Original Research Article

Diagnostic and Prognostic utility of Hormone receptors and KI67 in Proliferative Endometrial lesions

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Received: 21-09-2020 / Revised: 26-10-2020 / Accepted: 22-11-2020

Abstract

Background: The overall survival rate of patients with early-stage endometrial cancer is relatively high; how-ever, there are few treatment options for patients with advanced or recurrent endometrial cancer, and the prognosis of such patients remains poor. Recent progress in molecular-targeted therapies demonstrated that they have the potential to improve the long-term survival of cancer patients with appropriate biomarkers. In this study we evaluated 50 patients with endometrial proliferative lesions and their correlation with hormonal receptors and proliferative index, with emphasis on the utility of these markers in disease prognosis. **Objective**: To evaluate the association of expression of ER, PR and Ki67 in endometrial hyperplasia and carcinoma. **Material and Methods:** Small and large biopsy samples obtained from 50 female patients were evaluated by histopathology and immunohistochemistry by using specific markers. Statistical analysis was done to determine the P value. **Results:** Out of 50 cases studied 16 cases (32%) were endometrial hyperplasia and 34 cases (68%) were of endometrial carcinoma. Among the carcinoma cases 22 cases (65%) were grade 3, 8 (23%) were grade 2 and 4(12%) were grade 1 tumors. Assessment of myometrial invasion was possible in 23 cases out of which 20 cases showed >50% invasion and 3 cases showed <50% invasion. Cervical invasion was present in 17 cases and vascular invasion was seen in 10 cases. Negative expression of hormonal receptors was seen in grade 3 (high grade) endometrial carcinomas. P value was estimated at 0.0001. The proliferative index was high (>35%) in grade 3 (high grade) endometrial carcinoma. P value was 0.006.**Conclusion:** There is significant association of expression of ER, PR and Ki67 in endometrial carcinomas with respect to histological variants, grade, tumor invasion and metastasis.

Keywords: lesions,endometrium,receptors

Introduction

Over the last 15 years, there has been an increase in endometrial cancer related deaths, supporting the notion that therapies for endometrial cancer need to be urgently revised and improved.[1] Endometrial Adenocarcinoma is divided into two broad histological types. Type 1 includes Endometrioid and mucinous carcinoma accounting for about 80% of the cases wherein there is unopposed estrogen stimulation.[2] These tumors are often associated with precursor lesions such as Atypical Endometrial Hyperplasia (AEH) or Endometrial Intraepithelial Neoplasia (EIN) and presents as low grade tumor with distinct genetic abnormalities such as PTEN, PAX2and k-Ras mutation.[2] . Type 2 includes Serous Carcinoma, Clear Cell Carcinoma, undifferentiated carcinoma and carcinosarcoma accounting for about 10% of the cases less associated with estrogen stimulation, presenting with higher tumor grade and stage.[2,3] According to 2014 WHO classification, hyperplasias are classified as hyperplasia without atypia and AEH or EIN. Hyperplasia without atypia does not show relevant genetic alterations and less than 2% progress to carcinoma in case endocrine abnormality persists.[3] However, atypical endometrial hyperplasia progresses to endometrial adenocarcinoma in 23% of cases.[3] In

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order to improve the efficacy of endometrial carcinoma treatment, identification of high-risk prognostic factors is a high research priority.[3] Early assessment could enable conservative therapy in patients with favorable prognosis as well as reserve effective and more radical therapy for patients with aggressive forms of the tumor. [4]

The use of estrogen receptor (ER), progesterone receptor (PR), human epidermal growth factor receptor 2 (HER2), and Ki-67 have been routinely used in breast cancer and endometrial carcinoma cases for molecular sub typing and guiding treatment.[4] Numerous studies showed that prognosis for endometrial carcinoma is closely related to patient age, tumor grade, depth of invasion and/or cervical involvement, and the occurrence of lymph node metastases.[5]

Recently a panel of immuno histochemical markers (ER, PR, Her-2, and Ki-67) was tested to ascertain their relationships with the histopathological prognostic parameters of endometrial carcinoma. [5] Ki-67 is a nuclear antigen present only in proliferating cells except G0 phase of cell cycle.[5] Increase in antigenic expression during cell cycle in both benign and malignant cell lines assessing their proliferative status has also been shown.(5)Ki-67 score is now used to predict the prognosis, survival, and even the recurrences.[5]

A high level of Ki-67 proliferative index (PI) is associated with aggressive tumoral behavior and metastasis.[5] The over expression of HER2 and Ki-67 and low expression of ER and PR indicate a more malignant EC behavior.[6] In this study we intend to study the expression of immuno histochemical markers in proliferative lesions of endometrium and their utility for targeted therapy of endometrial carcinoma.

Aims and objectives

Aim of this work is to study the expression of hormone receptors (ER, PR) and proliferative index in correlation with proliferative lesions of the endometrium and study their prognostic significance in cases of endometrial carcinoma.

