EVALUATION OF ESTROGEN AND PROGESTERONE RECEPTORS (ER, PR) IN ENDOMETRIAL HYPERPLASIA AND CARCINOMA- AN IMMUNOHISTOCHEMICAL STUDY

Pathology

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ABSTRACT

The endometrium which lines the uterine cavity is one of the most dynamic tissues in the human body. Proliferation and differentiation of the endometrial glands and stroma are regulated by steroid hormones mainly estrogen and progesterone. Endometrial hyperplasia is a heterogeneous set of pathologic lesions that range from mild, reversible glandular proliferations to direct cancer precursors. Endometrial carcinoma is one of the most common gynecologic malignancies in industrialized and developing countries and is generally accepted to be an endocrine related neoplasm. Understanding the biochemical basis of endometrial responsiveness to hormones in such patients is fundamental to designing a successful medical therapy. The estrogen receptor (ER) and progesterone receptor (PR) are measured by biochemical and immunohistochemical methods. Immunohistochemistry (IHC) is a semiquantitative method for determination of protein expression. The technique is inexpensive and relatively quick to perform. Immunohistochemistry was done in this study to evaluate the expression of ER, PR in cases of Endometrial hyperplasia and Endometrial Carcinoma. A total of 42 cases were evaluated immunohistochemically which included 32 cases of Non atypical hyperplasia, 7 cases of Atypical hyperplasia and 3 cases of Endometrial carcinoma. The expression of ER/PR was maximal for cases of Non atypical hyperplasia, followed by atypical hyperplasia and least for endometrial carcinoma. Those can be benefited from hormonal therapy, especially atypical hyperplasia cases, whose progression can be halted by hormonal therapy.

KEYWORDS

Endometrial hyperplasia, Endometrial Carcinoma, Estrogen and Progesterone Receptors, Immunohistochemistry.

INTRODUCTION:

Endometrial hyperplasia is an important cause of abnormal bleeding and a frequent precursor to the most common type of endometrial carcinoma. Clinicopathologic and epidemiologic studies have supported the malignant potential of endometrial hyperplasia and the concept of a continuum of proliferative glandular lesions culminating, in some cases, in carcinoma.

The most current World Health Organization (WHO) classification recommends collapsing the four categories into two major categories which differ in appearance and in their propensity to progress to carcinoma: 1. Non-atypical hyperplasia and 2. Atypical hyperplasia. The cyclic change of endometrium is regulated elaborately by hormone. Estrogen promoted hyperplasia of gland and stroma; progesterone promoted gland transformation from proliferating phase to the secretory phase, with stromal decidual change. Estrogen and progesterone specific binding with their receptors are the most important step in their biological effects.²

Endometrial hyperplasia develops in a setting of estrogen excess. Potential precancerous lesions of endometrium such as complex hyperplasia with or without atypia may contain levels of estrogen and progesterone receptors that are distinct from carcinomas.³

Detection of high levels of steroid receptors denotes a good response to hormonal therapy (progesterone) in simple and complex hyperplasia. Decrease in receptor activity which is found in atypical hyperplasia results in low sensitivity to progesterone therapy. If a higher level is found in some rare incidence of atypical hyperplasia, they have a good chance of response to hormonal therapy and a radical surgery can be avoided. Higher positivity in malignant lesion usually correlates with better differentiation and better survival rates.⁴

Endometrial carcinoma is the most common invasive cancer of the female genital tract. Endometrial carcinomas also contain estrogen receptors (ER) and progesterone receptors (PR), but several studies have reported that the levels are reduced or absent as compared to normal cyclical endometrium and hyperplastic endometrium. Higher positivity of receptor expression in malignant lesions usually correlates with better differentiation and better survival rates.^{5,6}

This study aimed at the evaluation of estrogen and progesterone receptors in endometrial hyperplasia and carcinoma by

immunohistochemistry.

