Breast cytology

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Reporting:
- need to know age of the patient, clinical presentation, whether on hormone replacement therapy if post-menopausal
- always comment on cellularity and adequacy
- describe architecture and cytology
- give general category and specific diagnosis

ADEQUACY:
- no consensus reached; require clinical and radiological correlation
- for a suspected epithelial lesion, to exclude malignancy: at least 6 epithelial groups of 5-10 cells each (decrease false negative rate by 30% in one study)
- for suspected non-epithelial lesion: no minimal cell groups proposed
- presence of atypical cells, even in small numbers, is never considered unsatisfactory

Special circumstances:
Cyst - Features consistent with a benign cyst. Comment on paucity of ductal cells but not classified as unsatisfactory.

Similar situations with duct ectasia, mastitis, stromal fibrosis and fat necrosis.

Lipoma – If there is clinical and mammographic evidence of a lipoma, presence of fat without ductal cells is in support of the diagnosis and it is not unsatisfactory.

Intramammary lymph node – If there is mammographic evidence of a lymph node, the appropriate lymphoid background in absence of ductal cells is considered satisfactory.

REPORTING CATEGORIES:

Unsatisfactory
- broken slide, wrongly labelled slide
- poorly prepared (poorly spread smears: too thick; too vigorous smearing; prolonged air-drying; no fixation for Pap stain; poor staining: weak diluted staining solutions)
- too bloody obscuring diagnostic cells
- completely necrotic
- poorly cellular
- contaminants: ultrasound gel

Benign
- non-specific
- specific e.g. mastitis, fat necrosis, fibroadenoma

Atypical but favouring benign/reactive
- ductal cells are crowded, mild pleomorphism, occasional nucleoli
- not a lot of myoepithelial cells
- few dyscohesive ductal cells
- inflammatory background may be present
- these cases need further investigations (esp if there is also clinical and radiologic suspicion)

**Suspicious and favouring malignant**
- ductal cells with nuclear enlargement and prominent nucleoli but are in large sheets with no single cells
- only a few malignant cells are present
- malignant cells intermixed with bare bipolar nuclei
- these cases need further investigations – core biopsy

**Malignant**
- unequivocal evidence of malignancy is present
- needs to type whether carcinoma, lymphoma, sarcoma or melanoma
- may subclassify variants of invasive ductal carcinoma if the features are obvious
- mention whether ductal or lobular if there are sufficient diagnostic features
- may comment on the tumour grade (nuclear size, comparing to an erythrocyte; nucleoli; nuclear pleomorphism; chromatin pattern; nuclear contour; cell dyscohesion)
- may perform ER, PR and HER2 on the cell block if requested

**How to report smears from nipple discharge:**
- comment on presence or absence of RBC
- comment on presence or absence of ductal cells (Normal: a few small cohesive groups of ductal cells at the most; different from adequacy criteria for FNA)
- foam cells, heavy background proteinaceous material: duct ectasia
- increased numbers of ductal epithelial cells +/- RBC: intraductal papilloma, epithelial proliferation. Needs further investigations.

**Normal breast:**
- often scant cellularity (depends on age, hormonal status)
- small groups of ductal cells
- lobular structures may be seen
- myoepithelial cells in cell groups (as elongated nuclei) and in the background (ovoid nuclei stripped of the cytoplasm)
- adipose tissue and stroma

**Apocrine cells:**
- in cohesive flat sheets, papillary structures or as individual cells
- large, round to oval nuclei
- nuclear contours are smooth with evenly dispersed chromatin
- often there is a single prominent nucleolus
- occasional binucleation or multinucleation may be seen
- cytoplasm is finely granular and dense with well-defined cell borders
- cytoplasm is blue, purple or grey with Diff-Quik and pink or orange with Pap
Specific entities:

**Breast cyst:**
- 99% benign
- after complete aspiration of the cyst, it is important to repalpate the area to determine if a residual mass is present
- if a residual mass is found, a second aspiration should be performed
- benign cyst features: watery, yellow, clear to slightly opaque, few particles, no residual mass
- atypical cyst features: watery to viscous, brown to red, densely opaque, numerous particles, residual mass
- send cyst fluid for cytology if: blood stained, residual mass, recurrent cyst, complex cyst on radiology

**Cytology:**
- background of amorphous material
- degenerate cells and debris
- foamy macrophages
- ductal epithelial cells, often apocrine and balling-up
- myoepithelial cells may not be seen (do not overcall aspirate as malignant)

Always make a comment if there are not many epithelial cells present but the diagnosis is still consistent with benign cyst content.

