

Immunohistochemistry in breast carcinoma

Gireesh V. Achalkar

Associate Professor, Dept. of Pathology, Raichur Institute of Medical Sciences, Raichur, Karnataka, India

***Corresponding Author:**

Email: drgireesha@yahoo.com

Received: 8th February, 2018

Accepted: 13th April, 2018

Abstract

Introduction: Breast Carcinoma has emerged as most common type of malignant tumor in women surpassing cervical carcinoma which was commonest few decades ago. Hence a proper diagnostic protocol is very much necessary for diagnostic, management and prognostic purposes. Immunohistochemistry has helped to some extent in this aspect.

Materials and Methods: Both H&E stained sections and IHC sections for ER, PR and Her2 were studied. The Hormone receptor status was correlated with age, grade and type of tumor.

Results: Total cases analyzed in the present study were 50. Infiltrating ductal carcinoma was 45/50(90%) and others were 5/50(10%). IDC was the most common one and triple negative tumors being 34.83% Hormone receptor status was more in younger patients than older ones. And there was positive association with smaller size of the tumors. Their expression was higher in grade I tumors at 85% than in grade III tumors at 10%. The smaller tumors less than 2cm showed triple positivity more frequently than larger tumors more than 2cm.

Conclusion: In the present study the breast carcinoma in younger patients having smaller size of tumor was frequently positive for ER, PR and negative for Her2/neu than older patients with larger size of the tumor. Immunohistochemical study of breast carcinoma must be routine so as to help the clinician for better management of patients.

Keywords: Breast carcinoma, Infiltrating duct carcinoma, Immunohistochemistry (IHC), Estrogen receptor (ER), Progesterone receptor (PR), Human epidermal growth factor receptor-2(HER2).

Introduction

In the recent few decades, there have been significant advances in breast carcinoma diagnosis with earlier detection of disease and development of more effective patient management. Molecular and genetic study is very informative but is not affordable by many in our country. Immunohistochemistry markers like particularly estrogen receptor (ER), progesterone receptor (PR) and human epidermal growth factor receptor 2(HER2) proteins give almost equally valuable information and help in patient management.¹⁻³

The hormones act by attaching themselves to the receptors present on breast cells. They are useful for normal development and function of breast cells, pregnancy, childbirth and lactation. It has been well studied that anti hormonal therapy in breast carcinoma have dramatically improved the patient outcomes.

The expression of the hormone receptors in a patient with breast cancer is an example of a weak prognostic but strong predictive biomarker.^{4,5}

If a patients tumor express ER and PR as seen in approximately 70% of breast cancer we can predict they can benefit from endocrine therapy. Knowing the hormone receptor status of cancer patients helps the treatment options. The hormonal therapy doesn't work if the patient is hormone receptor negative.⁶

All invasive breast cancers must be tested for the hormone receptor status as well as Her-2/neu. DCIS should also be checked for this. Triple negative cancers of breast are known to grow and spread faster than

other types. Triple positive breast cancers benefit from hormonal therapy and targeted therapy to Her-2/neu.^{7,8}

Materials and Methods

The present cross sectional study was conducted on 50 paraffin blocks of with infiltrating duct carcinoma. Data regarding age, tumor size and grade were collected. Bloom-Richardson scoring was used for histology grading. Paraffin embedded blocks were used to prepare 3-4mm thick slides, the slides were incubated with Tris EDTA for 15 minutes. The slides were covered by primary antibody for 30 minutes at room temperature followed by incubation. Then they were covered by chromogen. ER, PR and HER2/neu were evaluated.

In all the cases both H and E sections and IHC slides were studied using light microscopy and the percentage and intensity of nuclear immunostaining was assessed with negative and positive controls. Staining of normal ductal epithelium was used as internal control for ER and PR staining. IHC was considered positive if >1% of tumor cell nuclei are immune reactive and negative if <1%.

Results

Hematoxylin and Eosin Stained sections along with IHC slides were studied. Majority of them were infiltrating duct carcinoma ER/PR positive were accounted for 36 cases (72%) HER2 Positive cases accounted for 20 cases (40%) Triple negative cases were 10 (20%).

The average age of the patient was 59 years. All of them were post menopausal patients. Stage I was most

common followed by stage II and Stage III.

