Nowadays the study of the causes of infertility and ways to overcome it is an important and urgent problem of modern biomedical science. Most researchers indicate that in the structure of infertile marriages male factor constitutes 20%, the cause of infertility cannot be detected in 15% of these families, and 25% of infertility index are caused by reproductive disorders in men and women [1, 4, 9].

Reduced spermatogenic and hormone-producing function of the testes can result from acute and chronic disorders of blood circulation (both local and systemic), endogenous intoxication, which occurs in many pathological conditions, and exposure to physical and chemical factors [2, 4, 8]. In recent years, environmental technological burden increases. As a result, the number of chemical substances and their metabolites, which have a negative influence on organs and body systems, including testicles, increase as well. Technogenic pollution with heavy metals and their derivatives is especially dangerous [2, 6, 8]. It should be noted that peculiarities of structural changes in testes under the influence of heavy metals are not sufficiently studied.

The aim is to study the morphological changes in testes under the influence of cadmium chloride.

Materials and Methods

Testicles of 20 mature white male rats were studied morphologically. Rats were divided into 2 groups. The first group included 9 healthy intact animals; the second group included 11 male rats that received cadmium chloride subcutaneously at a dose of 6 mg/kg for 4 weeks [6].

Euthanasia of animals was performed one month after the beginning of experiment via exsanguination under thiopental anaesthesia. Pieces of the testes were fixed in 10% neutral formalin solution and, after corresponding processing in ethyl alcohol of increasing concentrations, placed in wax. Microtome sections 5-7 microns thick were stained with hematoxylin-eosin, toluidine blue, by van Gieson, Mallory, and Veyhert [7]. Histological specimens were examined with the help of microscopes MBS-15 and Lyumam-P8.

Results and discussion

No deviations of the histological structure of testes were detected in animal specimens of the control group. In the intact testes, spermatogenic epithelium cells, which are at different stages of development and are arranged in several concentric layers, adjoin to the proper tunic of convoluted seminiferous tubules. Between the convoluted seminiferous tubules interstitial connective tissue is localized. In the testicular stroma there are small groups of Leydig cells, which are characterized by homogeneous cytoplasm and a nucleus located eccentrically, along the blood vessels of small calibre. The nuclei of these cells are mostly irregular in shape.

Histological examination of macro-preparations of the testes of experimental animals that received cadmium chloride revealed significant structural changes. In these experimental conditions, there was a slight reduction in the size of convoluted seminiferous tubules as compared to control animals.

Fig. 1. Stromal edema, partial reduction of layers and spermatogenic epithelium cells damage and desquamation, thickening of the membrane of convoluted seminal tubules in testicles of white rats under the influence of cadmium chloride. Staining by hematoxylin-eosin. Magnification: x 140.
Microscopically it was found out that most convoluted seminiferous tubules are in a state of devastation and degeneration of spermatogenic epithelial cells. Also, often areas of dystrophic and necrotic spermatogenic epithelium were observed (Fig. 1, 2).

Intesticle samples of white male rats that received cadmium chloride, in the lumen of convoluted seminiferous tubules products that appear as a result of the disintegration and necrosis of spermatogenic epithelium were found. In the testes areas with convoluted seminiferous tubules, the lumen of which was filled with cellular detritus, are detected microscopically. Some researchers consider that partial or complete obliteration of the seminiferous tubules leads to thickening of their proper tunics [3, 5].

In addition to the above described changes there was revealed the desquamation of spermatogenic epithelium in convoluted seminiferous tubules of experimental animals, which received cadmium chloride, and its dislocation into the lumen. Sometimes the cells of spermatogenic epithelium were placed on the basement membrane unevenly; in some seminiferous tubules cavities appeared between the cells. One, sometimes two layers of dystrophic spermatoblasts and spermatocytes were attached to the proper tunic of some tubules. Between the layers of myoid cells bundles of collagen fibres were observed.

Among the spermatogenic epithelial cells of convoluted seminiferous tubules the marked reduction of spermatocytes and early spermatids is detected. The nuclei of these cells are hyperchromic, and their cytoplasm is swollen and vacuolated. Basement membrane in most observations has signs of sclerosis. Microscopically we have observed structural changes in the interstitium of the testicles in the modelled pathology. Spaces between the convoluted seminal tubules are expanded as a result of edema and proliferation of connective tissue elements. These spaces are of different shapes and sizes, often with polymorphonuclear cell infiltration. In interstitium the connective tissue is replaced by rough fibrous, hyalinized and scleroid structures. These effects lead to deformation of vessels and convoluted seminiferous tubules. Isolated Sertoli cells were located at the proper tunic of convoluted seminiferous tubules of the testicles.

The nuclei of Sertoli cells, which are located mostly near the basement membrane of convoluted seminiferous tubules, are deformed. The cytoplasm of these cells in the majority of observations is granular and with signs of active phagocytosis.

The cytoplasm of Leydig cells is granular and often vacuolated; their nuclei are hypochromic. In marked proliferation of connective tissue Leydig cells lose their orientation, in some of them marked nuclei lysis is detected.

