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EVALUATION OF THE EFFECTIVENESS OF ANTI-ADHESIVE COATING ON A MODEL OF A LUNG WOUND IN AN EXPERIMENT

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ABSTARCT:

The problem of the adhesive process in surgery is still extremely relevant. The vast majority of studies of domestic and foreign authors are devoted to the formation of adhesions in the abdominal cavity. Interperitoneal fusion is still the cause of many diseases of the abdominal cavity [1,2,3]. First of all, these are directly peritoneal adhesions that occur both as a result of the inflammatory process and after surgery [4,5].

The question of the expediency of preventing adhesions in the abdominal cavity is now proven, and there is no doubt. At the same time, pleural adhesions in the modern classification of diseases, as an independent nosological unit, are absent. In this connection, a logical question arises: are the adhesions of the pleural cavity a pathological process, or is the adhesiogenesis in this area a compensatory-adaptive reaction [6,7,8]?

However, it is necessary to be sure that the induced increase in adhesiogenesis will not lead to the development of total intrapleural adhesions, weakening of the function of external respiration and the occurrence of respiratory failure. This fact requires close study and determines the relevance of the work.

INTRODUCTION:

Thus, a number of studies have been conducted on the prevention of spike formation, but their results are encouraging, but most of them are contradictory and were conducted on experimental models.

Materials and methods of research. Experiments on the formation of lung wounds with subsequent evaluation of the effectiveness

of the anti-adhesive coating of cellulose derivatives were performed on the basis of the State Institution "RSNPMTSH named after Academician V. Vakhidov", Department of Experimental Surgery in 2019. As experimental animals, white mongrel rats in the number of 32 individuals were used. In total, 2 series of experiments were performed: control and experimental groups.

Anesthesia was performed using the RO-6 anesthesia machine with oxygen supply. The ventilation mode was carried out with a frequency of 24 per minute and a volume of up to 30 ml. For mask anesthesia, a special rubber nozzle was used, which was put on the animal's muzzle and covered the area of the transition of the head to the neck hermetically. The volume of the mask is 50ml. The mask has a non-return valve for exhaling air.

THE PROCEDURE OF THE OPERATION:

Incision of the skin and superficial muscle in the region 6 of the intercostal space up to 3 cm long. The pectoral muscles were loosened along the course of the muscles (Fig. 1).

Fig. 1. Skin incision in the area of the right half of the chest

In area 6 of the intercostal space, a thoracotomy was performed using a mosquito – type blunt instrument (Fig. 2).

Fig. 2. Dilution of the chest muscles, allocation of the intercostal space

The wound was diluted to 1.5 cm and then hooks were inserted to expand the wound (Fig. 3).

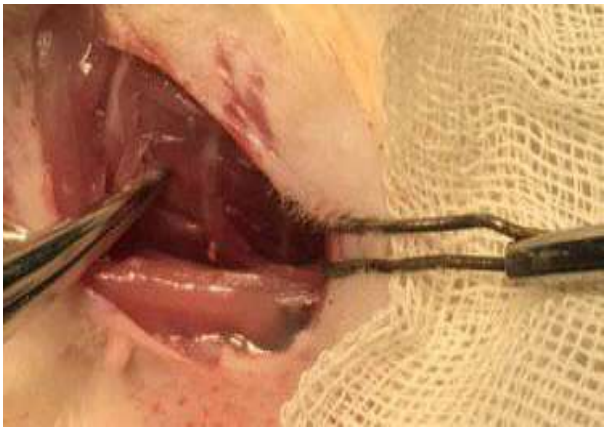


Fig. 3. Using a mosquito-type clamp to open the pleural cavity and dilute the intercostal space

The right lung is moderately collapsed, breathing during mask anesthesia, air. Using atraumatic micro-clamps, the right lung was removed into the thoracotomy wound. The anterior surface of the lung was damaged using a bipolar coagulator (Fig. 4).

Fig. 4. Damage to the lung using a bipolar coagulator

On a surface of 1 cm2, there are 5 damages, each measuring up to 2 mm in diameter. In the sample with the introduction of saline solution, the appearance of air bubbles was not noted.

Fig. 5. Application of anti-adhesive coating on the wound surface of the lung

In the control group of animals, the lung sank back into the right pleural cavity. Next, a microcatheter was left and the thoracotomy wound was closed hermetically at first by stitching the edges of the chest muscles and then the skin wound. After the sealing was achieved, air was sucked out of the pleural cavity through the catheter and the catheter was removed. Ventilation of the lungs was carried out with oxygen until the animal was fully awakened. Then the rat was moved to a separate cage for observation. For 3 days, water was given with the addition of pirprofen at the rate of 0.5 g per 100 ml of water.

