



Human Histology

REFERENCE: JUNQUEIRA'S BASIC HISTOLOGY, TEXT AND ATLAS, 15TH EDITION, BY ANTHONY L. MESCHER , CHAPTER 1.



COURSE OBJECTIVES

- DESCRIBE THE STRUCTURE AND FUNCTION OF MAJOR TISSUE TYPES.
- UNDERSTAND THE RELATIONSHIP BETWEEN STRUCTURE AND FUNCTION IN HISTOLOGICAL SAMPLES.
- IDENTIFY TISSUES AND ORGANS UNDER THE MICROSCOPE.
- APPLY HISTOLOGICAL KNOWLEDGE TO CLINICAL AND PATHOLOGICAL CONTEXTS.



HISTOLOGY

- MICROSCOPIC ANATOMY!
 - *HISTO= WEB OR TISSUE*
 - *LOGOS= STUDY*
 - THE STUDY OF CELLS AND THE EXTRACELLULAR MATRIX
- 

Histology

- Histology is the study of the tissues of the body and how these tissues are arranged to constitute organs.



Topics

Introduction to histology &
microscopy techniques

The cell: structure and organelles

Epithelial tissue

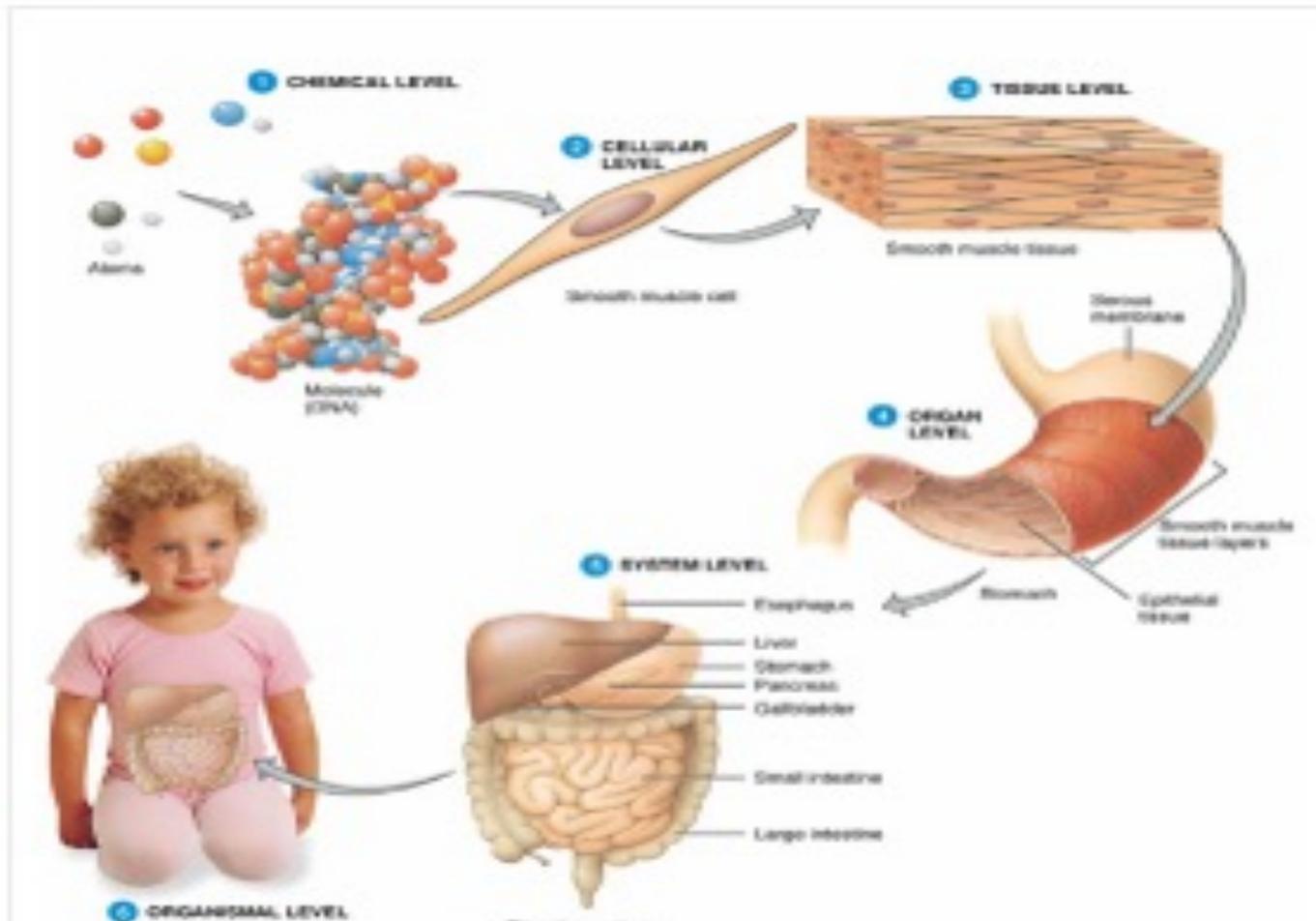
Connective tissue

Cartilage and bone

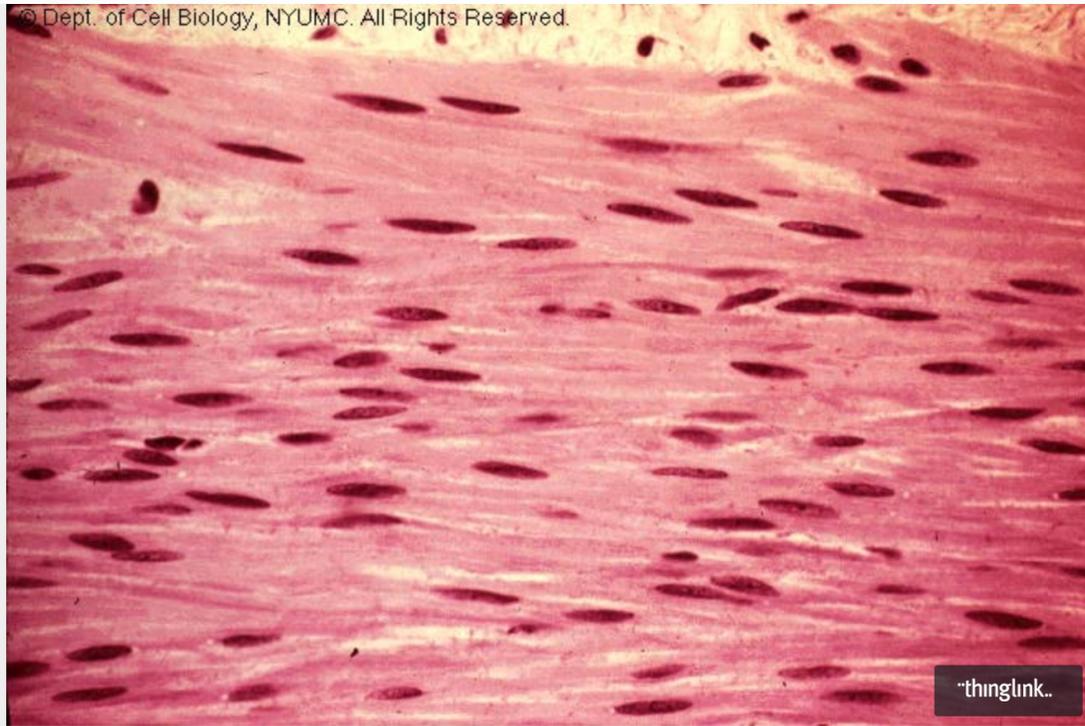
Nervous tissue

Muscle Tissue

Level Of Organization



What is this?
How did we get it?



