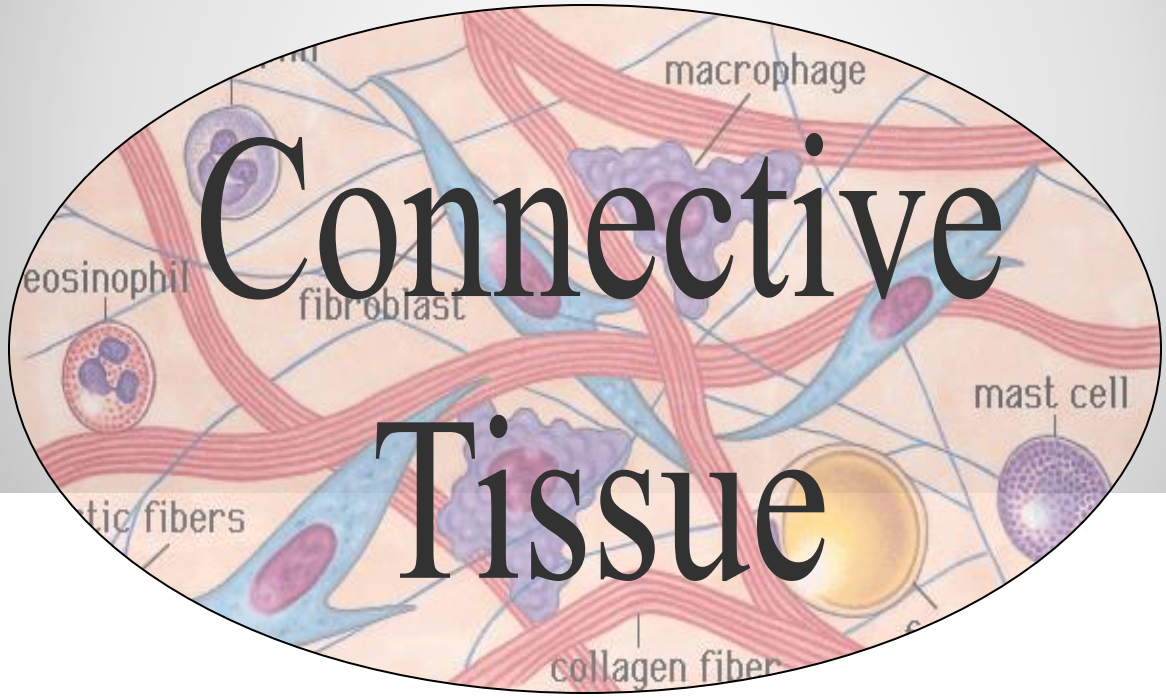
Connective Tissue



Dr. Mustafa Saad
(2024)

Connective tissue (CT) is a type of body tissue characterized by an abundant extracellular matrix within which are dispersed different types of cells and fibers.

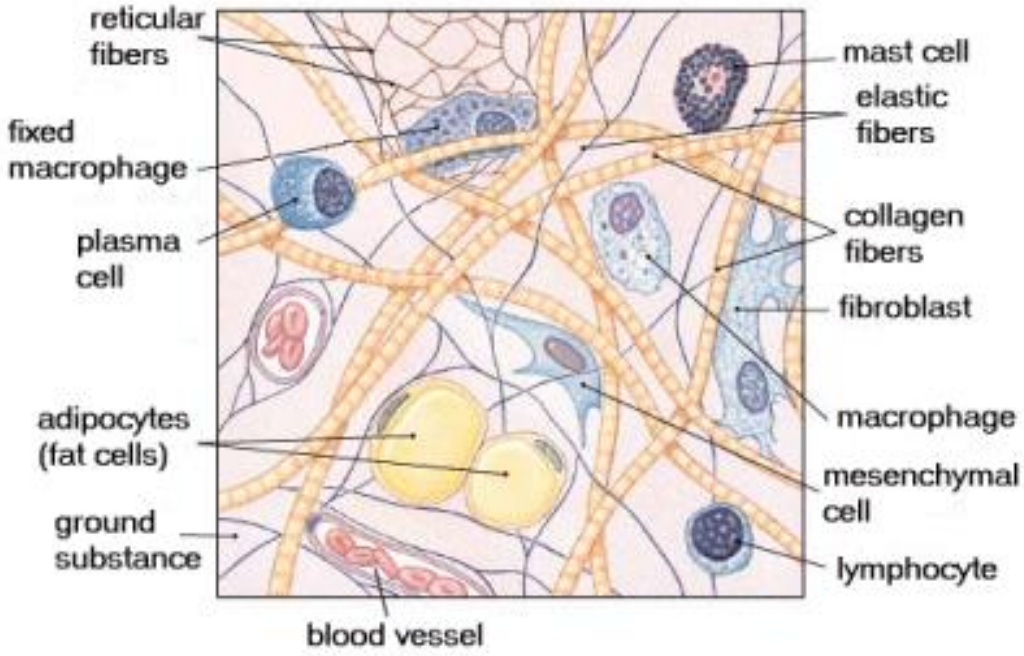


Fig.1: Image showing the components of CT: Cells, Fibers and Ground substance

Functions

1. Provide and maintain form of organs.
2. Support different tissues and organs.
3. Connect and bind different body regions.
4. Provide a medium for diffusion of nutrients and waste products.

The Cells Of The Connective Tissue

- *Cells of the CT are, usually, not regularly arranged.*
- The cells of CT could:
  - Originate and remain in the CT all their lives (fibroblasts).
  - Originate outside the CT and then come to the CT and remain in it for the rest of their long lives (mast cells).
  - Originate outside the CT and then come to the CT and remain in it for a short period (neutrophils).

1) Fibroblasts

- Most common cell in connective tissue.
- **Function:** ***Synthesizes fibers and produces components of extracellular matrix.***
- Active Fribroblasts and inactive Fibrocytes.
- Rarely divide. Mitosis resumes when they're needed under influence of several growth factors.

Fibro- = fiber. -blast = forming.

Fibroblasts:

- Abundant irregularly branched cytoplasm
- Large, pale-staining nucleus with prominent nucleolus
- Rich in RER (rough endoplasmic reticulum)
- Golgi apparatus well developed

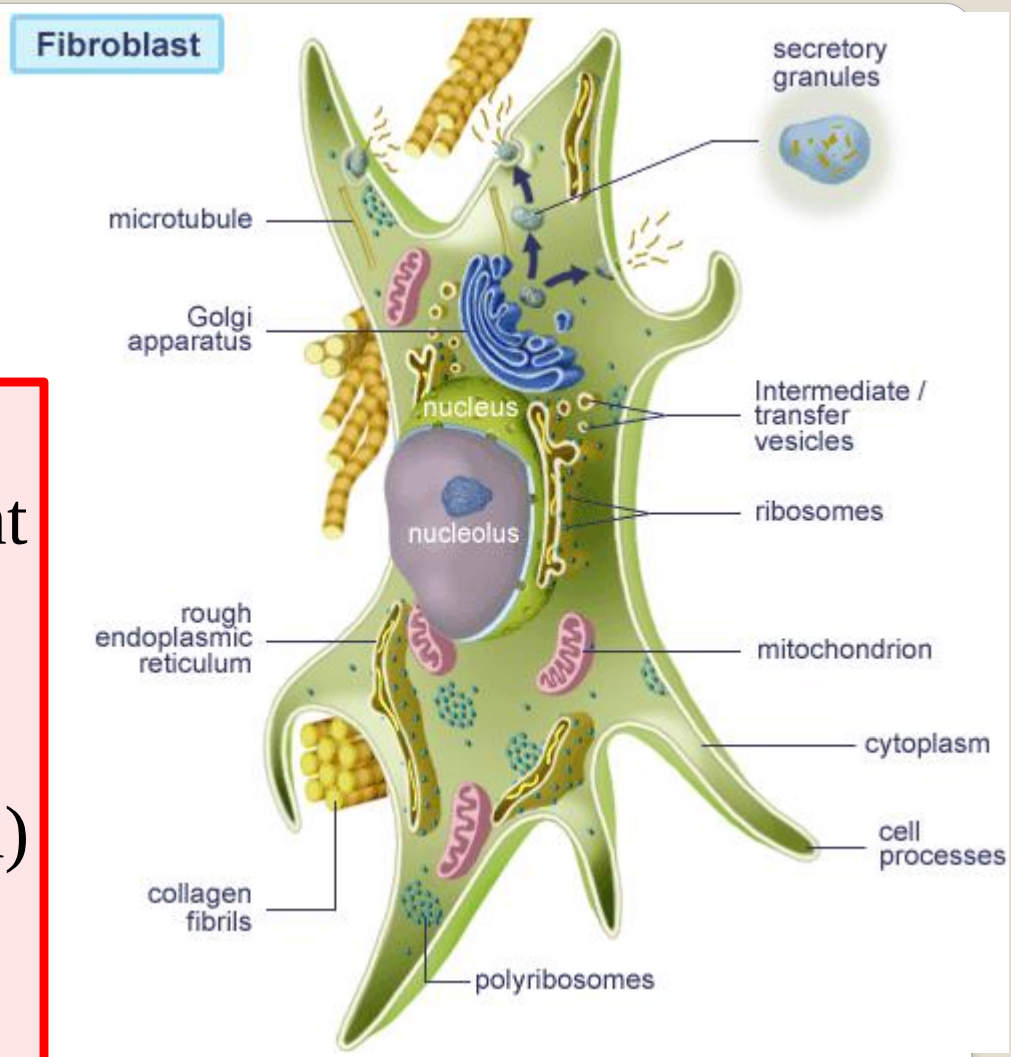


Fig.2: Histological features of fibroblasts.

Features of all protein producing cells

Fibrocytes:

- Smaller than fibroblasts.
- Less cytoplasmic processes.
- Nucleus smaller and darker.
- Less RER.

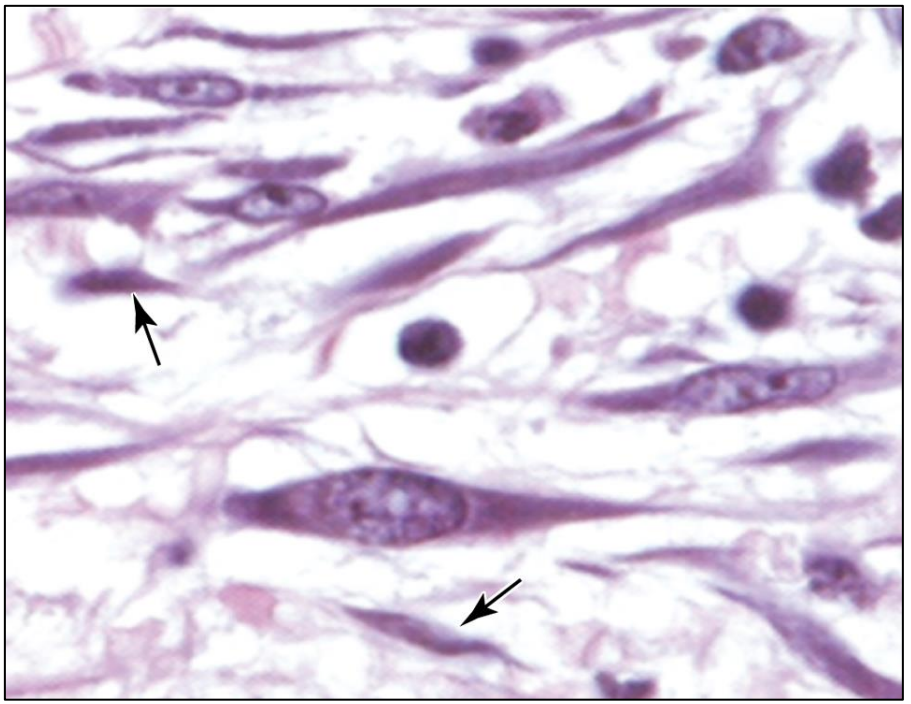


Fig.3: Fibroblasts and fibrocytes (arrows).

- ***Myofibroblasts:*** Fibroblast cells with contractile ability. Important in wound contraction.

-cyte = cell. Myo- = related to muscles, from greek *mys* = mouse (because movement of muscles resembles mice).

2) Macrophages and the Mononuclear phagocyte system

- Monocytes form in the bone marrow.
- Travel with blood and enter the connective tissues by passing through capillary walls.
- Activated monocytes will form several types of phagocytic cells in tissues.
- Macrophages in different tissues are given different names.

Macro- = large. Micro- = small. Phages = eaters. Mono- = one, single.

Macrophages

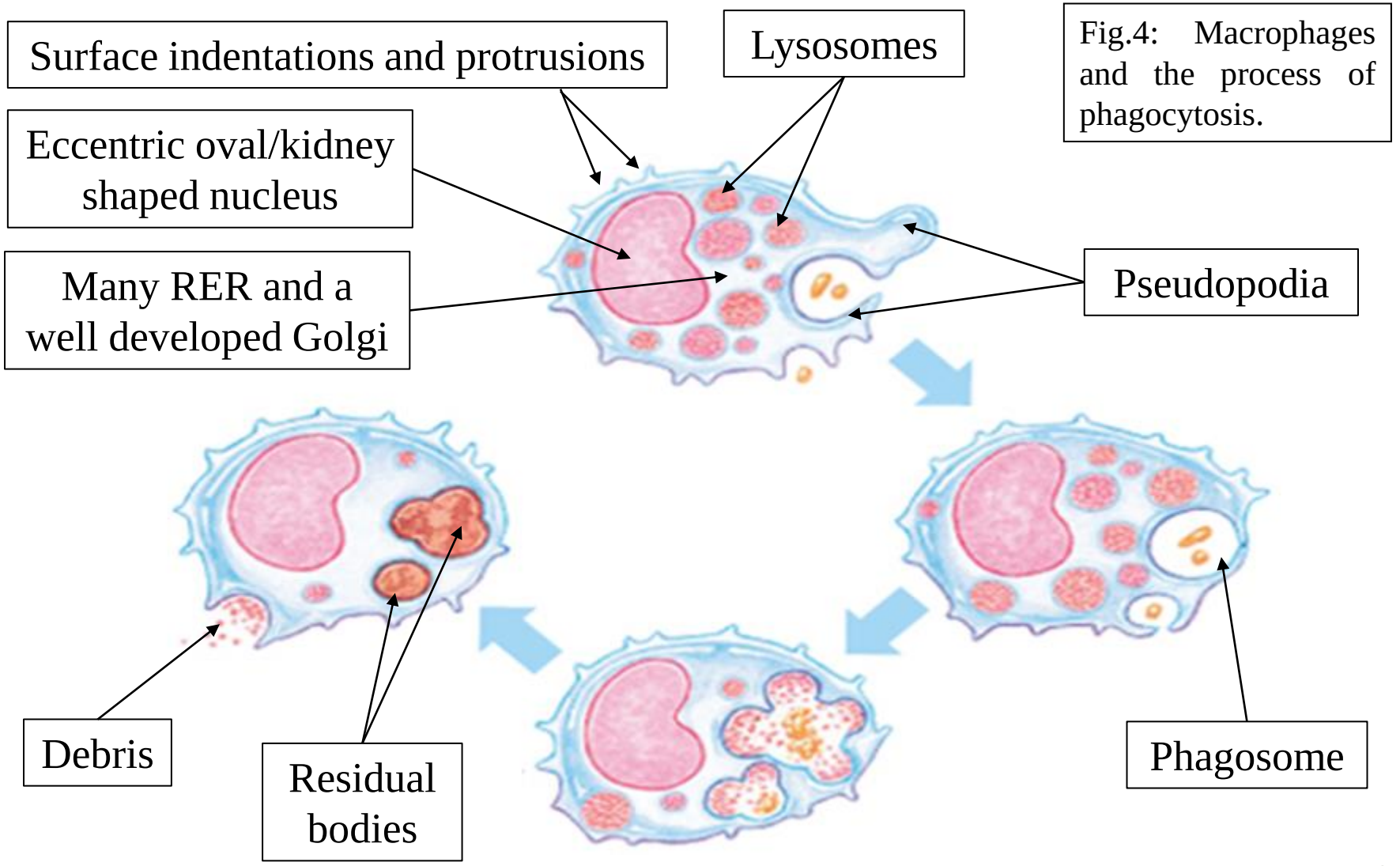


Fig.4: Macrophages and the process of phagocytosis.

Functions of Macrophages:

1) Phagocytosis (microorganisms, neoplastic cells, dead cells, debris, and abnormal extracellular elements).
2) Destruction of red blood cells (metabolism of iron and hemoglobin).
3) Antigen presentation to lymphocytes.
4) Release of cytokines and collagenases.

3) Mast Cells

- Large, oval or round cells. Cytoplasm filled with basophilic secretory granules.
- Nucleus small, spherical and centrally located (may be obscured by granules).

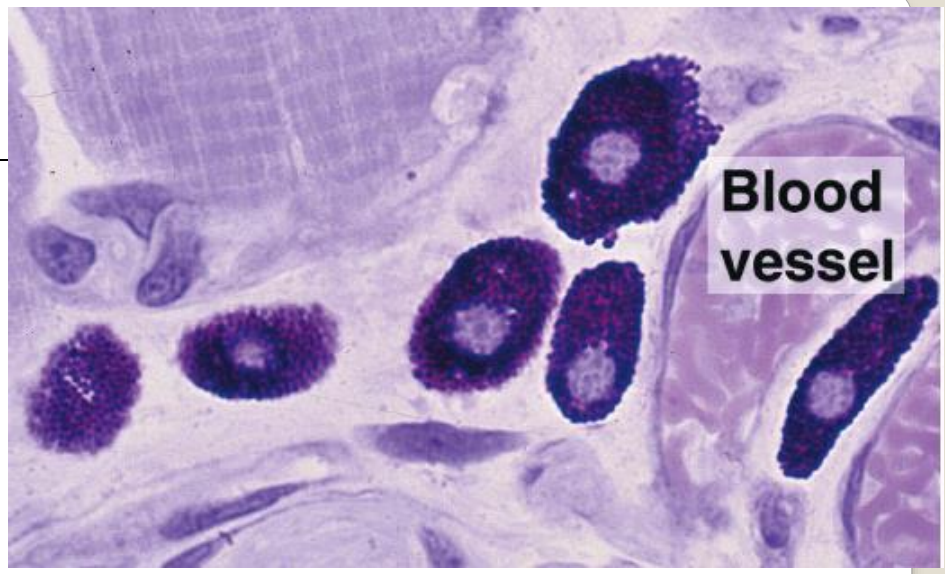


Fig.5: Mast cells. Note how the cytoplasm is intensely basophilic.

- Depending on what's contained in their secretory granules, mast cells may change the blue color of basic dyes into a different color – ***metachromasia***.
- **Function:** Release of heparin, histamine, and various inflammatory molecules. Important in inflammatory and allergic reactions.

Meta- = beyond. Chroma = color.

4) Plasma Cells

- Large, ovoid cells with basophilic cytoplasm because it's rich in RER (no secretory granules). Golgi and centrioles occupy a juxtanuclear position and appear pale.
- Nucleus spherical and eccentric. Has dark peripheral regions alternating with lighter regions (clock-face appearance).
- Short life span (10-20 days).
- Derived from B-Lymphocytes.
- Function:

  production of Antibodies.

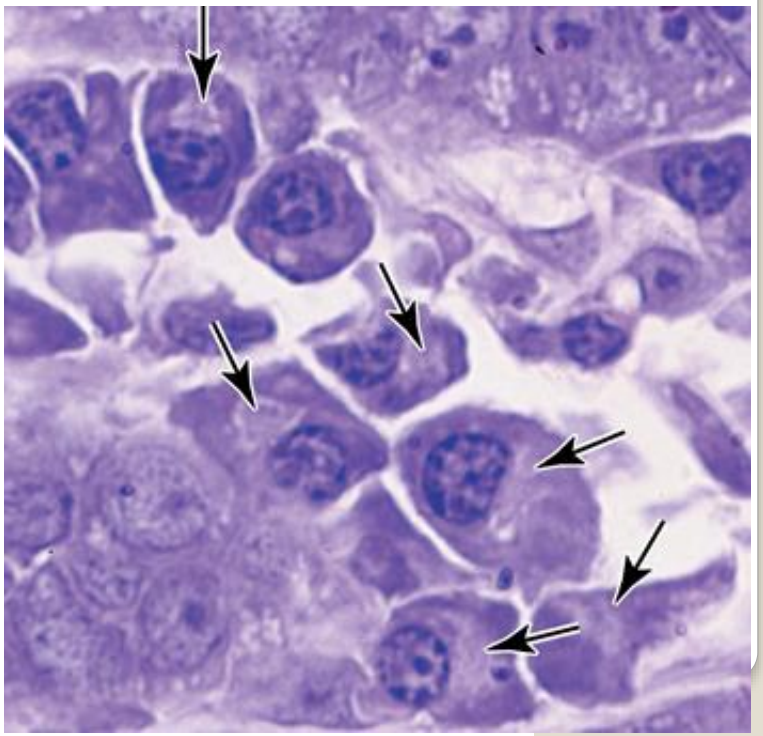


Fig.6: Plasma cells. Note: basophilic cytoplasm,. juxtanuclear pallor (arrows), and the clock-face appearance of the nucleus.

Juxta- = near, close to.

Fibers of the Extracellular Matrix

- Formed from proteins that polymerize into elongated structures.
- The 3 main types are:

1) *Collagen fibers* (from protein Collagen)
2) *Reticular fibers* (from protein Collagen)
3) *Elastic fibers* (from protein Elastin)

1) Collagen Fibers

- Present in different tissues: skin, bones, cartilage, basal lamina, ligaments, and tendons. They give strength to the tissue.
- Several types of collagen protein exists.
- Collagen turn-over is slow in some organs, like tendons where the collagen is stable. In the periodontal membrane, collagen has a high turn-over rate.
- Collagen fibers may be in the form of (1)thick bundles (as in tendons and ligaments), (2)fibrils (as those that anchor the basal lamina to underlying tissues), or (3)networks.

