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STUDY OF FREE PROLINE IN SOME AGRICULTURAL PLANTS WHICH SEEDS ARE EXPOSED TO STERILIZING γ -IRRADIATION

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Abstract: Herein, it has been presented the results of phenological observations, and also investigated the dynamics of dose-dependent changes in the amount of free proline in beans, eggplant, cucumber, tomato, as well as in potato, the seeds, and roots, which were processed with γ rays in the sterilization dose before sowing. It has been shown that pre-sowing irradiation in high doses increases the content of proline in all studied plants. It has been established that the growth and development of plants slow down as the dose increases. It has been shown that potato tubers irradiated in a sterilization dose are not suitable for subsequent sowing.

Keywords: presowing irradiation, sterilization dose, beans, eggplant, cucumber, tomato, potato, free proline

Food preservation is based on the ability of microorganisms to respond to the effects of physical, chemical and biological factors. It is possible to increase the shelf life of food products by changing the environmental conditions and exerting one or another impact on the product. Environmental conditions can be changed by sterilization, for example, by heat or chemical processing.

Sterilization is the elimination of viable microorganisms and their spores. One of the methods of sterilization is the radiation, which is carried out by irradiating the product with ionizing radiation. For this sterilization method, the absorption dose is set in the range of 0.03 kGy-0.1 kGy - in the case of processing potatoes and onions from germination [4], and 10 kGy - 50 kGy for sterilization of medicinal plant materials, medicinal herbal preparations, herbal medicines, etc. [1]. The use of other doses is allowed if it is previously proved that the chosen mode provides the necessary and reproducible level of mortality of microorganisms. Many developing countries often seek to effectively implement food radiation processing. Since the heat of food products does not occur during radiation processing, they retain their original freshness and physical condition.

The ability to slow down the germination of root crops prevents the reproduction of insects, eliminates viable microorganisms and their spores, as well as mold, radiation processing makes it possible to reduce the loss of harvested crops and get nutritious foods suitable for long-term storage [2].

The effect of increased doses of ionizing radiation leads to the disruption of cellular metabolism. One of the protective mechanisms for oxidative stress is a stress-dependent accumulation of low-molecular organic antioxidants in plants. These metabolites include proline.

The purpose of these studies was to study the dose-dependent content of proline for some agricultural plants, of which seeds were processed at a sterilization dose. We conducted pre-plant irradiation of seeds of agricultural plants, such as beans, eggplant, cucumber, and tomato, as well as potato tubers in different doses, studying their growth and development, and trying to investigate the effect of irradiation on the amount of free proline.

1. Materials and Methods

The object of study. Some agricultural plants were selected as objects of study: potato tubers, along with the seeds of beans, eggplant, cucumbers, and tomato, which were subjected to sterilization processing with γ -rays. Plant seeds and tubers were irradiated in the “RUXUND” device, with a source of irradiation ^{60}Co at absorption doses of 0.3 kGy, 0.5 kGy, 1 kGy, 1.5 kGy, 2 kGy (dose rate in all cases was 0.19 Gy/s). The irradiated seeds were planted at the experimental site of the Institute of Botany of the National Academy of Sciences of Azerbaijan.

Equipment. “RUXUND” device, with the γ -radiation source - ^{60}Co , HIMAC –CT 15 RE (United Kingdom) type centrifuge, JENWAY –67 Series (United Kingdom) type spectrophotometer.

Method for the free proline determination. The Bates method was used (Bates et al., 1973) to determine the content of free proline [3]. The method is based on the interaction of free proline with ninhydrin reagent, forming a pink-red color. A weighed portion of fresh leaf tissue was homogenized in 10 ml of a 3% sulfosalicylic acid solution. The homogenate was centrifuged at 5000 g for 10 min. 2 ml of ninhydrin reagent and 2 ml of glacial acetic acid were added to 2 ml of the supernatant in the reaction tube. The tubes were closed with a stopper with a reflux condenser and kept for 1 h in a boiling water bath. Then the samples were cooled up to room temperature, 4 ml of toluene was added to them and carefully mixed by shaking. The top layer of toluene, containing the colored complex of the proline-ninhydrin, was separated from the aqueous phase and colorimetricated on a spectrophotometer at 520 nm.

