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Clinico-pathological study of breast carcinoma with correlation to hormone receptor status & HER2/neu

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Abstract

Carcinoma of the breast is the most common malignant tumor and the most common cause of death from carcinoma in females all over the world. In recent years, interest in prognostic factors has been stimulated by the success of systemic adjuvant therapy for early stage of breast carcinoma. The important pathological prognostic factors in invasive breast carcinoma include patient's age, tumor size, lymph node metastasis, nuclear grade, histological grade, histological type, hormone receptor status & HER2/neu. Tumors that express both Estrogen Receptor(ER) and Progesterone Receptor(PR) have the greatest benefit from hormonal therapy. HER2/neu has its role as a prognostic indicator and as a predictor of response to Trastuzumab (Herceptin) therapy in breast cancer patients. The study was conducted in Department of Pathology, Kilpauk Medical College, Chennai from July 2008 to August 2011. 73 cases of mastectomy specimens were received and clinical staging, histological grading and NPI score were analysed for these cases. 55 cases were selected at random and ER, PR & HER2/neu status was analysed. Greater than 80% of the cases were 40 years and above and majority (56.2%) were postmenopausal. Maximum number of cases (65.8%) were stage 2 and grade 2(72.6%).

ER was positive in 30.9%, PR in 25.5%, HER2/neu in 30.9%. The prevalence of hormone receptor positivity was less & HER2/neu positivity was higher which was in concordance with the studies conducted in Asian population compared with western world. There was a statistically significant association between the two hormone receptor expressions but not with other prognostic factors.

Keywords: Breast carcinoma, Clinical staging, Histological grading, Hormone receptor status, HER2/neu

Introduction

Carcinoma of the breast is the most common malignant tumor and the most common cause of death from carcinoma in females all over the world. In our country incidence of breast cancer is second only to that of cancer cervix[1]. Commonest neoplasm of breast is the tumor arising from epithelial component of glandular element of breast. Infiltrating ductal carcinoma being the most common type of carcinoma (70%), Lobular carcinoma is the second most common followed by smaller groups such as Medullary, Mucinous, Comedo carcinoma, Papillary, Tubular and Inflammatory carcinoma. In recent years, interest in prognostic factors has been stimulated by the success of systemic adjuvant therapy for early stage of breast carcinoma. The important pathological prognostic factors in invasive breast carcinoma include patient's age, tumor size, lymph node metastasis, nuclear grade, histological grade, histological type, hormone receptor status & Her2/neu. A single parameter with the strongest prognostic significance is hormone receptor status[2]. Intracellular steroid-hormone receptor proteins, primarily estrogen receptor (ER) and progesterone receptor (PR), have received intensive study both as indicator of prognosis and as guide to hormone therapy[3]. Estrogen plays a central role in the growth and differentiation of normal breast epithelium, stimulating cell proliferation and regulating the expression of genes, including the progesterone receptor[4]. Hormone receptor status in breast cancer helps to determine a patient suitability for hormone therapy. Tumors that express both ER and PR have the greatest benefit from hormonal therapy, and those containing only ER or only PR still have significant response[5]. HER2/neu is an oncogene. The amplification and overexpression is seen with breast cancer. Recently, importance of HER2/neu in breast cancer has been a topic of considerable interest, both in its role as a prognostic indicator and as a predictor of response to therapy. With the advent of the drug Trastuzumab(Herceptin), a humanised monoclonal antibody directed against cells that express HER2/neu, assessment of HER2/neu status in patients with metastatic breast carcinoma has become an even more important clinical consideration[6]. These receptors are measured by immunohistochemical methods in formalin fixed paraffin embedded tissue. In summary, Hormone receptors and HER2/neu expression appear to hold the key to the understanding of breast cancer and ER, PR & HER2/neu status are important biomarkers that helps physicians to individualize therapy.

Aims and Objectives

- To study the clinico-pathological parameters of breast carcinoma.
- To grade the breast tumors based on Nottinghams modification of Bloom & Richardson grading system.

- To assess the hormone receptor status & HER2/neu by immunohistochemistry.
- To correlate clinico-pathological parameters with hormone receptor status & HER2/neu.

Material and Methods

The tissue material for the study was obtained from the patients admitted in Kilpauk Medical College Hospital, from the Department of Surgery between July 2008 and August 2011. A detailed history regarding age, parity, socio economic status, family history and menstrual history were reviewed in all cases.

Inclusion criteria: All female patients who underwent mastectomy irrespective of age and proved to be malignant histologically were included for study.

Exclusion criteria: Excision and incision biopsies, proven to be malignant histologically, were not included in the study.