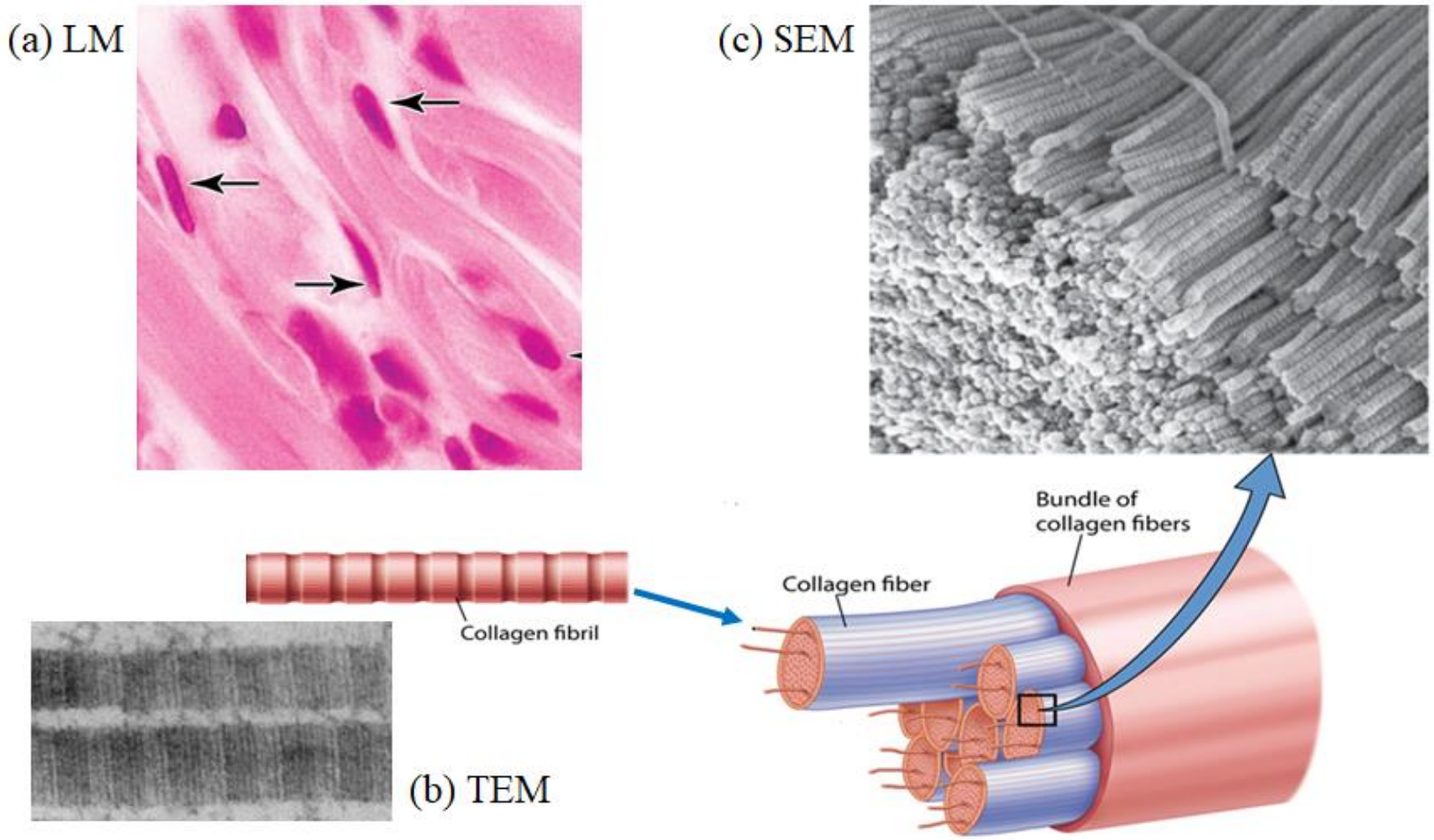


Fig.7: Collagen fibrils, fibers, and bundles. Molecules of collagen protein form collagen fibrils with alternating banding. Several fibrils form a fiber. In some organs, several of these fibers form a bundle. (a) Typical acidophilic appearance of collagen fibers under the LM (arrows indicate nuclei of fibroblasts). (b) TEM image of fibrils, note the banding. (c) SEM of the numerous fibrils in a fiber (the banding can also be seen).

Collagen disease

Disease	Pathology	Notes
Osteogenesis imperfecta	Genetic defect in collagen synthesis	Affects all body. A severe condition. May result in spontaneous bone fracture.
Scurvy	Vitamin C deficiency (this vitamin is important for collagen synthesis)	Periodontal membrane is mostly affected and the teeth fall off
Sclerosis	Accumulation of collagen	Affects all body
Keloid	Accumulation of collagen	In skin wounds

2) Reticular Fibers

- Formed by a type of collagen protein that is heavily glycosylated.
- Thinner than Collagen fibers.
- Stain black with silver impregnation (argyrophilia).
- Form a network of fibers that holds the parenchyma of several organs: liver, spleen, lymph nodes, bone marrow.
- Allow organs to stretch: arteries, uterus.

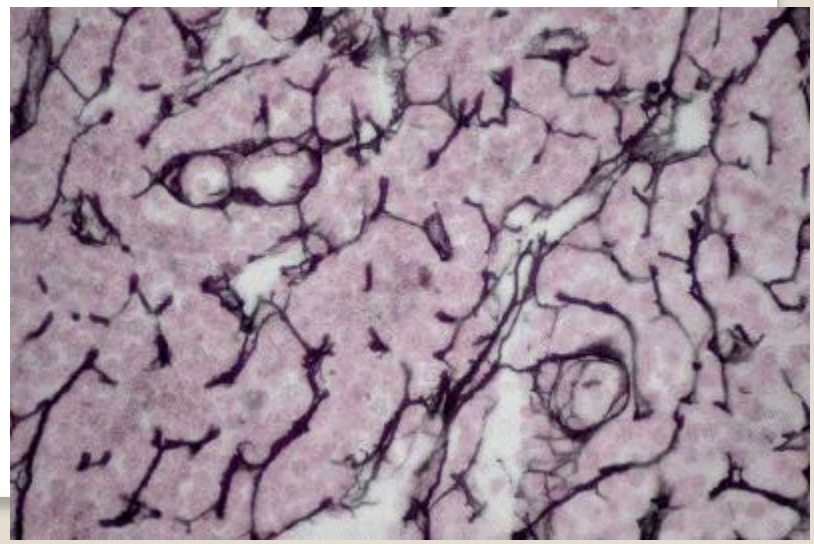


Fig.8: Reticular fibers in lymph node (silver impregnation).

3) Elastic Fibers

- Thinner than collagen fibers.
- Forms a network dispersed between collagen bundles in organs subject to stretching and bending. Elastic fibers may form fenestrated sheets in the walls of large blood vessels called Elastic lamellae.
- They provide Elasticity for the organ
- They’re synthesized by fibroblasts and smooth muscle cells.

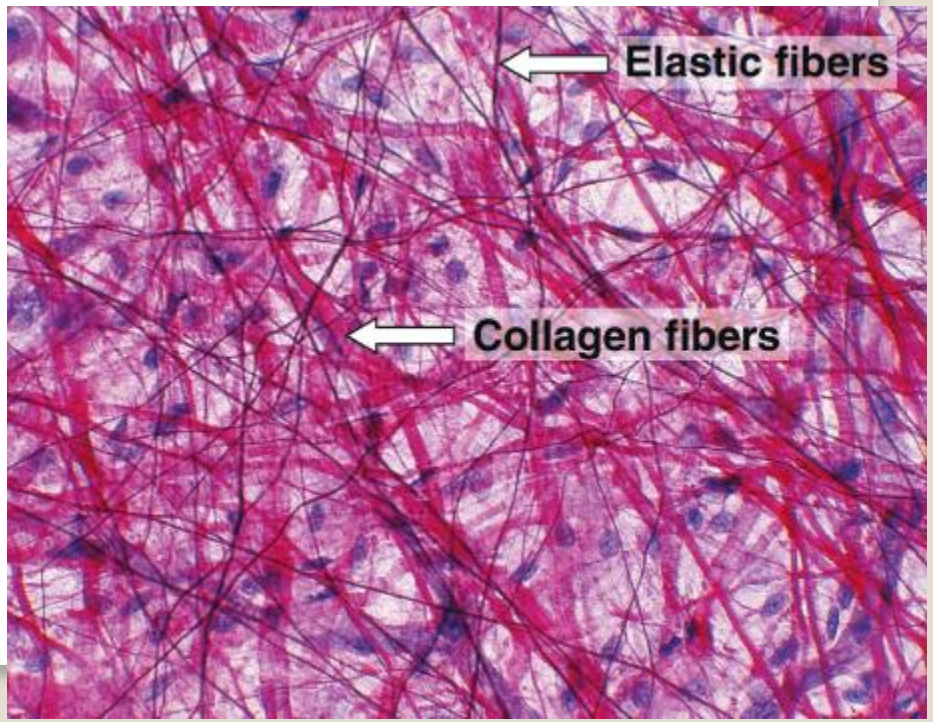


Fig.9: Two stains were used to differentiate between collagen fibers (which are red/pink) and elastic fibers (which are darker in color).

Ground Substance

- Is a hydrated mixture of complex macromelocules that fills the spaces between the cells and the fibers in the connective tissue.

- The macromolecules include:

1) Glycosaminoglycans (GAG) – example ***hyaluronic*** acid
2) Proteoglycans
3) Glycoprotein: Can bind to various components of the tissue and cell membrane. Example: ***Laminin*** of the basal lamina which adheres the basal lamina to epithelial cell membrane.

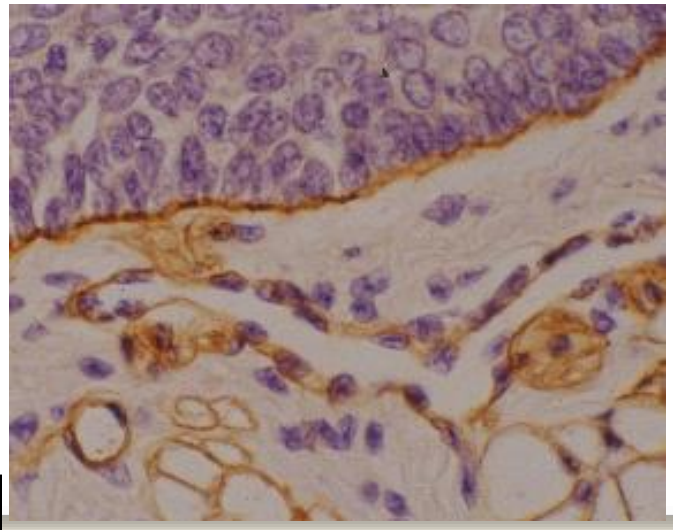
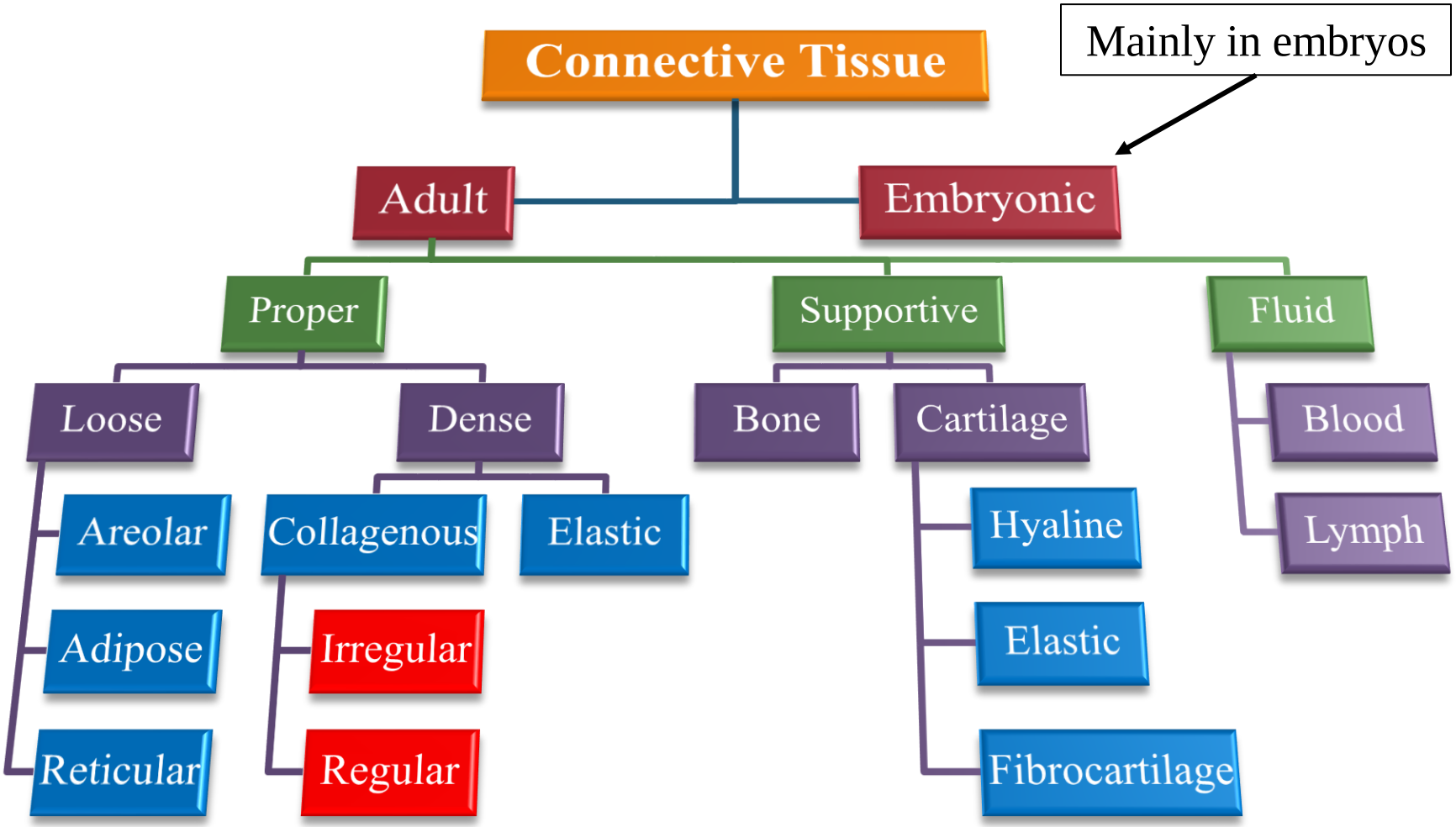


Fig.10: Laminin in the basal lamina.

Classification of Connective Tissue



- ***Proper connective tissue*** is the connective tissue in which the main type of cell that forms the ECM is the fibroblast.

- ***Loose connective tissue***: the fibers are loosely arranged forming a network.

- ***Dense connective tissue***: the fibers are densely packed.

Areolar Connective Tissue:

- A very common type of connective tissue. It gives some support to organs and tissues.

- Features:
  - All types of connective tissue cells (especially fibroblasts and macrophages) are present here.
  - Contain all three types of fibers arranged loosely.
  - All these components are embedded in an abundant semi-fluid ground substance.
  - It's highly vascular.

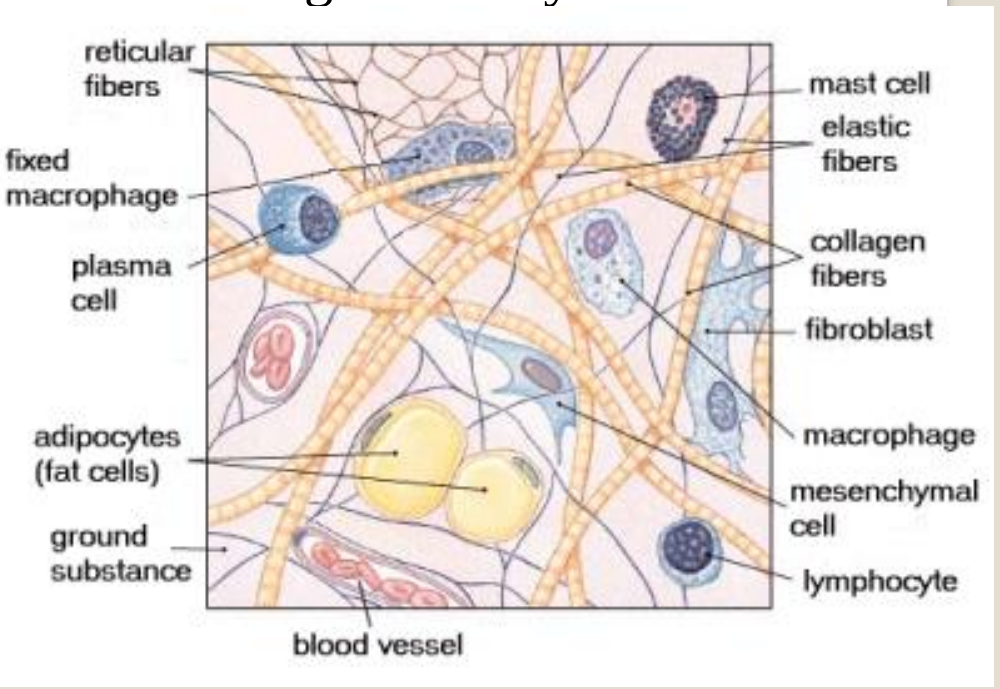


Fig.11: Areolar connective tissue.

Areolar = small spaces.

- Found:
  1. Under epithelia.
  2. Around glands.
  3. In the spaces between muscle and nerve fibers.
  4. Around blood and lymphatic vessels.
  5. It fills many small spaces making it the '*packing material*' of our body.

- Functions:
  1. It gives organs their shape.
  2. It is a medium for the diffusion of gases, nutrients, and waste product.
  3. It is usually the first tissue where microorganisms and foreign particles enter the body; therefore, it's an important site for immune and inflammatory responses.

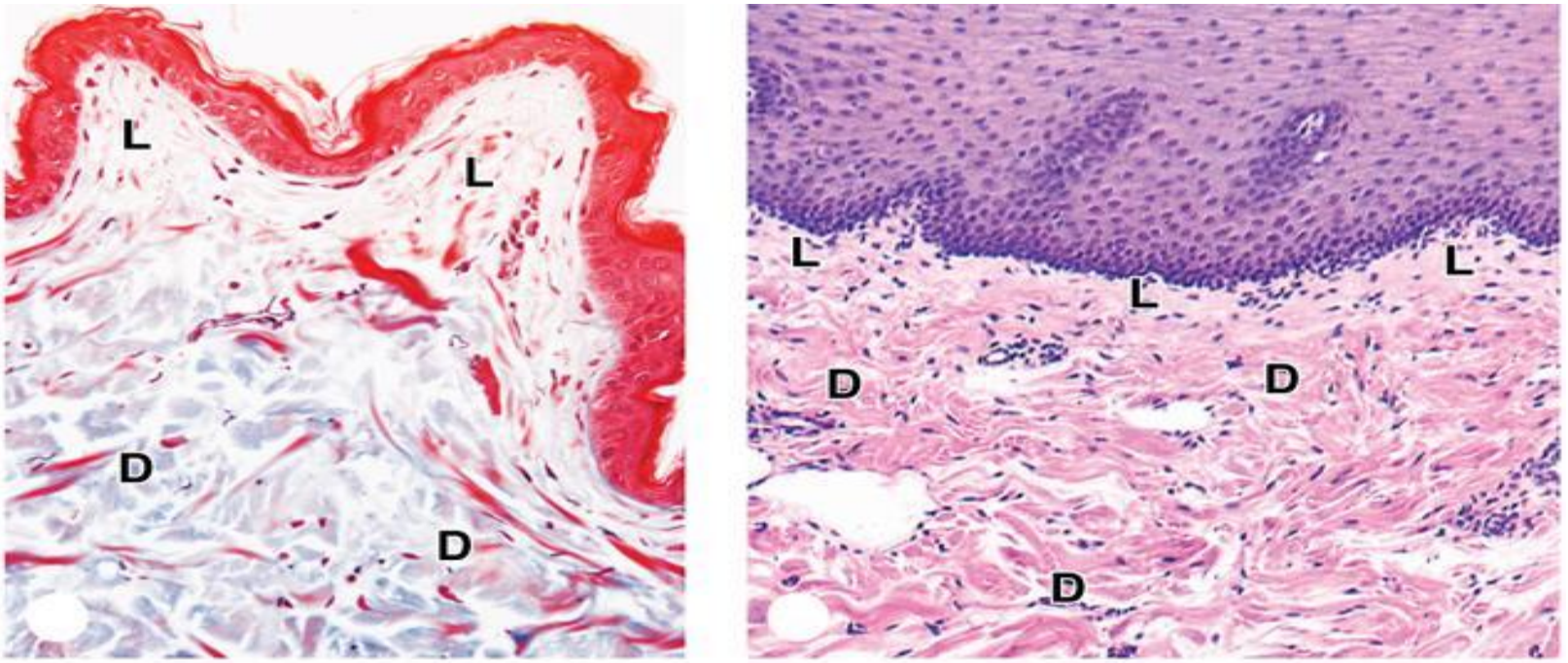


Fig.12: Left: Skin. Right: Esophagus. In both images, the loose areolar connective tissue is indicated by L. Note its position under the epithelium. (The Ds in both images indicate dense collagenous irregular connective tissue).

Reticular Tissue:

- Composed mainly of loosely arranged reticular fibers forming a network within hematopoietic organs (bone marrow, spleen) and lymphatic organs.
- Fibers formed by ***Reticular cells*** (modified fibroblasts). The paces between the fibers is filled with the parenchyma of that organ.
- Macrophages are also present in this tissue.

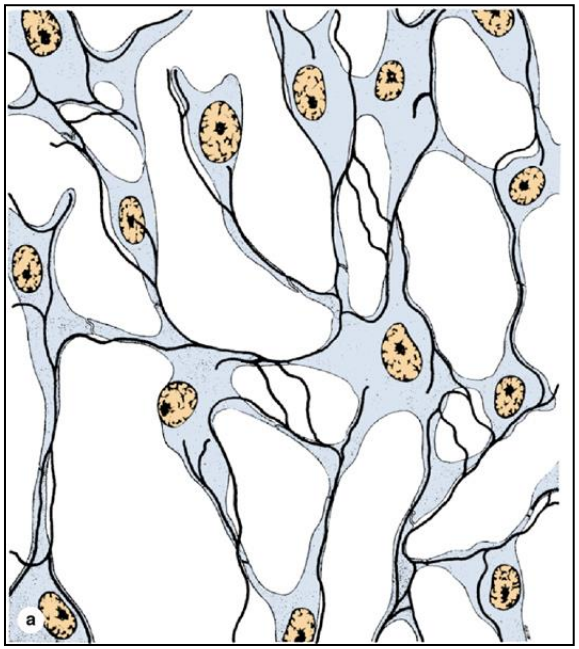


Fig.13: Reticular tissue. Note how there are several spaces in this tissue giving it a sponge-like appearance.

Dense Collagenous Connective Tissue:

- **A type of connective tissue that has numerous densely packed collagen fibers with few cells and ground substance**. Highly resistant to stress.
- Could be irregular or regular

1. ***Irregular***: fibers arranged in no specific orientation forming a 3-dimensional network that resist stress from all directions. It is found in organ subject to great stresses (see Figure 13).

2) ***Regular:*** fibers arranged parallel to each other in a specific orientation with flattened fibroblasts dispersed between them. Provides great resistance to traction forces. Found in tendons and some ligaments giving the living tissue a white color.