METHODS AND MATERIALS:

This prospective study was conducted in the Department of Pathology, Gauhati medical college and hospital, Guwahati from June 2018 to May 2019. Endometrial dilatation and curettage samples or hysterectomy specimens of the patients, collected or done in the premenstrual phase of the cycle or at random for cases presenting with irregular cycles, was sent to Pathology Department from Gynaecology Department. Endometrial dilatation and curettage samples or hysterectomy specimens received were fixed in 10% neutral buffered formalin and brought to the laboratory. The histological preparation was performed by the classical method for inclusion in paraffin, followed by Hematoxylin and eosin staining.

Immunohistochemical markers such as ER & PR were done from paraffin block of the patients who had a histological diagnosis of endometrial hyperplasia or carcinoma. Normal cyclical endometrium taken as positive control.

We have used the Immunoreactive score (IRS-score) which is calculated as follows: $IRS = SI \times PP$, where SI is the optical staining intensity (graded as 0 = no, 1 = weak, 2 = moderate and 3 = strong staining). PP is the percentage of positively-stained cells. The PP was estimated by counting approximately 200 cells and it was defined as 0 = no staining, 1 = 81%.⁷

RESULTS:

During this study period of one year a total of 42 cases were evaluated immunohistochemically which included 32 cases of Non atypical hyperplasia, 7 cases of Atypical hyperplasia and 3 cases of Endometrial carcinoma.

10 cases of normal cyclical endometrium (10 proliferative) were also studied for the receptor status. Positive staining of both ER and PR was seen as fine granular staining in nuclei of glands and stroma. Staining was scored semi quantitatively taking into consideration both intensity as well as percentage of cells stained in the glands and stroma.

The intensity and distribution of ER and PR was evaluated using a semi-quantitative method (IRS-score). The IRS score was calculated as follows: $IRS = SI \times PP$, where SI is the optical staining intensity (graded as 0 = no, 1 = weak, 2 = moderate and 3 = strong staining). PP is

the percentage of positively-stained cells. The PP was estimated by counting approximately 200 cells and it was defined as 0 = no staining, 1 = 81%. Unpaired Student's t-test was performed by using SPSS 21.0 and MS Excel softwares. p-value < 0.05 was taken as critical level of significance.

Table 1 : Table showing Comparison of ER and PR expression scores in glands and stroma of endometrium in control proliferative (n=10) and Non Atypical Hyperplasia (n=32) groups.

ER	Control Proliferative n= 10	Non Atypical Hyperplasia n=32	p-value
Gland	4.1± 0.25	9.69±0.92	<0.001
Stroma	3.3± 1.12	9.31±1.55	<0.001
Total Score	7.4±1.18	19.00±2.00	<0.001
PR	Control Proliferative n=10	Non Atypical Hyperplasia n=32	p-value
Gland	4.5±1.05	9.44±1.27	<0.001
Stroma	3.1±1.01	9.41±1.95	<0.001
Total Score	7.6±1.11	18.84±2.49	<0.001

Figure: 1

Figure:2

The ER and PR expression in both glands and stroma of the endometrium were significantly higher in Non-atypical hyperplasia (n=32) as compared to Control proliferative (n=10) (p-value < 0.001).

Table 2 : Table showing Comparison of ER and PR expression scores in glands and stroma of endometrium in Non-atypical hyperplasia (n=32) and Atypical hyperplasia (n=7) groups.

ER	Non atypical hyperplasia (n=32)	Atypical hyperplasia (n=7)	p-value
Gland	9.69±0.92	4.14±0.83	<0.001
Stroma	9.31±1.55	3.57±0.49	<0.001
Total Score	19.00±2.00	7.71±1.16	<0.001
PR	Non atypical hyperplasia (n=32)	Atypical hyperplasia (n=7)	p-value
Gland	9.44±1.27	3.43±1.29	<0.001
Stroma	9.41±1.95	3.57±0.49	<0.001
Total Score	18.84±2.49	7.00±1.41	<0.001

In Atypical hyperplasia (n=7), the ER and PR expressions in both glands and stroma of the endometrium were reduced. The ER and PR expression in both glands and stroma of the endometrium were significantly higher in Non-atypical hyperplasia (n=32) as compared to AUB Atypical hyperplasia (n=7) (p-value < 0.001).