**Mastitis and abscess formation:**
- numerous neutrophils and histiocytes
- granular background
- scanty reactive ductal cells: open chromatin, vesicular nuclei, central nucleoli

Most cases of mastitis contain very few epithelial cells. Their presence in significant number is a suspicious finding. Important differential is a carcinoma (or comedo DCIS) with heavily inflamed background, obscuring the tumour cells.

**Subareolar abscess:**
- specific clinicopathologic entity involving the lactiferous ducts in the subareolar region with inflammation, squamous metaplasia and duct obstruction
- anucleated squamous cells, keratinous debris, cholesterol crystals
- acute and chronic inflammatory cells
- foreign body type giant cells
- reactive ductal and squamous cells
- granulation tissue may be present
Idiopathic granulomatous mastitis:
-loose granulomas with epithelioid histiocytes
-other inflammatory cells: neutrophils, lymphocytes
-prominent branching capillaries
-epithelial cells rare
-needs to exclude infectious aetiology by culture and check for refractile silicone

Fat necrosis:
-usually not very cellular
-fragments of degenerate necrotic adipocytes with loss of nuclear staining
-ductal cells are generally scanty
-dirty background
-foamy macrophages with multinucleated giant cells
-calcium may be seen

Epidermal cyst:
-abundant anucleate squames and keratinous debris
-abundant inflammatory cells present if infected

Lactational adenoma:
-increased cellularity
-frothy secretory debris, fat cells (foamy, fatty background)
-many of the ductal cells are single
-cells have enlarged uniform nuclei, smooth nuclear contour, prominent nucleoli
-cytoplasmic foaminess or vacuolation

Vacuolated background may be seen when fat has been sampled. It does not always represent lactational change. Diffuse cytoplasmic foaminess is a more specific feature of lactation.

Gynaecomastia:
-many groups and clusters of epithelial cells with naked bare nuclei and stromal fragments (similar features as seen in a fibroadenoma)
-occasional cases may show epithelial atypia including presence of nucleoli and nuclear overlapping
-squamous metaplasia has been reported

Fibrocystic change:
-low to moderate cellularity
-proteinaceous background
-cohesive sheets of ductal cells in a honeycomb pattern
-bare bipolar nuclei dispersed in the background and within or attached to sheets of epithelial cells
variable numbers of apocrine cells and foam cells
-variable fat and stroma

Collagenous spherulosis:
-usually incidental but has been reported to present as a mass
-may be associated with intraductal papillomas, sclerosing adenosis, radial scars
-myoeipithelial cells surround hyaline globules
-benign ductal cells forming monolayered sheets and groups
-DDx: adenoid cystic carcinoma (forming a suspicious mass clinically; greater cellularity; more nuclear atypia; cribriform, tubular, finger-like and cup-shaped groups). In low grade cases, the morphology may be very similar to collagenous spherulosis, requiring histology for distinction.

Usual ductal hyperplasia:
-moderately cellular
-crowded sheets
-regular or irregular cell spacing
-associated bare bipolar nuclei
-some cytologic variation of cell size and shape
-single epithelial cells are absent or very scarce

Atypical ductal hyperplasia:
-highly cellular
-crowded groups consisting of cells with both benign and atypical features
-cells show greater degree of hyperchromasia and nucleoli can be seen
-occasional single atypical cells may be present
-bare bipolar nuclei are identified (usually absent in DCIS)

Fibroadenoma:
-moderate to high cellularity
-tightly cohesive branching antler-horn or finger-like projections of epithelial cells
-stromal fragments (metachromatic fibrillary matrix material)
-needs both ductal and stromal components to be diagnostic
-numerous bare bipolar nuclei, bordering and within epithelial clusters
-may see few foam cells or apocrine cells
-often mild nuclear atypia with prominent nucleoli, particularly in younger patients

Problems with fibroadenomas:
-necrosis and regenerative atypia associated with infarction in fibroadenomas
-bare bipolar nuclei may be nearly absent due to hyalinisation and poor cellularity of the stroma
-superimposed pregnancy / lactation effects
-fibroadenoma situated near a carcinoma
-fibroadenoma that contains a carcinoma / DCIS
-squamous or apocrine metaplasia in fibroadenomas

UDH, ADH and DCIS are difficult to be distinguished from each other on cytology. The term “epithelial proliferation” may be used.
Hamartoma:
-no specific diagnostic features
-diagnosis not made on cytology
-tends to have more intact rounded lobules
-stromal fragments uncommon
-abundant adipose tissue

