PALPABLE BREAST LESIONS – CYTOMORPHOLOGICAL ANALYSIS AND SCORING SYSTEM WITH HISTOPATHOLOGICAL CORRELATION

DISSEPTION SUBMITTED FOR

M.D. DEGREE EXAMINATION

BRANCH – III (PATHOLOGY)

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CERTIFICATE

This is to certify that Dr. D. SHEEBA is a bonafide student of the Department of Pathology, Kilpauk Medical College, Chennai and this study titled “Palpable Breast Lesions – Cytomorphological Analysis and Scoring system with Histopathological Correlation” is the original work done by her for her dissertation towards the partial fulfillment of requirements for the M.D(Pathology) Degree 2006 – 2009.

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Introduction
Introduction

Fine Needle Aspiration Cytology has been used to diagnose Breast Cancer for over 50 years. In recent years, it has gained acceptance as a diagnostic tool in assessment and management of mammary lesions.

With the advent of mammography and Ultrasound, these investigations were used to recognize breast lesions. Regardless of the sex of the patient and the method used to diagnose the lesion, cytological techniques play an important role in the diagnosis of breast lesions.

Fine Needle Aspiration Cytology is successful in identification of benign and malignant breast lesions, but its role in proliferative breast lesions is poorly defined. To expand the role of FNAC in diagnosing of proliferative breast lesions, the analysis of cytomorphological features of proliferative breast lesions in conjunction with cytological scoring system proposed by Masood et al and with histopathology was done.

Relative Risk of developing Invasive breast cancer from carcinoma in situ, from proliferative breast disease with atypia, proliferative breast disease without atypia and non proliferative breast disease is of the order of 8 – 10, 4 – 5 , 1.5 – 2 , and 1 respectively\(^1\). Hence, it is very important to identify proliferative breast disease.
This study was undertaken to categorise the breast lesions into four categories depending on nuclear dissociation, myoepithelial cells, pleomorphism of cells, anisonucleosis, nuclear chromatin and nucleoli. The four categories are

1. Non-Proliferative Breast Disease,
2. Proliferative Breast Disease without Atypia
3. Proliferative Breast Disease with Atypia
4. Carcinoma.

The categorization of proliferative breast lesions by FNA remains a challenge to the pathologist and the cytologic criteria need to be further defined and assessed. Decreasing the number of diagnostic categories is likely to improve the correlation between the cytologic and histologic diagnoses without compromising patient management.
Aims of the Study
Aims of the Study

The study aims to correlate the Cytomorphological Diagnosis and the Modified Masood’s scoring system with Histopathological Diagnosis in palpable Breast Lesions.

Age distribution of case under different categories of diagnoses are also studied.
Review of Literature
Review of Literature

The use of cytology for diagnosis of breast lesions dates back to the early 1930s, when Martin and Ellis first reported their experience with FNA at the Memorial Hospital for Cancer and Allied Diseases in New York\(^2\). This was followed in the late 1940’s and the early 1950’s by Adair and Godwin. However, not until the Europeans reported a large series of a number of FNAC of the breast was Aspiration Cytology shown to be a valuable and accurate diagnostic procedure.

The sensitivities of Open Biopsy and FNAC are comparable (99% for the former and 96.2% for the latter). The specificity of FNAC is better (98.8% vs 85.5% for open biopsy).\(^3\)

Aspiration biopsy of breast lesions has become a quasi-routine clinical procedure, often replacing per-op tissue biopsy. FNAC is the least expensive method and the most rapid and most versatile of all approaches.

FNAC can be used for palpable masses that may be either solid or cystic or nonpalpable lesions detected by Mammography, Ultrasound or MRI.

It is the consensus of most observers that in most cases, 2 to 4 passes of needle are required to harvest optimal diagnostic material. Most authorities define a numerical cut-off for cellularity of at least 6 epithelial cell groups with 5 to 10 cells per group for adequacy\(^1\).
**Embryology:**

The Human Mammary Gland develops during the 5\textsuperscript{th} week of Gestation at which time, thickenings of the ectoderm appear on the ventral surface of the fetus. These mammary ridges, also known as milk lines, extend from the axilla to the groin. Except for a small area in the pectoral region, the bulk of these ridges normally regress as the fetus continues to develop.

After the 15\textsuperscript{th} week of gestation, the developing breast exhibits transient sensitivity to testosterone, which acts on the mesenchyme. The mesenchyme condenses around an epithelial stalk on the chest wall to form the breast bud, the site of the mammary gland development.

Solid epithelial columns then develop within the mesenchyme, and these give rise to the lobes or segments of the mammary gland. Portions of the fetal papillary dermis encase the developing epithelial cords and give rise to the vascularised connective tissue that surrounds the mammary ducts and lobules. The collagen-rich reticular dermis extends into the breast to form the Suspensory Ligaments of Cooper.

Portions of the mesenchyme differentiate into fat between the 20\textsuperscript{th} and 32\textsuperscript{nd} weeks of Gestation. During the last eight weeks of Gestation, the epithelial cords canalise and branch forming lobulo alveolar structures as a result of mesenchymal paracrine effect.
A depression in the epidermis forms at the convergence of the lactiferous ducts. The nipple forms by evagination of the mammary pit near the time of birth.

