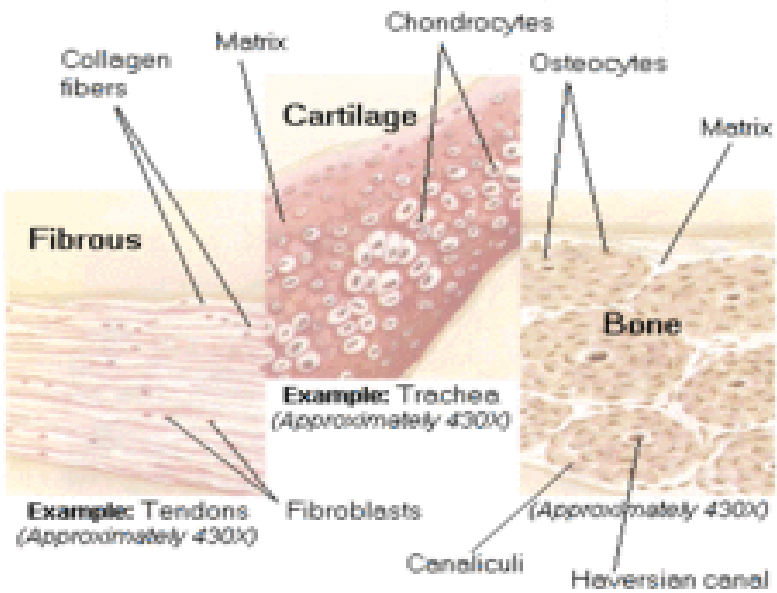


## Connective Tissues



# Connective tissue

## Connective tissue proper



**Cartilage**

**Bone**



# Origine and function of c.t.

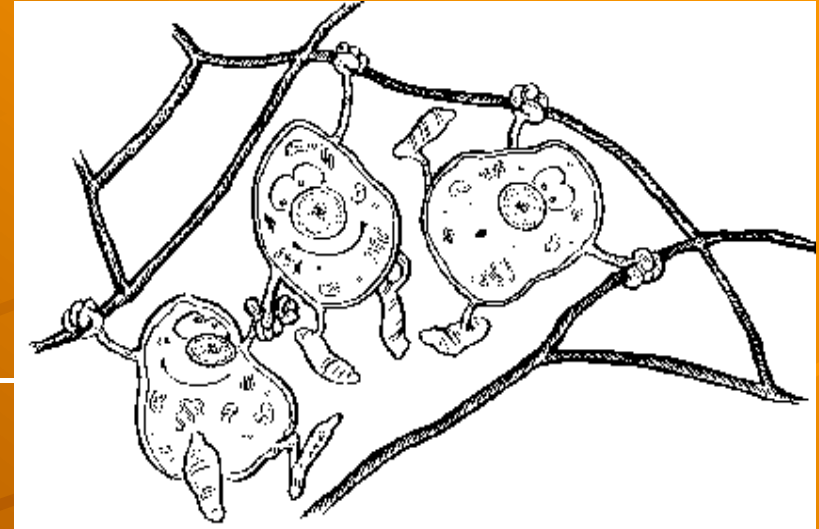
■ Origin – embryonic mesenchyme

■ Functions:

- **nutritive** (blood vessels and difusion of nutriens)
- **protective** – immunocompetent cells and antibodies production
- **connective** – between different tissues
- **mechanical** (supporting and mechanical protection of organ – *in skull, thorax, pelvis*)

# Connective tissue types

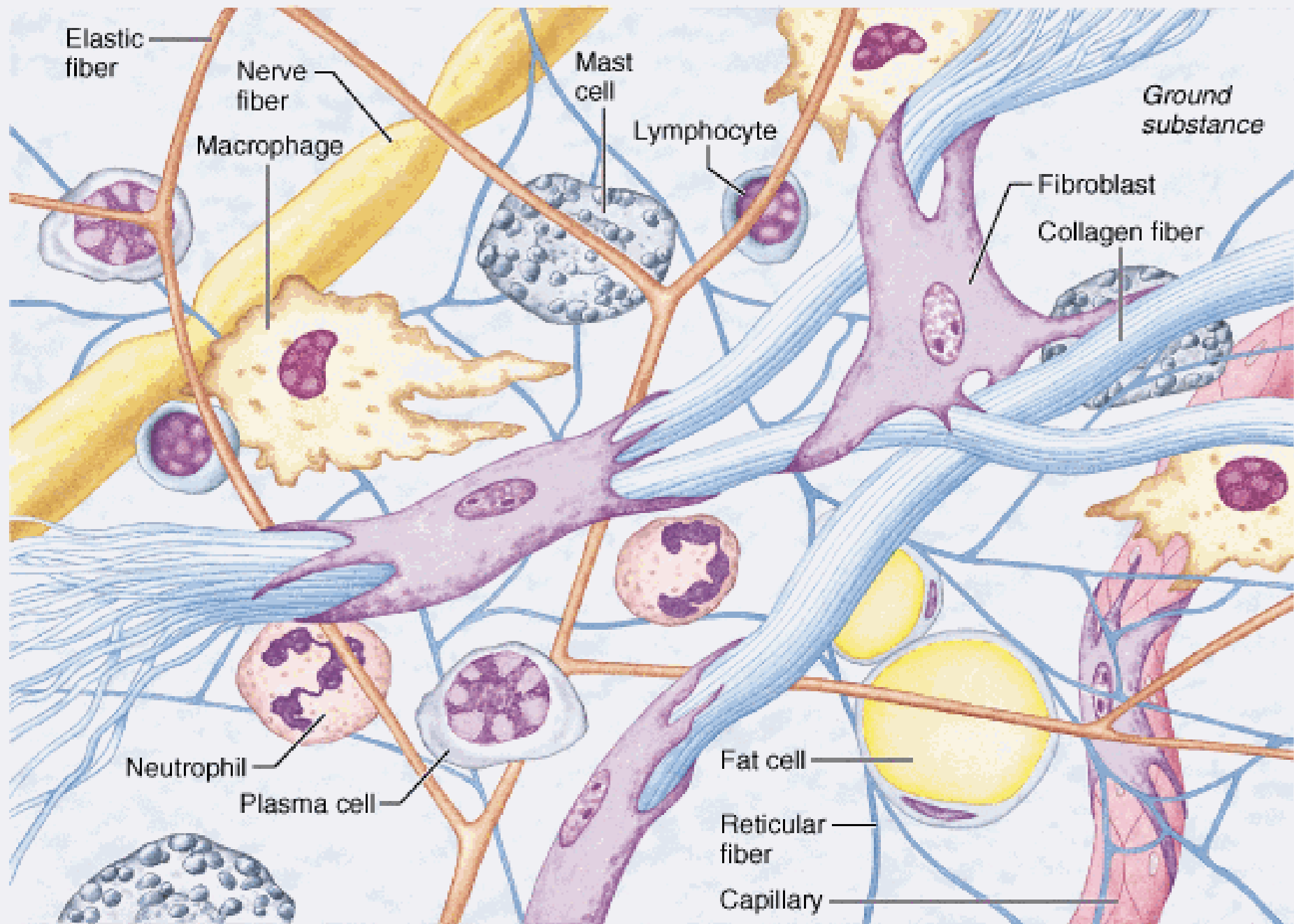
- ✦ C.t. proper
- ✦ Cartilage
- ✦ Bone



- ✦ general structure:
  - cells
  - intercellular matter

fibers

ground amorphous substance

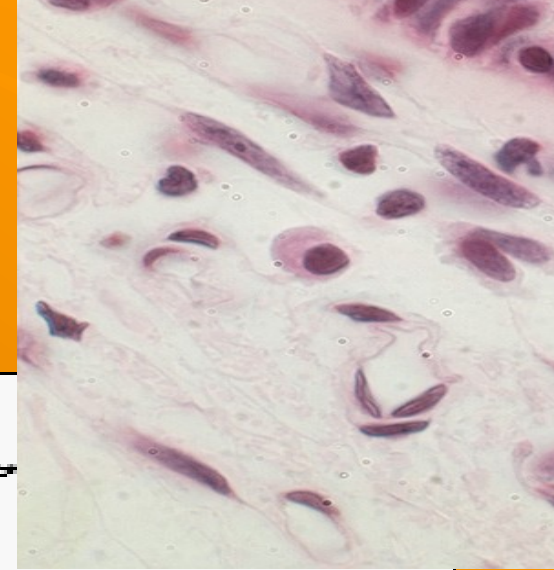


# Connective tissue proper - cells

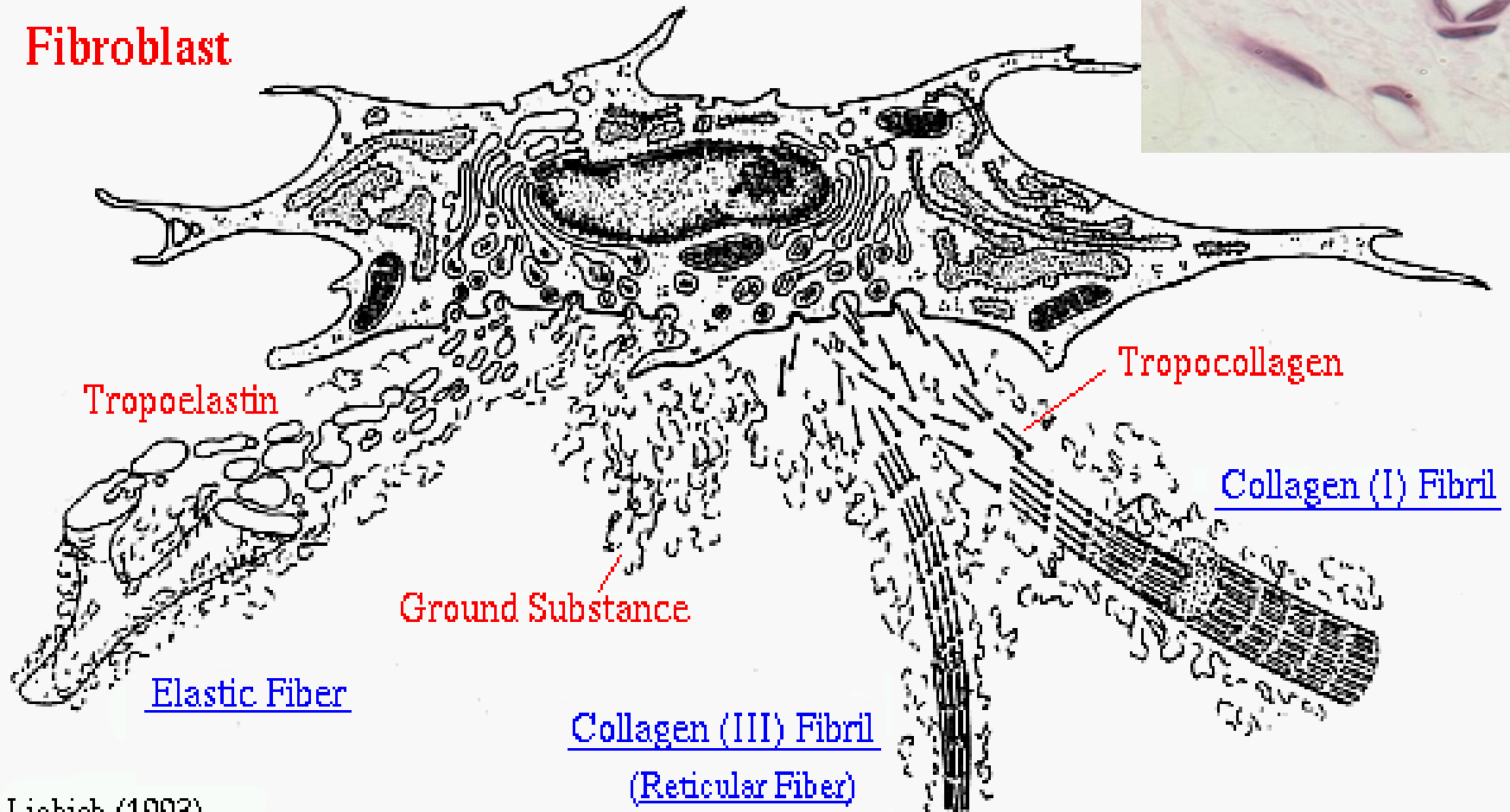
## FIXED CELLS

- ▣ Fibroblasts, fibrocytes
- ▣ Reticular cells
- ▣ Fat cells (univacuolar, *multivacuolar*)
- ▣ Pigment cells
- ▣ Undifferentiated cells

# Fibroblasts, fibrocytes



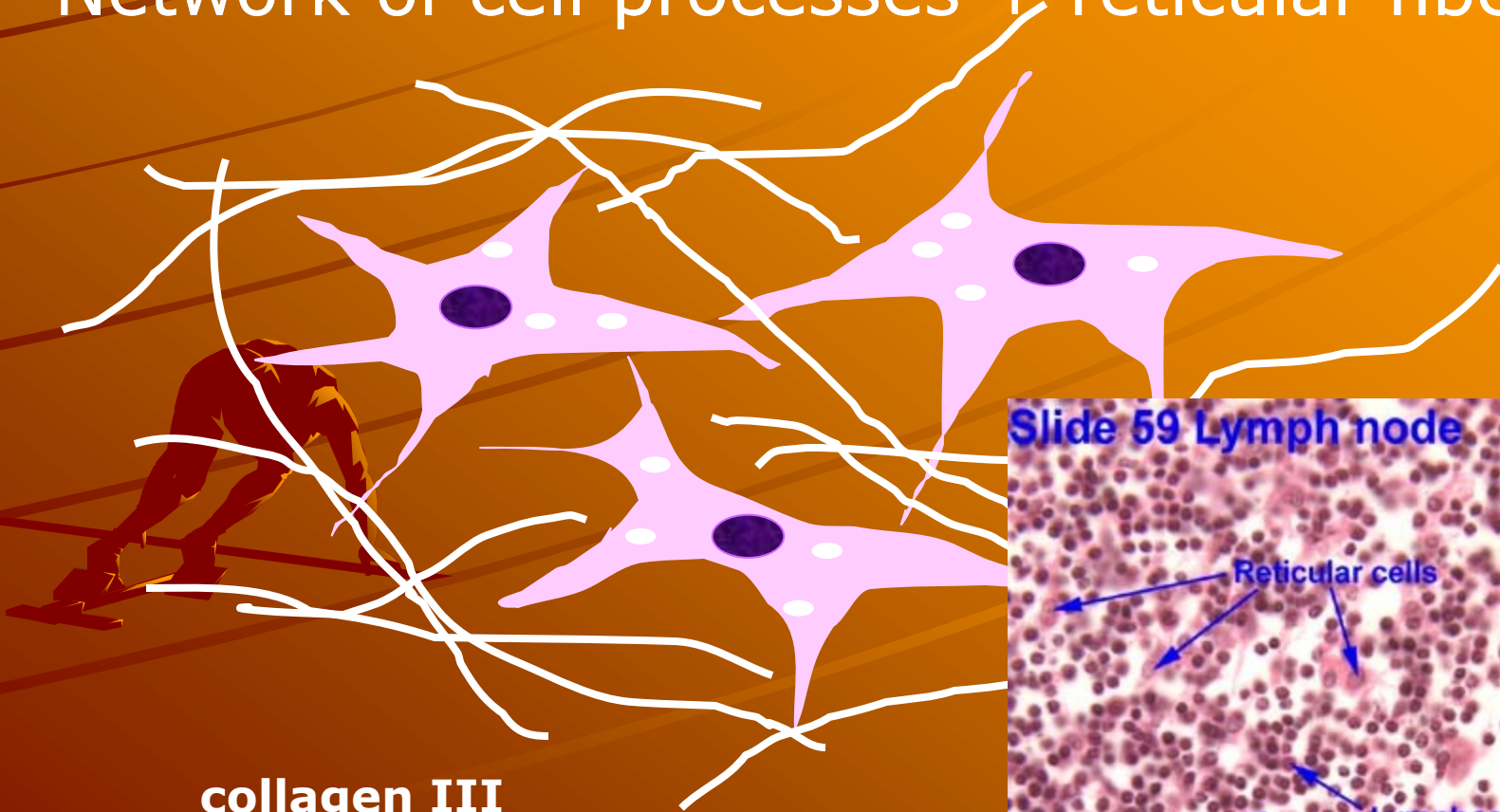
## Fibroblast



Liebich (1993)

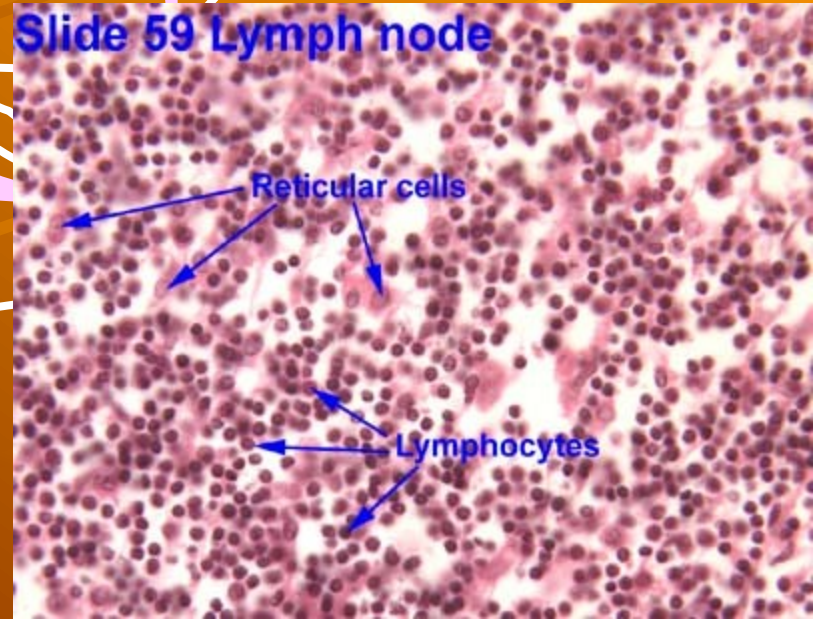
# Reticular cells

Network of cell processes + reticular fibers



collagen III

Slide 59 Lymph node



# Adipose (fat) cells

univacuolar  
(white adip.c.t.)



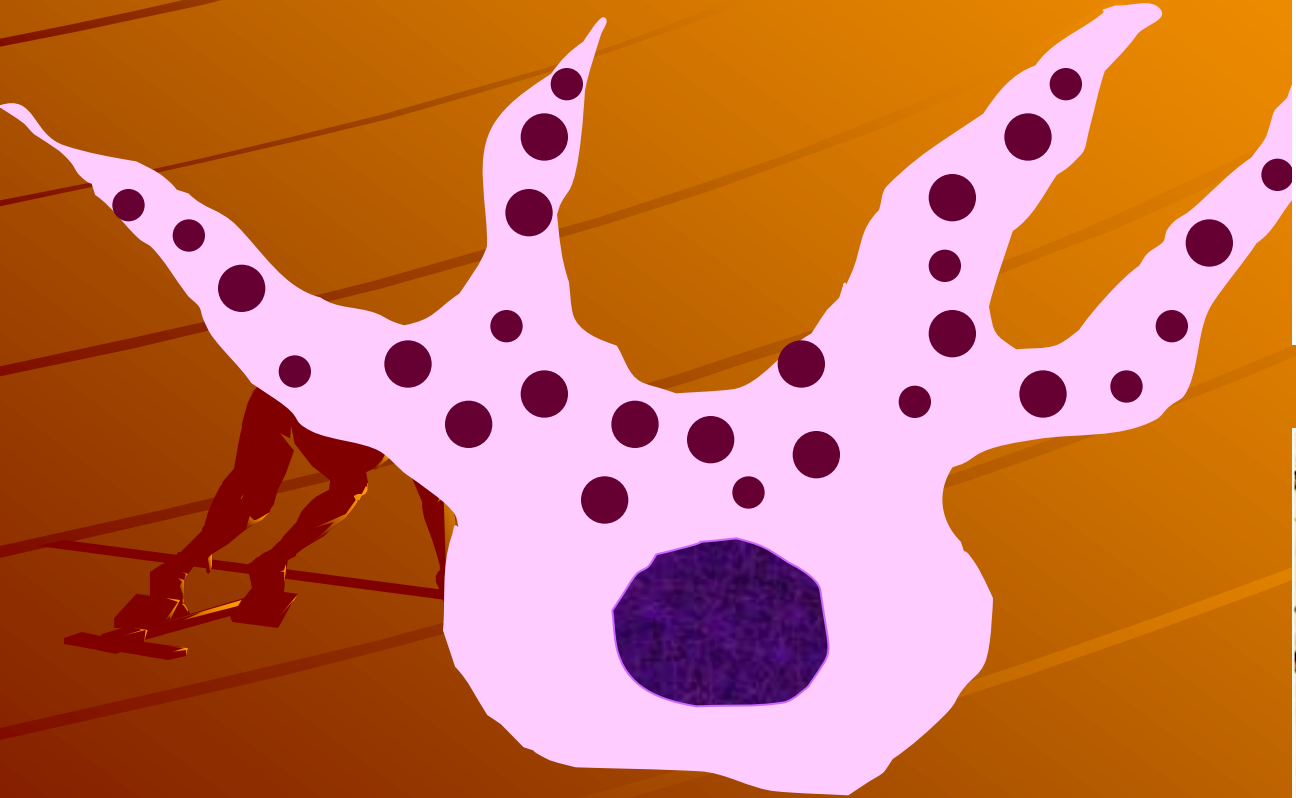
multivacuolar  
(brown adip.c.t.)



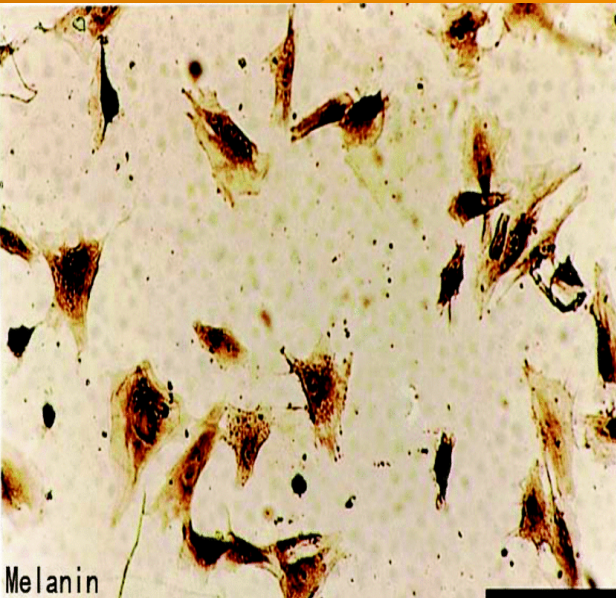
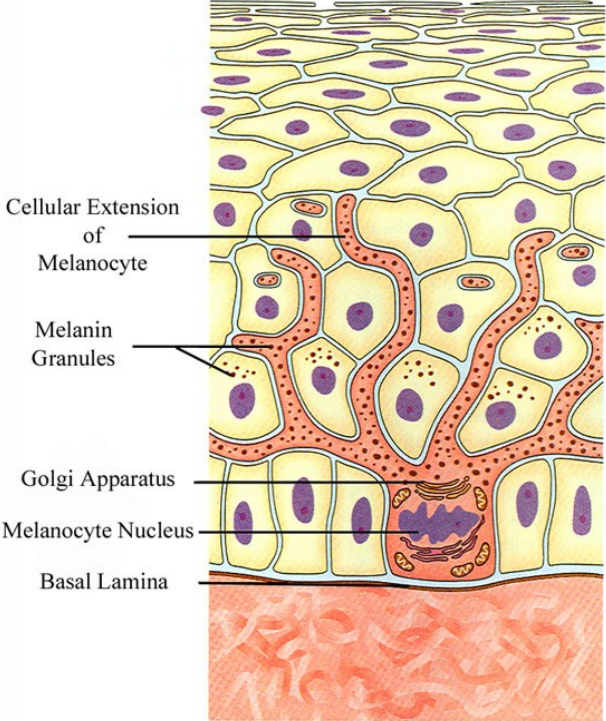
**Lipid droplets**



# Pigment cells



neuroectodermal origin



# Connective tissue proper - cells

## MOBILE CELLS

 Histiocytes  Macrophages

 Mast cells

 Plasma cells

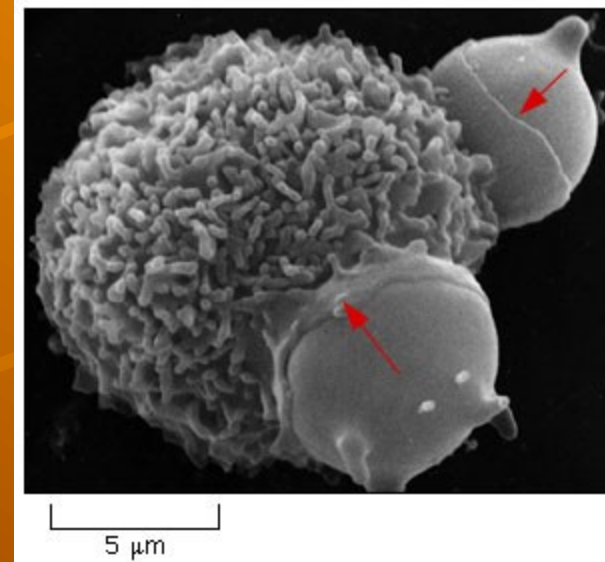
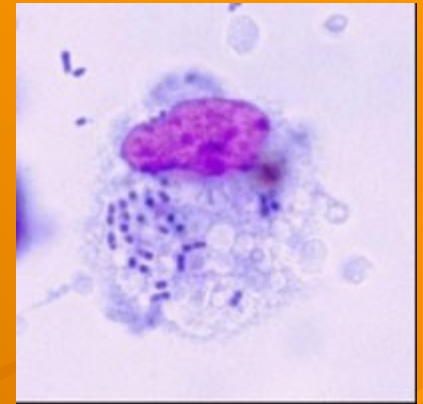
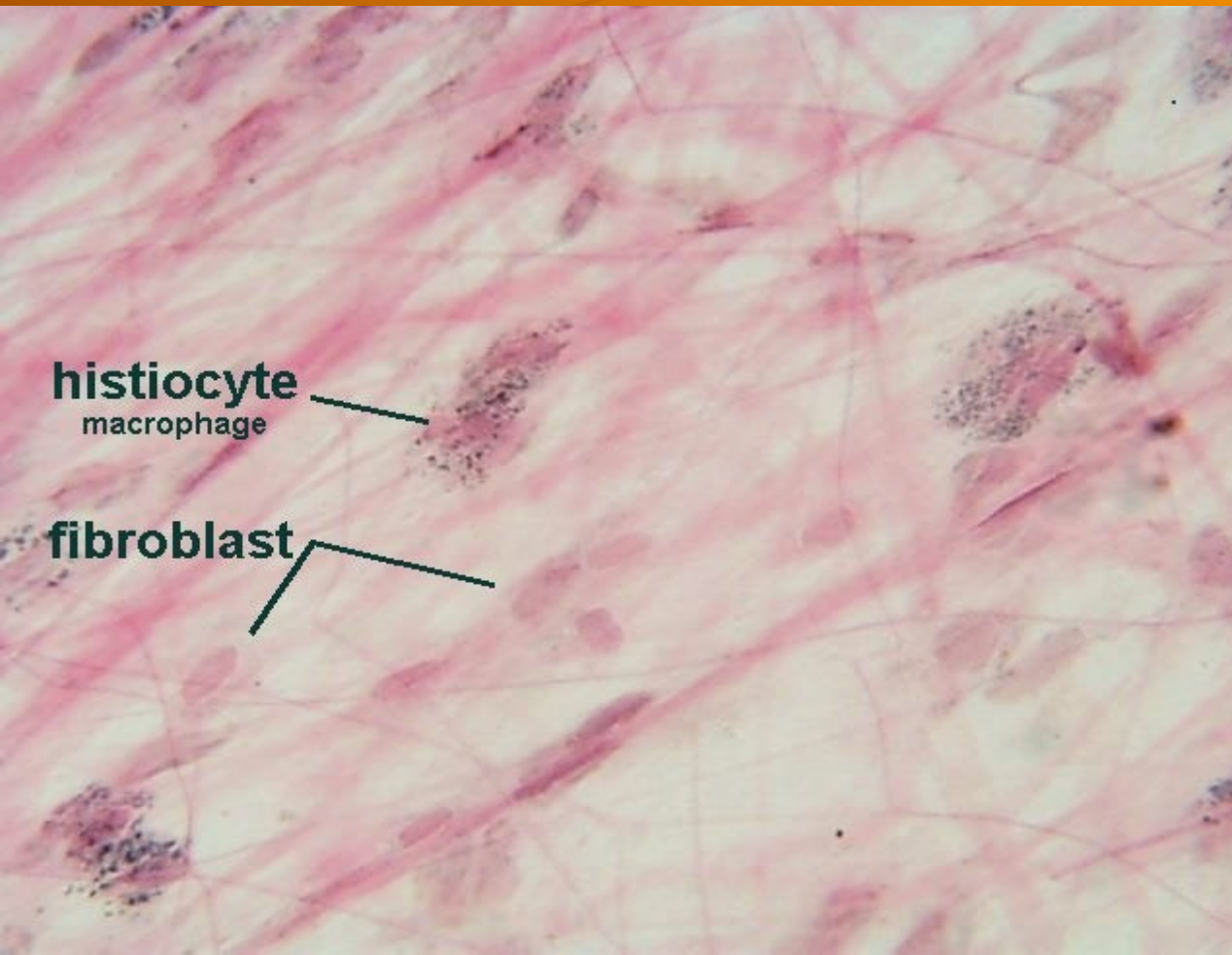


