

Histological Subtyping of Carcinoma Breast and Analysis of Ki67 Expression: A Single Institution Study in Northern India

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Abstract

Original Research Article

Breast cancers are widely prevalent in females worldwide. Diagnosis and prognosis of carcinoma breast is effectively facilitated by the analysis of various hormone receptors and certain biomarkers. Ki-67 is an important proliferative biomarker which is a nucleoprotein used for predicting these cancers. This study was carried out to review the distribution of histological subtypes and pattern of Ki-67 expression in them. A part retrospective and part prospective study was done on a total of 59 cases of carcinoma breast. All stained slides were reviewed and the morphological diagnosis based on H & E staining was considered. For Ki-67 immunohistochemistry, representative sections from paraffin blocks were taken on poly-L lysine coated glass slides. Most common histological subtype in our study was invasive ductal carcinoma followed by invasive papillary carcinoma. Ki67 score was maximum in metaplastic carcinoma followed by IDC with squamous metaplasia. Also ductal carcinomas had more ki67 proliferation activity than lobular ones. Further studies on large samples are required to correlate the Ki-67 to histological subtypes of breast cancer and to consider it as a significant prognostic factor of breast cancer.

Keywords: Breast cancer, immunohistochemistry, Ki-67 antigen, prognosis.

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INTRODUCTION

Breast cancers are widely prevalent in females worldwide. Histological types refer to the growth pattern of the tumours. The histological diversity of adenocarcinomas in the breast has long attracted pathologists, who have identified specific morphological and cytological patterns that were consistently associated with distinctive clinical course and outcomes. The commonest type of breast carcinomas the so-called invasive ductal carcinomas not otherwise specified (IDC-NOS) or of no special type (IDC-NST)[1], which is a diagnosis of exclusion and comprises adenocarcinomas that fail to exhibit sufficient characteristics to warrant their classification in one of the special types. Although grade identifies prognostic subgroups among special types of breast cancer, some entities that are by definition of high histological grade have a relatively good prognosis (eg. medullary carcinoma)[1]. Diagnosis and prognosis of carcinoma breast is effectively facilitated by the analysis of various hormone receptors and certain biomarkers. Ki67 is a monoclonal antibody directed against a nonhistone bound nuclear protein present only in proliferation cells. The immunostaining of malignant tumors with low and high proliferation rates showed that

the number of tumor cells stained with Ki-67 coincided with estimated proliferation rates of the tumors [2]. The exact determination of percentage of proliferation cells in neoplasia is of prognostic significance and may also be important for the choice of appropriate treatment. Immunostaining with Ki-67 has also been used in breast tumors. There have been quite a few studies which have tried to correlate Ki-67 immunostaining with other indicators of prognosis and have also found it to be an independent prognostic marker in these tumors [3]. Although it is reasonably well established that Ki-67 positivity is an independent predictor of ultimate prognosis and course of disease and its recurrence many lacunae exist. These studies in our own population are very scarce. As it is widely accepted that cancer management should be according to loco-regional profile and treatment strategy should be devised accordingly, however no large-scale cancer registry is available in developing countries. So ki67 may serve as a useful marker in tailoring treatment regimen as response to chemotherapy may be altered by the proliferative activity of cancer cells [4]. Our study was aimed to study histological profiles and pattern of ki67 expression in breast cancer subtypes.

SUBJECTS AND METHODS

This study was carried out in Department of pathology, Vardhman Mahavir Medical College and Safdarjung Hospital, New Delhi. A part retrospective and part prospective study was done on a total of 59 cases of carcinoma breast. Representative paraffin blocks of Modified Radical Mastectomy, lumpectomy or biopsies of carcinoma breast were included for the study.

EXCLUSION CRITERIA

1. All the patient's blocks with breast tumors other than carcinoma breast.
2. Tissue blocks with extensive necrosis of hemorrhage.
3. Tissue section with inadequate study material.

