

PATHOLOGY II PRACTICAL RECORDS

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Experiment No. 01

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Tissue Processing & Histology

1. Fixation and Fixative in histopathology section, the tissue are fixed before examination.

Aim:- To prevent the process of autolysis and attack of bacteria. Tissue which have been fixed do not change shape and volume during any subsequent procedure.

They should be in same condition which subsequently allow clear staining of the section. At time tissue should be closed to their leaving space ideally no small part should be lost.

Most important Section are those which stabilize the Profixative from by the cross links between the profixative forming gel. Ideally keeping everything in profixo reaction soluble and fixative to structural profixative and thus renders insoluble frame giving the whole structure some mechanical strength.

Fixation usually takes place in aqueous solution sometimes various other fixative are used those are 10% formaldehyde, 10% Normal saline, alcohol, Carnoy's Fixative, absolute alcohol, chloroform, glacial acetic acid, Zeller's Fluid (Fixed within 5 hrs.)

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1 mm slide is fixed with mercuric chloride containing fixative.

Advantage :- The tissue is left with advanced capacity of staining

Tissue Processing

definition :- It is the term used to refer to complete the reaction of tissue necessary to impregnate with solid medium to facilitate the reaction for processing.

Agents for impregnation are wax and resin impregnative material should be such that tissue can be easily stored for the section and can be easily produced on later days.

Labelling of the tissue :-

Lead pencil is used for labelling.

Embedding :- In this way gentle but complete dehydration of tissue to remove aqueous fluid and water.

Clearing with Substances :- which is totally reversible with both dehydrating and hydrating agents.

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Steps in automatic tissue processing :-

Jar Number	Reagents	duration
01	10% Formalin	12 hrs.
02	70% alcohol	2 hrs
03	85% alcohol	2 hrs
04	95% alcohol	2 hrs
05	absolute alcohol	2 hrs
06	absolute alcohol	2 hrs
07	absolute alcohol	2 hrs
08	Xylene	20 min
09	Xylene	20 min
10	Wax	2 hrs
11	Wax	2 hrs.

0.5 mm or 5 µ section is taken from the tissue.

Staining :- Various staining agents are used for staining the slide. The most common stain is used Hematoxylin and Eosin. Acidic component of tissue have affinity for basic dye and vice versa.

Result :- Nucleus and Mucin stain blue, Cytoplasm stain pink.

Special Stains :- For demonstration of muscle, Collagen, fibrin connective tissue

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Stain is used.

egs: von Zeimsa stain.

Masson's stain - Commonly used for staining Bone marrow giving a yellow colour.

Stain for elastic fibers:- 1. Virchow's stain
2. Orcein stain - black colour.

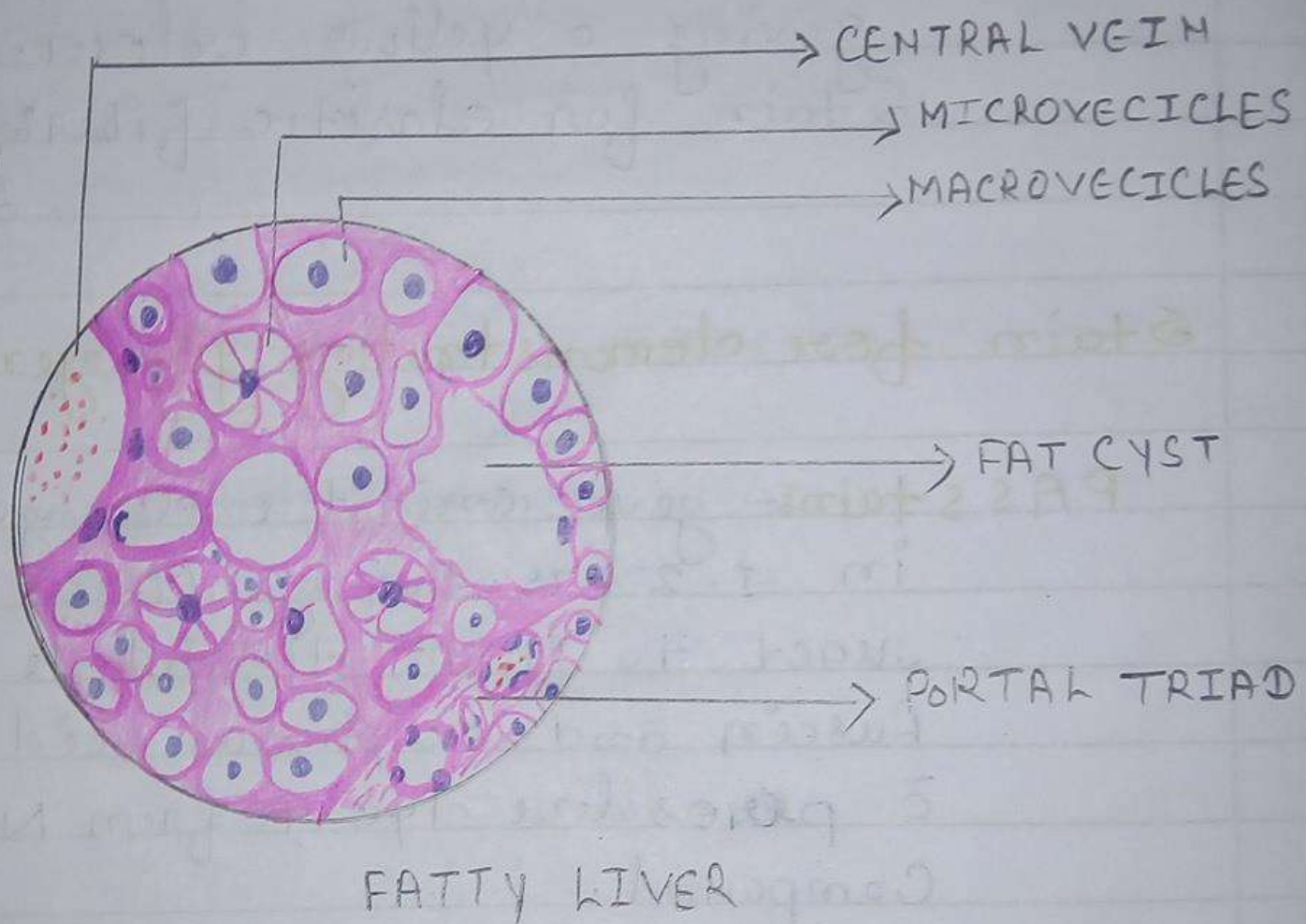
Stain for demonstrating glycogen, glycolipids mucins-

PAS stain:- gives oxidative cleavage of carbon bonds in 1-2 hrs. Glycogen/their anions/amino acid react to form dialdehyde. This reacts with fuchsin and sulphuric acid which combine to form magenta dye to form magenta coloured compound.

This stain is also used for fungus and large bacilli.

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Experiment No. 02

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Page No. 09Fatty Liver :-

Fatty changes most often seen in liver and appears as clear vacuoles in parenchymal cells. Intracellular accumulation of water / polysaccharide glycogen may produce clear vacuoles so the fat is demonstrated in frozen section and it stained with oil of Sudan black.

Gross Findings :-

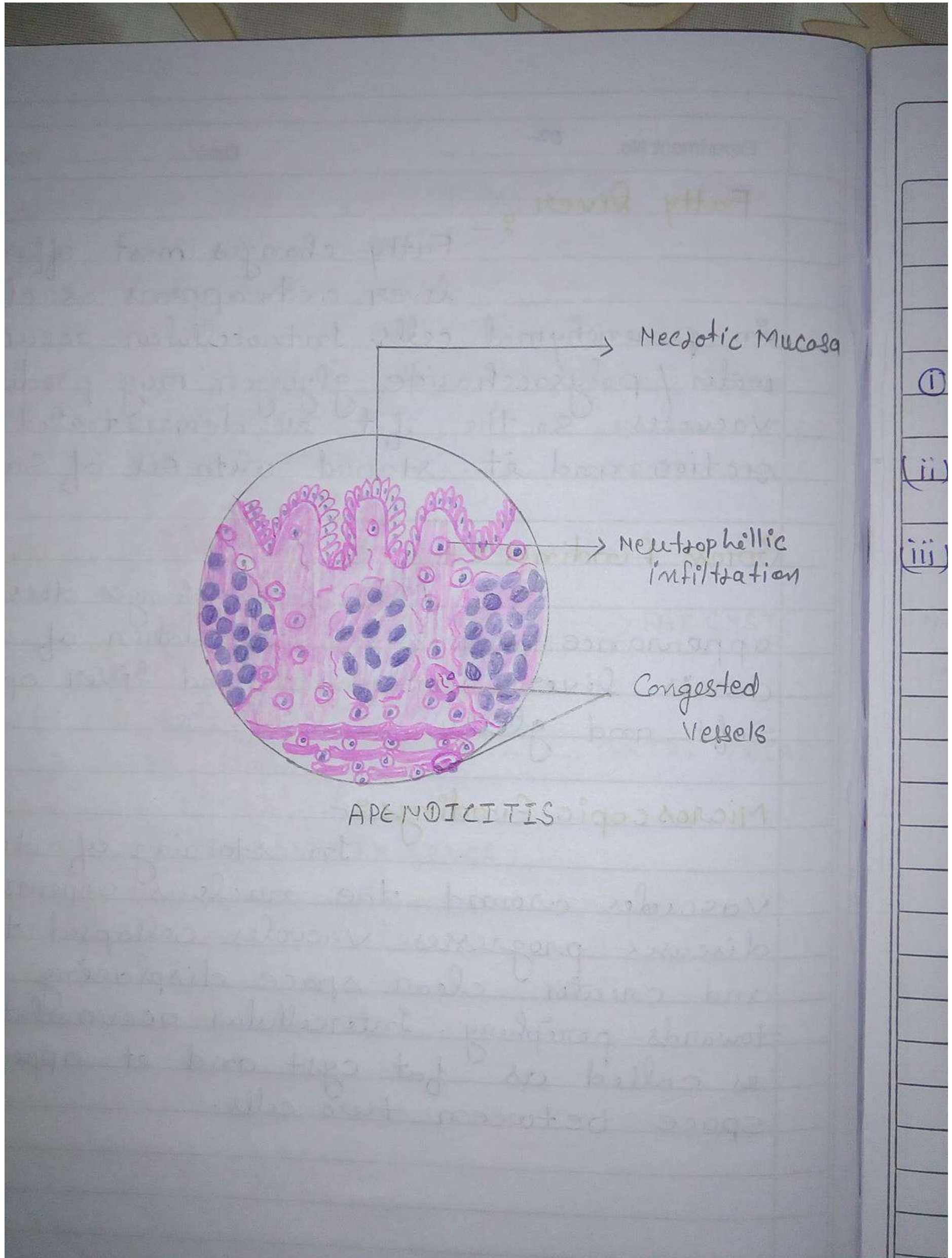
Mild fatty changes does not changes appearance progressive accumulation of fat cells causes liver enlargement and liver appears yellow, soft and glossy.