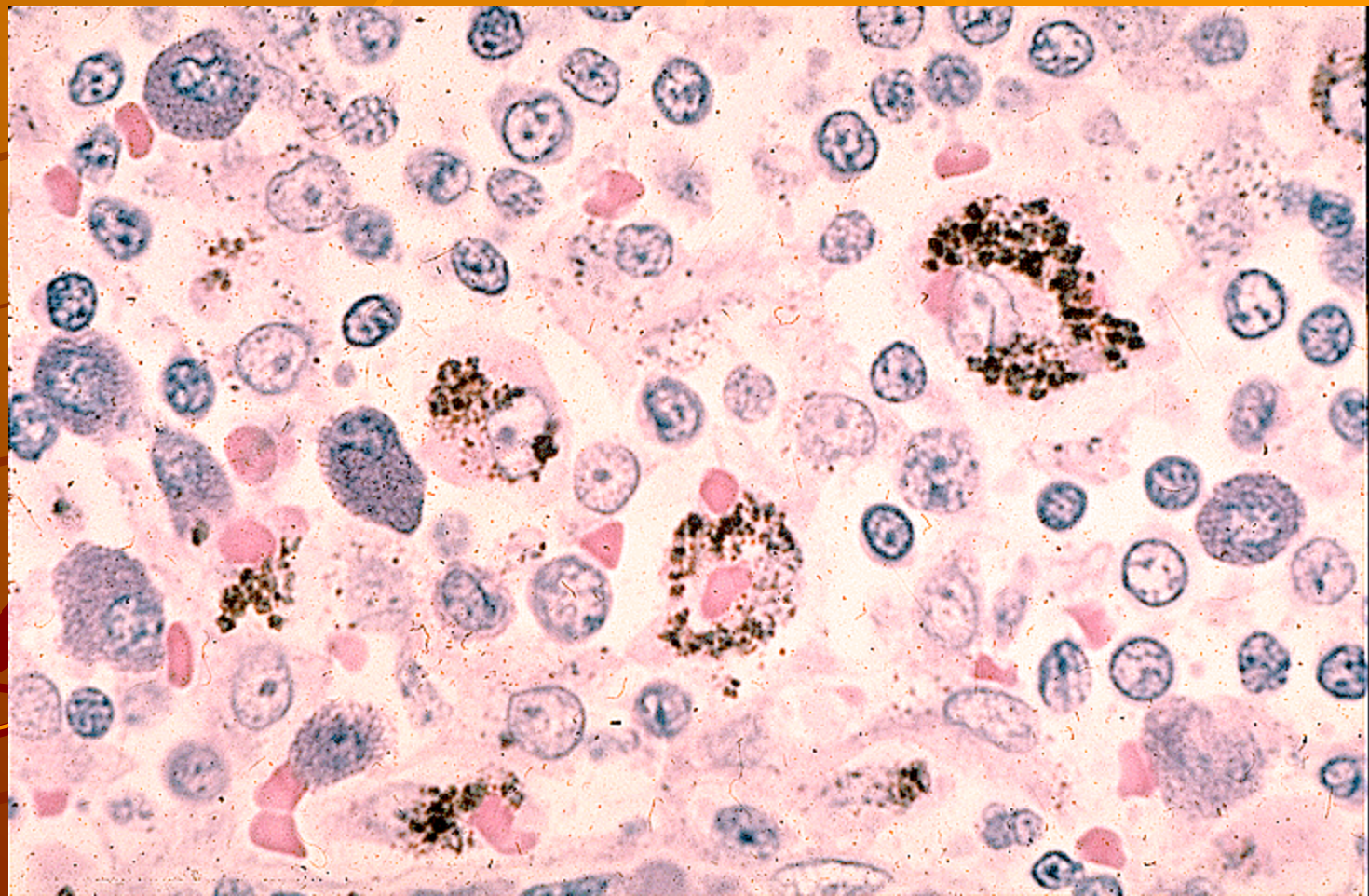
 Leukocytes



# Histiocytes $\Leftrightarrow$ macrophages (belong to monocyte-macrophage system)

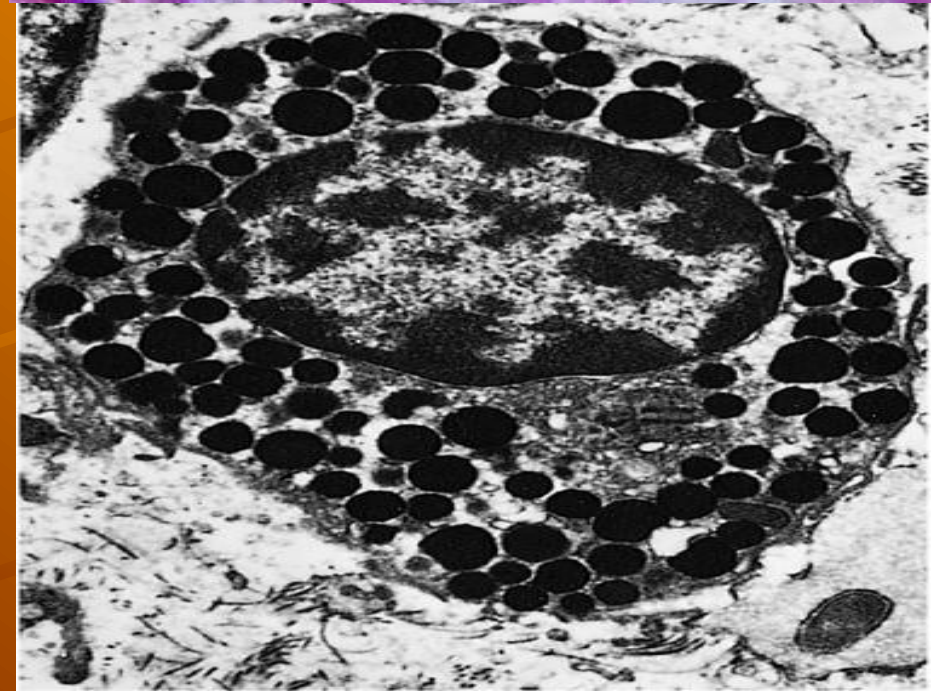
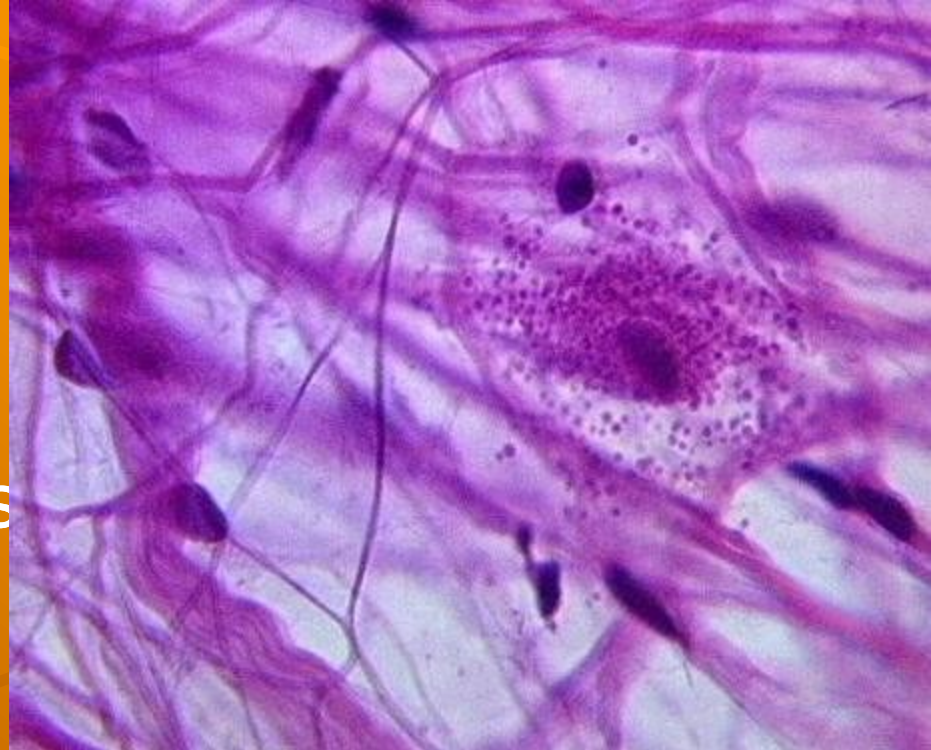
## phagocytosis





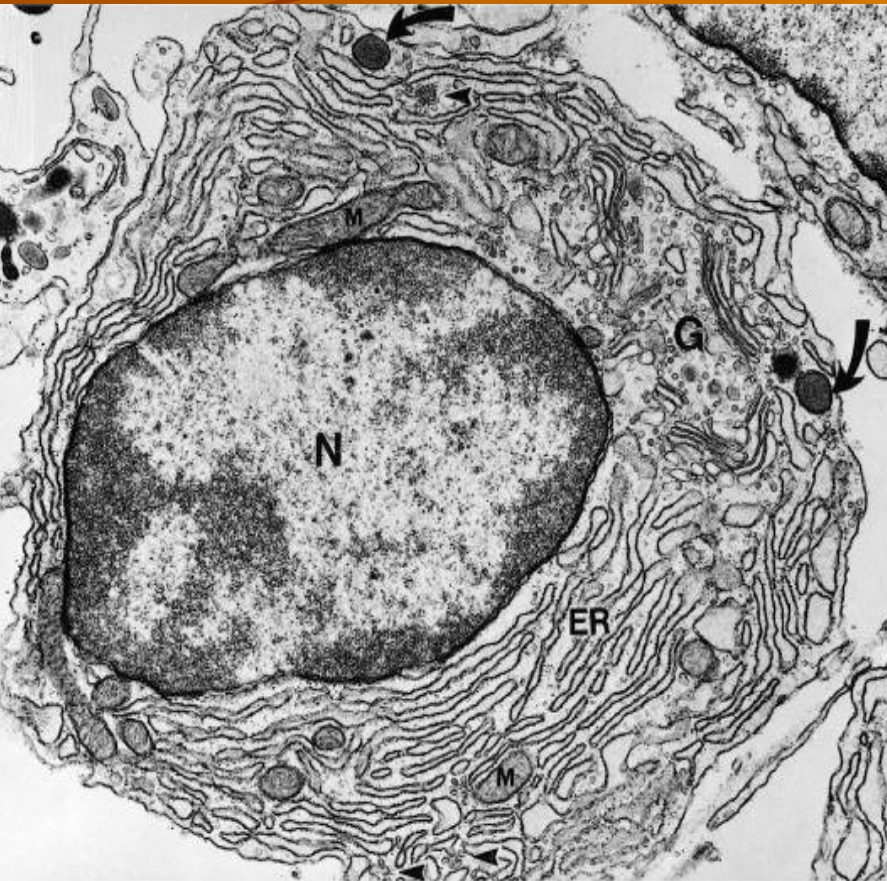
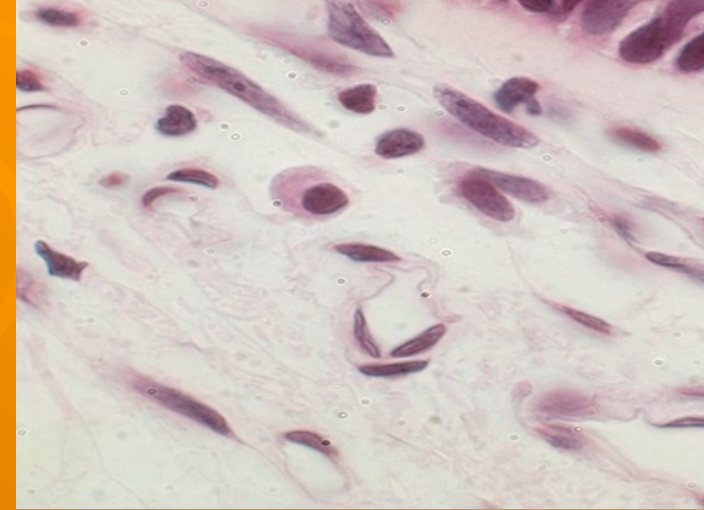
# Mast cells

heparin, histamin  
– inflammation mediators

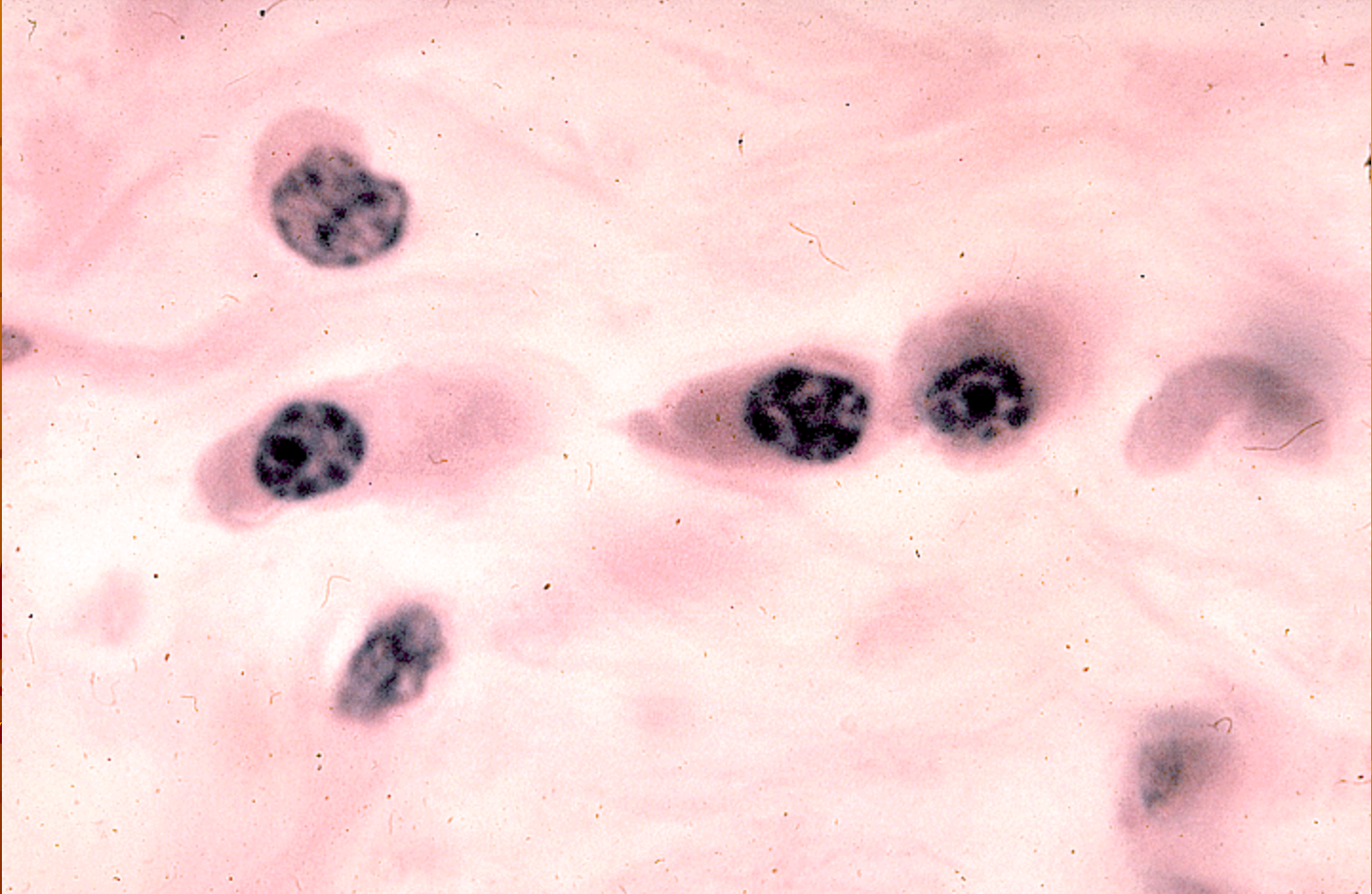


# Plasma cells (B-ly are precursors)

antibodies (immunoglobulins)  
production



**Rough ER**



# Intercellular (amorphous) ground substance

homogenous semifluid material

■ glycosaminoglycans (hyaluronic acid, chondroitinsulphate, dermatansulphate, keratansulphate, heparansulphate)

■ glycoproteins (fibronectin, laminin) – cell adhesive function

■ H<sub>2</sub>O, ions

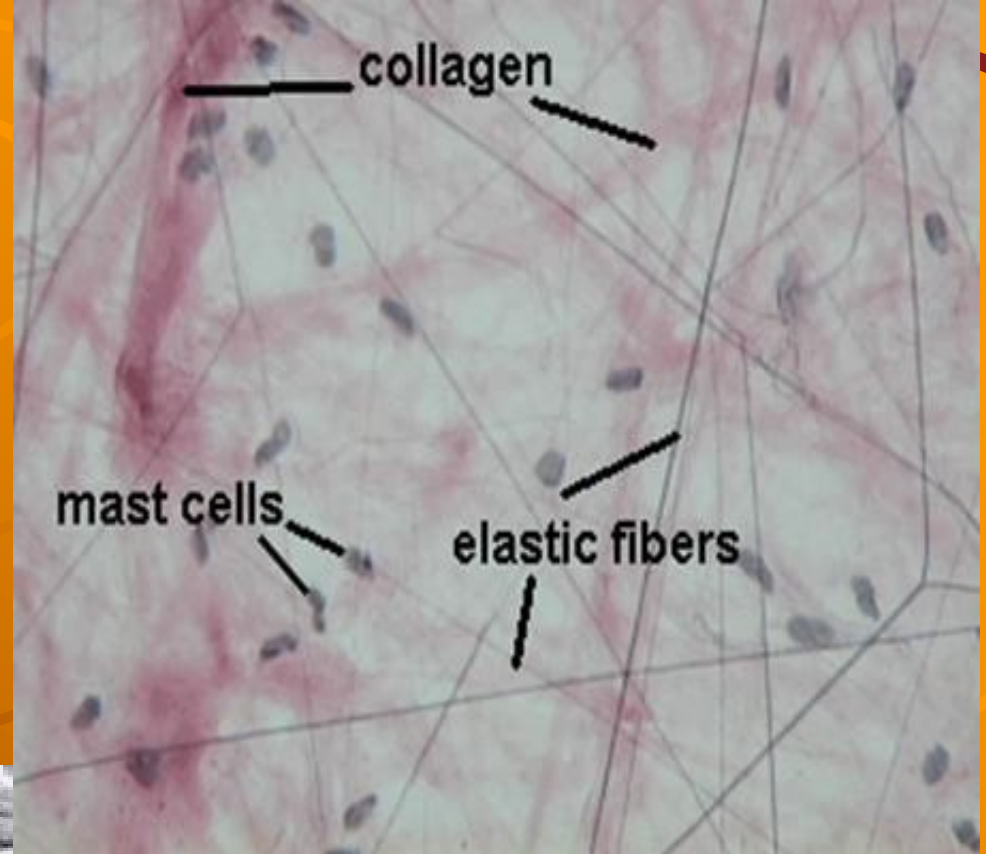


# Fibers

■ collagenous

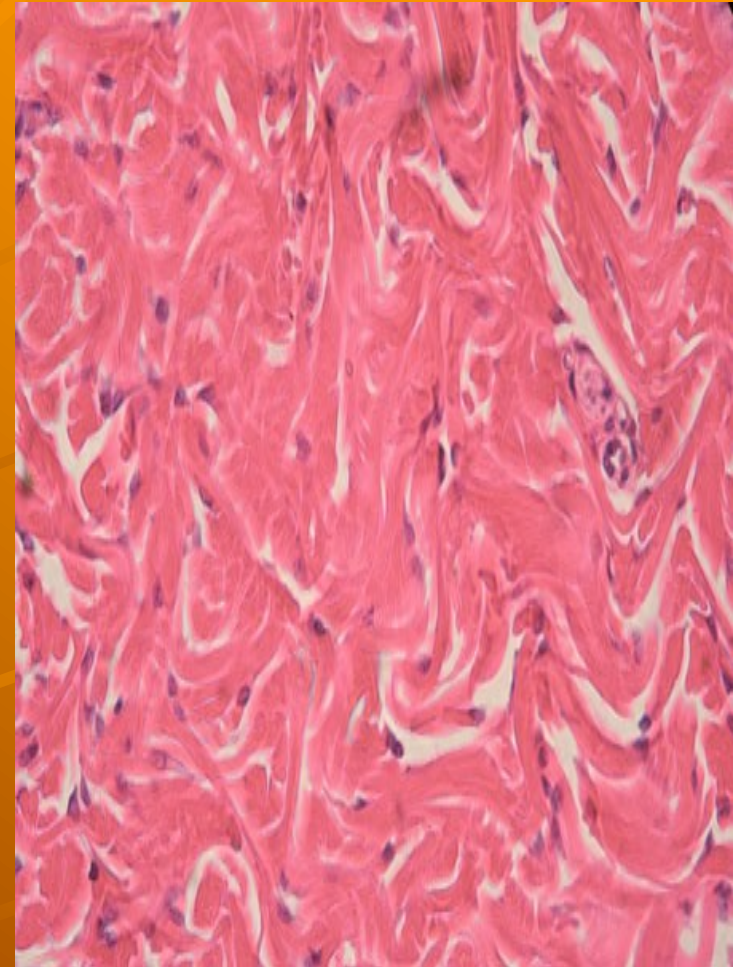
■ reticular

■ elastic



# Collagen fibers

- „white fibers“, solid and strong, but not elastic
- 1 – 20  $\mu$ m  $\varnothing$
- arranged into bundles
- collagenase – digestive enzyme
- in LM – acidophilic (pink in HE)



# Collagen fibers

amino acids  
-1 nm



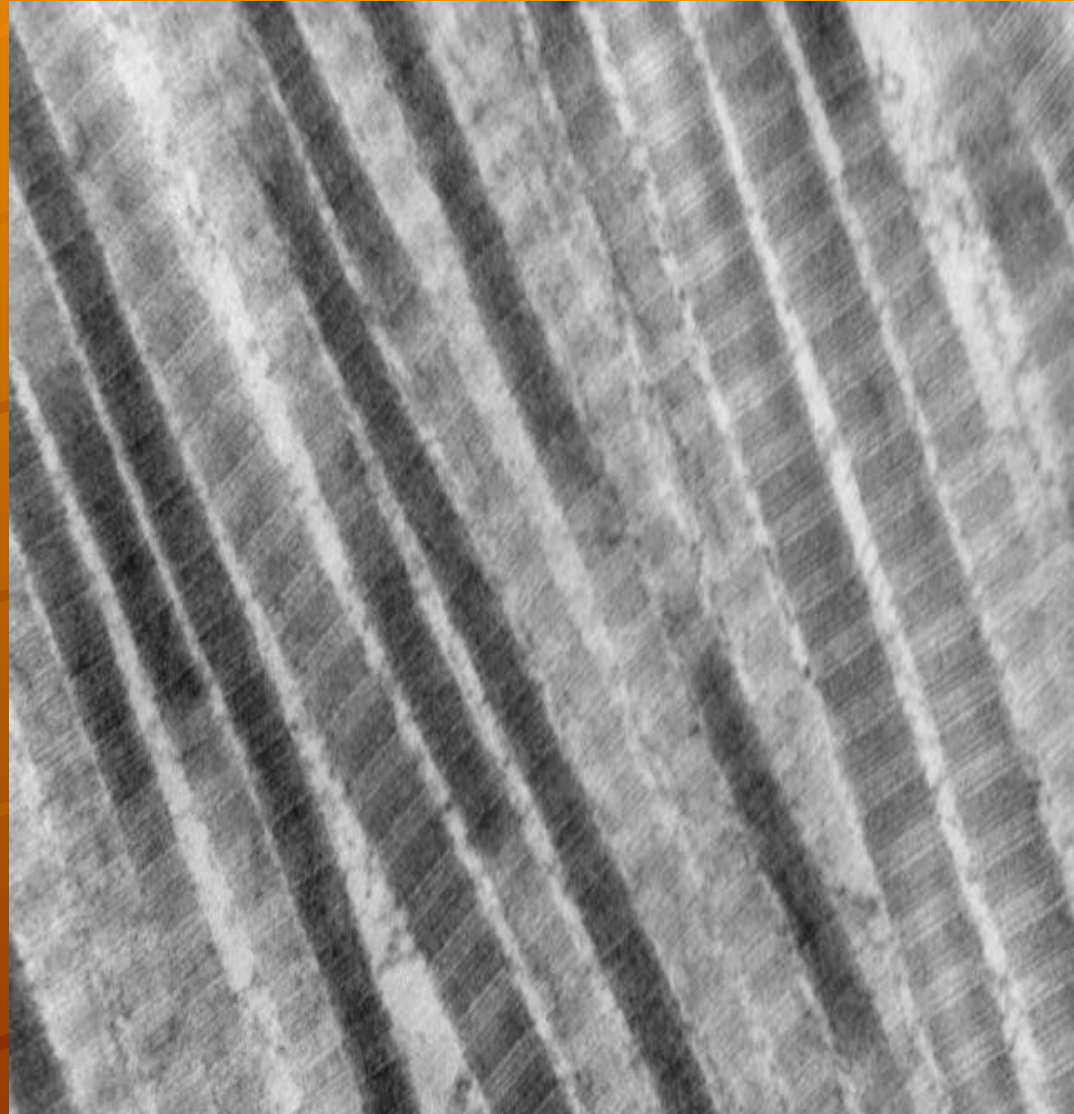
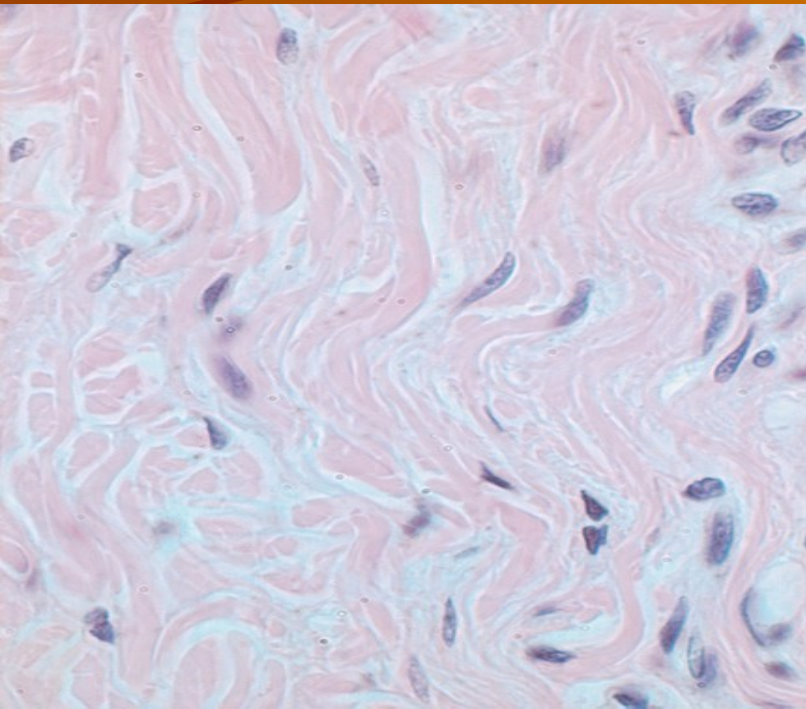
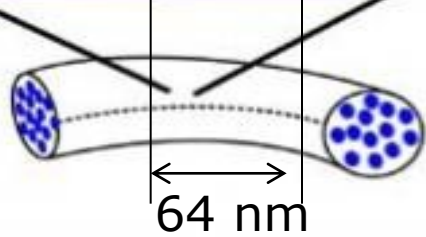
tropocollagen  
-300 nm



fibrils  
-1  $\mu\text{m}$



fibers  
-10  $\mu\text{m}$



# Collagen fiber types

## Collagen Type I

- most ubiquitous (cf.)