Micron thick sections were cut for H & E Staining, and for Ki-67 immunohistochemistry, section were on poly- L-lysine coated slides. Positive and negative controls were run with each batch of IHC stain. Positive controls unclouded section from invasive breast cancer known to express Ki-67. Negative control staining was obtained by substituting primary antibody with an antibody of irrelevant specificity. Ki-67 score is the percentage of tumor cells showing the nuclear positivity for Ki-67. We used random counting method for counting cells positive for Ki-67 immunostaining. Thus, percentage of tumor cells in 10 randomly chosen fields from most representative invasive areas with minimum 1000 cells were counted. The Ki-67 expression was then correlated with histological subtypes of carcinoma breast.

RESULTS

Table-1: Age distribution

AGE GROUP (YEARS)	NO. OF CASES	% AGE
11-20	1	1.69%
21-30	4	6.78%
31-40	14	23.73%
41-50	27	45.76%
51-60	8	13.56%
61-70	5	8.47%
GRAND TOTAL	59	100.00%

Table-2: sex distribution

SEX	NO. OF CASES	% AGE
FEMALE	55	93
MALE	4	7
GRAND TOTAL	59	100

Table-3: Type of specimen

TYPE OF SPECIMEN	NO. OF CASES	% AGE
MODIFIED RADICAL MASTECTOMY	39	66.10%
LUMPECTOMY	13	22.03%
BIOPSY	7	11.87%
GRAND TOTAL	59	100%

Table-4: Histologic subtypes

Code	Subtype	No. of Cases	% AGE
1	IDC WITH DCIS	1	1.69%
2	INVASIV DUCTAL CARCINOMANOS	52	88.14%
3	INVASIVE PAPILLARY CARCINOMA	2	3.39%
4	INVASIVE LOBULAR CARCINOMA	1	1.69%
5	MUCINOUS CARCINOMA	1	1.69%
6	IDC WITH SQUAMOUS METAPALSIA	1	1.69%
7	METAPLASIC CARCINOMA	1	1.69%
	GRAND TOTAL	59	100.00%

Table-5: Association between ki-67 and histologic subtypes

CODING	SUBTYPE	No. of CASES	KI-67 SCORE
1	IDC WITH DCIS	1	18
2	INVASIVE DUCTAL CARCINOMA	52	13.46
3	INVASIVE PAPILLARY CARCINOMA	2	15
4	INVASIVE LOBULAR CARCINOMA	1	11
5	MUCINOUS CARCINOMA	1	1
6	IDC WITH SQUAMOUS METAPLASIA	1	28
7	METAPLASTIC CARCINOMA	1	50

In present study, maximum number of cases was in age group of 41-50 years constituting 45.76% of call cases while cases were minimum in the age group of 11- 20 year (1.69%). Mean age of presentation was 45.71 years. Out of total cases, 93% were females. Table 3 represents the type of specimen submitted for histopathological and immunohistochemical study. Most of the specimens were of Modified Radical Mastectomy (MRM) constituting 66.10% followed by lumpectomy specimens (22.03%). Biopsy specimens were minimum i.e 11.87%. While males constituted only 7% of all cases. Present study consists of 88.14% of invasive ductal carcinoma showing that it is the most common type of breast carcinoma in present study. Invasive papillary carcinoma is second most common subtype and constituted 3.39% of all cases. Remaining subtype i.e invasive lobular carcinoma, mucinous carcinoma, metaplastic carcinoma, IDC with squamous metaplastic and IDC with DCIS contributed 1.69% each. Among the breast carcinoma cases, most common subtypes was Invasive ductal carcinoma – not otherwise specified (IDC-NOS) constituting 88.1% of total cases.

Remaining cases studied were

1. IDC with DCIS (1.69%)
2. Invasive papillary carcinoma (3.39%)
3. Mucinous carcinoma (1.69%)
4. Invasive lobular carcinoma (1.69%)
5. Metaplastic carcinoma (1.69%) and
6. Invasive duct carcinoma with focal squamous metaplasia (1.69).