Microscopic Findings :-

In staining of diseases small vacuoles around the nucleus appears, as the diseases progresses vacuoles collapsed to each other and creates clear space displacing the nucleus towards periphery. Intercellular accumulation of fat is called as fat cyst and it appears as open space between two cells.

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Experiment No. 01

Date _____ Page No. 11

Acute appendicitis:-

It is immediate and early response to any injurious agent. It has 3 components:-

- (i) Vasoconstriction in vascular caliber leads to an increased blood flow.
- (ii) Structural microvascular disruption that permits the plasma protein to leave the circulation.
- (iii) Emigration of Leucocytes from microcirculation and accumulation at the foci of injury.

Gross Features:-

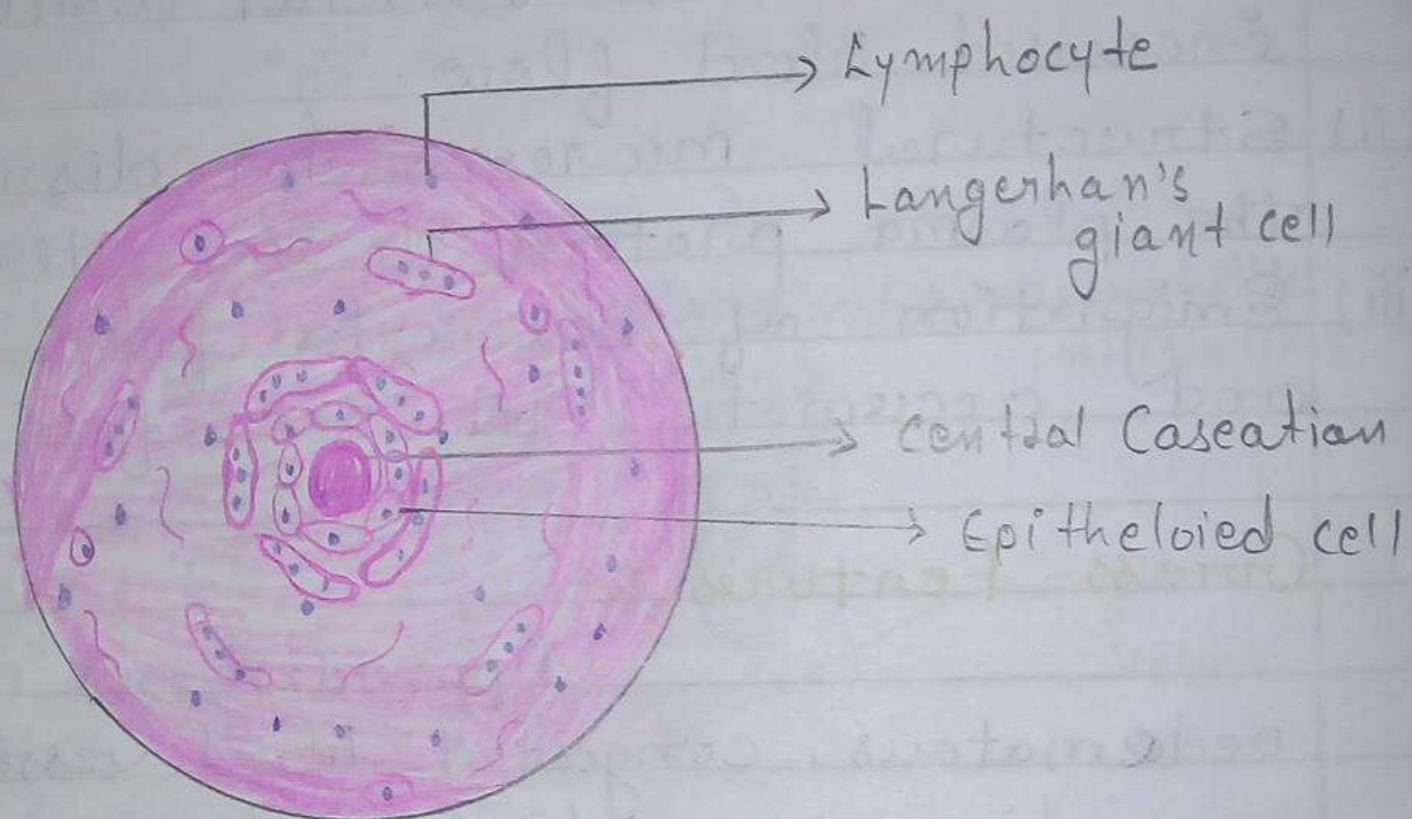
Appendix is swollen, serosa is oedematous, congested blood vessel, lumen of the appendix some tissue show exudate.

Microscopic Features:-

Acute inflammatory cell in all layers of appendix starting from serosa to mucosal layer. Dilated and congested blood vessels in mucosa.

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Tubercular Lymphadenitis

Experiment No. 04

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Tubercular lymphadenitis

It is distinct pattern of chronic inflammatory response in which predominant cell type is macrophage and lymphocyte with modified epithelial cell like appearance called as epithelioid cell. The epithelioid cell present as pale pink granular cytoplasm with indistinct cell boundary. They often appear as merged with each other appearing as multinucleated cell. These cells are surrounded by a collar of lymphocyte and plasma cells.

The fusion of epithelioid cell is known as giant cell / Langerhan's giant cell. They are mostly 40-50 μ in diameter. The giant cells are mostly arranged either peripherally or around the foreign body.

Gross features :-

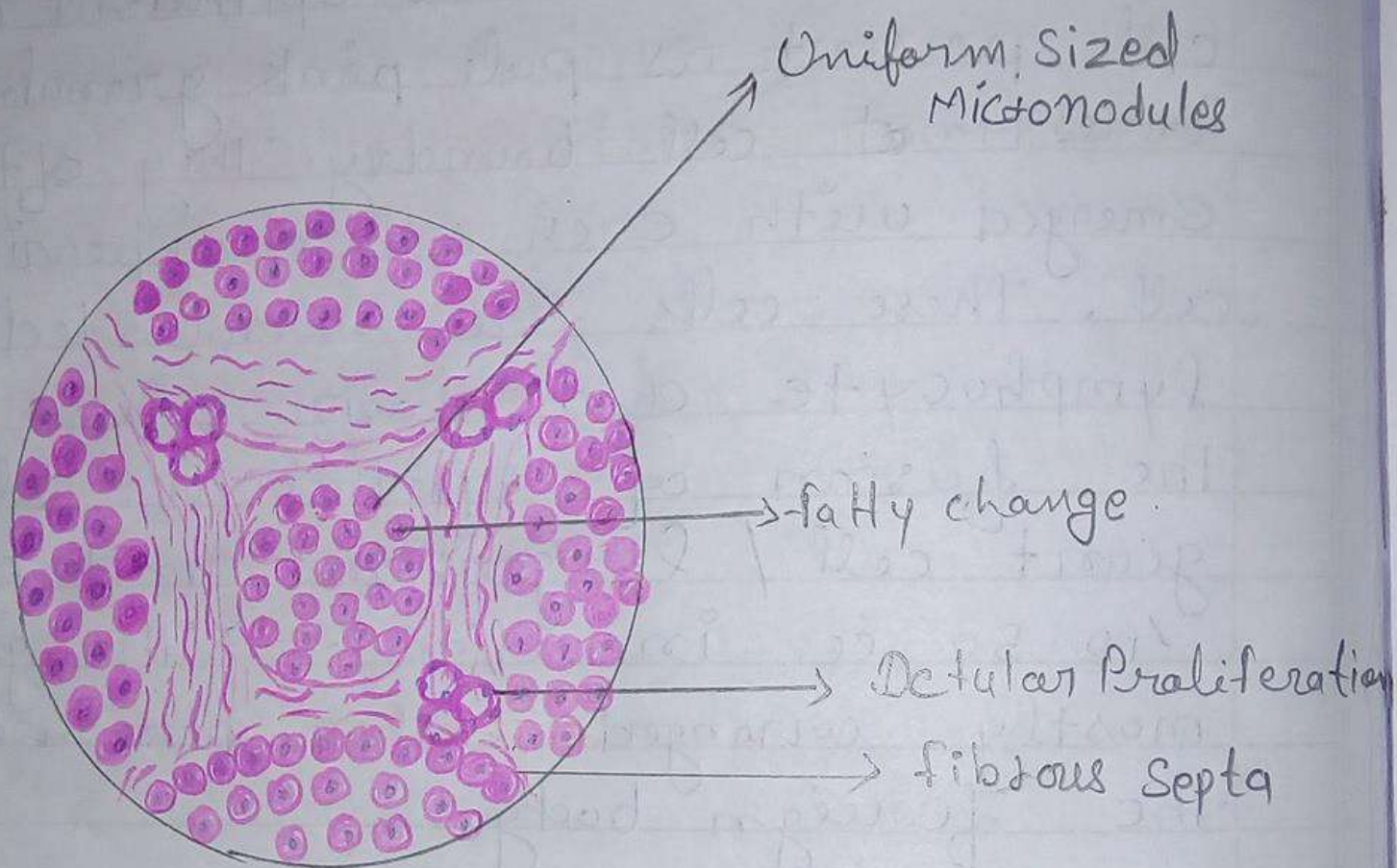
Lymph nodes are matted showing central necrosis and central caseation.

Microscopic features :-

Caseation in center and surround by epithelioid cell and lymphocyte along with Langerhan's giant cell.

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Experiment No. 05

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Cirrhosis of Liver

It is a diffuse disease having disorganised lobular architecture and formation of nodules which are separated from each other by irregular bands of fibrosis.

