ORIGINAL ARTICLE

AN ANALYSIS OF CHANGING TRENDS IN THE EPIDEMIOLOGICAL, PATHOLOGICAL AND RECEPTOR EXPRESSION OF BREAST CANCERS TREATED IN A TERTIARY CARE HOSPITAL IN SOUTH INDIA

Vidhya Lakshmi S[1], Seyed Rabiya[2]

ABSTRACT: Carcinoma breast is the commonest malignancy of females all over the world and second leading cause of death due to cancer among females. The incidence of breast carcinoma in India is on the rise. The prognosis of tumors depends upon the size of the tumour at presentation, histological type, grade, marginal clearance and lymph node status. The prevalence of hormone receptor-positive breast cancer in Asian countries has been found to be lower than the western world where more than 50% tumours express hormone receptors. The study aims at determining the incidence and prevalence of hormonal receptors and Her-2-neu in various subtypes of breast carcinoma and the distribution of expression among the epidemiological subgroups of patients of different age groups, parity, reproductive status etc. All cases of breast cancers attending the oncology OPD in a tertiary care hospital in South India during a 2 year period (2012-2013) were taken up for our study. The criteria of selection of patients included availability of histopathological diagnosis, hormone receptor studies, Her-2-neu expression status and follow up till the study period. Breast carcinomas tend to occur more frequently in older and post-menopausal females. However the incidence of breast carcinoma in females below 40 is on the rise. Greater awareness regarding self-examination of breast, screening tests like mammogram and ultrasound and fine needle aspiration cytology should be advocated for an early diagnosis and better prognosis. Risk factors of developing carcinoma breast include obesity, diabetes, estrogen exposure and nulliparity. Most tumors belong to Grade III, probably because patients present to the hospital a late stage of cancer. The prevalence of estrogen receptor positivity is 43.3%, progesterone receptor is 30% and Her 2 neu over expression is 56.6%. Hormone receptor status depends on size, histological type and grade of the tumour. In our study we found that there is no significant association between hormone receptor status and age, menopausal status or number of axillary lymph nodes showing tumor deposits.

KEYWORDS: Breast cancers, tumour grading, hormone receptors.

INTRODUCTION: Carcinoma breast is the commonest malignancy of females all over the world and second leading cause of death due to cancer among females. The incidence of breast carcinoma in India is on the rise. The risk factors of breast carcinoma include long estrogen exposure, nulliparity, obesity and family history of breast cancer. The incidence of breast carcinoma differs in various age groups of patients. Most cancers are diagnosed when patients find a lump during self-examination of their breasts. Mammograms are done as a screening test in many centers and the finding of micro calcifications in a breast lump alerts the treating physician that there is a high index of suspicion of malignancy.

ORIGINAL ARTICLE

Fine needle aspiration of the tumour can be done as an out-patient procedure to confirm malignancy. Neo adjuvant chemotherapies before surgeries have improved the over-all prognosis of operable breast cancers. The surgeries for a malignant breast swelling range from lumpectomy to modified radical mastectomy. Nevertheless, breast conservation surgery followed by chemotherapy and hormonal therapy produce almost equal results as radical surgery, thereby ensuring better quality of life after surgery.

The prognosis of tumors depends upon the size of the tumour at presentation, histological type, grade, marginal clearance and lymph node status. Immunohistochemical evaluation of estrogen receptor (ER) and progesterone receptor (PR) status of breast carcinoma has become a routine investigation to predict the response to adjuvant endocrine therapy. The prevalence of hormone receptor-positive breast cancer in Asian countries has been found to be lower than the western world where more than 50% tumours express hormone receptors.

HER-2/neu is the human homologue of the neu gene, called HER-2 or c-erbB-2, which shares extensive homology with epidermal growth factor receptor (EGFR). It is considered one of the poor prognostic factors in breast cancer. The introduction of Herceptin {Trastuzumab}, which is a monoclonal antibody which binds to Her 2 neu receptor has brought significant changes in the prognosis and survival of breast cancer patients.

