

erythropoiesis and granulomonocytopoiesis observed during stress, but also to changes characteristic of the general adaptation syndrome, observed in their proliferative status. Consequently, one manifestation of regulatory influences of lymphocytes of thymic origin on hematopoiesis is triggering of E-CFU<sub>c</sub> and GM-CFU<sub>c</sub> for proliferation, followed by an increase in their number, the basis for development of the phenomenon of hyperplasia of medullary hematopoiesis in stress.

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#### ACTION OF HELIUM-NEON LASER RADIATION ON THE TRACHEAL AND BRONCHIAL MUCOSA: AN EXPERIMENTAL STUDY

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Published reports on the use of low-intensity laser radiation in pulmonology [2, 4, 6] have described properties of coherent light such as ability to intensify metabolic processes, to accelerate wound healing, to relieve the pain syndrome, and to enhance immunity [1, 3, 7, 9]. Some publications have described the biosynthetic capacity of laser radiation in lung tissue culture [5, 8]. No experimental research into the action of energy of a helium-neon laser (HNL) on the bronchial and tracheal mucosa could be found in the accessible literature. The aim of the present investigation was to study the effect of exposure to coherent red light on the mucosa of the tracheobronchial tree and to choose optimal parameters of irradiation for clinical use.

#### EXPERIMENTAL METHOD

The LG-75 laser system with the following parameters was used: power at the output of the emitter 20 mW, power at the output of the light guide 10 mW, thickness of the quartz thread of the light-conducting cable 400  $\mu$ m, distance from the end of the light guide to the object 1 cm. The emission energy was calculated by the equation:  $E \text{ (in J)} = P \text{ (in mW)} \times T \text{ (in sec)}$ . The dose was calculated by the equation:  $W \text{ (J/cm}^2\text{)} = E \text{ (J)} / S \text{ (cm}^2\text{)}$ . In acute experiments (61) on 14 gray rabbits (weighing 2-5 kg) the mucosa of the trachea and bronchi was irradiated after preliminary tracheotomy and bronchotomy. Group 1 comprised seven animals, whose tracheal mucosa was irradiated. Under pentobarbital anesthesia (40 mg/kg body weight, intravenously) the trachea was exposed. The anterior wall of the trachea was removed 2 cm

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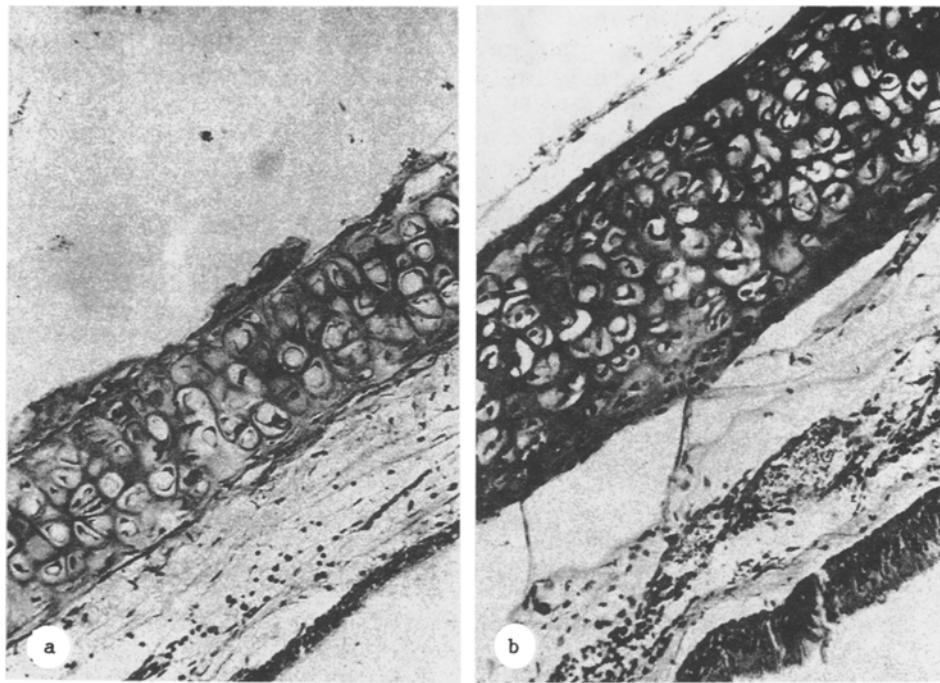