**Anatomy and Histology of Breast:**

The breast is a glandular tissue, surrounded by fibro-adipose tissue and covered by epidermis. Centrally located is the nipple, surrounded by a circular pigmented area, the areola. Tubercles of Montgomery, a specialised sebaceous gland of the areola, enlarge during pregnancy and lactation.

The arteries of the mammary gland are branches of the Internal Mammary, External Mammary and Intercostal Arteries. The veins are the Axillary, Internal Mammary and Intercostal.

The breast is composed of 15 to 25 lobes that converge on the nipple in a radial pattern. Each segment consists of a lactiferous duct, lactiferous sinus, segmental collecting duct, sub segmental duct, terminal duct and acini. The collecting ducts are lined by columnar cells which are multilayered in the larger ducts.

The terminal portions of the lactiferous sinus and the lactiferous ducts are lined by stratified squamous epithelium. The secretory acini consist of a single layer of cuboidal epithelial cells with surrounding elongated myoepithelial cells resting on a basement membrane. The acini are set within the loose specialised stroma that defines the lobular unit.
The surrounding lobular connective tissue contains increased number of capillaries and few lymphocytes, histiocytes, plasma cells and mast cells. The lobular connective tissue is sharply separated from the more dense periductal fibrous tissue and abundant fat that make up the majority of the breast.

During pregnancy, the breast undergoes lobular and ductal proliferation, with evidence of lactation. After menopause, the breast shows increased amount of fat, diminished connective tissue, persistence of the mammary ducts and disappearance of the lobules.

**Cytology of the Normal Breast:**

Cells derived from ducts have round to oval nuclei, 8 to 10 \( \mu \) in diameter, with a very small nucleolus or no visible nucleolus. The cytoplasm is scanty. The myoepithelial cells are recognized as having a small spindly curved dark homogenous bipolar nucleus with scant cytoplasm that may either adhere to the epithelial fragments or appear singly.

The responsiveness of the breast epithelium to cyclic hormonal influences have been shown in Fine Needle Aspiration specimens. Post ovulatory aspirates are characterised by an increase in the number of acinar cells with all cells including the ductal cells displaying more features of acinar cells. Peripheral orientation of the nuclear chromatin with clearing is seen. The cytoplasm is lacy and fragile. The
epithelial fragments show a multilayered arrangement with marked superimposition of cells.

In pre-ovulatory aspirates, the cell borders are more prominent, with the cytoplasm appearing more even in consistency and better delineated. The nucleus is small and more compact with evenly distributed chromatin. Epithelial fragments are arranged in single layered sheets. The stroma is composed of fat and loose or fibrous connective tissue.

The cytological reporting categories are:

- Malignant
- Suspicious
- Atypical
- Benign specific
- Benign – non-specific
- Unsatisfactory sample

A palpable breast lump is a common diagnostic problem. Excisional Biopsy was accepted practice in the past, but presently, Radiological Imaging in combination with needle biopsy makes it possible to reduce unnecessary surgical excision of benign breast lesions to a minimum.
Carter et al\textsuperscript{4} studied the relationship between benign breast disease and subsequent breast cancer in 16,692 women. Women were classified into one of the 5 benign breast disease categories:

- Atypical Hyperplasia
- Proliferative Disease without Atypia
- Non Proliferative Breast Disease
- Fibroadenoma
- Others

Relative risk estimates of breast cancer for women in the 5 benign breast disease categories compared with screened women who did not develop recognizable breast disease were computed using the proportional hazards model. Results indicated that the risk was associated with the degree of epithelial atypia.

Women with nonproliferative breast disease, proliferative breast disease without atypia and atypical hyperplasia displayed progressively increasing risk of 1.5, 1.9 and 3 respectively, compared with normal subjects with 95\% confidence intervals exceeding unity\textsuperscript{5}.

Dupont et al attempt to quantitate the relative risk of breast carcinoma with the degree of proliferation and atypia of intraductal epithelial proliferation of the breast.
Table 1: DuPont and Page: Relative Risk of Carcinoma in various categories

<table>
<thead>
<tr>
<th>Study</th>
<th>Study Design</th>
<th>Nonproliferative</th>
<th>Proliferative without atypia</th>
<th>Atypical hyperplasia</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nashville</td>
<td>Retrospective Cohort</td>
<td>1</td>
<td>1.9</td>
<td>5.3</td>
</tr>
<tr>
<td>Nurses Health Study</td>
<td>Case-Control</td>
<td>1</td>
<td>1.6</td>
<td>3.9</td>
</tr>
<tr>
<td>Breast Cancer detection and Demonstration Project</td>
<td>Case-Control</td>
<td>1</td>
<td>1.3</td>
<td>4.3</td>
</tr>
<tr>
<td>Florence, Italy</td>
<td>Case-Control</td>
<td>1</td>
<td>1.3</td>
<td>13.0</td>
</tr>
</tbody>
</table>

The cytological reporting category does not help us in assessing the relative risk of different lesions turning malignant. In the current study, Dupont & Page’s categorisation of breast lesions has been used.

The study showed a relative risk for cancer of 1 for Non-proliferative Breast Disease and a relative risk of 1.9 for proliferative Breast Disease without Atypia, and a risk of 5.3 for Proliferative Breast Disease with Atypia.

