DOI 10.26724/2079-8334-2023-1-83-193-196
UDC 616.37:612.018/.08

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MORPHOFUNCTIONAL CHARACTERISTICS OF THE STRUCTURAL COMPONENTS OF THE PANCREAS EXOCRINE PART IN CHEMICAL CASTRATION

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Throughout a person's life, the pancreas, which is the second largest gland (after the liver), is significantly affected by various adverse factors. It is a unique organ that combines external and internal secretory activity, playing an important role in the carbohydrate metabolism of the human body. Today, diseases of the pancreas are one of the most important problems in medicine. Established dystrophic changes of the gland, which were expressed by the formation of fibrous fields, the thickness of the capsule and interlobular connective tissue between the lobes visually increased, a decrease in the area of acini and exocrinocytes was observed, acini lost clear contours and shape due to swelling of the connective tissue. Exocrinocytes of acini lost apical eosinophilia and basal basophilia of the cytoplasm, there was a decrease in the area of the nuclei, which were changed in shape and location. The introduction of triptorelin causes destructive-degenerative changes in the exocrine apparatus of the pancreas, which are characterized by compensatory mechanisms in the early periods of observation, and in the later periods – dystrophic changes in the structural components of the gland are determined, which corresponds to the stage of partial decompensation, a decrease in the activity and functionality of the cellular component of the exocrine apparatus of the pancreas.

Key words: testosterone deprivation, pancreas, acini, exocrinocytes.

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МОРФОФУНКЦІОНАЛЬНА ХАРАКТЕРИСТИКА СТРУКТУРНИХ КОМПОНЕНТІВ ЕКЗОКРИННОЇ ЧАСТИНИ ПІДШЛУНКОВОЇ ЗАЛОЗИ ПРИ ХІМІЧНІЙ КАСТРАЦІЇ

Впродовж усього життя людини підшлункова залоза, яка є другою за розміром (після печінки) залозою, що піддається значному впливу різних несприятливих факторів. Це унікальний орган, який поєднує зовнішню і внутрішню секреторну діяльність відіграючи важливу роль у вуглеводному обміні організму людини. На сьогодні захворювання підшлункової залози є однією з найважливіших проблем у медицині. Встановлені дистрофічні зміни залози, які були виражені шляхом формування фіброзних полів, товщина капсули та міжчасточкової сполучної тканини між часточками візуально збільшувалася, спостерігали зменшення площі ацинусів і екзокриноцитів, ацинуси втрачали чіткі контури та форму за рахунок набряку сполучної тканини. Екзокриноцити ацинусів втрачали апікальну еозинофілію та базальну базофілію цитоплазми, спостерігалося зменшення площі ядер, які були змінені за формою та розташуванням. Введення трипторелін викликає деструктивні-дегенеративні зміни в екзокриному апараті підшлункової залози, що характеризуються на ранніх термінах спостереження компенсаторними механізмами, а на пізніх термінах ‒ визначаються дистрофічні зміни структурних компонентів залози, що відповідає стадії часткової декомпенсації, зменшенню активності та функціональності клітинної складової екзокринного апарату підшлункової залози.

Ключові слова: депривація тестостерону, підшлункова залоза, ацинуси, екзокриноцити.

The study is a fragment of the research project "Experimental and morphological study of the effect of cryopreserved preparations of cord blood and embryofetoplacental complex (EFPC), diferelin, ethanol and 1 % methacrylic acid ether on the morphofunctional state of a number of internal organs", state registration No. 0119U102925.

The pancreas is the most important organ of the human digestive system. Structurally, it consists of two parts, different in terms of morpho-functionality – exocrine and endocrine, the first component of the gland significantly predominates in the lobules. The exocrine function in the gland is related to the synthesis and release of enzymes involved in the disintegration of proteins, carbohydrates and fats [3].

In practice, in the treatment of prostate cancer by chemical castration, triptorelin embonate is used, which indirectly affects the structural components of the exocrine part of the pancreas [7]. It has a fairly quick and effective influence on the hypothalamic-pituitary-gonadal axis at the highest level of its regulation. In addition, the biphasic mechanism of action permits the use of two opposite therapeutic effects on the reproductive system: stimulating during short-term administration and inhibitory during long-term use. Gonadotropin-releasing hormone is a regulator of sexual and other endocrine glands, which remains a relevant object of modern scientific research [6, 12].

The study will permit to explore in more detail the causes of diseases associated with morphological changes of the gland, as a result of which the functioning of the organ is impaired, and to develop possible ways of correcting this pathological condition.