Papillary lesion:
-the term “papillary lesion” includes benign papilloma, papillary DCIS, encysted and invasive papillary carcinoma. It is often difficult to distinguish these entities on cytology. Further investigations are recommended for all papillary lesions.
-some lesions may appear “papillary” but turn out “non-papillary” on histology e.g. fibrocystic change, fibroadenoma, gynaecomastia

General features:
-usually cellular
-finger-like epithelial fragments with a structural border of a row of columnar cells without a stromal core suggest a papillary lesion
-palisades of columnar epithelial cells, even if only 4 or 5 in a row, is a useful sign of a papillary lesion
-3-D papillary clusters with fibrovascular cores
-think papilloma if cellular aspirate with not that many bare bipolar nuclei (contrast with that of a fibroadenoma)
-cell dyscohesion combined with high cellularity, nuclear atypia and mitotic activity in a benign papilloma can lead to a false positive diagnosis
-nuclear atypia may be present (mild nuclear enlargement, small nucleoli), especially in infarcted papillomas
-often foamy macrophages in the background

Features favouring benign papilloma:
-mild to moderate cellularity (less cellular as compared to papillary carcinoma)
-papillary structures with fibrovascular cores and monolayered sheets, containing ductal cells with maintained polarity
-myoeopithelial cells present within the cell groups and in the background
-occasional single ductal cells but not showing nuclear atypia
-no single bare tumour nuclei
-apocrine cells present

Features favouring papillary carcinoma:
-marked cellularity
-3D large complex papillary structures which may not have fibrovascular cores, containing multilayered ductal cells with loss of polarity and cellular crowding
-acinar and cribriform structures
-myoeopithelial cells not present within the cell groups and not in the background
-many single atypical ductal cells with cytoplasm

Stromal fragments may be somewhat cellular in young patients, mimicking a phyllodes. Look for atypical stromal cells in the background.
-many single bare tumour nuclei with nuclear atypia e.g. prominent nucleoli
-haemosiderin-laden macrophages
-no apocrine cells

**Granular cell tumour:**
- cellular
- scattered groups of cells with abundant granular cytoplasm and indistinct cell borders
- nuclei are oval to round and uniform in size
- evenly dispersed chromatin pattern
- occasionally nucleoli may be present
- cytoplasmic granules are red with Pap and PAS accentuates the granules

**Sclerosing adenosis:**
- thick fragments of stroma containing acinar structures
- ductal cells bland-appearing with bare bipolar nuclei

> Acinar structures can be seen in benign aspirates and they do not necessarily indicate malignancy.

**Radial scar / complex sclerosing lesion:**
- highly cellular
- plentiful large monolayered sheets, others more complex, folded or 3D
- minor component of dispersed cells
- myoepithelial cells identified within cell groups
- bare bipolar nuclei present in most cases in small numbers
- apocrine cells may be seen
- foamy macrophages and proteinaceous background often present
- cytology may appear benign, atypical or suspicious

> These lesions should be all completely excised, regardless of the cytology.

**Ductal carcinoma in-situ:**
- high cellularity
- large complex tissue fragments, some showing papillary or cribriform patterns
- crowding and overlapping of nuclei which tend to show a uniform degree of enlargement
- plentiful dispersed cells which may be atypical
- scant bare bipolar nuclei

> Features that may suggest invasion: tubular groups of malignant cells, cytoplasmic lumina, single tumour cells invading between adipocytes or within stroma.

**Invasive ductal carcinoma, of no special type:**
- usually very cellular
- disorganised, loosely cohesive groups
- single, polygonal, plasmacytoid epithelial cells (which can look deceptively bland)
- absence of bare bipolar nuclei
- cellular and nuclear pleomorphism (2-4x RBC)
- nuclear border irregularity
- hyperchromasia
- nucleoli
- there may be mucin vacuole / targetoid inclusion within the cytoplasm
- mitoses
- tumour diathesis

20% of the cancers have tissue fragments only with no dyscohesion. Look for nuclear atypia, absence of bare bipolar nuclei, abnormal mitoses, tumour diathesis.

