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COMPARISON OF THE EFFECT OF DIFFERENT MESH IMPLANTS ON THE REPRODUCTIVE SYSTEM IN EXPERIMENTAL MODELS OF GYNECOLOGICAL SURGERIES

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Genital prolapse complicates the lives of many women after the age of 50. The use of mesh implants is a promising method of surgical correction of this disease. The study on mice compared the effect of the most common surgical meshes on the morphofunctional state of the reproductive system of animals, the postoperative period, and the completeness of morphological and functional recovery of the defects in the lower abdominal wall. It was found that thinner meshes cause less inflammatory reactions, faster recovery and less atrophic phenomena in the internal genitals. The recovery rate also depends on the material and structure of the mesh.

Key words: gynecological surgeries, surgical meshes, genital prolapse, uterus, ovaries.

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ПОРІВНЯННЯ ВПЛИВУ РІЗНИХ СІТЧАСТИХ ІМПЛАНТІВ НА СТАТЕВУ СИСТЕМУ В ЕКСПЕРИМЕНТАЛЬНИХ МОДЕЛЯХ ГІНЕКОЛОГІЧНИХ ОПЕРАЦІЙ

Генітальний пролапс обтяжує життя багатьох жінок після 50 років. Перспективним методом хірургічної корекції цього захворювання є застосування сітчастих імплантів. В дослідженні на мишах порівняно вплив найбільш розповсюджених хірургічних сіток на морфофункціональний стан статевої системи тварин, перебіг післяопераційного періоду, повноту морфологічного та функційного відновлення дефекту нижнього відділу черевної стінки. Виявлено, що більш тонкі сітки з викликають менше запальних реакцій, швидше відновлення та менші атрофічні явища у внутрішніх статевих органах. Швидкість відновлення також залежить від матеріалу та структури сітки.

Ключові слова: гінекологічні операції, хірургічні сітки, пролапс геніталій, матка, яєчники.

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Genital prolapse significantly reduces the quality of life of many elderly women [8, 13]. The main complaints are omission and prolapse of the vagina and uterus, urinary incontinence, pain, inflammation, dryness and erosion of the mucous membranes, difficulties and unpleasant sensations during sexual life, reduced efficiency, and problems in the family [7]. Prerequisites for this pathology are abnormal childbirth, hereditary inferiority of the connective tissue elements of the pelvis and heavy physical activity. All these factors make it impossible to correct genital prolapse using only their own tissues due to their inferiority.

One of the ways to solve the problem is to use mesh surgical implants to strengthen the pelvic floor [4, 6, 7]. Mesh implants are widely used in surgery to treat hernias and close abdominal wall defects [1, 9]. However, infection, rejection, and organ dysfunction due to adhesions or inflammation are sometimes observed using implants [2, 3, 5]. The treatment effectiveness and the number and severity of complications may vary depending on the type of operation, the type of mesh used and the patient's condition [15]. Since most studies of the mesh's effectiveness have been performed in surgical pathology, the effect of surgery on the condition of the female reproductive system is not taken into account, which is most important when using meshes to correct genital prolapse.

The purpose of the study was to examine the effect of different mesh implants on the condition of the female reproductive system in the experiment.

Materials and methods. 101 female C57Bl/6J mice aged 6 months, weighing 21±0.5 g, with a regular estrous cycle, were used in the study. Rodents do not have the connected pelvic bones and the muscles that form the pelvic floor in humans enter the lower abdominal wall, tissue failure of the lower abdominal wall was simulated by the formation of a 4 mm diameter defect at the level of uterine discharge from the vagina. The defect was closed with a 1.0x1.0 cm mesh implant. A 1.0x1.0 cm mesh fragment was additionally placed in the abdominal cavity near the uterus and vagina to assess its effect on the reproductive system. The skin above the defect was sutured. The animals were divided into 5 groups: 1 – mice treated with Monomesh (M) polypropylene implants (Fiatos, Belarus), 2 – mice with Monomesh Light (ML) polypropylene implants (Fiatos, Belarus), which differ in structure, 3 – mice with Polymesh

