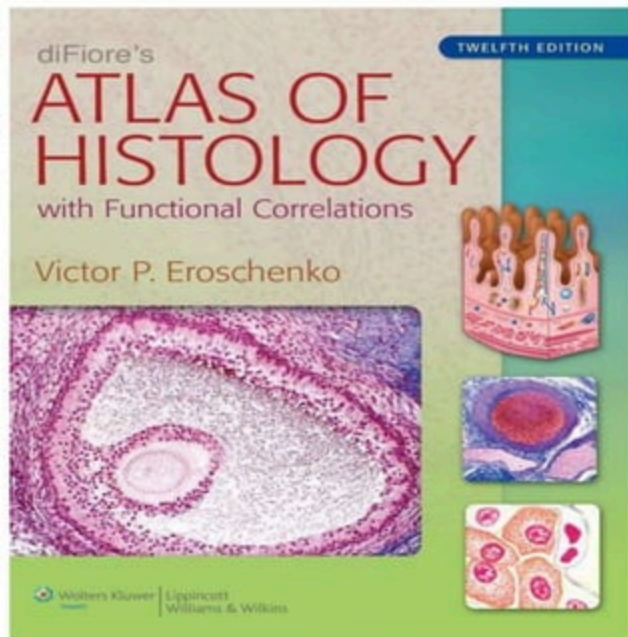


Introduction to Histology

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References

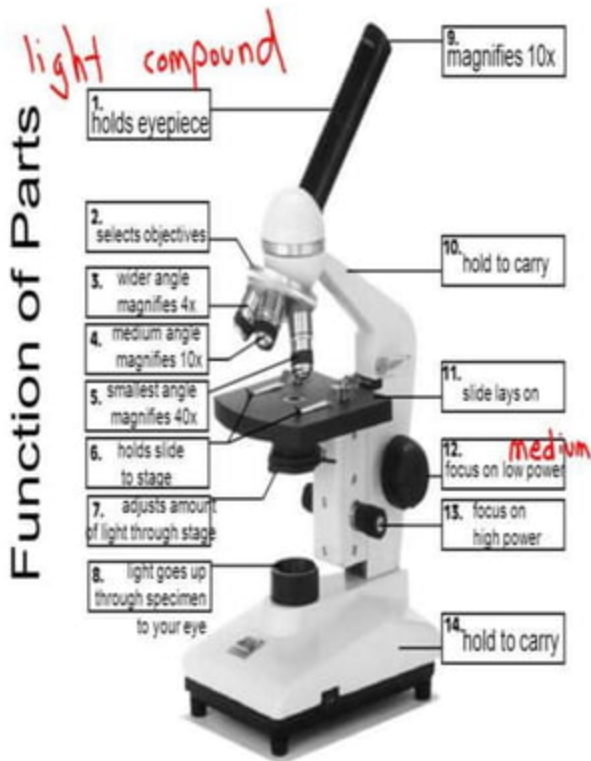
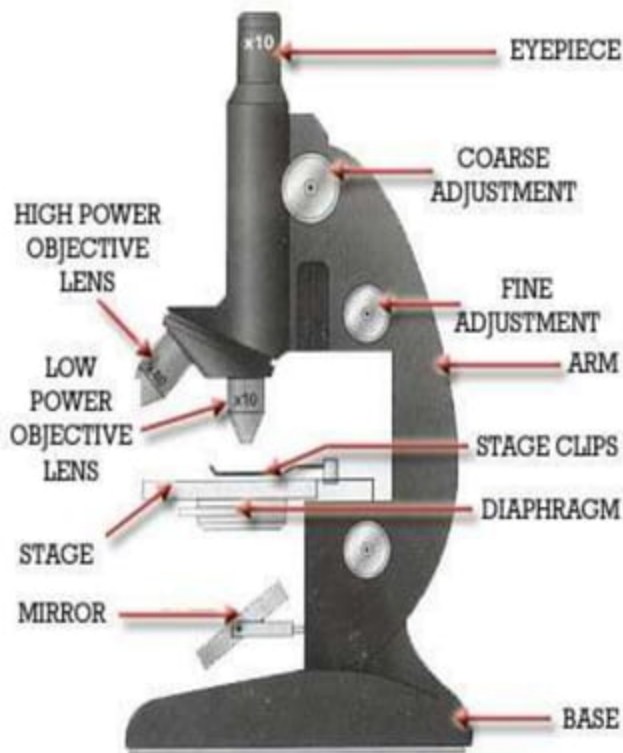
- <http://www.histology.leeds.ac.uk/what-is-histology/index.php>.
- http://histology.leeds.ac.uk/what-is-histology/The_electron_microscope.php
- The histology text book - Jonquiere's Basic Histology: Text and Atlas, Thirteenth Edition.
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Histology is the study of tissues and organs under the microscope

- It is the microscopic anatomy, to study the detailed structure with the aid of light microscope or electron microscope. Histology means the science of the tissues.
- histos is greek for web or tissue
- logia is greek for branch of learning
- Microscope was invented by Zacharias Janssen.
- A hand lens, spectacle serves as a simple microscope.
- A compound microscope is having system of lenses, that uses concave mirror for reflecting the light from the source and directs it to pass through the condenser and the ray of light pass through the slide, tissue and comes to the eye piece after passing through the objective piece.
- Robert Hooke discovered the cells.
- Cytology is the study of cell.

