Morphological changes in the kidney, liver and spleen during prolonged administration of iron nanoparticles

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Abstract. We determined the cytotoxic effect of iron nanoparticles of 70 nm, with a single per oral administration in an experiment on white outbred mice. Morphological changes were evaluated in the internal organs. Thus, changes depend on the concentration of nanoparticles at long-term per oral exposure: identified violations of the structure of the liver, kidneys and spleen as venous plethora and degeneration of cells at 250 and 500 mkg / kg dose of nanoparticles are reversible, changes in the organs were pronounced with a dosage of 1000 mkg / kg.

1. Introduction
Nanotechnologies have become a revolutionary step in the various fields of science and promising area for scientific research in coming years, as these studies are based on the application and use of substances at the molecular level, and structures of one nanometer (one billionth of a meter) and 100 nm. The possibilities of using nanoparticles in biomedical purposes were a rapidly developing field of nanotechnology, which allows to open new possibilities in the diagnostics and treatment of various diseases.

At present, nanostructures were established for biomedical applications in various fields, including visualization of various biological objects and delivering drugs to cells, tissues and organs. The unique features of nanoparticles, such as high surface activity, the stability of biomolecular absorption, changes in physical and chemical properties under the influence of physical fields, their small size, comparable to biomolecules, the severity of the magnetic properties and biocompatibility, open wide prospects for creation of new drugs based on nanopreparations, that can be used in the treatment of various diseases, including cancer.

It should be noted that only few studies in the field of magnetic nanoparticle delivery and their application for MRI and CT scans were carried with using of morphological methods. In addition, we found no published data of comparative analysis of structural and functional changes that occur in the kidneys, liver and spleen of laboratory animals at peroral administration of different doses of magnetic nanoparticles.

Objective: to study the morphological changes in the kidneys, liver and spleen of laboratory animals at peroral administration of Fe nanoparticles in experiment.

Materials and methods: Fe nanoparticles of 70 nm ± 10 nm, 28 white outbred male mice, organometric and morphological methods.

The design of the experiment: the animals were divided into three experimental groups of seven mice in each group, during 5 days they received administered per orally solution of iron nanoparticles at a concentration 250 mkg/kg, 500 mkg/kg, 1000 mkg/kg. Each mouse of control group received...
administered per orally saline during 5 days. Animals were derived from the experiment by decapitation the next day.

2. Results and their discussion

Organometric study.
The coefficient $K$ was introduced for increasing the objectivity of evaluation of organometric study, which was calculated as the ratio of the mass of organ to the weight of the animal body. Median was used in the statistical analysis due to the heterogeneity of the samples.

| Series of experiment | Coefficient (K) |
|----------------------|-----------------|
|                      | Fe              | Control        |
| 250 mg/kg            | 103,4±3,35      | 122,5±8,9      |
| 500 mg/kg            | 115,6±13,184    | 122,5±8,9      |
| 1000 mg/kg           | 113,2±14,33     | 122,5±8,9      |

Findings were the following: reduction in the $K$ (Tab. 1) was noted in mice compared with controls, suggesting an increase in weight of kidneys.

Pathological changes in organs.
The following changes have been identified due repeated per oral administration of iron nanoparticles: in the kidney - swelling of the kidney convoluted tubules was marked at a dosage of 250 mg/kg. Changes were represented by small granular dystrophy and swelling of the epithelium, the poorly defined plethora of vessels of the brain substance with the introduction of iron nanoparticles at a concentration of 500 mg/kg. Degeneration of convoluted tubules and a more pronounced hyperemia of vessels of the cortex and the glomeruli were observed at the introduction of iron nanoparticles in the maximum concentration of 1000 mg/kg. Other authors who have studied the morphology of the internal organs of rats with intravenous injection of $\text{Fe}_3\text{O}_4$, also note the venous plethora of vessels of the brain substance, but in our experiment there was no expansion of Shymlansky-Bowman capsule, which is possible due to other dimensions and method of introducing nanoparticles [4,7].

![Morphological changes](image)

Figure 1. Morphological changes in the kidneys with the introduction of iron nanoparticles at a concentration of 1000 mg/kg, which are represented by degeneration of convoluted tubules and expressed plethora of vessels of the cortex and glomeruli.

In the liver - a moderately pronounced hyperemia of blood vessels and degeneration of hepatocytes were indicated with a dosage of 250 mg/kg. The severity of plethora dystrophy was increasing with a dosage of 500 mg/kg and a large number of the black granules were observed in the blood and between hepatocytes. There were a very marked hyperemia of blood vessels and degeneration of cells with a dosage of 1000 mg/kg.
Figure 2. Morphological changes in the liver at doses of 1000 mg / kg: very pronounced plethora of vessels and degeneration of hepatocytes.

In the spleen - a large numbers of black granules were observed in the red pulp with all the doses. Large follicles with germinal centers active were observed at of 250 mg / kg and 500 mg / kg. The predominance of red pulp over white pulp was indicated at a dosage of 1000 mg / kg, white pulp was with indistinct outlines, the follicles were without light centers and large cells were found.

Figure 3. Morphological changes in the spleen at a dosage of 1000 mg / kg were presented a white pulp with indistinct outlines and follicles without light centers, large cells were found. Nanoparticles were not accumulated in the organs, which was confirmed by the lack of visualization on MRI.

3. Conclusions
Changes in varying degree were revealed in the analysis of micropreparations of the kidneys of laboratory animals of experimental group, depending on the solution concentration of iron nanoparticle. The maximum changes were observed at a dosage of 1000 mg / kg. The presence of vascular venous plethora was evidence of the reaction on the coming of Fe nanosized particles into the systemic circulation, and degenerative changes in the organs testify to the presence of the damaging effect of nanoparticles. These changes were not observed in the control group.

Acknowledgment
Thus, uncoated iron nanoparticles were not accumulate in the organs, during prolonged oral administration. Morphological changes depend on the concentration of nanoparticles: identified violations of the structure of the liver, kidneys and spleen as venous plethora and degeneration of cells at 250 and
500 mkg / kg dose of nanoparticles are reversible, changes in the organs were pronounced with a dosage of 1000 mkg / kg.

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