## Collagen Type II

- in cartilage

## Collagen Type III

- reticular fibers

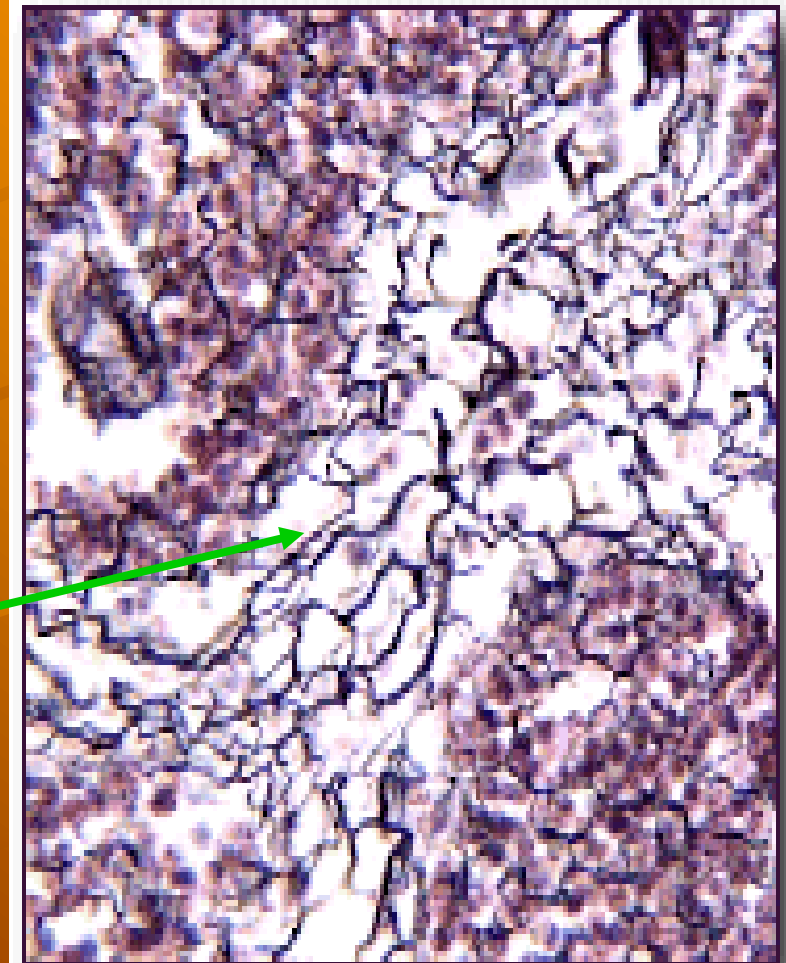
## Collagen Type IV

- "amorphous collagen" without a fiber structure, synthesized by epithelial cel

About 40 types of collagen are known

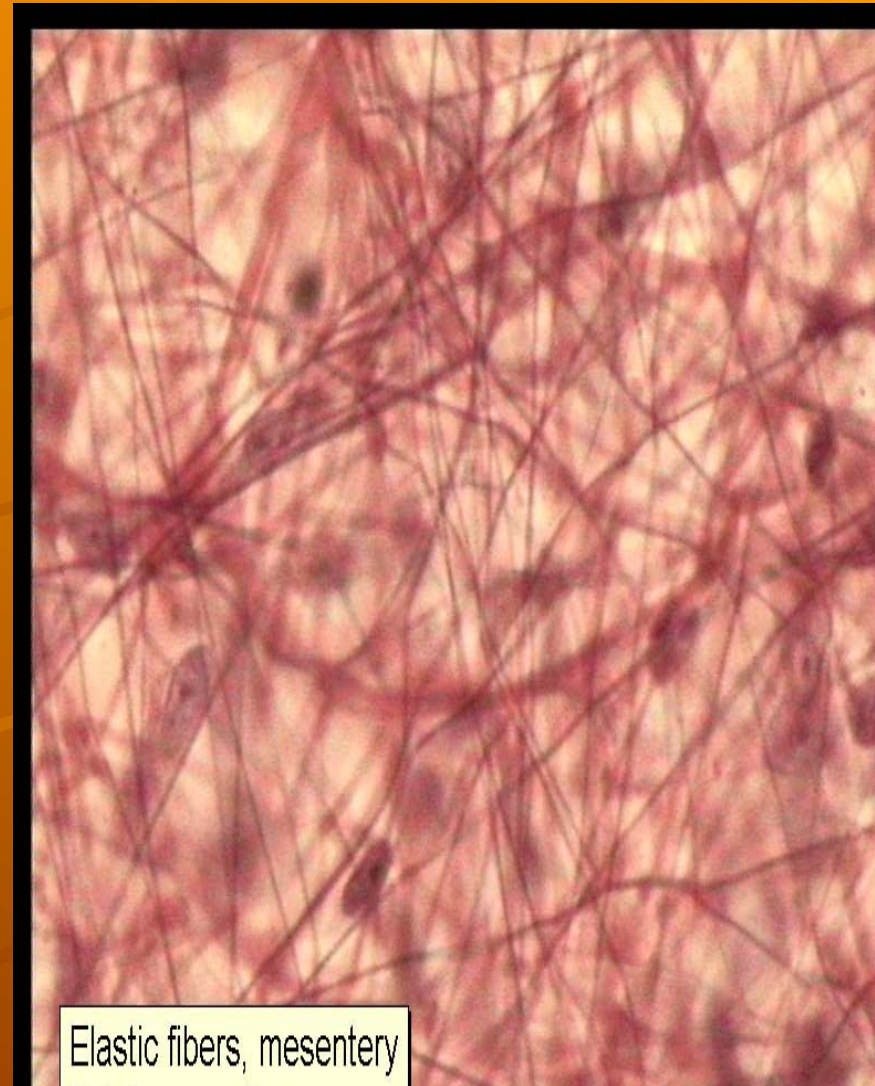
# Reticular fibers

- contain type III collagen
- form a fine meshwork (reticulum) - a supporting mesh in soft tissues such as liver, bone marrow, and lymph organs
- they are argyrophilic (silver staining)
- 0.5 – 2  $\mu$ m  $\varnothing$



# Elastic fibers

- „**yellow fibers**“ - bundles of protein (elastin) produced by fibroblasts and smooth muscle cells in blood vessels.
- these fibers can stretch up to 1.5 times their length, and snap back to their original length when relaxed.
- are branched and wavy
- special staining with orcein or resorcin-fuchsin



Elastic fibers, mesentery

# C.t.proper classification

- several types of c.t.proper:  
classification depends on cells, fibers and ground substance quantity and arrangement
- abundant ground subst. – „soft tissue“
- abundant fibers – „hard tissue“
- fibers can be arranged (ir)regularly

# CTP classification (types of c.t.)

■ Mesenchyme

■ Jelly-like c.t. (Wharton's jelly)

■ Collagenous c.t. → areolar (loose)  
→ dense → irregular  
→ regular

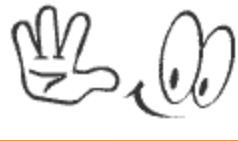
■ Reticular c.t.

■ Elastic c.t.

■ Adipose c.t. → white fat  
→ brown fat

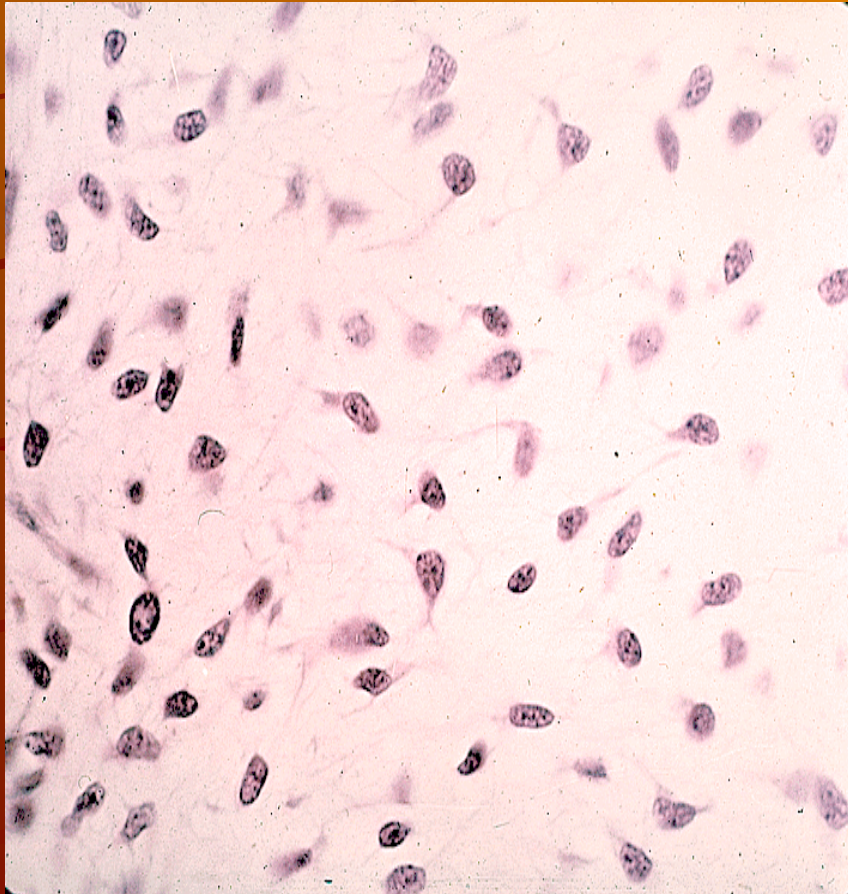






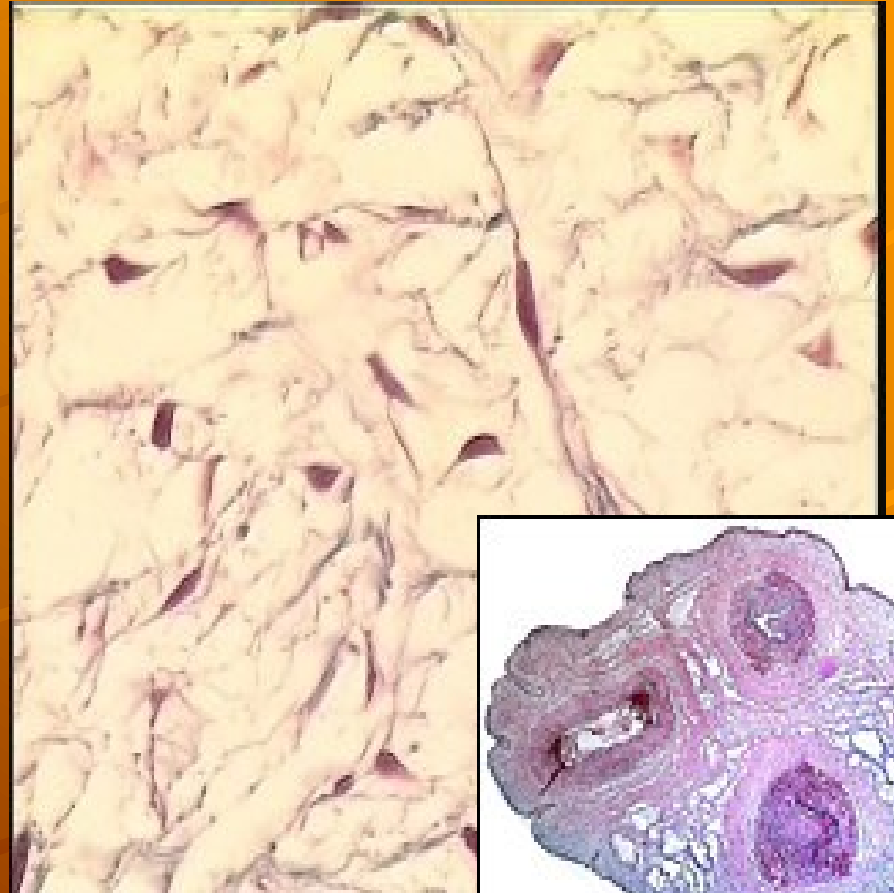
# 1. Mesenchyme

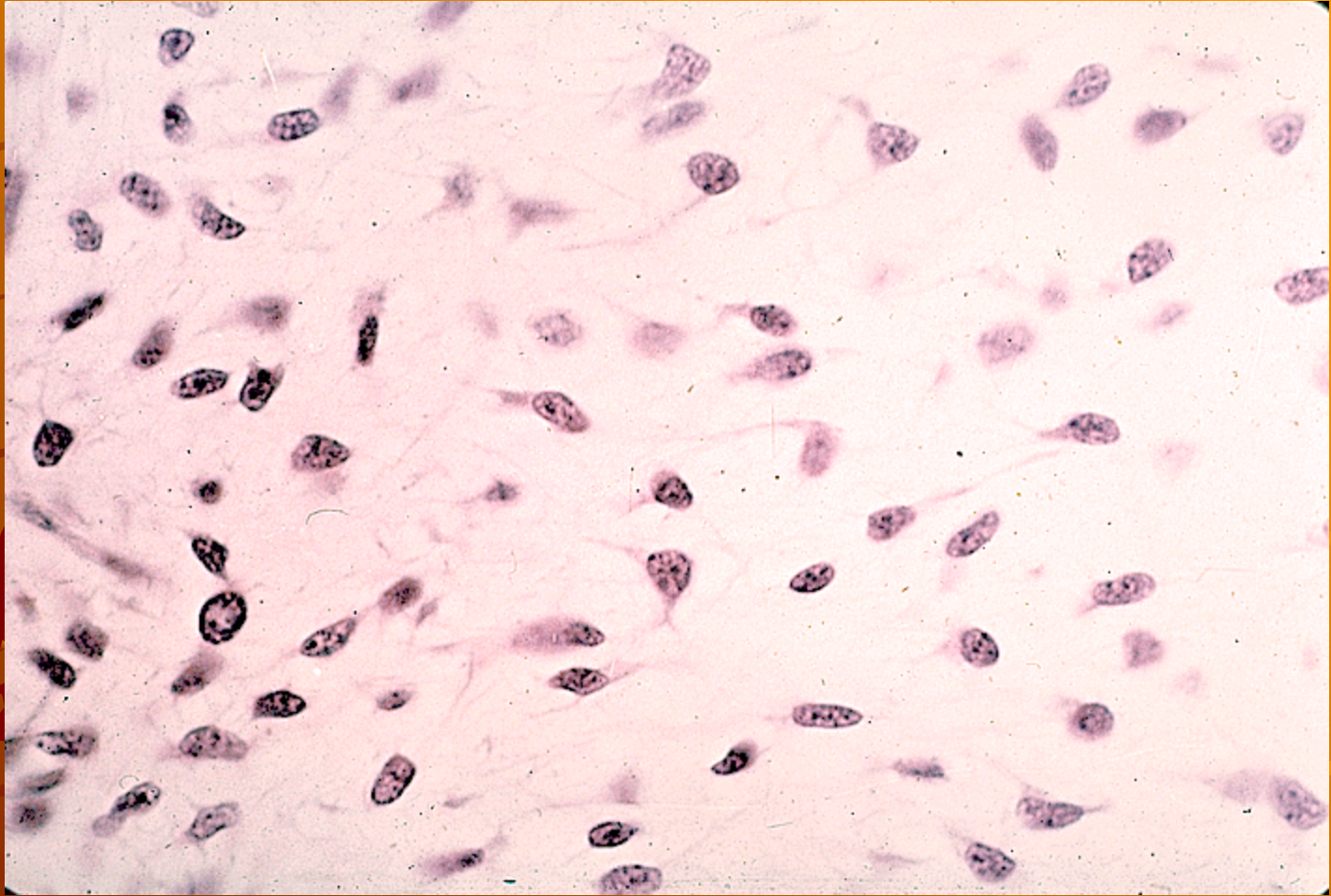
embryonic c.t.



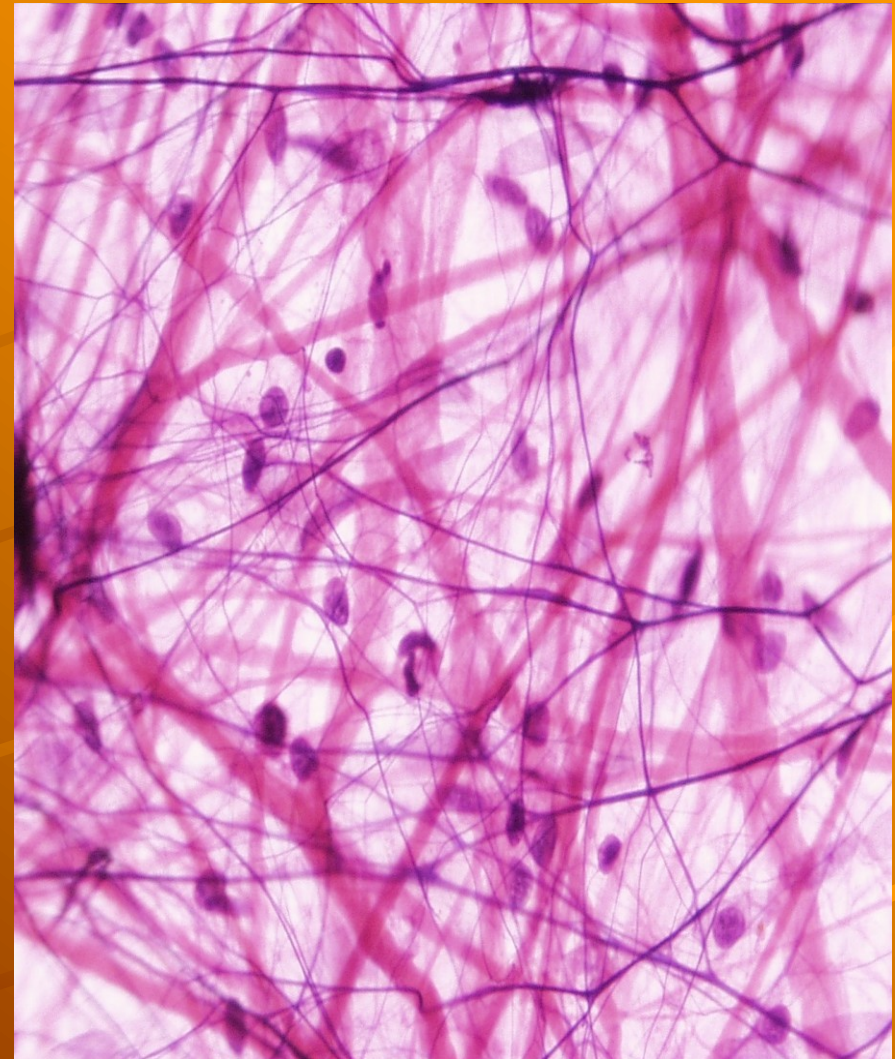
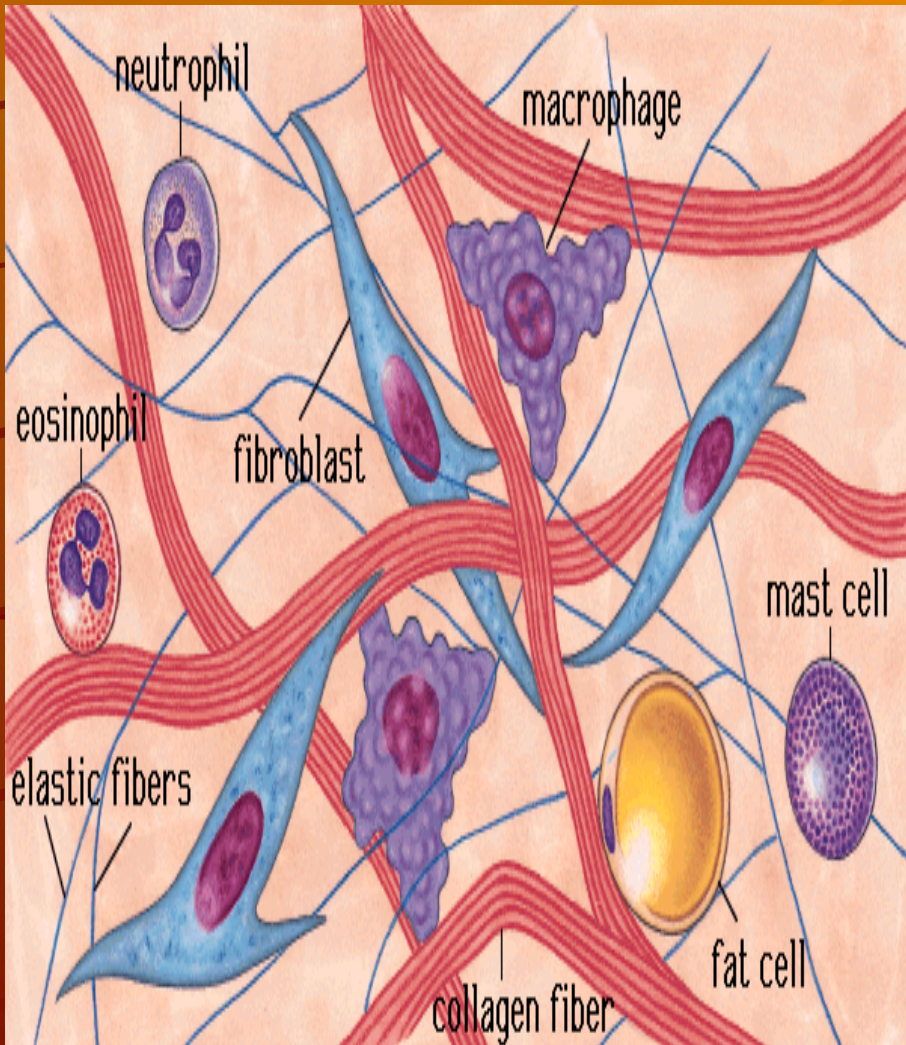
# 2. jelly-like c.t.

umbilical cord, dental pulp

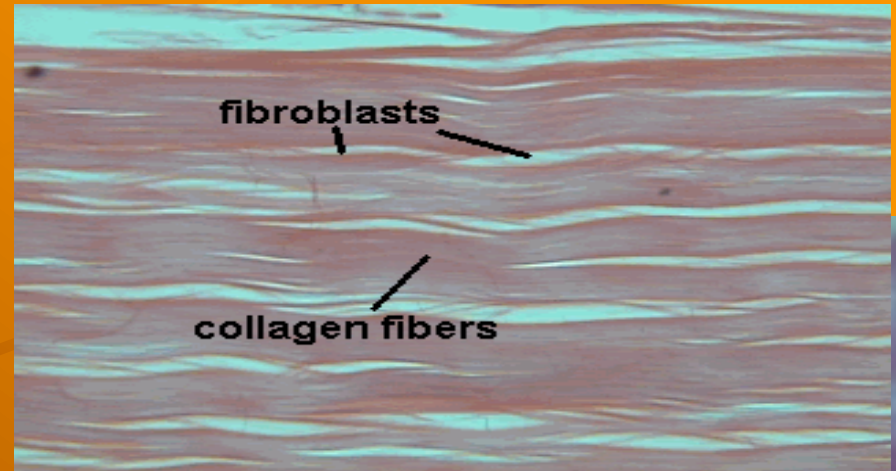
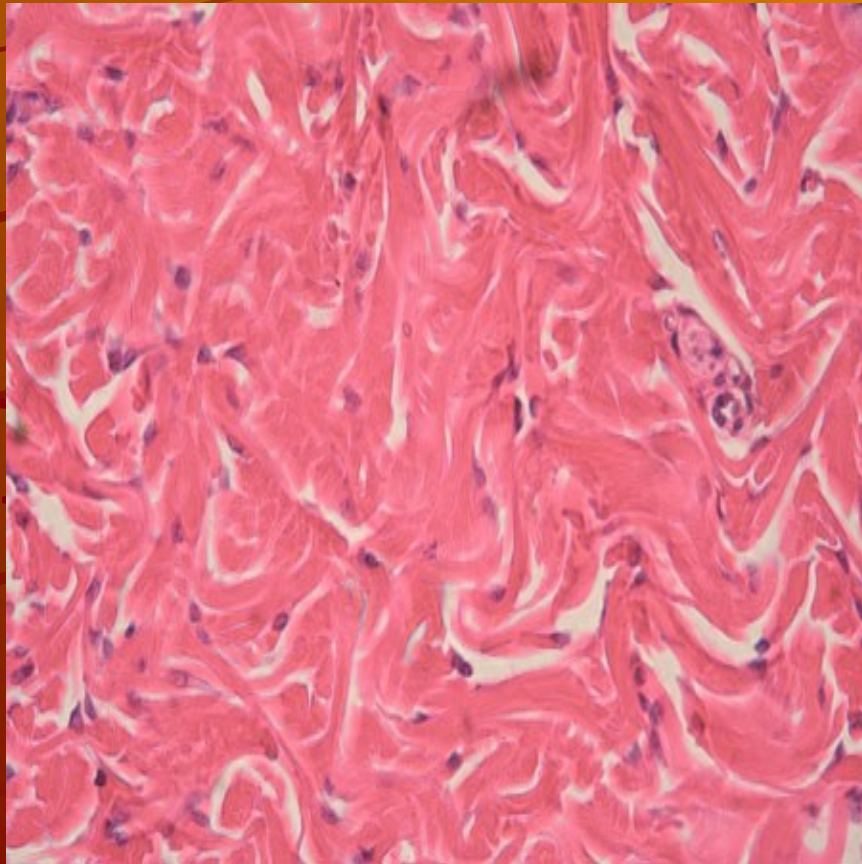