In the experimental group of animals, a special anti-adhesive coating made of cellulose derivatives was applied to the area of lung damage (Fig. 5)

At the present time, lung surgery is more focused not on the prevention of the adhesive process, but on their formation, in particular, to prevent the development of relapses of pneumothorax, etc. However, the purpose of this part of the experimental morphological study was to further confirm the effectiveness of the hemostatic coating of Geoprocessing in terms of preventing the development of postoperative adhesions. By analogy with the previous experiment, 32 white mongrel rats were used as experimental animals. In total, 2 series of experiments were performed: a control group (17 animals) and an experimental group (15 animals).

After the adhesion and uniform distribution of the coating on the surface of the lung, we proceeded to the next stage. To prevent the process of cellular inflammation, blood serum was applied to the adhesive coating using a syringe (Fig. 6). As a result, within 1-3 minutes, the coating passed into a state of a translucent film, soft-elastic consistency, did not prevent the lung from stretching during breathing and tightly adhered to the wound surface (Fig. 7).

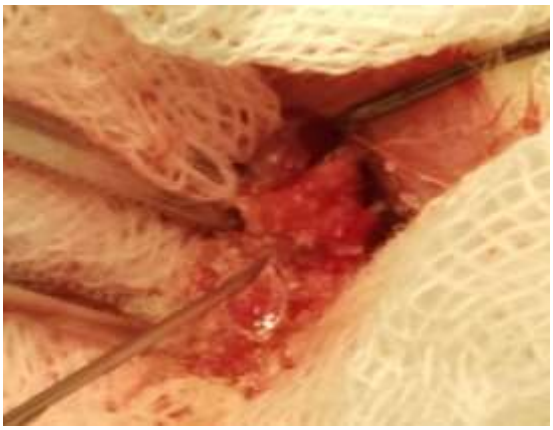


Fig. 6. Application of blood serum to the powder coating in order to form a translucent film on the surface of the wound.

Fig. 7. Formed coating on the surface of the lung wound.

When the coating is formed using blood, it takes on the character of a blood clot, which contributes to the formation of a denser coating with subsequent biodegradation by the type of

cellular inflammation during resorption of thrombotic masses (Fig. 8).

Fig. 8. The nature of the coating when using blood to form a hemostatic coating using a Geoprocel implant.

A similar coating was also formed on the wound surface of the parietal pleura, where access to the pleural cavity was provided. Then the operation was completed in the same way as in the control group.

During the experiments, a fatal outcome was observed in 2 cases. In the first case in the control group, the fatal outcome occurred due to depression of the heart in the early post narcosis period. In the second case, in the early postoperative period, mortality occurred as a result of lung collapse in the control group.

IN THE CONTROL GROUP:

1 day. After the operations were performed on the first day, all the other operated animals were alive, active, took food and drank water (with the addition of pirprofen for anesthesia). In the area of the postoperative wound, the phenomena of crepitation and pathological mobility of the chest were not observed. The stitches are good, there are no signs of inflammation. The right and left sides of the chest are actively involved in breathing.

3 days. The animals are active, moving around the cage. They drink water well and take food. There are no signs of inflammation in the area of the operating wound. The chest is evenly involved in the act of breathing. When taking the animals in their hands, there were no painful sensations in the area of the postoperative wound.

7 days. The condition of the operated animals without any special pathological changes. The postoperative wound healed in both groups of rats. On palpation, both halves of the chest and the wound area are painless. The weight of the animals changed slightly, the difference in both groups of animals is not statistically significant (Table 1).

14 days. The animals are in good condition. Active. They take food and drink water. The postoperative wound has healed. The stitches were removed from the wound, there is no discharge. Palpation of the wound area is painless. Breathing is performed. Pulse is within normal range.

During the observation of the operated 17 animals in the control group, signs of wound suppuration were noted in 2 cases. The average period of complete wound healing was 6 days.

THE MAIN GROUP OF ANIMALS:

In the main group of 15 animals, postoperative care and analgesia were performed in the same way as in the control group. There were no significant changes in behavior, physiological functions, or general condition. Complications in the form of superficial suppuration of the wound were noted in 1 of one rat, which was stopped after removal of the skin thread. The average wound healing time was 5-6 days

The dynamics of animal weight indicators are presented in Table 1.

