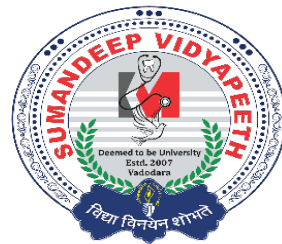


**BDS Year 3 Regular batch
Academic Year 2022-2023**

Subject: Oral Pathology & Microbiology
Topic: Biopsy & Cytology.1.2022

Dr. Vandana Shah
Professor & Head

Dept. of Oral Pathology and Microbiology



BIOPSY

DEFENITION

- ❖ The term “BIOPSY” (G. bios, life +opsis, vision) was coined by the French Dermatologist Ernest Henry Besnier in 1879
- ❖ Is the removal of tissue from the living organism for the purpose of microscopic examination and diagnosis

HISTOLOGICAL AND CYTOLOGICAL DIAGNOSIS

❖ HISTOLOGICAL

1. EXCISIONAL BIOPSY
2. INCISIONAL BIOPSY
3. PUNCH BIOPSY
4. SHAVE BIOPSY
5. TREPHINE BIOPSY
6. DRILL BIOPSY

CYTOLOGICAL SMEAR

- 1 . FNAC
2. EXFOLIATIVE CYTOLOGY
Tzanck smear
3. BRUSH BIOPSY
4. IMPRINT SMEAR
- 5 EXUDATE SMEAR

The Provisional clinical diagnosis is especially important in guiding the technique .

EXCISIONAL BIOPSY

- ❖ Complete removal of entire lesion with an adequate margin of normal tissue on all sides and made available for histopathologic examination.

- ❖ INDICATIONS

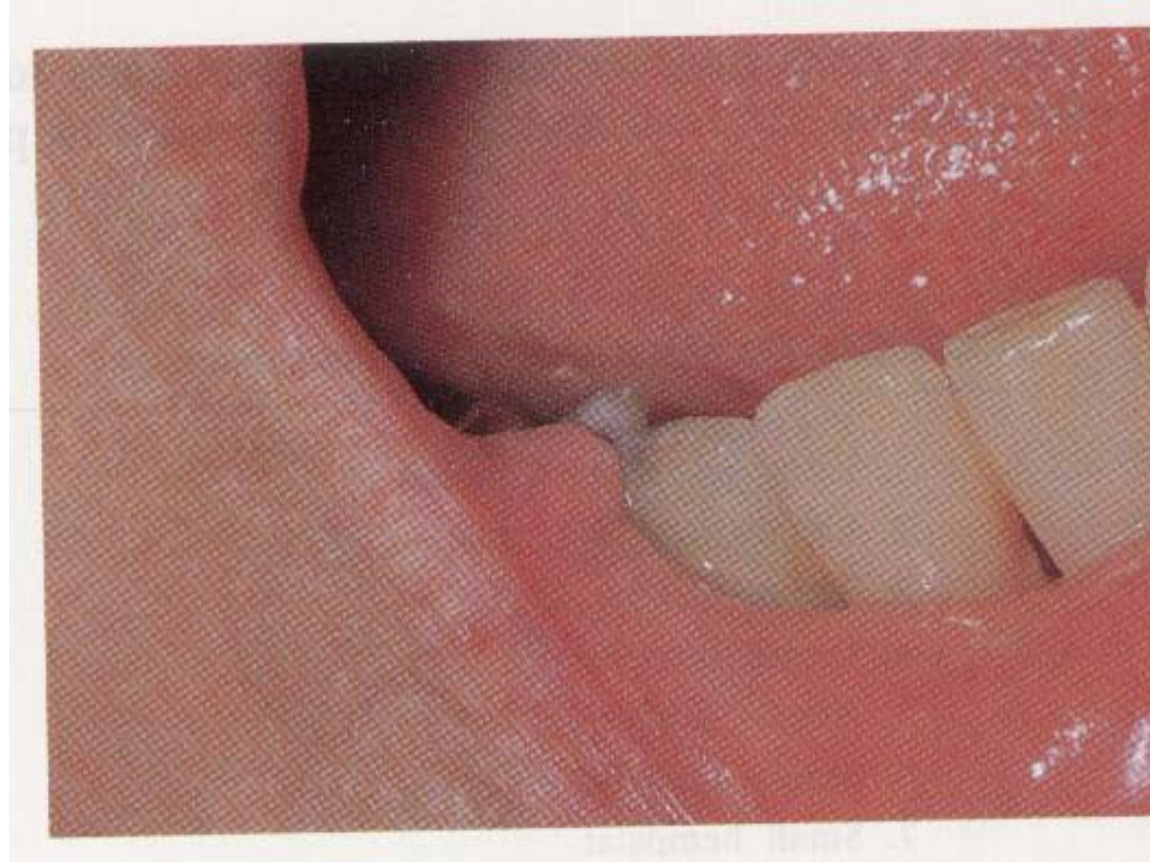
1. For discrete lesions which are less than 1 cm in diameter and on
2. C/E appears to be benign

Examples :Fibromas, Papillomas, mucocoeles, Pyogenic granulomas,verruca vulgaris etc

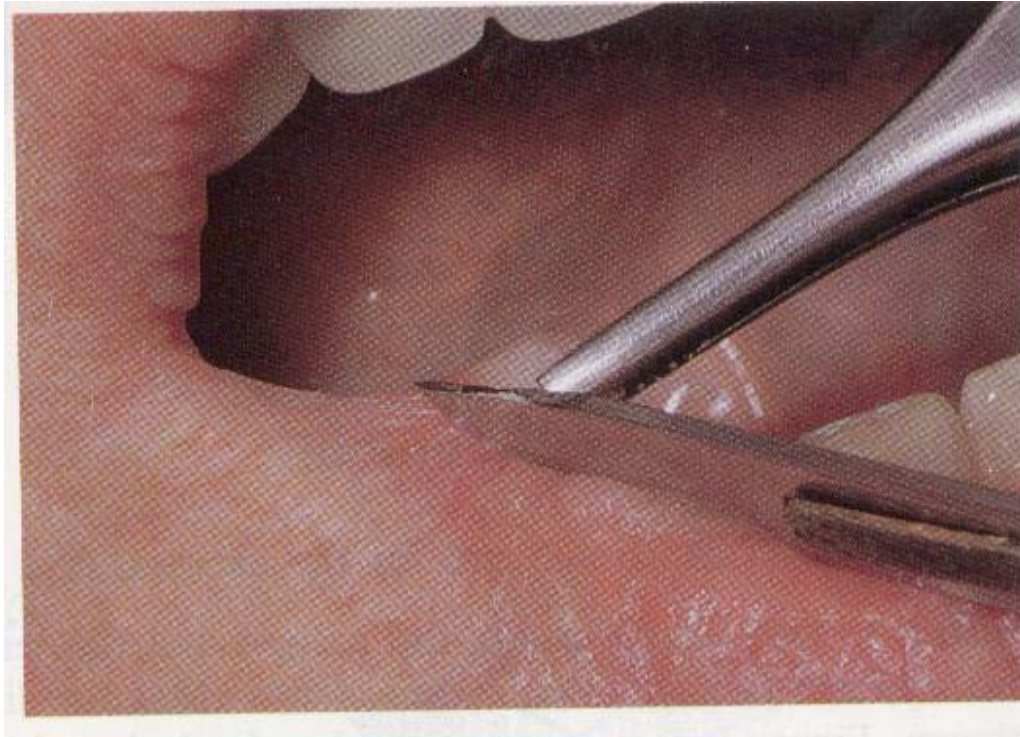
3. Pigmented and small vascular lesions.

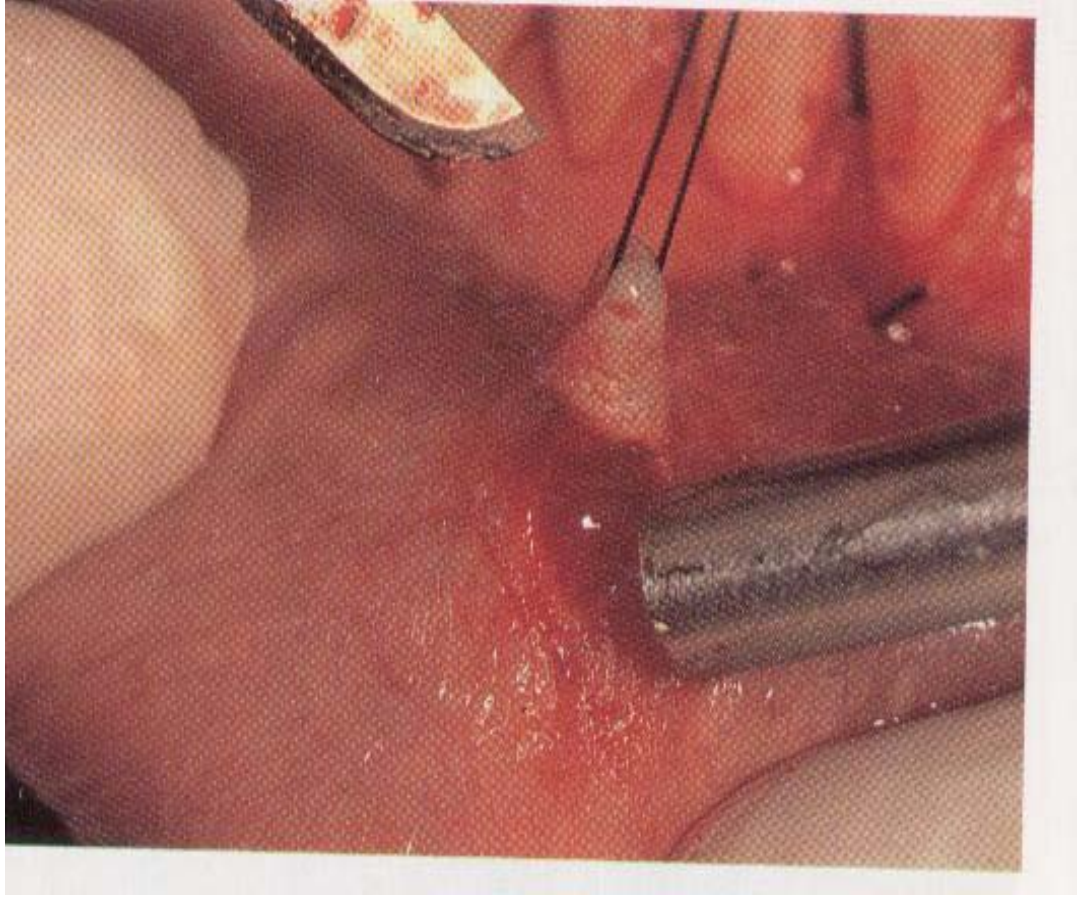
<http://www.webmd.com/cancer/what-is-a-biopsy>

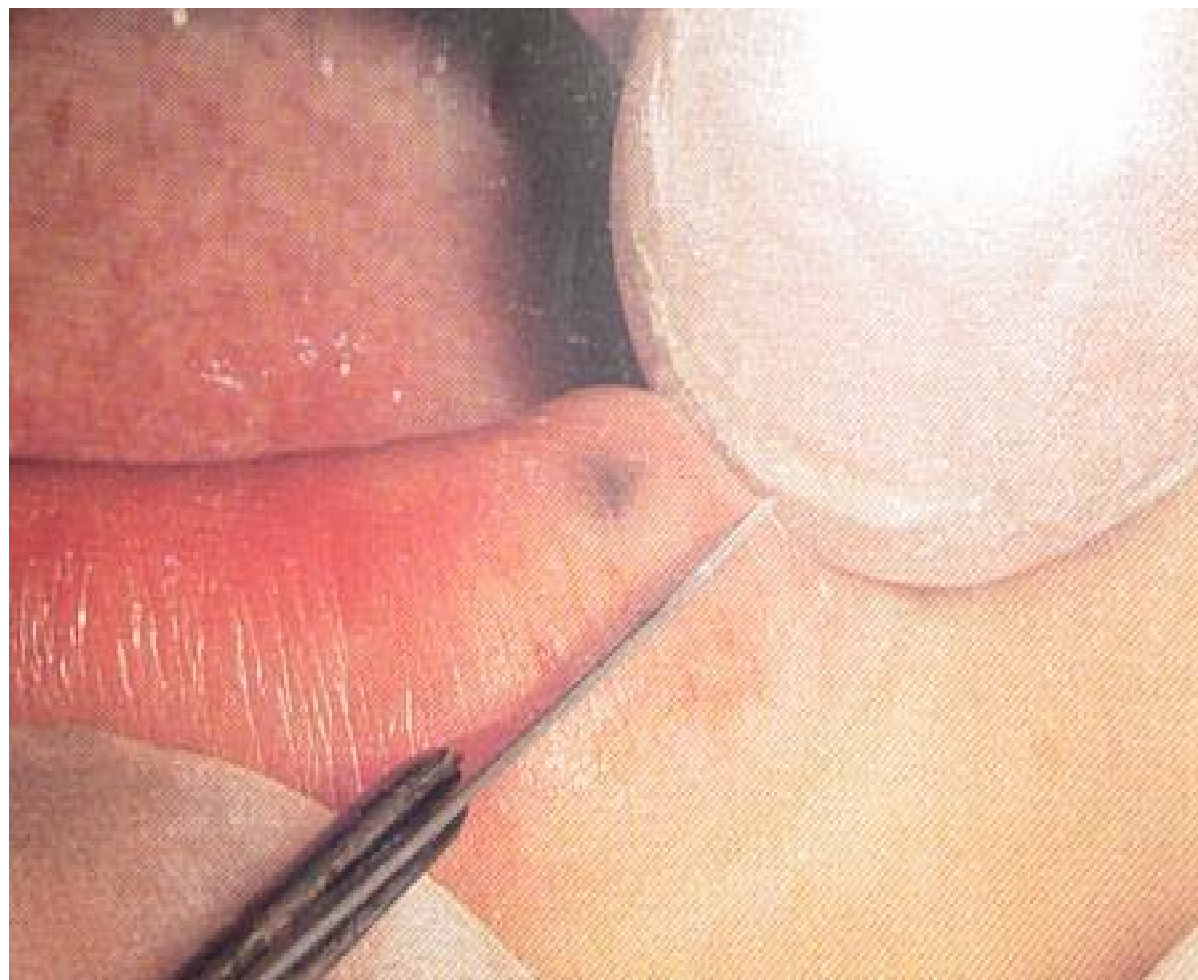
•TECHNIQUE



Applying traction to lesion

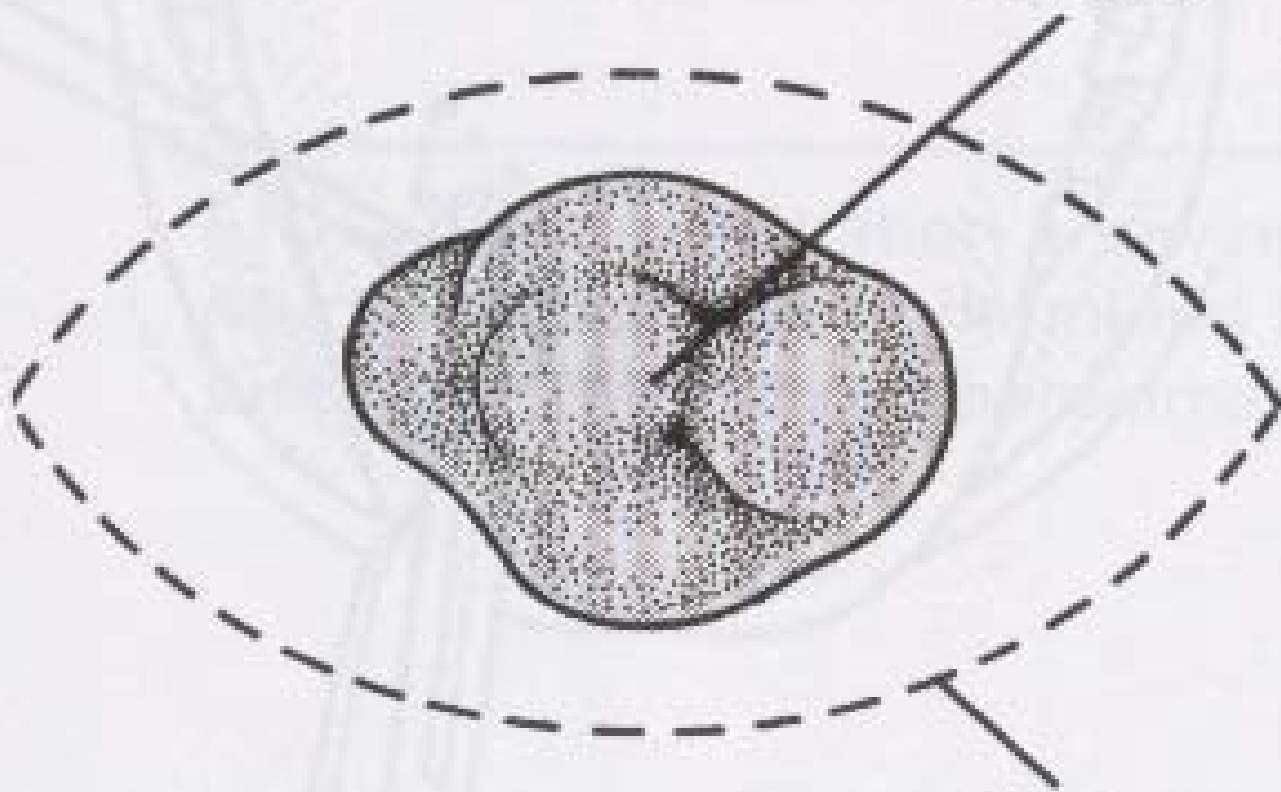






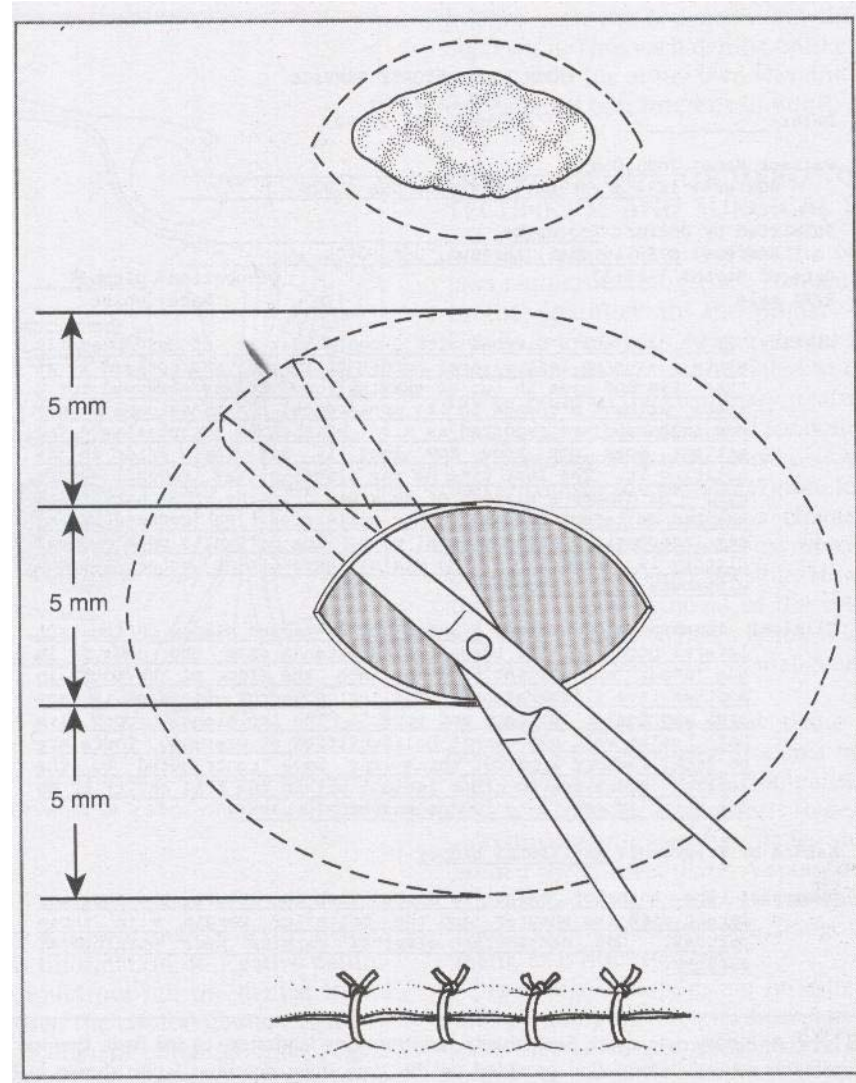
A

Lesion



Incision







h

i

❖ ADVANTAGES

- ❖ Offers dual advantage i.e., it is both diagnostic and curative.

❖ DISADVANTAGES

- ❖ Specimens likely to get crushed by using tissue forceps.

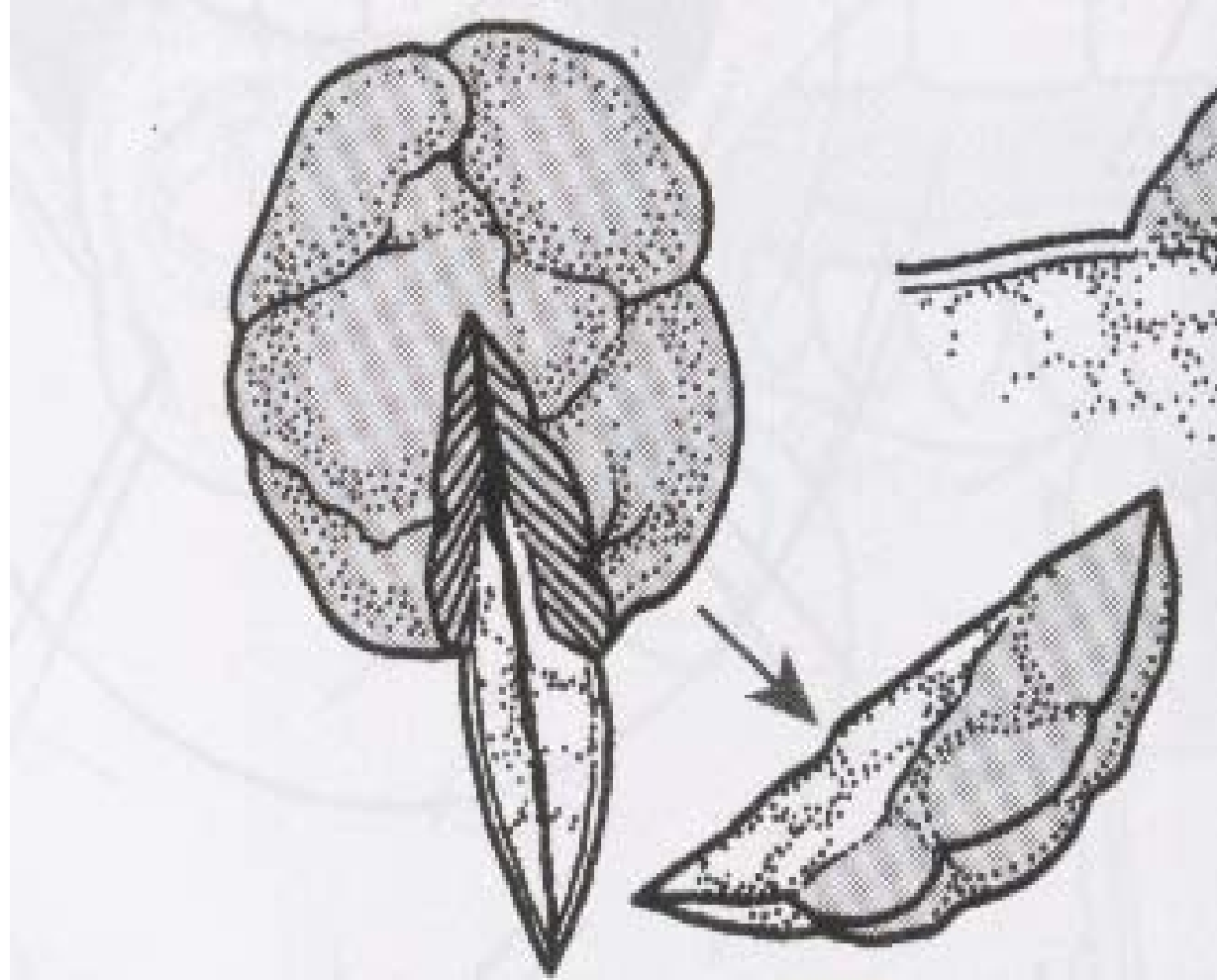
INCISIONAL BIOPSY

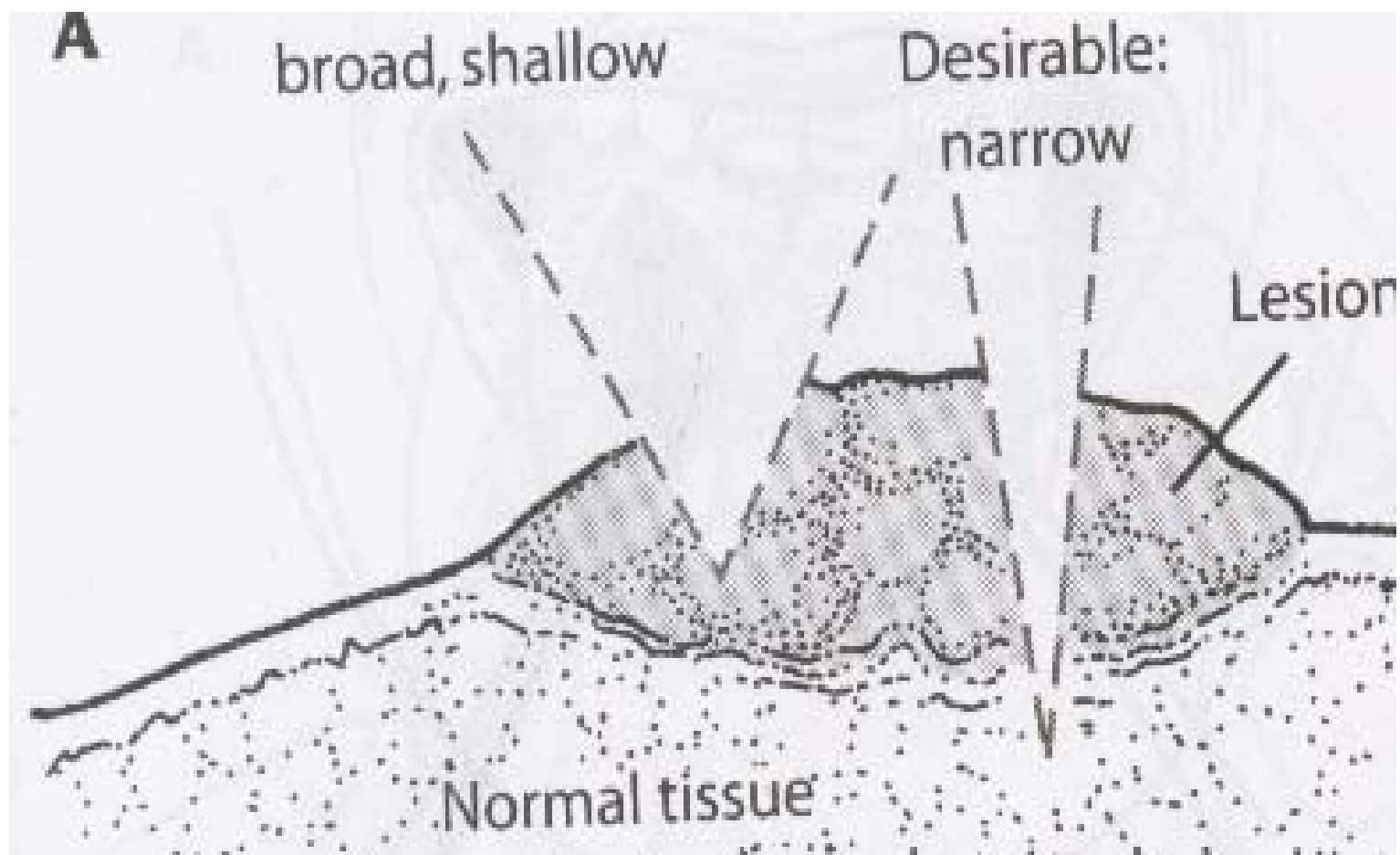
- ❖ Biopsy that samples only a particular or representative part of the lesion

INDICATIONS

- ❖ If the lesion is larger than 1 cm in diameter
- ❖ Hazardous location
- ❖ In suspected malignant lesions-Chronic ulcer or sq cell ca
- ❖ Mucocutaneous lesions
- ❖ Vesiculobullous lesions
- ❖ Precancerous lesions
- ❖ Intrabony lesions.

- ❖ A biopsy in a wedge fashion should be performed.
- ❖ It should be removed at the margin of the lesion.
- ❖ Area of necrosis should be avoided.
- ❖ Incision should be of sufficient length to include underlying tissues.
- ❖ A deep, narrow sample.







ADVANTAGES

1.

Gives representation of large areas of defects.



DISADVANTAGES

1.

Dissemination of cancer cells into circulation occurs by incisional biopsy of oral squamous cell carcinoma.

J Oral Pathol Med 2000:29:303-7

TOLUIDINE BLUE

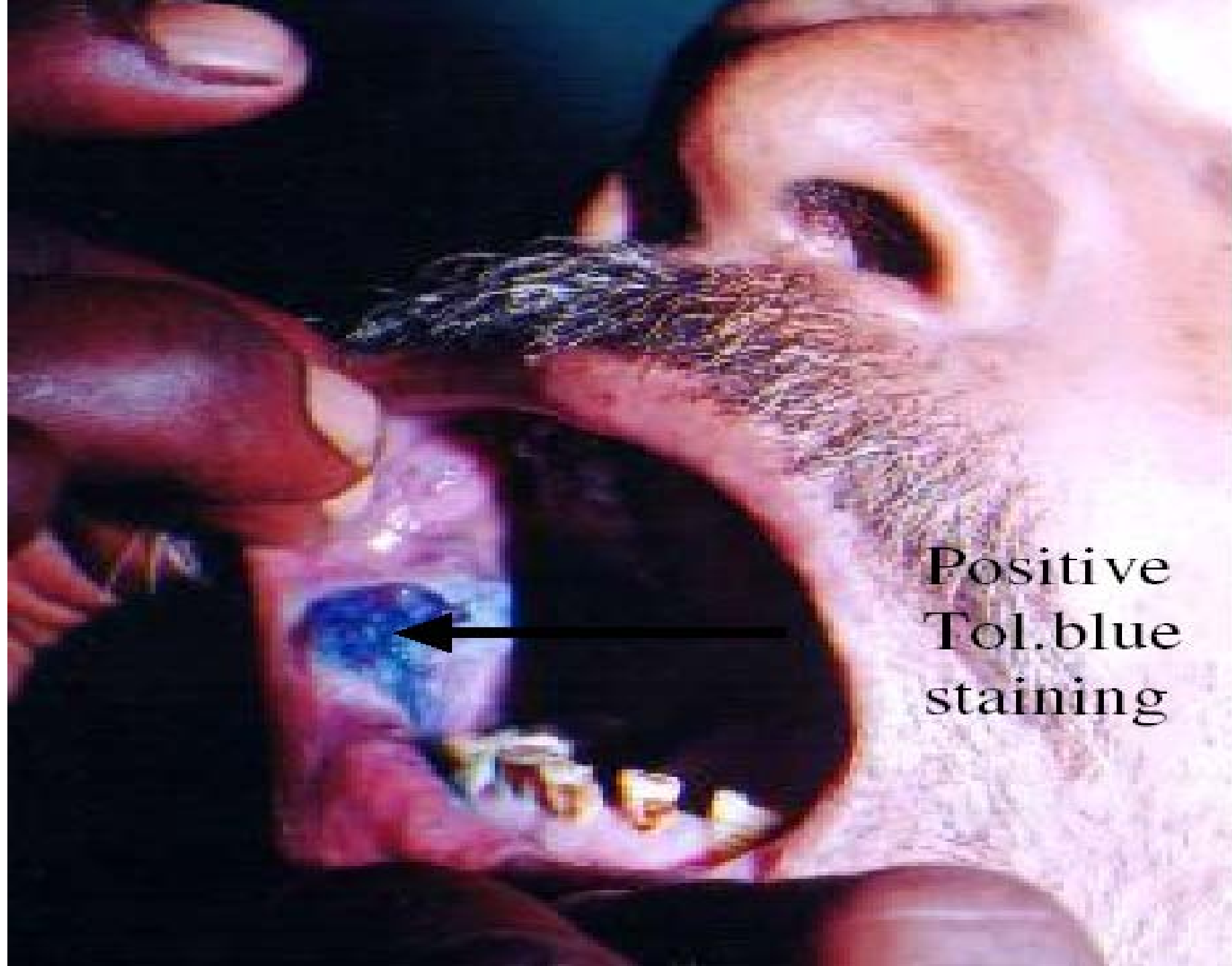


❖ ADVANTAGES

1. High sensitivity

❖ DISADVANTAGES

1. Not specific for ca.



Positive
Tol.blue
staining

PUNCH BIOPSY



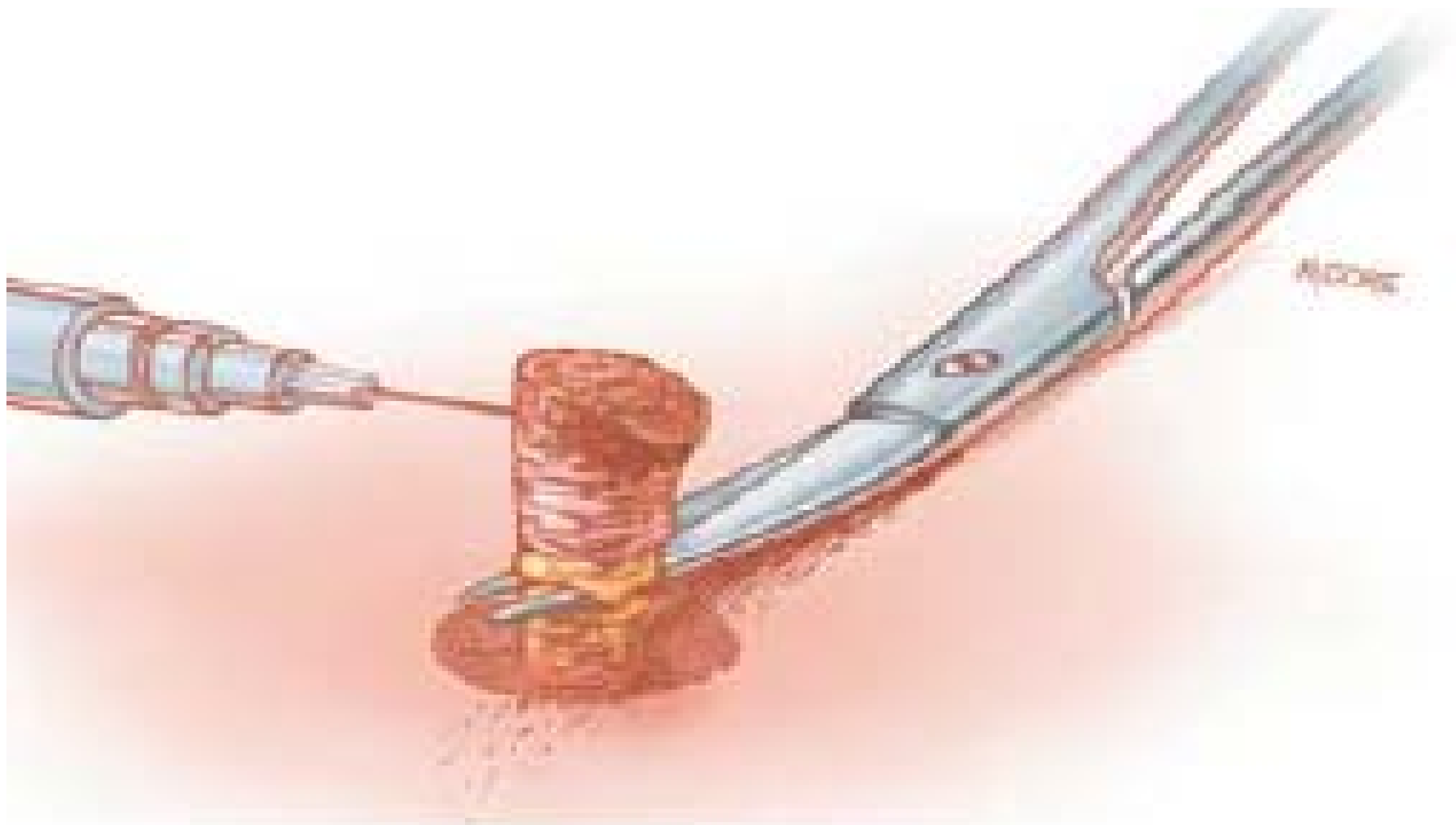
INDICATIONS

1. For surface lesions.
2. Considered the primary technique for obtaining diagnostic full thickness skin specimens.
3. Most commonly used procedure to obtain tissue from areas that are not readily accessible i.e., of tissues accessible to endoscopy

OWING TO THE ACCESSIBILITY OF LESIONS OF THE ORAL CAVITY, THE PUNCH BIOPSY IS RARELY INDICATED

Punch comprises a circular blade attached to a plastic handle





❖ ADVANTAGES

1. Technique is easy, useful in mass screening
2. Post surgical morbidity is minimal
3. Suturing is not required.

❖ DISADVANTAGES

1. Depth of penetration is limited
2. Technique cannot be employed on freely movable mucosa.
3. Palatal and gingival sites
4. Ulcerated areas may be difficult to punch.

SHAVE BIOPSY

- ❖ This is a technique that is commonly used for skin lesions that are raised or benign exophytic lesions and superficial inflammatory lesions.

Examples : Seborrheic keratosis, Benign nevi, Basal cell ca

Contraindicated for suspected melanomas.

ADVANTAGES

- ❖ The lower dermis will be left intact.

TREPHINE BIOPSY

- ❖ One of the two main types of bone marrow test
- ❖ A bone marrow trephine means removal of 1 – 2 cm core of bone marrow in one piece.
- ❖ Bone marrow tests are done for cancers that are most likely to affect the bone marrow, such as
 1. Lymphomas (non Hodgkin's lymphoma or Hodgkins lymphoma)
 2. Leukemias
 3. Myeloma

It is also used for maxillofacial reconstructive procedures that is harvesting iliac bone crest by trephination.

