

RADIOSENSITIVITY OF SOFT WHEAT SEEDS TO GAMMA RADIATION

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The study of the effect of different doses of γ -irradiation on the growth and development of seedlings of soft wheat seeds, resistance, and germination of seeds to irradiation has been carried out. The data show that the seeds of common wheat are resistant to ionizing radiation and up to 80% of the seeds remained viable. Under the influence of ionizing radiation, the functional and mitotic activity of the cell nucleus is disrupted, which has a significant effect on the growth parameters of the seedlings of irradiated seeds.

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Introduction. According to the UN FAO, annual losses of agricultural and food products reach 30%. The main causes of losses are associated with damage to crops by insect pests and diseases, premature germination of tubers and root crops, and bacterial spoilage of products. Reducing losses is one of the essential reserves for increasing production efficiency. To solve these problems, it is necessary to introduce environmentally friendly technologies, among which the most promising are technologies using physical factors, in particular, ionizing (IR) and non-ionizing radiation (NR). Irradiation technologies require less energy, replace or drastically reduce the use of chemicals, exclude environmental pollution, as well as the entry of toxicants into products. Irradiation technologies can be used to increase yields, increase the shelf life of products, destroy pathogenic microflora and insect pests, reduce losses during storage of fruits and vegetables and lengthen product sales. According to some sources, radiation technologies are able to solve many topical issues of food security, are cost-effective and environmentally safe. Over the past decade, there have been opportunities for their use in traditional technologies for the production, storage, and processing of agricultural and food products [1]. However, the issue of the safety of their use and the selection of the optimal radiation doses still requires a large-scale study. Thus, it is known that under the influence of small doses of IR, changes in expression occur in more than 500 genes of rice seedlings associated with cellular processes and signaling pathways involved in the synthesis of the cell wall and the biosynthesis of secondary metabolites [2]. In rice varieties

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obtained by mutagenesis, IR causes changes in the metabolism of carbohydrates and proteins [3], such changes often concern antioxidant systems [4, 5]. As studies show, high doses of radiation have a positive effect on seed productivity, influencing the chromosomal apparatus of wheat cells: on the nucleotide composition and the content of phosphorus compounds [6].

This study aimed to study the effect of γ -irradiation on germination of soft wheat seeds, in particular, to determine the degree of radiosensitivity of soft wheat seeds, and its influence on the morphological characteristics of seedlings of irradiated wheat seeds.

Materials and Methods.

Processing and Germination of Seeds. In this study, we used seeds of the common wheat variety Amby (d1D2D3; ch1rCh2r), which contains genes for hybrid dwarfism and hybrid red chlorosis. Dry wheat seeds were subjected to one-time γ -irradiation, which was carried out at the Institute of Physical Research of the National Academy of Sciences of the Republic of Armenia on an isotope emitter K120.000 Co60, with quantum energy of 1.20 MeV, at an irradiation power of 0.4 Gy/s. The treated and control seeds were soaked overnight, then the hatched seeds were sown in trays and then germinated for the next 6 days in a thermostat at the 26°C.

Radiation Sensitivity Test. Irradiated seeds, 20 seeds in each Petri dish, were sown on moistened filter paper and then germinated in a thermostat at 26°C under laboratory conditions. Seeds were considered germinated if they had a radical elongation >2 mm. Germinated seeds were counted daily for four days to determine the final percentage of germination. The final percentage of germination (FGP) was calculated using the formula [7] as follows:

$$\text{FGP} = \frac{N_T 100}{N},$$

where N_T is the proportion of germinated seeds for each treatment for the final measurement; N is the number of seeds used in the bioassay.

The length of the seedlings of the control and irradiated samples was measured on the 7th day after the start of the experiment. Seedling length data are arithmetic mean values of three independent biological replicates.

Results and Discussion. Ionizing radiation is subject to more research, as this radiation is considered an environmental factor that damages both humans and plant cells. However, at present, radiation effects are used to breed new forms of plants, for pre-sowing treatment and disinfection of seeds, storage of vegetables and products in warehouses, etc. Many studies have shown an increased radiosensitivity on the part of cells during division and differentiation. Since the seeds of cereals, and in particular, wheat, are more resistant to radiation compared to vegetative plants, the region of low doses shifts for them towards high absolute values of the absorbed dose, which makes seeds a convenient object from a methodological point of view. The study of the effect of γ -irradiation of seeds on the development of plants is of great practical importance since it is an integral part of the work on the scientific substantiation of the technology for stimulating the growth and development of agricultural crops using pre-sowing irradiation. In addition, there are studies

confirming the possibility of using radiation to obtain mutations that improve economically important traits of agricultural crops.

