UDK: 616.37:612.61+611.018

MORPHOLOGICAL AND MORPHOMETRIC EVALUATION OF STRUCTURAL CHANGES IN THE STOMACH OF EXPERIMENTAL ANIMALS WITH INDUCED COLITIS

Sanoyev Bakhtiyor Abdurasulovich - C.M.Sc., associate professor
Hamroyev Bobir Bakhtiyorovich - independent researcher
Bukhara state Medical University (Bukhara, Uzbekistan)

Abstract: *This scientific study investigates the morphological and morphometric alterations in the gastric mucosa under experimental ulcerative colitis (UC) conditions. An ulcerative colitis model was induced in laboratory rats using dextran sulfate sodium (DSS). Gastric tissue samples were collected and examined using standard histological techniques. Morphological analysis revealed erosions, glandular atrophy, inflammatory infiltration, and signs of epithelial regeneration in the gastric mucosa. Morphometric evaluation included measurements of mucosal thickness, glandular density, crypt depth, and the nucleus-to-cytoplasm ratio. Statistically significant differences were observed between the UC group and the control group, indicating that ulcerative colitis induces systemic effects not limited to the colon, but also involving upper gastrointestinal organs such as the stomach. The proposed method of combined morphological and morphometric assessment may serve as a reliable approach for better understanding the pathogenesis and early diagnosis of ulcerative colitis.*

Key words: *ulcerative colitis, gastric mucosa, DSS model, morphological changes, morphometry, laboratory experiment, inflammation*

KOLITGA INDUKTSIYALANGAN TAJRIBA HAYVONLARIDA OSHQOZON STRUKTURAVIY O‘ZGARISHLARINI MORFOLOGIK VA MORFOMETRIK BAHOLASH

Sanoyev Baxtiyor Abdurasulovich - t.f.n., dotsent
Hamroyev Bobir Baxtiyorovich - mustaqil izlanuvchi
Buxoro davlat tibbiyot instituti (Buxoro, O‘zbekiston)

Annotatsiya: *Mazkur ilmiy ishda eksperimental yarali kolit (YK) modelida oshqozon shilliq qavatida yuzaga keladigan morfologik va morfometrik o‘zgarishlar o‘rganildi. Tadqiqotda DSS (dekstran sulfat natriy) yordamida yarali kolit modeli yaratilib, laborator zichchalarda eksperiment o‘tkazildi. Histologik usullar asosida oshqozonning shilliq qavati mikroskopik darajada tahlil qilindi. Morfologik o‘rganishda shilliq qatlamda eroziya, glandular atrofiyalar, yallig‘lanish infiltratsiyasi va regenerativ jarayonlar aniqlandi. Morfometrik o‘lchovlar orqali shilliq qavat qalinligi, glandular zichlik, kript chuqurligi va yadro-sitoplazma nisbati aniqlandi hamda ular orasidagi statistik farqlar tahlil qilindi. Natijalar shuni ko‘rsatdiki, YK faqat kolonda emas, balki yuqori GI trakt, ya’ni oshqozonda ham tizimli o‘zgarishlarga olib keladi. Taklif etilgan morfologik va morfometrik baholash usullari yarali kolitning diagnostik va patogenetik mexanizmlarini chuqurroq o‘rganishda samarali vosita hisoblanadi.*

Kalit so‘zlar: *yarali kolit, oshqozon shilliq qavati, DSS modeli, morfologik o‘zgarishlar, morfometriya, laborator tajriba, yallig‘lanish*

МОРФОЛОГИЧЕСКАЯ И МОРФОМЕТРИЧЕСКАЯ ОЦЕНКА СТРУКТУРНЫХ ИЗМЕНЕНИЙ ЖЕЛУДКА У ЭКСПЕРИМЕНТАЛЬНЫХ ЖИВОТНЫХ С ИНДУЦИРОВАННЫМ КОЛИТОМ