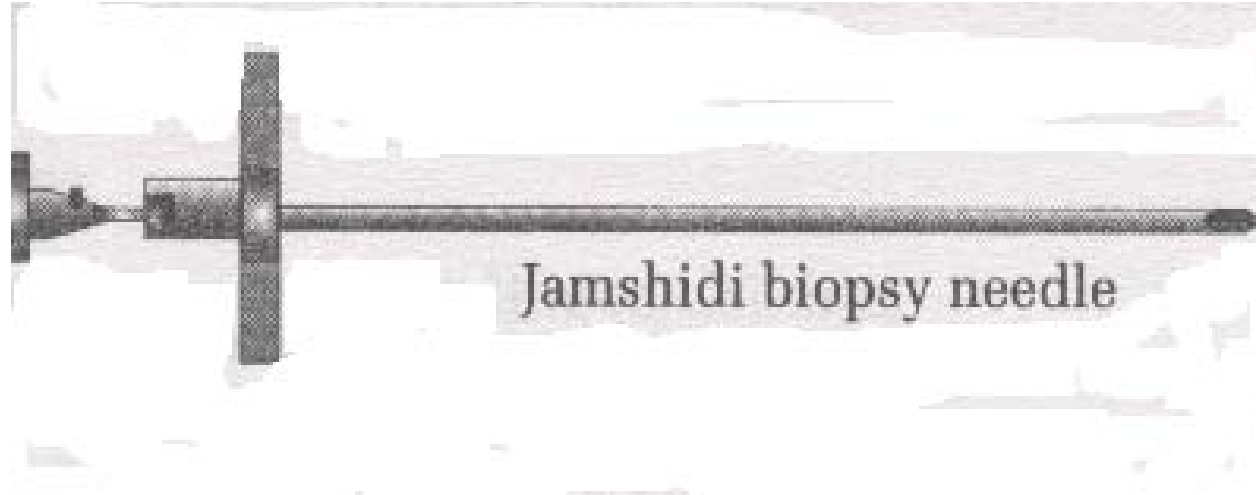
By: Logan, R. M.; Goss, A. N.. Australian Dental Journal, Jun2010 Supplement, Vol.

55, p9-13, 5p, 4 Color Photographs, 1 Chart; DOI: 10.1111/j.1834-7819.2010.01194.



INDICATIONS

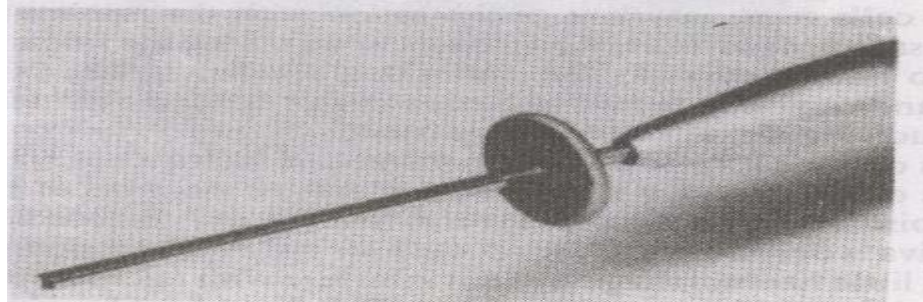
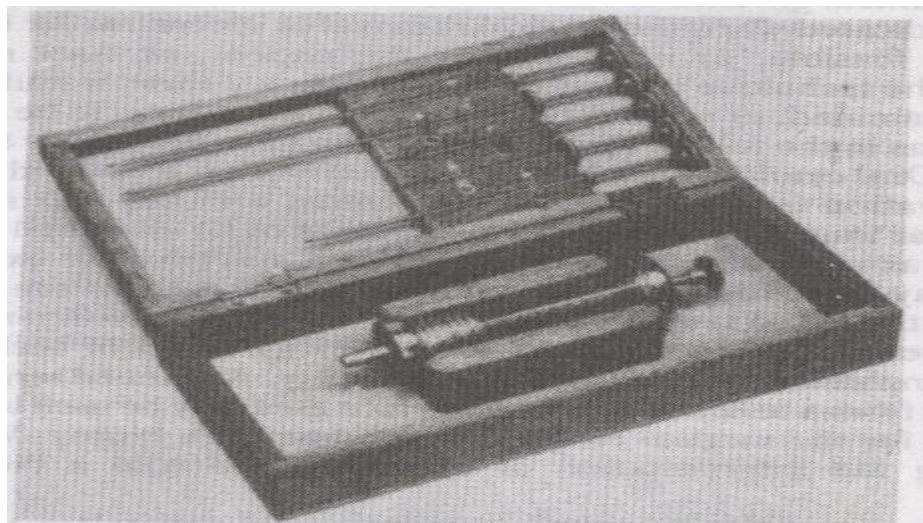
1. Lymphomas (non Hodgkin's lymphoma or Hodgkin's lymphoma)
2. Leukaemias
3. Myeloma



Jamshidi biopsy needle

DRILL BIOPSY

- ❖ Modified Ellis biopsy drill that fits into a straight handpiece
- ❖ Specimens 1.2 cms long and 1.4 mm in diameter.
- ❖ Indicated in intra osseous lesions.
- ❖ Disadvantage of generating heat during procedure.



NEEDLE/CORE BIOPSY

❖ Vim Silvermans Needle biopsy

❖ INDICATIONS

1. Deep seated lesions i.e., neoplasms of the salivary gland, neck masses
2. Liver biopsy
3. Kidney biopsy
4. Lung biopsy-rarely.

ADVANTAGES

Removes a strip of intact tissue.

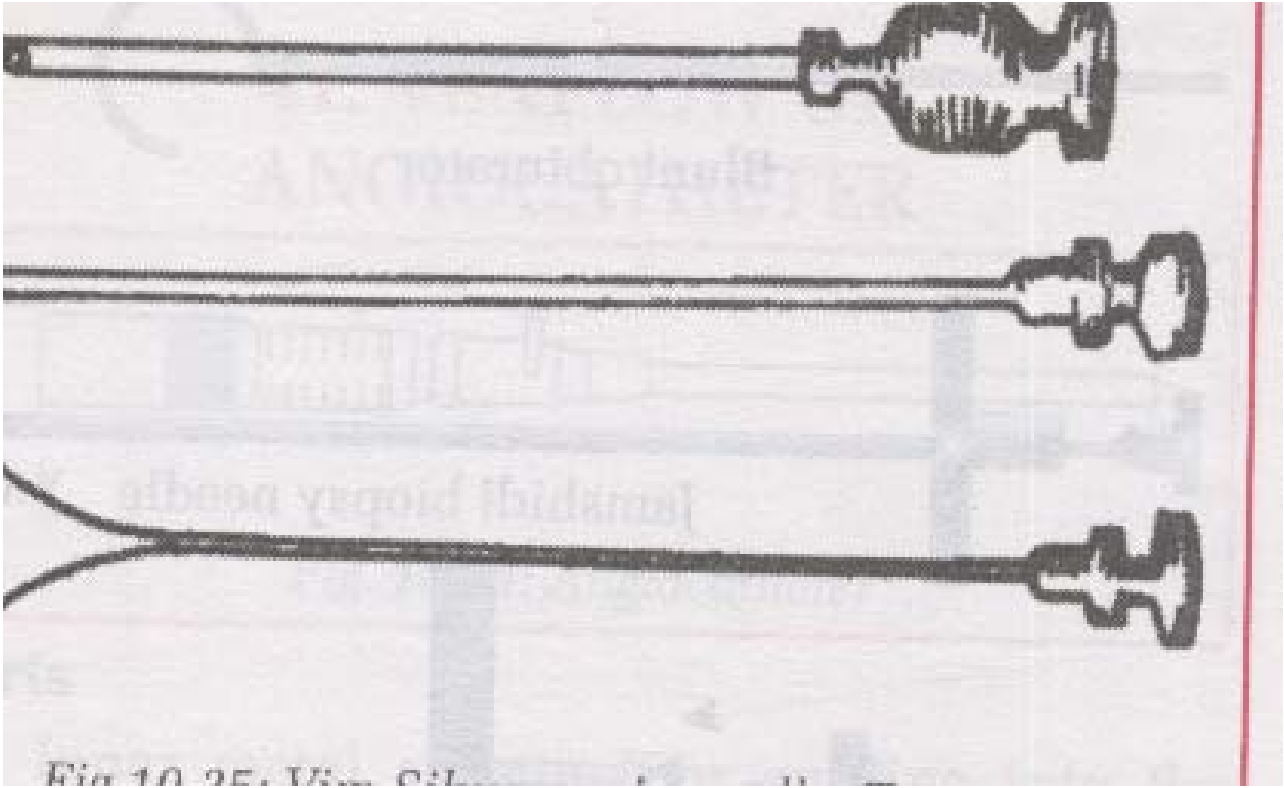
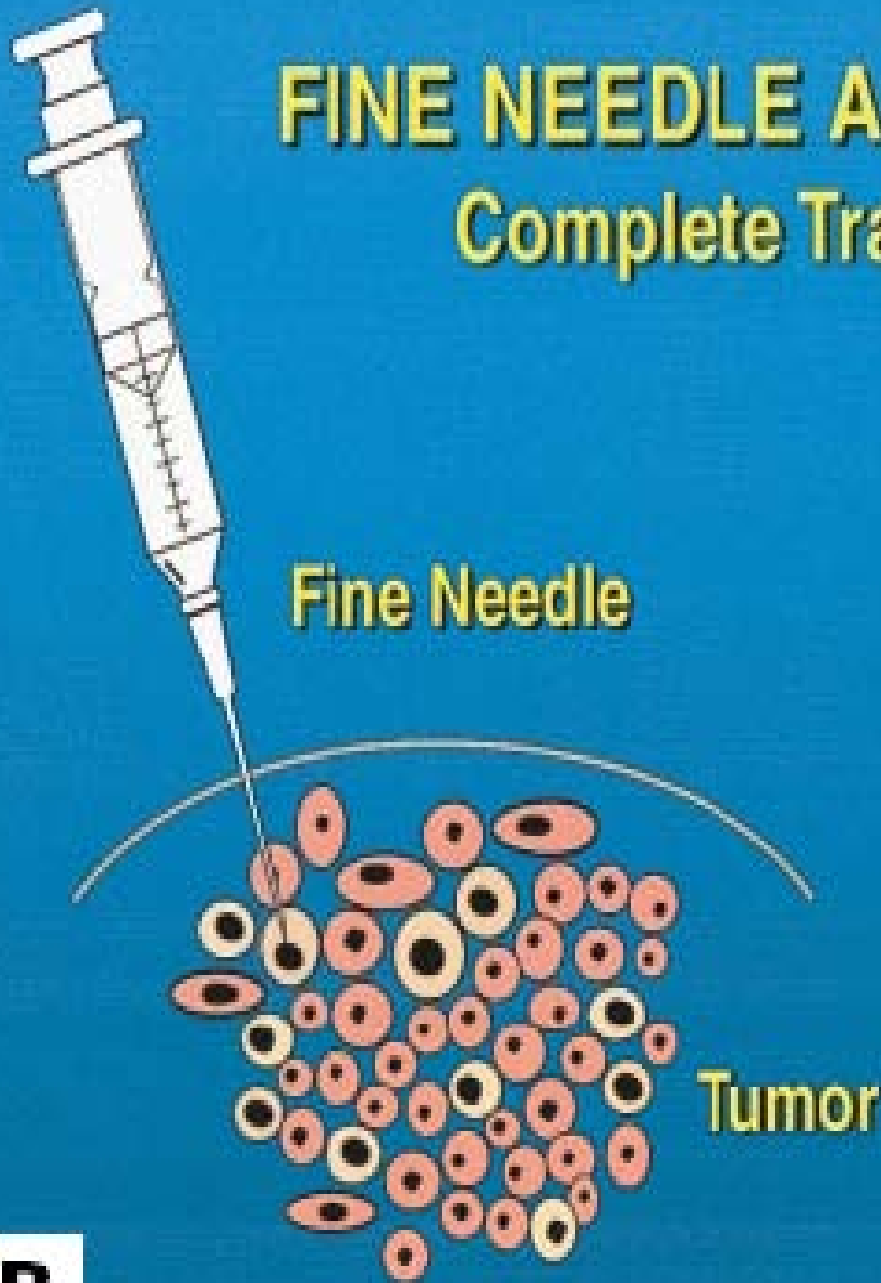


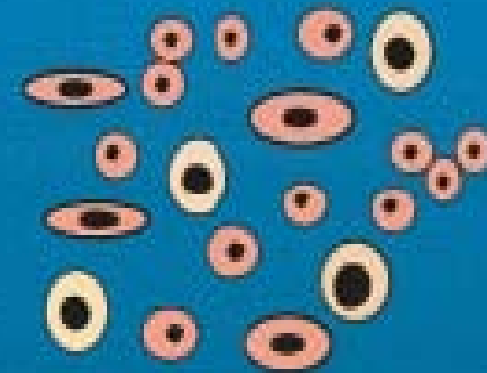
Fig. 10.25. Various types of ovaries.

FINE NEEDLE ASPIRATION BIOPSY

Complete Transtumor Sample



Specimen



B

Aspiration of Palpable Masses

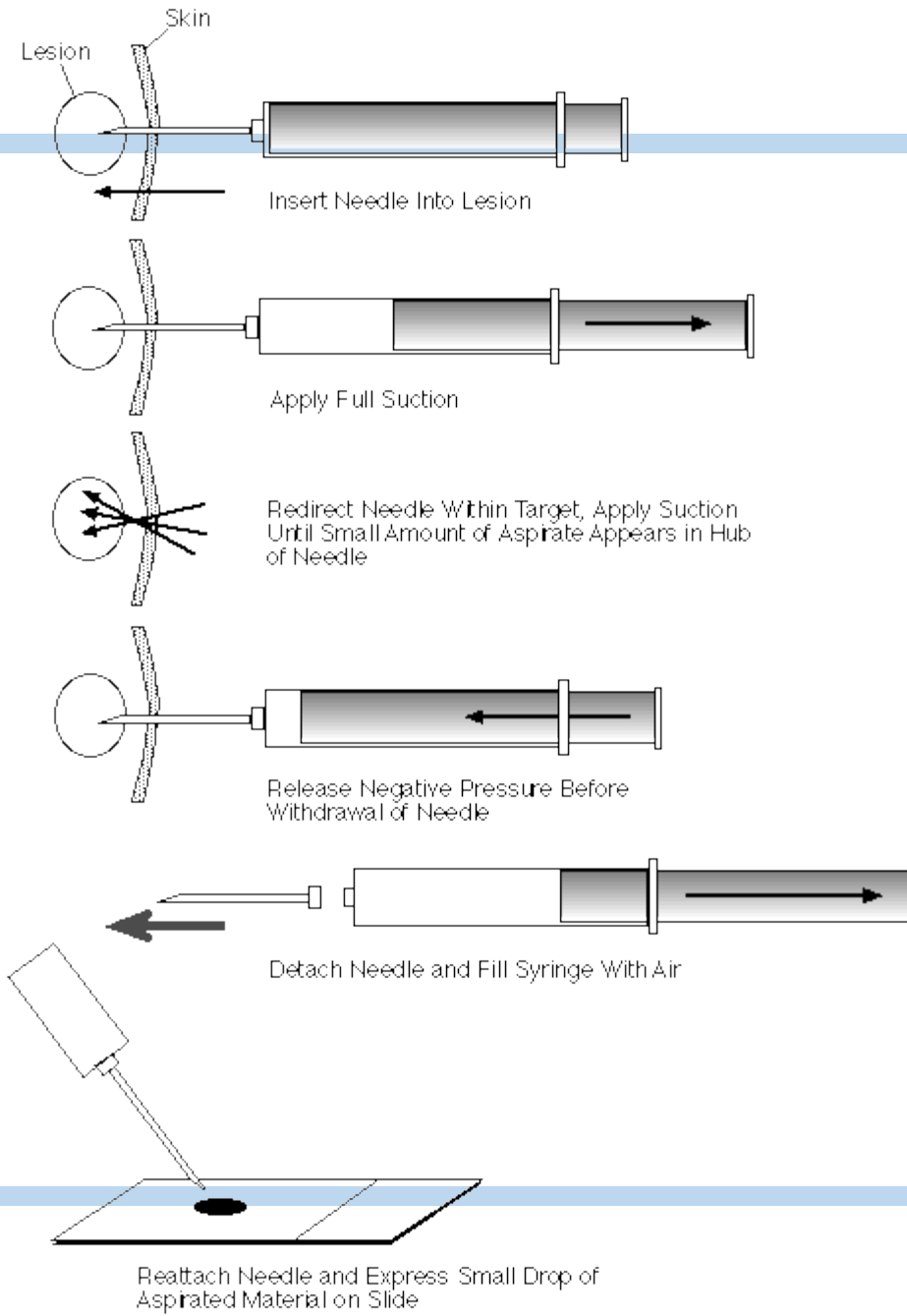




Figure 1-3 George N. Papanicolaou, 1954, in a photograph inscribed to the author.

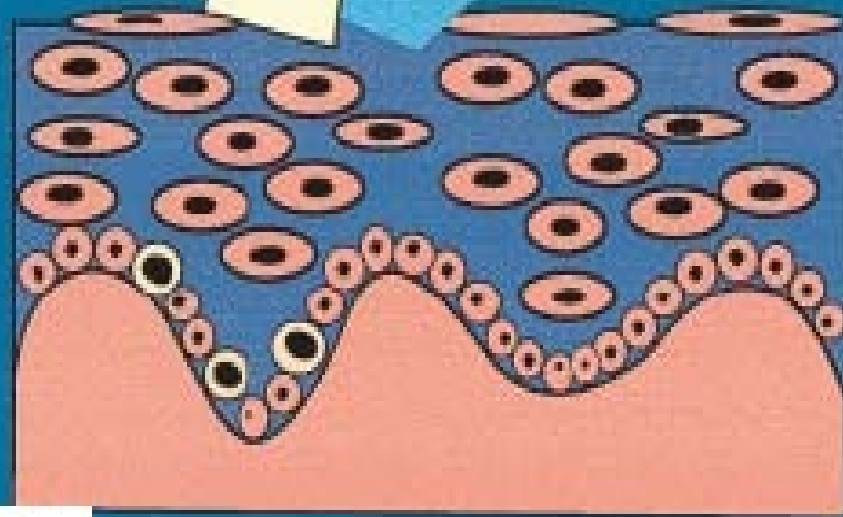
Copyright © 2006 Lippincott Williams and Wilkins.

EXFOLIATIVE CYTOLOGY

Only Surface Cells Captured

Broom sweep limited
to superficial cells

Specimen



Superficial
|
Intermediate
|
Basal



C

RESULTS OF EXFOLIATIVE CYTOLOGIC SMEAR

- ❖ **Class I (Normal)**
- ❖ **Class II (Atypical)**
- ❖ **Class III (Intermediate) – Biopsy is recommended.**
- ❖ **Class IV (Suggestive of cancer) – Biopsy is mandatory**
- ❖ **Class V (Positive for cancer) – Biopsy is mandatory.**

Negative cytology report does not rule out cancer and that a repeat smear or biopsy is indicated in all clinically suspicious lesions.

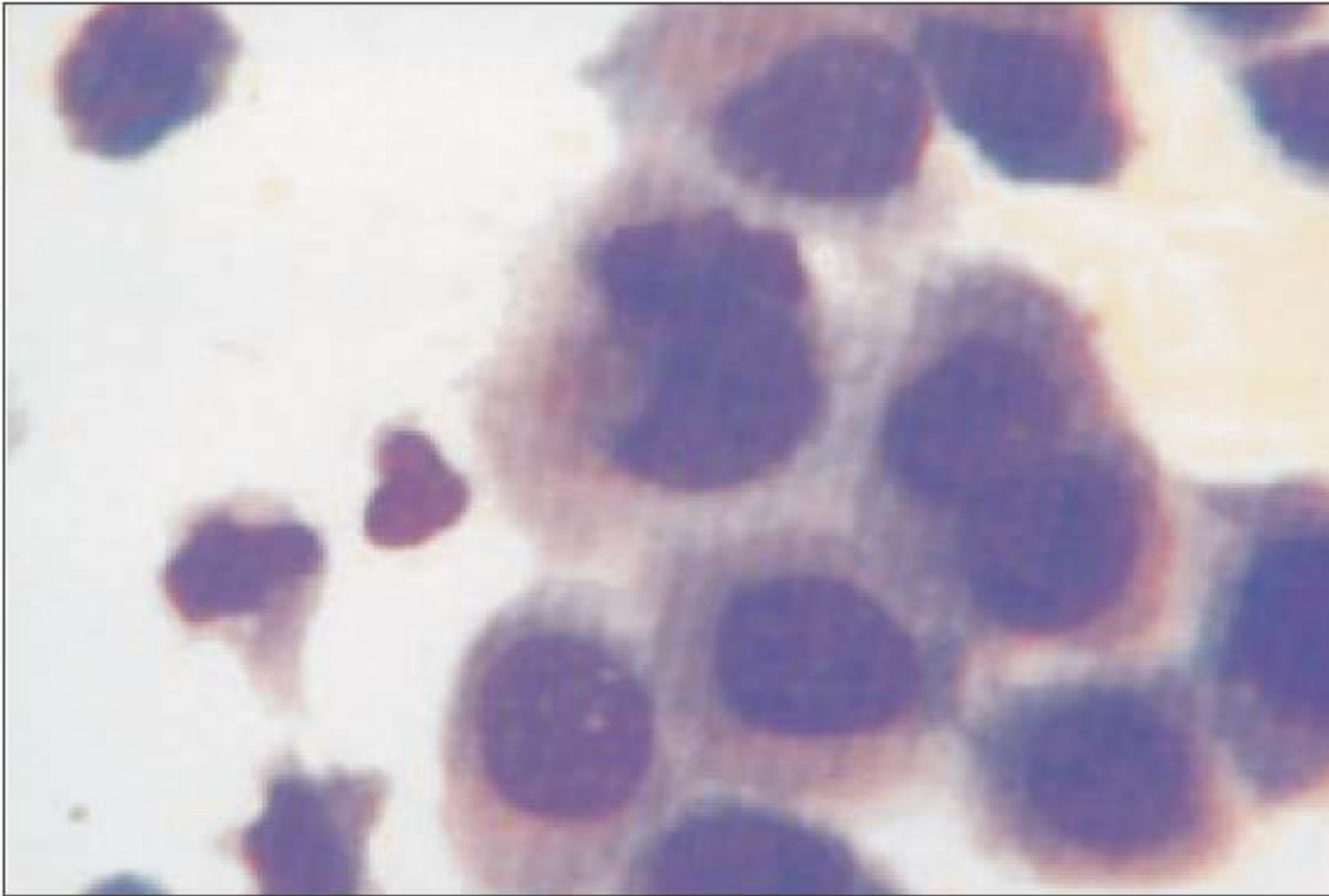
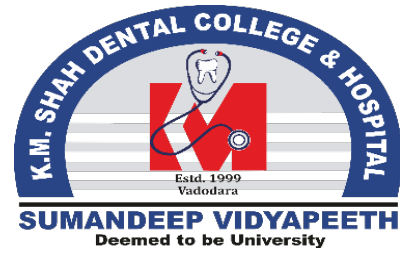
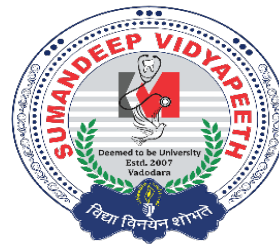


Figure 1: A typical acantholytic cell of pemphigus vulgaris



BRUSH BIOPSY

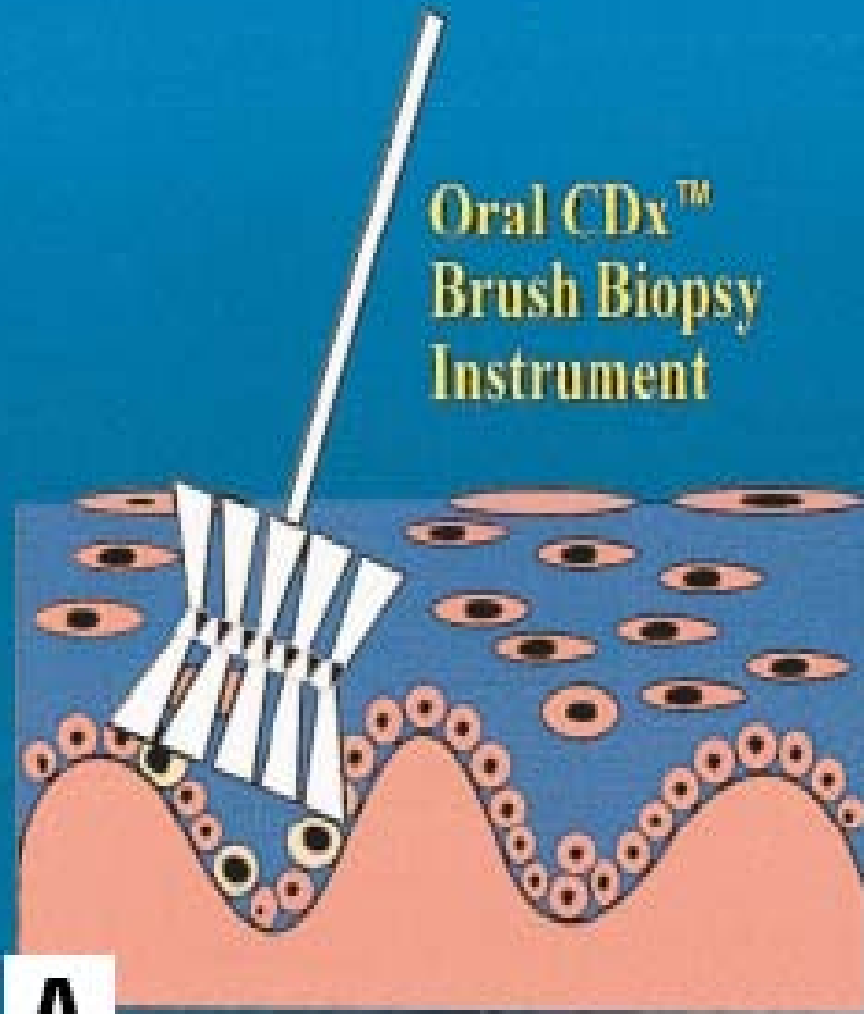
“AS A SCREENING TOOL”





BRUSH BIOPSY

Complete Transepithelial Sample

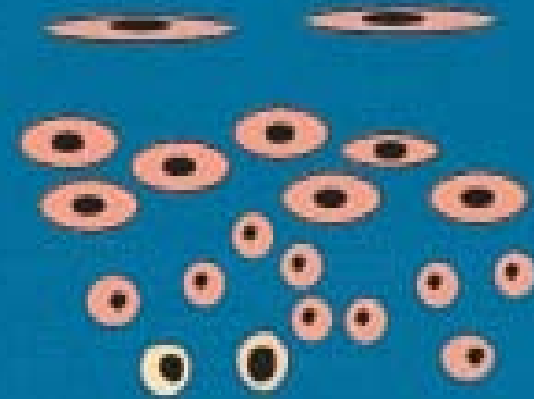


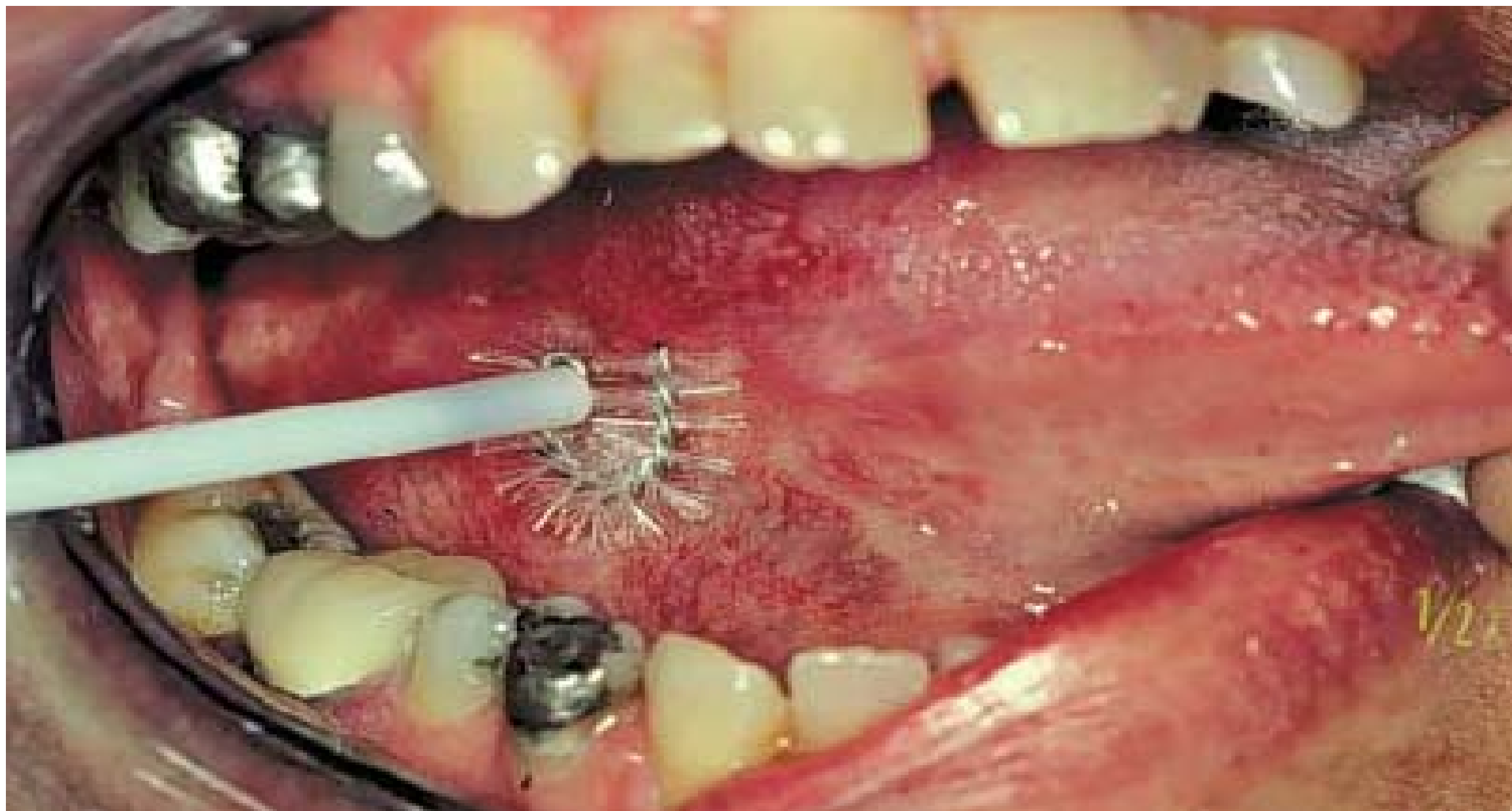
A

Oral CDx™
Brush Biopsy
Instrument

Specimen

Superficial
|
Intermediate
|
Basal





RESULTS OF BRUSH CYTOLOGY SPECIMENS

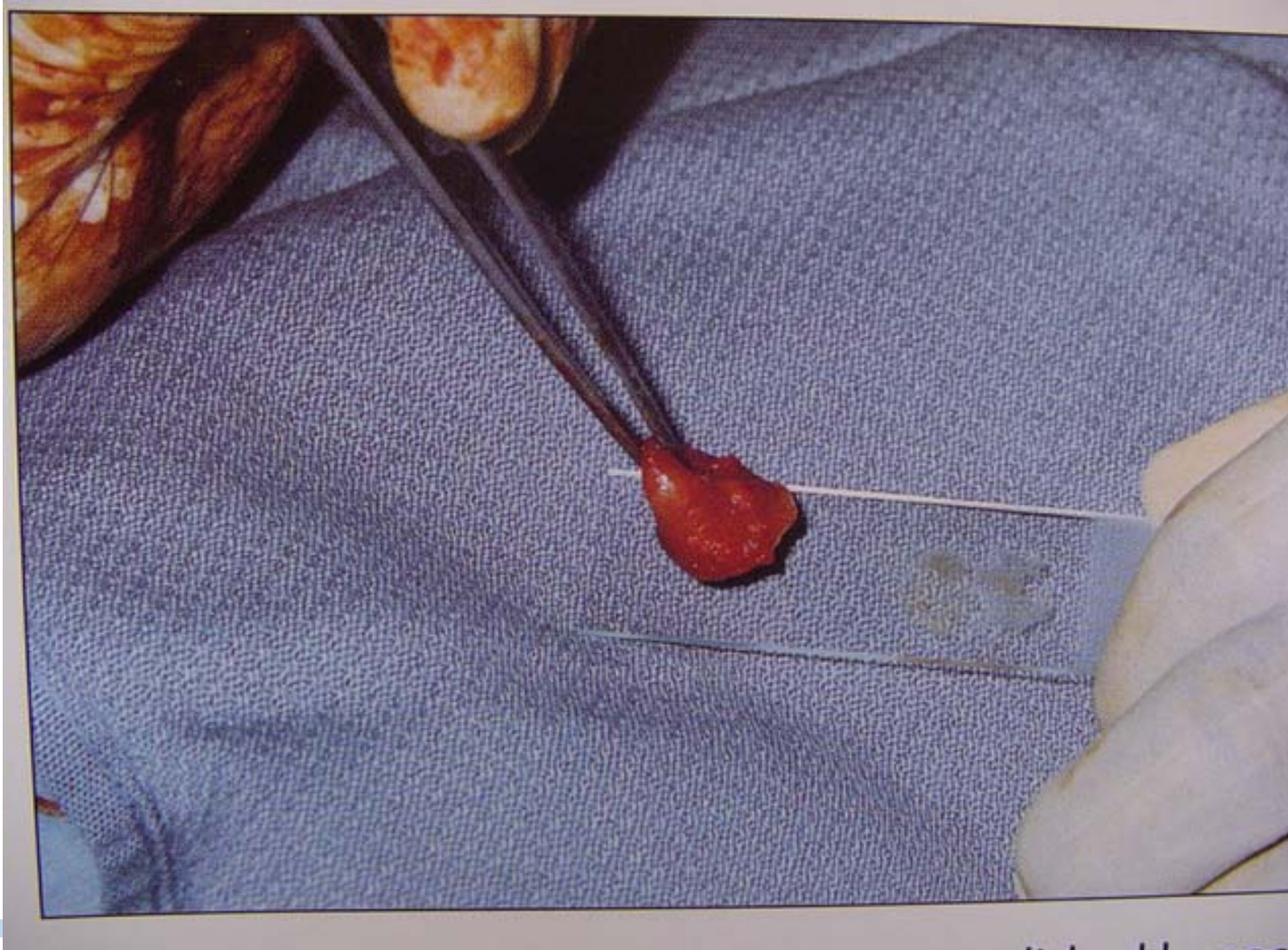
- ❖ Inadequate
- ❖ Negative
- ❖ Atypical
- ❖ Positive

For atypical or positive results, patient should receive follow – up scalpel biopsy.
Negative result, clinical follow-up of persistent oral lesions is recommended.



Pin-point
bleeding

TOUCH PREPARATION



FR

Fig 1-1b The firm brush is able to capture deeper cells to the level of the basement membrane. (Courtesy of Oral Scan Labs, New York.)

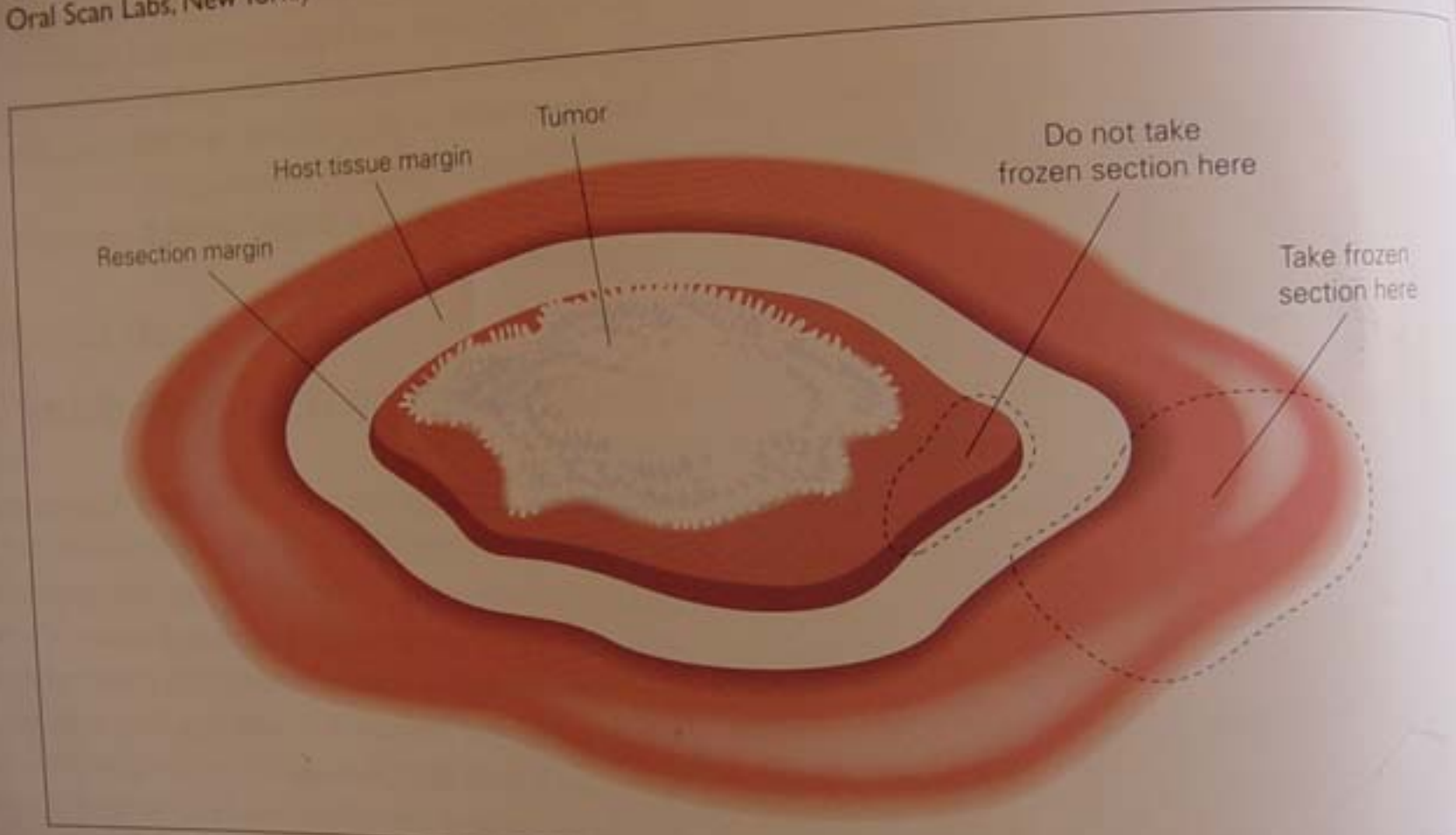


Fig 1-2 Frozen sections should be taken from the host tissue edge rather than from the specimen tissue edge.

reference

- ❖ <http://www.webmd.com/cancer/what-is-a-biopsy>
- ❖ By: Santos, Pedro Bello; Velásquez, Joel Omar Reyes. Medicina Oral (16656024), 2001, Vol. 3 Issue 4, p176-179, 4p; Language: Spanish
- ❖ Dos Santos, José Antonio Rossi; Capella, Diogo Lenzi; Rozza, Rafaela Elvira; Ferreira, Stefânia Jeronimo; Berti-Couto, Soraya de Azambuja; Sant'Ana-Filho, Manoel; De Lima, Antonio Adilson Soares; Westphalen, Fernando Henrique; Couto-Souza, Paulo Henrique. Journal of Oral & Maxillofacial Research, Apr2011, Vol. 2 Issue 2, Special section p1-6, 6p; DOI: 10.5037/jomr.2011.2203
- ❖ By: Logan, R. M.; Goss, A. N.. Australian Dental Journal, Jun2010 Supplement, Vol. 55, p9-13, 5p, 4 Color Photographs, 1 Chart; DOI: 10.1111/j.1834-7819.2010.01194.

