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Original Article

Histomorphological Study of a Spectrum of Breast Diseases in Association with Immunohistochemistry in Vadodara, Gujarat, India

Anamika Thakral[1], Meena Daveshwar[2]

[1]Resident, Department of Pathology, Medical College, Vadodara, Gujarat, India, [2]Associate Professor, Department of Pathology, Medical College, Vadodara, Gujarat, India

Abstract

Introduction: Breast lumps are a cause for concern, both for the patient and surgeon, because of the risk of cancer. In this study, a total of 350 cases of breast pathology were studied which included cases of all ages and histological types. The main purpose of this study was to analyze the spectrum of breast lesions in SSG Hospital, Vadodara, while highlighting the common uses of immunohistochemistry (ICH) in diagnostic breast pathology.

Materials and Methods: This observational study was conducted in histopathology section of the Department of Pathology, SSG Hospital, Vadodara. The specimens were grossly examined followed by processing, sectioning from paraffin-embedded blocks, and staining with hematoxylin and eosin. ICH markers were applied on the cases wherever applicable and required from a diagnostic point of view.

Results: The most common type of inflammatory lesion of breast was granulomatous mastitis, benign breast lesion was fibroadenoma, and breast malignancy was infiltrating ductal carcinoma-not otherwise specified type. Triple-positive breast cancers in this study were 1.7%. Triple-negative breast cancers in this study were 36.75%. E-cadherin was used in 8 cases. Loss of E-cadherin indicates lobular type. Myoepithelial markers were used in 8 cases in our study. Positivity of these markers confirms the benign nature of the lesion.

Conclusion: Microscopy has always been the mainstay of histological diagnosis in breast pathology. However, the growing use of ICH has proved to be advantageous and an incredible aid in the diagnosis. Several cases of diagnostic dilemma such as invasion or pseudoinvasion, ductal or lobular type, and *in situ* or invasion can be reliably diagnosed by the judicious use of ICH markers.

Key words: Breast diseases, Histomorphology, Immunohistochemistry

INTRODUCTION

Breast diseases are showing a rising trend worldwide. A number of studies have been done to know the magnitude of the problem. There is a wide variation in the spectrum of breast diseases in various countries or ethnic groups. Breast lumps are a cause for concern, both for the patient and surgeon, because of the risk of cancer.

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Benign breast diseases are more prevalent as compared to malignant and inflammatory lesions, as seen throughout the world. In India, breast cancer forms the second most common malignancy after cervical cancer and is detected in 20/1, 00,000 women.

Aims of the Study

1. To evaluate the histopathological profile of different breast diseases in SSG Hospital based on hematoxylin and eosin (H and E) sections.
2. To know the frequency of various breast diseases in relation to the age of occurrence in our institute.
3. To distinguish usual ductal hyperplasia from atypical ductal hyperplasia or low-grade carcinoma *in situ* using immunohistochemical (ICH) markers.
4. For the distinction of ductal carcinoma *in situ* (DCIS)

Corresponding Author: Dr. Anamika Thakral, Thakral Hospital, Kurukshetra Road, Kaithal, Haryana - 136 027, India. Phone: +91-8447074412. E-mail: anamikathakral@gmail.com

and lobular carcinoma *in situ* (LCIS), and of invasive ductal carcinoma-not otherwise specified (NOS) and invasive lobular carcinoma using ICH markers.
5. To differentiate *in situ* versus invasive carcinoma; invasion versus pseudoinvasion using myoepithelial markers.
6. To evaluate the prognosis of malignant breast lesions using ICH markers.

MATERIALS AND METHODS

The present study was conducted at the Department of Pathology, Medical College Baroda and SSG Hospital. The test population comprised patients with breast pathology, between September 2013 and October 2015. The protocol was approved by the local Ethics Committee.

Data for the study were obtained from the departmental records (for retrospective study) and tissue specimens received in the histopathology section (for prospective study) in the specified period of study.

Tissue Collection

The tissues of the test population submitted were evaluated by histopathological processing and examination (HPE). Pro forma designed to gather uniform necessary information was used for every case. After that, the most suitable tissue block was selected for IHC evaluation.

Tissue Processing

Tissues were fixed in 10% buffered formalin overnight, for an average period of 16-24 h. The tissue was grossed and representative blocks were processed in the histokinete with a cycle of 24 h, after which the processed tissue was embedded into paraffin wax blocks and then chucked onto wooden chucks. The wax blocks were trimmed using a rotary microtome. Sections were taken onto slides and stained by the routine H and E stain. During the HPE reporting, most of the cases were diagnosed by light microscopy and subsequently, estrogen receptor (ER), progesterone receptor (PR), and Her2 were done on the best section representing the tumor. Only in certain cases where there was diagnostic dilemma, other IHC markers were applied.

IHC Procedure

The selected tissue block sections were taken up on poly-l-lysine-coated slides for IHC procedure. The slides were deparaffinized in xylene, thereafter brought down to water after passing through increasing grades of alcohol. The peroxidase antiperoxidase method of IHC was followed. Biogenex reagents were used for the antigen retrieval and IHC staining process. The heating cycles followed in the Biogenex temperature-controlled microwave were two cycles of 10 min and 5 min each at 95°C, with intermittent refilling of the antigen retrieval solution.

After that, the slides were brought down to room temperature and taken through the steps of wash with TRIS buffer, peroxide block, power block, and monoclonal antibodies. After this, slides were again washed in TRIS buffer, the secondary antibody exhibited, thereafter DAB chromogen was added. The slides were then washed with water, counterstained with hematoxylin, and blued. Then, the slides were serially dehydrated in alcohol, cleared in xylene, and thereafter mounted using DPX. After drying, the test slides were examined along with the control sections stained simultaneously.

RESULTS

All the 340 cases were evaluated histopathologically, and 124 cases were subsequently evaluated ICH. The following observations were made.

In our study, the age of patients ranged from 13 to 85 years, with a mean age of 40.5 years. Maximum number of cases were seen in the age group of 41-50 years ($n = 80$, 23.53%) followed by the age group of 31-40 years ($n = 72$, 21.18%) (Table 1).

Totally 2 cases of borderline phyllodes were found which cannot be classified into either of the above-mentioned categories, resulting in little disparity in the total number of cases.

About 331 out of the 340 cases in our study were females, constituting 97.35% of the total patients (Table 2).

The most common inflammatory lesion in our study was found to be granulomatous mastitis ($n = 7$, 43.75% of the total inflammatory lesion cases) (Figure 3) followed by breast abscess ($n = 6$, 37.50%) (Figure 1) and duct ectasia ($n = 2$, 12.5%) (Figure 2). One case of chronic mastitis was found whereas we did not confront any case of fat necrosis in our study (Table 3).

Table 4 shows that most common benign breast disease in our study was fibroadenoma forming the major bulk with 103 cases out of the total 138 cases of benign breast diseases (74.64%) (Figures 5 and 6). It was followed by 8 cases of fibrocystic disease (6.02%) and 6 cases of gynecomastia (4.51%) (Figure 4). There were 4 cases of phyllodes tumor (Figure 13) (3%), 3 cases each of sclerosing adenosis (Figure 9), lactating adenoma (Figure 7), and intraductal papilloma (Figure 11), 2 cases each of ductal adenosis and galactocele (Figure 14), and 1 case each of tubular adenoma (Figure 8), florid ductal adenoma (Figure 10), atypical ductal hyperplasia, and radial scar (Figure 12). We did not encounter any case of lipoma, fibromatosis, and lobular hyperplasia in our study.

