Peritoneal Adhesions and Possible Mechanisms of Their Formation

I. M. Baibekov, K. Madartov, and V. A. Khoroshaev

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 122, No. 11, pp. 589-593, November, 1996
Original article submitted September 10, 1995

Morphological study of peritoneal adhesions induced mechanically or with high-energy CO_2 and Nd-YAG lasers shows that adhesion formation does not occur in all cases, the process being dependent on the severity of damage and the rate at which the damaged surface is closed by mesothelial cells. The occurrence of adhesions is lower after mechanical or CO_2 laser-induced injury than after irradiation with an Nd-YAG laser. The structure of adhesions forming after various types of peritoneal injury is virtually the same. Several stages are distinguished in the process of adhesion formation.

Key Words: *peritoneum; high-intensity lasers; adhesions; morphology*

The structure and formation of adhesions have attracted the attention of scientists for a long time [6-8]. The peritoneum plays a key role in resorption and transudation of peritoneal fluid and is capable of forming adhesions in response to various stimuli [1]. Adhesions curb the infection, on the one hand, and induce the development of so-called adhesion disease, on the other [2,4,5].

The morphology and formation of adhesions have not been studied in detail since the description of the morphology of intestinal obstruction resulting from adhesions, classification of adhesions, and description of their structure [3]. Adhesions were never investigated by scanning and transmission electron microscopy (SEM and TEM, respectively). Meanwhile, adhesions and adhesion disease remain the major complications in abdominal surgery.

Our objective was to examine adhesions by SEM and TEM and to establish the cause of their formation.

MATERIALS AND METHODS

Male Wistar rats weighing 120-150 g were used in the study. Parietal and visceral peritoneum was perforated mechanically or with the use of high-energy CO_2 or Nd-YAG laser. Median laparotomy was performed under ether anesthesia, and the peritoneum of the lateral abdominal wall and the small intestine loop was injured. The animals were sacrificed by decapitation. Material for optic, scanning, and transmission electron microscopy was collected 2 h up to 2 weeks after the injury.

RESULTS

Three patterns of wound healing were distinguished: 1) rapid closure of the defect by mesothelial cells without formation of adhesions between peritoneal surfaces, 2) closure of the wound with the membranous part of the greater omentum, and 3) formation of adhesions between the injured peritoneal leaves.

The mesothelial coating remained disrupted during the first 24 h after injury; the wound was covered with the peritoneal exudation cells, primarily macrophages and lymphoid cells, some erythrocytes, and fibrin fibers (Fig. 1, *a*). The capillaries adjacent to the wound were plethoric, the interstitial space was edematous, and the connective tissue was infiltrated with macrophages, occasional neutrophil leukocytes, and lymphoid cells. Mesothelial cells boarding the wound were cube-shaped, the apical surface of their

Department of Pathological Anatomy, Surgery Research Center, Ministry of Health, Republic of Uzbekistan, Tashkent

0007-4888/96/0011-1161$15.00

Fig. 1. Reaction of the peritoneum to mechanical and high-energy laser exposure. *a*) mechanical exposure (day 1): accumulation of the peritoneal fluid cells on the wound surface; fibrin precipitation, ×1000, SEM; *b*) closure of the wound by mesothelial cells after irradiation with an Nd-YAG laser (day 3), ×700, SEM; *c*) fixation of the membranous part of the greater omentum to the wound surface, day 3 after mechanical injury, ×100, hematoxylin and eosin staining; *d*) fixation of the membranous part of the greater omentum to the site of injury, day 3 after exposure to a CO_2 laser, ×100. Semithin sections stained with methylene blue and fuchsin.

cytoplasmic membrane protruded into the lumen of the peritoneal cavity, the number of microvilli decreased, the microvilli became shorter, and cell-to-cell spaces were dilated.

On day 3 after injury, in some animals the wound was completely covered with elongated or spindle-shaped mesothelial cells without or with few microvilli (Fig. 1, *b*). Electron microscopy revealed widened rough endoplasmic reticulum and oval or elongated nuclei with invaginations into the cytoplasm. The mitochondria were small and had electron-dense matrix and few cristae; mitoses were observed. Cellular infiltration of the connective tissue peritoneum increased, with macrophages and lymphoid cells pre-