The study aims at determining the incidence and prevalence of hormonal receptors and Her-2-neu in various subtypes of breast carcinoma and the distribution of expression among the epidemiological subgroups of patients of different age groups, parity, reproductive status etc. The hormonal status has definite prognostic implications and so enables the clinicians to initiate various ancillary modalities of therapy.

AIMS AND OBJECTIVES:
To identify the risk factors of breast Carcinomas:
1. To classify breast carcinomas based on histopathological features
2. To document the estrogen and progesterone receptor (ER & PR) status and Her-2-neu expression of breast cancers
3. To correlate the steroid receptor status of breast cancer with all relevant patient and tumour characteristics.

MATERIALS AND METHODS: All cases of breast cancers attending the oncology OPD in a tertiary care hospital in South India during a 2 year period (2012-2013) were taken up for our study. The criteria of selection of patients included availability of histopathological diagnosis, hormone receptor studies, Her-2-neu expression status and follow up till the study period. The clinical data regarding age, family history, reproductive status, exogenous hormone administration and parity were gathered from the hospital case records. Special care was taken to note the presence of Diabetes mellitus, hypertension, History of tobacco, alcohol and use of oral contraceptives. An approval from the Institutional Human Ethics Committee was obtained prior to our study.

The clinical presentation of the tumour, the size of the swelling, axillary lymph node status, the presence of sub cutaneous lymphatic involvement, ulceration and adherence to the chest wall etc were noted.

ORIGINAL ARTICLE

A detailed record of all pre-operative investigations like mammogram, ultrasound, Fine needle aspiration cytology and true cut biopsies was made. Some patients had received pre-operative neo adjuvant chemotherapy and the complete record of it was made too.

The details of surgery performed like radical mastectomies, breast conservation surgeries or toilet mastectomies and presence of any post-operative complications were noted.

The gross specimen reports were reviewed to note the size of the tumour, marginal clearance, involvement of the nipple and areola and presence of ulceration. The number of axillary lymph nodes dissected was recorded in each case.

The histopathological slides were reviewed and the tumour was classified into the following histological types namely infiltrating ductal carcinoma, lobular carcinoma, colloid or mucinous carcinoma, medullary carcinoma, tubular carcinoma and other rarer type of tumors like sarcomas.

The tumours were graded according to the Nottingham modification of Scarff Bloom Richardson (SBR) grading system into Grade I, II and III based on tubule formation, mitotic activity and nuclear pleomorphism.

The margin clearance status, necrosis and lymphovascular invasion were observed. The exact number of axillary nodes showing tumor deposits was recorded.

The paraffin blocks were taken up for immunohistochemical studies using the Avidin Biotin immunoassay method. The markers for Estrogen and Progesterone receptors and Her-2-neu over expression were used. Evaluation of the staining and assigning a score was performed. The staining was evaluated on the invasive component only. Best-preserved and best-stained areas of the sections were assessed. Nuclear staining was assessed for ER and PR while membrane staining was assessed for Her 2.

A score for the proportion of stained cells (0 = no nuclear staining, 1 = <1% nuclear staining, 2 = 1-10% nuclear staining, 3 = 11-33% nuclear staining, 4 = 34-66% nuclear staining and 5 = 67-100% nuclear staining) and the intensity of staining (0 = no staining, 1 = weak staining, 2 = moderate staining, 3 = strong staining) were assigned to each tumour. Tumors that showed strong complete membrane staining in >10% of the tumour cells were considered positive.

The observations in each of these were tabulated and critically analyzed. Graphs and charts were made to study the expression of the hormone receptors and Her-2-neu among different histological types, high & low grade tumors. These results were compared with other high risk epidemiological factors.

OBSERVATIONS AND RESULTS: 30 patients were included in our study. The age of the patients varied from 27 to 78. The median age was 54. The distribution of the tumour in various age groups is given below.

7 patients were pre-menopausal and 23 of them were post-menopausal. 2 patients gave a positive history of breast cancer in mother and maternal aunt respectively.

The average parity of the patients was 2.8. Only one patient was nulliparous and she had a grade III tumour with metastatic deposits in the liver. All the 30 patients denied use of tobacco or alcohol. 5 patients had been sterilized and none of them had taken oral contraceptives.