Out of the 184 cases of malignant lesion in our study, the most common (150) was infiltrating ductal carcinoma (IDC)-NOS (Figure 16), forming 80.64% of the total malignant cases. There were 6 cases of mucinous carcinoma (3.47%) (Figure 18), 5 cases of DCIS (2.89%) (Figure 15), 4 cases of metaplastic carcinoma (2.31%) (Figure 23), 2 cases each of mixed IDC and ILC (Figure 17), medullary carcinoma, metastatic adenocarcinoma (Figure 27), and stromal sarcoma, (Figure 26) and 1 case each of invasive micropapillary carcinoma (Figure 22), papillary carcinoma (Figure 21), secretory carcinoma (Figure 19), and malignant phyllodes (Figure 25). There were no cases of LCIS, ILC, and lymphoma in our study. (Table 5)

Table 1 and 6 shows that most of the benign breast diseases occurred in the age group of 21-30 years (n = 49). Of these, 44 were females and 5 were males. It was closely followed by 43 benign cases in the age group of 11-20 years. (Tables 1 and 2)

Whereas, the most common age group facing the malignant breast lesion was 41-50 years (n = 64), of which all were females. The age group with second highest frequency of malignant lesions was 51-60 years (n = 44), of which 1 was male (diagnosed as IDC-NOS). (Tables 1 and 2)

Thus, out of the total neoplastic cases studied, 93.98% of benign and 99.52% of malignant lesions were seen in females.

Table 1: Distribution of cases according to age

Age group	Total (%)	Inflammatory (%)	Benign (%)	Malignant (%)
11-20	43 (12.65)	0 (0)	43 (100)	0 (0)
21-30	64 (18.82)	9 (14.06)	49 (76.56)	16 (25.00)
31-40	72 (21.18)	3 (4.17)	30 (41.67)	39 (54.17)
41-50	80 (23.53)	2 (2.50)	12 (15.0)	64 (82.50)
51-60	44 (12.94)	0 (0)	0 (0)	44 (100)
61-70	30 (8.82)	2 (6.67)	3 (10.00)	25 (83.33)
71-80	6 (1.76)	0 (0)	1 (16.67)	5 (83.34)
81-90	1 (0.29)	0 (0)	0 (0)	1 (100)
Total	340	16 (4.70)	138 (40.59)	184 (54.71)

Table 2: Distribution of cases according to gender

Gender	Number of cases (%)
Male	9 (2.65)
Female	331 (97.35)

Table 3: Incidence of inflammatory lesions

Type of lesion	Number of cases (%)
Breast abscess	6 (37.50)
Chronic mastitis	1 (6.25)
Granulomatous mastitis	7 (43.75)
Duct ectasia	2 (12.50)
Fat necrosis	0 (0)

Table 7 shows that out of the total 117 cases, those which came out to be ER positive in this study were 37 (31.62%), total PR-positive cases were 35 (29.91%), and Her2neu-positive cases were 36 (30.77%). Whereas, the total number

Table 4: Incidence of benign lesions

Type of lesion	Number of cases (%)
Fibroadenoma	103 (74.64)
Fibrocystic disease	8 (6.02)
Sclerosing adenosis	3 (2.26)
Ductal adenosis	2 (1.50)
Lactating adenoma	3 (2.26)
Tubular adenoma	1 (0.75)
Florid ductal adenoma	1 (0.75)
Benign phyllodes tumor	4 (3)
Intraductal papilloma	3 (2.26)
Lipoma	0 (0)
Atypical ductal hyperplasia	1 (0.75)
Fibromatosis	0 (0)
Galactocele	2 (1.50)
Lobular hyperplasia	0 (0)
Radial scar	1 (0.75)
Gynecomastia	6 (4.51)

Table 5: Incidence of malignant lesions

Type of lesion	Number of cases (%)
Intraductal carcinoma *in situ*	5 (2.89)
Lobular carcinoma *in situ*	0 (0)
Infiltrating duct carcinoma-NOS	150 (80.64)
Invasive lobular carcinoma	0 (0)
IDC+ILC	2 (1.16)
Invasive micropapillary carcinoma	1 (0.58)
Papillary carcinoma	1 (0.58)
Mucinous carcinoma	6 (3.47)
Secretory carcinoma	1 (0.58)
Medullary carcinoma	2 (1.16)
Metaplastic carcinoma	4 (2.31)
Malignant phyllodes	1 (0.58)
Stromal sarcoma	2 (1.16)
Metastatic adenocarcinoma	2 (1.16)
Lymphoma	0 (0)

IDC: Infiltrating ductal carcinoma, NOS: Not otherwise specified

Table 6: Age and sex distribution in benign and malignant neoplasm

Age interval (in years)	Benign neoplasm			Malignant neoplasm		
	Number of cases (%)			Number of cases (%)		
	Female	Male	Total cases	Female	Male	Total cases
11-20	41 (95.35)	2 (4.65)	43	0 (0)	0 (0)	0
21-30	44 (89.79)	5 (10.20)	49	16 (100)	0 (0)	16
31-40	29 (96.67)	1 (3.33)	30	39 (100)	0 (0)	39
41-50	12 (100)	0 (0)	12	64 (100)	0 (0)	64
51-60	0 (0)	0 (0)	0	43 (97.73)	1 (2.27)	44
61-70	3 (100)	0 (0)	3	25 (100)	0 (0)	25
71-80	1 (100)	0 (0)	1	5 (100)	0 (0)	5
81-90	0 (0)	0 (0)	0	1 (100)	0 (0)	1
Total	130 (93.98)	8 (6.02)	138	183 (99.52)	1 (0.48)	184

of cases negative for ER, PR, and Her2neu were 80, 82, and 81, respectively.

Table 8 shows the distribution of malignant lesions according to the combined ER/PR status. The number of tumors positive for both ER and PR was 30 (25.64%). Those which were positive for either ER or PR were very few. However, majority of the breast carcinomas in this study came out to be negative for both ER and PR.

Table 7: Distribution of cases according to ER, PR, and Her2neu status

IHC marker	Positive cases (%)	Negative cases (%)
ER	37 (31.62)	80 (68.38)
PR	35 (29.91)	82 (70.09)
Her2neu	36 (30.77)	81 (69.23)

IHC: Immunohistochemistry, ER: Estrogen receptor, PR: Progesterone receptor

Table 8: Distribution of cases according to ER and PR status

ER/PR status	Number of cases (%)
ER+PR+	30 (25.64)
ER+PR−	7 (5.98)
ER-PR+	6 (5.13)
ER-PR−	74 (63.25)

PR: Progesterone receptor, ER: Estrogen receptor

Table 9: Distribution of cases as per IHC group results

IHC status	Number of cases
ER+, PR+, Her2+	2 (1.70)
ER+, PR+, Her2−	27 (23.08)
ER+, PR−, Her2+	3 (2.56)
ER+, PR−, Her2−	4 (3.42)
ER−, PR+, Her2+	1 (0.85)
ER−, PR+, Her2−	7 (5.98)
ER−, PR−, Her2+	30 (25.64)
ER−, PR−, Her2−	43 (36.75)

IHC: Immunohistochemistry, PR: Progesterone receptor, ER: Estrogen receptor

Table 10: Comparison of distribution of cases according to age

Criteria	Mansoor (2001)[1]	Prajapati *et al*. (2014)[2]	Present study (2015)
Mean age in years	35.7 years	37.5 years	40.5 years

Table 11: Comparison of distribution of cases according to gender

Sex	Prajapati *et al*. (2014)[2]	Rahman *et al*. (2014)[3]	Present study (2015)
Male (%)	0.4	1	2.65
Female (%)	99.6	99	97.35

From Table 9, it can be concluded that majority of the cases in our study were negative for all the three receptors ($n = 43$, 36.75%), referred to as triple-negative breast carcinomas. The second majority group was ER−/PR−/Her2+ which comprised 30 cases forming a total of 25.64%. It was followed by ER+/PR+/Her2− group of tumors which occupied 23.08% ($n = 27$). Rest of the tumors constituted about 12% in total.