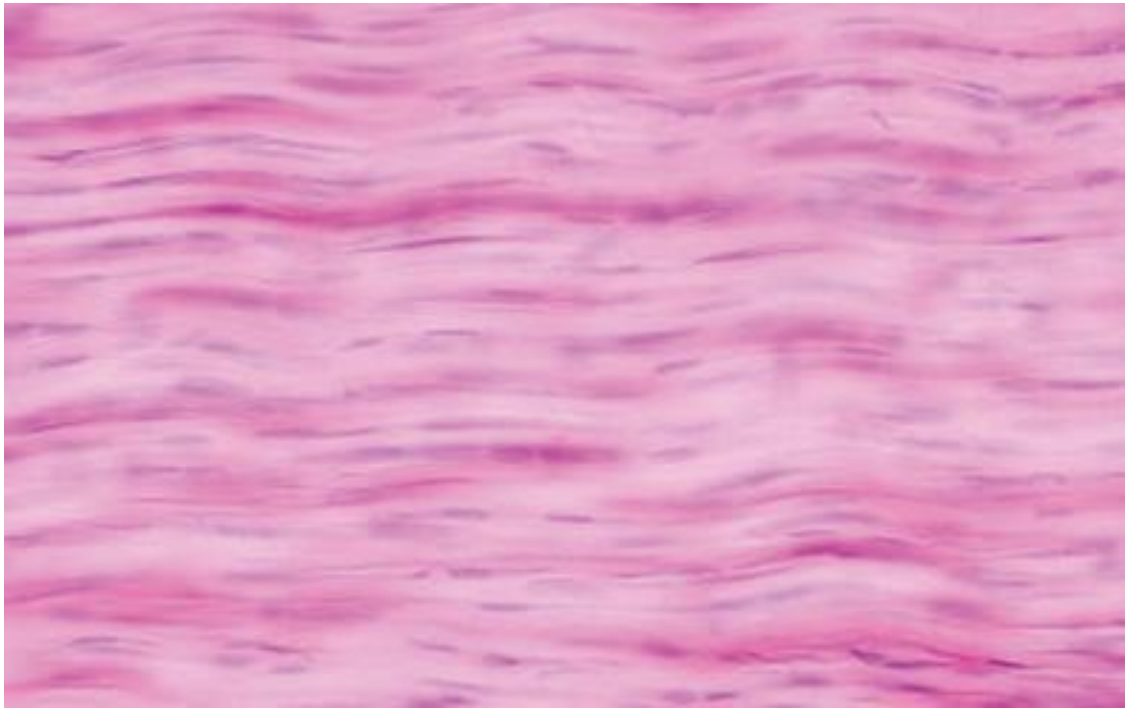


Fig.14: Dense collagenous regular connective tissue of a tendon (longitudinal section).

Dense Elastic Tissue:

➢ Composed mainly of elastic fibers. Found in aorta, some ligaments, and true vocal cords. Gives the organ elasticity.

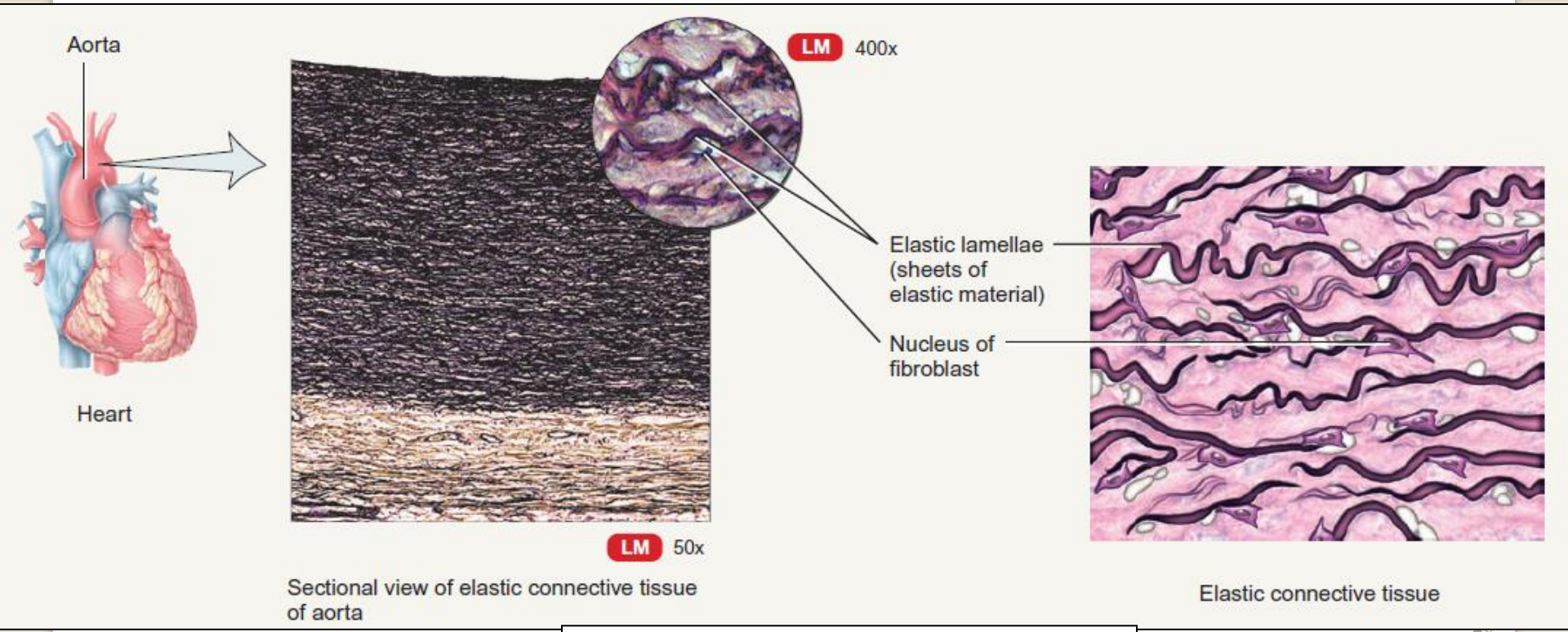
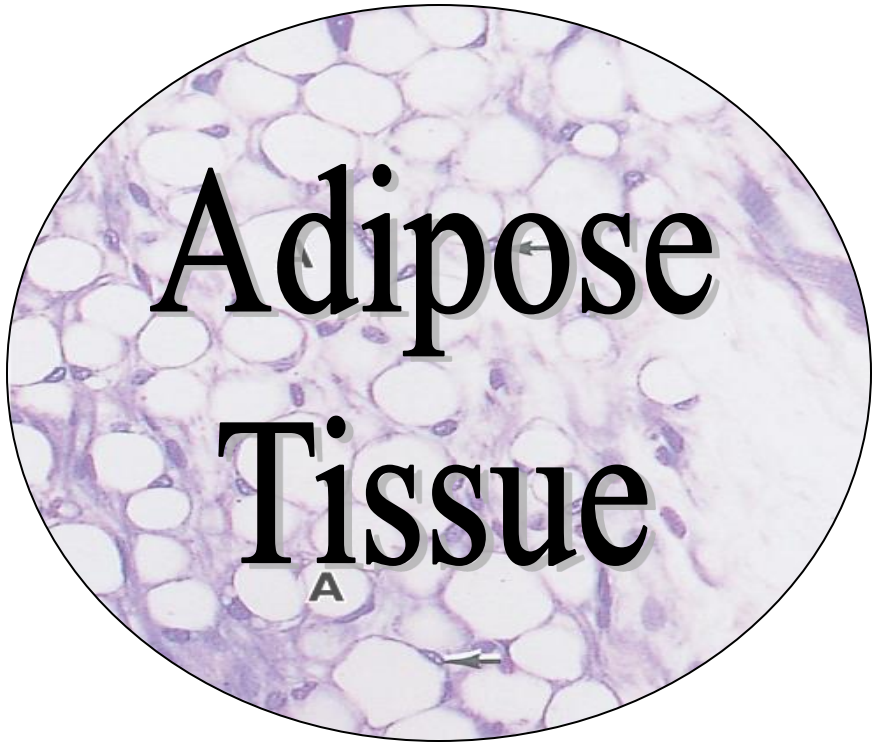


Fig.15: Elastic tissue in the aorta.


Adipose
Tissue
A

- Is a type of loose connective tissue in which adipocytes predominate.
- It’s present throughout the body.
- It constitute about 15-20% of the body weight of males with normal weight, and 20-25% of females body weight.
- It could ***White*** (WAT) or ***Brown*** *(BAT)*.

Functions of Adipose Tissue

1) Storage of energy in the form of Triglycerides.
2) Endocrine role by the release of certain hormones and cytokines.
3) Insulator, because it's a poor conductor of heat.
4) Fills the large spaces between tissues and keeps some organs in place.
5) Subcutaneous fat helps shape the surface of the body.
6) Fat pads act as shock absorbers (palms and soles).
7) Warming of blood (brown fat).

White Adipose Tissue

- Specialized in energy storage in white adipose cells.

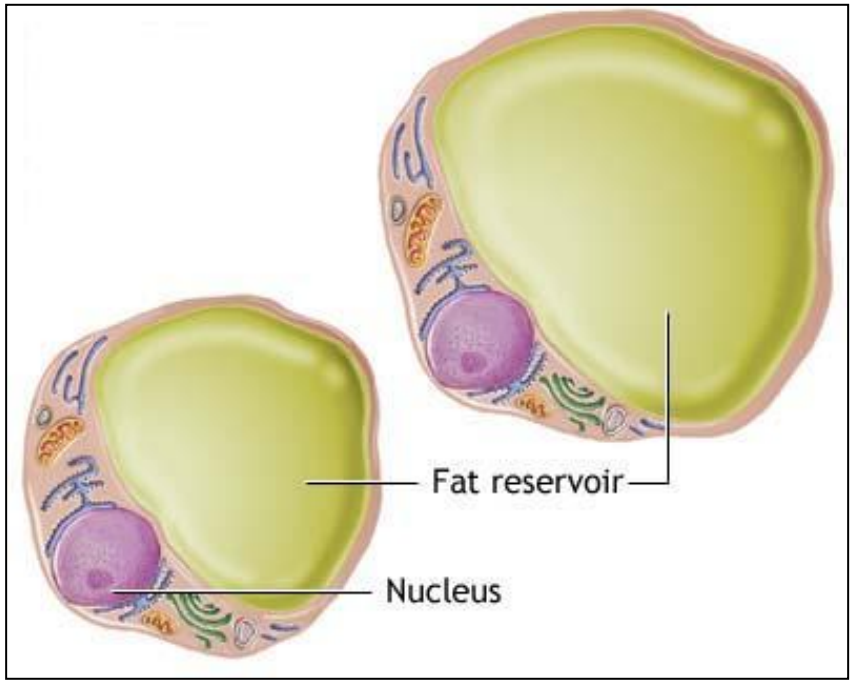
- Depending on diet, its color varies from white to bright-yellow.

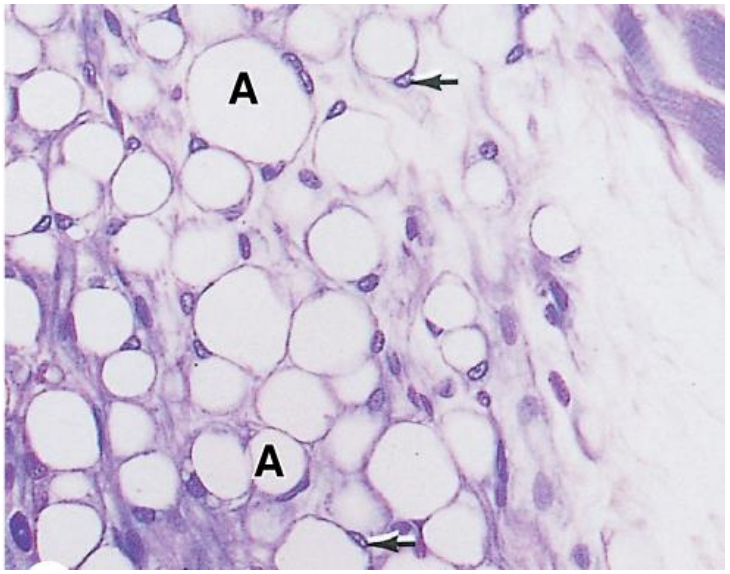
- ***Features of white adipose tissue:***

1) Fibroblasts and macrophages are present in the tissue.
2) Reticular fibers form a network that supports individual adipose cells.
3) Divided by connective tissue partitions into incomplete lobules.
4) Highly vascularized.

Histological features of White Adipocytes

1. Large spherical cells with a single large fat droplet (unilocular).
2. Flattened nucleus on one side (pushed by the droplet).
3. A thin film of cytoplasm around the droplet containing well-developed SER (smooth endoplasmic reticula) and pinocytotic vesicles.
4. Thicker cytoplasm around the nucleus containing several mitochondria, Golgi apparatus, polyribosomes, and poorly developed RER.
5. The droplets are surrounded by Vimentin intermediate filament.
6. The cell is surrounded by a thin external lamina (similar to basal lamina).





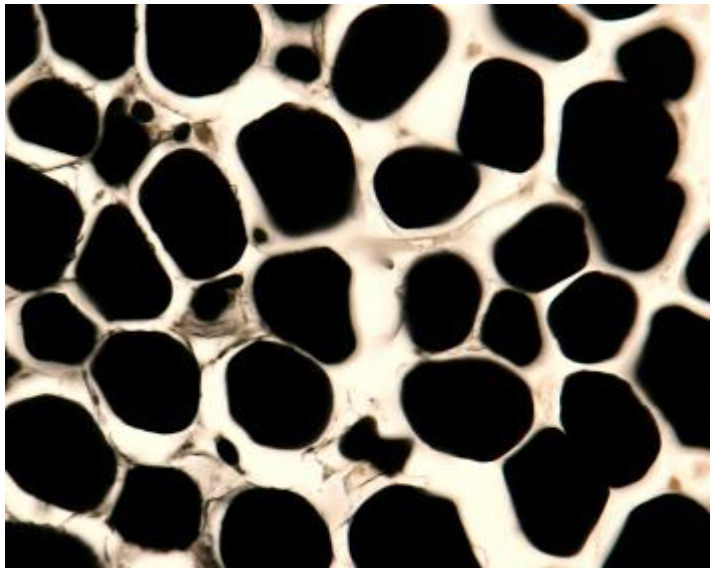


Fig.16: The image above shows the features of white adipocyte. Top-right, the image shows the typical appearance of fatty tissue in a routine preparation. Fat droplets dissolve during tissue preparation and the cell appears as a thin ring with the nucleus projecting on one side (the ***signet-ring appearance***). Bottom-right, special stain was used to preserve the fat droplet.

Clinical aspects of White adipose tissue

1) WAT secretes the hormone ***Leptin*** which is a '***Satiety Factor***' → Could obesity be treated by hormonal therapy?

2) ***Adiponectin*** is released by adipocytes. The larger the adipocyte is, the less adiponectine it releases. This hormone protects against diabetes and other diseases.

3) ***Obesity is characterized by a state of chronic mild inflammation*** because WAT secretes several inflammatory factors → Could these be related to the cardiovascular or diabetic complications of obesity?

4) Although histologically similar, visceral and subcutaneous WAT have different gene expression. The visceral WAT is more dangerous to health → Could obesity be treated by gene therapy?

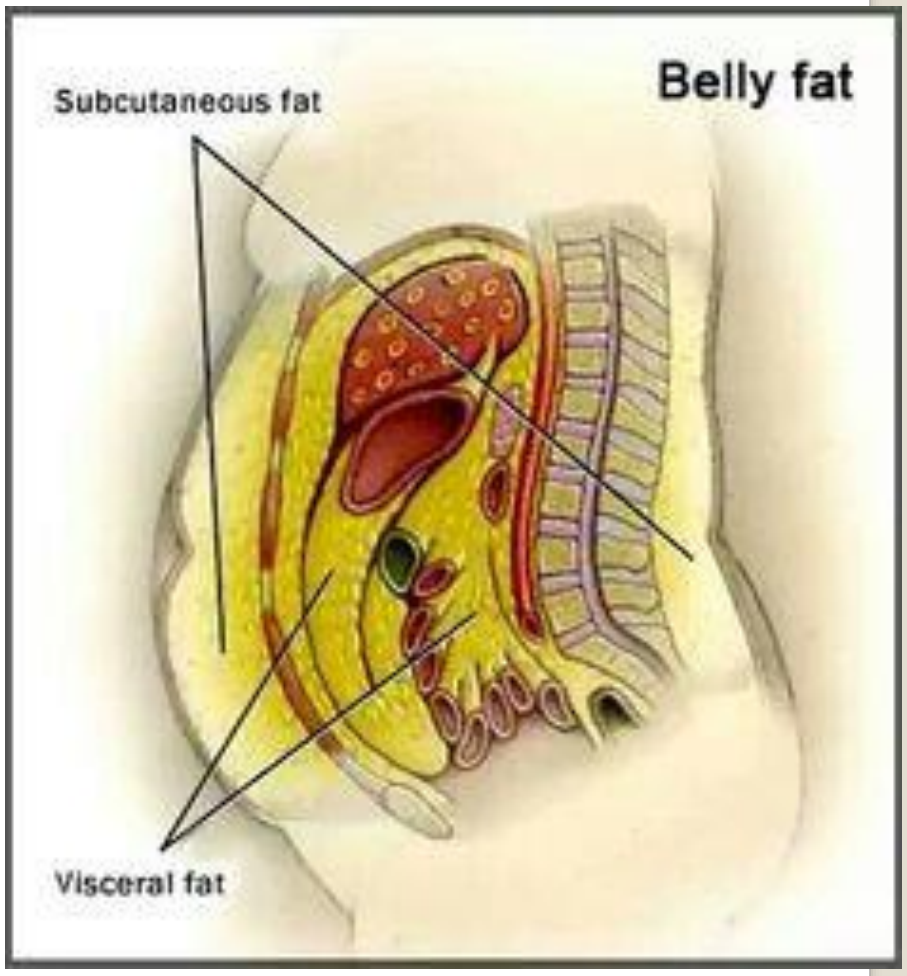


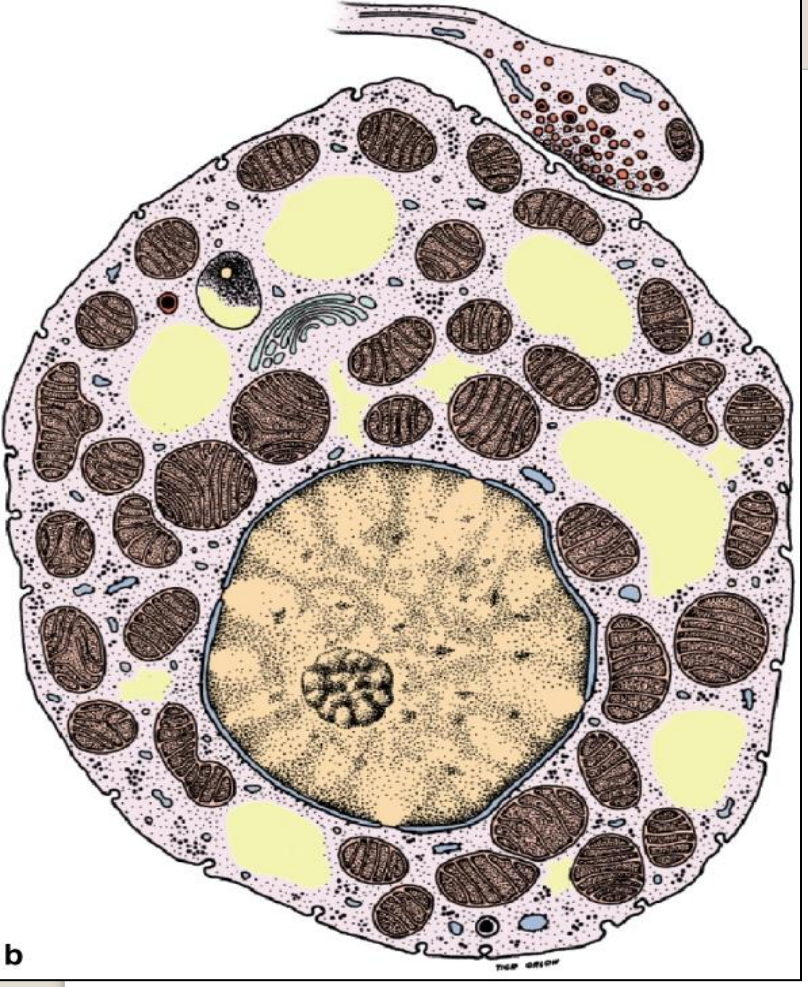
Fig.17: Subcutaneous and visceral fat.

5) At birth, fat stores are already formed and distribution and density varies with age and gender.

- *Obesity* in *adults* is *hypertrophic* (results from increase in size of the already present adipocytes). In *Children*, the obesity could be *hyperplastic* (increase in the number of cells) because new adipocytes can be formed from precursor cells that are still present at this age. Such obese children are liable to develop a more severe hypertrophic obesity because they have more adipocytes. → ***Treat/prevent obesity at an early age.***

Brown Adipose Tissue

- Specialized in heat production.
- Brown adipocytes are smaller than white adipocytes, polygonal, with multiple fat droplets (multilocular). They have numerous mitochondria and a central spherical nucleus.
- Cells arranged in an almost epithelial arrangement around a blood capillary. The tissue is divided into lobules by connective tissue partitions.
- The brown color is due to the mitochondria and the blood vessels.