The purpose of the study was to establish the morphofunctional changes in the structural components of the exocrine part of the pancreas during central chemical castration.

Materials and methods. The work was performed on 35 sexually mature male white outbred rats, which were divided into 4 groups: group I consisted of 5 intact animals, group II (1 month), group III (3 months), group IV (6 months) consisted of 10 animals for each term, which were injected subcutaneously with "Diferelin" (triptorelin embonate, IPSEN PHARMA, France), at a dose of 0.3 mg/kg of the active substance per animal for 180 days [8].

The animals were kept under standard conditions in the vivarium of Poltava State Medical University. The euthanasia of experimental animals took place at the appropriate times – on the 1st, 3rd and 6th months, (n=30) by overdose of ether anesthesia and was carried out in strict accordance with the provisions of the "European Convention for the Protection of Vertebrate Animals Used in Experimental and for other scientific purposes"; (Strasbourg, 1986), as well as in accordance with the "General ethical principles of animal experiments", adopted by the First National Congress on Bioethics (Kyiv, 2001) [8].

The material for microscopic examination of the organ was taken immediately after euthanasia of the animals, embedded in paraffin, according to the generally accepted method, and semi-thin sections were made, which were stained with hematoxylin and eosin [1].

The morphometric study of histologic specimens and their photomicrography was carried out using a Biorex-3 VM-500T microscope with a DCM 900 digital microphoto nozzle with programs adapted for the research data. The mean cross-sectional diameters of the acini were measured, the number and height of the exocrinocytes forming the acinus wall, and the diameter of their nucleus were measured.

Quantitative analysis of the results of morphometric study and statistical processing of morphometric data was carried out with generally accepted statistical methods using the Exel software [4].

Results of the study and their discussion. The results of the performed microscopic examination of an intact group of animals showed that the pancreas in rats is covered by a capsule of dense, irregular connective tissue, from which connective tissue fibers depart into the gland, dividing the parenchyma into lobules of different sizes. Blood vessels, nerve fibers and excretory interlobular ducts of the gland were found in the interlobular layers of the connective tissue.

In the intact group of rats, the exocrine part in the form of acini and ducts of various diameters significantly prevailed. The acini consisted of 8_10 cone-shaped cells, which were located on the basement membrane, in the center of the acinus, several small centroacinous cells were visualized, granularity was observed in their cytoplasm, the nucleus was located basally, where the granularity was expressed to a lesser extent. Microscopically, it was found that exocrinocytes had basally located 1_2 nuclei, their basal part was homogeneous and stained more intensely than the apical part, which had a granular appearance.

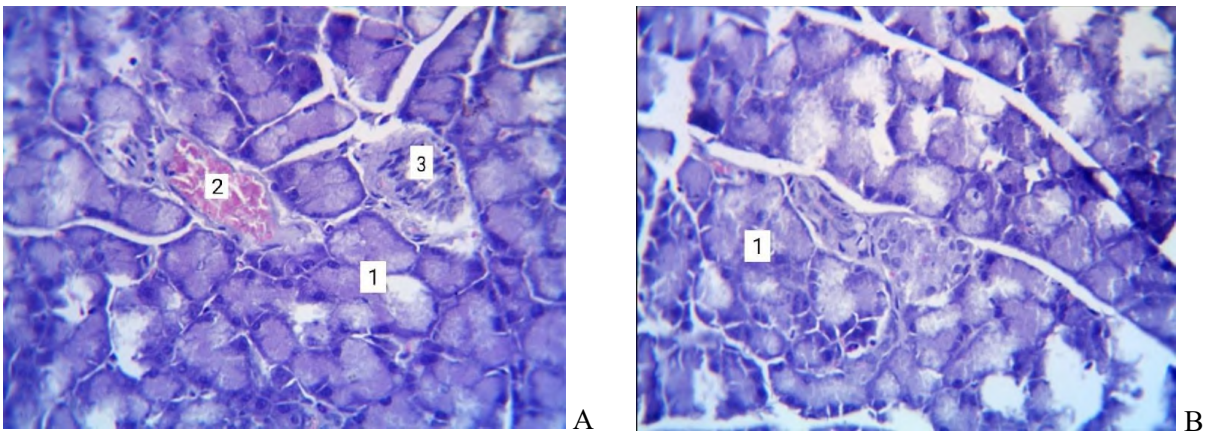
When examining the sectioned slides in the IInd group of animals, which were simulated pathological condition in the 1st month, microscopically, dystrophic changes in the structure of the pancreas were established, which were characterized by visual changes in both the parenchymal and stromal components. The thickness of the capsule and interlobular connective tissue of the pancreas compared to the previous term visually decreased, the lobular structure of the gland was preserved. There were changes in the acini, which were manifested in a decrease in their area, compared to the intact group of animals, and the basal membrane was swollen in some places.