Table 5 represents Ki67 score in different subtypes of breast carcinoma. Ki67 score was highest in metaplastic carcinoma (60), followed by IDC with squamous metaplasia (28), IDC with DCIS (18), Invasive papillary carcinoma (15), Invasive ductal carcinoma (13.46) and Invasive lobular carcinoma (11) showed Ki-67 score in decreasing order while the score was minimum for mucinous carcinoma (1).

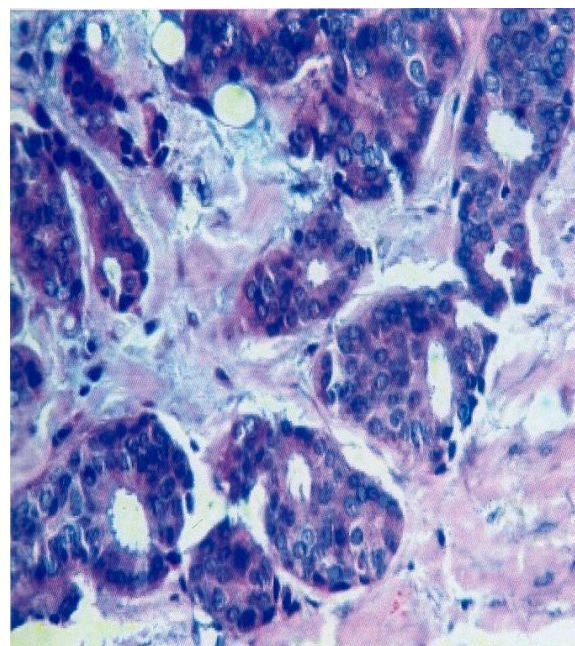


Fig-1: Photomicrograph showing invasive ductal carcinoma (H&E; x400)

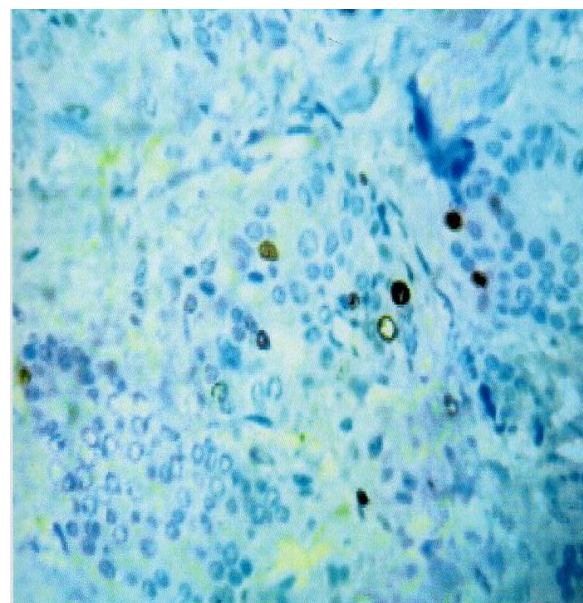


Fig-2: KI-67 Immunostaining in invasive ductal carcinoma (H&E; x400)

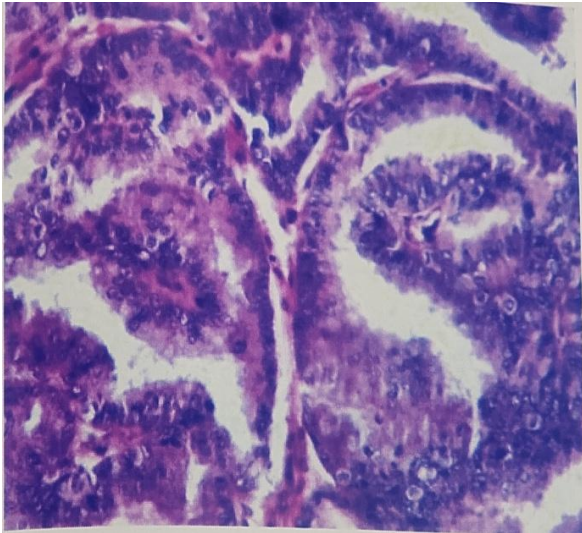


Fig-3: Photomicrograph showing invasive papillary carcinoma (H&E; x400)