- In 17 cases of diagnostic dilemma from the histopathological point of view, IHC markers other than ER, PR, and Her2neu were applied. These were as follows:
  1. Use of E-cadherin was done in 8 cases for distinguishing between ductal and lobular carcinoma. Of these, 6 came out to be IDC-NOS while 2 came out to be mixed ductal and lobular carcinoma (Figure 28).
  2. Myoepithelial markers were applied in 8 cases to confirm invasion or pseudoinvasion in cases. These were 2 cases of breast papilloma and one each of ductal adenosis, radial scar, and fibroadenoma. In all these cases, myoepithelial markers were positive. In 3 cases, these markers were negative and were diagnosed as IDC (Figures 11, 12 and 15).
  3. There was one case of secretory carcinoma which was confirmed with the help of IHC markers. The results were positive for S100, polyCEA, and EMA whereas CK5/6, Type IV collagen, and gross cystic disease fluid protein (GCDFP) were negative (Figure 19).
  4. In one case of triple-negative IDC, basal cell IHC markers were applied and they came out to be positive, confirming the basal-like carcinoma of breast (Figure 24).
  5. Two cases had a differential diagnosis of lymphoma, melanoma, and neuroendocrine carcinoma. However, the respective markers, i.e., CD45, CD20, HMB45, synaptophysin, and chromogranin were found to be negative. Hence, the diagnosis of IDC-NOS was made (Figure 29).

DISCUSSION

In the present study, the age of patients ranged from 13 to 85 years, with a mean age group of 40.5 years (Table 10). Maximum number of cases were seen in the age group of 41-50 years (23.53%). This is probably because, in our study, malignant cases were more than the benign and inflammatory lesions combined. Hence, the load of breast lesions is tilted toward the higher age.

In the present study, out of 340 cases, only 9 (2.65%) cases were males and the remaining 331 (97.35%) cases were females (Table 11). The male-to-female ratio was 1:37.

Table 12: Comparison of incidence of inflammatory lesions

Criteria	Mansoor (2001)[1]	Aslam *et al*. (2013)[4]	Rahman *et al*. (2014)[3]	Present study (2015)
Percentage of cases (%)	10.7	11.8	19.24	4.70

Table 13: Comparison of distribution of benign lesions

Criteria	Mansoor (2001)[1]	Shanthi *et al*. (2011)[5]	Aslam *et al*. (2013)[4]	Rahman *et al*. (2014)[3]	Present study (2015)
Percentage of cases out of the total number of cases (%)	56.87	68	75.30	76.66	40.59
Most common (percentage of total benign cases)	Fibroadenoma (66.86%)	Fibroadenoma (70.83%)	Fibroadenoma (71.3%)	Fibroadenoma (70.88%)	Fibroadenoma (74.64%)

Table 14: Comparison of distribution of malignant breast lesions

Criteria	Mansoor (2001)[1]	Shanthi *et al*. (2011)[5]	Aslam *et al*. (2013)[4]	Rahman *et al*. (2014)[3]	Present study (2015)
Percentage of cases out of the total number of cases	32.42	24	11.80	23.34	54.71
Most common (percentage of total malignant cases)	IDC-NOS (87.70)	IDC-NOS (78.57)	IDC-NOS (100)	IDC-NOS (94.59)	IDC-NOS (80.64)

NOS: Not otherwise specified, IDC: Infiltrating ductal carcinoma

Table 15: Comparison of distribution of cases according to ER, PR, and Her2neu status

Marker	Ambroise *et al*. (2010)[6]	Ali *et al*. (2014)[8]	Doval *et al*. (2015)[7]	Present study
ER+ (%)	59	78.7	30.12	31.62
PR+ (%)	51	76.4	27.95	29.91
Her2neu+ (%)	27	13.2	23.0	30.77
Triple negative (%)	20.67	15.5	23.8	36.75

PR: Progesterone receptor, ER: Estrogen receptor

Universally, the breast cancer gender ratio of male:female is in the range of 1:99. The present study had one case of male breast cancer which automatically pushed the incidence to above 2%. This is very much in accordance with the past studies done by Prajapati *et al*.[2] in 2014 and Rahman *et al*.[3] in 2014.

In the present study, out of the 340 cases studied, inflammatory lesions' group contained 16 cases (4.70%) (Table 12). Of these, the most common inflammatory lesion was granulomatous mastitis ($n = 7$, 43.75%) and the most common age group was 21-30 years. The probable reason behind this is that most of the inflammatory lesions in our tertiary setup are diagnosed by fine-needle aspiration cytology and biopsy is not sent to the laboratory.

In the present study, out of the 340 cases, 138 were benign (40.59%). Most of the benign cases fell in the age group of 21-30 years ($n = 49$) closely followed by 11-20 years ($n = 43$). In accordance with the past studies, in the present study also, the most common benign breast lesion was found to be fibroadenoma constituting a total of 103 cases (74.64% of the total benign breast diseases and 30.29% of the total breast lesions). Fibroadenoma occupied 66.86% of the total benign cases studied by Mansoor, 70.83% in a study by Shanthi *et al*., 71.3% in a study by Aslam *et al*., and 70.88% in a study carried out by Rahman *et al*. This clearly concludes that fibroadenoma is the most common benign breast lesion. (Table 13)

In the present study, out of the 340 cases, 186 were malignant (54.71%). The finding that breast cancer was slightly more common than benign breast lesions in this study is at variance with most studies, probably because being a tertiary health-care facility, most of the patients admitted here are referred as malignant cases from the periphery and the growing environment of private-owned hospitals by general practitioners where most of the benign lesions are probably managed. In this study, most of the malignant cases were found to be in the age group of 41-50 years ($n = 64$, 82.5%). The most common breast cancer was found to be IDC-NOS, constituting a total of 150 cases (80.64%). This is similar to the past studies conducted by Mansoor in 2001, Shanthi *et al*. in 2011, Aslam *et al*. in 2013, and Rahman *et al*. in 2014. From the above data, we can conclude that IDC-NOS is the most common breast carcinoma. However, a large variety of cases were seen in this study in variable numbers. (Table 14)

Expression of both ER and PR was specifically nuclear while positive expression of HER2 was demonstrated as continuous membranous immunoreaction. In our study, total cases positive for ER, PR, and Her2 were 31.62%, 29.91%, and 30.77%, respectively. These results were 59%, 51%, and 27% in the study conducted by Ambroise *et al*., 78.7%, 76.4%, and 13.2% as per Ali *et al*., and 30.12%,