Parts of a light compound microscope



The electron microscope

- The actual power or magnification of a compound optical microscope is the product of the powers of the ocular (eyepiece) and the objective lens.
- The maximum normal magnifications of the ocular and objective are $10\times$ and $100\times$ respectively, giving a final magnification of $1,000\times$.
- Electron Microscopes can have magnifications of $\times 500,000$.
- Resolution is the smallest distance below which two discrete objects will be seen as one.
- It is the amount of information that can be seen in the image.
- The limit of resolution of the light microscope is $0.2\ \mu\text{m}$ (greatest magnification is $\times 1,400$).



Light Microscopy

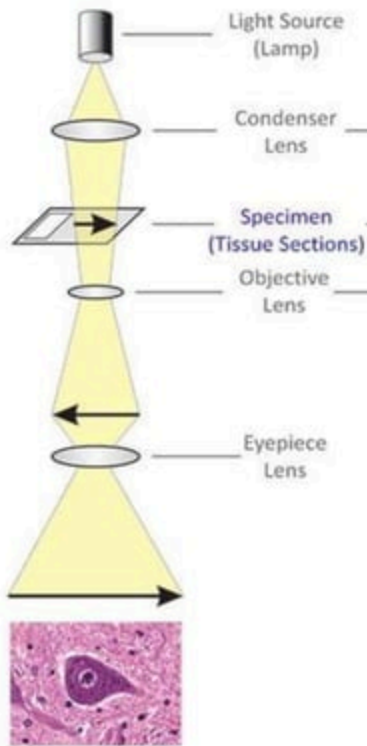


Image Viewed Directly

Transmission Electron Microscopy

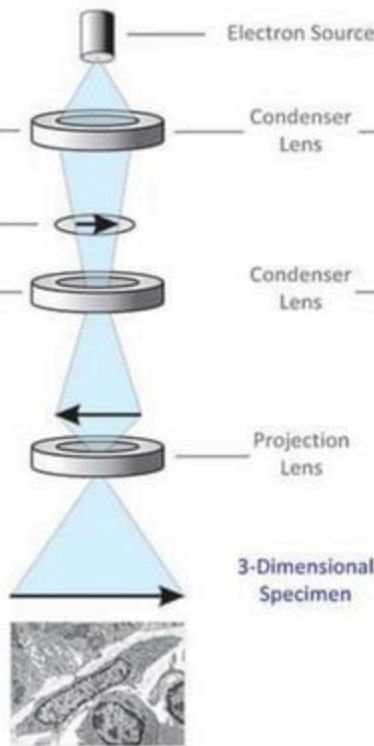


Image Viewed on Fluorescent Screen

Scanning Electron Microscopy

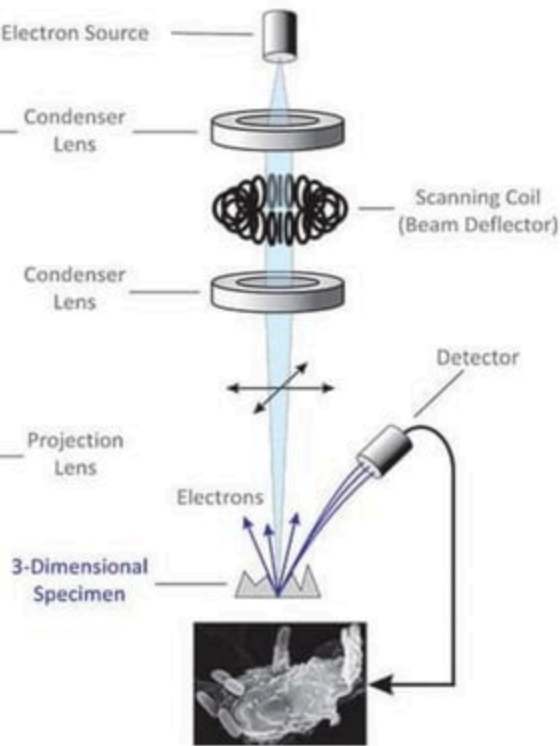


Image Viewed on Monitor

Differences between Light Microscope and Electron Microscope

Light Microscope	Electron Microscope
Illuminating source is the Light .	Illuminating source is the beam of electrons .
Specimen preparation takes usually few minutes to hours .	Specimen preparation takes usually takes few days .
Live or Dead specimen may be seen.	Only Dead or Dried specimens are seen.
Condenser, Objective and eye piece lenses are made up of glasses .	All lenses are electromagnetic .
It has low resolving power ($0.25\mu\text{m}$ to $0.3\mu\text{m}$).	It has high resolving power ($0.001\mu\text{m}$), about 250 times higher than light microscope.
It has a magnification of 500X to 1500X .	It has a magnification of 100,000X to 300,000X .
The object is $5\mu\text{m}$ or thicker.	The object is $0.1\mu\text{m}$ or thinner.
Image is Colored .	Image is Black and White .
Vacuum is not required.	Vacuum is essential for its operation.
There is no need of high voltage electricity.	High voltage electric current is required (50,000 Volts and above).
There is no cooling system.	It has a cooling system to take out heat generated by high electric current.
Filament is not used.	Tungsten filament is used to produce electrons.
Radiation risk is absent.	There is risk of radiation leakage.
