

EXPERIMENTAL BIOLOGY

ACTION OF PROGESTERONE ON THE MITOTIC INDEX AND NUMBER OF DNA-SYNTHESIZING CELLS IN THE EPITHELIUM OF REPRODUCTIVE ORGANS

N. A. Roslyakova

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A single dose of progesterone causes a maximal increase in the number of DNA-synthesizing cells in the epithelium of the uterine glands 14 h after injection of the hormone, and in the epithelium of the uterine cavity and vagina after 18 h. In all three organs an increase in mitotic activity was observed 8 h after stimulation of DNA synthesis.

The mechanism of regulation of cell division is increasingly attracting the attention of investigators. The role of sex hormones in the regulation of cell division of "target" organs is being studied particularly actively [2, 5, 6]. It has now been shown that administration of progesterone without preliminary administration of estrogens causes considerable proliferation in the epithelium of the reproductive organs [4]. The effect of progesterone on processes preceding mitosis has been studied very inadequately.

Experiments [7] have shown that after a single injection of progesterone, an increase in the level of DNA synthesis in the epithelium of the endometrium and vagina begins after 7 h and reaches a plateau after 14 h.

A comparative study of DNA synthesis and mitotic activity appeared to be indicated. There are reports in the literature that the epithelia of different parts of the reproductive tract of female mice differ in their sensitivity to sex hormones [1, 3]. An attempt was therefore made to discover whether injection of the hormone causes an increase in the index of labeled nuclei and mitotic activity simultaneously in the epithelial cells of the uterine cavity, uterine glands, and vagina, or whether the various types of epithelium differ in their reactivity.

EXPERIMENTAL METHOD

Experiments were carried out on 82 C₅₇BL mice weighing 20 g. All mice were ovariectomized 12 days before the experiment began. Progesterone from the Rostov factory (batch 30.368) was used in the experiments.

A single dose (1 mg) of the hormone was injected subcutaneously into 77 mice at 9 A.M. and the animals were sacrificed 5, 7, 9, 14, 18, 22, 26, 30, 34, and 48 h later. The groups of animals sacrificed after 5, 7, and 48 h included 7 mice, and the remaining groups contained 8 mice. Five control animals (ovariectomy, without injection of hormone) were sacrificed simultaneously with the first group.

An injection of thymidine-H³ in a dose of 1 μ Ci/g body weight was given to the animals 45 min before sacrifice. Sections, 4 μ in thickness, were coated with type M radiosensitive emulsion. The exposure was two weeks.

To determine the number of mitoses and labeled nuclei, three thousand cells in the epithelium of the uterine cavity and vagina and 1000 cells in the epithelium of the uterine crypts were examined. The mitotic

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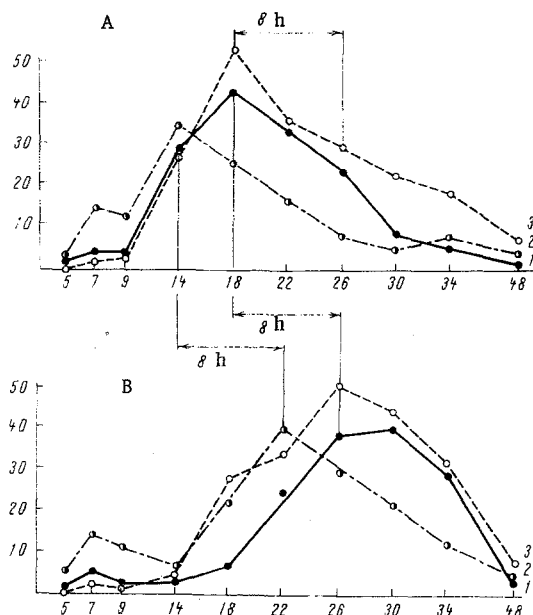


Fig. 1. Changes in index of labeled nuclei (A; in %) and mitotic index (B; %). 1) Epithelium of uterine cavity; 2) epithelium of uterine glands; 3) vaginal epithelium. Abscissa: time after single injection of progesterone (in h).