FINE NEEDLE ASPIRATION CYTOLOGY



CONTENTS

- ❖ Introduction
- ❖ History
- ❖ Definition
- ❖ Equipment
- ❖ Aspiration biopsy procedures
- ❖ Smear preparation
- ❖ FNAC staining procedure



CONTENTS

- ❖ FNAC of salivary glands
- ❖ FNAC of lymph nodes
- ❖ FNAC of common head and neck lesion
- ❖ Complications
- ❖ Newer techniques
- ❖ Conclusion



Introduction

Rubin (1994) has classified cytological methods into three types namely.

- ❖ Exfoliative cytology (examination of cells that spontaneously shed into body fluids / secretions)
- ❖ Abrasive cytology (examination of cells that are dislodged from body surfaces by various tools: Artificial surface exfoliation)
- ❖ Fine needle aspiration cytology (Deep micro biopsy) [Examination of cells that are obtained by means of a fine gauge needle].

- ❖ The complex and often disfiguring operations necessary for the treatment of diseases can be confirmed by a simple method of biopsy,
- ❖ If possible one that could be carried out in OPD for histological confirmation of the diagnosis before contemplating on an operation.
- ❖ Thus developed the technique of **NEEDLE BIOPSY**.

- ❖ fine needle biopsy is an effective tool in evaluating and diagnosing suspected lumps or masses.
- ❖ A quick diagnosis can mean that cancer is detected early, giving more options for treatment

Historical aspect



- ❖ 1833: Leyden, first used needle and syringe to obtain specimen from the pneumonia of the lungs.
- ❖ In Great Britain 1927, Dudgeon and Patrick proposed the needling of tumors as a means of rapid microscopic diagnosis.
- ❖ 1930 : Dr. H Martin – radiotherapist – First published paper on needle aspiration method using 18 gauge needle

- ❖ 1933 : Stewart -- American pathologist – became the first pathologist to interpret aspiration smears
- ❖ 1950 : Netherland and Sweden –physicians and hematologist started actual use of fine needle for aspiration
- ❖ 1960 – 1970 – use of 18 gauge needle was completely stopped
- ❖ 1980 : when interest in FNAC was fueled by radiologist to evolve ultrasound guided FNAC
- ❖ Field of FNAC continuous to evolve

NOMENCLATURE

The needle biopsy is known as

**FINE NEEDLE ASPIRATION BIOPSY (FNAB) OR
FINE NEEDLE ASPIRATION CYTOLOGY (FNAC)**

- ❖ **Thomson** has referred to this as “Thin needle selective sampling”.
- ❖ **Svante, Gregory** refer the whole art of obtaining the specimen as **FNAC /FNAB** and to the material obtained as either samples, smears or aspirations.
- ❖ **Trott** refers to the whole art of obtaining the specimens and the together as **FNAC / FNAB**.

Terminology

Cytopathology

“It is the art and science of interpretation of cell from the human body, that either exfoliate (desquamate) freely from the epithelial surface or are removed from the various tissue sources by various clinical procedures such as washings of organs with saline, brushing with help of fibroptic instruments and needle aspiration of tissue”.

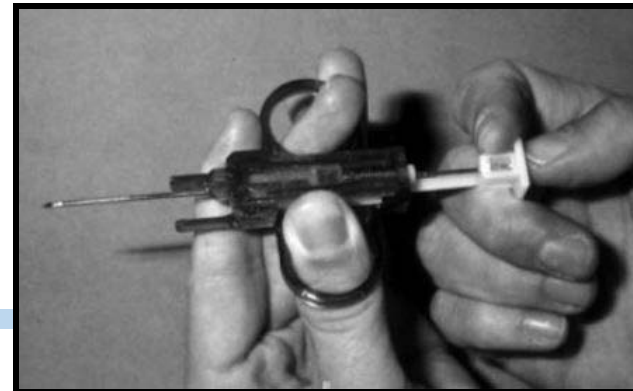
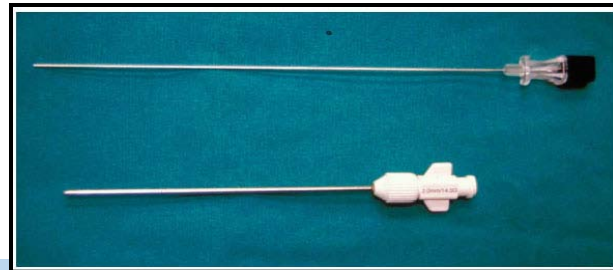
(As per Leopold Koss)

“As the name **Fine needle aspiration cytology** is a minimally invasive procedure used for the diagnosis of a mass”

- ❖ For FNA biopsy, most use “fine” or “thin” (22 to 27gauge) needles; most commonly used is a 25-gauge needle.
- ❖ External diameter 0.6 to 1.0 mm

Needle Core Biopsy

- ❖ **Core biopsy** is another method of 'tissue diagnosis' - that is, a way of sampling the cells in a suspicious lump or mass..
a more invasive procedure than FNA,
- ❖ Involves making a small incision (cut) in the skin.
- ❖ A large needle is then passed through this incision

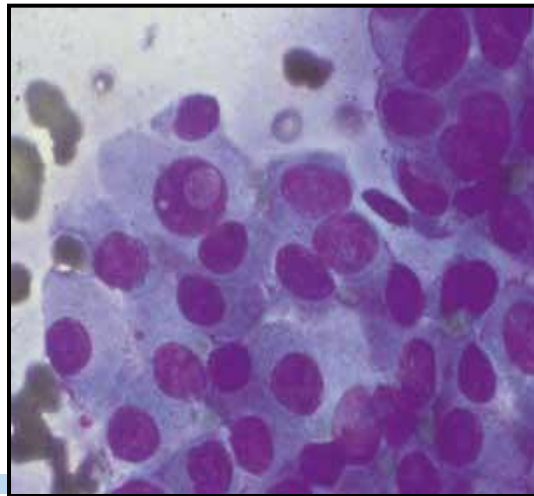


- ❖ **Needle core biopsy is generally safe and quick, and can be performed on an awake cooperative patient.**
- ❖ **The Histopathological results are generally more accurate than fine needle aspirate cytology, but not as accurate as biopsy**

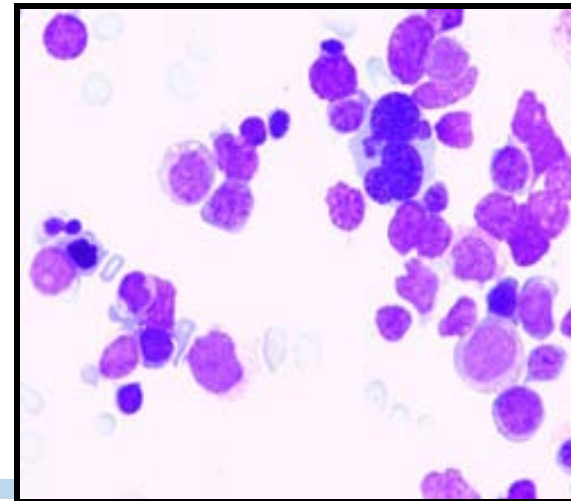
The samples of tissue taken differ from those taken during FNA.

- ❖ The cells from a fine needle aspiration biopsy are sucked up randomly into the needle -- disorganized jumble of cells
- ❖ core biopsy sample-- the larger needle allows the cells to be removed with their relationship to each other intact.--- clump of cell

FNAC



CORE BIOPSY



FNA Vs Tissue Core Biopsy

FEATURE	FNA	TISSUE CORE BIOPSY
Speed of reporting	Results possible on same day	In general, result obtained the following day
Anaesthesia	Not needed (generally)	May be needed
Complications of Procedure	Usually minor	May be serious
Inadequate specimens	Common	Less common
Cost	Low	Higher
Cellular Sampling	Large no. of cells	Smaller no. of cells
Tissue architecture	Not preserved	Preserved
Assessment of adequacy of Sample	Can be done at bedside	Difficult at time of biopsy
Spare tissue for special tests	Not usually	Usually

Indication in head and neck lesions

- SALIVARY GLAND LESIONS
- THYROID LESIONS
- PARATHYROID LESIONS
- LYMPH NODE
- SKIN AND SOFT TISSUE
- BONE

Salivary gland

INFLAMMATORY LESIONS

- Acute supportive sialadenitis
- Sub acute and chronic sialadenitis
- Granulomatous sialadenitis

CYST

- Retention cyst
- Salivary Cyst

AUTOIMMUNE DISORDERS

- Sjogrens syndrome
- Mickulickz diseases
- Benign lymphoepithelial lesions

NEOPLASMS

- Pleomorphic adenoma
- Monomorphic adenoma
- Adenolymphoma
- Oncytoma
- Hemangioma
- Adenoidcystic carcinoma
- MEC
- Clear cell carcinoma
- Adenocarcinoma

LYMPH NODES

INFECTIOUS

- Tuberculosis
- Sarcoidosis
- Granulomatous infections

METASTATIC MALIGNANCIES

- SCC
- Renal cell carcinoma
- Prostate carcinoma
- Malignant melanoma

LYMPHOPROLIFERATIVE

- infectious mononucleosis

IMMUNOLOGICAL DISEASES

- Lymphomas
- Hodgkin's lymphoma

THYROID

NON NEOPLASTIC

- Goiter
- Hyperthyroidism
- Thyroiditis

NEOPLASTIC LESIONS

- Follicular carcinoma
- Papillary carcinoma
- Medullary carcinoma
- Anaplastic carcinoma

SKIN AND SOFT TISSUE

CYSTS

- Epidermal cysts
- Sebaceous cysts

NEOPLASTIC LESIONS

- BCC
- SCC
- Malignant Lymphoma
- Granular cell tumors
- Lipoma
- Fibro sarcoma

CONTRAINDICATIONS FOR FNAC

- ❖ Inexperienced operator.
- ❖ FNAC of liver is contraindicated in patients who are severely jaun-diced and hence have interference with blood clotting. FNAC in such patients can be done once the jaundice subsides.

Certain highly malignant tumors

- ❖ Melanomas
- ❖ germ cell tumors of testis and
- ❖ Ovarian cystadenocarcinoma

As they are better managed surgically without FNAC when they are localized.

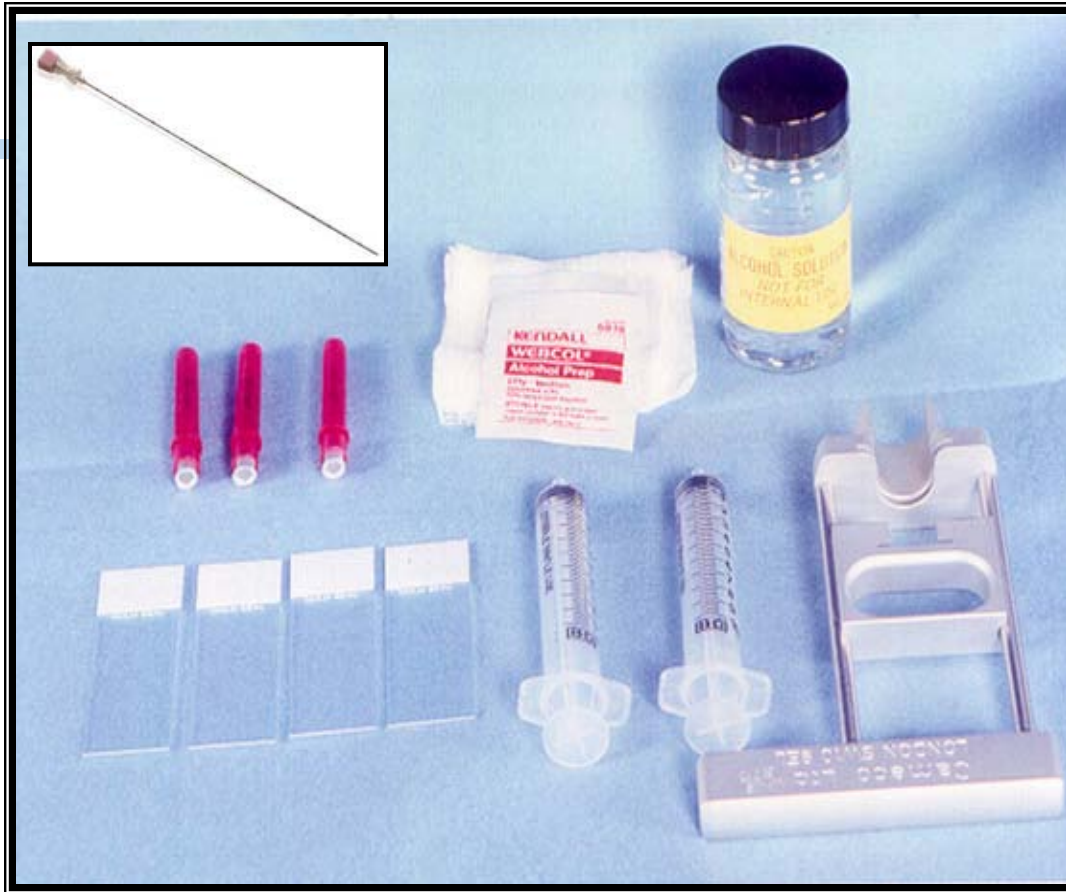
TECHNIQUES OF FNA CYTOLOGY

- I. Patient selection;
- II. Preparation for biopsy
- III. Patient preparation
- IV. Sterilization
- V. Anaesthesia
- VI. Biopsy procedure
- VII. Preparing the aspirate
 - a. Direct smearing
 - b. Indirect smearing
- VIII. Fixation and staining.

PATIENT SELECTION:

I. Patient with localized, clearly defined by either clinical examination or radiological imaging of the disease are usually selected.

However occasionally patients with diffuse disease process, are also chosen mainly to obtain material for microbiological examination.



Preparation for biopsy

- ♦ Alcohol wipe, 4×4-inch gauze pads,
- ♦ 10-ml plastic syringes,
- ♦ 25-gauge 1 1/2-inch stiff, noncutting, bevel-edged needles,
- ♦ Glass slides, alcohol bottles,
- ♦ pistol-grip mechanical syringe holder.

III. PATIENT PREPARATION:

- ❖ Explanation of procedure to the patient before doing it, ensures patients co-operation
- ❖ Important to take patients consent before performing the procedure.
- ❖ It can be done with patient being in supine position on an ordinary examination couch.
- ❖ However for head and neck biopsies chair with head rest is essen-tial.

IV. STERILIZATION:

- Use pre packed swabs for skin disinfections for superficial and few deep tissues.
- For transpleural, retroperitoneal and bone biopsies surgical skin disinfectants, fenestrated sterile cloth and sterile surgical gloves are needed.

V. ANAESTHESIA:

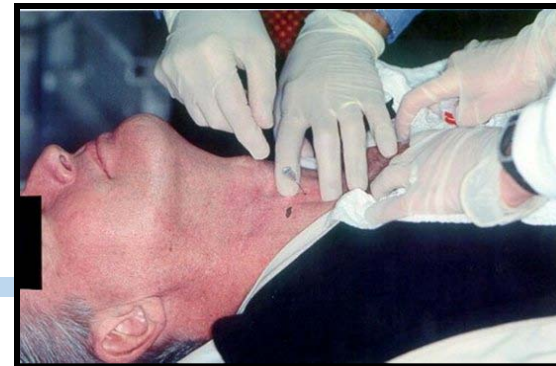
- Prebiopsy sedation is usually not required, except in the deep aspiratoins in very anxious patients.
- In some instances FNA can be co-ordinated with other operative procedures which require general anaesthesia.
- LA is hardly ever necessary when 22 – 25 gauge needles are used.

VI. Biopsy procedure

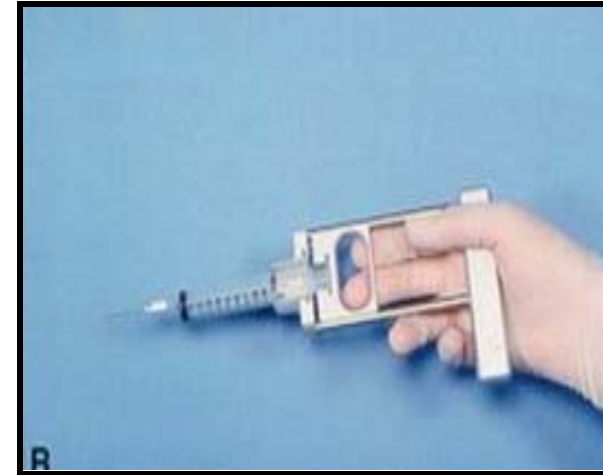
- 1) Place the patient in comfortable position – with mass readily palpable and easily grasped



- 2) Palpation of lesion – attempt to determine the location and surrounding tissue
- 3) Grasp the lesion with one hand usually between two fingers.



- 4) Lay the syringe pistol with attached needle against the surface of lesion
at determined puncture site and angle



- 5) Insert the needle quickly and advance it in to the mass
6) Apply the suction to syringe and , about the one third the length of
syringe barrel

- 7) Observe at the junction of hub and needle for appearance of any specimen
- 8) At first appearance of any sample at junction – release the syringe pistol and let the vacuum equate to normal



- 9) With draw the needle slowly
- 10) Apply the pressure to the puncture site sterile gauze pad

VII. Preparing the aspirate

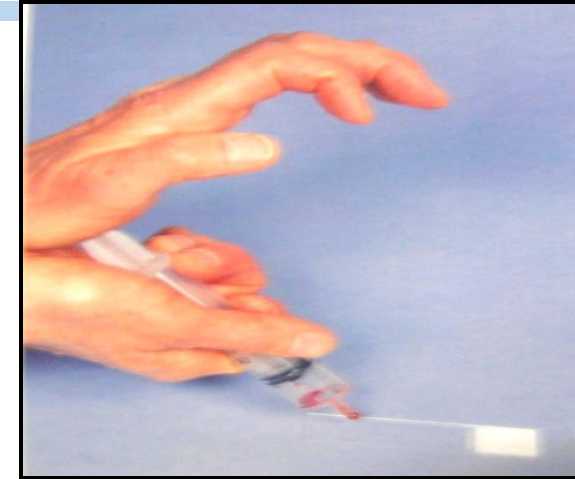
- ❖ The needle is removed quickly from the syringe.



- ❖ Five milliliters of air is aspirated into the syringe, and the needle is placed back on the syringe.



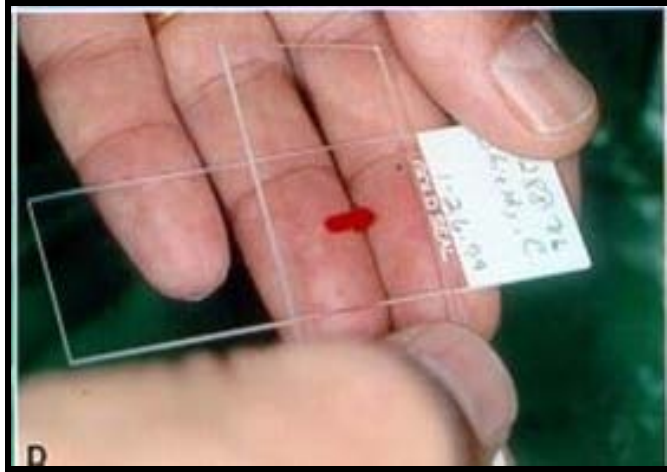
- C.** With the needle bevel facing down, 1 drop of aspirated material is expelled onto each of several glass slides.



- D.** With a second slide, smears are prepared in a manner similar to that for blood smears.

Slides are then immediately wet-fixed by placing them in an alcohol bottle.

Preparation of smear



A drop of aspirate is placed in the centre of plain glass slide.

A second slide is inverted over the drop, and the slides are gently pulled apart vertically or horizontally once

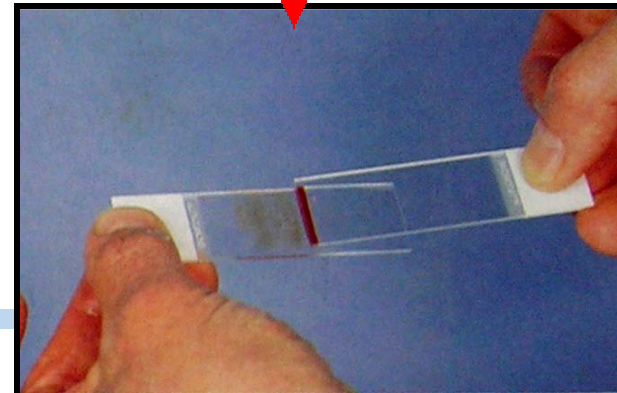
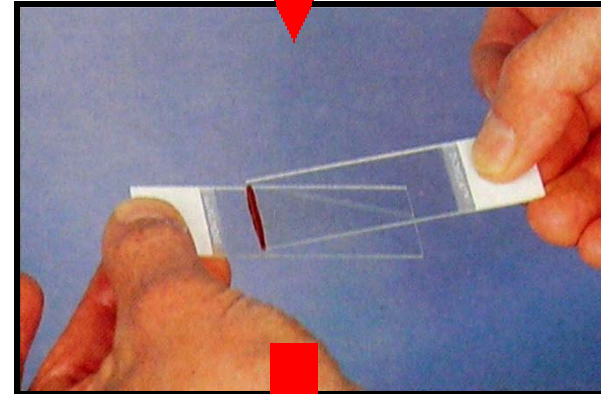
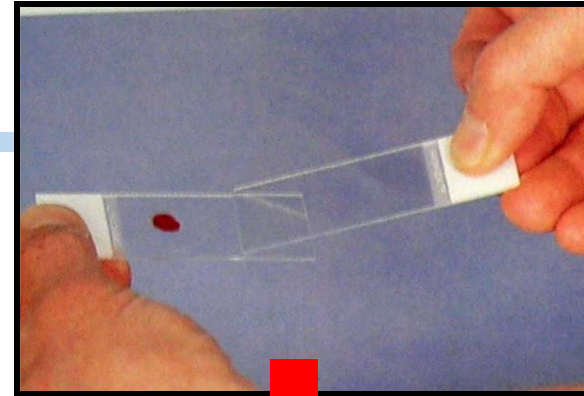
A drop placed near the frosted end of slide held in left hand



Bloody material is spread along the edge of the slide held in right hand



Material is pulled gently down the slide in a manner of making a blood smear



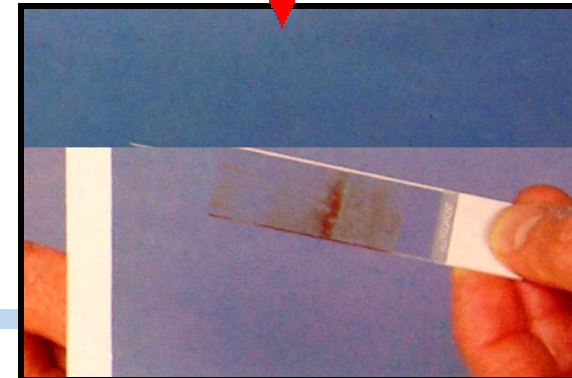
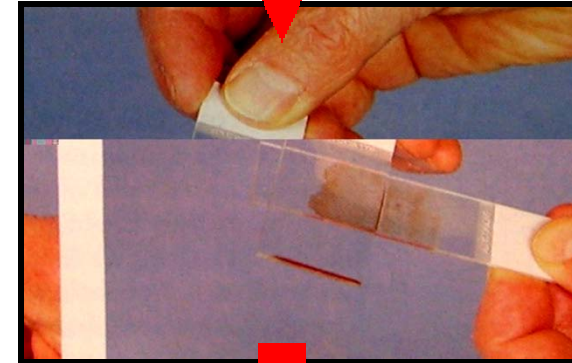
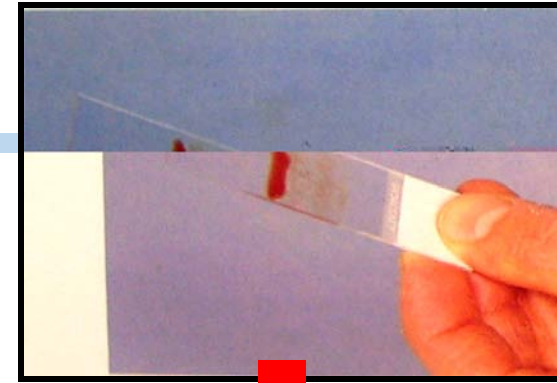
Slide with partially made smear is tilted towards the frosted end so that aspirate runs back towards that end



The long edge of the slide in the right hand is then used to continue making the smear from the leading edge of original smear



Finished smear will have any cell fragments present away from frosted end of slide

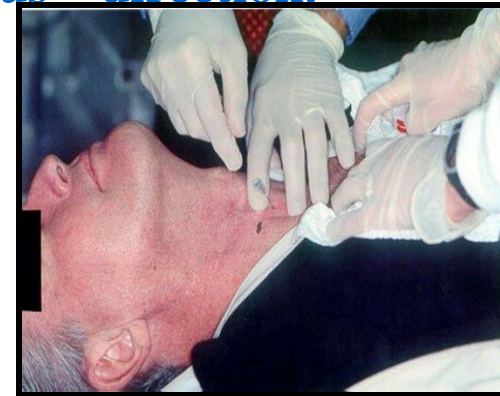


NEEDLE BIOPSY WITHOUT ASPIRATION:

- ❖ The negative pressure plays a relatively minor role .
- ❖ Zajdela recommended needle biopsy without aspiration based on capillary pressure in a fine needle is sufficient to keep the scraped cells inside the lumen.
- ❖ 25 gauge needle is held directly with fingersteps and is inserted into the target lesion and is moved back and forth in various direction.

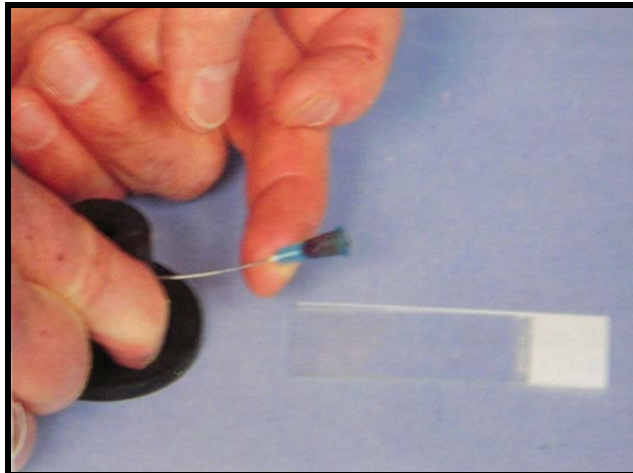
Advantages

- ❖ consistency of the tissues felt much better.
- ❖ Very valuable for tiny lymphnode.
- ❖ Admixture with blood is less than with aspiration.



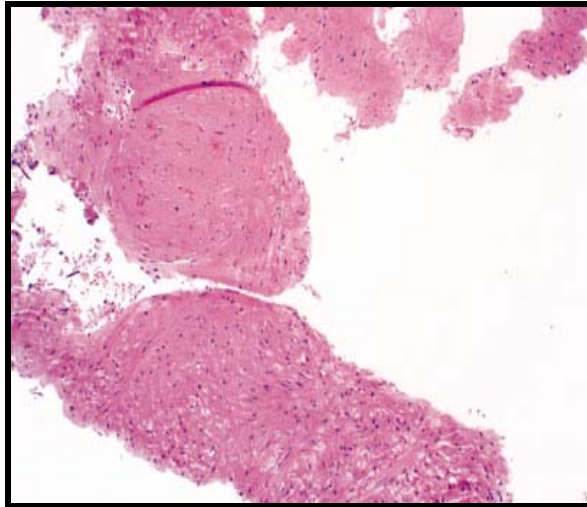
Cell block preparation

- ❖ Valuable in some aspiration biopsy
- ❖ Reinforce some tissue patterns that may be seen on smears – aids in specific diagnosis
- ❖ Provide more consistency for immunoperoxidase staining



- ❖ Do not attempt to smear these small cores --will tear and become distorted

- ❖ **They should not be crushed but they should be gently rolled over slide to make touch preparation**
- ❖ **The material on slide should be alcohol fixed**
- ❖ **After fixation, it can be gently detached and processed as a cell block**



Cell block preparation of an esophageal leiomyoma (H&E, x100).

Fixatives

- ❖ 70 to 90% ethanol for wet fixation of smears is preferable to spray fixatives.
- ❖ Carnoys fixative has the advantage of lysing red blood cells.
- ❖ Glutaraldehyde and 10% buffered isotonic formalin for EM or for paraffin embedding

- ❖ Two fundamentally different methods of fixation and staining are used in FNA cytology.
- ❖ Air drying followed by staining with hematological stain such as May-Grunwald Giemsa (MGG) stain, Jennes – Giemsa, Wright stain or Diff Quick, Romanowsky stain.
- ❖ Alcohol fixation and staining with PAP or H and E.

Individual choice

- ❖ Pathologist with gynaecological cytology background – prefer wet fixation and PAP or H and E stain
- ❖ Pathologist with background in hematology – prefer air dried Giemsa stained smears
- ❖ Surgical pathologist with limited experience in cytology may feel more comfortable with H & E stain
- ❖ Experienced cytopathologist will prefer Papanicolaou stain – transparency for viewing nucleus

- ❖ In air drying – both cytoplasm and nucleus, is flattened on the glass surfaces and appears larger than a cell fixed in ethanol.
- ❖ [Air drying is therefore a helpful ‘artefact’ in cytological diagnosis .
- ❖ It exaggerates nuclear enlargement, one of most important criteria of malignancy].
- ❖ Air dried smears are sterilized by fixation with methanol soon after drying to prevent cross infection,
- ❖ Nuclear chromatin patterns can be studied in Giemsa smears, but the appearances are different to those of the PAP smear.

Stains for FNAC

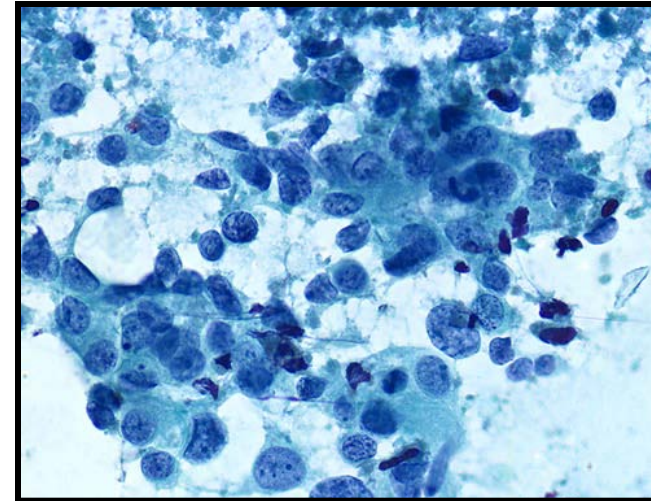
- ❖ PAPINICOLAOU STAIN
- ❖ RAPID PAPINICOLAOU STAIN
- ❖ DIFF QUICK STAIN / MODIFIED WRIGHT STAIN
- ❖ MAY GRUNAWALD GIEMSA
- ❖ MODIFIED WRIGHT'S STAIN
- ❖ HEAMTOXYLIN AND EOSIN STAIN

Conventional PAP stain



after preparation to prevent air drying

- ❖ Spray fixation with 95% ethyl/methyl/isopropyl alcohol
- ❖ The spray fixatives should be held at least 30.5cm (1ft.) away from the slide to prevent freezing artifact result from propellant
- ❖ After fixation – dry it for 1 hr. before staining



Ductal carcinoma.
Fine-needle aspiration.
Papanicolaou stain.

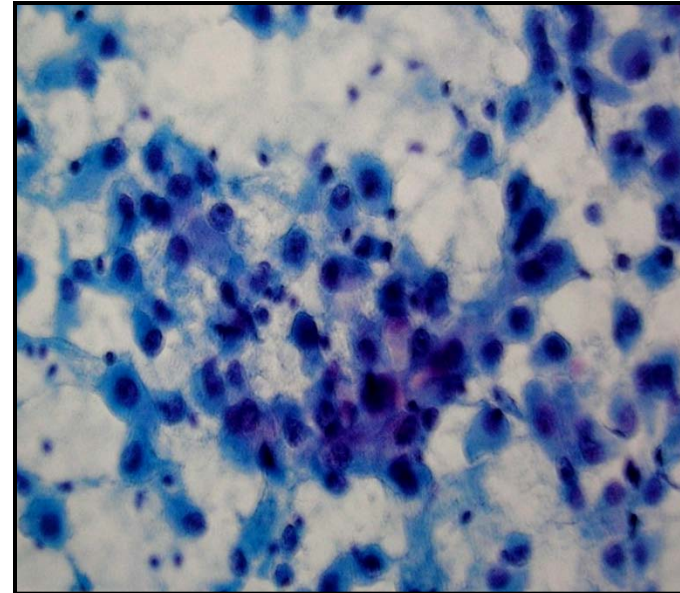
Rapid Papanicolou stain

- ❖ 90 second staining protocol
- ❖ Reduces blood in the back ground of aspiration

Procedure

- ❑ Normal saline --30 seconds
- ❑ Alcohol formalin --10 seconds
- ❑ Water-- 6 slow dips
- ❑ Hematoxylin-- 2 slow dips
- ❑ Water --6 slow dips
- ❑ Richard Allen cyto stain --4slow dips
- ❑ 95 %ethanol --6 slow dips
- ❑ 100% ethanol --6 slow dips
- ❑ xylene
- ❑ Mount in mounting media

Mucoepidermoid carcinoma



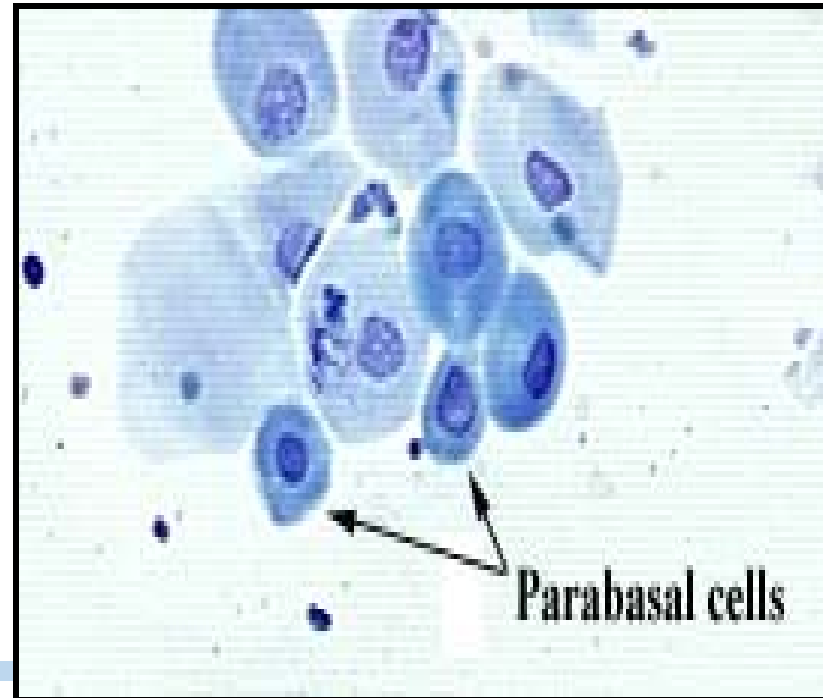
FNAC

Author and year	Study design & level of evidence	Aim and Method	Results and conclusion
Khandekar M.M etal 2006	EXPERIMENTAL CASE CONTROL Level III	<p>Analysis of salivary gland lesions by FNAC and correlation with histopathology. To evaluate utility of FNAC in salivary gland lesions.</p> <p>Seventy patients were studied prospectively over two years. FNAC was done using 10 cc syringes and 20-22 no. needle. liistomorphology was assessed on 'routine H & E (haemotsylin and eosin) stained paraffin sections. SPAR' (periodic acid Schiff) and mucicarmine satins were also done</p>	<p>Results : 80 ' of the lesions were neoplastic (61 %a benign, 31 % malignant) and 20% were neoplastic. Pleomorphic adenoma was they most frequent benign 'neoplasm while inucoepidermoid carcinoma was the most frequent malignant lesion. Among the non neoplastic lesions, the maximum number of eases were of chronic sialadentis. In the present study, ;FNAC has a sensitivity of 94,545/ specificity of 80.95cc for neoplastic lesions. Conclusions: FN C was found to be a useful diagnostic tool in the evaluation of salivary gland lesions because of its simplicity, excellent patient compliance and rapid diagnosis. This cost effective tool is invaluable in planning the surgical management of the patient.</p>

Classification of Epithelial Cells

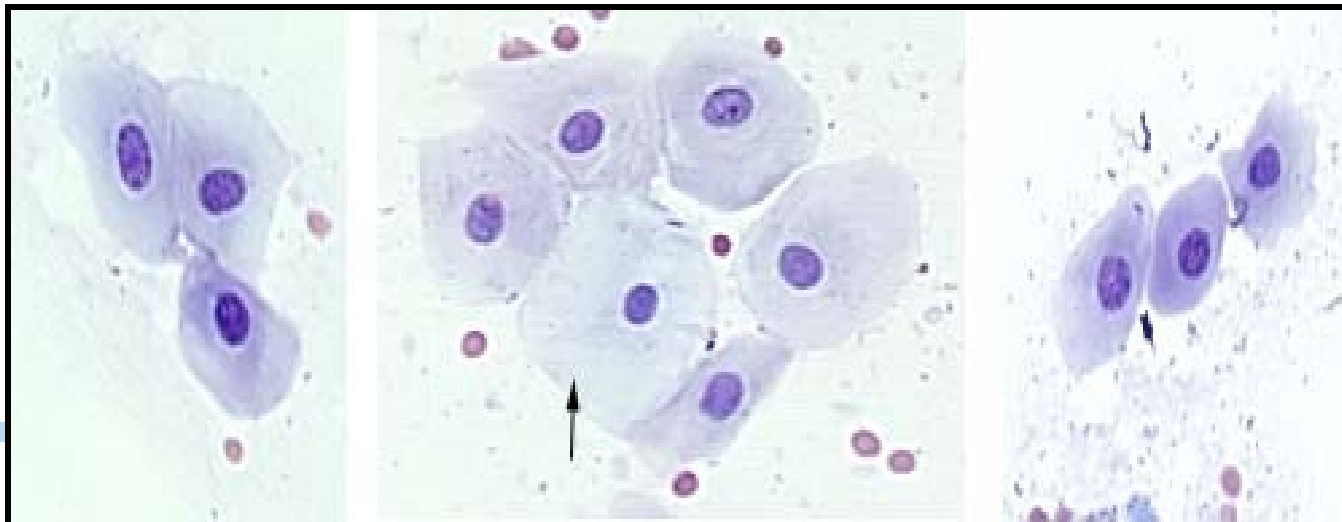
PARABASAL CELLS

- ❖ Parabasal cells are the smallest epithelial cells
- ❖ They are round or nearly round and have a high nuclear to cytoplasmic ratio.



