

5. A. G. Khomenko, V. V. Erokhin, G. M. Nikolaeva, et al., *Probl. Tuberk.*, No. 1, 34 (1984).
6. A. J. Barrett, *Lysosomes*, Amsterdam (1972), pp. 46-135.
7. R. M. Du Bois, *Thorax*, 40, 321 (1985).
8. N. V. Herscowitz, *Ann. Allergy*, 55, 634 (1985).
9. O. H. Lowry, N. J. Rosebrough, A. L. Farr, et al., *J. Biol. Chem.*, 193, 265 (1951).
10. A. R. Nath, A. V. Hegde, and P. J. Mehta, *J. Assoc. Physicians India*, 33, 287 (1985).
11. L. Visser and E. R. Bout, *Biochim. Biophys. Acta*, 268, 257 (1972).

MORPHOLOGICAL EVALUATION OF RECONSTRUCTIVE OPERATIONS ON THE BRONCHI USING EXPERIMENTAL MICROSURGICAL TECHNIQUES

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Of all reconstructive operations on the bronchi 5-10% are undertaken on patients with cancer and 70% on patients with benign tumors [1, 5, 7, 11, 12]; suture failure, and granulations and cicatricial stenosis of the anastomoses develop in 5-10% of patients [4, 8-10]. As a rule, interrupted sutures passing through all layers of the bronchial wall are used in the formation of bronchial anastomoses. A high-precision method of formation of the bronchial anastomosis by a microsurgical technique must create the most favorable conditions for healing by first intention. The aim of the present investigation was accordingly a morphological comparison of the processes of healing of a bronchial anastomosis formed by a microsurgical technique and by the traditional method with through-and-through sutures.

EXPERIMENTAL METHOD

Experiments were carried out on 30 mongrel dogs. An anastomosis between the left main and lower-lobe bronchi was formed after upper bilobectomy, with circular resection of the main bronchus. The anastomosis in 18 dogs of the experimental series was formed by a method developed by ourselves: after removal of the upper and cardiac lobes of the left lung, in order to create an excess of mucous membrane, the mucosa was separated, under an OM-2 operating microscope with magnification of 6 times, from the extreme cartilaginous half-ring of the main bronchus with a razor blade, fixed in a holder, and half the circumference of the extreme cartilaginous half-ring was excised. A similar manipulation was performed on the lower lobe bronchus. Next, without the operating microscope, three external Lavsan thread 3/0 external sutures were applied to the posterior half-circumference of the anastomosis, between the mucous membrane and cartilage or muscular layer of the membranous part. Under the operating microscope with magnification of 6 times, a continuous suture of 7/0 thread was applied to the mucous membrane. Next, external sutures were applied to the anterior half-circumference of the anastomosis with 3/0 thread, not including the mucous membrane. On 12 dogs of the control series the anastomosis was formed by interrupted sutures passing through all layers of the bronchial wall. In the postoperative period bronchoscopy was performed on all the animals at various times — from 2 days to 1 month. The animals were removed from the experiment 2-5, 7-8, 14-16, and 28-30 days after the operation. At autopsy, the state of the anastomosis was assessed macroscopically, after which the region of anastomosis of the bronchus was removed en bloc and fixed in 10% buffered formalin by Lillie's method. Pieces of tissue were excised from different parts of the anastomosis and embedded in paraffin wax. Sections were stained with hematoxylin and eosin, with picrofuchsin-fuchselin, by the PAS reaction, and by Gomori's impregnation method. Pieces of mucosa from the anastomosis measuring 0.3×0.3 cm were processed by Rostovshchikov's method for scanning electron microscopy [6] and examined in the Hitachi S 500 microscope.