DATA FROM MACROSCOPIC STUDIES:

As planned, on the 7th day after the operation, 5 rats from each group were removed from the experiment to assess the condition in the pleural cavity after modeling a lung wound (2 animals in the control group died in the early postoperative period).

Table 1. The dynamics of weight of the animals after exposure

Group	Prior to exposure	7 day	14 day	21 day
Control system	176 ± 12	150 ± 13	160 ± 14	175 ± 12
Experienced	174 ± 14	165 ± 15	171 ± 15	178 ± 14
t-criterion	0,43 P>0,05	1,69 P>0,05	1,20 P>0,05	0,36 P>0,05

In the control group for the 7th day. When opening the chest after euthanasia, according to the provisions on the humane treatment of animals, it was revealed: the left pleural cavity is intact, the lungs are dormant after air intake, air. The pleural cavity is clean. No adhesions or pathological manifestations were detected. Right pleural cavity (from the side of the performed operation), without pathological effusion. There are massive junctions between the lungs and the parietal pleura. The area of surgical access in the 6th intercostal space is also located in the junctions with the right lung (Fig. 9).

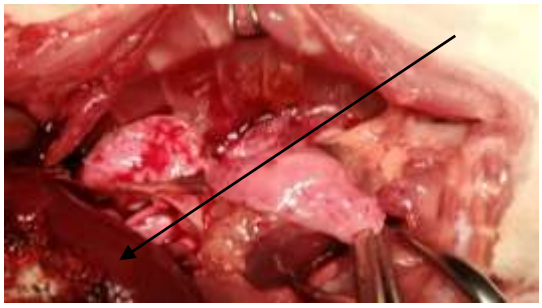


Fig. 8. 7 day control. Planar junction of the lower lobe of the right lung with the parietal pleura in the area of the thoracotomy wound.

The adhesions are separated from the lung with blunt dilution, in places there is a tear of the visceral pleura. When calculating the number of specks, it was revealed that in 5 points of the defect, in most cases, adhesions with the parietal pleura were formed. In the area of the wound, there are spikes along the entire course of the surgical access. Of the 5 rats studied, in all cases there was an adhesive process, which was in most cases planar in nature. The area of the soft tissues of the chest and skin had no signs of infection, there was no discharge.

In the main group, 5 animals were also removed from the experiment on day 7. As in the control group, no pathological changes were detected in the left pleural cavity. When opening the right pleural cavity, there is a loose adhesive process in the area of applying point defects in the area of the anterior surface of the lung. Of the 5 animals studied, 3 spikes out of 5 injuries occurred in 2 cases and a single spike out of 5 injuries occurred in one case. Visually, there are no signs of the presence of a coating, the visceral surface of the lung is covered with a thin, shiny film without color. The adhesions are easily separated without damaging the visceral pleura. The lungs are airy, not deformed. In the area of the surgical wound in the 6th intercostal space, there are also planar loose adhesions in all cases out of 5, easily separated.

On the 14th day after the operation, in the control group, when opening the chest, it was revealed: the left pleural cavity was intact, the lungs were asleep after air intake, air. The pleural cavity is clean. No adhesions or pathological manifestations were detected. The postoperative wound in the area of the right half of the chest is clean, without infiltration. Only in 1 case there was suppuration of soft tissues with the formation of an abscess in the ligature area. Right pleural cavity without pathological effusion. There are adhesions between the lungs and the parietal pleura (Fig. 9).

Fig. 9. 14 days. Adhesions between the visceral and parietal pleura in the form of cords in the control group

The area of surgical access in the 6th intercostal space is also located in the junctions with the right lung. Adhesions are poorly separated from the lung with blunt dilution, there is a tear of the visceral pleura. Calculating the number of specks is difficult due to the fusion of the adhesive process. Most of the adhesions are represented as strands in 2 cases in view of the planar adhesions of the visceral and parietal pleura (Fig. 10). In the area of a previously performed thoracotomy wound, there is an adhesive process with visceral pleura in the form of thin cords.

