Introduction to Histology

By:
Musab Nureldeen Husein
MBBS, MSc

Definition

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

Tissue components

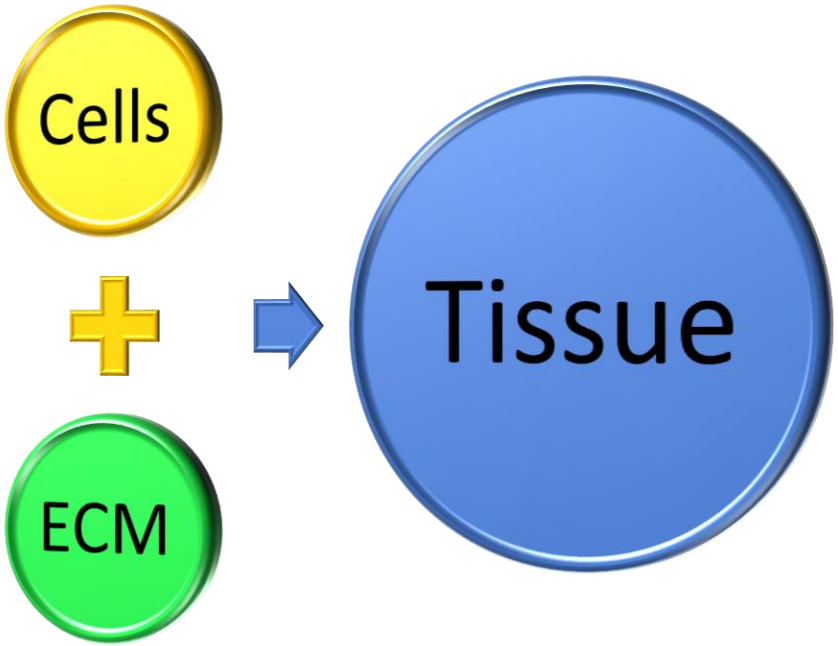
- Tissues have two interacting components:

1- cells

2- Extracellular matrix (ECM)

- The cells consists of organelles (e.g. Nucleus and endoplasmic reticulum). The ECM consists of many kinds of macromolecules that form complex structures (e.g. collagen fibrils and basement membranes).

Cells produce ECM while the ECM supports the cells and the fluid that transports nutrients to the cells, and carries away their catabolites and secretory products.



From cell to human body

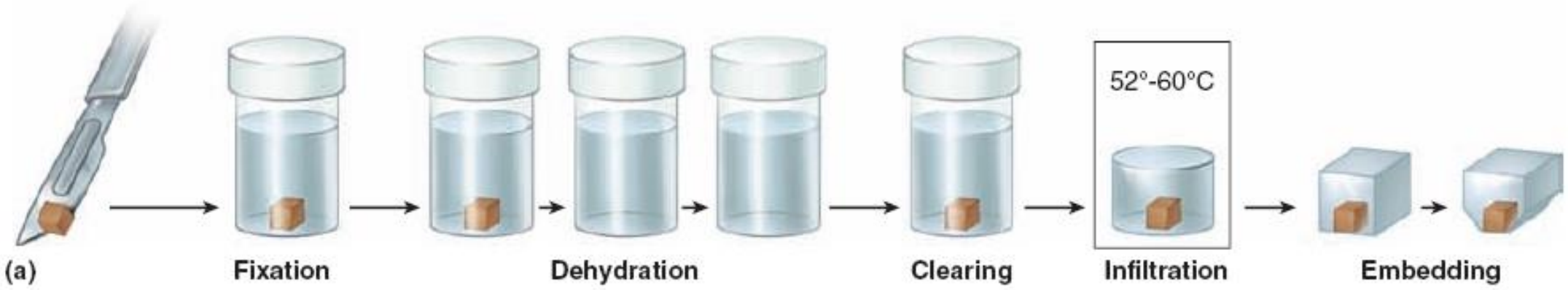
- Cells constitute tissues
- Tissues constitute organs
- Organs constitute systems
- Systems constitute body

Types of tissue

1. Epithelial tissue.
2. Connective tissue
3. Muscular tissue.
4. Nervous tissue.

Tissue Preparation

1. Fixation
2. Dehydration
3. Clearing
4. Infiltration
5. Embedding
6. Trimming
7. Sectioning
8. Staining



1- Fixation

- It is the process of immersion of tissue in solutions of stabilizing or crosslinking compounds called **fixatives**.
- Purpose: To avoid tissue digestion by degradative enzymes within the cells (autolysis) or bacteria.
- Examples: Formaldehyde (Formalin) and glutaraldehyde.

2- Dehydration

- It is the process of extraction of water from the fixed tissues by successive transfer through a graded series of ethanol (alcohol) and water mixtures usually from 70% to 100%.

3- Clearing

- It is the process of removal of alcohol in toluene or other agents to make the tissue transparent.

4- Infiltration

- It is the process when the tissue is placed in melted paraffin until it becomes completely infiltrated with this substance.

5- Embedding

- It is the process when the Paraffin infiltrated tissue is placed in a small mold with melted paraffin and allowed to harden.
- Embedding materials include paraffin and plastic resins

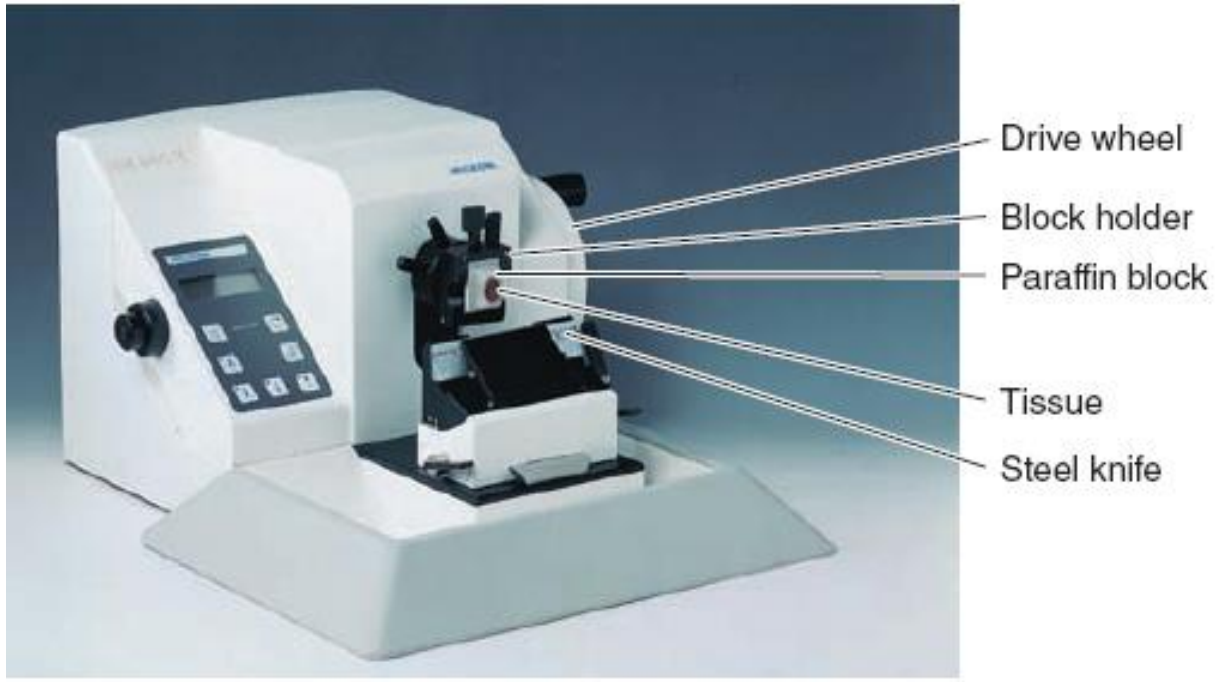
6- Trimming

- It is the process when the resulting paraffin block is trimmed (remove excess and decorate) to expose the tissue for sectioning (slicing).

