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ABSTRACT

of the dissertation for the degree of Doctor of Philosophy (Doctor of Science)

STUDY OF PHYSIOLOGICAL ADAPTATION FEATURES OF COTTON PLANT TO γ-IRRADIATION AND SALT STRESS CONDITIONS

Speciality:	2411.02 – Plant physiology
Field of science:	Biology
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GENERAL NATURE OF THE WORK

Relevance of the topic and degree of development. As a result of human activities, especially the use of fossil fuels, deforestation for industrial purposes and other similar processes, the release of greenhouse gases such as carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O) into the atmosphere leads to climate changes on a global scale. Climate change has a serious impact on ecosystems, a group of closely related living organisms and the environment in which they live. If these processes continue at the current rate, some plant species in nature may lose their ability to adapt to changing ecological conditions. In order to increase the resistance of plants to stress factors under climate change conditions, interest in studying physiological processes has recently increased[1]. In this regard, studying the impact of unfavorable environmental factors on physiological and biochemical processes occurring in cells is one of the priority directions of biological science. In modern times, the mechanism of resistance to environmental stress factors in plants has not been fully elucidated[2]. It is believed that the main cause of soil salinization is the aggravation of ecological factors and improper implementation of agrotechnical measures in agriculture. It is noted that if necessary measures are not taken, salinization of arable lands may reach ~50% by 2050. According to recent data, ~1.4 million hectares of irrigated lands in the Republic of Azerbaijan are to some extent saline. It is shown that in the near future this figure may increase to ~1.9 million hectares, which is about half of the agriculturally suitable area of our country (~49.8%). It is known that currently radiation technologies are widely used as a new physical method in various fields of agriculture. Studying and evaluating the

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[1] Chaudhry, S., Sidhu, G.P.S. Climate change regulated abiotic stress mechanisms in plants: a comprehensive review. / S.Chaudhry , G.P.S. Sidhu, //Plant Cell Reports 2022,- 41, -p. 1–31 -. https://doi.org/10.1007/s00299-021-02759-5

[2] Cramer, G.R. Effects of abiotic stress on plants: a systems biology perspective / G.R. Cramer, K. Urano, S. Delort, M. Pezzotti, K. Shinozaki // BMC Plant Biology, -2011, v. 11, - p. 163

stimulating effects of radiation and various doses and concentrations of salt is also one of the important problems facing modern science. In order to reduce the negative effects of salt stress, many studies have been conducted to improve the salt tolerance and physiological adaptation properties of plants using γ-radiation as a physical mutagen[3]. The mentioned problems require studying the mechanism of changes in metabolic processes within the cell due to the effect of salinization. In order to understand the mechanism of response reactions in plants to the effects of abiotic factors of the environment, their adaptive properties should be studied at different levels of organization of organisms[4].

Cotton is a technical plant of great strategic importance, cultivated in large areas in Azerbaijan. On July 14, 2010, the law "On Cotton Growing" was adopted in our republic, which envisaged the application of new technologies in the cultivation and production of cotton, which is considered a valuable raw material for the textile industry and many other fields[5]. State support for the development of cotton growing in our country, approved by the Order No. 3082 of the President of the Republic of Azerbaijan Ilham Aliyev dated July 13, 2017 entitled "State Program on the Development of Cotton Growing in the Republic of Azerbaijan for 2017-2022", is aimed at solving problems in this area. Cotton growing is one of the production areas in our country that has wide opportunities in meeting the population's demand for food products, raising the standard of living of the rural population, and developing the feed base in animal husbandry.

From this point of view, the treatment of seeds intended for sowing with gamma rays before sowing, as well as the investigation of the physiological and biochemical mechanisms of adaptation in

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[3] Azizov, G.Z. Classification of saline soils of Azerbaijan by salinity degree and type / G.Z. Azizov // -Baku: -2002. -29 p

[4] Jafarov, E.S. Plant radiobiology / E. S. Jafarov // Textbook – Baku: Elm, - 2023, – 276 p.

[5] Aliyev, I.H. The Republic of Azerbaijan, Law "On Cotton Growing" / I.H. Aliyev- Xalq newspaper, -Baku, - July 14, 2010.

cotton plants under the influence of various types of salts, and the creation of highly productive, stress-resistant and economically efficient local cotton varieties using these results are among the most urgent problems facing modern biology and agricultural science.

Object and subject of the research. Various research objects were used to study in depth the physiology of adaptation properties of plants to environmental stress factors. In the dissertation work, preference was given to the light and heat-loving technical cotton plant, which is typical for our Republic and is called the "white gold" of Azerbaijan. The local Ganja 182 variety of cotton was taken as the object of research. During the research, the amount of photosynthetic pigments in the leaves of the cotton plant, gas exchange parameters, and some antioxidant system enzymes in the leaves and roots were studied.

Goals and objectives of the research. The main objective of the research is to study the process of adaptation of cotton plants to different doses of γ-irradiation before sowing, as well as the stimulating effect of different concentrations of salt on cotton plants. To achieve this goal, the following tasks were set:

Cotton plants grown under γ-irradiation and salt stress:

1) Determination of the amount of photosynthetic pigments in their leaves;

2) Determination of gas exchange parameters and the amount of intermediate metabolites (H_2O_2, proline, MDA) in ontogenesis;

3) Study of the dynamics of changes in the amount of adenine nucleotides and nicotine coenzymes in leaves during ontogenesis;

4) Study of the dynamics of changes in the amount of proteins, fats and carbohydrates during ontogenesis;

5) The effect of radioactive radiation and salt stress on the activities of some carbon, nitrogen and antioxidant system enzymes in roots and leaves;

6) The effect of γ-irradiation and salt stress on the activity of nitrate reductase and carbonic anhydrase enzymes in leaves and roots during the ontogenesis of cotton plants.

Research methods. During the studies, the amount of photosynthetic pigments was determined according to Sims and Gamon. Gas exchange parameters were measured using an infrared gas analyzer (Bioscience, USA)[6]. The amount of proteins was determined according to Bradford [Bradford, 1976][7]. Enzyme activity was measured using a spectrophotometer. A number of analytical and preparative methods were used during the experiments.

The main provisions put forward for defense:

• In cotton plants, γ-irradiation and low doses (5-50 Gy) and concentrations (5-50 mM) of salts have a stimulating effect on the physiological and biochemical processes occurring in the cell, while at high doses and concentrations they have an inhibitory effect, minimizing the adaptive capacity of the plant.

• In the roots and leaves of cotton plants, the antioxidant defense system and carbon and nitrogen metabolism enzymes work in coordination with proton pumps localized in the root cells under stress conditions.

• In cotton plants, the effectiveness of stress factors, such as γ-irradiation and salinity, depends on the phase of ontogenesis, the dose of radiation, the ionic composition and concentration of salts.

• The activity of the KA enzyme highly depends on the concentration of Zn^{2+} ions in the environment.