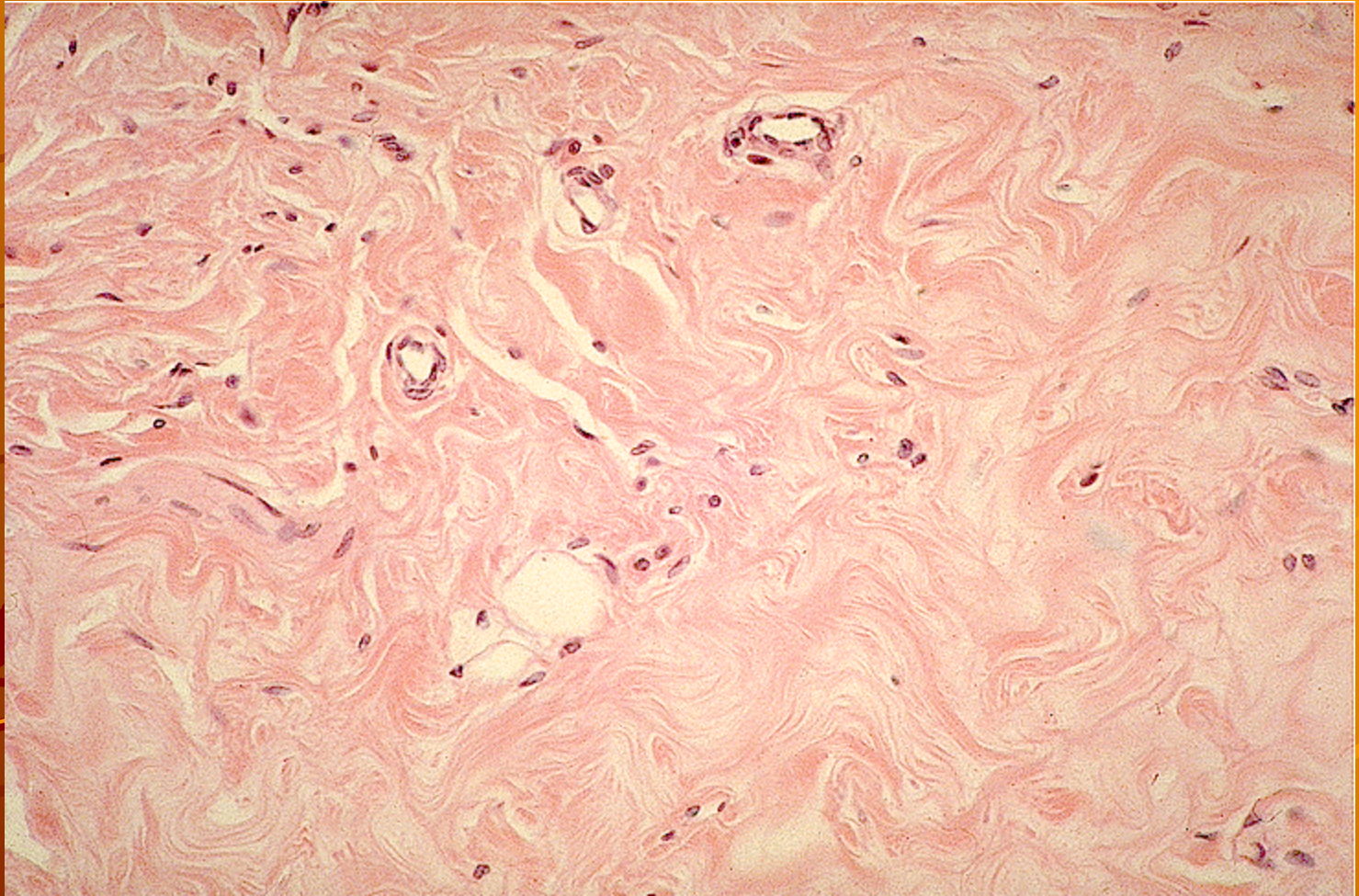


# 3a. collagenous loose (areolar) c.t.



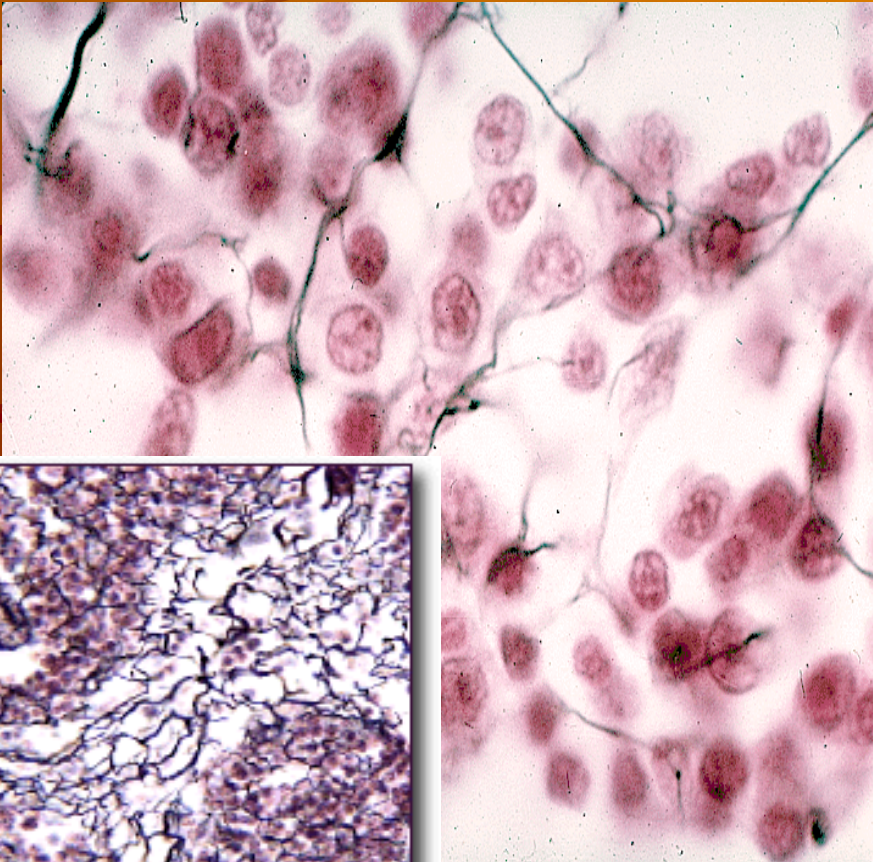
# 3b. collagenous dense c.t. → irregular → regular





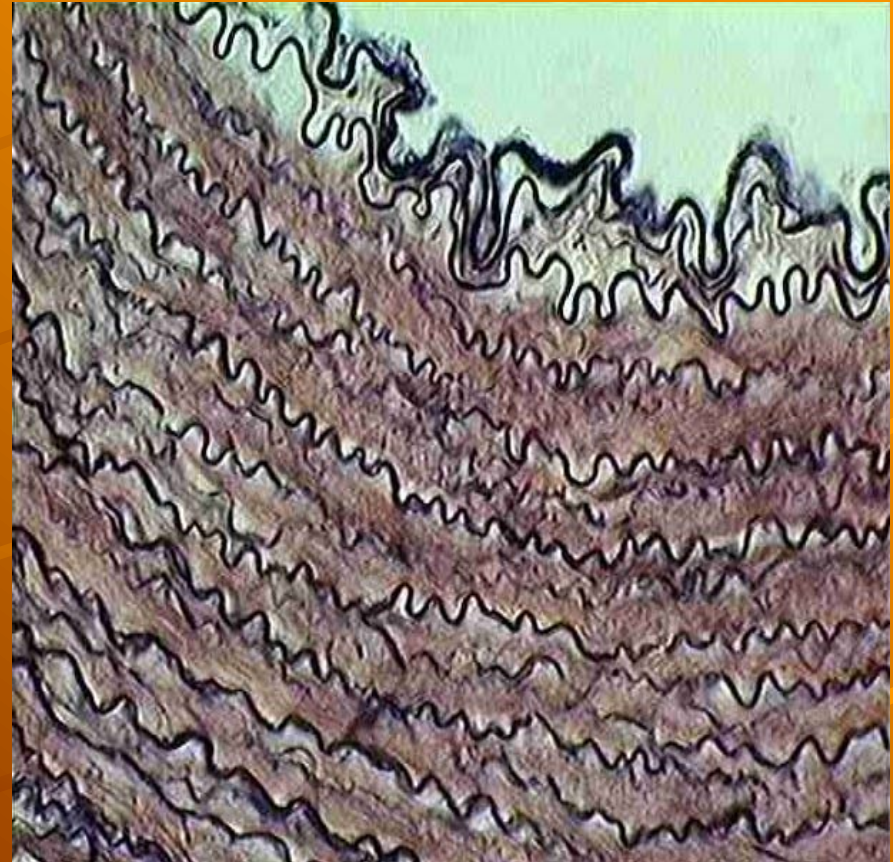
## 4. reticular c.t

supporting tissue in  
lymph organs (*impregn.*)



## 5. elastic.c.t.

membranes in blood  
vessels wall(*orcein*)

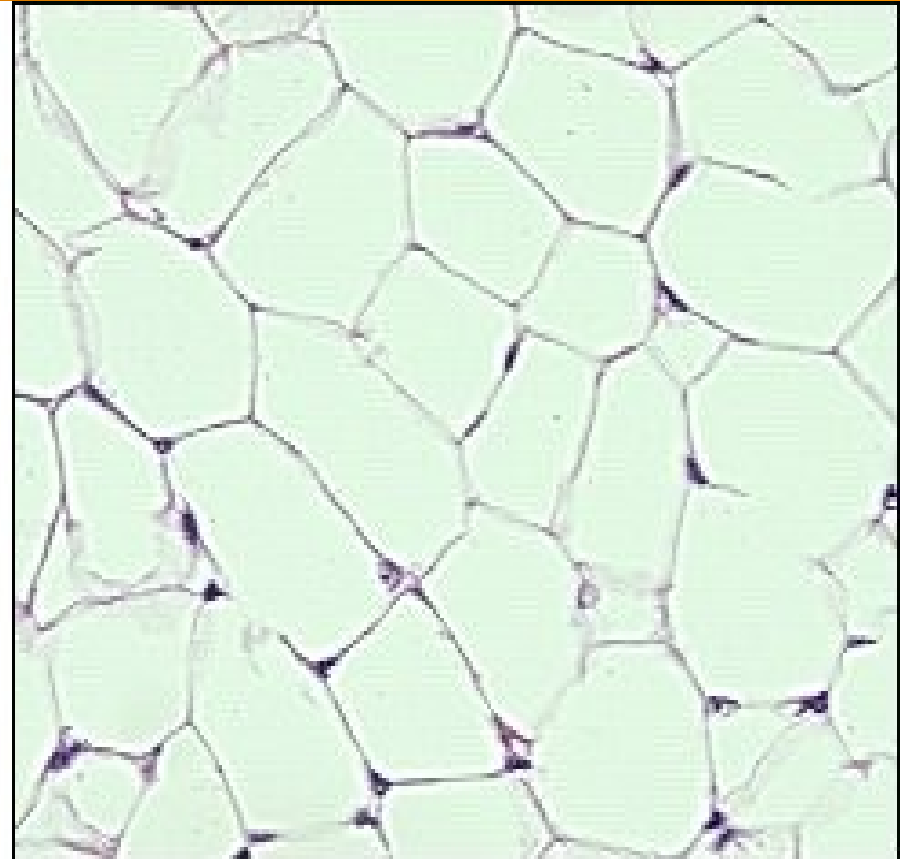
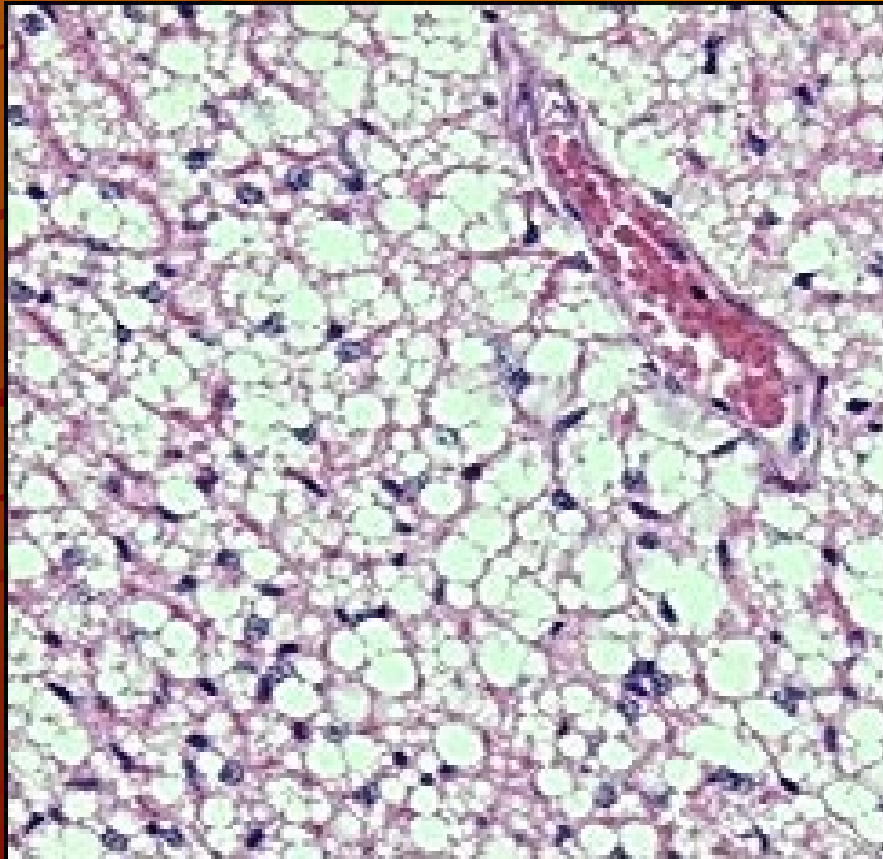


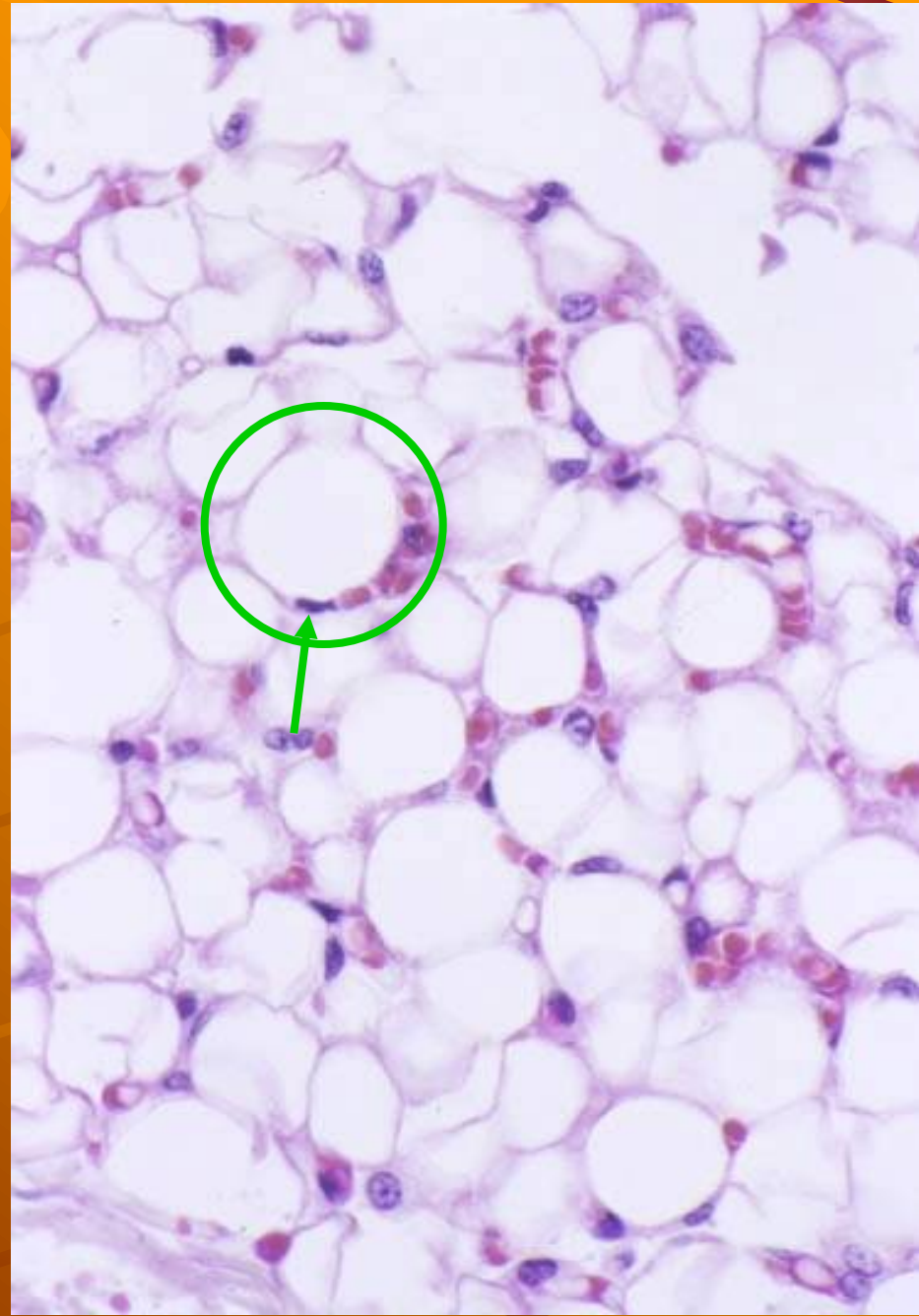
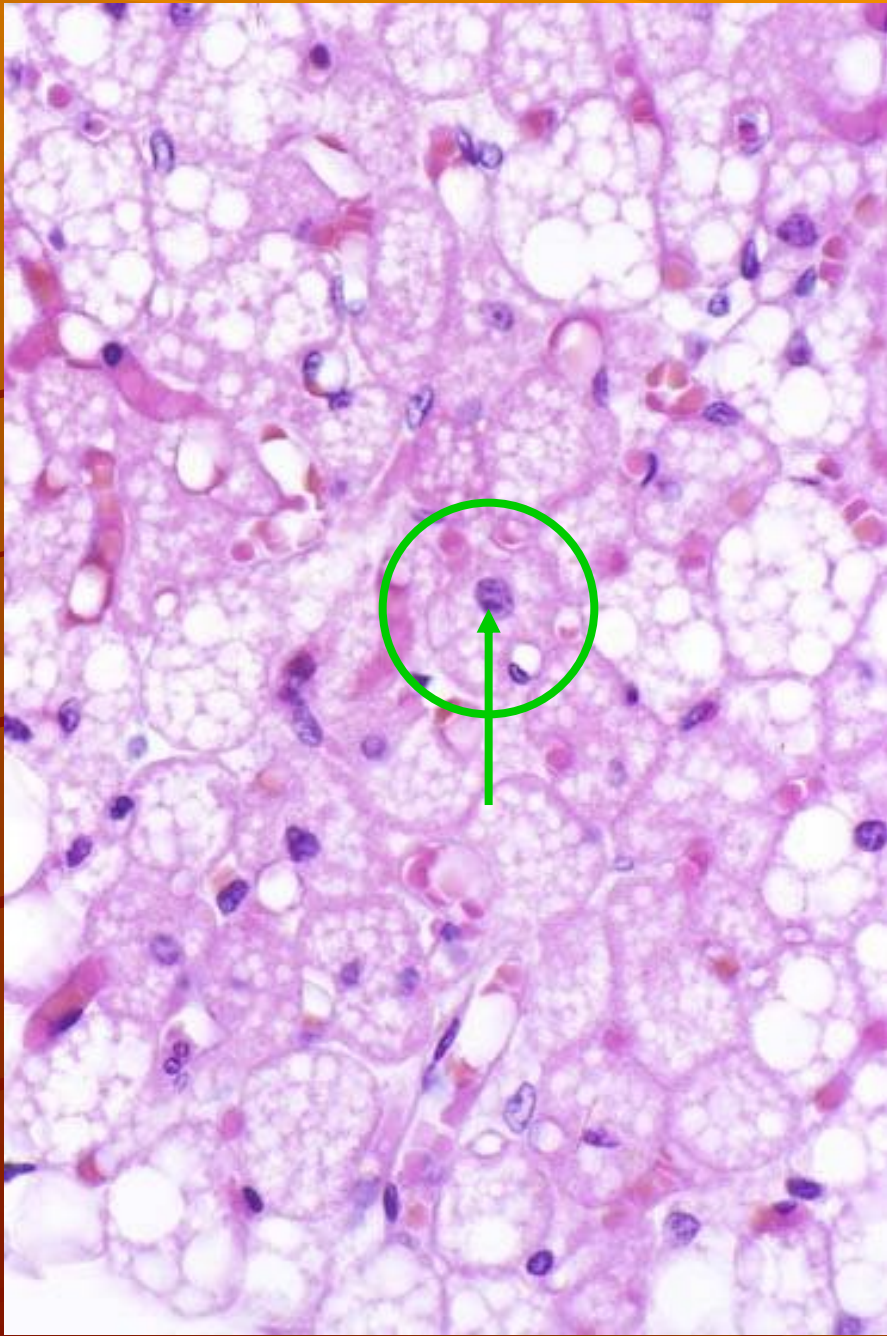
# 6. adipose tissue

brown fat

white fat

fetal adipose tissue  
(thermoregulation)

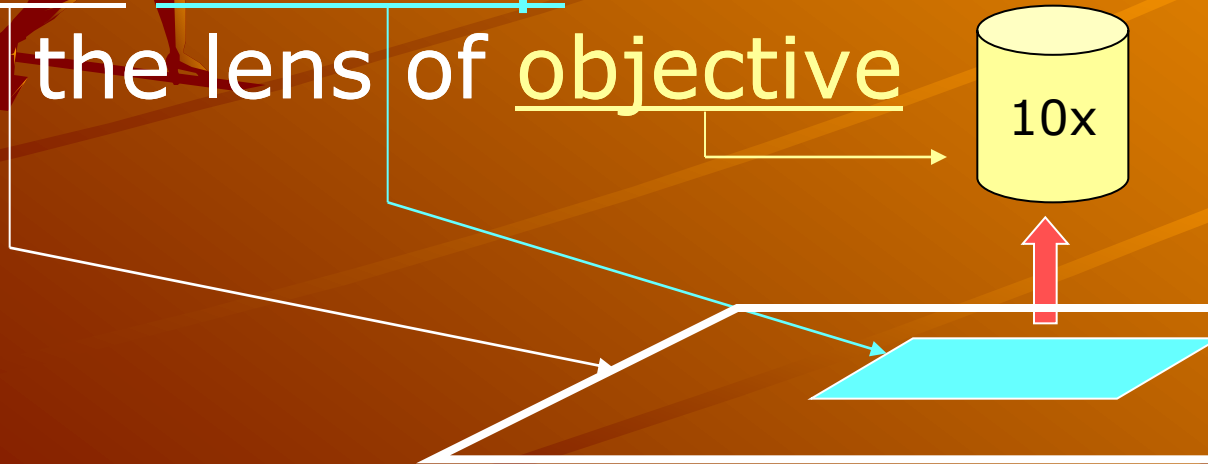






# Light microscopy

- Occular magnif.: 10x
- Objectives magnif. 4x, 10x, 40x
- Total magnif.: 40x, 100x, 400x
- **Don't use immersion objective!**
- Slide cover slip must be situated up to the lens of objective



# Supporting connective tissue

■ Cartilage

■ Bone



# Cartilage

- general characteristic -

- Avascular tissue (without blood vessels); nutrients are transported to the cells by diffusion from perichondrium
- Perichondrium\* – dense connective tissue capsule with blood vessels
- Mesenchymal origin

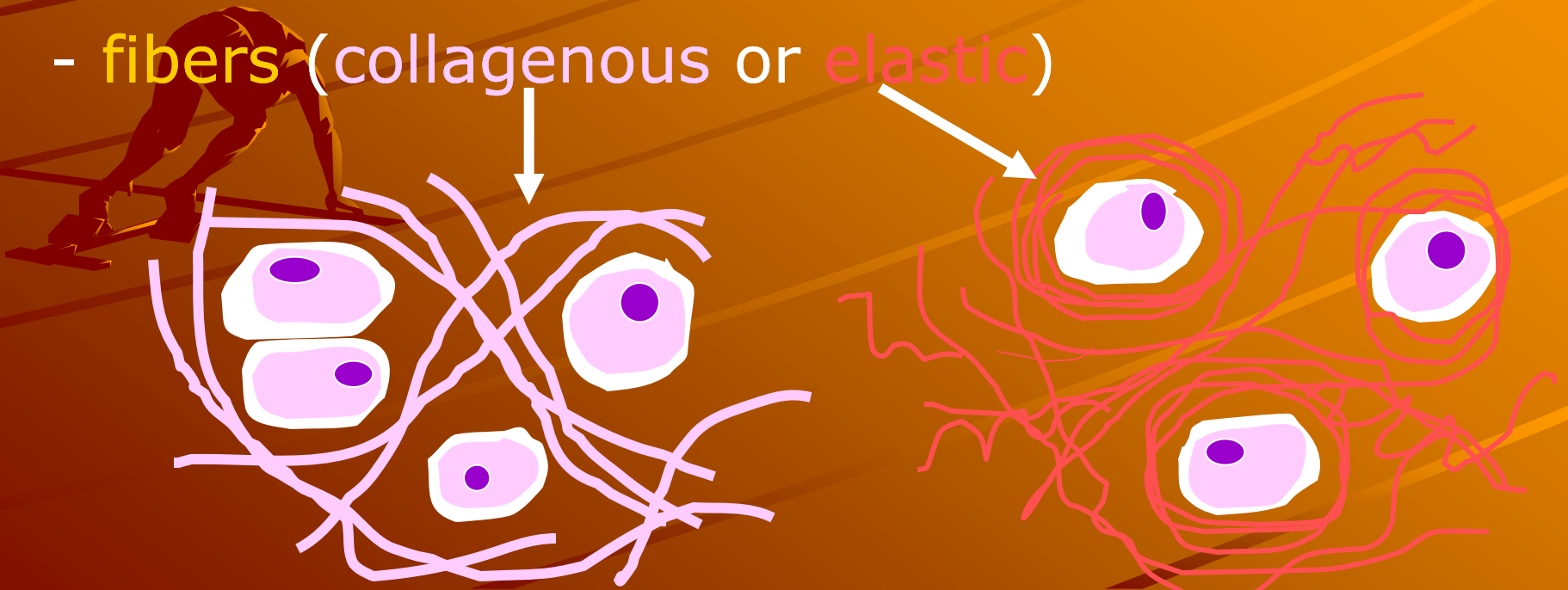
\* *Is not present on the articular surfaces of cartilage and on the surface of fibrocartilage*

# General structure of the cartilage

➤ Cells – chondrocytes in lacunae

➤ Intercellular matter:

- **ground substance**
  - chondroitinsulphates
  - chondronectin
- **fibers** (collagenous or **elastic**)