The tumours were classified based on size into 3 groups; < 5cm, 5 to 10 cm and > 30cm. There were 11, 13 and 6 cases in each of these sub groups respectively.

The size of the tumour did not correlate with the grade or receptor status.

ORIGINAL ARTICLE

There were 24 cases of infiltrating ductal carcinoma, 2 cases of invasive lobular carcinoma, 1 case of tubulolobular carcinoma, 2 cases of colloid/mucinous carcinoma and 1 case of ductal carcinoma in situ.

Grading of the tumours was done using the Nottingham modification of Scarff Bloom Richardson grading system and tumours were grouped in to 3 categories based on tubule formation, mitotic rates and nuclear pleomorphism. 5 tumours were graded as Grade I, 11 as grade II and 14 as grade III.

Grade of the tumour	Total cases	ER +	PR +	Her 2 neu +
I	5	5	3	4
II	11	4	2	7
III	14	4	4	6

The number of axillary nodes showing metastasis ranged from 0 to 8.

13 patients received neoadjuvant chemotherapy prior to surgery. 20 of them received chemotherapy after surgery also. The patients with estrogen receptor positive tumours received Tamoxifen.

The details of follow up of all these 30 patients were obtained from the hospital case files. 3 patients developed metastases in the liver within a period of 1 year. 1 patient had a cystic metastasis in the brain. 3 of these patients had a primary grade III tumour whereas one had grade II tumour.

3 patients developed a contra lateral tumour in the opposite breast. Interestingly, a patient with ductal carcinoma in situ had carcinomatous deposits in all 20 lymph nodes detected and in due course she developed invasive tumour in the opposite breast.

DISCUSSION: In India there is concern about the rising incidence of breast cancers in younger women. Premenopausal women comprised 50% of the cohort in an urban study conducted by Raina et al.[1] The median age at presentation in their study was 47 years. In our study only 7 patients were premenopausal. The median age of presentation in our study was 54. However 3 patients were below the age of 35. Primary breast carcinomas arising before the age of 40 are far more aggressive and the likelihood of metastasis is higher than those arising in older patients regardless of the hormone status. These issues raise the concern about the lifestyle, use of exogenous hormone administration and the need to implement screening mammograms at an early age.

As mammographic facilities are not widely available here and there is no nation-wide breast-screening program, the commonest mode of presentation remains a lump in the breast, 96% of the patients in the present series presented with a breast lump.

In our study we found that modified radical mastectomy was the most preferred surgery followed by lumpectomy. Raina et al[1] proposed that patient preference for mastectomy is an important reason for the under-utilization of breast conservation therapy. The advanced stage and lack of facility for radiotherapy provoke the treating physicians to opt for radical surgeries. The majority of patients included in the present study had high-grade tumors, which explains the low prevalence of hormone receptor expression. It is often observed that high-grade tumors tend to be negative for hormone receptor expression.

ORIGINAL ARTICLE

Thriveni et al[2] proposed that grade of the tumour was directly proportional to the number of axillary lymph nodes. But in our study, there was no significant association between the grade of the tumor and number of lymph nodes.

Estrogen receptor is a well-established predictive and prognostic factor in breast cancer. In the recent International consensus on the treatment of early breast cancer, the panel affirmed that the first consideration was endocrine responsiveness of the tumour and suggested categorization of the tumours into endocrine responsive, endocrine non-responsive and tumours of uncertain endocrine responsiveness. Therefore testing for estrogen and progesterone receptors status is critical to plan optimal treatment in breast cancers.

According to Desai et al[3] the receptor positivity rates are lower in Indian population than in Western countries. This is probably due to the high prevalence of high-grade tumors among the study population. The Nottingham grade and the mitotic count have a significant inverse correlation with the Quick Score of hormone receptor expression. They reported ER and PR positivity rates of 32.6% and 46.1% respectively. Additionally, not all patients in India undergo hormone testing due to financial constraints. In our study ER positivity was found in 43.3% of the cases and PR positivity in 30% of the cases.