Fig. 1. Action of HNL on bronchial wall: a) edema of bronchial submucosa with solitary inflammatory cells. Here and in Fig. 2, stained with hematoxylin and eosin, 250 $\times$ . Exposure 4 min, dose 17.12 J/cm<sup>2</sup>; b) congestion of blood vessels, perivascular accumulation of inflammatory cells. Exposure 22 min, dose 105.16 J/cm<sup>2</sup>.

below the thyroid cartilage, leaving a defect measuring 0.5  $\times$  1.5 cm. The posterior wall of the trachea was washed free from mucus and blood with warm sterile physiological saline, followed by aspiration. The light guide was applied at a right angle to the posterior wall of the trachea at a distance of 10 mm. The diameter of the spot was 4 mm, with a halo of 2 cm<sup>2</sup>. The power density under these conditions was 7.9 mW/cm<sup>2</sup>. Films were obtained from the irradiated surfaces, squash preparations were made on slides warmed to 37°C, because cold glass induced negative taxis in the mobile cells and deprived them of their adhesion properties. The first squash preparation was taken before irradiation. During irradiation, squash preparations were taken after exposures of: 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 15, 20, 21, 22, 25, 30, 40, 45, 50, 60, and 75 min. Group 2 also comprised seven animals whose bronchial mucosa was irradiated (bronchi of the 1st, 2nd, and 3rd orders). Tracheostomy was performed 2 cm below the thyroid cartilage of the larynx and the trachea was intubated. The PVC intubation tube was advanced into the right main bronchus and connected to the artificial respiration apparatus. Under controlled breathing, thoractomy was performed in the 4th left intercostal space. After separation of the corresponding bronchi (by blunt dissection) from the lung tissue, bronchotomy was performed. The edges of the bronchotomy wound and the irradiated bronchus were securely fixed so that the light guide was located perpendicularly to the irradiated surface and at a distance of 10 mm from it. Blood and mucus were removed from the lumen. Squash preparations were taken after the same exposures as in group 1. The preparations obtained were dried, then stained with a 20% alcoholic solution of hematoxylin and eosin. They were then examined under the light microscope with immersion objective and under a magnification of 90. Material taken intravitaly for histological investigation was kept in 10% formalin solution. Paraffin sections were stained with hematoxylin and eosin and with picrofuchsin by Van Gieson's method.

#### EXPERIMENTAL RESULTS

During irradiation of the tracheal and bronchial mucosa with HNL no change in color, consistency, shape of the tissues, or intensity of secretion or bleeding was detected visually.

Under the influence of irradiation of HNL with a power of 10 mW and with exposures of 5 to 75 min (energy varying from 3 to 45 J, corresponding to doses of 23.9 to 358.5 J/cm<sup>2</sup> on the tracheal and bronchial mucosa of healthy animals destructive changes took place in the

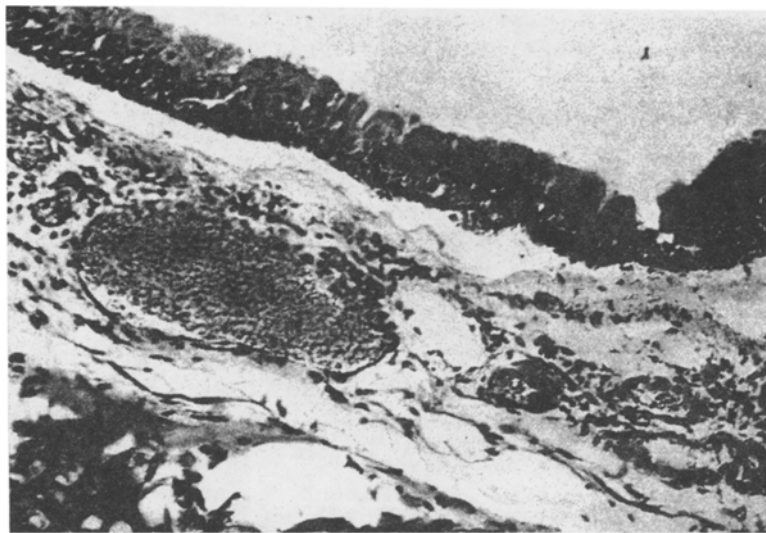


Fig. 2. Action of HNL on wall of a large bronchus. Picture of hyperemia and sludging in vessels of submucosa of main bronchus with numerous inflammatory cells. Exposure 45 min, dose 215.1 J/cm<sup>2</sup>.