Mutagenesis and mutant cultivars of various crops registered in Bulgaria (retrieved from [8])

Crops species	Commonly used mutagens and range of doses	Number of mutant cultivars developed**	Main mutant characteristics reported
Barley	^{60}Co γ rays (20-100 Gy); UV-C light (0.5-5 J/cm ²)	3 (2) ● Rectangular Snip	Increased productivity; improved grain characteristics; spike fertility; short and lodging resistant stalks; short vegetation period; high hardness to dip; high cold tolerance; resistance to powdery mildew and to brown, black and stem rust
Common wheat	^{60}Co γ rays (50 Gy); NaN_3 (0.1, 1-10 mM); EMS (0.1-0.3%)	4 (1)	High grain yield; high and stable productivity; ecological adaptability, drought and cold tolerance; improved tolerance to lodging and shedding; resistance to brown rust; quality index - softening of dough and energy for dough deformation
Durum wheat	^{60}Co γ rays (50-200 Gy); EMS (0.1-3.5%)	8 (1)	High yield, high productivity; high cold tolerance; short stem; awlessness; new leaf shape - spherococcum, erectum, compactum, spread rosette
Maize	NEU (0.001-0.01%); NMU (0.001-0.01%); Dioxane (<1%); DMS (0.1-0.3%); DES (0.1-0.3%); EMS (0.1-0.3%)	(26)	High grain yield and productivity; tolerance to dense sowing; early ripening; drought tolerance; high protein content; high biomass dry matter; shifts in the flowering time; white-colour grain; strong stem; altered ear length; increased number of rows
Sunflower	^{60}Co γ rays (20-500 Gy); ^{137}Cs γ rays (8-540 Gy); Ultrasound (0.5-25.5 w/cm ²); EMS (0.2 - 0.8%)	(3)	Higher seed oil content; higher plants; larger leaves; greater number of branches; larger diameter of stem and plant head; altered size, position, shape, colour of leaves, seeds and inflorescence; increased seed weight; shortened vegetation period; small stalk height; increased oil contents; improved oil seed composition; cytoplasmic male sterility; better combining ability; resistance to <i>Orobanche cumana</i>

Above presented a small part of a table from the article [8], indicating the receipt of new mutants of some agricultural importance plants with improved properties as a result of irradiation with gamma radiation.

As is known, in plants, cellular macromolecules are modified by direct ionization and the reactivity of high-energy particles formed as a result of water radiolysis (indirect ionization effects), influencing, according to some estimates, 2000 events of primary ionization [9]. In plants, light reactions of photosynthesis are initiated by photolysis of water, a process with the same products as radiolysis of water, and which can lead to the formation of a huge number of oxidative radicals, which plants are usually able to neutralize due to their high production of antioxidants [10].

Previously, we also studied the effect of γ -irradiation of ^{60}Co at doses of 50 Gy, 100 Gy, 150 Gy, and 200 Gy on seeds of common wheat cultivar Frisco L-2 (D1d2D3; ch1rCh2r), which has hybrid depression. However, treatment with such high doses of ionizing radiation with aim to obtain mutations in the direction of weakening genes of depression and thus overcoming hybrid depression in wheat, we have not been achieved [11].

In the presented work, seeds of common wheat Amby, also possessing hybrid depression, were used as the object of research. Wheat hybrid depression is especially difficult to overcome when strong alleles are combined. According to some authors, the most effective method for overcoming depression in strongly necrotic hybrids (F1) is γ -irradiation with a radioactive isotope (^{60}Co). Cobalt-60 is the longest-lived of the radioactive isotopes of cobalt; it has important practical

applications, especially for the production of new varieties and breeds in agriculture. Investigation of the effect of IR on wheat seeds with hybrid depression, firstly, revealed that seeds germinate in dark conditions in an unnatural way for control seeds, and the ability to lodge was manifested. This indicates the occurrence of mutations in the genes Rht-B1b, Rht-D1b, Rht8, Rht11 lodging resistance in seeds of common wheat [12]. As you know, wheat lodging is one of the main problems of reducing the yield and grain quality of winter and spring wheat. Fig. 1 shows 4-day-old seedlings of control seeds and seeds irradiated with ionizing radiation, where lodging of seedlings and a change in their color are seen.



