ONCOLOGY

Relationship between Proliferation of Ehrlich Ascitic Tumor Cells and Status of the Chalone System under Conditions of Modified Photoregimen

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The effects of permanent darkness on proliferation of Ehrlich ascitic tumor cells and status of the chalone system in the tumor were studied. Chalone-containing preparations from animals exposed to different light conditions exhibited different biological effects on cell proliferation in this tumor. A relationship between biological activity of chalone-containing preparations and sensitivity of tumor cells to these preparations under conditions of modified photoregimen was revealed.

Key Words: *chalones; photoregimen; permanent darkness; Ehrlich ascitic tumor*

Study of the temporal regularities of cell proliferation is an important modern trend of research [3,4]. Photoperiodicity is essential for proliferation of cells, including cells of Ehrlich ascitic tumor (EAT) [7,10]. Of factors involved in local (tissue) regulation of the proliferative system, chalones, tissue inhibitors of cell proliferation, play the key role [2,5,6,8,9]. However, the interactions between these levels of regulation of temporal organization of the proliferative system are little studied.

Here we studied the relationship between biological activity of chalone-containing preparations and the sensitivity of tumor cells to these preparations under conditions of modified photoregimen.

MATERIALS AND METHODS

The study was carried out on 270 outbred male mice. A group of 5-7 animals was examined per time point. The object of the study was a diploid strain of EAT. Four types of chalone-containing preparations (CCP) were used. CCP-1 was isolated at 14.00 from animals with EAT kept under standard light:darkness (L:D=12:12 h) conditions with the light period from 6.00 to 18.00. CCP-3 was collected from the same mice at 2.00. CCP-2 was obtained at 14.00 from mice with EAT exposed to permanent darkness (D:D), and CCP-4 at 2.00 from the same mice. Experimental animals were distributed into 22 groups.

Mice of groups 1-10 and 21 (L:D) were kept under L:D=12:12 h (light from 6.00 to 18.00) for 21 days before the experiment. Subsequent operations with these animals were carried out under the same conditions. On day 5 after EAT transplantation, the animals were injected at 14.00 with saline (0.5 ml/mouse; group 1), CCP-1 (10 mg/0.5 ml saline/mouse; group 2); CCP-2 (10 mg/0.5 ml saline/mouse; group 3), CCP-3 (10 mg/0.5 ml saline/mouse; group 4), or CCP-4 (10 mg/0.5 ml saline/mouse; group 5). Five to seven animals from each group were sacrificed 2

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and 4 h after the injection (at 16.00 and 18.00), EAT preparations were made, and total mitotic index (TMI) and mitosis phase indexes were analyzed.

Identical manipulations were carried out with animals of groups 6-10 at 2.00; the material was collected at 4.00 and 6.00.

Intact mice (group 21) served as the control for groups 1-10. These animals were kept under standard light conditions (light at 6.00-18.00). Group 1 mice and group 6 mice served as control 2 for groups 2-5 and 7-10, respectively.

Animals of groups 11-20 and 22 were kept at permanent darkness for 21 days before the experiment. All subsequent manipulations on these animals were carried out under the same conditions as in groups 1-10.

The results were evaluated by estimation of the TMI, by graphic parametrical method for studies of biological rhythms [7], analysis of correlations, and standard methods of mathematical processing.

RESULTS

Study of the relationship between TMI changes in EAT after injection of CCP and biological activity of CCP and tumor cell sensitivity to this preparation under conditions of permanent darkness showed that tumor cells of L:D animals are more sensitive to all 4 CCP types injected at 14.00 (when mitotic activity of the tumor decreases) and less sensitive to the preparations injected at 2.00 (when mitotic activity increases; Fig. 1).

These findings confirm previously detected [1] regularity indicating higher effect of chalones during the period of reduced number of mitoses in the tumor.

The tumors of animals exposed to permanent darkness were similarly sensitive to these CCP types injected at 14.00 and 2.00, mitotic activity reducing in response to treatment (Fig. 2). Hence, the decrease in proliferative activity of EAT cells in animals exposed to permanent darkness can be due to their higher sensitivity to chalones realized through their mitosis-inhibitory effect. These data attest to coordination of circadian rhythm phases of mitoses in EAT and tumor cell sensitivity to CCP treatment in mice exposed to standard L:D conditions and to permanent darkness.

