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COMPARATIVE ANALYSIS OF CHANGES IN THE MORPHOLOGICAL AND MORPHOMETRIC PARAMETERS OF THE PANCREAS IN 9-MONTH-OLD WHITE OUTBRED RATS UNDER THE INFLUENCE OF EXPERIMENTAL ALCOHOL INTOXICATION, COMPARED WITH THE CONTROL GROUP

[1]Akhtamov Azizbek Avaz ugli,
[2]Radjabov Akhtam Boltaevich
[1]PhD., Associate Professor, Department of Anatomy, Clinical Anatomy, Bukhara State Medical Institute named after Abu Ali ibn Sino, City of Bukhara, 200118, Gijduvan street, 23, Uzbekistan
[2]DSc., Professor, Head of the Department of Anatomy, Clinical Anatomy, Bukhara State Medical Institute named after Abu Ali ibn Sino, City of Bukhara, 200118, Gijduvan street, 23, Uzbekistan
E-mail: azizbekakhtamov@mail.ru

Abstract

Background: Alcohol intoxication remains a major etiological factor contributing to structural and functional impairment of the pancreas due to its high sensitivity to toxic influences. Morphological and morphometric evaluation is essential for understanding the degree of tissue damage and adaptive changes under toxic conditions.

Objective: To perform a comparative analysis of morphological and morphometric changes in the pancreas of 9-month-old white outbred rats under experimental alcohol intoxication versus a control group.

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Materials and Methods: The study was conducted on 30 white outbred rats divided into control (n=15) and experimental (n=15) groups. Alcohol intoxication was induced by daily administration of 40% ethanol (7 g/kg body weight) via gastric tube for 30 days, combined with replacement of drinking water with a 5% ethanol solution. Pancreatic tissues were processed using hematoxylin-eosin and Van Gieson staining. Morphological and morphometric parameters were evaluated using standard histological methods.

Results: Experimental alcohol exposure caused pronounced structural remodeling of the pancreas. In the exocrine part, acinar disorganization, reduction of acinar diameter, and decreased acinar area were observed. Exocrinocytes demonstrated atrophic and dystrophic changes with an increased nucleus-to-cytoplasm ratio, indicating reduced synthetic activity. Fibrotic changes with increased collagen deposition were also detected. In the endocrine portion, a significant decrease in Langerhans islet area, number, and endocrine cell density was observed, accompanied by vacuolization, pyknosis, and karyorrhexis.

Conclusion: Experimental alcohol intoxication induces significant morphological and morphometric alterations in the pancreas, characterized by degeneration of exocrine and endocrine components, increased fibrosis, and activation of apoptotic and necrobiotic processes. These changes lead to impaired pancreatic structural integrity and functional activity, providing important insights into the pathogenesis of alcohol-induced pancreatic damage.

Keywords: Pancreas, alcohol intoxication, morphometry, acinus, exocrinocyte, endocrinocyte.

Introduction

Alcohol intoxication remains one of the significant causes of structural and functional disorders of the pancreas, which indicates the high sensitivity of this gland to toxic influences [1]. In recent years, at the modern stage of

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morphological science development, particular attention has been paid to the comprehensive analysis of morphological and morphometric changes that reflect the degree of damage to tissue and cellular structures [2,11].

The pancreas is a complex parenchymal organ consisting of an exocrine acinar portion and an endocrine islet apparatus. Acinar cells are characterized by high secretory activity and are distinguished by a well-developed rough endoplasmic reticulum and zymogen granules located in the apical part of the cell [3,13]. Under ethanol exposure, ultrastructural changes such as disorganization of membrane organelles, dilation of endoplasmic reticulum cisternae, and a decrease in the number of mitochondria are observed in pancreatic cells [4].

During chronic alcohol intoxication, cytological changes manifest as cytoplasmic vacuolization, karyopyknosis, karyorrhexis, and karyolysis, indicating the development of apoptotic and necrobiotic processes in pancreatic cells [15]. At the same time, disruption of cell polarity, reduction of zymogen granules, and decreased secretory activity are observed, reflecting functional insufficiency of the exocrine apparatus [12].

One of the key pathogenetic factors is oxidative stress, which leads to lipid peroxidation of cell membranes, damage to the cytoskeleton, and disruption of nucleus-cytoplasm interactions [8]. This process is accompanied by activation of intracellular signaling pathways that induce apoptosis and necrosis, morphologically manifested by destruction of acinar structures and formation of pancreatic necrosis foci [12,18].