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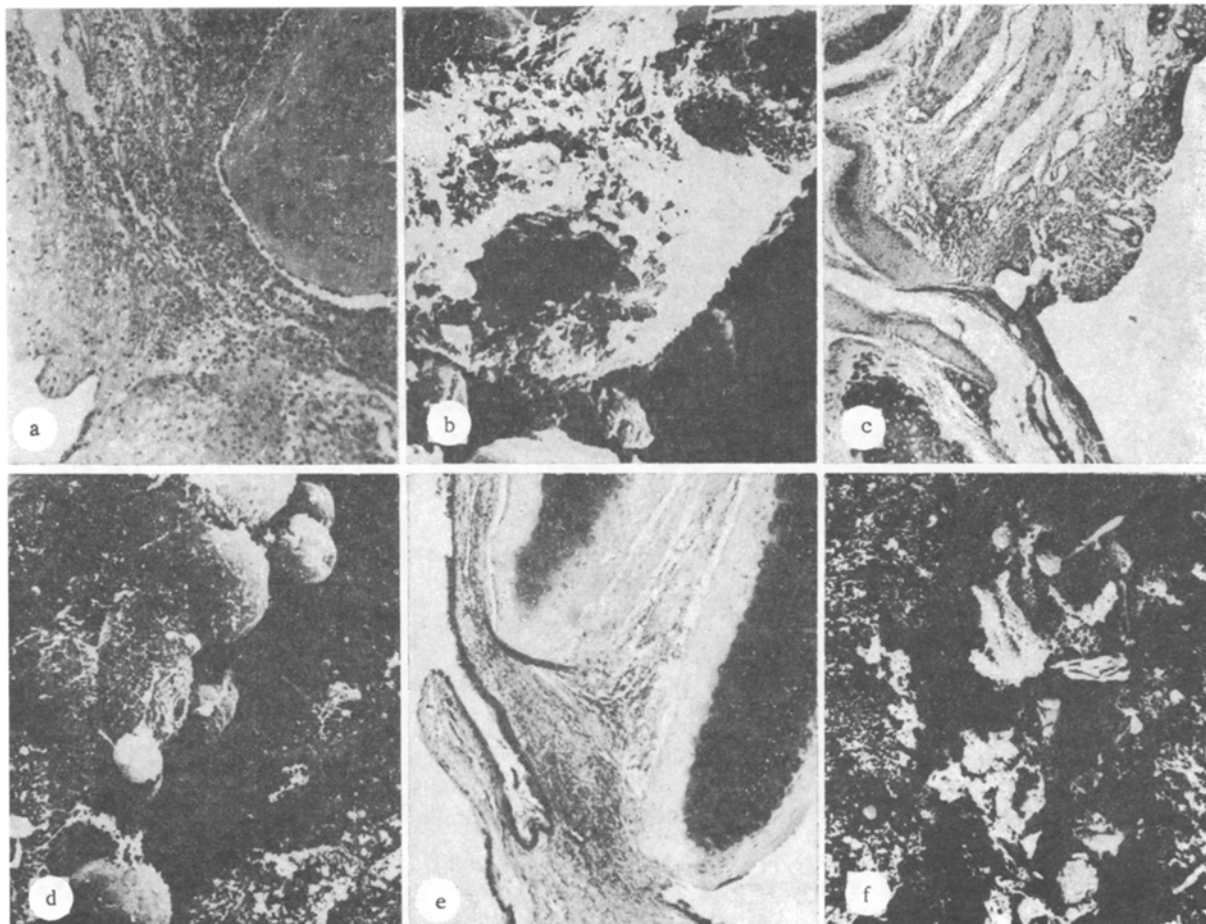


Fig. 1. Morphological picture (a, c, e) and stereoscopic ultrastructure of surface (b, d, f) of anastomosis formed by means of interrupted sutures passing through all layers of bronchial wall, at different times after operation. Hematoxylin and eosin. a) 2nd day, 160 \times ; b) 2nd day SEM, 2500 \times ; c) 8th day, 63 \times ; d) 8th day, SEM, 2000 \times ; e) 27th day, 63 \times ; f) 27th day, SEM, 600 \times .

EXPERIMENTAL RESULTS

Control Series. At bronchoscopy 2 days after the operation the mucous membrane in the region of anastomosis was hyperemic and edematous, with fibrin deposits. Partial epithelization of the fibrin film with one or two rows of flattened epithelium was observed histologically (Fig. 1a). Edema and inflammatory infiltration with polymorphs in the region of anastomosis spread to all layers of the bronchial wall. The PAS reaction revealed no goblet cells in the epithelial layer. On scanning electron microscopy (SEM) deposition of fibrin and leukocytes was observed (Fig. 1b).

After 1 week the edema and hyperemia of the mucous membrane were less marked than after 2 days. The anastomosis was covered in some places by fibrin films and mucus. Histologically, proliferation of granulation tissue, invading the lumen of the bronchus, could often be seen (Fig. 1c). Inflammatory infiltration with polymorphs, lymphocytes, plasma cells, and histiocytes was well marked along the course of the anastomosis and in the lamina propria of the mucous membrane throughout. The stereoscopic ultrastructure of the surface of the mucosa was uneven and often modular. Escape of leukocytes onto the surface was observed through the widened intracellular spaces (Fig. 1d). After 2 weeks, at sites of adequate apposition of the mucous membrane no inflammatory phenomena could be observed. At sites of incomplete apposition, fibrin deposits and granulations were present. Deformation of the mucosa in the form of folds was frequently observed. Focal fibrosis was well marked. After 3 weeks the anastomosis appeared at bronchoscopy as a whitish scar with threads of suture material showing through the mucous membrane. Histologically the epithelial layer in the region of the anastomosis was completely restored. The PAS reaction revealed a concentration of goblet cells in the epithelial layer. At bronchoscopy after 1 month the anastomosis had