Scientific novelty of the work. For the first time, the functional changes in the physiological parameters of cotton plants under the separate and combined effects of γ-irradiation and salt stress have been comprehensively and comparatively studied. It was found that irradiation at doses of 5, 10 and 50 Gy has a stimulating effect on plant growth and development. Regardless of the phase of ontogenesis, the amount of biopolymers (fats and proteins) increases

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[6] Sims, D.A. Relationships between leaf pigment content and spectral reflectance across a wide range of species, leaf structures and developmental stages / D.A.Sims, J.A.Gamon // Remote Sensing of Environment, 2002, v. 81, p. 337-354

[7] Bradford, M.M. A rapid and sensitive method for the quantitation of micro-gram quantities of protein utilizing the principle of protein-dye binding / M.M. Bradford // AnalBiochemistry, - 1976. №72,- p. 248–254

under salt stress conditions. The rate of photosynthesis (Pn) decreased by 29%, 38% and 62%, respectively, compared to the control variant, under the influence of stress at the stages of budding (BF), flowering (FP) and opening boll (OBP). The activity of KAT and APO antioxidant enzymes increases at doses of 5-50 Gy of γ-irradiation, and gradually decreases at higher doses (100, 200 Gy). Concentrations of NaCl, $FeCl_3$, Na_2SO_4, $ZnSO_4$ salts of 5-50 mM have a stimulating effect on the activity of CA and APO, while at concentrations of 100 and 200 mM, inhibition of the activity of their enzymes occurs. Unlike chloride salts, sulfate-containing salts (Na_2SO_4, $ZnSO_4$) have a stimulating effect on the activity of CA, and at concentrations of 50-100 mM, the activity of the enzyme increases depending on time (10, 20 and 30-day-old plants). It has been shown that the activity of the KA enzyme is highly dependent on the concentration of Zn^{2+} ions in the environment. The dynamics of changes in the amount of photosynthetic pigments, biometric indicators of photosynthesis intensity and gas exchange parameters during radiation and salt stress can be explained as an indicator of the formation of high resistance of plants to the effects of stress factors.

Theoretical and practical significance of the work. In modern times, the reduction of land areas suitable for cultivation as a result of salinization increases the demand of people all over the world for technical plants of high economic importance. Since such problems are also important and relevant for the population of our country, it is very relevant to conduct scientific research in this field. The cotton plant, which is taken as the object of research, occupies one of the most important places in the world in terms of its production, sale and use as a technical plant. The results of scientific research, in addition to being theoretical and fundamental in nature, allow us to understand the physiological and biochemical mechanisms of the adaptation process that occurs in the cells of the leaf and root system during the effects of radiation and high salt concentrations on the cotton plant.

❖ The results of the adaptation process that occurs in plants against the effects of stress factors can be used to determine the limiting limits and stimulating doses of radiation and salt stress. The results we have obtained can be used as a marker trait in selection work aimed at increasing the intensity of photosynthesis and creating new salt-resistant varieties.

❖ The results of the dissertation work allow us to clarify the mechanisms of the impact of radioactive radiation, one of the main abiotic factors of the environment, on living organisms. The results of the dissertation work can be used as a scientific-theoretical-practical source in the teaching of the subjects “Photosynthesis”, “Respiration”, “Physiology and Biochemistry of Plants” at the faculties of Biology, Ecology, Agronomy and Soil Science of universities, and in bioecological research. The recommendations can also be used in farms, organizations and enterprises engaged in the production of environmentally friendly products.

Publication of the results of the dissertation: In accordance with the content of the research topic, 27 scientific works (9 articles in periodicals, 2 articles in conference materials, 16 theses) were published in periodicals, in materials of scientific conferences, congresses and symposiums of republican and international scale. Two of them were published in Scopus, one in Agris international databases, another in a journal included in the database of the Russian Academy of Sciences, and five in journals included in the database of the Azerbaijani Academy of Sciences.

Approbation of the dissertation work. The results obtained during the dissertation research were discussed at the following scientific meetings:

Международная научная конференция «Становление и развитие экспериментальной биологии в Таджикистане» (Душанбе, 2022); Всероссийская научно-практичесая конференция имени профессора М.Х. Ханиева (Россия, Нальчик, 2022); Ümummilli lider Heydər Əliyevin anadan olmasının 100-cü ildönümünə həsr olunmuş “Qeyri-neft sektoru və qlobal ərzaq təhlükəsizliyi problemləri” mövzusunda beynəlxalq elmi-praktik konfransda (Gəncə, ADAU, 2023); Ümummilli Lider Heydər Əliyevin anadan olmasının 100 illiyinə həsr olunmuş “Biologiyanın aktual

problemləri: Davamlı inkişaf kontekstində” elmi-praktik konfransda (Bakı, BDU, 2023); professor A.M.Əfəndiyevin 80 illik yubileyinə həsr edilmiş ″Biokimya və təbabətin aktual problemləri″ elmi-praktik konfransında (Bakı, ATU, 2023); V International Scientific and Practical Conference (Ukraine, 2023); akademik Rəhim Rəhimovun 100 illiyinə həsr olunmuş Beynəlxalq Elmi-Praktiki konfransda (Bakı, ATU, 2023); “Təbabətin aktual problemləri” mövzusunda beynəlxalq elmi-praktiki konqresdə (Bakı, 2023); “Müasir təbiət və iqtisad elmlərinin aktual problemləri” mövzusunda elmi konfransda (Gəncə, GDU, 2023); Международная научная конференция молодых ученых ″Фундаментальные и прикладные исследования в области молекулярной биологии, биохимии, биотехнологии″ (Алматы, 2023); Heydər Əliyevin anadan olmasının 100 illiyinə həsr olunmuş ″Avrasiyada biomüxtəliflik-SEAB-2023″ VI Beynəlxalq Simpoziumda (Mərdəkan, 2023); “Heydər Əliyev və Azərbaycan təbiəti” mövzusunda beynəlxalq konfransda (Bakı, 2023); V Международная научно-практическая конференция «Современные вопросы естествознания и экономики» (Проковьевск, КГУ, 2023); II Международной молодёжной конференции «Генетические и радиационные технологии в сельском хозяйстве» (Обнинск, 2023)
In the laboratories, conferences and seminars of the Azerbaijan State Agrarian University of the Ministry of Agriculture of the Republic of Azerbaijan and the Institute of Radiation Problems of the Ministry of Science and Education of the Republic of Azerbaijan.

Organization where the dissertation work was carried out. The dissertation work was carried out in the Plant Physiology Laboratory named after Academician Jalal Aliyev of the Biology Department of the Azerbaijan State Agrarian University of the Ministry of Agriculture of the Republic of Azerbaijan and in the Radiobiology Laboratory of the Institute of Radiation Problems of the Ministry of Science and Education of the Republic of Azerbaijan. Field experiments were conducted on a private farm located in the territory of the Alibeyli municipality of the Aghdam region, as well as on the Teaching and Experimental Farm of the Agrarian University.

Thesis size and structure: The thesis consists of 176 printed pages (318932 characters) and consists of “Introduction” (18007 characters), “Literature Review” (86580 characters), “Materials and Methods” (33565 characters), “Results and Discussion” (78664 characters), “Conclusion” (13477 characters), “Conclusions” (4048 characters), “Recommendations for Production” (874 characters), “List of References” (52955 characters) and “List of Abbreviations” (1529 characters). The author referred to a total of 260 literature sources, including 16 local and 244 foreign literature, of which ~70% cover recent years. During the interpretation and analysis of the results obtained in the dissertation work, 16 tables, 17 figures were used. The total text part of the dissertation (excluding figures, tables, graphs, appendices and bibliography) is 118 pages of computer text or 235220 characters. The research work was carried out in accordance with the thematic plan of the Azerbaijan State Agrarian University, Ministry of Agriculture of the Republic of Azerbaijan.

Personal contribution of the researcher: The idea of selecting the local cotton plant variety Ganja-182 as the object of the research was given by the author. The author closely participated in the planning of the research work to be conducted and demonstrated modern scientific approaches. The research methodology was fully adopted by the author, appropriate innovative changes were made and it was carried out in a free form. The author directly participated in the statistical analysis of the results obtained, discussions, and writing of the articles.

