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REACTIVE CHANGES IN THE SPLEEN OF EXPERIMENTAL ANIMALS WHEN EXPOSED TO GENETICALLY MODIFIED PRODUCT

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ABSTRACT
In the experimental group of laboratory animals, visible changes in the spleen are noted, characterized by an increase in size, average weight, changes in the structure and color of the organ under study. A high synchronicity of quantitative changes in the central and peripheral part of the lymphoid nodule and the red pulp of the spleen under the conditions of GMO use was revealed. It is characterized by inter-organ linear correlation relationships between the same and dissimilar types of cells and in some cases even corresponds to the values within the organs. A genetically modified product-soy flour-negatively affects the condition of the spleen.

Keywords: genetically modified product, experimental animals, spleen, morphology.

The influence of various pathogenic factors on human health leads to morphological changes in tissues, disruption of the function of individual organs, and in particularly severe cases, the whole organism as a whole. All this forces scientists to pay close attention to the organs of immunogenesis that provide the body's defense mechanisms. The spleen, in response to the effects of various pathogenic agents, forms a generalized immune response that ensures the maintenance of immune homeostasis and the necessary level of adaptive potential of the body [1, 3, 5].
GMOs are plant or animal organisms that have been modified in an unnatural way for nature with the help of genetic engineering to give the genotype new properties of the organism. Various transgenic plants (soybeans, corn, sugar beet, potatoes, cotton) with resistance to viruses, the Colorado potato beetle and other insects, as well as to pesticides have been obtained with the help of genetic engineering [4,13].
According to the World Health Organization, one of the most important factors shaping human health is a proper diet. The creation of genetically modified food sources is an inevitable way to solve many nutrition and health problems [15].
Crop improvement through breeding was largely based on genetic diversity, which emerged as a result of a long history of natural mutations. As the demand for food increases and the pressure on crop selection continues to grow, the need to create gene diversity through rapid mutation becomes urgent [15,16].

The term "genetically modified organisms" (GMOs) has appeared recently [6,11]. There are very few studies on the biomedical safety of GM products from potatoes, corn, soybeans, rice, cucumbers, tomatoes, sweet peppers, peas and rapeseed for the production of food and feed, conducted by independent scientists, and not by interested manufacturing companies.

While transgenic technologies marked a new era in crop improvement, some problems prevented their widespread spread [16].

McCann et al. [14] found that the nutrient composition of several varieties of commercial glyphosate-tolerant soybeans obtained after 3 years of breeding remained equivalent to the composition of conventional soybeans.

Kim S.H. et al. [11] revealed that the allergenicity of extracts obtained from conventional types of beans and GM soybeans was identical in the adult group of people studied. Nevertheless, despite the positive results in the study of glyphosate-tolerant soybeans, other authors concluded that more thorough studies are needed to assess the allergenicity of GM soybeans and other GM foods, including a wide selection of controlled samples of GM soybeans [2, 5, 11].

Soybeans are the number one genetically modified crop in the world. Some genetically modified soybean varieties have been modified specifically for the production of oil and yield three times more oleic acid than conventional varieties. GMOs increase yields, create resistance of crops to diseases, produce insecticides against pests. But mutations of viruses that cause genetic changes in plants bring diseases to animals and people. Everything artificially created is inferior in quality to natural analogues [8, 9, 10]. The spleen reacts quickly to a variety of external factors. Morphological transformations of the spleen after exposure to various substances of chemical and biological nature, including genetically modified organisms, have been widely studied [6,12]. Despite the abundance of numerous studies devoted to the study of the immune apparatus of the human spleen and in experimental animals, almost no attention is paid to the issues of microanatomy and the cellular composition of the lymphoid structures of this organ [7]

A detailed study of the morphogenesis of the spleen with the use of GMOs will allow the correction of possible immunological lesions to be scientifically justified.