Fig. 1. Photo of seedlings of seeds of control and treated with γ -irradiation at doses of 50 Gy.

The test for radiation sensitivity of wheat seeds after gamma irradiation of 50 Gy and 100 Gy showed that the maximum percentage of germination is observed in control seedlings. As shown in Fig. 2, the final percentage of germination decreased with an increase in the dose of gamma-radiation by 3% and 11%, respectively, relative to the control seeds, as shown in Fig. 2.

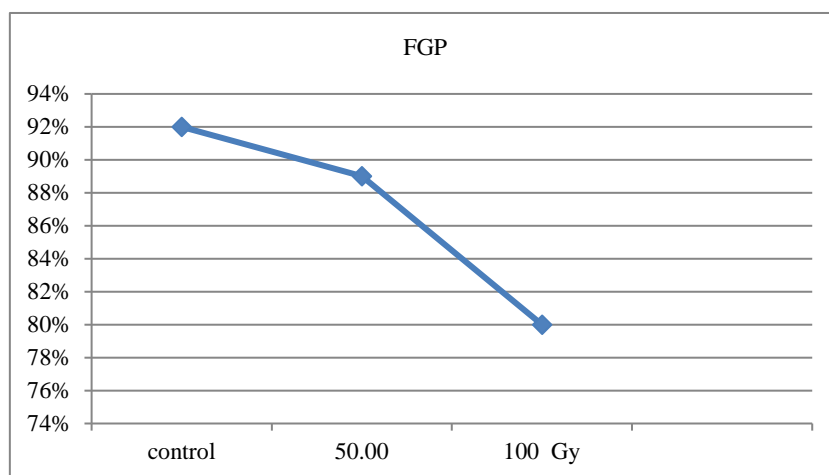


Fig. 2. Influence of gamma irradiation on the final percentage of seed germination.

With the absorption of smaller doses of radiation, when the cell is still alive, more or less significant changes occur in its organelles, especially in the cell nucleus, which is reflected in many physiological and molecular-biochemical parameters. Therefore, we carried out studies on the effect of irradiation of wheat seeds with gamma radiation of the radioactive isotope ^{60}Co at a dose of 50–200 Gy on the physiological parameters of seedlings of irradiated seeds.

We have investigated the effects of ionizing radiation at doses of 50–200 Gy in the early stages of seed germination, since low doses of ionizing radiation are believed to have various biological effects through DNA damage, some of which are beneficial for plant growth [8, 13]. As evidenced by the data presented in Fig. 3 IR-treated seeds that survived and germinated in all treatments were inferior in growth to the control seeds.

The dependence of changes in the morphophysiological characteristics of germination on the dose of irradiation of seeds with a radioactive isotope ^{60}Co is presented for 7 day old seedlings of control and irradiated wheat seeds (during the formation of the first leaf), the data are presented in Fig. 3. From the data obtained, it follows that with an increase in the irradiation dose, the length of seedlings decreases in an inverse proportional relationship. According to our data, it can be judged that the studied radiation doses are stressful for the studied common wheat with hybrid depression, which is possibly the main reason for the suppression of growth. However, further research on the length of the roots, on the total weight of the seedlings, may reveal improved indicators. Since, although in terms of the morphological parameter – the length of the seedling, the irradiated seeds are inferior to the control ones, however, outwardly the seedlings look larger than the control ones.