After 14 h a sharp rise in MI began, to reach a maximum at 22 h (by comparison with 14 h, $P = 0.01$). MI was close to the control level after 48 h.

In the epithelium of the uterine cavity 5 and 7 h after injection of the hormone a slight increase in ILN was observed in 3 animals. There was still no significant increase in ILN after 14 h (compared with 9 h, $P = 0.078$). The maximal increase in ILN occurred at 18 h (compared with 9 h, $P = 0.002$). A significant decline in the curve was observed after 34 h (compared with 8 h, $P = 0.008$).

In the epithelium of the uterine cavity a significant increase of MI to 38 ‰ was observed after 26 h (the highest value of MI in this group was 66.6 ‰). By 30 h, no significant change in MI had occurred. After 48 h, the decrease in MI was significant, but its mean value was still little higher than in the control. Hence, the increase in DNA synthesis and the increase in mitotic activity took place in the epithelium of the uterine glands 4 h sooner than in the epithelium of the uterine cavity.

In the vaginal epithelium the ILN curve was close to that of the epithelium of the uterine cavity (Fig. 1). After 9 h the curve began to rise and it reached its maximum at 18 h (ILN 59%). Later, although the ILN of the uterine cavity fell, ILN of the vagina still remained high. A significant decrease was observed only after 48 h.

MI for the vaginal epithelium began to increase after 14 h and reached its maximum after 26 h (compared with 14 h, $P = 0.001$). MI was still high after 48 h, in some animals being 10 times higher than in the control, and only in one animal was it close to the control value.

Hence, the increase in DNA synthesis in the epithelium of the uterine glands of ovariectomized mice was observed 14 h after a single injection of progesterone, compared with 18 h in the epithelium of the uterine cavity and vagina. An increase in MI in the epithelium of the uterine crypts was found 22 h after injection, and in the epithelium of the uterine cavity and vagina 26 h after injection of the hormone, i.e., in all three organs the increase in mitotic activity took place 8 h after the increase in DNA synthesis. Variations of ± 2 h are possible because the animals were sacrificed at 4-hour intervals.

It was also noted that 18 h after injection of the hormone, when DNA synthesis in the epithelium of the uterine cavity and uterine glands was already appreciably reduced, in the vaginal epithelium it still remained at a high level.

index (MI) in promille and the index of labeled nuclei (ILN) in percent were determined. The numerical results were analyzed statistically by the Fisher - Student method.

EXPERIMENTAL RESULTS

The mean value of ILN for the epithelium of the uterine glands in the control animals was 0.8% (0.4-1.2%). A marked increase in ILN (4.2, 5.2, 6.4%) was observed in 3 of the 7 animals 5 h after injection of the hormone. After 7 h the increase in the ILN curve was significant (compared with the control, $P = 0.003$). In this group and in the group sacrificed 9 h after injection, one animal with a low ILN value remained: 0.6 and 1.2%. By 14 h the ILN had reached its maximum, with a mean value of 34% (12-54%) (compared with ILN after 9 h, $P = 0.001$). After 26 h, the decline of the curve was statistically significant (compared with 14 h, $P = 0.01$). By 48 h, the ILN in 2 animals was higher than in the control (2.6 and 14%); in the rest it was at the control level.

The mean value of MI for the epithelium of the uterine glands in the control was 1.4 ‰. MI of one animal 5 h after injection of the hormone was 24 ‰, and in the rest between 0.1 and 4 ‰. An increase in MI, from 10 to 28 ‰, was also observed after 7 h in 6 animals. MI of the remaining 2 animals were indistinguishable from the control. The mean value of MI after 14 h was 6.7 ‰.

In all groups, especially in the first few hours after injection of the hormone, a wide scatter of the ILN and MI values was observed. The reasons for this were evidently, first, differences in the individual sensitivity of the animal to progesterone, and second, differences in the intensity of absorption and excretion of the hormone.

The ILN curve drawn from the results of these experiments differs to some extent from that given by Beato [7]. This may be because, when calculating ILN for the uterine epithelium, Beato did not determine it separately for the epithelium of the uterine cavity and the epithelium of the uterine glands, in which the increase begins sooner.

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