Gross appearance :-

Cirrhosis is

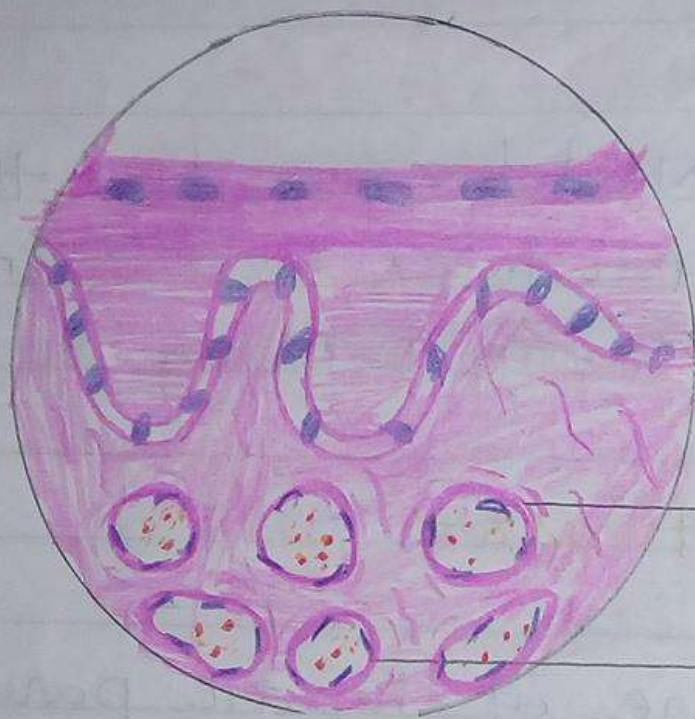
- (i) Micromodular - Nodules are less than 3mm.
- (ii) Macromodular - Nodules size $> 3\text{mm}$
- (iii) Mixed - Having both micro and macromodular

Microscopic features :-

- (i) Lobular architecture of hepatic parenchyma is lost and central veins are hard to find.
- (ii) Fibrous septa divide the hepatic parenchyma into nodules.
- (iii) Hepatocytes in surviving parenchyma form regenerative module having disorganised masses of hepatocyte.
- (iv) Fibrous septa contain mononuclear inflammatory cell infiltrate and proliferative bile ductles.

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plump endothelium

lobule capillaries

Capillary Haemangioma

Experiment No. 06

Date _____ Page No. 17

Capillary Haemangioma

It is a neoplasm characterised by ↑ no. of normal and abnormal vessels. This may be difficult to distinguish from malformation to haemangioma. Most commonly localised but may involve distal segments of extremities. Malignant transformation is rare :-

There are two types :-

- (i) Capillary haemangioma.
- (ii) Cavernous haemangioma.

Capillary haemangioma :-

Usually occurs in skin, subcutaneous tissue. They may also occur in viscera and mucous membrane of oral cavity. Strawberry type of haemangioma is extremely common.

Gross Features :-

It may vary few mm to several cms. Lesions are bright red to blue in colour, may be in level of skin or slightly elevated, usually having intact covering of epidermis.

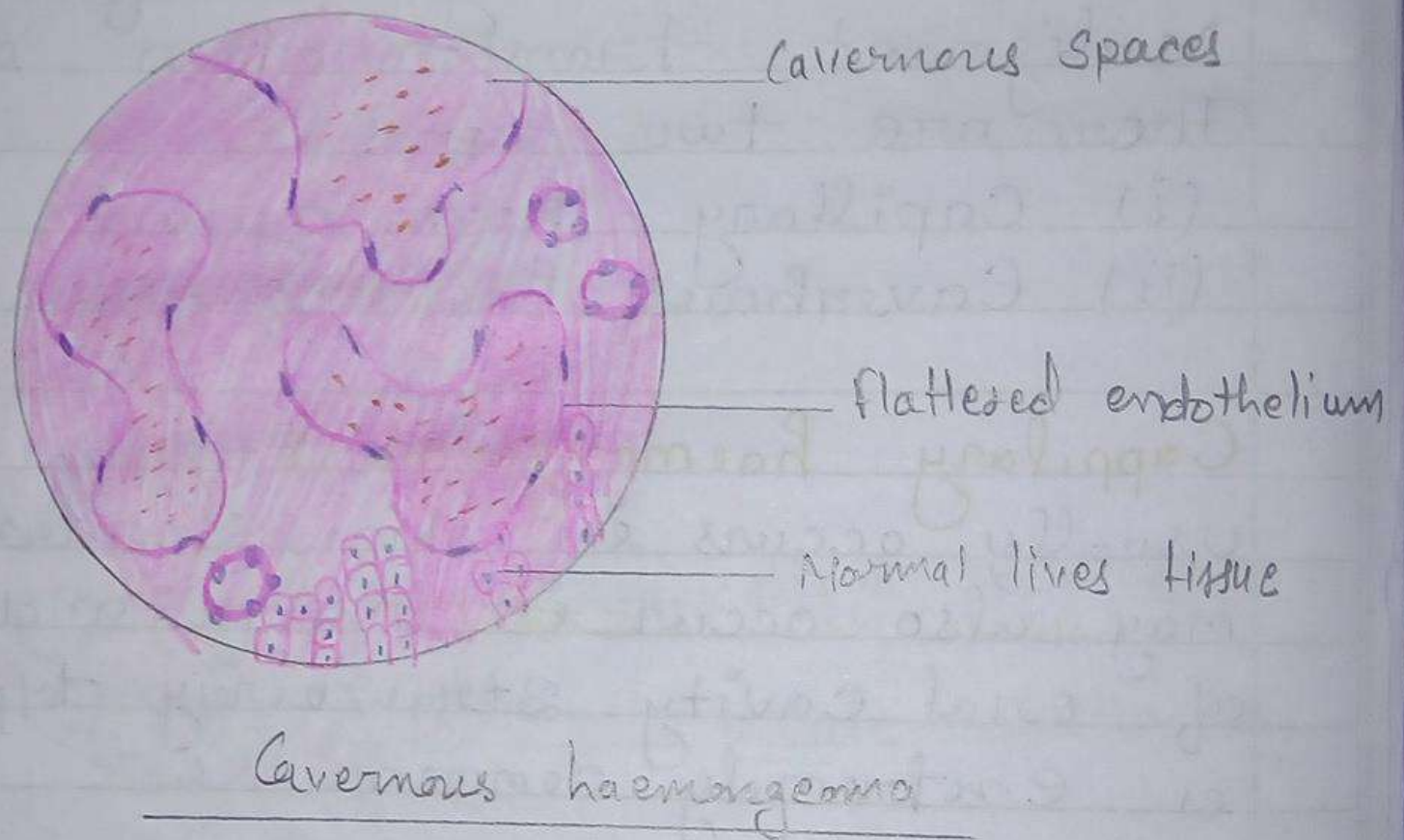
Microscopic Features :-

Usually lobulated but encapsulated haemangioma show aggregate of closely packed involved capillaries.

Usually blood is filled in endothelium and separated by scanty connective tissue stroma.

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Experiment No. 07

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Cavernous haemangioma :-

Less common than capillary haemangioma.
distinguished by formation of vast vascular channels usually large and less circumscribed.
More frequently involves deep structures than capillary haemangioma.

Gross Features :-

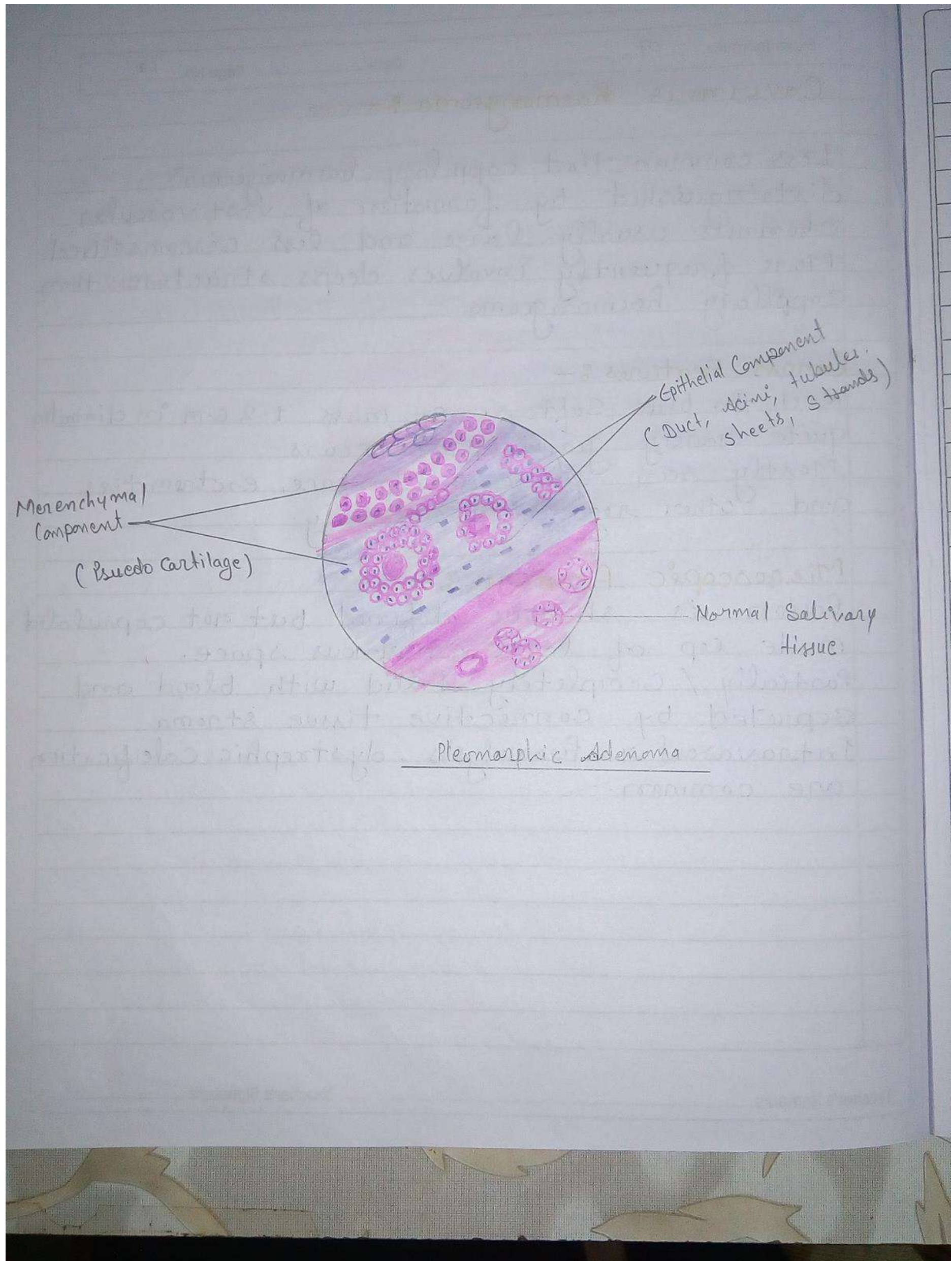
Red to blue soft spongy mass, 1-2 cm in diameter
quite rarely giant form occurs.
Mostly seen in areas of face, extremities
and other regions of the body.

Microscopic Features :-

lesion is sharply defined but not capsulated
made up of large cavernous space.
Partially / Completely Filled with blood and
separated by connective tissue stroma.
Intravascular haemolysis, dystrophic calcification
are common.

