Immunohistochemical Expression of KI-67 in Ovarian Surface Epithelial Tumors and its Correlation

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ABSTRACT

The ovaries are unique in the variety of lesions that can arise from them. The complex anatomy and physiology of ovary with constant changes from puberty to menopausal age give rise to number of different cells, which are capable of giving rise to tumors. The present study is a two years, prospective study carried out in a tertiary care hospital. It is a descriptive study which included all specimens of ovarian tumors, diagnosed as surface epithelial origin on histopathological studies. Most patients presented with non-specific and vague symptoms over a period of time. Patient's education and awareness among primary care physicians with timely attention and further investigations, can help in early detection of ovarian malignancies in this subset of cases, thereby reducing morbidity and mortality associated with diagnosis in advanced stages.

Keywords: Immunohistochemical, Ki-67, Histopathologic, Ovarian tumor, endosalpingiosis

INTRODUCTION

The ovaries are unique in the variety of lesions that can arise from them. The complex anatomy and physiology of ovary with constant changes from puberty to menopausal age give rise to number of different cells, which are capable of giving rise to tumors. Both primary as well as metastatic tumors of the ovaries are relatively frequent.[2] Ovarian malignancy is the fifth common malignancy among women and second most common gynaecologic malignancy. It is the most common cause of mortality associated with female genital tract cancers.[3] Ovarian tumors represent about 30% of all cancers of FGT.[4] As per data collected in 2010 by American Cancer Society, each year over 22,000 women are diagnosed worldwide with ovarian cancers and 15,000 die of it.[5] Asian countries have rate of 2-6 new cases of ovarian cancer per 1,00,000 women per year. In India, the ovary is the important seat of cancer of female genital tract next to cervix.[6]

Ovarian cancer remains enigmatic disease especially surface epithelial ovarian tumors. Their behaviour, pathogenesis and their clinical management is controversial. Ovarian surface epithelial tumors constitute two third of all ovarian tumors and malignant SETs account for 90% of all ovarian malignancies.[8] These are present with different histological patterns with varying amount of aggressiveness.[9] At diagnosis, nearly 70% to 75% of the surface epithelial cancers have spread beyond ovary.[10] Rapid pre-operative diagnosis of the ovarian masses is important for surgeons to conserve the other ovary in young patient with benign tumors and for early institution of chemotherapy in malignant tumors.[11]

The high proliferation rate has been associated with tumor aggressiveness and correlates well with the prognosis and other known clinico-pathological features of the tumor. Ki-67 is a stronger and robust indicator for diagnosis of uncontrolled cellular proliferation than the routinely used mitotic activity on routine H&E stained sections.[12]

The present study titled as Study of immunohistochemical expression of Ki-67 in surface epithelial ovarian tumors and its correlation with clinic-pathological factors was undertaken.

AIM & OBJECTIVE

AIM

To study immunohistochemical expression of Ki-67 in ovarian surface epithelial tumors and correlation of Ki-67 expression with clinico-pathological factors.

OBJECTIVES

To diagnose ovarian surface epithelial tumors by histopathological study of excised ovarian tumors, according to WHO (2014) histopathological classification of ovarian tumors. To study ovarian surface epithelial tumors with clinico-pathological factors viz. age, tumor type, grade, stage, ascites and serum CA-125 level etc. To study immunohistochemical expression of Ki-67 in ovarian surface epithelial tumors. To study biological significance of Ki-67 expression in ovarian surface epithelial tumors.

REVIEW OF LITERATURE

The ovaries which Hippocrates mentioned as 'Female Testes', was first identified as a female reproductive organ in 300 BC. The term "Ovary" was coined in the 7th Century AD. Sushruta described tumor of the ovary in ancient India in his literature of excellence on medical knowledge "Sushruta Samhita".[15]

Embryology[16,17] : As emphasized by Harold N et al in his study, in order to understand the broad range of ovarian tumors and the clinical significance of each tumor, it is essential to review embryological origins of various histological sub-types.[14]

Glazuvan in 1937 postulated that serous cystadenomas can arise in the foci of endosalpingiosis, term coined by Barzilai in 1943 for the same was Endosalpingioma.[1,7]

Micro-papillary variant is histologically composed of cellular proliferation that arises from the surface of fibrous papillae without hierarchical branching. It was first observed in 1996. It was a spectrum of serous borderline tumor associated with micro-papillary pattern and worse prognosis than usual serous borderline tumor. The term micro-papillary serous carcinoma was considered by WHO in 2000. Majority clinicians preferred to call it as micro-papillary serous borderline tumor over carcinoma.