73 mastectomy specimens were included in the study. Of the 73 cases, ER, PR, HER2/neu study was done for 55 cases. All the mastectomy specimens received were properly sliced and fixed in 10% formalin for 18 - 24 hours. Detailed gross examination pertaining to overall size of the specimen, nipple and areola, tumor size, margin status and nodal status were carefully studied. The tissue was processed for routine histopathological examination and stained with Hematoxylin and eosin. All H&E stained tissue sections were first classified according to WHO and Histological grading was done by modified Bloom and Richardson scoring system. Suitable blocks were chosen for Immunohistochemistry which were cut in microtome using disposable blades. Slides coated with chrome alum were used. Sections were subjected to antigen retrieval by pressure cooker using citrate antigen retrieval solution (pH 6). Peroxidase-antiperoxidase method was used. The slides were stained with ER, PR & HER2/neu antibodies. Scoring for ER, PR was done by Quick Score System which takes the proportion and intensity of nuclear staining into account. Scoring for HER2/neu was done by denoting the membranous reactivity of the tumor cells. Data was compiled and associations were analysed by Chi square test, Logistic regression test using SPSS software.

Observation and Results

The patients in the present study were all females aged between 2nd and 8th decade. The youngest patient was 28 years old and the oldest patient was 80 years old. Maximum number of cases were seen in 41-50 years age group. Mean age was 50.18 years. 43.8% of the cases were premenopausal and 56.2% of cases were postmenopausal. The maximum number of cases were in clinical stage 2, histopathological grade 2 and lymph node stage 1(Table 1).

Table 1: Clinico-pathological parameters of breast carcinomas

Stage/grade	1	2	3	4
Clinical stage	6 (8.2%)	48 (65.8%)	19 (26%)	0 (0%)
Histological grade	7 (9.6%)	53 (72.6%)	13 (17.8%)	-
Lymph node stage	30 (41.1%)	25 (34.2%)	18 (24.7%)	-

The majority of the cases 46(63%) were having moderate prognosis, 5(6.9%) had good prognosis and 22(30.1%) had poor prognosis by NPI scoring. 17 cases(30.9%) were ER positive. 14(25.5%) were PR positive. 17(30.9%) were HER2/neu positive.

There is a statistically significant association between the two hormone receptor expressions with the p value of 0.002 but not with histological grading, tumor size, lymph node stage, clinical stage and NPI score. (Table 2)

Fig. 1: 50/F, hard palpable mass in right breast involving all quadrants and skin