Chlebowski et al[4] published a data indicating a different pattern of association for most factors with ER-positive disease than with ER-negative disease. For ER-negative cancers, odds ratios for age, family history, age at menopause, greater parity, smoking, and alcohol use were all close to 1.0. The association between breast cancer risk and chronologic age, race or ethnicity, and age at menopause differed statistically significantly across ER disease subtypes. Age at menarche was not statistically significantly associated with either ER-positive or ER-negative breast cancer.

Our study is in concordance with the above data.

PR⁻ breast cancers, even if they are ER⁺, are more aggressive than the PR⁺ phenotype and are more likely to be of high grade than PR⁺ tumours. ER and PR were inversely related to HER-2/neu over expression, whereas tumour grade was positively associated with HER-2/neu over expression according to the study undertaken by Huang et al.[5] In our study also, the PR⁻ tumours were all grade III and 2 of them showed muscle invasion and lymphovascular invasion. Unlike ER, there was only a low tendency for PR to be associated with smaller carcinomas and with older patients, but this tendency was not statistically significant.

Almasri et al[6] said that ER expressing breast carcinomas were, on average, 1.6 cm smaller than carcinomas lacking ER expression. Similarly, 57% of T2 tumors were ER positive as opposed to only 30% of T3 tumors. Our study did not show any significant relationship between ER expression and size of the tumour.

The presence of necrosis and lymphovascular invasion showed an inverse relationship with ER and PR reactivity according to Desai et al.[3] In our study, 2 tumors showed perineural/ lymphovascular invasion and both of these were negative for ER and PR but showed strong membrane positivity for Her 2 neu.

Swain et al[7] declared that whether contralateral breast cancer represents a second primary versus recurrent or metastatic disease clearly has implications for treatment. For example, if contralateral breast cancer represents a second, biologically distinct primary cancer, treatment with Tamoxifen may be beneficial for women with an ER-negative primary cancer who are at risk for

ORIGINAL ARTICLE

developing contralateral disease. In our project, only one of the tumours which metastasized to the opposite breast was ER positive and she was treated with Tamoxifen.

Naeem et al[8] reported that Her 2 neu over expression tends to decrease with age. But in our study there was no significant difference in Her 2 neu over expression among patients of different age groups.

Various studies have shown that Her 2 neu over expression is significantly associated with ER and PR negativity. In our study, Her 2 neu was positive in 13.3% cases with positive ER receptor status.

Almasri et al[6] observed that Her 2 neu over expression is directly proportional to the size of the tumour and the number of positive axillary lymph nodes. But in our study we found that there was no relation between the Her 2 neu expression status and the number of positive axillary lymph nodes. Also Her 2 neu over expression was more prevalent in grade II tumours.

In the present study, the overall grade of the tumour correlated well not only with the proportion of cells stained but also the intensity of staining, the two components of the Quick Score that justify the assessment of the intensity of staining in giving a score in an article that was published by Lakmini et al.[9]

Onody et al[10] suggested The HER-2/neu gene status may be useful not only as a prognostic factor in determining clinical outcome, but also as a predictive factor in projecting the response to adjuvant therapies. In our study, most of the Her 2 neu positive cases were given neo adjuvant chemotherapy and were disease free during our follow up period.

Dujaily et al[11] reported that most of the ductal carcinoma cases were purely invasive ductal carcinomas of nonspecific type (74.4%) that showed significant HER-2/neu over expression (65.6%). This provides more evidence of the hypothesis that aggressive tumors seem to show significant HER-2/neu over expression and demonstrates the association between the nature of the biological expression of HER-2/neu by the tumor and its degree of malignancy since it has been argued that nonspecific type ductal carcinomas are the most aggressive variants of breast cancer. The current study demonstrated that 58.3% of invasive ductal carcinoma cases exhibited HER-2/neu expression whereas 100% of invasive lobular carcinomas were Her2 neu positive.

The present study found no significant association between HER-2/neu over expression and age of the patients, which is in concordance with the publication of Dujaily et al.[11]

Li et al[12] proved that lobular and ductal/lobular tumours tended to be diagnosed at a more advanced stage and to be both >5.0 cm and node positive, but they were also much more likely to be hormone receptor positive. In contrast, come do and medullary carcinomas were less likely to have an advanced stage at diagnosis and to be node positive, but more likely to be hormone receptor negative and to have a high grade. Our study reproduced similar inferences.