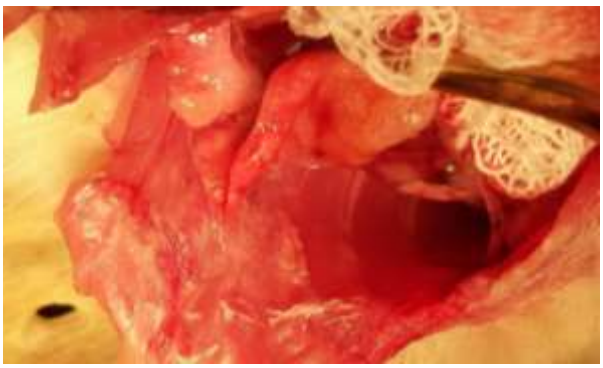


Fig. 10. 14 days. Planar adhesions between the visceral and parietal pleura in the control group.

In the main group, 5 animals were also removed from the experiment on day 14. As in the control group, no pathological changes were detected in the left pleural cavity. When opening the right pleural cavity, there is a single adhesive process in the area of applying point defects in the area of the anterior surface of the lung. Of the 5 animals studied, only 1 animal had adhesions; in other cases, no adhesions were detected. In the area of a previously performed thoracotomy wound, no adhesions were detected in any case (Fig. 11). The area of lung damage is determined by the barely noticeable scars and compaction of the lung tissue. The lung tour is not disturbed. Visual indication of the presence of the coating is not revealed. In the soft tissue area where the thoracotomy was performed, the inflammatory process was not detected.

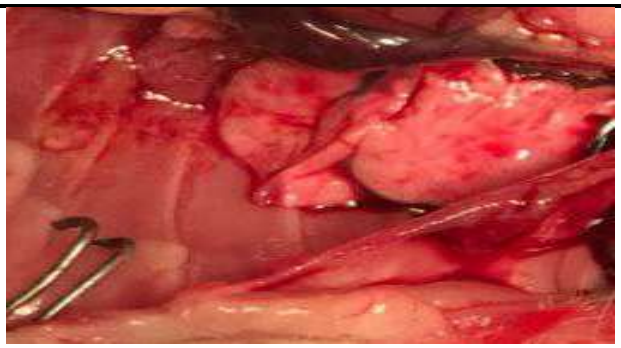


Fig. 11. 14 days. Absence of lung adhesions in the experimental group of operated animals

21 days after the operation. When opening the chest in the control group, it was revealed: the left pleural cavity is intact, the lungs are dormant after air intake, air. The pleural cavity is clean. No adhesions or pathological manifestations were detected. When opening the right pleural cavity, there is no pathological effusion. There are splices between the lungs and the parietal pleura in the form of strands (Fig. 12).

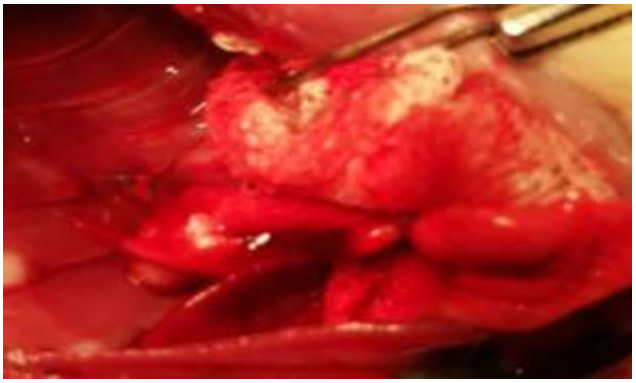


Fig. 12. Control 21 days. There is an adhesive process between the visceral and parietal pleura in the form of dense strands and planar adhesions.

In the area of surgical access in the 6th intercostal space, adhesions in the form of thin cords with the right lung were also revealed. The adhesions are separated from the lung with the formation of tears in the visceral pleura. Of the 5 rats studied, there was an adhesive process in all cases. The area of the soft tissues of the chest and skin had no signs of infection, there was no discharge.

In the main group, 5 animals were removed from the experiment for 21 days. No pathological changes were detected in the left pleural cavity. When opening the right pleural cavity, the lungs subside, there are no signs of formed adhesions between the visceral and parietal pleura. Only in 1 case there was a thin layer between the area of the surgical wound and the visceral pleura (Fig. 13). At the site of the lung injury – small whitish scars, the lung tissue is airy, the elastic surgical wound is completely healed, there is no infiltration, the layers of tissue are distinguishable.