7- Sectioning

- Because most tissues and organs are too thick for light to pass through them, they must be sliced to obtain thin, translucent sections that are attached to glass slides for microscopic examination.

- This is done by **Microtome**



8- Staining

- Most cells and extracellular material are completely colorless, and to be studied microscopically sections must typically be stained (dyed).
- Purpose: Visualization and permit distinctions to be made between the various tissue components.

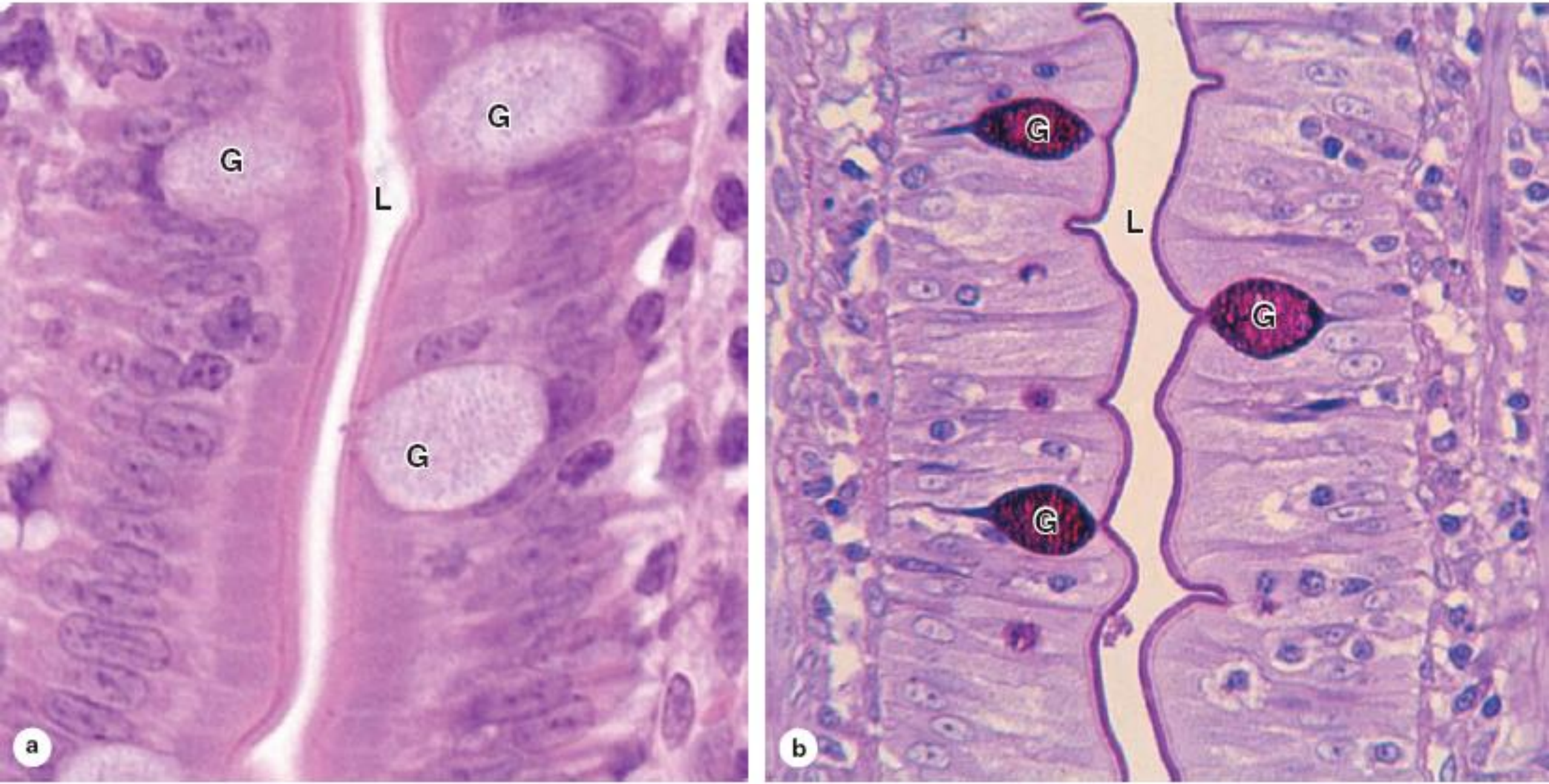
Stain types

1. Basic dyes: E.g. Hematoxylin, toluidine blue, alcian blue and methylene blue
2. Acid dyes: E.g. Eosin, orange G, and acid fuchsin
3. Periodic acid Schiff (PAS)
4. Lipid soluble dyes: E.g. Sudan black
5. Metal impregnation techniques

