Research Article

Spectrum of epithelial ovarian tumors with HER2/neu expression by the carcinomas among patients admitted in a tertiary care hospital in Eastern India

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Abstract

Background: Women in India constitute a neglected part of the society and their sufferings are least highlighted. Ovarian cancer is a fatal disease that affects them silently. Its incidence is rising with progressively enlarging elderly population. Early diagnosis is challenging, since majority of the cases of ovarian carcinomas are detected in advanced stage with nonspecific symptoms. Neo-adjuvant chemotherapy, a useful therapeutic modality, is limited by toxicity and resistance. Hence, targeted therapy is now being proposed to overcome these hurdles.

Objectives: 1. To study the histopathological spectrum of epithelial ovarian tumors. 2. To study the expression of HER2/neu among carcinomas on immunohistochemistry.

Materials and Methods: It was an observational, descriptive, cross-sectional study carried out among 68 patients admitted in the hospital, who were newly diagnosed as having primary epithelial ovarian tumor. Postoperatively, the surgical specimens were processed accordingly. Histopathological sections were examined under Hematoxylin and Eosin stain. Histopathologically confirmed carcinomas were analyzed for HER2/neu expression status on immunohistochemistry. Statistical analysis was carried out using *SPSS software, version 20*.

Results: Of the total cancer cases, 64.71% were benign, 5.88% borderline, 29.41% malignant. Serous type constituted majority for both benign and malignant variants (59.09% and 70% respectively). Overall, serous type constituted 58.82% of total population studied. Risk of malignancy index score was >200 for 95% of carcinomas in sharp contrast to benign tumors (15.91%). Grade 3+ HER2/neu expression was found in 35% of malignant cases, of which 85.71% were at advanced stage (FIGO III and IV).

Conclusion: Serous type was the most common type of epithelial ovarian tumor. Risk of malignancy index score had strongly correlated with the malignant nature of tumor ($p < 0.001$). Grade 3+ HER2/neu expression was associated with advanced stage carcinomas.

KEY WORDS: HER2/neu, ovarian tumor, risk of malignancy index score

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Introduction

Ovarian cancer, traditionally known as the silent killer, is a dreaded disease because of its vague, nonspecific symptoms and late presentation. Worldwide, it is the seventh most common cause of cancer and eighth most common cause of cancer-related death among women.[1] The annual age standardized incidence rate varies widely across the globe, ranging from >11/100,000 women in mid- and eastern

Europe to <5/100,000 women in Africa.[1] Incidence rates are 11.7/100,000 women in the UK, 8.0/100,000 women in the USA, 5.2/100,000 women in Brazil, and 4.1/100,000 women in China.[1] It is considered to be a disease of developed and industrialized countries where parity of women is low. However, striking international difference has been described in Japan where despite industrialization and low parity incidence of ovarian cancer is lower than that of the Western countries.[2] Literature says that about 80–90% of all ovarian cancers are epithelial in origin.[3–5] Other than invasive epithelial ovarian tumors, there exist a group of benign epithelial ovarian tumors and another group of low malignant potential, referred to as borderline epithelial tumors. In contrast to invasive tumors, these borderline tumors lack the invasive pattern of growth. But they have the potential to metastasize and recur. The prognosis largely depends on the extent of disease at the time of diagnosis that is usually advanced. Incidence of ovarian cancer in India is lower than that of the Western countries and it affects post-menopausal women in their sixties.[6,7] But it is becoming a threat to life due to changes in life style habit, rise in age at marriage, delay in first child birth, reduction in parity. Though benign ovarian tumors can be managed by simple cystectomy/hysterectomy, carcinomas require exploratory laparotomy with chemotherapy. Borderline tumors have the tendency to recur and metastasize. Hence, appropriate operative planning might save the patient from a second surgery. Risk of malignancy index score (RMI) can predict the clinical behavior of the tumor.[8] RMI score is derived from USG findings, patient's menopausal status, and CA125 level. If RMI score is >200, risk of malignancy is high, and if the score is <200 then risk is low. Two treatment options, available for ovarian cancers in advanced stage, are either a primary surgical cytoreduction or chemotherapy in an attempt to downstage the tumor followed by surgery. Though platinum-based therapy has produced an impressive result, chemoresistance and toxicity are creating hurdles. To overcome this, monoclonal antibodies targeting HER2/neu, a transmembrane protein homologous to epidermal growth factor receptor, has been introduced recently that might bring a new era of management.[9] But, HER2/neu overexpression in epithelial ovarian carcinomas has not been studied as extensively as it has been studied in case of breast carcinomas, especially in Indian scenario. Hence, this study was intended to find out the extent of HER2/neu expression in epithelial ovarian carcinomas.