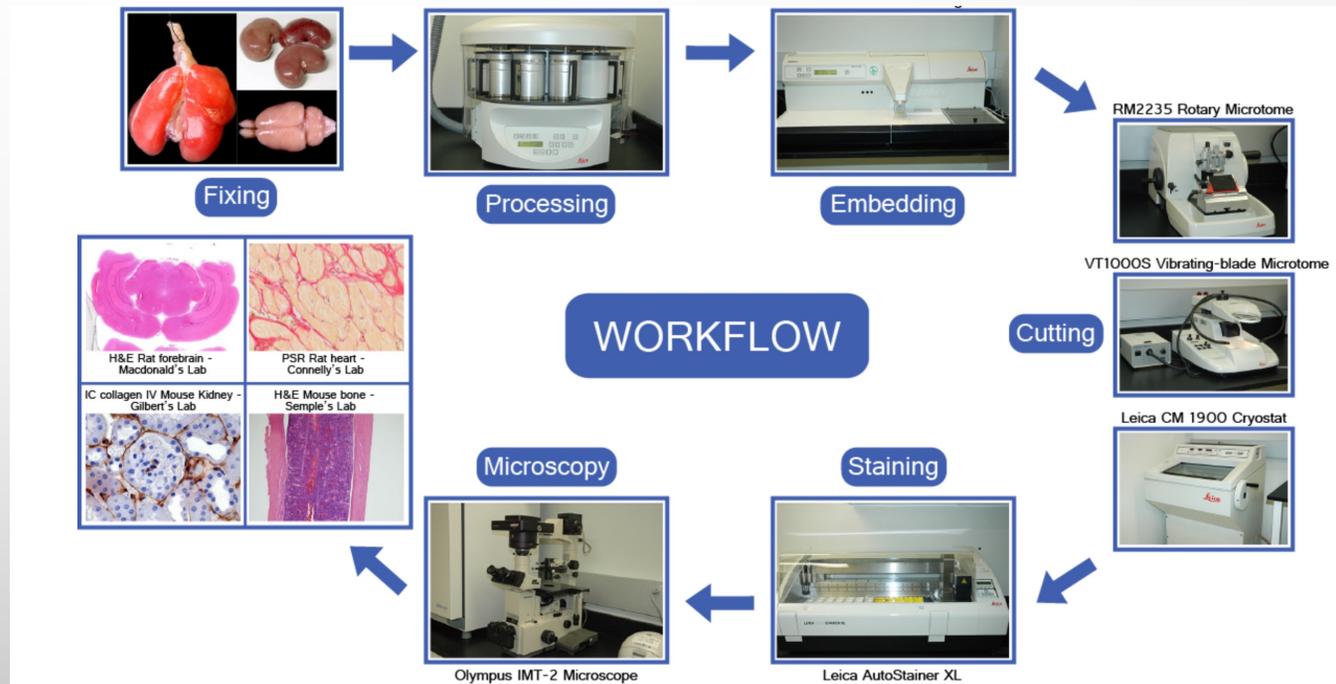
Tissue Processing For Histology



<https://www.youtube.com/watch?v=4DJm4NLEcQs>

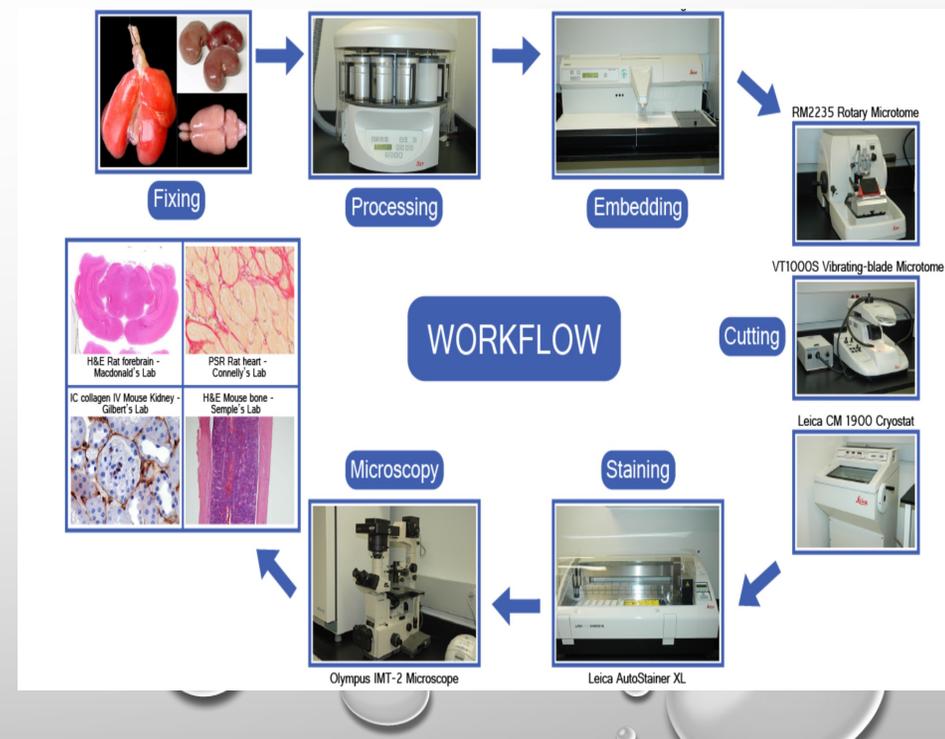


Paraffin block



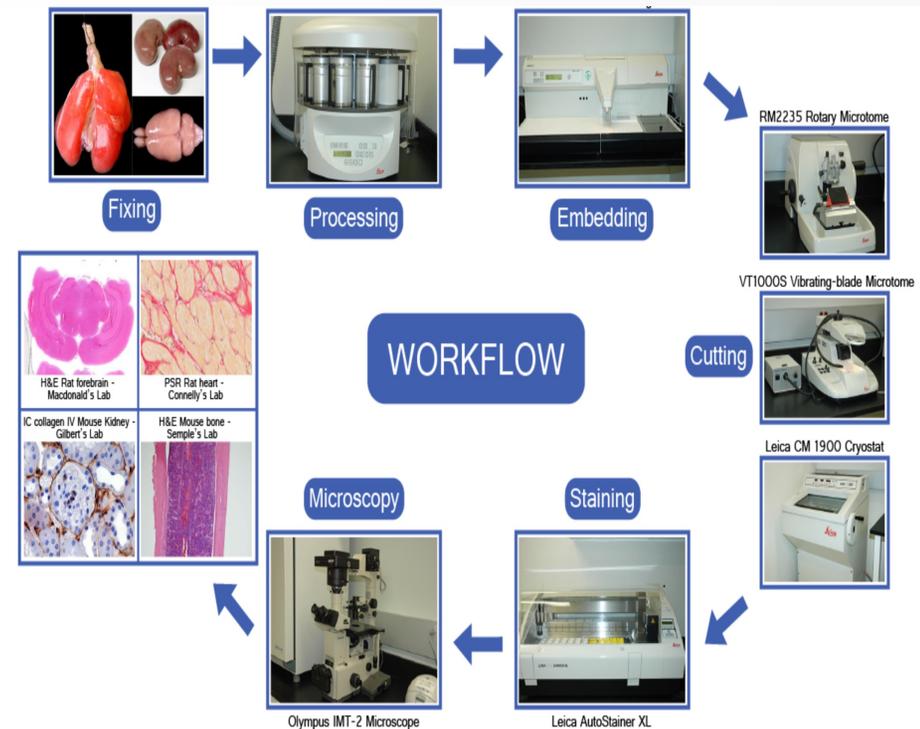
Tissue Preparation For Light Microscopy

- **Fixation:** small pieces of tissue are placed in solutions of chemicals that cross-link proteins and inactivate degradative enzymes, which preserves cell and tissue structure.
- **Dehydration:** the tissue is transferred through a series of increasingly concentrated alcohol solutions, ending in 100%, which removes all water.