CCP-3 obtained from L:D animals during maximum mitotic activity in the tumor less markedly modified cell proliferation in EAT than CCP-1 obtained from these animals during the period of reduced mitotic activity (Figs. 1 and 2). These data indicate synchronous changes in the biological rhythms of CCP and cell division in EAT (that is, the phases of higher biological activity of CCP and of mitotic activity decrease coincide).

The same regularity is confirmed by virtually identical effects of CCP-1, CCP-2, and CCP-4 obtained at different time from L:D and D:D animals with identical levels of mitotic activity. Hence, the rhythms of mitotic activity in the tumor and of biological activity of CCP obtained from it are synchronous.

Thus, the interaction between the photoperiodical and tissue (chalone) regulation of EAT cell proliferation was revealed: changes in photoperiodicity affect biological activity of CCP and reactivity of EAT cell to CCP.

Hence, coordination of circadian rhythms of mitoses in EAT, biological activities of CCP obtained from

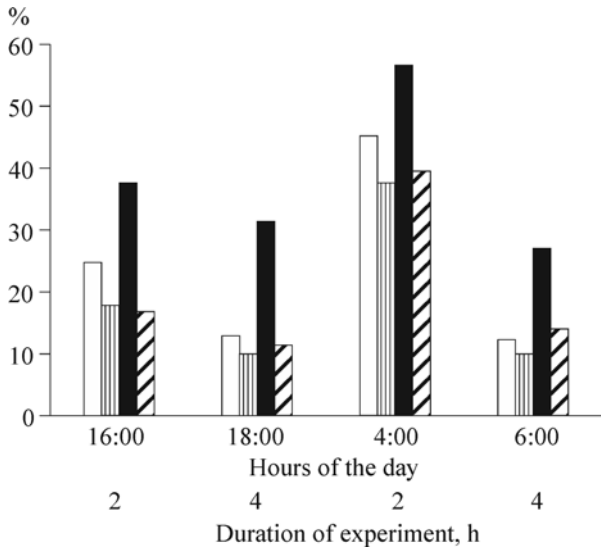

Fig. 1. Changes in TMI in EAT cells of L:D animals after injection of different CCP (in comparison with respective changes in L:D animals injected with saline). Here and in Fig. 2: light bars: CCP-1; vertically hatched bars: CCP-3; dark bars: CCP-2; obliquely hatched bars: CCP-4.

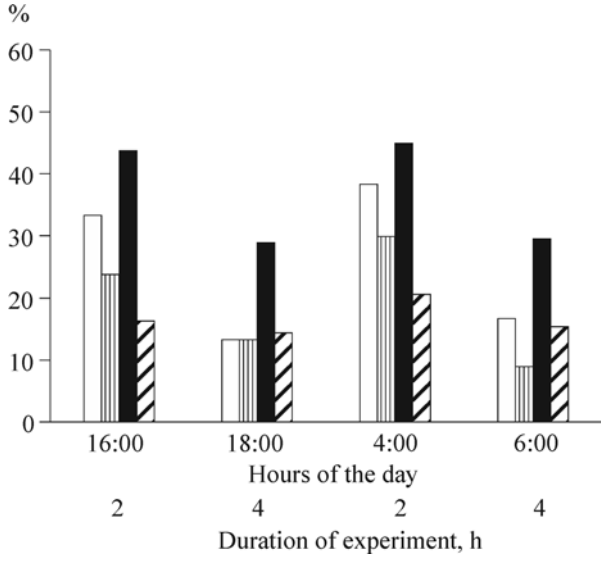

Fig. 2. Changes in TMI in EAT cells of animals exposed to permanent darkness after injection of various CCP in comparison with the respective changes in similar animals injected with saline.

the tumor, and tumor cell sensitivity to these preparations was revealed in animals exposed to standard photoregimen and to permanent darkness.

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