Objectives:

1. To study the histopathological spectrum of epithelial ovarian tumors.
2. To study the expression of HER2/neu among carcinomas on immunohistochemistry. With these in mind the study was started.

Materials and Methods

It was an observational, descriptive, cross-sectional study carried out among 68 patients admitted in the hospital, who were newly diagnosed as having primary epithelial ovarian tumor. After approval from institutional ethical board, the study was proceeded for a duration of 1 year from April 2013 to March 2014. Newly diagnosed cases of primary epithelial ovarian tumors were included in the study. Patients with malignancy in other organ systems, with recurrent ovarian tumors, with non-epithelial tumors of ovary, and with metastatic tumors to ovary were excluded from the study. All the cases admitted in this hospital from April 2013 to March 2014 that fulfilled requirement criteria were included in this study. Patients were thoroughly assessed clinically. After the operative procedure, the surgical specimens were processed to prepare formalin-fixed, paraffin-embedded blocks. All specimens were examined by a single pathologist and diagnosis was made following WHO classification of ovarian tumors. The difficult cases were discussed with supervisor and definitive diagnosis was made. Histological typing, histological grading, FIGO staging (1988 classification) were carried out.

RMI scoring was done following RMI 2 recommendations for each case.[8] USG features contain multilocular cyst, solid areas, bilateral lesions, ascites, intra-abdominal metastasis; USG score were 0 = none, 1 = one abnormality, 4 = ≥two abnormalities; premenopausal score = 1, postmenopausal score = 4; CA125 level: value in U/mL; and final RMI score = USG score X menopausal score X CA125 value.

HER2/neu Expression Determination

In this study, HER2/neu expression was evaluated using anti-mouse monoclonal antibody (CELL MARQUE, CB 11, 1 mL prediluted, pH 7.3–7.7, protein base and preserved with sodium azide, Cat. no. 237M-17, 6600 Sierra College Blvd. Rocklin, CA 95677, USA, 800-665-7284; Key Code CMC23721030) following heat retrieval method of antigen retrieval. HER2/neu expression was scored on a scale of 0 to 3+, where 0 = no membrane staining, 1+ = complete membrane staining in <10% of the expressing cells, 2+ = faint complete membrane staining in >10% of the expressing cells, 3+ = strong complete membrane staining in >10% of expressing cells.[10] Ovary with normal histology was taken as negative control to emphasize the expression by carcinoma cells. For positive control, known case of HER2/neu positive breast carcinoma block was taken.

Statistics

At the end of the study, all the data were compiled and analyzed using *SPSS software, version 20* (Chicago, IL).

Results

Patient characteristics and histopathological findings are depicted in Table 1. Overall the mean age of presentation was 41.03 years, median was 40 years. For benign tumors, mean and median ages were 38.39 and 35 years; for carcinomas, these values were 46.3 and 46 years, respectively. The tumors were more prevalent in patients with lower

Table 1: Patient demography and tumor characteristics (*N* = 68)

Parameter	Benign (*n* = 44)	Borderline (*n* = 4)	Malignant (*n* = 20)
Age (years)			
Mean	38.39	43.75	46.3
Median	35	41	46
Most common chief complaint	Pain abdomen (47.73%)	Pain abdomen (50%)	>1 complaint (40%)
Menarche			
12 years	30 (68.18%)	2 (50%)	9 (45%)
13 years	14 (31.82%)	2 (50%)	9 (45%)
14 years	0 (0%)	0 (0%)	2 (10%)
Gross appearance of tumor			
Solid	0 (0%)	0 (0%)	5 (25%)
Cystic	41 (93.18%)	3 (75%)	2 (10%)
Both	3 (6.82%)	1 (25%)	13 (65%)
Tumor pathology			
Serous	26 (59.09%)	0 (0%)	14 (70%)
Mucinous	17 (38.64%)	4 (100%)	4 (20%)
Brenner	1 (2.27%)	0 (0%)	
Endometrioid	0 (0%)	0 (0%)	1 (5%)
Transitional cell carcinoma			1 (5%)

Table 1 shows the distribution of total population according to clinicopathological aspects.