Саноев Бахтиёр Абдурасулович - д.м.н., профессор
Хамроев Бобир Бахтиярович - независимый исследователь
Бухарский государственный медицинский институт (Бухара, Узбекистан)

Аннотация: *В данной научной работе исследованы морфологические и морфометрические изменения слизистой оболочки желудка в условиях экспериментального язвенного колита. Модель язвенного колита была индуцирована с помощью декстансульфата натрия (DSS) у лабораторных крыс. Гистологическим методом проведено микроскопическое изучение слизистой оболочки желудка. Морфологический анализ выявил наличие эрозий, атрофических изменений желез, воспалительных инфильтратов и признаки регенерации. Морфометрические измерения включали определение толщины слизистой оболочки, плотности желез, глубины крипт и соотношения ядро/цитоплазма. Полученные*

данные продемонстрировали статистически значимые изменения в группе с язвенным колитом по сравнению с контролем. Это подтверждает, что язвенный колит вызывает не только локальные, но и системные изменения, затрагивающие верхние отделы желудочно-кишечного тракта. Представленный метод морфологической и морфометрической оценки может быть полезным инструментом для патогенетического изучения и ранней диагностики язвенного колита.

Ключевые слова: *язвенный колит, слизистая желудка, модель DSS, морфологические изменения, морфометрия.*

Introduction.

Inflammatory bowel diseases (IBD), particularly ulcerative colitis (UC), have emerged in recent years as one of the most pressing challenges in global healthcare. These chronic inflammatory conditions not only affect the structural and functional integrity of the intestinal tract but may also extend their pathological impact to other organs involved in the digestive process, including the stomach. Both clinical observations and experimental studies on animal models have demonstrated that, against the background of ulcerative colitis, the stomach may undergo a range of pathological changes such as inflammatory infiltrates, erosions, mucosal atrophy, and dysplasia of epithelial cells.

Furthermore, it has been shown that the systemic increase in inflammatory markers and pro-inflammatory cytokines associated with the pathogenesis of UC can adversely affect the gastric mucosa, potentially exacerbating tissue damage and altering normal gastric function. These findings suggest a broader systemic impact of UC beyond the colon itself and underscore the importance of studying associated changes in other digestive organs.

Given these considerations, it becomes crucial to evaluate the morphological and morphometric alterations occurring in the gastric tissues under conditions of experimentally induced colitis. Such investigations not only enhance our understanding of the multisystemic nature of IBD but also contribute to the development of more comprehensive therapeutic approaches aimed at protecting and restoring the structure and function of the gastrointestinal tract as a whole.

Materials and Methods.

For our experiment, a total of 30 nine-month-old albino (non-breed-specific) rats of both sexes were selected. All laboratory rats were housed together in a single vivarium under controlled environmental conditions. The animals were provided with an adequate supply of water and a balanced nutritional diet. In preparing and conducting the experimental research, special attention was paid to the proper care and maintenance of the laboratory animals, as this is a critical factor influencing the reliability of experimental outcomes.

Strict adherence was maintained to the feeding schedule and dietary regimen, and hygienic protocols were observed during feeding times to ensure the well-being of the animals.

The rats were divided into two groups based on the design of the experiment:

- **Group I (Control Group - Intact):** This group consisted of albino rats that were fed a standard laboratory diet without any intervention. These animals served as the control group, providing a baseline for comparison with the experimental group.
- **Group II (Experimental Group):** In this group, experimental ulcerative colitis was induced by rectally administering 1 ml of an 8% acetic acid solution via a special probe once daily for 20 consecutive days. This method aimed to mimic conditions of chemically induced ulcerative colitis in laboratory rats. *(See Table 1 below for animal distribution.)*

Group (n-control, t-experimental)	Description of Experimental Condition	Age of Animals	Total Number of Animals (*Number of Deaths)
I (n) – Control	Standard diet, no intervention	9 months	15
II (t) – Experimental	Ulcerative colitis induced via rectal administration of 1 ml 8% acetic acid for 20 days	9 months	15 (*5)
Total			30 (*5)

The experimental animals were euthanized at appropriate time points during the study. Euthanasia was carried out in the morning hours on an empty stomach using brief decapitation under ether anesthesia, ensuring minimal suffering.

