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FEATURES OF ULTRASTRUCTURAL REARRANGEMENT OF TESTICULAR TISSUES OF RATS AFTER THYROIDECTOMY

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ABSTRACT
In an experiment in rats treated with thyroidectomy, by using the electron microscopy, it was found that in the postoperative period progressively increase the dystrophic-destructive changes in stromal cells and in parenchymal cells of testicular epithelium. They consist of picnotial deformation of the nuclei, enhanced electron density of the karyoplasm, formation of lysis sites and different sizes of vacuole structures in the cytoplasm with the expansion and fragmentation of the granulum endoplasmic mesh tubules and tubules of the Golgi complex. At the same time, there was a decrease in the content of lipid inclusions and hormonal granules and mitochondria that had an enlightened matrix and often – damaged crystals. Due to the consolidation of the fibrous layer and the cytoplasm of the myoid cells, the walls of the convoluted seminal canals dusted. The basal plate acquired an uneven thickness, microfilaments in it were poorly contoured. Significantly increased the intercellular gaps. All these changes occurred on the background of violation of hemomycrocycle circulation. Moreover, the first day was characterized by the narrowing of the lumen of the capillaries with a decrease in their bandwidth. Later on, the lumen of the capillaries markedly expanded and hemomycrocycle disorders became stagnant, which could be the cause of ischemia of the organ as one of the important morphogenetic factors in the development of dystrophic-destructive processes in the parenchymal-stromal components of the testicles. The estimated features of structural reorganization of tissues of the testicle can form the basis of functional insufficiency of this organ.

KEY WORDS: Thyroidectomy, Testicle, Ultrastructure.

INTRODUCTION
Postoperative hypothyroidism, as known, develops in 35-48% of patients undergoing the surgery on thyroid gland. And thyroidectomy in 100% of cases is accompanied by persistent Hypothyroidism.[1]

At the same time, it has now been estimated that the deficiency of thyroid hormones may lead to an impaired spermatogenous and sucrose function of the testicles in adult males.[2,5] The reason for this is the decrease in testosterone levels in the blood, which develops as a result of a violation of the enzyme systems responsible for the synthesis of androgens in the testicles.[3, 4] There is also an opinion that subclinical hypothyroidism, as well as the obvious, is associated with hyperandrogenism in men. However, there are alternative data indicating hypoandrogenesis in this contingent of patients.[5]

Therefore, to clarify the pathogenesis of male sexual dysfunction in conditions of hypothyroidism, morphological studies may be important.

MATERIALS AND METHODS
Experiments were carried out on 18 sexually mature white laboratory male rats with a body weight of 160-180 g. Of these, 3 animals composed the control group. Another 15 rats under ketamine anaesthesia underwent the thyroidectomy. The material for electron microscopic studies was taken after 1, 3, 7, 14 and 28 days after surgery in accordance with the generally accepted method. Ultra-thin sections made on ultramicrotome LKB-3 (Sweden) were contrasted with 1% aqueous uranyl acetate solution and lead citrate according to Reynolds method and studied by the Illuminating Electronic Microscope – 125K.

All experimental studies were carried out in accordance with the principles of bioethics set forth in the Helsinki Declaration and the Law of Ukraine "On the Protection of Animals from Cruel Treatment" (No. 1759-VI of 15.12.2009), which was confirmed by the Commission on Bioethics of the Ternopil I. Horbachevskyi State Medical University (protocol No. 34 of 01.03.2016).

RESULTS AND DISCUSSION

Electron microscopic examination of testicles of white laboratory rats in the early days (1-3 days) after experimental thyroidectomy allowed to estimate a certain reorganization of their structural components.

One day after the thyroidectomy was characterized by a decrease in the capacity of hemocapillaries due to a noticeable narrowing of their lumen. This was due to the swelling of endothelial cells and, in particular, of their nuclei, which, together with a thin layer of the cytoplasm and the cytolymum, broke out into the lumen of the microvessels. In the lumen of the hemocapillars, the accumulation of ovally deformed erythrocytes was detected. After 3 days there was a partial expansion of the capillaries in which the lumen continued to remain ovally deformed red blood cells.