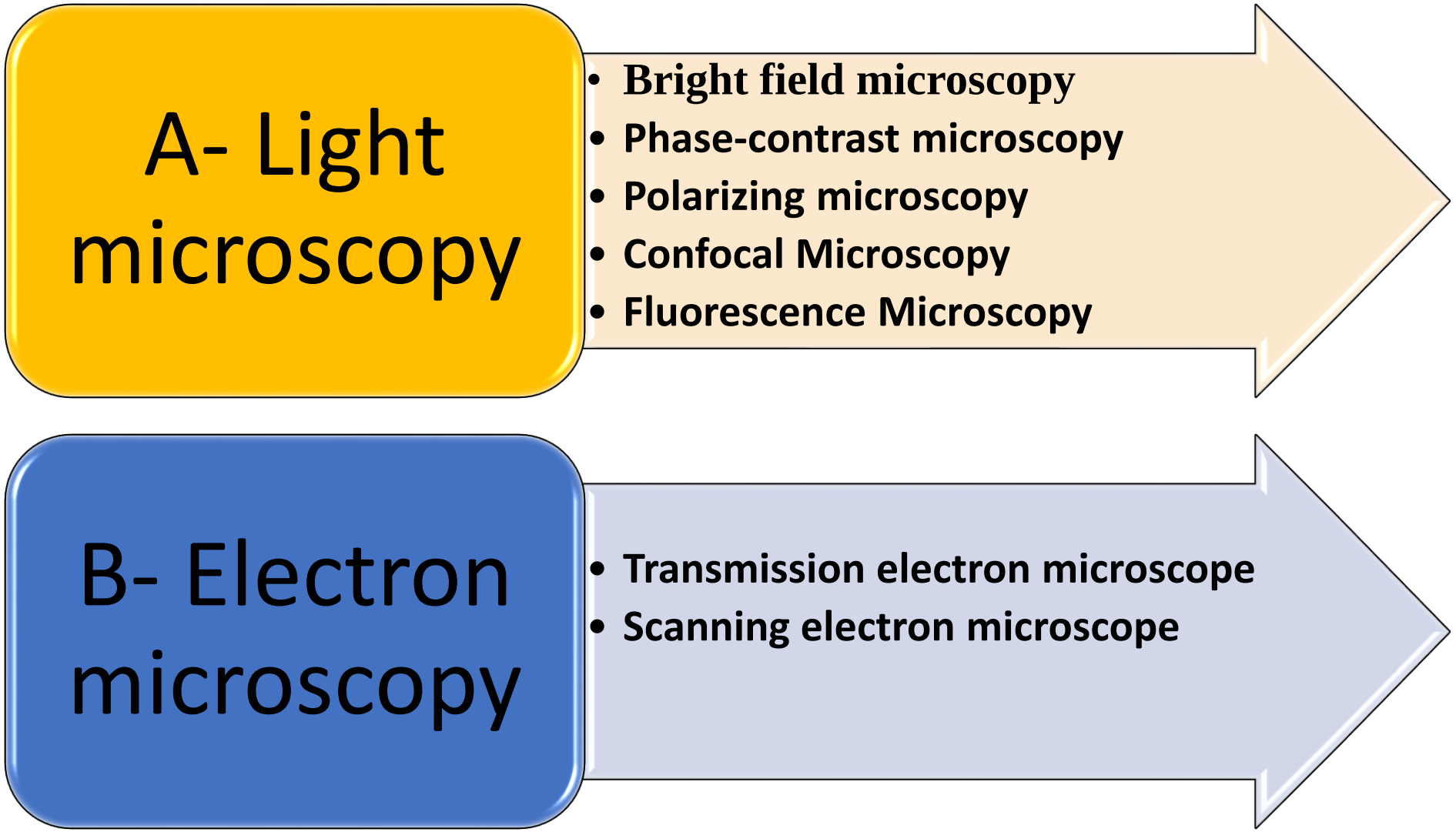
Hematoxylin and eosin (H&E)

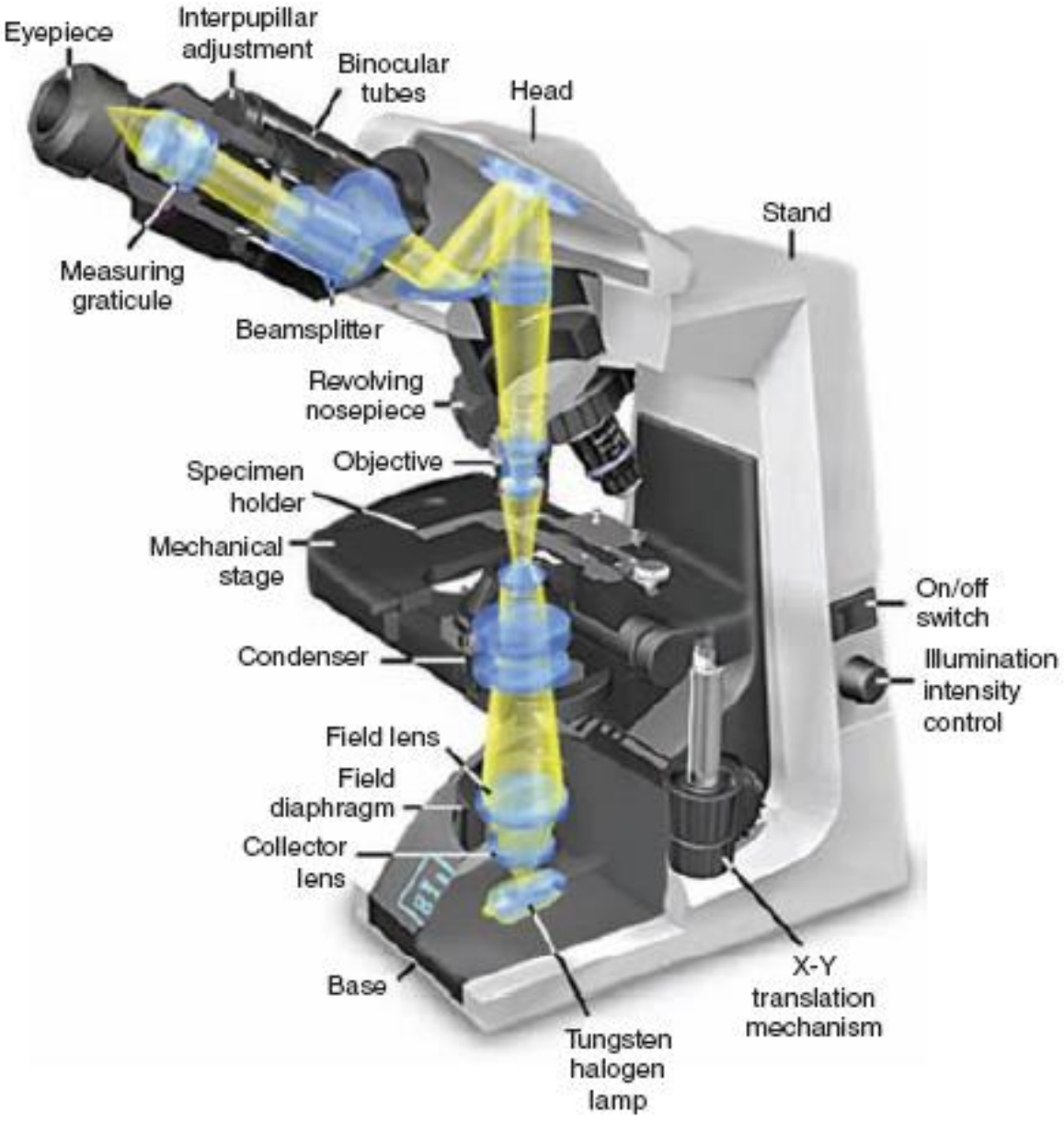
- This simple combination of hematoxylin and eosin (H&E) is used most commonly.
- Hematoxylin produces a dark blue or purple color, staining DNA in the cell nucleus and other acidic structures (such as RNA-rich portions of the cytoplasm and the matrix of cartilage).
- Eosin stains other cytoplasmic components and collagen with pink color.