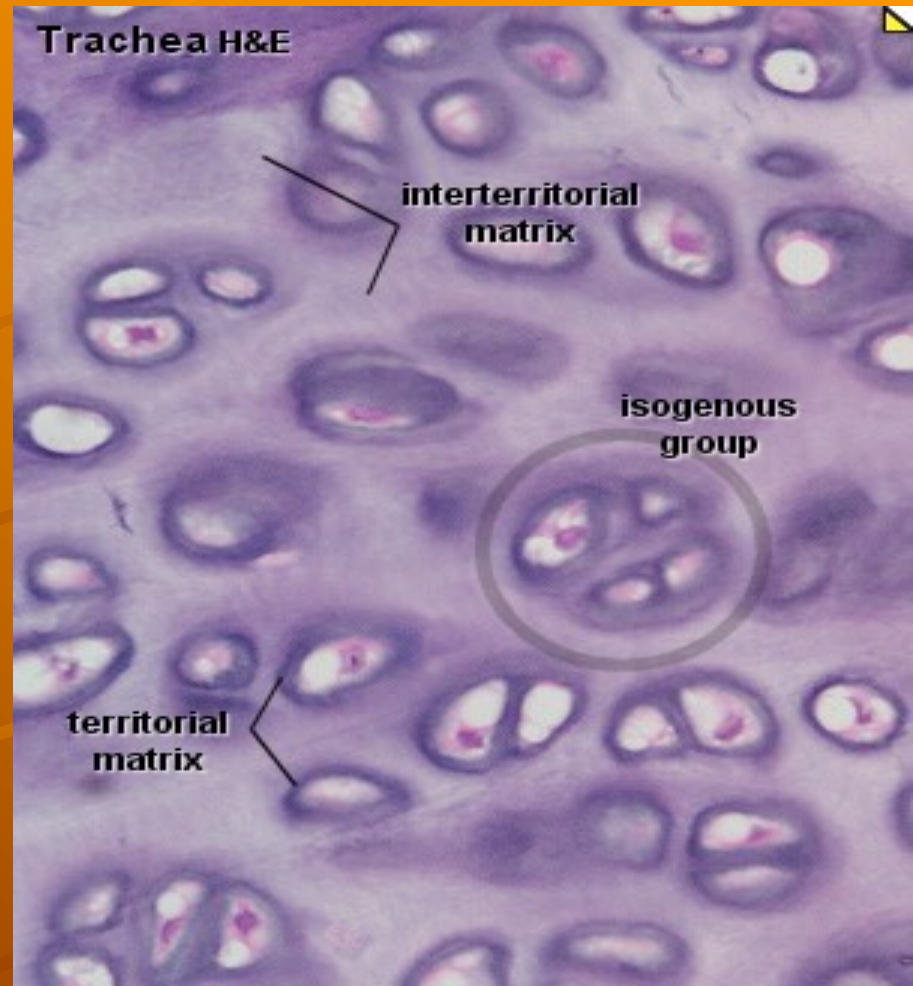
# Cartilage cells: chondroblasts, chondrocytes

- in lacunae
- produce intercellular matter:
  - collagen or elastic fibers
  - ground substance: 1) main glycosaminoglycans are **chondroitinsulfates**, keratansulfate = acid substances cause basophilia of cartilage, 2) glycoprotein **chondronectin** (increases adhesion of cells and matrix)
- cells are involved in cartilage growth:
  - **interstitial growth (*isogenic groups*)**
  - **appositional growth (from perichondrium)**

# Hyaline cartilage

(example: free surfaces of joints, epiphyseal growth plate)

- Chondrocytes form isogenous groups (in basophilic capsule)
- Collagenous fibers form bundles
- Ground substance is abundant and covers the fibers

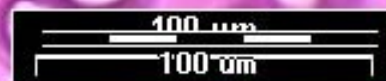


Hyaline cartilage, trachea

Perichondrium

Chondroblasts

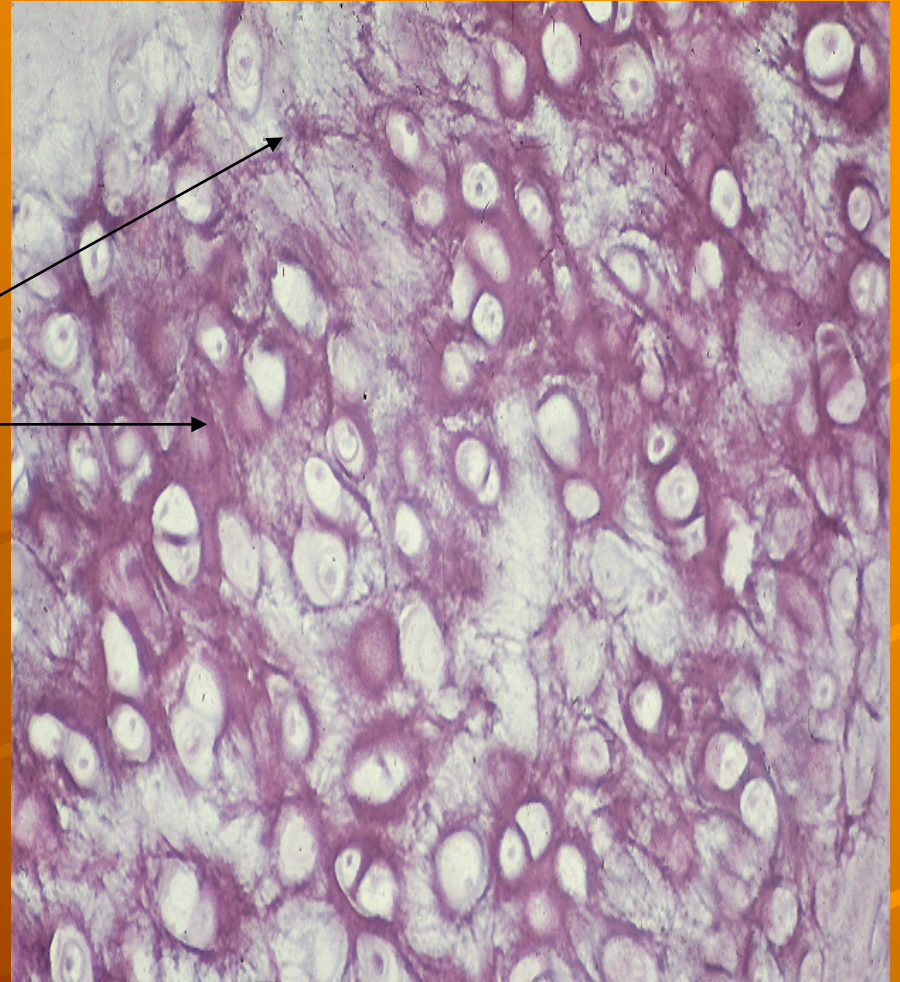
Isogenous (nest) cells



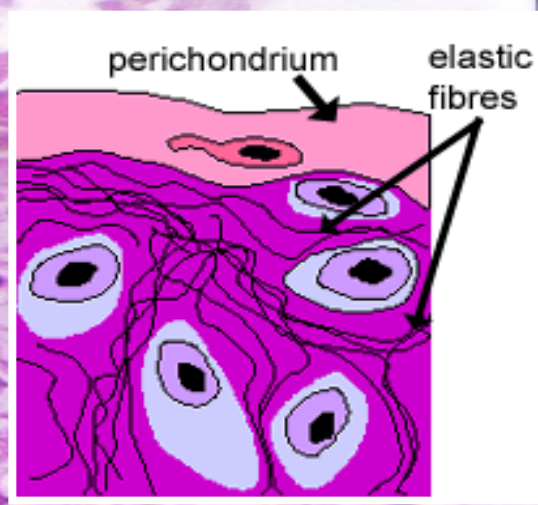
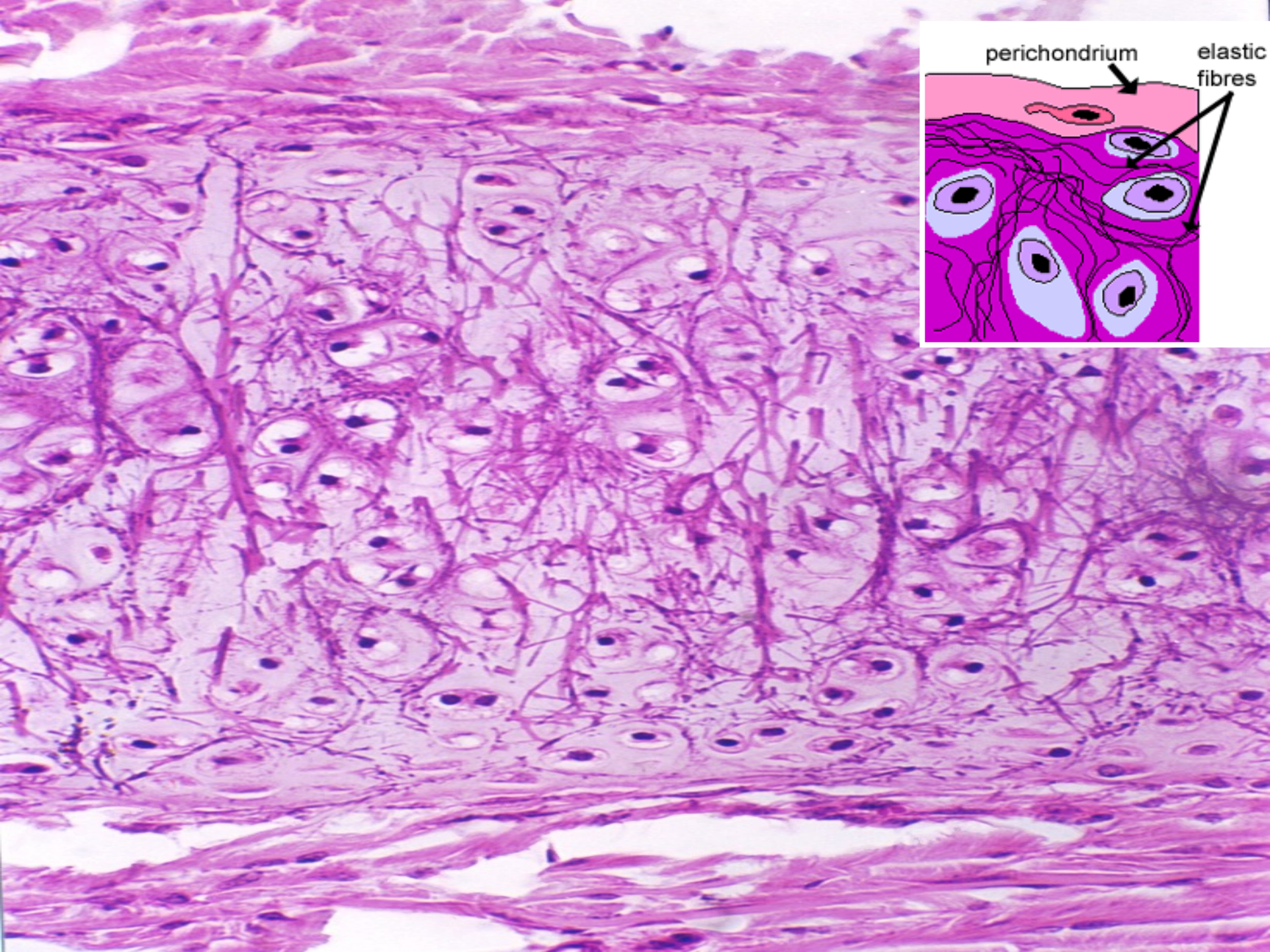
# Elastic cartilage

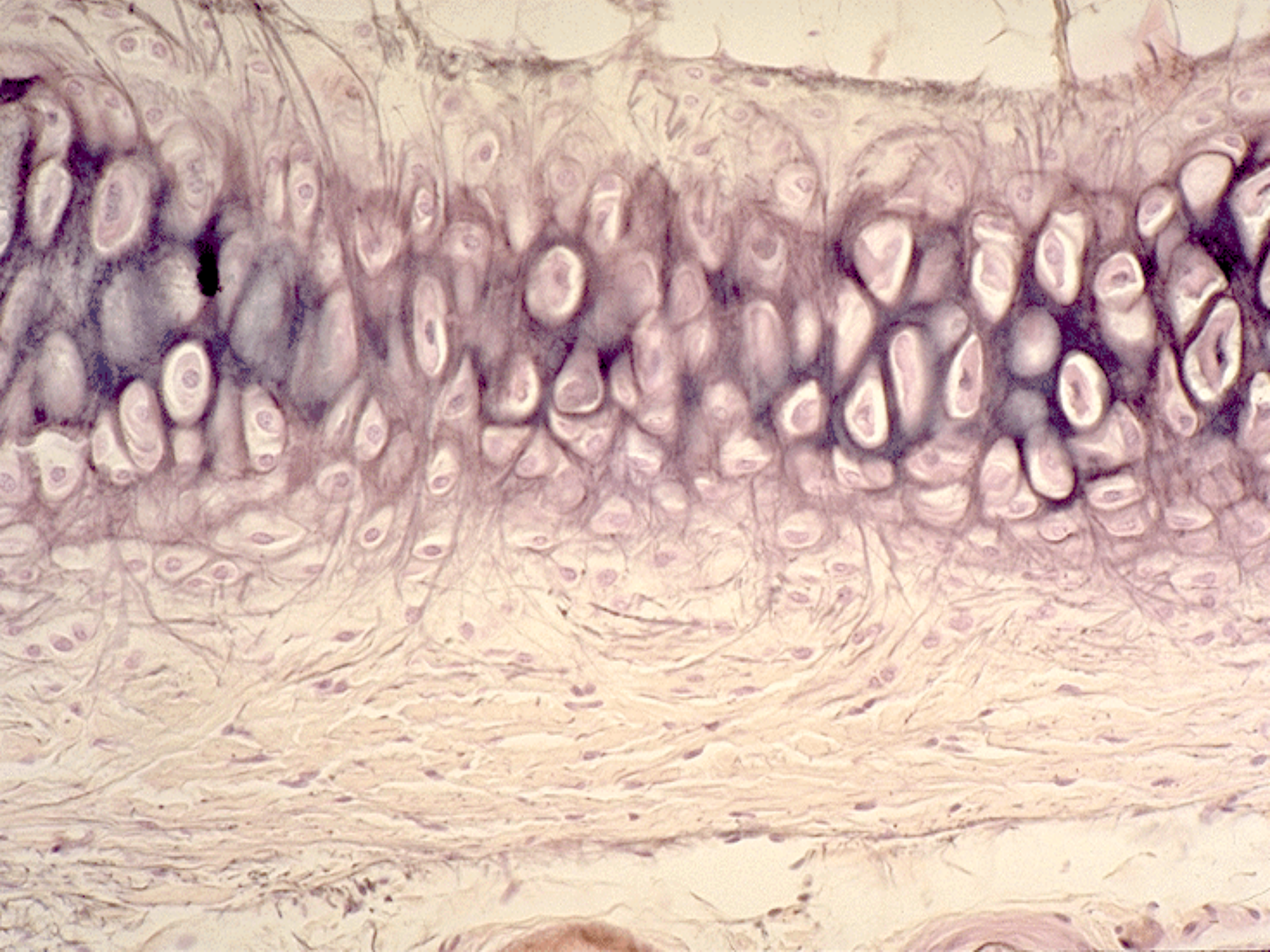
(example: auricle, epiglottis)

- Chondrocytes in lacunae are dispersed:  
NO isogenic groups
- Elastic fibers
- Ground substance







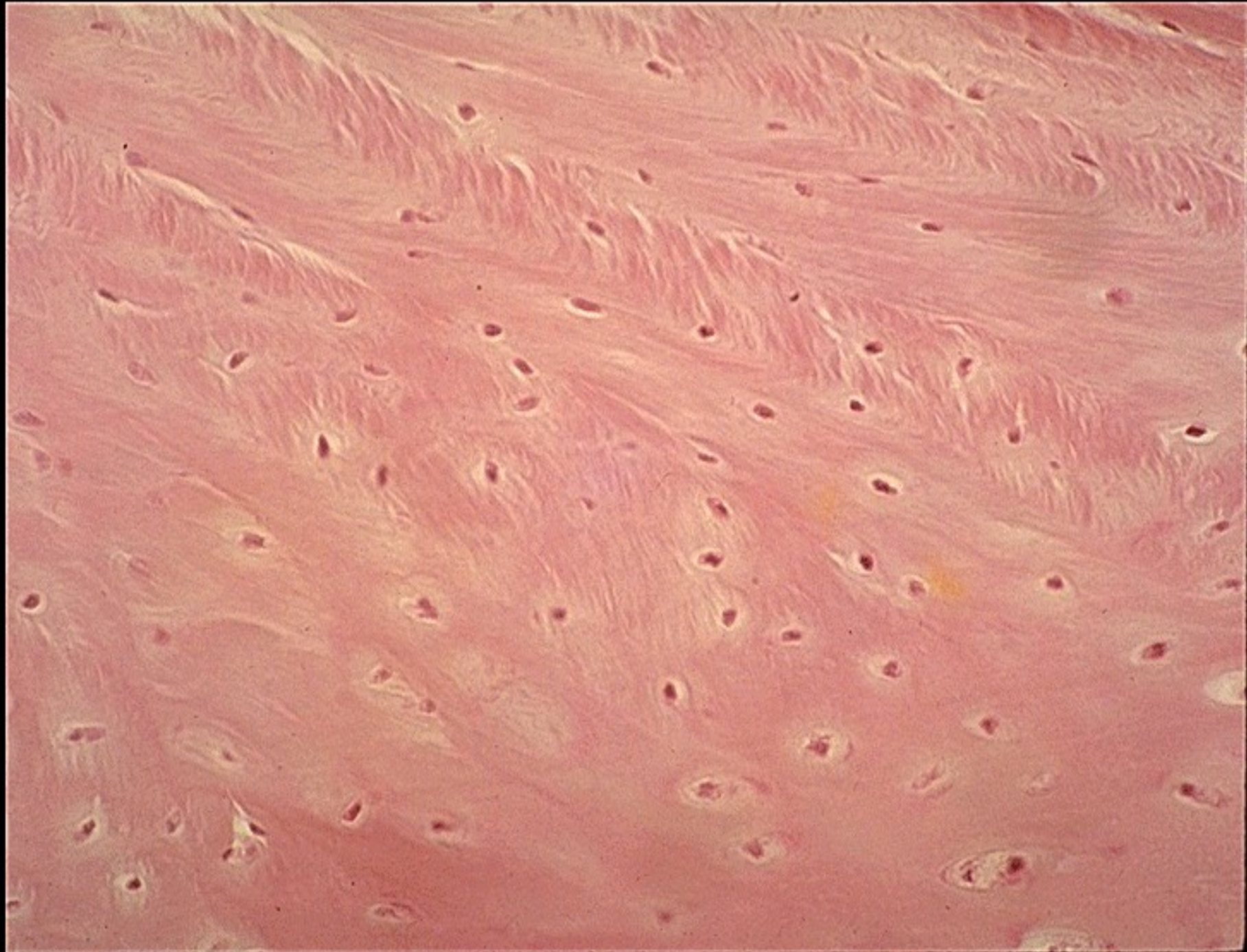


# Fibrocartilage

(example: symphysis, intervertebral discs)

- Small and flattened chondrocytes
- Thick bundles of collagenous fibers
- Small amount of ground substance
- No perichondrium





# Bone tissue

## Organic component

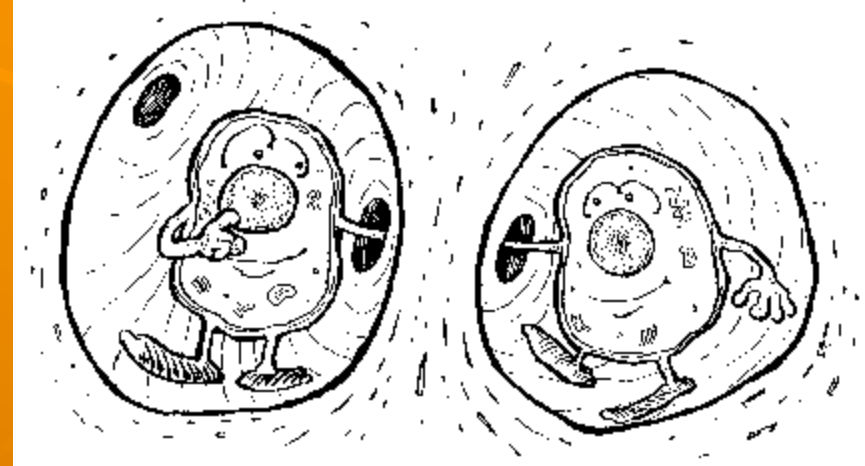
▣ Cells

▣ Collagenous fibers

▣ Ground substance is mineralized

## Inorganic component

▣ Minerals – hydroxyapatite crystals



# Intercellular matrix

## ✦ **Organic component:**

- collagen fibers (collagen type I)
- ground substance: 1) main glycosaminoglycan are chondroitin-sulfate, keratan-sulfate, 2) **fibronectin**, **osteopontin** (increase adhesion of cells and matrix), **osteocalcin** (binds Ca)

## ✦ **Inorganic component:** crystals of hydroxyapatite

# Bone cells

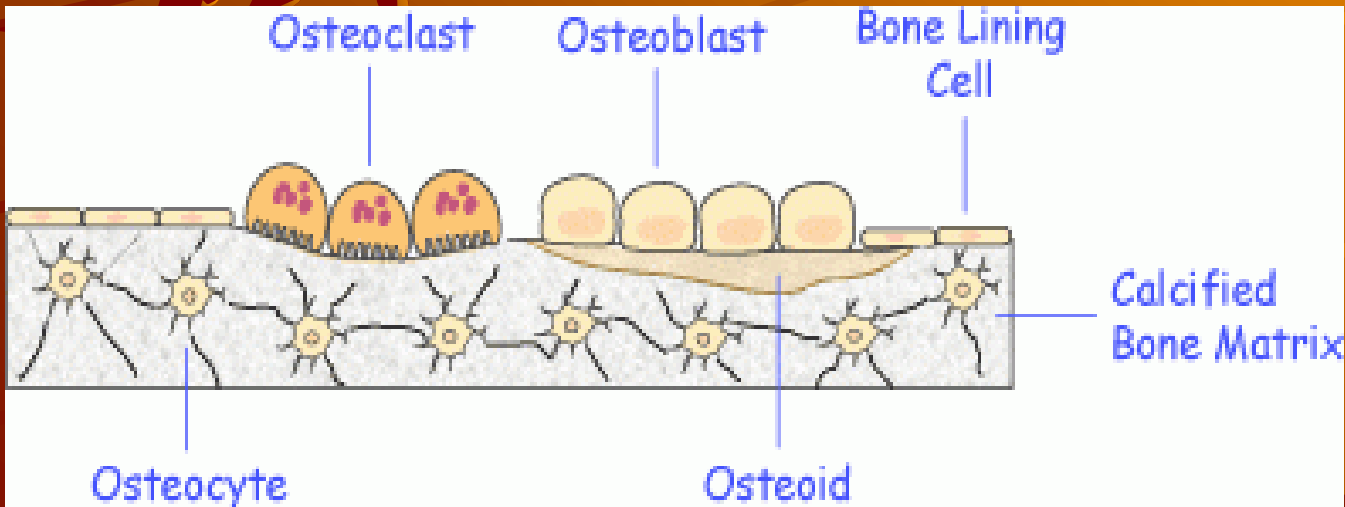
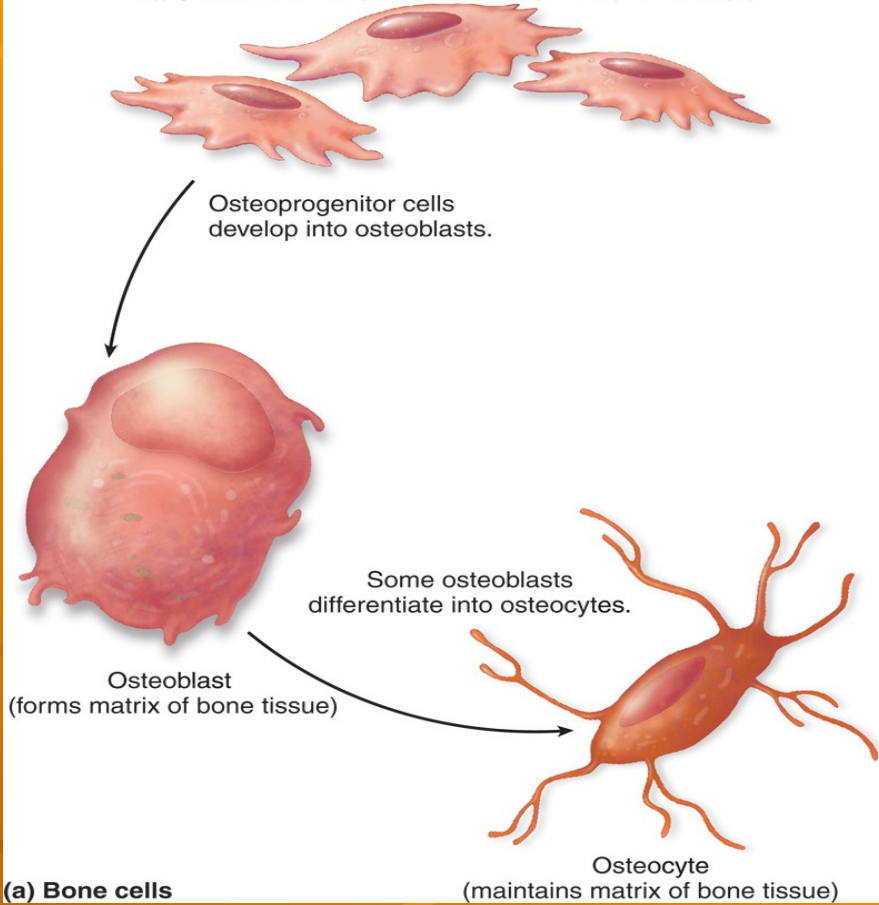
osteoprogenitor cells

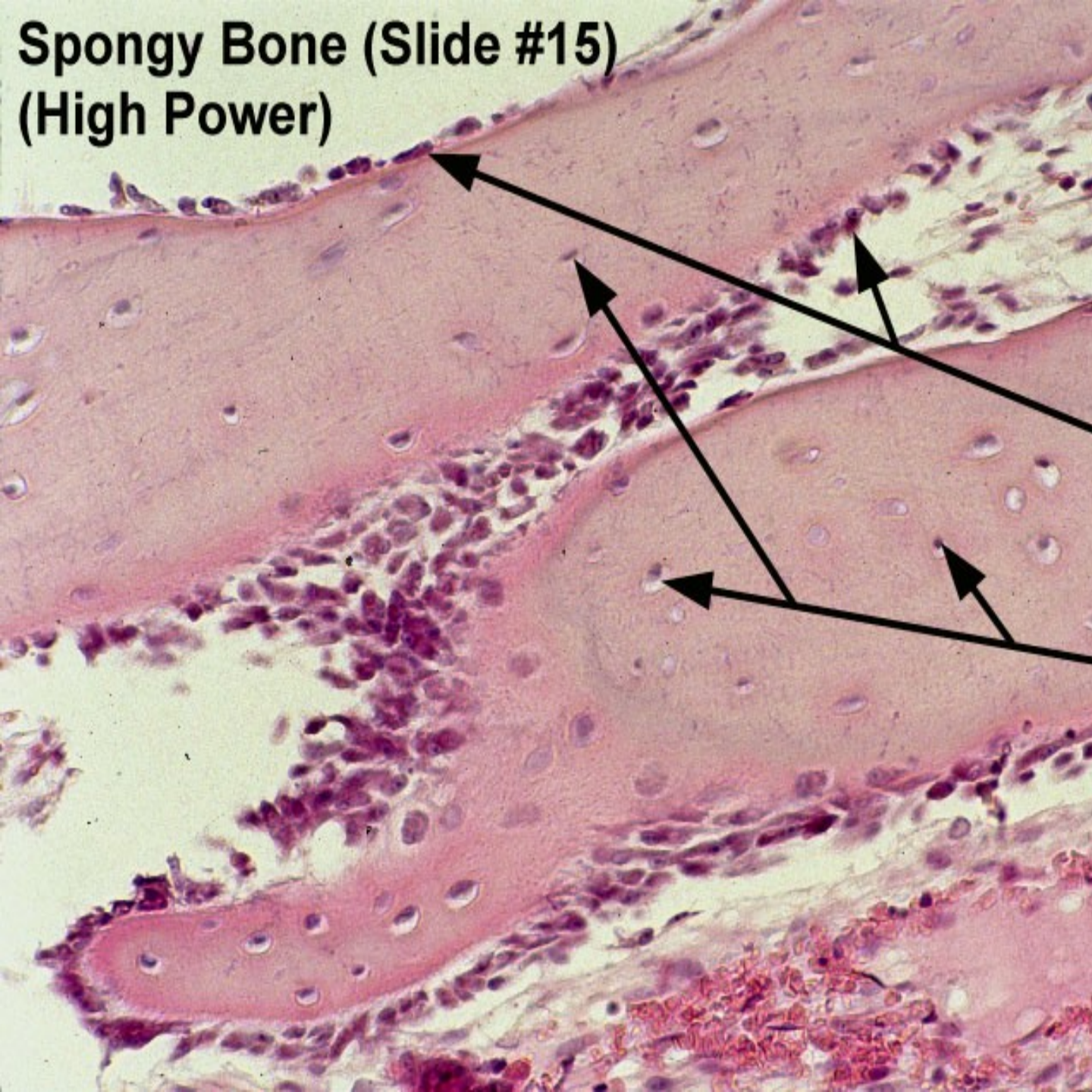
osteoblasts

osteocytes

osteoclasts

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**Spongy Bone (Slide #15)  
(High Power)**