Intermediate cells

- ❖ Vary in size and shape,
 - ❖ Typically have a diameter two to three times that of parabasal cells.
- subclassify these cells into:
- ❖ *small intermediates*: nearly round or oval shape with large, prominent nuclei
 - ❖ *large intermediates*: polygonal shape with a small nuclear/cytoplasmic ratio

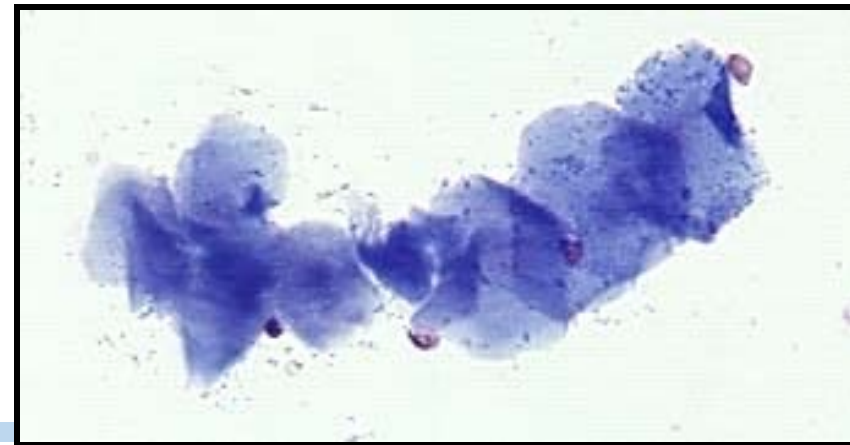
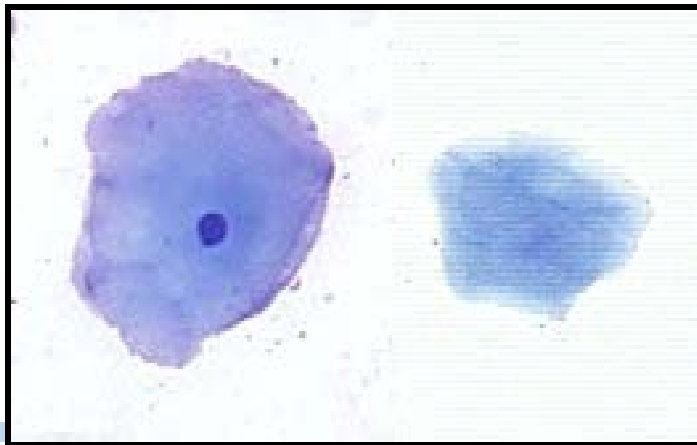


Superficial Cells



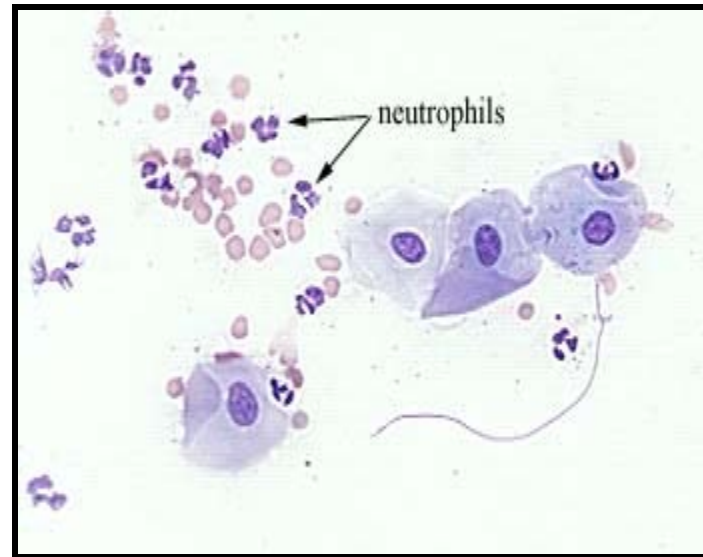
- ❖ They are polygonal in shape and distinctly flat,
- ❖ sometimes having the appearance of being rolled up.
- ❖ Their nuclei are either absent or pyknotic (very small and dark).
Superficial cells without nuclei are often referred to as being "fully cornified".

Superficial cells are often seen in large sheets or strings, as seen below with fully cornified cells.



Other Cells

- ❖ Erythrocytes
- ❖ Neutrophils
- ❖ "Foam cells" is a term given to non-descript epithelial cells containing numerous vacuoles



Difference Quick

❖ Difference Quick is a rapid haematology type stain

❖ Three steps to Diff –Quick stain

1) fixation in methyl alcohol

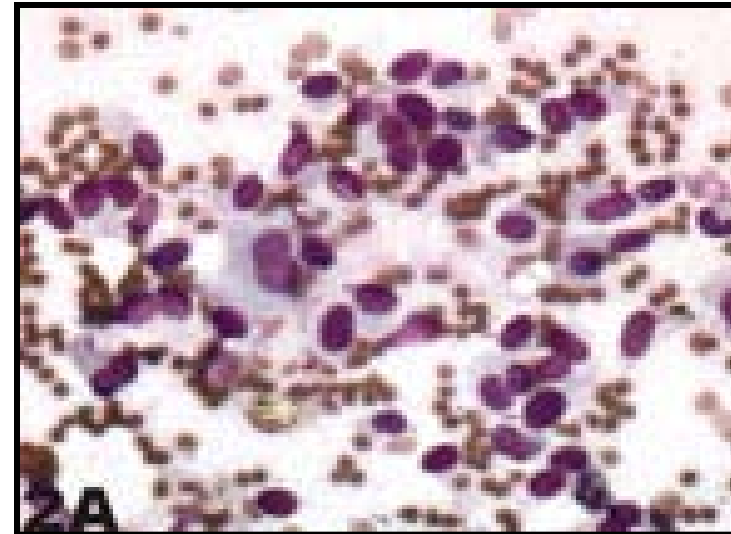
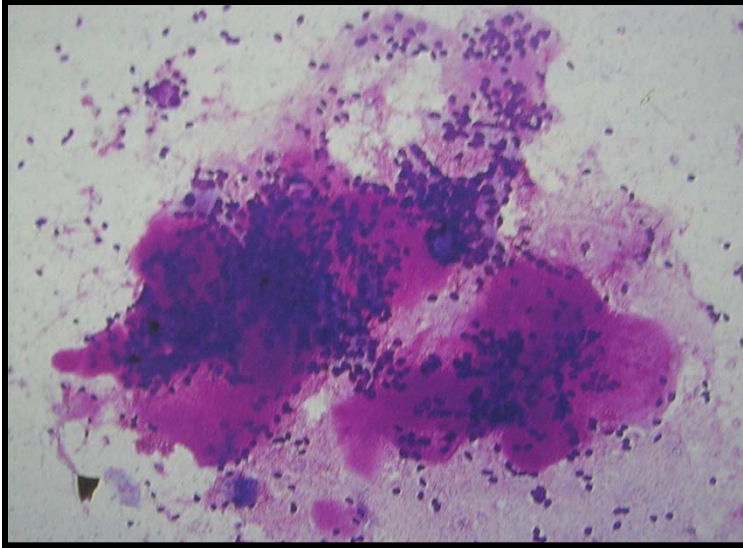
2) Staining in eosin Y

3) Staining in Azure A

The procedure will take less than 20 seconds



**sheets of epithelial cells embedded in dense metachromatic stroma
-- pleomorphic adenoma**

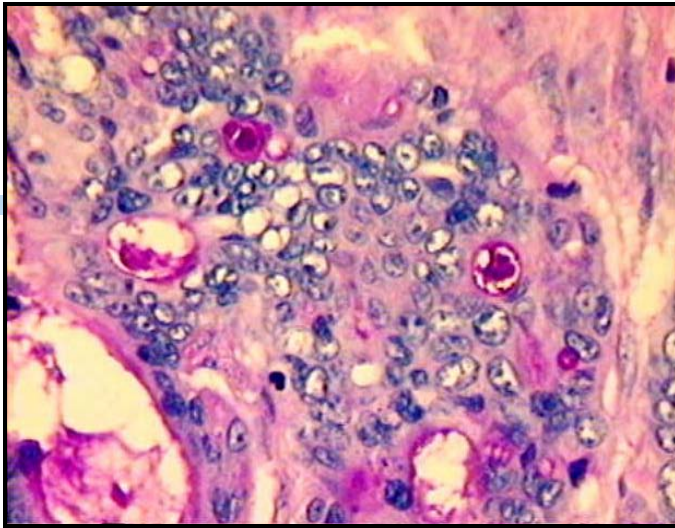


Diff Quik stain reveals a fairly cellular aspirate with round to ovoid, monotonous loosely aggregated cell clusters (DQ stain; 400).

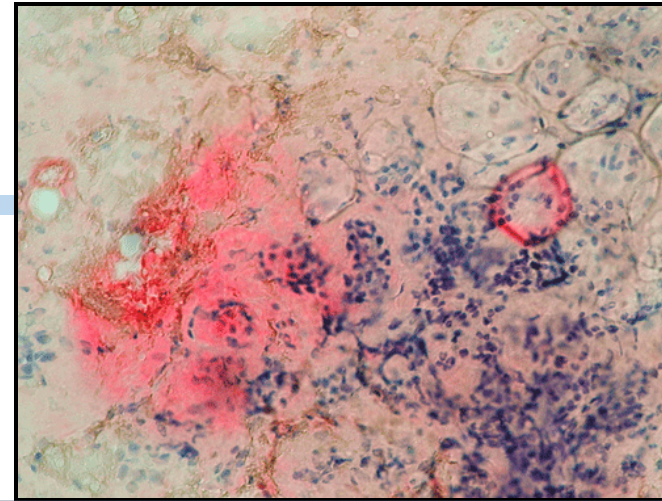
SPECIAL STAINS

For both air dried or wet fixed smears it can be done:

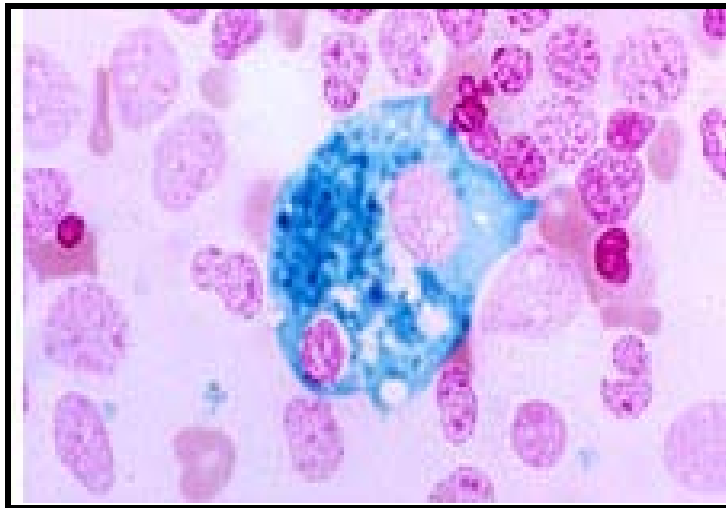
- ❖ 1. PAS / diastase or Alcian blue - Mucins.
- ❖ 2. Prussian blue - Iron
- ❖ 3. Masson – Fontana - Melanin
- ❖ 4. Grimellus - Argyrophilic granules.
- ❖ 5. Congo red - Amyloid
- ❖ 6. Gram, PAS or Gomoris silver stain - Micro-organism
- ❖ 7. Ziehl-Neelsen - AFB
- ❖ 8. Oil red O or sudan black - Fat



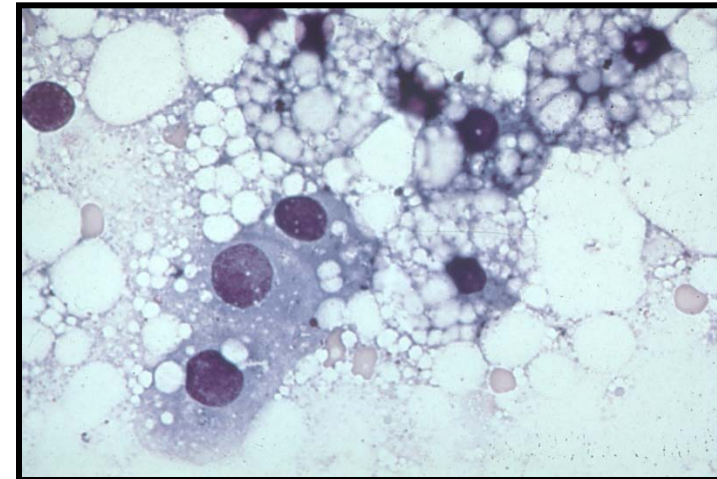
PAS - Mucins.



These amorphous substance stains orange on Congo red stain

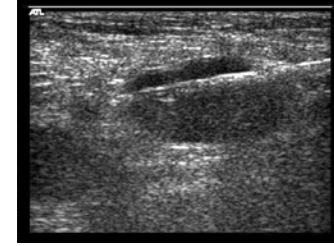
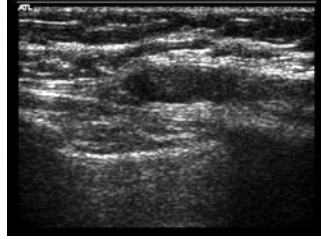


Prussian blue stain. Macrophage packed with iron (blue colored reaction)



Lipid can be confirmed with an Oil-Red-O stain

ORGAN IMAGING FOR THE GUIDANCE OF THE BIOPSY NEEDLES:



- ❖ There has been continuous improvements in needle biopsy guides and mechanical biopsy devices together with technological advances
- ❖ It is done in order to allow the precise localization of the aspiration biopsy needles, within the lesions.
- ❖ Image-guided FNAC has a pivotal role for establishing tissue diagnosis.
- ❖ fine needle aspiration corresponded with the final diagnosis in **69%** of patients.

Bahrain Med Bull 2004;26(1):

❖ The major imaging methods

- Plain radiography
- Fluoroscopy
- Fluoroscopy and contrast radiography;
- Radionuclide scanning
- Ultrasound
- Computerised tomography.
- MRI



❖ All the above methods can be used singly or in combination

- ❖ The most recent method is STERIOSTATIC GUIDANCE especially for brain and breast lesion
- ❖ Can be used for FNAC of fetal blood

ADVANTAGES AND DISADVANTAGES OF IMAGING TECHNIQUES

METHOD	ADVANTAGES	DISADVANTAGES
FLUOROSCOPY	Needle easily visible	<ul style="list-style-type: none">▪ Radiation exposure▪ Difficulty in visualize lesion▪ Difficulty to demonstrate depth
Ultra sound	Real time imaging	<ul style="list-style-type: none">▪ Poor needle visibility▪ Limited by gas or bone
Computed tomography	Easy visibility and precise localization	Expensive Radiation exposure
MRI	Suitable for deep lesions	Expensive requires Special needles

REFERENCES



- ❑ **Fine-Needle Aspiration Cytology in Tumors and Tumor-Like Conditions of the Oral and Maxillofacial Region. Cancer (Cancer Cyto pathol) 1997;81:238– 52.**
- ❑ **Manual and atlas of fine needle aspiration cytology. S. Orel ,G Sterrett.**
- ❑ **GOOGL IMAGE SEARCH**



FINE NEEDLE ASPIRATION CYTOLOGY- PART II

CONTENTS

- ❖ Needles used for FNAC
- ❖ Papanicolaou stain
- ❖ FNAC of salivary glands
- ❖ FNAC of lymph nodes
- ❖ FNAC of common head and neck lesion
- ❖ Complications
- ❖ Conclusion



Needles used for FNAC

- ❖ Standard disposable 27-22 gauge (0.4-0.7) 30-50mm needles are suitable for superficial palpable lesions
 - ❖ 23-22 gauge needle for hypocellular, fibrotic, and desmoplastic lesions in the soft tissues
 - ❖ 27 gauge needle for children and sensitive areas such as orbit and eyelids
 - ❖ 22 gauge 90mm lumbar puncture needles with trocar are convenient for most deep biopsies
- (the needle is sufficiently rigid and trocar prevents contamination during the passage through surrounding tissues)

- ❖ If a needle of greater length is required a 22 gauge 150-200mm Chiba needle is used
- ❖ If the purpose of the biopsy is to obtain a core of tissue for paraffin embedding and sectioning a cutting core needle of 22-18 gauge is used

The Papanicolaou stain

- ❖ The PAP stain utilizes Harris hematoxylin as the optimum nuclear stain and the combination of orangeG6 and EA50 gives the subtle range of green, blue and pink hues to the cell cytoplasm according to the amount of keratin contained in the cell

Composition of PAP stain

❖ Harris hematoxylin

- ❖ Hematoxylin, Absolute alcohol, Ammonium alum, Distilled water, Mercuric oxide, Glacial acetic acid

❖ Orange G

- ❖ Orange G, Alcohol, Phosphotungstic acid

❖ EA 50

- ❖ Light green, Eosin Y, Phosphotungstic acid, Alcohol, Methanol, Glacial acetic acid

Staining procedure

- ❖ Fix smears in 95% alcohol - 15 minutes
- ❖ Rinse smears in distilled water
- ❖ Stain in Harri's haematoxylin for 4 minutes
- ❖ Wash in tap water for 1-2 minutes
- ❖ Differentiate in acid alcohol (0.25% HCL in 70% alcohol)
- ❖ Blue in tap water or 1.5% sodium bicarbonate
- ❖ Rinse in distilled water

- ❖ **Transfer to 70% alcohol, then 95% alcohol for a few seconds**
- ❖ **Stain in Orange G6 for 1-2 minutes**
- ❖ **Rinse in 3 changes of 95% alcohol for a few seconds**
- ❖ **Stain in EA 50 for 3-5 minutes**
- ❖ **Rinse in 3 changes of 95% alcohol for a few seconds**
- ❖ **Clear and mount**

Results

- ❖ Nuclei – blue
- ❖ Acidophilic cells – red to orange
- ❖ Basophilic cells – green to blue green
- ❖ Cells or fragments of tissue penetrated by blood – orange to orange green

Cytologic classification

- ❖ **Class I (Normal cytology)**
 - ❖ Indicates that only normal cells are observed
- ❖ **Class II (Atypical cytology)**
 - ❖ Indicates the presence of minor atypia due to inflammation
 - ❖ No evidence of malignancy
- ❖ **Class III (Intermediate cytology)**
 - ❖ The cells display wider atypia, which may be suggestive of severe dysplasia, carcinoma in-situ or cancer. A repeat is advised after treating or removing any possible pre-disposing factor. If the cytologic features remain unchanged, biopsy is recommended.

❖ Class IV (Suggestive of cancer)

- ❖ Cytology shows few epithelial cells with malignant changes.
- ❖ Few epithelial cells may show borderline characteristics.
- ❖ Biopsy is mandatory

❖ Class V (Positive for cancer)

- ❖ Cells show characteristic malignant features
- ❖ Biopsy is mandatory

❖ Class I Cytology (Normal cells)

- ❖ Anucleated orthokeratinized squamous cells are polygonal in shape and the cytoplasm stains pale orange to yellow
- ❖ Some cells may show pyknotic nuclei
- ❖ Parakeratotic cells are also polygonal in shape, with a tendency to curl and fold
- ❖ The cytoplasm is eosinophilic
- ❖ The nucleus is round with smooth outline

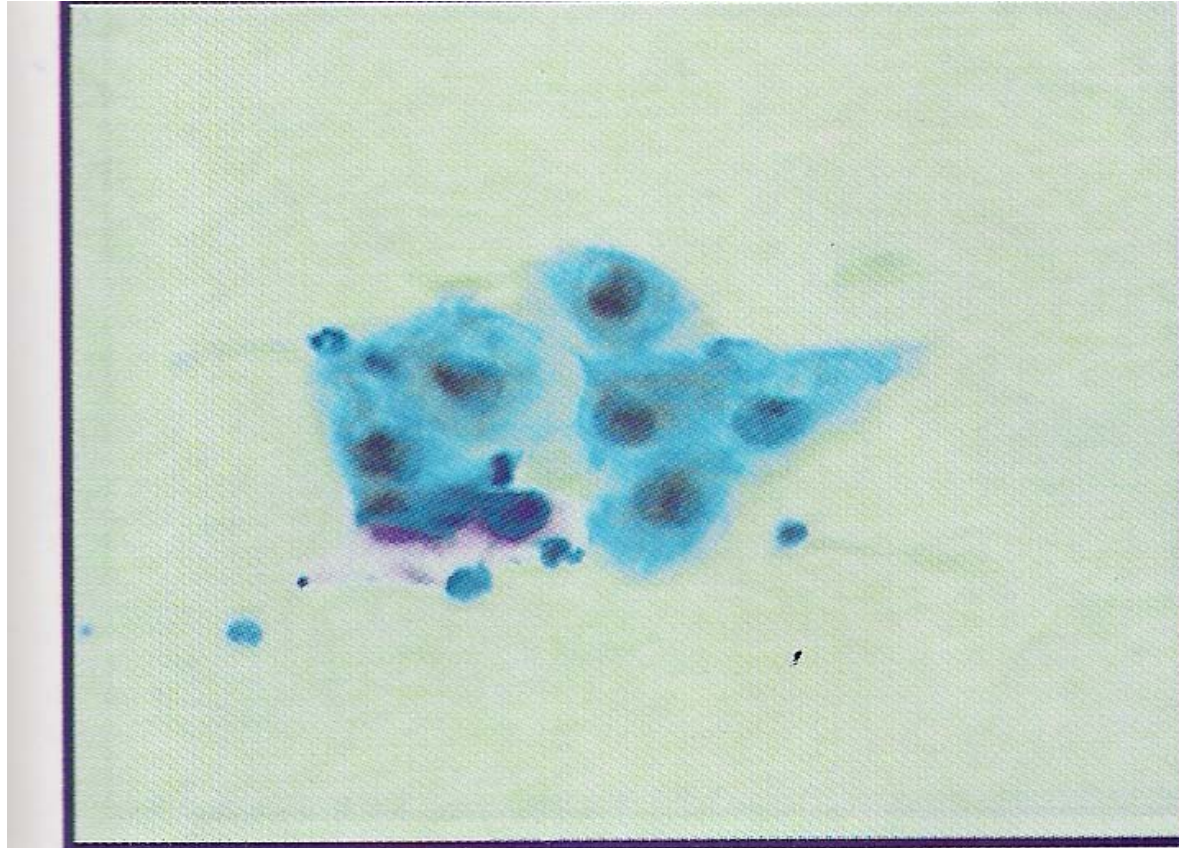


Fig 1. Normal Basal Cells X 400. Small Cells with blue stained cytoplasm, slightly large nuclei, maintaining uniformity in size and shape (class I)

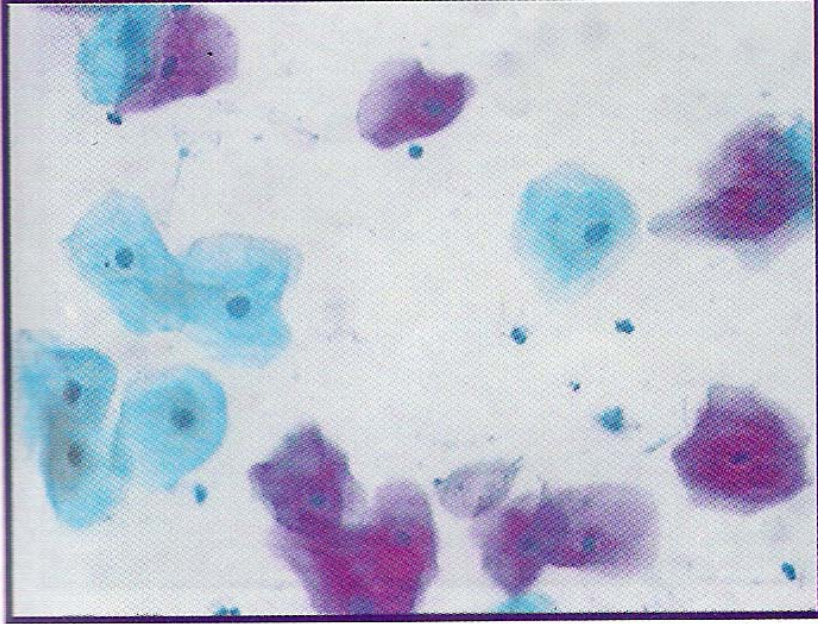


Fig 2. Normal precornified cells X 400. Flat polyhedral cells with blue stained cytoplasm. Pink stained cytoplasm indicates progressive cornification. (class I)



Fig 3. Progressive stages of Cornification (maturation) epithelial cells from blue precornified cell, pink cornifying and yellow stained cornified cell X 400. (class I)

- ❖ **Class II cytology (Atypical cells)**
- ❖ **The cells and the nucleus show proportionate enlargement**
- ❖ **The cytoplasm may show bacterial colonization**
- ❖ **Presence of many free or naked nuclei**
- ❖ **Cells affected by virus will show ballooning degeneration and inclusion bodies**
- ❖ **In fungal infections presence of yeast cells or hyphae**

- ❖ In pemphigus, the polygonal shape of the squamous cell will be lost. The cells appear more rounded and small due to shrinkage and because a greater percentage are obtained from basal/parabasal layer of epithelium

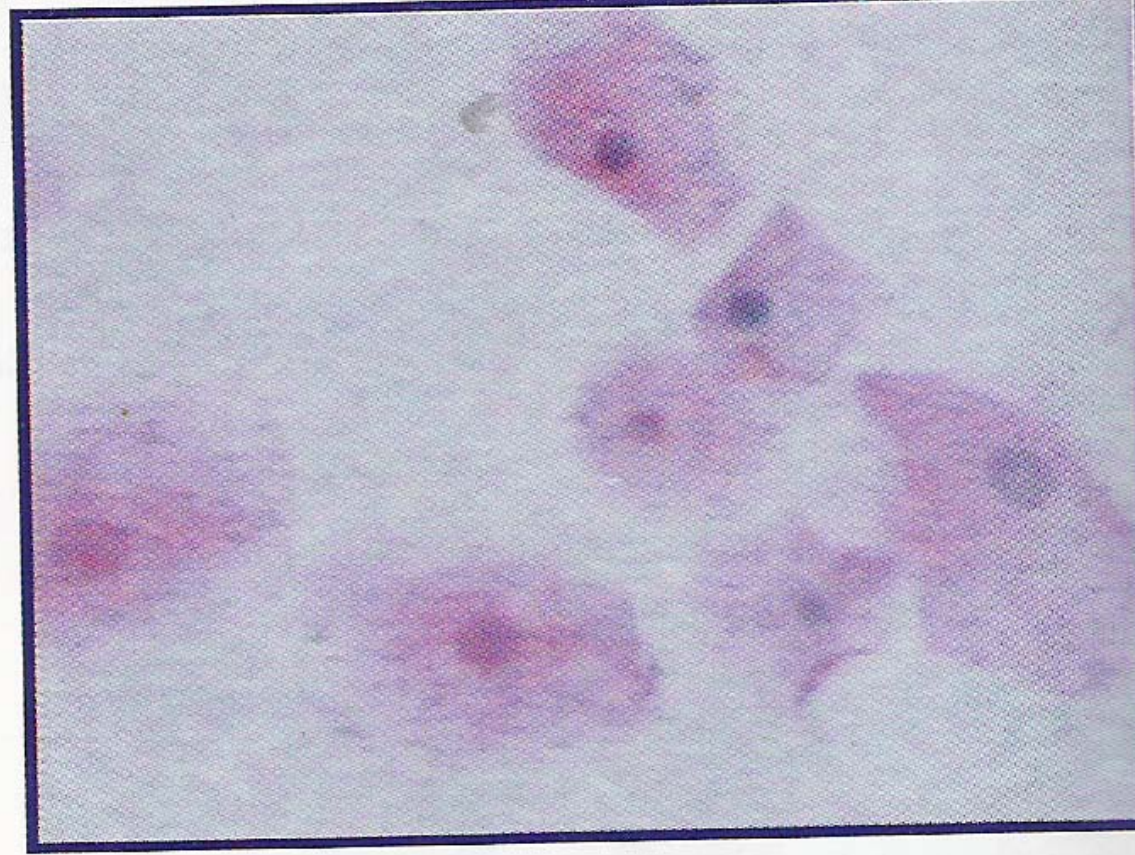


Fig 4. Cornifying epithelial cells showing Nuclear enlargement and Variation in the size & shape of the nuclei X 400 (classical)

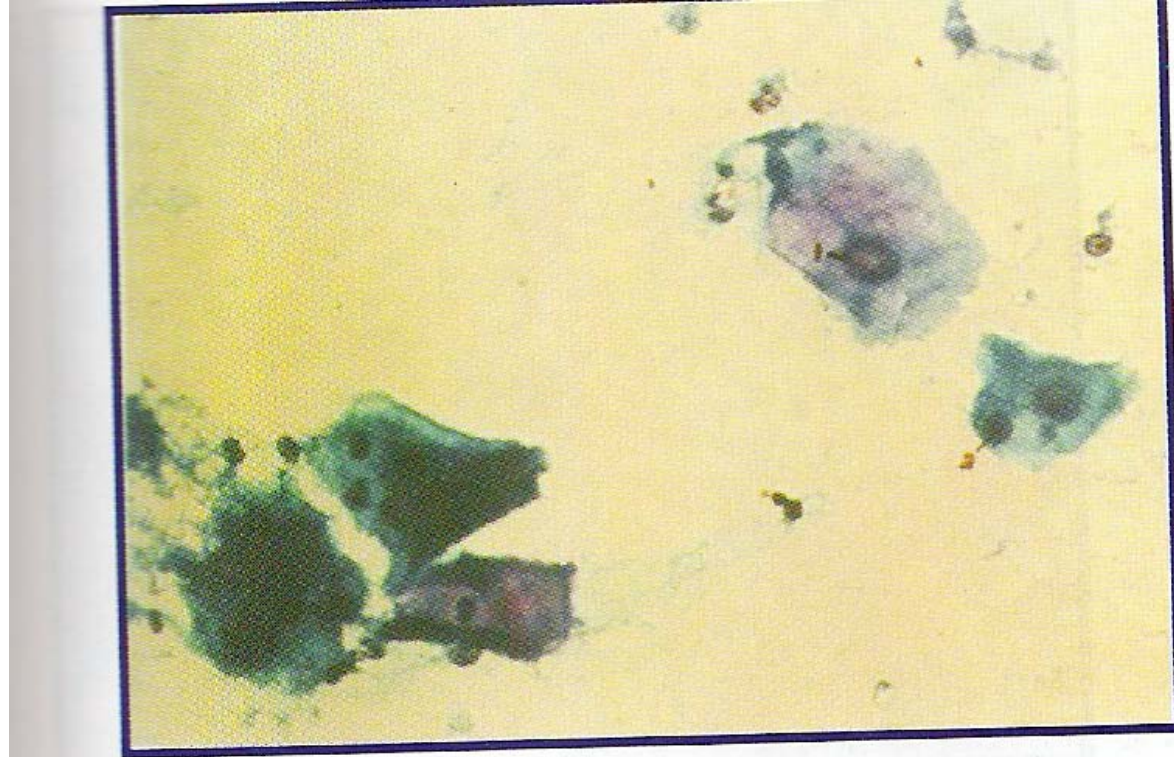


Fig 5. Cornifying cells showing Perinuclear condensation of chromatin and Binucleated cell with folded cytoplasm X 400 (Class III)

❖ Cytopathology of oral carcinoma

❖ Nuclear abnormalities

❖ Size of the nucleus

❖ Anisonucleosis

❖ Irregular shapes

❖ Outline may be crenated with sharply indented edges. Extreme elongation, sharp contours, angular shapes, budding and spheroidal projections from the nuclei are common

❖ Mutinucleation

❖ Nuclei are hyperchromatic, tend to be closer together and difficult to distinguish from one another

- ❖ **Abnormal mitosis**
- ❖ **Nuclear hyperchromatism**
- ❖ **Aberrant chromatin pattern**
 - ❖ Irregular distribution of chromatin, coarse aggregates frequently at the periphery of the nucleus are common
- ❖ **Prominent or multiple nucleoli**
- ❖ **Nuclear predominance**
- ❖ **Altered nuclear cytoplasmic ratio**
- ❖ **Degenerative changes of the nuclei**

❖ Cytoplasmic abnormalities

- ❖ Scanty cytoplasm
- ❖ Vacuolization and inclusions
- ❖ Altered staining characteristics

❖ Cell as a whole

- ❖ Enlargement
 - ❖ Anisocytosis, Anisonucleosis
- ❖ Bizarre shapes
 - ❖ Elongated squamous cells have a fibroblast like morphology
Tadpole shaped cells
 - ❖ Cannibalism- engulfment of one cell by another, due to abnormal behavior of cancer cells or modification of multinucleation

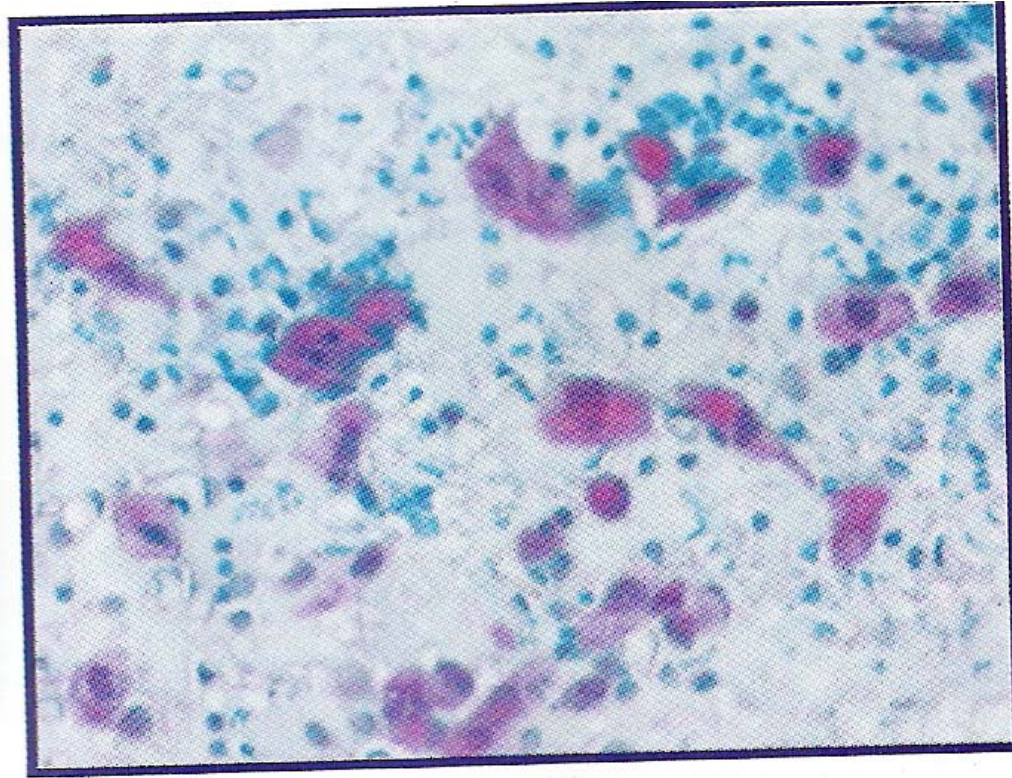


Fig 6. Variation in the size & shape of nuclei. Cells with giant nucleus, irregular nuclear and cytoplasmic outline X 200 (class IV)