- **Clearing:** alcohol is removed in organic solvents in which both alcohol and paraffin are miscible.

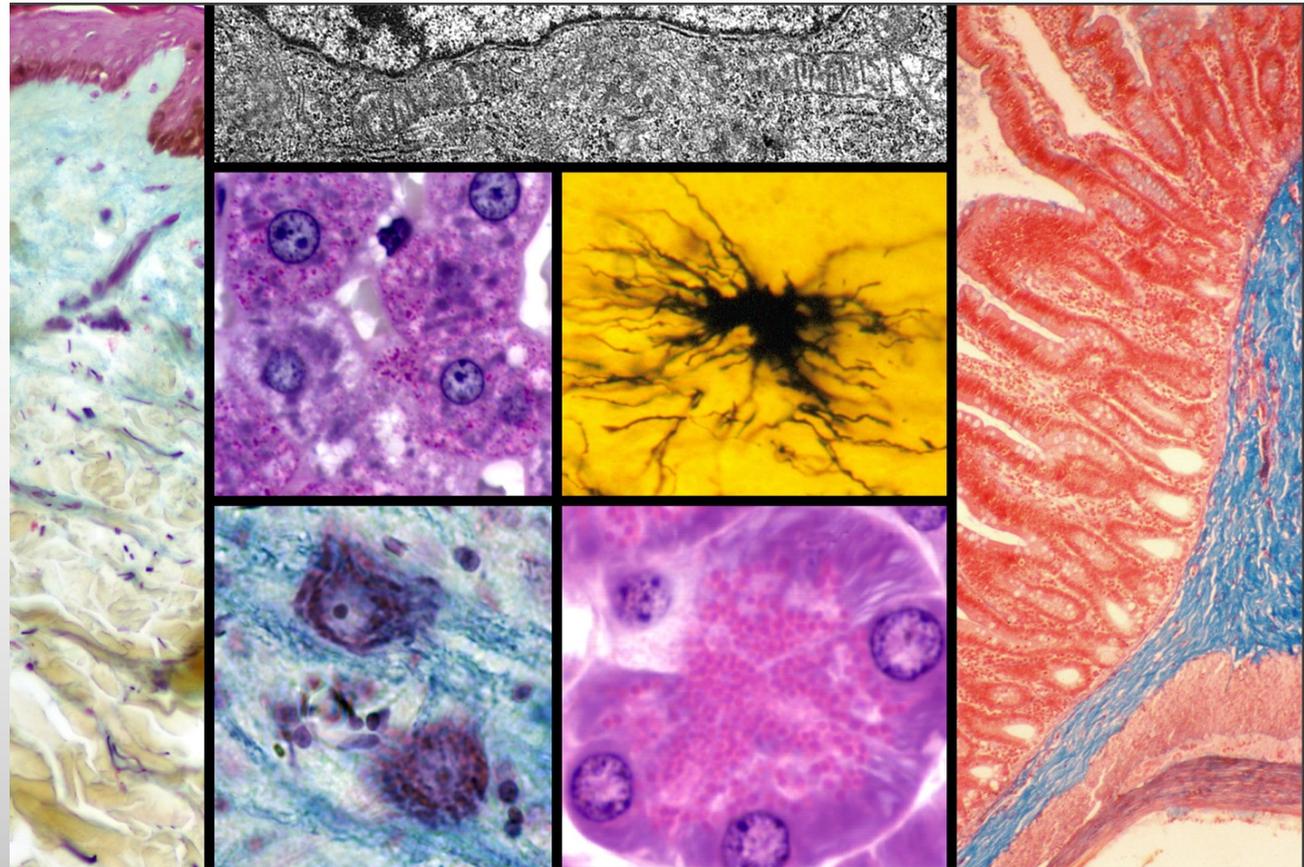


Tissue Preparation For Light Microscopy

- **Infiltration:** the tissue is then placed in melted paraffin until it becomes completely infiltrated with this substance.
- **Embedding:** the paraffin-infiltrated tissue is placed in a small mold with melted paraffin and allowed to harden.
- **Trimming:** the resulting paraffin block is trimmed to expose the tissue for sectioning (slicing) on a microtome.



