FUNCTIONAL MORPHOLOGY OF THE ADRENAL CORTEX DURING PHYSIOLOGICAL RHYTHM OF SEX STEROID SYNTHESIS

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To determine the degree to which the various zones of the adrenal cortex participate in formation of the physiological rhythm of steroid synthesis, the method of quantitative histoenzymologic analysis was used. Activity of 3β-OH-steroid dehydrogenase, glucose-6-phosphate dehydrogenase, NAD- and NADP-diaphorases, acid and alkaline phosphatases, and nonspecific esterase was investigated. The results of the histoenzymologic investigation were compared with those of biochemical analysis of corticosteroids in the peripheral blood. The results showed that under physiological conditions steroid formation is stimulated by mobilization of individual groups of cells and not of the whole adrenal parenchyma. The physiological rhythm of steroid synthesis is maintained by the integrated function of all the zones; however, structural heterogeneity for the duration and quantitative expression of secretory activity is observed in the adrenal cortex.

KEY WORDS: histoenzymologic analysis; adrenal cortex; sex cycle.

Until recently biochemical analysis has played the leading role in the study of the adrenal cortex. However, biochemical investigations give an idea only of cortical secretion as a whole and do not show the morphological structures responsible for it. Histoenzymic analysis is a method that can reveal correlation between secretory activity and particular morphological substrates. Its use to study the adrenal cortex is particularly promising, for the biosynthesis of steroids has been adequately studied and methods worked out for detecting enzymes responsible for various stages of steroid synthesis. The subjective element introduced by visual assessment of histoenzymic tests can be overcome by the use of microspectrophotometry [1, 2].

The object of this investigation was to determine the degree to which various morphological structures of the adrenal cortex participate in maintenance of the physiological rhythms of steroid synthesis. The sex cycle of animals, during which fluctuation of the corticosteroid level both in the adrenal tissue and in the peripheral blood has been established biochemically [3, 6, 8], was used as the model. Quantitative histoenzymic analysis was used to estimate the functional state of the cortical cells.

EXPERIMENTAL METHOD

Experiments were carried out on 20 female albino rats in different phases of the estrus cycle (5 animals in each phase). The activity of the following enzymes was determined in 10-μ cryostat sections: glucose-6-phosphate dehydrogenase (G6PD), 3β-OH-steroid dehydrogenase (3β-OH-SD), NAD- and NADP-diaphorases, acid and alkaline phosphatases, and nonspecific esterase. For quantitative assessment of the intensity of the histoenzymic tests the MUF-5 microspectrophotometer was used, with scanning in visible monochromatic light. The optical density of the reaction products was measured in seven parts of the cortex (Fig. 1). The concentration of 11-hydroxycorticosteroids (11-HCS) was determined simultaneously in the peripheral blood by gel filtration, enabling both the secretion of free corticosteroids and the degree of

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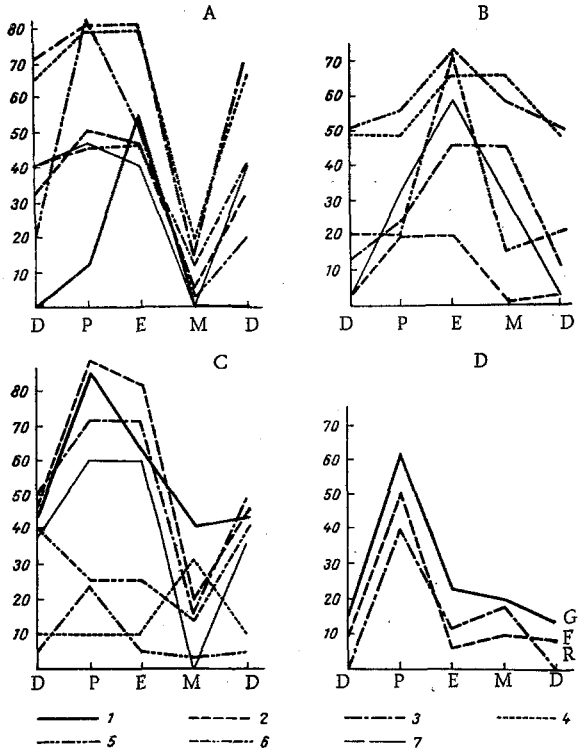

Fig. 1. Dynamics of intensity of histoenzymic reactions in adrenal cortex during estrus cycle. A) Glucose-6-phosphate dehydrogenase; B) 3β-OH-steroid dehydrogenase; C) NAD-diaphorase; D) acid phosphatase. In A, B, C: 1) outer zona glomerulosa; 2) inner zona glomerulosa; 3) outer zone fasciculata; 4) middle z. fasciculata; 5) inner z. fasciculata; 6) outer z. reticularis; 7) inner z. reticularis. In D: G) zona glomerulosa; F) z. fasciculata; R) z. reticularis. Abscissa, phases of cycle: D) diestrus; P) proestrus; E) estrus; M) metestrus; ordinate, percentage of cells with above -average density of reaction products.

their binding with the plasma proteins to be studied [4, 5].

EXPERIMENTAL RESULTS

Biochemical analysis revealed cyclic changes in steroid concentration with a maximum in the phase of proestrus (32.7 ± 2.4) and a minimum in estrus–metestrus (25.7 ± 0.9), in agreement with the findings of other workers [6, 7, 8]. Microphotometric investigations revealed considerable fluctuations (by 1.5-2 times) in the intensity of the reactions in different cells of the same part of the cortex. The conclusion that enzyme activity was changed was based on a significant (with a probability of not less than 99%) increase in the number of cells with an above-average intensity of the reaction. All the enzymes tested were detected in all zones of the cortex. In the zona glomerulosa maximal activity was attained by NAD-diaphorase and the hydrolytic enzymes, whereas activity of 3β-OH-SD, G6PD, and NADP-diaphorase was minimal in that zone. In the zone fasciculata and zona reticularis high dehydrogenase activity and low phosphatase activity were observed. Depending on the functional state of the adrenal cortex as a whole, relations between the intensities of the histoenzymic reactions varied in different zones. For instance, with minimal synthesis of steroids (estrus), 3β-OH-SD activity was most marked in the inner third of the zona fasciculata and the outer part of the zona reticularis, but during maximal steroid-synthesizing function of the cortex (proestrus) high activity of the enzyme was shifted into the middle and inner part of the zone fasciculata, and it was sharply reduced in the zone reticularis.

The dynamics of the intensity of the histoenzymic reactions during the estrus cycle (Fig. 1) correlated with the fluctuations in the blood 11-HCS level. Cyclic changes in intensity of the enzymic reactions were observed in all zones and were manifested as an increase in activity of the enzymes in proestrus–estrus and a decrease in metestrus–diestrus. Meanwhile, in different zones activation of the enzymic reactions were not synchronized and differences in their quantitative assessment were observed.

Quantitative analysis of the activity of a group of intracellular enzymes of the adrenal cortex thus reflects the level of synthesis of corticosteroids. The physiological rhythm of steroid synthesis in the adrenal cortex is maintained by the integrated function of all zones; however, in the duration and quantitative expression of secretory activity, marked structural heterogeneity is found in the cortex. Under physiological conditions stimulation of steroid synthesis is brought about by mobilization of only individual groups of cells and not of the adrenal parenchyma as a whole.

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