The purpose of this study was to study and evaluate the effect of the GM product on the morphological parameters of the spleen of laboratory animals in an experiment.

Materials and Methods

Commercial soy flour was used as a GM product. Experimental studies were conducted on white mongrel rats.

All laboratory animals were divided into 3 groups: the experimental group-animals that included soy flour in the total diet (at a dose of 0.02-0.03 g per 1 rat weighing 130-150 g for 30 days (n=30); the control group - animals that received only a total diet, without soy flour (n=30). Group 3-intact animals (n=30) kept in standard vivarium conditions.

Soy was used as a GM product in the experiments. Using the PCR method, the presence of the 35S+FMV promoter in the studied GM soy was revealed, which proves that the studied soy is a GM product. There is no such promoter in ordinary soy.

To study the morphological parameters of the spleen, a macroscopic method (anatomical dissection) was used.

After cutting the material, fixation was carried out in 10% buffered formalin, then rinsing in water and dehydration in alcohols and compaction with benzene. After that, sections 4-6 microns thick were poured into paraffin and prepared, which were stained with hematoxylin and eosin. The sections were examined morphometrically using an eyepiece micrometer DN-107T/ Model CM001 CYAN cope (Belgium).

Mathematical processing was performed directly from the general data matrix "Excel 7.0" with the involvement of the capabilities of the program "STTGRAPH 5.1", the standard deviation and representativeness errors were determined. When organizing and conducting research, the principles of evidence-based medicine were observed.

Research Results and Discussion

The parameters of the spleen of laboratory animals of the experimental and control groups also significantly differed (Table 1). Comparative changes in the spleen concerned size, structure (looseness) and color (dull).

Table 1 Results of macroscopic examination of the spleen of experimental animals of the experimental and control groups

Indicators	Control group	Main group
The spleen is enlarged, %	0	28,8±6,4*
Average weight of the spleen, g	0,65±0,12	0,80±0,10*
Relative weight of the spleen, g/100 g of body weight	0,38±0,03	0,49±0,05*
Change in structure, %	0	7,6±4,7*
Change in color, %	0	13,4±4,6*

Note: * - the reliability of the differences in relation to the control.

If the animals of the control group did not have an increase, changes in the structure and color of the spleen, then in the experimental groups these indicators were markedly different compared to the control.

The revealed changes in the central and peripheral parts of the lymphoid nodule and the red pulp of the spleen of mice after two weeks of GMO exposure indicate that against the background of a decrease in the number of small lymphocytes (1.3 – 2.5 times,

P<0.01) and mature plasmocytes (more than 6 times, P<0.01), an increase in the number of cell forms is observed in the state of destruction (1.8 - 4.57 times, P<0.01) and reticular cells 2.5-5.3 times, P<0.001). At the same time, the content of less differentiated lymphocytes (blasts, large lymphocytes) does not significantly differ from the control. An increase in destructive processes and a decrease in the number of differentiated lymphoid cells, plasma cells are signs of decompensation in the specified zone of the spleen and indicate an inhibition of the processes of differentiation of B-lymphocytes.

Pathological changes in the spleen noted in the experimental group indicate that this GM product negatively affects the condition of these organs in experimental animals. The absence of carcinogenic effect of GM soy flour on animals of the experimental group was apparently due to the short period of exposure to this food product.

Conclusions:

In the experimental group of animals, visible changes in the spleen are noted, characterized by an increase in size, average weight, as well as changes in the structure and color of this organ. This means that the GM product-soy flour has a negative effect on the condition of the spleen. In the spleen, the correlation of cytological profile indicators (absolute values characterizing the number of cells of different types) did not increase, but decreased. The maximum decrease in conjugation was found in the central part of the lymphoid nodule. This indicates a significant increase in cellular autonomy and morphofunctional disorganization of individual parts of the organ, which makes it possible to attribute the spleen and, especially, its lymphoid nodules to the "weak links" of the immune system.

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