When this type of tumor shows stromal invasion exceeding 5 mm, then they are considered as invasive micro-papillary carcinoma, which is categorized under low grade serous carcinoma[13]. When these micro-papillary serous tumors are associated with >75% of psammoma bodies, the term psammocarcinoma is used. 22% population with serous ovarian tumors shows invasive implants. They are generally associated with stage III tumors. These kind of implants are associated with haphazard growth with or without cribriform glandular pattern with mild cytological atypia with occasional mitosis.[13]

Zollacus (1952) coined the term pseudo-mucin and proved that it can be differentiated from true mucin as it is not precipitated by acetic acid[18]. Mucinous tumors are 2nd most common and are bilateral in only 10- 20% cases. These occur during the 3rd-6th decades.[19]

These are large, unilateral, multilocular and contain viscous mucoid material. Sometimes, they even show solid components, as in case of Cystadenofibroma.

Samption (1925) first described this type of SET, which resembles uterine endometrial carcinoma. The tumor arises from the metaplastic transformation of coelomic epithelium to the endometrial type. Ovarian endometrioid carcinomas comprise 10-25% of all primary ovarian carcinomas[20]. It is most commonly observed around peri-menopausal age group and almost 10-20% cases show association with endometriosis. These are partly solid, partly cystic, tan cystic lesions ranging 8-10 cm in diameter. It reveals cysts lined by benign appearing endometrial type of cells, sometimes associated with squamous differentiation.[21]

It reveals highly compact area of endometrioid glands or cysts with cytological atypia, without any stromal invasion. Few areas of low mitotic activity are also noted. Surrounding ovary shows focus of endometriosis in many cases.[22,23]

MATERIAL AND METHODS

Source of data

The present study is a two years, prospective study carried out in a tertiary care hospital. It is a descriptive study which included all specimens of ovarian tumors, diagnosed as surface epithelial origin on histopathological studies from June 2016 to May 2018. Histopathologically confirmed cases were further evaluated for clinico-pathological factors and investigated for their Ki-67 labeling index.

Method of Data Collection

10% Formalin preserved specimens were received after scrutiny of patients demographic and clinical details along with details of surgical procedures performed. The specimens were fixed in 10% neutral buffered formalin for 12-24 hours after serial sectioning and noticing gross external and internal details. Proper care was taken so that specimen were not left in formalin for more than 24 hrs as antigen retrieval procedure of IHC staining get affected with over-fixation.

OBSERVATIONS AND RESULTS

In two years from June 2016 to May 2018, total 98 ovarian tumor specimens were received in histopathology section of department of Pathology of tertiary care hospital.

Table 1: WHO categorization of Surface Epithelial Tumors

Surface Epithelial Tumor	No. of cases	Percentage (%)
Benign	46	54.76
Borderline	4	4.76
Malignant	34	40.48
Total	84	100

Out of these 84 cases of SETs studied, 54.76% (46/84) were benign, 4.76% (04/84) were borderline and 40.48% (34/84) were malignant.

Table 2: Age-wise distribution of cases with SETs

Age (in years)	Benign	Borderline	Malignant	Total
0-10	0	0	0	0 (0%)
11-20	1	0	0	1 (1.64%)
21-30	8	1	0	9 (14.75%)
31-40	10	1	2	13 (21.31%)
41-50	9	2	6	17 (27.87%)
51-60	3	0	3	6 (9.84%)
> 60	6	0	9	15 (24.59%)
Total	37	4	20	61 (100%)

Maximum number of the cases (17/61, 27.87%) in the present study presented in the fifth decade. Younger patients in 20-40 years age group had benign tumors more often (18/22, 81.82%); while those presenting in the 6th decade and later showed a greater predilection for malignant tumors (12/21, 57.14%).

Table 3: Distribution of cases as per clinical presentations

Chief presenting complaint	Benign	Borderline	Malignant	Total
Abdominal Pain	25	2	15	42 (68.85%)
Abdominal mass/lump	3	1	2	6 (9.84%)
Abnormal bleeding	3	0	2	5 (8.20%)
Abdominal Fullness	2	1	0	3 (4.92%)
Difficulty in micturition	2	0	1	3 (4.92%)
Irregular menses	1	0	0	1 (1.64%)
Asymptomatic	1	0	0	1 (1.64%)
Total	37	4	20	61 (100%)

Remarkably, 60/61 (98.36%) cases were symptomatic, only 1/61 was asymptomatic. Majority of the cases (42/61, 68.85%) presented with chief complaint of pain in abdomen which was confined to lower abdominal quadrant and was of dull or dragging type with no radiating pattern. Abdominal lump/mass was the second most common clinical presentation in 9.83% (6/61) cases. A single case, who was asymptomatic, was a pregnant female with 5 months amenorrhea. She was incidentally diagnosed with ovarian cyst on ultra-sonography, which turned out to be benign mucinous cystadenoma on histopathological study.