Based on this study, Breast Lesions were categorised as follows:
Categorisation of Breast Lesions according to the criteria of Dupont, Page and Rogers:

Non Proliferative

Cysts

Papillary apocrine change

Epithelial-related Calcifications

Mild hyperplasia of the usual type

Duct Ectasia

Proliferative lesions without atypia

Moderate or Florid ductal hyperplasia of the usual type (usual ductal hyperplasia)

Intraductal papilloma

Sclerosing adenosis

Fibroadenoma

Atypical hyperplasia

Atypical ductal hyperplasia

Atypical lobular hyperplasia
Carcinoma

Non-Invasive

Intraductal Carcinoma with Paget’s Disease

Lobular Carcinoma insitu

Invasive

Invasive Ductal Carcinoma with Paget’s Disease

Invasive Ductal Carcinoma with predominant Intraductal Component

Invasive Lobular Carcinoma

Medullary Carcinoma

Mucinous Carcinoma

Invasive Papillary Carcinoma

Tubular Carcinoma

Adenoid Cystic Carcinoma

Secretory Carcinoma

Apocrine Carcinoma

Metaplastic Carcinoma

Inflammatory Carcinoma
Clinical and Cytological Findings

Cyst:

- Age > 30, Multifocal and Bilateral
- Poorly defined lumpiness on palpation with a shotty feeling
- Scanty, watery or fatty smear
- Benign or Uncertain Mammogram
- Low to Moderate cellularity
- Apocrine Cells in Variable Cellularity
- Foam Cells (Macrophage or Epithelial origin)
- Sheets or Fragments of Non apocrine Ductal Epithelium with bland nuclei arranged in honey comb pattern with admixed myoepithelial cells
- Dispersed stromal bipolar nuclei
- Fat or Fibrous Stroma in variable quantities
- Haagensen states that the initial insult of fibrocystic diseases is periductal mastitis, resulting in periductal scarring\textsuperscript{10}.
- Proliferative lesions accompanying Fibrocystic Disease are
  - Sclerosing Adenosis
  - Collagenous Spherulosis
  - Papillomatosis
  - Ductal Hyperplasia
- Atypical Ductal Hyperplasia

- Differential Diagnosis
  - Adenoid Cystic Carcinoma
  - Collagenous Spherulosis
  - Signet-ring cell Carcinoma

**Mastitis**

**Acute Mastitis or Breast Abscess:**

- Cytology shows Neutrophils, Foamy Macrophages, Cell debris in the background
- Atypical epithelial cells with features of regeneration and repair including nuclear enlargement and prominent nucleoli
- Microorganisms (Infectious Mastitis)

**Granulomatous Mastitis:**

Differential Diagnosis of Granulomas in the breast include Infectious Granulomas (Tuberculosis, Fungi, Leprosy, Brucella), Sarcoid, Tumour, Fat Necrosis, Foreign Body reaction and Idiopathic Granulomatous Mastitis.

- Granulomas show clusters of Epithelioid Histiocytes, with or without Multinucleated Giant Cells, Lymphocytes and Plasma Cells.
Fat Necrosis:

- History of Trauma with or without bruising of the skin
- Tender on Palpation
- Foamy Macrophages and multinucleated giant cells with foamy cytoplasm
- Fragments of Normal Adipose tissue
- Variable number of other inflammatory cells.
- Few epithelial cells
- Free Lipid Droplets
- Granular Background

Papillary Apocrine Change:

- Papillary proliferation of ductal epithelial cells in which all of the cells show Apocrine features

Epithelial Hyperplasia:

Intraductal Epithelial proliferation includes a spectrum ranging from intraductal hyperplasia without atypia to atypical ductal hyperplasia to ductal carcinoma in situ.

- Clinical picture is usually benign but may be suspicious
• Low or Moderate Cellularity with small epithelial groups suggest Fibroadenosis, Sclerosing Adenosis or Other Sclerosing lesions

• High Cellularity with large flat or folded sheets of cohesive regular monolayered cells suggest Epitheliosis when there is regular spacing of nuclei within the sheets

• Adenosis lesions frequently show a microacinar appearance in smears

• Nuclei may be enlarged and nucleoli are visible but inconspicuous

• The Groups contain smaller, darker, ovoid nuclei of myoepithelial cells

• Variable number of stromal cell bipolar nuclei are seen between the groups. If stromal cells are frequent, a sclerosing lesion may be suspected

• If separate epithelial cells are present, they should have a fine chromatin pattern and small nucleoli.

• The Nuclear Membrane appears smooth

• Macrophages and apocrine cells may be present

• An absence of nuclear atypia, widespread poor cell cohesion or necrotic debris.

**Mild Hyperplasia:**

• Increase in the number of epithelial cells within a duct that is less than 4 epithelial cells in depth

• Epithelial cells do not cross the lumen
**Moderate or Florid Hyperplasias:**

- Intraductal Epithelial proliferations are more than 4 epithelial cells in depth
- They bridge and distend the space
- Cytologically, the cells are benign and variable in shape, size and orientation
- Arranged in a swirling pattern
- 2 distinct cell populations seen

According to Sneige and Staerkel\(^{11}\) aspirates from Ductal Epithelial Hyperplasia show groups of epithelial cells admixed with myoepithelial cells and stromal cells arranged in a complex or cribriform fashion. Cell streaming with overriding nuclei or tapered intercellular bridges is a feature of ductal hyperplasia.