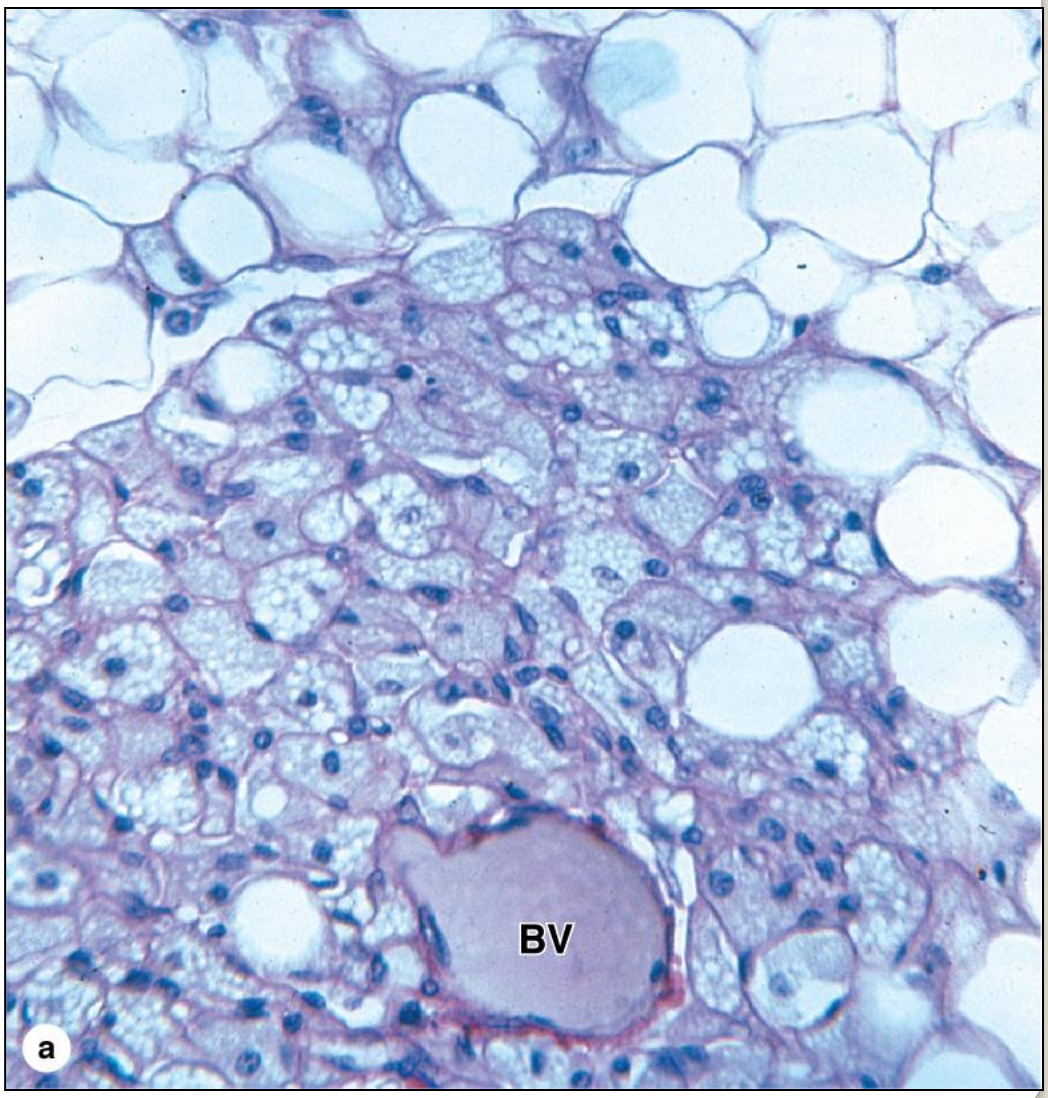
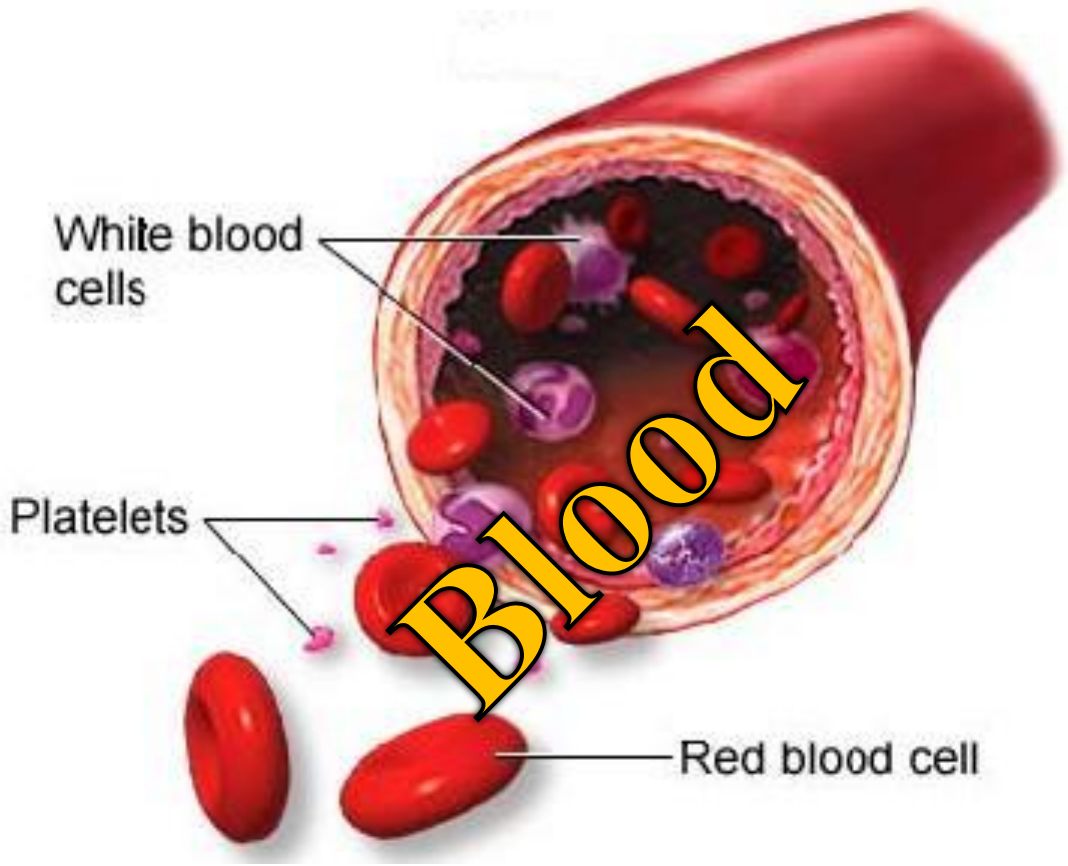


Fig.18: Above, a brown adipocyte, note the several small fat droplets. Right, brown adipose tissue, note how the cells surround a blood vessel (BV).

- At birth, brown adipose tissue is maximal for body weight. It then decreases with age.
- In adults, it’s found in scattered areas especially *around the kidneys, the adrenals, the aorta, and in the mediastinum*.
- It increases during cold adaptation.


White blood cells
Platelets
Red blood cell
Blood

- Blood Is a fluid type of connective tissue characterized by having a liquid extracellular matrix (plasma) in which are dispersed the formed elements of blood: (1) Red blood cells (erythrocytes), (2) White blood cells (leukocytes) and (3) Platelets (thrombocytes).

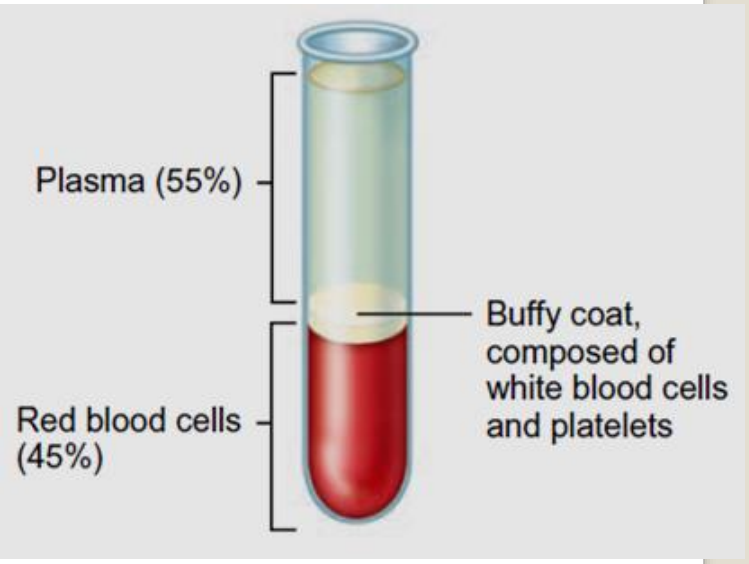


Fig.19: Appearance of centrifuged blood.

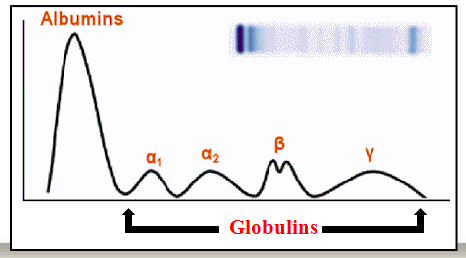
- Functions of blood:

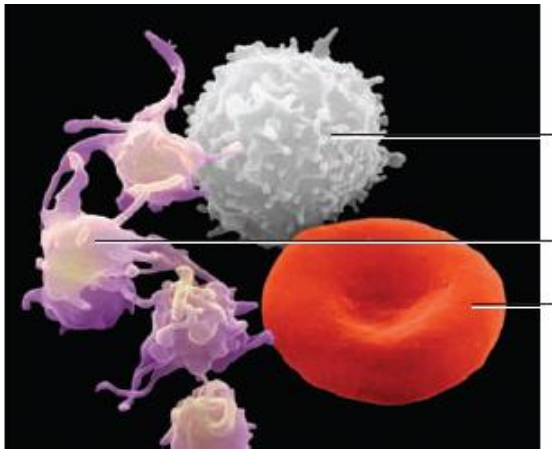
1) *Transportation:* Gases, nutrients, waste products, hormones.
2) *Regulation:* pH, body temperature.
3) *Protection:* Clotting, white blood cells, proteins (antibodies).

Components of Blood

Blood

- **Plasma** — 55% of blood
  - Water — 91% of plasma
  - Solutes (mostly protein)
    - Proteins (Mostly Albumin)
    - Other Solutes
- **Formed Elements** — 45% of blood
  - Red Blood Cells
  - White Blood Cells
  - Platelets

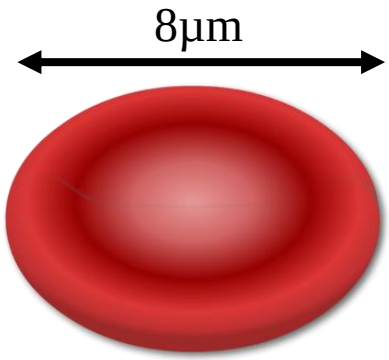




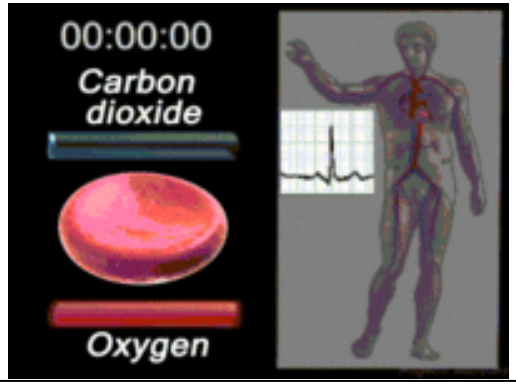
Formed Elements of Blood

Erythrocytes (Red Blood Cells)

- The most abundant type of cell in blood
- *This cell is normally only present in blood inside the blood vessels.*
- Biconcave disc in shape. This increases surface area.
- Lack nucleus and other organelles. Cytoplasm is filled with the oxygen-carrying protein hemoglobin. Because it has no mitochondria, it doesn't use oxygen.



- Strong, flexible plasma membrane. This allows the cell to change its shape without rupturing as it passes through narrow capillaries.



The flow rate in this animation has been tripled. An average cycle actually takes about 60 seconds.

- Life span about 120 days.

Functions of the red blood cells

1) The hemoglobin in the RBCs functions in the transportation of:
   - Oxygen – this is the main function of RBCs
   - CO_2
   - Nitric Oxide (NO) – this gas is a vasodilator that helps in increasing blood flow

2) Glycolipids in plasma membrane are responsible for ABO and Rh blood groups.

3) When RBCs are destroyed by some microorganism, they release substances that can kill the microorganism.

Leukocytes (White Blood Cells)

- Wandering cells: formed in bone marrow, circulate in blood and enter tissues.
- Respond to local factors in inflammation.

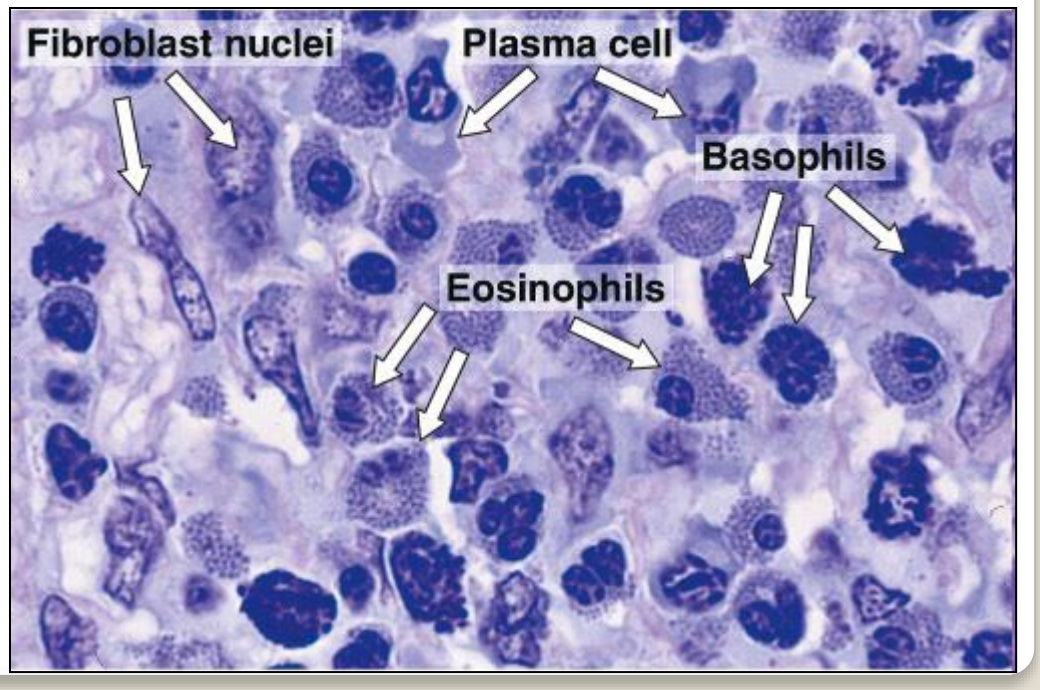
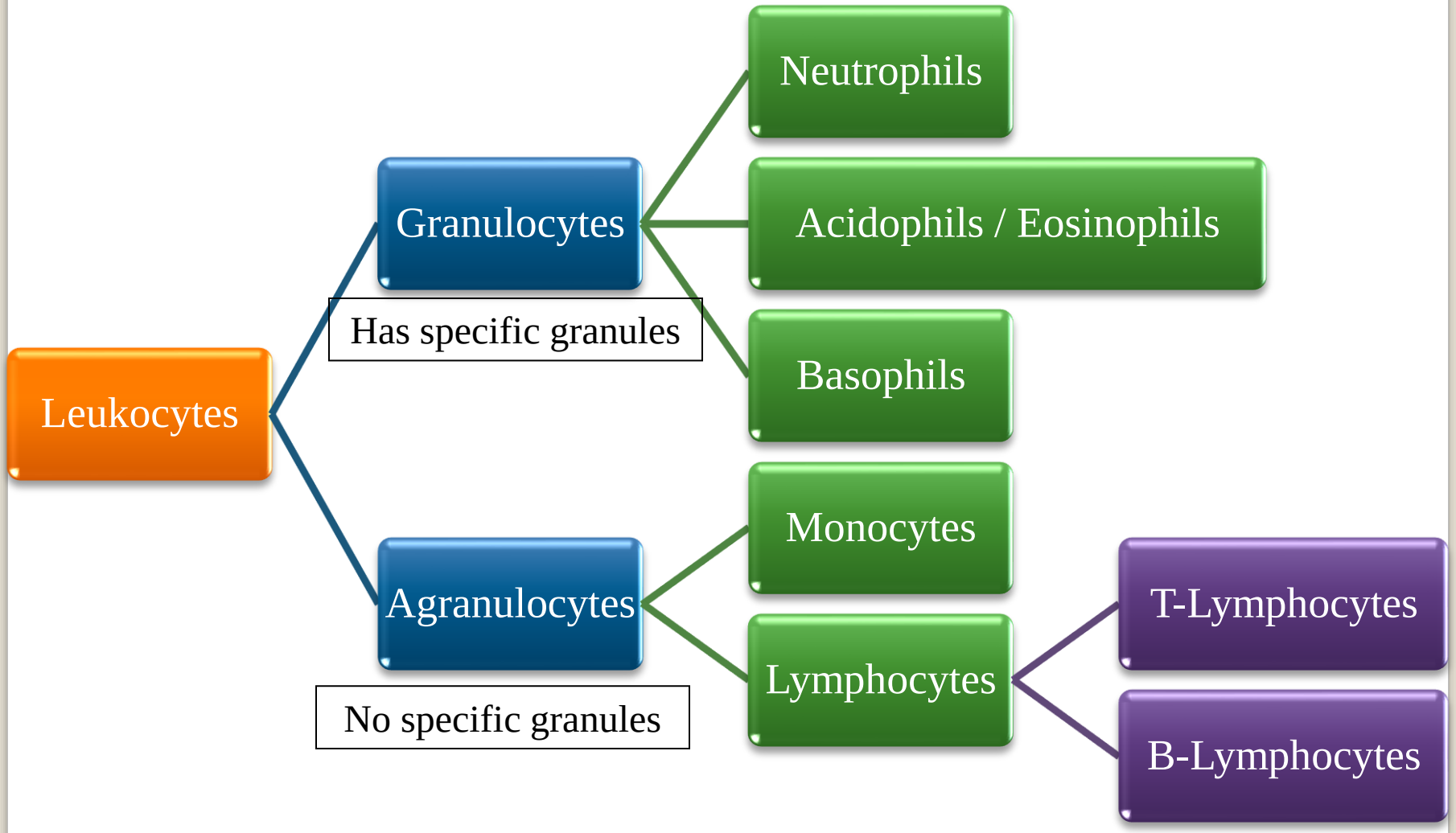
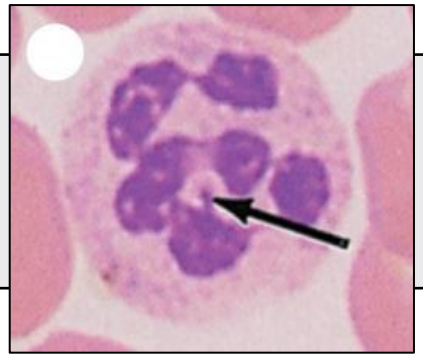
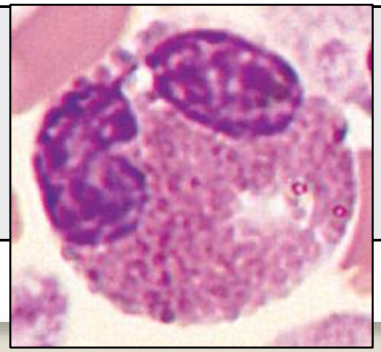
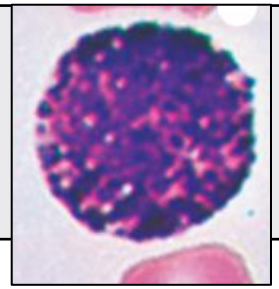


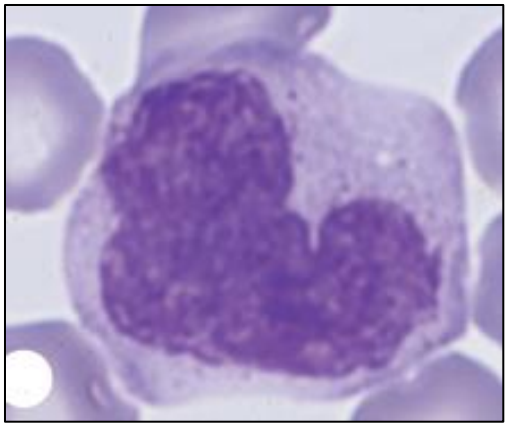
Fig.20: Leukocytes in inflamed tissue.

Classification of Leukocytes



Granulocytes	Neutrophils	Eosinophils	Basophils
Abundance (% of leukocytes)	60-70%	2-4%	0.5%
Nucleus	Multilobed (with inactive X-chromosome of females appearing as a drumstick appendage)	Bilobed	S shaped (obscured by granules)
Granules	Sparse and stain variably	Large eosinophilic	Large basophilic
Function	Phagocytosis	• Defense against parasitic infection • Allergic reactions	Release of inflammatory molecules
			

Agranulocytes:

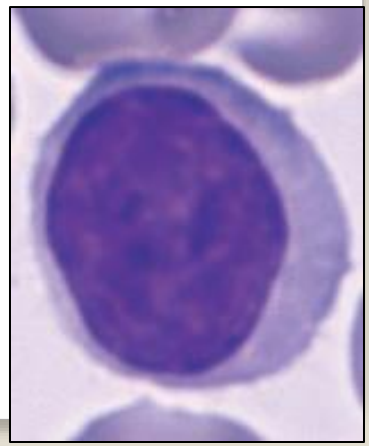


Monocytes (5%):

- Kidney or U-shaped nucleus.
- Cytoplasm basophilic.
- Function: formation of macrophages.

Lymphocytes (28%):

- Variable in size.
- Nucleus very dark and occupies most of the cell.
- Functions:
  - T-cells → Cell mediated immunity
  - B-cells → Antibody-mediated immunity



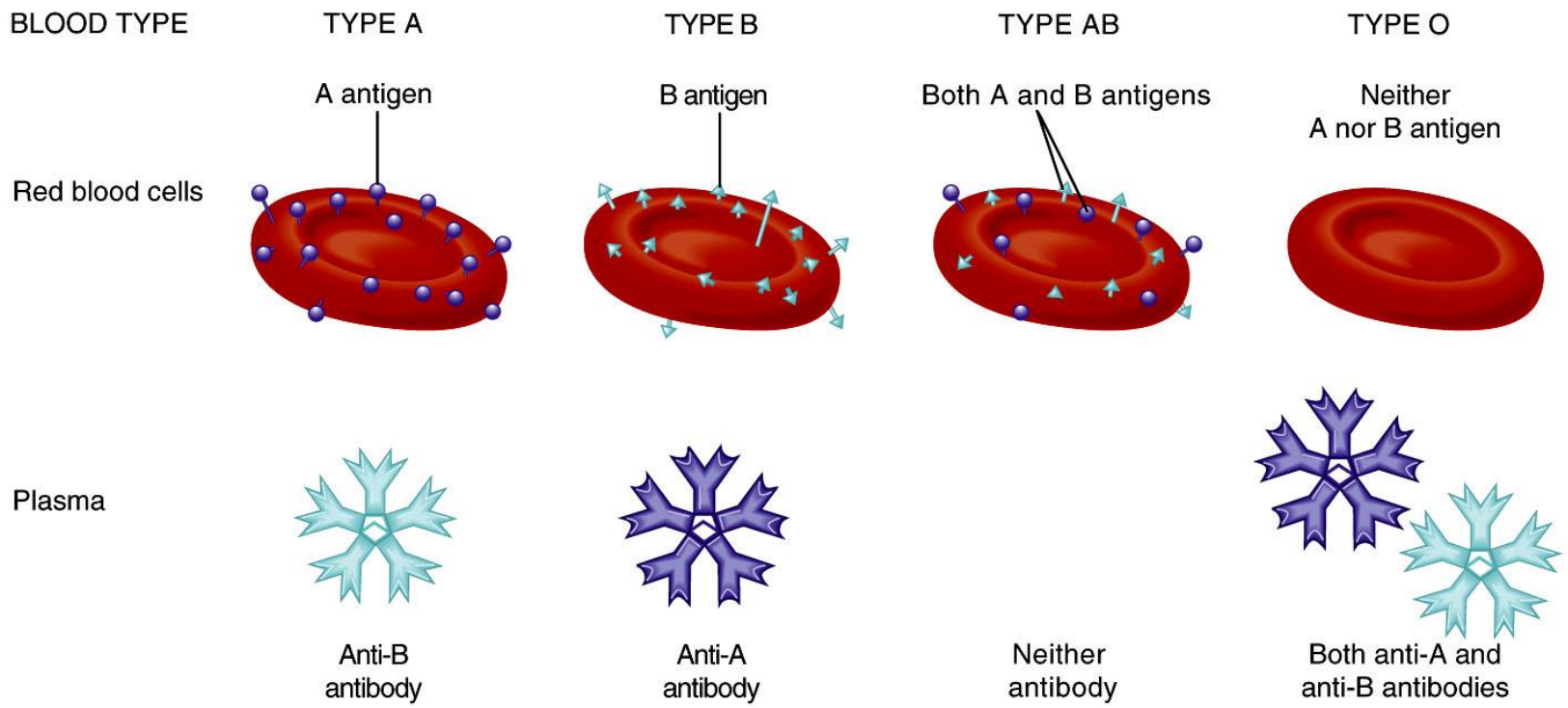
Platelets/ Thrombocytes

- Large cells in the bone marrow called ***Megakaryocytes*** send processes into blood vessels. These processes will splinter into small fragments called Platelets.
- This process continues until each megakaryocyte gives rise to about 2000 platelets.
- Each platelet is a disc-shaped structure surrounded by cell membrane and containing no nucleus but numerous vesicles containing blood-clotting promoting factors.
- Short life span: 5-9 days.
- Function: Stops bleeding by the formation of (1) platelet plug and (2) blood clot.

