ACTION OF ESTRADIOL DIPROPIONATE ON PROTEIN SYNTHESIS IN A CELL-FREE SYSTEM FROM THE LIVER OF OVARIECTOMIZED RATS

A. I. Maiskii, N. N. Vedernikova, and V. A. Volov

UDC 612.351.015.36:547.96],014.46:615.256.51]-06:612.616.1

Intraperitoneal injection of estradiol dipropionate (10 and 25 μg/100 g body weight) increases the incorporation of glycine-C^{14} and phenylalanine-H^3 into liver proteins. The maximum of stimulation is observed after 15 h. Estrogens exert their action on the protein-synthesizing system of the liver through activation of the microsomal and pH-5 fractions.

Estrogens have a marked stimulant action on the synthesis of nucleic acids and proteins in target organs [1, 3, 5]. At the same time evidence has been obtained of the anabolic effect of estrogens in the liver also [2, 4], although no specific receptors have yet been described in it. The dynamics of estrogen accumulation in the uterus and liver suggests that estrogens stimulate protein synthesis in these organs by different mechanisms.

An investigation was therefore carried out to study the mechanism of action of estradiol on protein synthesis in a cell-free system from the liver microsomes of ovariectomized rats.

EXPERIMENTAL METHOD

Altogether 158 noninbred female albino rats were used. The sexually mature animals weighing 150-180 g were ovariectomized under superficial ether anesthesia and used in the experiments not earlier than 3 weeks after the operation. Estradiol dipropionate was injected intraperitoneally as an oily solution in doses of 10 and 25 μg/100 g body weight. The animals were killed 12, 15, 18, 24, and 36 h after injection of the hormone. The control rats received olive oil. The cell-free protein-synthesizing system of the liver was constituted in the usual way [8]. To detect the role of the microsomal component and the pH-5 fraction in the mechanism of the effect of the estrogen on protein synthesis, Croft experiments were carried out in which microsomes isolated from the liver of the experimental animals were incubated with the pH-5 fraction from the liver of the control animals, and vice versa.

EXPERIMENTAL RESULTS AND DISCUSSION

Estradiol dipropionate in both doses stimulated incorporation on glycine-C^{14} and phenylalanine-H^3 into protein by the cell-free system of the rat liver. The effect of the hormone began to appear 12 h after its administration. The maximal increase in protein biosynthesis was observed after 15 h, when its level stood at 81.1 ± 7.8 and 62.4 ± 2.8% of the control for glycine-C^{14} and phenylalanine-H^3, respectively (dose of the hormone 25 μg/100 g). After 18 h the activity had fallen a little but it still remained high until 24 h (Table 1). No effect of the hormone was seen 36 h after its injection. Estradiol in a dose of 25 μg/100 g body weight had a stronger stimulant action on the incorporation of glycine-C^{14} than of phenylalanine-H^3, suggesting specificity of the composition of the protein synthesized.

Department of Molecular Pharmacology and Radiobiology, Medico-Biological Faculty, N. I. Pirogov Second Moscow Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR N. N. Zhukov-Verezhinkov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 77, No. 6, pp. 95-97, June, 1974. Original article submitted June 25, 1973.

TABLE 1. Effect of Estradiol Dipropionate on Incorporation of Glycine-C^{14} and Phenylalanine-H^3 (in % of control) in a Cell-Free Protein-Synthesizing System from the Liver of Ovariectomized Rats (M ± m)

Incubated sample	Estradiol in dose of 25 μg/100 g body weight				Estradiol in dose of 10 μg/100 g body weight			
	time after injection of hormone (in h)							
	12	15	18	24	12	15	18	24
Control microsomes	100,0±9,7	100,0±14,1	100,0±10,1	100,0±12,5	100,0±4,8	100,0±12,1	100,0±9,2	100,0±6,4
+ Control pH-5 fraction	100,0±3,0	100,0±5,3	100,0±9,1	100,0±19,0	100,0±16,8	100,0±14,3	100,0±5,3	100,0±5,9
Control microsomes	152,0±5,4*	141,5±5,6*	118,8±18,4	114,6±15,4	103,5±5,8	82,5±7,9	138,6±6,7	118,2±1,9
+ Experimental pH-5 fraction	121,8±13,9	103,3±8,4	115,1±11,3	117,7±12,9	100,0±11,3	135,7±10,1	112,3±11,3	100,1±2,4
Experimental microsomes	126,8±9,5*	122,6±3,9	139,5±10,1	119,4±8,7	106,0±9,2	126,1±11,2	123,2±11,4	115,1±6,1
+ Control pH-5 fraction	117,5±17,2	148,2±7,3*	119,2±15,3	129,3±15,9	116,8±28,1	124,1±11,0	119,4±7,8	104,2±2,7
Experimental microsomes	166,7±4,7*	181,1±7,8*	150,7±5,3*	157,7±3,5	116,1±20,0	142,9±5,1*	136,5±8,5*	136,0±6,9
+ Experimental pH-5 fraction	141,6±5,3*	162,4±2,8*	131,5±3,2*	130,7±5,9*	133,7±8,6*	140,2±15,4*	130,5±8,7*	106,3±4,9