**Atypical Ductal hyperplasia:**

- Cell-rich smears, large sheets of cohesive epithelial cells, few single cells
- Focal Crowding and overlapping of nuclei, holes suggestive of cribriform pattern
- Mild to moderate Nuclear Atypia
- Few Naked Bipolar and Myoepithelial nuclei
It is generally agreed that Fine Needle Aspiration Cytology cannot reliably distinguish atypical Ductal Hyperplasia from a Non-Comedo type of Ductal Carcinoma in situ. Surgical Biopsy confirmation is required whenever atypical ductal hyperplasia or ductal carcinoma in situ, non-comedo type is suggested by the cytological findings.

**Atypical Lobular Hyperplasia:**

- Monomorphic cells, Evenly spaced and dyshesive with round or oval eccentric nuclei, pale cytoplasm with intracytoplasmic vacuoles

**Intraductal Papillomas:**

- Age range of 50 – 60 years
- Clinically presents with nipple discharge and a mass identifiable only after careful palpation
- Moderate to high cellularity
- Epithelial cells are often dispersed or in small groups with papillary clusters
- Small number of stromal cells
- Apocrine cells may be present
- Small amount of debris and macrophages may be present
- Differential Diagnosis : Fibroadenoma
Fibroadenoma:

- Common in age group of 20 – 35 years
- 5 – 30 mm diameter, mobile lump
- Benign Mammographic appearance of a round, well-defined lesion
- Moderate or High Cellularity
- Cohesive Sheets with an antler like appearance
- Many naked bipolar cell nuclei
- Apocrine or Foam cells may also be present
- Fragments of fibromyxoid stroma

Bottles et al\textsuperscript{12}, using stepwise logistic regression analysis, demonstrated that stromal fragments, antler horn clusters and marked cellularity were the three most useful cytological variables to distinguish fibroadenoma from fibrocystic disease.

Dejmek and Lindholm \textsuperscript{13} applied Bottle’s criteria to a series of fibroadenomas and noted that stromal fragments were found in only 57% of the cases, antler horn clusters in 90% and honey-comb sheets in 81%.

Stanley et al\textsuperscript{14} state that Fine-needle aspiration cytology of fibroadenomas with atypia could mimic carcinoma. The atypia was due to multifactorial causes including hormonal stimulation, inflammation, metaplastic changes and pre-neoplastic atypia.
Phyllodes tumour

- Cellular Smear
- Biphasic population of Epithelial and Stromal Cells
- Hypercellular stromal fragments consisting of spindle shaped cells present singly and enmeshed in metachromatically staining stroma.
- Stromal cell atypia is a feature of Malignant Phyllodes
- Epithelial hyperplasia can be present
- Numerous bipolar naked nuclei

Complex Sclerosing Lesions / Radial Scars:

- Features similar to fibrocystic change
- Epithelial hyperplasia with or without atypia
- Angular groups of epithelial cells with mild nuclear atypia
- Fragments of fibrotic and elastotic stroma

Sclerosing Adenosis / Adenosis Tumour:

- Age range of 20 – 67 years
- Average size of 2.5 cm
• Epithelial aggregates may show microacinar pattern

• There may be some loss of cell cohesion and mild nuclear atypia, but single bipolar nuclei are usually present

• Stromal fibrosis and changes of proliferative fibrocystic disease

• Differential Diagnosis: Myo-epithelioma, which shows cohesive irregular clusters of spindle shaped cells.

Orell\textsuperscript{15} reported a significant false positive rate of 4.3 % in radial scar / complex sclerosing lesions collected from Breast Carcinoma screening.

\textbf{Adenoma:}

\textit{Adenoma of Nipple}

• Cellular smear consisting of clusters of uniform ductal epithelial cells and dissociative bipolar naked nuclei\textsuperscript{16, 17}.

Papillary adenoma, Eccrine Spiradenoma and Ductal Carcinoma show features similar to proliferative Fibrocystic Change.

\textbf{Lactating Adenoma:}

• Cell-rich smear containing an uniform population of epithelial cells, which are dispersed with occasional cell clusters\textsuperscript{18}
- Epithelial cells with fragile, frayed, granular to foamy to vacuolated cytoplasm
- Mildly enlarged, Well Dispersed, Hyperchromatic nuclei with prominent nucleoli
- Greater numbers of stripped epithelial nuclei present
- Dirty background of cytoplasmic fragments and secretory material
- False positive diagnosis of malignancy is possible owing to the pattern of dissociated epithelial cells stripped of cytoplasm coupled with larger epithelial cells, demonstrating nuclear atypicality and prominent irregular nucleoli.

Granular Cell Tumour

- Cellular Aspirate
- Groups of Cells with abundant granular cytoplasm and indistinct cell borders
- The nuclei are oval to round and uniform in size with an evenly dispersed chromatin pattern
- Grossly and Clinically it mimics Scirrhou Carcinoma.
Carcinoma of Breast:

The criteria to distinguish between benign and malignant lesions are:

1. The Cellularity of the Specimen
2. Dispersal of Cells
3. Biphasic Pattern
4. Nuclear Size and Pleomorphism
5. Nucleolar Size
6. Nuclear Membrane Irregularity
7. Nuclear Cytoplasmic Ratio
8. Chromatin texture
9. Nuclear Fragility
10. Mitotic Figures
11. Contents of the Background

Cytological Findings:

- Cell-rich smears
- Single population of Epithelial Cells, No Myoepithelial cells
- Variable loss of Cell Cohesion
- Moderate to severe Nuclear Atypia
- Fibroblasts and fragments of Collagen (stromal desmoplasia)
- Intracytoplasmic neolumina
- Necrosis Unusual
Medullary Carcinoma:

- Cellular smear
- Loose syncytial aggregates and single cells
- Bizarre Tumour cells with prominent nucleoli and occasional stripped tumour nuclei.
- Benign lymphoid cells with occasional plasma cells
- Differential Diagnosis: Metastatic Melanoma, Malignant Lymphoma, High grade Ductal Carcinoma in situ

Mucinous Carcinoma:

- Abundant pools or strands of mucin
- Aggregates and cell balls of tumour cells
- Occasional Signet-ring malignant cells
- Moderate Nuclear Atypia
- Chicken-wire blood vessels
- Differential Diagnosis: Mucinous Ductal Carcinoma in situ or Atypical Ductal Hyperplasia, Mucocele like lesions, Mucinous Fibroadenoma, Metastatic Carcinoma

Tubular Carcinoma:

- Low to moderate cellularity
• Angulated, pointed, open tubules and glands with comma shaped projections
• Little or No cellular Atypia
• Bipolar Naked nuclei occasionally seen
• Fibroblastic cells; fragments of fibromyxoid or elastotic stroma

Papillary Carcinoma:

• Cellular smear
• Three dimensional papillary clusters of uniform atypical cells
• Tall columnar cells
• Naked, enlarged, Atypical epithelial nuclei
• Blood and Hemosiderin laden macrophages

Lobular Carcinoma:

It accounts for 3 – 15 % of all breast carcinomas

• Low to moderate cellularity
• Single cells and small clusters, cords and strands of Atypical cells
• Mildly atypical cells with increased nuclear-cytoplasmic ratio hypochromatic to Hyperchromatic, oval to irregular nuclei and small nucleoli
• Signet-ring cells and intracytoplasmic mucin
• No Bipolar nuclei
The most common cause of false negative diagnosis if the breast malignancy is accurately sampled is aspiration from lobular carcinoma. Apocrine carcinoma cytologically shows both individually scattered and syncytial fragments of cells with apocrine features. Shinagawa et al. state that grape-like clusters of vacuolated cells may be a helpful cytological feature for the diagnosis of secretory carcinoma.

Sidawy classified the nonproliferative breast lesions according to:

<table>
<thead>
<tr>
<th>Cellularity</th>
<th>Low (&lt; 15 epithelial groups per slide)</th>
<th>Moderate (15 – 30 epithelial groups)</th>
<th>High (&gt; 30 epithelial groups)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Size of Epithelial Groups</td>
<td>Small (&lt; 3000 cells)</td>
<td>Large (&gt; 3000 cells)</td>
<td></td>
</tr>
<tr>
<td>Intact Lobules</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cellular Arrangement</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non–Complex</td>
<td>(Simple sheets with little folding or branching)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Complex</td>
<td>(More significant folding, branching, lumen formation or three dimensionality)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
• Number of single epithelial cells
  
  o Grade I – < 10%
  o Grade II – 10 – 20%
  o Grade III – 20 – 30%

• Size of the nucleus
  
  o Grade I - < 1.5 times the size of RBC
  o Grade II - 1.5 to 2.5
  o Grade III - 2.5 to 3

Sidawy et al\textsuperscript{23} state that using univariate analysis, they were able to identify 6 cytological features that differed between non-proliferative breast lesions and proliferative breast lesions. Proliferative breast lesions show more complexity of epithelial groups, slit like spaces, mixture of apocrine metaplasia and nuclear pleomorphism, large epithelial groups and cell swirling streaming. The latter feature was the only one that showed significance in both air dried and alcohol fixed smears.

The study showed the limited role of FNA in distinguishing nonproliferative breast disease from proliferative breast disease and it demonstrated the spectrum of cytomorphological features in nonproliferative breast disease

Masood et al\textsuperscript{24, 25} proposed a cytological scoring system in which a value of 1 to 4 was given for each of the following features:

Cellular Arrangement

Cellular Pleomorphism
Presence of Myoepithelial cells

Anisonucleosis

Nucleoli

Chromatin clumping

A score derived from the sum of these values was used to classify each FNA sample. Non Proliferative Breast Disease was diagnosed with a score of 6 to 9, Proliferative Breast Disease with a score of 10 to 14. The results showed 29 of 34 (85%) FNA specimens diagnosed as Nonproliferative Breast Disease and 15 of 17 (88%) FNA specimens diagnosed as Proliferative Breast Disease were found to correlate with histological Diagnosis.

Maygarden et al.\textsuperscript{26} evaluated the cytological features of 99 FNA specimens of histologically proven proliferative and nonproliferative fibrocystic change. They found no parameter that reached statistical significance in distinguishing between these two entities.

According to McDivitt et al.\textsuperscript{27} benign breast lesions can be categorised by a modification of the Black-Chabon Grading system which differentiates hyperplasia and atypia. When compared with women who had never had a breast biopsy, women with benign breast disease without hyperplasia had an odds ratio of 1.5 (95% Confidence Limits 1.3 – 1.9), women with hyperplasia without atypia had an odds ratio of 1.8 (CL =1.3, 2.4) and women with hyperplasia and atypia had an odds
ratio of 2.6 (CL = 1.6, 4.1). Fibroadenoma was an independent risk factor (OR = 1.7; CL= 1.1,2.5).