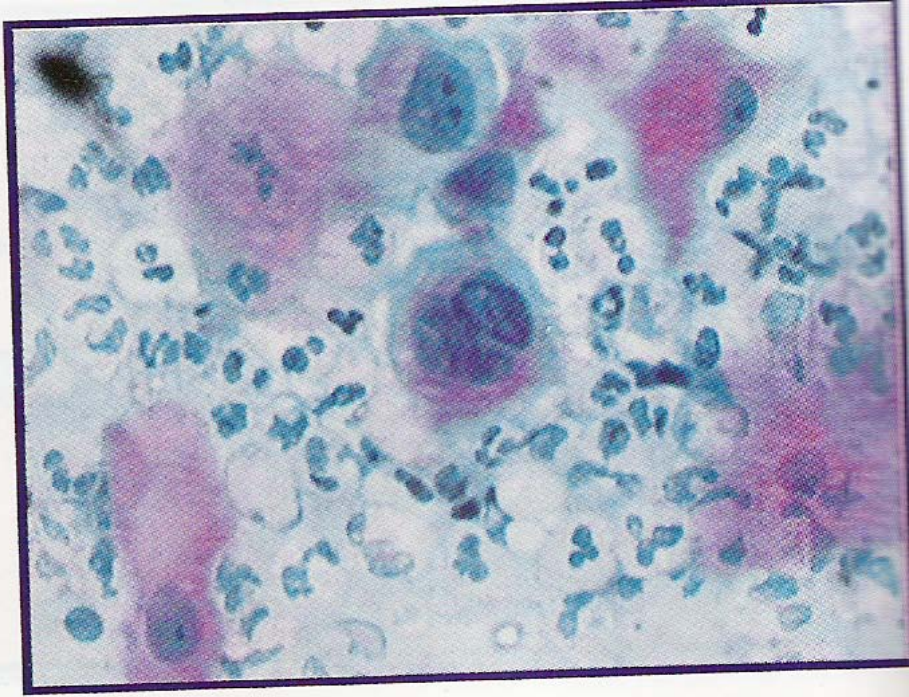


Fig 7. Cells showing Altered Nuclear Cytoplasm ratio
Prominent nucleoli and Binucleation X 400 (class V)

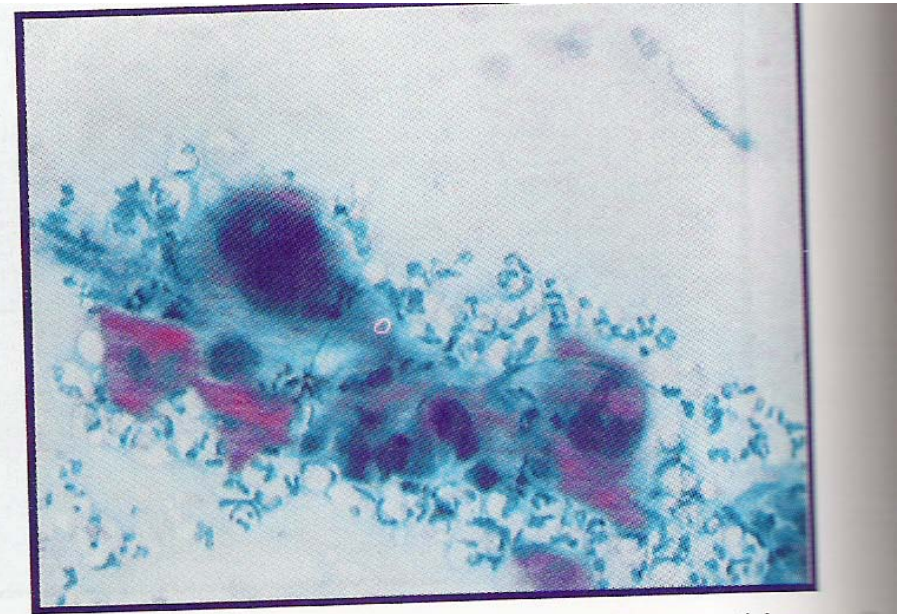


Fig 8. Malignant cells showing nuclear pleomorphism and
giant nuclei X 200 (class V)

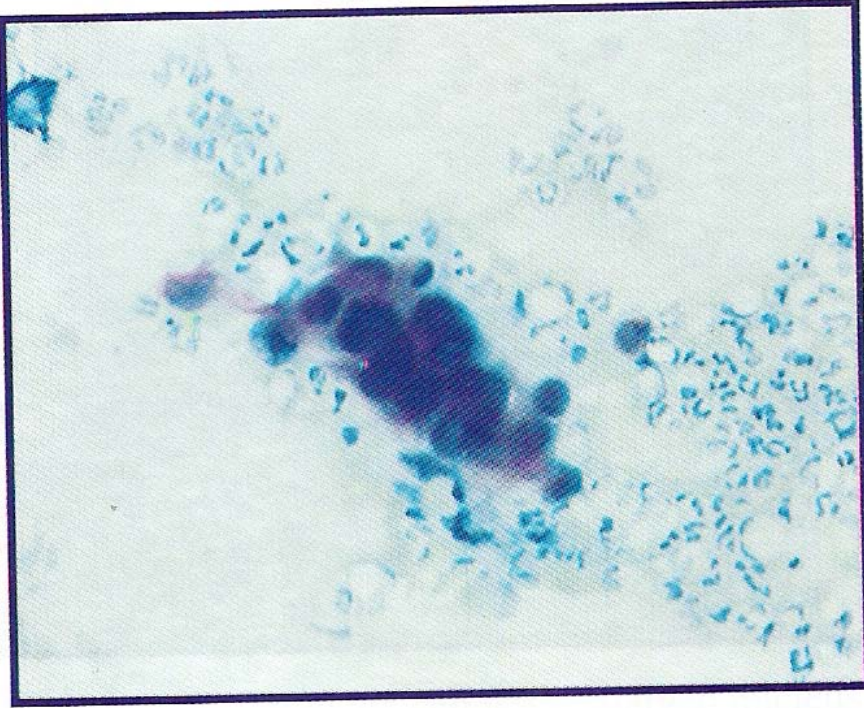


Fig 9. Cluster of nuclei with little or no cytoplasm (free nuclei) X 400 (Class V)

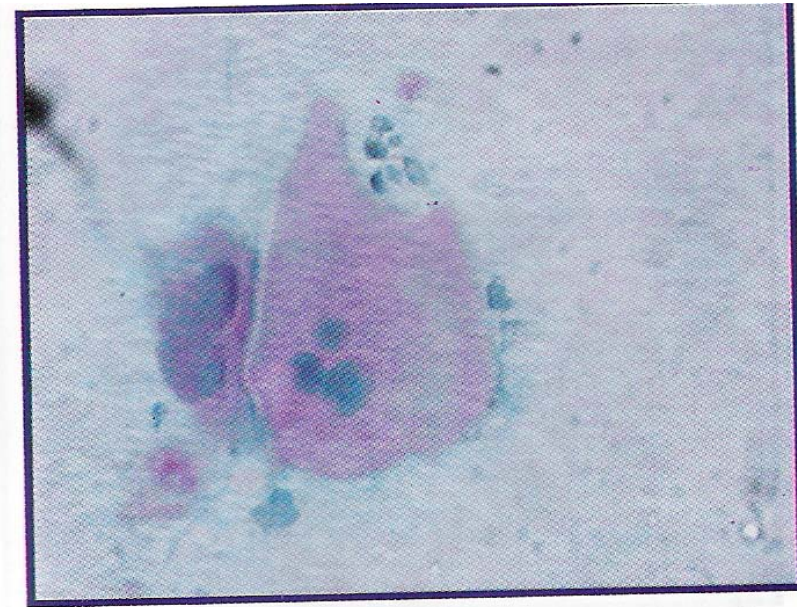


Fig 10. A cornifying cell showing multi-nucleation with prominent nucleoli X 500 (class V)

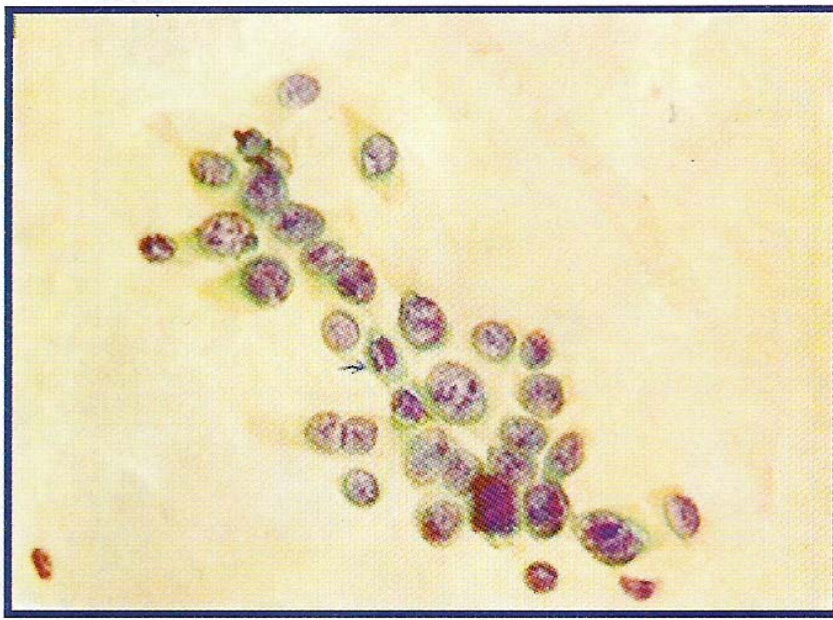


Fig 11. Altered N/C ratio, irregular size and shape of nuclei with prominent nucleoli. Arrow showing mitotic figure X 200 (class V)

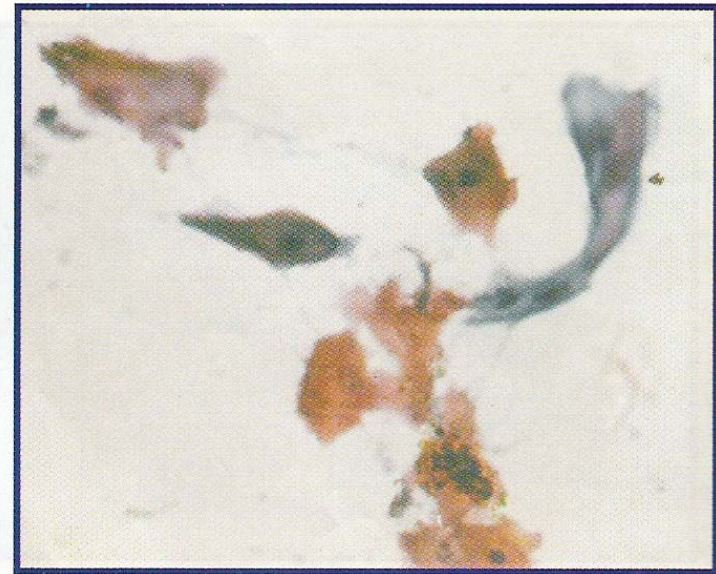


Fig 12. Bizarre shapes of cells with excessive cornification. Elongated cornified cell containing hyperchromatic, irregular shaped nucleus (Arrow) X 200 (class V)



Fig 14. Cannibalism (cell in cell) Orange stained cell with crescentic nuclear is enclosed within a multinucleated cell X 400 (class V)

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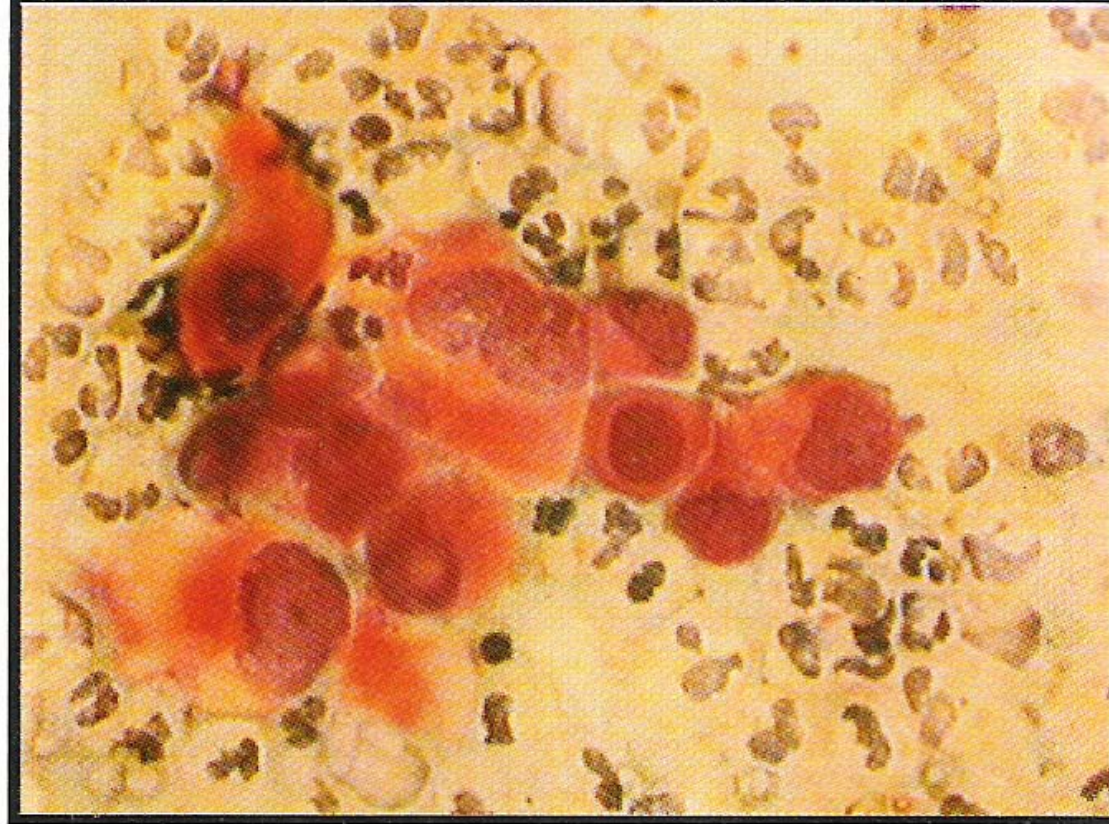
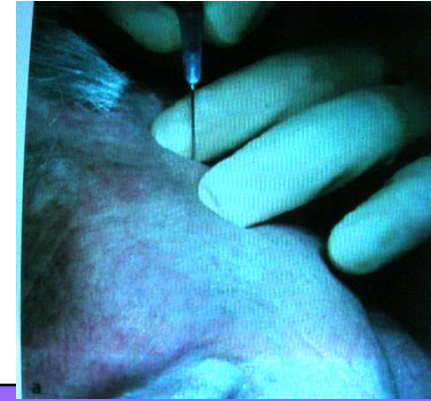
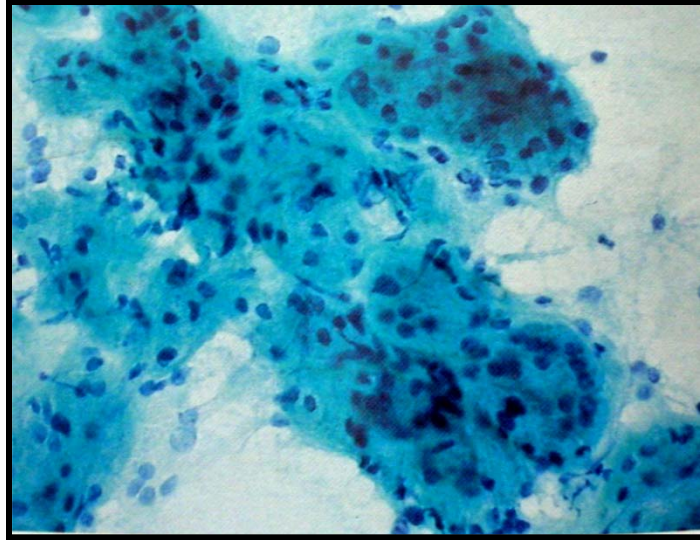


Fig 15. Excessive and abnormal cornification. Precornified cells showing deep orange cytoplasm X 400 (class V)

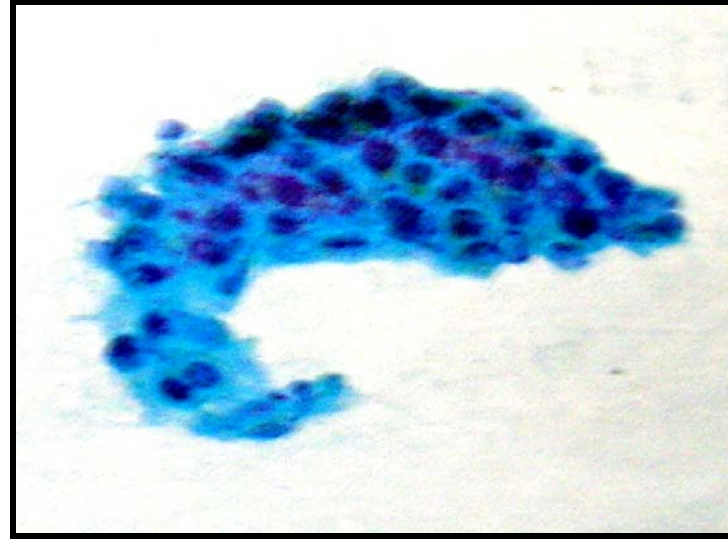


FNAC OF SALIVARY GLAND LESIONS

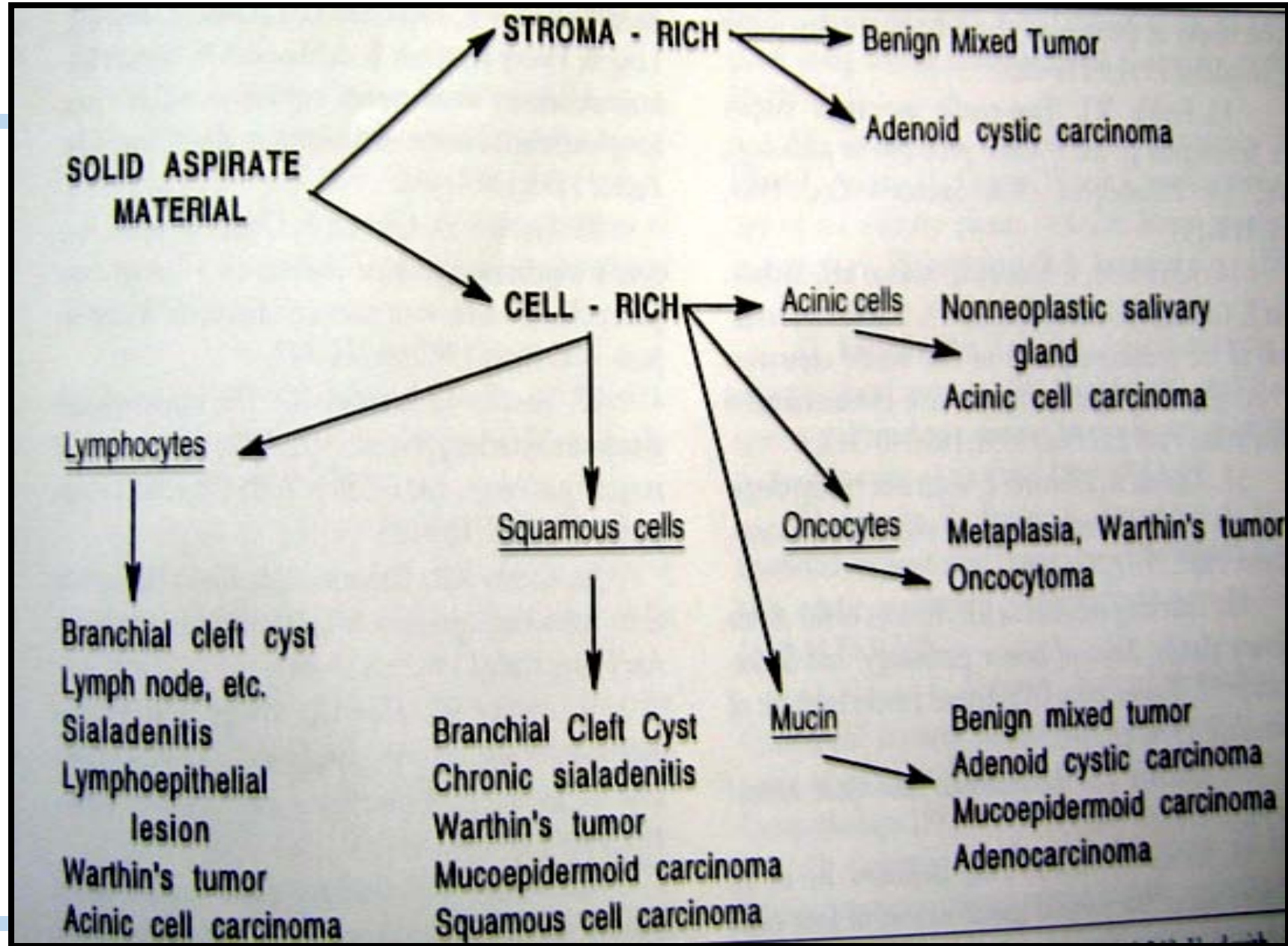
NORMAL SALIVARY GLAND



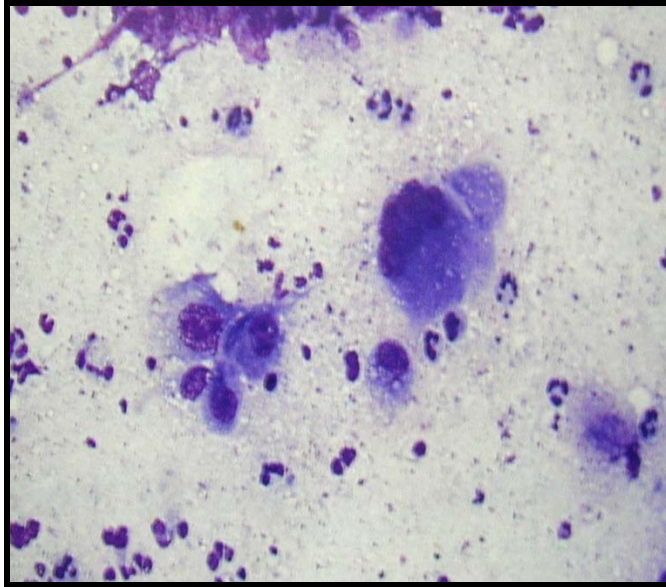
Scattered clear cells represents the mucous cells surrounded by larger numbers of serous cells



Small sheets intercalated ducts.

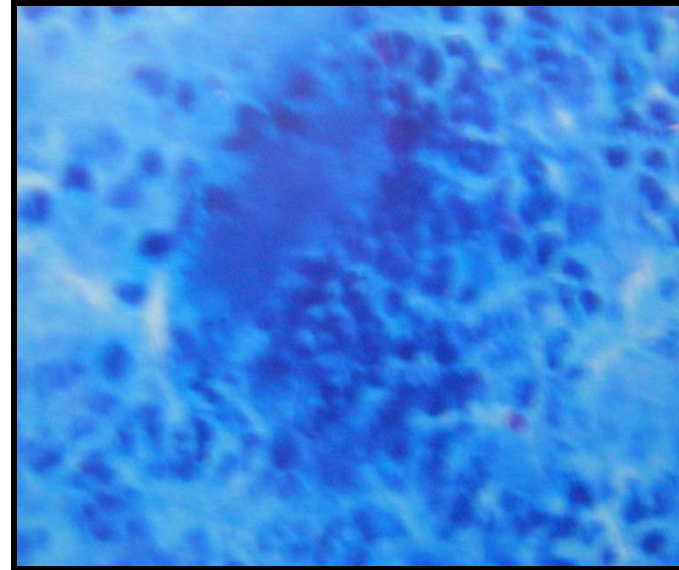


Acutely inflamed parotid gland



- Scattered atypical duct cells in a background of neutrophils
- degenerating acinar cells

Actinomycotic infection



- Organism of Actinomycotic in the centre of field surrounded by acute inflammatory cells

RETENTION CYST

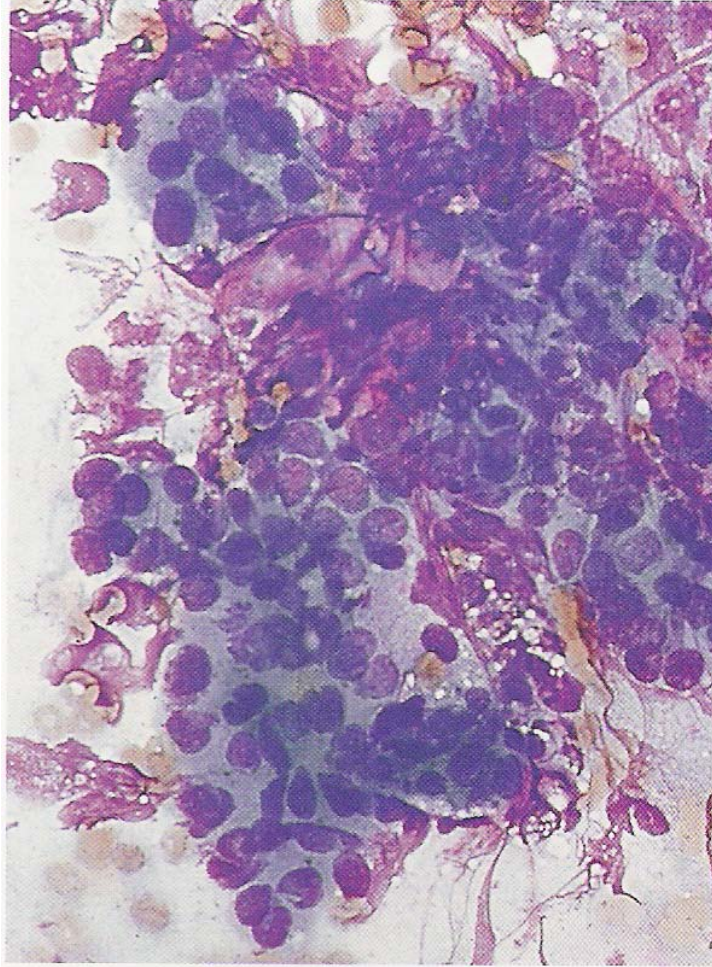


Fig. 4.17 Retention cyst

Debris and ductal epithelium showing degenerative and regenerative atypia (MGG, HP).

Criteria for diagnosis for Pleomorphic adenoma

- ❖ Fibrillary chondromyxoid ground substance
- ❖ Epithelial cells single & poorly cohesive clusters and sheets
- ❖ Regular ovoid nuclei with bland nuclear chromatin, well defined cytoplasm
- ❖ Spindle shaped 'mesenchymal' cells mainly in stromal matrix

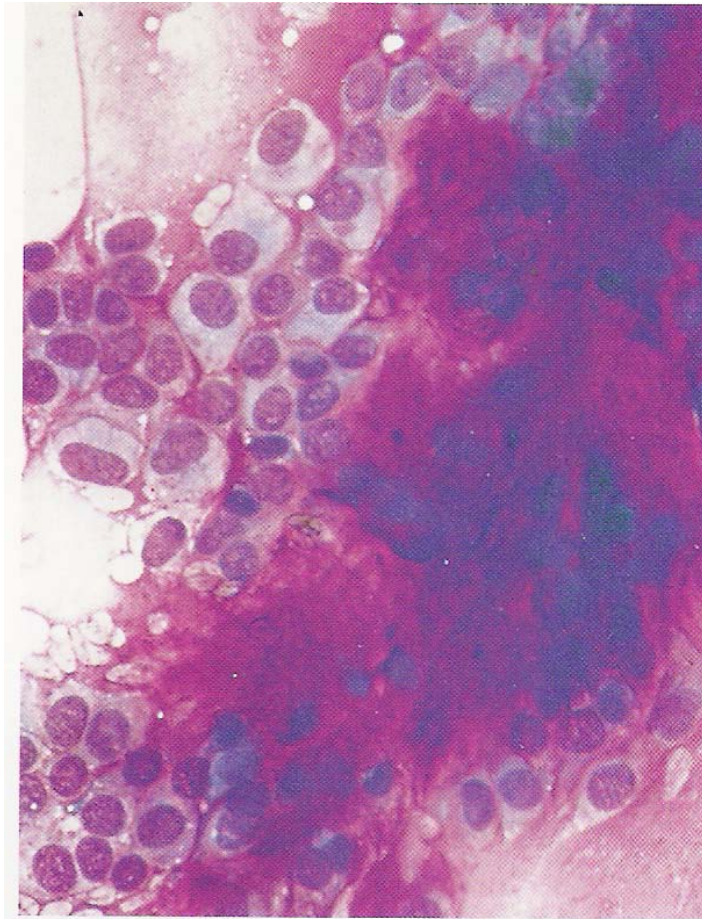


Fig. 4.23 Pleomorphic adenoma
Mixture of epithelial cells and myxoid stroma. Note the well-defined cytoplasm in the epithelial cells (MGG, HP).



Fig. 4.26 Pleomorphic adenoma

This is the most striking example of hyaline stromal globules that we have seen in a pleomorphic adenoma. Note bland appearance of epithelial cell nuclei (MGG, IP).

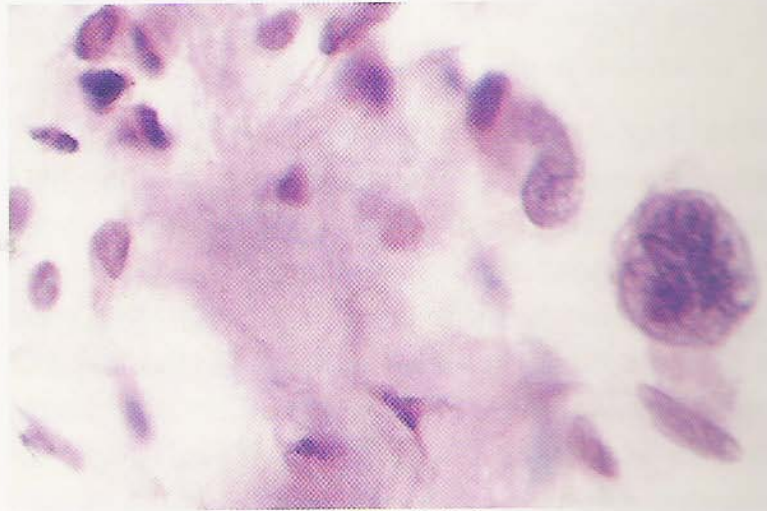


Fig. 4.27 Pleomorphic adenoma

Several pleomorphic bizarre epithelial cells among fragments of myxoid stroma; histology showed no evidence of malignancy (H & E, HP).

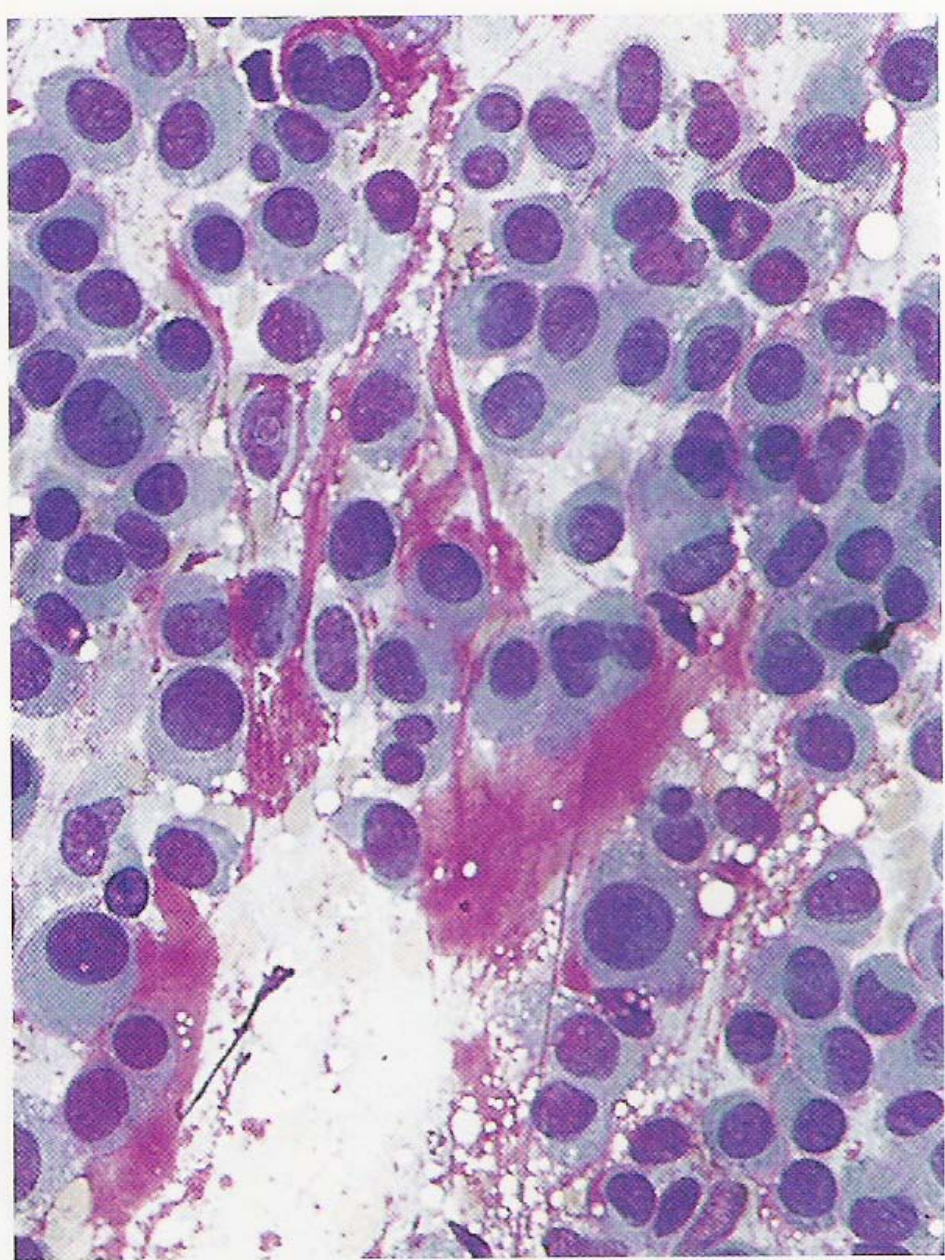


Fig. 4.25 Pleomorphic adenoma

Epithelial cell predominance; the degree of nuclear pleomorphism here should not suggest malignancy (MGG, HP).

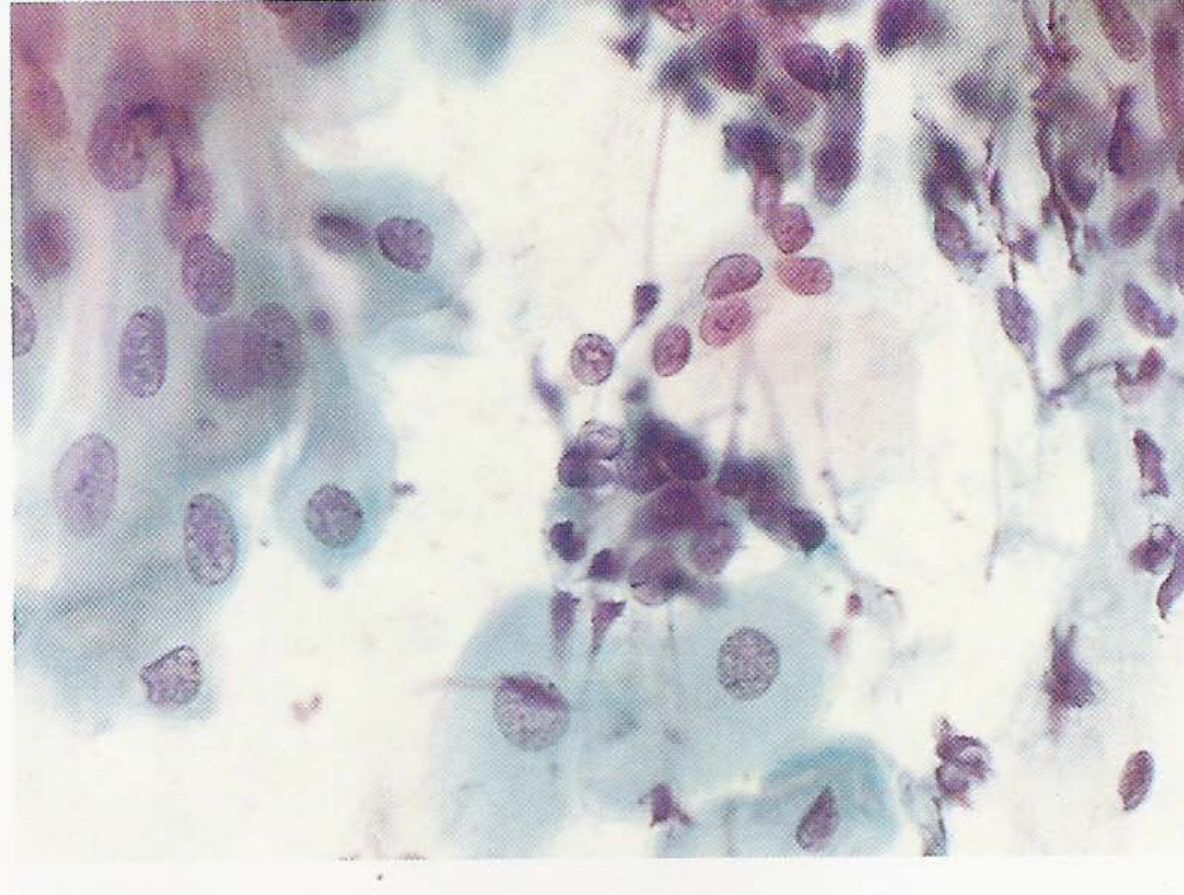


Fig. 4.31 Pleomorphic adenoma
Prominent component of squamous epithelial cells (left and centre); some small epithelial cells and fragments of myxoid stroma (upper right) reveal the nature of the lesion (Pap, HP).

