

CHANGES IN CELL DIVISION AND DNA SYNTHESIS
IN THE FUNDAL GLANDS OF THE RAT STOMACH
PRODUCED BY RADIUM EMANATION

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Diurnal changes in the mitotic index in the fundal glands of the rat stomach reach a high level between 7 A.M. and 4 P.M. and at 10 P.M. The diurnal rhythm of the index of labeled nuclei shows maxima at 10 A.M., 1 P.M. and 10 P.M. During oral administration of radon waters of different concentrations the α -radiation affects the diurnal changes in mitotic index and index of labeled nuclei in the fundal glands of the rat stomach. The degree of the change in proliferation and in the level of DNA synthesis depends on the radon concentration in the water.

The pattern of mitosis in the gastric mucosa of rats in various functional states has been adequately studied [3].

This paper describes an investigation of the relationship between the diurnal rhythm of mitosis and DNA synthesis in the epithelium of the fundal glands of the rat stomach and also of the character of the effect of α -radiation on changes in the level of DNA synthesis and the mitotic index in the gastric mucosa of rats receiving radon waters by mouth.

EXPERIMENTAL METHOD

Experiments to determine diurnal changes in DNA synthesis and in the mitotic index were carried out on 48 noninbred male albino rats with a mean weight of 150-170 g. The animals were kept in artificial illumination from 8 A.M. to 4 P.M. They received a natural diet ad lib. at 10-11 A.M. The rats were decapitated during the 24-h period in groups every 3 h. All the animals received an intraperitoneal injection of thymidine- H^3 1 h before sacrifice in a dose of $0.3 \mu\text{Ci/g}$ body weight. The stomach was fixed, filled with Carnoy's mixture, and embedded in paraffin wax; sections 4μ in thickness were coated with type R liquid emulsion. The exposure lasted 20 days. After development the preparations were stained by the Dominici-Kedrovskii method. Mitoses and labeled cells were counted in the fundal region of the stomach in 100 longitudinally divided stomachs, in samplings of 10,730 cells. Cells were regarded as labeled if there were at least 4-5 grains of silver above the nucleus. The mitotic index (MI) was expressed in promille, and the index of labeled nuclei (ILN) in percent. The numerical results were subjected to statistical analysis by the Fisher-Student method.

Experiments to determine changes in the level of DNA synthesis and proliferative activity during administration of radon waters by mouth were carried out on 140 noninbred male rats weighing 200-230 g. Chloride-free and radon waters were introduced into the stomach through a gastric tube daily at 11 A.M. for 21 days. The animals were divided into 4 groups: the animals of group 1 (the control) received chloride-free water, while the rest received radon in doses of $0.01 \mu\text{Ci}$ (group 2), $2.31 \mu\text{Ci}$ (group 3), and $23.1 \mu\text{Ci}$ (group 4) per course. An intraperitoneal injection of thymidine- H^3 was given to the animals 1 h

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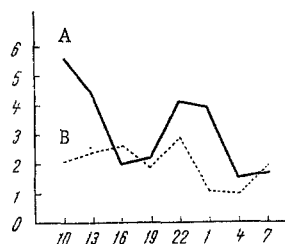


Fig. 1

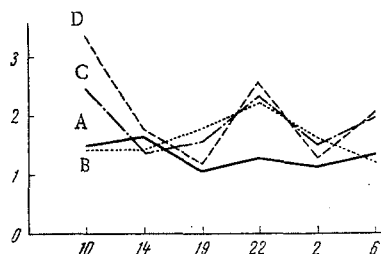


Fig. 2

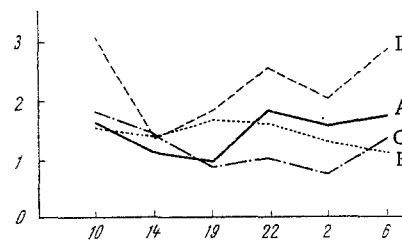


Fig. 3

Fig. 1. Diurnal changes in index of labeled nuclei (A) and mitotic index (B) in cells of fundal glands of the stomach of intact rats. Here and in Figs. 2 and 3: abscissa, time of day; ordinate, MI (in ‰) and ILN (in percent).

Fig. 2. Diurnal changes in index of labeled nuclei in cells of fundal glands of the rat stomach during administration of radon waters by mouth. Ordinate, ILN (in %); A) control (chloride-free water); B, C, D) radon water (0.01, 2.31, and 23.1 μCi per course).

Fig. 3. Diurnal changes in mitotic index in cells of fundal glands of rat stomach during administration of radon waters by mouth. Ordinate, MI (in ‰).

before sacrifice in a dose of 0.3 $\mu\text{Ci/g}$ body weight. The rats were sacrificed in groups every 4 h during the 24-h period. The numerical results were analyzed as indicated above.