MAIN CONTENT OF THE WORK

The introduction explains the relevance of the topic, its object and subject, the goals and objectives of the research, its methods, the main provisions put forward for defense, and the scientific novelty and practical significance of the results obtained.

CHAPTER I. LITERATURE REVIEW.

In the literature review, literature sources on the topic of the dissertation were examined. Scientific research works related to stress and its study, scientific results obtained, all issues related to the topic of the dissertation were studied and analyzed on the basis of a wide range of existing literature sources[8]. At the same time, the effect of gamma radiation on the physiological adaptation properties of plants was thoroughly investigated[9]. The mechanisms of the effect of small doses of radiation on plant cells were also studied in detail[10].

CHAPTER II. METHODS AND CONDITIONS OF THE EXPERIMENT.

The object of the study was the local Ganja-182 variety of the common cotton plant *Gossypium hirsutum* L. species. First of all, the morphophysiological and biometric indicators of the plant were measured. To determine the amount of photosynthetic pigments in the active stages of plant development, samples were taken from its leaves and the amount of pigments was determined by the Sims and Gamon method. The rate of photosynthesis (P_n), stomatal permeability (g_s), the concentration of CO_2 in the intercellular spaces (C_i) and the rate of transpiration (T_r) were measured using an infrared gas analyzer (Li-COR 6400 XT Biosciences, Lincoln, USA). The amount of fats was determined in a Soxhlet apparatus [Ермаков et al., 1972]. The amount of proteins was determined according to the Bradford method [Bradford, 1976].

[8] Eckardt, N.A. 2023. Focus on climate change and plant abiotic stress biology / N.A. Eckardt, S. Cutler, T.E. Juenger, [et al.] // The Plant Cell, - 2023. №35(1), p. 1-3.

[9] Трофимова, Е. А. Влияние γ-излучения на развитие растений из облученных семян и проростков Allium Cepa L / Трофимова Е. А. , Дементьев Д. В., Болсуновский А. Я. // Радиационная биология. Радиоэкология, - 2019. Т. 59, № 3, - стр. 293-299.

[10] Болсуновский, А.Я. Влияние гамма-излучения в малых дозах на развитие растений из облучённых проростков Pisum sativum в лабораторных экспериментах / А.Я. Болсуновский, Е.А. Трофимова, О.П. Орешникова // Вестник НГАУ. – 2024. № (3) – с. 13-21.

The activity of the nitrate reductase enzyme was measured calorimetrically by the method of Krasilnikova [Красильникова и др., 2004]. The activity of other enzymes was measured spectrophotometrically. The activity of the carbonic anhydrase enzyme was determined electrometrically according to Anderson [Anderson N.G., Wilbur K.M., 1948][11]. The amount of H_2O_2 was determined spectrophotometrically based on the oxidation of Fe^{+2} ions to Fe^{+3} ions with xylene orange. The amount of malondialdehyde (MDA) was determined in the presence of thiobarbituric acid (TBT). The amount of proline was determined by the method of Trol and Lindsley [Trol and Lindsley, 1955] [12]. In a cotton plant sample, the growth rate (V_y) and length (L_y), root length (L_k), and the ratio of leaf length to root length (L_y/L_k) were determined . The values in the tables, diagrams and graphs given in the dissertation show the average mathematical expression of several repetitions. During the analysis of the results, the average mathematical errors and deviations (M±m) were taken into account, and $p<0.05$ was accepted as the reliability coefficient. The obtained results were analyzed using the computer programs "Statistica for Windows 10.0" and "Microsoft Office Excel 2010".

CHAPTER III. RESULTS OBTAINED AND THEIR DISCUSSION.

3.1. The effect of radioactive radiation and salinity on biometric indicators and morphophysiological properties of cotton plants. It is known that root systems formed in the first days of germination are

[11] Anderson N.G., Wilbur K.M. (1948) Electrometric and colorimetric determination of carbonic anhydrase. J. Biol. Chem., 176, 147-154

[12] Troll W. and Lindsley J., 1955. A photometric method for the determination of proline. J. Biol. Chem., 215, p. 655-660

an optimal object for studying the mechanisms underlying the resistance of plants to salt stress[13].

In this regard, first of all, the percentage of seed germination and morphophysiological changes in seedlings under the influence of stress were studied in the early stages of plant development (Fig. 1).

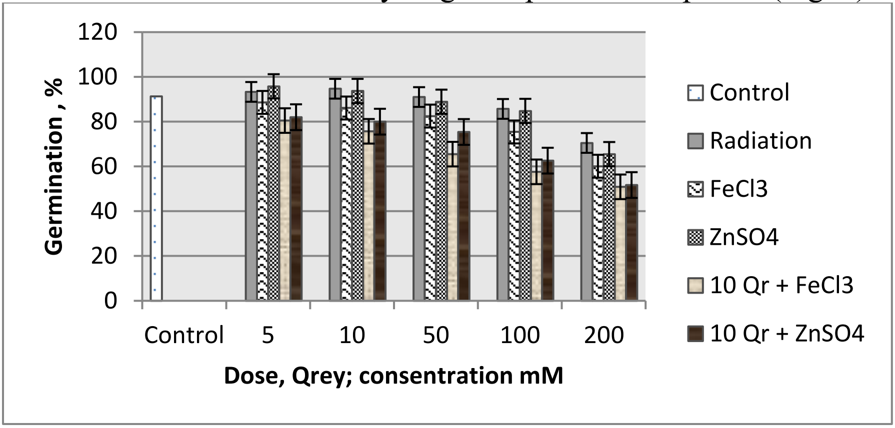

Figure 1. Effect of γ-irradiation and salinity on the germination of cotton seeds

As can be seen from the figure, the highest germination percentage is observed at doses of 5-50 Gy of radiation, as well as at concentrations of 5-50 mM of $FeCl_3$ and $ZnSO_4$ salts. When seeds are exposed to different concentrations of $FeCl_3$ and $ZnSO_4$ after irradiation at a dose of 50 Gy, the germination percentage of seeds decreases due to the effect of both salts[14].

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[13] Khalequzzaman, U.H. Seed priming improves germination, yield, and water productivity of cotton under drought stress / U.H. Khalequzzaman, S.K. Himanshu, N. Islam [et al.] // J Soil Sci Plant Nutr. – 2023. № 23(2), - p. 2418–2430.

[14] Алакбарова, Ш.Э. Влияние различных концентраций $FeCl_3$ на физиологические процессы у культуры хлопчатника, выращенных из семян, облученных γ – лучами // «Современные вопросы естествознания и экономики» Сборник трудов V Международной научно-практической конференции. Кузбасский Государственный Технический Университета г. Прокопьевск: - 16 марта 2023. - с. 528-530.

That is, the combined effect of radiation and salts (2-fold stress) reduces the germination capacity of seeds, which leads to the gradual weakening of plants and their death towards the end of the vegetative development phases.