Table 1

	ER/PR+	ER/PR+	ER/PR-	ER/PR-	P. Value
	Her2+ 16(32%)	Her2- 20(40%)	Her2+ 04(08%)	Her2-10(40%)	
Age(Median)	54.2	64.0	56.1	59.1	<0.001

Tumor Stage					
I	5(31.25%)	12(60.0%)	02(50.0%)	02(30.0%)	<0.001
II	7(43.75%)	04(20.0%)	01(25.0%)	05(50.0%)	
III	4(25.00%)	04(20.0%)	01(25.0%)	02(20.0%)	

Cancer Type				
Ductal	14(87.50%)	17(85.0%)	04(100.0%)	07(70.0%)
Lobular	02(12.50%)	03(15.0%)	Nil	03(30.0%)

Final analysis included 50 breast cancer patients from Jan, 1 2016 to Dec, 31, 2017. The mean age of all subjects was 59.1 year. Of 50 patients 16(32.0%) were triple positive, 20 (40%) were ER/PR+Her2-, 04(08.0%) were ER/PR-Her2+ and the rest 10(20.0%) were classified as triple negative.

Discussion

Table 2: IHC Markers used in breast carcinoma

Diagnosis	Prediction
Myoepithelial Markers	Estrogen receptor
Smooth Muscle actin	Progesterone Receptor
CK 14	PS2 Protein
Pcadherin	
Calponin	
Caldesmon	

CK 5/6	HER2 Protein
Basement Membrane Collagen type IV	Epidermal Growth factor receptor
Epithelial Membrane Antigen	MIB-1
E cadherin	P53 Protein

The breast carcinoma is a heterogenous tumor with multifaceted features. There are different subtypes with varied natural history. It has been recognized that the clinicopathological features gave information with regard to different therapeutic and prognostic features.^{9,10} Present study reinforces the above fact and reveals statistically significant features in pathological findings and treatment outcomes. The estimated survival differences have important implications for both pathologist and oncologist. This should be complemented with other variables like age, tumor size, grade and stage.¹¹

IHC based study of both ER/PR and Her2/neu status is better than the previous studies where only one of 3 tests were used and is better at communication. Recent research studies have shown that newer molecular classification also have important prognostic value where microarrays are used for gene expression analysis.^{12,13}

The IHC based classification is very much useful in clinical practice and correlates well with molecular study.^{14,15} This study finding is same as that of other studies where there is positive correlation between triple negative breast carcinoma and poor prognosis. Arenas et al in a study of 25 patients reported no significant differences in expression patterns of ER, PR or Her2 following neoadjuvant chemotherapy when compared with other studies. A similar trend was reported by Adams et al.^{16,17}

In contrast, statistically significant change in hormone receptor status of breast carcinoma following neoadjuvant chemotherapy have been reported.^{18,19}

Breast carcinoma is presently the most common type of malignancy in women and is a major cause of morbidity and mortality. In India it accounts for 1,44,937 newly detected cases. As per the population based cancer registry data, location wise Bengaluru ranks the top most position (36.6%). Dunnwald et al (2007) observed that the higher relative risks of mortality was associated with having an ER+/PR-, ER-/PR+ or ER-/PR-. In a study from Kerala the ER positivity was found to be 52.0%. ER positivity in the present study was 72%.²⁰

Breast carcinomas are molecularly classified into four types as per their gene expression profiling. Luminal A type are ER positive and/ or PR positive and HER2 negative, with low ki67. Luminal B type are ER positive and/ or PR positive and HER2 positive or if HER2 negative have high ki67 (>14%). They have higher histological grade than luminal A. Triple negative types are negative for all three above markers.

Luminal A and B types can be given endocrine treatment but the response in the luminal B is not good and shows response to chemotherapy.²⁰

Conclusion

Breast carcinoma being the most common type of malignant tumor needs proper workup. Immunohistochemical study in addition to routine H and E sections gives significant information about the hormone receptor status which can be managed with anti hormonal therapy such as tamoxifen. Molecular study will give very valuable information with regard to therapy and prognosis, but it is expensive and not available in all pathology laboratories. Given the limited treatment options for triple negative tumors, it is important to encourage early diagnosis of tumor through mass screening.