After dissection, the abdominal cavity was opened, and the large intestine and stomach were carefully isolated. Subsequently, the width of the stomach was measured using a millimeter ruler to assess morphological changes. The human stomach is typically divided into the cardiac and pyloric regions, the body in between them, and the fundus or dome-shaped region. In male albino rats, the majority of the stomach lies beneath the liver. The rat's stomach is firmly secured in place by several ligaments: superiorly by the phrenic-gastric ligament,

inferiorly by the pancreatic-gastric ligament, medially (at the lesser curvature) by the hepato-gastric ligament, and laterally (at the greater curvature) by the gastro-splenic and gastro-colic ligaments.

The stomach of the male albino rat shares the same anatomical divisions as the human stomach; however, in rats, the most prominent part is the fundus, which connects to the abdominal portion of the esophagus at the middle of the short lesser curvature. The rat stomach is more curved and resembles a hook in shape. It forms an angle with the duodenum, opening caudally.

After isolating the stomach from its surrounding ligaments and tissues, we measured the organ's total length, width, thickness, and the lengths of both the lesser and greater curvatures using a Vernier caliper (shtanger cirkul). These measurements were recorded systematically in a laboratory notebook.

The following methods were applied in our research: **organometric, histomorphometric, microscopic, and statistical** analyses.

Morphometric indicators were examined from histological preparations, and the thickness of the stomach walls was measured using an ocular micrometer. For microscopy, we used a trinocular microscope manufactured in China, model **DN-107t / NLSD-307b (Roman, China)**.

From the **cardiac, fundic, pyloric, and body** regions of the stomach, we selected and analyzed the following morphological parameters:

- Height of the gastric mucosa (μm)
- Height of the gastric folds (μm)
- Depth between gastric folds (μm)
- Thickness of the submucosal layer (μm)
- Thickness of the muscular layer (μm)
- Total thickness of the gastric wall ($x10^3$ μm^2)
- Glandular tissue height (μm)

These parameters were measured in preparations obtained from both the control group and the experimental group of albino rats with induced ulcerative colitis.

In rats with experimentally induced ulcerative colitis, histological evidence of colitis was first confirmed using preparations obtained from the large intestine.

Morphological data collected during the research were processed mathematically using the general matrix of the Microsoft Office data package in **Excel 7.0** on a Pentium-IV personal computer. The **"STTGRAF 5.1"** software was used to calculate standard deviations and standard errors.

The human stomach is typically divided into the cardiac and pyloric regions, the body in between them, and the fundus or dome-shaped region. In male albino rats, the majority of the stomach lies beneath the liver. The rat's stomach is firmly secured in place by several ligaments: superiorly by the phrenic-gastric ligament, inferiorly by the pancreatic-gastric ligament, medially (at the lesser curvature) by the hepato-gastric ligament, and laterally (at the greater curvature) by the gastro-splenic and gastro-colic ligaments.

The stomach of the male albino rat shares the same anatomical divisions as the human stomach; however, in rats, the most prominent part is the fundus, which connects to the abdominal portion of the esophagus at the middle of the short lesser curvature. The rat stomach is more curved and resembles a hook in shape. It forms an angle with the duodenum, opening caudally.

After isolating the stomach from its surrounding ligaments and tissues, we measured the organ's total length, width, thickness, and the lengths of both the lesser and greater curvatures using a Vernier caliper (shtanger cirkul). These measurements were recorded systematically in a laboratory notebook.

The following methods were applied in our research: **organometric, histomorphometric, microscopic, and statistical** analyses.

Morphometric indicators were examined from histological preparations, and the thickness of the stomach walls was measured using an ocular micrometer. For microscopy, we used a trinocular microscope manufactured in China, model **DN-107t / NLSD-307b (Roman, China)**.

