

Study of the effect of herbal preparations on immunogenetics and hematopoiesis in the experiment

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Abstract. When creating an experimental radiation sickness in animals, the development of deep immunodeficiency is observed, which manifests itself in the inhibition of the production of antibodies to the thymus-dependent antigen EB. Introduction to animals with radiation sickness Ginger, Detoxioma, and Gulzar balm help to increase the immune mechanisms of antibody production. Plant preparations can increase thymus nucleated cells in irradiated mice. Ginger, Detoxioma, and Gulzar balm significantly increase the level of erythrocytes and leukocytes in the blood of irradiated animals.

1 Introduction

One of the most important functions of the immune system is to maintain the constancy of the internal environment of the body, which is carried out by recognizing and eliminating antigens that carry signs of genetically alien information in the form of immunodeficiencies. Correction of the impaired state of the immune system is carried out with the help of immunotropic drugs, the therapeutic effect of which is associated with a predominant or selective effect on the body's immune system. Individual immunomodulators can selectively affect the corresponding link of the immune system, but the final effect is multifaceted, since the functional immune system [1-6].

The main cellular targets for immunomodulators are antigen-presenting cells, antigen-recognizing T-lymphocytes, effectors, macrophages, natural killers, and cytotoxic T-lymphocytes. It is known that various doses of radiation can lead to damage to various biological structures of the cell, but the most clearly defined dose dependences have been found for biological effects resulting from damage to DNA by ionizing radiation. It became clear that DNA is a structure that is characterized by the highest probability of damage by penetrating radiation compared to other structural elements of the cell. Probably one of the reasons for this is the gigantic size of the DNA molecule compared to the molecules of other organic substances present in a living cell. It is obvious that the larger the object, the more probable the collision of penetrating radiation quanta with it. Therefore, the theory of "targets" and the principle of "hit", proposed by N.V. Timofeev-Resovsky, To stimulate the immune system, herbal preparations are widely used, in particular, various derivatives of

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Echinacea purpurea, registered as immunomodulators (immunal, *echinacea*) with immunomodulatory, antiseptic, antiviral, antibacterial and antioxidant properties. Essential oils of carrot, calendula, rosehip, and sea buckthorn: stimulate phagocytosis, the activity of natural killers, and cytotoxic T-lymphocytes oils of lavender, sage, thyme, thawed, and lemon [7].

Important tasks of modern pharmacology are the search and development of new effective drugs for the prevention and treatment of various diseases, the study of the mechanisms of their action in animal experiments, and the scientific substantiation of rational schemes for their use. To solve these problems, a search for new biologically active pharmacological substances is carried out among natural and synthetic compounds, products of biotechnology, and genetic engineering, their effectiveness and safety are assessed on experimental models of pathological conditions, and individual sensitivity to existing and developed drugs is studied [3-6, 8].

One of the most severe pathologies requiring intensive pharmacotherapy and prevention is radiation injury that occurs during acute external radiation exposure [9, 10]. The occurrence of such injuries is possible in case of accidents and catastrophes at nuclear power facilities, in medical institutions using sources of ionizing radiation for diagnostic or therapeutic purposes, and in the case of sabotage or terrorist use of radionuclides [11-16]. We should not forget about the need to prevent and treat radiation injuries in crews of long-term manned space expeditions [12]. Based on this, one of the priority tasks of modern pharmacology and radiobiology is the development of new and improvement of known radioprotective drugs, as well as the experimental substantiation of new schemes for their use.

Currently, ecdysteroid-containing plants, which do not have side effects from plant sources, are of great interest as plant raw materials for obtaining new immunostimulating drugs to expand the arsenal of existing drugs [13, 14].

Recent advances in clinical disciplines and, in particular, immunology show that the pathogenesis of many diseases is to some extent related to the functioning of the human immune system. Modern studies are increasingly showing that various environmental factors lead to the inevitable disruption of the functioning of the immune system and, as a result, a change in the immune status of the body. This is due to the fact that the immune system is very vulnerable when exposed to damaging environmental factors and is the main target of a significant number of xenobiotics. Despite the great success in the creation of chemical medicines, interest remains in herbal remedies and their active component with immunotropic activity, including for the treatment of chronic and long-term diseases. Recently, rapidly developing technologies, research in medicine and pharmacology confirms the presence of phytopreparations with unique properties that affect the body in a complex manner with low toxicity and high efficiency, which allows them to be used not only for treatment, but also for the prevention of diseases.

