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Comparative Analysis of Progesterone and Estradiol Reception in Human Myoma

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Cytosolic and plasma membrane receptors for progesterone and estradiol are studied in myomatous nodes (MN) and in histologically unaltered myometrium (HUM) against the background of myoma. The level of cytosolic receptors for both hormones is higher in the myoma cells than in the essentially healthy myometrium. In the plasma membranes the progesterone reception is reduced and the estradiol reception is unchanged compared with HUM.

Key Words: *progesterone receptors; estradiol receptors; plasma membrane; myoma*

The role of steroid hormones in the formation and development of certain tumors has been studied for a long time. The presence of steroid receptors, their number, and the tissue ratio are fundamental for the diagnosis and choice of therapy in a number of oncological diseases. At the same time, the presence of cytoplasmic and nuclear receptors for sex steroids and their characteristics are by no means always indicative of the hormonal dependence of a pathological process [2,7,11]. New data are being published on the structure of receptors of the cells that are targets for steroid hormones; specific binding sites for steroids have been found on the plasma membranes (PM) [5,6], mitochondria [4], and lysosomes [10]. We have shown that the PM of human endometrial and myometrial cells has specific binding sites for 17β-estradiol; these binding sites have a high affinity for the ligand and change their characteristics according to the stage of tumor growth [3]. Since the relative plasma content of estradiol and progesterone, rather than the absolute concentration of estrogens [1,8],

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TABLE 1. Specific Binding of Estradiol and Progesterone to PM of Myometrial Cells ($M \pm m$)

Type of tissue	Estradiol		Progesterone		B estradiol/ B progesterone
	K_{ass}, nM^{-1}	B_{max}, fmol/mg protein	K_{ass}, nM^{-1}	B_{max}, fmol/mg protein	
HUM	0.20±0.06	38.0±9.0	0.39±0.08	57.0±6.1	0.667
Myomatous node	0.69±0.23	33.0±10.2	0.76±0.06*	19.6±4.5*	1.684

Note. Here and in Table 2 an asterisk indicates values statistically different from the parameters for HUM at $p<0.05$.

is the crucial parameter in predicting the development of estrogen-dependent hyperplasia, a parallel study of the reception of progesterone and estradiol in the uterus in health and pathology is relevant.

In this study we compared the specific binding of 17β-estradiol and progesterone to the PM and cytosol receptors in HUM against the background of myoma and in MN of the human uterus.

MATERIALS AND METHODS

The myometrium (the tissue of the myomatous node and histologically unaltered myometrium) from 18 patients (aged 41-63 years) who had undergone surgery for myoma of the uterus was provided by our colleagues from the Facultyof Obstetrics and Gynecology (Pediatric Department, Russian State Medical University). The estradiol and progesterone receptors in the cytosol fraction were studied as described elsewhere [1]. Plasma membranes were isolated as described [8]. Tritiated progesterone (Russia) and estradiol (Amersham) were used in the binding experiments. The plasma membrane suspension was incubated with [^{3}H]progesterone at 4°C for 45 min with a 100-fold excess of hydrocortisone; incubation with [^{3}H]estradiol was perfomed at 25°C for 60 min. The incubation mixtures were then passed through GF/F filters (Whatman), and the filters were counted in an Intertechnique SL-38 scintillation counter. For the evaluation of the specific binding of estradiol and progesterone PM were incubated with a 200-fold excess of unlabeled 17β-estradiol (Merck) or with unlabeled progesterone (Russia), respectively. The results were analyzed in Scatchard plots [12]. Protein was measured by the method of Lowry [9]. The data were analyzed with the use of Student's t test.

RESULTS

Analysis of the number of binding sites for progesterone in HUM and in MN of patients with myoma of the uterus revealed that PM of MN contain a smaller number of progesterone receptors compared with PM of HUM (Table 1). However, the number of cytosol receptors for progesterone in myoma is higher than in HUM (Table 2). This finding may indicate a more active translocation of the hormone-receptor complex in the cells of an MN.

The number of binding sites for estradiol on the PM of myoma cells practically did not differ from this parameter in HUM (Table 1), while the content of cytosol receptors for estradiol was higher in the MN cells (Table 2).

The association constant (K_{ass}) for both hormones was higher in MN cells than in unaltered tissue (Table 1). This indicates that myomatous tissue differs from HUM tissue in a high level of cytosol receptors for sex steroids. At the same time, it should be noted that the reception of steroids in the PM of a myoma cell differs in having a reduced binding capacity for progesterone with no changes in estradiol binding. This is clearly seen from the ratio of B_{max} for estradiol to B_{max} for progesterone. This ratio is 2.5-fold higher in PM of myoma cells than in unaltered tissue.

The term cytosol receptors refers to the unstably bound (predominantly with the nuclear fraction) steroid receptors which are readily washed out by the soluble fraction during preparative procedures [4]. Consequently, the changes in the content of cytosol and membrane receptors for sex steroids can be interpreted as a disruption of the nucleus-cytoplasm relationships determining cell division and differentiation. An increase in the content of progesterone and estradiol receptors may

TABLE 2. Content of Cytosol Progesterone and Estradiol Receptors in Human Myometrium ($M \pm m$)

Type of tissue	Estradiol receptors, fmol/mg protein	Progesterone receptors, fmol/mg protein	Estradiol receptors/ progesterone receptors
HUM	37.4±6.4	64.0±11.6	0.584
Myomatous mode	109.6±7.8*	149.6±17.2*	0.733

result from a more active cell metabolism in the myometrium. "Interference" in the plasma membrane reception of sex steroids during myoma growth characterizes the abnormal sensitivity of tumor cells to regulatory agents.

We have found that the recognition of tropic hormones at the PM level is impaired during tumor growth. Changes in the membrane reception of estrogens and progestins are the most informative criterion of the development of hyperplastic processes in the myometrium. A decrease in the exposure of the progesterone receptors on the cell surface causes a diminution of tissue sensitivity to gestagens.

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Experimental Validation for a Clinical Study of GABA-Positive Substances in Threatened Abortion

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The barouterography method revealed that intravenous phenibut in a dose of 50 mg/kg and phenazepam in a dose of 2.5 mg/kg have a suppressive effect on the contractile isometric (fetus-expelling) activity of the uterus in nonpregnant and pregnant rabbits. Phenibut (150 mg/kg) and phenazepam (3 mg/kg) do not have an adverse effect on fetal development in rats. Clinical trials of phenibut and phenazepam as gravidoprotectors in threatened abortions are recommended.

Key Words: *tocolytics; phenibut; phenazepam*

Spontaneous abortions have a negative impact on female reproductive function and on the health of the progeny, being one of the principal causes of childhood morbidity and mortality [5,8]. There is thus a pressing need to develop effective and safe pregnancy protectors as means of etiopathogenetic therapy.

We searched for potential tocolytics among the agents activating the GABAergic system. Published data on γ-aminobutyric acid (GABA) as the prin-

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