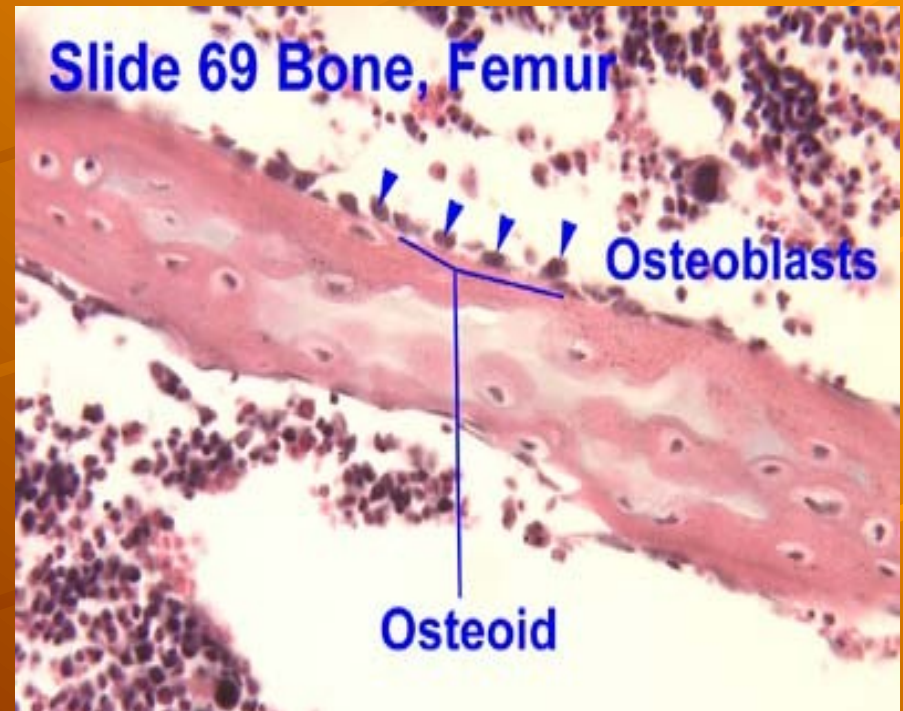
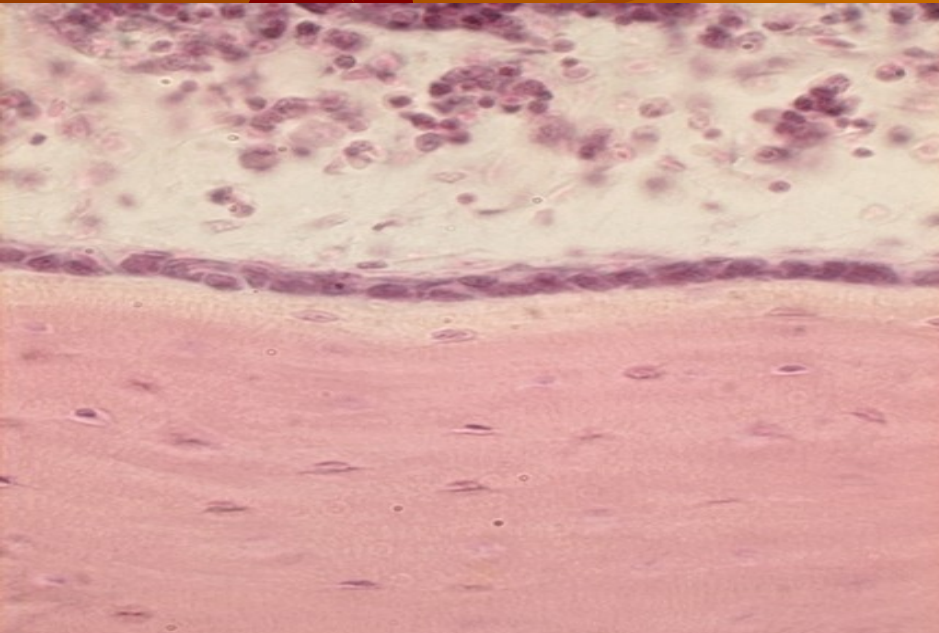
**Osteoblasts**  
*on the surface  
of the bone*

**Osteocytes**  
*inside of the  
bone matrix  
(in lacunae)*



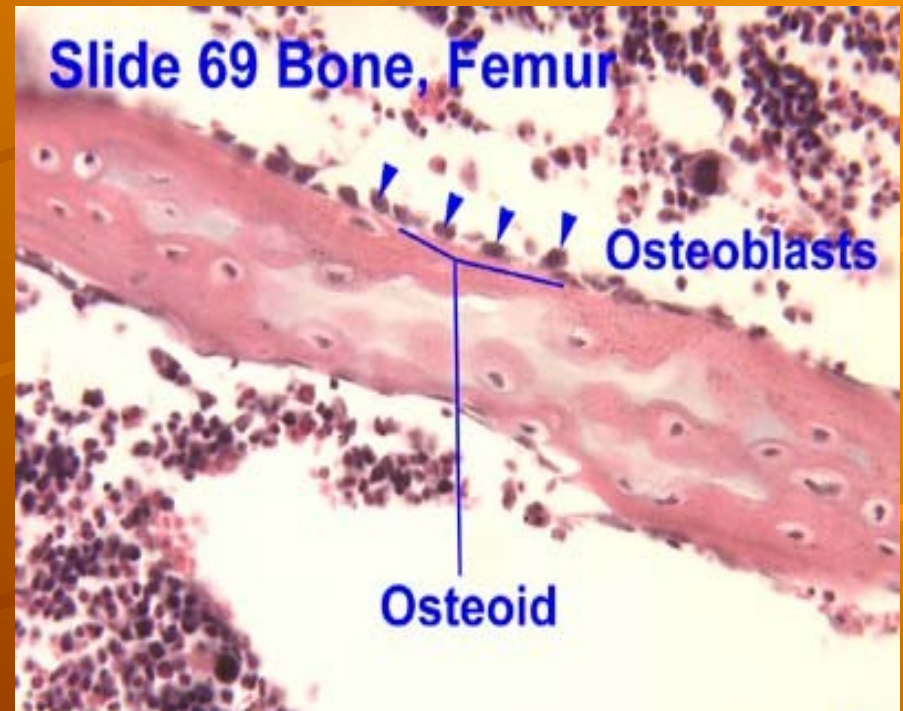
# osteoblasts

- on the surface of bone (epitheloid arrangement)
- synthesize non-mineralized intercellular matrix = **osteoid** (ground substance + collagen)
- periosteal osteoblasts – growth of bone by apposition



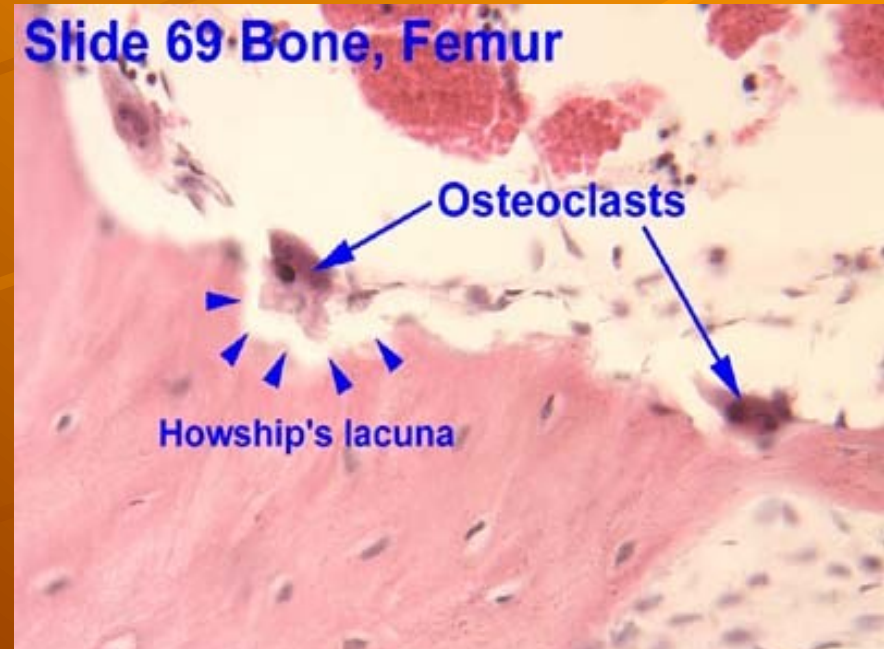
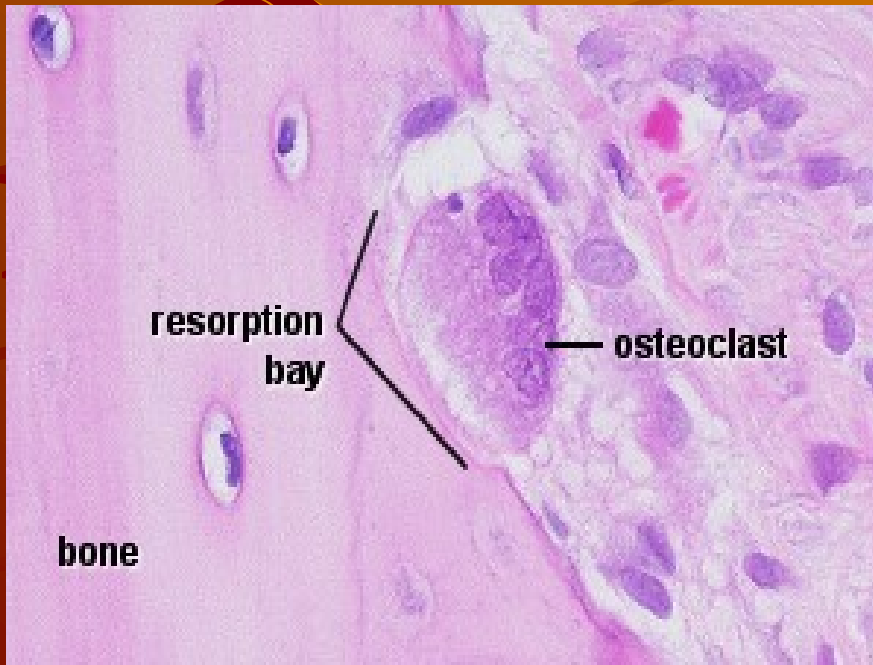
# osteocytes

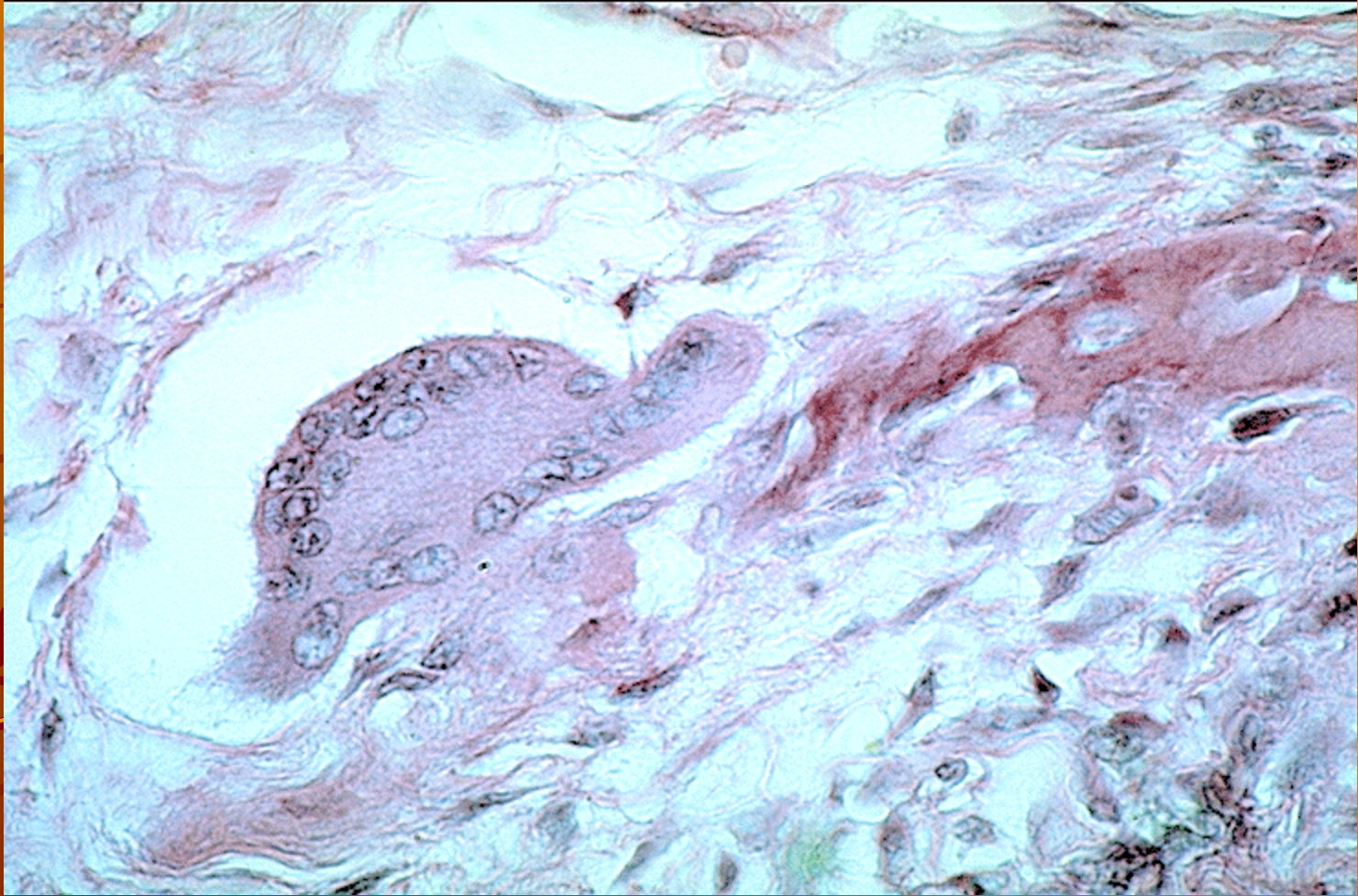
- osteoblasts entrapped in mineralized bone matrix = osteocytes (cytoplasmic processes in canaliculi ossium ■■■■ triation of bone lamellae)
- transport of minerals from blood into the matrix



# osteoclasts

- in Howship's lacunae
- large multinucleated cells (up to 50 nuclei) – arise by fusion of monocytes
- produce acid phosphatase – enzyme involved in bone resorption during ossification, bone remodeling or reparation





# Periosteum – endosteum

## Periosteum

dense c.t.capsule attached to bone by **Sharpey's fibers** (collagen); is composed of two layers: 1) **outer fibrous layer** of dense irregular c.t. and 2) **inner cellular layer** composed of cells that give rise to osteoblasts.

## Endosteum

thin layer of connective tissue containing osteoprogenitor cells and osteoblasts



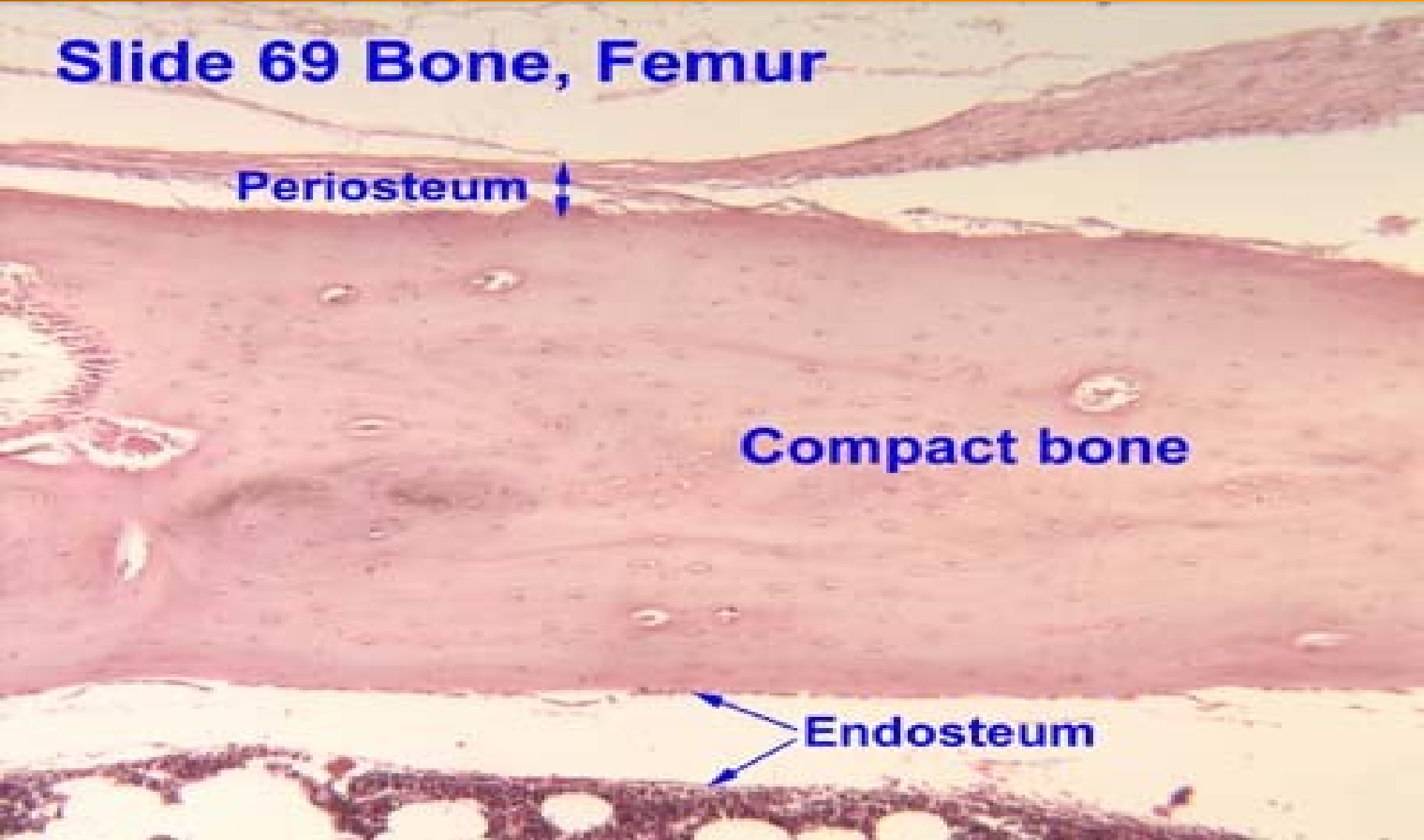
# Periosteum – endosteum

## Slide 69 Bone, Femur

Periosteum

Compact bone

Endosteum



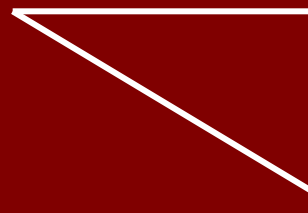
# Types of bone tissue

**Fibrillar**  
(woven)

**Lamellar**  
(cancellous)

**spongy**

**compact**



# Bone types

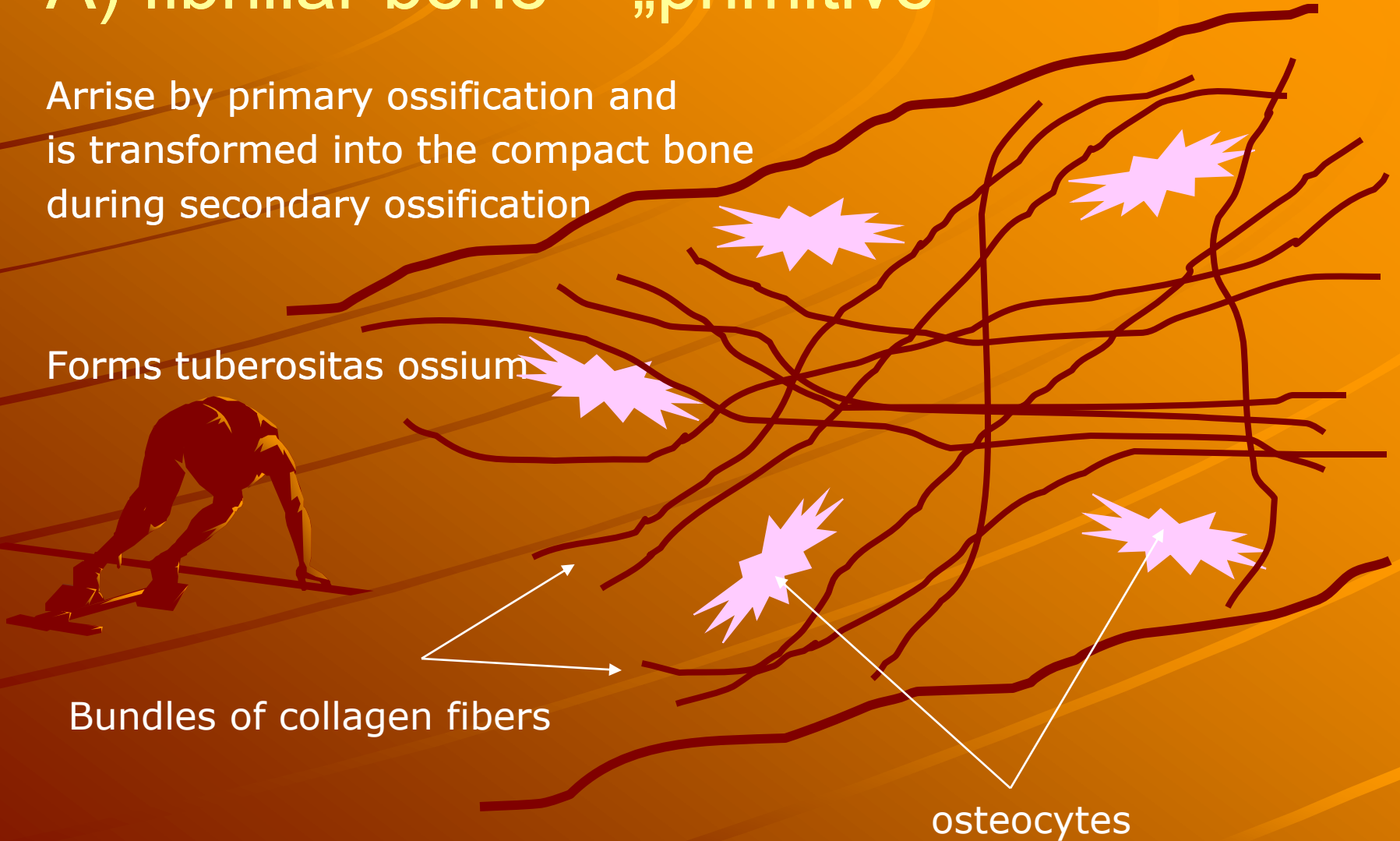
## A) fibrillar bone – „primitive“

Arise by primary ossification and is transformed into the compact bone during secondary ossification