According to WHO (2019), about 130 countries in the world have official programs involving traditional medicine for the treatment of diseases. The study of substances used for medicinal purposes in traditional medicine of various ethnic or cultural groups makes a significant contribution to the discovery and development of modern methods of treatment. Herbal immunomodulators serve as an alternative therapy for various diseases, especially in cases of a weakened immune response and when discriminatory immunosuppression occurs, for example, in the case of autoimmune syndromes. In light of recent events, the use of herbal immunomodulators, including for the treatment of patients with COVID-19, both the plants themselves, such as betel and turmeric leaves, and the biologically active substances contained in them.

In the Republic of Uzbekistan, natural and synthetic immunomodulators have begun to be widely used for the treatment of secondary immunodeficiency states. Several herbal

medicines used throughout the world are well known for their anti-infective effects, not only by direct action on the pathogen, but also by stimulating the host's natural defense mechanisms.

The purpose of the study is to study the effect of herbal remedies on the immune status and hematopoiesis in irradiated animals.

Object and methods of research: In the experiments, outbred mice of 2-3 months of age, weighing 18-20 grams, were used. Irradiation was induced totally at a sub lethal dose of 5.0 Gray on a RUM-17 device. Filters AL-0.5 mm, Cu 0.5 mm, current strength 10 mA, power 180 kV. During irradiation, mice were placed in special boxes. On the 8th day, they were intraperitoneally immunized once with ram erythrocytes (EB) at a dose of 2×10^8 /ml. Before immunization, EB was washed twice in medium No. 199, in a centrifuge at 1000 rpm for 10 minutes. Then, the number of erythrocytes was counted in the Goryaev's chamber. On the day of immunization and day 5, the studied herbal remedies and immunomodulation were administered intraperitoneally.

Herbal remedies:

1. Ginger. Ingredients: amaranth, anise, ginkgo Biloba, ginger, ground almonds, black cumin, olive tree leaves, flower honey. Ginger was administered intraperitoneally at a dose of 0.005 ml/mouse.

2. Detoxyoma. Ingredients: papaya, guava, pomegranate, olive, goji, bitter watermelon. Detoxyoma was administered at a dose of 0.005 ml/mouse.

3. Gulzar balm. Ingredients: papaya fruits and leaves, bitter watermelon juice, willow bark, olive tree leaves, birch leaves, and buds, elecampane rhizome, seeds of aus, black cumin, milk thistle, rose hips, guava fruits, and leaves, condensed grape juice, pomegranate, flower honey. Gulzar balm was administered intraperitoneally at a dose of 0.005 ml/mouse.

4. Immunomodulin. Developed under the guidance of F.Yu. Garib et al. (patent No. 1758., 1990, Tashkent). at a dose of 0.0002 ml/mouse.

On the 6th day after immunization, antibody-forming cells (AFC) in the spleens were determined by the direct method of local hemolysis according to Jerne NK and Nordin AA [15]. dishes (to cultivate tissues with a diameter of 40 mm, 1.0 ml of agarose solution, 0.03 ml of 20% EB solution, and 0.1 ml of spleen cell suspension were poured. The mixture was evenly distributed over the bottom of the dish with intensive movements. After that, the dishes were placed on a thermostat at + 37°C for 1.5 hours. Then, 1 ml of guinea pig complement, diluted in medium No. 199 at a ratio of 1:5, was added to the dish. this, the number of AFCs per 1 million spleen nucleated cells (NSCs) (relative indicator) was determined. For this, the number of AFCs for the entire organ was divided by the total cellularity of the spleen. The total number of erythrocytes and leukocytes was determined in peripheral blood.

Animals were divided into 5 groups of 6 heads. The first group is intact (healthy animals). The second group - is irradiation (control). The third group - irradiation + Balm Gulzar. The fourth group is irradiation + Ginger + Detoxioma + Gulzar Balm. The fifth group - irradiation + immunomodulation.

2 Results and discussion

As can be seen from Table 1, on the 6th day after immunization, 8341.7 ± 36.8 antibody-forming cells are formed in the spleen in the control group. In animals that received irradiation, antibody genesis in the spleen significantly decreased by 5.7 times, which indicates the development of a secondary immunodeficiency state. When calculating the number of antibody-forming cells per 1 million splenocytes, it decreased by 4.4 times and spleen nucleated cells (NSCs) decreased by 1.3 times compared to the control group.