Table 4: Surface Epithelial Ovarian Tumors according to Menopausal status

Menopausal status	Benign	Borderline	Malignant	Total
Premenopausal	19	2	2	23 (37.70%)
Perimenopausal	9	2	6	17 (27.87%)
Postmenopausal	9	0	12	21 (34.43%)
Total	37	4	20	61 (100%)

The proportion of cases presenting in the pre-menopausal (23/61, 37.70%) age group was slightly higher than that of post-menopausal (21/61, 34.42%) and peri-menopausal (17/61, 27.87%) age group. Major proportion of cases with benign surface epithelial tumors were in pre-menopausal (19/37, 51.35%) age group, while majority (12/20, 60%) malignant SETs were diagnosed in post-menopausal age group. No case with borderline SET was detected in post-menopausal age group. Borderline variety showed equal incidence in pre-menopausal and peri- menopausal age group (2 cases each).

Table 5: Distribution of SETs according to the surgical procedure adopted for management

Procedure Performed	Benign	Borderline	Malignant	Total surgeries
Panhysterectomy	6	1	9	16 (26.23%)
Total Hysterectomy with bilateral Salpingo- oophorectmy	5	1	7	13 (21.31%)
Salpingo-oophorectomy	4	0	0	4 (6.56%)
Oophorectmy	17	2	2	21 (34.43%)
Cystectomy	5	0	0	5 (8.20%)

Excisional Biopsy	0	0	2	2 (3.28%)
Total	37	4	20	61 (100%)

Oophorectomy was the most common (17/37, 45.94%) procedure performed for benign surface epithelial ovarian tumors; while Panhysterectomy (9/20, 45%) followed by total hysterectomy (7/20, 35%) were the procedures of choice for malignant surface epithelial tumors of ovary. In cases with surgically unresectable tumors, excisional biopsies were performed and were almost exclusively performed on malignant tumors. In the present study excisional biopsy was performed for 2 cases with malignant SETs.

Table 6: Correlation of Ki-67 labeling index and outcome

Ki-67 Labeling Index	Disease Free	Living with Disease	Died	Total	Chi Square value - 32.16; P-value <0.001
Low	21	0	0	21 (53.85%)	
Intermediate	3	3	1	7 (17.95%)	
High	0	4	7	11 (28.21%)	
Total	24	7	8	39 (100%)	

In the present study, out of 39 cases followed, majority of cases who were disease-free, were of low Ki-67 index (21/24, 87.50%) and were of benign variety of SETs. Sizeable number (7/11, 63.64%) of malignant cases with high labeling index lost their lives and 4/11 (36.37%) are alive with disease burden. In borderline tumor variety, 3 out of 4 (75%) cases were followed up; of which, 2/3 (66.66%) lost their lives and 1/3 (33.33%) was leading a disease free life. Out of 2 cases who lost their life, 1 was belonging to high and 1 belonged to intermediate Ki-67 labeling index group.

DISCUSSION

Ovaries are common sites of non-neoplastic and neoplastic lesions. Throughout the life span, from puberty to menopause, ovaries go through many cyclical changes during this time with different cell types. Each of these cells has potential to develop varieties of tumors.[21] Determination of proliferative activity of a tumor plays very important role in prognostication of a tumor. There are various methods available for determining proliferative activity of a tumor e.g. proliferative cell nuclear antigen, Bromodeoxyuridine as well as traditional microscopic mitotic count method.[24]

Ki-67 is a nuclear non-histone protein. It acts as a proliferative marker during all stages of cell cycle except G0 phase because of its complete expression in tissue even in neo-adjuvant chemotherapy received cases. Its gene is situated in humans at long arm of chromosome 10.[25]

It has been shown that Ki-67, a proliferative marker, is overexpressed and amplified in ovarian malignancy. It is associated with prognosis and biological behaviour of ovarian tumor. [24,25]

Determination of proliferative activity of a tumor plays very important role in prognostication of a tumor. There are various methods available for determining proliferative activity of a tumor e.g. proliferative cell nuclear antigen, Bromodeoxyuridine as well as traditional microscopic mitotic count method.

CONCLUSIONS

Results from this study of Ki-67 expression in ovarian surface epithelial tumors show that-Histopathological study is the gold standard for basic tumor diagnosis, but evolution of ancillary techniques like Immunohistochemistry will help for better understanding of tumor behaviour, disease progression and its outcome. Detail study of demographic data, clinical presentation, detail histopathological examination for typing of ovarian surface epithelial tumors, as per WHO classification, has significant impact on patient management. Ki-67 a cell proliferative marker correlates well with tumor type and its clinical stage. So, it can be included with routine histopathological study of ovarian surface epithelial tumors, especially in cases of borderline tumor variety for defining their biological behaviour and further treatment guidelines. Majority of the surface ovarian malignancies in this study were diagnosed in post-menopausal women. Most patients presented with non-specific and vague symptoms over a period of time. Patient's education and awareness among primary care physicians with timely attention and further investigations, can help in early detection of ovarian malignancies in this subset of cases, thereby reducing morbidity and mortality associated with diagnosis in advanced stages.

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