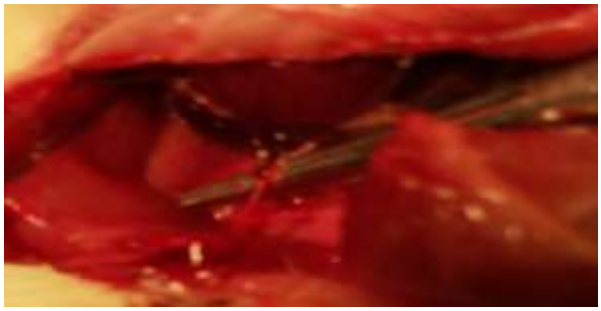
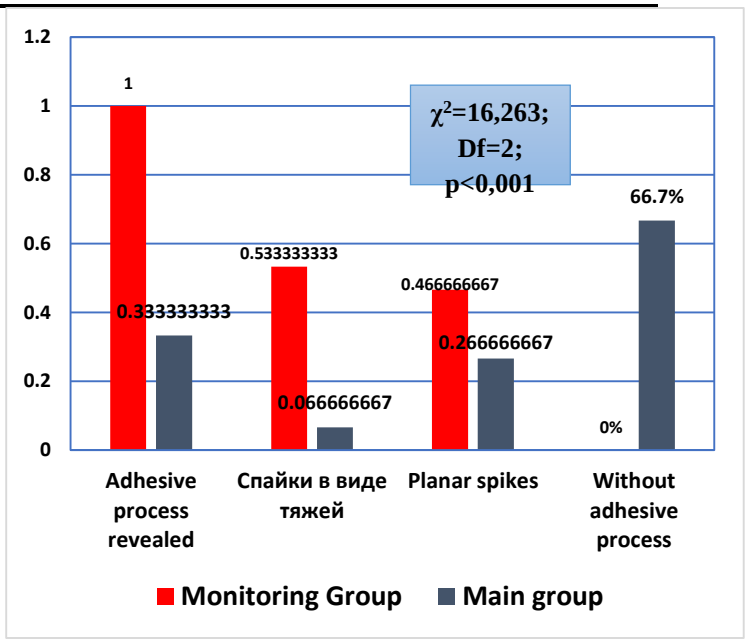


Fig. 13. The thin layer between the parietal and visceral pleura in the postoperative wound area in the main group

In this study, it was possible to demonstrate that the use of anti-adhesive coating allowed to reduce the risk of adhesions and the nature of their development (planar or in the form of strands) (Table. 2; fig. 14).

Table 2 Indicators of the formation of the adhesive process in the experiment

Indicator	Monitoring Group		Main group	
	abs	%	abs	%
7 days				
Adhesive process revealed	5	100,0%	3	60,0%
Spikes in the form of strands	0	0,0%		0,0%
Planar spikes	5	20,0%	3	12,0%
14 days				
Adhesive process revealed	5	100,0%	1	20,0%
Spikes in the form of strands	3	60,0%	0	0,0%
Planar spikes	2	40,0%	1	20,0%
21 days				
Adhesive process revealed	5	100,0%	1	20,0%
Spikes in the form of strands	5	100,0%	1	20,0%
Planar spikes	0	0,0%	0	0,0%



14. Summary results of experimental studies on the formation of adhesions in the abdominal cavity

Thus, the task of experimental studies was to create a model of lung damage that would allow to cause an adhesive process in the pleural cavity, while not causing such complications as pneumothorax, pleurisy and pleural empyema. The model should be feasible and reproducible, and not be accompanied by severe complications and death. The second task was to evaluate the effectiveness of the hemostatic domestic implant Geoprocel in preventing the formation of adhesions in the pleural cavity by a new method of forming an elastic coating using blood serum instead of blood. At the same time, the resulting coating does not have the character of a blood clot, i.e. it does not contain red blood cells, and therefore allows you to prevent the presence of blood cell elements in the process of biodegradation, thereby preventing the formation of adhesions.

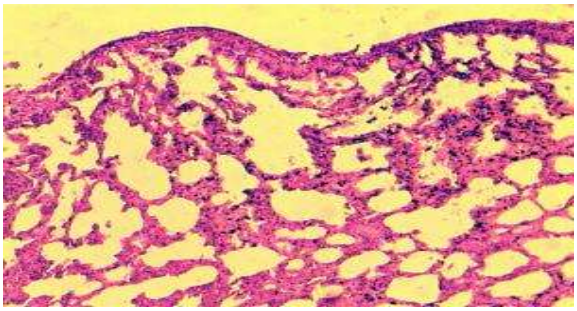
The model of lung damage in the form of a right-sided thoracotomy with minimal tissue bleeding due to blunt separation of soft tissues and intercostal muscles made it possible to exclude the use of physical and chemical methods of hemostasis. Damage to a specific

surface of the right lung allowed us to form a standardized model of lung damage. At the same time, the left pleural cavity served as a control. The operations were performed by the original mask anesthesia using halothane vapors with oxygen in a special evaporator.

