

Study of the Effect of Pomegranate Oil on the Immunological State in Experimental Animals

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Annotation: The article presents morphometric parameters of the histostructure of the spleen of Wistar rats of both sexes of mature age in normal, after a single total irradiation in a sublethal dose, as well as after correction of these conditions with pomegranate oil. The data obtained by us indicate that total irradiation leads to a significant suppression of the white pulp of the spleen, the condition of which normalizes after 30 days. At the same time, the introduction of pomegranate oil leads to a significant acceleration of repair processes and restoration of organ functions. This suggests the possibility of using pomegranate oil in immunodeficiency conditions.

Keywords: pomegranate oil, spleen, irradiation.

Immunodeficiency conditions are attracting increasing attention from researchers and clinicians. An active search for new immunotropic drugs is underway [1-4]. A possible biological source of such a biological product can be considered pomegranate oil. The presence of a large number of biologically active substances in it, such as immunoglobulins, interferons, hormones, suggest an active effect of pomegranate oil on the recipient's immune system. The absence of specific markers makes it possible to administer pomegranate oil to any animal species, including humans, without the risk of causing severe allergic reactions or rejection [5]. At the Department of Normal Human Anatomy of the Abu Ali Ibn Sina Medical Institute in Bukhara, a number of studies are being conducted on the effect of pomegranate oil on the immune organs of rats. Thus, the peculiarities of the effect of pomegranate oil on the red bone marrow, thymus, mesenteric lymph nodes of rats were clarified [6-8]. The purpose of this work is to determine the features of the parenchymal component of the spleen of sexually mature rats after the introduction of pomegranate oil in normal and immunodeficient condition.

Material and methods: The experiment was conducted on Wistar rats of both sexes aged 10 months. The animals were divided into control and experimental groups. The control group consisted of rats receiving saline at a dose of 2 ml/kg of live weight. Experimental animals were injected with pomegranate oil in the same dosage 3 and 10 times every third day. A separate experimental group consisted of rats that were subjected to a single total irradiation at a dose of 5 Gy in order to create a model of immunodeficiency. The irradiation was carried out at the Teratron installation. Half of them received liquor after irradiation three- and tenfold. A day after the last injection, the animals were decapitated under ether anesthesia for organ sampling. The spleen was measured, weighed and fixed in a ten percent formalin solution for further studies. After the manufacture of paraffin blocks, transverse thin sections were obtained at the level of the organ gate,

stained with hematoxylin-eosin to measure the morphological parameters of the white pulp. Statistical processing of the obtained results was carried out in the licensed Statistica 6.0 program using the nonparametric Mann-Whitney U-test at $p < 0.01$.

Results. In mature rats, after three injections of liquor, the proportion of stroma elements was 2.1% of the cut area, the red pulp occupied 60.1%, the proportion of white pulp was 35.5%. The capsule of the organ is dense, smooth. The red pulp of the spleen has normal blood filling. A slight increase in the number of primary lymph nodes (LU) was revealed in comparison with the control. At the same time, LU containing germinative centers significantly outnumber those in the control group by 36.1%. Their diameter also increased by 27.9%. This was due to an increase in the marginal zone of the LU. Thus, its relative area in the control group was 44.1% of the total area of the LU, in the experimental group this value increased to 51.6%.

Tenfold administration of oil led to an increase in the proportion of white pulp on the cross sections of the spleen by 31.2% and a decrease in the proportion of red pulp by 17.1%, while the proportion of stromal elements did not change. The number of primary nodules accounted for a third of the total, but the size of the secondary nodes and their zones increased significantly, this is especially pronounced in the marginal zone.

Thus, the LU area increased by 91.1% relative to the control group, and their diameter – by 31.1%. The area of the marginal zone exceeded the control values by 124%, and its relative area also increased. At the same time, the relative areas of the remaining zones of the LU significantly decreased. A single total irradiation of rats in a sublethal dose of 5 Gy led to significant changes in the spleen, which primarily affected its pulp. The red pulp is anemic and amounted to 78.9% of the cut area. The stroma elements accounted for 4.1%, due to the proliferation of trabeculae and vessel walls. The white pulp occupied only 17.5% and was represented by periarterial lymphatic vaginas and small LU. Their area significantly decreased by 40.9%, and their diameter by 47.9%. The marginal sine is weakly expressed. The marginal zone is devastated. The periarterial zone is also often characterized by a low density of cells. The vast majority of LU does not contain germinative centers (GC).

After 30 days, the morphometric parameters of the rat spleen pulp began to approach the control values. The area of lymph nodes increased to 211093.7 ± 3244.2 mm², their diameter was 579.1 ± 12.1 mm. These values have no significant differences from the control. Most of the nodules have germinative centers. All nodule zones are well defined, including the marginal one. Its relative area was $47.3 \pm 2.4\%$, which practically does not differ from the control values. When correcting irradiation with injections of pomegranate oil three- and ten-fold, we observed the following changes. After three injections of oil to irradiated rats, the proportion of red pulp decreased slightly (to 74%) due to an increase in white pulp (to 21%). The red pulp is still anemic. There was abundant branching of arterioles in the LU and in the red pulp of the spleen. The LU area increased by 73.8% and the diameter by 112.2% compared to irradiated rats that did not receive CSF. The marginal zone and marginal sinus are still difficult to determine in the LU, but the periarterial zone and HZ, which are present in most LU, are clearly visible. The area of the HZ was 11085.1 ± 870.8 microns, and the diameter was 149.0 ± 6.8 microns, which is twice as large as those in the control group.

After tenfold administration of oil to irradiated rats, the ratio of the areas of red and white pulp was equal to those in the first experimental group on day 30. The red pulp had normal blood filling. The number of LU increased, while more than half of them contained germinative centers. The LU area and their diameter had no significant differences from the control values.

It is widely known that ionizing radiation has a detrimental effect on living cells, especially those in the stage of proliferation. For this reason, there is a total emptying of the red bone marrow, followed by peripheral organs of immunogenesis, such as lymph nodes and spleen. We observed depletion of the red pulp and depletion of the lymphoid component of the spleen, which does not contradict the literature data [9-11]. In

general, by the 30th day of the experiment, the morphofunctional state of the spleen was approaching normal. However, the introduction of liquor significantly accelerated this process. So, after three injections, there was an increase in the number of LU, an increase in their size, in most cases the nodules contained HZ. At the same time, if the numerical parameters of the size of the LU were normal and after correction of oil irradiation did not statistically differ from each other, then morphologically the condition of the white pulp of the spleen in these groups was significantly different. If, normally, the diameter of the LU and its area are largely determined by the width of the marginal zone, then after irradiation, a significant proportion of the area falls on the germinative center. In the first case, we observe an active white pulp, in the second case, the process of restoring the potencies of the organ is clearly expressed.

Thus, the data obtained by us indicate that total irradiation leads to significant changes in the structure of the rat spleen. On the seventh day of the experiment, there is a decrease in the percentage of white pulp of the spleen, a decrease in the number and size of moat of lymph nodes. The normal functional state of the spleen is restored by 30 days. At the same time, the introduction of pomegranate oil leads to a significant acceleration of repair processes and restoration of organ functions. This suggests the possibility of using pomegranate oil in immunodeficiency conditions.

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