

of development, when investigated 3 days after the operation, remained capable of metabolizing [3 H]progesterone *in vitro* at the control level [12], is noteworthy. After decapitation no change was found in the weight of the testes, but their testosterone content was sharply reduced compared with the control (3.00 ± 0.36 pmoles/testis in the control; 0.63 ± 0.08 pmole/testis after decapitation [11]). Injection of luteinizing hormone into decapitated fetuses led to an increase in volume of the testis and in the number of Leydig cells up to the level of these parameters in control fetuses, whereas injection of LHRH into decapitated fetuses did not abolish the effect of decapitation [6, 7].

The data obtained in the present investigation, showing a decrease in the testosterone concentration in the testes after encephalectomy and the abolition of this effect by injection of LHRH into the encephalectomized fetuses, as well as data in the literature cited above, are evidence that function of the gonads in male rats is under the control of the fetal hypothalamus by the end of the prenatal period of development. This conclusion is in agreement with the recently published observations of Daikoku et al. [5], concerning the role of endogenous LHRH in the gonadotrophic function of the rat fetal pituitary gland.

LITERATURE CITED

1. M. S. Mitskevich, O. N. Rummyantseva, E. V. Proshlyakova, et al., *Ontogenez*, 1, 631 (1970).
2. S. A. Chiappa and G. Fink, *J. Endocrinol.*, 72, 211 (1977).
3. M. Chowdhury and E. Steinberger, *J. Endocrinol.*, 69, 381 (1976).
4. S. Daikoku, M. Kinutani, and Y. G. Watanabe, *Neuroendocrinology*, 11, 284 (1973).
5. S. Daikoku, T. Adachi, H. Kawano, et al., *Experientia*, 37, 1346 (1981).
6. Y. Eguchi, Y. Sakamoto, K. Arishima, et al., *Endocrinology*, 96, 504 (1975).
7. Y. Eguchi, K. Arishima, T. Nasu, et al., *Anat. Rec.*, 190, 679 (1978).
8. J. E. Jirasek, *Human Fetal Endocrines*, The Hague (1980).
9. H. Kawano, Y. G. Watanabe, and S. Daikoku, *Cell Tissue Res.*, 213, 465 (1980).
10. S. Naessany, R. Habert, and R. Picon, *J. Endocrinol.*, 88, 359 (1981).
11. T. Neomura, J. Weisz, and C. W. Lloyd, *Endocrinology*, 78, 245 (1966).
12. L. H. Zondek and T. Zondek, in: *Endocrinologie Sexuelle de la Période Périnatale*, M. G. Forest and J. Bertrand, eds., Paris (1974), p. 79.

EFFECT OF A HELIUM-NEON LASER ON CELL ULTRASTRUCTURE AND PROLIFERATION OF THE EPITHELIUM OF THE DUODENAL MUCOSA

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KEY WORDS: laser irradiation; duodenum; ultrastructure; cell proliferation.

The widespread use of various types of lasers in medicine has aroused interest in the study of morphological changes produced by coherent radiation in organs and tissues [1-3, 5]. Lasers generating low-power radiation and, in particular, helium-neon lasers, are known not to give rise to severe pathomorphological changes in the tissues, but under the influence of the low-energy radiation activity of metabolic processes is substantially modified [6]. Hence, there is a stimulating effect of helium-neon lasers on the course of repair processes, which is evidently primarily associated with an increase in proliferative activity of the cells under the influence of laser irradiation.