Other cytological Breast Cancer scoring systems used are the Robinson system, the Moriquand system and Fisher’s system.

Robinson’s system:

Robinson’s system of scoring assesses 6 features, i.e, cell dissociation, nuclear margin, cell size, cell uniformity, nucleoli and chromatin. A grade of 1 is given for a score of 6 – 11, grade 2 for a score of 12 – 14, grade 3 for a score of 15 – 18.

Moriquand system:

Moriquand scoring assesses 4 features, i.e, cellular characters, nuclear features, nucleoli and mitoses. A score of 0 – 3 is given for each feature and a grade 1 is given for a score ≤ 5. Scores of 6 – 9 are Graded 2, and Scores of >10 are graded 3.

Fisher’s system:

Fisher’s Modification of Nuclear Grading depends on 4 features : nuclear size, nuclear membrane, Chromatin and Positive or Negative nucleoli.

Giard and Hermans\textsuperscript{28} found a false positive rate of 0.15% with cytomorphological analysis. Dupont and Page\textsuperscript{29} et al observed a strong association between breast carcinoma risk and Atypical Hyperplasia.
Jersey et al state that women with proliferative breast disease have an increased relative risk of subsequent invasive carcinoma. The risk is stratified according to the degree of epithelial proliferation and is elevated in women with a family history of breast cancer.

Frost et al\textsuperscript{30} evaluated 12 cytological features in 51 benign breast aspirates and found that only the presence of a swirling pattern reached statistical significance in distinguishing proliferative breast disease and non-proliferative breast disease.
Materials and Methods
Materials and Methods

The records of the cytopathology and histopathology laboratory of Kilpauk Medical College Chennai were analysed over a period of 2 years (from June 2006 to September 2008). One Hundred FNAC cases were collected and the smears were stained with Hematoxylin & Eosin and their histological confirmation were included in the study.

Inclusion Criteria:

All palpable breast lumps from Female patients with an adequate smear showing 5 to 6 ductal epithelial groups were included in the study.

Exclusion Criteria:

Inflammatory smears and smears with no ductal epithelial cells were excluded.

All aspirates were performed as an outpatient procedure without imaging or mammographic guidance using a 23 G Needle and a 5 ml syringe. The smears were fixed in Absolute alcohol for 20 minutes and then stained by Hematoxylin & Eosin method. Aspirates were evaluated according to the Modified Masood Scoring System.

Masood et al proposed a cytological scoring system in which a value of 1 to 4 was given for each of the following features:
1. Cellular Arrangement

2. Presence of Myoepithelial cells

3. Anisonucleosis

4. Cellular Pleomorphism

5. Nucleoli and

6. Chromatin Clumping

A score derived from the sum of these values was used to classify each FNA sample. A score for each category was assigned as follows:

1. Non Proliferative Breast Disease – Score of 6 to 9

2. Proliferative Breast Disease without Atypia – Score of 10 to 14

3. Proliferative Breast Disease with Atypia – Score of 15 to 18

4. Carcinoma – Score of 19 to 24

Masood’s score was modified with a score of 6 to 10 denoting Nonproliferative breast disease instead of the original score of 6 to 9.

Cytomorphological Diagnosis and Histopathological Diagnosis were also categorised to four groups:

1. Non Proliferative Breast Disease

2. Proliferative Breast Disease without Atypia

3. Proliferative Breast Disease with Atypia

4. Carcinoma
Correlation was calculated using Spearman’s rho and Kendall’s tau coefficients. SPSS Statistics version 17 was used for the statistical Analysis. The Tables of data were constructed in Microsoft Office Excel 2007.

This scoring system yielded very promising results when applied prospectively by the authors in a study evaluating 100 radiographically directed FNAs. Their results show that 29 of 34 FNA specimens diagnosed as Non Proliferative Breast lesions (85%) and 15 of 17 FNA specimens diagnosed as Proliferative Breast lesions were found to correlate with the histological Diagnosis.

The scoring system requires good sampling techniques, good cellular yield and non-fibrotic stroma for optimum results.
Fig. 1. Cellular arrangement
1 = monolayer
2 = nuclear overriding
3 = overriding & clustering
4 = loss of cell cohesion
(Papanicolaou, x 200)

Fig. 2. Presence of myoepithelial cells
1 = many
2 = moderate
3 = few
4 = absent
(Papanicolaou, x 200)

Fig. 3. Cellular pleomorphism
1 = absent
2 = mild
3 = moderate
4 = conspicuous
(Papanicolaou, x 400)

Fig. 4. Anisonucleosis
1 = Absent
2 = Mild
3 = Moderate
4 = Conspicuous
(Papanicolaou, x 400)
Results and Observations
Results and Observations

The cases included in the study were all females with an Age Range of 15 – 65 years. The study included 100 palpable breast lesions. Results were tabulated and analysed.

Masood’s score was modified[^32] with a score of 6 to 10 denoting Nonproliferative breast disease instead of the original score of 6 to 9.

The scoring system and cytomorphology correlated with histological diagnosis for 77 cases. In 12 cases, the cytomorphological diagnosis correlated well with histopathological diagnosis. In 11 cases, the modified Masood scoring system correlated well with the histological Diagnosis.