STAINING

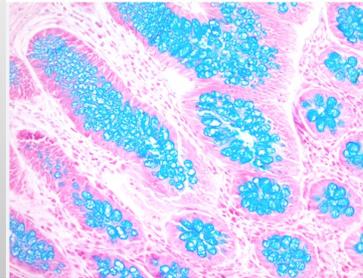
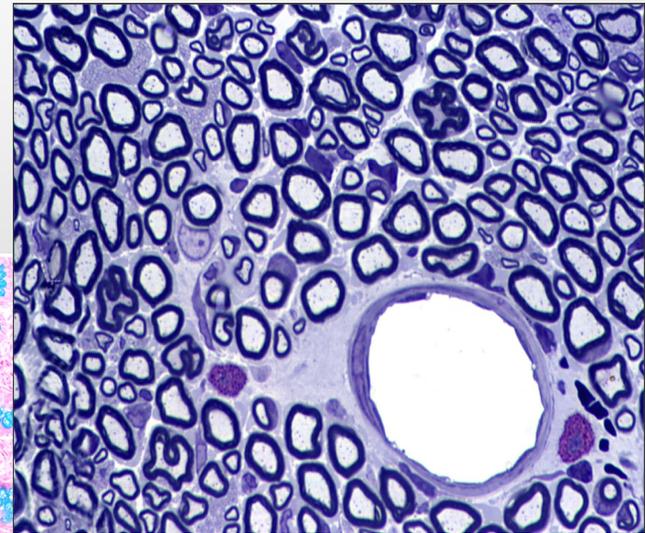
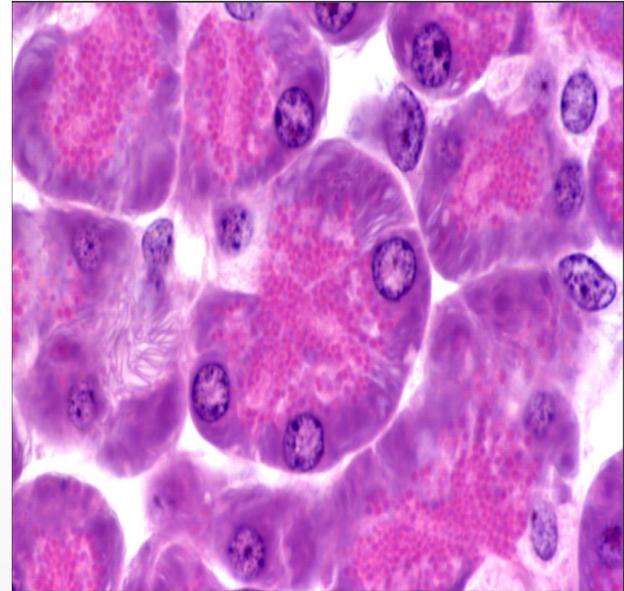


Staining And Stains

- Most cells and extracellular material are completely colorless!
- Dyes forming electrostatic (salt) linkages with ionizable radicals of macromolecules in tissues.
- Cell components such as nucleic acids with a net negative charge (anionic) have an affinity for basic dyes and are termed **basophilic**.
- Cationic components, such as proteins with many ionized amino groups, stain more readily with acidic dyes and are termed **acidophilic**.

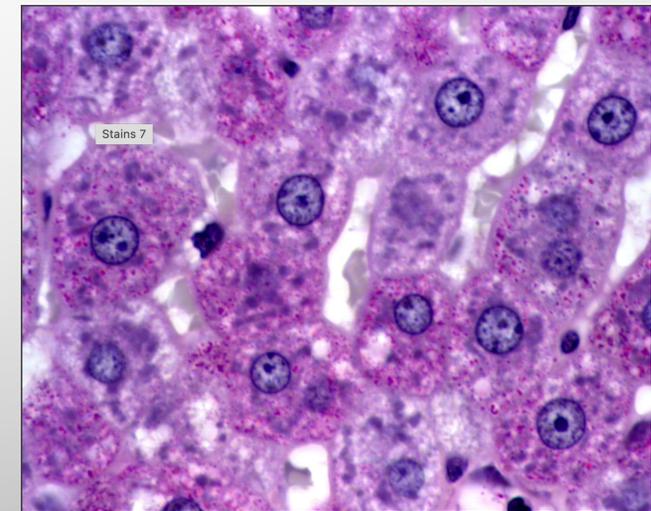
Staining and Stains

- Basic dyes include toluidine blue, alcian blue, and methylene blue.
- **Hematoxylin** behaves like a basic dye, staining basophilic tissue components.
- DNA, RNA, and glycosaminoglycans: ionize and react with basic dyes do so because of acids in their composition
- Acid dyes: **eosin**, orange g, and acid fuchsin stains mitochondria, secretory granules, and collagen are acidic.



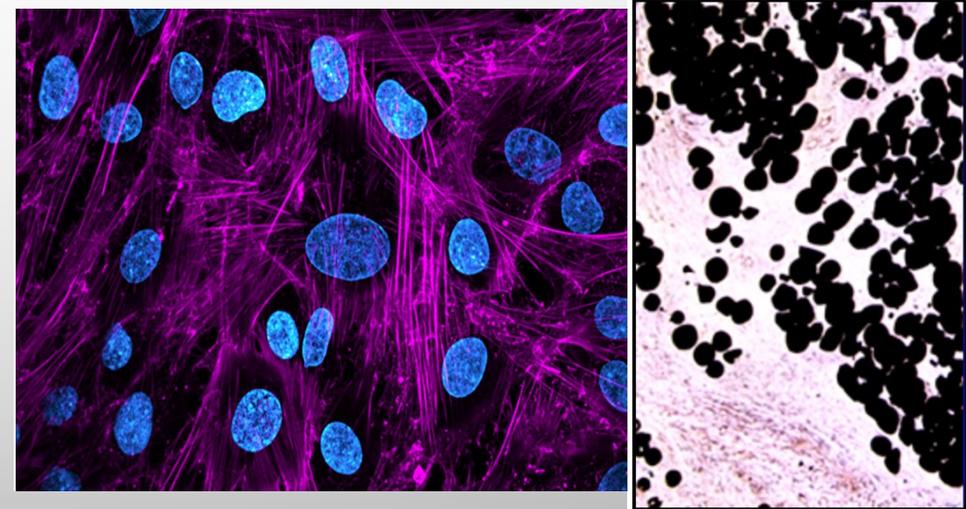
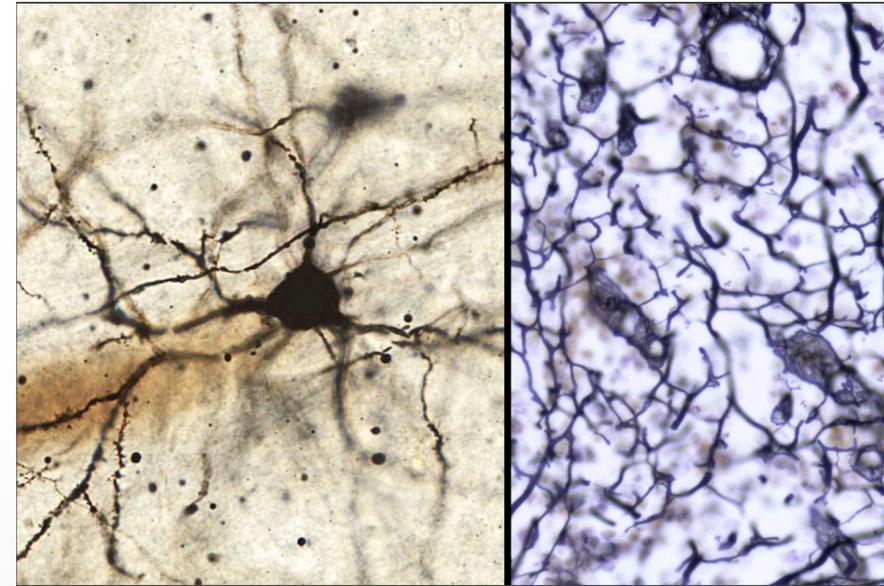
Staining and Stains-special stains

- **Trichrome** stains allow greater distinctions among various extracellular tissue components, e.g., Masson trichrome.
- **The periodic acid-Schiff (PAS)** reaction utilizes the hexose rings of polysaccharides and other carbohydrate-rich tissue structures and stains such macromolecules distinctly purple or magenta.