membrane, as shown by the presence of a floccular, finely granular, protein-lipid component in the tissue debris, of lipid crystals and drops, and also of their aggregation with fragments of damaged cells. Inflammatory cells were absent. In the squash preparations from 5 of the 14 animals pathological forms of erythrocytes were seen, with swellings and with intracellular foaming.

We saw fine structural changes in the cells as a result of the action of the laser beam on the tissues with exposures of 1 to 4 min (energy from 0.6 to 2.4 J), which corresponds to doses of between 4.78 and 17.12 J/cm<sup>2</sup>. A microscopic study of the cell composition of the squash preparations revealed activation of phagocytes, expressed as positive taxis toward the foci of irradiation, an increase in the number of pseudopodia, potentiation of phagocytic activity, and the ability of the cells to cooperate, and to respond actively to foreign agents. Alveolar macrophages appeared in the foci of irradiation and proliferative activity of histiocytes and fibroblasts was increased. Meanwhile, in individual animals (four rabbits) pathological forms of erythrocytes were found at the 2nd minute, and in three animals erythrocytes aggregated into "rouleaux." Not only damage to cell surface membranes, but also lysis of intracellular organelles, with vacuolation and edema of the cells, were observed in the phagocytes of these animals (if motor activity was present). These types of changes in the membranes we interpreted as functional and reversible, for the cells remained structurally intact. In all the experimental animals, after irradiation for 1 min a marked cellular response was observed, with structural reorganization of the membranes, maintaining their functional activity.

The study of histological preparations of the tracheobronchial structures of animals after irradiation for not more than 15 min showed that changes were not present in the zone of the ciliated epithelium. In the submucosa of the trachea and bronchi a picture of marked edema was observed, with the presence of single inflammatory cells (Fig. 1a).

In the submucosa of the tracheobronchial tree of animals irradiated for 21, 22, and 25 min edema was moderate, the blood vessels were congested, and perivascular accumulation of inflammatory cells was observed (Fig. 1b). In animals irradiated for 30, 45, 60, and 75 min, a picture of a marked sludge syndrome was observed in the vessels of the submucosal layer, and many inflammatory cells could be seen in the tissues (Fig. 2).

Thus exposure of the mucosa of tracheobronchial structures to energy of the HNL in a dose of up to 358.5 J/cm<sup>2</sup> did not cause irreversible changes in the tissues. Exposure of these formations to a dose of between 17.12 and 71.35 J/cm<sup>2</sup> stimulated metabolism, and this was reflected in congestion of the blood vessels and an increase in the number of inflammatory cells in the tissues.

Examination of squash preparations under the microscope showed that laser irradiation of the tracheal and broncheal mucosa of a healthy animal with an energy of 3 J (dose 17.12

J/cm<sup>2</sup>) or higher, corresponding to an exposure of more than 5 min, caused cytolysis of the mobile inflammatory cells in the focus of irradiation. The stimulating effect of laser radiation with coherent red light, under the conditions stipulated above, was exhibited (without any marked cytolytic action) on cells of healthy animals, when their bronchial and tracheal mucosa was irradiated with energy of between 0.6 and 1.8 J (doses from 4.78 to 14.34 J/cm<sup>2</sup>), corresponding to an exposure of between 1 and 3 min.

The use of HNL to treat children with chronic nonspecific lung diseases compelled us to restrict the exposure to 1 min (energy 0.6 J, dose 4.78 J/cm<sup>2</sup>). This energy, in our opinion, is adequate to give a distinct therapeutic effect. The existence of this minimum of the duration of irradiation in the course of bronchologic treatment means that manipulations under anesthesia need not be prolonged.

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