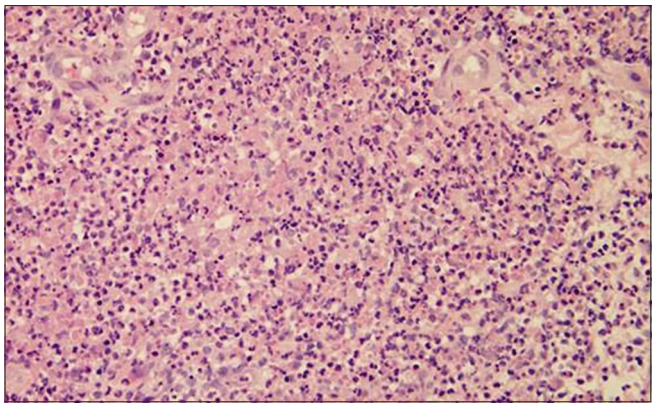

Figure 1: Breast abscess showing plenty of neutrophils and obiteration of lobular pattern (H and E; ×100)

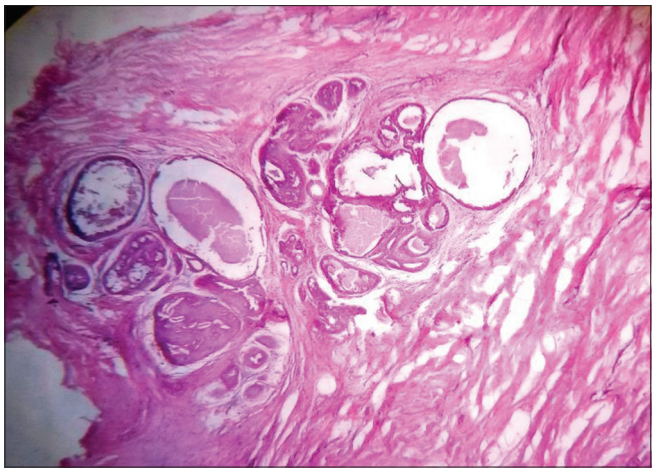

Figure 2: Duct ectasia showing dilated large ducts with fatty detritus in lumen and fibrous thickening of wall (H and E; ×100)

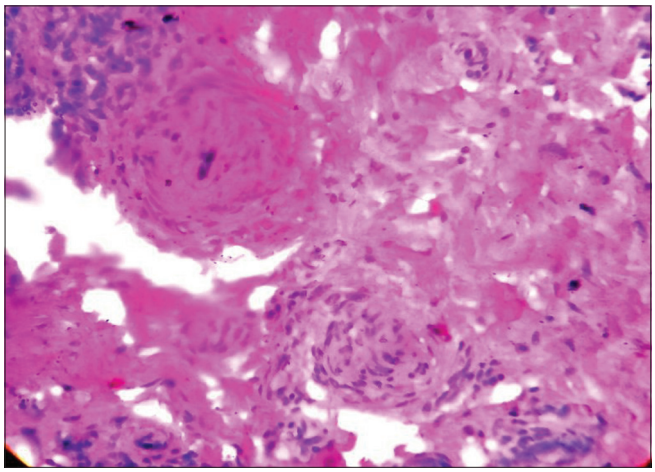

Figure 3: Granulomatous mastitis showing foreign body granulomas (H and E; ×100)

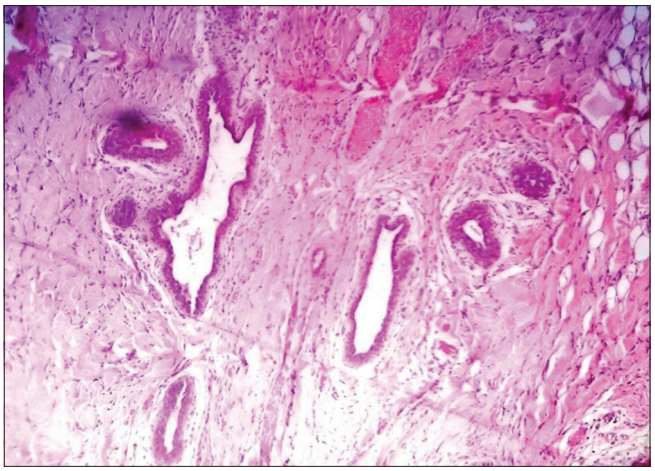

Figure 4: Gynecomastia showing epithelial proliferation surrounded by edematous stroma (H and E; ×100)

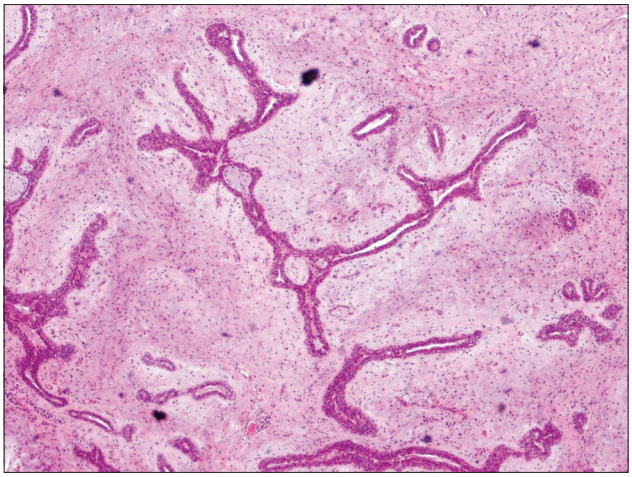

Figure 5: Fibroadenoma showing tubular glandular formations lined by ductal epithelial and myoepithelial cells surrounded by loose connective tissue (H and E; ×100)

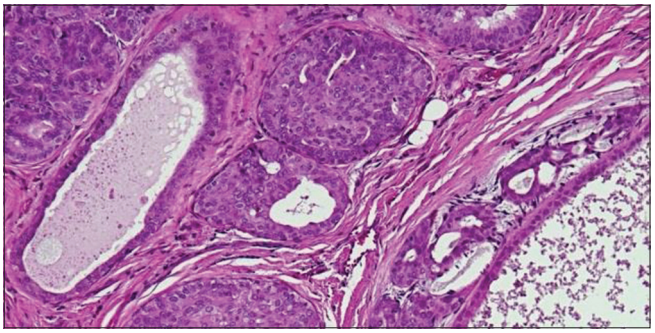

Figure 6: Fibrocystic disease of breast showing cystic dilation, apocrine metaplasia and florid ductal hyperplasia (H and E; ×400)

27.95%, and 23% in the study done by Doval *et al.* for ER, PR, and Her2 positivity, respectively. (Table 15)

In our study, as far as triple-negative breast carcinomas are concerned, 36.75% of the total breast malignancies were negative for ER, PR, as well as Her2neu. They presented with a mean age group of 50 years, and majority of the patients were aged between 40 and 65 years. Similar results were obtained by Doval *et al.* (2015)[7]. According to their study, the patients with triple-negative tumors presented at a mean age of 50 years and majority of the patients (51.5%)

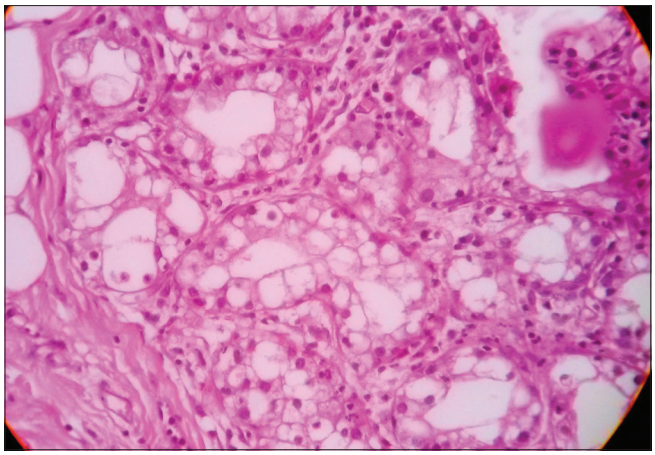

Figure 7: Lactating adenoma of breast showing proliferated glands lined by actively secreting cuboidal cells with marked cytoplasmic vacuolation (H and E; ×400)