Guarneri et al[13] proposed that the molecular classification of breast cancer represents the foundation of treatment selection for early and advanced disease. The more clinically useful classification recognizes three subtypes: HR positive (ER and/or PR ≥10%), HER-2 positive (IHC 3+, irrespective of HR expression), and triple-negative tumors (ER, PR, and HER-2 negative). Endocrine manipulation and anti–HER-2 agents represent the foundation of treatment in cases of HR-positive and HER-2–positive tumors, respectively, while, so far, chemotherapy is the only available option in triple-negative tumors. The practice of obtaining biopsies of metastatic lesions varies considerably across centers; therefore, the clinical management of the majority of patients is still based on the

ORIGINAL ARTICLE

initial assessment. In our study all the 3 tumours which metastasized to the liver were triple negative, whereas the tumour that produced a cystic metastasis in the brain was strongly positive for ER and PR.

Hawkins et al[14] suggested that tumour type, soluble protein content, type of surgery, tumour stage, type of systemic therapy and tumour size (clinical/pathological)] appeared to relate to risk of any recurrence. Eight of these factors also appeared to relate by univariate analysis to risk of distant metastases. And seven (ER concentration, node status, tumour grade, clinical and pathological tumour size, type of surgery and soluble protein content) appeared to relate to risk of death. For risk of local recurrence, however, only tumour grade and ER concentration appeared important. The tumors which metastasized to liver and brain in our study were all grade II/III, size above 5cm and triple negative (except one). But there was no difference in the type of therapy or surgery given to these patients. This was similar to the study done by Sambasivaiah et al [15] in 2007.

CONCLUSION: Breast carcinomas tend to occur more frequently in older and post-menopausal females. However the incidence of breast carcinoma in females below 40 is on the rise. Greater awareness regarding self-examination of breast, screening tests like mammogram and ultrasound and fine needle aspiration cytology should be advocated for an early diagnosis and better prognosis. Risk factors of developing carcinoma breast include obesity, diabetes, estrogen exposure and nulliparity:

- Infiltrating ductal carcinoma is the commonest histological type of breast carcinoma.
- Most tumors belong to Grade III, probably because patients present to the hospital a late stage of cancer.
- The prevalence of estrogen receptor positivity is 43.3%, progesterone receptor is 30% and Her 2 neu over expression is 56.6%
- Hormone receptor status depends on size, histological type and grade of the tumour. In our study we found that there is no significant association between hormone receptor status and age, menopausal status or number of axillary lymph nodes showing tumor deposits.
- Her 2 neu over expression status is inversely proportional to hormone receptor positivity. Poorly differentiated tumours, larger tumours and tumours with a more aggressive histological type express stronger Her 2 neu membrane staining.
- There is no significant association between Her 2 neu over expression and parameters like age, menopausal status and number of axillary lymph nodes.
- Widely metastasizing tumours are usually negative for hormone receptors and Her 2 neu genes.
- The introduction of neo adjuvant chemotherapy has brought about significant improvement in the prognosis of operable breast cancers.
- A regular follow up and better patient awareness will bring about drastic reduction in mortality due to breast carcinoma.
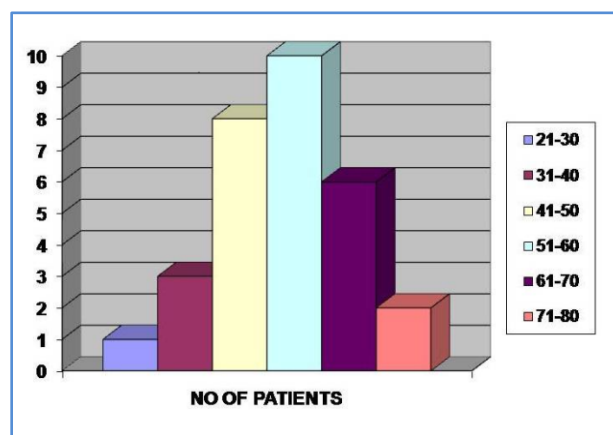
ORIGINAL ARTICLE

SUMMARY:

Clinical features (Figures in parenthesis show percentage)	
Age (Median)	54 years
Range	(27–78)
Grade	
I	5 (16.6)
II	11 (36.6)
III	14 (46.6)
Metastatic	4 (13.3)
Bilateral Breast Ca	3 (10.0)
Axillary Lymph node	
Positive	17 (56.6)
Negative	13 (43.4)
Estrogen Receptor	
Negative	13(43.3)
Positive	17 (56.6)
Progesterone Receptor	
Positive	9 (30.0)
Negative	21 (70.0)
Her 2 neu	
Positive	17(56.6)
Negative	13 (43.3)
Chemotherapy	
Neoadjuvant	13
Post-surgery	20

REFERENCES:

1. Raina V, Bhutani M, Bedi R, Sharma A, Deo SV, Shukla NK, Mohanti BK, Rath GK: Clinical features and prognostic factors of early breast cancer at a major cancer center in North India. Indian J Cancer 2005, 42:40-45.
2. Thriveni K, Vijayalakshmi Deshmane, P.P.Bapsy.Clinical utility of human epidermal growth factor receptor-2 neu detection in breast cancer patients. Indian J of Med Res: 125, 2007, pp 137-142.
3. Desai SB, Moonim MT, Gill AK, Punia RS, Naresh KN, Chinoy RF. Hormone receptor status of breast cancer in India: A study of 798 tumours. Breast 2000; 9: 267-70.
4. Rowan T. Chlebowski, Garnet L. Anderson, Dorothy S. Lane, Predicting Risk of Breast Cancer in Postmenopausal Women by Hormone Receptor Status. Oxford journal 2007:99; 1695-1705.
5. H J Huang, P Neven, M Drijkoningen. Association between tumour characteristics and HER-2/neu by immunohistochemistry in 1362 women with primary operable breast cancer. J Clin Pathol 2005; 58: 611-616.

ORIGINAL ARTICLE

6. Nidal M Almasri and Mohammad Al Hamad. Immunohistochemical evaluation of human epidermal growth factor receptor 2 and estrogen and progesterone receptors in breast carcinoma in Jordan. Breast cancer research 2005: 7; 598-694.

7. Sandra M. Swain, John W. Wilson, Eleftherios P. Mamounas. Estrogen Receptor Status of Primary Breast Cancer Is Predictive of Estrogen Receptor Status of Contralateral Breast Cancer, Oxford journal 2004: 96 (13); 1041.

8. Mohammad Naeem, Amir Nasir, Zahid Aman, Tariq Ahmad, Ambreen Samad. Frequency of her-2/neu receptor positivity and its association with other features of breast cancer. J Ayub Med Coll Abbottabad 2008; 20(3).

9. Mudduwa LK. Quick score of hormone receptor status of breast carcinoma: Correlation with the other clinicopathological prognostic parameters. Indian J Pathol Microbiol 2009;52:159-63

10. Peter Onody, BS, Françoise Bertrand, MD, Françoise Muzeau, BS, Ivan Bièche, PhD, and Rosette Lidereau, PhD. Fluorescence In Situ Hybridization and Immunohistochemical Assays for HER-2/neu Status Determination; Application to Node-Negative Breast Cancer. Archives of Pathology & Laboratory Medicine: Vol. 125, No. 6, pp. 746-750.

11. AL-Dujaily EA, Al-Janabi AA, Pierscionek T, Yasseen AA. High prevalence of HER-2/neu overexpression in female breast cancer among an Iraqi population exposed to depleted uranium. J Carcinog 2008;7.

12. C I Li, D J Uribe, and J R Daling Clinical characteristics of different histologic types of breast cancer. British Journal of cancer 2005. 93; 1046-56.

13. Valentina Guarneri, Simona Giovannelli, Guido Ficarra. Comparison of HER-2 and Hormone Receptor Expression in Primary Breast Cancers and Asynchronous Paired Metastases: Impact on Patient Management. The Oncologist 2008. Vol. 13, No. 8, 838-844.

14. RA Hawkins1, AL Tesdalel, ME Killen. Prospective evaluation of prognostic factors in operable breast cancer. Br J Cancer. 1996 November; 74(9): 1469–1478.