Teacher's Remarks

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Experiment No.

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Pleomorphic Adenoma

This is the commonest tumour of salivary gland.

Gross

Tumour is generally circumscribed Pseudo in Capsulated, Rounded, occasionally multilobulated with variegated appearance.

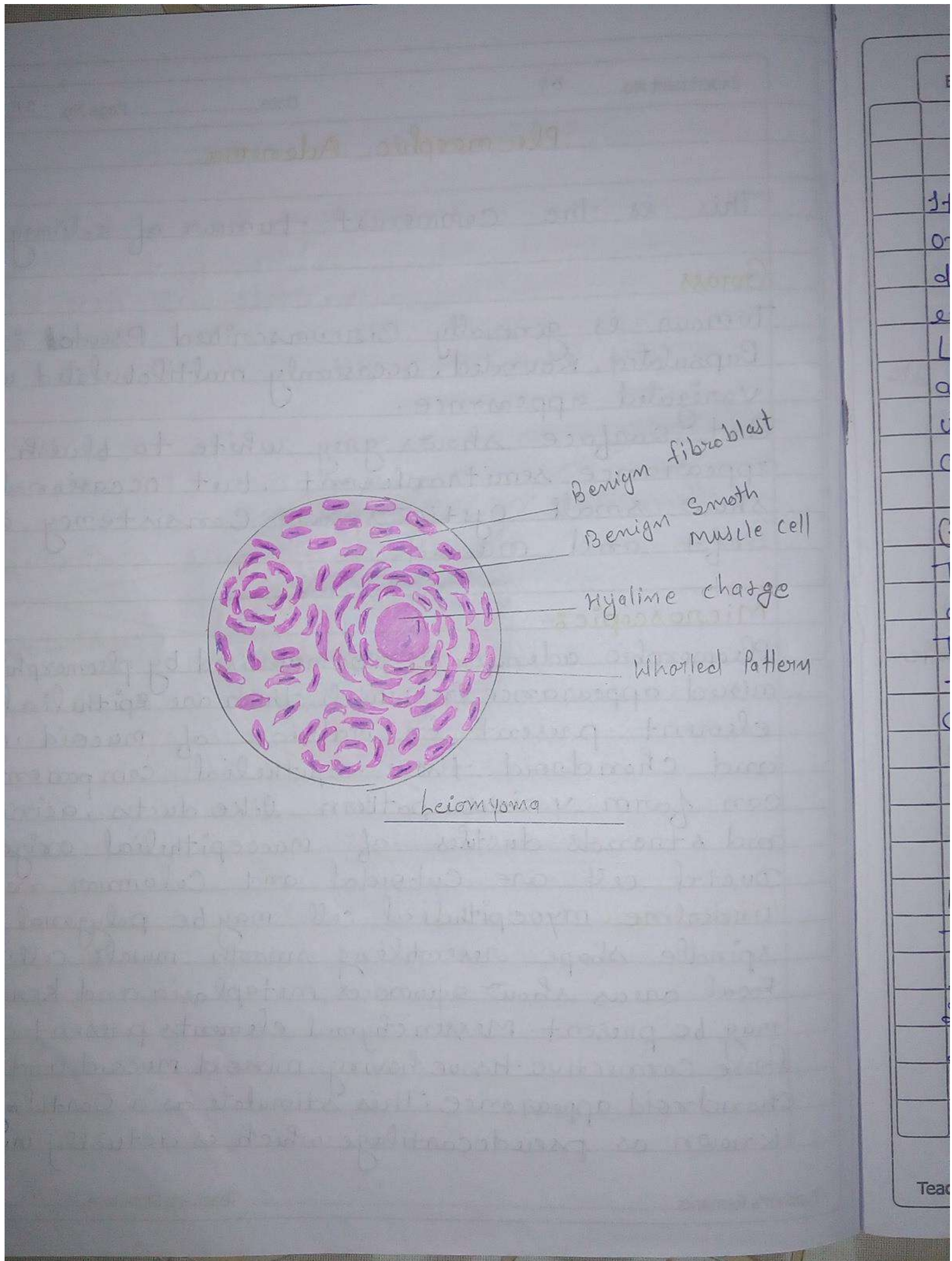
Cut surface shows grey white to bluish variegated appearance semitranslucent, but occasionally show small cystic spaces Consistency is soft and mucoid.

Microscopic:-

Pleomorphic adenoma is characterised by pleomorphic or mixed appearance in which there are epithelial element present in matrix of mucoid, myxoid and chondroid tissue. Epithelial components can form various pattern like ducts, acini, sheets and strands ductules of mucoepithelial origin. Ductal cell are cuboidal and columnar and underlying myoepithelial cell may be polygonal or spindle shape resembling smooth muscle cell. Focal areas show squamous metaplasia and keratinisation may be present. Mesenchymal elements present as loose connective tissue having myxoid, mucoid and chondroid appearance. This simulate as a cartilage also known as pseudocartilage which is actually mucin.

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Experiment No. 09

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Leiomyoma

It is smooth muscle tumour, benign in nature mostly originating in uterus. Appears involve of blood vessels deep soft tissue, Subcutaneous tissue, skin, erector pili muscle and scrotum.

Leiomyoma of erector pili muscle a multiple in no and painful.

uterine leiomyoma is also called as fibroid. most commonly found in women of reproductive age group.

Gross

They are well circumscribed, discrete, round, firm and white in colour.

Tumour may vary from small to large visible nodule to massive tumour that fills the pelvic cavity.

They occur in 3 forms.

Intramural, submucosal and subserosal.

Cut section show characteristic whorl pattern.

Microscopic Feature:-

They essentially composed of 2 tissue elements:

1. whorled bundles of smooth muscle cells.

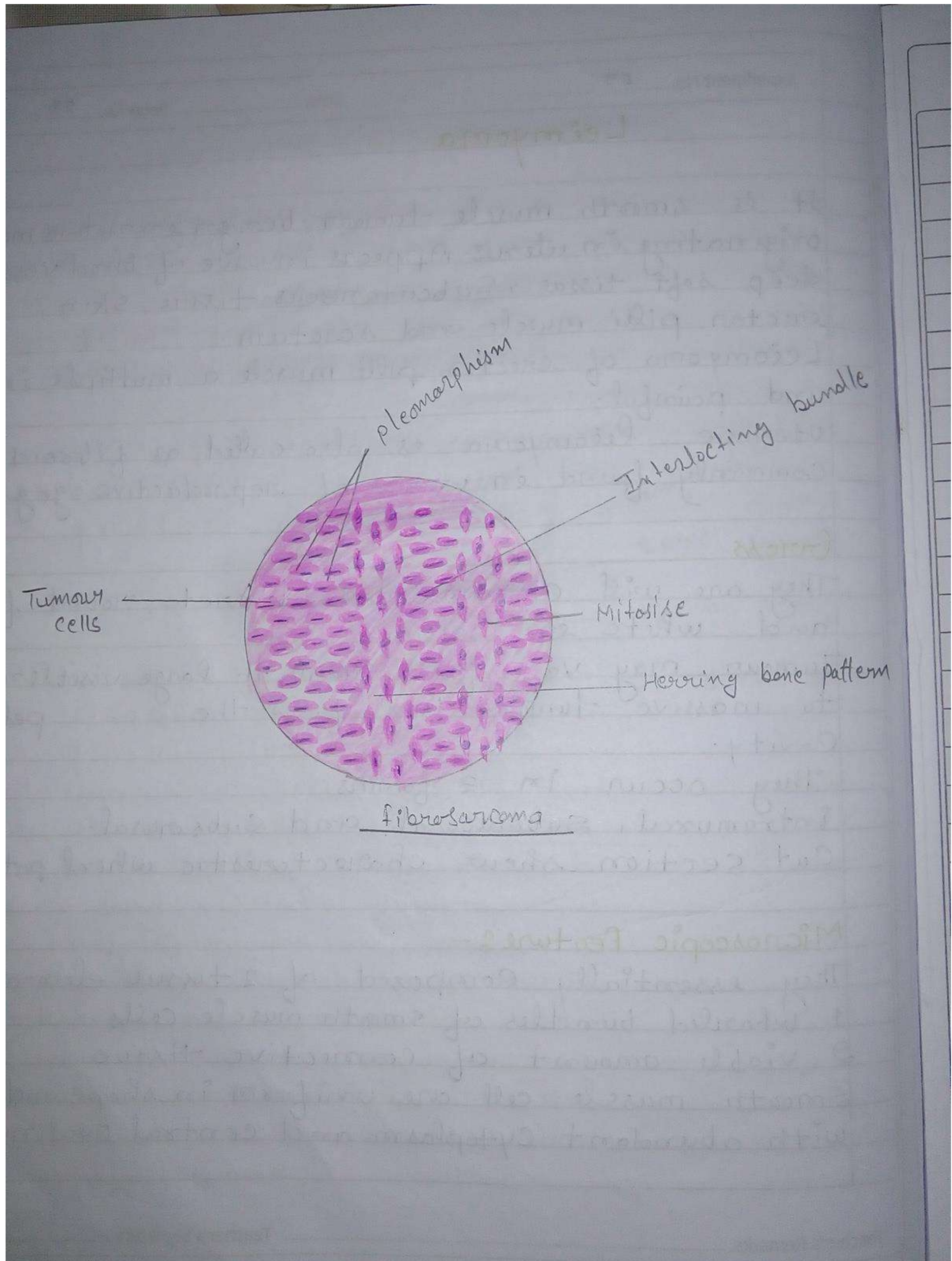
2. viable amount of connective tissue

Smooth muscle cell are uniform in shape and size with abundant cytoplasm and central oval nuclei.

Teacher's Remarks

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Experiment No. 10

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Fibrosarcoma

It is slow growing tumour, affective the adults b/w 4th to 7th decade of life.

Most common location are lower extremities sp. thigh and around the knee, upper extremities trunk, head and neck and retroperitoneum.

The tumour is capable of metastasis chiefly via the blood stream.

Gross :-

Grey-white, firm lobulated and characteristically circumscribed mass. Cut surface shows soft having fish flash like appearance with foci of necrosis and hemorrhage.