ABO Blood Groups

- Blood group is type of blood designated to a person based on the presence/absence of an antigen on the surface of RBCs.
- The ABO blood groups are based on the A and B antigens.
- Reason for antibodies presence not clear.

Blood Type	Antigen on RBCs	Antibody in Plasma
A	A	Anti-B
B	B	Anti-A
AB	A & B	None
O	None	Anti-A & Anti-B



- Each blood group can give to itself
- ***AB*** is the ***Universal Recipient***
- ***O*** Is the ***Universal Donor***

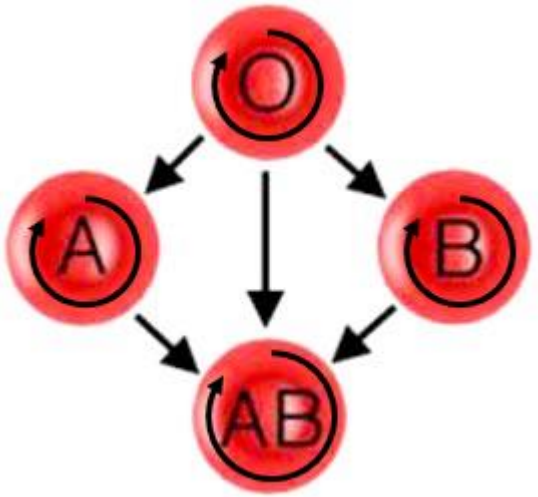


Fig.21: ABO blood groups. Top image: Antigens and antibodies. Right image: Blood donation.

Thank You

There are two ways of spreading light: to be the candle or the mirror that reflects it.

كن عالما او متعلما و لا تكن ثالثهما فتخسر

Cartilage

Dr. Mustafa Saad

(2024)

- ***Cartilage is a supportive type of connective tissue whose ECM is of a firm consistency which allows the cartilage to bear mechanical stresses.***

- Cartilage is formed of Chondrocytes and ECM. This ECM is composed of fibers and ground substance synthesized by the chondrocytes.

- According to the relative contents of the ECM, cartilage can be divided into three types:

1. Hyaline Cartilage.
2. Elastic Cartilage.
3. Fibrocartilage.

- ***Cartilage contain no blood vessels or nerves***. Nutrients and stimuli reach this tissue by diffusion from the perichondrium or from the nearby synovial fluid.

- **Perichondrium:**

  Is a layer of dense connective tissue that covers all hyaline cartilages (except in joints) and all elastic cartilages. It's richly vascular and contains collagen fibers and fibroblasts. It's essential for the nourishment of the cartilage.

Peri- = around. Chondro- = related to cartilage.

Functions of Cartilage:

1. Support of soft tissues, as in the larynx and trachea.
2. Acts as a shock absorber as in the intervertebral disc.
3. Important in the development and growth of bones before and after birth.

Chondrocytes:

- Oval or round cells.
- Located inside spaces in the surrounding matrix called ***lacunae***.
- Each lacuna may contain 1-8 chondrocytes.
- Function: production of the ECM including the fibers and large molecules of the ground substance.

Lacuna (plural, lacunae) = small space, gap

Hyaline Cartilage

- The most common type of cartilage. Fresh hyaline cartilage is bluish-white in color.
- The chondrocytes are oval or round.
- The ECM is rich in collagen fibers.
- The ground substance is rich in ***hyaluronic acid***.

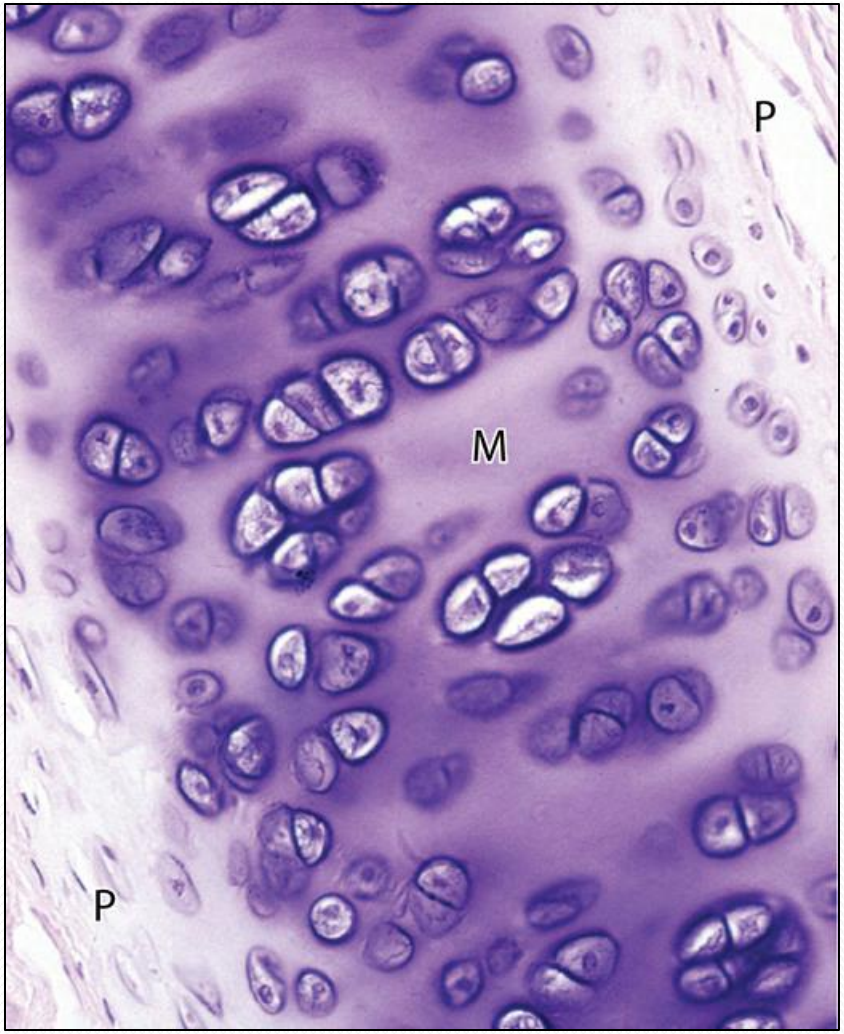


Fig.1: Hyaline Cartilage. P = perichondrium. M = ECM.

- During development, it serves as a template for the formation of long bones.

- Location (in adults):
  1. Articular surfaces of bones in some joints.
  2. Some cartilages of the airway passages (nose, thyroid and cricoid cartilages of the larynx, and trachea).
  3. Costal cartilages.
  4. The epiphyseal growth plate.

Elastic Cartilage

- The ECM of this type of cartilage is rich in *elastic fibers* which gives the fresh cartilage a yellow color.

- Location:
  1. Auricle of the ear.
  2. Wall of external auditory canal.
  3. Auditory tube.
  4. Epiglottis and cuneiform cartilages of the larynx.

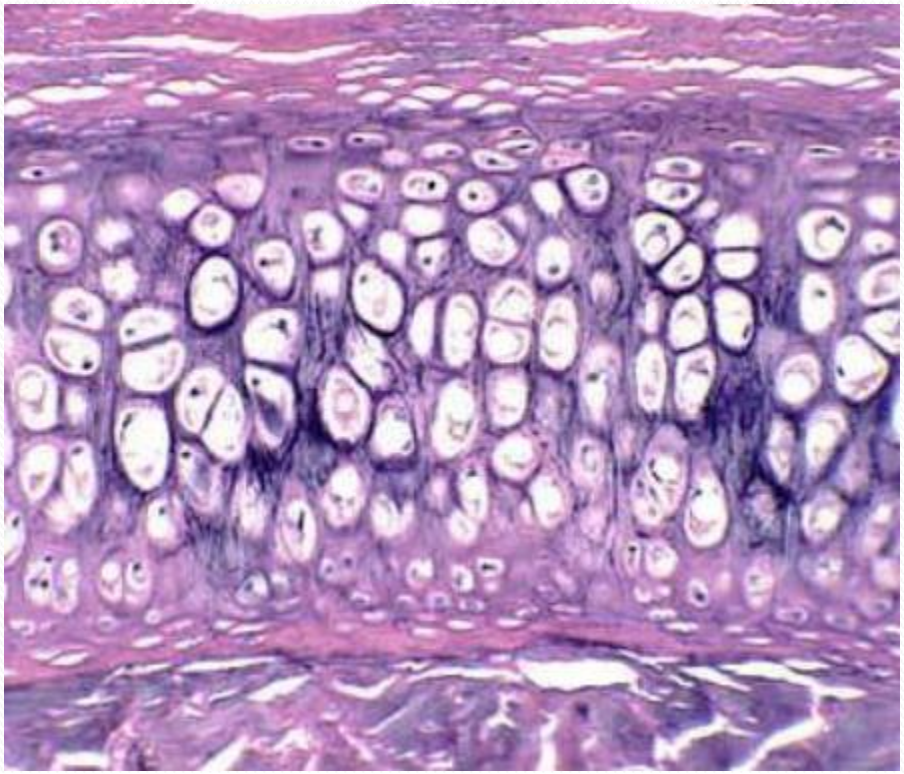


Fig.2: Elastic Cartilage. Special stain was used for the elastic fibers. Note the perichondrium on both sides.

Fibrocartilage

- ✓ The chondrocytes in the lacunae are arranged axially.
- ✓ The matrix is rich in collagen fibers.
- ✓ *This type of cartilage possesses no perichondrium.*

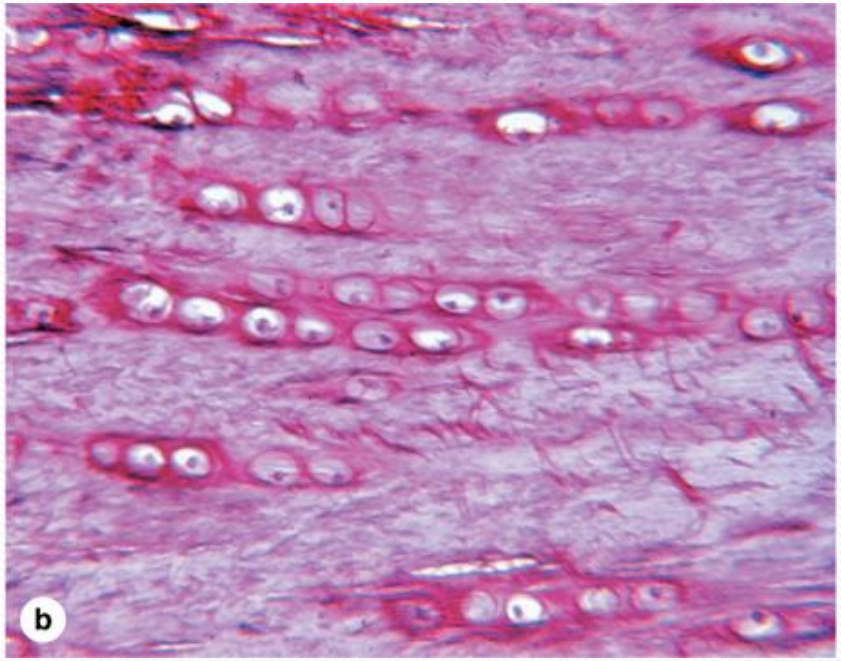


Fig.3: Fibrocartilage. Note the axial arrangement of the chondrocytes and the acidophilic matrix.

- ✓ Location: Intervertebral discs, pubic symphysis, and the menisci of the knee joint.

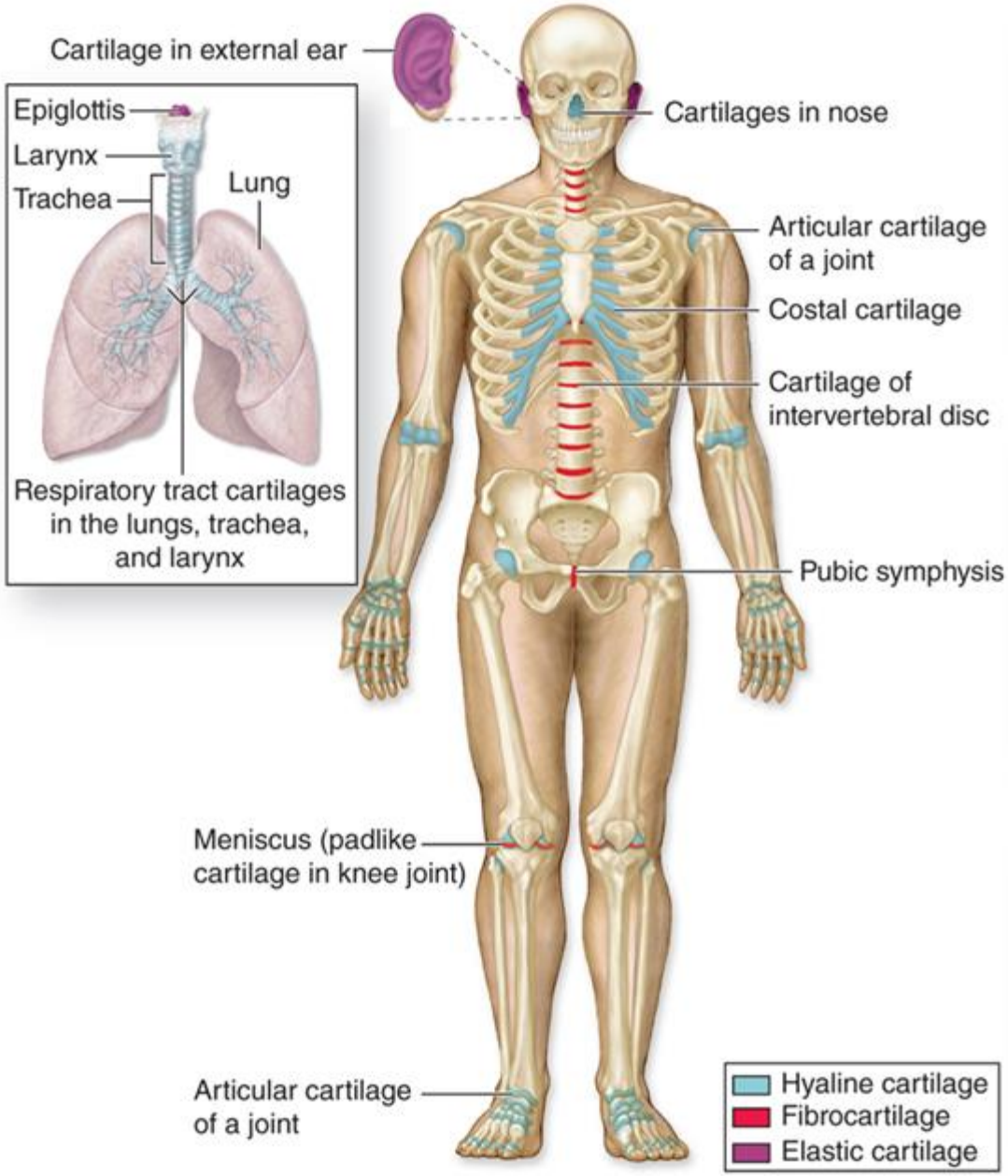


Fig.4: Location of the different types of cartilage.

Intervertebral Discs

- These discs are located between the bodies of two adjacent vertebrae.

- They’re formed of two parts:

1. Outer annulus fibrosus.
2. Inner nucleus pulposus.

- They act as cushions and shock absorbers for the vertebrae.

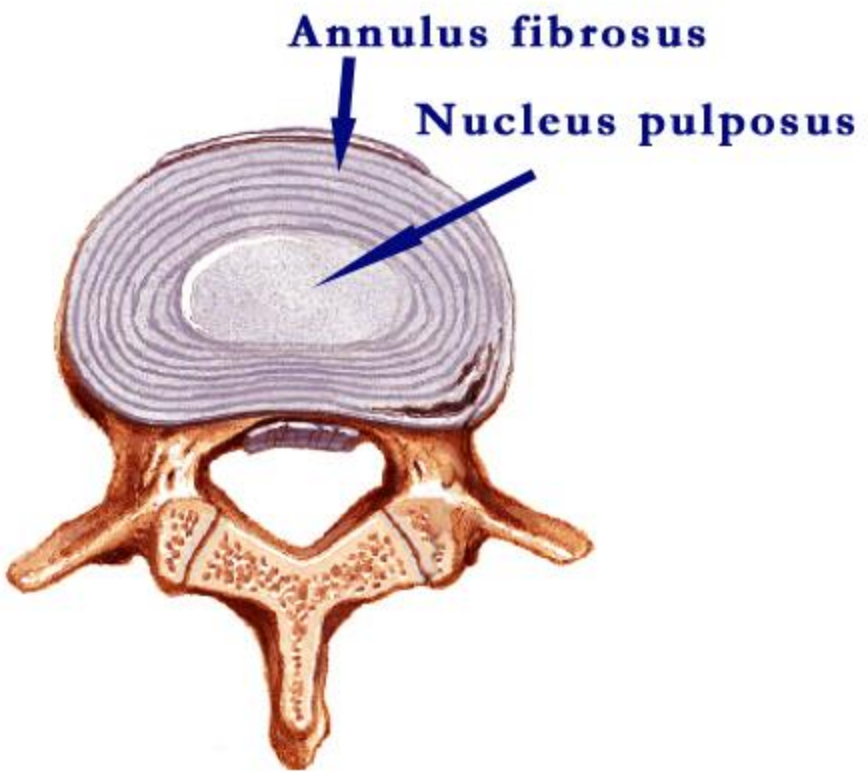
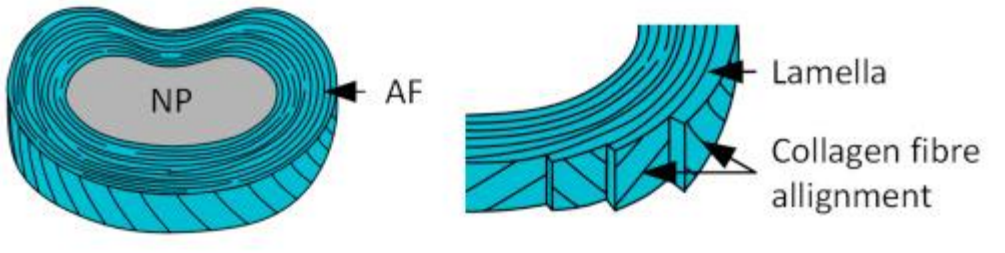


Fig.5: Parts of the intervertebral disc.

Annulus fibrosus:

- It's formed of an external layer of dense collagenous irregular connective tissue. Internally, there are multiple layers of fibrocartilage. The direction of the collagen fibers in any layer is 90º to that of the adjacent layer.



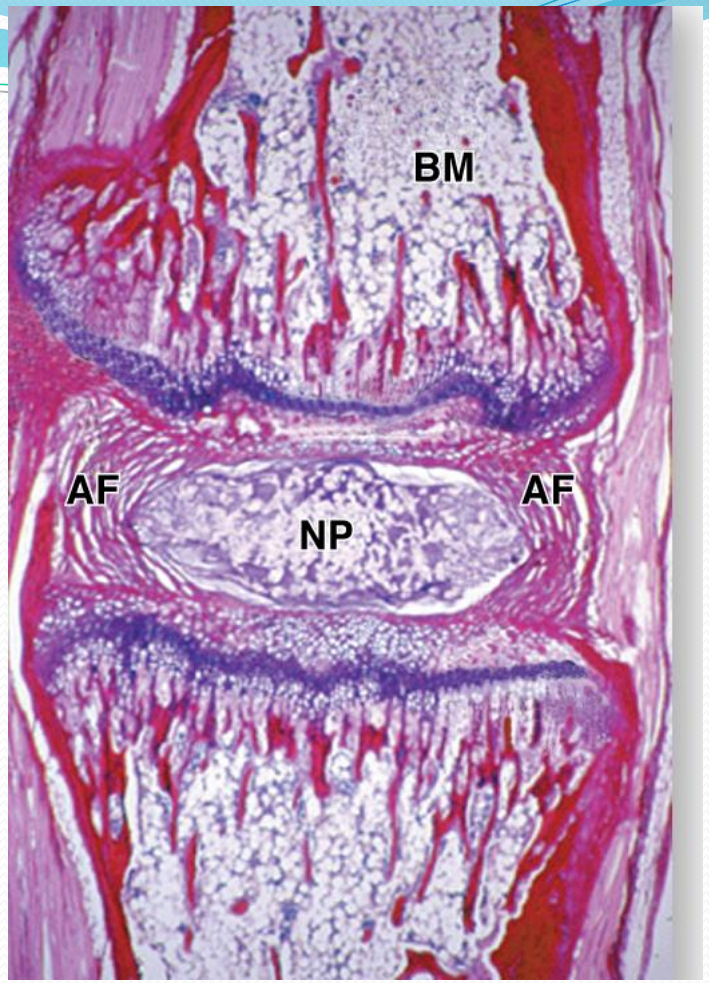


Fig.6: The annulus and its layers.

- This arrangement makes the annulus resilient and enables it to act as a support for the vertebrae.

Nucleus pulposus:

- It's formed of a gel-like substance rich in hyaluronic acid and some collagen fibers.
- It may contain some cells.
- It has a high water content.
- It acts as a shock absorber.

