



Cytomorphological determinants for diagnosing breast lesions

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ABSTRACT

To find out combinations of the most effective cytomorphologic features to diagnose a case of breast lesion. To measure accuracy of combination of criteria used to diagnose a specific breast lesion. To determine the utility of FNAC and its continuing role in diagnosing breast lesions. Retrospective analysis of 460 patients with breast lesion referred to the Department of Pathology, Mahavir Cancer Institute and Research Centre, Patna, for FNAC and later on confirmed by HPE were included in the study, irrespective of age. Lumpectomy provided the diagnosis in 107 cases. In the present study we found that the combination of criteria assessed, enabled the segregation of breast lesions into different categories with high significance values, which will be more effective in diagnosis of disease. Combined criteria provide better results and a practical way of expressing our opinion as more objective and precise diagnosis which is good from clinician's point of view too.

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Introduction

Cytology has an established role in diagnosing the symptomatic breast diseases. Fine needle aspiration cytology (FNAC) was first introduced in 1930 [1]. FNAC has been found to have sensitivity ranging from 82% to 97.5% and specificity of more than 99% [2,3,4]. FNAC of breast lumps is an important part of triple assessment (clinical examination, imaging, and FNAC of palpable breast lumps). Sometimes it is difficult to determine whether a suspicious lump is benign or malignant simply from clinical examination. Therefore a method of definitive diagnosis of patients who present with breast lumps at the outpatient clinic is needed. This method must be accurate, easy to perform and reproducible. It must also be acceptable to the patient, can be carried out in a busy clinic setting and must not require too much preparation or expensive equipment. FNAC of breast lump is an accepted and established method to determine the nature of the lump and it may play an important role when it is difficult to determine the nature of breast lump by clinical examination. Though histo-pathological diagnosis is a universally accepted confirmatory mode of diagnosis but FNAC has proved to be a very rapid, simple, non-invasive and cost-effective procedure in comparison. Thus, a good diagnostic tool for various breast diseases. Self-assessment, clinical examination, mammography and FNAC are proven pillars of screening program in different parts of the world [5,6,7]. But very little recorded information is available about the definitive cytomorphological features required for diagnosing a specific disease entity.

There is a need to set up standard protocol for diagnosing breast disease by cytomorphological variables. We present the spectrum of cytological features from 460 breast lesion cases with an aim to define the combinations of features most effective in diagnosing a particular entity.

Materials and Methods:

This cross-sectional study was conducted from Sep 2011 to Feb 2013 over a period of 18 months in the Department of Pathology, Mahavir Cancer Institute and Research Centre,

Patna. Retrospective analysis of 460 patients with breast lesion referred to the Department of Pathology for FNAC and later on confirmed by HPE were included in the study, irrespective of age. Lumpectomy provided the diagnosis in 107 cases. Altogether eight cytomorphological variables were counted for the evaluation of each case.

FNAC is a rapid and non-invasive procedure widely used as an alternative to excision biopsy of palpable breast lesions [8,9,10]. The reason for loss of popularity of FNAC includes high error rate due to lack of experienced hands. There is always a chance of misinterpreting a case of breast disease as "negative for disease", because aspirated material may not represent the diseased site correctly. Thus, applying the correct technique of aspiration is as important as correct interpretation of the sample. However, in experienced hands, FNAC is a highly accurate diagnostic procedure.

FNAC was done with 21 gauge disposable needle and a 10cc disposable syringe. The aspirates were deposited on to clean glass slides, smeared, air-dried and stained with May-Grunwald Giemsa (MGG) stain. Smears wet-fixed with 95% ethanol were stained with Papanicolaou stain. Smears were then studied for cytological details.

Histological material obtained from lumpectomy or mastectomy specimens were fixed in formalin and processed routinely. They were stained with Hematoxylin and Eosin and examined.

