HISTOLOGY

Histology is the study of the tissue

Tissue: Cells + ECM
The Extracellular Matrix

* Atoms → Molecule → Organ → system → Body

* Tissue Preparation for Light Microscopy:

Fixation:
- function:
  1. Cross-link proteins
  2. inactive degradative enzymes
- aim: to preserve cell and tissu structure

Dehydration:
- the Process: transfer the tissue through a series of increasingly concentrated alcohol solution.
  20% → 40% → 60% → 80% → 100% (the End)
- aim: remove the water from the Tissue

Alcohol & paraffin are miscible

Clearing:
- aim: remove alcohol and replace it with organic solvents.

Infiltration:
- aim: the tessiue is completely infiltrated in paraffin

Embedding:
- aim: allow the pareffin to harden.

Trimming aim → Slicing the block

Finally:

Staining:

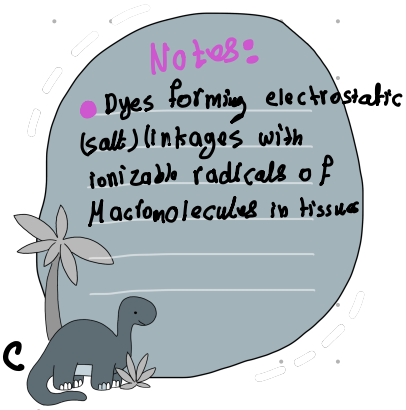
Staining and Stains:

most cells & extracellular material are completely colorless.

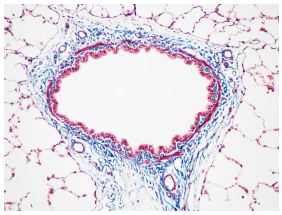
Cell components

basophilic
- has a negative charge
- has affinity for basic dyes ex: toluidine blue, alcian blue methylene blue -Hematoxylin
- Such as nucleic acids (DNA / RNA / glycosaminoglycans)

acidophilic
- has a Positive charge
- has affinity for acidic dyes ex: eosin, orange g fuchsin (Mitochondria, secretory granules, collagen)



Special stains:



- Tri chrome: allow greater distinction in extra cellular components ex: Masson tri chrome
- The Periodic acid schiff (PAS): reaction utilizes the hexose rings of uses Polysaccharides and other carbohydrate-rish tissue ex: macro molecules distinctly purple or magent
- Sudan black: (lipid-soluble dyes) we avoid the steps of removing lipids ex: treatment with heat and Organic solvents
- Metal impregnation: using solution of silver salts to visual certain ECM fibers and specific cellular element in nervous tissue.
- Immunostaining: immunofluorescence and immunohistochemistry.
  - the least common

Microscopes

Light Microscope: L.M

- Interaction light with tissue.
- resolution around 3 Mm
- Bright-field/Fluorescence/Phase-contrast confocal/polarizing

Electron Microscope:

- interaction of tissue with beams of electrons.
- resolution around 3 Nm
- Transmission/scanning

Resolution: the smallest distance between two object as separated objects.

any object is smaller than or thinner than 0.2 Mm cannot be distinguished

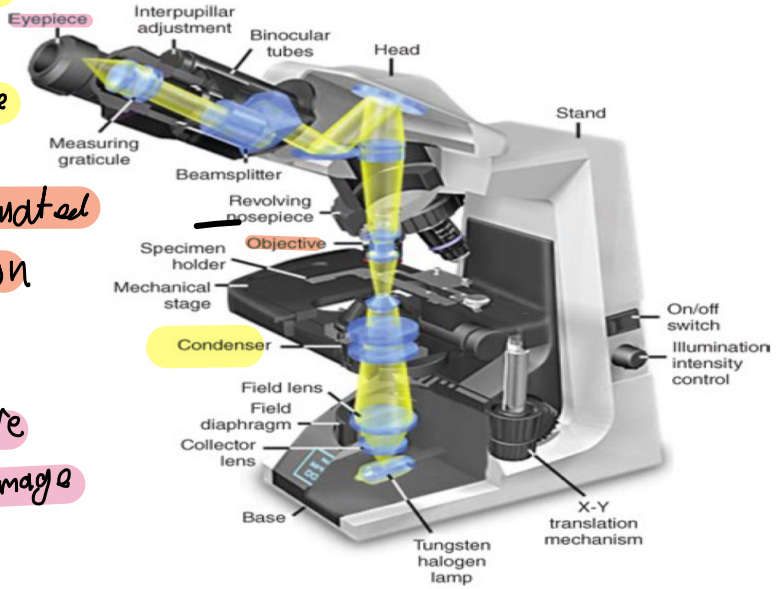
The max resolving power for L.M 0.2 Mm

magnified 1000-1500 times

Light Microscope (Bright-field)

* Collects and focuses a cone of light that illuminates the tissue
* enlarge the illuminated image of the obj. on eyepiece
* 2 Eyepiece: more magnifity for the image

Components and light path of a bright-field microscope.