Among the exocrinocytes, destructively changed forms were observed, their shell had a blurred outline, the demarcation into homogeneous and zymogenous zones was not visualized, and few zymogen granules were observed in the apical part. Nuclei of exocrinocytes were intensely basophilically stained with clear nucleoli. The excretory ducts were determined by the expansion of the internal lumen, with a visual increase in their outer diameter, morphometric changes were characterized by a significant decrease in the areas of the exocrine part in the lobes from the terminal secretory departments (fig. 1B).

When studying histological preparations in the IIIrd experimental group of animals, normalization of the structural components of the exocrine part of the pancreas was observed. The capsule surrounded the organ and groups of acini with elements of the hemomicrocirculatory bed. The shape of the acini was quite diverse: round, oval, oblong-elongated. From the inside, the acini are lined with cone-shaped exocrinocytes, the more narrowed pole (tip) of them is directed to the center of the acinus, and the opposite expanded (base) – outwards. The cytoplasm of the cells had well-defined granularity, especially towards the apical pole. Nuclei were located at the base, where granularity was less pronounced, and contained nucleoli. The epithelium of the interlobular excretory ducts also underwent destructive changes, by thinning, single desquamated epitheliocytes were visualized in the lumens.

Such a cellular reaction of the structural components of the pancreas'es exocrine part in the 3rd month of the study testifies to the reactivation of the synthetic process in the cells and strengthening of the metabolic processes in them in comparison with the previous period of observation, and morphological changes in the cells of the acini with a violation of the adaptive mechanisms to pathological changes due to their own mechanisms (fig. 2 A).

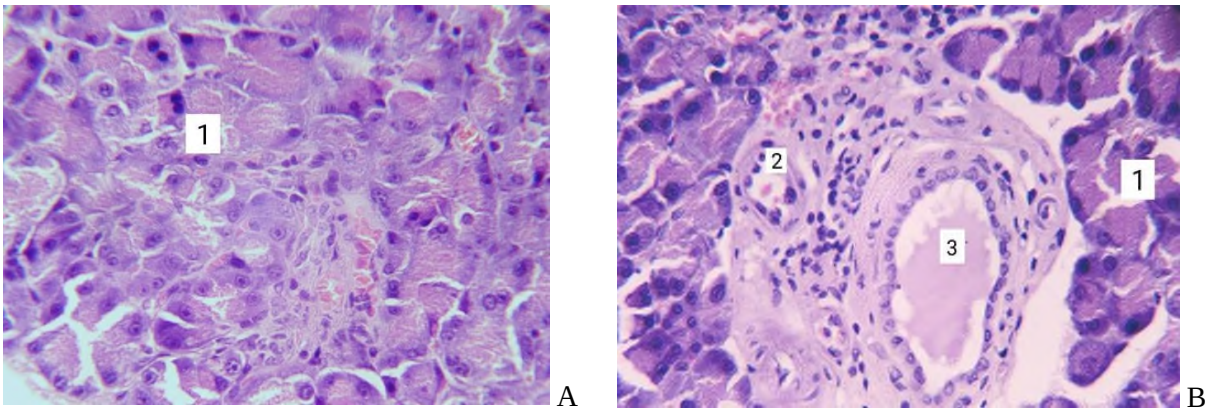
The IVth group of animals, in which the pathological process was simulated during the 6-month study, was characterized by a decrease in indices compared to both the previous periods of the experiment and the indices of the intact group of animals. Microscopically, dystrophic changes of the gland were established, which were more pronounced by the formation of fibrous fields that separated the lobes.



A B

Fig. 1. Exocrine part of the rat pancreas: A – intact group; B – the 1st month of the experiment. 1 – acini, 2 – vein, 3 – interlobular excretory duct. Hematoxylin and eosin staining. Magnification: approx.:10; vol. 40

The thickness of the capsule and interlobular connective tissue between the lobules of the pancreas, compared to the previous term, visually increased, accordingly, a decrease in the area of acini and exocrinocytes was observed, acini lost clear contours and shape due to swelling of the connective tissue. Exocrinocytes of acini lost apical eosinophilia and basal basophilia of the cytoplasm, there was a decrease in the area of the nuclei, which were changed in shape and location (figs. 2B, 3).