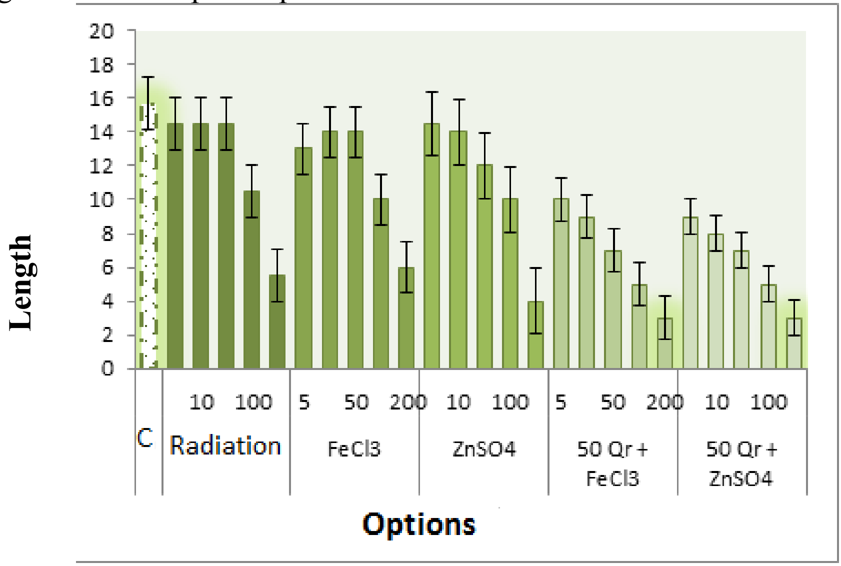

Figure 2. Effect of stress on root length of seven-day-old cotton seedlings in different variants (cm).

As can be seen from Figure 2, the experiments were conducted under standard conditions, and the biometric dimensions of all plant organs were measured depending on its developmental phases.

3.2 The effect of radiation and salt stress on the amount of photosynthetic pigments in the leaves of cotton plants. In the research work, the amount of photosynthetic pigments was measured in different phases of ontogenesis in cotton plants obtained from seeds exposed to γ-irradiation in the dose range of 5-200 Gy. It was found that the amount of carotenoids in the leaves, unlike chlorophylls, increases even at high radiation doses and until the final stages of ontogenesis, which can be associated with their adaptive functions resulting from the activation of appropriate metabolic processes in plant organisms.

Table 1.

Effect of radiation and salt stress on the amount of pigments in leaves during the ontogenesis of cotton plants

On-to.	C	Radiation, Qy					$FeCl_3$, mM					$ZnSO_4$, mM				
		5	10	50	100	200	5	10	50	100	200	5	10	50	100	200
Chlorophyll a (mq/q wet mass)																
LP	0,89	0,86	0,58	0,62	0,19	0,18	0,69	0,59	0,42	0,38	0,21	0,61	0,62	0,45	0,27	0,11
YP	0,65	0,81	0,83	0,74	0,49	0,19	0,74	0,79	0,79	0,41	0,22	0,77	0,84	0,81	0,24	0,20
QP	0,83	0,88	1,01	0,96	0,82	0,18	0,82	0,84	0,81	0,53	0,33	0,90	0,92	0,77	0,38	0,15
FP	0,87	0,90	1,71	1,48	0,96	0,09	0,99	1,07	0,99	0,49	0,28	1,04	1,11	0,95	0,40	0,18
BOP	0,98	0,96	1,23	1,33	0,66	0,07	0,99	1,05	0,96	0,61	0,21	1,05	1,13	0,95	0,41	0,27
Chlorophyll b (mq/q wet mass)																
LP	0,46	0,42	0,35	0,30	0,08	0,01	0,49	0,36	0,24	0,11	0,09	0,34	0,35	0,20	0,11	0,09
YP	0,31	0,44	0,58	0,50	0,28	0,09	0,39	0,47	0,48	0,17	0,11	0,36	0,41	0,44	0,13	0,11
QP	0,49	0,45	0,64	0,64	0,53	0,11	0,61	0,61	0,42	0,38	0,20	0,37	0,51	0,39	0,19	0,04
FP	0,52	0,51	0,96	0,73	0,59	0,06	0,64	0,63	0,59	0,34	0,12	0,49	0,60	0,63	0,25	0,08
BOP	0,54	0,76	0,78	0,79	0,21	0,03	0,54	0,75	0,87	0,1	0,11	0,5	0,60	0,68	0,20	0,07
Chlorophyll (a+b) (mq/q wet mass)																
LP	1,35	1,28	0,93	0,92	0,27	0,19	1,18	0,95	0,66	0,49	0,30	0,95	0,97	0,65	0,38	0,20
YP	0,96	1,25	1,41	1,24	0,77	0,28	1,13	1,26	1,27	0,58	0,33	1,13	1,25	1,25	0,37	0,31
QP	1,32	1,33	1,65	1,60	1,35	0,29	1,43	1,45	1,23	0,91	0,53	1,27	1,43	1,16	0,57	0,19
FP	1,39	1,41	2,67	2,21	1,55	0,15	1,63	1,70	1,58	0,83	0,40	1,53	1,71	1,58	0,65	0,26
BOP	1,52	1,72	2,01	2,12	0,87	0,28	1,53	1,80	1,83	0,71	0,32	1,55	1,73	1,63	0,61	0,34
Carotenoid (mq/q wet mass)																
LP	1,21	1,49	1,61	1,62	1,28	1,21	1,38	1,53	1,62	1,23	1,21	1,5	1,59	1,67	1,75	1,8
YP	1,28	1,5	1,62	1,68	1,44	1,3	1,39	1,6	1,69	1,31	1,28	1,55	1,68	1,75	1,89	1,9
QP	1,33	1,58	1,71	1,80	1,38	1,3	1,45	1,67	1,78	1,35	1,33	1,53	1,7	1,83	1,99	2,2
FP	1,46	1,65	1,84	1,97	1,51	1,46	1,68	1,87	1,99	1,52	1,47	1,64	1,82	1,95	2,01	2,1
BOP	1,58	1,7	1,91	2,1	1,93	1,85	1,72	1,89	2,03	1,67	1,59	1,72	1,87	2,2	2,23	2,2
Chlorophyll (a+b)/Carotenoid																
LP	1,11	0,65	0,64	0,44	0,15	0,02	1,74	0,56	0,35	0,24	0,08	0,63	0,61	0,39	0,22	0,10
YP	0,75	0,77	0,87	0,74	0,53	0,22	0,81	0,79	0,75	0,44	0,26	0,72	0,74	0,71	0,3	0,16
QP	0,99	0,84	0,96	0,89	0,85	0,25	0,99	0,93	0,75	0,75	0,32	0,83	0,85	0,63	0,34	0,12
FP	0,95	0,85	1,45	0,89	0,89	0,1	1,03	1,14	0,95	0,61	0,24	0,93	0,94	0,81	0,19	0,1
BOP	1,03	0,43	1,1	0,68	0,35	0,04	1,03	1,06	0,9	0,25	0,14	0,9	0,92	0,83	0,36	0,15

As can be seen from Table 1, the effect of $ZnSO_4$ salt on the dynamics of changes in the amount of photosynthetic pigments was more intense than the effect of other factors. It was established that the amount of chlorophyll a and chlorophyll b changes in a similar spectrum under the influence of radiation, chlorine and sulfate salts. Thus, up to the flowering phase (FF) of ontogenesis, the amount of photosynthetic pigments increased at low doses of these factors, and decreased at a radiation dose of 50-100 Gy and at salt concentrations of 50-100 mM. A sharp decrease in the amount of pigments occurred under the influence of 200 Gy radiation and 200 mM $FeCl_3$ and $ZnSO_4$ salts. The change in the amount of chlorophyll a under the influence of all 3 factors occurred with a similar trend in the phases of ontogenesis. It was established that, unlike chlorophyll a and b, the amount of carotenoids increases during the vegetation period, regardless of the plant development phases and the dose and concentration of stress factors. Such an increase in the amount of carotenoids can be explained by their protective function under stress conditions.

3.3. Study of the effect of radiation and salt stresses on gas exchange parameters in cotton leaves. It can be noted that the biometric indicators and productivity of higher plant organs are directly related to photosynthesis and its Pn indicator. Photosynthetic pigments play an important role in this process[15].