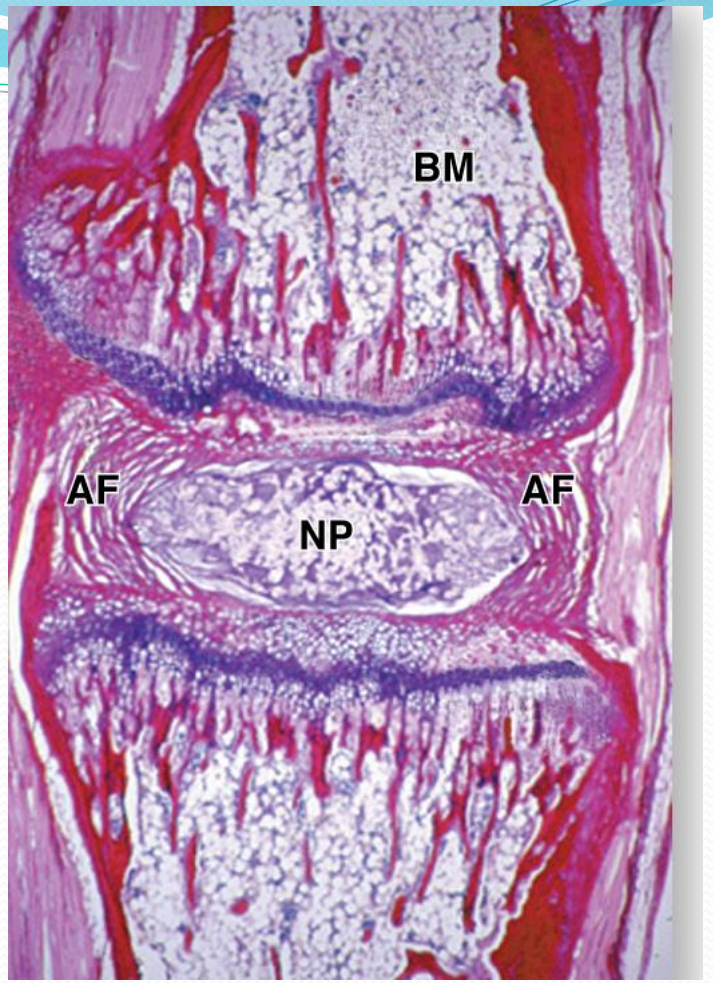


Fig.7: The nucleus pulposus and how it acts as a shock absorber.

Disc prolapse

- If the annulus weakens, the nucleus will protrude outside the disc → Disc prolapse/herniation.
- This may cause compression on the spinal cord or the roots of the spinal nerves.

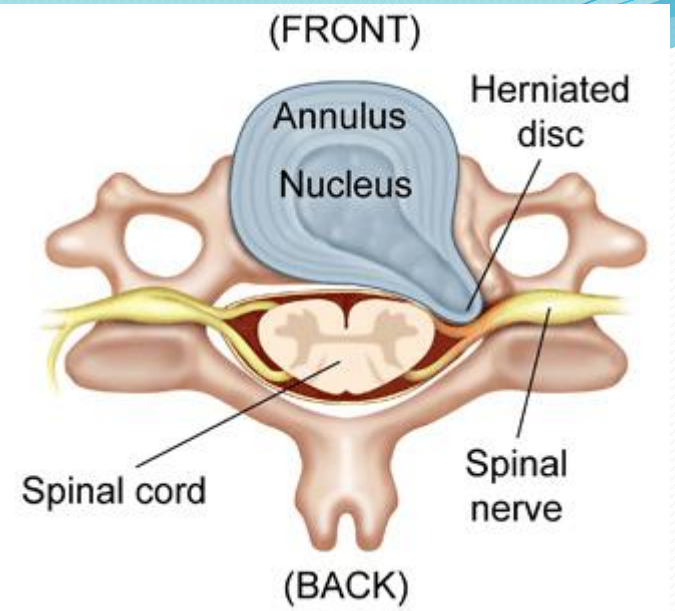


Fig.8: Disc prolapse.

- *Percutaneous Laser Disc Decompression*
  - An optical fiber through a hollow needle is inserted into the herniated disc
  - Laser beam is delivered to the nucleus pulposus through the fiber. This will evaporate some of the water in the nucleus creating a vacuum.
  - The herniated disc will recede back into this vacuum relieving the compression.

Cartilage Formation, Growth and Repair

- Formation of cartilage is from precursor cells called Chondroblasts.

- Growth of cartilage is by two methods:

1) *Interstitial growth:* in which the chondrocytes of the cartilage divide to form new cells.
2) *Appositional growth*: in which the cells of the surrounding perichondrium differentiate into chondrocytes.

- Cartilage repair is usually slow and incomplete due to the avascularity of this tissue.

Bone Tissue And Ossification

DR. MUSTAFA SAAD

(2024)

Bone is a structural type of connective tissue characterized by the presence of a ***calcified extracellular matrix*** (called Bone Matrix) with 3 types of cells:

1. Osteoblasts 2. Osteocytes 3. Osteoclasts

Functions of Bones

1. Support fleshy tissues.
2. Protect vital organs: Skull protects the brain, thoracic cage protects the heart and lungs, vertebral column protects the spinal cord.
3. Store and release of Ca^{2+} and PO_4^{3-} ions.
4. Some bones contain red marrow which is the site of formation of blood cells.
5. Act as levers that multiply force of contraction of muscles.

Osteo- = related to bone. –clast = break. Bone tissue = osseous tissue.

Bone Cells

1) Osteoblasts

- Osteoblasts are responsible for the formation of the organic matrix of bone and the subsequent deposition of minerals.
- They form a single layer of cells on the surface of bones.
- Active cells are cuboidal or low columnar with basophilic cytoplasm. Inactive cells are flattened and less basophilic.

- Osteoblasts secrete the organic matrix from its surface in contact with old bone, creating an area of yet unmineralized bone called ***Osteoid.***

- Later on, osteoblasts will deposit the inorganic components to form the bone matrix.

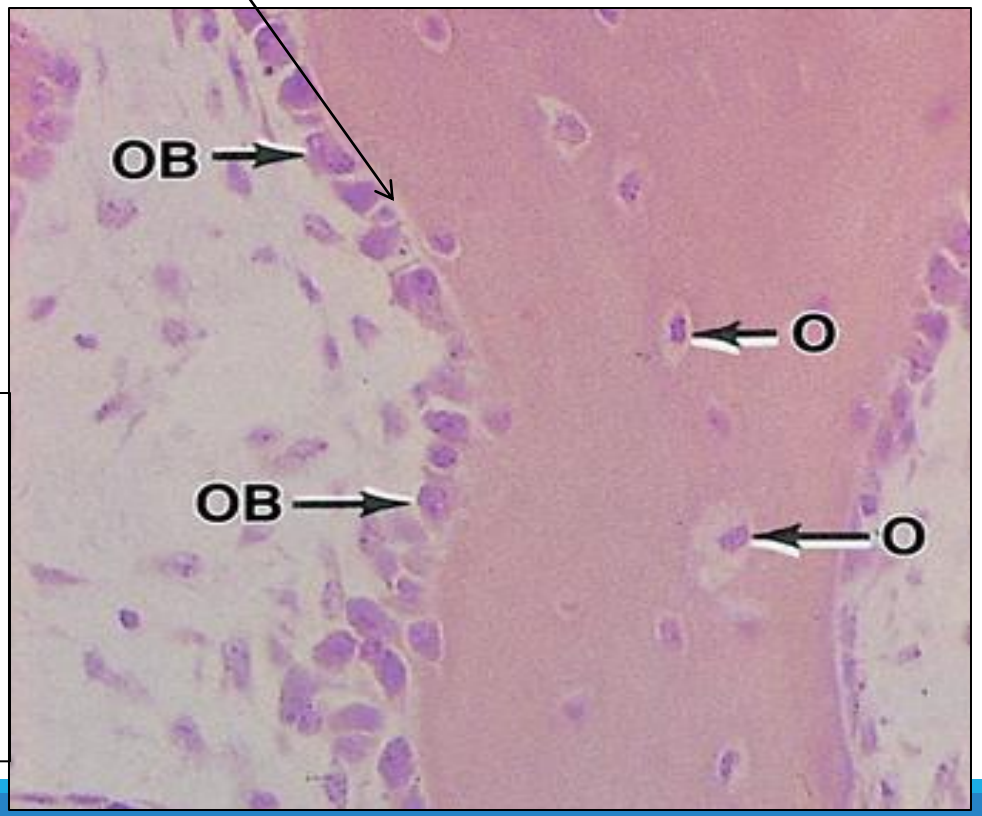


Fig.9: Osteoblasts (OB). Note how they form as single layer. The bone tissue is red in color due to high collagen content. The narrow faint line adjacent to the osteoblasts is the newly formed osteoid.

-oid = similar to, like.

2) Osteocytes

- An osteoblast will eventually be surrounded by the matrix it produced and it'll convert into an Osteocyte. Osteocytes are flattened, almond-shaped cells featuring cytoplasmic processes with reduced rough endoplasmic reticulum and Golgi complex and darker nuclei.

- They're involved in the maintenance of the bony matrix.

- Each *osteocyte* is located within a *lacuna*. Its *processes* are located in bony canals called *canaliculi*.

- Processes of osteocytes are connected with each other by gap junctions, allowing transport of nutrients between cells. This is vital because the passage of nutrients through the calcified matrix is difficult.

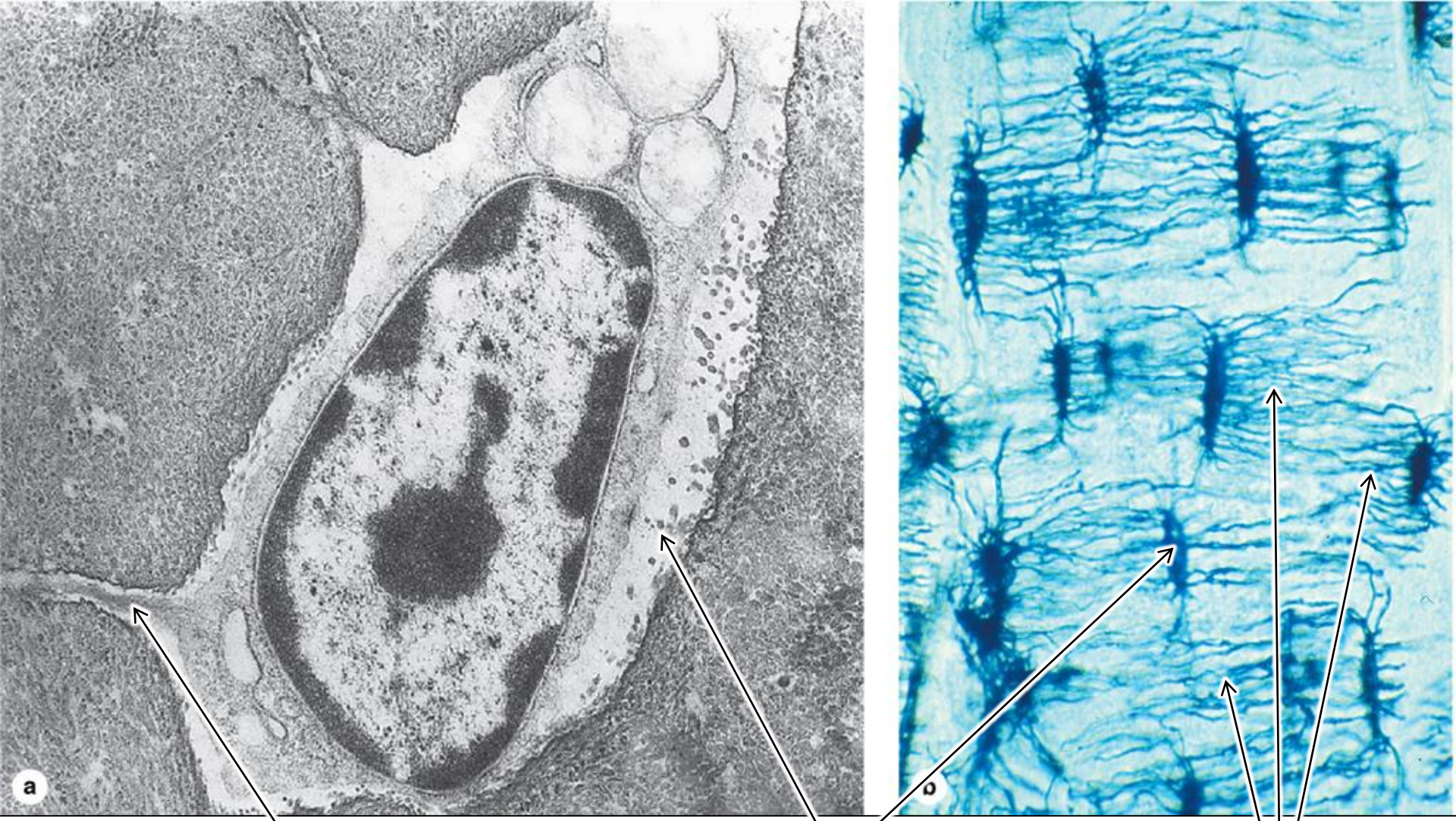


Fig.10: Osteocytes. The EM image to the left clearly shows the lacuna, the cell processes and the canaliculi.

3) Osteoclasts

- Osteoclasts are large, motile, multinucleated cells.
- They're formed by the union of several bone-marrow derived mononucleated cells.
- They're responsible for the resorption of bone.

Fig.11: Osteoclast in action.

Process of Bone Resorption:

1) Osteoclast works in a specified depression called ***Resorption Bay*** (Howship's Lacuna).

2) Their cell membrane facing the matrix is thrown into folds called the ***Ruffled Border*** (to increase surface area).

3) Around the ruffled border the cytoplasm is rich in actin filaments which help in adhering the cell to the matrix (this area is called the ***Circumferential adhesion/clear zone***).

4) Into the subcellular space thus formed, H^+ ions are pumped and lysosomes fuse with the cell membrane and release their secretions (including collagenases) to the outside. In this way, the collagen and hydroxyapatite of the matrix are dissolved.

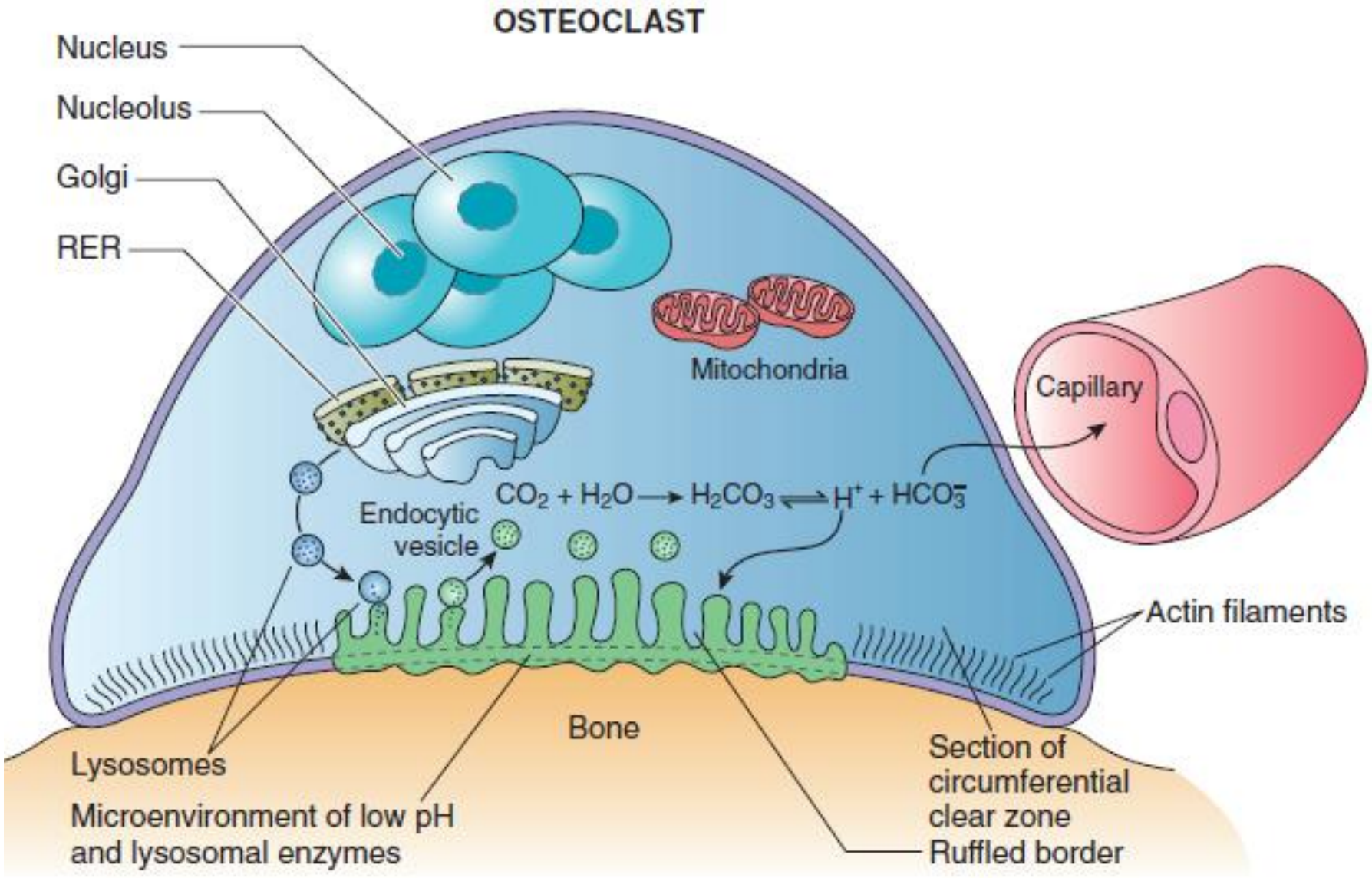


Fig.12: Bone resorption by osteoclasts

Disease	Pathology	Notes
Osteitis fibrosa cystica	Increased level of PTH causes excessive stimulation of osteoclasts that leads to increased resorption. Cysts are formed within the bone.	– Bones are decalcified and liable for fracture. – High Ca^{2+} level in blood increases risk of renal stones.
Osteopetrosis (Marble bone disease)	Genetic disorder in which there's abnormality in osteoclasts that leads to decreased resorption.	– Bones are thicker and appear denser on X-rays. – The bone marrow cavity is narrowed → anemia and increased risk of infection.

PTH = Parathyroid Hormone. –itis = inflammation. Petrosis = stone.

Bone Matrix

a) Inorganic Components: (50% of dry weight of bone)
- Mainly Hydroxyapatite crystal.
- Various ions and compounds.

b) Organic Component:
- Fibers: Collagen.
- Ground substance: Proteoglycans and multiadhesive Glycoproteins.
- Ca^{2+} binding proteins.
- Alakaline Phsophatase in matrix vesicles (which increase PO_4^{3-} concentration).

- ✓ In the matrix, the association of minerals with collagen fibers is responsible for the hardness and resistance of bones.
- ✓ If Ca^{2+} is removed, the bone will maintain its shape but become flexible as a tendon.
- ✓ If collagen is removed, the bone will maintain its shape but becomes fragile and easily broken.

Periosteum and Endosteum

- ***Periosteum***: A connective tissue layer that covers the outer surface of the bone. It consists of an outer layer of dense fibrous tissue with fibroblast, and an inner single layer of osteoprogenitor cells. A number of collagen fibers pass from this layer to the bone matrix attaching them together (these are called *perforating fibers*).

- ***Endosteum***: A thin layer that lines the inner surface of the bone. Formed of a single layer of osteoprogenitor cells with osteoblasts.

Peri- = around. Endo- = inside.

Fig.13: Periosteum (P) and Endosteum (E). The spaces between the bone tissue are filled with blood vessels and blood elements. The perforating fibers cannot be seen by routine LM study. Also note the osteon (O).

Classification of Bone

- **Gross Morphology of section**
  - Compact
  - Cancellous
- **Histological Features**
  - Primary
  - Secondary
- **Shape of bone**
  - Long
  - Short
  - Flat
  - Irregular

Cancellous = latticed, porous.

According to Gross Morphology:

- In a section, a part of the bone appears as a dense area with generally no cavities. This is called *compact bone*.

- Another part has several interconnected cavities. This is called *spongy (cancellous) bone*.

- Histologically, both the compact bone and the trabeculae of the spongy bone have the same features.

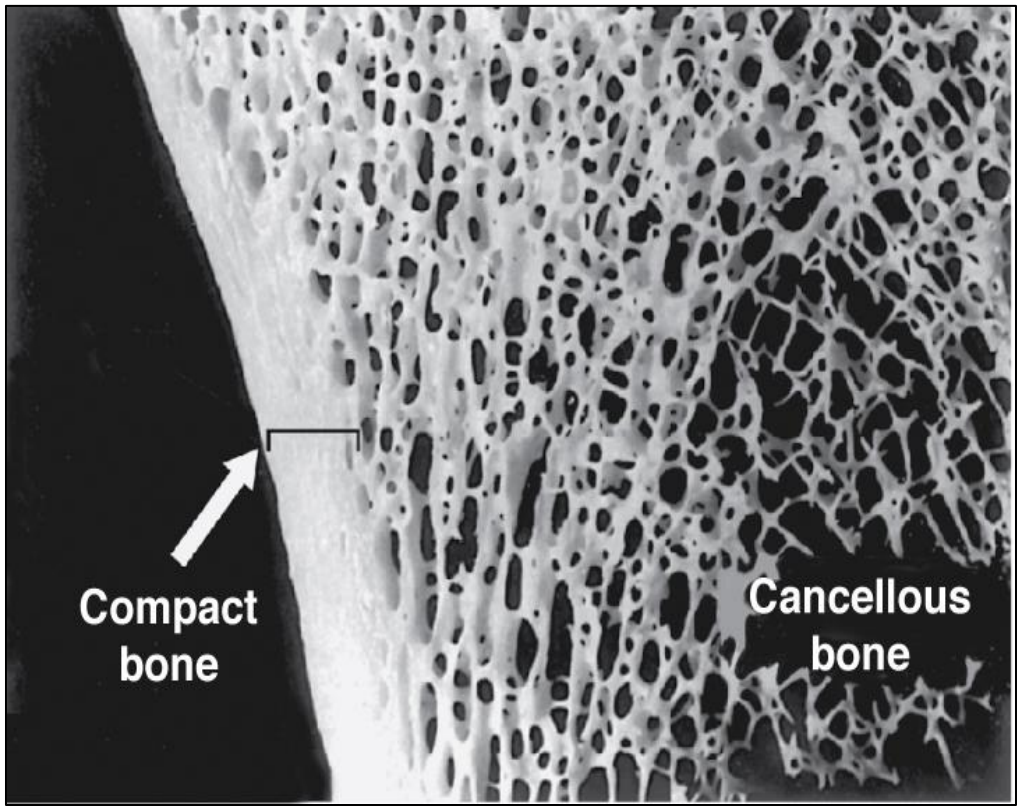


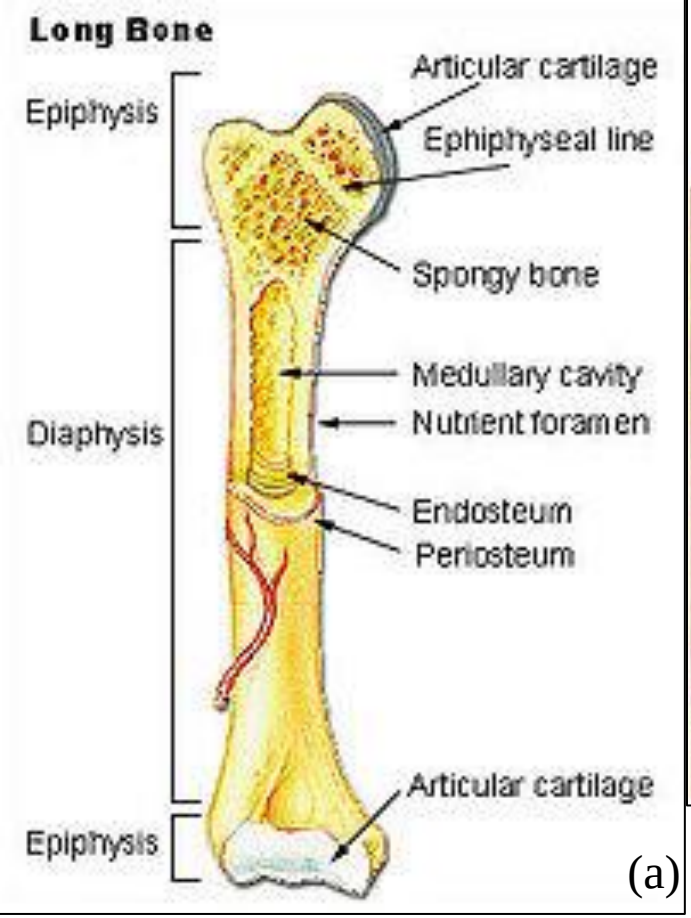
Fig.14: Compact and Cancellous bones.