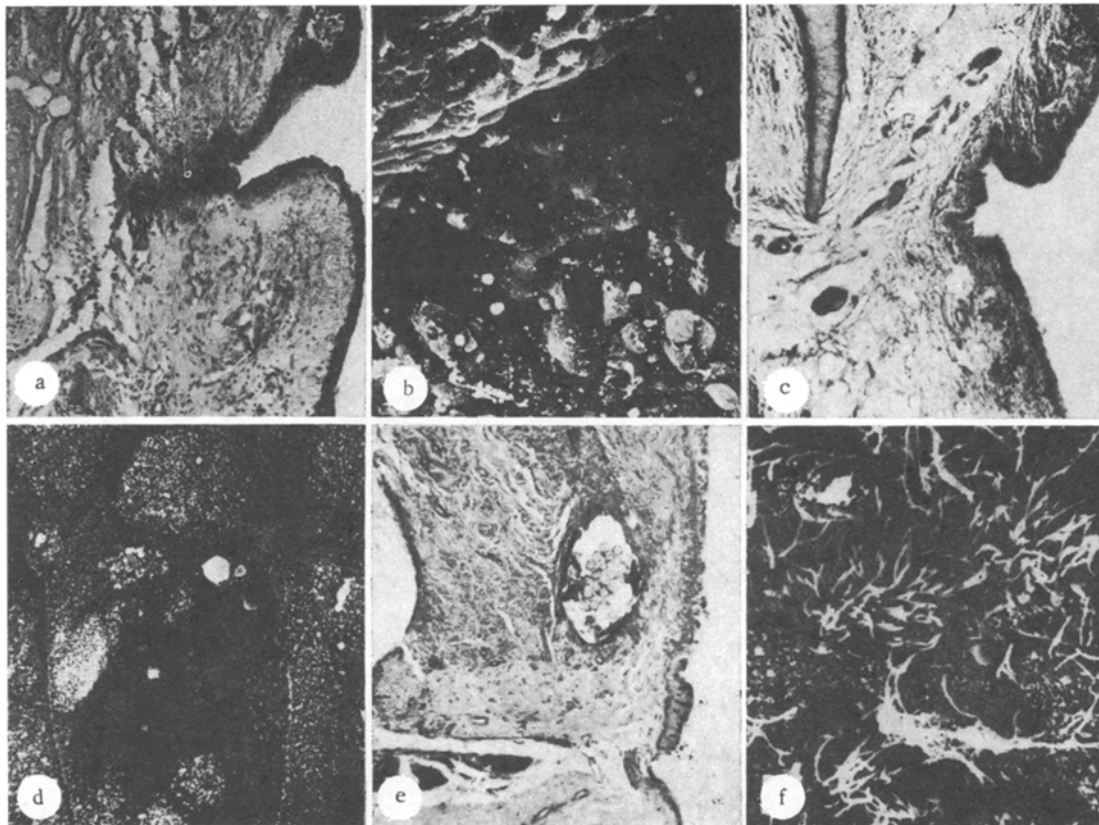


Fig. 2. Morphological picture (a, c, e) and stereoscopic ultrastructure of surface (b, d, f) of anastomosis formed by a microsurgical technique, at different times after operation. a) 2nd day, 160 \times ; b) 2nd day, SEM, 2500 \times ; c) 8th day, stained with picrofuchsin-fuchselin, 63 \times ; d) 8th day, SEM, 2500 \times ; e) 21st day, PAS reaction, 160 \times ; f) 21st day, SEM, 3000 \times .

the appearance of a delicate scar; where apposition of the mucous membrane was incomplete, the development of scar tissue was observed at the site of the previous granulations. Histologically, folding of the mucous membrane was observed in the region of anastomosis. Although the epithelial layer was completely restored, in some places it was loose in texture and partly detached. The lamina propria of the mucous membrane showed fibrous changes with focal infiltration of lymphocytes, plasma cells, histiocytes, and fibroblasts (Fig. 1e). The stereoscopic ultrastructure of the surface of the mucosa was uneven, with deposits of desquamated epithelial cells and mucus (Fig. 1f).

A characteristic feature of the histological picture of the control series at all times of observation was disorganization of the complex structure of the cartilaginous laminae, which sometimes protruded into the mucous membrane, deforming it.