The values of gas exchange parameters were measured in the active developmental phases of ontogenesis under the influence of different doses of γ-rays, chlorine and sulfate salts with different chemical compositions. The results obtained during the conducted studies are given in Table 2.

As can be seen from the table, the energy metabolism of the plant is stimulated at a radiation dose of 50 Gy. As a result of increasing the concentration of $FeCl_3$ to 100 mM, sharp changes occur in the parameters of photosynthesis rate (P_n), stomatal permeability (g_s), CO_2 concentration in intercellular spaces (C_i) and transpiration rate (T_r) in the leaves of the plant.

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[15] Manikandan, A. Cotton response to differential salt stress / Sahu, D.K., Blaise, D., Shukla, P.K // International Journal of Agriculture Sciences -2019, 11(6), - p. 8059-8065

Table 2.

Effect of radiation, chlorine and sulfate salts on gas exchange parameters in active developmental phases of cotton plant ontogenesis

Op-tions	Control	Chlorine salts, 50 mM		Sulfate salts, 50 mM		Radiation 50 Qr
		NaCl	$FeCl_3$	Na_2SO_4	$ZnSO_4$	
Leaf phase (LP)						
P_n	39,3±0,74	39,8±4,1	41,3±0,74	45,8±4,83	46,7±4,22	49,9±1,09
C_i	229±19,6	226±21,5	231±17,6	243±20,4	249±14,7	264±21,7
g_s	5,3±0,78	6,5±1,11	4,1±0,68	6,1±1,00	5,1±0,77	4,01±1,01
T_r	6,0±0,89	2,99±0,97	6,1±0,69	5,9±1,12	6,55±0,83	6,02±1,43
Budding phase (BP)						
P_n	45,8±1,9	31,1±3,9	36,0±2,33	35,9±2,58	39,8±2,3	49,8±1,5
C_i	283±24,8	238±19,3	247±20,1	254±19,8	263±14,7	282±24,8
g_s	5,2±0,98	4,5±1,28	3,9±0,84	5,5±1,23	5,4±1,12	4,91±0,93
T_r	5,44±0,85	3,9±0,95	4,2±0,67	4,18±0,94	4,21±0,99	6,14±0,95
Flowering phase (FP)						
P_n	29,6±2,11	27,0±4,3	27,6±1,11	28,1±3,88	28,1±2,1	54,1±7,63
C_i	276±20,77	246±19,3	272±19,67	266±19,81	283±19,51	269±20,71
g_s	4,1±0,93	4,0±1,22	3,1±0,83	4,8±1,89	5,71±0,42	5,55±1,22
T_r	2,91±0,27	4,4±0,99	2,21±0,37	1,12±0,45	1,83±0,18	2,4±0,44

It was found that the rate of transpiration in green leaves is intensively regulated by the intensity of photosynthesis and stomatal permeability. Accordingly, as the rate of photosynthesis increases, the concentration of CO_2 in the intercellular spaces decreases. The intensity of photosynthesis decreased by 29, 38 and 62%, respectively, compared to the control at the stages of budding, flowering and opening of bolls.

3.4. Comparative analysis of the effect of radiation and salt stresses on the total amount of lipids and proteins in cotton leaves. It is known that due to the influence of radiation and salt stresses, as a result of the disruption of the activity of proteolytic, lipolytic and amylolytic enzymes in plants, serious changes occur in the biosynthesis and accumulation of high-energy organic substances in the cell, such as fats, proteins, etc. Taking into account the above, adaptive processes occurring in cotton plants under the influence of radiation doses and various types of salts were studied. The results obtained are given in Table 3.

Table 3.

Effect of radiation and salt stress on the amount of biopolymers in leaves during the early phases of ontogenesis of cotton plants

Conc.	Phases of the onthogenesis					
	LP		BP		FP	
	Protein	Lipid	Protein	Lipid	Protein	Lipid
Radiation, Qy						
C	10,5±1,4	8,9±1,3	16,7±4,4	11,7±2,8	15,5±2,4	18,3±2,9
5	12,3±1,5	15,3±3,3	18,3±3,2	18,3±2,5	19,3±2,6	23,3±4,2
10	15,1±1,7	17,3±2,9	19,1±3,8	22,1±3,0	20,0±3,1	26,1±3,8
50	18,0±2,1	20,4±3,1	21,8±3,8	28,4±3,6	21,2±2,9	30,9±7,6
100	17,5±2,0	19,9±2,9	22,0±2,9	27,9±4,0	21,9±3,5	19,4±1,4
200	14,2±1,9	15,0±2,2	19,9±3,9	23,3±3,8	21,1±3,2	9,1±1,6
$FeCl_3$, mM						
C	10,5±1,4	8,9±0,9	16,7±1,3	11,7±2,8	15,5±2,4	18,3±2,9
5	13,1±2,0	11,7±1,9	16,9±2,3	14,2±1,8	16,8±2,2	14,6±3,7
10	14,1±1,9	14,2±2,0	17,2±3,0	15,8±3,5	17,7±2,9	15,8±3,9
50	18,6±2,4	15,1±2,1	17,9±2,8	16,9±4,0	18,3±3,1	16,8±3,7
100	17,2±2,1	15,0±2,3	16,6±2,1	16,3±3,8	17,5±2,1	16,4±3,9
200	9,89±1,1	11,1±1,7	13,4±2,9	11,5±1,7	15,8±2,2	9,5±1,8
$ZnSO_4$, mM						
C	10,5±1,4	8,9±1,3	16,7±4,4	11,7±2,8	15,5±2,4	18,3±2,9
5	14,7±1,8	18,8±2,1	19,2±3,7	19,9±4,8	18,9±3,4	23,0±4,2
10	16,0±3,1	21,4±3,3	21,6±3,9	24,2±3,5	22,4±4,2	24,2±4,8
50	20,5±3,8	26,8±3,9	23,7±3,6	29,3±4,1	24,8±4,9	31,1±5,3
100	18,2±3,4	26,0±5,8	21,5±3,1	31,0±4,6	25,5±4,7	36,5±5,1
200	12,3±2,9	21,1±4,0	20,2±3,9	26,7±4,2	24,7±4,9	31,5±4,6

As can be seen from the table, during salt stress, the amount of total proteins in plants changes according to the degree of salinity of the environment. It was found that as salinity increases, the amount of soluble proteins also increases.

3.5. Study of the effect of radiation and salt stresses on the activity of adenine nucleotides and nicotine coenzymes in cotton leaves. To study the dynamics of changes in the amount of ATP and ADF in cotton leaves cultivated under irradiation and different salt concentrations, experiments were conducted on control, 10, 20 and 30-day-old plants. The effect of radiation and salt stress lasted for 10 days, and the study samples were taken from the same layer of leaves according to each stage. The stress environment was created with 50, 100 and 200 Gy doses of γ-irradiation, and salt stress was created with 50, 100 and 200 mM concentrations of NaCl, $FeCl_3$ and Na_2SO_4, $ZnSO_4$ salts. Quantitative analysis of nucleotides and nicotine coenzymes was carried out every 10 days. As can be seen from the table, the decrease in ATP was more noticeable during Na_2SO_4 stress. The effect of salt stress on the amount of ADF occurs in a similar trend to the effect on ATP. Signs adequate to this were also observed when determining the amount of $NADF^+$.

The amount of $NADF^+$ in leaf samples taken from the control increased from 58.5% to 67.8% within 20 days. Thus, the studies conducted showed that due to the effects of radiation and salt stress in the leaves of cotton plants, there was a decrease in the amount of ATP and the amount of nicotine coenzymes during the duration of the stress.

