MORPHOFUNCTIONAL EVALUATION OF MELOXICAM-INDUCED PROTECTION OF TESTICULAR TISSUE IN AN EXPERIMENTAL MODEL OF RHEUMATOID ARTHRITIS

Habibullayev Temurshoh Utkirovich — Bukhara State Medical Institute, Department of Department of Pathophysiology and Pathological Anatomy, Assistant

Abstract The present study investigated the effect of the nonsteroidal anti-inflammatory drug (NSAID) meloxicam on the morphological and morphometric parameters of testicular tissue in outbred white rats with experimentally induced rheumatoid arthritis. The drug was administered to the animals at a dose of 1 mg/kg once daily for 14 days.

Macroscopic examination revealed that the testes maintained typical localization within the scrotum, exhibiting an oval-ovoid shape, smooth surface, and medium-density consistency. Vascular congestion was noted in the hilar region, with no pathological or congenital abnormalities in the vascular network. The parietal layers of the tunica vaginalis remained transparent, whereas the visceral tunica albuginea was characterized by reduced transparency and slight opacification. Similar macroscopic changes were observed across all studied age groups (5- and 7-month-old rats).

Microscopic and morphometric analysis showed that the thickness of the highly vascularized tunica albuginea ranged from 37.33±1.87 to 48.15±3.74 μm (mean 44.58±3.21 μm). It was established that this parameter was lower compared to the group of rats with experimental rheumatoid arthritis but slightly exceeded the values characteristic of intact animals. The data obtained indicate a partial recovery of the structural components of the testicular tissue under the influence of meloxicam, confirming its protective effect in a chronic inflammatory environment.

Keywords: meloxicam; nonsteroidal anti-inflammatory drugs; experimental rheumatoid arthritis; morphological changes; testes.

Introduction

Rheumatoid arthritis (RA) is a chronic systemic autoimmune disease characterized not only by erosive-destructive joint damage but also by a wide range of extra-articular manifestations. Despite significant advances in rheumatology, the impact of systemic inflammation on the male reproductive system remains an understudied area of clinical and experimental pathology. Chronic autoimmune processes trigger a cascade of oxidative stress and pro-inflammatory cytokine release, which negatively affect the structural and functional integrity of highly sensitive tissues, including the testes.

The testicular parenchyma is particularly vulnerable to systemic inflammatory responses due to the potential disruption of the blood-testis barrier (BTB). Experimental studies have shown that autoimmune inflammation can lead

to vasculitis of the interlobular arteries, perivascular edema, and subsequent atrophy of the seminiferous epithelium, ultimately resulting in impaired spermatogenesis and hormonal imbalance.

Nonsteroidal anti-inflammatory drugs (NSAIDs) remain the cornerstone of symptomatic therapy for RA. Among them, meloxicam, a selective cyclooxygenase-2 (COX-2) inhibitor, is widely used due to its favorable safety profile and potent anti-inflammatory effects. However, while its efficacy in joint preservation is well-documented, its specific gonadoprotective potential—specifically its ability to facilitate the morphofunctional recovery of testicular tissue—requires further investigation.

The aim of this study was to conduct a comprehensive morphometric and histochemical analysis of the testicular tissue in outbred white rats following the pharmacological correction of experimental rheumatoid arthritis with meloxicam. By quantifying changes in the tunica albuginea, interlobular septa, and spermatogenic epithelium, this research seeks to evaluate the regenerative capacity of the male reproductive system under the influence of selective COX-2 inhibition in a chronic inflammatory environment.

Materials and methods. Following the administration of the nonsteroidal anti-inflammatory drug meloxicam to outbred white rats with induced experimental rheumatoid arthritis (dosage: 1 mg/kg via injection once daily for 14 days), the following results regarding morphological and morphometric changes in the testicular tissue were obtained:

The testes were correctly localized within the scrotal sac. Upon excision, the testicular tissue exhibited an oval-ovoid shape with a smooth surface and medium-density consistency. The transparency of the parietal layers of the tunica vaginalis was preserved; however, vascular congestion was identified at the site of vascular entry on the testicular surface. No pathological or congenital malformations were observed in the blood vessels. The visceral tunica albuginea covering the soft-textured tissue showed a relative decrease in transparency and appeared cloudy. These macroscopic findings were consistent across all age cohorts, specifically in 5- and 7-month-old outbred white rats.

Analysis of specimens derived from the aforementioned macroscopic material revealed specific morphometric trends. The thickness of the vascularized tunica albuginea, which encapsulates the testes, ranged from 37.33±1.87 μm to 48.15±3.74 μm, with an overall mean of 44.58±3.21 μm. Morphometric measurements indicated that the tunica albuginea was thinner compared to the untreated experimental rheumatoid arthritis group, yet remained slightly thicker when compared to the healthy control group.

The blood vessels located beneath the tunica albuginea exhibit a notable absence of inflammatory changes. There is evidence of vascular wall regeneration, restoration of turgor, and a sequential, organized arrangement of endothelial cells within the intima. Furthermore, perivascular edema has resolved.