In 7 cases, the Cytomorphological Diagnosis and Histopathological Diagnosis correlated well and categorised the patients under Proliferative Breast Disease, while the scoring system categorised them under Non-Proliferative Breast disease.

In one case, the cytomorphological Diagnosis categorised the case under Poliferative Breast Disease without Atypia, while the Scoring system brought it under Proliferative breast Disease with Atypia and the Histopathological Diagnosis was Carcinoma.
In 2 cases the cytomorphological Diagnosis categorised the case under Proliferative Breast Disease without Atypia, whereas the scoring system overdiagnosed it as Proliferative Breast Disease with Atypia.

In 3 cases, the Cytomorphological diagnosis overdiagnosed and categorised the cases under proliferative Breast with Atypia, while the scoring system correlated with the histopathological Diagnosis of Proliferative Breast Disease without Atypia.

In 2 cases, the Cytomorphological Diagnosis categorised the cases under Proliferative Breast Disease without Atypia, while the Scoring system correlated with the Histopathological Diagnosis of Non-Proliferative Breast Disease.

The study shows 2 cyt morphological positives correlated with histopathological positivity under the category of nonproliferative breast disease, when compared to 4 cases in the scoring system. 61 cases correlated with histopathological diagnosis when compared to 56 cases in the scoring system under the category of proliferative breast disease without atypia.

Out of the 5 cases of Proliferative Breast Disease with Atypia under Modified Masood Scoring system, 3 cases were found to be carcinomas by histology and 2 cases were found to be fibroadenomas with atypia. The cytomorphological diagnosis brought 2 cases under carcinoma, and three cases under proliferative breast disease without atypia.
Under the category of proliferative breast disease with atypia, there were no correlating cases. 25 cases correlated with the cytomorphological diagnosis when compared to 24 cases in the scoring system under the category of Carcinoma.

Cytohistological correlation was 88% while correlation of the Modified Masood score with Histology was 84%. The correlation was statistically significant with a correlation coefficient of 0.832 for Cytology-Histology and 0.821 for Modified Masood-Histology. Both the correlation coefficients were significant at the 0.01 level (1-tailed).

Sensitivity of the scoring system was found to be 80%, Specificity 100% and Positive Predictive Value was 100%, Negative Predictive Value was 92% for a diagnosis of Carcinoma.

Sensitivity of FNAC was 83%, Specificity was 100%, Positive Predictive Value was 100% and Negative Predictive Value was 93%.

The peak age of incidence of nonproliferative breast disease was found to be the third decade. The peak age of incidence of proliferative breast disease was found to be the third decade and carcinomas peaked at the 6th decade.
Figure 1: Age Distribution of Study Population

Figure 2: Distribution of cases by FNAC Score
Figure 3: Distribution of cases by Masood Score

Figure 4: Distribution of cases by Biopsy Scores
Figure 5: Concordance of FNAC and Masood Scores with Biopsy Scores

Figure 6: Age distribution of cases according to Modified Masood Scoring
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Table 3: Biopsy vs. Modified Masood Scoring
Figure 7: Age Distribution of cases according to Biopsy Score

Figure 8: Age Distribution of cases according to FNAC Score
### Correlations

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**. Correlation is significant at the 0.01 level (1-tailed).

*Table 4: Non parametric correlation coefficients*
Figure 9: Distribution of cases by Histopathology

Distribution of cases under various categories by Histopathology

- Non-Proliferative
- Proliferative without atypia
- Proliferative with atypia
- Carcinoma

Figure 9: Distribution of cases by Histopathology
Pic 1: FNAC of Phyllodes tumour, H&E, 40 X with a score of 9

Pic 2: Biopsy of Phyllodes Tumour
Pic 3: FNAC Diagnosis of Phyllodes with a score of 5

Pic 4: Biopsy showing Fibroadenoma
Pic 5 : FNAC Diagnosis of Proliferative Breast Disease with a score of 12

Pic 6 : Biopsy Diagnosis – Infiltrating Ductal Carcinoma
Pic 7: FNAC Diagnosis - Proliferative Breast Disease with a score of 9

Pic 8: Biopsy - Tuberculous Mastitis
Pic 9: FNAC - Sclerosing Adenosis with a score of 13

Pic 10: Biopsy - Sclerosing Adenosis
Pic 11: FNAC - Intraductal Carcinoma with a score of 20

Pic 12: Biopsy - Intraductal Carcinoma
Pic 13: FNAC Diagnosis of Fibroadenoma with a score of 16

Pic 14: Biopsy showing Intraductal Carcinoma
Pic 15: FNAC Diagnosis of Fibroadenoma with a score of 11

Pic 16: Biopsy Diagnosis of Tubular Adenoma
Pic 17: FNAC Diagnosis of Fibrocystic Disease with a score of 13

Inset: Cyst Macrophages

Pic 18: Biopsy showing Infiltrating Papillary Adenocarcinoma
Discussion
Discussion

The study was initiated to evaluate the applicability of Modified Masood Scoring in Cytological Diagnosis of Palpable Breast masses and to compare the scoring system with Cytomorphological Diagnosis. The Histopathological Diagnosis was considered to be the Gold Standard.

According to Barrows\textsuperscript{33}, the FNAC positivity for Breast Cancer varies between 48 to 88%. To increase the diagnostic yield of the FNAC, Modified Masood scoring of the aspirates were done. The study helps to categorise the lesion so that aspiration of minimally suspicious lesions is helpful in initiating excisional biopsy.