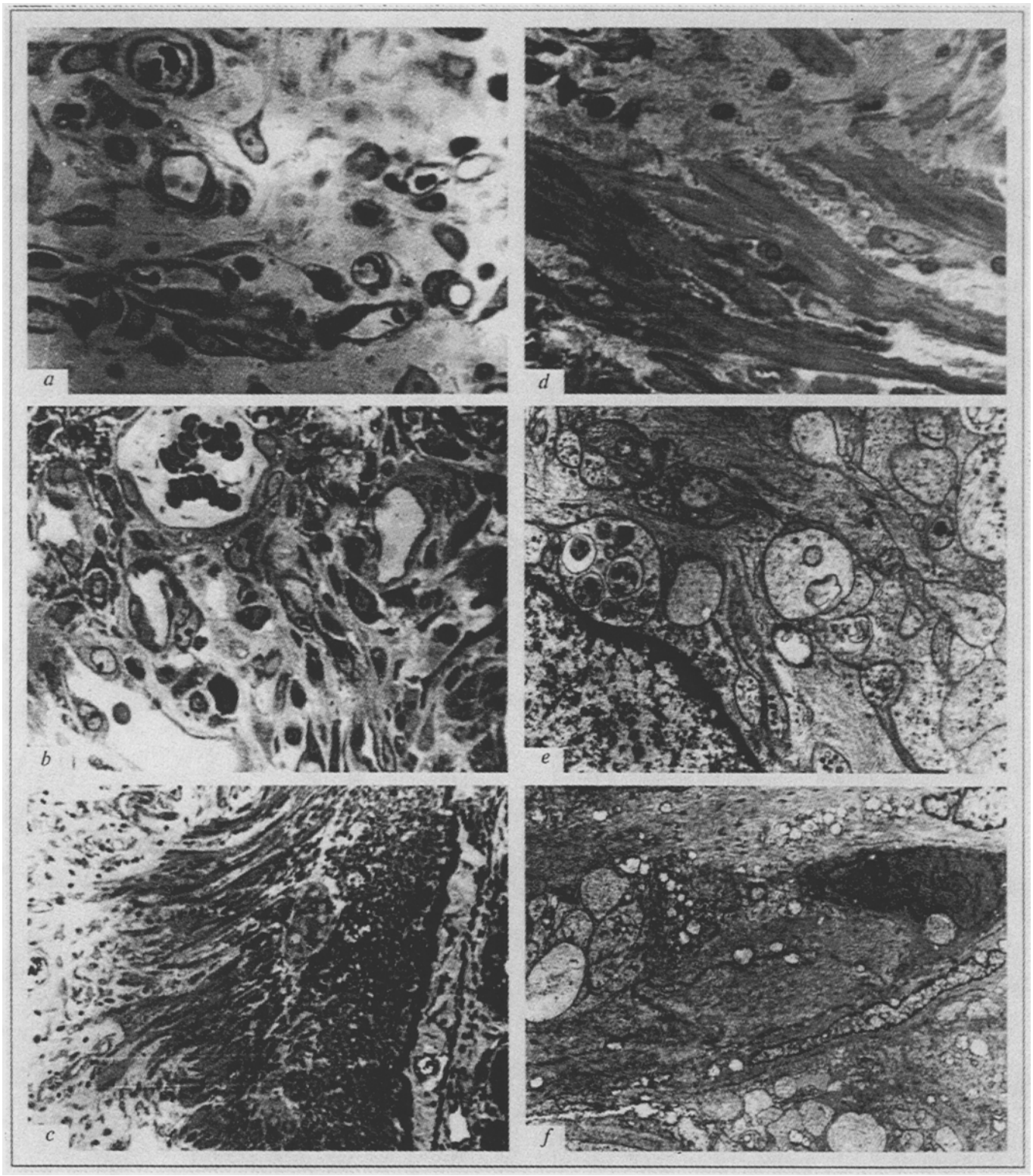

Fig. 2. Structure of peritoneal adhesions after mechanical or laser-induced injury. *a*) blood capillaries, poorly differentiated cells, and young fibroblasts in adhesion 3 days after irradiation with a CO_2 laser, ×400. Semithin section, methylene blue and fuchsin staining; *b*) increased amount of fibroblasts and collagen fibrils in adhesion, day 7 after irradiation with a CO_2 laser, ×400. Semithin section, methylene blue and fuchsin staining; c) penetration of smooth-muscle cells from the tunica media of the small intestine into adhesion, day 7 after exposure to an Nd-YAG laser, ×100. Semithin section, methylene blue and fuchsin staining; *d*) smooth muscle cells in adhesion, day 14 after irradiation with a CO_2 laser, ×400. Semithin section, methylene blue and fuchsin staining; *e*) smooth muscle cells in adhesion. Invagination of the nuclear membrane, decreased amount of myofilaments in the cytoplasm, day 14 after irradiation with a CO_2 laser, ×1500. TEM; *f*) bundles of unmyelinated fibers in adhesion, day 14 after irradiation with a CO_2 laser, ×5000. TEM.

dominating. After 1-2 weeks, in some animals the wound was completely covered with ultrastructurally normal mesothelial cells; under SEM the wound was virtually indistinguishable from intact peritoneum. Fibrosis was discernible in the connective tissue.