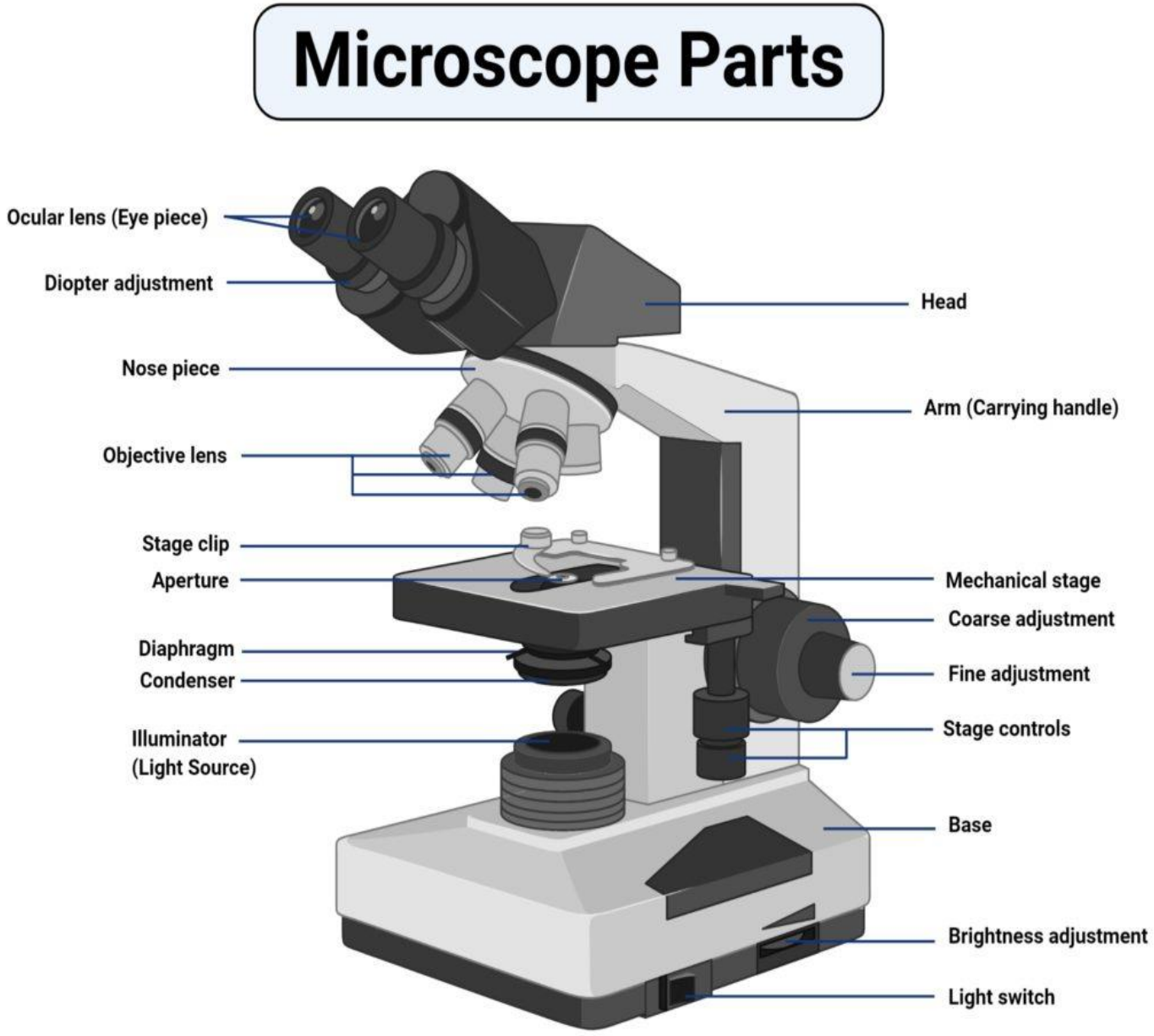
FIGURE **1–2** **Hematoxylin and eosin (H&E) and periodic acid-Schiff (PAS) staining.**