Table 1. Indicators of immunogenetics in irradiated animals.

Experiment steel groups (n=6)	A drug	Number of JSCS x106	IP	Number of AOK per			
				spleen	IP	106 spleen cells	IP
Intact	-	616.8±15.8		8341.7±36.8		13.6±0.4	
Control (Irradiation)	-	467.8±12.8a	-1.3	1460.0±24.5a	-5.7	3.1±0.1	-4.4
Irradiation	Balm Gulzar	600.3±18.5b	+1.3	3291.7±27.1b	+2.3	5.5±0.2b	+1.8
Irradiation	Ginger + Detoxyoma + Gulzar Balm	502.6±17.9	+1.1	4760.0±18.7b	+3.3	9.5±0.4b	+2.9
Irradiation	Immunomodulation	492.6±23.3	+1.1	3580.0±37.4b	+2.5	7.3±0.4b	+2.4

Note: NSC-nucleated spleen cells, IS-ratio index, a-significant compared with intact animals, b-significant compared with the 2nd group, (n=6)-number of animals in the group.

Administration of Balsam Gulzar to irradiated animals significantly increased the immune response to ram erythrocytes by 2.3 times. 7. In mice treated with immunomodulation, the number of AFC per spleen significantly increased by 2.5 times compared to the control - 3580.0 ± 37.4 . Therefore, all drugs can significantly increase the number of AFC in the spleen (absolute indicator).

When calculating the AFC per 1 million spleen cells, it was found that this indicator in the intact group was 13.6 ± 0.4 , under the action of X-rays this indicator significantly decreased by 4.4 times, and in animals receiving Balsam Gulzar, it significantly increased by 1.8 times. Similar results were obtained for Ginger + Detoxyoma + Gulzar Balm and immunomodulation in irradiated animals.

Thus, when calculating AFC for the entire spleen (absolute indicator), and for 1 million spleen cells (relative index) studied herbal remedies can increase immunological parameters in irradiated animals.

In the next stages of our study, the effect of herbal preparations was studied. Balsam Gulzar, complex preparations (Ginger + Detoxyoma + Balsam Gulzar) for hematopoiesis. Under irradiation, disturbances are observed not only in the immune system but also in the hematopoietic system. irradiation $7.6 \pm 0.1 \times 10^9/\text{ml}$ (Table 2).

Introduction to irradiated animals Gulzar Balm, a complex of preparations (Ginger + Detoxyoma + Gulzar Balm) increases the number of erythrocytes by 1.2 times.

When irradiated, leukopenia develops. Thus, if in intact animals the number of leukocytes is $12.9 \pm 0.2 \times 10^6 / \text{ml}$, then in irradiated mice it significantly decreases by 2.3 times. times respectively.

The results obtained indicate the ability of the studied herbal preparations to correct disorders in the immune status and hematopoietic system in irradiated animals (Table 2).

Table 2. Indicators of hematopoiesis in irradiated animals.

experimental groups (n=6)	A drug	red blood cells x 10 ⁹ /ml	IP	Leukocytes x 10 ⁶ /ml	IP
Intact	-	8.6±0.1		12.9±0.2	
Control (Irradiation)	-	7.6±0.1a	-1.1	5.7±0.3a	-2.3
Irradiation	Balm Gulzar	8.8±0.1b	+1.2	8.9±0.3b	+2.3
Irradiation	Ginger + Detoxioma + Gulzar Balm	9.0±0.1b	+1.2	8.9±0.2b	+1.6
Irradiation	Immunomodulation	8.6±0.3b	+1.1	7.0±0.3b	1.2

Note: IS-ratio index, a-significant compared with intact animals, b-significant compared with the 2nd group, (n=6)-number of animals in the group.

3 Conclusion

1. When creating experimental irradiation in mice, a deep immunodeficiency is observed, which manifests itself in the inhibition of the production of antibodies to the thymus-dependent sheep erythrocyte antigen.

2. Introduction to irradiated animals Gulzar Balm, a complex of preparations (Ginger + Detoxyoma + Gulzar Balm) helps to restore the immune mechanisms of antibody formation.

3. Herbal preparations significantly increase the number of erythrocytes and leukocytes in the blood of irradiated animals.

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