Note. 1) Top of figures in each group represents incorporation of glycine-C^{14}, bottom line represents phenylalanine-H^3; asterisk denotes that results differ by a statistically significant degree from the control $P < 0.05$).

An important role in the mechanism of action of estradiol on protein synthesis in the rat liver was played by its effect on the activity of the microsomal and pH-5 fractions. After incubation of the control pH-5 fraction with the experimental microsomes, for instance, the degree of stimulation was 26.8 ± 9.5 and 22.6 ± 3.9%, whereas the use of a system with control microsomes and the experimental pH-5 fraction increased the level of stimulation to 52.0 ± 5.4 and 41.5 ± 5.6% (12 and 15 h respectively after administration of 25 μ g/100 g estradiol). These results apply to the incorporation of glycine-C^{14}. The results with phenylalanine-H^3 were less clear.

However, it must be emphasized that the hormone exhibited its maximal stimulant effect only if both experimental fractions were incubated together.

The increased incorporation of labeled amino acids into liver protein in vitro, depending on the pH-5 fraction, in the writers' opinion can be associated with the increased synthesis of aminoacyl-tRNA-synthetase or of enzymes methylating tRNA. This hypothesis agrees with the increase in methylase activity in the rat liver, the change in the relative quantities of the iso-forms of the aminoacyl-tRNA complex, and the increased incorporation of the individual amino acids into that complex [6, 9].

Meanwhile the action of estrogens on protein synthesis in the uterus takes place through activation of the microsomal fraction but not of the supernatant fraction, and the effect of the estrogens is more marked than in the liver [7, 10, 11]. This difference may arise because the endometrial cells contain specific receptor molecules responsible for the greater accumulation and the longer intracellular retention of the hormone, and also because these cells have no systems capable of metabolizing estradiol. The role of estrogen-dependent pyridine-nucleotide transdehydrogenases must also be taken into account: activation of these enzymes in the cells of the uterus recruits reduced NADP for the intensification of biosynthesis [12]. The results of these experiments show that estrogens can increase the rate of protein biosynthesis in the liver cells despite the organ specificity of the cellular mechanism; unlike in the uterus, activation of the protein-synthesizing apparatus in the liver also involved the action of the hormone on the pH-5 fraction.

LITERATURE CITED

1. L. M. Gol'ber and R. S. Tishenina, Dokl. Akad. Nauk SSSR, 190, No. 4, 1001 (1970).
2. N. P. Saksonov, P. V. Sergeev, R. D. Seifulla, et al., Farmakol. i Toksikol., No. 5, 572 (1971).
3. P. V. Sergeev, R. D. Seifulla, and A. I. Maiskii, Molecular Aspects of the Action of Steroid Hormones [in Russian], Moscow (1971), p. 149.
4. P. V. Sergeev, R. D. Seifulla, and N. P. Saksonov, Byull. Éksperim. Biol. i Med., No. 7, 36 (1971).
5. R. S. Tishenina, Probl. Éndokrinol., No. 6, 89 (1967).
6. G. Beck, D. Hentren, and J. P. Ebel, Biochim. Biophys. Acta, 213, 55 (1970).
7. D. L. Greenman and F. T. Kenney, Arch. Biochem., 107, 1 (1964).
8. R. Kato, W. R. Jondorf, L. A. Loeb, et al., Molec. Farmacol., 2, 171 (1966).
9. O. K. Sharma and E. Borek, Biochemistry (Washington), 9, 2507 (1970).
10. C. S. Teng and T. V. Hamilton, Biochem. J., 105, 1101 (1967).
11. C. S. Teng and T. V. Hamilton, Biochem. J., 118, 341 (1970).
12. C. A. Villee, in: J. M. Allen and H. A. McGraw (editors), The Molecular Control of Cellular Activity, New York (1962), p. 297.