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Hif-1 α and IGF Expression in Endometrial Hyperplasia

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Abstract

The objective: to assess the effect of insulin resistance and hypoxia on the risk of endometrial hyperplasia development. The research was conducted on the base of Municipal Maternity House №5 (Odessa, Ukraine). The data obtained from the sample of 417 reproductive, perimenopausal, and postmenopausal age women who referred for medical care to the gynecological department, were analyzed. All the women under examination were carried out medical and diagnostic scraping of the uterine cavity. **Results.** The intensity of hypoxia increases with age and closely correlates ($r=0,78$) with the level of insulin resistance. IGF demonstrates reverse dynamics – its rates has been diminished with age ($r=-0,63$). **Conclusion.** It was demonstrated that insulin resistance and hypoxia play significant role in the pathogenesis of endometrial hyperplasia. Indices of IGF and HIF could be recommended as valuable biomarkers of prognosis in the case of endometrial hyperplasia.

Key words: endometrial hyperplasia; IGF; Hif-1 α ; malignancy

Introduction

Endometrial hyperplasia (EH) is a chronic progressive disease with a high prevalence among women of any age [1, 2]. The pathogenesis of the disease is multifactorial, however, the role of hormonal homeostasis in the occurrence and development of hyperplastic processes in the endometrium is undeniable. Thus, in many studies, the hypothesis of

estrogen-dependent state is preferred, but according to other researches up to 20-30% of endometrial adenocarcinomas are not associated with hormonal response [1, 3]. Endometrial hyperplasia could result in the malignization if untreated with progression to cancer [4-6]. The prevalence of EH in the general population reaches 30%, and the percentage of transformation into endometrial cancer ranges from 5 to 50% [1, 2, 7].

In the pathogenesis of hyperplasia, a significant role is played by hyperestrogenism, impaired endometrial receptivity, inadequate response of endometrial cells to external influences [3, 8]. A common characteristic of all the above processes leading to cell transformation is hypoxia [8, 9]. Intracellular hypoxia significantly worsens the prognosis of the disease by stabilizing the hypoxia transcription factor HIF-1 α (hypoxia-inducible factor) [10-12]. Stabilization of HIF-1 α promotes adaptation of cells under hypoxic conditions by increasing cell proliferation, neovascularization, antiapoptosis [3, 8, 10, 11]. However, the processes of hypoxic damage and neovascularization of the endometrium in its hyperplasia remain poorly understood.

Another factor closely related to hypoxia is insulin resistance (IR) [3, 13]. According to some authors, IR is the most important adaptive mechanism of the body. It is assumed that the reason for the existence of IR is the constant need of the body for plastic and energy substances, which the body receives mainly from its own depots. In a young healthy organism in a state of rest or normal activity, it is absent when food is used for its needs, but IR is always available when spending its own reserves of nutrients [13].

The effect of insulin on the genetic apparatus of a cell is modulated by intracellular glucose metabolism. The selectivity of the effect of the hormone on individual genes is probably due to the existence of special insulin-sensitive DNA elements that transactivate the promoters of such genes [3, 8, 13, 14]. The mitogenic effect is the slowest effect of insulin. It is assumed that in a number of cells this effect is realized through the interaction of the hormone with the receptors of insulin-like factors, while in other cells (for example, hepatocytes) the mitogenic effect of insulin is mediated by its own receptors. The mitogenic effect of insulin consists in the activation of prenyl transferases (farnesyltransferase, geranyltransferase), in an increase in the farnesylated protein p21, which copresses RAS, RHO, Rab oncogenes and, as a consequence, in an increase in the cellular response to IGF-1, epidermal growth factor, factor platelet growth, the ability of the mutated p21 protein to initiate cell proliferation and transform normal cells into malignant cells, etc. [3, 14]. This leads to the accumulation of molecular damage and the development of obesity,

atherosclerosis, various vascular events, an increase in the risk of developing cancerous tumors, and aging of the body as a whole [14, 15].