Table 3 : Table showing Comparison of ER and PR expression scores in glands and stroma of endometrium in Non-atypical hyperplasia (n=32) and Endometrial carcinoma(n=3)

ER	Non atypical hyperplasia (n=32)	Endometrial Carcinoma (n=3)	p-value
Gland	9.69±0.92	5.00±1.41	0.01
Stroma	9.31±1.55	4.67±0.94	0.03
Total Score	19.00±2.00	9.67±2.05	0.018
PR	Non atypical hyperplasia (n=32)	Endometrial Carcinoma (n=3)	p-value
Gland	9.44±1.27	5.00±2.16	<0.001
Stroma	9.41±1.95	5.33±1.89	0.036
Total Score	18.84±2.49	10.33±4.03	0.041

The ER and PR expression in both glands and stroma of the endometrium were significantly higher in AUB-Non atypical hyperplasia (n=32) as compared to Endometrial Carcinoma (n=3) (p-value < 0.05).

DISCUSSION:

In our study The ER and PR expression in both glands and stroma of the endometrium were significantly higher in Non-atypical hyperplasia (n=32) as compared to Control proliferative (n=10) (p-value < 0.001) which is in concordance with the study of Chakraborty S et al 8 (2004),

Chakravarthy VK et al 9 (2013), Jiang W et al 10 (2015) , Singh P et al 11 (2016), Mostafa AM et al 12 (2018) .

In the present study ,In Atypical hyperplasia (n=7), the ER and PR expressions in both glands and stroma of the endometrium were reduced. The ER and PR expression in both glands and stroma of the endometrium were significantly higher in Non-atypical hyperplasia (n=32) as compared to AUB Atypical hyperplasia (n=7) (p-value < 0.001) which is in concordance to the study by Teleman S et al 13 , (1991), Nyholm et al 3 ,(1992), Chakraborty S et al 8 (2004), Johnsy MJ et al 14 (2015).

The ER and PR expression in both glands and stroma of the endometrium were significantly higher in AUB-Non atypical hyperplasia (n=32) as compared to Endometrial Carcinoma (n=3) (p-value < 0.05) in our study which is similar to the findings of Mylonas et al 7,(2005), Ilie D et al 15 (2011), Vellanki S et al 16 (2017).

Photomicrograph 1: Proliferative endometrium showing ER positivity in glands and stroma

Photomicrograph 2 : Proliferative endometrium showing PR positivity in glands and stroma

Photomicrograph 3 showing strong ER positivity in glands and stroma of Non atypical hyperplasia

Photomicrograph 4 showing strong PR positivity in glands and stroma of Non atypical hyperplasia

Photomicrograph 5 : Endometrioid carcinoma showing ER positivity

Photomicrograph 6: Endometrioid carcinoma showing PR positivity

Photomicrograph 7: Endometrioid carcinoma (H &E)

Photomicrograph 8: Atypical hyperplasia(H&E)

Photomicrograph 9: Non atypical hyperplasia (H&E)

Photomicrograph 10 : Proliferative endometrium

CONCLUSION:

Evaluation of endometrial histology can be made more meaningful with the study of Estrogen Receptor (ER) and Progesterone Receptor (PR).

Endometrial hyperplasia and carcinoma that show positive ER/PR expression could benefit from hormonal therapy, especially hyperplasia cases, whose progression can be halted by hormonal therapy and in case of atypical hyperplasia an invasive surgery can be avoided.

This study is a short term study with limited number of subjects. ER, PR are very important immune-markers. Thus the role of Estrogen Receptor (ER), Progesterone Receptor (PR) in Endometrial hyperplasia and Endometrial Carcinoma should be further explored with larger sample size. It may be helpful to assess the prognosis and in providing evidence based treatment.

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