The endocrinocytes, the sutentocytes and the cells of the spermatogenic epithelium during this observation period retained the rounded form of their nuclei. Euchromatin prevailed in their karyoplasm, although small granules with their predominant localization in the peripheral zones were detected. Nuclear membranes were clearly contoured; perinuclear spaces were moderately expanded. However, the cytoplasm of the cells as a result of edema was electron-permeated, containing a small amount of organelles, vacuoles and small amounts of osmophilic mitochondria and lysosomes. In the part of spermatogonia there were signs of mitosis in its various phases. Sometimes the spermathetes revealed a violation of the structure of acrosomes.

The myoid cells of the walls of the convoluted tubules had an oblong form of the nucleus and a small volume of the cytoplasm. The basement membrane was sometimes contoured. It contained small sized spermatogonia with a rounded form of the nucleus and a small area of the cytoplasm.

At 7-14 days of the experiment there was a further expansion of the lumen of the hemocapillaries with their marked blood supply. The thickness of the cytoplasm of the endothelial cells, especially in the peripheral parts, decreased somewhat. In endocrinocytes for a given period of observation, there was a change in the shape of the nuclei due to shallow invaginations of the carriel. The karyoplasma of such cells acquired a moderate electron density and became homogeneous. In their cytoplasm there was a relatively small number of lipid inclusions and only isolated hormone-releasing secretory granules. There were also relatively few granular endoplasmic grenade tubes and tanks of the Golgi complex, as well as mitochondria. The peripheral areas of the cytoplasm looked locally enlightened or included different sizes of vacuoles (Fig. 1).

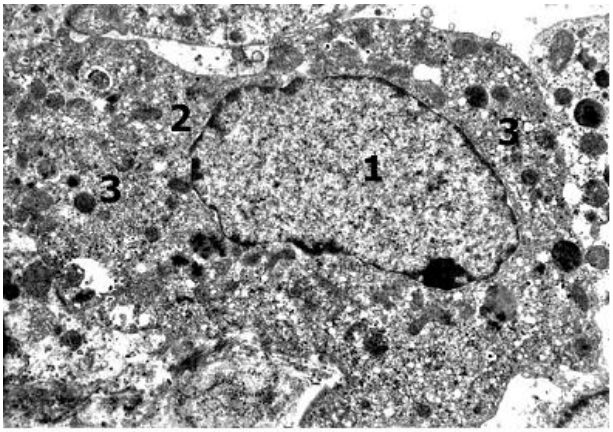

Figure. 1: Ultrastructural changes in rat testicular endocrinocite 14 days after the experimental thyroidectomy. x 12,000. The nucleus of the endocrinocyte – 1, the endocrinocyte cytoplasm – 2, the endocrine granules – 3.

It was also noted the damage to the structure of the wall of the spermatic tubules and the spermatogenic epithelium. At the same time, the walls of the convoluted seminal tubules looked thickened due to the consolidating of the fibrous layer and reducing the thickness of the cytoplasm of the myoid cells. Basal plate was uneven in thickness, homogeneous in appearance and included osmiofilic areas. Microfilaments were not very clearly contoured in it.

Destructive changes in the sustentocyte manifested by a change in their form. The cytoplasm of these cells acquired the elevated electron density, it contained a small amount of organelles. There were quite large sizes of vacuoles and necrotic areas. The shape of the nuclei has broken down, their area has decreased. Karioplasma acquired the increased osmophilicity and looked homogeneous. Spermatogonia lost their contact with the basal membrane. Separate spermatogonia showed signs of mitosis. Plasmolemia and cariolomas of the cells were unclearly contoured, often locally damaged. Intercellular spaces looked considerably enlarged (Fig. 2).