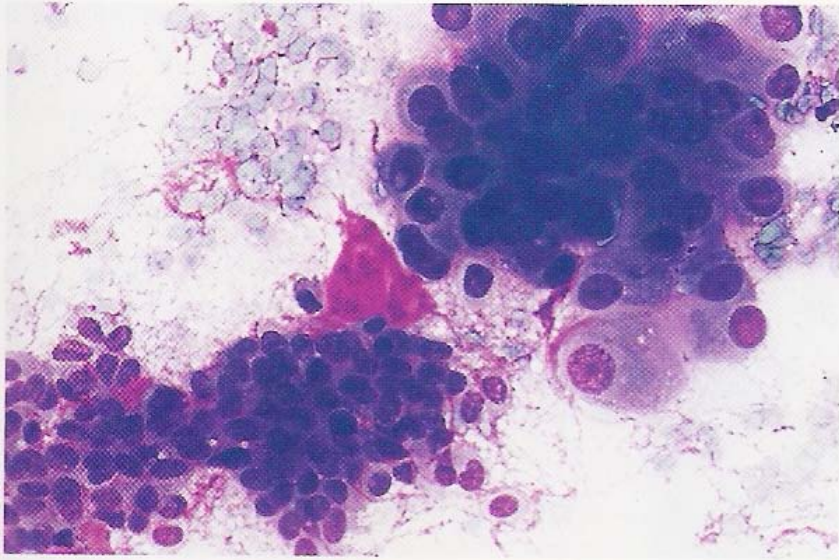


Fig. 4.28 Carcinoma arising in pleomorphic adenoma
The epithelial cell cluster upper right shows prominent nuclear enlargement and atypia, the clusters at lower left are of the usual benign appearance with a fragment of myxoid stroma (MGG, HP).

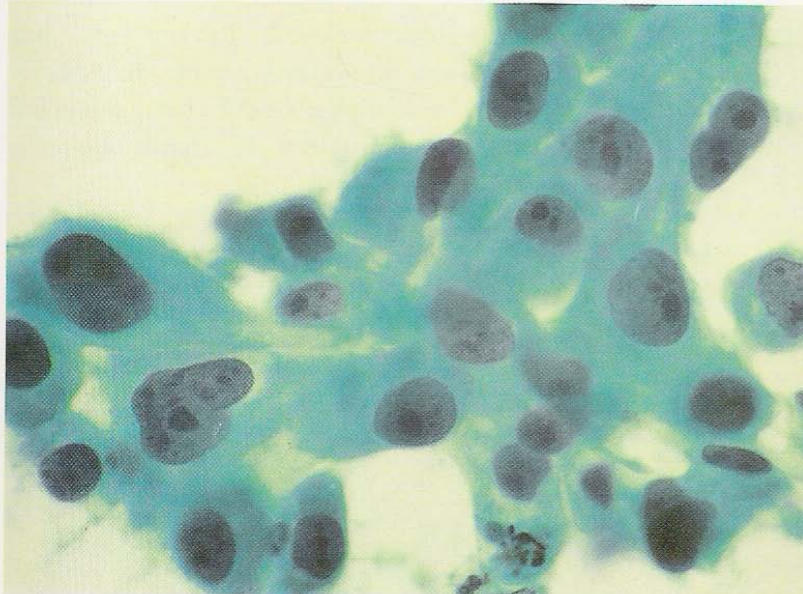


Fig. 4.30 Carcinoma in pleomorphic adenoma
Large, poorly differentiated malignant cells. Histology showed poorly differentiated carcinoma and small foci of residual typical pleomorphic adenoma (Pap, HP).

BASAL CELL ADENOMA

Criteria for diagnosis

- ❖ Numerous cell clusters with few dissociated cells
- ❖ Regular round or oval nuclei and sparse cytoplasm
- ❖ Bland, granular nuclear chromatin
- ❖ Variable amounts of stromal material probably of basement origin

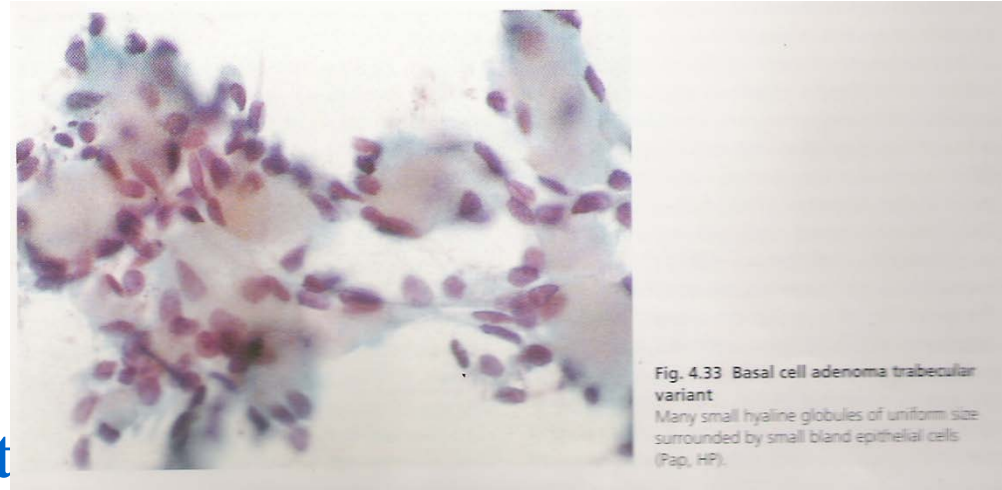


Fig. 4.33 Basal cell adenoma trabecular variant
Many small hyaline globules of uniform size surrounded by small bland epithelial cells (Pap, HP).

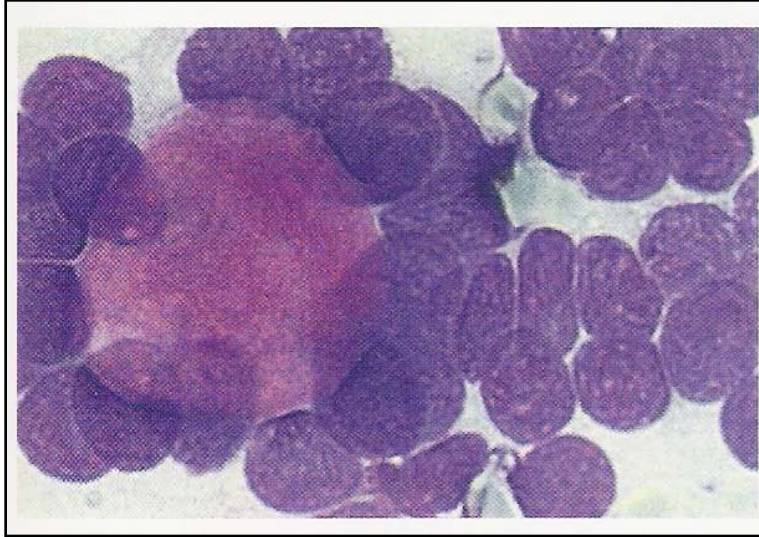


Fig. 4.34 Basal cell adenoma trabecular variant

Small hyaline globules, small epithelial cells with bland granular chromatin (MGG, HP oil). (Courtesy Dr K. Lindholm, Malmö General Hospital)

Warthin's tumor

- ❖ **Criteria for diagnosis**
- ❖ **Aspirate of mucoid, murky fluid**
- ❖ **Background of amorphous and granular debris**
- ❖ **Bland oncocytic cells in cohesive, monolayered sheets**
- ❖ **Many lymphoid cells**

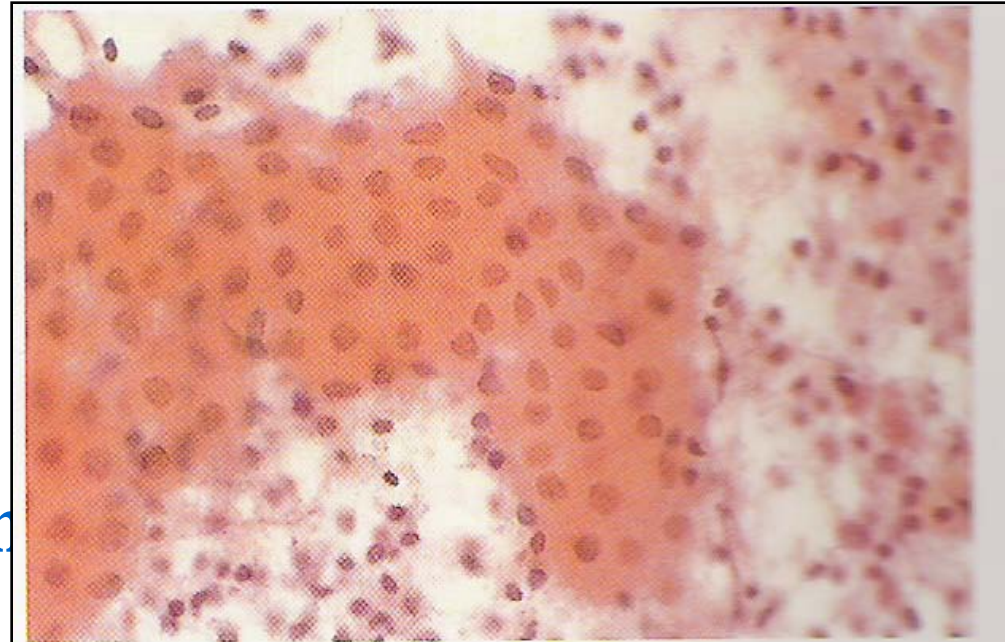


Fig. 4.38 Warthin's tumour

Monolayered sheet of uniform epithelial cells of oncocytic type; background of lymphocytes and debris (Pap, HP).

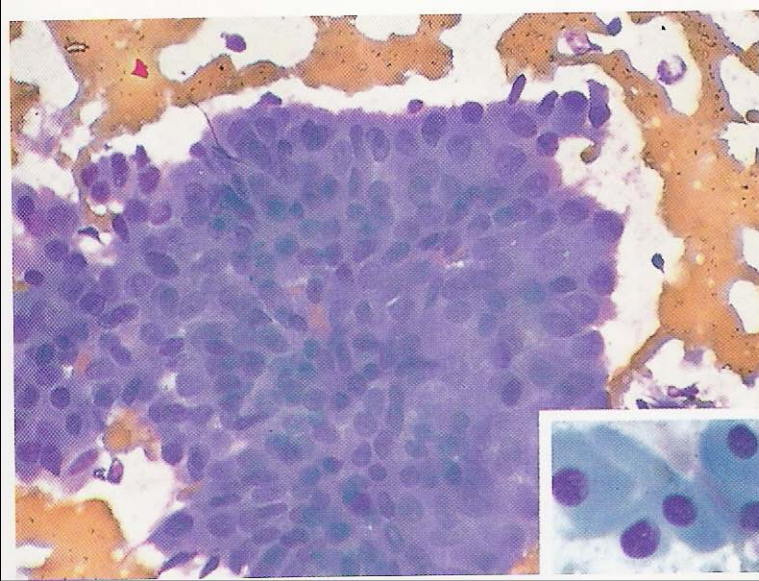


Fig. 4.39 Warthin's tumour
Monolayered sheet of oncocytes; note the peripheral palisading (MGG, HP). Inset: squamoid oncocytes with dense blue cytoplasm (MGG, HP).

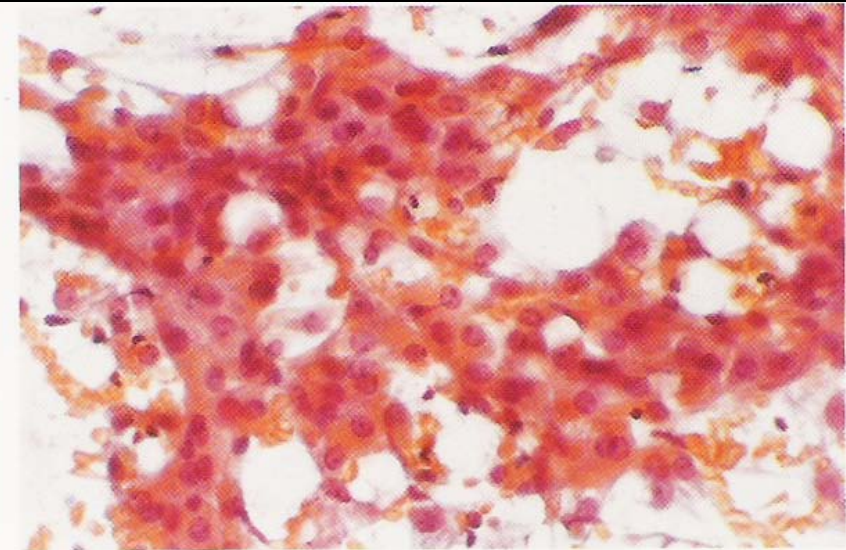
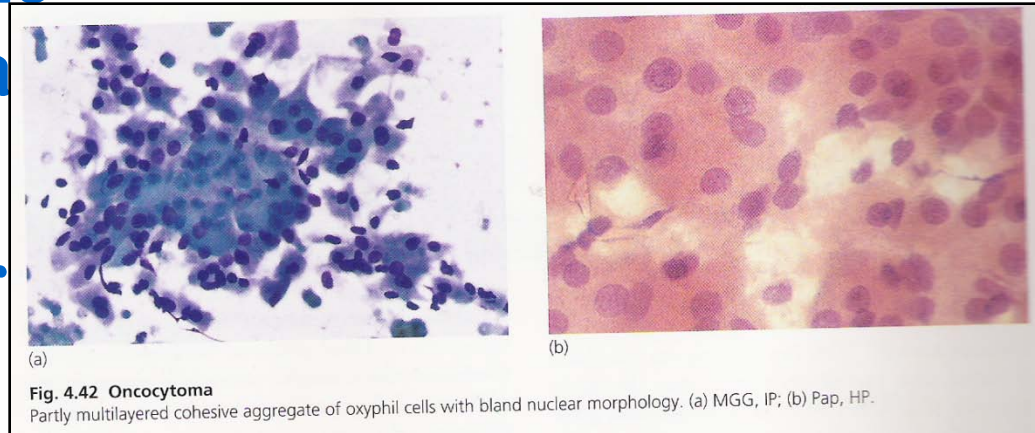


Fig. 4.40 Warthin's tumour
This aggregate of squamous metaplastic cells showing mild atypia and a few cells with intracytoplasmic vacuoles could be mistaken for low-grade mucoepidermoid tumour. The smear, however, showed mainly typical features of Warthin's tumour (Pap, HP).

Oncocytoma

Criteria for diagnosis

- ❖ Cohesive multilayered aggregates of oncocytic cells with small regular nuclei
- ❖ Absence of fluid, debris and lymphoid cells



Other benign neoplasms

❖ Myoepithelial adenoma

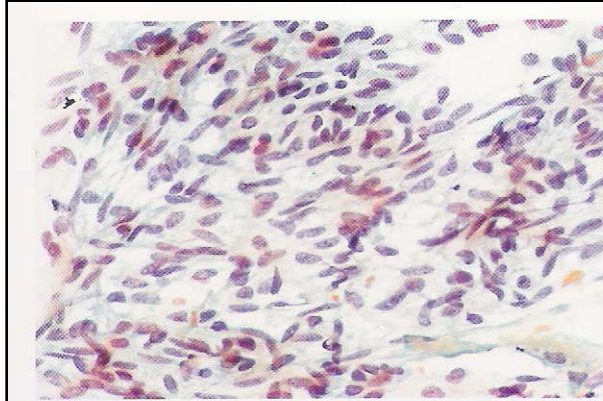


Fig. 4.43 Myoepithelial adenoma, spindle cell type

This pattern of bland spindle cells could be mistaken for a benign soft tissue tumour (Pap, HP).

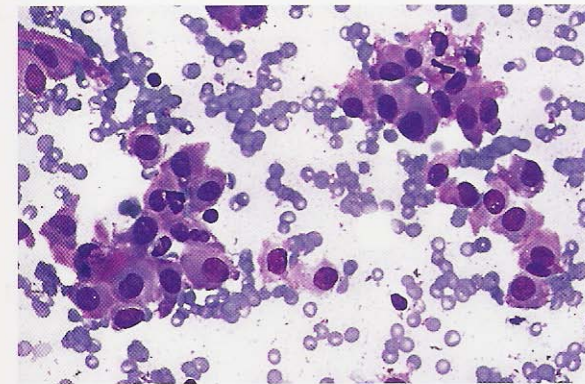


Fig. 4.44 Myoepithelial adenoma, plasmacytoid type

Loosely cohesive clusters of cells with abundant cytoplasm and eccentric, mildly atypical nuclei (MGG, HP).

Malignant neoplasms

Acinic cell carcinoma

Criteria for diagnosis

- ❖ Abundant cell material in a clean background
- ❖ Cohesive clusters of cells sometimes with central fibrovascular cores
- ❖ Poorly formed microacinar groupings
- ❖ Abundant finely vacuolated or dense oncocyte-like cytoplasm
- ❖ Mildly pleomorphic medium-size nuclei, bare, round, lymphocyte-like

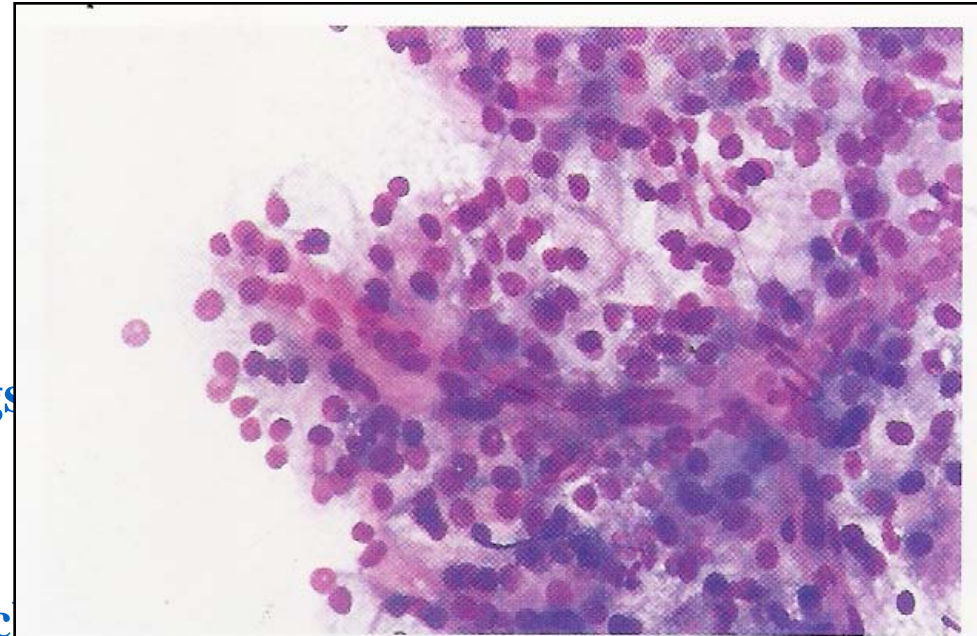
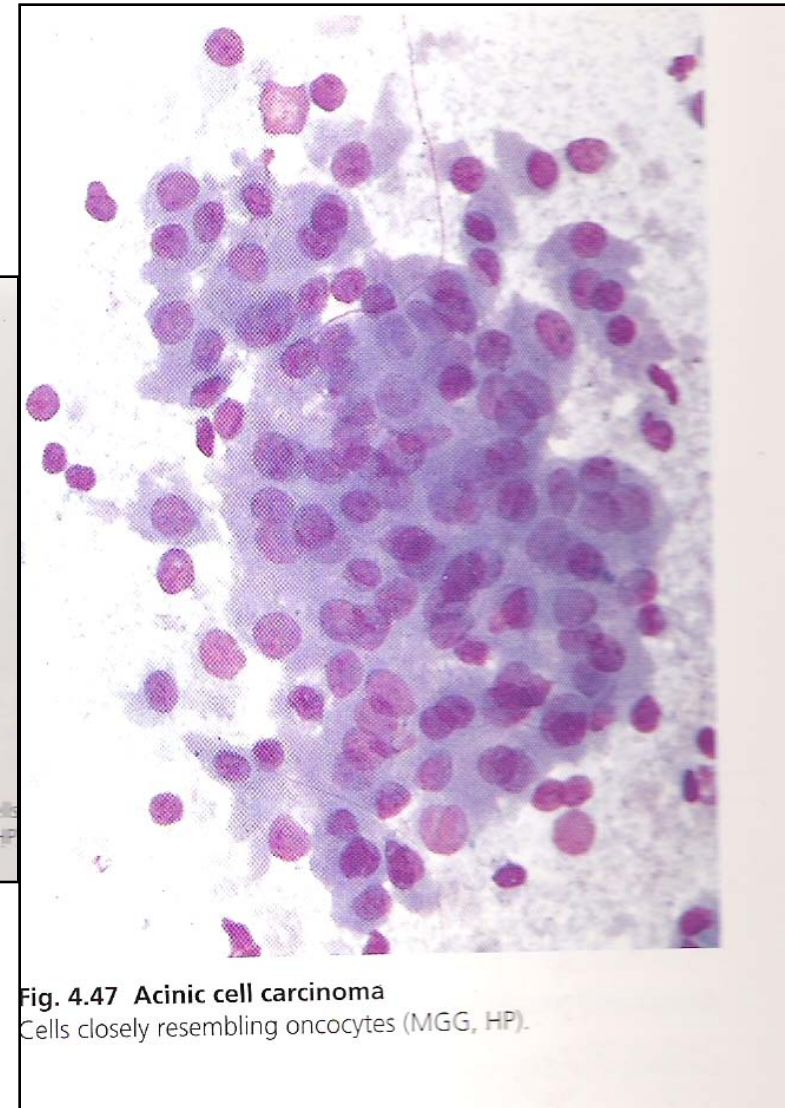
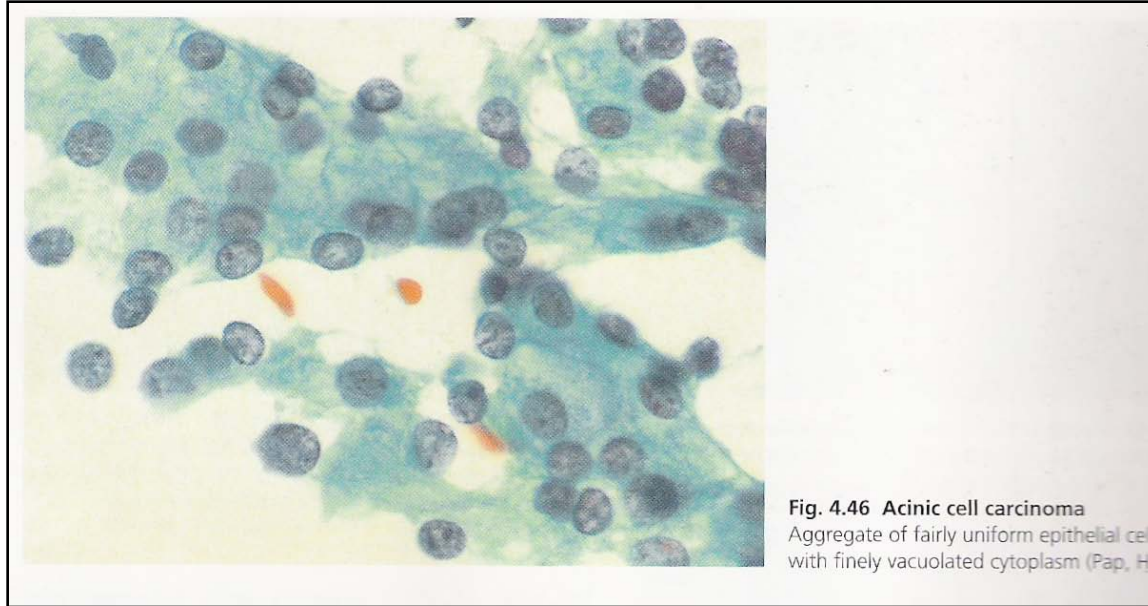


Fig. 4.45 Acinic cell carcinoma

Cohesive fragment composed of rather bland vacuolated cells resembling normal acinar cells adherent to a thin fibrovascular stroma. Note absence of well-formed acinar structures (MGG, IP).



Adenoid cystic carcinoma

Criteria for diagnosis

1. Hyaline spherical globules of varying size (basement membrane material) surrounded by tumour cells.
2. Finger-like or beaded hyaline stroma between cell clusters. Hyaline stromal material may be absent in poorly differentiated tumours.
3. Dense aggregates of small cells with uniform rounded or oval nuclei and scanty cytoplasm.
4. Hyperchromatic nuclei, coarse nuclear chromatin, nucleoli.

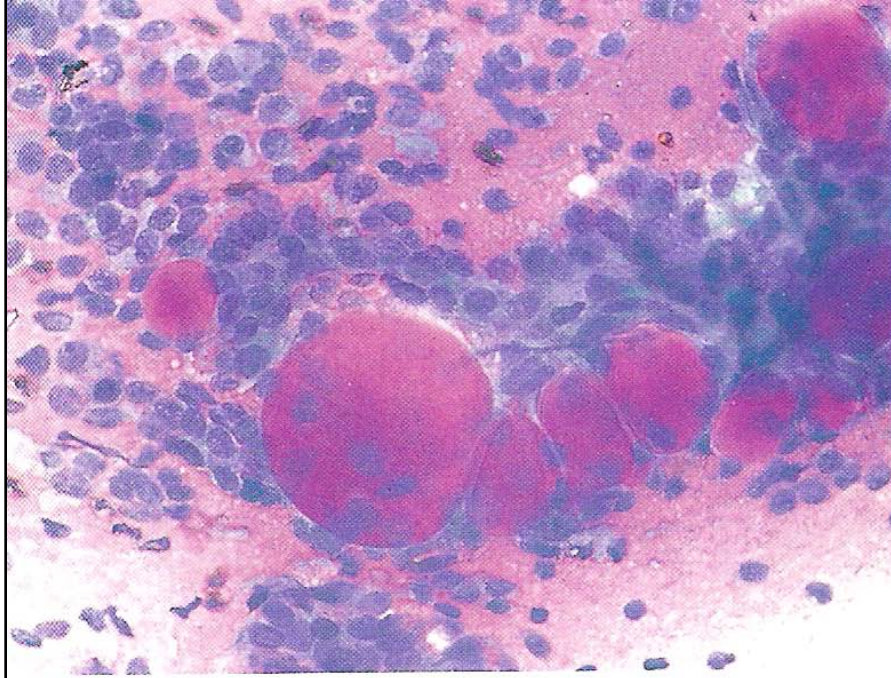


Fig. 4.49 Adenoid cystic carcinoma
Small uniform epithelial cells with hyperchromatic nuclei, hyaline stromal globules (MGG, HP).

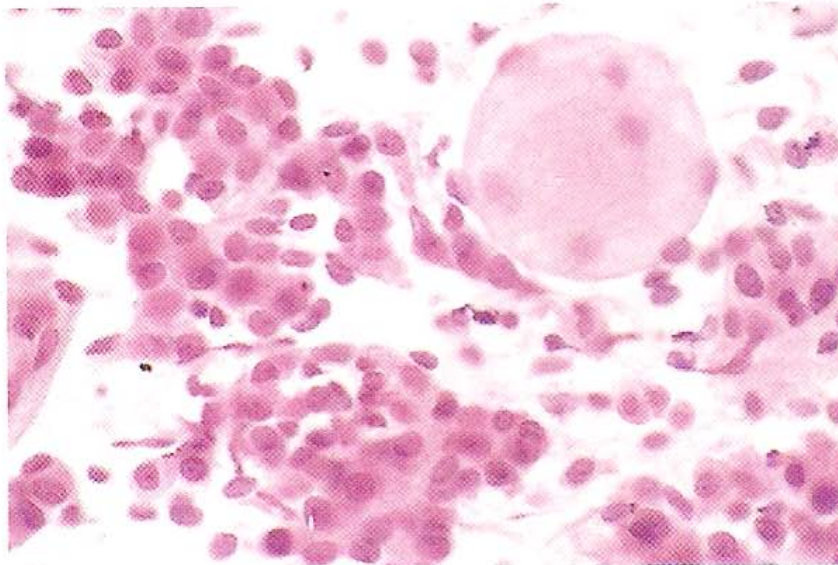


Fig. 4.50 Adenoid cystic carcinoma
Small uniform epithelial cells and a translucent hyaline globule (H & E, HP).

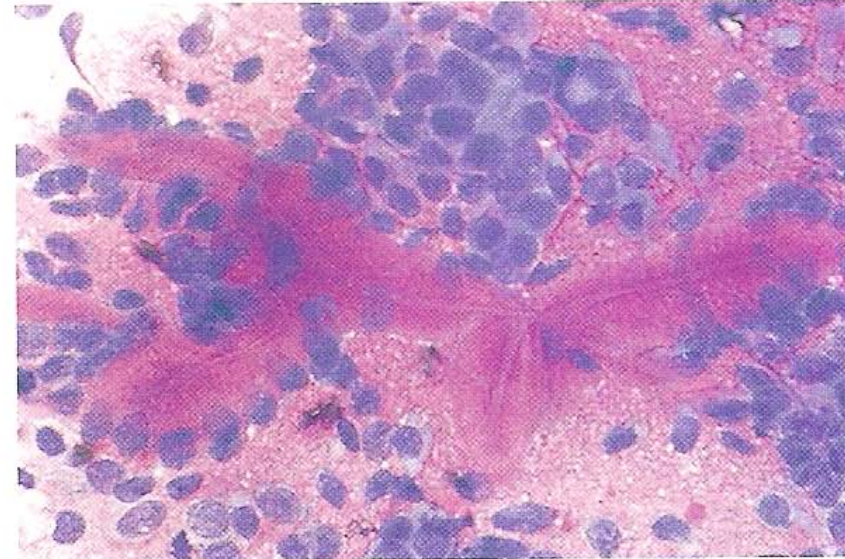


Fig. 4.51 Adenoid cystic carcinoma
Basement membrane material in finger-like structures (MGG, HP).

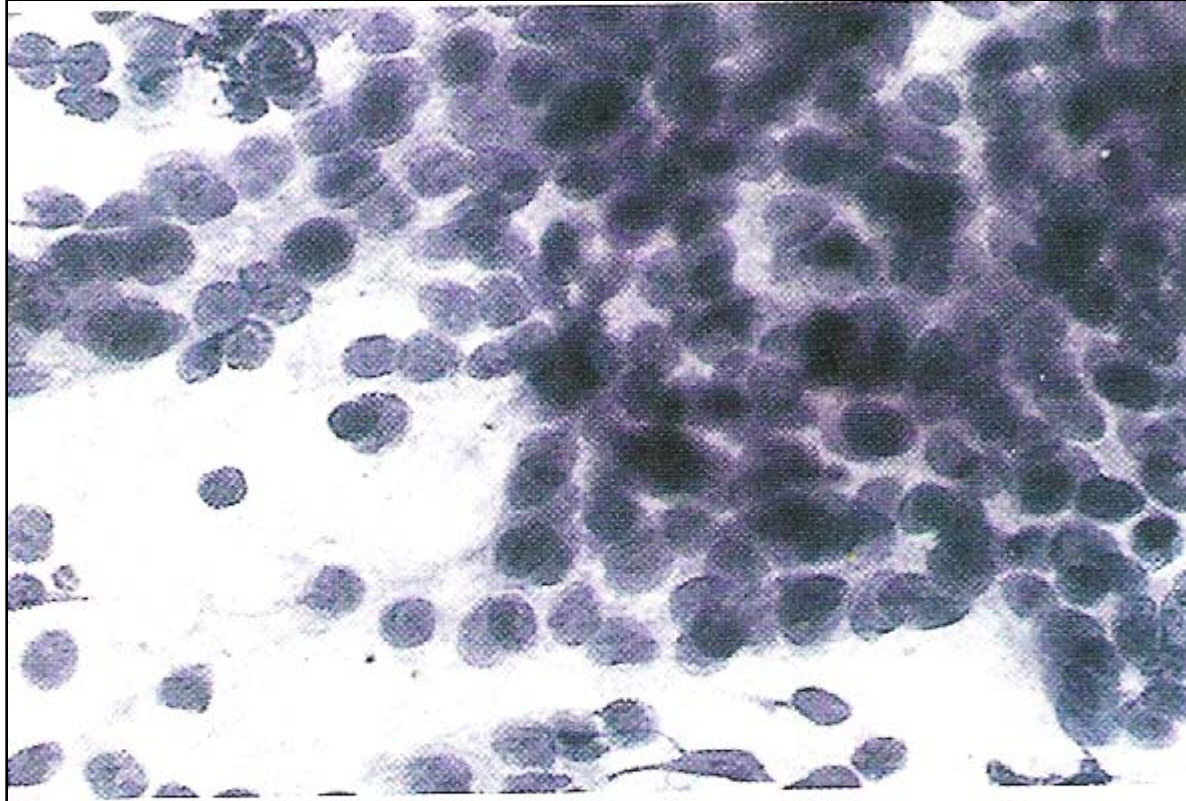


Fig. 4.52 Adenoid cystic carcinoma, poorly differentiated
Numerous small epithelial cells, single and in dense clusters;
hyperchromatic nuclei with coarse chromatin, stromal elements
absent (Pap, HP).

Mucoepidermoid tumour

Criteria for diagnosis

1. A dirty background of mucus and debris.
2. Cohesive clumps and sheets of cells together with small streams of cells within mucus.
3. Variation in cell type – intermediate, squamous, mucin secreting – most with abundant cytoplasm.
4. Rather regular nuclei, prominent nucleoli in some cells.

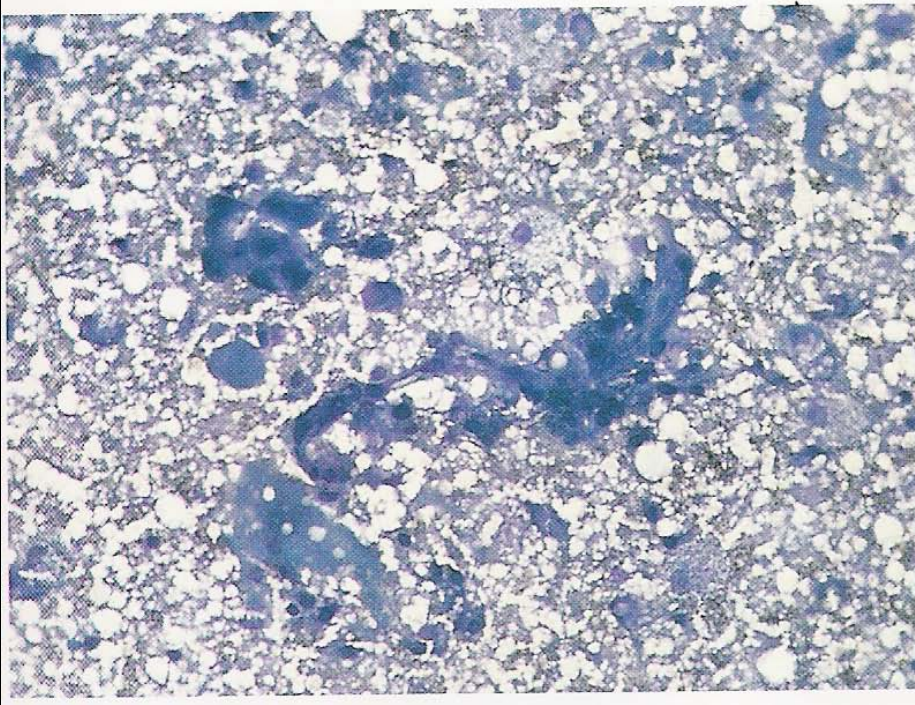


Fig. 4.53 Mucoepidermoid tumour
Degenerating squamoid cells partially obscured by background debris and mucus (MGG, IP).

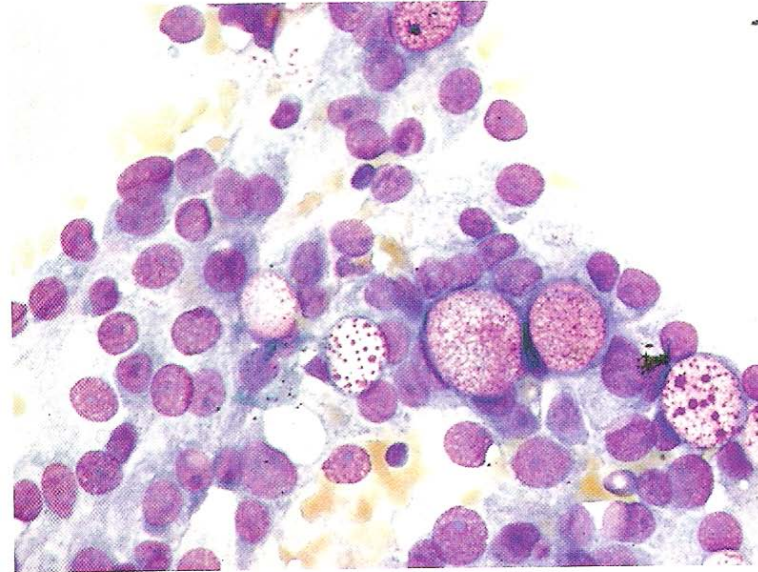


Fig. 4.54 Mucoepidermoid tumour
Intracytoplasmic mucin vacuoles (MGG, HP).

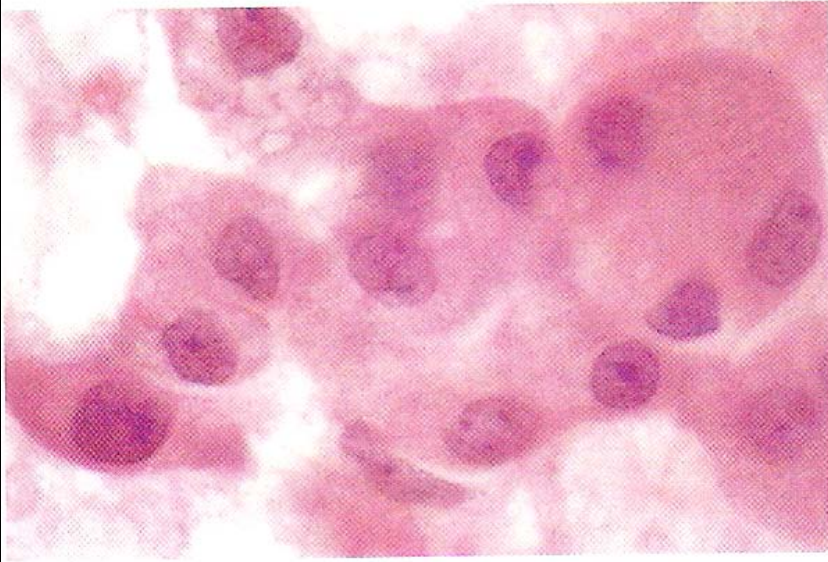


Fig. 4.55 Mucoepidermoid tumour
Intermediate cells with some cytoplasmic vacuolation and mild nuclear atypia (H & E, HP oil).