Microscopic :-

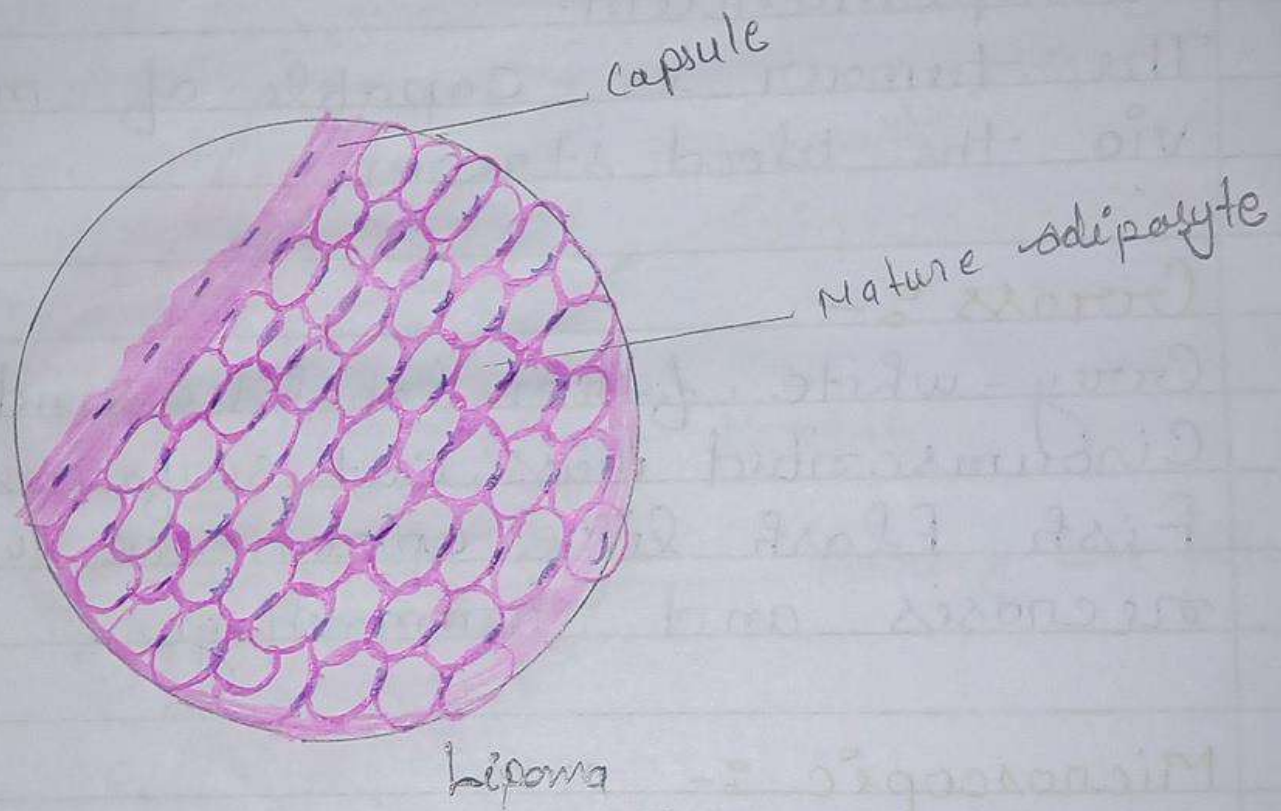
Tumour is composed of uniform spindle shape fibroblast arranged in intersecting fascicles.

Well differentiation tumour shows a herring bone pattern of fibroblast.

Poorly diff. fibrosarcoma has high pleomorphic appearance with frequent mitosis and bizarre cells.

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Experiment No. II

Date

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Lipoma

Most common benign soft tissue mass caused due to well defined proliferation of fat cells.

Gross :-

Yellowish, greasy, lobulated, well circumscribed, round to oval in shape, smooth and delicately encapsulated.

Microscopic

Normal fat cells with lobules of mature adipose cells, separated by thin delicate fibrous septa.

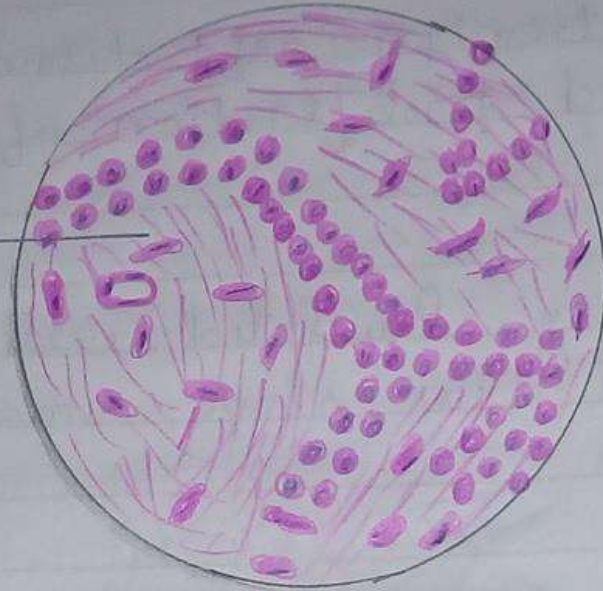
Fat cells are mostly seen as round cells.

Teacher's Remarks

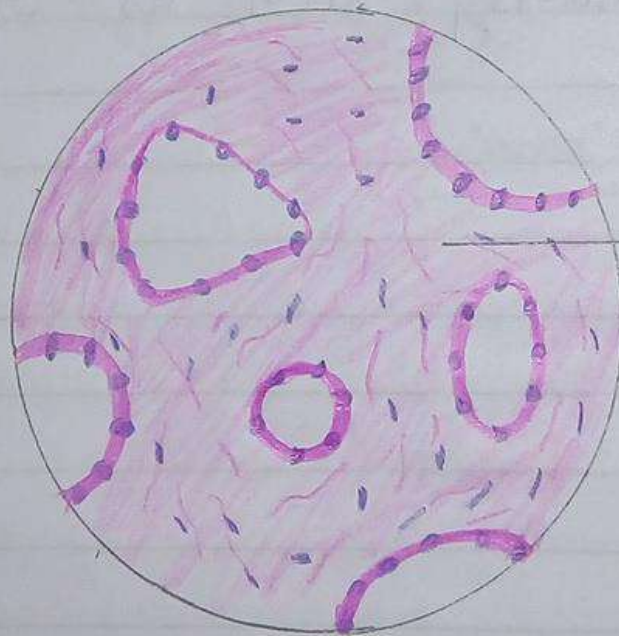
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Collagenic
stroma



Intra can



Collagenic
stroma

Pericanalicular pattern

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Fibroadenoma

It is a benign tumour of fibrous and epithelial elements of breast. Most common benign tumour of breast in reproductive life.

Gross :-

- It is small, solitary, well encapsulated, spherical or discoid mass.
- Cut surface firm, grey white, slightly myxoid and shows slit like spaces.

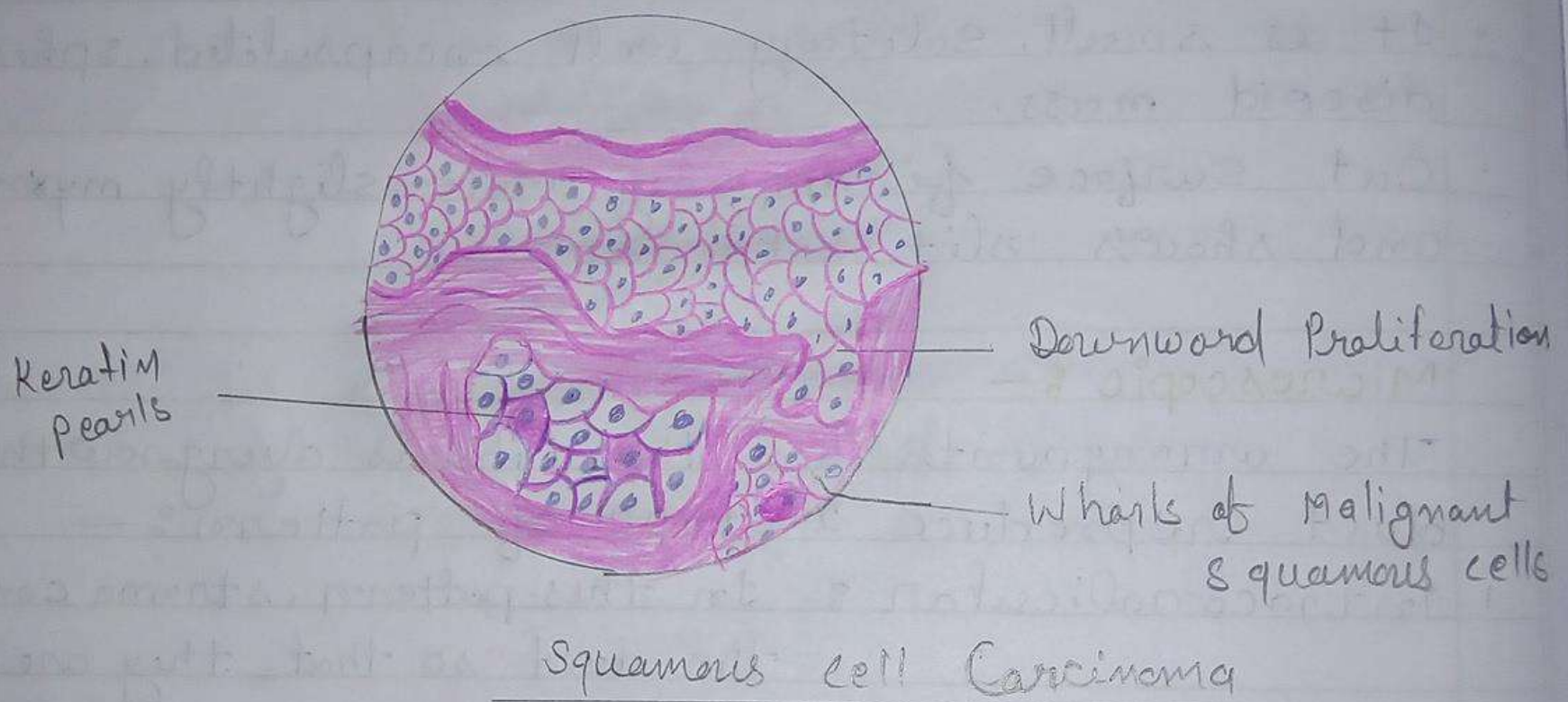
Microscopic :-

The arrangements b/w the fibrous overgrowth and duct reproduce 2 type of pattern :-

1. Intracanalicular :- In this pattern, stroma compresses the duct so that they are reduced to slit like spaces lined by ductal epithelium may appear as cord of epithelial elements surrounding the mass of fibrous tissue.
2. Pericanalicular :- This pattern is characterised by encircling mass of fibrous tissue around patent or dilated duct.

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Experiment No. 17

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Squamous Cell Carcinoma

It occurs most common in skin, oral cavity, oesophagus uterine cervix and at the edges of chronic ulcer.

Gross

Tumour is either in form of lobular and ulcerative growth with fungation or polyploid mass without ulceration.

The margin of growth is elevated and indurated cut section shows grey white endophytic as well as exophytic tumour.