A B

Fig. 2. The exocrine part of the rat pancreas: A – the 3rd month of the experiment; B – the 6th month of the experiment. 1 – acini, 2 – arteriole, 3 – interlobular duct. Hematoxylin and eosin staining. Magnification: approx.:10; vol. 40

Under the influence of triptorelin administration, there was a decrease in morphometric parameters, which was accompanied by a decrease in the area of acini, the area of the exocrinocytes' cytoplasm , the area of their nuclei, and the thinning of the interlobular excretory ducts' epithelium, both in comparison with the previous group of animals and with intact animals, which corresponds to decompensation, namely, a decrease in activity and functionality of its cellular component. At the same time, wide fields were formed that separated the particles associated with the adaptive reaction of the pancreas'es structural components.

The processes of decreasing the area of acini by 1.93 %, nuclei of exocrinocytes by 34.85 %, cross-sectional area of the cytoplasm by 19.92 % of the pancreas in the IVth experimental group of animals compared to the intact group of animals indicate a decrease in the functional activity of the exocrine apparatus of the gland with pronounced fibrous and atrophic changes.

The data we obtained regarding the decrease in the area of the exocrinocytes' cytoplasm when triptorelin embonate is administered leads to a decrease in the area of the structural components of the exocrine part of the pancreas, which is consistent with the data in earlier studies, where the reaction of acini was observed, which changed their shape from rounded to slightly elongated, while the area of acini decreased by 32.64 %. The acinus cells themselves were characterized by dystrophic changes. The number of cells that form acini decreased by 10.40 %, which has a significant impact on the quantitative values of secretory activity aimed at ensuring functional activity, exocrinocytes dystrophically changed [2].

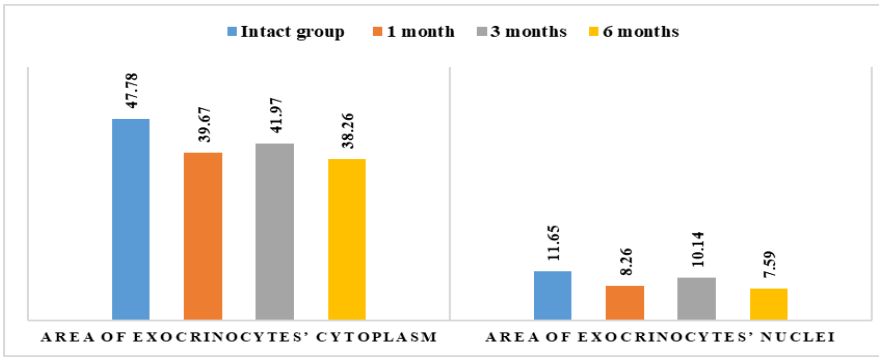


Fig. 3. Morphometric indices of the exocrine part of the pancreas in the experiment.

The conducted experimental study of the exocrine apparatus of the pancreas affects the variability of the structural and functional parameters of exocrinocytes. The obtained results coincide with the literature data of other authors, which set out the basic principles and regularities of the pancreas cells population and changes in them [5], showed significant swelling and an increase in the area of the connective tissue component of the gland. Growth of inter-acinar connective tissue with increasing fibrous changes was observed. There were areas of atrophy of the exocrine part of the organ's parenchyma, and in some places, for months, exocrinocytes were located chaotically. The area of acini and, accordingly, pancreatocytes decreased visually. The most characteristic feature was the loss of apical eosinophilia and basal basophilia of the cytoplasm, pyknosis of the nuclei and their irregular arrangement.

Thus, androgens affect the system of the gastrointestinal tract, which leads to specific differences in the manifestations of many diseases depending on gender. The reaction to the introduction of triptorelin in our experiment on the 1st month of the experiment shows a decrease in the activity and functionality of the cells of the exocrine apparatus of the pancreas. Thus, the diameters in exocrinocytes and the diameter of their nuclei are significantly reduced. As in another phase I study, the safety, efficacy, and pharmacodynamics of androgen deprivation with the gonadotropin-releasing hormone (GnRH) agonist triptorelin in combination with nivolumab were evaluated in male patients with anti-PD-1-resistant melanoma [9, 10, 11].

Conclusion

Triptorelin causes changes in the structure of the pancreas, and actually in the exocrine apparatus. The 1st and 3rd months of observation are characterized by compensatory mechanisms in the cells and tissues of the pancreas when they are damaged in comparison with the intact group of animals. On the 6th month of observation, dystrophic changes of exocrinocytes are determined, which corresponds to the stage of partial decompensation, a decrease in the activity and functionality of the cellular component of the exocrine apparatus of the pancreas.

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Стаття надійшла 20.01.2022 р.