3.6. The effect of radiation and salt stress on the activity of catalase (CAT) and ascorbate peroxidase (APO) enzymes in cotton plants. The dynamics of changes in the activity of CAT and APO enzymes in the leaves of cotton plants depending on the type, level of stress factors and the phases of ontogenesis were studied.

For this purpose, a part of the cotton seeds was irradiated with γ-rays at doses of 5, 10, 50, 100, 200 Gy and sown, and the second part was sown separately in concentrations of NaCl, $FeCl_3$, Na_2SO_4 and $ZnSO_4$ salts at 5, 10, 50, 100 and 200 mM, and the activity of CAT and APO enzymes was studied at different stages of plant development (Figure 3 and Figure 4).

CAT activity

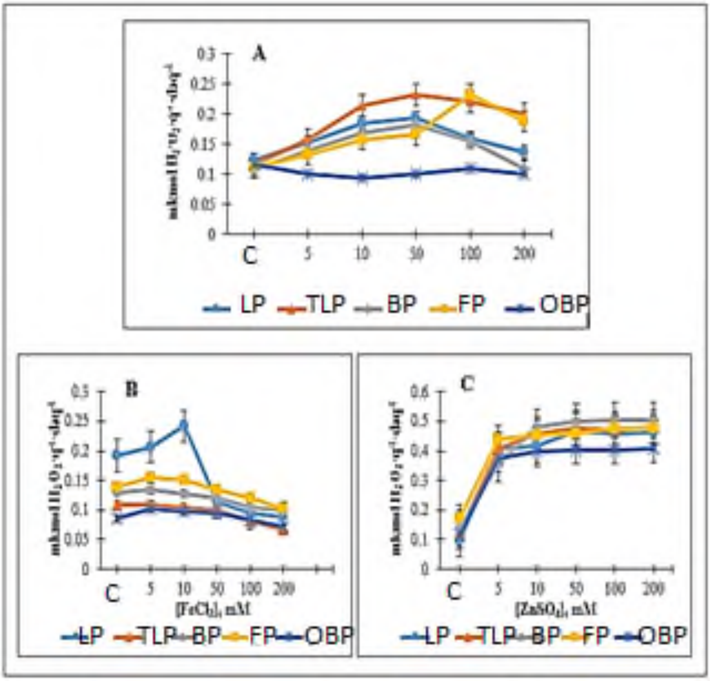

Figure 3. Effect of radiation, $FeCl_3$ and $ZnSO_4$ on catalase activity at different doses and concentrations during ontogenesis of cotton plant. LP-core leaf phase, TLP-true leaf formation phase, BP-budding phase, FP-flowering phase, OBP- opening boll phase. C-Control, A-radiation, B-$FeCl_3$, C-$ZnSO_4$

The activities of the enzymes were also studied based on the results obtained at the optimal concentrations of radiation and salts of 50 Gy and 50 mM. It was determined that the activities of the studied enzymes increase at doses of 5-50 Gy of γ-irradiation, while at higher doses of radiation (100, 200 Gy) the activity of the enzymes gradually decreases. In the presence of NaCl, $FeCl_3$, Na_2SO_4 and $ZnSO_4$ salts in the medium, an increase in the activities of CAT and APO enzymes was observed at low - 5, 10 mM concentrations of salts compared to the control variant.

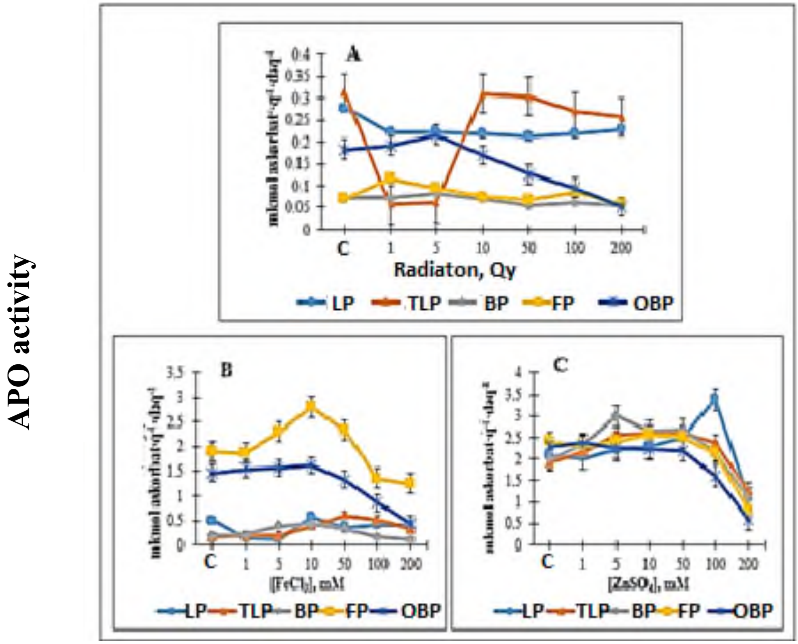

Figure 4. The effect of radiation, $FeCl_3$ and $ZnSO_4$ salts of different doses and concentrations on ascorbate peroxidase activity in the ontogenesis of cotton plants. LP-core leaf phase, TLP-true leaf formation phase, BP-budding phase, FP-flowering phase, OBP-opening boll phase. C-Control, A-radiation, B-$FeCl_3$, C-$ZnSO_4$

Inhibition of enzyme activities was shown at salt concentrations above 50 mM, 100 mM and 200 mM. Based on the results obtained, it can be said that the CAT enzyme significantly retains its activity at higher concentrations of $ZnSO_4$ salt compared to other salts (Figure 3).

3.7. The role of CA and NR-ase enzymes in the regulation of carbon and nitrogen metabolism in cotton plants under the influence of radiation and salt stresses. As is known, when plants are in normal growth and development, the metabolism in the cell, the energy supply of organs and tissues is at a high level. In the research work, the activity of carbonic anhydrase (CA) and nitrate reductase (NR-ase) enzymes was measured in the extract obtained from cotton plant leaves grown in different concentrations of salt (Figure 5).

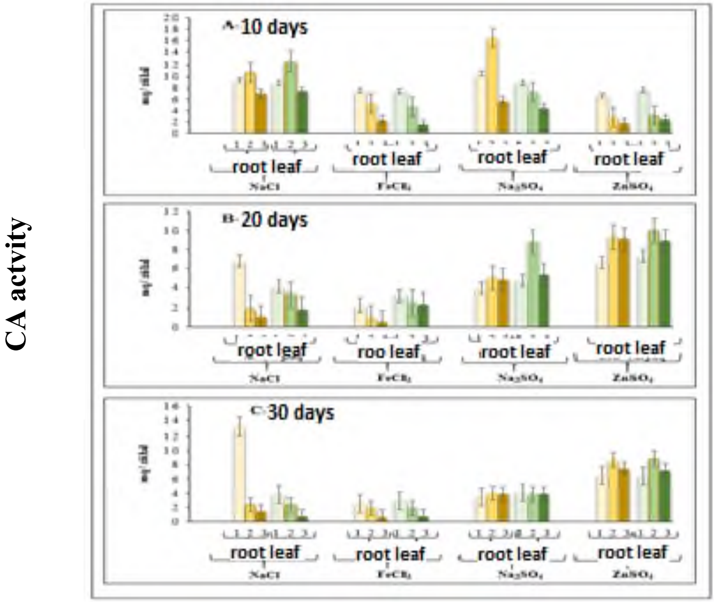

Figure 5. The effect of chloride and sulfate salts on the dynamics of changes in CA activity in cotton plant leaves.1-Control; 2-50 mM salt; 3-100 mM salt