Immuno-staining of cytosmears would prove to be a good diagnostic adjunct to morphological diagnosis of malignant lesions. Molecular testing can be used as an aid in the assessment of breast lesion cases. Various evidences have emerged in support of the fact that unique molecular findings in case of Breast Cancer influence the prognosis and therapeutic decisions to a great extent [11, 12, 13]. Molecular testing can be applied on cytological smears too. Like for patients with T1 and T2 Breast Carcinoma, a positive cytological diagnosis avoids sentinel lymph node procedures [14,15] and helps in monitoring therapeutic response in Breast Carcinoma patients receiving

neo-adjuvant chemotherapy. Statistical analysis was performed with Graph Pad software. The data were analyzed for mean±SEM and P value.

Results

The study population included 460 females, irrespective of their age. The mean age of patient was 43.93± 0.708 years (Mean ± SEM), as shown in Figure 1. Various diagnostic categories used in FNAC of the breast along with the number of cases reported in each subtype are summarized in Table-1.

Table 1: Number of cases reported under each diagnostic category

DIAGNOSIS	NO. OF CASES
1.Benign Non-proliferative Breast Disease -Fibrocystic disease & Simple Cyst	38
2.Benign Proliferative Disease Without Atypia	32
3.Benign Proliferative Disease With Atypia	28
4.Benign Tumor -Fibroadenoma	63
-Phyllodes tumor, benign	05
5. Malignant Tumor -Ductal Carcinoma	285
-Medullary Carcinoma	04
-Apocrine Carcinoma	02
-Others	03
TOTAL CASES	460

Table 2: Percentage of each cytomorphological variable found in particular diagnostic category. BNPD: Benign Non-proliferative breast Disease; BPD: Benign Proliferative Disease without Atypia; BPDWA: Benign Proliferative Disease Without Atypia; BENIGN: Benign tumor (Fibroadenoma); MALIGNANT: Ductal Carcinoma

	BNPD	BPD	BPDWA	BENIGN	MALIGNANT
Cellularity-low		83			
Cellularity-increased			71	51	89
Cellularity-markedly increased					86
Apocrine cells		96			
Foam cells		80			
Cell pattern-poorly cohesive cluster					86
Cell pattern-streaming pattern			88		
Cell pattern-cribriform pattern				66	
Background-bare bipolar nuclei			71	77	81
Background-fibromyxoid stroma					80
Myoepithelial cell -seen		35	71	66	87
Myoepithelial cell -not seen					75
Nuclear atypia- mild			11		12
Nuclear atypia- moderate to severe				74	3
Background- single tumor cell					71

Due to the lack of definitive cytologic features to distinguish between in-situ and invasive breast carcinoma [16-18], we omitted the use of ‘in-situ’ and ‘invasive’ terms while categorizing the malignant lesions on FNAC. The relationship between cytological parameters and the disease diagnosed is summarized in Table-2, more specifically the percentage of cases having a particular criteria is tabulated. Only ductal carcinoma cases are considered in morpho-analysis of malignant lesions because of lesser number of cases of other malignant subtypes.

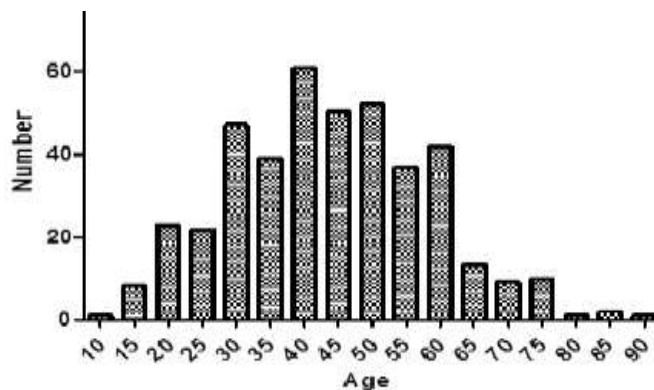


FIGURE 1: Age groups involved by breast diseases.