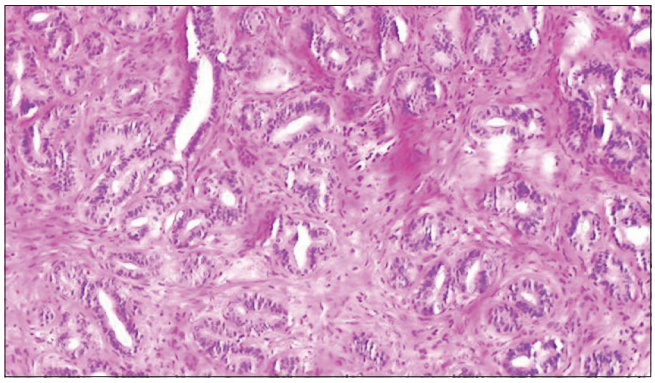

Figure 8: Tubular adenoma showing closely packed uniform small tubules lined by epithelial and myoepithelial cells (H and E; ×100)

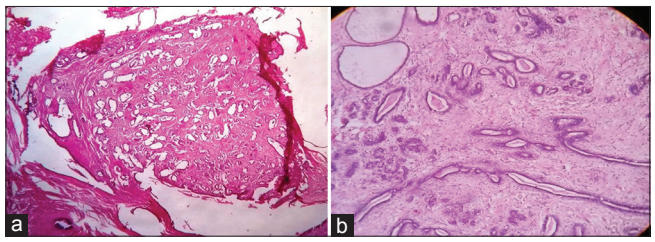

Figure 9: (a) Histomorphology of sclerosing adenosis of breast- showing nodular architecture (H and E; ×40), and (b) atrophy of epithelial cells, cystically dilated glands and increased intralobular stroma (H and E; ×100)

were in the age group of 20-50 years as compared to the non-triple-negative tumor group. In the study conducted by Basu *et al.* (2008)[9], "triple-negative" group comprised 14.08% of the total study population. The age of the patients with this subtype of tumor ranged from 33 years to 75 years.

- There were 17 cases in our study where IHC markers other than ER, PR, and Her2neu were applied. These are discussed as follows:

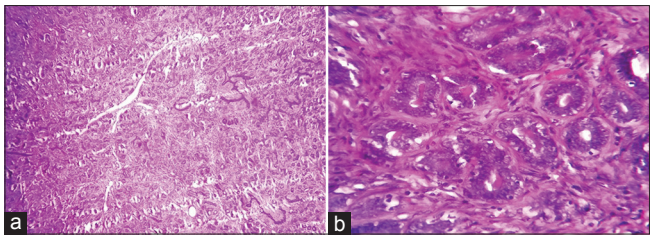

Figure 10: (a and b) Histomorphology of florid ductal adenoma of breast- showing effaced architecture due to increased proliferation of glands (H and E; a - ×100 and b - ×400)

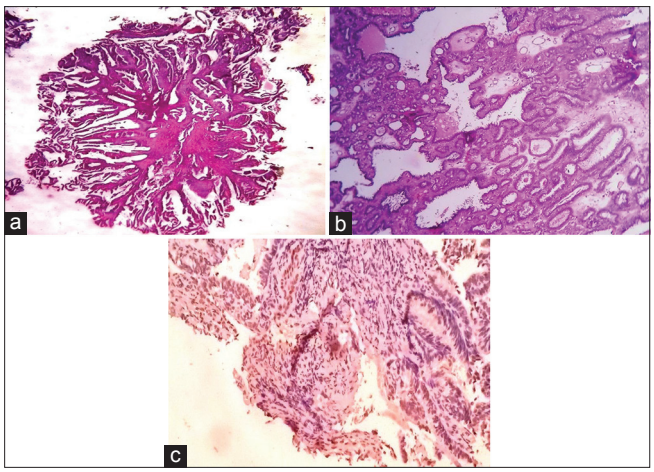

Figure 11: (a) Histomorphology of benign intraductal papilloma of breast- showing complex arborizing architecture (H and E; ×40), and (b) well defined dual cell composition (H and E; ×100), and (c) well defined row of myoepithelial cells highlighted by p63 positivity

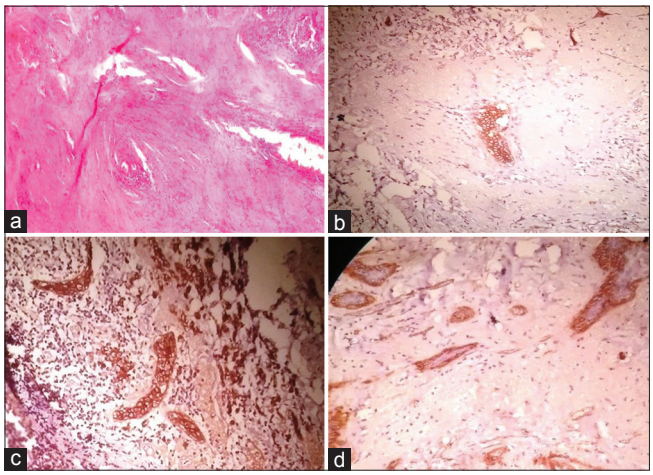

Figure 12: (a) Histomorphology of radial scar of breast- showing stellate shaped central fibrous core and variable degree of epithelial distortion and proliferation (H and E; ×100), (b) The entrapped ductular cells show positivity for CK7, (c) Pankeratin, and (d) myoepithelial cells stain for S100

I. In 8 cases, E-cadherin was applied to differentiate between ductal and lobular carcinoma. Of these, 6 came out to be IDC-NOS while 2 came out to be mixed ductal and lobular carcinoma. According

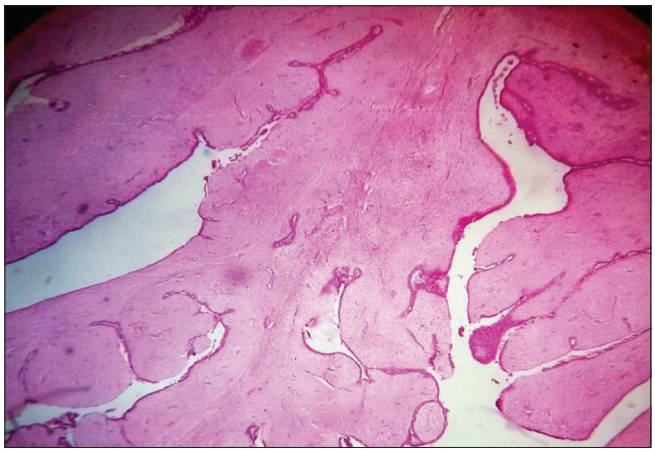

Figure 13: Benign phylloides tumor of breast showing stromal hypercellularity and benign glandular elements (H and E; ×100)