Types of microscopy





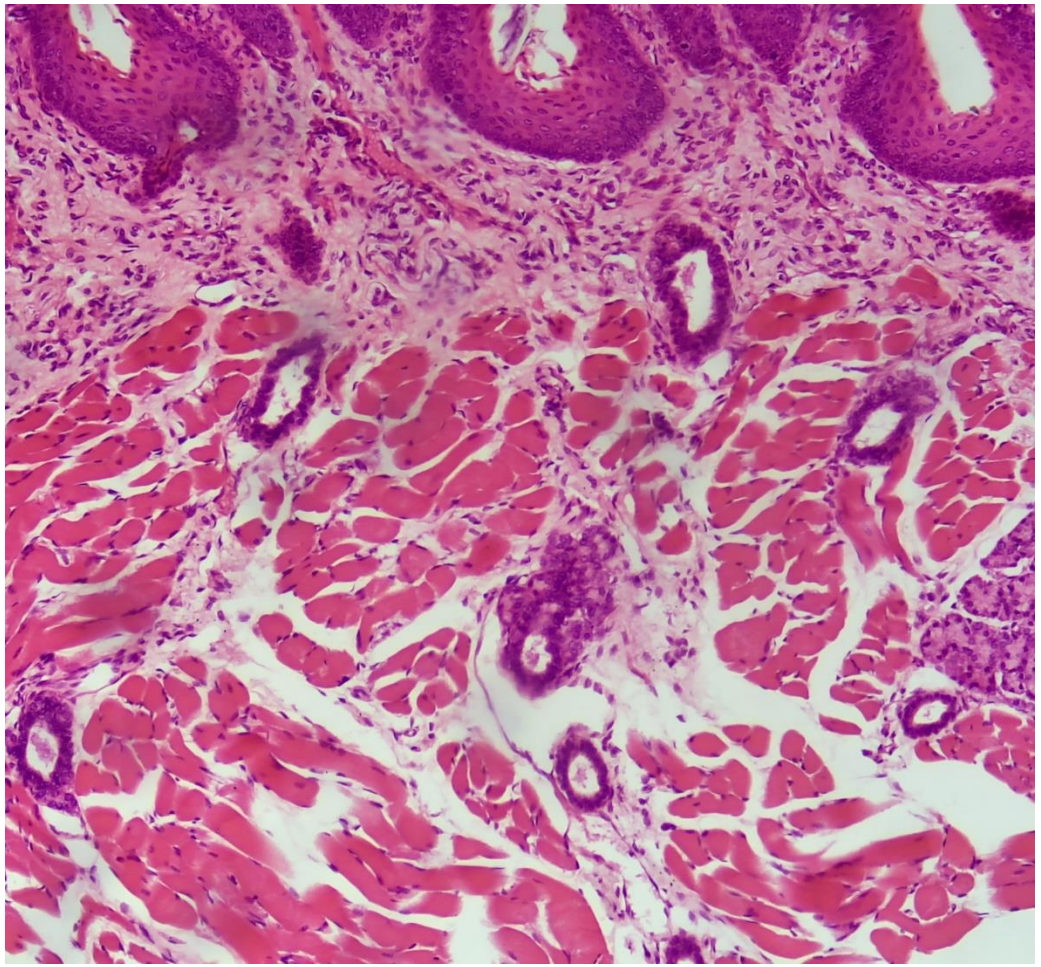


Light microscope

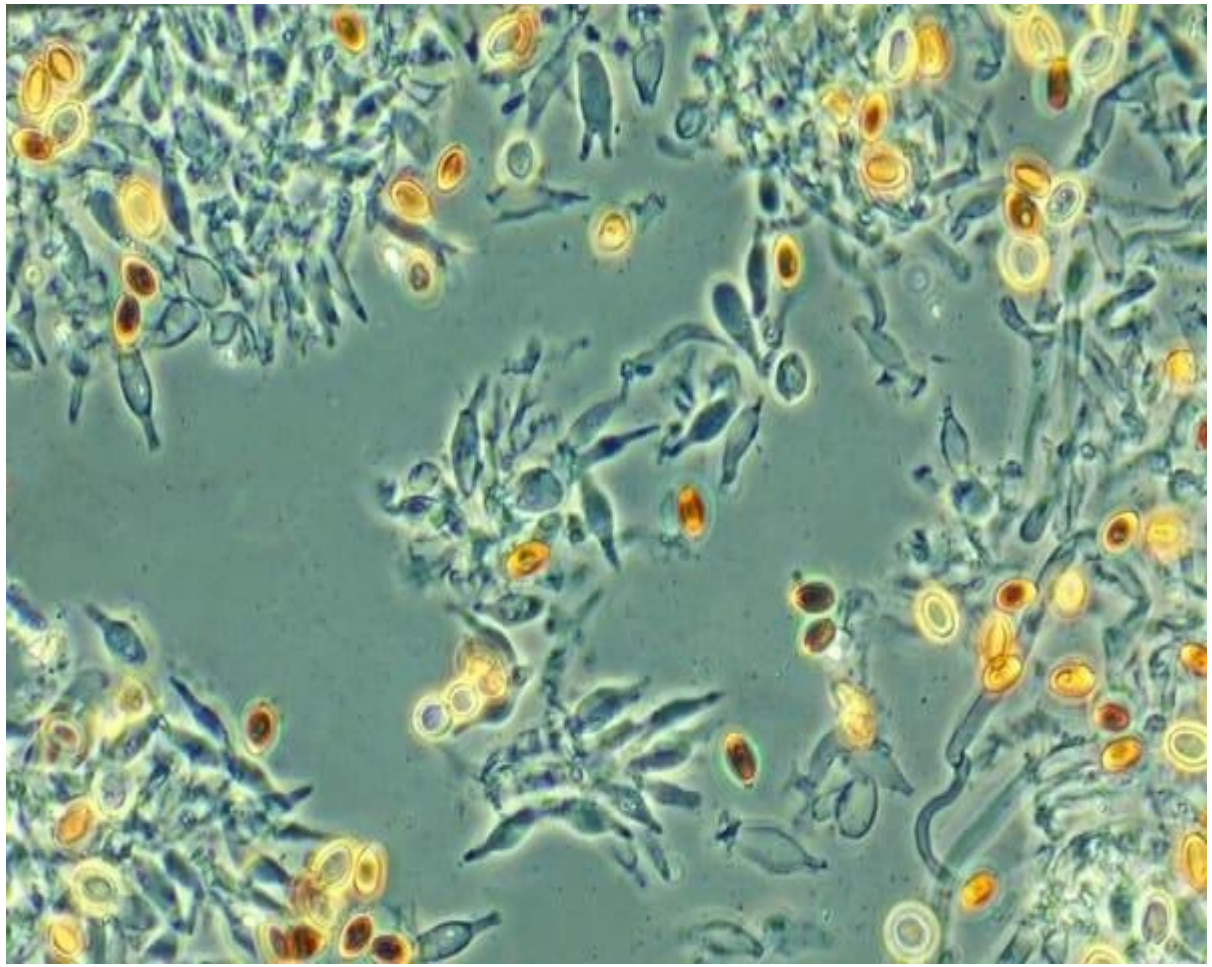
Types of Light microscopy:

1. **Bright field microscopy:** widely used by students in which stained preparations are examined by means of ordinary light that passes through the specimen.
2. **Phase-contrast microscopy**: light changes speed when passing through cellular structures with different refractive indices.

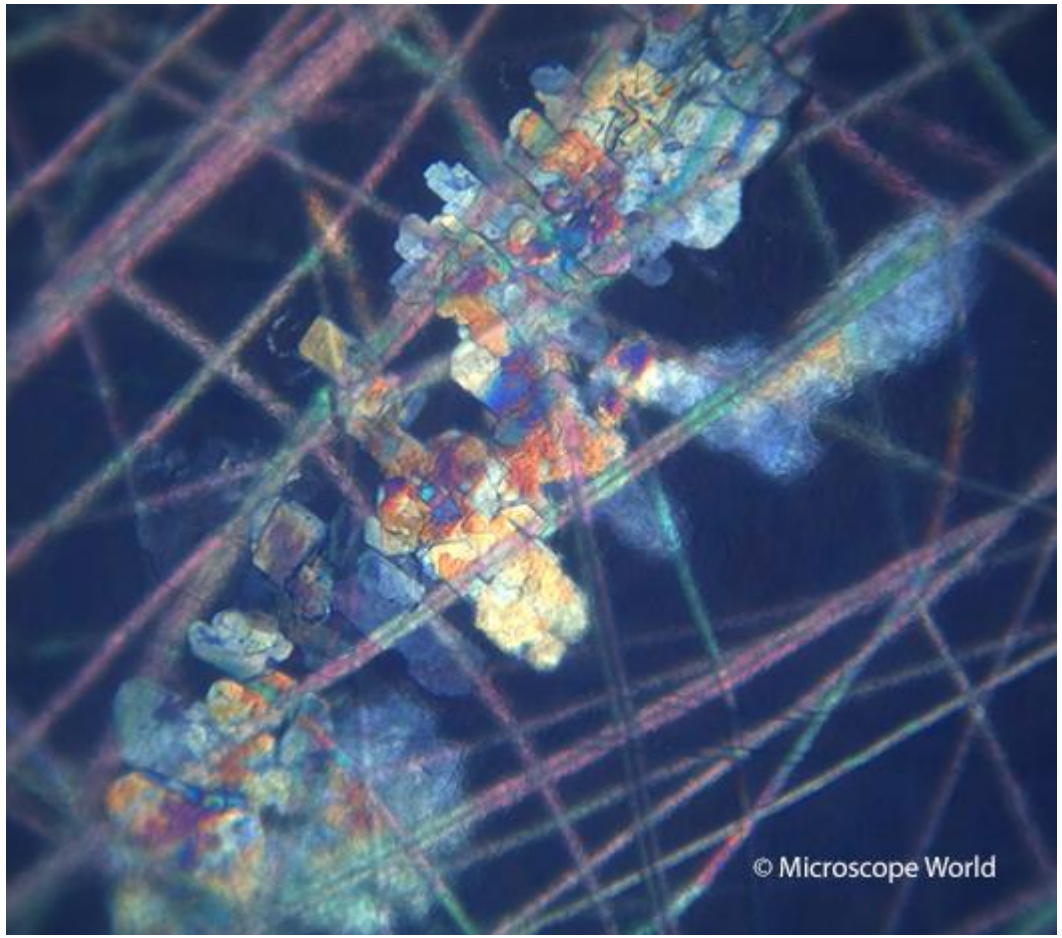
3. **Polarizing microscopy**: for structures that have highly organized molecules (e.g. collagen, microtubules, and microfilaments), these structures have the ability affect the polarized light.
4. **Confocal Microscopy:** only one very thin plane of the specimen is focused at a time.
5. **Fluorescence Microscopy:** using ultraviolet (UV) light or laser to irradiate tissues, the structure appears bright on dark background.