The content of free proline was determined by a calibration curve constructed using proline solutions in the concentration range from 50 to 150 $\mu\text{g/ml}$, and expressed in micrograms per 1 g wet weight.

2. Discussion of results

The results of phenological observations are presented in Figure 1.

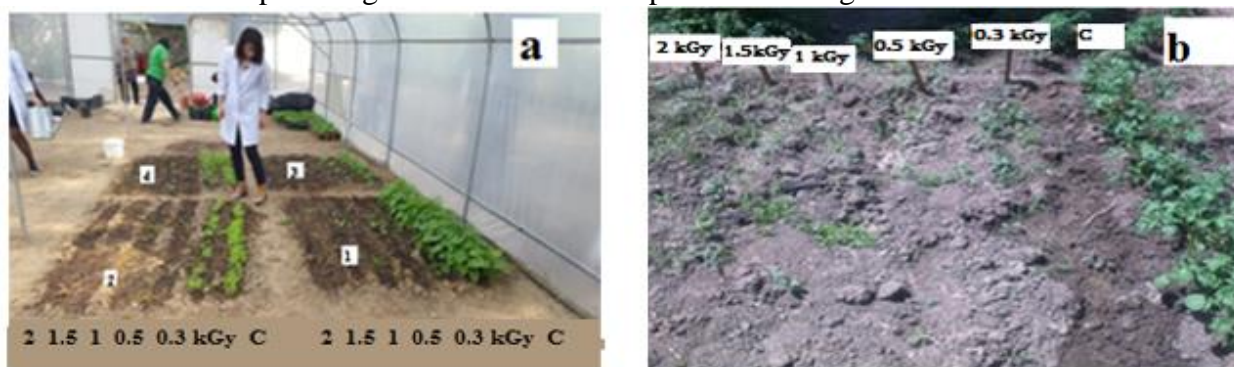


Fig.1. Three-week-old seedlings of plants of the bean, eggplant, cucumber, tomato (at the site, respectively 1, 2, 3, 4) and potatoes (at the site b).

As a result of phenological observations, it has been determined that non-irradiated (control - K) seeds of plants of beans, eggplant, cucumber, and tomato, as well as irradiated at doses of 0.3 kGy and 0.5 kGy sprouted (Fig.1a). The non-irradiated seeds sprouted one week after planting, and seeds irradiated at a dose of 0.3 kGy after three days and irradiated at a dose of 0.5 kGy six days after the control samples. However, most seedlings of seeds irradiated at a dose of 0.5 kGy, were unviable and wilted after a week.

Tubers of potatoes irradiated at all doses did not grow (Fig. 1b). From this, we can conclude that although irradiated potato tubers suitable for use, they are not suitable for subsequent sowing.

Counting the number of flowering plants at the beginning of flowering showed that in all irradiated samples, the number of flowering plants at the day of counting was 3-4 times higher than the plants in control. The same tendency was observed in the phase of fruit set, that is, the fruits on irradiation variants were significantly higher (Fig.2).



Fig.2. Observation of the flowering and fruit set in cucumbers, tomato, and eggplant.

As they grew, plants from seeds irradiated at a dose of 0.3 kGy became noticeably different from the plants in control. So, they were greener, higher and their leaves and fruits were larger (Fig. 3).