According to Shape:

- ***Long Bones*** have a tubular shaft, the diaphysis, and an expanded epiphysis at each end. The shaft has a central cavity for the bone marrow (medullary cavity). The shaft is mostly composed of compact bone with a thin layer of spongy bone surrounding the cavity. The epiphyses are composed of cancellous bone surrounded by a thin layer of compact bone.

- ***Short bones*** are composed of spongy bone completely surrounded by a thin layer of compact bone.

- ***Flat bones*** consists of two thin layers of compact bones (plates, tables) separated by a layer of spongy bone called diploë.



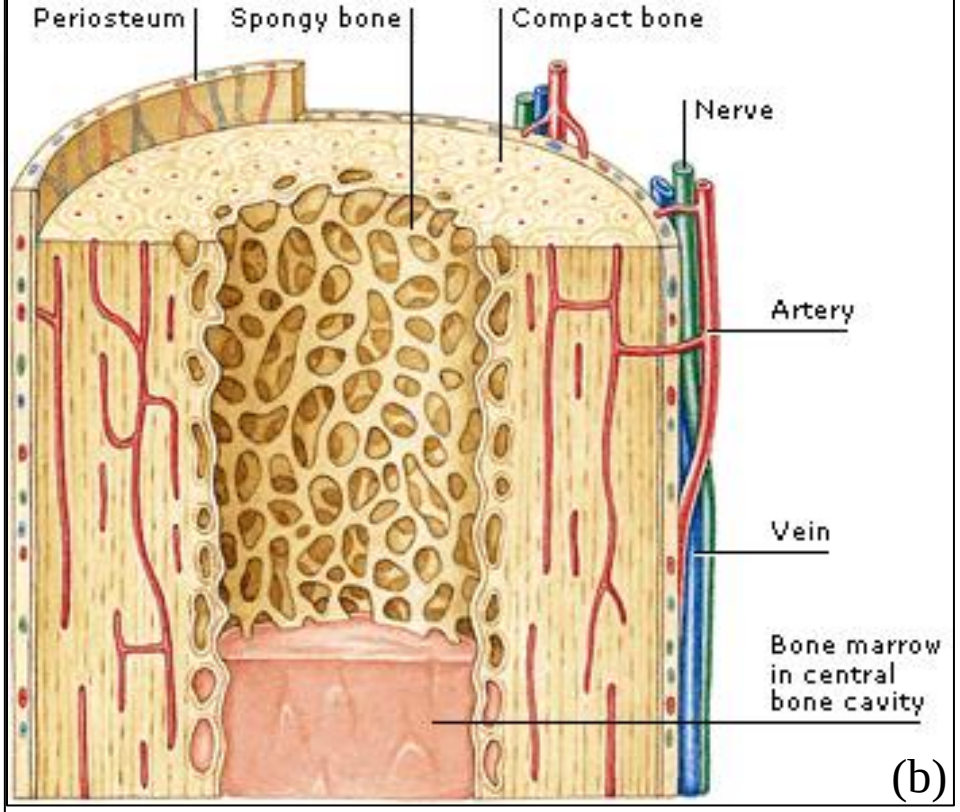
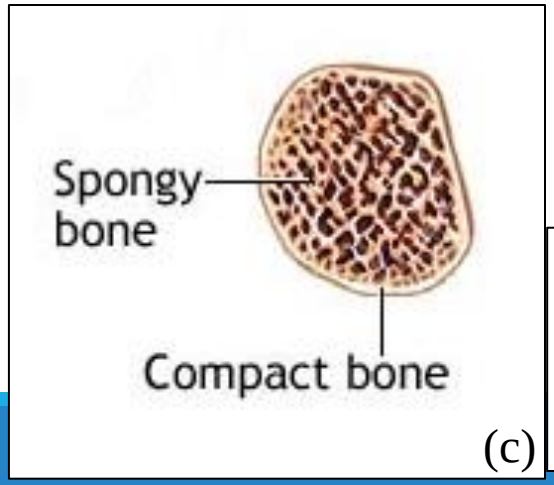


Fig.15: Composition of various types of bone. (a) Long bone. (b) Diaphysis of long bones. (c) Short bones. (d) Flat bones of the skull.



According to Histological Features:

1) Primary (Woven) Bone:

- Characterized by the irregular arrangement of its collagen fibers. Osteocytes are more abundant. And it appears less dense on X-Rays due to less mineral content.
- It's the first type of bone to appear during embryonic development and in fracture repair.
- It's replaced by secondary bone, except in areas of *tendon attachment*, *tooth sockets*, *and near the sutures of the skull bones*.

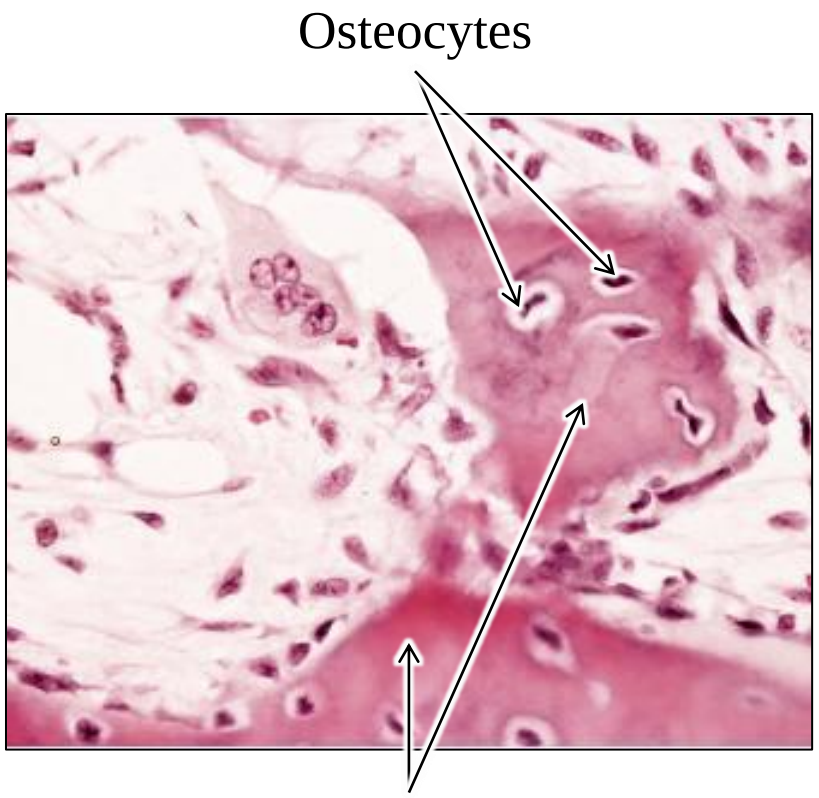


Fig.16: Primary bone.

2) Secondary (Lamellar) Bone:

- Characterized by the arrangement of the matrix into multiple layers called ***lamellae***. The osteocytes are located inside lacunae found between the lamellae.

The lamellae could be arranged as:

a) Parallel layers just inside the periosteum (*the external circumferential lamellae*) or around the bone marrow cavity (*the internal circumferential lamellae*)

Lamellae (singular = lamella) = Thin plates, layers.

b) Concentric circles around a central canal forming an *Osteon* (*Haversian System*). This canal contains blood vessels, nerves, and loose areolar connective tissue. The outer layer of the osteon is rich in collagen and is called the *cement line*.

- The collagen fibers in each lamella are parallel to each other and helically arranged. The collagen fibers in adjacent lamellae are at right angles to each other.

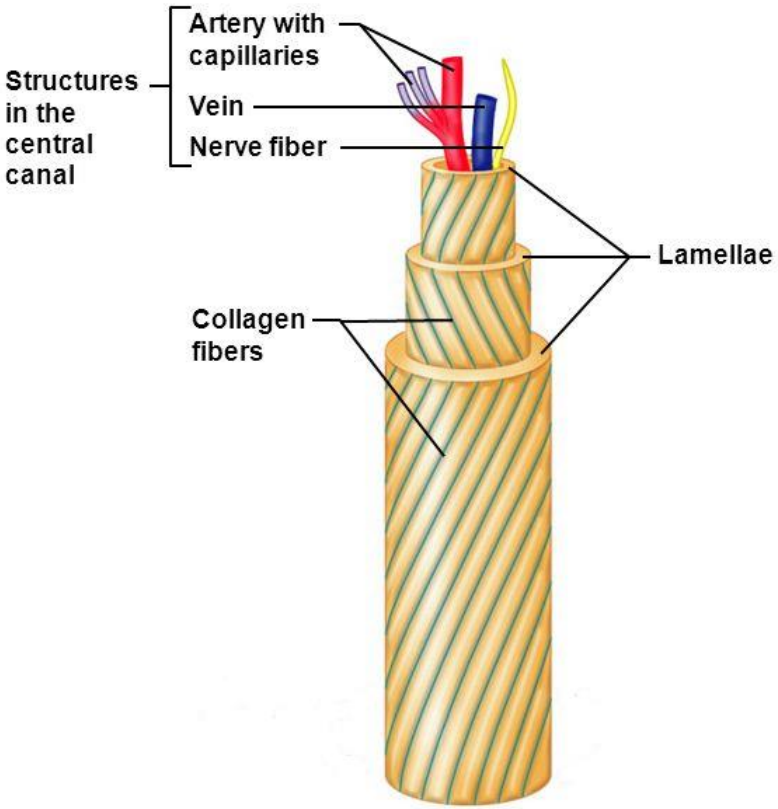


Fig.17: An osteon.

- The central canals are connected to the periosteum, the bone marrow cavity, and to each other by transverse (or oblique) *Perforating canals*.

c) Irregularly shaped groups of lamellae called *interstitial lamellae*. They are found between the previous two and represent the remnants of osteons that have been resorbed.

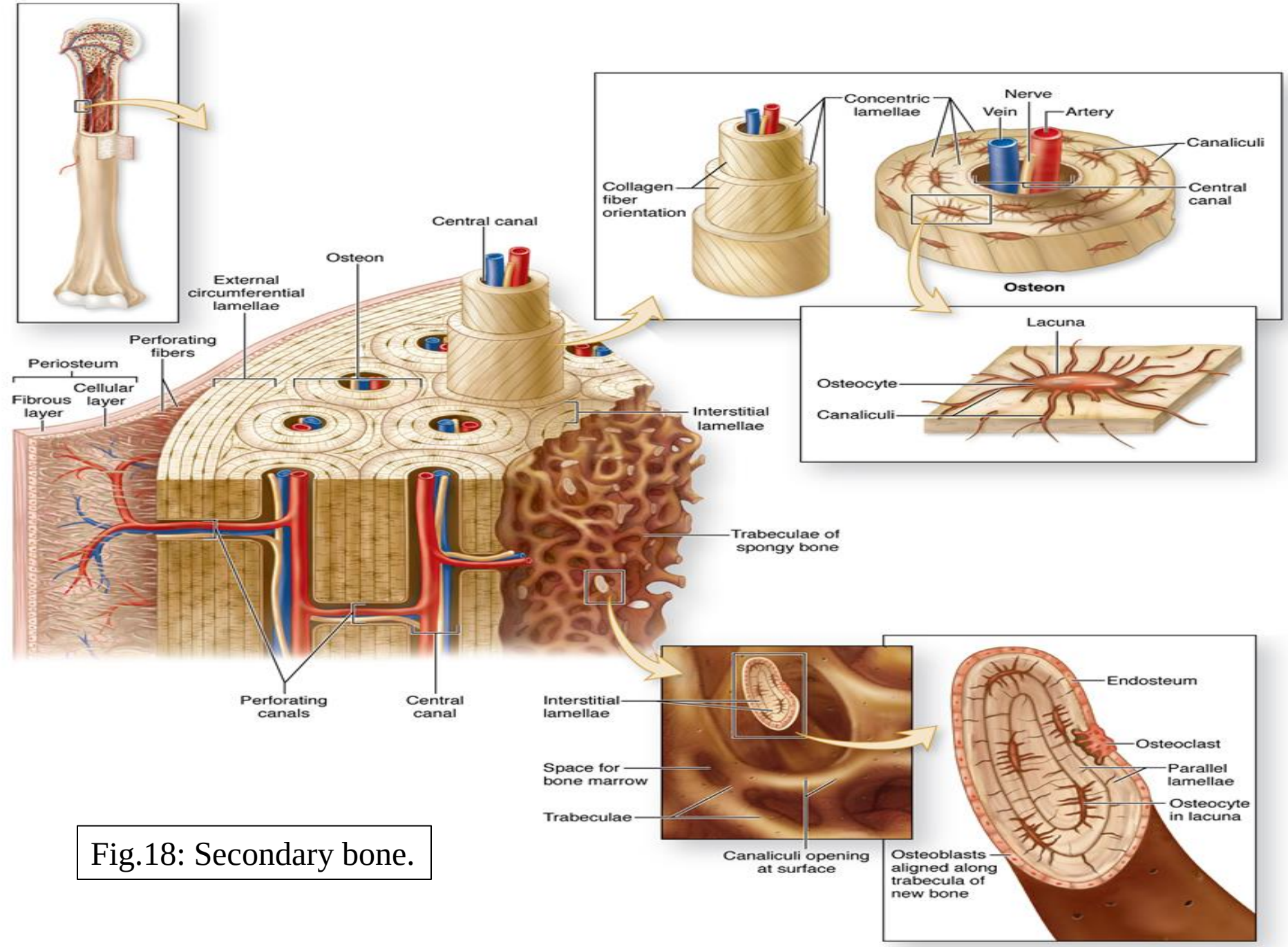
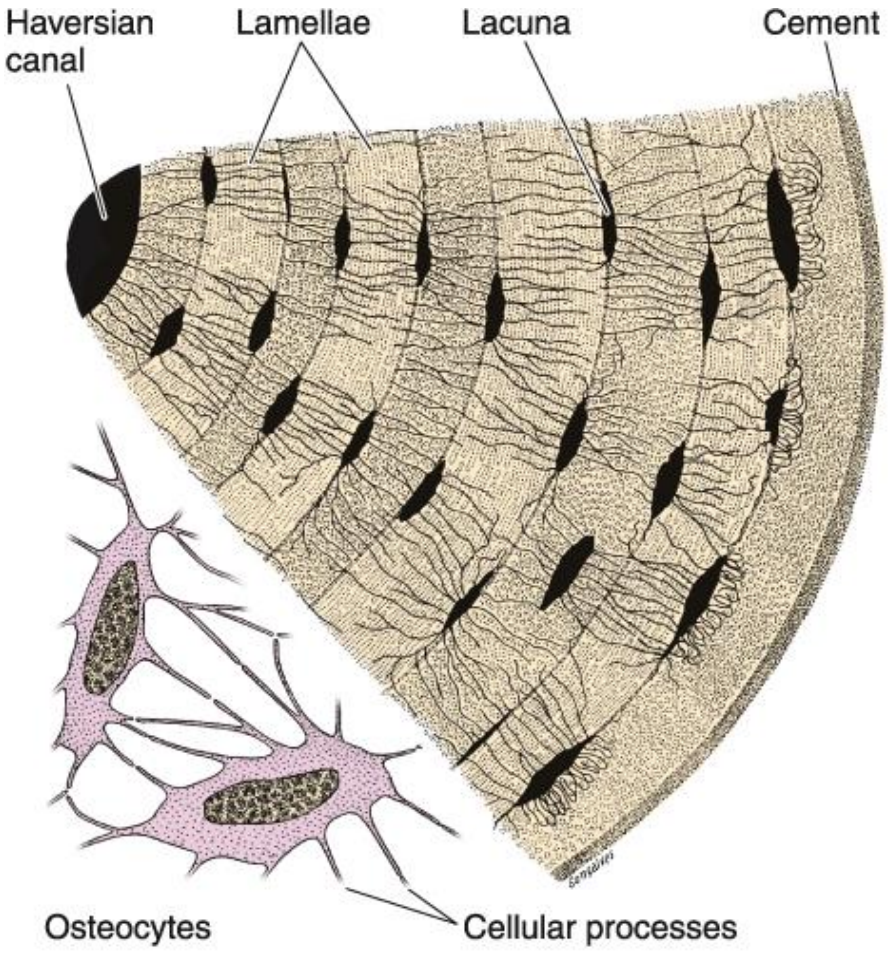


Fig.18: Secondary bone.



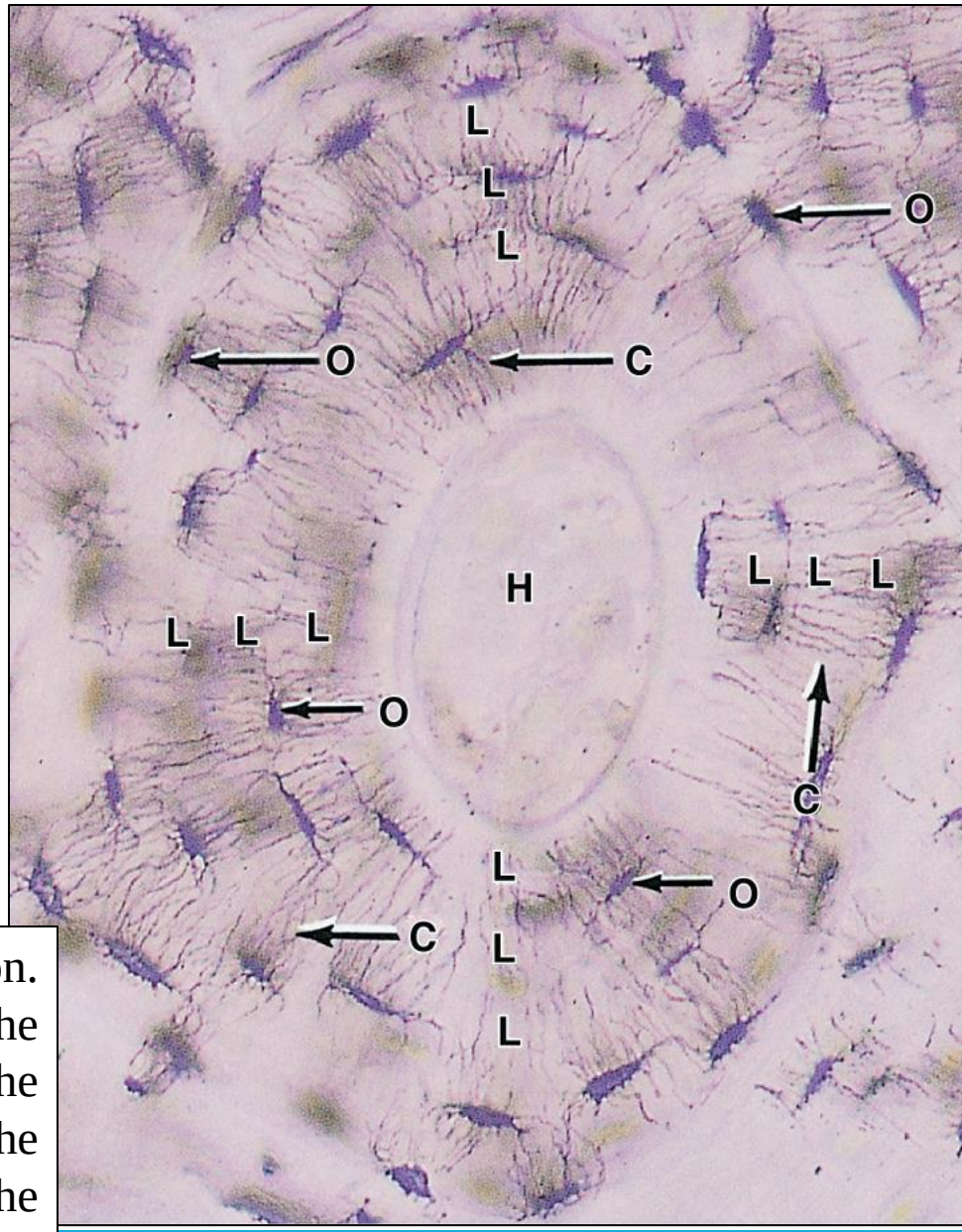


Fig.19: To the right, section through an Osteon. Note the concentric arrangement of the lamellae (L). The central canal (H) is in the center. (O) = osteocytes. (C) = Canaliculi. The image above shows part of an osteon. Note the cement line.

Ossification

- The process by which new bone tissue is formed.

- It's of two types:

1. Intramembranous: Is the formation of bone from a group (membrane) of mesenchymal cells. It's the process by which most of the flat bones are formed.

2. Endochondral: Is the formation of bone from the matrix of a pre-existing hyaline cartilage model of the bone. Long and short bones are mostly formed by this method.

Intramembranous Ossification:

a) In ***Ossification Centers***, some mesenchymal cells differentiate into osteoblasts.

b) These osteoblasts will form osteoid which will later become calcified. Some osteoblasts will be surrounded by the bone matrix forming osteocytes in lacunae.