Specimen is stained by colored dyes.	Specimen is coated with heavy metals in order to reflect electrons.
Image is seen by eyes through ocular lens .	Image is received in Zinc Sulphate Fluorescent Screen or Photographic Plate .
It is used for the study of detailed gross internal structure.	It is used in the study of external surface, ultra structure of cell and very small organisms.

The microscopic measurements

- One millimeter is equal to 1000 micrometer/micron.
- One micrometer = $1/1000000$ meter or 0.001 millimeter
- One micrometer = 1000 Nanometer
- One Nano meter = 0.001 micrometer
- Normal RBCs have a diameter of 6 - 8 μm .
- The diameter of a capillary vary from 5-10 micrometers.



Top View shows RBC to be circular



Side view shows RBC to be a biconcaved disc



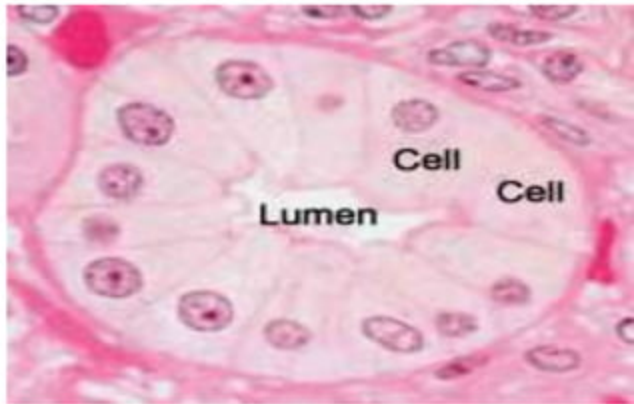
The common stains used to study tissues

- Acidic stains
- H&E stains
- Basic stains
- Neutral stains
- Special stains

Basic dyes	
Methyl green	Green
Methylene blue	Blue
Pyronin G	Red
Toluidine blue	Blue
Acidic dyes	
Acid fuschin	Red
Aniline Blue	Blue
Eosin	Red
Orange G	Orange

What is H & E. staining?

- The most commonly used staining system is called H&E (Haematoxylin and Eosin).
- H&E contains the two dyes haematoxylin and eosin.
- Eosin is an acidic dye: It stains basic (or acidophilic) structures red or pink. This is also sometimes termed 'eosinophilic'.
- Thus the cytoplasm is stained pink.
- Haematoxylin can be considered as a basic dye.
- It is used to stain acidic (or basophilic) structures a purplish blue.
- Thus the nucleus is stained purple .

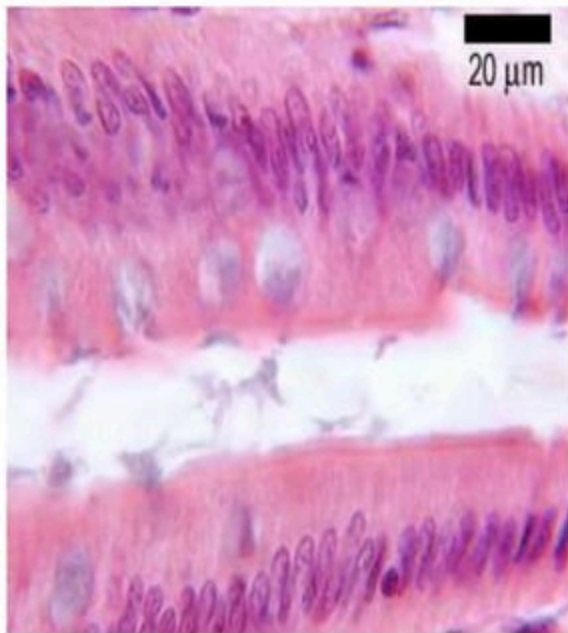


Structures taking stains

- What structures are stained purple (basophilic)?
- DNA (heterochromatin and the nucleolus) in the nucleus, and RNA in ribosomes and in the rough endoplasmic reticulum are both acidic, and so haemotoxylin binds to them and stains them purple.
- Some extracellular materials (i.e. carbohydrates in cartilage) are also basophilic.
- What structures are stained pink (eosinophilic or acidophilic)?
- Most proteins in the cytoplasm are basic, and so eosin binds to these proteins and stains them pink.
- This includes cytoplasmic filaments in muscle cells, intracellular membranes, and extracellular fibres.