From the **cardiac, fundic, pyloric, and body** regions of the stomach, we selected and analyzed the following morphological parameters:

- Height of the gastric mucosa (μm)
- Height of the gastric folds (μm)
- Depth between gastric folds (μm)
- Thickness of the submucosal layer (μm)
- Thickness of the muscular layer (μm)
- Total thickness of the gastric wall ($x10^3$ μm^2)
- Glandular tissue height (μm)

These parameters were measured in preparations obtained from both the control group and the experimental group of albino rats with induced ulcerative colitis.

In rats with experimentally induced ulcerative colitis, histological evidence of colitis was first confirmed using preparations obtained from the large intestine.

Morphological data collected during the research were processed mathematically using the general matrix of the Microsoft Office data package in **Excel 7.0** on a Pentium-IV personal computer. The **"STTGRAF 5.1"** software was used to calculate standard deviations and standard errors.

Results

The stomach is entirely covered by the peritoneum. Anatomically, it consists of four regions: the cardiac, fundus, body, and pyloric parts. Histologically, the stomach wall is composed of the following layers:

1.Tunica mucosa – mucosal layer

2.Tela submucosa – submucosal layer

3.Tunica muscularis – muscular layer

4.Tunica serosa – serous layer

The mucosal layer (tunica mucosa) lines the inner surface of the stomach and is covered by a single-layer columnar epithelium. The color of the mucosa ranges from pale pink to dark red depending on the blood content and vascularization. Its primary function is to support the digestion of food in an acidic environment.

The surface of the mucosa is uneven, with numerous large and small gastric folds (plicae gastricae). The number and orientation of these folds vary and have significant clinical relevance. Two folds run parallel to the lesser curvature, directing substances straight toward the duodenum. This path is referred to as the "gastric canal". Additionally, the mucosa contains gastric areas (areae gastricae) separated by gastric pits (foveolae gastricae) into which the gastric glands open.

The columnar epithelial cells constantly secrete mucous, which protects the stomach wall from the harmful effects of gastric acid and food particles. Within the lamina propria of the mucosa lie the gastric glands, surrounded by loose connective tissue containing lymphocytes and lymphoid follicles, which play an immune defense role.

Based on their location and function, gastric glands are divided into three types:

1.Cardiac glands (glandulae cardiacae)

2.Proper gastric glands (glandulae gastricae propriae) – located in the body and fundus; contain chief cells that produce pepsinogen and parietal cells that secrete hydrochloric acid

3.Pyloric glands (glandulae pyloricae) – mainly secrete mucus

In addition to these, scattered lymphoid follicles (folliculi lymphatici) can be found in the mucosa. At the junction of the stomach and duodenum, the mucosa forms a circular fold, functioning as a valve (valvula pylorica) that separates the acidic stomach contents from the alkaline environment of the duodenum.

The submucosal layer (tela submucosa) consists of loose connective tissue rich in elastic fibers and connects the mucosa with the muscularis layer. It contains blood vessels, lymphatic vessels, and Meissner's nerve plexuses, as well as MALT (mucosa-associated lymphoid tissue), which contributes to immune function.

The muscular layer (tunica muscularis), the third layer of the stomach wall, is composed of smooth muscle fibers arranged in three directions:

- Outer longitudinal layer (stratum longitudinale) – well developed along the greater and lesser curvatures
- Middle circular layer (stratum circulare) – prominent throughout, especially at the pylorus, forming the pyloric sphincter (m. sphincter pylori)
- Inner oblique layer (stratum obliquae) – primarily developed in the body and fundus

These layers contribute to stomach motility, moving contents toward the duodenum in coordinated contractions.

The serous layer (tunica serosa) is the visceral layer of the peritoneum and consists of a single layer of mesothelial cells overlying loose connective tissue. This layer envelops the stomach from all sides.

Gastric glands are classified into three types based on their location and secretory products:

- Fundic (proper) glands
- Pyloric glands
- Cardiac glands

Fundic glands are the most numerous and are found in the body and fundus. These glands are simple or branched tubular in structure and consist of three parts: neck (isthmus), body (corpus), and base (fundus). The body and base constitute the secretory portion, while the neck functions as the excretory duct, opening into the gastric pits.