Modified Masood’s scoring gives additional information by eliminating benign cases and improves the diagnostic yield. Application of scoring in a step-wise manner in atypical aspirates can help in selection of cases suitable for biopsy.

The risk of developing subsequent invasive breast cancer is stratified according to the degree of epithelial proliferation and atypia. The risk is 1-fold in women with Non-proliferative Breast disease, 1.9 fold in women with proliferative breast disease and 5.3 in women with carcinoma insitu.

Histological criteria that allow the distinction of these various breast lesions are established. Sneige and Staerkel\textsuperscript{7} introduced the concept of using architectural features cytologically and concluded that the application of both cytological and
architectural criteria is more reliable than cytology alone in separating proliferative breast lesions.

Dawson et al\textsuperscript{34} showed that applying both architectural and cytological criteria enhanced diagnostic accuracy. Cytologically, the architectural features of proliferative breast lesions may be apparent in the larger breast fragments and recapitulate the histologic appearance of these lesions. Slit like lumens, swirling and streaming are noted in proliferative breast lesions without atypia. Round spaces may be seen in proliferative breast lesions without atypia and with atypia.

Rigid sublumina and a micropapillary architecture are features of DCIS. Thomas et al\textsuperscript{35} demonstrated that experience and fine tuning of cytological criteria increased the concordance with the histological findings.

All these studies emphasise the importance of adequate sampling to minimise, in particular, underdiagnosis.

Criteria for the cytological diagnosis of Fibroadenoma and Carcinoma are well established. The sensitivity and specificity of Fine Needle Aspiration Cytology for the diagnosis of Fibroadenoma is 86.9 % and 93.8% respectively, while for Carcinomas, Sensitivity is 89 – 98 % and Specificity 93 – 98 %.

Proliferative Breast lesions on cytology are categorised into Proliferative Breast Disease without atypia, and Proliferative breast disease with Atypia because it not possible to delineate all the histological entities on FNAC. However, the
diagnostic accuracy in this distinction is still unclear, and the cytological features of proliferative breast disease are not well established.

In this study, I have confined myself to palpable breast lesions referred for routine FNAC. A high degree of concordance was found between FNA Cytology and Histological Diagnosis in cases of proliferative breast disease without atypia and carcinoma. In these groups, the scoring system did not contribute any addition information.

The peak age of incidence of nonproliferative breast disease was found to be the third decade. The peak age of incidence of proliferative breast disease was found to be the third decade and carcinomas peaked at the 6th decade.

Use of the scoring system can reduce the number of atypical reports and hence limit unnecessary procedures performed on patients.

The study has found that in cases showing benign ductal cells and suspected to be proliferative breast disease without atypia, the scoring system did not add substantially in the evaluation of FNAC. This category is likely to form the majority of cases on FNAC and in this group, the laborious application of the scoring system can be avoided.

The study found that the diagnosis of atypia or routine cytomorphological assessment was influenced more by nuclear pleomorphism than by architectural
details. Application of the scoring system evaluated both the nuclear atypia and the cytoarchitectural features.

So, the scoring system should be applied in a stepwise manner after cytomorphological assessment. In cases with cytological diagnosis of proliferative breast disease without atypia, and carcinoma, the scoring system offers no advantage over cytomorphology.

The scoring system is useful in aspirates with cytological diagnosis of proliferative breast disease with atypia.
Summary and Conclusions
Summary and Conclusions

100 cases during a period of two years from September 2006 to September 2008, of palpable breast lesions were studied. The cytomorphological analysis by FNAC and Modified Masood system were taken and correlated with histopathological diagnosis. Out of the 100 cases, 4 cases were diagnosed as Non proliferative breast Disease and 66 cases were diagnosed as Proliferative breast disease without atypia. There were no cases under proliferative breast disease with atypia. 30 cases were diagnosed as carcinoma by histopathology.

With Modified Masood’s scoring, 12 cases were categorized as Nonproliferative breast disease, 59 cases were categorized as proliferative breast disease without atypia, 5 cases were categorized as proliferative breast disease with atypia, and 24 cases were categorized as Carcinoma.

With FNAC, 4 cases were categorized as nonproliferative breast disease, 25 cases diagnosed as carcinoma, 66 cases categorized as proliferative breast disease without atypia and 5 cases were categorized as proliferative breast disease with atypia.

Sensitivity of the scoring system was found to be 80%, Specificity 100% and Positive Predictive Value was 100%, Negative Predictive Value was 92% for a diagnosis of Carcinoma.
Sensitivity of FNAC was 83%, Specificity was 100%, Positive Predictive Value was 100% and Negative Predictive Value was 93%.

Both FNAC and Modified Masood’s scoring were found to correlate highly with each other and with Histopathology.

In conclusion, Modified Masood’s scoring system can be done to categorize the breast lesions into the four categories which correlate highly with FNAC and histopathology.
Bibliography
Bibliography


3 Koss LG, Melamed MR. Koss' Diagnostic Cytology and its Histopathological Bases – 5th edition – Volume One. Lippincott Williams & Wilkins


Sidawy et al. The Spectrum of cytologic Features in Non proliferative breast lesions. Cancer Cytopathology, 2001 93(2) : 140-45


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