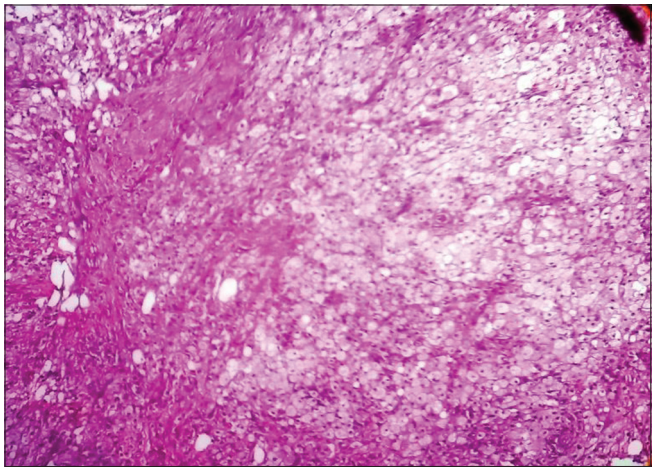

Figure 14: Galactocoele showing dilated, anastomosing, epithelium lined channels with secretory activity (H and E; ×400)

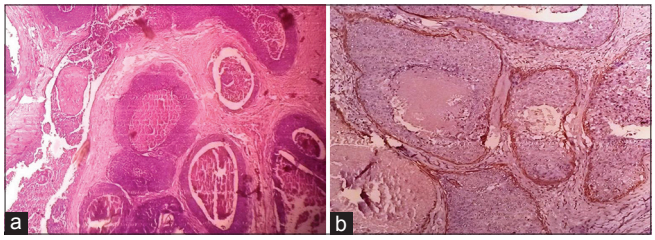

Figure 15: (a) Histomorphology of ductal carcinoma *in situ* of breast, Comedo type showing ducts with solid growth of large, pleomorphic tumor cells alongwith comedo type of central necrosis (H and E; ×100), and (b) presence of myoepithelial cells confirmed by calponin

to an article published in the Arch Pathology Laboratory Medicine, 2008[10], E-cadherin, a cell-cohesion protein encoded by a gene on chromosome 16q22.1, is the current marker of choice to help discriminate between lobular and ductal carcinoma. The majority of usual ductal carcinomas express cytoplasmic E-cadherin, whereas most *in situ* and

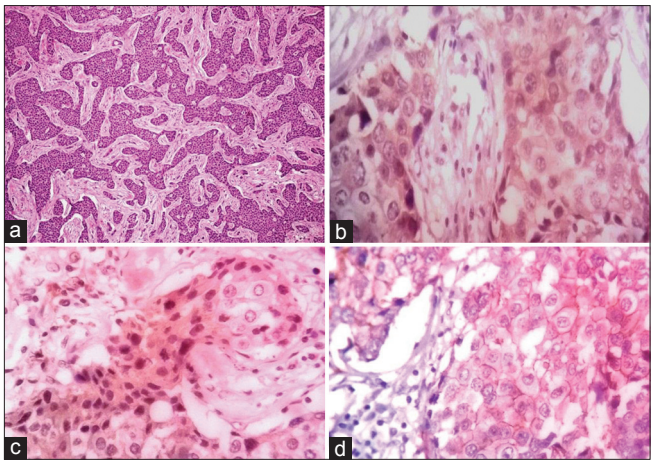

Figure 16: (a) Histomorphology of invasive ductal carcinoma of breast- not otherwise specified showing large pleomorphic ductal epithelial cells in sheets and cords having prominent nuclei with surrounding desmoplasia (H and E; ×100), (b) Estrogen receptor positivity, (c) Progesterone receptor positivity, and (d) Her2 positivity

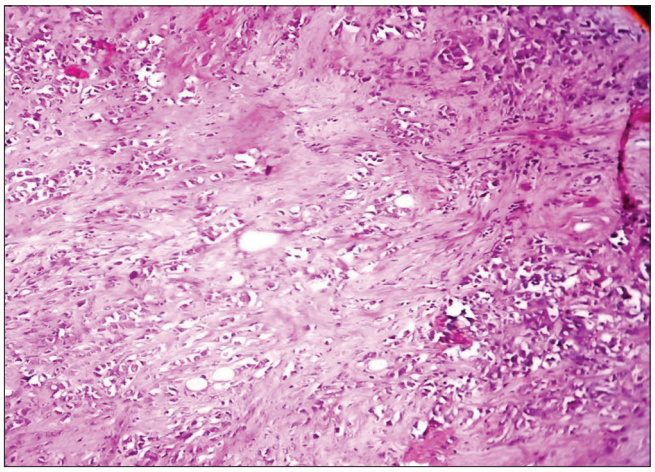

Figure 17: Mixed infiltrating ductal carcinoma and ILC of breast- right corner shows infiltrating ductal carcinoma component and central part shows ILC component

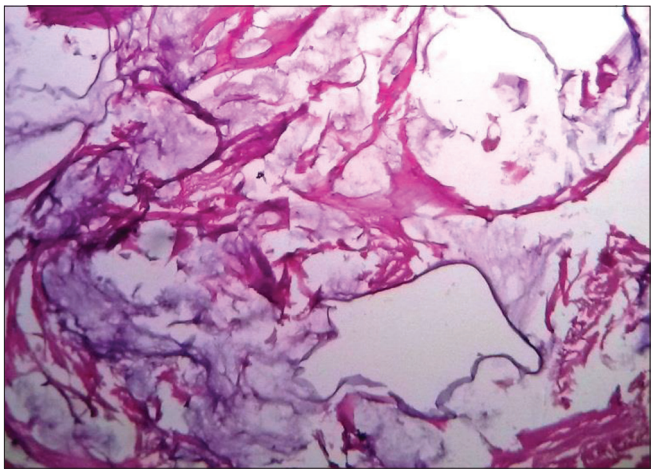

Figure 18: Mucinous carcinoma of breast showing tumor cells in pools of extracellular mucin (H and E; ×100)

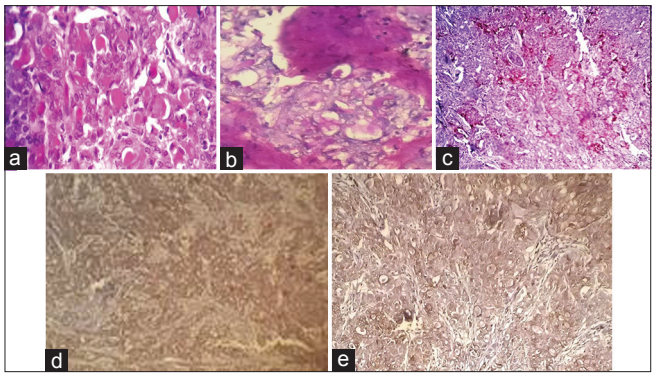

Figure 19: (a) Histomorphology of Secretory carcinoma of breast- showing tubuloalveolar formations lined by cells with vacuolated cytoplasm forming lumina filled with eosinophilic PAS positive secretions (A; H and E; ×400), (b) PAS stain, (c) Strong reactivity for S100, (d), polyCEA, and (e) EMA

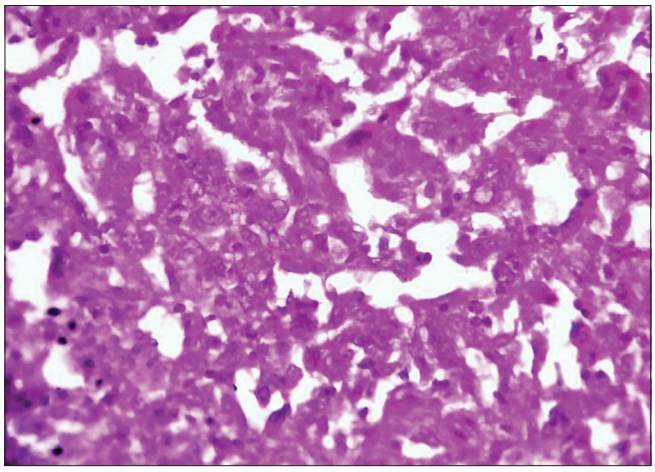

Figure 20: Medullary carcinoma of breast showing large tumor cells in syncytial pattern surrounded by stroma infilterated by lymphocytes (H and E; ×400)