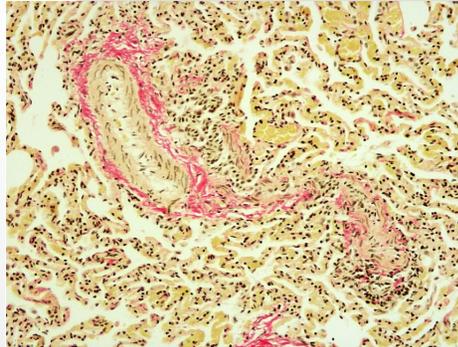
Staining and Stains-special stains

- **Sudan black:** lipid-soluble dyes --stains lipids; avoiding the processing steps that remove lipids, such as treatment with heat and organic solvents which can be useful in diagnosis
- **Metal impregnation:** less common methods. Using solutions of silver salts to visualize certain ECM fibers and specific cellular elements in nervous tissue.
- **Immunostaining:** immunofluorescence and immunohistochemistry.

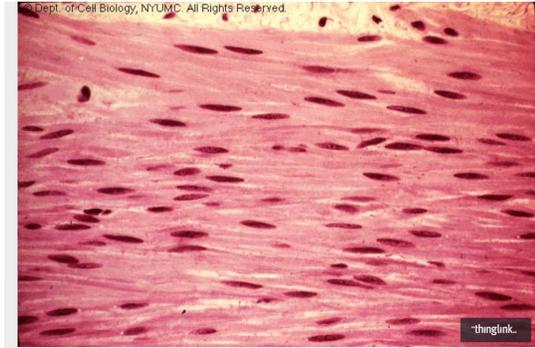
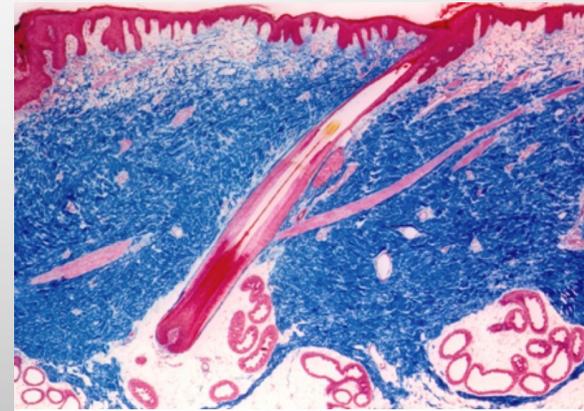


Examples Of Commonly Used Histological Stains

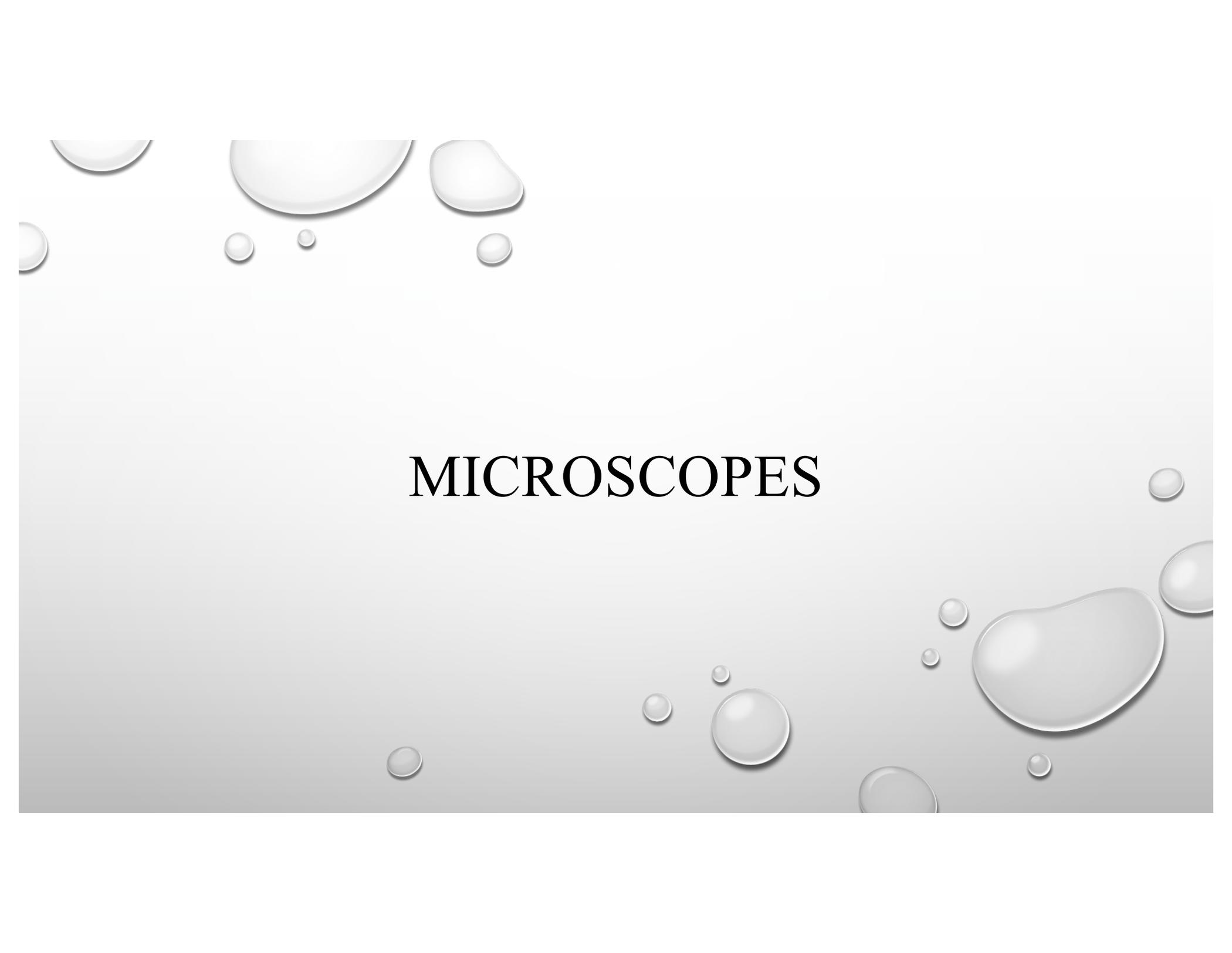
- Van Gieson method: collagen/pink, muscle/yellow.



- Trichrome method: three color system to emphasize support fibers: connective tissue/blue, cytoplasm/pink, nuclei/dark brown.



- Hematoxylin and eosin (H&E): nucleus/blue, cytoplasm/pink

The background of the slide is a light gray gradient. It is decorated with several realistic water droplets of various sizes and shapes, scattered across the top and bottom edges. The droplets have highlights and shadows, giving them a three-dimensional appearance.