The postoperative period was uneventful. The study of the state of animals, physiological parameters, as well as macroscopic changes during the withdrawal of animals from the experiment allowed us to come to the following conclusion. The domestic hemostatic implant Geoprocel adheres well to the tissues of the damaged lung, as well as to the edges of the surgical wound. The use of blood serum makes it possible to achieve the formation of a soft-elastic coating on the surface of the lung wound, which does not deform the lung and stretches when the lungs are opened. In comparison with the control group of animals, the new coating almost completely prevents the pronounced adhesive process in the pleural cavity, thereby preserving the physiological excursion of the lungs after wound exposure. The process of biodegradation is not accompanied by an inflammatory reaction of the tissues and effusion in the pleural cavity. The wound heals in the usual time frame. The behavior of animals and their condition does not undergo significant changes. The weight gain of animals begins as early as 14 days after the operation. Thus, a new method of coating formation using a domestic Geoprocel implant can be applied after performing surgical interventions on the lungs in order to restore the most complete physiological parameters of respiration.

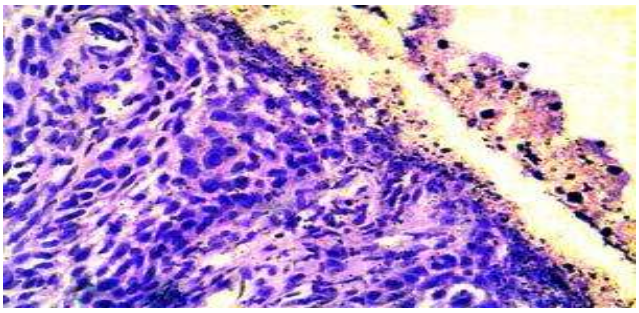
Morphological features of the anti-adhesive effect in the experimental model on the thoracic cavity

In the groups of animals in the norm (left lung), the pulmonary alveoli were airy. No pathological elements or proliferative process were detected (Fig. 15).

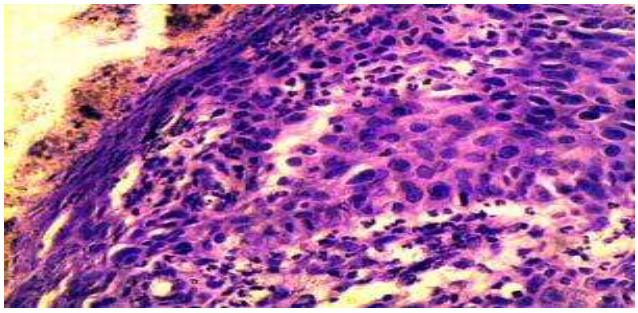


15. Visceral hymen and pulmonary parenchyma with air alveoli. The norm of G-E is 10x10.

On the side of the wound defect (right lung) on Day 7, the mesothelial layer is destroyed, connective tissue cells are detected on the surface, the lung parenchyma is edematous (Fig. 16-17).



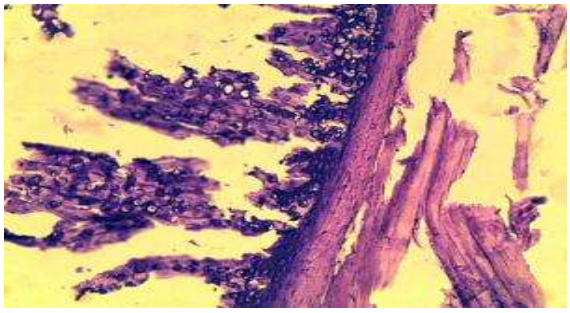
16. Violation of the integrity of the visceral pleura by numerous connective tissue cells on its surface. G-E. 10 x 40



17. Atelectasis of the alveoli with numerous connective tissue cells on its surface. G-E. 10 x 40

At the time of Day 14, the development of granulation tissue is noted, in the thickness of which connective tissue strands are formed (Fig. 18) with the development of coarse -

fibrous connective tissue-the formation of adhesions (Fig. 19), while quite numerous fibroblasts are located between the fibers of the connective tissue (Fig. 20).