Discussion

FNAC of breast lump is an accepted and established method to determine the nature of breast lump with high degree of accuracy [19-22]. The application of FNA for the diagnosis of palpable breast masses was first introduced by Martin and Ellis [23] in 1930, and since then, it has been established as an important tool in the evaluation of breast lesions. Now-a-days, FNAC is being performed as a pre-operative test to evaluate the breast lump. A study of Khatun et al. [24] in the year 2000 evaluated the accuracy of FNAC on 310 patients presented with palpable breast lumps showed a very high sensitivity, specificity and accuracy. FNAC can prevent unnecessary surgery also. 95% accuracy in preoperative diagnosis of mammary cancer by clinic-cytological combination was reported in a study [25]. But still there is controversy in the literature regarding the exact features needed for diagnosing a particular cytological entity.

In our study we found that with regard to benign non-proliferative breast disease if both the criteria of poorly cellular smear and foam cells along with apocrine cells are counted in combination, there is high possibility of diagnosing it.

The most useful cytological detail associated with BPD without atypia is presence of ductal cell cluster in streaming pattern (i.e. with no holes) with bare bipolar nuclei in background ($p < 0.01$). The diagnostic significance is increased if criterion of mild-mod atypia and presence of myoepithelial cells is used in combination.

If the criteria of increased cellularity with cribriform pattern ($p < 0.0001$) is seen in combination with mod-severe nuclear atypia, there is more possibility of a BPD with atypia.

Presence of myoepithelial cells interspersed in ductal cell cluster with fibromyxoid stroma in background ($p < 0.0001$) and increased cellularity of the smear ($p < 0.001$) were selected as the highly significant cytomorphological variables for diagnosis of fibroadenoma (Figure 2, 3). Fibroadenoma usually display sheets of ductal epithelium and myoepithelial cells reflecting the histological features. Occasionally, fibroadenomas may display cytological atypia [26].

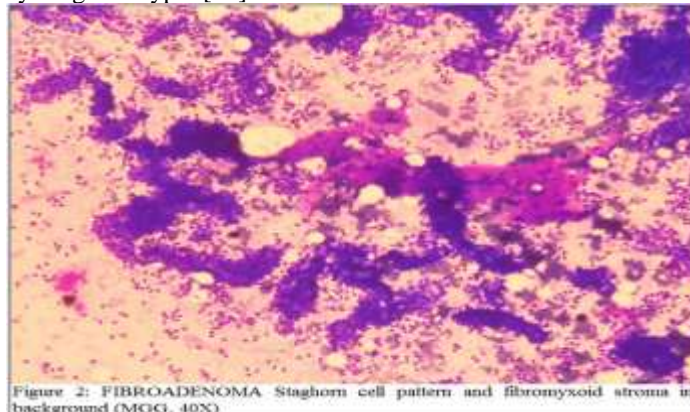


Figure 2: FIBROADENOMA Staghorn cell pattern and fibromyxoid stroma in background (MGG, 40X)

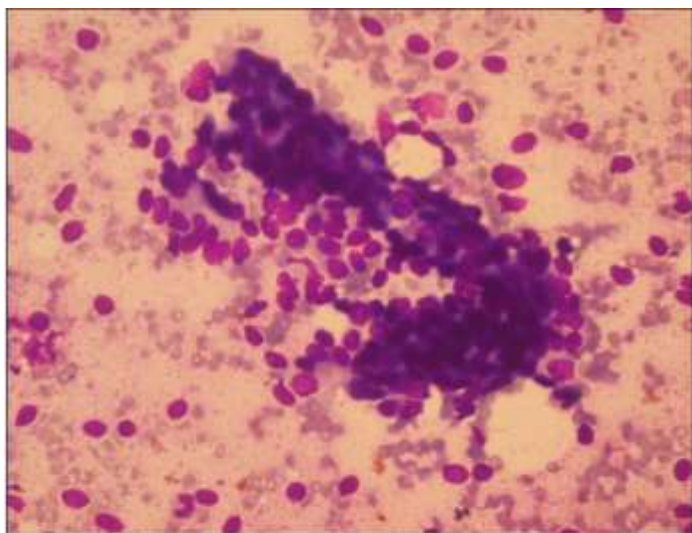


Figure 3:FIBROADENOMA Myoepithelial cell interspersed in ductal cell cluster (MGG,400X)