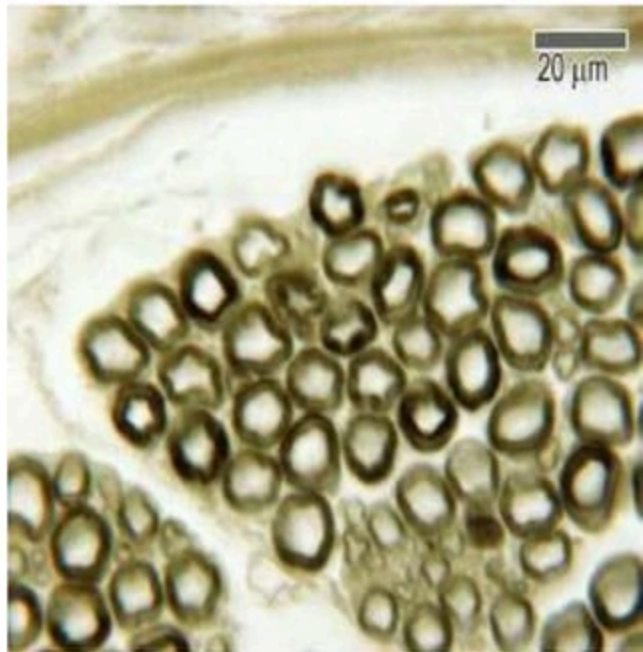
Periodic acid-Schiff reaction (PAS)

- It is a basic stain. PAS stains carbohydrates and carbohydrate rich macromolecules a **deep red colour (magenta)**.
- PAS will therefore stain up:
- Glycogen the intracellular storage form of carbohydrate in cells.
- Mucus in cells and tissues, Basement membranes, and Brush borders of kidney tubules and small and large intestines.
- Reticular fibres (i.e. collagen) in connective tissue and Cartilage.
- The mucin produced by goblet cells is stained a purple colour by this stain.



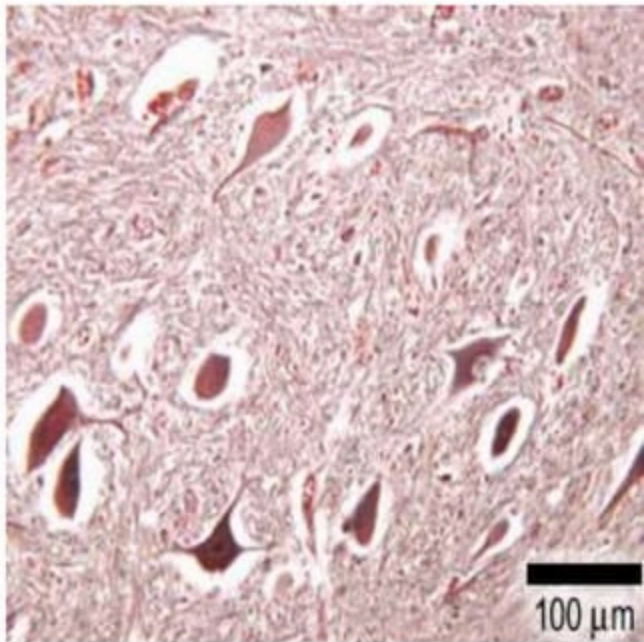
Special stains

- Sudan Black and osmium.
- These dyes stain lipid-containing structures such as myelin a brownish-black colour.



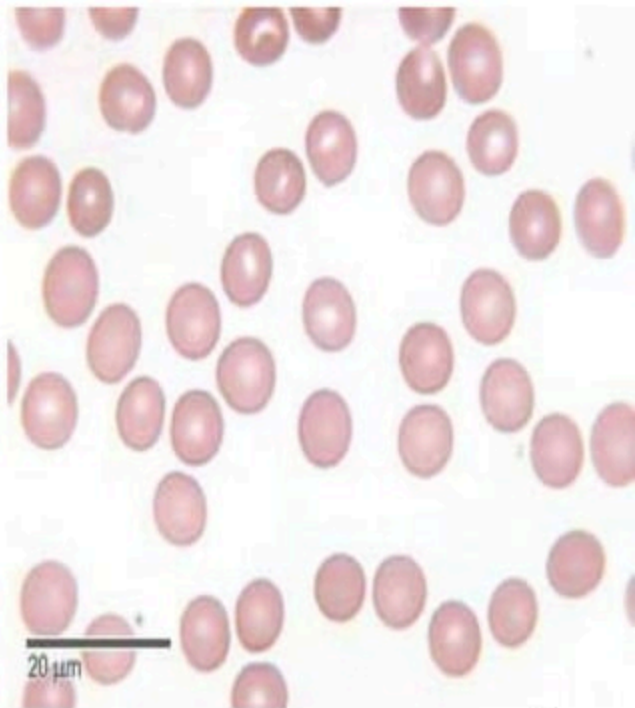
Nissl and methylene blue

- A basic dye used to stain the rough ER in neurons.