MICROSCOPES

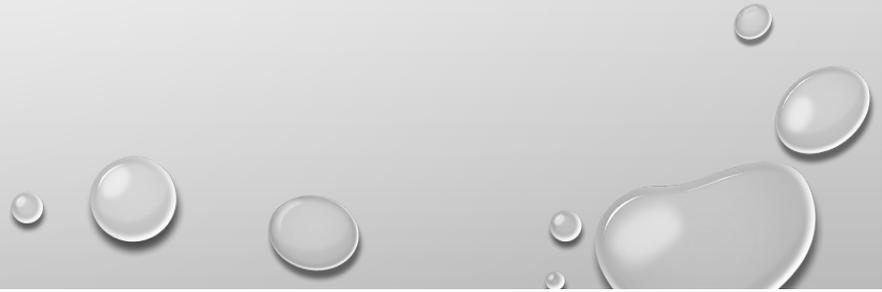


Types of microscope

- **Light microscope.**

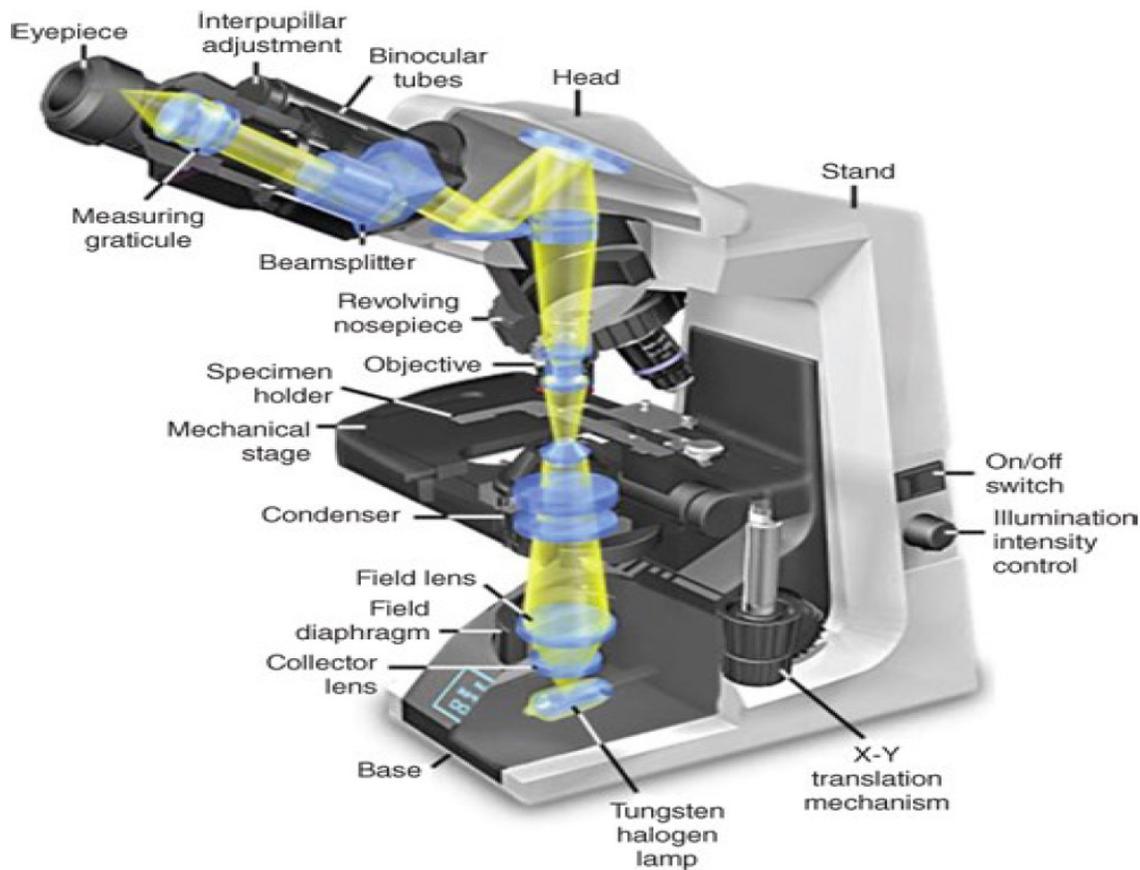
1. Bright-field microscopy
2. Fluorescence microscopy
3. Phase-contrast microscopy
4. Confocal microscopy
5. Polarizing microscopy

- **Electron microscope**

1. Transmission electron microscopy
 2. Scanning electron microscopy
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Light Microscope (Bright-field)

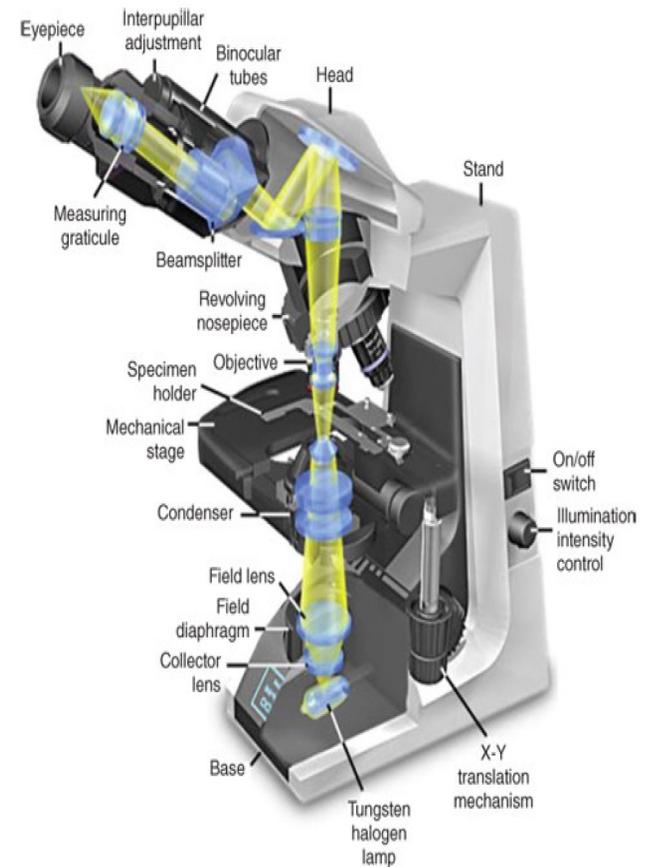
Components and light path of a bright-field microscope.