In some animals, the greater omentum was fixed to the wound surface as early as on day 1 after injury (Fig. 1, *c*). This process involved predominantly the membranous portion of the omentum adjacent to the wound. It was paralleled by "dissociation" of mesothelial cells and connective tissue elements and plethora of capillaries located in the connective tissue septae. Macrophages and poorly differentiated cells infiltrated the wound area. Three days after injury, vascularization of the membranous part of the greater omentum fixed to the wound was intensified, and the number of cells increased: young fibroblasts and macrophages appeared in the wound bottom (Fig. 1, *d*).

After 1-2 weeks, the membranous portion of the greater omentum fixed to the wound looked like a thin bundle lined with mesothelial cells. The bundle was composed of collagen fibrils and blood vessels between them.

About 30% of animals developed adhesions between the damaged sites of the peritoneum. The damaged sites contacted with each other on days 1-3 with formation of fibrin fibers. Pronounced edemas were seen at the contacts, the amounts of macrophages and monocytes increased; occasional lymphoid cells and neutrophilic polymorphonuclear leukocytes were seen. Microvessels adjacent to the wound were plethoric. On day 3, fibrosis of the adhesion zone was noted: young fibroblasts with well-developed granular endoplasmic reticulum and Golgi complex were seen. The amount of collagen fibrils was increased. New blood capillaries were formed (Fig. 2, *a*). Seven days after injury, adhesions consisted predominantly of fine collagen fibrils and blood vessels (capillaries and postcapillary venules) with a thin vascular wall (Fig. 2, *b*). The amount of fibroblasts remained high. This period of adhesion formation was characterized by the appearance of occasional smooth-muscle cells originating from the tunica media of the small intestine. The tunica media "dissociated", and smooth-muscle cells migrated into the depth of the connective tissue (Fig. 2, *c*).

Two weeks after injury, adhesions consisted predominantly of the connective tissue and smooth-muscle cells (Fig. 2, *d*). In contrast to the intestinal walls, the cells were loosely packed. The surface of adhesions was lined with a massive layer of mesothelial cells with normal structure, their apex being covered with numerous microvilli. The connective tissue contained fewer capillaries than at previous times after injury. Cellular infiltration was less pronounced. This period was characterized by the appearance of unmyelinated nerve fibers in adhesions. The fibers were arranged in bundles or lay separately (Fig. 2, *e*). The ultrastructure of smooth muscle cells differed from that of the small intestine smooth muscle cells: the cells contained fewer microfilaments, the cisternae of rough endoplasmic reticulum and Golgi complex as well as the perinuclear space were dilated, and the nuclear membrane was invaginated (Fig. 2, *f*). As a result, the cytoplasm of the majority of leiomyocytes was transparent. The amount of connective tissue in adhesions decreased, the tissue became compact due to formation of collagen bundles. Cell composition of adhesions also changed: the amount of macrophages and lymphoid cells decreased, Golgi complex and rough endoplasmic reticulum in the fibroblasts shrunk, and tissue basophils containing secretory granules appeared.

Thus, adhesions did not form in all animals; this process depended on the severity of injury and the rate at which mesothelial cells closed the damaged surface. The occurrence of adhesions was higher after mechanical and CO_2 laser-induced damage than after that caused by irradiation with an Nd-YAG laser. The structure of adhesions resulting from different types of injury was virtually the same.

The process of adhesion formation includes the following stages:

1. Adhering of the damaged sites of the peritoneum as a result of fibrin precipitation.
2. Migration of poorly differentiated cells to the wound and subsequent appearance of collagen-producing fibroblasts and growth of blood capillaries.
3. Migration of smooth muscle cells from the small intestine wall into adhesions.
4. Gradual replacement of connective tissue by smooth muscle cells in parallel with recalibration of microvessels and growth of unmyelinated nerve fibers.

REFERENCES

1. M. A. Baron, *Reactive Structures of the Internal Membranes* [in Russian], Leningrad (1949).
2. R. A. Zhenchevskii, *Adhesion Disease* [in Russian], Moscow (1989).
3. A. I. Strukov (Ed.), *Manual of Pathological Anatomy* [in Russian], Vol. 4, Books 1, 2, Moscow (1956, 1957).
4. K. S. Simonyan, *Adhesion Disease* [in Russian], Moscow (1966).
5. D. P. Chukhrienko, I. S. Belyi, and V. A. Bondarenko, *Adhesion Disease* [in Russian], Kiev (1972).
6. H. Ellis, *Br. J. Surg.*, **50**, 10-16 (1962).
7. H. Myllarniemi and V. Karppienen, *Ibid.*, **55**, 606-608 (1968).
8. D. S. Shade and J. R. Williamson, *Ann. Surg.*, **167**, 500-510 (1968).