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# Changes in the Interepithelial Lymphocytes of the Mucous Membrane of the Small Intestine when Exposed to Herbicides

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**Abstract:** The article analyzes the quantitative composition of lymphocytes of the epithelial cover of the mucous membrane of the small intestine in normal and under the influence of the herbicide kotorana. In the experiment, when exposed to cotoran, an increase in the number of large and medium lymphocytes is noticeable, and by the age of 3 months, the number of large, medium and small lymphocytes approaches the indicators of the control group.

**Keywords:** Peyer's plaques, cotoran, lymphocytes, germinative center.

**Introduction.** Among the immune formations of the digestive system, lymphoid nodules (Peyer's plaques) of the small intestine play an exceptional role. They, like the thymus, amygdala, appendix of mammals, belong to the lymphoepithelial organs in which lymphopoiesis takes place and are in close interaction with the reticular tissue, the epithelium. [1,17,18]

The primary element of the immune defense of the digestive tract is the lymphoid tissue of the intestine, which makes up a quarter of the entire mass of the immune system, which includes lymphoid nodules of the small intestine [6,7].The importance of lymphoid organs of the small intestine increases with biopsy, diagnosis, transplantation, cytodiagnostics, lymphoid tissue hyperplasia, enterosorption, endoscopy and other medical interventions in the field of ga- stroenterology[3,4].

The digestive system occupies an important place in the relationship of the body with the external environment. The mucous membrane of the digestive organs is affected by a wide variety of substances that make up food, and it becomes clear that it is not by chance that the mucous membrane and the submucosal base have their own lymphoid formations, which are organs of immunogenesis [9, 10]. On the one hand, the mucous membrane of the digestive system is a barrier structure that prevents the penetration of various agents of the external world into the body, and on the other hand, it participates in metabolic processes between the external and internal environment of the body [5,11].

**Materials and methods.** To achieve the goal and solve the tasks set, we conducted a study on 120 sexually mature mongrel white rats.

Experimental animals of all age periods were divided into the following groups:

I - intact (control), mothers (female rats) of which were injected intragastrically through a probe with 1 ml of distilled water once a day for 20 days after the birth of baby rats;

II - animals whose mothers (female rats) were injected intragastrically with cotoran at a dose of 5 MDU (0.05 dry matter dissolved in 1 ml of distilled water). Subclavian catheters No. 1 were used as a probe for female rats.

Experimental animals were kept under normal conditions. Feeding of animals of both experimental and control groups was the same.

The slaughter of rats was carried out at the age of 1, 30, 60 and 90 days of postnatal development under ether anesthesia. After opening the abdominal cavity, the small intestine was extracted, the total length and diameter of the parts along its length were measured. The mesenteric part of the small intestine was conditionally divided into the initial, middle and final sections. For a detailed study of

morphological parameters, pieces were taken from the proximal, middle and distal mesenteric part of the small intestine. Pieces of the small intestine were fixed in a 12% formalin solution, then carried out over alcohol batteries and poured into paraffin. Sections 5-10 microns thick were made from the blocks on the microtome. Sections were stained with hematoxylin-eosin, according to Van Gieson.

On the NOVEL Model NLCD-307 microscope manufactured in China, 2016, morphometric measurements were made in micro-preparations using an ocular ruler on four sides of the small intestine wall: two lateral, mesenteric and mesenteric. The total thickness of the wall, the thickness of each layer separately, the depth of the crypts, the height of the villi, the width, and the distance between them were measured throughout the small intestine. The cellular composition of lymphoid structures in various parts of the small intestine and their changes under the action of cotoran were studied.In the field of view of the microscope (about 40 x approx.7) the number of microvessels was determined, the diameter and wall thickness of these vessels were measured using an ocular ruler. The cells were counted using a morphometric grid mounted in the eyepiece of the lens -100, eyepiece-10. The number of small, medium and large lymphocytes in the center of reproduction of lymphoid follicles was calculated under a microscope.

The degree of infiltration of the integumentary epithelium of the small intestine per 100 epithelial cells of the villi was determined.

The data obtained during the study were subjected to statistical processing on a Pentium IV personal computer using the Microsoft Office Excel - 2012 software package, including the use of built-in statistical processing functions.

**Results and discussions.** Lymphoid formations of the digestive system, in particular, Peyer's plaques, take part in the formation of an immune response, lymphopoiesis, and lymphocyte recirculation. They are clusters of lymphoid nodules that are devoid of a closed connective tissue case, with germinative centers (B-zones), domes located above them and interstitial areas (T-zones) [4].

Functionally, the immune system of the small intestine is divided into 2 sections: inductive, including Peyer's plaques and regional lymph nodes, and effector, which includes the mucous membrane and its own plate. [ 5,10]. The recognition and presentation of the antigen occurs in the inductive region, and the accumulation of activated T- and B-lymphocytes occurs in the effector region.

Structurally, the immune system of the small intestine is divided into several links:

- ✓ primary, including the epithelium and its own plate of the mucous membrane;
- ✓ secondary - the lymphoid tissue of the posterior plaque;
- ✓ tertiary, represented by regional mesenteric lymph nodes [2].

R.M. Khaitov, B. V. Pinegin (2003) believe that one of the main components of the immune system of the mucous membranes are interepithelial lymphocytes, the number of which increases after birth, which is associated with the ingestion of various antigens.

Our studies have shown that weak lymphocytic infiltration is noted in the epithelial cover of the villi of newborns, where the number of lymphocytes per 100 epithelial cells ranges from 6 to 11, averaging - 6.9 ± 0.6. The average number of small lymphocytes is 3.8+0.25, medium lymphocytes - 2.2± 0.2, large - 1.6± 0.3. In our opinion, such a small number of lymphocytes can be explained by the sterility of the intestine, the absence of microbes at this age.