Material and Methods

A prospective and observational study was done in a tertiary care hospital with a sample size of 50 cases. Female patients attending gynecology OPD with complains of abnormal uterine bleeding, peri menopausal symptoms and increased endometrial thickness on radiology were included in the studied. Samples (small biopsy and hysterectomy specimen) from such patients were evaluated for histopathological examination. 50 cases of endometrial hyperplasia and carcinoma were included in our study. Exclusion criteria were cases diagnosed as inflammatory lesion, polyp, progesterone effect and secretory changes. All tumor lesions were evaluated for histological grade, tumor type, depth myometrial invasion and metastasis. Hormonal receptors(ER, PR) and proliferative index (Ki67) was assessed using appropriate immuno histochemical markers. Statistical analysis was done to find the association of immunoexpression of ER, PR and Ki67 with endometrial hyperplasia and carcinoma.

Results

Among 50 cases the mean age of hyper plastic lesions was 51.5 years and 53.5 years in cases of carcinoma. The chief complaint was abnormal uterine bleeding (72%) followed by pain abdomen in 12% cases. Eight cases (16%) had history of unopposed estrogen exposure. On Transvaginal UGG thickness of >=5 mm was seen in 90% cases. Out of 50 specimens received 21(42%) were hysterectomy specimens, 17 (34%) were D/C specimen and 12(26%) were endometrial biopsies. On histopathological examination 16 cases were of endometrial hyperplasia and 34 cases were of endometrial carcinoma.(Table 1) Among the carcinoma cases 22 cases(65%) were grade 3, 8 (23%) were grade 2 and 4(12%) were grade 1 tumors. Assessment of myometrial invasion was possible in 23 cases out of which 20 cases showed >50% invasion and 3 cases showed <50% invasion. Cervical invasion was present in 17 cases and vascular invasion was seen in 10 cases.

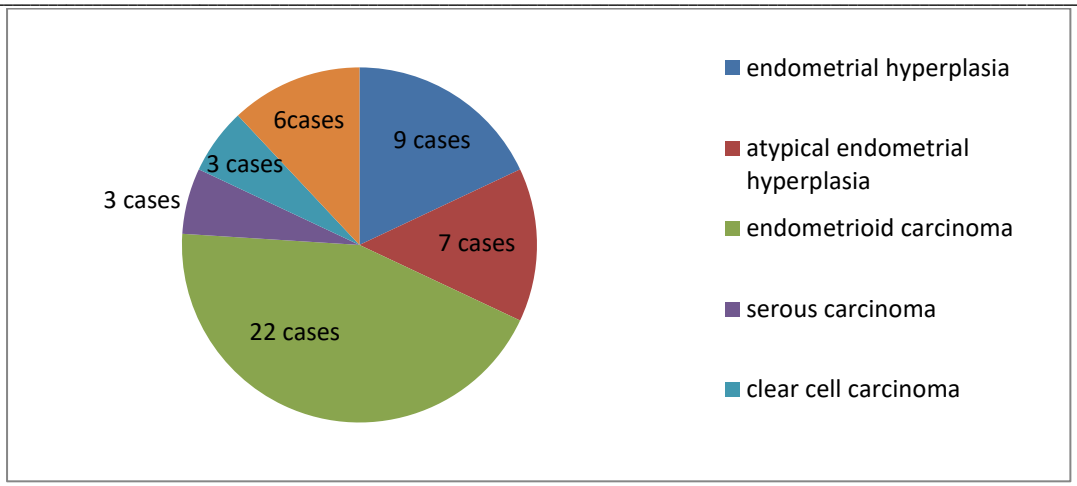

Fig1: Distribution of histological variants of endometrial lesions (n=50)

Examination of hormonal expression among the cases of endometrial hyperplasia showed positive ER expression among 8 cases (50%) of simple hyperplasia and 6 cases (37.5%) of atypical endometrial hyperplasia.

Positive PR expression was seen in 7 cases (43.75%) of simple hyperplasia and 6 cases (37.5%) cases of atypical endometrial hyperplasia. IHC study for proliferative index (Ki 67) shows >35% in 1 case (6.25%) of simple hyperplasia and 1 case (6.25%) of atypical endometrial hyperplasia. Among the 34 cases of endometrial carcinoma ER expression was positive in 14 cases (41.2%) of endometrioid carcinoma. PR expression was positive in 16 cases (47.05%) of endometrioid carcinoma. Ki67 index showed >35% staining in 6 cases(17.64%) of undifferentiated carcinoma, 3 cases (08.82%) of serous carcinoma and 3 cases (08.82%) of clear cell carcinoma.(Table 1)

Table 1: Distribution of expression of hormonal receptors and proliferative index among endometrial hyperplasia and carcinoma (n=50)

