing radiation: Effective physical agents for economic crop seed priming and the underlying physiological mechanisms. International Journal of Molecular Sciences 23(23):15212. https://doi.org/10.3390/ijms232315212

Wang, N., Zhang, Y., Wang, Y., Liu, Z., Li, C. & Feng, H. (2020). Assessment of a stay-green mutant for variety improvement in Pakchoi (*Brassica campestris* L. ssp. *chinensis*). Scientia Horticulturae 266, 109261. https://doi.org/10.1016/j.scienta.2020. 109261.

Xu, D., Li, Y., Yao, Z. Yin, Y., Feng, H. & Wei, Z. (2019). Study on the relationship between seed absorbed dose and seed composition of ^{252}Cf neutron source irradiated bean seed. Sci Rep 9, 9635. https://doi.org/10.1038/s41598-019-45829-1

Yarar, G., Kocak, M., Denli, N., Cavagnaro, P. F. & Yıldız, M. (2021). Determination of the effective radiation dose for mutation breeding in purple carrot (*Daucus carota* L.) and possible variations formed. Molecular Biology Reports 49(6):5219-5228. https://doi.org/10.1007/s11033-021-06618-0