(P) implants (Fiatos, Belarus), which contains polyglycaprolactone, 4 – mice whose defect (D) was not covered with a mesh, 5 – Control Group (C) operated without defect formation. In dynamics, the temperature, weight of animals, physical strength, edema and soreness of the surgical site, exudation, and regularity of the estrous cycle were studied using colpocytological examination. On days 7, 14, 30, and 60, 6 animals from the group were sacrificed, and the severity of the adhesive process after surgery was assessed; the uterus, ovaries, and vagina were fixed in formalin sections stained with hematoxylin-eosin, and histological examination was performed. A Primo Star microscope (Carl Zeiss, Germany) with an integrated camera and Zen software (Carl Zeiss, Germany) was used to analyze preparations.

On day 60, fragments of meshes were removed from the abdominal cavity, cleaned, and their strength was measured. For this purpose, they were fixed with threads on both sides and stretched until rupture. The same mesh fragments were kept for 60 days in phosphate buffer (BioWest, France) and 0.25% trypsin solution (BioWest, France) and tested for strength.

Strength was assessed by fixing the animal to the back of the torso as it held the front paws of the mesh with a dynamometer, recording the maximum value.

The degree of edema at the surgical site was assessed on a scale of 0–3: 0 – no edema, 1 – mild edema, 2 – moderate edema, and 3 – severe edema. The degree of exudation was assessed on a scale of 1–4: 1 – absence (dry wound), 2 – light exudates (the wound is wet, does not ooze when pressed), 3 – moderate exudates (the wound is wet), 4 – extreme exudates (exudates are visible) [10]. Painfulness was assessed by palpating the wound based on the animals' reactions.

Adhesions were evaluated as 0 – absence, 1 - thin adhesions that are easily removed, 2 – thick adhesions limited to one area, and 3 – thick and wide adhesions involving the anterior or posterior abdominal wall and viscera [10].

The experiments were performed according to the General Principles of Animal Experiments, approved by the V Congress on Bioethics (Kyiv, 2013) and by the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (Strasbourg, 1986). Mann-Whitney U-test and Student's t-test were used to obtain statistically significant conclusions. Past V.3.15 software (Hammer, Natural History Museum, University of Oslo) was used for statistical calculations and data processing.

Results of the study and their discussion. All animals had anaesthesia recovery for the first 20 minutes after surgery and moved freely but were inhibited during the day. The next day, mice from all groups ate freely, drank water and were active.

When studying rectal temperature, an increase on the 2nd day was detected in animals of all groups. At the same time, the temperature was significantly higher in groups of animals implanted with completely non-biodegradable meshes (Monomesh and Monomesh Light). On Day 3, the temperature in animals of all groups implanted with meshes was higher than in the control groups, in which the temperature decreased to physiological. On day 4, the temperature in the animals implanted with Polymesh meshes did not differ from the control one. Starting from day 5, the temperature of animals in all groups did not differ from preoperative rates.

The weight of all animals remained unchanged for a week after surgery. After 2 weeks, the weight of animals with implants began to decrease. One month later, the weight of all animals, except those with false surgery, was significantly lower than the initial values, in particular animals without implants. At the same time, animals that did not have implants lost the least weight. Complete weight recovery was observed after 2 months.

An animal strength study found that in the first 3 days after surgery, all study groups were likely to have decreased physical strength associated with postoperative trauma. On days 7 and 14, the strength in animals with an open defect and in animals with a Monomesh mesh remained reduced, while in other groups it increased significantly. On days 30 and 60, the physical strength of the animals with the Polymesh and Monomesh Light probably did not differ from the control and was maximal. At the same time, the strength of animals with Monomesh and an open defect was lower.

Edema at the site of surgery was observed for 3 days in animals with an abdominal wall defect and with implants. Exudation was not observed in any animal. Pain response was observed in some animals of all groups for 1–3 days. There was no significant difference between the pain response in animals from different groups.