IHC	Status	Endometrial hyperplasia (n=16)		P value	Endometrial carcinoma (n=34)	P value
		SH	AEH			
ER	Negative	1	1	1	20	0.0047
	Positive	8	6		14	
PR	Negative	2	1	1	18	0.0009
	Positive	7	6		16	
Ki 67	<35%	8	6	1	22	
	>35%	1	1		12	0.0001

On histological examination, endometrial carcinomas were graded as per their features. Negative expression of hormonal receptors was seen in grade 3 (high grade) endometrial carcinomas. P value was estimated at 0.0001. The proliferative index was high (>35%) in grade 3 (high grade) endometrial carcinoma. P value was 0.006. With reference to myometrial and vascular invasion loss of hormone receptors was seen in tumors with more than half of myometrial invasion (16 cases) and presence of vascular invasion (9 cases). High proliferative index (>35%) was seen in 14 cases of carcinoma with >1/2 myometrial invasion and 9 cases with vascular invasion. P value was significant with a value of 0.02. Our results showed that histologically high grade endometrial tumors with myometrial and vascular invasion have loss of hormone receptors and high proliferative index, because of which survival rate of patients with early-stage endometrial cancer is relatively high.

Discussion

Salama A et al in 2019 studied immuno histochemical markers among 109 cases of endometrial carcinoma

and found mean age of patients in this study was 59.8 ± 8.2 years.[7] Low ER and PR expression scores and high Ki-67 expression showed highly significant associations with non-endometrioid histology (p = .007,p<.001,and p<.001,respectively) and poor differentiation(p =.007,p<.001,and p<.001,respectively). [7]

Low PR score showed a significant association with advanced stage (p =.009).[7] Masjeed et al in 2017 studied 85 cases of endometrial hyperplasia and 28 cases of endometrial carcinoma, the peak incidence of EH was seen in 41-50 years.[8] Mean age was 44.52±7.3 years and the peak incidence of endometrial carcinomas was seen in 51-60 years of age.[8] Mean age was 58.14±9.57 years. ER expression was seen more in Simple hyperplasia (65/70, 92.85%) than in AEH (85.71, 12/14) and endometrial carcinoma (17/28, 60.71%). This was statistically significant ($\chi 2$=15.357; p<0.01).(8) PR expression was seen more in Simple hyperplasia (63/70, 90%) than in AEH (12/14, 85.71%) and endometrial carcinoma (18/28, 64.28%). This was statistically significant ($\chi 2$=9.470; p<0.01).[8] This shows that ER and PR expression has inverse correlation with the severity of endometrial lesion. The progesterone therapy for endometrial hyperplasia and neoplasia is based on its ability to inhibit DNA synthesis and to induce regression of abnormal endometrial proliferation.[9]The estrogen-agonist effect on the uterus is manifested by polypoid endometrial proliferation with glandular hyperplasia ranging from simple to complex and atypical.[9] Combination of progesterone and tamoxifen therapy in high grade endometrial carcinoma has been reported in recent literature.[9] Ki67 positivity increased as the severity of endometrial lesions increased from EH to endometrial carcinoma which was statistically significant ($\chi 2$=6.106; p<0.05).[8] Mean Ki67 increased from 8.4% in simple hyperplasia to 9.8% in atypical endometrial hyperplasia.[8] In all cases of endometrial carcinoma, mean Ki67 was increasing from a value of 22.52 % to 40% as the grade increased[8]. In our study mean age of hyper plastic lesion were 51.5 years and 53.5 years in cases of carcinoma. ER and PR expression was more in simple hyperplasia cases compared to atypical hyperplasia. Negative hormonal receptor expression was seen in high grade tumors with myometrial and vascular invasion. Ki 67 index was >35% in grade 3 tumors predominantly undifferentiated type. The presence of hormone receptors in ECs correlates with the clinical disease stage, histological grade, and overall survival. The absence of hormone receptors is considered to be indicative of aggressive tumor behavior and poor prognosis. Increased Ki-67 expression indicates higher mitotic activity and greater tumor cell proliferation.[9] Kitson et al in 2017 studied biopsy specimens of 179 women and found that Ki-67 scores positively correlated with grade, stage and depth of myometrial invasion (P-values all o0.03).[10] By univariate analysis, higher Ki-67 scores were associated with a significant reduction in cancer-specific survival (P≤0.05); however, this effect was substantially attenuated in the multivariate model.[10,11]

Conclusion

There are few choices of useful treatments for advanced or recurrent endometrial carcinoma other than radiotherapy for localized lesions. The establishment of a new strategy, for treatment based on the genetic background or tumor micro environment of Endometrial Carcinoma, is needed. In terms of biomarkers, studies showed that classical immuno staining; serum tumor markers and gene expression testing all have been useful in identifying effective new prognostic and predictive biomarkers to improve the therapeutic decision-making process.

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Conflict of Interest: Nil
Source of support:Nil