Giemsa stain

- Usually used for staining blood and bone-marrow smears.
- Nuclei are stained dark-blue to violet, cytoplasm pale blue, erythrocytes pale pink.



The fundamental tissues of our body

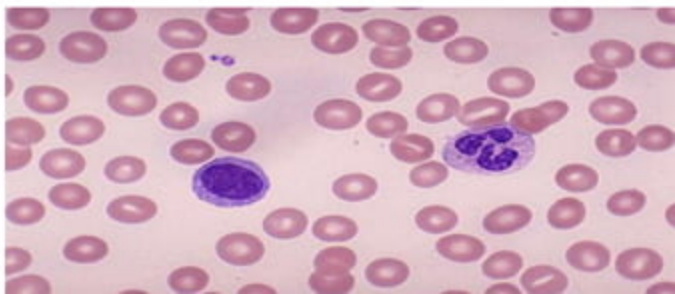
- The entire body is made up of four basic tissues, they are:-
 - 1. Epithelial tissue
 - 2. Connective tissue
 - 3. Muscle
 - 4. Nervous tissue
- Tissues are made up of
 - cells
 - extracellular (outside cells) matrix
 - body fluids.

Activities to do in the histology lab

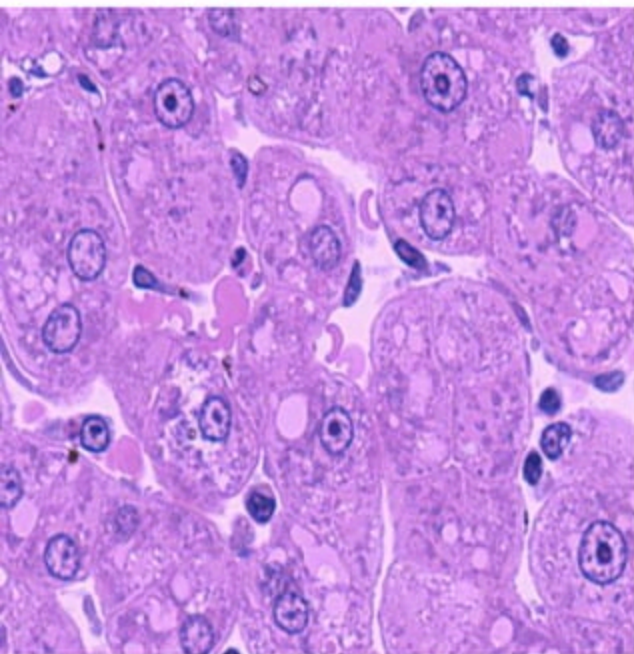
- The entire batch is divided into
- (A) batch=roll no. 1-33
- (B) batch=roll no. 34-37
- (C) batch=roll no. 38-till last.
- (A) batch will have histology lab on Monday from 2.00-4.00pm
- (B) batch will come on Tuesday same timings.
- (C) batch will be having histology lab on Wednesday from 2.00-4.00pm.
- The students have to bring the histology record, pink, blue and HB2 lead pencils.
- They have to attend the prelab and study the slides under microscope and draw the tissue and label it.
- Discuss the slide and identification points.
- Relate the structures with their intended functions.
- All should wear white lab coats during lab activities.

Cytology is the study of cells

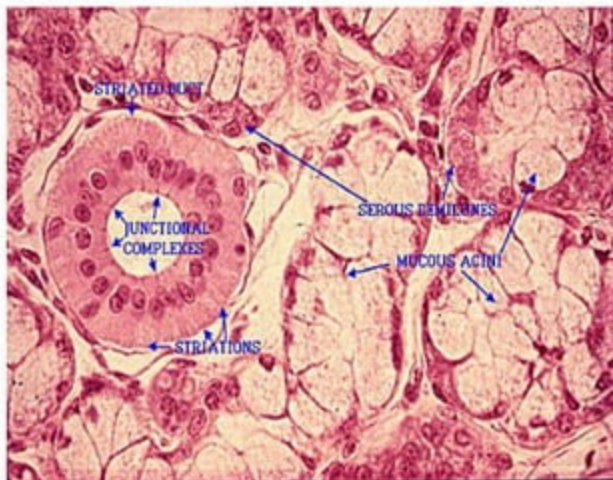
- A cell is a structural and functional unit of all living things.
- It has a cell membrane, the boundary that limit the cell area.
- The cell has smaller components called as cell organelles like nucleus, mitochondria, rough endoplasmic reticulum, Golgi bodies and vesicles.
- **Cell membrane** is the membrane that limits the cell periphery. Below is a blood smear, we observe RBCs, monocyte, polymorph and platelets.
- The cell membrane separates one cell from another.