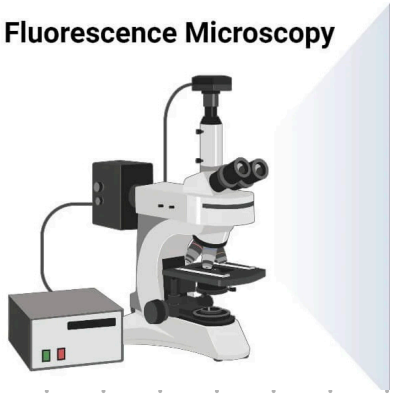
The quality of the pic. depends on objective lens

The total magnification =

M. Eyepiece * M. Objective lens

is always X10

Fluorescence: when a certain cellular substance absorb light with a proper wavelength they emit light with longer wavelength



"The tissue sections are irradiated with UV light and the emission light is in the visible portion of the spectrum"

Phase-contrast Microscopy:

- Study unstained cells and tissue (colorless, similar optical densities) and it can use living cultured cells and it can produces images from transparent objects.

The way of working:

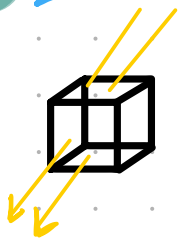
the light changes its speed when passing through cellular and extra cellular structures with different refractive indices appear lighter or darker

اللَّهُ نُورُ السَّمَاوَاتِ وَالْأَرْضِ ۚ مَثَلُ نُورِهِ كَمِشْكَاةٍ فِيهَا مِصْبَاحٌ ۖ
الْمِصْبَاحُ فِي زُجَاجَةٍ ۖ الزُّجَاجَةُ كَأَنَّهَا كَوْكَبٌ دُرِّيٌّ يُوقَدُ مِن شَجَرَةٍ
مُّبَارَكَةٍ زَيْتُونَةٍ لَّا شَرْقِيَّةٍ وَلَا غَرْبِيَّةٍ يَكَادُ زَيْتُهَا يُضِيءُ وَلَوْ لَمْ
تَمْسَسْهُ نَارٌ ۚ نُّورٌ عَلَىٰ نُورٍ ۗ يَهْدِي اللَّهُ لِنُورِهِ مَن يَشَاءُ ۚ وَيَضْرِبُ
اللَّهُ الْأَمْثَالَ لِلنَّاسِ ۗ وَاللَّهُ بِكُلِّ شَيْءٍ عَلِيمٌ ﴿٣٥﴾

Electron Microscope

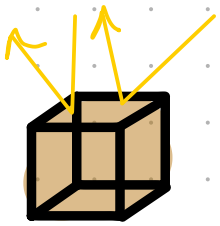
TEM

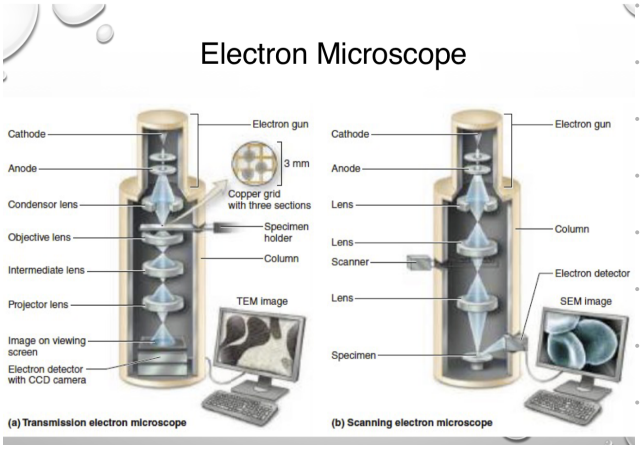
- The electron beam passes the tissue
- Very hight Magnification
- Very thin sections 40-90 nm
- $\bar{e}$ interact with tissue producing black white shades of gray images.
- 2D images.



SEM

- The electron beam doesn't passe the tissue
- the surface of cells and tissue is coated with heavy metals gold → which reflect the $\bar{e}$ → producing 3D images → which is recording of the specimen topography





Done By Noor Marzoog.