Forms tuberositas ossium

Bundles of collagen fibers

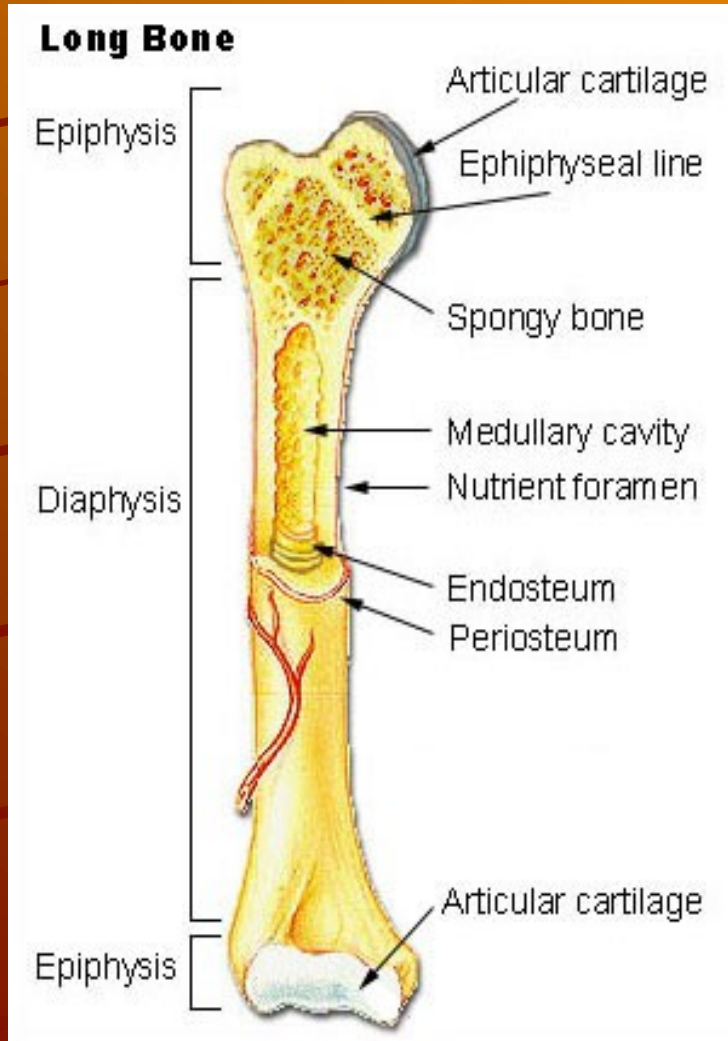
osteocytes



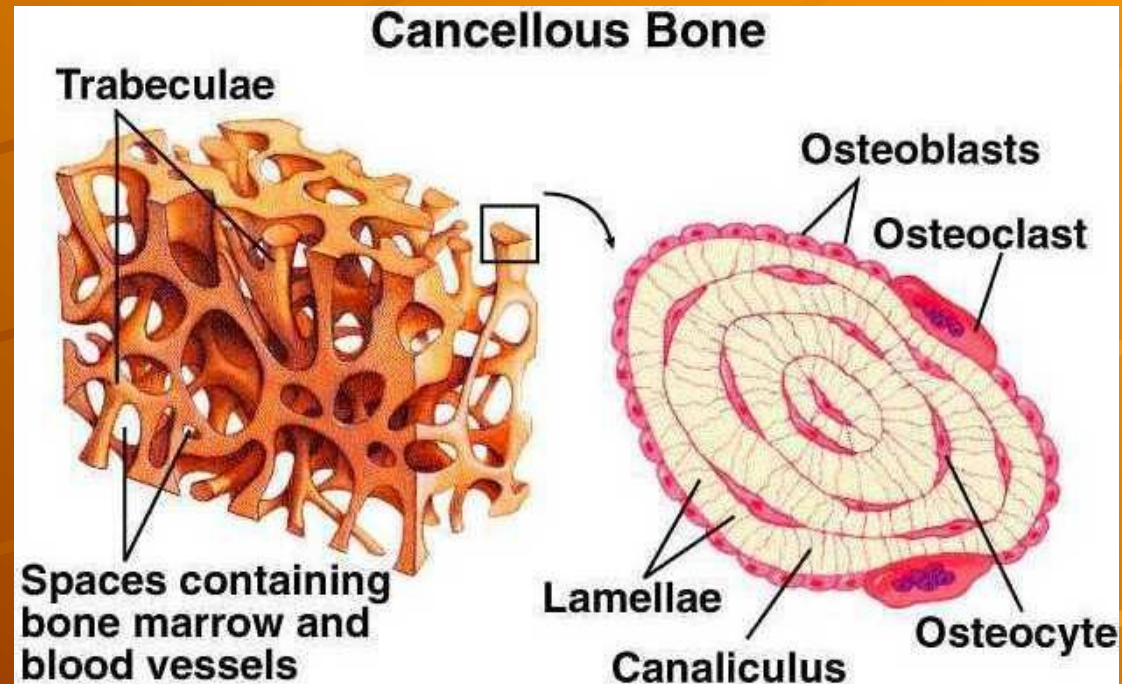


# Bone types

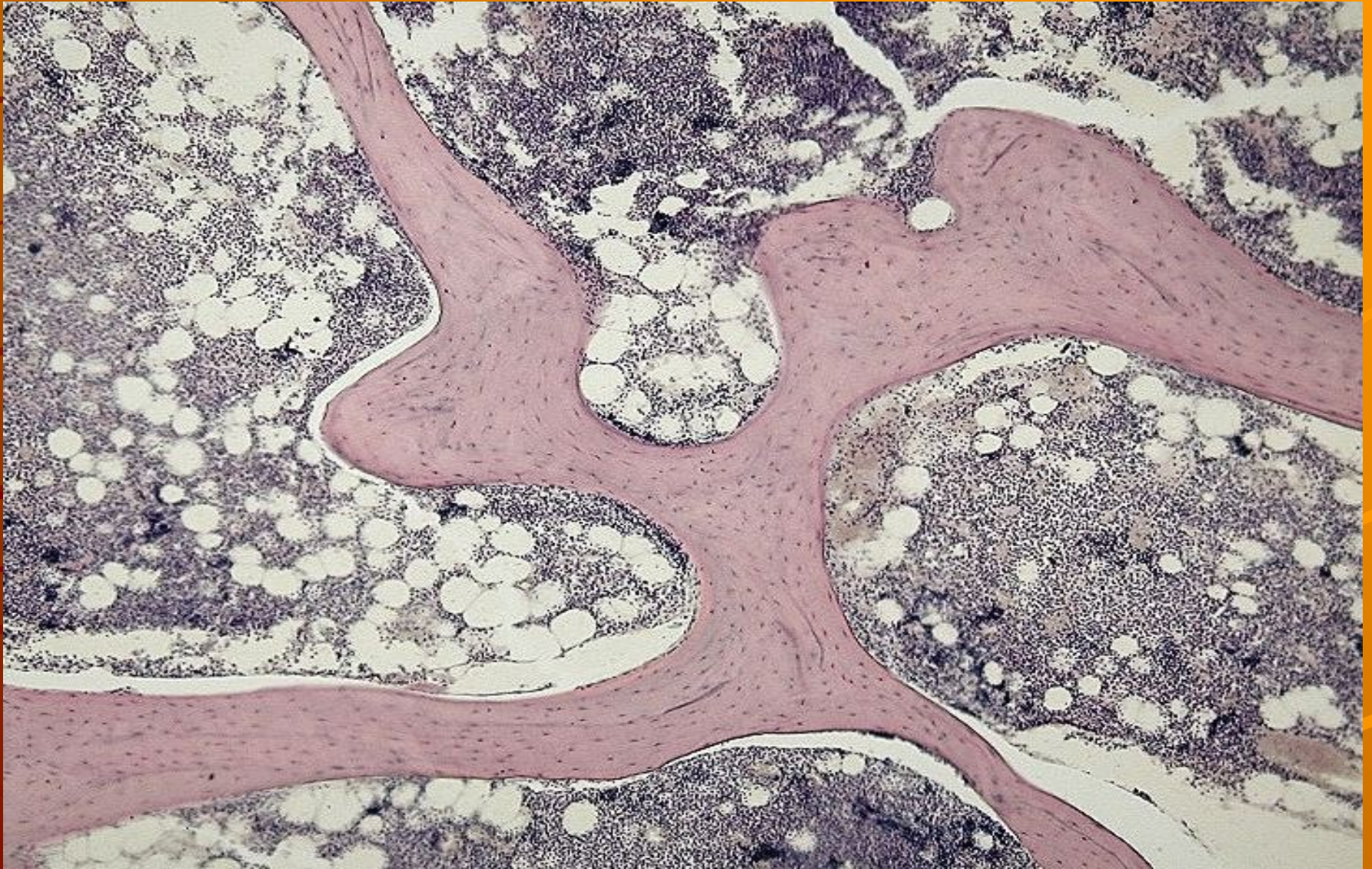
## B) lamellar (cancellous) bone



**spongy - compact**



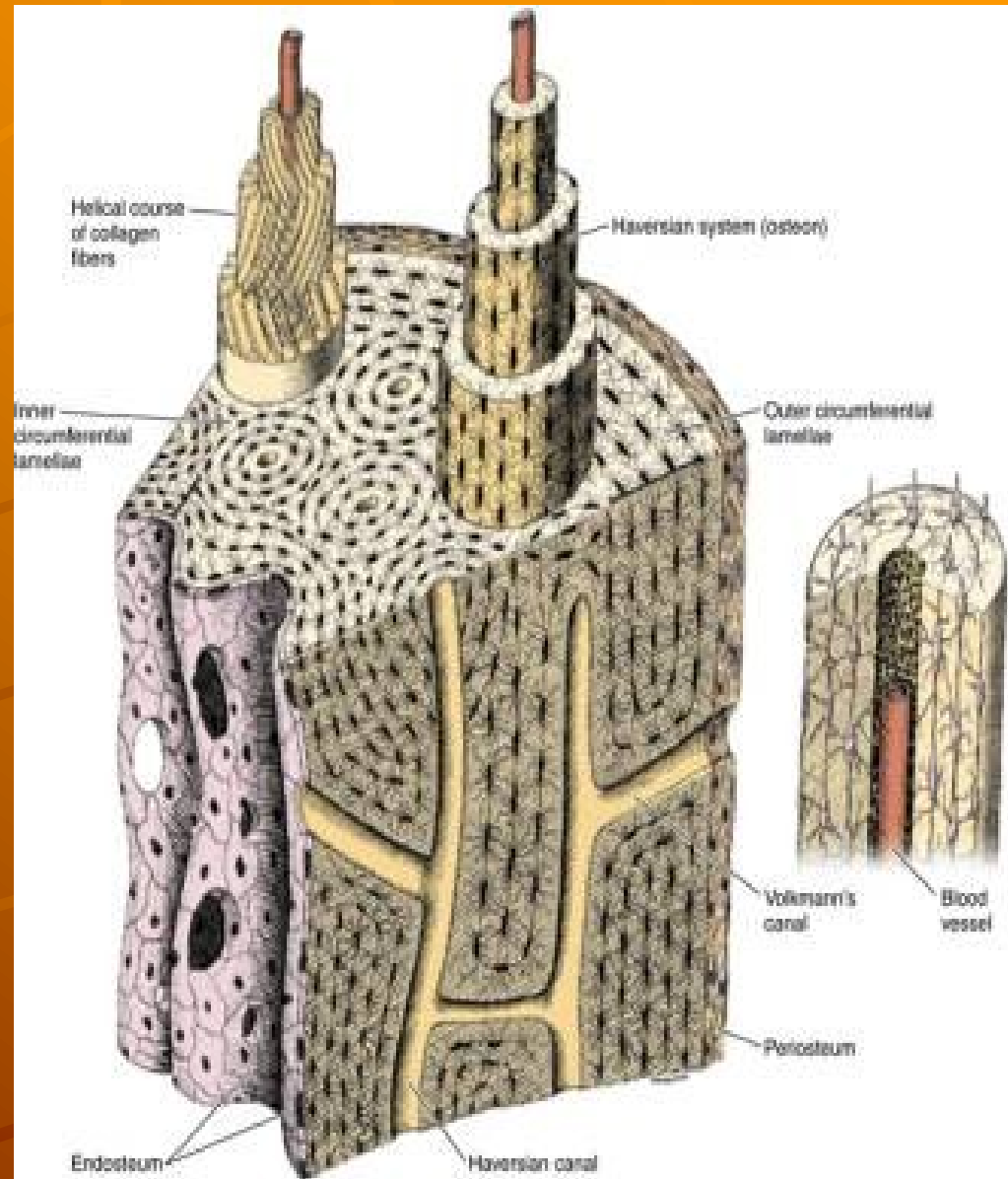
# Spongy (trabecular) bone



# compact bone

collagen fibers in mineralized ground substance form lamellae, which are arranged as:

- Haversian lamellae = osteon
- interstitial lamellae
- circumferential lamellae (inner, outer)



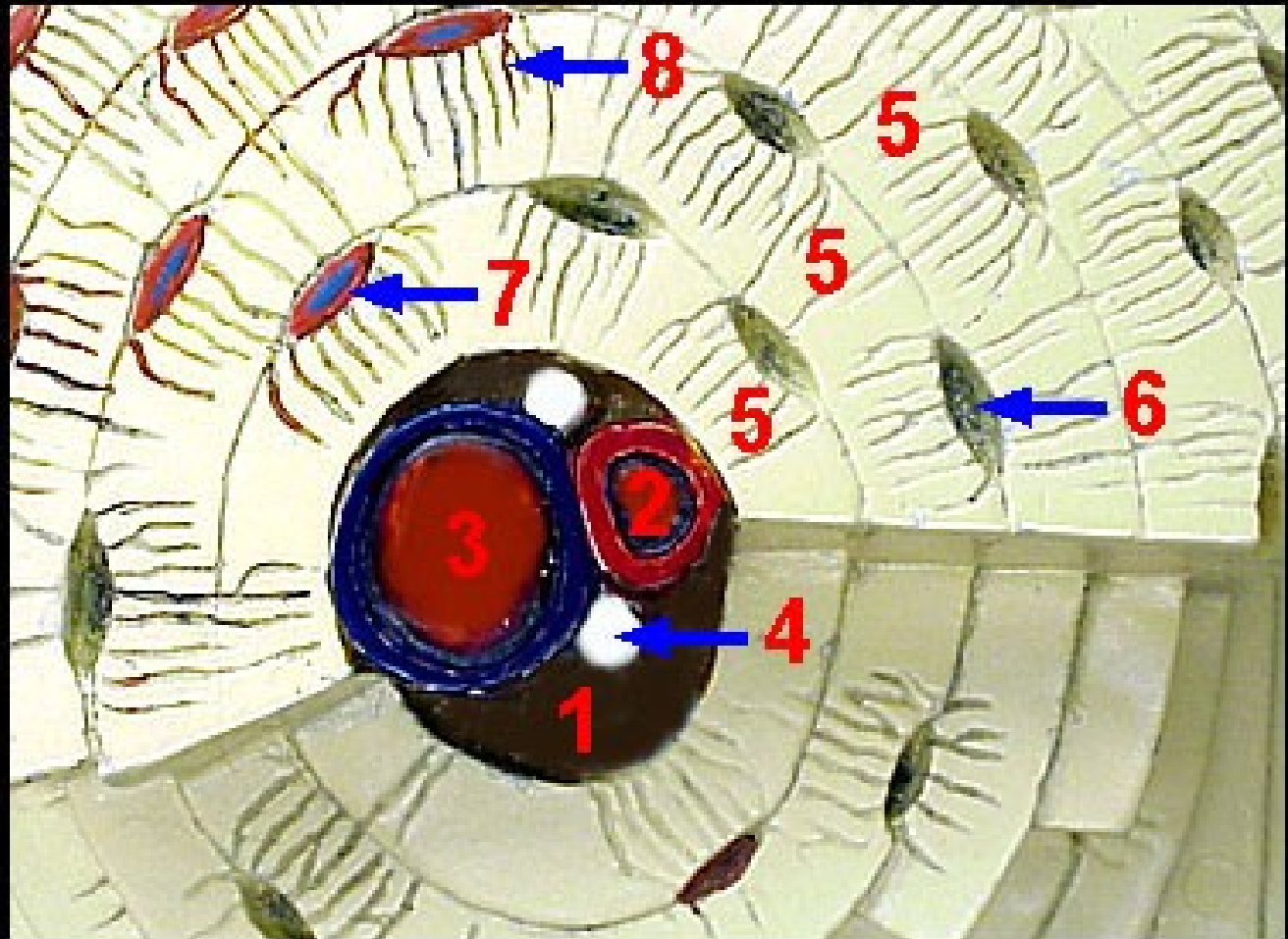
## OSTEON:

1 – Haversian canal contains 2, 3 – blood vessels, 4 – nerves in loose connective tissue;

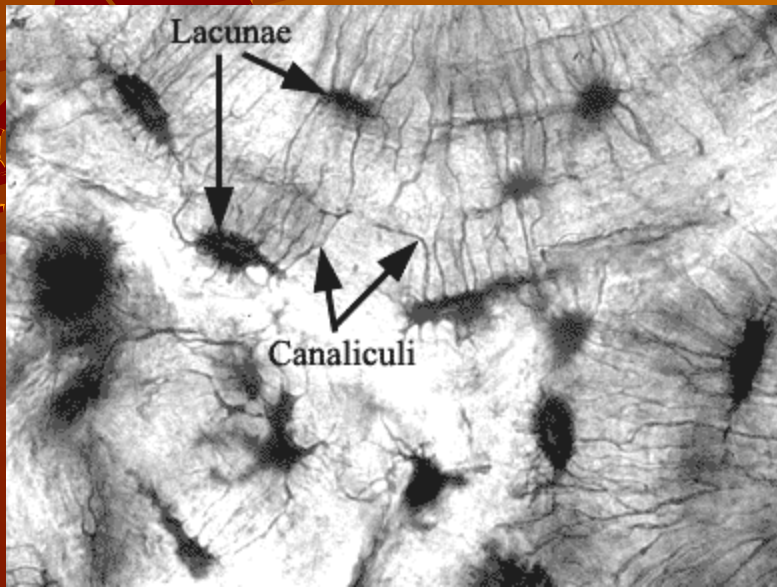
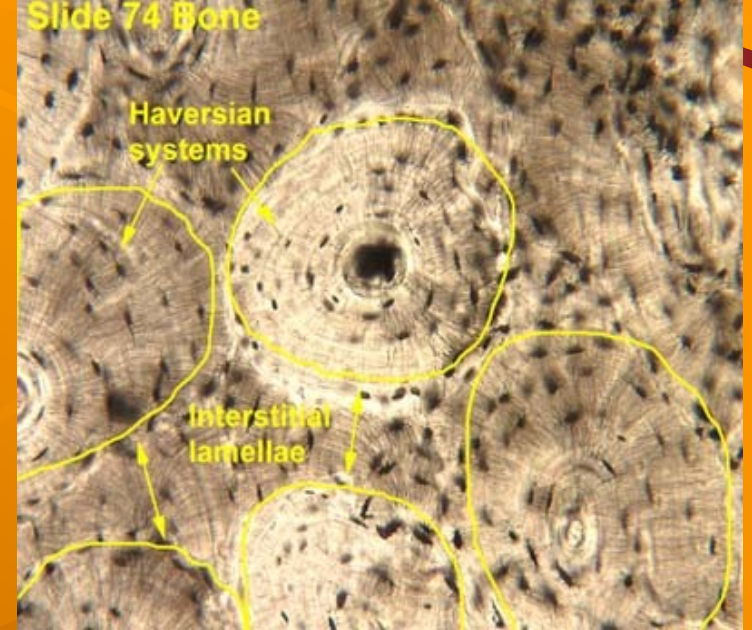
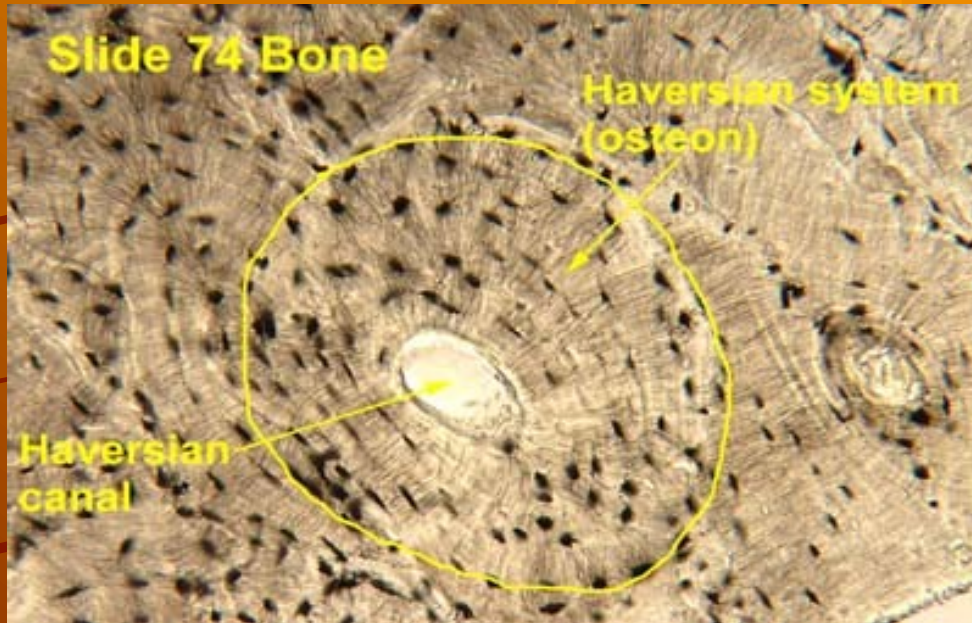
5 – lamellae

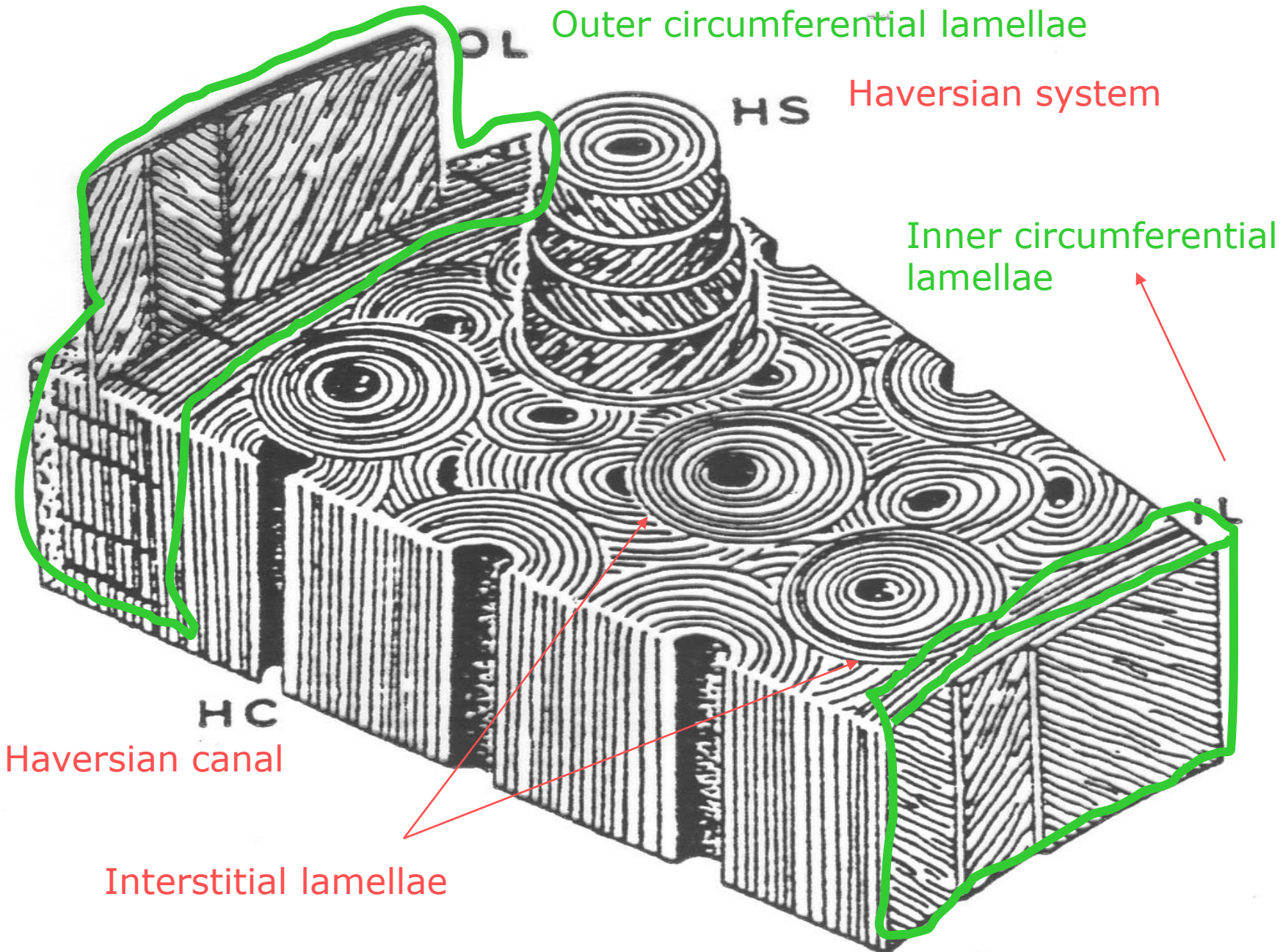
7 – osteocytes in lacunae (6)

8 – cytoplasmic processes in canaliculi ossium



# Osteon





Outer circumferential lamellae


Haversian system

Inner circumferential lamellae

HC  
Haversian canal

Interstitial lamellae

# Ossification – bone formation

- PERIOSTEUM contains **blood vessels** and **osteoprogenitor cells** osteogenic potential
-  **osteoblasts** (produce inter-cellular matrix OSTEOID)
- Osteoid + minerals (**from blood**) = OSSEIN
- Osteoblasts entrapped in ossein **osteocytes**,  
(osteocytes + ossein = BONE)
- The result of primary ossification is fibrillar bone
- Resorption, remodelation of fibrillar bone by **osteoclasts**
- The result of secondary ossification is compact bone  
(are involved in destruction of woven bone)

# Ossification types

## Intramembranous ossification

- bone develops in mesenchymal membrane  
flattened bones

## Endochondral ossification

- bone develops on a cartilage model  
long bones

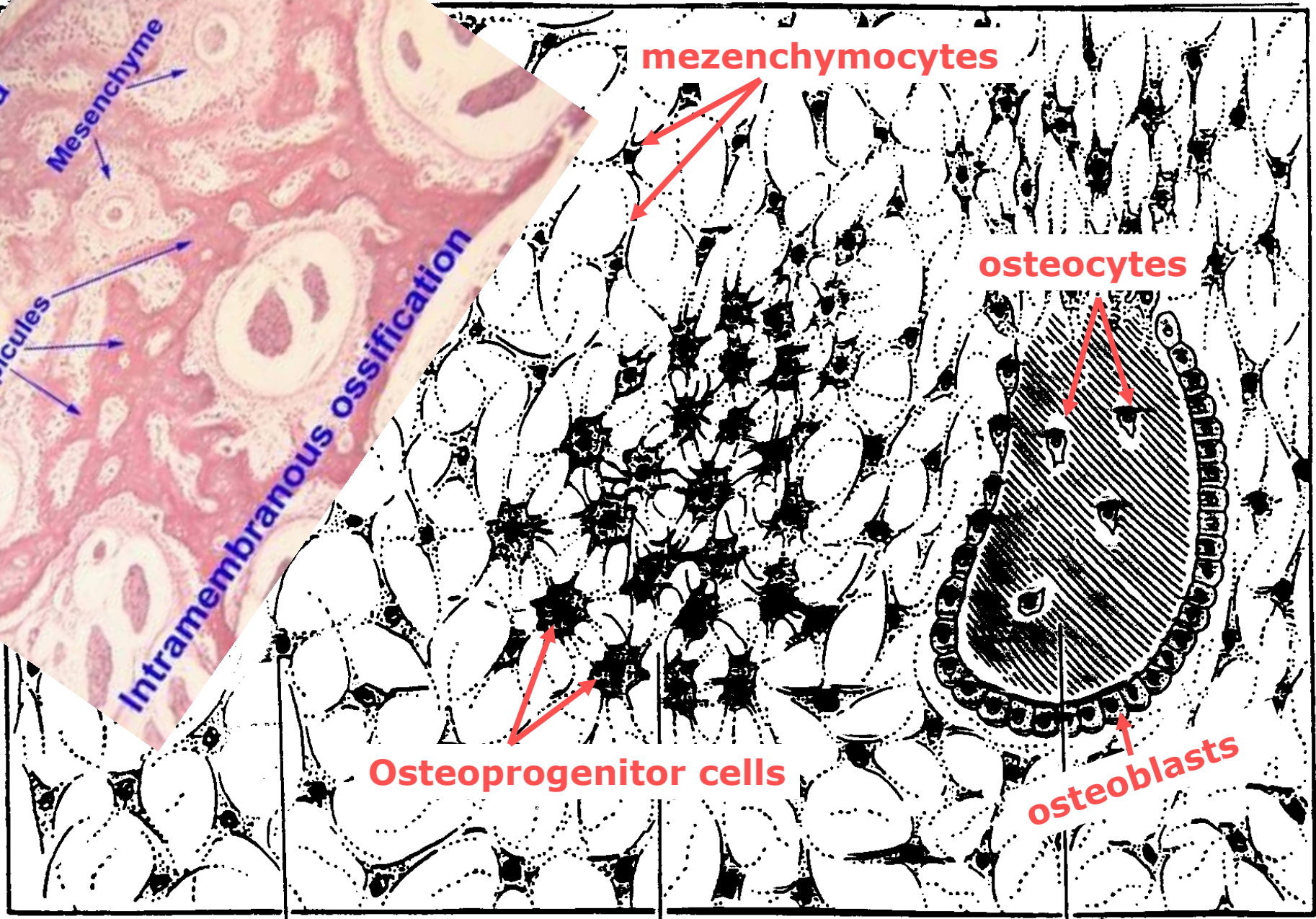
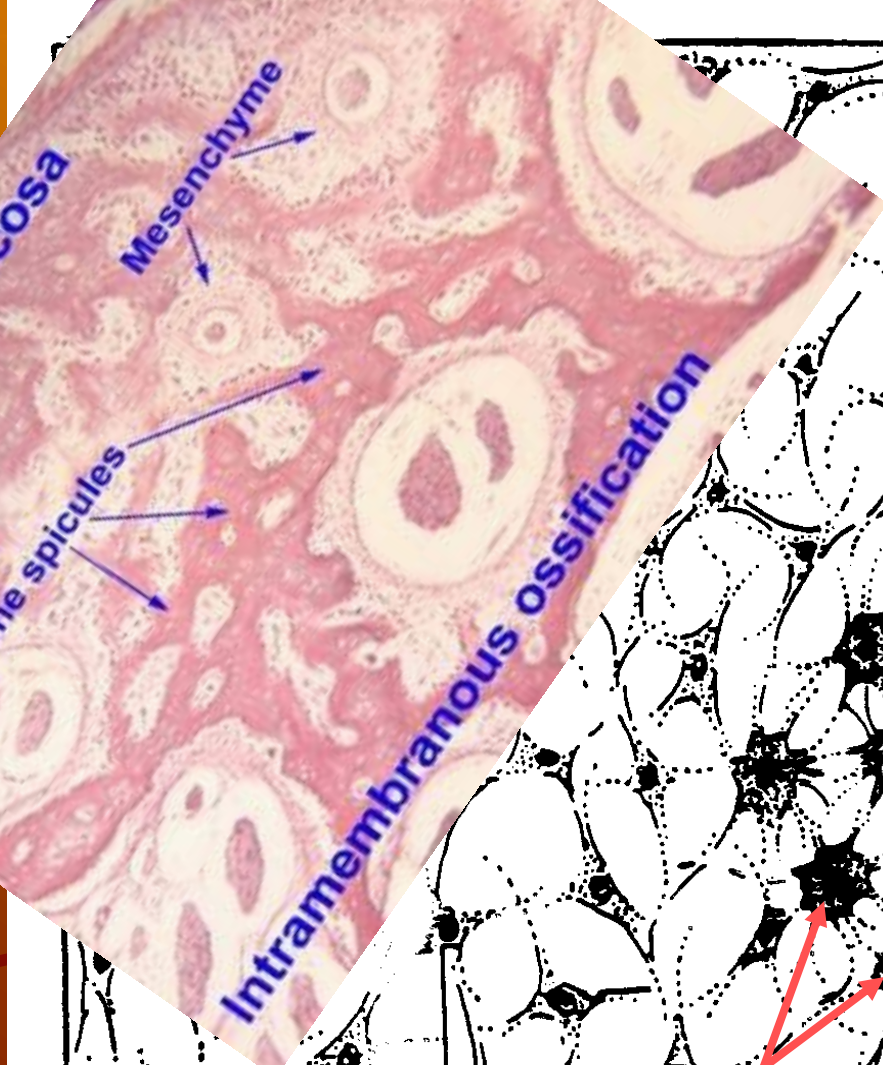


# Intramembranous ossification

Formation of bone directly from mesenchyme.