Fig. 2: Modified radical mastectomy specimen showing a growth measuring 5x4cm

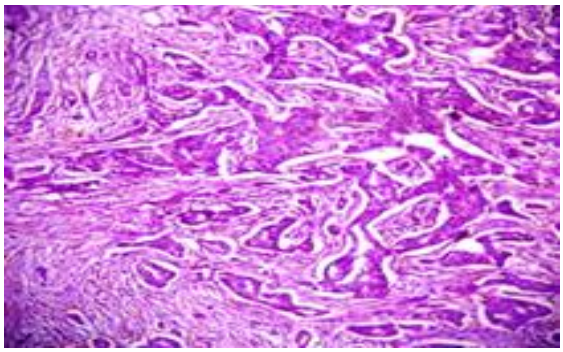

Fig. 3: IDC-NOS, Grade2, low power

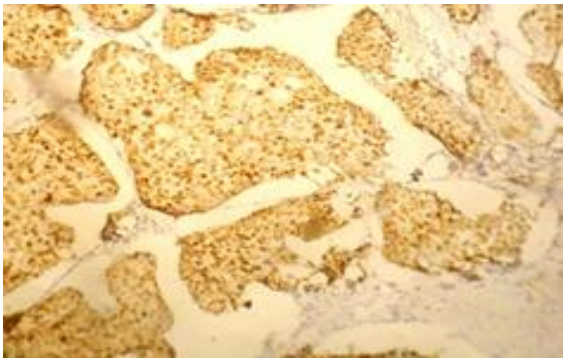

Fig. 5: IHC, PR postivity-score 8, low power

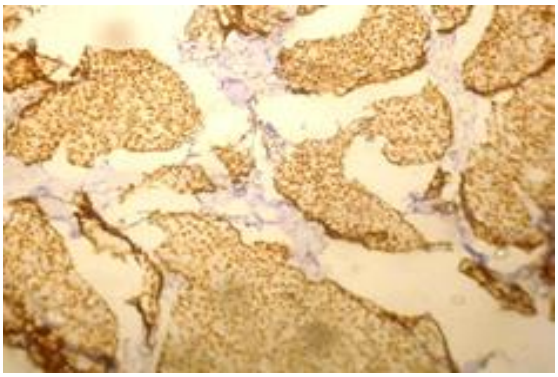

Fig. 4: IHC, ER Positivity-score 8, low power

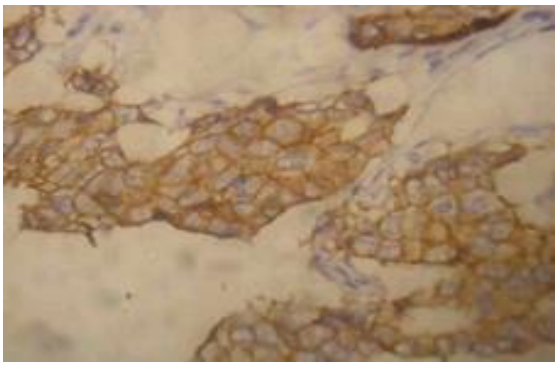

Fig. 6: IHC, HER2/neu Strong positivity, high power

Table 2: Correlation of clinico-pathological parameters with hormone receptor and her2/neu status

		No. of cases	ER + Cases	PR+ Cases	HER2/neu + Cases
Grade	1	7	3(42.8%)	3(42.8%)	1(14.2%)
	2	40	11(27.5%)	8(20%)	13(32.5%)
	3	8	3(37.5%)	3(37.5%)	3(37.5%)
Tumor size	1-2 cm	11	4(7.3%)	4(7.3%)	3(5.5%)
	2-5 cm	36	13(23.6%)	10(18.2%)	13(23.6%)
	>5 cm	1	0(0%)	0(0%)	1(1.8%)
Lymph node stage	1	17	6(10.9%)	4(7.3%)	7(12.7%)
	2	15	7(12.7%)	5(9.1%)	3(5.5%)
	3	16	4(7.3%)	5(9.1%)	7(12.7%)
Clinical stage	1	5	2(40%)	2(40%)	2(40%)
	2	38	11(28.9%)	8(21%)	10(26.3%)
	3	12	4(33.3%)	4(33.3%)	5(41.6%)
	4	Nil	Nil	Nil	Nil
NPI Score	Good prognosis	6	3(5.5%)	1(1.8%)	2(3.6%)
	Moderate prognosis	28	9(16.4%)	10(18.2%)	9(16.4%)
	Poor prognosis	14	5(9.1%)	3(5.5%)	6(10.9%)

Table 3 shows multivariate analysis of all the factor using Logistic regression. It showed a statistically significant correlation with number of lymph nodes.

Table 3: Multivariate analysis

S. No	Factor	P-Value
1	Age	0.27
2	Menopausal state	0.95
3	Tumor size	0.06
4	Number of lymph nodes	0.02
5	Grade	0.96
6	ER positive	0.23
7	PR positive	0.09
8	HER2/neu positive	0.294

Table 4: Comparative study of the ER, PR & HER2/neu positivity

Sl. no	Study	No. of cases	ER Positive	PR Positive	HER2/neu positive
1	Priti Lal et al at New York[11]	3655	71.6%	47.4%	26.89%
2	Sughayer et al at Jordan[12].	267	50.8%	57.5%	17.5%
3	Onitilo et al at Weston[13].	1134	77.9%	59.1%	17.7%
4	Ljiljana Hulpic et al in Croatia[15].	242	37.5%	40.6%,	18.7%
5	Azizun Nisa et al at Karachi[16].	150	32.7%,	25.3%,	24.7%
6	Desai SB et al in India[17].	798	32.6%	46.1%	
7	Dutta V et al at Army Hospital and Research centre in New Delhi[9].	75	24%	30%	57.2%
8	Present study	55	30.9%	25.5%	30.9%

Discussion

Lesions of the breast constitute a very important chunk of the causes of morbidity and mortality in women worldwide. Prognosis is related to clinical parameters, histological type, grade and lymph node metastasis. Estrogen, Progesterone receptors & HER2/neu receptors, have with increasing importance, influenced the management of this malignancy. Recently there has been increasing interest in the early detection of the malignant breast lesions as early therapeutic intervention improves survival.

Age distribution: The mean age of patients included in our study was 50.18years and maximum number of cases were in the age group of 41-50years. This is less than the observation made by Rhodes. D.J, who found more than 75% of the cases were above 50years and the mean age was 64 years[7]. But usually in Asian countries breast carcinoma occurs a decade earlier. Our results are in concordance with the study conducted by Lakmini. K.B. Mudduwa in which mean age was 52.5 years and 85.7% of the patients were more than 40years.[14]

Menstrual status: In our study 56.2% of the patients were postmenopausal women. This is in concordance with the study conducted by Louis. W.C. Chow et al, in which 52% of the women were postmenopausal[8] and Dutta V et al, in which 59% of the cases were postmenopausal women[9].