Bright-field Light Microscope

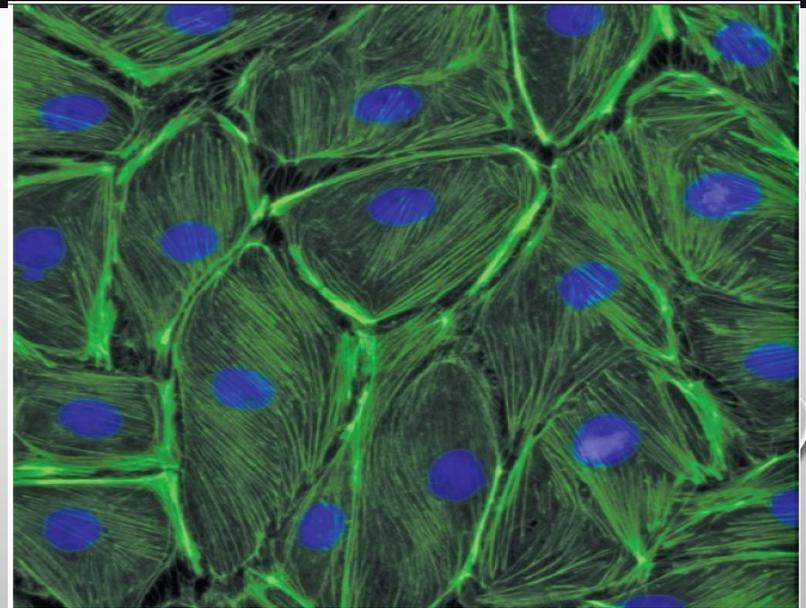
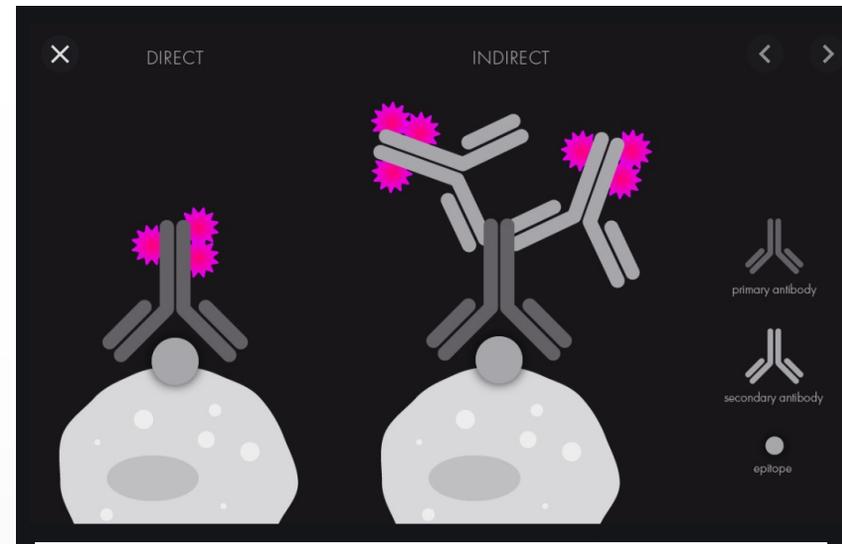
- Stained tissue is examined with ordinary light passing through the preparation.
- Includes an optical system and mechanisms to move and focus the specimen.
- The **condenser** collects and focuses a cone of light that illuminates the tissue slide on the stage.
- **Objective** lenses enlarge and project the illuminated image of the object toward the eyepiece.
- The two **eyepieces** or oculars magnify this image another 10X and project it to the viewer, yielding a total magnification of 40X, 100X, or 400X.

Components and light path of a bright-field microscope.



Fluorescence Microscopy

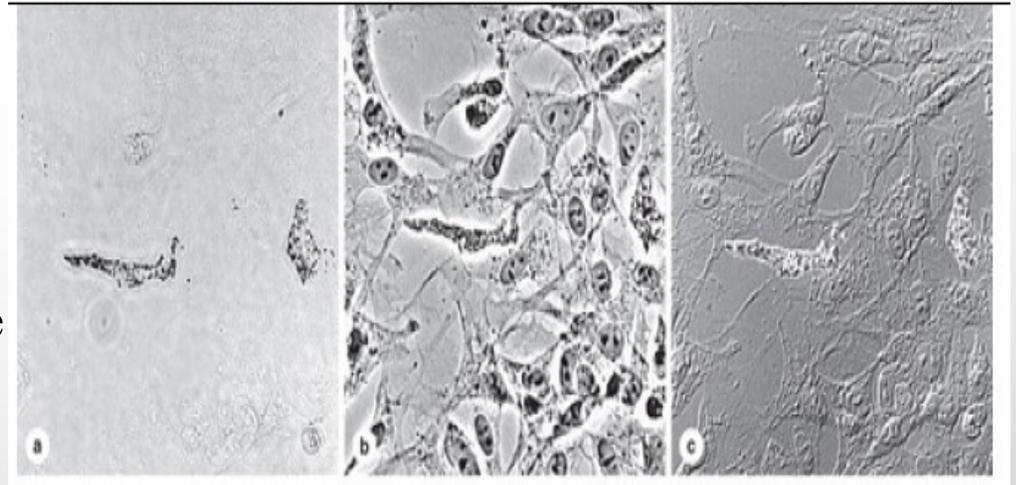
- **Fluorescence:** when certain cellular substances are irradiated by light of a proper wavelength, they emit light with a longer wavelength.
- In fluorescence microscopy, tissue sections are irradiated with
- Ultraviolet (UV) light and the emission is in the visible portion of the spectrum.
- The fluorescent substances appear bright on a dark background.
- For fluorescent microscopy the instrument has a source of UV or other light and filters that select rays of different wavelengths emitted by the substances to be visualized.



Phase-contrast Microscopy



- Study unstained cells and tissue sections (colorless; similar optical densities).
- Uses a lens system that produces visible images from transparent objects and can be used with living, cultured cells.
- Is based on the principle that light changes its speed when passing through cellular and extracellular structures with different refractive indices--- appear lighter or darker in relation to each other.



Electron Microscope

- Interaction of tissue with a beam of electrons.