**Mucinous carcinoma:**
- often present clinically or radiologically as a round well-defined lesion, misinterpreting as a benign lesion
- cohesive clusters of cells, along with single cells
- cells uniform but have increased N/C ratio
- mucin (non-fibrillary, thinner than stroma)
- tumour cells in mucin
- signet-ring cells may be seen
- absence of bare bipolar nuclei
- only diagnose mucinous carcinoma when the tumour nuclei are low grade (otherwise call it invasive ductal carcinoma with mucinous differentiation)

Differential is mucocoele-like lesion: scant cellularity, lack of single ductal cells, epithelial cells in sheets, lack of nuclear atypia. Needs excision.

**Tubular carcinoma:**
- beware: epithelial cohesiveness is generally maintained
- usually very cellular
- dispersed intact cells frequently found
- cells often arranged in tubular, angulated or comma-shaped configurations
- mild nuclear atypia
- intracytoplasmic vacuoles may be present

Bare bipolar nuclei found in up to 25% of cases of tubular carcinomas. Prominent angulated tubules a helpful clue.

**Medullary carcinoma:**
- very cellular
- large cells with pleomorphic nuclei and macronucleoli
- single cells or in syncytia
- numerous lymphocytes and plasma cells in the background
- diagnosis is made on histology
**Metaplastic carcinoma:**
- numerous malignant spindle cells
- stromal fragments seen
- myxoid or chondromyxoid background
- osteoclast-like giant cells
- features of adenocarcinoma may be present
- squamous cells may be seen

**Invasive lobular carcinoma:**
- often low cellularity
- small cuboidal monomorphic cells
- singly, small clusters or short chains
- scant cytoplasm
- useful feature: single cells with high N/C ratio and round nuclei. The cytoplasm may be stripped (do not confuse with myoepithelial cells - ovoid nuclei)
- often eccentric hyperchromatic nuclei with 1-2 micronucleoli
- nuclear moulding is suggestive of lobular carcinoma
- there may be intracytoplasmic vacuoles and targetoid inclusions (vacuoles containing a dot-like globule of secretion)
- signet-ring cells may be seen
- no bare bipolar nuclei

**Phyllodes tumour:**
- variably cellular
- biphasic population of benign epithelial tissue fragments and atypical stromal cells
- hypercellular stromal fragments with spindle-shaped cells present singly and enmeshed in metachromatically staining stroma
- leaf-shaped stromal fragments are not specific and can be seen in fibroadenomas
- stromal cell atypia, with variation in nuclear size and shape (cells more pleomorphic in malignant phyllodes)

**Radiation / Chemotherapy effect:**
- usually hypocellular smears
- epithelial cells with variation in nuclear size but N/C ratio is not increased
- no hyperchromasia, chromatin may be degenerate and smudged
- vacuolated cytoplasm with polychromasia
- bare bipolar nuclei present
- no tumour diathesis
radiation fibroblasts: delicate, vacuolated cytoplasm with numerous cytoplasmic projections. Nuclear chromatin is finely granular.

FALSE POSITIVES:
- fibroadenoma with hypercellularity, atypia
- "dyscohesive cells" from smearing the cellular material too hard
- florid UDH, ADH
- intraductal papilloma
- mucocoele-like lesion
- gynaecomastia with epithelial atypia
- hormonally stimulated glandular tissue, esp. in pregnancy, lactation, HRT
- mastitis, fat necrosis with regenerative atypia
- post-radiation atypia
- atypical apocrine cells in cysts
- granular cell tumour, nodular fasciitis and fibromatosis

FALSE NEGATIVES:
- low grade invasive carcinoma e.g. tubular carcinoma: hypocellular, minimal atypia, cohesive groups
- low grade DCIS
- highly desmoplastic carcinomas
- lobular carcinoma: hypocellular
- mucinous carcinoma: hypocellular, mildly atypical cells, mucin overlooked
- papillary carcinoma: mild atypia, difficult to differentiate from papilloma
- necrotic carcinoma with no viable tumour cells

PRACTICAL TIPS:

1. Always correlate with radiology. Know the clinical history. Be cautious about making a malignant diagnosis especially when the radiology is reported as benign.

2. Beware ultrasound gel mimicking necrosis. There is absence of cellular debris and inflammatory cells.

3. Beware smears which include a substantial component of benign cells (apocrine cells, bare bipolar nuclei) in addition to the atypical cells. Do not diagnose malignancy unless there is unequivocal evidence.

4. Beware dyscohesive ductal cells if cells are smeared too hard (such as in a fibroadenoma). Usually see these cells in the tail of the smear with streak artefact.
5. Relative uniformity of the ductal cells does not always mean benignity. Low grade carcinomas can show single cells with no significant nuclear pleomorphism.