Hormone receptor & HER-2/neu receptor status in breast carcinomas: Hormone receptor status is an important prognostic factor in breast cancer and help to determine a patient's suitability for endocrine therapy. Breast cancer patients that are estrogen receptor positive and/ or progesterone receptor positive have lower rate of mortality after their diagnosis as compared to the women with estrogen receptor/progesterone receptor negative disease (Lisa K Dunnwald et al. 2007)[10]. In a study conducted by Priti Lal et al at New York with 3655 breast carcinomas, ER was positive in 71.6%, PR in 47.4% & HER2/neu in 26.89%[11]. Sughayer et al found in Jordan with 267 breast cancers, that ER was positive in 50.8%, PR was positive in 57.5%, & HER2 neu was positive in 17.5% of the cases[12]. Onitilo et al from Weston conducted a study between 1998 and 2005 and found ER positivity in 77.9%, PR positivity in 59.1%, HER2 neu positivity in 17.7%[13]. These are some of the studies conducted in western population. According to Lakhmini. K.B. Mudduwa the prevelance of hormone receptor positive breast cancer in Asian countries is found to be lower

than western world where more than 50% tumors express hormone receptors[14]. However the number of studies performed on this topic is much less in the Asian communities compared with the western world. Ljiljana Hulpic et al conducted a study in Croatia with 242 cases and found ER positivity in 37.5%, PR positivity in 40.6%, HER2 neu positivity in 18.7% of the cases[15]. Azizun Nisa et al studied 150 cases in Karachi and found ER positivity in 32.7%, PR positivity in 25.3%, HER2 neu positivity in 24.7% of the cases respectively[16]. In a study conducted by Desai SB et al in India of 798 cases ER was positive in 32.6% of the tumors and PR was positive in 46.1% of the cases[17]. Dutta V et al conducted a study in Army Hospital and Research centre in New Delhi and found that out of 75 cases, 24% were ER positive and 30% were PR positive, 57.2% were HER2 neu positive[9].

The results of studies conducted in Asia are not in concordance with the studies in western population. The results of our study are in concordance with studies conducted in Asian population. The overall positivity rate for ER and PR is lower possibly because of the difference in techniques of evaluation[18] high tumour grades and majority being menopausal women in our study. Higher Her-2/neu immunoreactivity may be inherent in breast tumours in Indian women[9]. Nulliparity, late age at first birth, early age at menarche, higher body mass index and the use of hormone replacement therapy have all been associated with increased risk of developing an ER positive tumour. Young patients have high levels of circulating oestrogens and a correspondingly low expression of steroid receptors, which is reflected in their tumours. There appears to be a variation in steroid receptor positivity in the Asian population[9].

Correlation of hormone receptor and HER-2/neu receptor status positivity with other prognostic variables

In this study there is a statistically significant association between ER & PR status. Expression of Hormone receptor(ER, PR) decreases with increasing tumor size though there is no statistical significance, most probably due to smaller sample size. No significant association of ER, PR, and HER2/neu status with clinical staging and NPI score noted. Lakmini. K.B. Mudduwa has found a significant inverse relationship with the grade and ER, PR expression in his study. His study also shows no significant association of hormone receptor status with tumor size[14]. Ana Lucia Amaral Eisenberg et al in Brazil also has established a significant correlation between ER, PR status and histological grade[19]. Ljiljana Hlupic has found no statistically significant association between ER, PR status and NPI score in concordance with our study but in contrast to our study there is a significant association between NPI index and HER-2/neu status[15]. Kenneth McCarty et al and Rosemary. R. Millis have obtained association between ER, PR status and histological grade but no association with other prognostic variables[20,21]. Ur Rahman E in Pakistan has obtained no correlation of ER, PR & HER2/neu status with clinical parameters[22].

Summary and Conclusion

73 cases of mastectomy specimens were received and clinical staging, histological grading and NPI score were analysed for these cases. 55 cases were selected at random for ER, PR & HER2/neu status was analysed. Greater than 80% of the cases were 40 years and above and majority (56.2%) were postmenopausal. Maximum number of cases (65.8%) were stage 2 and grade 2(72.6%). ER was positive in 30.9%, PR in 25.5%, HER2/neu in 30.9%. The prevalence of hormone receptor positivity was less & HER2/neu positivity was higher which was in concordance with the studies conducted in Asian population compared with western world. There was a statistically significant association between the two hormone receptor expressions but not with other prognostic factors. Presence of hormone receptors correlates well with response to hormone therapy. There is a significant decrease in mortality and tumor recurrences with hormone therapy. HER-2/neu has significant prognostic value as a marker since it can predict resistance to hormonal therapy & response to Trastuzumab (Herceptin). So, determination of ER, PR & HER2/neu status is essential in all cases irrespective of clinical staging and lymph node metastasis.

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