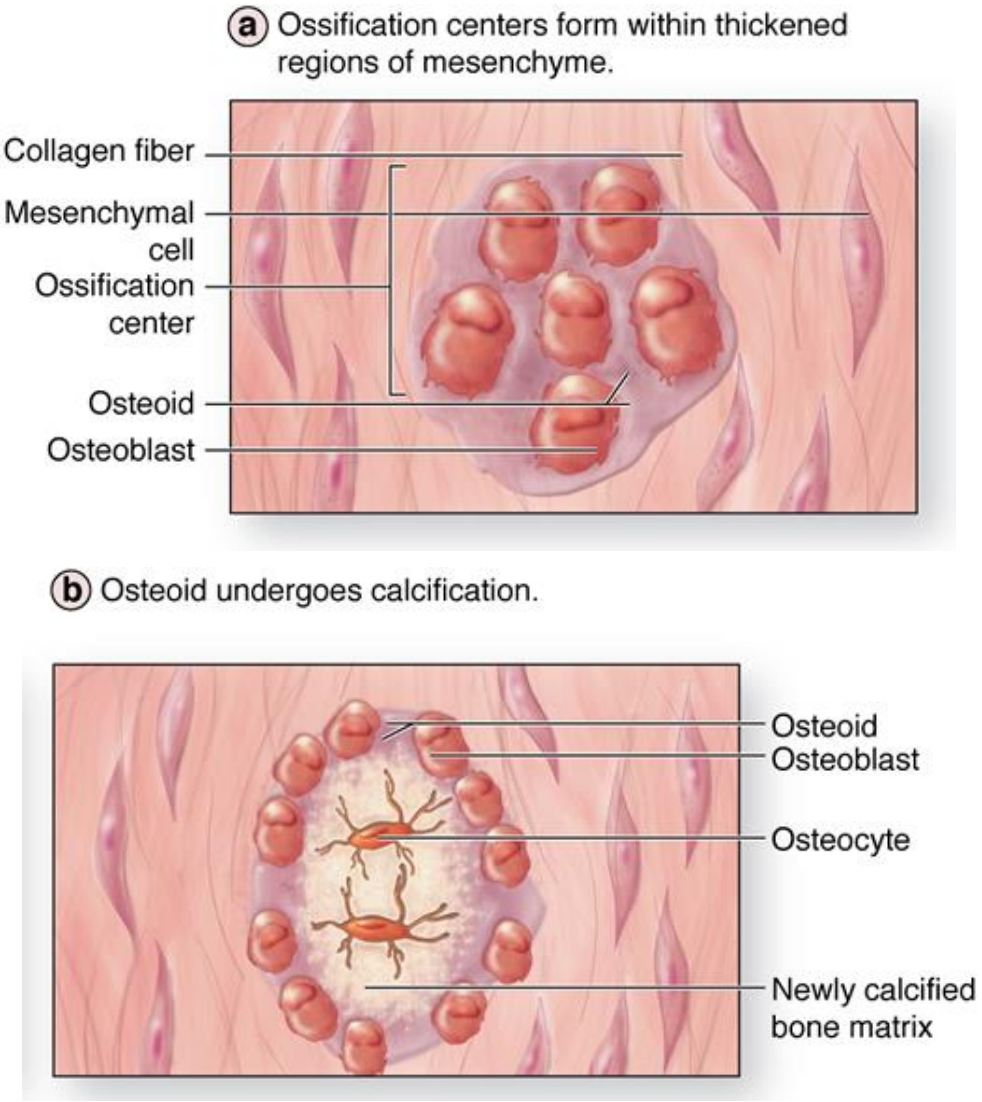


Fig.20: Intramembranous ossification.

c) Woven bone is formed at first. Several centers will fuse forming trabeculae surrounding cavities containing blood-forming and mesenchymal cells.

d) Lamellar bone replaces the woven bone as compact bone surrounds the spongy bone.

e) The remaining non-calcified mesenchymal tissue will form the periosteum and endosteum.

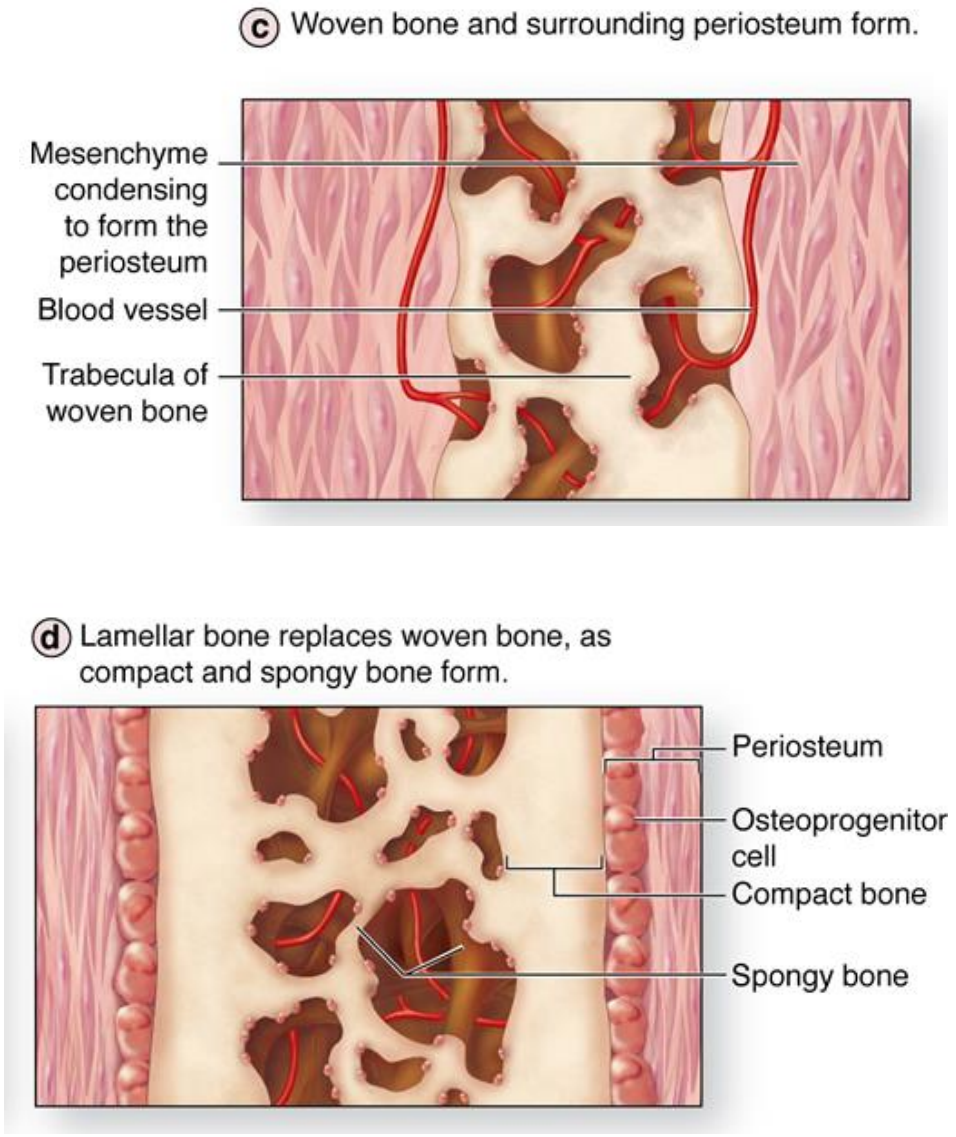


Fig.20 (continued): Intramembranous ossification.

Endochondral Ossification:

1) A hyaline cartilage model of the bone is formed. Cells in the perichondrium will differentiate to osteoblasts that will form a ***bone collar*** around the diaphysis of the model. This will prevent passage of nutrients from the perichondrium to the chondrocytes.
2) Chondrocytes will hypertrophy, enlarging their lacunae and compressing the cartilage matrix between them. The compressed matrix will be calcified and the chondrocytes will die forming a porous structure.
3) Osteoclasts will dig tunnels through the calcified matrix. Through these tunnels, blood vessels and osteoprogenitor cells from the perichondrium (which is now called periosteum) will reach the matrix.

4. The osteoprogenitor cells will form osteoblasts that line the cavities of the porous structure. Osteoblasts will produce primary bone which will later convert into secondary bone.
5. Ossification in the diaphysis is called the ***primary ossification center.*** Later in life, ***secondary ossification centers*** appear in the epiphyses by a similar process.
6. Cavities will be formed during this process and these will be filled with bone marrow.

- The hyaline cartilage remains in two areas:
  - The ***articular cartilage***. This persists throughout life.
  - The ***epiphyseal plate***, which disappears during adulthood.

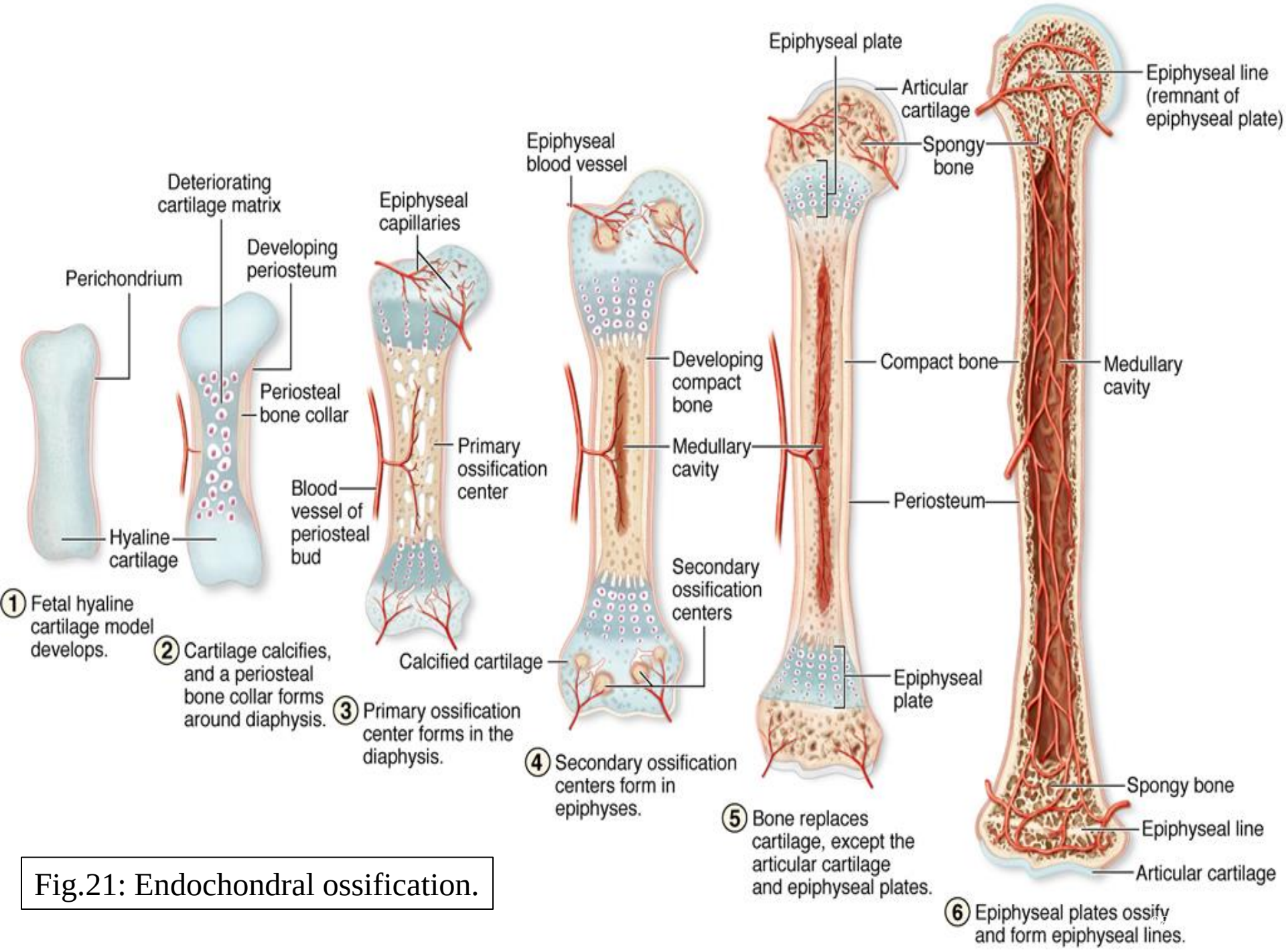


Fig.21: Endochondral ossification.

Bone Remodeling (Remodeling Cone):

- It occurs throughout life.

- Osteoclasts resorb old bone forming a tunnel (cutting cone).
- The tunnel is invaded by osteoprogenitor cells and sprouting loops of capillaries from the endosteum or periosteum.
- Osteoblasts develop, line the wall of the tunnel, and begin to secrete osteoid in a cyclic manner, forming a new lamella to the inside of the older ones trapping osteocytes inside lacunae (closing cone).
- The tunnel becomes constricted with multiple concentric layers of matrix, and its lumen finally becomes the central canal of a mature osteon.

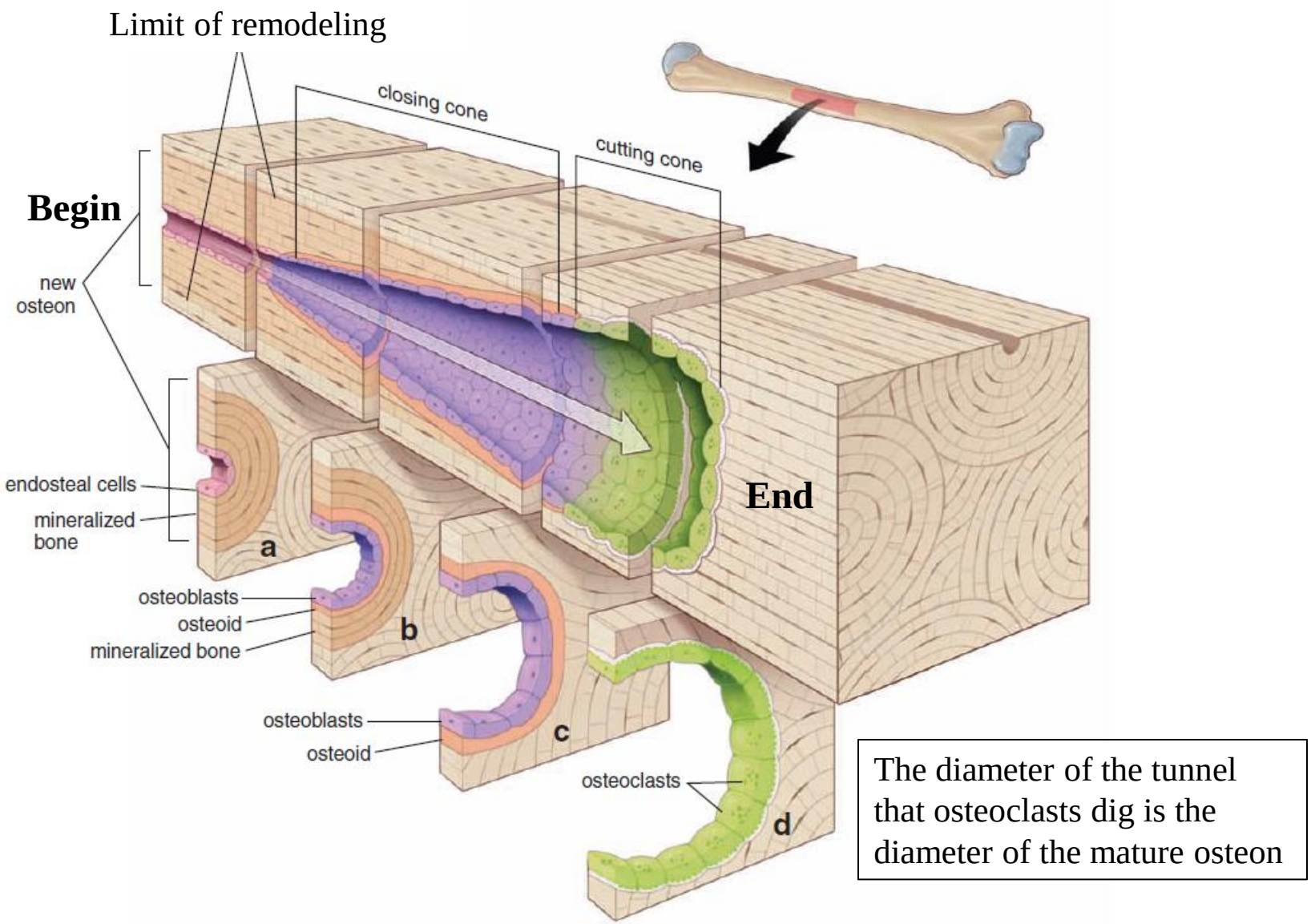


Fig.22: Bone remodeling cone. The process begins by osteoclasts (green cells) digging a tunnel (section d). Osteoblasts (purple cells) will line the tunnel and produce osteoid (orange) that will later mineralize (sections c and b). More layers are formed until a mature osteon is formed (section a) with a central canal lined by endosteal cells (pink cells).

Bone Growth & Repair

- Increase in length of bones occur at site of epiphyseal plate before they're closed. After closure of the plates during adulthood, no further increase in bone length can occur. The time of closure of the plate is specific for the bone. This can be used to determine the age of the person.
- Increase in width of bone can occur throughout life by appositional growth from the periosteum.
- Bone growth is affected by several hormones in the body, like growth hormone.

- Repair of bone is usually very well because bones are well vascularized.

Tetracycline and Bones:

- Tetracycline binds with great affinity with Ca^{2+} in recently deposited bone matrix. Based on this interaction, a method was developed to measure the rate of bone apposition, an important parameter in the study of bone growth and the diagnosis of bone growth diseases.

- Tetracycline is administered twice to a patient, with an interval of 5 days between injections. A bone biopsy is then performed and the sections are studied by means of fluorescence microscopy. The distance between the two fluorescent layers is proportional to the rate of bone apposition.

Tetracycline must not be given to a pregnant or lactating women or to a child whose teeth are erupting, because it may bind to Ca^{2+} of the newly forming teeth of the child leading to the permanent discoloration of the teeth.

Fig.23: Staining of teeth as a side effect of tetracycline use.

The Epiphyseal plate

- Epiphyseal cartilage is divided into five zones starting from the epiphyseal side of cartilage:

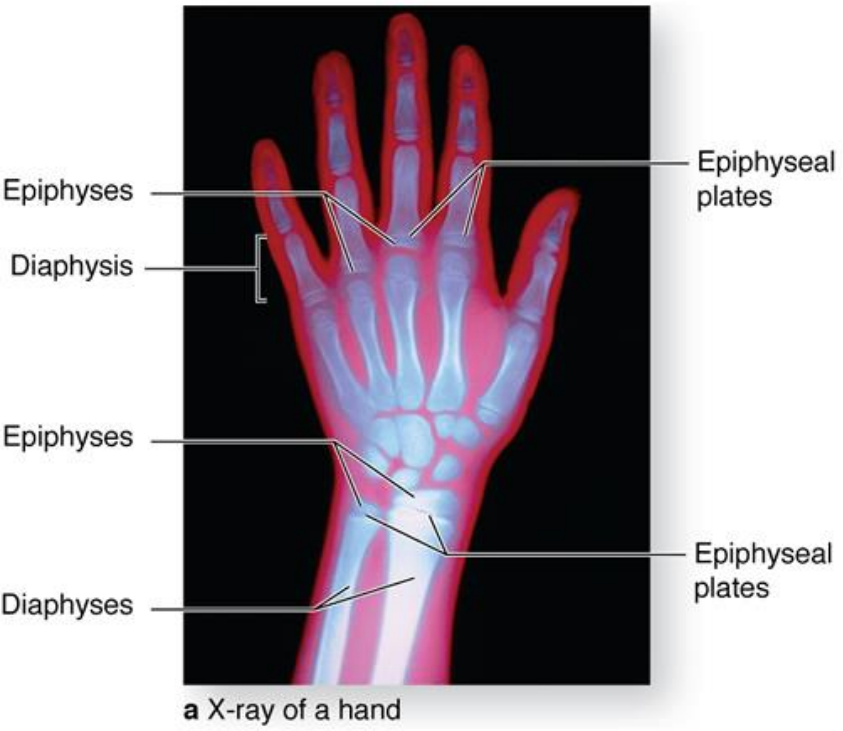
1. **The resting zone:** consists of hyaline cartilage with typical chondrocytes.

2. **Proliferative zone**: chondrocytes divide rapidly and form columns of stacked cells parallel to the long axis of the bone.

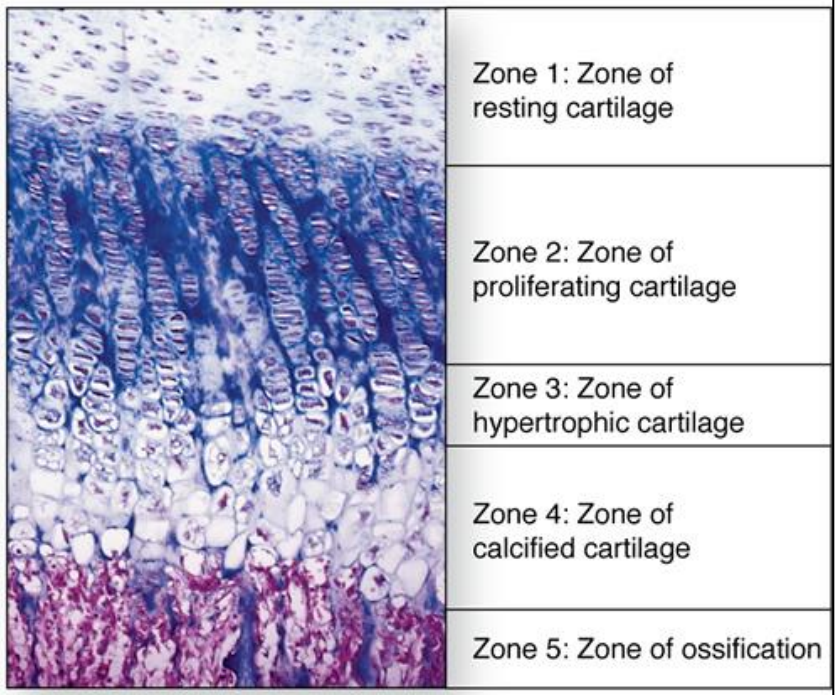
3. **The hypertrophic zone:** contains enlarged chondrocytes. The matrix is reduced to thin septa between the chondrocytes.

4. **The calcified zone**: Death of chondrocyte with calcification of the thin septa of cartilage matrix.

5. **The ossification zone:** Blood capillaries and osteoprogenitor cells originating from the periosteum invade the cavities left by the chondrocytes. The osteoprogenitor cells form osteoblasts which will deposit bone matrix.



a X-ray of a hand



b Epiphyseal plate

Fig.24: The epiphyseal plate

- Before closure, each layer converts to the next at the same rate, with the formation of new cartilage in zone 1 and new bone in zone 5. Therefore, there's no change in the relative size of the plate. The plate moves away from the center of the bone thus increasing bone length. The chondrocytes in the plate will, eventually, start to die without forming new cartilage. The dead cartilage is replaced by bone until all the plate becomes ossified at the time of closure.

Thank You

Do not despise a snake for having no horns; for it may one day become a dragon.