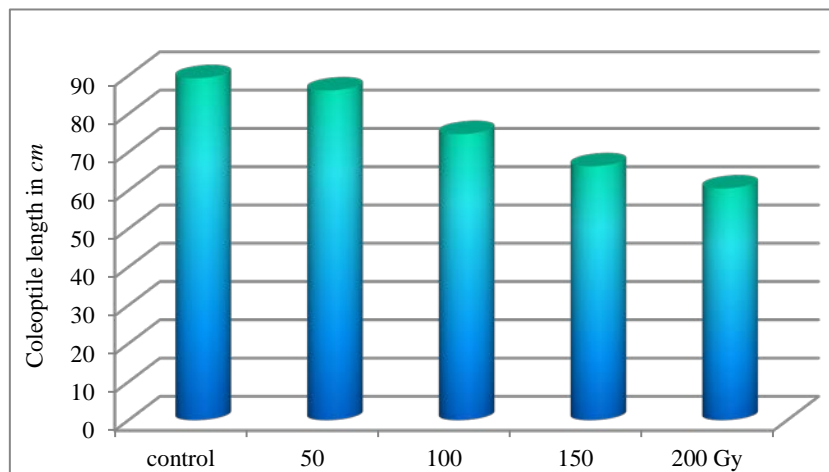


Fig. 3. Dependence of changes in the length of the coleoptile of wheat seedlings on the radiation dose.

Conclusion. Wheat is an agricultural valuable crop, the seeds of which are more resistant to irradiation compared to other vegetative plants, therefore the region of low doses shifts for them towards high absolute values of the absorbed dose, which makes seeds a convenient object from the methodological point of view. The study of the effect of γ -irradiation of seeds on plant development is of great practical

importance, since it is an integral part of the work on the scientific substantiation of the technology for stimulating the growth and development of agricultural crops using pre-sowing irradiation. AI affects the germination of wheat seeds, as a result of which some of the seeds lose their ability to germinate and die, but the seeds that have adapted germinate and may have better characteristics. As shown, Experimental data indicate that as a result of chain reactions arising from the absorption of radiation energy, the functional activity of subcellular structures, macromolecules (DNA, RNA, proteins), ATP, coenzymes changes, and there is also a change in the growth and color of seedlings [4, 6, 8]. Under irradiation in relatively low doses, a temporary arrest of mitosis is observed. Disruption of the normal course of mitosis is accompanied by chromosomal rearrangements, the emergence of mutations leading to shifts in the genetic apparatus of the cell, therefore, to a change in subsequent cell generations (cytogenetic effect), which we have shown earlier [11]. As this study has shown, AI affects the germination of wheat seeds, some of the seeds lose their ability to germinate and die, but the final percentage of germination at the investigated doses of AI reaches 80%. Changes at the cellular level are reflected in the length of coleoptiles; there is a slight suppression of plant growth in length. However, seeds adapted to new growing conditions may have better characteristics of economically important traits. Further studies will show how the effect of AI on the biochemical parameters of the seedling nucleus of irradiated seeds is reflected, on the morphological parameters, the content of nucleic acids and protein.

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ՓՈՓՈԻԿ ՑՈՐԵՆԻ ՍԵՐՄԵՐԻ ՌԱԴԻՈՉՖՈՒՆԿՈՒԹՅՈՒՆԸ ԳԱՄՄԱ-ՃԱՌԱԳԱՅԹՄԱՆ ՆԿԱՏՄԱՄԲ

Իրականացվել է փափուկ ցորենի սերմերի ծիլերի աճի և զարգացման վրա γ -ճառագայթման տարբեր դոզաների ազդեցության, ճառագայթման նկատմամբ սերմերի կայունության և ծլեցման հետազոտություն: Տվյալները ցույց են տալիս, որ փափուկ ցորենի սերմերը դիմացկուն են իոնացնող ճառագայթման նկատմամբ, և սերմերի մինչև 80%-ը պահպանել է կենսունակությունը: Իոնացնող ճառագայթման ազդեցության տակ խախտվում է բջջային կորիզի ֆունկցիոնալ և միտոտիկ ակտիվությունը, ինչն էական ազդեցություն է թողնում ճառագայթահարված սերմերի ծիլերի աճի պարամետրերի վրա:

Л. А. МИНАСБЕКЯН, И. А. АВАГЯН

РАДИОЧУВСТВИТЕЛЬНОСТЬ СЕМЯН МЯГКОЙ ПШЕНИЦЫ К ГАММА-ОБЛУЧЕНИЮ

Проведено исследование влияния различных доз γ -облучения на рост и развитие проростков семян мягкой пшеницы, устойчивости и всхожести семян к облучению. Данные показывают, что семена мягкой пшеницы устойчивы к ионизирующему излучению и до 80% семян сохраняли всхожесть. Под действием ионизирующего излучения происходит нарушение функциональной и митотической активностей клеточного ядра, что оказывает значительное влияние на ростовые параметры проростков облученных семян.