EXPERIMENTAL RESULTS

The results of the experiments on intact animals are shown in Fig. 1. The level of DNA synthesis in the cells of the fundal glands of the stomach showed significant changes during the 24-h period. The curve of these changes was bimodal in character with significant maxima at 10 A.M., 1 P.M. and 10 P.M. - 1 A.M. The mean daily ILN was 3.2%. The curve of the changes in the number of mitoses during the 24-h period was much less demonstrative in character, although it was approximately bimodal. The number of mitoses reached a maximum at 10 P.M., with a significant decrease in mitotic activity toward 1 A.M. A second significant increase in MI was observed between 7 A.M. and 4 P.M. The mean diurnal MI was 2 ‰. The ratio between ILN and MI was 16. According to the literature [3] curves of diurnal changes in the number of mitoses in cells of the fundal glands of the rat stomach may be either unimodal, with a maximum between 2 and 6 A.M., or bimodal with maxima between 7 A.M. and 1 P.M. and between 7 and 10 P.M.

Two maxima of the dividing cells also were obtained in the present investigation, at about the same times. The mean MI for the 24-h period also was similar. The interval between the maximum of DNA synthesis and the peak of mitoses in these experiments was 12 h, while in mice [1, 2] it was 6 h. These differences can probably be attributed to differences in the conditions of illumination under which the animals were kept.

The results of the experiments in which radon waters were given are shown in Figs. 2 and 3.

In the control animals of group 1, which received chloride-free water, there were few synthesizing cells and the amplitude of the fluctuations in the diurnal changes of ILN was low. The differences were significant between the ILN levels at 2 and 6 P.M. ($P=0.02$). MI for the control animals was highest between 10 P.M. and 10 A.M. The number of mitoses reached a minimum between 10 A.M. and 6 P.M.

Changes in the level of DNA synthesis in the experimental groups during administration of radon waters compared with the control were largely similar in direction. In all animals of these groups there was a significant increase in the number of cells synthesizing DNA at 10 P.M., while for the animals of groups 3 and 4, receiving radon water with the highest concentration of radon, this increase was observed at 10 A.M. The higher the radon concentration, the greater the increase in the labeling index. In the animals of group 2, which received radon water with the lowest radon concentration, no increase in the level of DNA synthesis was observed at 10 A.M. In all the experimental groups the diurnal rhythm of changes in ILN continued in the animals.

TABLE 1. Mean Diurnal Changes in ILN and MI in Gastric Mucosa of Rats Receiving Radon Waters Internally

Type of water	ILN	MI	ILN/MI
Chloride-free water	1.34	1.48	9.0
Radon water 0.01 μ Ci	1.68	1.48	11.3
Radon water 2.31 μ Ci	1.88	1.22	15.4
Radon water 23.1 μ Ci	2.03	2.30	9.0

Changes in mitotic activity under the influence of α -radiation of the radon waters were less marked. In the animals of group 4, receiving radon water in high concentration, the diurnal rhythm of MI persisted and there was a significant increase in the number of dividing cells by comparison with the control at 10 A.M., 6 P.M., 6 A.M., and 10 P.M. ($P < 0.001$, 0.002 , < 0.001 , and 0.05 respectively), with a tendency for cell division to increase in intensity at the other times of day. In the animals of group 2, receiving radon water with the lowest radon concentration, the diurnal changes in MI were slight, but compared with the control group an increase in the number of dividing cells also was observed at 6 P.M. ($P = 0.02$).

Somewhat different indices were obtained for the animals of group 3 receiving radon water with an average concentration of radon (2.31 μ Ci per course). In these animals diurnal changes in mitotic activity were observed with the number of mitoses reaching a maximum at 10 A.M. compared with the control animals; they showed a significant decrease in MI at 10 P.M. and at 2 and 6 A.M., i.e., at times of high values of the mitotic index in the control animals.

Comparison of the mean diurnal changes in ILN and MI (Table 1) shows that radon water with an activity of 23.1 μ Ci per course led to an increase in ILN and MI which was close to significant ($P = 0.06$ and 0.02 respectively), while radon water with an activity of 2.3 μ Ci per course increased the level of DNA synthesis ($P = 0.02$). The mean diurnal ratio between ILN and MI varied among the different groups from 9 to 15. It is difficult to assess the relationship between the ILN and MI indices in the animals of the experimental groups. It was mainly the increases in ILN that were responsible for the increases in mitotic activity taking place largely at the same times of day. The α -radiation of the radon waters can be considered to have had some effect on the synchronization of synthesis and mitotic division. As a result of the action of radon water changes took place in the level of DNA synthesis and in proliferative activity in the fundal glands of the stomach at certain times of day, largely at the times of an increase in ILN and MI. It must be emphasized that the character of the effect of these doses differed depending on the radon concentration.

LITERATURE CITED

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