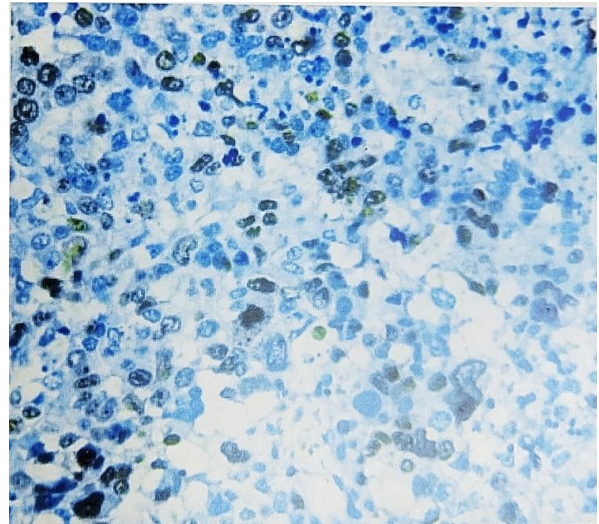


Fig-6 – KI-67 Immunostaining in invasive metaplastic carcinoma (H&E; x400)

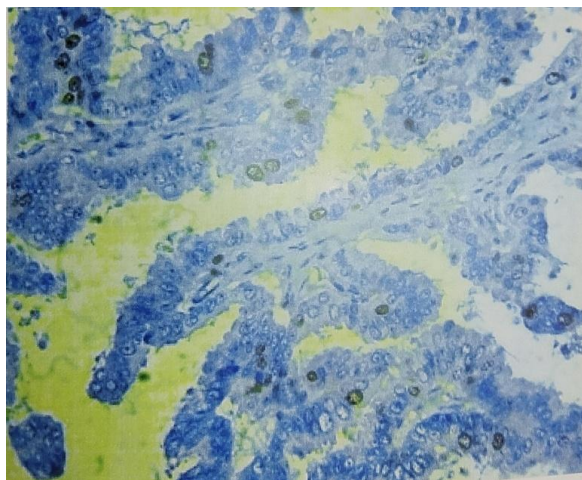


Fig-4: KI-67 Immunostaining in invasive papillary carcinoma (H&E; x400)

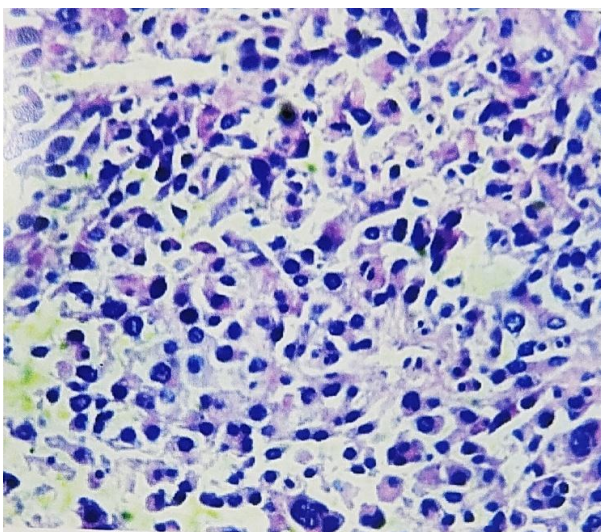


Fig-5: Photomicrograph showing invasive metaplastic carcinoma (H&E; x400)