Microscopic :-

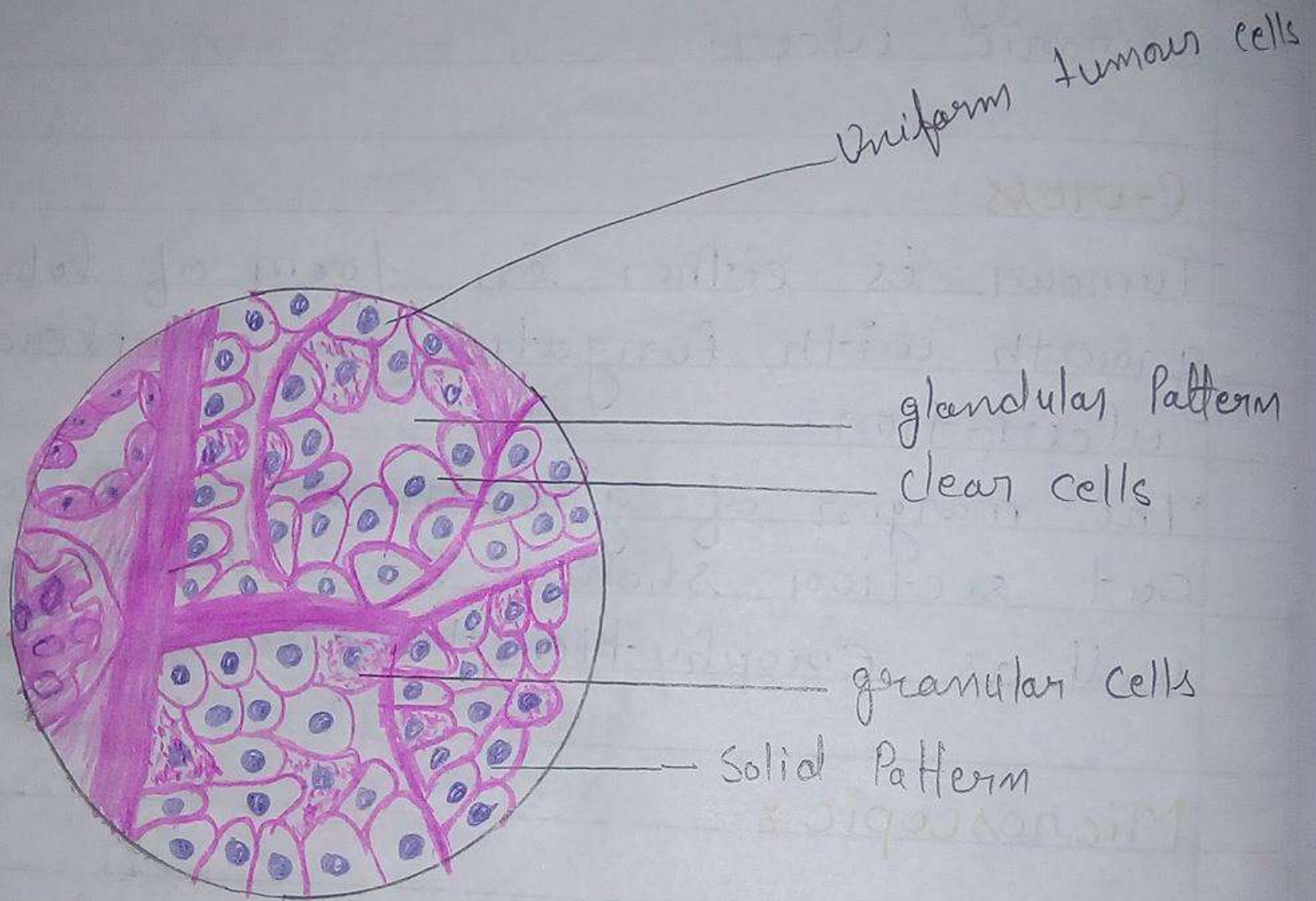
Tumour is characterised by malignant cell which may show variable degree of differentiation.

The mass of tumour cell invade through the B.Mo into the dermis.

In differentiated tumour, the cell are arranged in connective tissue layer called "Epithelial pearls" and contain keratin material in the center of the cell mass. The mass of tumour cell are separated by lymphocytes.

Teacher's Remarks.....

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Renal cell Carcinoma

Experiment No. 14

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Renal cell Carcinoma / Adenocarcinoma / Hypernephroma of kidney

It consists about 70-80% of all renal cancer and occurs mostly at the age of 50-70 years.

Gross

Renal cell carcinoma mostly arises as a tumour from a pole of kidney, mostly upper pole of kidney as a solitary and unilateral tumour.

The tumour appears golden yellow and well circumscribed. Cut section of tumour shows large areas of ischaemic necrosis, cystic changes and foci of haemorrhage. Another feature frequently present is RCC is tumour thrombus of renal vein.

Microscopic :-

A variety of patterns of tumour cells are seen such as solid, acinar, tubular, trabecular, cord and papillary arrangements.

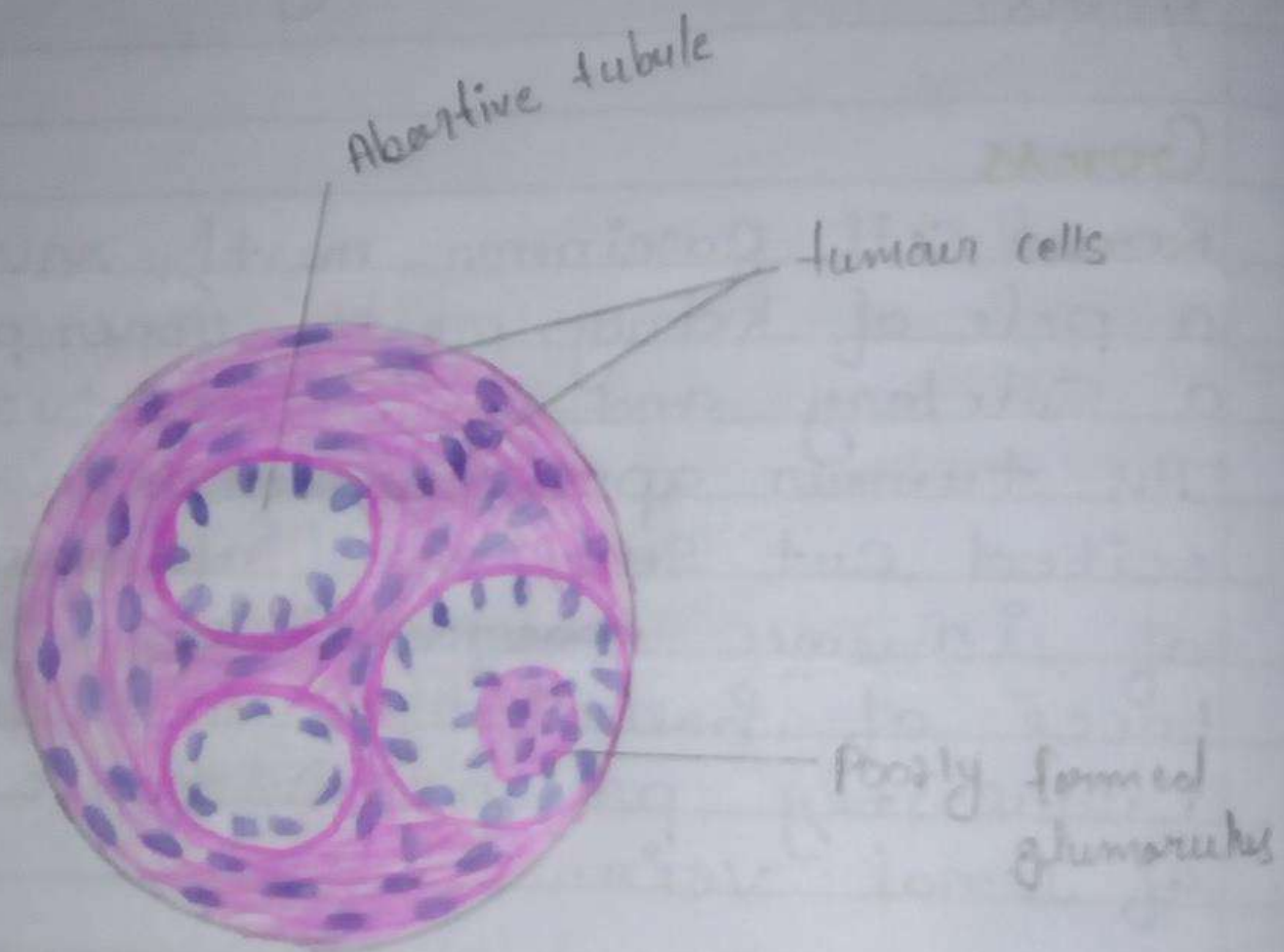
Tumour cells generally consist of 2 types :-

- (i) clear cells :- comprising 70% of Renal cell carcinoma. These are large cells and have well defined borders with abundant clear cytoplasm and regular pyknotic nuclei.
- (ii) granular cells :- These are also type large cells having well defined borders with moderate amount of pink granular cytoplasm.

Teacher's Remarks

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Wilms' tumor

Experiment No. 15

Date _____ Page No. 35

Wilms' tumour

It is an embryogenic tumour commonly seen in children of 1-6 years of age.

The tumour is large, spheroidal and replaces most of the kidney.