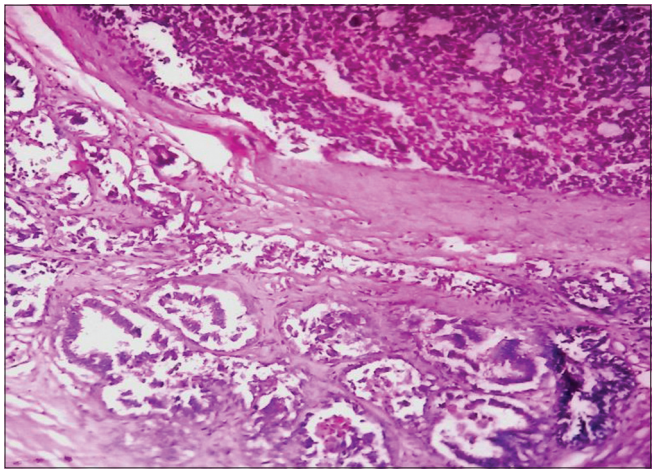

Figure 21: Papillary carcinoma of breast showing papillary formations with fibrovascular core (H and E; ×400)

invasive lobular carcinomas, both classic and pleomorphic types, lack expression.

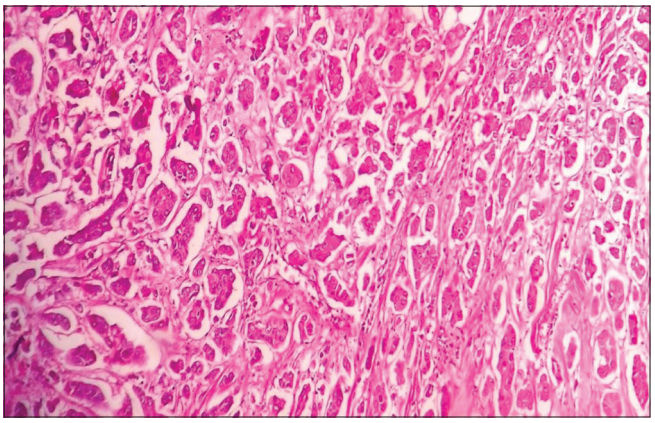

Figure 22: Invasive micropapillary carcinoma of breast showing pseudopapillary structures lacking fibrovascular core and tubular structures free floating in clear empty spaces (H and E; ×400)

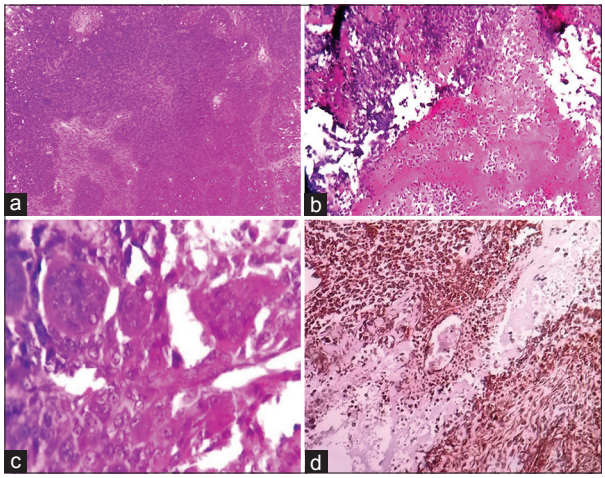

Figure 23: (a) Histomorphology of Metaplastic carcinoma of breast- showing squmaous differentiation (H and E; ×100), (b) chondroid differentiation (H and E; ×100), (c) osteoclastic giant cells (H and E; ×400), and (d) vimentin positivity

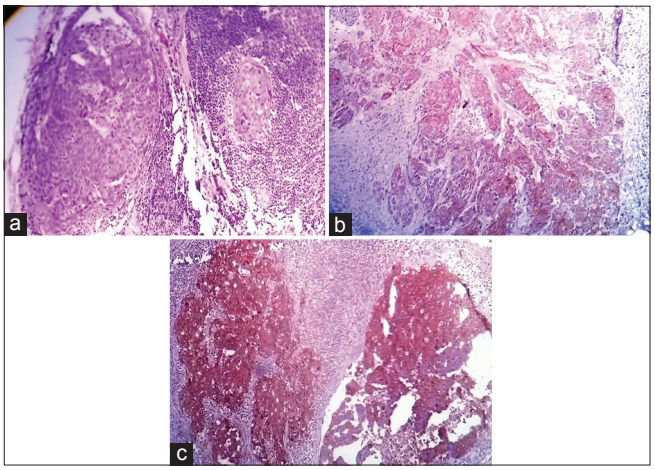

Figure 24: (a) Histomorphology of Basal like infiltrating ductal carcinoma of breast- showing high grade tumor with necrosis and lymphocytic response (H and E; ×100), and (b) positivity for basal like keratins CK5/6, and (c) EGFR, (d) These are triple negative tumors

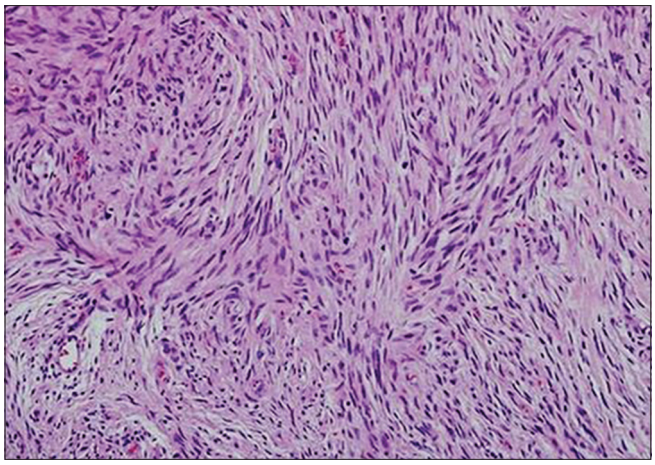

Figure 25: Malignant phylloides tumor of breast showing markedly hypercellular stroma (H and E; ×100)