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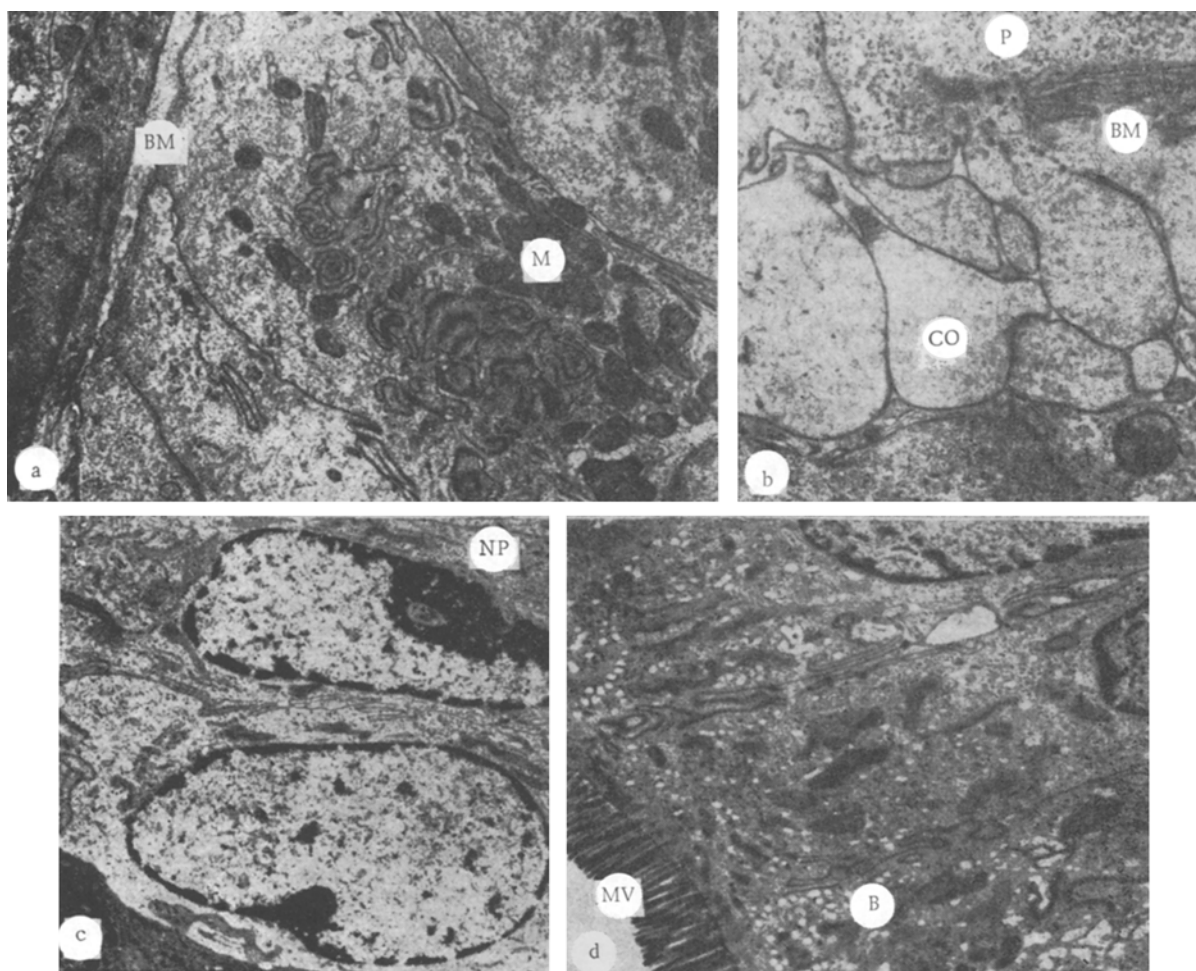


Fig. 1. Epitheliocytes of duodenal mucosa after irradiation by helium-neon laser. a) Basal part of enterocytes, irradiation for 3 min, 5000 \times ; b) basal part of enterocytes, irradiation for 5 min, 8000 \times ; c) nuclei of enterocytes, irradiation for 5 min, 4000 \times ; d) apical part of enterocytes, irradiation for 5 min, 5000 \times . M) Mitochondria; BM) basement membrane; P) polysomes; CO) cytoplasmic outgrowths; NP) nuclear pores; MV) microvilli; V) vesicles.