The nucleus of cell is the largest cell organelle



- The striated ducts of salivary glands have basal striation due to piles of mitochondria.

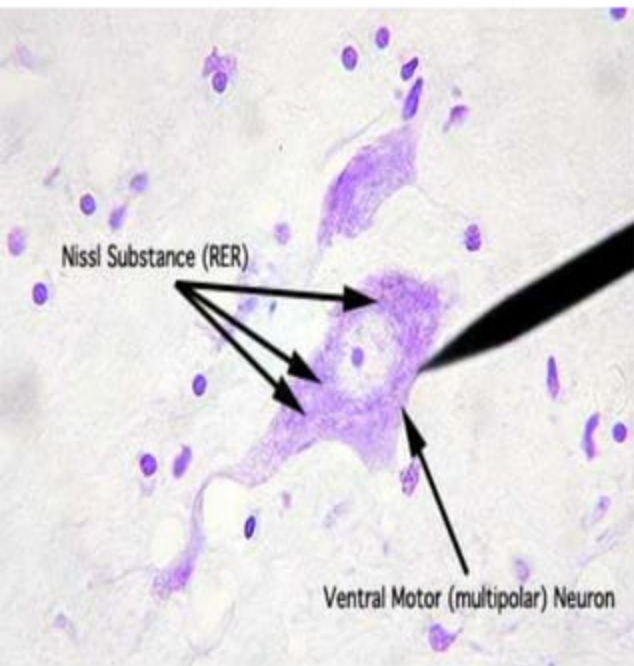


The secretory granules



- This oil immersion view of the pancreatic acinar cell highlights the appearance of another cytoplasmic constituent, the secretory granule .
- Granules may have different staining properties depending on the nature of their constituent product.
- These granules are noticeably eosinophilic.

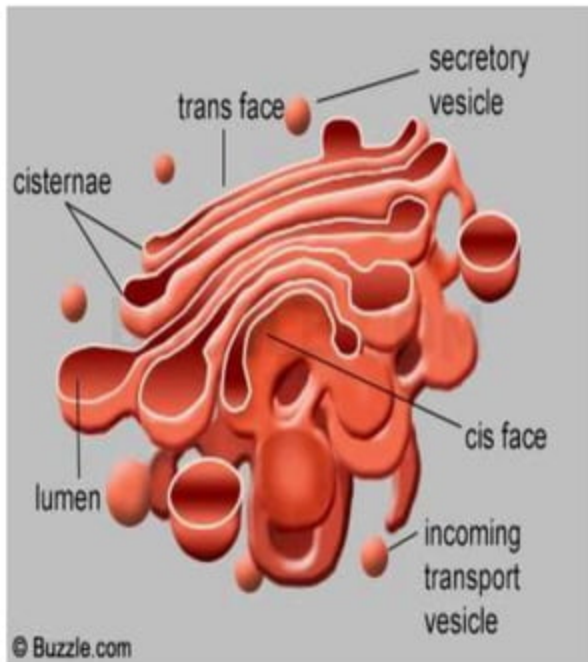
The rough endoplasmic reticulum



- The rough endoplasmic reticulum consists of endoplasmic reticulum with ribosomes attached.
- The RER is active in synthesizing proteins destined for export or bound for another intracellular organelle.

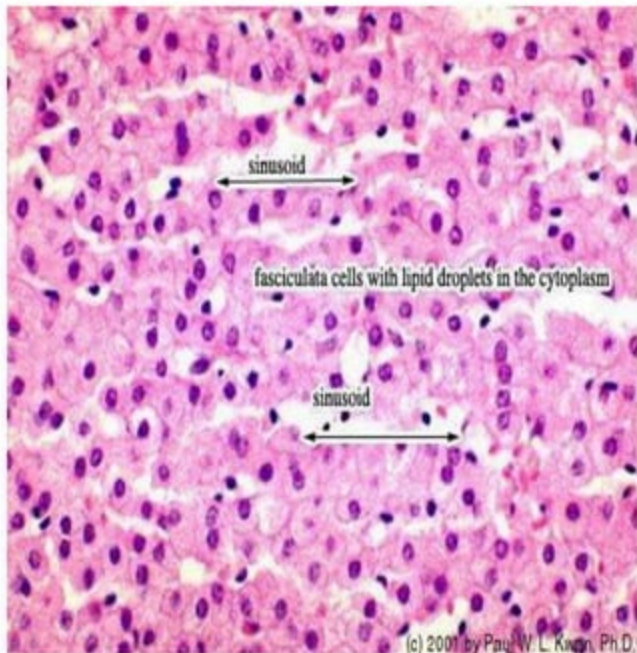
The Golgi complex

- Under light microscope is not visible, it will not take up stain-negatively stained area above nucleus is Golgi complex.
- Golgi complex packs proteins and transfer them.