The estrous cycle was monophasic in all animals of the Monomesh group within a week after surgery, and recovery was observed after 2 weeks. In animals implanted with a lightweight mesh or Polymesh, recovery was observed one week after surgery. No cycle disorders were observed in false-operated animals and animals with an undisguised defect.

When studying the adhesive process on day 7 after surgery, a pronounced adhesive process with conglomerates and edema in the abdominal cavity was observed in most animals implanted with Monomesh and Monomesh Light (fig. 1). A similar situation remained for up to 14 days. On Days 30 and 60, the adhesive process was lesser but involved the abdominal, intestinal, liver, and spleen organs.

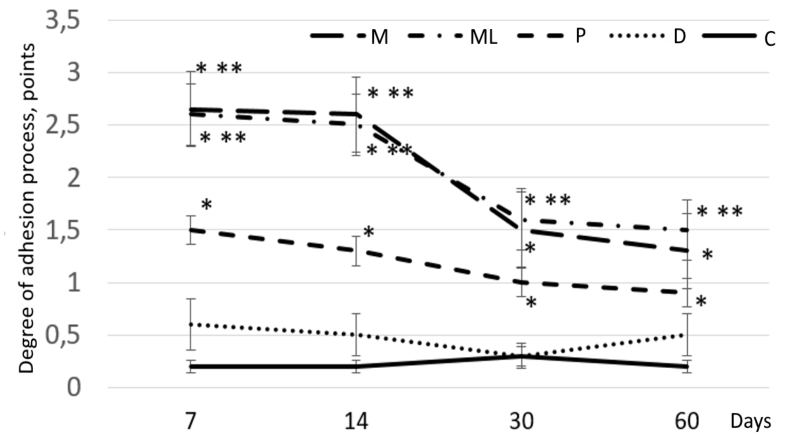

Fig. 1. The degree of adhesive process in the studied groups. * – probability of differences with the control group p<0.05, ** – probability of differences with the group implanted with the Polymesh p<0.05.

The adhesion process in the animals implanted with Polymesh was probably lower. Most of the sacrificed animals after 7 and 14 days did not have conglomerates but had edema and isolated adhesions. On Days 30 and 60, edema almost was not observed. Most adhesions were attached to the site of surgery in the abdominal wall. The adhesion process in falsely operated animals and animals with a defect of the abdominal wall was much smaller. Still, it was noteworthy that the defect of the abdominal wall did not last even after 2 months, which explained the decrease in the strength of the animals. Some mesh fragments left in the abdominal cavity rarely caused adhesions and were freely located between the internal organs.

At the site of Monomesh implantation on day 7, histological examination observed a large amount of connective tissue with leukocyte infiltration, which thickness reached 1.5±0.08 mm. The mesh structures in the section had a homogeneous structure. The mesh fibres were surrounded more closely by connective tissue fibres and cells (Fig. 2a). After a month, the connective tissue became less dense, the infiltration disappeared, and tissue compaction was observed only around the fibres (fig. 2b).