DISCUSSION

In the study of Grethe Albrektsen *et al.* [5], ductal tumors, the most common histological type accounted for 81.4% of all cases, followed by lobular tumors (6.3%) and unspecified carcinoma (5.5%). Other subtypes accounted for 0.4% - 1.5% of the cases Page and Anderson [6] have found invasive duct carcinoma (NOS) to be the commonest to the malignant tumors of breast. In the long term study by Toikkanen *et al.* from Finland [7], the percentage of various categories of malignant tumors of breast observed was – infiltrating ductal carcinoma (72.2%) and lobular carcinoma (16.1%). Similarly, in an analysis of breast tumors in a larger series by Eisenberg *et al.* [8], the investigator reported the incidences of various types of malignant tumors as infiltrating duct carcinoma – 89%, medullary carcinoma – 3.1% Lobular carcinoma – 59%, other – 2.0%.

Clark *et al.* [9] stated that invasive Lobular carcinoma (ILC) of the breast is far less common than invasive ductal carcinoma (IDC), consisting about 8 - 14% of all breast carcinoma. Lobular carcinoma is more likely to occur in older patients, (with median age of 64.6 years).

One case of papillary carcinoma was also observed in the study. Age at diagnosis was 65 years. Stalsberg and Thomas [10] reported that, while the relative frequency of ductal carcinoma is essentially constant by age, the frequencies of papillary and mucinous carcinomas tend to increase with age. Another case was of metaplastic carcinoma. Age at diagnosis was not mentioned. In the study of Huvos *et al.* [11], they found that metaplastic breast carcinoma constitute mammary carcinoma. Taylor *et al.* [12] stated that metaplastic carcinomas are usually seen in women who are more than 50 years old. In our study, single case of mucinous carcinoma was also seen which occurred in 52 year old male. It usually occurs in

postmenopausal women. It is widely accepted that at least 75% of the tumour have a mucinous growth pattern. Pure mucinous carcinoma of the male breast is an extremely rare neoplasm [13].

In present study, mean Ki-67 score for various subtypes of breast carcinoma was measured as 13.46%, 15%, 1%, 28%, 11%, 18% and 60% respectively for IDC (NOS), invasive papillary carcinoma, mucinous carcinoma, metaplastic carcinoma, and invasive lobular carcinoma, IDC with DCIS and IDC with focal squamous metaplasia. Kuenen- Bounmeester *et al.* [14] studied 34 cases of invasive breast carcinoma and analysed them for heterogeneity of Ki-67 in various histological subtypes. He found ductal carcinomas showing the highest.

Ki-67 reactivity of the 34, the mucinous the lowest. Mucinous and lobular carcinomas were subdivided according to their dominant growth pattern. Carcinoma with a solid or comedo growth pattern showed the highest proliferative activity. Lelle *et al.* compared the growth fraction, as determined by Ki-67, of ductal and lobular carcinomas and found lower values in the latter [15].

CONCLUSION

Breast carcinoma is the most common malignant tumor in women. Carcinoma of the breast is a truly complex disease with a large intertumoral and intratumoral heterogeneity characteristics leading to uncertain clinical course and varied response to treatment modalities. There are only a few data that have explained the heterogeneity of cells within a tumor; in general, this feature is not fully understood. The translation of molecular approaches into daily clinical practice to support morphological features and immunohistochemical profile is need of time and challenging [16]. Most common histological subtype in our study was invasive ductal carcinoma followed by invasive papillary carcinoma. Ki67 score was maximum in metaplastic carcinoma followed by IDC with squamous metaplasia. Also ductal carcinomas had more ki67 proliferation activity than lobular ones. This agrees with previous studies that anaplastic carcinoma is most aggressive and ductal carcinomas are more aggressive than lobular ones. Size of the present study is small to conclude about the value of Ki-67 in various histological subtypes of breast cancer. Further studies on large samples are required to correlate the Ki-67 to histological subtypes of breast cancer and to consider it as a significant prognostic factor of breast cancer.

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