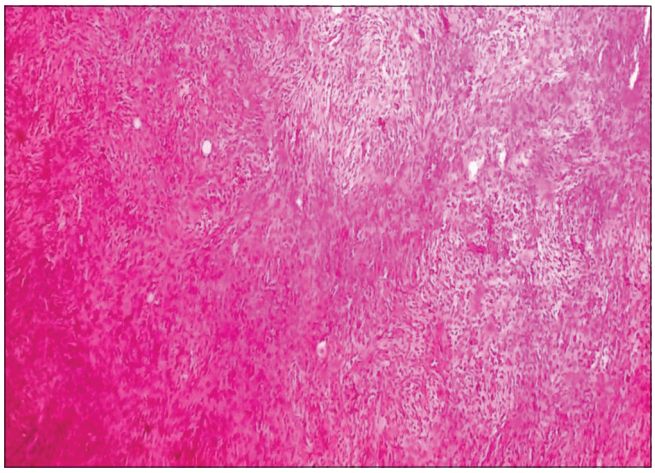

Figure 26: Stromal sarcoma of breast

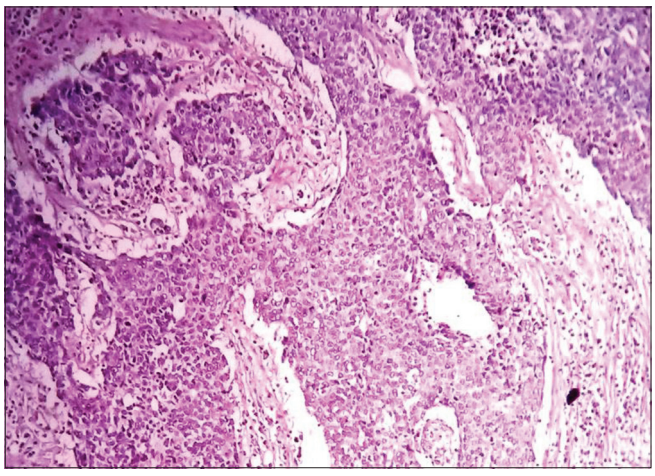

Figure 27: Metastatic adenosquamous carcinoma of breast

II. Myoepithelial markers were applied in 8 cases to confirm invasion or pseudoinvasion in cases. About 5 cases showed the presence of myoepithelial cells confirming the benign nature of the lesion. These were 2 cases of breast papilloma and one case each of ductal adenosis, radial scar, and fibroadenoma. In 3 cases, these markers were negative and were diagnosed as IDC. In addition to penetrating basement membrane, invasive carcinomas lack the mucoepidermoid carcinoma layer that normally surrounds benign breast glands. This observation is the basis of using myoepithelial markers to distinguish invasive carcinoma from infiltrative mimics.[11,12]

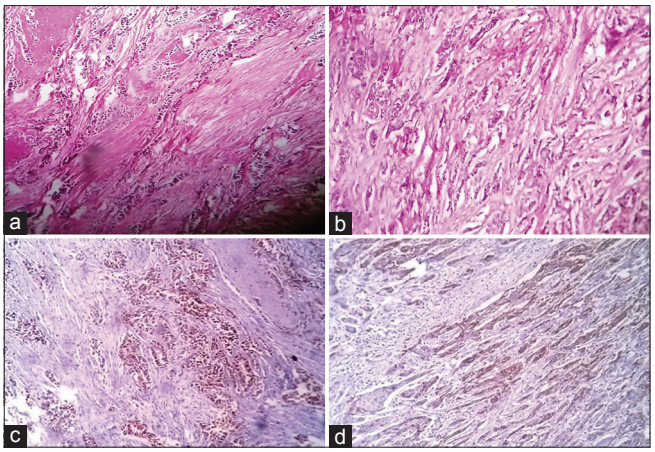

Figure 28: Diagnostic dilemma between infiltrating ductal carcinoma and ILC of breast (H and E; a and b), solved by E cadherin positivity, (c and d) which represents presence of cohesiveness in infiltrating ductal carcinoma

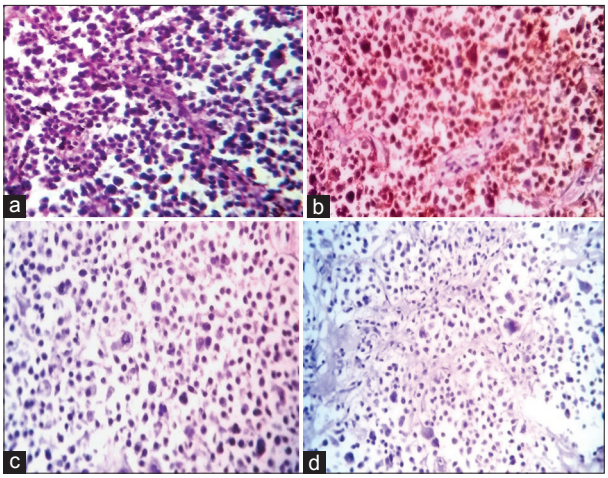

Figure 29: (a) Diagnostic dilemma between poorly differentiated infiltrating ductal carcinoma of breast and melanoma or lymphoma (H and E; ×400), (b) was solved by E cadherin positivity, (c), lack of HMB45 reactivity, (d) lack of CD20 reactivity

III. One case of secretory carcinoma was confirmed by positive S100, polyCEA, and EMA. GCDFP usually shows variable positivity in secretory carcinoma. However, in our case, it was negative. In the past, some of the cases of microglandular adenosis were misdiagnosed as secretory carcinoma. Hence, Type IV collagen and CK5/6 were applied but they came out to be negative, confirming the absence of basement membrane.[13]

IV. In another case which was negative for ER, PR, and Her2 (triple negative), epidermal growth factor receptor and CK5/6 were applied which showed positive results. Such kind of ICH profile is shown by basal-like carcinomas. According to a study done by Abd El-Rehim *et al.*[14] on the expression of luminal and basal cytokeratins in human breast carcinoma, the tumors expressing a basal phenotype or a mixed basal/luminal phenotype have a worse prognosis. The basal phenotype correlates with high-grade IDC-NOS, as well as with medullary carcinomas, adenoid cystic carcinomas, and squamous differentiation (metaplastic carcinomas).

V. High-grade IDC-NOS can be difficult to distinguish from lymphoma, melanoma, and neuroendocrine carcinoma. In this study, 2 such cases were confirmed immunohistochemically by their respective markers, i.e., CD45, CD20, HMB45, synaptophysin, and chromogranin. These all were found to be negative.

CONCLUSION

This study comprised 340 cases of breast lesion. The cases presented to SSG Hospital and Medical College Baroda. The surgical specimens were then evaluated histopathologically, and 124 cases were analyzed ICH.

Based on the above study, the following conclusions were drawn:

- The age group of patients ranged from 13 to 85 years, with a mean age of 40.5 years.
- Male-to-female ratio in this study was 1:37.
- The incidence rate of inflammatory lesions was 4.7%.
- The most common presenting age group for inflammatory lesions was 21-30 years (9 cases).
- The most common type of inflammatory lesion of breast was granulomatous mastitis (7 cases, 43.75% of the total cases of inflammatory lesions).
- The incidence rate of benign lesion was 40.59%.
- The most common presenting age group for benign breast lesion was 21-30 years (49 cases).
- The most common histopathological type of benign breast lesion was fibroadenoma (103 cases, 74.64% of the total benign breast diseases).
- The incidence rate of malignant lesions was 54.71%.
- Most of the malignant cases presented at the age group of 41-50 years (64 cases).
- The most common histopathological subtype of breast malignancy was IDC-NOS type (150 cases, 80.64% of the total cases with malignant lesion).
- Two cases of borderline phyllodes were found.
- Percentage of ER positivity in this study was 31.62%.
- Percentage of PR positivity in this study was 29.91%.
- Percentage of Her2neu positivity in this study was 30.77%.
- Triple-positive breast cancers in this study were 1.7%.
- Triple-negative breast cancers in this study were 36.75%.
- E-cadherin is helpful in differentiating IDC from ILC as in 8 cases in our study. Loss of E-cadherin indicates lobular type.
- Myoepithelial markers can aid in ruling out invasion or pseudoinvasion. This was used in 8 cases in our study. Positivity of these markers confirms the benign nature of the lesion.

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