18. Formation of connective tissue strands on the visceral pleura. G-E 10x10.

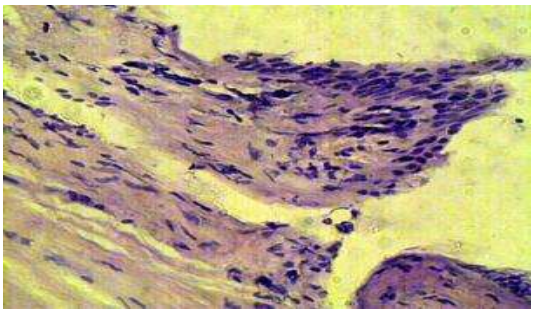


Fig. 19. Fragment of a spike on the visceral pleura. G-E 10x 40.

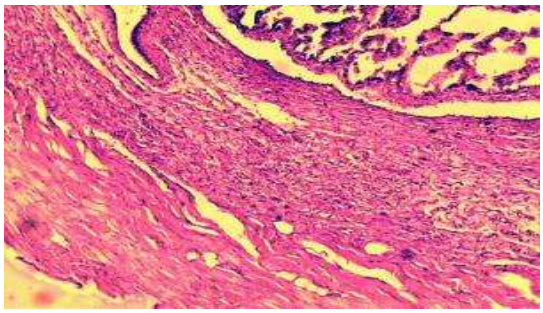
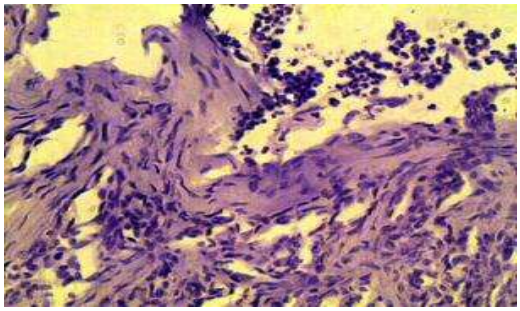


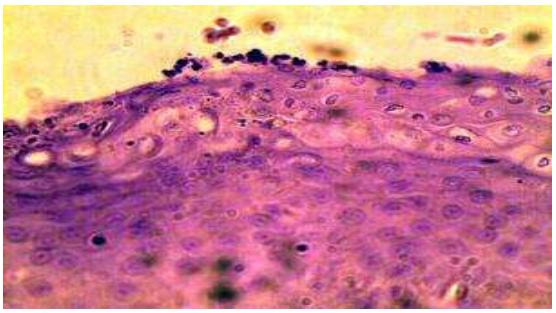
Fig. 20. Fragment of a spike on the visceral pleura. G-E 10x 10.

On the 21st day of observation, the connective tissue fibers become coarser. In the pulmonary parenchyma, the phenomena of atelectasis of the alveoli and the presence of exudate in their lumen are noted.

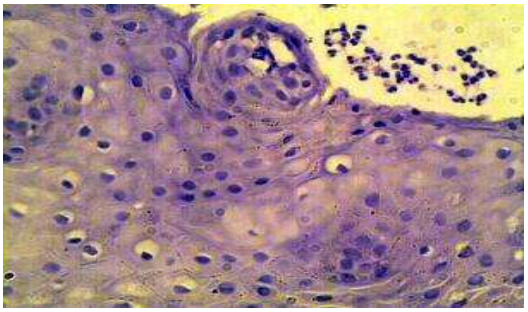
In the early stages of Geprocel application, the accumulation of fibroblasts is also noted (Fig. 21), but in the more distant periods, the number of migrated cells on the surface of the adhesions becomes smaller (Fig. 22, 23). And at the time of 21 days of observation on the background of the use of Hepracel, the migrated cells on the surface of the adhesions are no longer detected (Fig. 24).



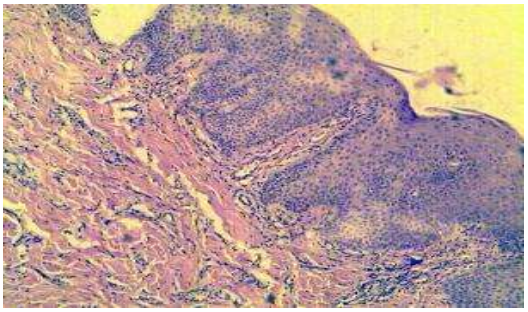
21. Accumulation of connective tissue cells of the inflammatory infiltrate on the surface of the adhesions. G-E 10x40.