Gross feature :-

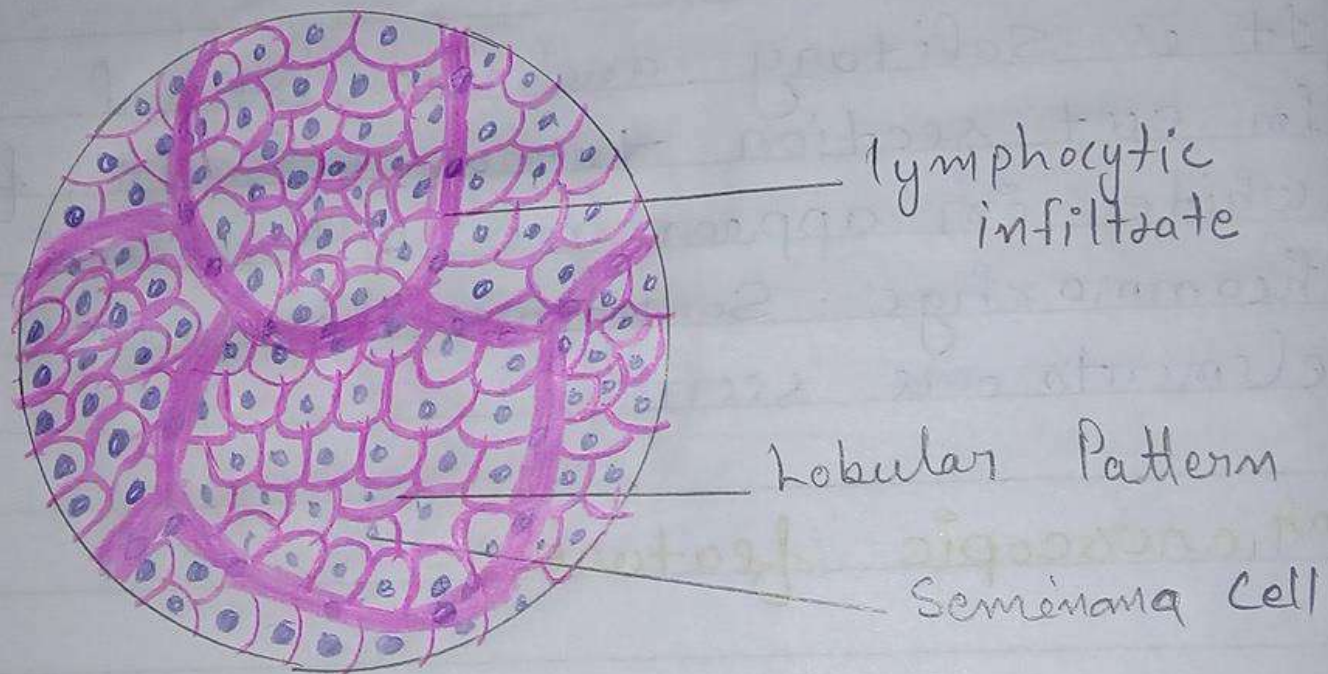
It is solitary and unilateral. In cut section, tumour is soft, fish flesh, grey white in appearance with foci of necrosis and haemorrhage. Sometimes myxomatous and cartilaginous elements are seen.

Microscopic feature :-

Tumour shows primitive epithelial and mesenchymal elements. Tumour largely consists of small, round to spindle, anaplastic, sarcomatoid tumour cells. Aborting tubules and poorly formed glomeruli are seen. Sometimes mesenchymal elements such as smooth muscle, cartilage and bone also seen.

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Seminoma

Experiment No. 16

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Seminoma

It is the most common malignant tumour of testis comprising 45% of all testicular germ cell tumours. This corresponds to dysgerminoma of female genital organ.

Gross :-

Involved testis enlarge upto 10 times. But it tends to maintain the normal colour of the testis as the tumour rarely invades the tunica.

Cut section shows homogenous, greyish white lobular appearance.

Microscopic :-

The Seminoma cells generally lying in cords, sheets and columns forming lobules.

The tumour cells are uniform in size with clear cytoplasm and well defined cell border.

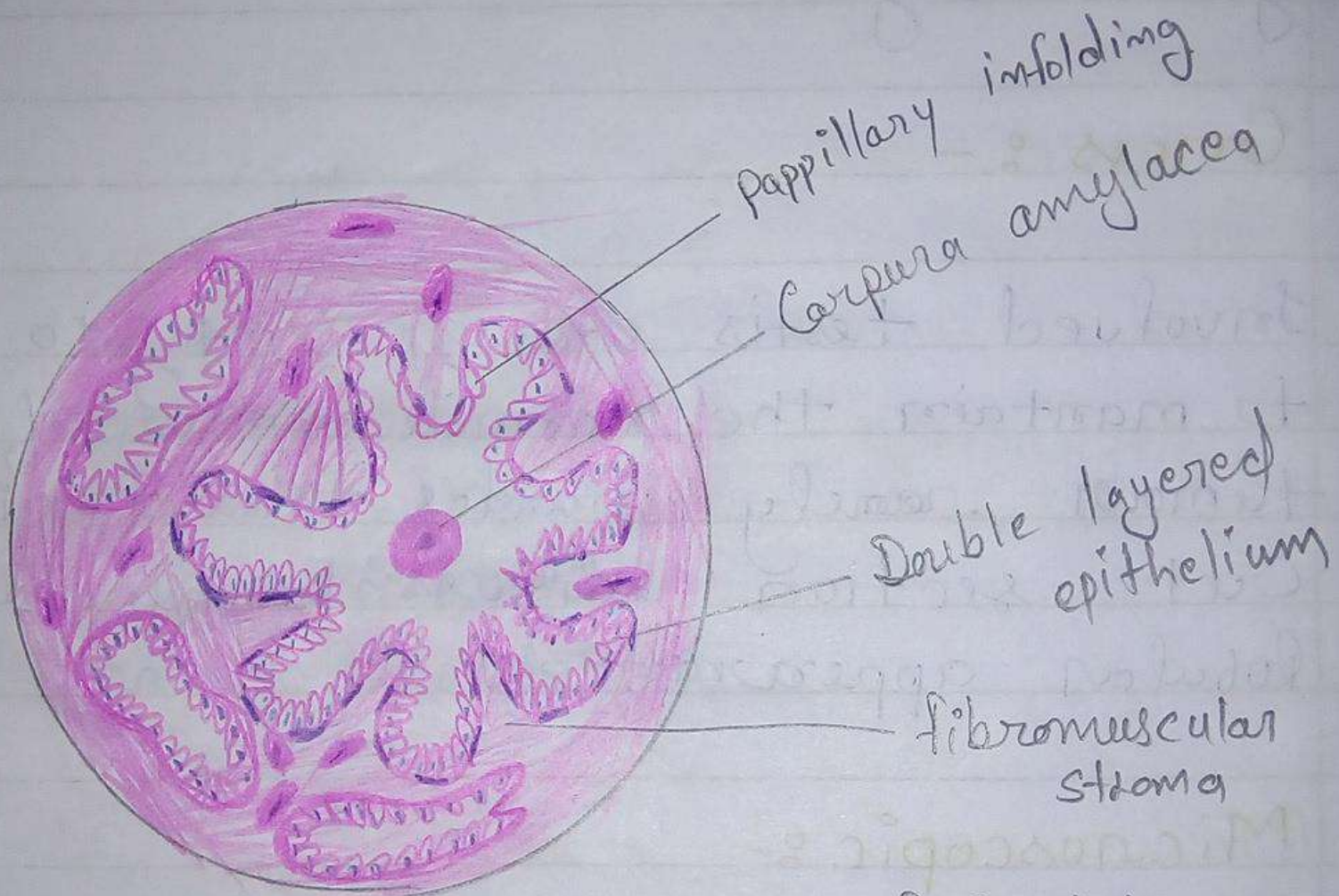
The nuclei are central, large, hypochromatic and contained 1-2 prominent nucleoli.

Delicate fibrous tissue which divide the tumour in lobules are called as stroma.

The lob tumour shows characteristic infiltration.

Teacher's Remarks _____

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Benign hypertrophy of Prostate

Experiment No. 17

Date

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Benign hypertrophy of Prostate

It is the enlargement of prostate in male, frequently above the age of 50.

Gross

The prostate appears nodular, smooth and raised 2-3 times the normal prostate size.

Normal prostate wt. 20 gm.

Cut section shows nodularity having varying admixture of yellowish, pink, soft honeycomb appearance and firm homogenous appearance.

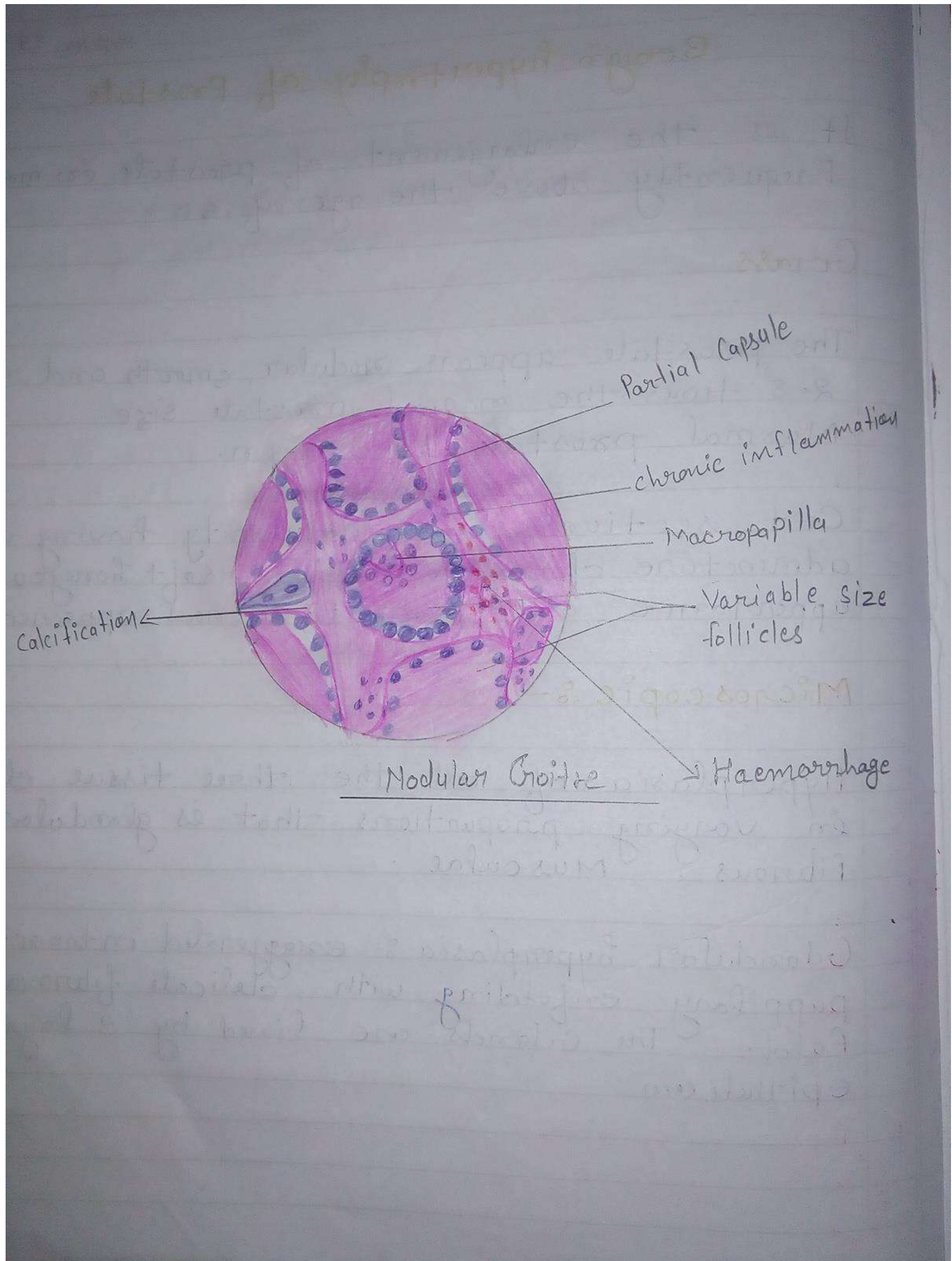
Microscopic :-

Hyperplasia of all the three tissue elements in varying proportions that is glandular, fibrous, muscular.