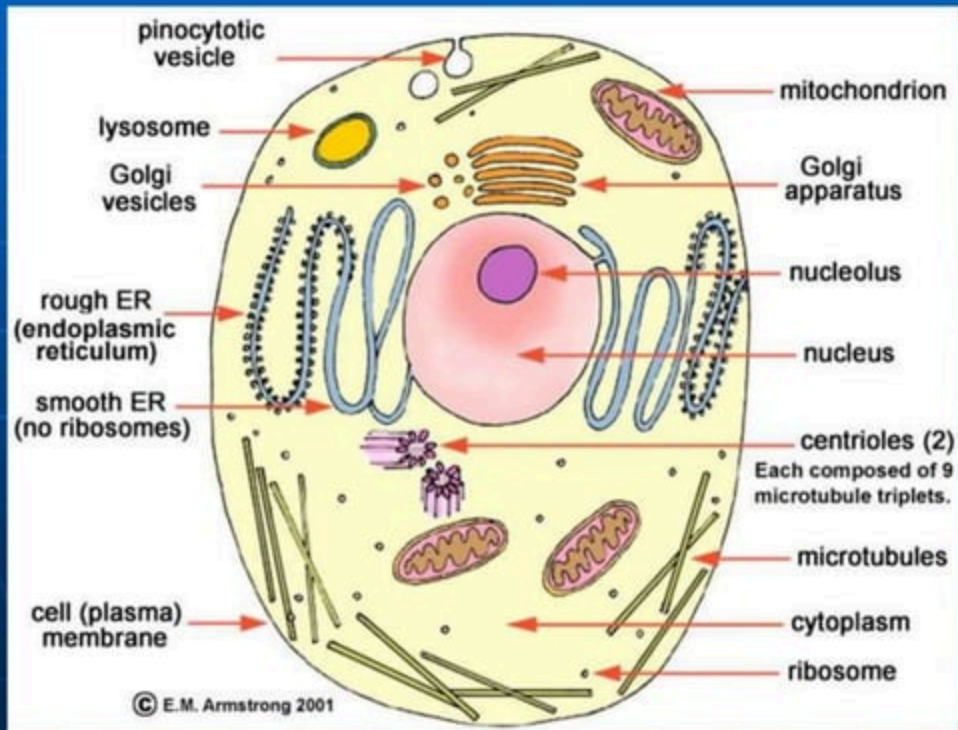


The lipid droplets

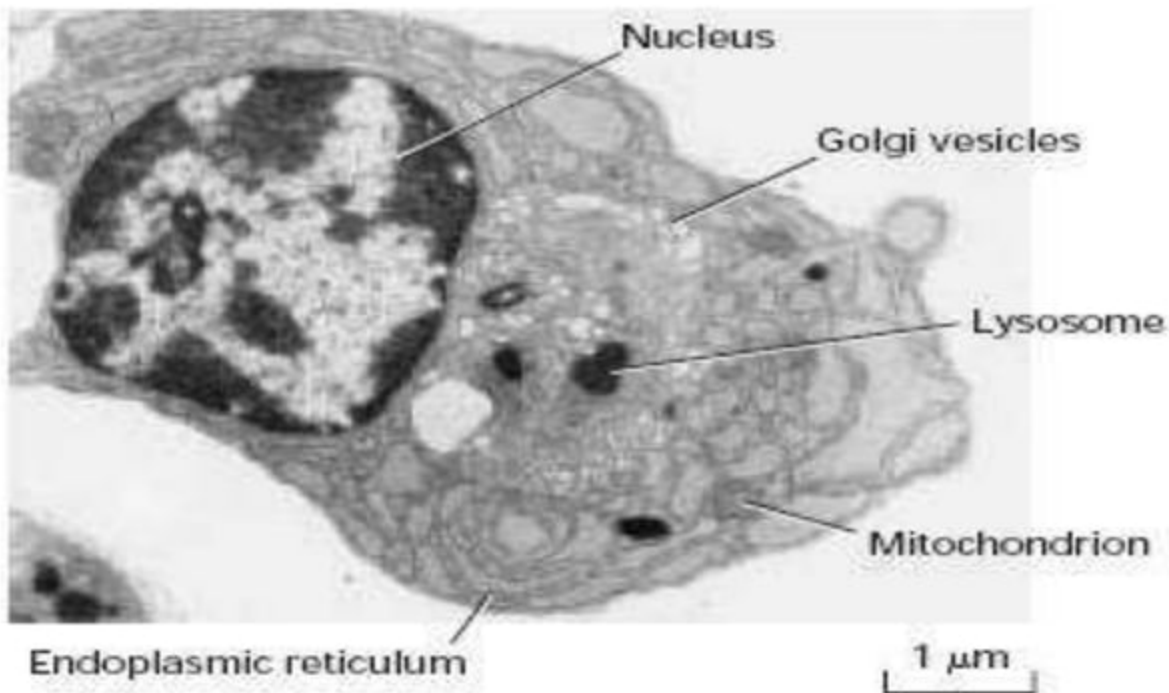
- These droplets represent an intracellular storage form, in this case, for precursors necessary for the synthesis of steroid hormone.
- It is a feature of steroid secreting cells-adrenal gland cortex.