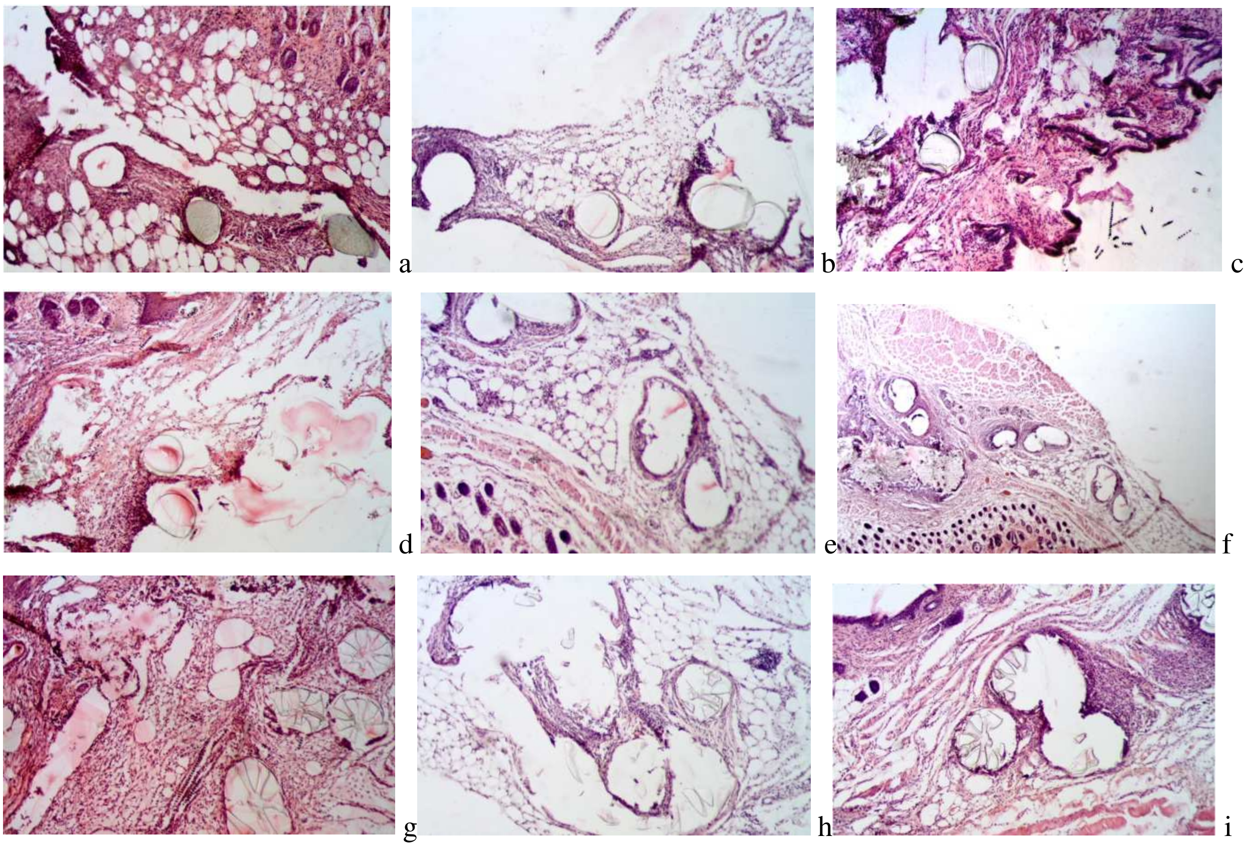

Fig. 2. Condition of implants and surgery sites in the study groups: Monomesh (a, b, c); Monomesh Light (d, e, f), Polymesh (g, h, i) a, d, g – 7 days after surgery, b, e, h – 30 days after surgery, c, f, i – 60 days after surgery. Scale bar 100 µm.

After 2 months, the connective tissue was fully formed, and there was a mesh (fig. 2c). Animals implanted with Monomesh Light had a thinner zone of inflammation of 0.7±0.05 mm on day 7, and infiltration was also observed around the mesh (fig. 2d). The mesh polymer was dense, unchanged. After

1 month, infiltrative changes remained only around the mesh fibres (fig. 2e). At the end of the experiment, the mesh was located in the formed scar (fig. 2f). Polymesh caused less accumulation of connective tissue and inflammation around the fibres, but the thickness of the scar was as with the Monomesh Light – 0.8±0.07 mm (fig. 2g, 2h, 2i). In the section, the mesh was star-shaped, but connective tissue did not fall between its particles throughout the experiment. Within 2 months, the number of connective tissue and leukocytes decreased, and a scar was formed. The structure of the mesh itself did not change.