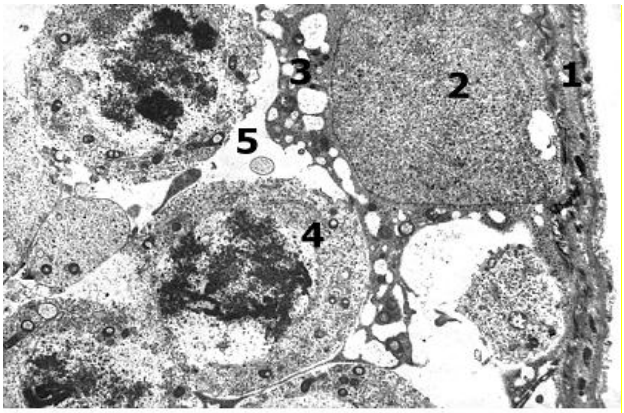

Figure. 2: Submicroscopic changes in the wall of the twist tubule of the testicle and spermatogenic rat epithelium at 14 days after the experimental thyroidectomy. x 9,000. The cytoplasm of the myoid

cell – 1, the nucleus of the susentocyte – 2, the cytoplasm of the susentocyte –s 3, the spermatogonia – 4, the intercellular space – 5.

Some primary and secondary spermatocytes acquired a modified form due to the swelling of the cytoplasm, a significant increase in intercellular gaps and deformation with plasmol. The nuclei of the spermatocytes remained to be mostly round, but their karyolema sometimes looked damaged. In the karyoplasm, both euchromatin and osmiofilic lumps of heterochromatin of different localization were observed. There were relatively few organelles in the cytoplasm. Often there were homogeneous, destructively altered sites.

In many spermatids and sperm at that time, there was an obscure vague contour of acrosomes, or even its absence, which combined with high electron density of nuclei and local damage to carriel membranes. Compared to earlier terms, lesser content of mitochondria was observed. Their membranes were often subjected to de-structuring.

In late terms (21-28 days) of the experiment at an electron microscopic examination, the lumen of hemocapillaries looked already noticeably extended and often overwhelmingly over the diameter of erythrocytes, which, as a result, acquired a clear rounded form. As part of the walls of hemocapillaries, endothelial cells that had electron-cytoplasm and picnotically deformed osmiofilic nuclei were found. There were not much organelles in the cytoplasm and they were often significantly damaged, and the number of pinocytosis cysts was sharply reduced. The basal plate of hemocapillaries became fuzzy contoured and acquired an uneven thickness. In extended perivascular spaces, signs of sclerotic changes were manifested, as evidenced by the presence of both individual collagen fibrils and their entire beams.

On the background of the increasing of changes in the structure of the blood capillaries in the part of the endocrinocytes, there were also significantly altered nuclei, which acquired an irregular shape, became picnotic, and their karyoplasma acquired the elevated electron density. In the cytoplasm of the endocrinocytes, at that time, there were lysis areas, visualized vacuole-like structures of different sizes, as well as expanded fragmented tubules of the granular endoplasmic net and tanker of the Golgi complex. Lipid inclusions and hormonal granules were comparatively few. Mitochondria had an enlightened matrix and often damaged crystals (Fig. 3).