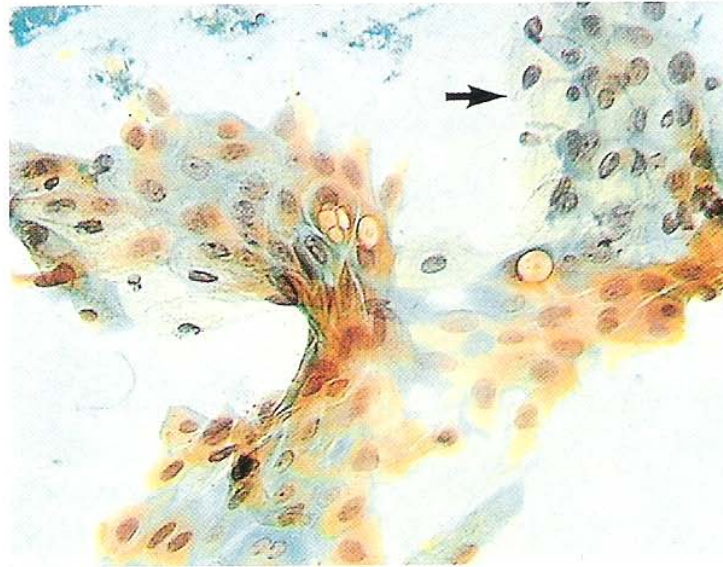


Fig. 4.57 Mucoepidermoid tumour
Moderately cohesive aggregate of atypical epithelial cells – squamous, intermediate and with intracytoplasmic mucin vacuoles; note cells with abundant foamy cytoplasm and small nuclei resembling macrophages (arrow); intermediate grade tumour (Pap, HP).

ADENOCARCINOMA

Criteria for diagnosis

1. Nuclear features of malignancy.
2. Some glandular differentiation (variable).
3. Intracellular and/or extracellular mucus secretion.

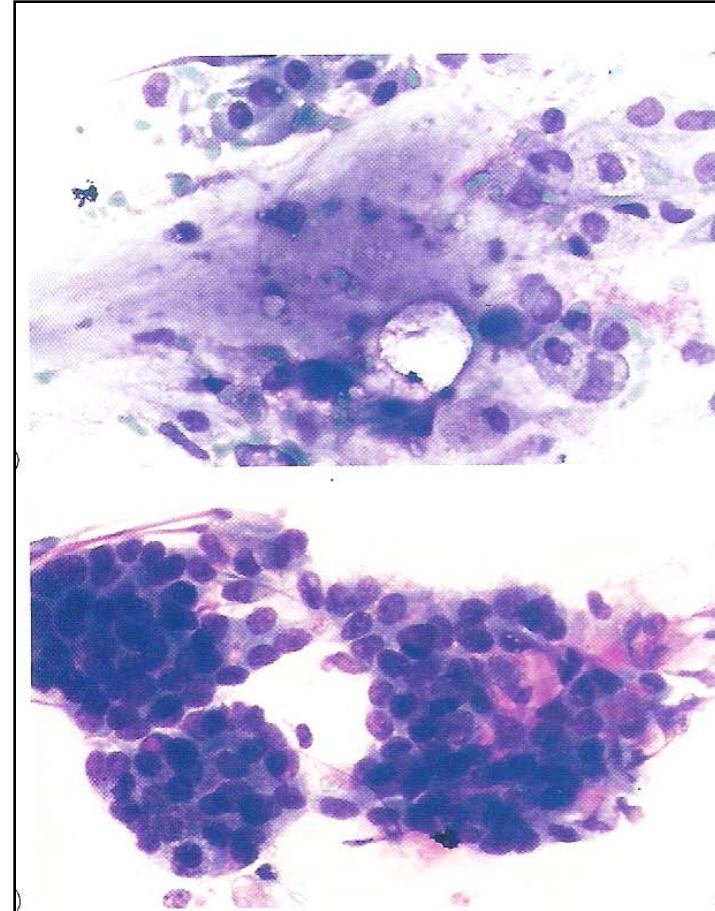


Fig. 4.59 Mucus-secreting 'adenopapillary' carcinoma
(a) Poorly cohesive, mildly atypical epithelial cells with a background of mucus. (b) Cohesive papillary clusters or similar epithelial cells, some with intracytoplasmic mucus (MGG, HP).

OTHER MALIGNANT NEOPLASMS

- ❖ Polymorphous low-grade adenocarcinoma
- ❖ Epithelial myoepithelial carcinoma
- ❖ Renal cell carcinoma metastatic to parotid
- ❖ Salivary duct carcinoma

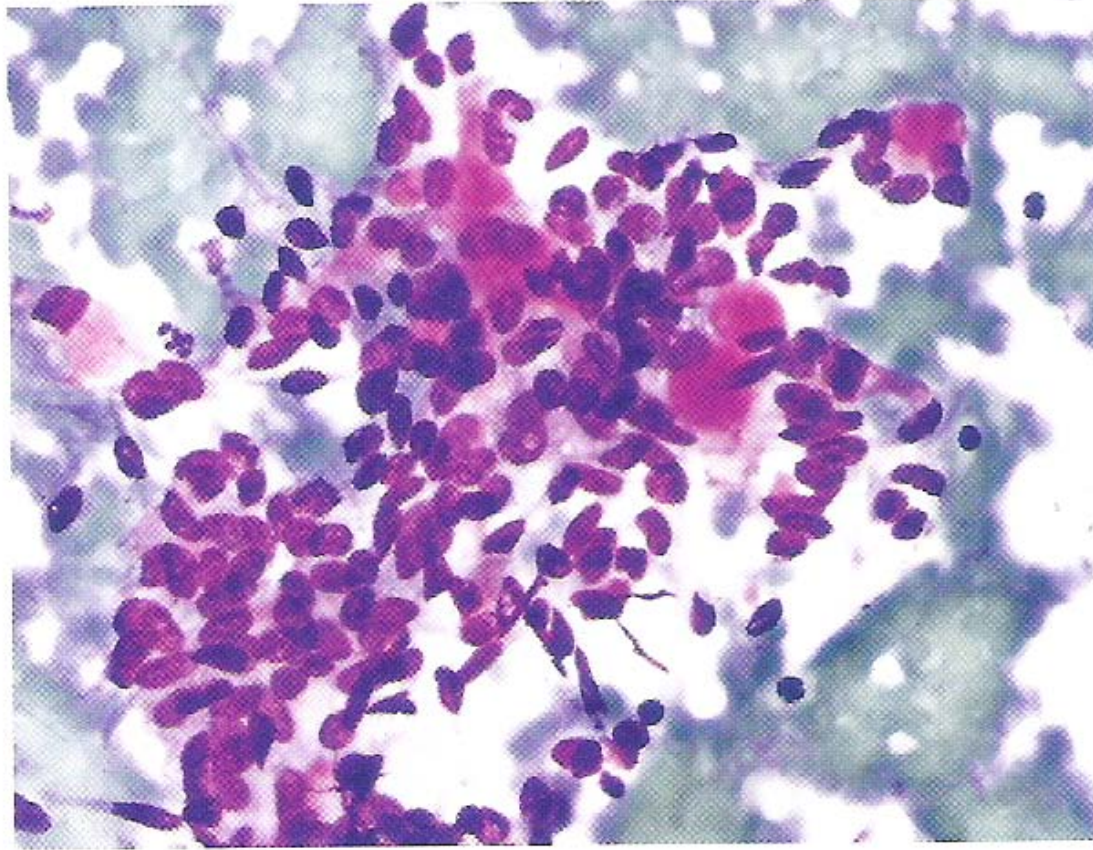


Fig. 4.61 Polymorphous low-grade adenocarcinoma

Poorly cohesive cluster of small cells with oval nuclei and a few small hyaline stromal globules. Some resemblance to adenoid cystic carcinoma, but nuclear chromatin bland (MGG, HP).

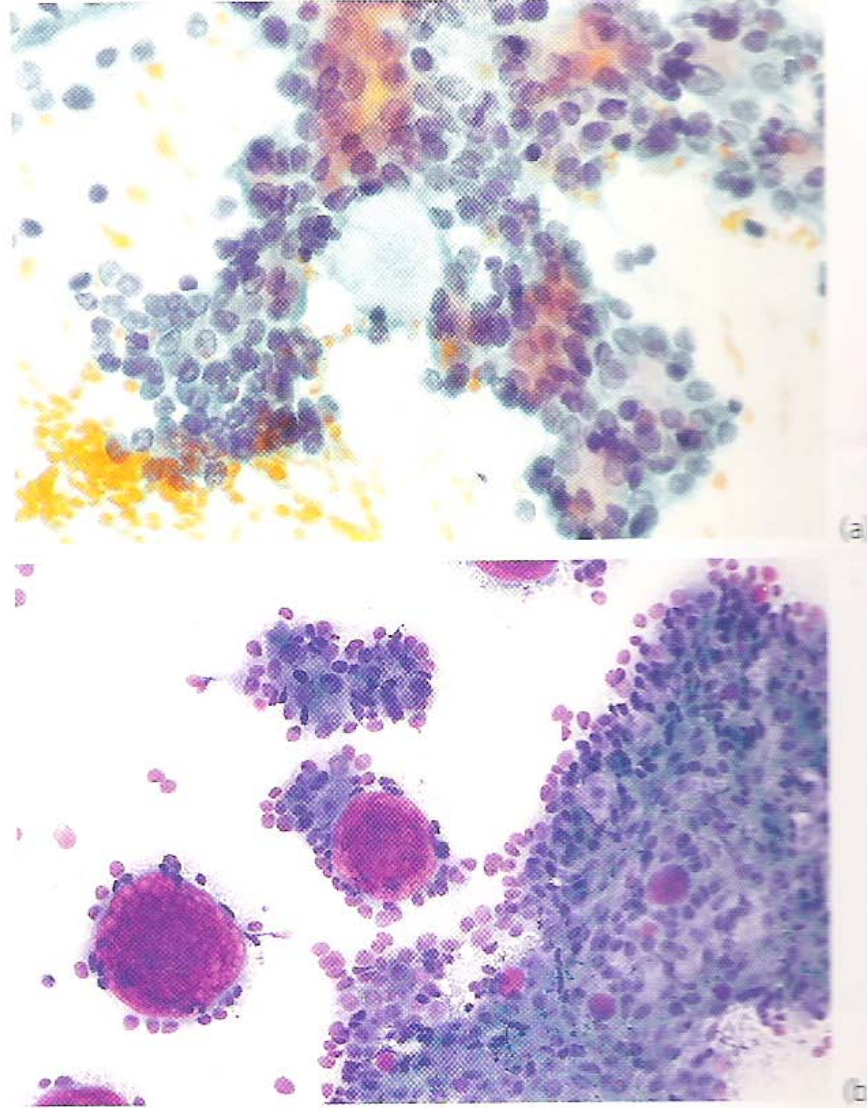


Fig. 4.63 Epithelial-myoeplithelial carcinoma

(a) Rounded clusters of epithelial cells, some with scanty cytoplasm, some with more abundant cytoplasm, and a suggestion of tubular structures. Single naked nuclei are also present (Pap, HP). (b) Similar rounded clusters of epithelial cells plus large hyaline stromal globules (MGG, IP). (Courtesy Dr J. Wright, Gribbles Pathology, Adelaide)

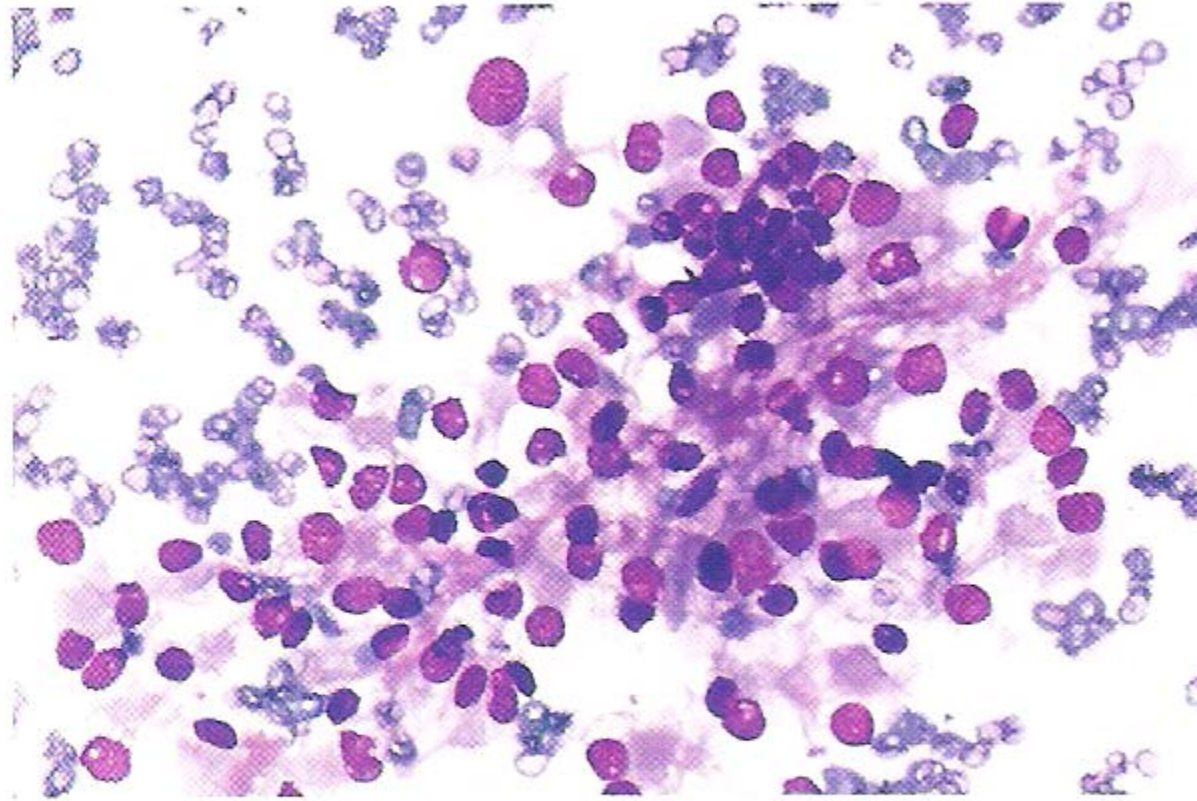


Fig. 4.64 Renal cell carcinoma metastatic to parotid
Aggregate of epithelial cells with abundant pale vacuolated ('clear') cytoplasm adherent to strands of vascular basement membrane (MGG, HP).

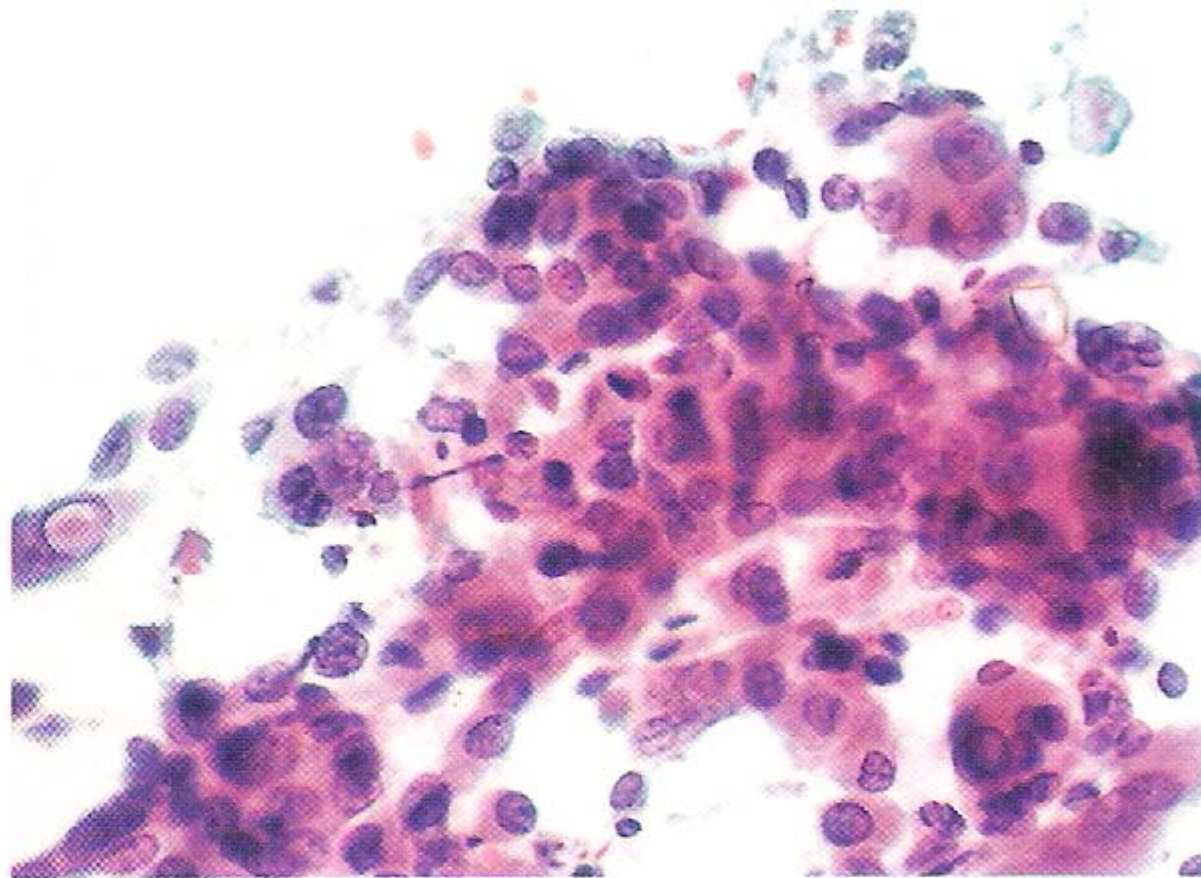
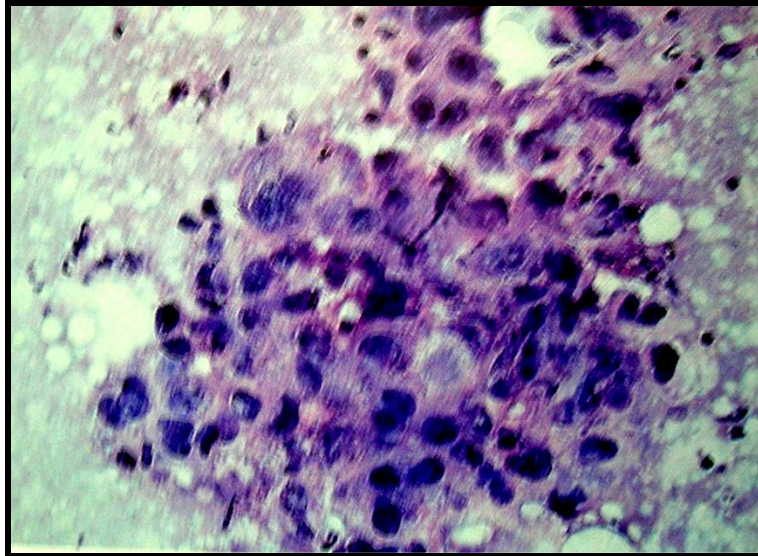


Fig. 4.65 Salivary duct carcinoma

Obviously malignant cells with large pleomorphic nuclei and abundant cytoplasm resembling ductal carcinoma of breast, some necrotic debris present (Pap, HP).

Squamous cell carcinoma



Poorly differentiated squamous cells

How much reliable???

FNAC

- ❖ Is useful in the diagnosis of salivary gland swellings especially in benign conditions with sensitivity of 90% and specificity of 100%.

Kathmandu University Medical Journal (2008), Vol. 6, No. 2, Issue 22, 204-208
Role of FNAC in the diagnosis of salivary gland swellings



FNAC OF LYMPH NODE

The Reactive node

Criteria for diagnosis

1. A mixed population of lymphoid cells.
2. A predominance of small lymphocytes.
3. Centroblasts, centrocytes, immunoblasts and plasma cells in variable but 'logical' proportions.
4. Dendritic reticulum cells associated with centroblasts and centrocytes (representing germinal centres).
5. Scattered histiocytes with intracytoplasmic nuclear debris (tingible body macrophages).
6. Pale histiocytes, interdigitating cells, endothelial cells, eosinophils, neutrophils (variable).

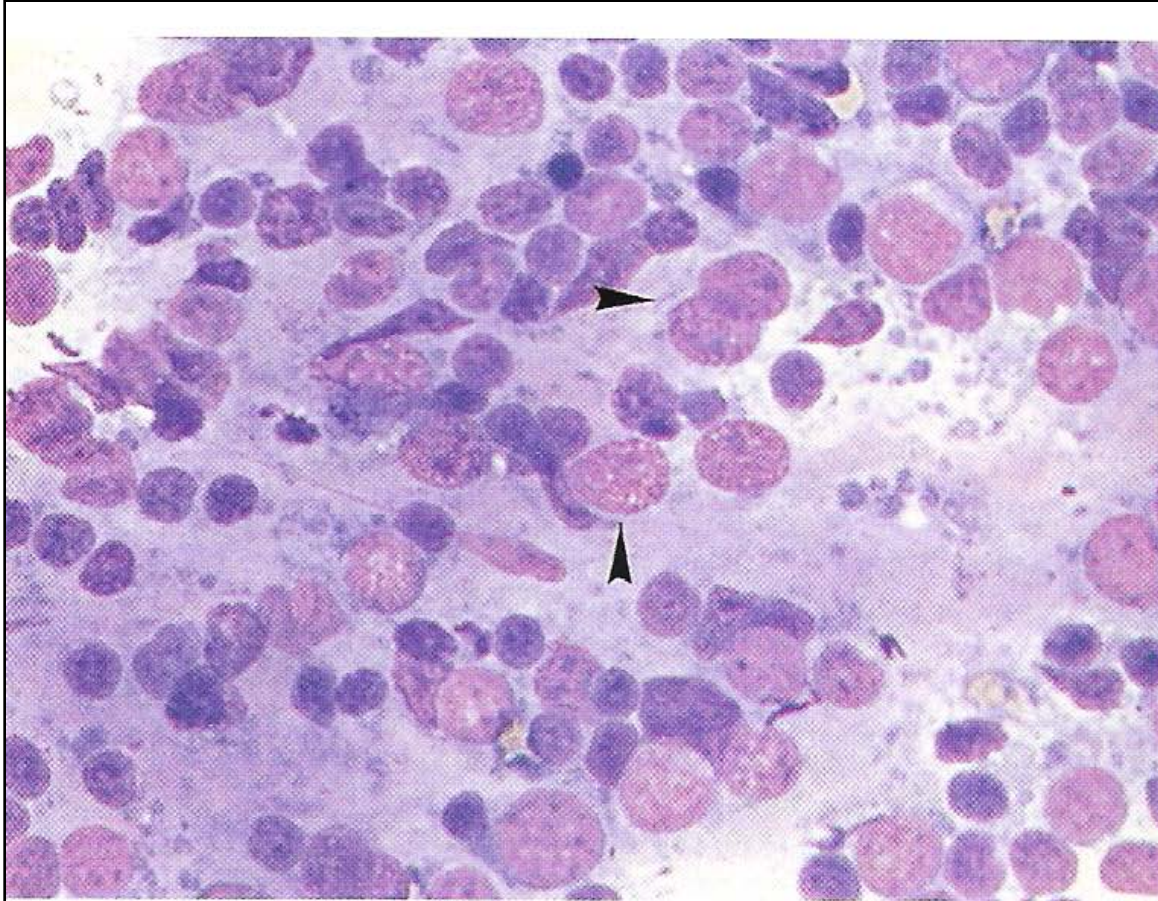


Fig. 5.6 Reactive lymphadenopathy
Smear derived from germinal centre; loose tissue fragment of dendritic reticulum cells (arrows), centroblasts, centrocytes and some lymphocytes (MGG, HP).

Granulomatous lymphadenitis

Criteria for diagnosis

- ❖ Histiocytes of epithelioid type forming cohesive clusters
- ❖ Multinucleated giant cells of Langhans type

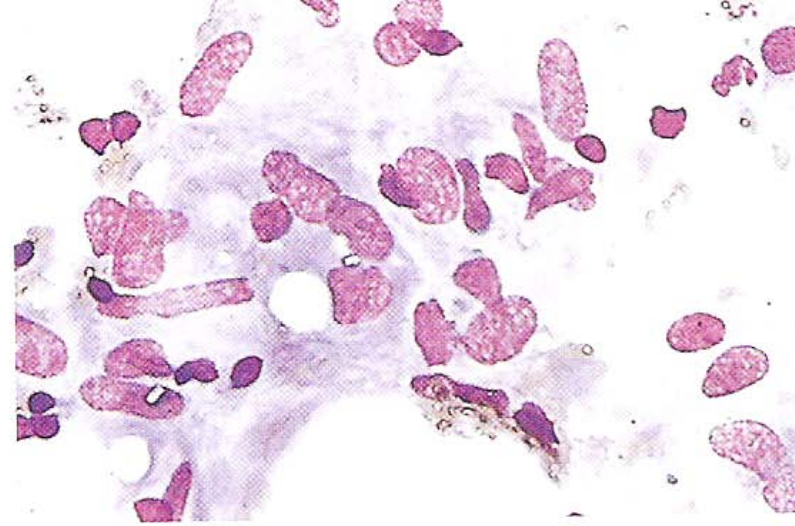


Fig. 5.17 Granulomatous lymphadenitis (sarcoidosis)
Cluster of cohesive epithelioid histiocytes (MGG, HP).

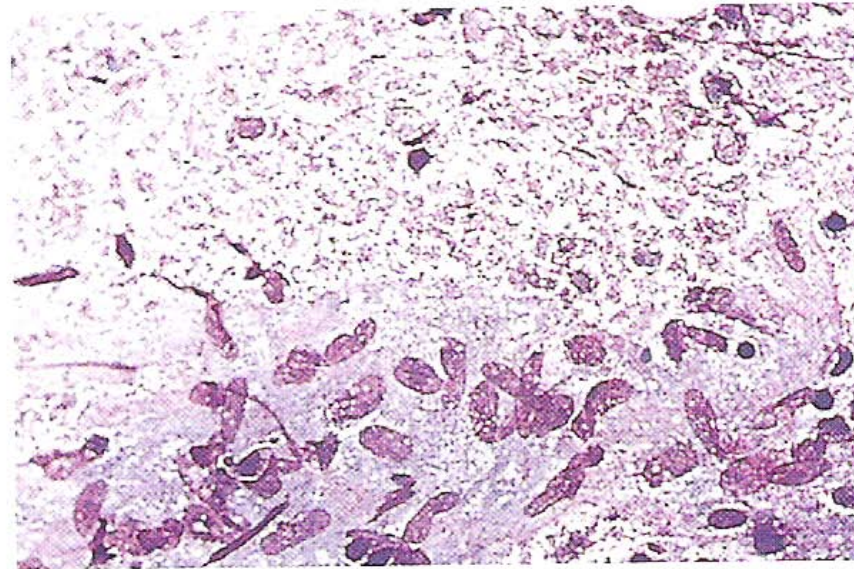


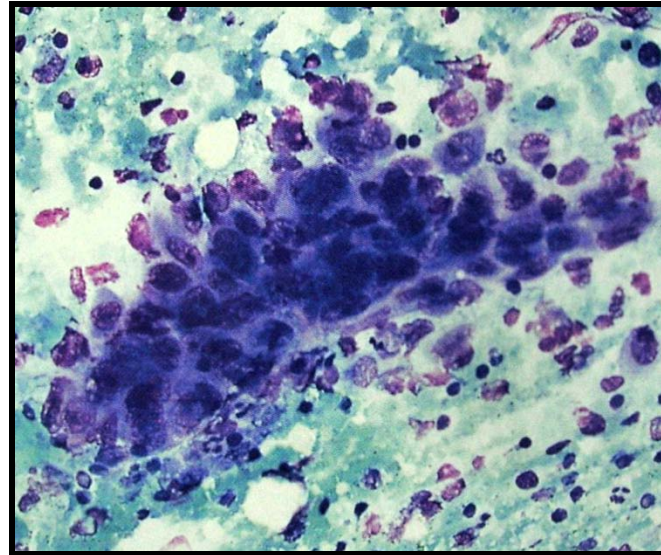
Fig. 5.18 Granulomatous lymphadenitis (tuberculosis)
Small epithelioid cell collection in a background of caseous necrosis (MGG, IP).

Metastatic malignancy

Criteria for diagnosis

- ❖ Foreign cells amongst normal/reactive lymphoid cells
- ❖ Cytological criteria for malignancy

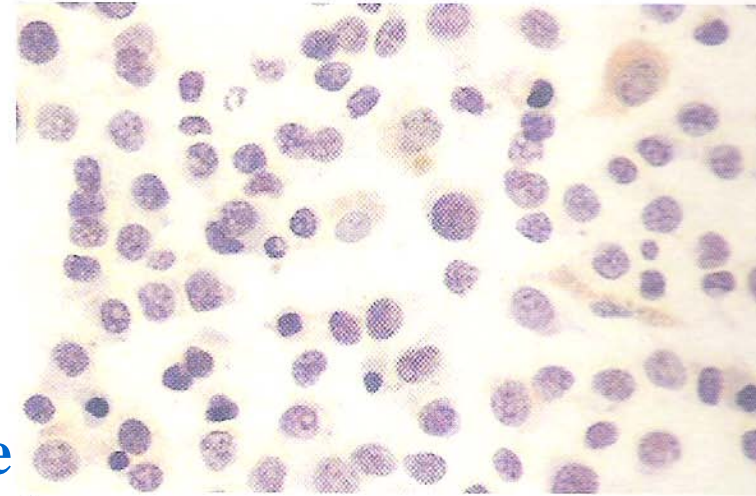
Squamous cell carcinoma



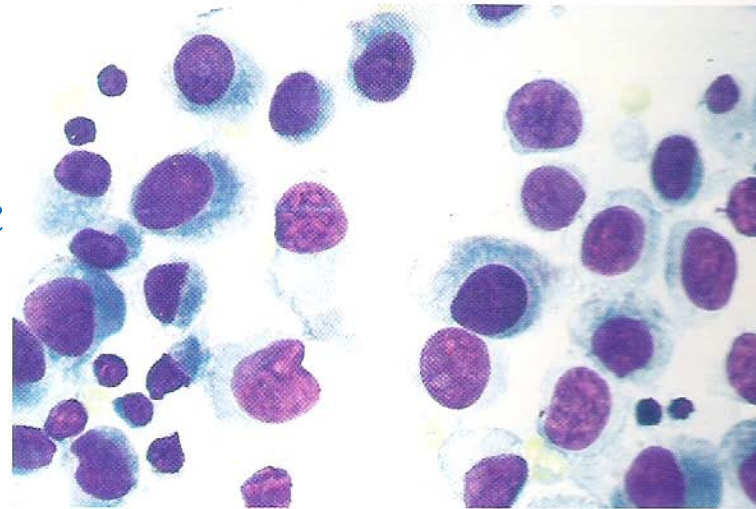
clusters of poorly differentiated squamoid cells with hyperchromatic nucleus

Malignant melanoma

Smears of malignant melanoma may show total dissociation of cells, well defined cytoplasm, eccentric nuclei, prominent anisokaryosis, a uniformly dense chromatin which does not vary much from nucleus to nucleus, often large nucleoli, binucleate cells, intranuclear vacuoles and some cells with intracytoplasmic pigment



(a)



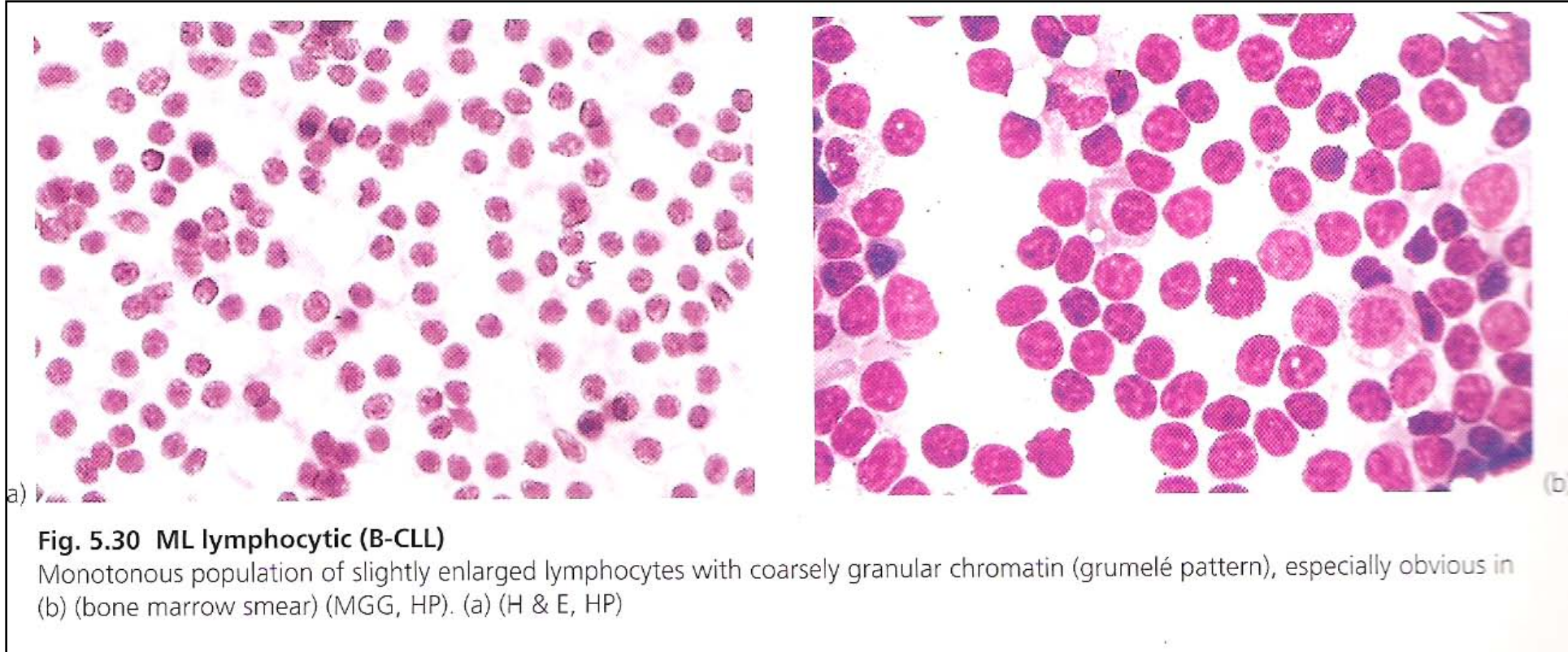
(b)

Fig. 5.27 Metastatic melanoma mimicking lymphoma
(a) Dispersed malignant cells with eccentric nuclei and pale cytoplasm; lymphoid globules absent (Pap, HP). (b) Amelanotic melanoma showing dense smooth chromatin and characteristic paranuclear dark area in tumour cells (MGG, HP).

Malignant lymphomas

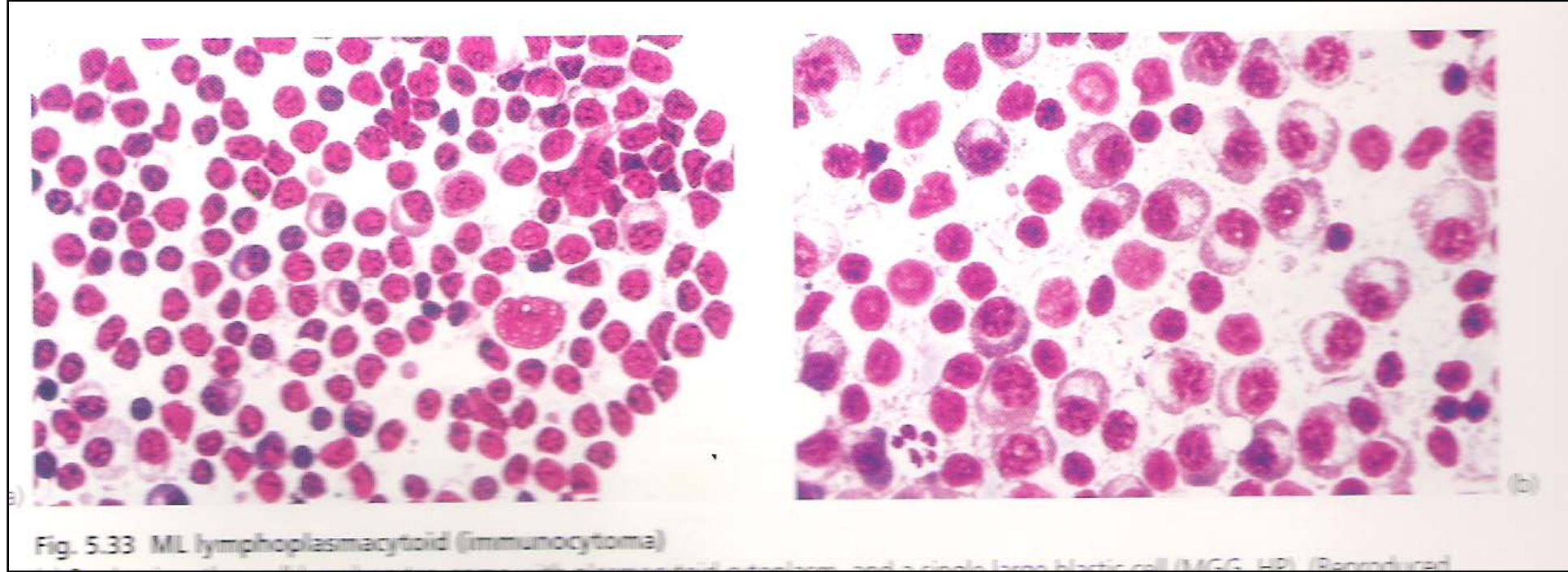
❖ Criteria for diagnosis

1. A monotonous population of small lymphoid cells.
2. Mainly round nuclei slightly larger than those of normal small lymphocytes.
3. Characteristically coarse granular nuclear chromatin (grumelé); nucleoli absent.
4. A varying number of prolymphocytes: larger size, more cytoplasm, pale chromatin, single central nucleolus ().