Figure 5 shows the effect of chloride and sulfate salts on the dynamics of changes in the activity of the CA enzyme in the leaves and roots of cotton plants depending on time. As can be seen from the figure, the activity of CA gradually increases in the roots of 10, 20 and 30-day-old plants in the control variant, while on the contrary, it decreases in the leaves of the plant. On the 20th and 30th days of plant development, 50 and 100 mM NaCl concentration causes a decrease in the activity of CA in the roots and leaves. It was determined that under the influence of 50 - 100 mM $FeCl_3$ in 10, 20 and 30-day-old plants, CA activity in the roots and leaves decreases in parallel compared to the control variant. The CA enzyme reacts differently to the effects of salts with different chemical compositions and concentrations. As can be seen from the figure, carbonic anhydrase loses its activity more quickly under the influence of chlorine-containing salts. All this shows that NaCl and $FeCl_3$ have an inhibitory effect on carbonic anhydrase activity in roots and leaves at concentrations of 50 and 100 mM. Such an effect of chlorine salts on carboanhydraze activity in roots and leaves of cotton plants can be attributed to the decrease in the activity of H^+- pumps and the disruption of osmotic processes in cells as a result of the increase in the amount and concentration of salts in the rhizosphere, which is the environment around the roots. Unlike chloride salts, different results were obtained in carboanhydraze activity when sulfate-containing salts were applied to cotton plants. It was found that sulfate salts had a more stimulating effect on the growth and development of cotton plants at a concentration of 50 mM. Of the sulfate salts, CA activity increased depending on time at concentrations of 50 and 100 mM of Na_2SO_4 and $ZnSO_4$ salts, and had the highest value in 20-day-old plants.

The value of CA activity in the roots and leaves of 30-day-old experimental plants in the control and experimental variants was almost the same (Figure 5). However, it should be noted that the activity of the CA enzyme is highly dependent on the concentration of Zn^{2+} ions. Studies have shown that as the concentration of $ZnSO_4$ salt in the medium increases to 50 and 100 mM, the activity of carboanhydraze also increases. Further increase in $ZnSO_4$ leads to a gradual decrease in the activity of the enzyme. As is known, orthophenanthroline (OFT) has the property of forming a complex with heavy metal atoms. It forms a complex with Zn atoms in the active center of carboanhydraze and in the environment, precipitating them, and as a result, the activity of the carboanhydraze enzyme is completely inhibited. According to the literature, the carboanhydraze enzyme, which is localized in various organelles of cells and tissues of C3 plants and performs various physiological and biochemical functions, has an oligomeric structure and each monomer contains a

Zn atom. In order to clarify the role of zinc ion (Zn^{2+}) in the activity of the carboanhydraze enzyme localized in the leaf cells of cotton plants, the effect of orthophenanthroline, which forms a complex with heavy metal atoms, on the activity of carbonic anhydrase was studied (Figure 6). It was found that the inhibition of the enzyme by orthophenanthroline depends strictly on the concentration of the complex with metals and the pH of the reaction medium. Thus, at alkaline pH, orthophenanthroline has a very weak effect on the activity of carbonic anhydrase. The results obtained are shown in Figure 6.

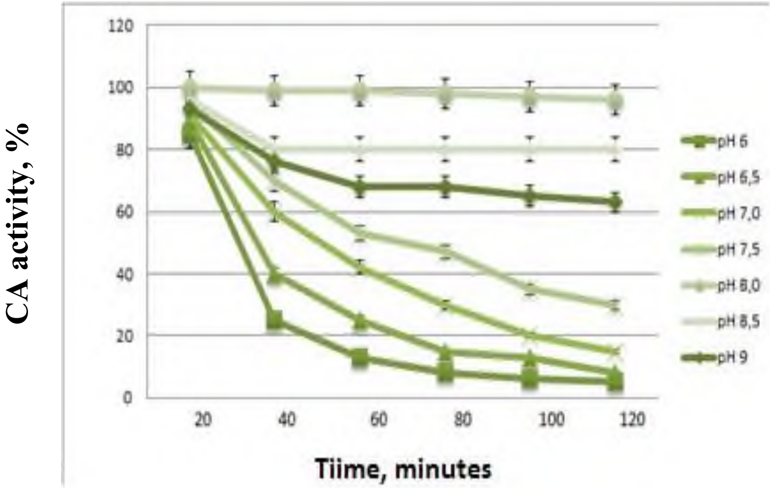

Figure 6. Effect of $ZnSO_4$ on the activity of orthophenanthroline carbonic anhydrase enzyme in leaves at the flowering phase (FP) of ontogenesis of cotton plants cultivated at different concentrations.

Currently, the study of carbon exchange enzymes, along with nitrogen exchange enzymes, is of great importance in increasing the resistance of plants to stress factors. Figure 7 shows the dynamics of the change in the activity of the nitrate reductase enzyme in the leaves of cultivated cotton plants at different doses of γ-irradiation (N, 5, 10, 50, 100 and 200 Gy) and different concentrations of salts (NaCl, $FeCl_3$, Na_2SO_4 and $ZnSO_4$) (N, 5, 10, 50, 100 and 200 mM) depending on the stages of ontogenesis. If we look at Figure 7, we can see that nitrate reductase activity increases at all stages of ontogenesis up to a dose of 50 Gy, while at subsequent doses of irradiation (100 and 200 Gy) the activity of the enzyme gradually decreases. At this time, the greatest decrease occurs in the core leaf phase (LF) and bal opening (OBP) phases.

At the same time, Figure 7 shows the results obtained during the influence of different concentrations of NaCl, $FeCl_3$, Na_2SO_4 and $ZnSO_4$ salts on nitrate reductase activity. As can be seen from the figures, at 5-50 mM salt concentrations, the nitrate reductase enzyme shows the highest activity compared to the control variant. It was determined that the NR-ase enzyme showed high activity in each variant during the budding (BP) and flowering (FP) phases

These processes increase with increasing plant age in flowering phase. After this phase, it remains relatively unchanged for some time, then increases slightly in FP, and then weakens as the vegetation approaches the end.

The processes occurring in the metabolism of cotton plants under the influence of radiation and various types of salts: interrelated changes in gas exchange parameters, mineral nutrition of plants, and the activity and functional diversity of carbonic anhydrase enzyme isoforms localized in roots and leaves can be considered as one of the components of the physiological and biochemical mechanisms of plant adaptation to stress. Sulfate salts have a more stimulating effect on the activity of carboanhydraze enzyme compared to chloride salts.

The active and stimulating effect of $ZnSO_4$ can be explained by the presence of Zn^{2+} ion in its composition. Since Zn^{2+} ion is a coenzyme of the carboanhydraze enzyme, the carbonic anhydrase enzyme localized in root and leaf cells becomes even more active in the presence of this ion, ensuring a high rate of photosynthesis. The decrease in carbonic anhydrase activity at low concentrations of $ZnSO_4$ leads to a weakening of CO_2 diffusion.