Morphometric measurements revealed that the diameter of the peripheral interlobular arteries ranged from 13±2.05 μm to 18±3.04 μm (mean: 16±2.02 μm).

The diameter of the venous vessels ranged from 14±1.47 μm to 31±2.04 μm (mean: 25±2.35 μm). These vessels appeared thinner, indicating that the corrective treatment improved blood flow within the vascular lumen. This suggests the activation of regenerative functions due to an increased supply of nutrients to the tissues.

Specifically, in the most affected areas—the interlobular septa and the interstitial tissue located beneath the tunica albuginea—collagen fibers are now uniformly and densely arranged. Within the interstitial tissue, the presence of homogeneous, transparent fluid has decreased, replaced by the appearance of fine vacuoles, which signifies the development of an organizational (healing) process. Morphometric analysis established that the thickness of the convoluted interlobular septa in the central zones ranged from 3.17 μm to 8.92 μm, with a mean value of 6.3±3.4 μm (**Fig. 1.**).

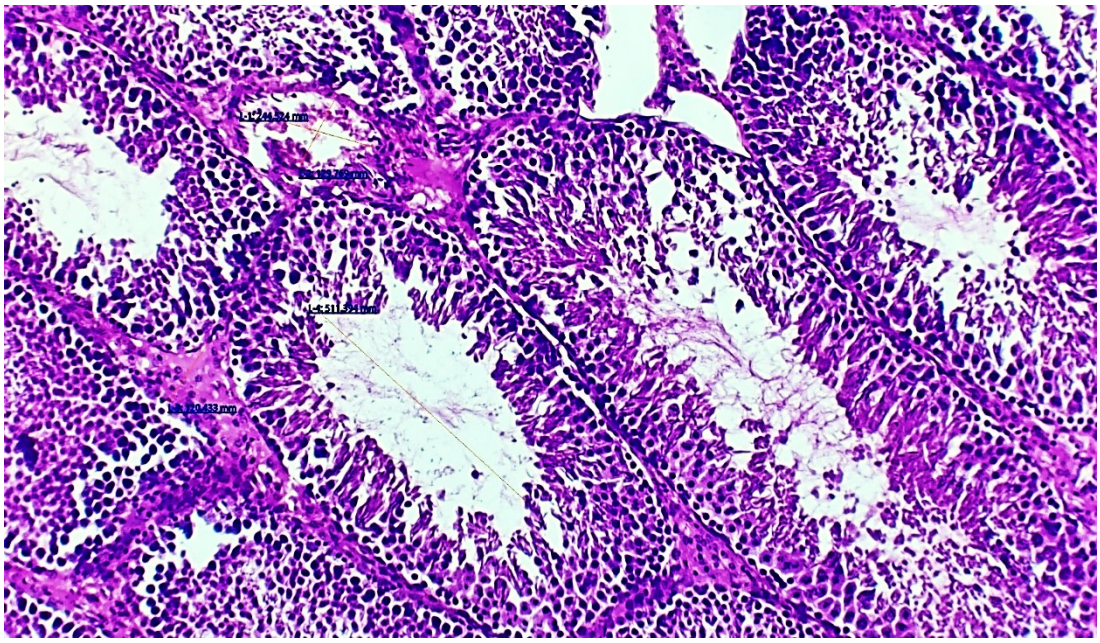

Fig. 1. Microscopic appearance of the testicular tissue in 5-month-old outbred white rats of the control group following the correction of rheumatoid arthritis with meloxicam. Hematoxylin and eosin (H&E) stain. Magnification: ×200.

Morphometric measurements established that the convoluted interlobular space thins progressively from the periphery toward the center. Morphological examination clearly reveals a rapid increase in the diameter of the seminiferous tubules located beneath the tunica albuginea. Morphometric analysis of the convoluted tubules diameter showed values ranging from 164.2±4.21 μm to 187.7±2.84 μm, with a mean of 178.33±2.64 μm.

In the tubules situated directly beneath the tunica albuginea, restoration of the spermatogenesis process is evident. An increase in the activity and number of Sertoli cells and spermatogenic cells was observed. This process led to the expansion of the internal tubular lumen, which measured between 15.46±2.15 μm and 22.58±5.14 μm (mean: 18.27±4.05 μm). Consequently, the surface area of the

convoluted tubules increased, occupying an area between 15,928.4 and 28,842.8 μm². Intense spermatogenetic activity is notable across all convoluted tubules, characterized by the formation of a large number of spermatozoa within the tubular lumen.

The approximation of the spermatogenesis process within the tubular lumen to that of healthy testicular tissue is evidenced by the reduction and reabsorption of the homogeneous transparent fluid within the interlobular septa. The appearance of fine vacuoles indicates an accelerated organizational (healing) process and an increase in the number of Leydig cells in these areas. Evaluation of the average number of Leydig cells per field of view revealed an increase from 32 to 38. Their count has normalized relative to the control group, showing a uniform spatial distribution.