Malignant lymphoplasmacytoid

- ❖ **Criteria for diagnosis**
- ❖ **A mixed population of lymphocytes, plasma cells and occasionally some blasts**
- ❖ **A variable number of lymphocytes with plasmacytoid features**



ML plasmacytic/plasmacytoma

- ❖ Cells resembling mature or immature (Nucleoli-containing) plasma cells

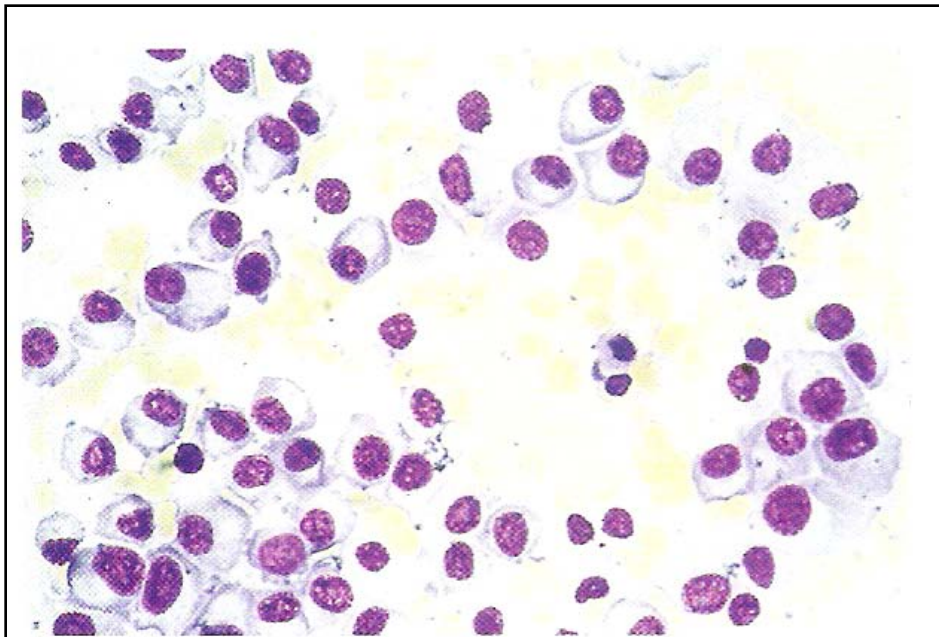
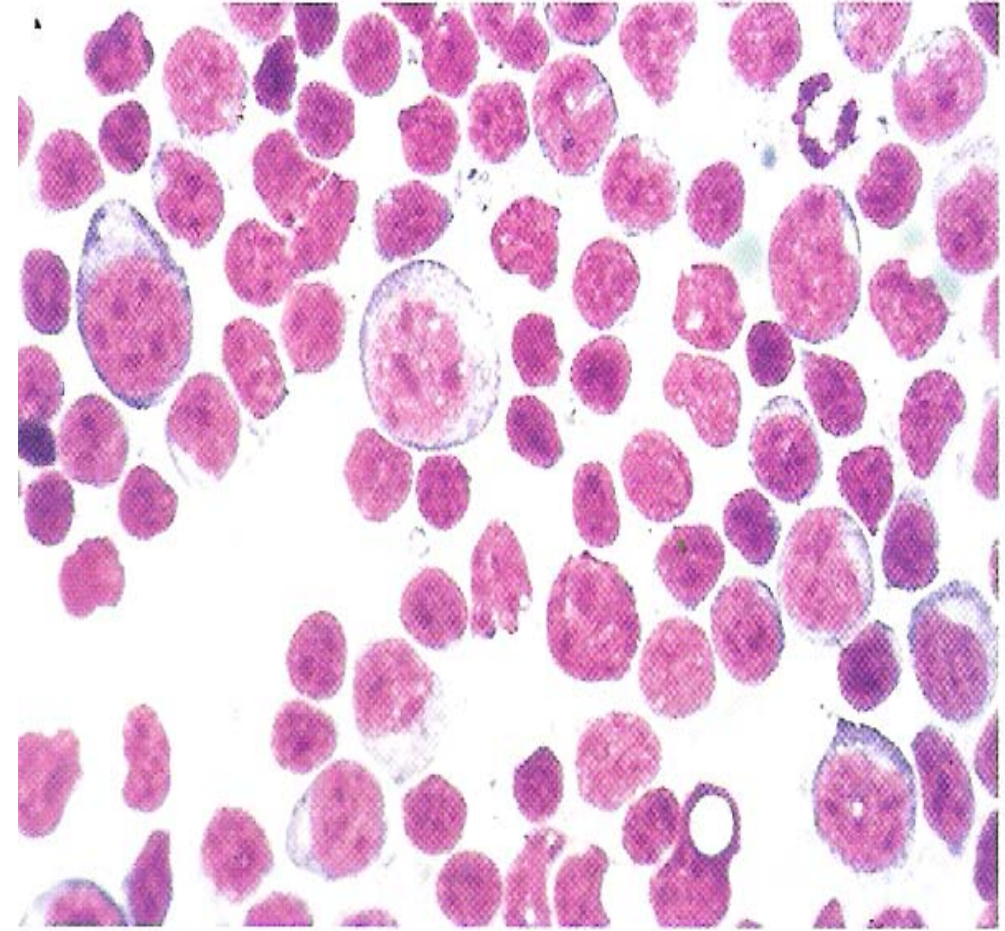


Fig. 5.34 ML plasmacytic
Pure population of well-differentiated plasma cells (MG, HP).

ML centroblastic/centrocytic

1. A variable proportion of centroblasts with large round or slightly irregular nuclei, small nucleoli and basophilic cytoplasm.
 2. Cell population dominated by centrocytes, small to medium-sized, with irregular or cleaved nuclei, inconspicuous nucleoli and little cytoplasm and a tendency to form loose clusters.
-
3. Usually a relatively low number of small lymphocytes.

- ❖ **Predominance of medium-sized cells with irregular, sometimes cleaved nuclei**
- ❖ **Relatively few lymphocytes**



(a)

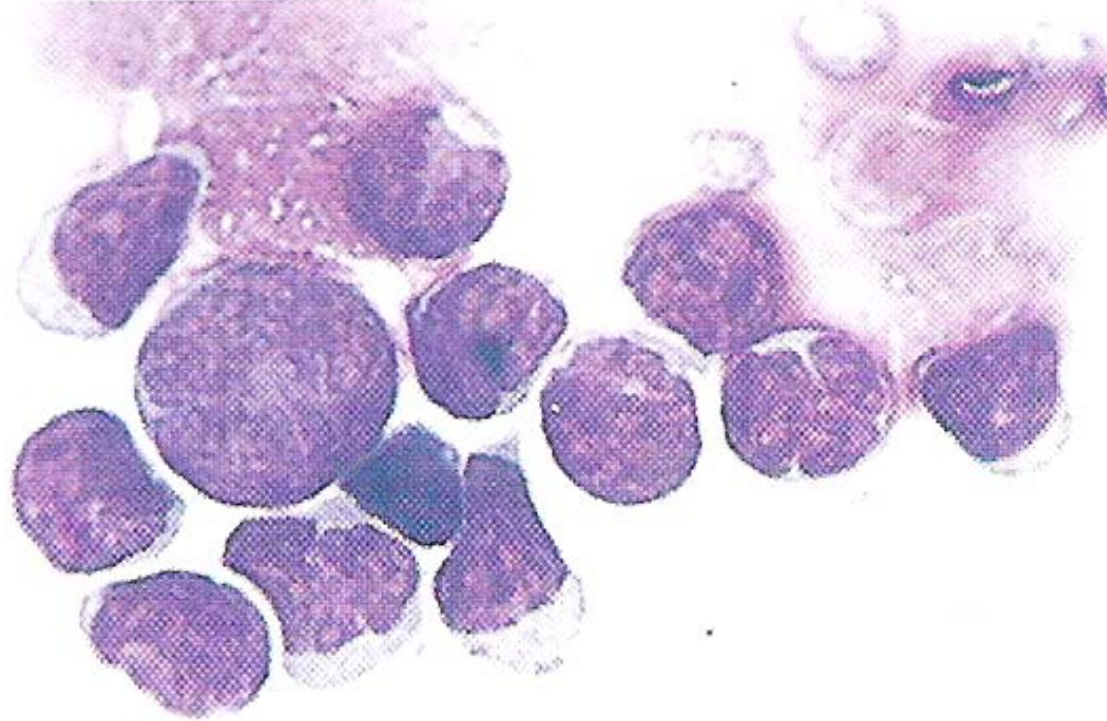
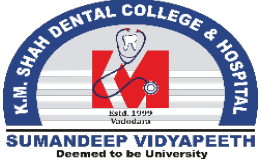


Fig. 5.38 ML centroblastic/centrocytic
Cytocentrifuge preparation; centrocytes showing cleaved
nuclei; one centroblast (MGG, HP oil).

Burkitt's lymphoma

1. A relatively uniform cell population with a high mitotic rate.
2. Rounded nuclei of variable but predominantly intermediate size.
3. A granular or speckled chromatin pattern; multiple small but prominent nucleoli.
4. A variable, mostly thin rim of dense blue cytoplasm with small lipid vacuoles (MGG).
5. Starry sky macrophages often prominent.



SUMANDEEP VIDYAPEETH
K M SHAH DENTAL COLLEGE AND HOSPITAL



Hodgkin's lymphoma

- ❖ **Criteria for diagnosis**
- ❖ **Reed-Sternberg cells**
- ❖ **Atypical mononuclear cells**
- ❖ **A variable number of eosinophils, plasma cells and histiocytes**
- ❖ **A background population of lymphocytes**

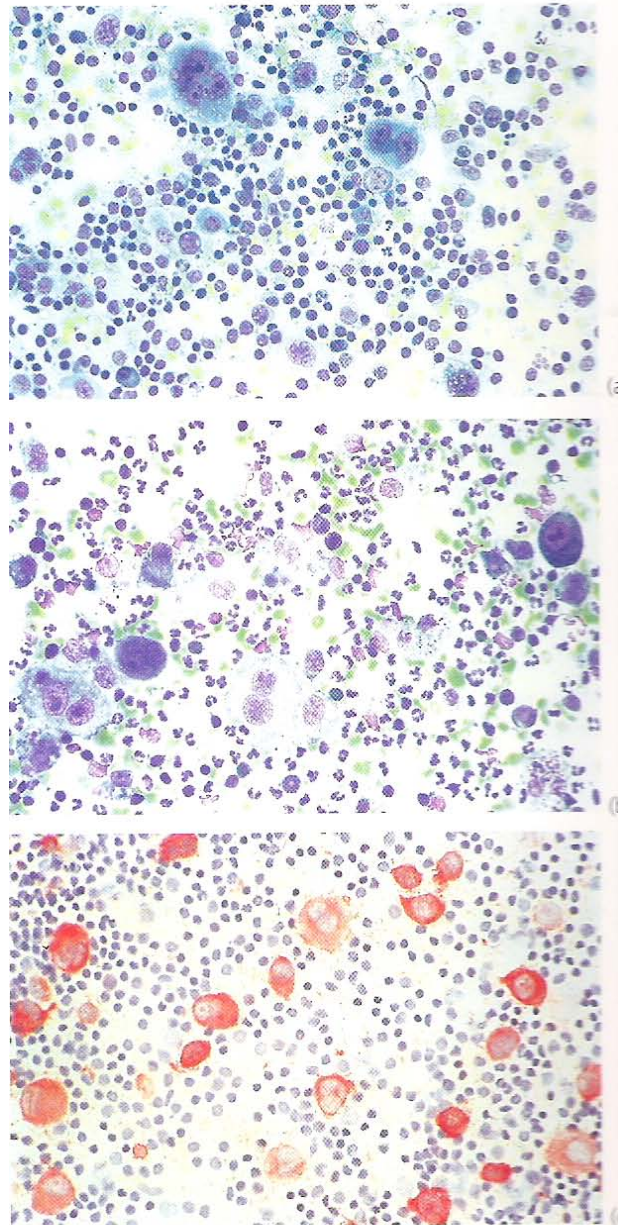


Fig. 5.57 Hodgkin's lymphoma

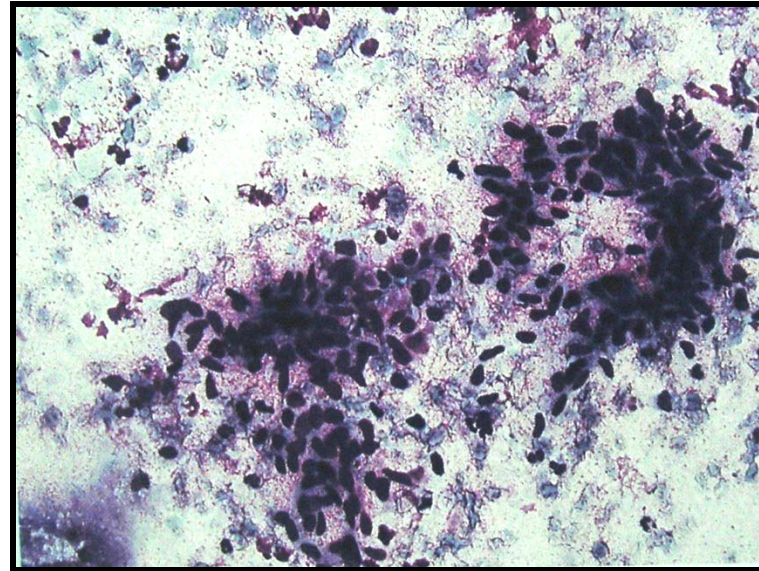
Binuclear Reed-Sternberg cells and mononuclear 'Hodgkin's cells' in a background of mainly small lymphocytes (a) and/or granulocytes (b). Varying basophilia. Characteristic CD30 positivity (a. MGG, IP; b. MGG, IP; c. immunostain CD30, IP).

How much reliable??

- ❖ The diagnostic sensitivity of metastatic and recurring malignancy reported in literature is usually above 95%
- ❖ Conflicting opinions are expressed in the literature regarding the accuracy of cytological diagnosis and typing of malignant lymphoma
- ❖ It is found to be significantly lower for lymphoma than for metastatic malignancy

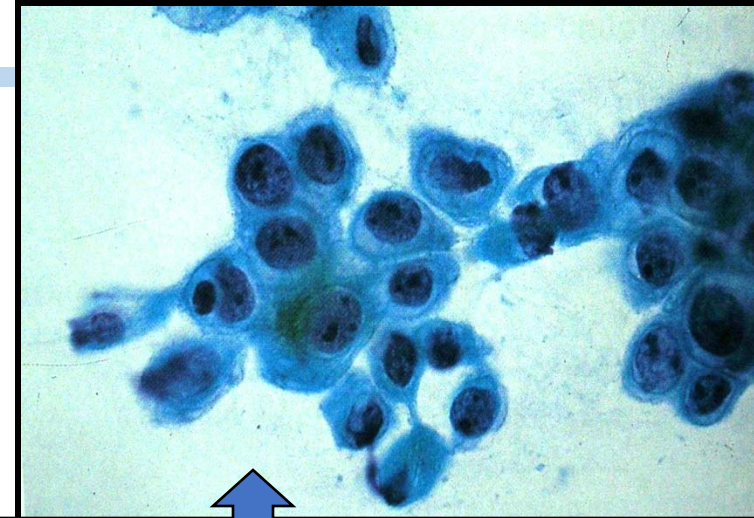
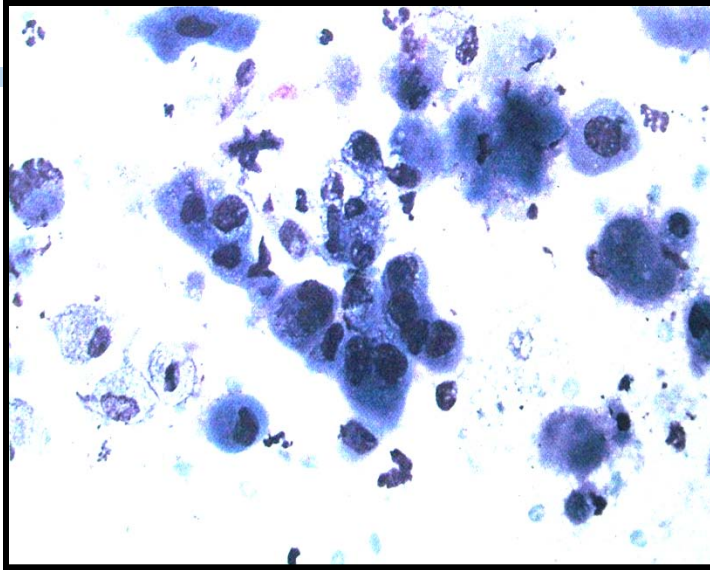
FNAC OF OTHER HEAD AND NECK TUMOUR

BASAL CELL CARCINOMA

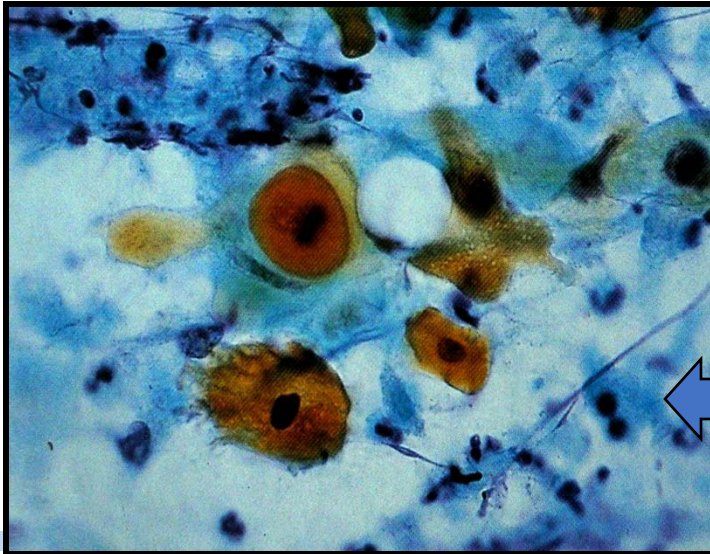


- Tight cell aggregates with alternating smooth and spiculate outline
- Palisading of nuclei along the edge of aggregates
- Scanty cytoplasm
- Small hyperchromatic ovoid nuclei; indistinct nucleoli

Squamous cell carcinoma

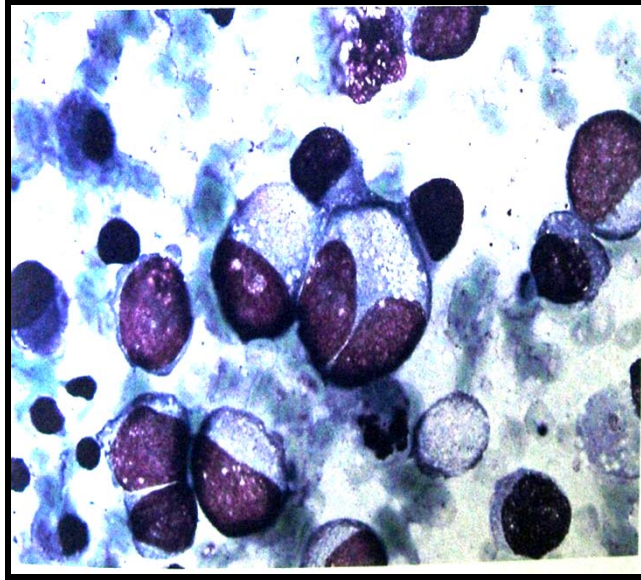


- Squamous cells with high nuclear to cytoplasmic ratio
- Dense blue cytoplasm



Neoplastic keratinized squamous cells with pyknotic nuclei and orangeophilic cytoplasm

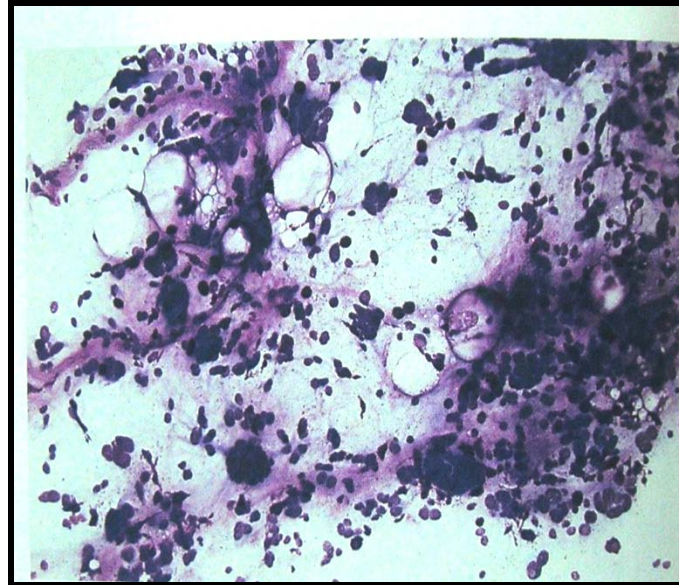
MALIGNANT MELANOMA



Malignant Melanoma

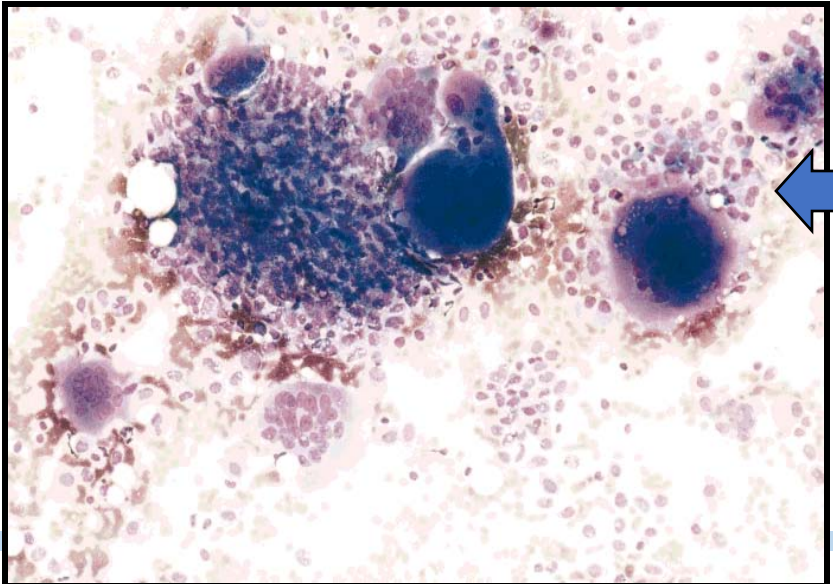
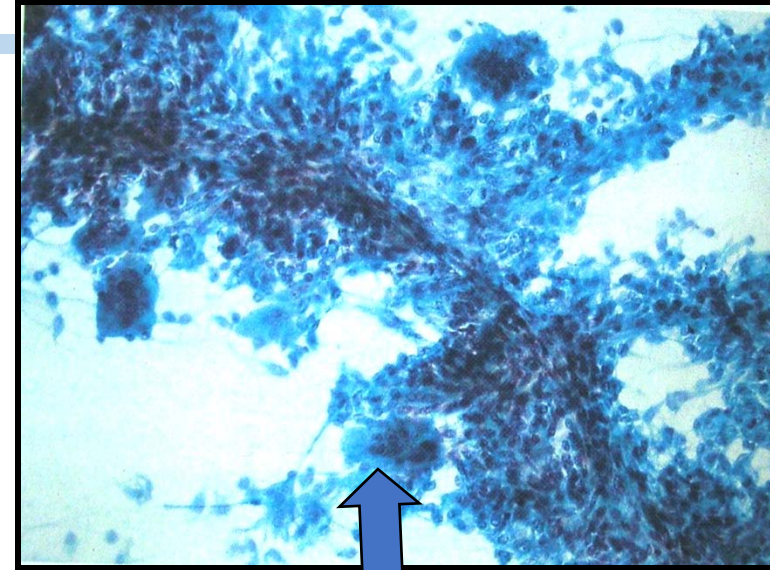
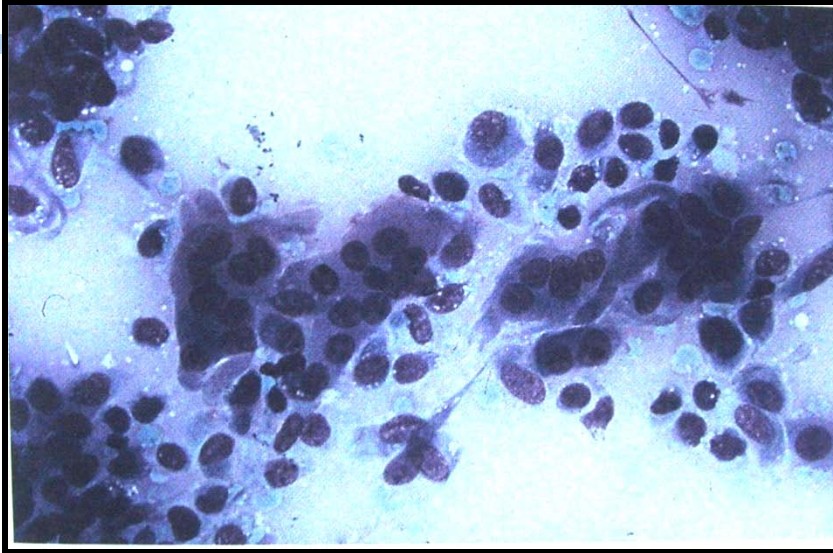
- discohesive often vacuolated, cytoplasm
- Mirror image nuclei
- Pigment laden macrophages

Lipoma



---Metachromatic stroma with clear adipose cells

GIANT CELL TUMOR



Cohesive fragments of spindle cells and scattered multinucleated giant cells

M

C

C

C

C

C

C

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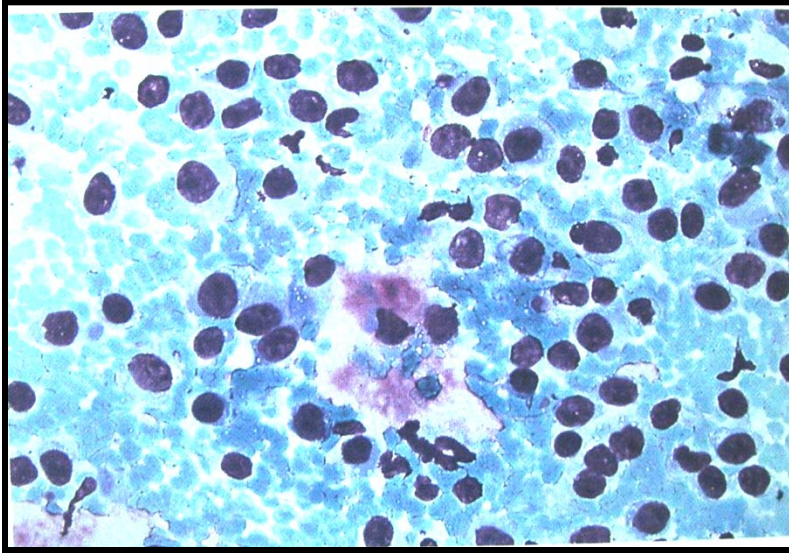
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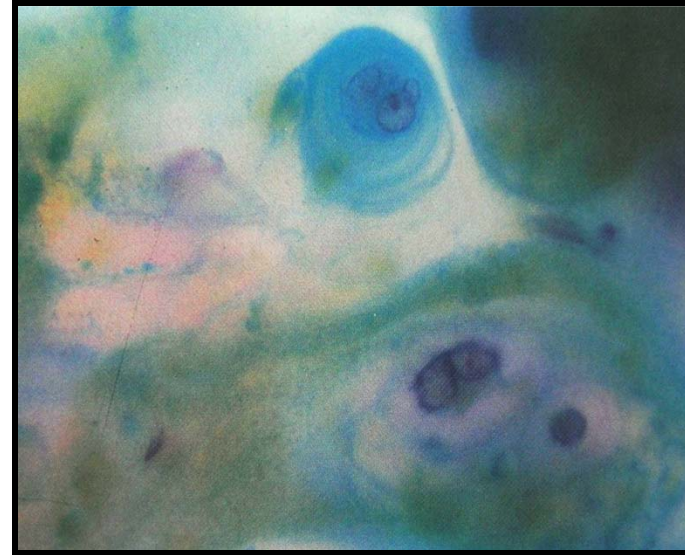
Giemsa stain,)

OSTEOSARCOMA



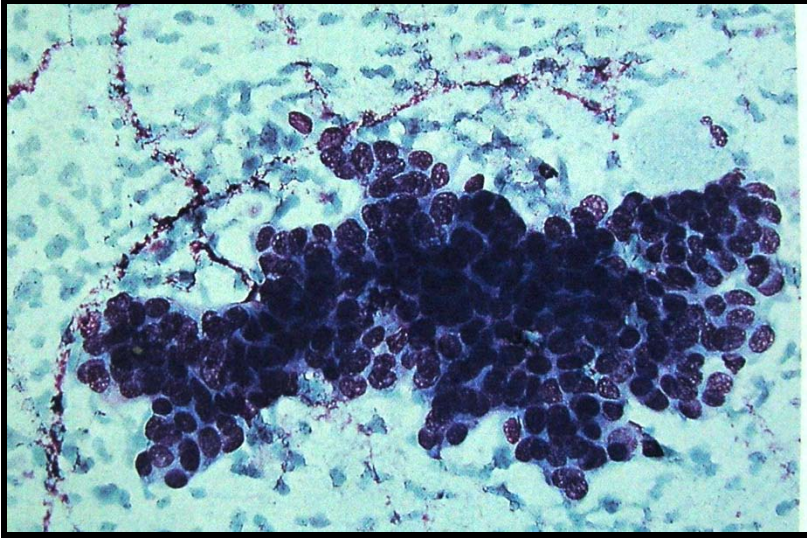
---loose cluster of pleomorphic, often multinucleate cells; the fragments of pink amorphous material may represent osteoid

CHONDROSARCOMA



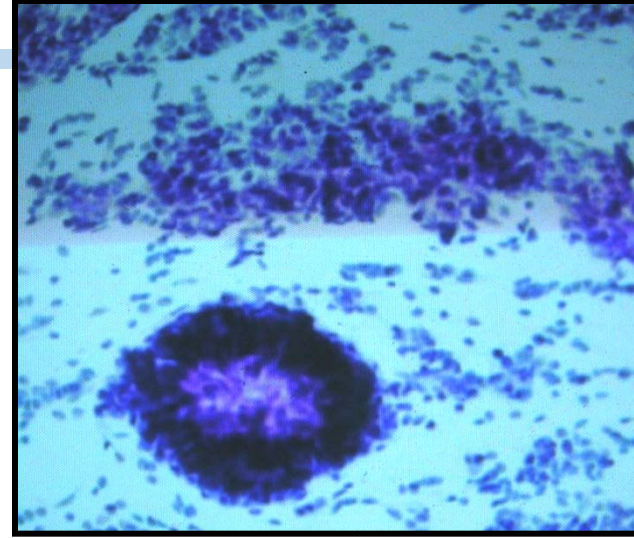
---Rounded cells outlined by thick cytoplasmic and stromal interface
--Binucleate chondrocyte

AMEOBLASTIC CARCINOMA



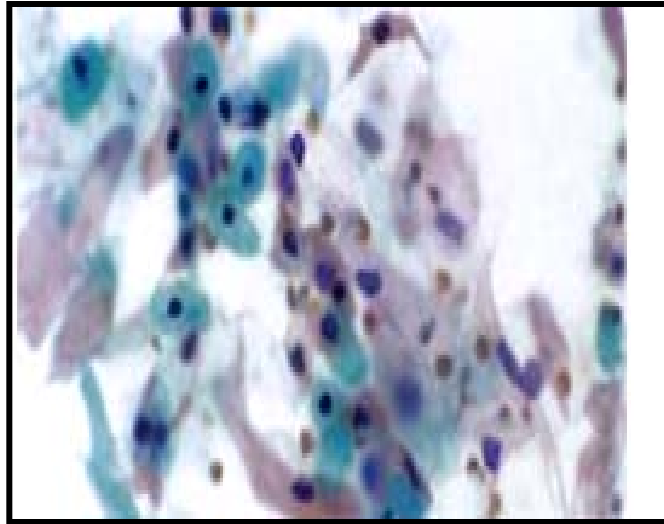
---Islands of hyperchromatic cohesive atypical epithelioid cells resembling ameloblasts are seen in the background of metachromatic stroma
--Absence of stellate reticulum

Ameloblastoma



---Ameloblastoma: cohesive clusters of cells surrounded by palisading columnar cells will be seen.
--Presence of stellate reticulum

Odontogenic keratocyst



---keratinized squamous cells
showing flakes of keratin

Tumours of smooth muscle- benign and malignant

- ❖ Very variable degree of cellularity
- ❖ Cells more often in cohesive clusters of fascicular fragments than single
- ❖ Cells in parallel bundles
- ❖ Abundant eosinophilic cytoplasm best visualised in single cells; often distinct cell borders
- ❖ Variable predominantly cigar shaped, sometimes truncated nuclei; intranuclear vacuoles (pseudonucleoli)
- ❖ Finely granular barred chromatin
- ❖ One or more small, distinct nucleoli
- ❖ Many multinucleated giant tumour cells in high grade leiomyosarcoma

Leiomyoma and Leiomyosarcoma

Tumors of striated muscles

- ❖ Cells with eosinophilic cytoplasm and an eccentric nucleus, triangular, strap shaped or tadpole-like may be recognized as rhabdomyoblasts
- ❖ Sometimes spindle-like with elongated nuclei and fusiform cytoplasm are seen

Rhabdomyosarcoma

- ❖ Loose arrangement of small cells with irregular eccentric nuclei; few cells have abundant cytoplasm

Neurogenic tumours

- ❖ The needling of neurofibroma or of a neurilemmoma may trigger a sharp pain radiating along the nerve and this a valuable diagnostic sign
- ❖ The amount of material obtained varies from case to case
- ❖ If Antony A areas are sampled, tissue fragments of cohesive cells are more commonly seen than single cells
- ❖ Samples from Antony B areas often show mainly dispersed cells and a myxoid background in fragments

Neurilemmoma

- ❖ The most typical feature is the **fibrillar appearance** of the background in fragments
- ❖ Nuclear palisading may occur
- ❖ Nuclei tend to be long and slender with pointed ends
- ❖ There is often a moderate degree of pleomorphism, but the chromatin pattern is uniformly bland

Granular cell tumor

- ❖ It is mainly found in the subcutaneous and submucosal tissues most commonly in the head and neck region
- ❖ FNB smears show many neoplastic cells in syncytial clusters and single
- ❖ Stripped nuclei are commonly seen
- ❖ The cytoplasm of intact cells is abundant and relatively dense, with a prominent granularity, eosinophilic in H & E and dark blue in MGG
- ❖ The nuclei are predominantly small, round or ovoid, uniform and bland chromatin and small but prominent nucleoli

BENEFITS

- ❖ A fine needle biopsy is a quick and effective test for determining the status of suspect tissue.
- ❖ Compared to a surgical biopsy, fine needle aspiration biopsy has low possibility of scarring, infection or pain, and has a significantly shorter recovery time.
- ❖ It is also extremely useful in the diagnosis and treatment of cysts.



COMPLICATIONS:

- ❖ Seen mainly due to damage of normal structures eg., bleeding because of damage to blood vessels, rupture of the limiting capsules.
- ❖ Infection of the skin wound.
- ❖ Dissemination of malignancy along the needle track or via the damaged venous and lymphatic supply is thought to be an important complication.
- ❖ Because an FNA biopsy can only sample a small number of cells from a mass or lump, there is a risk that any abnormal cells may be missed and not detected

❖

- ❖ The tissue obtained may not be representative of the tumor mass.

- ❖ **Needle trauma**

- ❖ **GRANULATION TISSUE FORMATION**

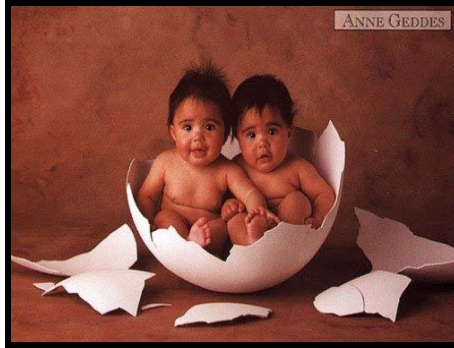
- ❖ **SARCOMA LIKE CHANGES**

- ❖ **NEEDLE LINEAR TRACT HEMORRHAGE**

- ❖ **TISSUE NECROSIS**

- ❖ **NEEDLE TRACK SEEDING**

- ❖ **Hematoma**



CONCLUSION

- ❑ IT is accepted that FNAC has reached high diagnostic accuracies of 90%.

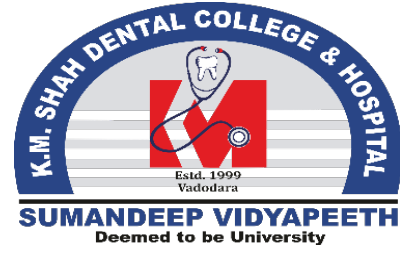
Increasing this accuracy requires more effort than has been made so far to bring this method to today's state of the art.

- ❑ Further prospective and retrospective studies are required to evaluate the validity of these proposals.
- ❑ It is apparent that cooperation between clinicians and cytologists contributes significantly to the understanding of each others efforts to improve diagnosis and treatment, with the final outcome being undoubtedly for the benefit of the patient.

REFERENCES



- ❑ Role of FNAC in the diagnosis of salivary gland swellings Kathmandu University Medical Journal (2008), Vol. 6, No. 2, Issue 22, 204-208
- ❑ Fine-Needle Aspiration Cytology in Tumors and Tumor-Like Conditions of the Oral and Maxillofacial Region. Cancer (Cancer Cyto pathol) 1997;81:238– 52.
- ❑ Manual and atlas of fine needle aspiration cytology. S. Orel ,G Sterrett.
- ❑ Oral Exfoliative Cytology by T.R. Saraswati et al
- ❑ GOOGLE IMAGE SEARCH



CONCLUSION