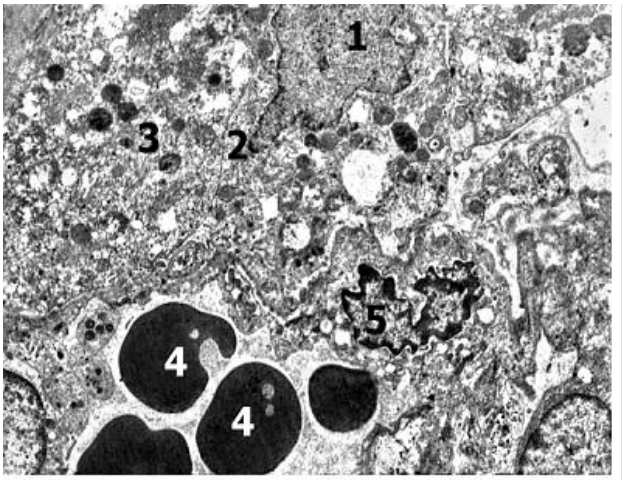

Figure. 3: Ultrastructural changes of hemocapillary and testicular endocrinocyte rat at 21 days after experimental thyroidectomy. x 9,000. Endocrinocyte nucleus – 1, endocrinocyte cytoplasm – 2, endocrine granules – 3, red blood cells in the lumen of the capillary – 4, core of the endothelitis – 5.

As in previous terms, there were significant submicroscopic changes in the structural organization of the wall of the spermatic tubules and the spermatogenic epithelium. The walls of the convoluted seminal canals were further enlarged, due to the consolidation of their fibrous layer and the cytoplasm of the myoid cells. In the latter, the nuclei have been significantly modified. They acquired a picnographic form and contained osmiumophilic karyoplasm. The basal plate acquired an uneven thickness, microfilaments in it were poorly contoured.

Most of the supporting cells also changed their shape. Their cytoplasm became osmiophilic, the number and size of vacuoles in it increased and determined the areas of destruction. The form and area of the nuclei changed due to their picnicking. Karyoplasm has acquired a significant electronic density.

Some of the spermatogonium lost their contact with the basal membrane. Separate spermatogonia were with signs of mitosis. Intercellular spaces thus significantly expanded (Fig. 4).

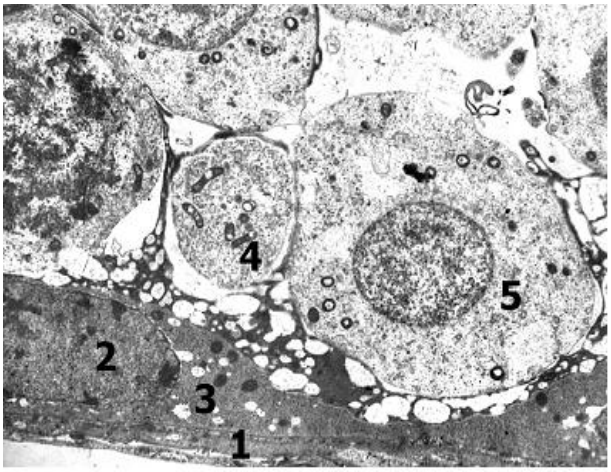

Figure. 4: Submicroscopic changes in the wall of the twisted tubule of the testicle and spermatogenic rat epithelium for 28 days after the experimental thyroidectomy. x 9,000. Cytoplasm of the myoid cell – 1, the nucleus of the susentocyte – 2, the cytoplasm of the susentocyte – 3, the spermatogonia – 4, the primary spermatocyte – 5.

In the cells of spermatogenic epithelium were noticed the destructive changes in both primary and secondary spermatocytes. Some of these cells differed in the deformation of plasmolars, which arose as a result of the expansion of intercellular spaces due to edema. Quite often the local lesions of nuclear karyoles were observed. This was especially true for secondary sperm cells. Their karyoplasm was characterized by the accumulation of osmiophilic lumps of heterochromatin that most were located disordered due to the abuse of the process of meiosis. In the cytoplasm was often observed the light areas of lysis that was simultaneously accompanied by a decrease in the total number of intracellular organelles (Fig. 5).