References

1. Böcker W, Bier B, Freytag G, Brömmelkamp B, Jarasch ED, Edel G, Dockhorn-Dworniczak B, Schmid KW. An immunohistochemical study of the breast using antibodies to basal and luminal keratins, alpha-smooth muscle actin, vimentin, collagen IV and laminin. Part II: Epitheliosis and ductal carcinoma in situ. *Virchows Arch A Pathol Anat Histopathol*. 1992;421:323–330.
2. Ichihara S, Koshikawa T, Nakamura S, Yatabe Y, Kato K. Epithelial hyperplasia of usual type expresses both S100-alpha and S100-beta in a heterogeneous pattern but ductal carcinoma in situ can express only S100-alpha in a monotonous pattern. *Histopathology*. 1997;30:533–541.
3. Yeh IT, Mies C. Application of Immunohistochemistry to Breast Lesions. *Arch Pathol Lab Med*. 2008;132:349–358.
4. Nayar R, Breland C, Bedrossian U, Masood S, DeFrias D, Bedrossian CW. Immunoreactivity of ductal cells with putative myoepithelial markers: A potential pitfall in breast carcinoma. *Ann Diagn Pathol*. 1999;3:165–173.
5. Barbareschi M, Pecciarini L, Cangi MG, Macri E, Rizzo A, Viale G, Doglioni C. p63, a p53 homologue, is a selective nuclear marker of myoepithelial cells of the human breast. *Am J Surg Pathol*. 2001;25:1054–1060.
6. Barnes DM, Harris WH, Smith P, Millis RR, Rubens RD. Immunohistochemical determination of oestrogen receptor: comparison of different methods of assessment of staining and correlation with clinical outcome of breast cancer patients. *Br J Cancer*. 1996;74:1445–1451.
7. Fisher ER, Anderson S, Dean S, Dabbs D, Fisher B, Siderits R, Pritchard J, Pereira T, Geyer C, Wolmark N. Solving the dilemma of the immunohistochemical and other methods used for scoring estrogen receptor and progesterone receptor in patients with invasive breast carcinoma. *Cancer*. 2005;103:164–173.
8. Allred DC, Bustamante MA, Daniel CO, Gaskill HV, Cruz AB. Immunocytochemical analysis of estrogen receptors in human breast carcinomas. Evaluation of 130 cases and review of the literature regarding concordance with biochemical assay and clinical relevance. *Arch Surg*. 1990;125:107–113.
9. Goldhirsch A, Coates AS, Gelber RD, Glick JH, Thürlimann B, Senn HJ. First--select the target: better choice of adjuvant treatments for breast cancer patients. *Ann Oncol*. 2006;17:1772–1776.
10. Goldhirsch A, Glick JH, Gelber RD, Coates AS, Thürlimann B, Senn HJ. Meeting highlights: international expert consensus on the primary therapy of early breast cancer 2005. *Ann Oncol*. 2005;16:1569–1583.
11. Fisher B, Dignam J, Wolmark N, Wickerham DL, Fisher ER, Mamounas E, Smith R, Begovic M, Dimitrov NV, Margolese RG, et al. Tamoxifen in treatment of intraductal breast cancer: National Surgical Adjuvant Breast and Bowel Project B-24 randomised controlled trial. *Lancet*. 1999;353:1993–2000.
12. Hicks DG, Linda Schiffhauer, Standardized Assessment of the HER2 Status in Breast Cancer by Immunohistochemistry. *Lab Medicine*. 2011;42:459–467.
13. Wolff AC, Hammond ME, Schwartz JN, Hagerty KL, Allred DC, Cote RJ, Dowsett M, Fitzgibbons PL, Hanna WM, Langer A, et al. American Society of Clinical Oncology/College of American Pathologists guideline recommendations for human epidermal growth factor receptor 2 testing in breast cancer. *J Clin Oncol*. 2007;25:118–145.
14. de Melo Rêgo MJ, Cordeiro MF, Cavalcanti Cde L, de Carvalho Junior LB, Beltrão EI. Immunohistochemiluminescence detection: a quantitative tool in breast cancer HER-2 status evaluation. *Dis Markers*. 2013;34:373–377.
15. Allred DC, Harvey JM, Berardo M, Clark GM. Prognostic and predictive factors in breast cancer by immunohistochemical analysis. *Mod Pathol*. 1998;11:155–168.
16. Choudhury KR, Yagle KJ, Swanson PE, Krohn KA, Rajendran JG. A robust automated measure of average antibody staining in immunohistochemistry images. *J Histochem Cytochem*. 2010;58:95–107.
17. Adams J, Carder PJ, Downey S, Forbes MA, MacLennan K, Allgar V, Kaufman S, Hallam S, Bicknell R, Walker JJ, et al. Vascular endothelial growth factor (VEGF) in breast cancer: comparison of plasma, serum, and tissue VEGF and microvessel density and effects of tamoxifen. *Cancer Res*. 2000;60:2898–2905.
18. Harbeck N, Thomssen C, Gnant M. St. Gallen 2013: brief preliminary summary of the consensus discussion. *Breast Care (Basel)* 2013;8:102–109.
19. Jonat W, Arnold N. Is the Ki-67 labelling index ready for clinical use? *Ann Oncol*. 2011;22:500–502.
20. Inwald EC, Klinkhammer-Schalke M, Hofstädter F, Zeman F, Koller M, Gerstenhauer M, Ortmann O. Ki-67 is a prognostic parameter in breast cancer patients: results of a large population-based cohort of a cancer registry. *Breast Cancer Res Treat*. 2013;139:539–552.

How to cite this article: Achalkar GV. Immunohistochemistry in breast carcinoma. *Ind J Pathol Oncol*, 2018;5(3):415-417.