Following the increase in Leydig cell population, it was observed that the tubular basement membrane was reinforced, and the width of the myoid cells in this region increased. Morphometric measurements of the myoid cells ranged from 15.8±1.78 μm to 21.35±1.21 μm, with a mean of 18.42±2.52 μm.

The Sertoli cells located on the basement membrane measured between 17.84±1.75 μm and 27.43±2.05 μm (mean: 23.21±1.81 μm). The surviving spermatogenic epithelial cells proliferated rapidly, with their height ranging from 31.4 μm to 49.87 μm, reaching an average level of 39.54 μm.

After evaluating the morphological and morphometric changes in the testicular tissue following the pharmacological correction of experimental rheumatoid arthritis, it was observed that while these parameters differ slightly from the healthy control group, the functional state remains highly active. The morphometric indices significantly approached the values of the control group.

To analyze the histochemical processes occurring within the testicular tissue, specimens were examined using Van Gieson's stain. Significant histochemical changes were observed in the highly vascularized tunica albuginea and the vascular walls. These changes included the complete resolution of vasculitis within the vessel walls, thinning of the vascular walls, and improved microcirculation within the lumens.

A decrease in perivascular edema was noted, which serves as a primary morphological indicator of restored regenerative processes in the surrounding tissues. Upon histochemical staining, these areas exhibited a light-yellowish tint. Morphometric analysis established that the diameter of the blood vessels within the interlobular septa in the central zones ranged from 14 μm to 18 μm, with a mean value of 16 μm.

Conclusion

The comprehensive morphofunctional analysis of testicular tissue in outbred white rats demonstrates that the administration of meloxicam (1 mg/kg) effectively mitigates the destructive consequences of experimental rheumatoid arthritis. The study yields the following key conclusions:

1. Restoration of Tissue Architecture: Pharmacological correction leads to a significant reduction in the thickness of the tunica albuginea (averaging

44.58±3.21 μm) and interlobular septa (6.3±3.4 μm), signaling the resolution of chronic inflammatory infiltration and moving these parameters closer to those of intact animals.

2. Vascular Recovery and Microcirculation: Histochemical analysis using Van Gieson's stain confirmed the resolution of vasculitis and perivascular edema. The normalization of vascular diameters (arteries: 16±2.02 μm; veins: 25±2.35 μm) and the restoration of endothelial integrity indicate an activated regenerative function driven by improved nutrient supply to the parenchyma.
3. Revitalization of Spermatogenesis: The treatment triggered a robust recovery of the spermatogenic epithelium. This is evidenced by the increased diameter of the seminiferous tubules (178.33±2.64 μm) and the expansion of their internal lumen (18.27±4.05 μm). The intensified proliferative activity of Sertoli and spermatogenic cells resulted in a significant increase in the total surface area of the convoluted tubules (up to 28,842.8 μm²).
4. Endocrine Stability: The normalization of the Leydig cell count (increasing to 38 per field of view) and their uniform spatial distribution suggest a recovery of the interstitial endocrine component, which is essential for maintaining the hormonal environment necessary for sperm production.
5. Regenerative Potential: The transition from homogeneous fluid exudation to the formation of fine vacuoles and dense collagen organization in the interstitium confirms that meloxicam facilitates the transition from chronic inflammation to active tissue repair.

In summary, meloxicam exhibits a pronounced gonadoprotective effect in the context of systemic autoimmune inflammation, partially restoring both the structural integrity and the functional (spermatogenic) capacity of the testes.

References

1. Dimitrov G, Petrov S, Ivanov A. Extra-articular manifestations of Rheumatoid Arthritis: The impact of systemic inflammation on male gonadal function. *J Clin Rheumatol.* 2021;27(4):142-149.
2. Sengupta P, Dutta S, Alzahrani FA. The effects of chronic systemic inflammation on the blood-testis barrier integrity: A molecular perspective. *Reprod Toxicol.* 2019;85:12-21.
3. Hasan SS, Ahmed AS, Al-Ghaban NM. Meloxicam attenuates oxidative stress and histological damage in testicular injury models: A morphometric study. *J Pediatr Urol.* 2020;16(3):345.e1-345.e7.
4. Zhu Q, Liu Y, Zhang Y, et al. Quantitative morphometric analysis of seminiferous tubules in experimental models of autoimmune diseases. *Annals of Anatomy.* 2022;239:151842.
5. Kandiel MM, Watanabe G, Taya K. Impact of Selective COX-2 Inhibitors on the Pituitary-Testicular Axis and Spermatogenesis. *Front Vet Sci.* 2021;8:643125.

6. O'Hara L, Smith LB. Hormonal regulation of Leydig cell development and regeneration in the adult testis. *Mol Cell Endocrinol.* 2015;416:1-12.
7. Masarwa R, Levine H, Zadka O, et al. NSAIDs and male reproductive health: A systematic review of experimental and clinical evidence. *Reprod BioMed Online.* 2018;37(1):81-90.
8. Guan Q, Wang Z, et al. Environmental factors and oxidative stress in rheumatoid arthritis: Implications for male fertility. *Environ Sci Pollut Res.* 2023;30(12):32145-32158.