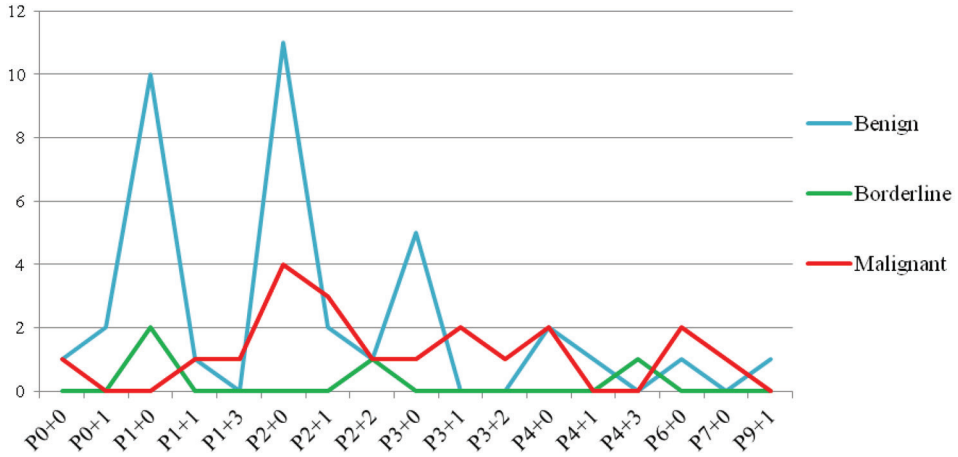

Figure 1: Line diagram shows trends of different types of tumors in relation to parity in total population (*N* = 68), with parity plotted in *X*-axis and number of cases in *Y*-axis. It clearly demonstrated that number of cases decreased with increase in parity. [Red: malignant (*n* = 20), green: borderline (*n* = 4), blue: benign (*n* = 44)].

parity. The number of cases decreased as parity increased (Figure 1). We analyzed RMI score for each case and tried to find out relation with the tumor types, if any (Table 2). RMI was <200 in 84.09% of benign tumors, whereas in case of carcinomas 95% had RMI >200. For borderline tumors only 25% cases had RMI >200. The RMI score was not normally distributed in the study population as elicited through normalcy test, Kolmogorov-Smirnov Test (*p*-value <0.001). Of the total cases studied, 64.71% cases were benign, 5.88% were borderline, 29.41% cases were malignant. Serous type of tumor was more prevalent overall (58.82%), among which 65% were benign and 35% were malignant. Among the benign serous tumors, most common type was serous cystadenoma (73.08%), followed by cystadenofibroma (19.23%) and papillary cystadenoma (7.69%). Next most common type was mucinous type (36.76%) of which 68% were benign, 16% were borderline, 16% were malignant. Of the total, 60% of the carcinomas were in advanced stage (FIGO III & IV). Among the serous carcinomas, 71.43% were in advanced stage, in contrast to mucinous carcinomas where only 25% were in advanced stage. Majority of the serous carcinomas were of high grade (92.86%), in contrast to mucinous carcinomas (25%). We found that high grade serous carcinomas were mostly of advanced stage (71.43%). We studied the carcinomas for HER2/neu expression and found that 35% had 3+ expression. In this study, we found only serous carcinomas were expressing HER2/neu. Among the advanced stage carcinomas, 50% had 3+ expressions (Table 3).

Table 2: Distribution of RMI score in benign, borderline and malignant group of tumors (*N* = 68).

Parameter	Group of tumors	Total patients (*N* = 68)	Mean	Standard deviation	Standard error mean	*p*-Value
RMI score	Benign	44	91.863636	219.5815885	33.1031700	
	Borderline and malignant	24	4842.320000	5558.8854995	1134.7027510	.000 (<.001)

Table 2 depicts that RMI score was not normally distributed in the study population. This distribution was statistically significant with *p*-value 0.000 (<0.001).