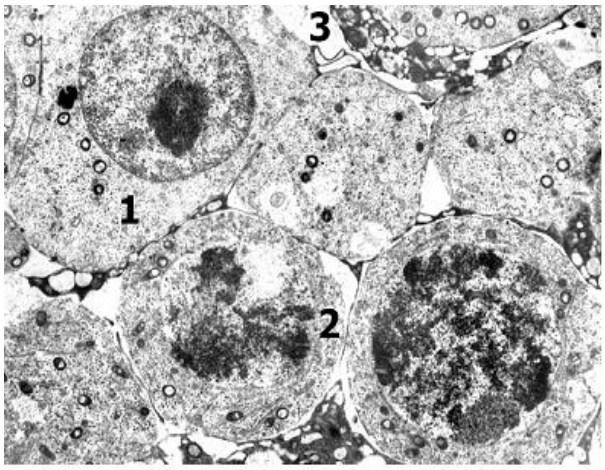

Figure. 5: Submicroscopic changes in the spermatogenic epithelium of the twisted tubule of the testicle of the rat at 28 days after the experimental thyroidectomy. x 9,000. Primary spermatocyte – 1, secondary spermatocyte – 2, intercellular space – 3.

Due to the violation of the formation phase in the spermatids and spermatozoa, the shape and area of the nuclei changed, they acquired a high electron density and had fuzzy contours of the karyolema. The acrosomes were unclearly contouring and around them, electron-illuminated areas were formed (Fig. 6).

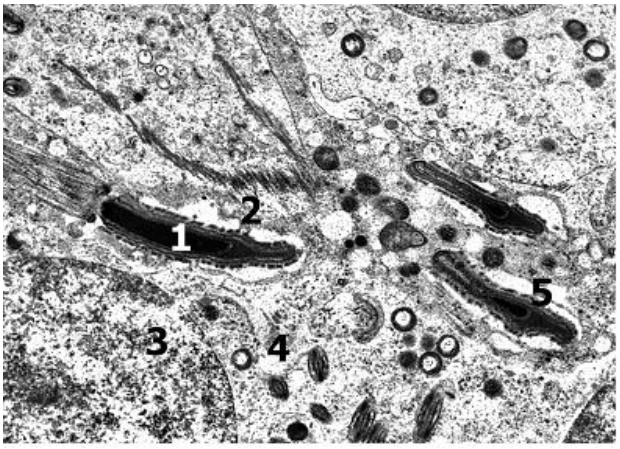

Figure. 6: Ultrastructure of the spermatogenic epithelium of the twisted tubule of the testicle of the rat at 21 days after the experimental thyroidectomy. x 12,000. The nucleus of spermatide – 1, and the cytoplasm of spermatide – 2, spermatocyte nucleus – 3, spermatocyte cytoplasm – 4, electron-light area – 5.

Thus, the results of this ultrastructural study allowed to establish a significant reorganization of all structural components of the testicles of male rats after thyroidectomy. The revealed changes consisted of a progressive increase in degenerative-destructive processes, both in stromal cells and in parenchymal cells of the epithelium. They consisted of picnotial deformation of nuclei, enhanced electron density of karyoplasma, formation of lysis sites and different sizes of vacuole structures in the cytoplasm with the expansion and fragmentation of the tubules of the granular endoplasmic net and tanks of the Golgi complex, reducing the content of lipid inclusions and hormonal granules and mitochondria that had an enlightened matrix and often - damaged membranes. Due to the consolidation of the fibrous layer and the cytoplasm of the myoid cells, the walls of the twisted seminal canals were thinned. The basal plate acquired an uneven thickness, microfilaments were poorly contoured in it. Significantly increased the intercellular gaps. All these changes occurred on the background of a violation of hemomicrocirculation of stagnant nature, which may be the cause of ischemia of the organ as one of the important morphogenetic factors in the development of dystrophic-destructive processes in the parenchymal and stromal components of the testicles, which can form the basis of the functional organ failure.

CONCLUSIONS

1. After the thyroidectomy in rats in parenchymal-stromal testicles components there is a progressive

increase in degenerative-destructive changes that may form the basis of functional organ failure.

2. The detected changes in the degenerative-destructive nature develop on the background of a violation of hemomicrocirculation of stagnant nature, which may be an important link in the morphogenesis of the development of functional organ failure.

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