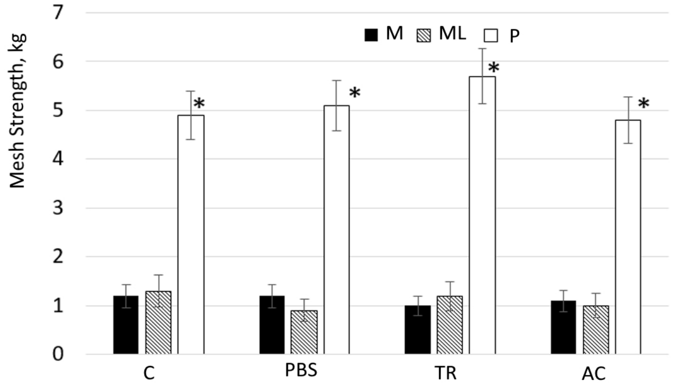

Fig. 3. Strength of different types of meshes after 60 days. M – Monomesh, ML – Monomesh Light, P – Polymesh, C– Control (meshes without influence), PBS – meshes in phosphate buffer solution, TR – meshes in trypsin, AC – meshes in the abdominal cavity.

When studying the strength of meshes, it was found that Polymesh meshes are significantly harder than Monomesh and Monomesh Light meshes (fig. 3). The incubation of all meshes for 2 months in phosphate buffer solution, in trypsin or in the abdominal cavity, did not affect the tensile strength of the meshes. However, since the strength was measured on a small piece of mesh 1.0x1.0 cm, and the distance between the fixing ligatures was less than 0.5 cm, this indicates that they are all able to withstand loads from 2 to 10 kg per 1 cm^2, which is sufficient for fixing abdominal organs.

Histological examination of the genitals of animals implanted with Monomesh and Monomesh Light on the 7th day showed atrophic and inflammatory changes in the ovaries. They were manifested in reduced organ size, lack of mature follicles and leukocyte infiltration around the organ (fig. 4a). The uterus was also atrophic and reduced with leukocyte infiltration and thinned endometrium. The glands were not clearly visualized (fig. 4d). The vagina was also reduced with thinned epithelium (fig. 4g).

After 30 days, mature follicles appeared in the ovaries, the walls of the uterus thickened slightly, and single glands appeared (figs. 4b, 4e, 4h). After 60 days, the ovaries recovered had follicles of all stages, a large number of glands were observed in the uterus, the endometrium was thickened, and the vaginal wall was also of average thickness, with glands (figs. 4c, 4f, 4i).