There are different types of IR [13]: Physiological IR, Metabolic IR, Endocrine IR, Non-endocrine IR.

Thus, IR occurs not only in type 2 diabetes but also in many other diseases and physiological conditions. IR occurs in more than 25% of practically healthy individuals without obesity, while its severity is comparable to the severity of IR observed in patients with type 2 diabetes

One of the regulatory systems involved in the development of metabolic adaptation is recognized insulin-like growth factor (IGF) system [16-18]. Insulin-like growth factor-1 (IGF-1) - peptide, structurally similar to insulin, but different in effects. At the metabolic level of IGF-1 coordinates the action of growth hormone and insulin and directly affects the intermediate metabolism, is an important stimulator of protein synthesis, regulator of carbohydrate and lipid metabolism [16].

The IGF-1R / PI3K / Akt signaling pathway also mediates the action of IGF as potent mitogens that regulate prenatal and postnatal growth, cell proliferation, differentiation, prevent the mechanisms of cell death, which plays a key role in tissue growth and remodeling, including promoting hypertrophic and tumor growth [16, 17].

The objective: to assess the effect of insulin resistance and hypoxia on the risk of endometrial hyperplasia development.

Material and methods: the study was conducted in Municipal Maternity House №5 (Odessa, Ukraine). The data obtained from the sample of 417 women of reproductive, perimenopausal, and postmenopausal age were analyzed. All the women referred for medical care to the gynecological department for medical and diagnostic scraping of the uterine cavity.

Following clinical groups were formed: Group I consisted of 57 women of reproductive age with hyperplastic processes without atypia, the average age was 35.25 ± 0.37 years; Group II - 48 patients of reproductive age with atypical endometrial hyperplasia (34.48 ± 0.42 years); Group III, the control group consisted of 49 women of reproductive age with proliferative endometrium, who planned pregnancy by ART (33.22 ± 0.48); Group IV - 62 women of perimenopausal period with simple endometrial hyperplasia (48.10 ± 0.44); Group V - 58 patients of perimenopausal age with atypical hyperplasia (48.53 ± 0.48); Group VI - 54 patients of postmenopausal age with simple endometrial hyperplasia (52.78 ± 0.42); Group VII consisted of 43 postmenopausal patients with atypical endometrial hyperplasia ($54.05 \pm$

0.61 years); Group VIII consisted of 46 postmenopausal patients with atrophic endometrium (53.39 ± 0.39 years).

Analysis of IGF, Hif-1 α expression were performed at the mRNA level by polymerase chain reaction of cDNA obtained by reverse transcription. The mRNA level of the test gene was determined by the number of conventional units of the fluorescent signal using the number of um. from fluorescent signal of the 36B4 gene to standardize the original amount of RNA. Changes in expression were calculated by the $\Delta\Delta C_t$ method [19, 20].

Insuline resistance was assessed by HOMA-IR index [21].

Statistical processing was conducted by ANOVA method using software Statistica 13.0 (TISCO, USA) [22].

Results and discussion

Early onset of menarche (under 11 years of age) was noted in 24 (5.8%) patients, after 17 years - in 7 (1.7%), average age - 13.43 ± 0.34 years; duration of menstruation ranged from 3 to 8 days, on average- 4.52 ± 0.18 days; the duration of the menstrual cycle - from 21 to 32 days, on average - 27.12 ± 0.32 days.

History of anovulatory cycles occurred in 35 (10.5%) patients. No birth history in 10 (9.5%) women, abortions in 11 (8.4%) cases.

Many patients have extragenital comorbidities, e.g. obesity was found in 76 (18.2%) patients, metabolic syndrome –in 176 (42.2%), arterial hypertension – in 181 (43.4%), diseases of the gastrointestinal tract - in 66 (15.8%), diseases of thyroid gland – in 38 (9,1%), diabetes mellitus - in 31 (7.4%) cases. History of uterine myoma had 97 (23.3%) patients, pelvic inflammatory diseases - in 65(15.6%), menstrual irregularities - in 79 (18.9%), infertility - in 33 (7.9%).