Experimental Series. At bronchoscopy after 2 days the anastomosis line was even; hyperemia and edema were present, but less marked than in the control. Adequate apposition of the edges was observed histologically. Circulatory disturbances and inflammatory infiltration with polymorphs were observed along the course of the anastomosis, but everywhere they were less marked than in the control. On the 5th day complete restoration of the stratified cylindrical epithelium was observed (Fig. 2a). In the region of the anastomosis edema and moderate infiltration with polymorphs, lymphocytes, plasma cells, and histiocytes were detected. On SEM the epithelial layer appeared normal but no ciliary apparatus could be identified (Fig. 2b). At bronchoscopy after 1 week, small submucous hemorrhages could be seen along the course of the anastomosis. Histologically, the line of the anastomosis could not always be identified (Fig. 2c). The epithelial layer was restored and inflammatory infiltration was weak. The stereoscopic ultrastructure of the surface of the epithelial layer was even and consisted of cells with microvilli, but the cilia were not yet restored (Fig. 2d). At bronchoscopy after 2 weeks the anastomosis was free from inflammatory phenomena and the mucous membrane was pale pink and glistening. Histologically, the epithelial layer was stratified and contained

goblet cells; sometimes small foci of fibrosis without inflammatory infiltration could be detected in the lamina propria. After 3 and 4 weeks on visual examination the anastomosis consisted of a whitish scar against the pink background of the mucous membrane. Histologically, the epithelium was stratified and the PAS reaction revealed goblet cells in it (Fig. 2e). On SEM the surface of the epithelial layer in the region of the anastomosis consisted of differentiated ciliated and goblet cells (Fig. 2f).

The investigations revealed some particular features of the course of repair processes in a bronchial anastomosis formed by different methods. In the control group some degree of inadequate apposition of the sutured segments could be observed. This was reflected in superposition of one edge on the other (as a rule, the distal edge on the proximal), and disorganization of the complex structure of the cartilaginous laminae. The reasons for inadequate apposition, in our opinion, are differences in the diameters of the sutured segments and the compressing force of the sutures, which deform the cartilaginous rings. The diastasis arising between the edges of the mucous membrane as a result of inadequate apposition of the segments is filled with granulation tissue, and this considerably delays epithelization. In cases of better apposition of the edges of the mucous membrane epithelium covers the wound by the 4th-5th days, in agreement with data published by other workers [2, 3]. In the experimental group, in which the anastomosis was formed by a microsurgical technique, apposition of the edges of the mucous membrane was more adequate, for they were firmly sutured together by thin thread, and the external sutures on the cartilaginous skeleton were tightened minimally, just sufficiently to produce contact of the cartilaginous half-rings. Close contact between the edges of the mucous membrane led to rapid epithelization of the anastomosis. The less severe trauma to the mucous membrane by microsurgical instruments induced a weaker inflammatory reaction, which disappeared earlier.

Comparative analysis of the results of bronchoscopy and of light and electron microscopy thus shows that a bronchial anastomosis formed by a microsurgical technique gives more adequate apposition of the edges of the segments to be sutured than in the control. Epithelization of a bronchial anastomosis formed by a microsurgical technique takes place earlier than in the control. The inflammatory reaction in the region of an anastomosis formed by a microsurgical technique is weaker and disappears earlier than in the control.

LITERATURE CITED

1. Yu. V. Biryukov, S. P. Grigor'eva, and S. R. Dobrovol'skii, *Surgery of the Trachea and Bronchi* [in Russian], Moscow (1986), p. 21.
2. Yu. E. Vyrenkov, *Important Problems in Contemporary Surgery* [in Russian], Petrozavodsk (1974), p. 23.
3. A. P. Kuz'michev, *Éksp. Khir.*, No. 2, 18 (1963).
4. M. I. Perel'man, S. P. Grigor'eva, and S. R. Dobrovol'skii, *Grudn. Khir.*, No. 2, 36 (1981).
5. B. V. Petrovskii, M. I. Perel'man, and N. S. Koroleva, *Tracheobronchial Surgery* [in Russian], Moscow (1978).
6. A. S. Rostovshchikov, "Pathomorphological analysis of the nasal mucous membrane at high altitudes," Author's abstract of dissertation for the degree of Candidate of Medical Sciences, Moscow (1983).
7. V. V. Utkin, Ya. Ya. Bashko, and M. Ya. Yudin, *Surgery of the Trachea and Bronchi* [in Russian], Moscow (1986), p. 111.
8. V. P. Kharchenko, V. D. Chkhikvadze, I. V. Kuz'min, et al., *Surgery of the Trachea and Bronchi* [in Russian], Moscow (1986), p. 118.
9. S. Attar, J. Miller, J. Hankins, et al., *Ann. Thorac. Surg.*, 40, 126 (1985).
10. H. Ayabe, Y. Nakamura, T. Miura, et al., *World J. Surg.*, 6, 433 (1982).
11. J. Lowe, A. Bridgemann, and D. Sabiston, *J. Thorac. Cardiovasc. Surg.*, 83, 227 (1982).
12. I. Ungar, I. Gyeney, E. Sherer, et al., *Thorac. Cardiovasc. Surg.*, 29, 41 (1981).