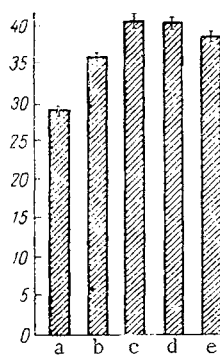


Fig. 2. Incorporation of $[^3\text{H}]$ thymidine into epithelial cells of crypts of duodenum during irradiation by helium-neon laser. a) Normal; b) control; c) irradiation for 1 min, d) for 3 min, e) for 5 min.

The study of proliferation of enterocytes in the duodenal epithelium, which have the highest rate of cell renewal in the mammalian body [4, 7], and also of ultrastructural changes in the mucosa following irradiation by a helium-neon laser, is particularly interesting.

EXPERIMENTAL METHOD

Experiments were carried out on 36 male Wistar albino rats weighing 120-130 g. For the autoradiographic study, at 10 a.m. the animals were given an intraperitoneal injection of [^3H]thymidine in a dose of 18.5 kBq (0.5 μCi)/g body weight. Laparotomy was performed under ether anesthesia and the anterior wall of the duodenum was irradiated by an LG-51-I helium-neon laser. Irradiation began 10 min after injection of [^3H]thymidine. Its duration was 1, 3, and 5 min, wavelength 0.63 μ , dose rate 8 MW, beam diameter 0.6 mm, and diameter of the zone of irradiation 3 mm. The irradiation energy density for a duration of 1 min was 6.78 J/cm 2 , 3 min 20.34 J/cm 2 , and 5 min 33.9 J/cm 2 . Intact animals, and also rats whose duodenum was irradiated with white light under similar conditions served as the control. The animals were killed instantly by decapitation 1 h after injection of [^3H]thymidine. Pieces of the duodenal wall from the zone of irradiation were fixed with 10% formalin by Lillie's method. Paraffin sections were coated with type M emulsion. The index of labeled nuclei (ILN) was calculated by counting at least 1000 cells in longitudinal sections through the crypts. Tissue for electron-microscopic study from the corresponding regions was fixed with glutaraldehyde and OsO_4 and embedded in a mixture of Epon and Araldite. Ultrathin sections were examined in the ÉVM-100L and Hitachi-600H electron microscopes.

EXPERIMENTAL RESULTS

Irradiation of the duodenum with white light caused no marked changes in the mucosa, and only congestion of the microvessels with blood was observed. Histological changes after laser irradiation for 1 min were expressed as congestion of the microvessels of the stroma of the villi and crypts and an increase in volume of the goblet cells as a result of distension with secretion. Irradiation for 3 min and especially for 5 min caused marked dilatation of the stromal microvessels in the villi and crypts, especially the lymphatic capillaries of the villi.

The electron-microscopic studies showed that irradiation of the duodenum by helium-neon laser for 1 min caused definite changes in ultrastructure of the stroma of the villi and epithelial cells. In the stroma there was an increase in size of the intercellular spaces, translucency of the peripheral regions of cytoplasm of the eosinophils, with the formation of pinocytotic vacuoles in them and outflow of secretory granules. In the mast cells, located mainly around the microvessels, degranulation was observed. The basement membrane of the epithelium was rather loose in structure.

Irradiation of the duodenal wall for 3 min by helium-neon laser caused an increase in intercellular edema of the stroma and considerable dilatation of and, in particular, lymphatic capillaries. Intensified migration of eosinophils and mast cells into the epithelial lining was observed. Widening of the intercellular spaces between the enterocytes was noted. The translucent basal parts of the enterocytes contained finely granular substance of average electron density and few organelles (Fig. 1a). The basement membrane was loose in structure and its integrity disturbed in some places. Enlargement of the vesicles and outgrowths from the apical (translucent) surface was observed in the capillary endothelium.

Irradiation of the duodenum from the side of the serous membrane by helium-neon laser for 3 min caused no visible ultrastructural changes in the apical parts of the enterocytes. The microvilli and organelles had their usual structure. No changes likewise were found in the ultrastructure of the goblet cells.