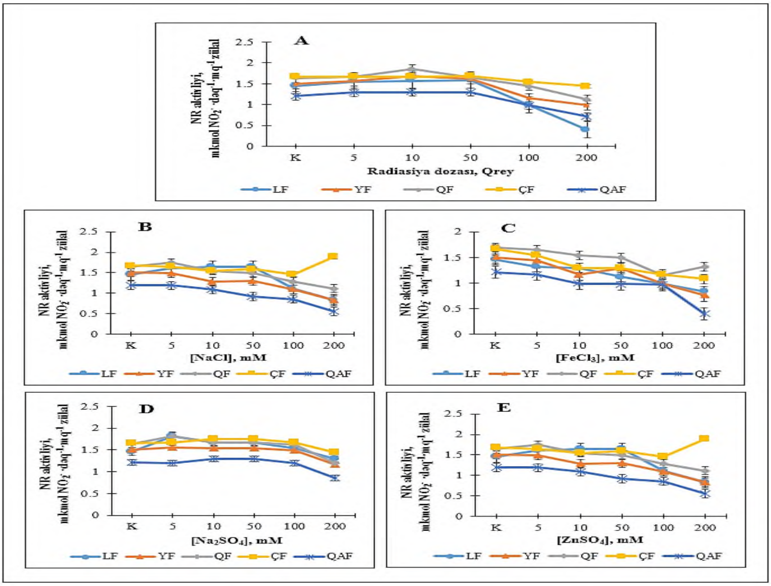

Figure 8. The effect of irradiation, chlorine and sulfate salts on the dynamics of changes in the activity of the NR-ase enzyme during the ontogenesis of cotton plants. A-irradiation, B-$FeCl_3$, C-NaCl, D-Na_2SO_4, E-$ZnSO_4$

3.8. Economic efficiency of cotton plants grown from irradiated seeds. Economic efficiency reflects the degree of realization of economic interests and is measured by a system of cost indicators characterizing the efficiency of production and sale of products.

In accordance with the agrotechnical measures carried out during the ontogenesis of the plant in field conditions, the economic efficiency of planting Ganja-182 cotton variety was compiled on the basis of technological maps. An average of 1140 manats was spent on the cultivation and harvesting of one hectare of cotton field, and the selling price of 1 kg of raw cotton was calculated based on the average yield of 0.70 AZN. The obtained price indicators are given in table 4.

Table 4

Effect of cotton planted with irradiated seeds on economic efficiency

Options		Productivity (cen/he)	Total product value (azn)	Production costs (azn)	Net income (azn)	Profitability level (%)
Sort	Radiation (Qy)					
Control		40	2800	1140	1660	145,6
Ganja -182	5	43.8	3066	1140	1926	168.9
	10	44.9	3143	1140	2003	175,7
	50	46	3220	1140	2080	182,4
	100	23	1610	1140	470	41,2
	200	20	1400	1140	260	22,8

As can be seen from the table, irradiating seeds with 10–50 Gy has a stimulating effect on them. The yield per hectare, profitability level, net income and other economic indicators of cotton plants grown from seeds irradiated with γ-irradiation are higher than those of the control variant. In this regard, pre-irradiation treatment of cotton seeds with small doses (up to 50 Gy) before sowing in farms will allow increasing productivity.

RESULTS

1.During the exposure of radioactive radiation and various salts to cotton seeds, it was determined that the highest germination percentage due to stress occurs at doses of 5-50 Gy of radiation, and at concentrations of 5-50 mM of $FeCl_3$ and $ZnSO_4$ salts. When seeds are irradiated at a dose of 50 Gy and then transferred to different concentrations of salt, that is, under the influence of stress 2, the germination percentage of seeds decreases sharply. It has been shown that the amount of pigments increases at low doses and concentrations of radiation and salt until the flowering phase of ontogenesis, while at higher doses and concentrations (200 Gy and 200 mM) there is a sharp decrease in the amount of pigments. Unlike chlorophyll a and chlorophyll b, the amount of carotenoids increases throughout the vegetation period, regardless of the phase of plant development and the effects of stress factors [2,4,7,8,14,16].

2. The values of gas exchange parameters were measured in the active developmental phases of ontogenesis in the presence of different doses of γ-irradiation and chemical salts of different composition. Under the influence of 100 mM $FeCl_3$ salt, sharp changes occur in the parameters of photosynthesis rate (P_n), stomatal permeability (C_s), CO_2 concentration in intercellular spaces (C_i) and transpiration rate (T_r) in the leaves of the plant. The photosynthesis rate (P_n) decreased by 29%, 38% and 62%, respectively, compared to the control variant at the stages of budding, flowering and opening of bolls [1].

3. During the study of the effect of radiation and salt stress on the amount of biopolymers in the leaves of cotton plants, it was determined that, regardless of the phase of ontogenesis, the amount of fats and proteins increases in parallel under the influence of stress. It has been shown that the amount of total protein in plants during salt stress changes according to the degree of salinity of the environment and that the amount of soluble proteins increases as salinity increases [9,13,15,27].

4. The dynamics of changes in the amount of adenine nucleotides (ATF, ADF) and nicotine coenzymes (NAD^+, $NADF^+$) in cotton leaves during γ-irradiation and salt stress were studied as a function of time. It was shown that in the control sample, the amount of ATP increased from 52 µg to 61 µg within 30 days. While this indicator was 37 µg on the 10th day of development, the amount of ADF reached 44 µg on the 30th day. Due to the effects of irradiation and high doses and concentrations of salts, the amount of ATP and ADF decreased [20,21].

5. It was found that the activity of CAT and APO enzymes increases at doses of 5-50 Gy of γ-irradiation, while at higher doses (100, 200 Gy) the activity of the enzymes gradually decreases. At low concentrations of Na_2SO_4, $ZnSO_4$, NaCl and $FeCl_3$ salts (5-50 mM), the activity of the enzymes increases compared to the control variant,

while at higher concentrations of salt (100, 200 mM), the activity of the enzymes is inhibited. Compared to other salts, the CAT enzyme significantly retains its activity under the influence of $ZnSO_4$ salt [5,10,11].

6. The dynamics of the change in the activity of the CA enzyme in the leaves of the cotton plant depending on time was measured under salt stress conditions. It was found that at concentrations of NaCl and $FeCl_3$ at 50-100 mM, the activity of the CA enzyme in the roots and leaves decreases compared to the control variant. Unlike chloride salts, sulfate salts (Na_2SO_4 and $ZnSO_4$) have a stimulating effect on the activity of KA, and at concentrations of 50-100 mM, the activity of the enzyme increased in a time-dependent manner and reached its highest value in 20-day-old plants. The activity of the CA enzyme was found to be highly dependent on the concentration of Zn^{2+} ions in the environment [6,19,25,26].

7. The dynamics of changes in NR-ase activity in cotton plant leaves under conditions of γ-irradiation and different salt concentrations was studied during ontogenesis. It was found that the activity of the enzyme increases at all stages of ontogenesis up to a dose of 50 Gy, while at high doses of irradiation (100 and 200 Gy) the activity of the enzyme gradually decreases. When examining the effect of salts on NR-ase activity, it was shown that at concentrations of 5-50 mM, the NR-ase enzyme exhibits high activity, and the highest activity of the enzyme is observed in the BP and FP of ontogenesis [9,18].

PRODUCTION RECOMMENDATIONS

1. Treatment of seeds with γ-irradiation doses of 5-50 Gy has a stimulating effect on them, and the yield, profitability level, net income and other economic indicators of cotton plants grown from such seeds were higher than the control variant. Treatment of cotton seeds with small doses (up to 50 Gy) of radiation before sowing on farms allows increasing productivity.

2. The results of physiological adaptation to the effects of stress factors in plants can be used to determine the limiting limits of radiation and salt stress and stimulating doses, and it is recommended to use these results in selection as initial material for the creation of new salt-resistant cotton varieties. The results of the dissertation are applied in private farming (reference is attached).

List of narrowly printed scientific publications on the topic of the dissertation

1. Алакбарова, Ш.Э. Изучение влияния высокой концентрации $FeCl_3$ на параметры газообмена в листьях хлопчатника сорта Гянджа-182 // Материалы Международной научной конференции «Становление и развитие экспериментальной биологии в Таджикистане» - Душанбе: - 2022. - с. 23-26.
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