Glandular hyperplasia :- exaggerated intracinar, papillary enfolding with delicate fibrovascular fold. The glands are lined by 2 layers of epithelium.

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Experiment No. 18

Date

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Nodular Goitre

It is a end stage of long standing simple goitre characterised by tumour like enlargement of thyroid and having characteristic nodularity.

Gross features:-

Thyroid shows asymmetric and extreme enlargement wt. up to 100-500 gms.

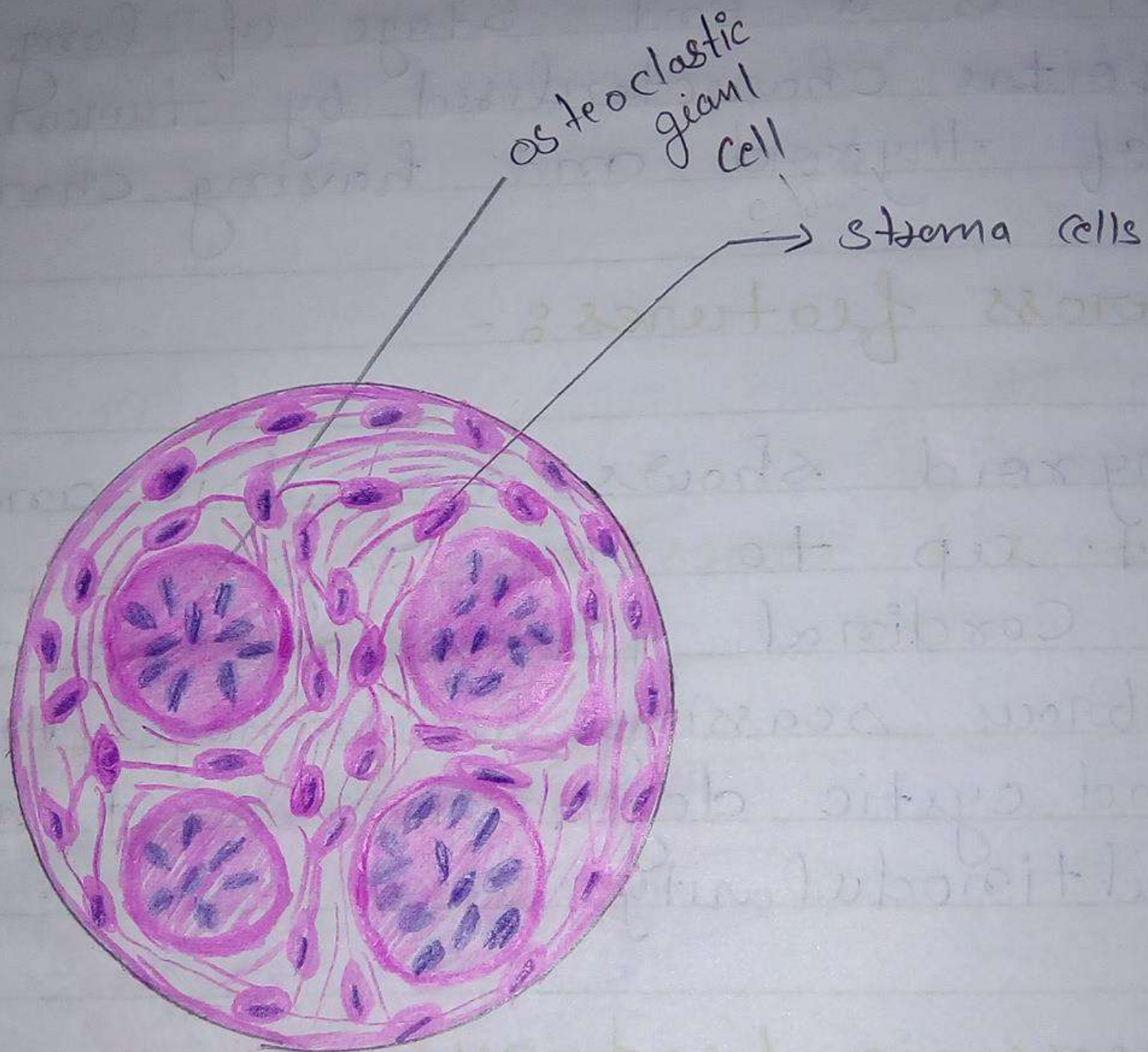
5 Cardinal gross features are - Nodularity, Fibrous scarring, Hemorrhage, focal calcification and cystic degeneration. Cut surface shows multinodularity.

Microscopic features:-

It shows follicles of varying size from small to large lined by flat epithelium and filled with colloid. Area of hemorrhage, fibrous scarring, calcification and cystic degeneration may be seen.

Teacher's Remarks

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Osteoclastoma

Experiment No. 19

Date

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Osteoclastoma / Giant cell tumour

It is a tumour arising from epiphysis of long bones. more common in the age groups of 20-40 years. Commonest site of involvement are lower end of femur, upper end of tibia, lower end of radius and upper end of fibula.

Gross features:-

Osteoclastoma is eccentrically located in epiphyseal end of long bone which expands in time. The tumour is well circumscribed, dark tan in colour and covered by thin layer of periosteal bone. Surface shows honeycomb appearance & area of hemorrhage and necrosis.

Microscopic:-

Large no. of osteoclast are regularly scattered through out the stroma.

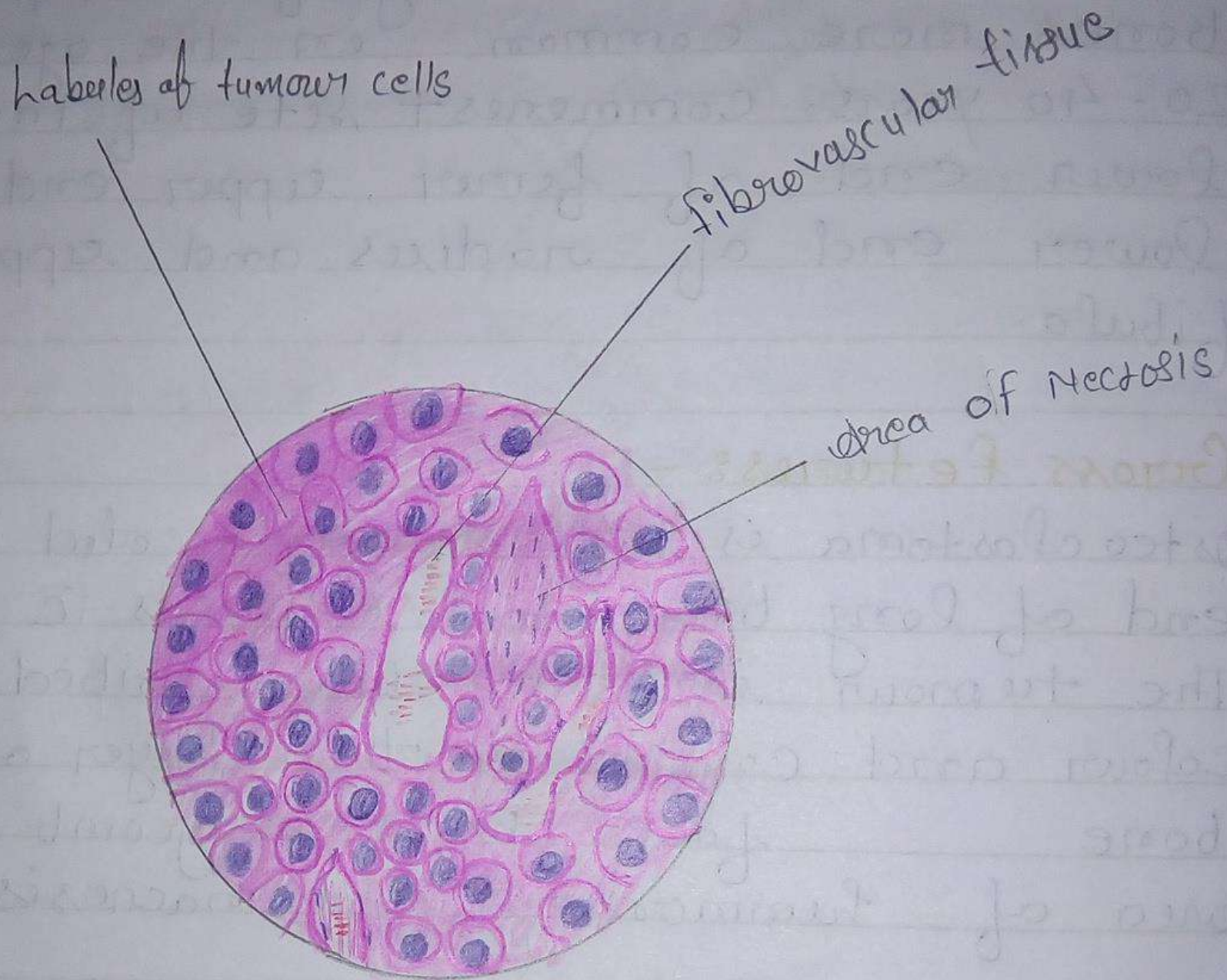
Giant cell contains many benign nuclei and are similar to normal osteoclast.

Stromal cells are mononuclear and they are the real tumour cells.

They are reniform plump spindle shape or round to oval but may have varying degree of atypical mitosis.

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Ewing's Sarcoma

Experiment No. 20

Date

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Ewing's Sarcoma

It is highly malignant, round cell tumour, arising in medullary canal of diaphysis or metaphysis. Normal site - shaft of long bone such as femur, tibia, fibula and humerus.

Common age of occurrence is 15-20 years of age with predilection for females are more.

Gross features:-

Typically located in medullary cavity and produce expansion of affected epiphysis and diaphysis. The tumour often extends into the adjacent soft tissue.

Cut surface shows greywhite soft friable mass.

Microscopic features:-

The tumour is divided by fibrous septa into irregular lobules of closely packed tumour cells. Tumour cells of the lobules are small, uniform appear as lymphocytes and have round nucleus with frequent mitosis.

Cytoplasm is scanty and contain glycogen which is positive for P.PAS.

The tumour cells arranged around the blood vessels forming pseudorosettes.

Teacher's Remarks

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