Clinical manifestations of endometrial hyperplasia in the examined patients are presented in fig. 1.

Interesting results were obtained when analyzing the effect of the severity of hypoxia and insulin resistance on the functional state of the endometrium. As can be seen from Table 1 below, the intensity of hypoxia increases with age and closely correlates ($r=0,78$) with the level of insulin resistance.

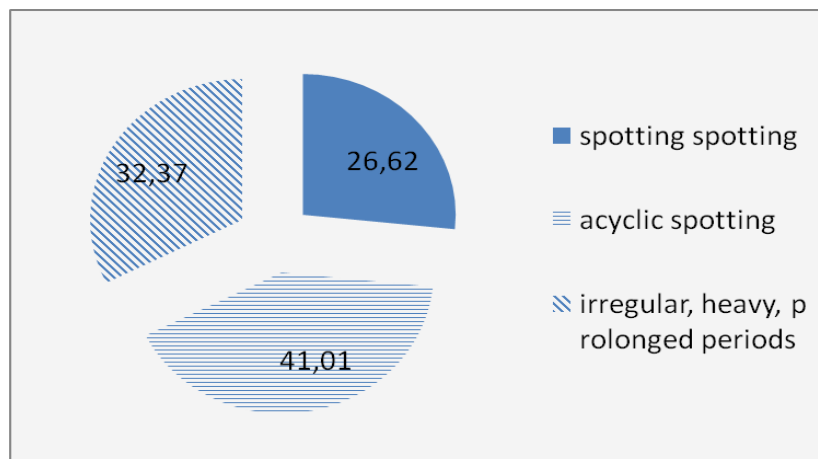


Fig. 1. Clinical manifestations, %

Table 1

Metabolic profile of examined women (M±m)

Indices	I group (n=57)	II group (n=48)	III group (n=49)	IV group (n=62)	V group (n=58)	VI group (n=54)	VII group (n=43)	VIII group (n=46)
HOMA-IR	2,13±0,10	2,24±0,11	1,99±0,11	2,34±0,24	2,59±0,19	2,55±0,17	2,46±0,28	2,86±0,22
IGF, (RU)	0,92±0,08	0,94±0,09	1,70±0,11	1,22±0,22	1,14±0,18	0,34±0,03	0,43±0,04	0,46±0,05
Hif-1α, (RU)	2,76±0,12	2,45±0,09	2,54±0,11	2,62±0,08	2,57±0,11	2,97±0,17	2,89±0,22	3,11±0,24

However IGF demonstrates reverse dynamics – its rates has been diminished with age ($r=-0.63$). We consider that this fact could be useful for prognosis of malignization risk and assessment of clinical course. Actually, endometrial hyperplasia is rare in women under the age of 30 but its prevalence and incidence drastically increasing in peri- and postmenopausal age. Thus endometrial hyperplasia being found to be more common in early postmenopausal women (within 5 years of menopause) compared to late postmenopausal women (over 5 years from menopause) [1, 2, 5].

There is an imbalance between proliferation processes and programmed death cells - apoptosis is sharply suppressed. At the same time, conditions are formed for the stability of pathological foci of the endometrium to apoptosis and cell resistance to hypoxia. It may be additional factors for progressing endometrial hyperplasia to adenocarcinoma. Even in the early stages of the pathological process, active processes of stroma formation and neoangiogenesis are noted. They lead to chronic hypoxia and formation of vicious circle.

Conclusion:

1. Both insulin resistance and hypoxia play significant role in the pathogenesis of endometrial hyperplasia
2. Indices of IGF and HIF could be recommended as valuable biomarkers of prognosis in cases of endometrial hyperplasia

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