And in the control group, from the newborn to 90 days of age, the number of interepithelial lymphocytes of the small intestine mucosa increases by 1.07 times. The highest growth rate of large lymphocytes is observed at 60 days of age (26.3%), medium lymphocytes at 30 days of age (16%), small lymphocytes at 30 days of age (34%), and the lowest growth rate of large (8.3%) and medium lymphocytes (9.38%) occurs at 90-day-old, small lymphocytes at the 30-day age of postnatal development (Fig. 1).

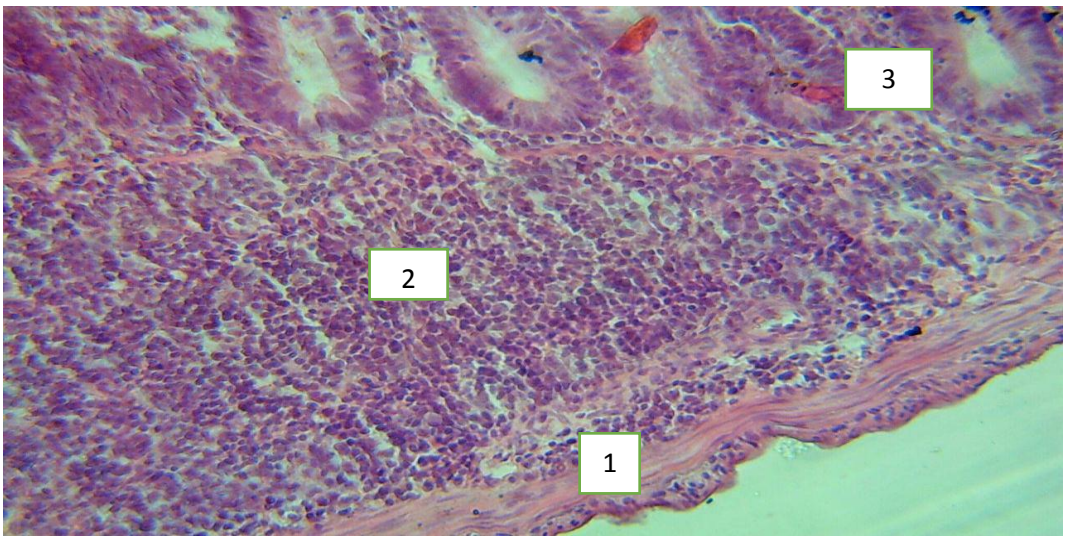


**Fig. 1. Lymphoid nodule of the plaque of the terminal part of the small intestine of a 30-day-old rat. 1 – the muscular lining of the small intestine; 2 –the lymphoid nodule; 3 – the crypts of the mucous membrane. Staining with hematoxylin-eosin. Approx. 10 x 20 vol.**

In the experimental group of rats from 30 days to 90 days of age, the interepithelial lymphocytes of the mucous membrane of the small intestine increase by 1.07 times. The highest growth rate of large lymphocytes is observed at 60 days of age (26.3%), medium lymphocytes - at 30 days of age (16%), small lymphocytes – at 30 days of age (34%), and the lowest growth rate of large (8.3%) and medium lymphocytes (9.38%) 90 days The age of small lymphocytes was noted at the 30-day age of postnatal development.

Our data on the cellular composition of interepithelial lymphocytes of the mucous membrane of the small intestine corresponds to the data of G.G. Aminov and co-authors (1992), who indicate that the main inhabiting cells of interepithelial lymphocytes are small lymphocytes. V.A. Kryzhanovsky (2000) agrees with this opinion, who also believes that the cellular composition of diffuse lymphoid tissue of the mucous membrane is represented by reticular cells, a small number of plasma cells, macrophages and numerous small lymphocytes.

A number of authors - Guanxiang Liang, Nilusha Malmuthuge (2016), Lisa Chedik, Dominique Mias-Lucquin, Arnaud Bruyere (2017), Lopes FM, Varela Junior AS (2014), Manuela Buettner and Matthias Lochner (2016) - single lymphoid follicles of the intestine began to be considered as an independently functioning part of the intestinal immune system.

Lymphoid follicles from the newborn period to 90 days of age are fully formed. Already at the age of 30 days, the reproduction center, dome and mantle zone can be distinguished in the lymphoid follicles.

The study of the cellular composition of the lymphoid plaque reproduction center showed that in newborn rats, more than half of the lymphocytes are small (50.5± 0.35) lymphocytes, and the proportion of large (14.7± 0.2) and medium (33.9± 0.3) lymphocytes is slightly less than 50%. With age, this ratio changes: from 5 to 10%, the number of small lymphocytes increases at 30 days of age, up to 10% at 90 days of age. The number of large lymphocytes at 60 days of age increases by only 2.2% compared to newborns.

And in the number of average lymphocytes, there is a decrease in their number with age: at 30 days of age they are -29.1 ± 0.22, at 60 days of age -26.5 ± 0.33, and at 90 days of age even less -24.0 ± 0.31.

Our data correspond to the data of M.H. Rakhmatova (2018), according to which all zones of the lymphoid nodule in the small intestine are fully formed by puberty.

Thus, when lymphocytes infiltrate the epithelial cover of the villi of the small intestine in the early stages, when exposed to cotoran, an increase in the number of large and medium lymphocytes is observed, and by the age of 3 months, the number of large, medium and small lymphocytes approaches

the indicators of the control group. This suggests that in the early stages, the intestine responds to the effects of kotoran by increasing the number of immature (large) lymphocytes, and at 3 months of age, the lymphocytic balance is restored, and lymphocytic infiltration from the proximal to the distal increases.

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