Under stress conditions, acinar structures undergo fragmentation, accompanied by interstitial edema, stromal infiltration, and fibrosis [7,14]. An increase in connective tissue proportion leads to disruption of parenchymal architecture, impairment of organ organization, and reduction of functional elements [6].

In morphological studies, morphometric analysis of the organ is of particular importance, as it allows quantitative assessment of pathological changes. The most informative parameters include acinar area, acinar cell diameter and area,

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nucleus-to-cytoplasm ratio, area of Langerhans islets, and relative proportion of stromal components [4,10,19]. A decrease in these parameters indicates progression of parenchymal atrophy and reduced functional activity of the organ. Under toxic influence, endocrine structures also undergo significant changes, including a decrease in β-cell number, nuclear structural abnormalities, hypochromia, and reduced cytoplasmic granularity [16,20]. Morphometrically, a reduction in the area of Langerhans islets reflects impaired hormonal regulation of carbohydrate metabolism.

The age factor is also important in determining the degree of morphological changes. In 9-month-old rats, corresponding to mature age, tissue regenerative capacity decreases, cell renewal slows down, and dystrophic processes increase, leading to pronounced structural damage under toxic influence [5,17].

Despite numerous studies on alcohol-induced pancreatic injury, the interrelationship between morphological and morphometric changes during intoxication has not been sufficiently investigated [6,11]. A comprehensive evaluation of these parameters allows for a deeper understanding of the mechanisms of organ damage and adaptation.

Aim of the study

To perform a comparative analysis of morphological and morphometric changes in the pancreas of 9-month-old white outbred rats under experimental alcohol intoxication versus the control group.

Materials and methods

The study was conducted on 30 white outbred rats, divided into two groups: control group (n=15) and experimental group (n=15). Laboratory animals were kept under standard recommended conditions (Zapadnyuk I.P., Zapadnyuk V.I., Zakharija E.A., 1983) in special cages. Each cage contained information about birth date, body weight, start of experiment, and age of experimental animals.

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In the experimental group, alcohol intoxication was modeled using forced alcohol administration. A 40% ethanol solution was used (Sidorov P.I., 2002), administered once daily via a gastric tube at a dose of 7 g/kg body weight for 1 month.

In addition, an artificial polydipsia model was applied: drinking water was replaced with a 5% ethanol solution (Knyshova L.P., 2016). The solution was sweetened with sucrose (5 g sugar per 100 ml of 5% ethanol solution).

At the end of the experiment, animals were euthanized under ether anesthesia (Sidorov P.I., 2002; Rybakova A.V., 2015; Koptyaeva K.E., 2018). Obtained specimens were stained using hematoxylin-eosin and Van Gieson methods.

Results

The morphological and morphometric changes in the pancreas of 9-month-old rats under 30-day experimental alcohol intoxication were analyzed in comparison with the control group. Histological examination of sections stained with hematoxylin-eosin and Van Gieson revealed complex remodeling processes in pancreatic tissue.

In the exocrine part, disorganization of acinar structures, loss of clear acinar boundaries and shape integrity, as well as altered parenchyma-to-stroma ratio were observed. In the control group, the relative area of exocrine parenchyma was 73.0 ± 1.5%, whereas in the experimental group this parameter significantly decreased to 65-67%, which was associated with a relative increase in stromal components.

Morphometric analysis of acini demonstrated a reduction in their volumetric parameters. Specifically, acinar diameter decreased from 30.0 ± 0.7 μm to 25.0-27.0 μm, while acinar area decreased from 900.0 ± 35.0 μm² to an average of 700.0-750.0 μm². Exocrinocytes forming the acini showed predominant atrophic and dystrophic changes: epithelial height decreased from 12.0 ± 0.4 μm to 9.0-10.0 μm, and exocrinocyte area decreased from 124.0 ± 5.0 μm² to 100.0-105.0

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μm².

Analysis of nuclear and cytoplasmic parameters revealed an increased nucleus-to-cytoplasm ratio due to cytoplasmic reduction. In the control group, this ratio was 0.18 ± 0.01, whereas in the experimental group it increased to 0.20-0.22. This indicates a decrease in protein synthesis activity and intensification of degenerative processes. Additionally, the number of exocrinocytes per acinus decreased from 6.9 ± 0.2 to 5.0-6.0 cells.