In our study, absence of myoepithelial cells interspersing in cell cluster, associated with presence of single tumor cells in background, poorly cohesive cell arrangement (Figure 4, 5) and moderate-severe atypia favored the diagnosis of ductal carcinoma. When the aforesaid 4 variables were used in combination there was less chance of missing a diagnosis of ductal carcinoma. High cellularity, poorly cohesive cells, pleomorphism, hyperchromasia, prominent nucleoli, and necrosis may strongly suggest cancer [27]. Complete sensitivity of FNAC diagnosis of grade 1 breast carcinomas is approximately 93% [28], which is only slightly lower than reported in all materials [29]. Thus FNAC is an effective method of diagnosing carcinoma of the breast and can prevent unnecessary surgery for benign disease. This framework provided a practical way of expressing our opinion as more objective and precise diagnosis which is good from clinician's point of view too, in reference of providing better management services. The role of other ancillary techniques like immunohistochemistry can't be underestimated, since they have an important role to play in deciding management protocols for the patient [30].

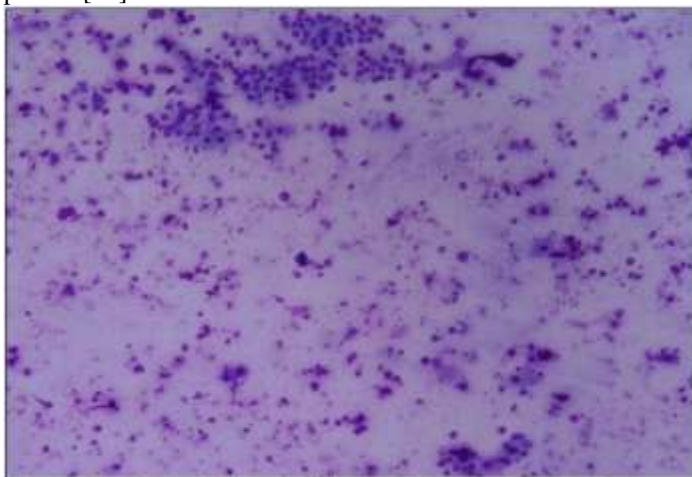


Figure 4: Poorly cohesive ductal cells in ductal carcinoma (MGG, 40X)

In the present study we found that the combination of criteria assessed, enabled the segregation of breast lesions into different categories with high significance values. Though for diagnosing a particular entity one of the single variable was found useful. This methodology has great potential for improving the evaluation of breast disease, especially borderline cases.

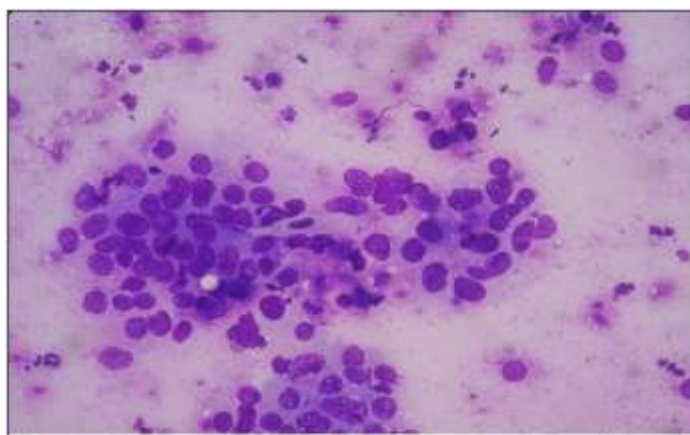


Figure 5: Absence of myoepithelial cells in poorly cohesive cluster of malignant ductal cells (MGG, 400X)

We conclude that the breast disease categorization into objective and precise entities ensures better management services too. In addition to the role in diagnosis, the cytosmears may prove beneficial for molecular analysis. It would not be far-fetched to say that molecular analysis of cyto-smears will become a routine procedure certainly in future.

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