Simple structure of a eukaryotic cell



The ultrastructure of Eukaryotic cell

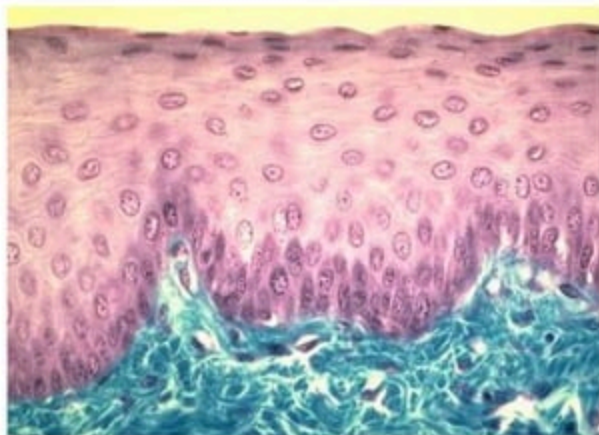


The histology is being classified as

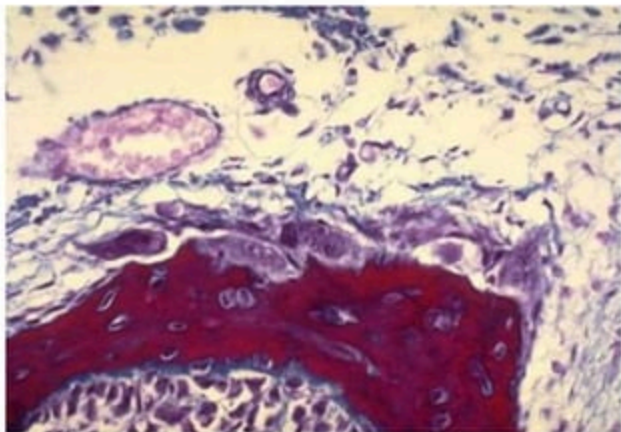
- General histology
- Deals with the fundamental tissues of the body.
- The epithelial tissue
- The connective tissue
- The muscular tissue
- And the nervous tissue
- Systemic histology
- Our entire body is made up of all four basic tissues.
- The systemic histology deals with microscopic study of various systems of our body like, musculoskeletal system, cardiovascular system, excretory system and other systems.

Basic tissues

- Epithelium



- Connective tissue

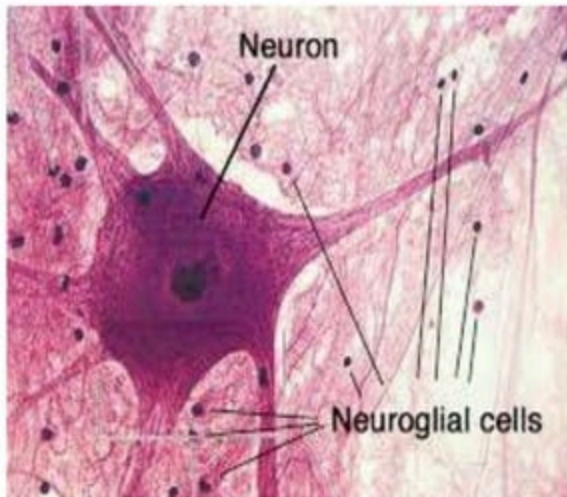


The basic tissues

- Muscular tissue



- Nervous tissue

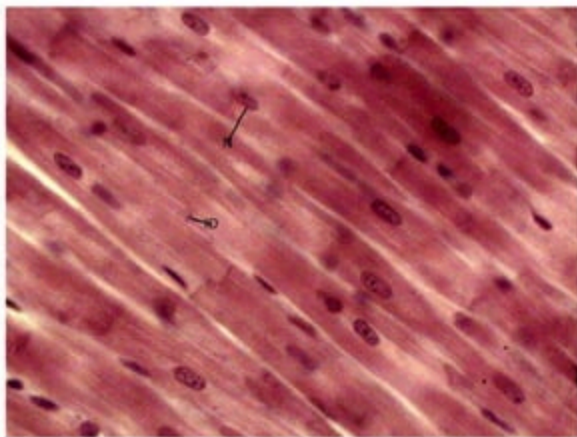


Systemic histology

- Histology of a blood vessel



- Histology of heart



Histology of endocrine glands

- Thyroid gland histology
- Histology of kidney