Intralobular ducts showed a number of reactive changes. Their diameter increased from 12.0 ± 0.6 μm in the control group to 13.0-15.0 μm in the experimental group, and in some cases epithelial hyperplasia and desquamation were observed. Van Gieson staining clearly demonstrated an increase in collagen fibers within the stroma, confirming the development of fibrotic processes.

Analysis of the endocrine portion revealed reduction changes in the islet apparatus. The relative area of the endocrine part decreased from 2.2 ± 0.1% to 1.7–1.9%. The number of islets decreased from 1.55 ± 0.10 to 1.2-1.3 conventional units, indicating islet reduction and partial degeneration.

Morphometric parameters of the islets also changed significantly: islet area decreased from 13.6 ± 0.5 $\times 10^3$ μm² to 10.0-11.0 $\times 10^3$ μm², and diameter decreased from 118.0 ± 7.0 μm to 95.0-105.0 μm. The number of endocrine cells decreased from 102.0 ± 4.0 to 80.0-90.0, while their density decreased from 7.5 ± 0.3 $\times 10^{-3}$/μm² to 6.5–7.0 $\times 10^{-3}$/μm². A reduction in individual endocrine cell area was also observed (from 69.0 ± 3.0 μm² to 60.0-63.0 μm²).

Microscopically, indistinct cell boundaries within islets, cytoplasmic vacuolization, as well as nuclear pyknosis and karyorrhexis were identified. These findings indicate activation of apoptotic and necrobiotic processes in endocrine cells.

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Discussion

The obtained results confirm that experimental alcohol intoxication induces systemic morphological and morphometric changes in the pancreas. The observed disorganization of acinar structures in the exocrine portion, as well as atrophic and dystrophic alterations in exocrinocytes, can be explained by the direct cytotoxic effect of ethanol. These findings are consistent with the studies of Ivanov and Petrov (2019), as well as Sidorov (2020).

The reduction in acinar diameter and area, together with an increased nucleus-to-cytoplasm ratio, indicates a decline in synthetic activity within the cells. These changes are in agreement with the data reported by Kuznetsov (2021) and Zakharov (2025). The increase in stromal components and development of fibrosis, as noted by Fedorov (2024) and Garcia et al. (2022), reflects intensified sclerotic processes in pancreatic tissue.

In the endocrine portion, the reduction of islet structures and the decrease in endocrine cells correspond to β-cell dysfunction described by Kim and Park (2023). Furthermore, the enhanced oxidative stress (Lebedev, 2019; Wang and Chen, 2018) forms a key pathogenetic mechanism underlying the observed morphological alterations by promoting cellular destruction, apoptosis, and necrobiotic processes.

The age factor also plays an important role. As noted by Smirnov (2022), 9-month-old rats demonstrate reduced regenerative capacity, which increases their susceptibility to toxic influences. Overall, the findings indicate a predominance of destructive and fibrotic processes in the pancreas under alcohol intoxication.

Conclusion

Experimental alcohol intoxication induces complex morphological and morphometric changes in the pancreas of 9-month-old rats. These changes are characterized by disorganization of acinar structures in the exocrine part, reduction in acinar size, predominance of atrophic and dystrophic processes in

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exocrinocytes, increased stromal components, and development of fibrosis. Morphometric analysis demonstrates a decrease in acinar diameter and area, a reduction in the number of exocrinocytes, cytoplasmic shrinkage, and an increased nucleus-to-cytoplasm ratio, indicating impaired cellular metabolism and synthetic activity.

At the same time, in the endocrine part, a decrease in the number and area of Langerhans islets, reduction of endocrine cells, and the presence of necrobiotic changes such as vacuolization, pyknosis, and karyorrhexis suggest disturbances in hormonal regulatory mechanisms. The proliferation of collagen fibers and progression of fibrosis in the stroma lead to disruption of organ architecture, reduction of parenchymal elements, and decreased functional reserves.

Overall, the identified morphological and morphometric alterations are closely associated with the activation of oxidative stress, apoptosis, and destructive processes under conditions of alcohol intoxication. These changes impair both exocrine and endocrine functions of the pancreas and provide an important scientific basis for a deeper understanding of the pathogenesis of this condition.

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