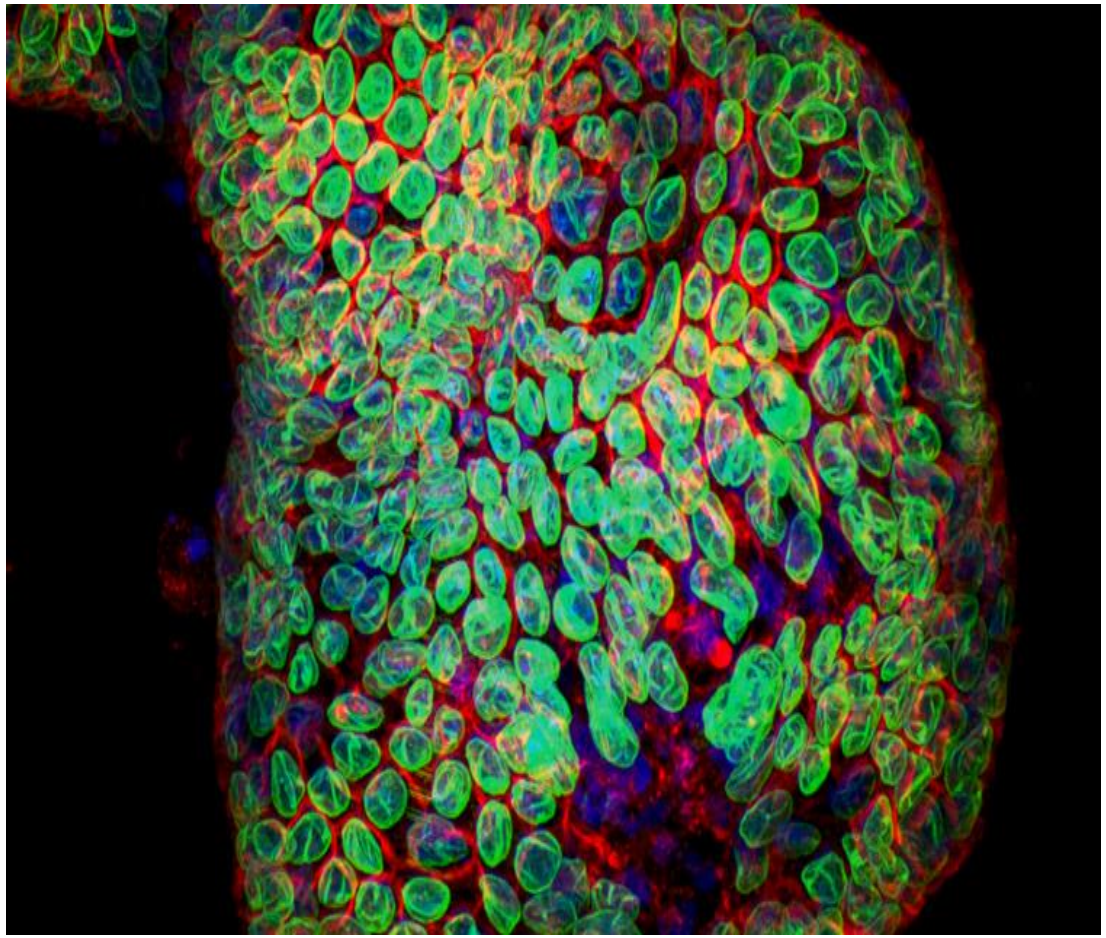
Bright field microscopy



Phase contrast microscopy

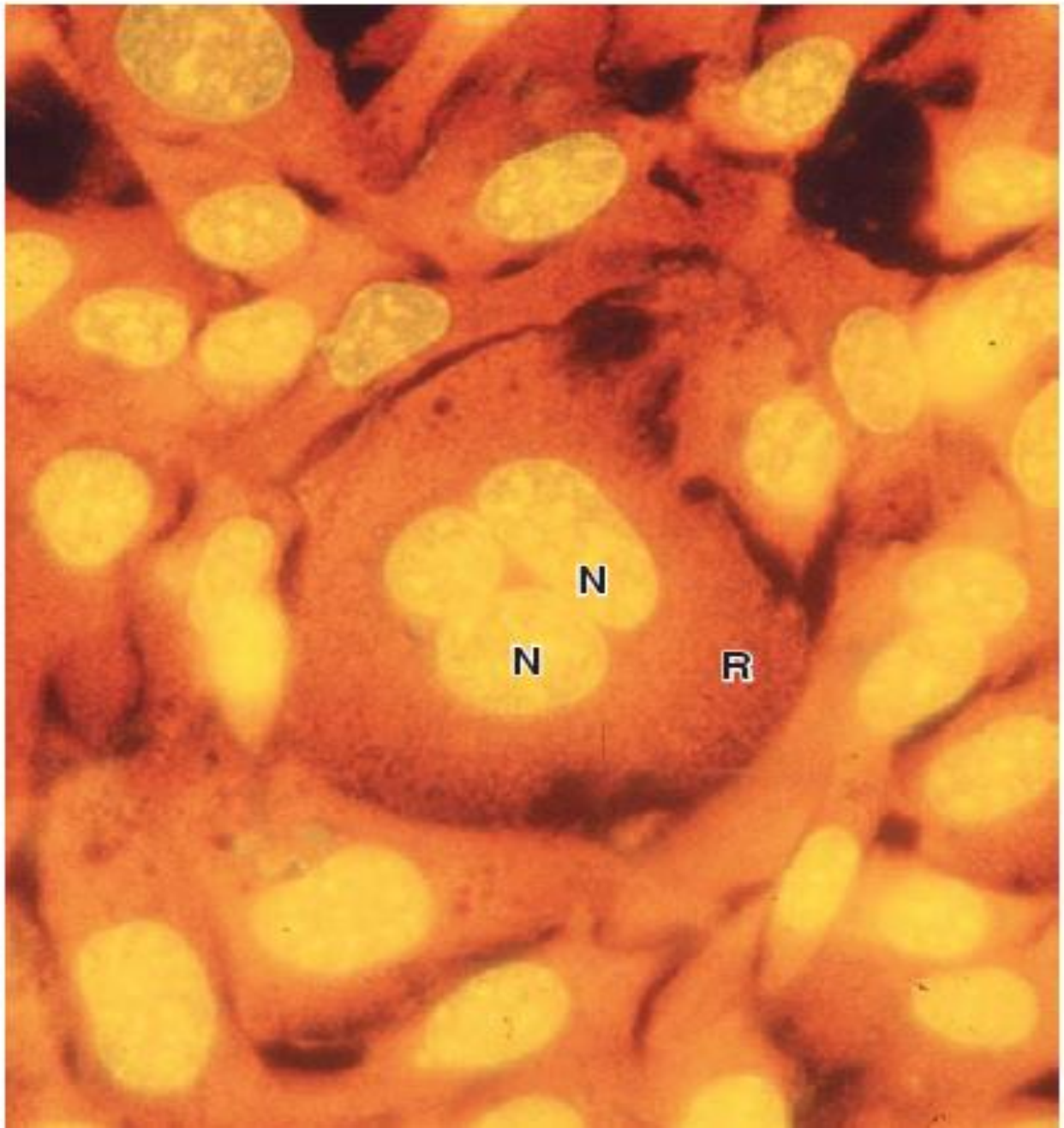


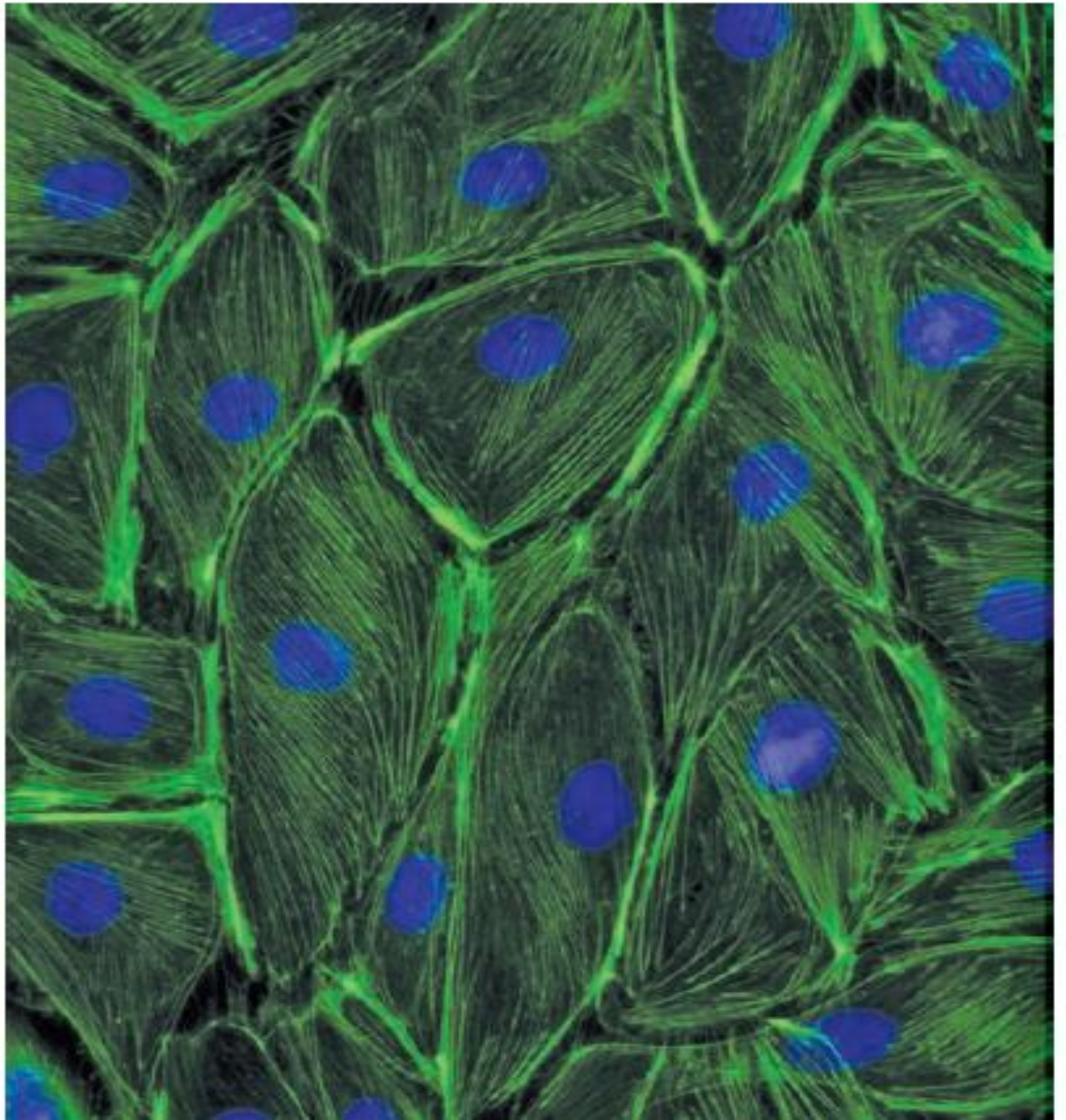
Polarizing microscopy



Confocal Microscopy

FIGURE **1–4** **Appearance of cells with fluorescent microscopy.**





Electron microscopy

- **Technique**: Based on the interaction of tissue components with beams of electrons.
- The wavelength in the electron beam is much shorter than that of light, allowing a 1000-fold increase in resolution.