22. Reduction of the number of connective tissue cells of the inflammatory infiltrate on the surface of the adhesions. G-E 10x40



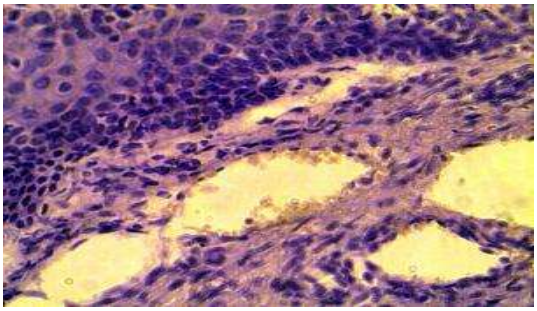
23. Reduction of the number of connective tissue cells of the inflammatory infiltrate on the surface of the adhesions. G-E 10x40



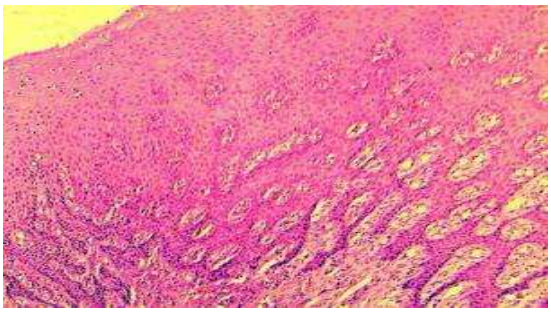
24. Absence of connective tissue cells of the inflammatory infiltrate on the surface of the adhesions. G-E 10x40.

At the same time, the number of atelectasis alveoli in the lung parenchyma decreases and their lumen does not contain exudate (Fig. 25-26).

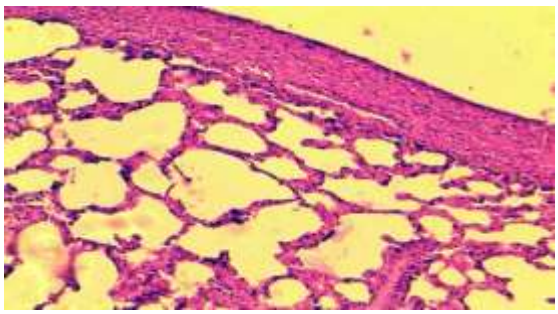
Upon completion of the course impact Garazel in the pleural cavity can not be detected spikes. The visceral pleura is thin and does not contain connective tissue cells (Fig. 27).



25. Restoration of alveolar airiness by the end of the course of Geoprocel exposure . G-E 10x40.



26. Absence of connective tissue cells of the inflammatory infiltrate on the surface of the adhesions. G-E 10x10.



27. Restoration of alveolar airiness by the end of the course of Geoprocel exposure. G-E 10x10.

The conducted studies have shown the positive effect of Hepracel as an anti-adhesive agent. Hepracel has a multi-sided effect on the anti-adhesive process. The appearance of adhesions is promoted by a pronounced inflammatory process in the pleural cavity. The inflammatory process causes manifestations of atelectasis of the alveoli and the accumulation of exudation fluid in the lumen of non-atelectatic alveoli. Hepracel, on the one hand, contributes to a pronounced reduction in the inflammatory effect of agents that cause the formation of adhesions, and on the other hand, reduces the manifestations of alveolar atelectasis, contributing to the reduction of adhesions and the restoration of lung excursion.

CONCLUSIONS:

The conducted research allowed to draw the following conclusion: in the formation of adhesions in the pleural cavity in the experiment established that in the control group in all cases is the development of adhesion, with 53.3 percent of the gross adhesions are formed of strands, and in 46.7% of the plane fusion, in turn, the use of domestic means Gabriel activated by the addition of serum helped reduce the frequency of postoperative adhesion formation to 33.3%, of which only 6.7% was in the form of strands and 26.7% - planar adhesions ($\chi 2=16,263$; $Df=2$; $p<0.001$). Morphological studies have shown a positive effect of Hepracel as an anti-adhesive agent after operations on the pleural cavity, characterized by a regression of the activity of the inflammatory process and local phenomena of alveolar atelectasis with the accumulation of exudation fluid in their lumen. This reduces the number of fibroblasts in the damaged area and prevents the formation of adhesions.

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