Fundic glands contain five main cell types:

- Chief cells (exocrinocyti principales) – mainly in the body and base; produce pepsinogen (converted into pepsin in the presence of HCl) and chymosin (involved in milk coagulation)
- Parietal cells
- Mucous neck cells
- Additional (undifferentiated) cells
- Endocrine cells
-
- Conclusion

1.In experimentally induced ulcerative colitis, the gastric mucosa and other stomach layers undergo significant morphological changes.

2.Morphometric changes identified through measurements can serve as diagnostic markers in clinical assessment.

3.The study's findings are useful for predicting the course of inflammatory gastrointestinal diseases in gastroenterological practice.

REFERENCES

1. Abaturov S.D. *Forage resources, food security, and viability of herbivorous mammal populations.* Zoological Journal. 2005; 84(10):1251–1271.

2. Adilbekova D.S., Chorieva Z.Yu., Ismatullaeva G.Kh., Khaitmuradova G.P. *Histomorphological changes in the gastrointestinal tract of offspring born to mothers with chronic toxic hepatitis.* Eurasian Pediatric Bulletin. 2020; (1)[4]:211–221.

3. Aleksandrova V.A. *Fundamentals of the gastrointestinal immune system.* St. Petersburg: MALO, 2006. p. 44.

4. Aliseyko E.A., Gromov I.N. *Effect of lithium carbonate on the lymphoid morphology of the digestive organs in chickens vaccinated against infectious bursal disease.* Actual Issues of Intensive Livestock Development. 2011; (14)[2]:101–107.

5. Al-Rayaashi Salim Nassir. *Morphological changes of gastric lymphoid formations in experimental hemorrhagic stroke.* Ph.D. Thesis Summary. Moscow, 2006. 25 p.

6. Aminova G.G. *Cytoarchitecture of lymphoid tissue associated with the wall of the human cecum in adolescence.* Morphology. 2002; (4):53–55.

7. Andrushchenko V.V. *Structural-functional features of gastric mucosa in rats of different age periods with altered immune status.* Ph.D. Thesis Summary. 2006. 128 p.

8. Arutyunyan A.D., Kyalyan G.A. *Age-related changes in the muscular layer of the human stomach.* Morphology. 2004; (4):126–134.

9. Balukova E.V. *NSAID-induced gastropathy: from understanding mechanisms to prevention and treatment strategies.* Russian Medical Journal. 2017; (10):697–702.

10. Bekov T.A., Kosim-Khodzhaev I.K. *Changes in human gastric mucosa during postnatal ontogenesis.* Morphology. 2004; 126(4):19–23.

11. Belyaeva E.V., Gushchin Ya.A. *Visualization methods for studying gut-associated lymphoid tissue in laboratory animals.* Laboratory Animals for Scientific Research. 2020; (3):68–74.

12. Bikkina G.M., Safuanov A.R. *Adverse drug reactions of nonsteroidal anti-inflammatory drugs.* Young Scientist. 2015; (7)[87]:269–272.

13. Bilich G.L., Zigalova E.Yu. *Human Anatomy: Russian-Latin Atlas.* Eksmo Publishing, 2nd ed., 2018. 453 p.

14. Bondarchuk A.O., Fomina L.V., Gavrilyuk A.A., Mnikhovich M.V., Zherebyateva S.R. *Regeneration of small intestine mucosa under cyclophosphamide and corrective therapies.* Russian Medical-Biological Bulletin of I.P. Pavlov. 2015; (1):15–24.

15. Borodin Yu.I. *Lymphology as an integrative biomedical science.* Bulletin of Lymphology. 2011; (2):4–7.

16. Burtseva A.S., Atyakshin D.A. *Mast cells in the digestive organs as objects of study in space gastroenterology.* Morphology. 2018; 153(3):51–52.