Types of electron microscopy

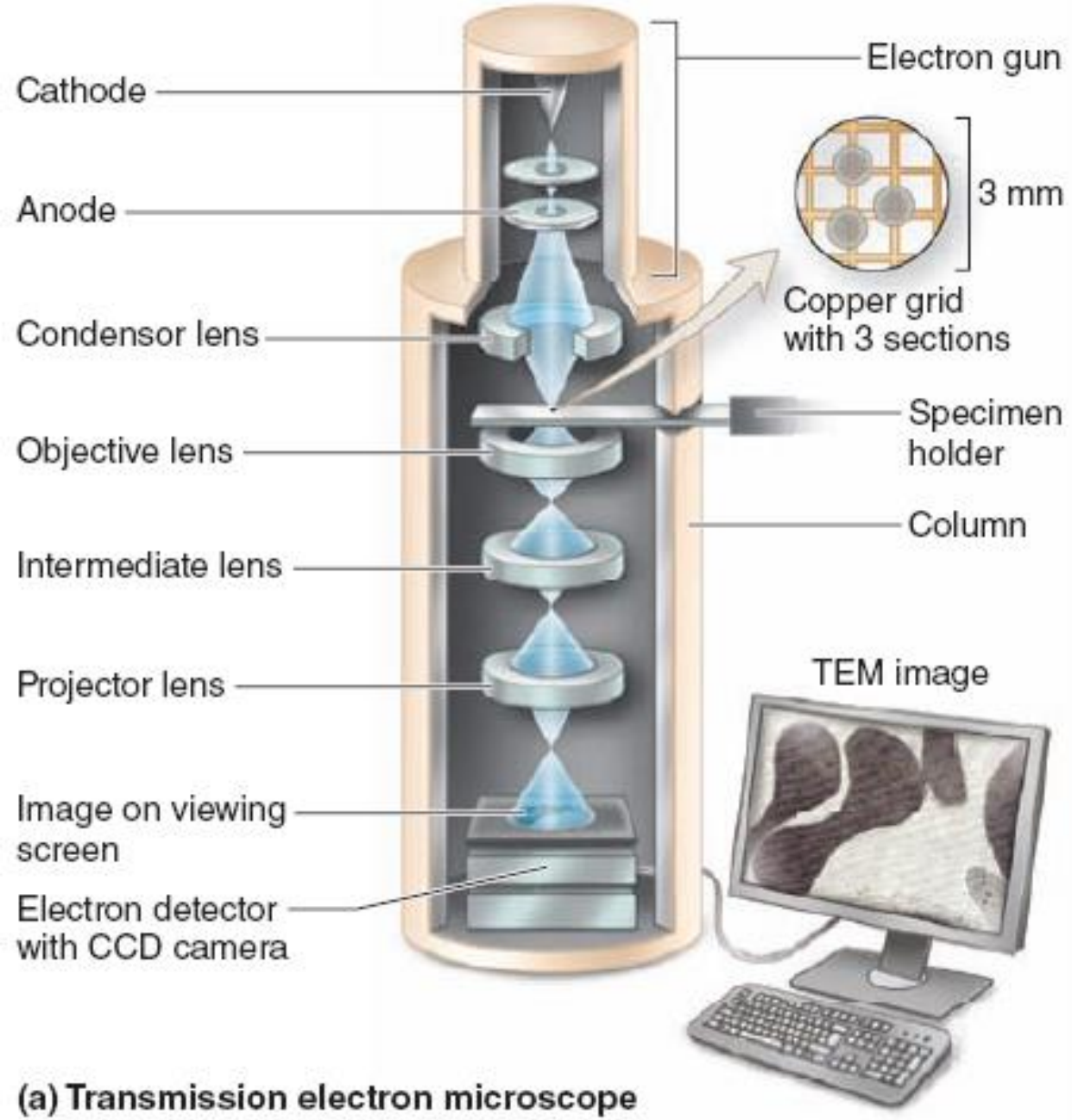
1- Transmission Electron Microscope (TEM):

A metallic filament cathode emits electrons that move toward an anode, a metal plate with a central hole that forms a beam of electrons. Electrons transmitted through the specimen reach the objective lens, which forms a focused, magnified image that is then magnified further through other lenses and captured on a viewing screen.

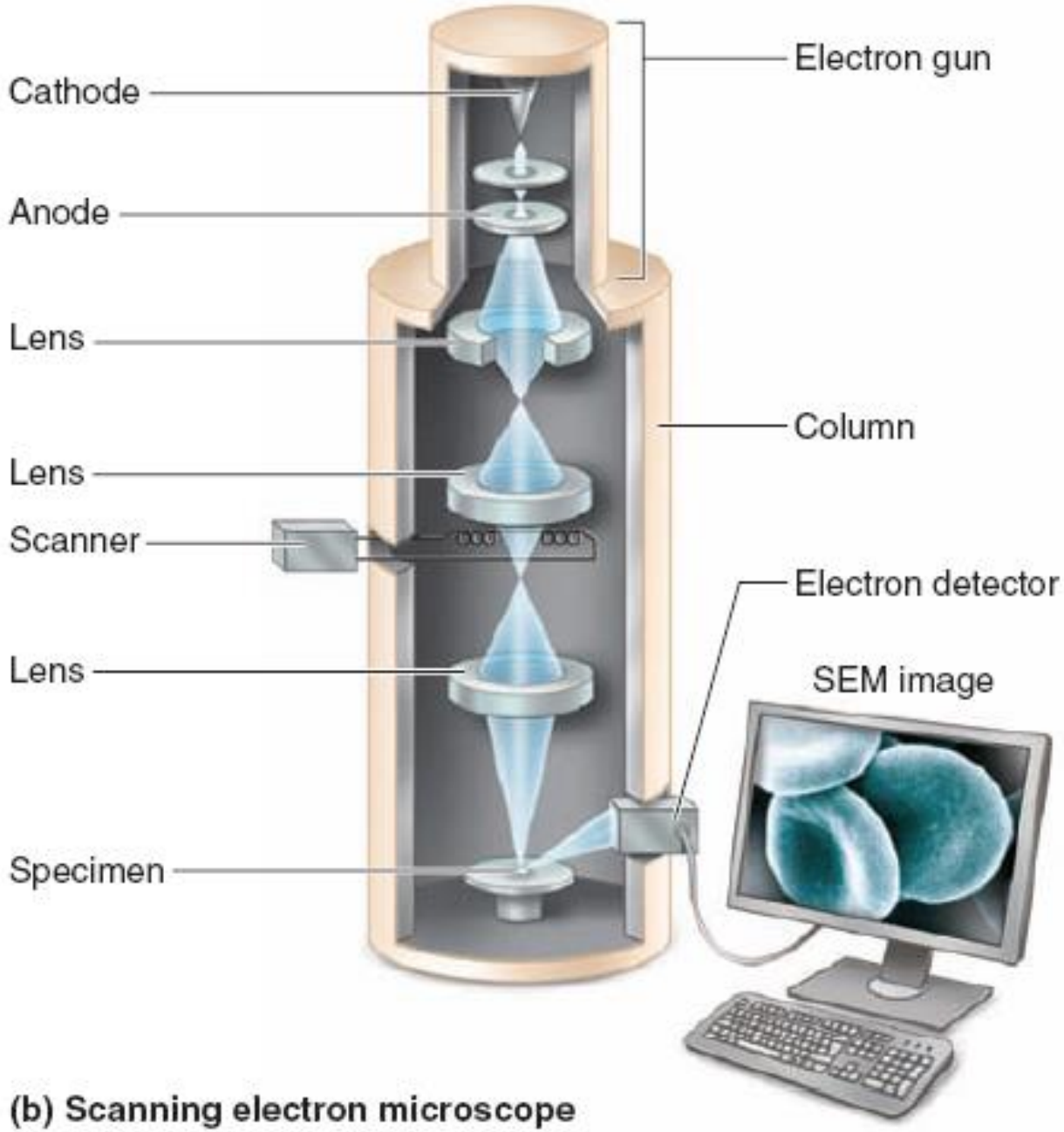
Types of electron microscopy

2- Scanning electron microscopy (SEM):

The beam does not pass through the specimen. Instead, the surface of the specimen is first dried and spray-coated with a very thin layer of heavy metal (often gold) through which electrons do not pass readily. The beam is scanned across the specimen, it interacts with the metal atoms and produces reflected (secondary) electrons emitted from the metal to produce 3D view.



(a) Transmission electron microscope



(b) Scanning electron microscope

Reference

- Anthony L. Mescher, Junqueira Basic Histology 13th edition McGraw – Hill’s 2013.