After irradiation of the duodenum for 5 min marked intercellular edema of the stroma was present in the villi and crypts, as well as signs of stasis in the blood and lymphatic capillaries. Substantial ultrastructural changes were observed in the basal parts of the enterocytes. The overwhelming majority of them had translucent areas of cytoplasm in this zone, resembling intracellular edema. Outgrowths of the basal part penetrated into the basement membrane and lay in the stroma of the villi, pinched off from the cells (Fig. 1b). Irradiation for 5 min caused changes also in the apical parts of the enterocytes. Vesicles appeared there in quite large numbers, and the intercellular spaces were widened. In the cell nuclei of both crypts and villi many nuclear pores could be observed (Fig. 1c, d).

The autoradiographic study showed that irradiation of the duodenal wall by helium-neon laser causes a significant increase in [^3H]thymidine incorporation into the epitheliocytes of the crypts. In intact rats ILN for the duodenal crypts was $29.5 \pm 1.0\%$. Irradiation of the duodenal wall for 5 min by white light caused an increase in [^3H]thymidine incorporation into enterocytes of the crypts (ILN was $35.9 \pm 0.9\%$). Irradiation of the duodenal wall by a helium-neon laser for 1 min caused an increase in ILN of the crypts to $40.6 \pm 1.0\%$. Irradiation by laser for 3 min did not cause an increase in ILN of the crypts ($40.4 \pm 0.7\%$). Irradiation for 5 min led to some decrease in [^3H]thymidine incorporation into the enterocytes: ILN was $38.5 \pm 0.7\%$ (Fig. 2).

Irradiation of the duodenal wall for 1-5 min by a helium-neon laser thus causes ultra-structural changes in the enterocytes and connective tissue cells which increase in severity with an increase in the total power of laser radiation, and it also causes an increase in proliferative activity of the crypt cells.

LITERATURE CITED

1. I. M. Baibekov and É. Musaev, Byull. Éksp. Biol. Med., No. 10, 501 (1981).
2. V. N. Galankin, A. A. Vishnevskii, A. I. Golovnya, et al., Arkh. Patol., No. 5, 45 (1979).
3. N. F. Gamaleya, Lasers in Experimental and Clinical Medicine [in Russian], Moscow (1972).
4. K. A. Zufarov, I. M. Baibekov, and A. A. Khodzhimetov, Compensatory and Adaptive Processes in the Intestine [in Russian], Moscow (1974).
5. Yu. G. Parkhomenko, O. K. Skobelkin, and E. I. Brekhov, Arkh. Patol., No. 3, 30 (1979).
6. S. D. Pletnev (editor), Lasers in Clinical Medicine [in Russian], Moscow (1981).
7. T. B. Timashkevich, Ways and Mechanisms of Regeneration of the Digestive Tract in Vertebrates [in Russian], Moscow (1978).

PRESERVATION OF THE ISOLATED KIDNEY UNDER NORMO-THERMIC CONDITIONS BY PERFUSION WITH PERFLUORO-TRIBUTYLAMINE EMULSION

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Two methods are used to preserve kidneys before transplantation: immersion, using a saline intracellular solution, and perfusion, with an extracellular solution. Preservation of the kidneys in these solutions is carried out at low temperature ($2-7^\circ\text{C}$). In recent years reports have been published of perfusion preservation of kidneys under normothermic and hypothermic conditions both experimentally and clinically, with the use of perfluorocarbon emulsions [2, 3].

The object of this investigation was to study the state of the isolated kidneys of dogs during normothermic perfusion with perfluorotributylamine (PFTBA) emulsion, developed at the Central Research Institute of Hematology and Blood Transfusion, Ministry of Health of the USSR [1].

EXPERIMENTAL METHOD

Experiments were carried out on nine mongrel dogs weighing 10-15 kg. Under hexobarbital anesthesia both kidneys were removed from the animals, washed free from blood, and connected

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