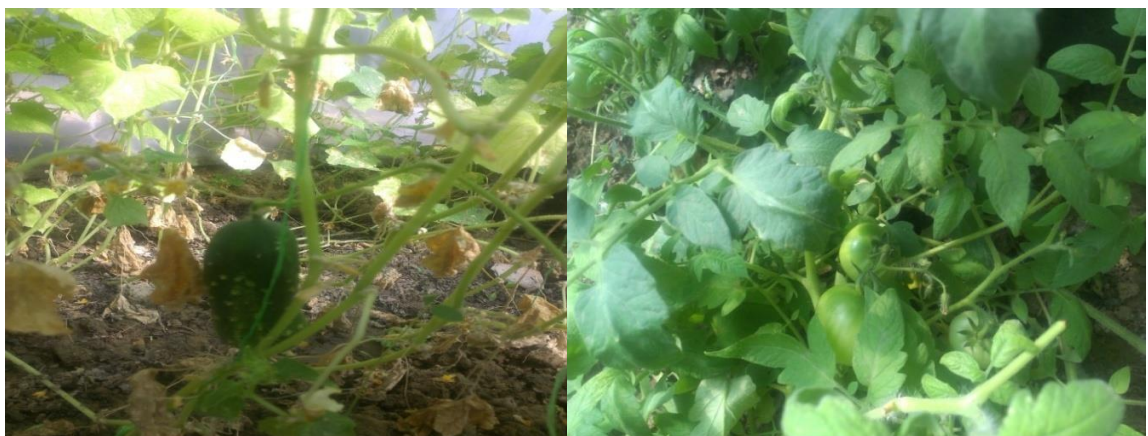


Fig.3. Fruits of cucumber and tomato plants irradiated with a dose of 0.3 kGy.

3. Results

The results of the dose-dependent content of proline in plants are presented in Figure 4. Since the studied plants (except potatoes) grew only at the doses of 0.3 and 0.5 kGy, the figure shows the data on these doses. Results were compared with non-irradiated control plant samples.

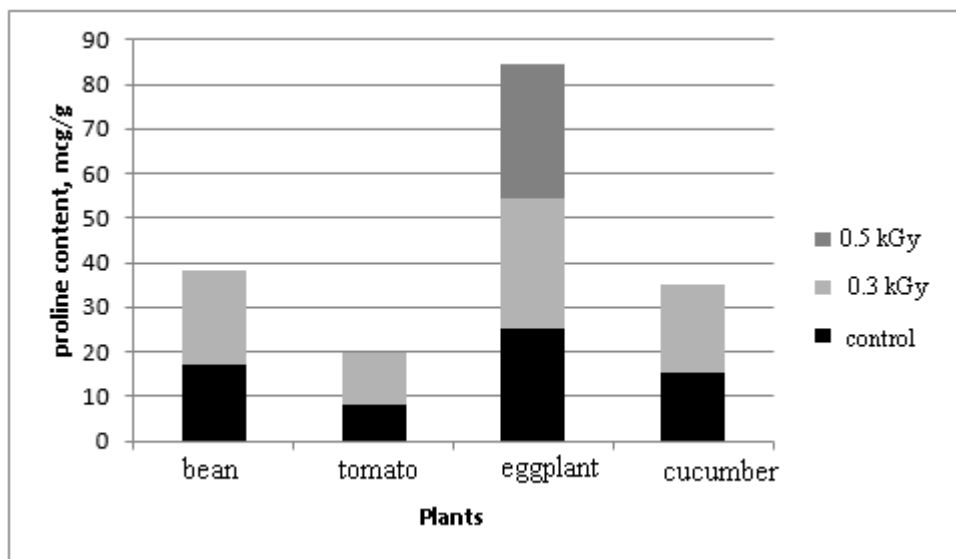


Fig. 4. Dose-dependent content of proline in plants.

From the obtained results it becomes clear that the control samples of all the presented plants differ from each other in the content of the proline. So, this amount is the highest in eggplant, a little less in beans and cucumber, and the lowest in tomato.

Presowing processing with gamma rays increases the content of free proline in all presented plants. This increase is ~25% in beans, ~50% in tomato, ~17% in eggplant and ~25% in cucumber.

Interestingly, even in the case of seed processing at a dose of 0.5 kGy, eggplant has a high proline composition.

As can be seen from the figure, all the presented plants differ from each other in the content of the proline. So, the amount of proline is the highest in eggplant and the smallest in the tomato. Pre-sowing seed processing with γ -rays increases the amount of free proline in all presented plants.

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