In the simulated experimental conditions we have observed thickening of the proper tunic of convoluted seminiferous tubules. The basement membrane of convoluted seminiferous tubules proper tunic is wavyly deformed, and unevenly expanded. It should be noted that the most common cause of thickening of convoluted seminal tubules proper tunic was the proliferation of connective tissue elements, sclerosis and hyalinization.

In the modelled pathology, arteries and veins of the testes underwent restructuring as well (Fig. 2, 3). The walls of the arteries are thickened, their lumen is narrowed;
some endotheliocytes have got dystrophic and necrobiotic changes and signs of desquamation. Within this experiment we have observed plethora, stasis of blood and stagnation in venules and veins of the testicles. The contours of the venous walls are indistinct and thickened. There are alternating zones of edema and sclerosis with the wiggly and crooked contours; lumen of the veins is deformed. Endotheliocytes of the venous vessels are increased in size with signs of edema, dystrophy, and necrosis. There were islands of endothelial cells desquamation, pycnosis and lysis of their nuclei. It is known that chemicals circulating in the blood can damage endotheliocytes, which was revealed at histological examination of the testicle micro-preparations of animals exposed to cadmium chloride. It is considered that the endothelium that covers the inner surface of blood vessels is an important autocrine, paracrine and endocrine organ with numerous regulatory functions. It is known that normally functioning endotheliocytes produce nitric oxide (NO), which regulates vascular tone, affects the vascular wall remodelling processes, determines the system of antioxidant defence and peroxidation aggression, inhibits platelet aggregation and macrophages adhesion, and proliferation processes in the vascular wall. Damage of a significant number of endothelial cells in the investigated arteries can lead to endothelial dysfunction, which plays a leading role in vascular remodelling and morphogenesis of the studied pathology [4, 9]. In the perivenous spaces there are marked swelling and sclerosis of perivascular tissues. The detected changes point to the disorders of the testicular venous drainage system, which were complicated by venous plethora, hypoxia, dystrophic and necrobiotic changes of cells and tissues, infiltration and sclerosis. So, the prolonged exposure of studied animals to cadmium chloride causes severe structural reorganization in parenchyma and stroma of testicles, which is characterized by severe cardiovascular disorders, reduction of layers, degenerative and necrobiotic changes of spermatogenic epithelium and endotheliocytes, stromal infiltration, sclerosis and hyalinization in the investigated organ.

**References:**

1. Antonechko F.F. The role of varicocele and its surgical treatment in violation of reproductive function., F.F. Antonechko, D.A. Scherbovskaya, S.A. Lelchuk., Reproductive health of children and adolescents. – 2009., No. 3., pp. 77-84.
2. Voloshin I.M. Assessment of the impact of heavy metals on human health., I.M. Voloshin., Modern ecological environment and human environmental pathology. – Lviv., B.L., 2007., pp. 10-12.
3. Hlodan O.J. Morphofunctional changes in the testis in terms of clamping blood vessels of the spermatic cord., O.J. Hlodan., Galician Drug Herald. – 2008., Vol. 15, No. 1. pp. 12-14.
4. Hrytsulyak B.V. The structure of the testes in acute ischemia., B.V. Hrytsulyak., Scientific Bulletin of the Uzhgorod University. The series "Medicine". – 2000., Issue 12., pp. 27-38.
5. Hrytsulyak B.V. Cytological changes in the test is under conditions of blockade of blood outflow in experiment., B.V. Hrytsulyak, V.B. Hrytsulyak, O.J. Hlodan., Bulletin Carpathian National University by V. Stefaniak. Series Biology. – 2011., Issue XV., pp. 201-204.
6. Pryshlyak A.M. Atrial cardiomyocytes secretory activity in experimental animals with different resistance to hypoxia in conditions of cadmium intoxication., A.M. Pryshlyak, M.S. Gnatyuk., Bulletin of scientific research. – 2006., No.3., pp. 62-64.
7. Sorochinnikov A.G. Histological and microscopic appliances., A.G. Sorochinnikov, A.E. Doroshevich. – Moscow., Medicine, 1997. – 448 p.
8. Trachtenberg I.M. Heavy metals as chemical pollutants of industrial and surrounding environment., I.M. Trachtenberg., Environment and Health. – 2005., No. 2., pp. 48-51.
9. Bong G.W. The adolescent varicocele: to treat or not to treat., G.W. Bong, H.P. Koo., Urol. Clin. North. Am. – 2004., No. 31 (3), pp. 509–515. https://doi.org/10.1016/j. ucl.2004.04.012
10. Jarow J.P. Induction of spermatogenesis in azoospermic men after varicocele repair., J.P. Jarow., J. Urol. – 2003. – 170 (2 Pt 1), pp. 667–678.

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**Conclusions**

Thus, in the course of the study it was found out that long-term administration of cadmium chloride to white male rats leads to significant structural reorganization in parenchyma and stroma of testicles, which is characterized by severe cardiovascular disorders, reduction of layers, degenerative and necrobiotic changes of spermatogenic epithelium and endotheliocytes, stromal infiltration, sclerosis and hyalinization in the investigated organ.