6. Beware dyscohesive cells in a papilloma. Overall features are usually not malignant. There are myoepithelial cells and apocrine cells.

7. If the atypia is beyond that allowed for reactive change in an inflamed background, think alternative diagnosis e.g. DCIS with comedo-necrosis, IDC with inflamed stroma. The epithelial to inflammatory cell ratio is increased.

8. In a postmenopausal woman not on HRT, hypercellularity alone, even with no significant nuclear atypia, needs further investigation.


10. Some tubular carcinomas may only present in cohesive groups with no single ductal cells and may even have bare bipolar nuclei in the background.
Fig 1. Normal ductal cells in a monolayered cohesive group.

Fig 2. Benign ductal group.
Fig 3. Normal apocrine cells with abundant granular cytoplasm and prominent central nucleoli.

Fig 4. Normal myoepithelial cells – Bare bipolar nuclei.
Fig 5. Normal myoepithelial cells within a cell group – Elongated darker cells (Arrows)

Fig 6. Fat necrosis with foamy macrophages.
Fig 7. Mastitis with marked reactive changes. Note round nuclei and central nucleoli.

Fig 8. Fibroadenoma. Branching ductal groups.
Fig 9. Fibroadenoma with benign ductal groups, myxoid stroma and bare bipolar nuclei in the background.

Fig 10. Fibroadenoma with magenta-staining fibrillary myxoid stroma.
Fig 11. Fibroadenoma with cellular stroma. No atypical stromal cells are seen elsewhere. The histology confirms fibroadenoma.

Fig 12. Fibroadenoma with occasional atypical cells (Arrows).
Fig 13. Fibroadenoma with occasional atypical cells (Arrows). Myoepithelial cells readily identified. Do not misinterpret as a carcinoma.

Fig 14. Hard smearing of a fibroadenoma with streak artefact showing dyscohesive cells.
Fig 15. Gynaecomastia. Cohesive ductal groups and stromal fragments. Resembles a fibroadenoma.

Fig 16. Lactational adenoma.
Fig 17. Lactational adenoma with vacuolated ductal cells.

Fig 18. Lactational adenoma with reactive changes. Note single cells with smooth nuclear outline and central nucleoli.
Fig 19. Phyllodes tumour. Hypercellular stromal fragments with atypical spindle cells.

Fig 20. Phyllodes tumour with atypical stromal cells (not bare bipolars).
Fig 21. Papillary lesion (Papilloma).

Fig 22. Papillary lesion (Papilloma). Note palisaded edge (Arrow).
Fig 23. Diagnostic trap: Dyscohesive cells in a papillary lesion (Papilloma).

Fig 24. Papillary lesion (Papillary DCIS). Difficult to distinguish from a papilloma.
Fig 25. Papillary lesion (Papillary DCIS).

Fig 26. Usual ductal hyperplasia. If the patient is post-menopausal and not on HRT, this degree of hypercellularity warrants further investigations.
Fig 27. Usual ductal hyperplasia.

Fig 28. Collagenous spherulosis with hyaline globules.
Fig 29. DCIS. Compare with normal ductal cells (Arrow).

Fig 30. DCIS with comedo-necrosis.
Fig 31. Apocrine DCIS. Compare with normal apocrine cells (Arrow).

Fig 32. Apocrine DCIS. These cells have much more nuclear atypia as compared with normal apocrine cells.
Fig 33. Invasive ductal carcinoma.

Fig 34. Invasive ductal carcinoma.
Fig 35. Invasive ductal carcinoma. Not all vacuolated background means lactation.

Fig 36. Low grade invasive ductal carcinoma with neoplastic tubules (Arrow).
Fig 37. Mucinous carcinoma.

Fig 38. Mucin – thin, non-fibrillary and basophilic. Compare this with myxoid stroma (Fig 10).
Fig 39. Invasive lobular carcinoma. Note hypocellularity.

Fig 40. Invasive lobular carcinoma. Tumour cells are uniform and they have round nuclei. They may mimic bare bipolar nuclei.
Fig 41. Invasive lobular carcinoma. Indian file with nuclear moulding.

Fig 42. Invasive carcinoma. Note targetoid inclusion(s): cytoplasmic vacuole with mucin droplet (Arrow).
Fig 43. Radiotherapy effect. Atypical cell with low N/C ratio, bizarre shaped and vacuolated cytoplasm.
References


Orell SR, Farshid G. False-positive reports in fine needle biopsy of breast lesions. *Pathology* 2001;33:428-436