15. Sambasivaiah Kuraparthy, Kumaraswamy M Reddy, Lakshmi Amancharla Yadagiri. Epidemiology and patterns of care for invasive breast carcinoma at a community hospital in Southern India. World Journal of Surgical Oncology 2007, 5:56.
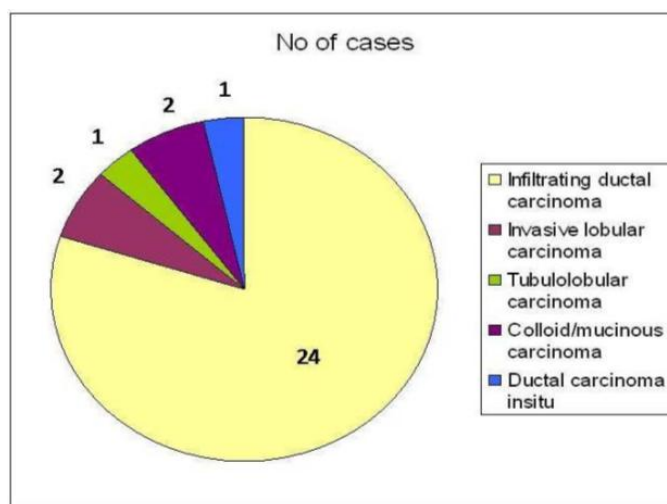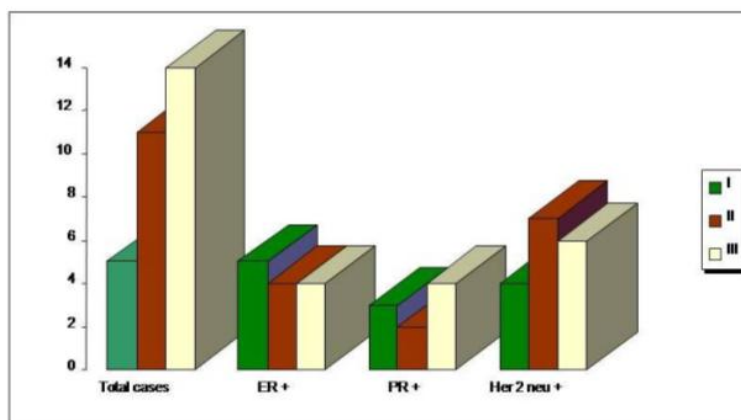
Slide 1

ORIGINAL ARTICLE
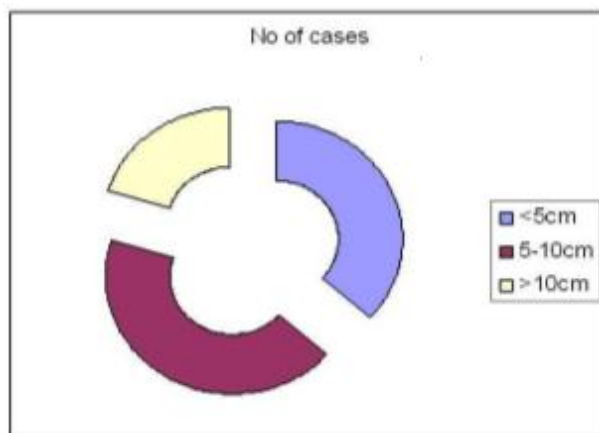
Slide 2

Slide 3

Slide 4

ORIGINAL ARTICLE

AUTHORS:
1. Vidhya Lakshmi S.
2. Seyed Rabiya

PARTICULARS OF CONTRIBUTORS:
1. Associate Professor, Department of Pathology, PSG Institute of Medical Sciences & Research, Coimbatore.
2. Post Graduate Student, Department of Pathology, PSG Institute of Medical Sciences & Research, Coimbatore.

NAME ADDRESS EMAIL ID OF THE CORRESPONDING AUTHOR:
Dr. S. Vidhya Lakshmi,
Associate Professor,
Department of Pathology,
PSG Institute of Medical Sciences and Research,
Coimbatore-641004.
Email: drvidhyalakshmi@yahoo.co.in