Table 3: Features of carcinomas (*n* = 20)

Parameters	Serous (*n* = 14)	Mucinous (*n* = 4)	Endometrioid (*n* = 1)	TCC* (*n* = 1)
HER2/neu expression				
0	3 (21.43%)	4 (100%)	1 (100%)	1 (100%)
1+	3 (21.43%)	0 (0%)	0 (0%)	0 (0%)
2+	1 (7.14%)	0 (0%)	0 (0%)	0 (0%)
3+	7 (50%)	0 (0%)	0 (0%)	0 (0%)
FIGO staging				
I	3 (21.43%)	3 (75%)	0 (0%)	0 (0%)
II	1 (7.14%)	0 (0%)	0 (0%)	1 (100%)
III	9 (64.29%)	0 (0%)	1 (100%)	0 (0%)
IV	1 (7.14%)	1 (25%)	0 (0%)	0 (0%)
Grading of serous				
High	13 (92.86%)			
Low	1 (7.14%)			
Grading of others				
I		3 (75%)	1 (100%)	1 (100%)
II		0 (0%)	0 (0%)	0 (0%)
III		1 (25%)	0 (0%)	0 (0%)

*TCC, Transitional cell carcinoma.

Table 3 shows the distribution of carcinomas according to HER2/neu expression status, FIGO staging and grading. Only serous carcinomas expressed HER2/neu ranging from 1+ to 3+. They were mostly of advanced stage and high grade compared to other carcinomas.

Discussion

In this study, the spectrum of epithelial ovarian tumor was very wide. The malignant tumors created a terrible situation by majority being of high grade and advanced stage though benign tumors outnumbered borderline and malignant tumors. Overall, serous tumor was the most common epithelial ovarian tumor. Most of the carcinomas were of advanced stage (60%). Among the high-grade serous carcinomas, stage IIIA was the most prevalent stage at presentation (46.15%). Some other studies have also described similar results. Kawamoto et al.[11] describes 60% of the epithelial ovarian tumors as benign, 35% as malignant, 5% as low-malignant-potential (borderline) type. Marinas et al.[12] shows that 57.7% of the serous tumors as benign, 7.7% as borderline, and 34.6% as malignant. The study by Phukan et al.[13] shows 59.1% carcinomas and that by Maxim et al.[14] shows 53.9% carcinomas at advanced stage (III and IV) at the time of presentation.

Like other authors,[8,15–18] we also found statistically significant correlation between RMI score and the histopathological nature of the tumor ($p < 0.001$). Therefore it emphasizes the role of RMI score in accurate preoperative assessment of epithelial ovarian tumors.

In this study, we found that 55% of the carcinomas were expressing HER2/neu ranging from grade 1+ to grade 3+. Here, 35% carcinomas had 3+ expression, 5% had 2+ expression, and 15% had 1+ expression. Our data revealed that HER2/neu expression was more intense for high grade, advanced stage serous carcinomas, similar to the results shown by Lanitis et al.[19] and Berchuck et al.[20] Literature shows varying degrees of expression of HER2/neu by the carcinomas.[21,22] Considerable methodological difference, antibody type used, varied scoring criteria might be contributing to it. Results using anti-mouse monoclonal antibody differed from those using polyclonal antibody. Standardization might overcome this difficulty. In this study, none of the mucinous carcinoma cases were HER2/neu expressing. It resembled those in the literature.[23,24]

Strengths and limitations of the study

The tertiary care hospital setup having supportive infrastructure was our main strength that enabled us to study wide varieties of epithelial ovarian tumor patients from various socio-economic background, having diverse mode of presentation. We were able to do the necessary investigations in every case. But we were severely constrained by the limited study duration that led us to have small population size. Further, we were unable to have follow-up results. Limited financial support restricted us from assessing HER2/neu expression in all the cases. We had analyzed only the carcinomas for HER2/neu expression.

Conclusion

Epithelial ovarian carcinomas are a disease of older post-menopausal patients. Risk of malignancy index score can predict the nature reliably. HER2/neu expression by epithelial ovarian carcinomas increases with rise in stage and grade of tumor. Our study results confirm the literature that HER2/neu overexpression is really present in epithelial ovarian carcinomas. Only serous epithelial carcinomas have expressed it consistently. But, a multicentric, large population-based study with facility of follow-up will be needed so that the dependency of anti-HER2/neu therapy in epithelial ovarian carcinoma and its prognostic implication gets more significance.

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