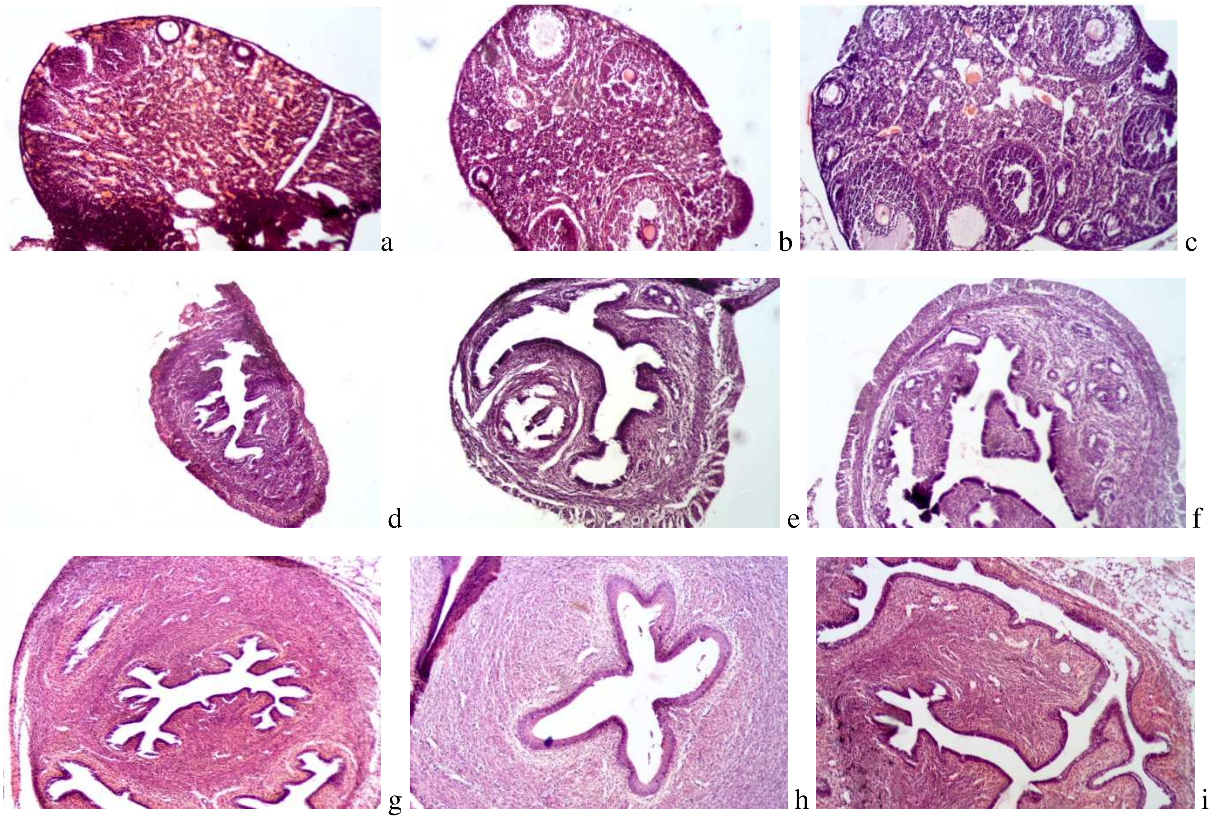

Fig. 4. The condition of the internal genitals in the study groups. a, d, g – 7 days after surgery; b, e, h – 30 days after surgery; c, f, i – 60 days after surgery; a, b, c – ovaries,d,e, f – uterus, g, h, i – vagina. Scale bar 100 µm.

In animals implanted with the Polymesh, inflammatory changes were observed for a week. After 1 month, the condition of the ovaries, vaginas, and uterus did not differ from the control groups. In falsely operated animals and animals with abdominal defects, pathological changes in the histological examination of the uterus, vagina and ovaries were not detected.

Insufficient pelvic floor muscles, ligaments and fascia in women can lead to genital prolapse, significantly reducing women's quality of life. Manifestations of prolapse can be the omission of the genitals, difficulties in sexual life, urinary incontinence, and ulcers [11]. In case of tissue insufficiency, the promising treatment method is the implantation of meshes used for successful hernia repair surgery [14]. However, their use in gynecology raises several questions related to the peculiarities of surgical techniques, the possible impact on the genitals and the choice of the meshes themselves [11]. The paper experimentally analyzes the effectiveness of using different meshes and their impact on different localization. It has been found that full recovery of a significant tissue defect and the force of contraction of the corresponding muscles is possible only with mesh. Any mesh causes an inflammatory response when interacting with the wound. Coarser meshes cause a more significant and extended response. The inflammatory reaction, in turn, leads to an adhesive process and inflammatory changes in the ovaries and uterus. Violation of the estrous cycle can occur due to the inflammatory process [5]. It has also been found that different polypropylene meshes have other strength characteristics that change little after being in a liquid, enzyme solution, or in the abdominal cavity of animals. Thus, it can be considered that for the effective treatment of genital prolapse, it is advisable to use polypropylene meshes, choosing lighter, more robust and preventing inflammatory and adhesive processes.

Conclusions

1. The main possible reactions to polypropylene surgical meshes are inflammatory changes that lead to weight loss, strength, violation of the estrous cycle, discirculation, inflammation and genital atrophy, which occur in the first weeks after implantation is reversible, except for the adhesive process. The phenomena of the inflammatory process probably cause the reaction of the genitals to the meshes.

2. The degree of reaction to the mesh depends on its structure and the material and is more pronounced when suturing the mesh into the wound than when the mesh is freely placed in the abdominal cavity.

3. The structure and strength of polypropylene meshes do not change after being in a liquid, enzyme solution, or animal body.

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