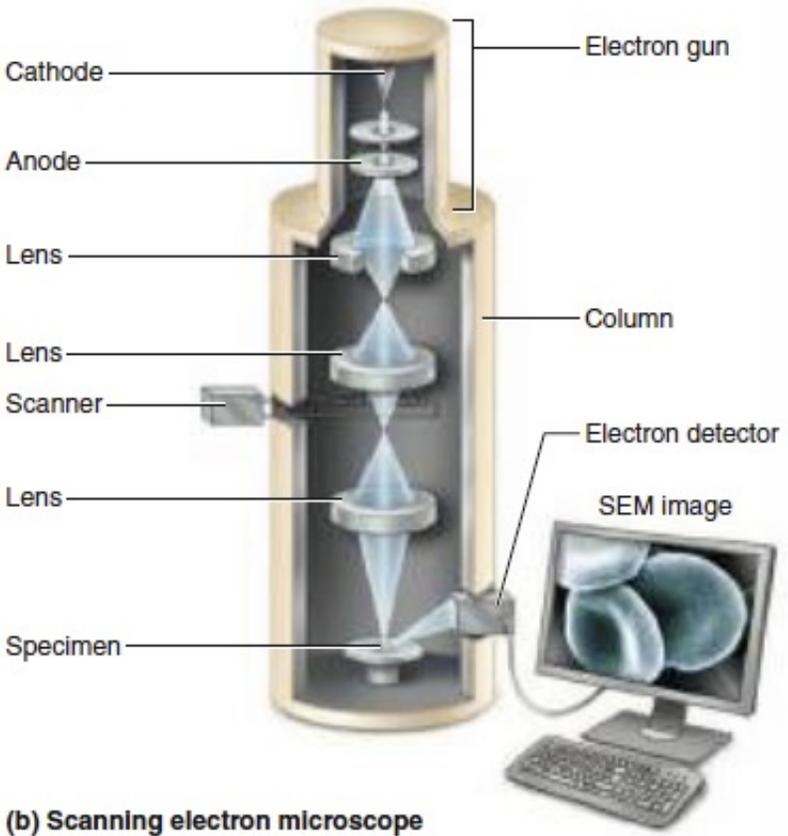
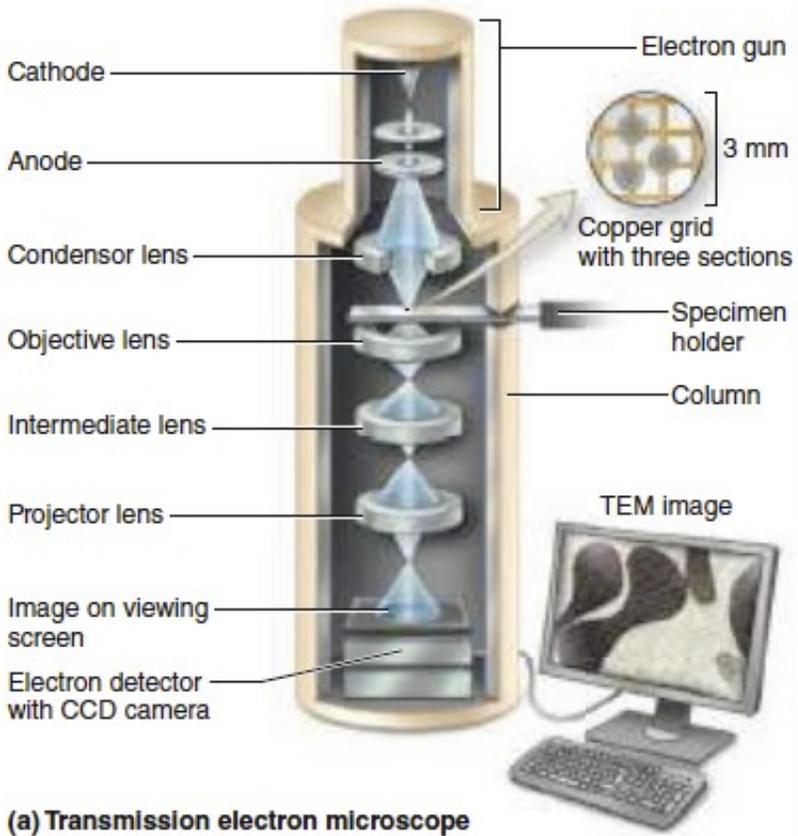
TEM

- The electron beam passes the tissue.
- Very high magnification
- Very thin sections, 40-90 nm.
- Electron beam interact with tissue producing black, white and shades of gray images.

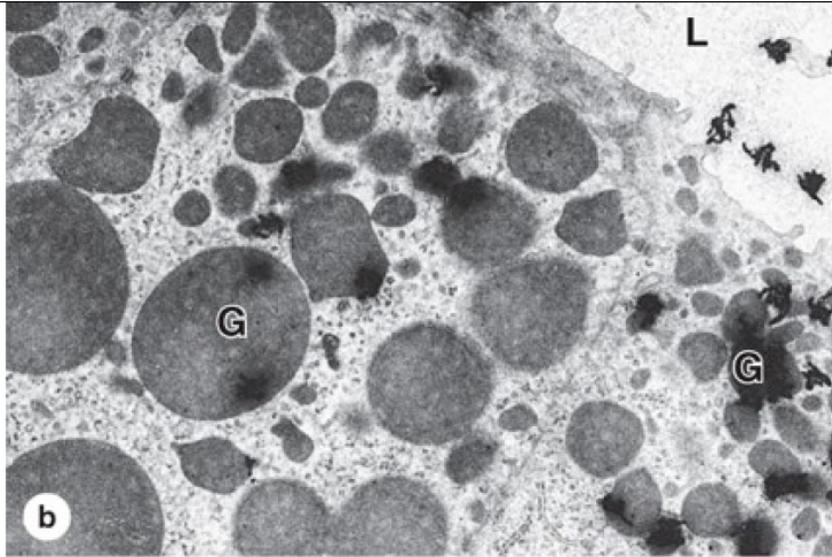
SEM.

- The electron beam does not pass the tissue.
- The surface of cells and tissue is coated with heavy metals (gold)---which reflect the electrons---producing 3D images which is a recording of the specimen topography.

Electron Microscope

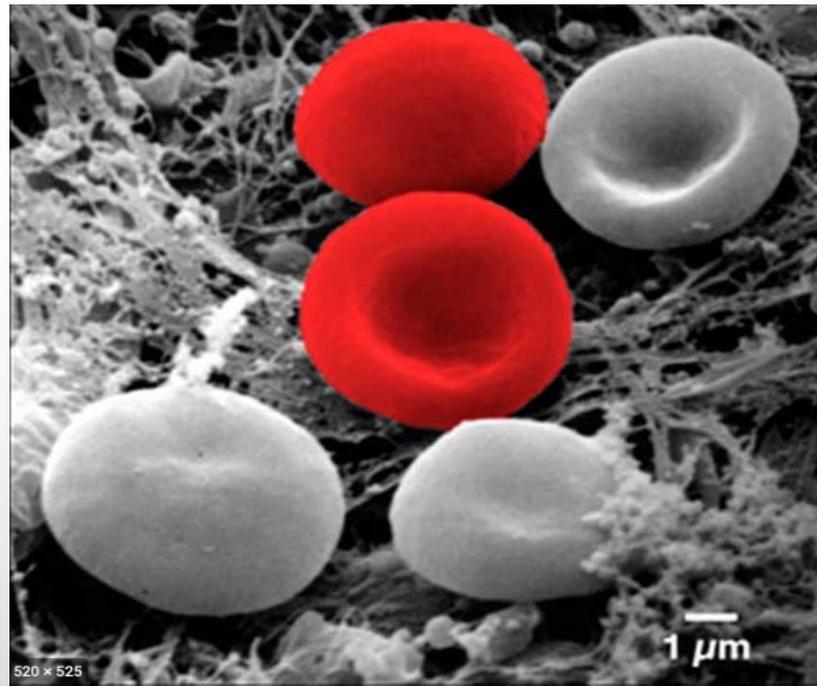


TEM



Source: Anthony L. Mescher: Junqueira's Basic
Histology: Text and Atlas, 15th Edition.
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SEM



Resolution

- **Resolving power:** the smallest distance between two structures at which they can be seen as separate objects.
- The maximal resolving power of the light microscope is approximately $0.2\ \mu\text{m}$ --- can permit clear images magnified 1000-1500 times.
- Objects smaller or thinner than $0.2\ \mu\text{m}$ (such as a single ribosome or cytoplasmic microfilament) cannot be distinguished.
- The microscope's resolving power determines the quality of the image, its clarity and richness of detail, and depends mainly on the quality of its objective lens.
- Magnification is of value only when accompanied by high resolution.
- Resolving of TEM is $3\ \text{nm}$ (electron wavelength is shorter than that of light).