- Increased **vascularity** of mesenchyme
- Mesenchyme cells  osteoprogenitor cells   
**osteoblasts**
- Osteoblasts produce osteoid and transform into **osteocytes**.
- **Periosteum + endosteum** (dura mater in skull)

NEUROCRANIUM and other flat bones



Mesenchyme

Bone blastema

Primary bone tissue

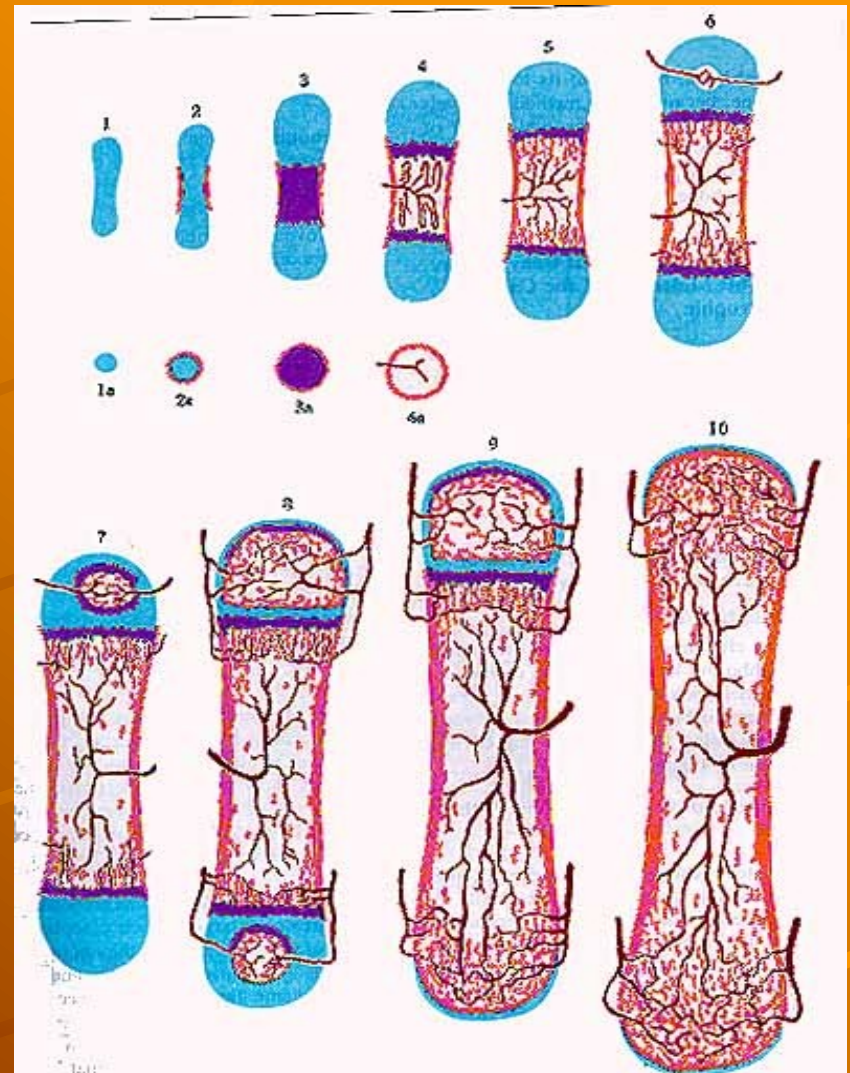
*Lacunae*

# Endochondral ossification

➡ **PRIMARY OSSIFICATION CENTER:** Occurs in the center of the diaphysis, and extends toward both epiphyses.

➡ **SECONDARY OSSIFICATION CENTER:** in the center of the epiphyses.

➡ **EPIPHYSEAL PLATE:** is hyaline cartilage which allows bone growing in the length.



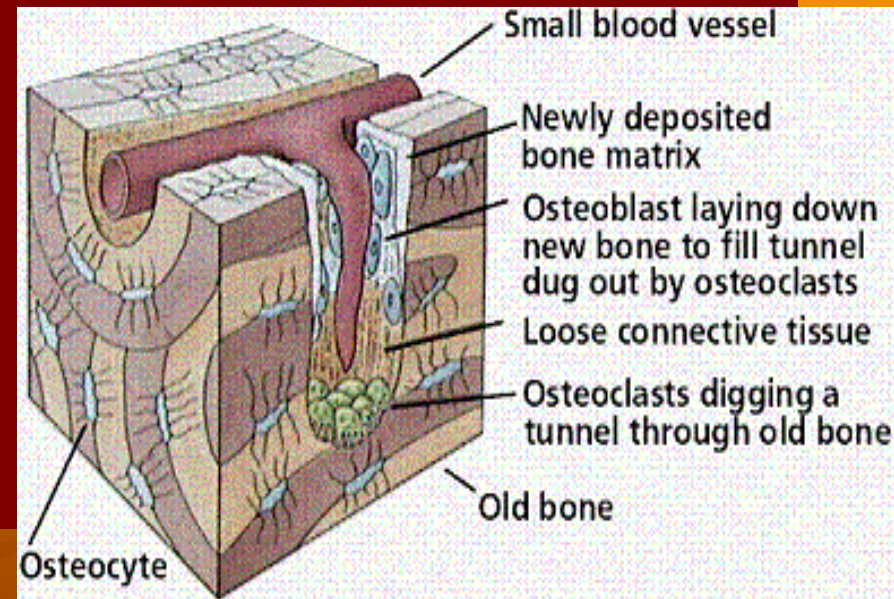
# Endochondral ossification

- ❏ Perichondrium around diaphysis transforms into **periosteum** (fibroblasts ████████ osteoprogenitor cells)
- ❏ Blood vessels invade the cartilage and bring **osteoprogenitor cells**, blood cells, bone marrow cells, **macrophages**, endothelial cells.
- ❏ Ca and other ions are transported by blood and intercellular matrix around the chondrocytes is calcified – nutrients cannot diffuse to them – cells degenerate and die

<to be continued>

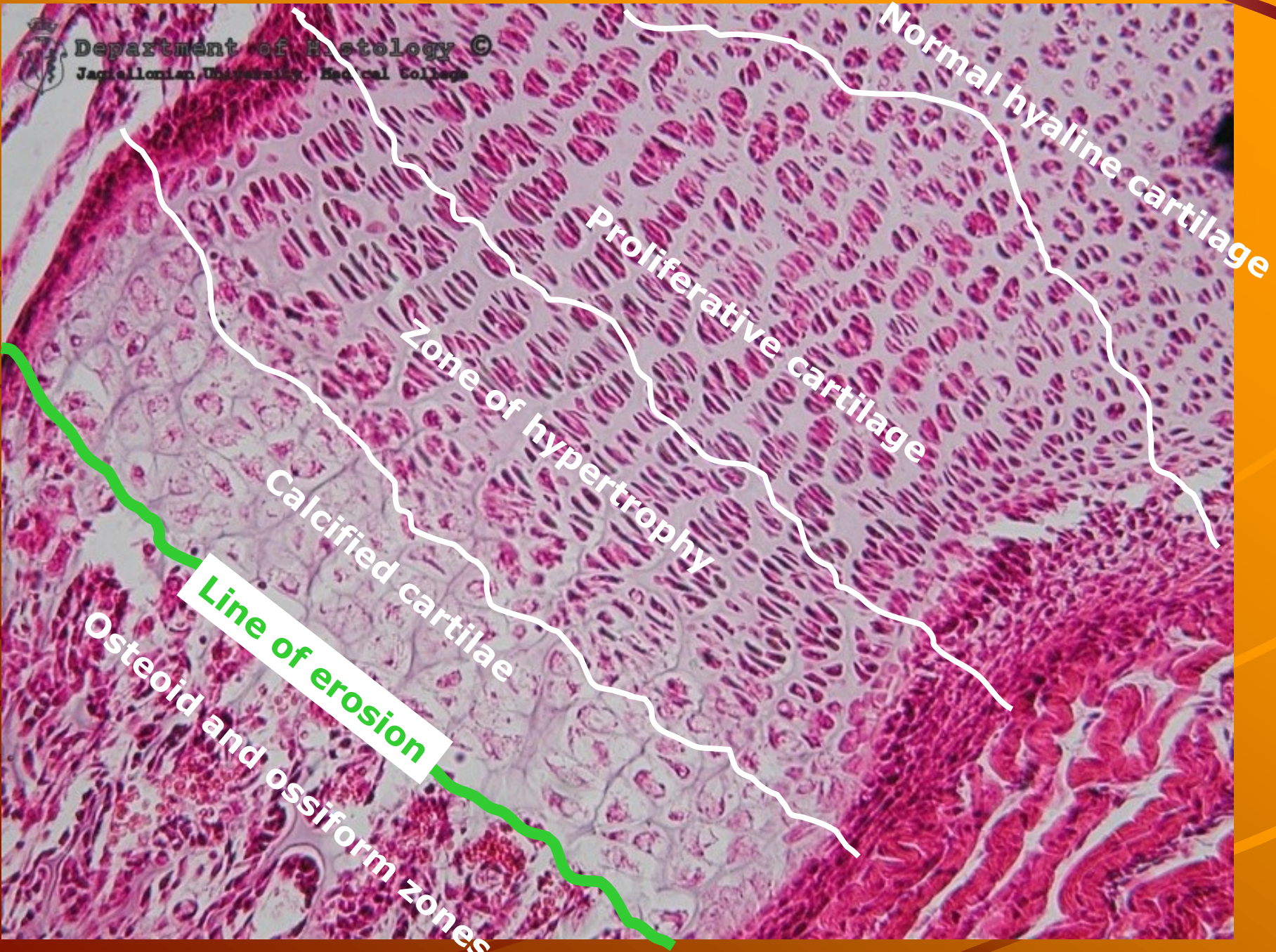
# Endochondral ossification

- ✦ after chondrocyte death mineralized intercellular matrix forms **spicules** (rests of cartilage)
- ✦ they are occupied by **osteoblasts** producing **osteoid**
- ✦ osteoid surrounds osteoblasts and is **mineralized** by ions from blood; osteoblasts **osteocytes**
- ✦ **primary ossification**  
= fibrillar bone
- ✦ resorption - **osteoclasts**
- ✦ **secondary ossification**  
= lamellar bone



# OSSIFICATION ZONES

- **RESERVE ZONE**: normal hyaline cartilage
- **PROLIFERATIVE ZONE**: chondrocytes in columns
- **ZONE of HYPERTROPHY**: enlarged chondrocytes
- **CALCIFIED ZONE**: (*different staining of intercellular matter in light microscope*)
- „**LINE of erosion**“ – imaginary line
- **OSSIFORM ZONE**: spicules, osteoid, ossein in regions of bone formation



Normal hyaline cartilage

Proliferative cartilage

Zone of hypertrophy

Calcified cartilage

**Line of erosion**

Osteoid and ossiform zones

# Cartilage

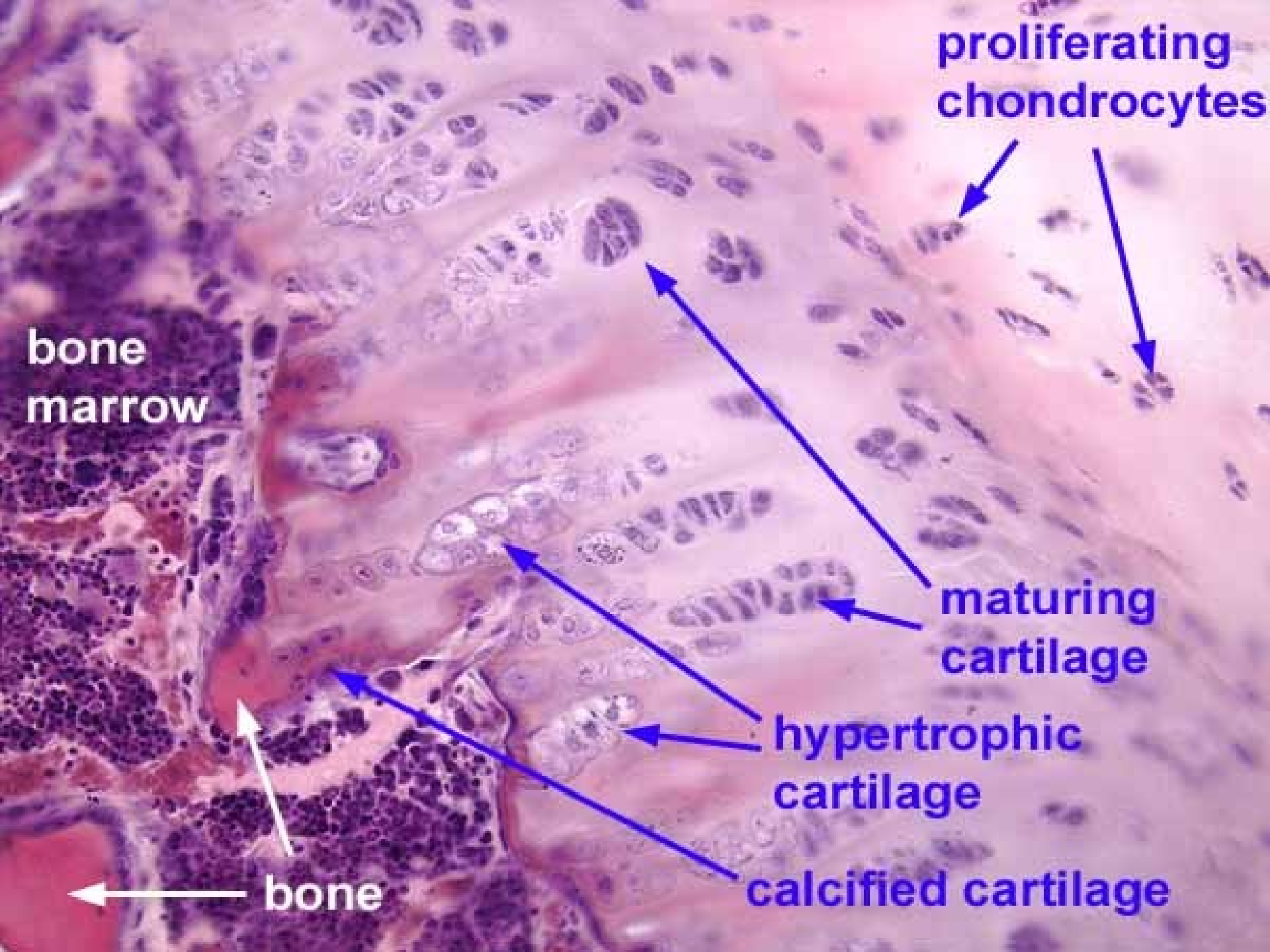
## Bone

### Slides:

- Hyaline cartilage (28. Trachea, HE)
- Elastic cartilage (26. Epiglottis, HE)  
(27. Elastic cartilage, orcein)
- Lamellar bone (...)
- Endochondral ossification (...)

### Atlas EM:





**proliferating  
chondrocytes**

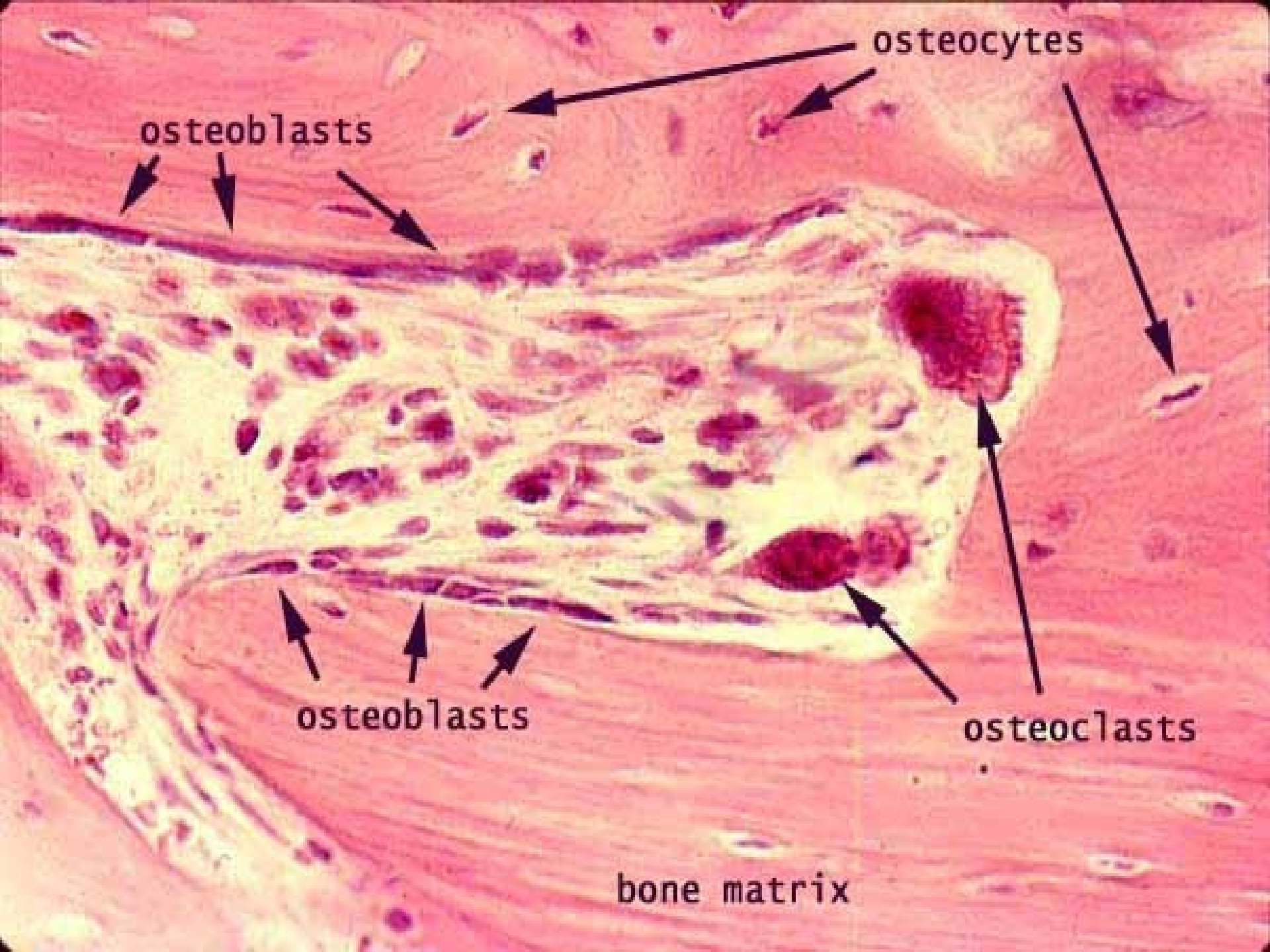
**bone  
marrow**

**maturing  
cartilage**

**hypertrophic  
cartilage**

**calcified cartilage**

**bone**



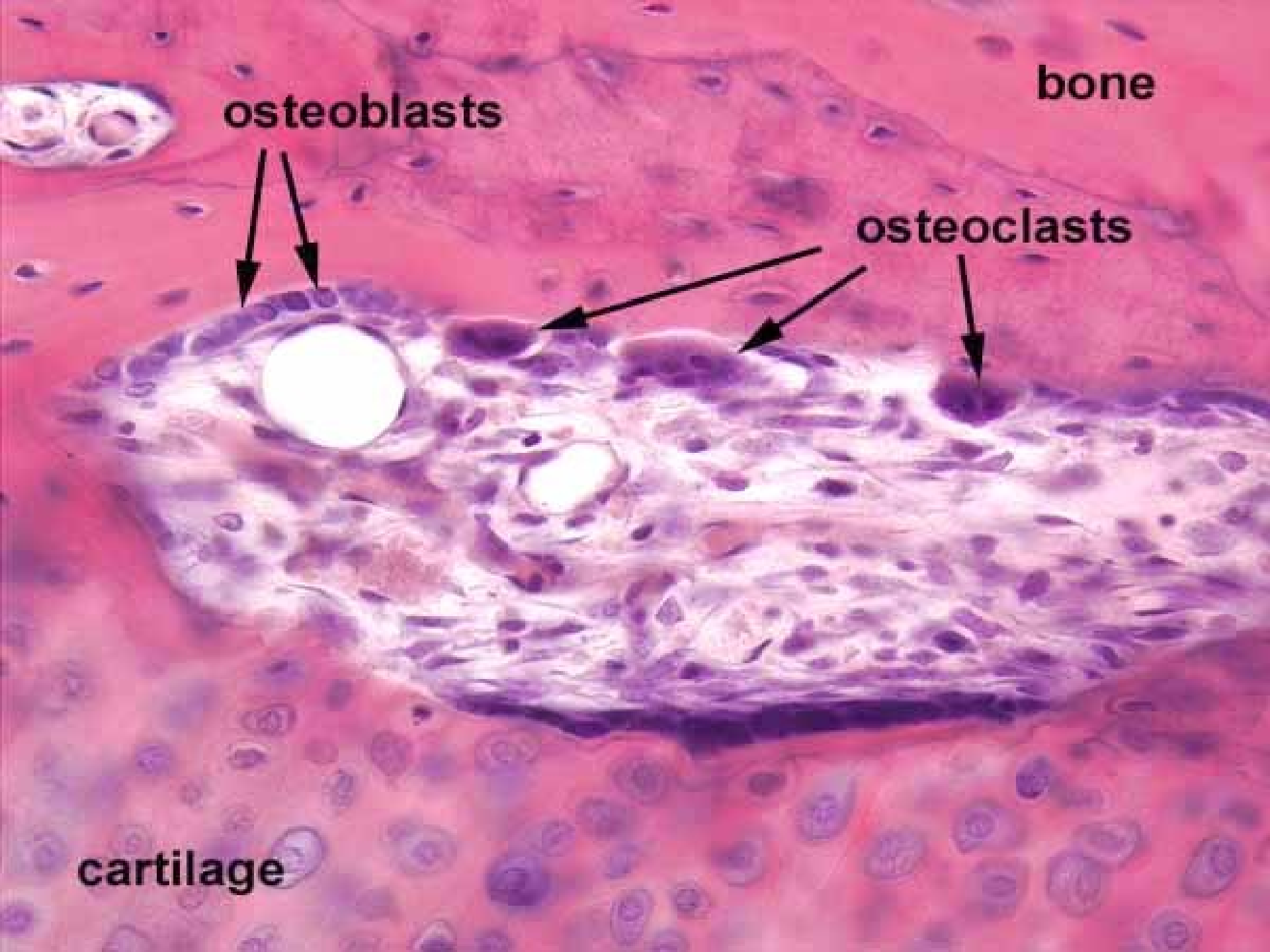
osteocytes

osteoblasts

osteoblasts

osteoclasts

bone matrix



**osteoblasts**

**bone**

**osteoclasts**

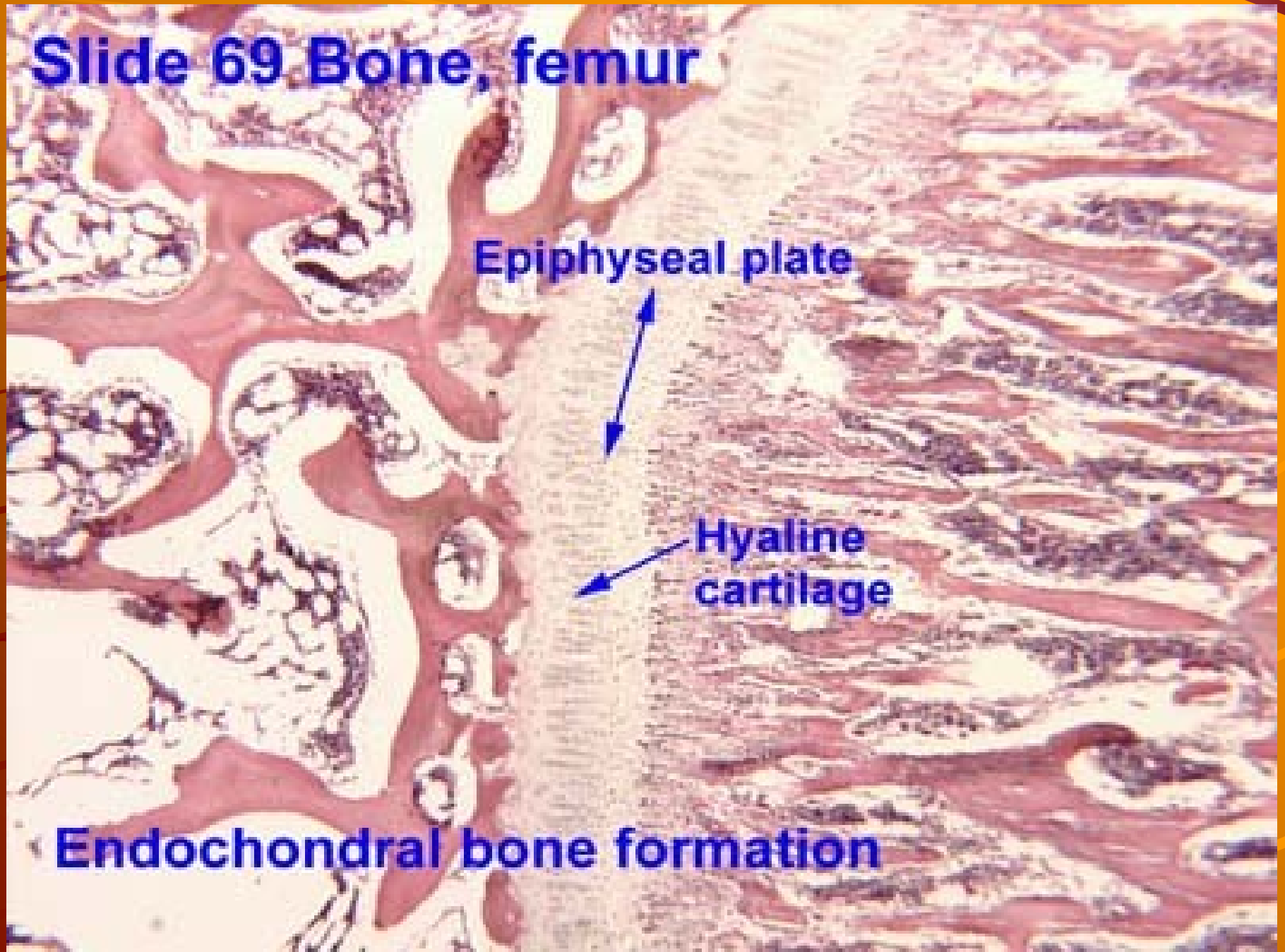
**cartilage**

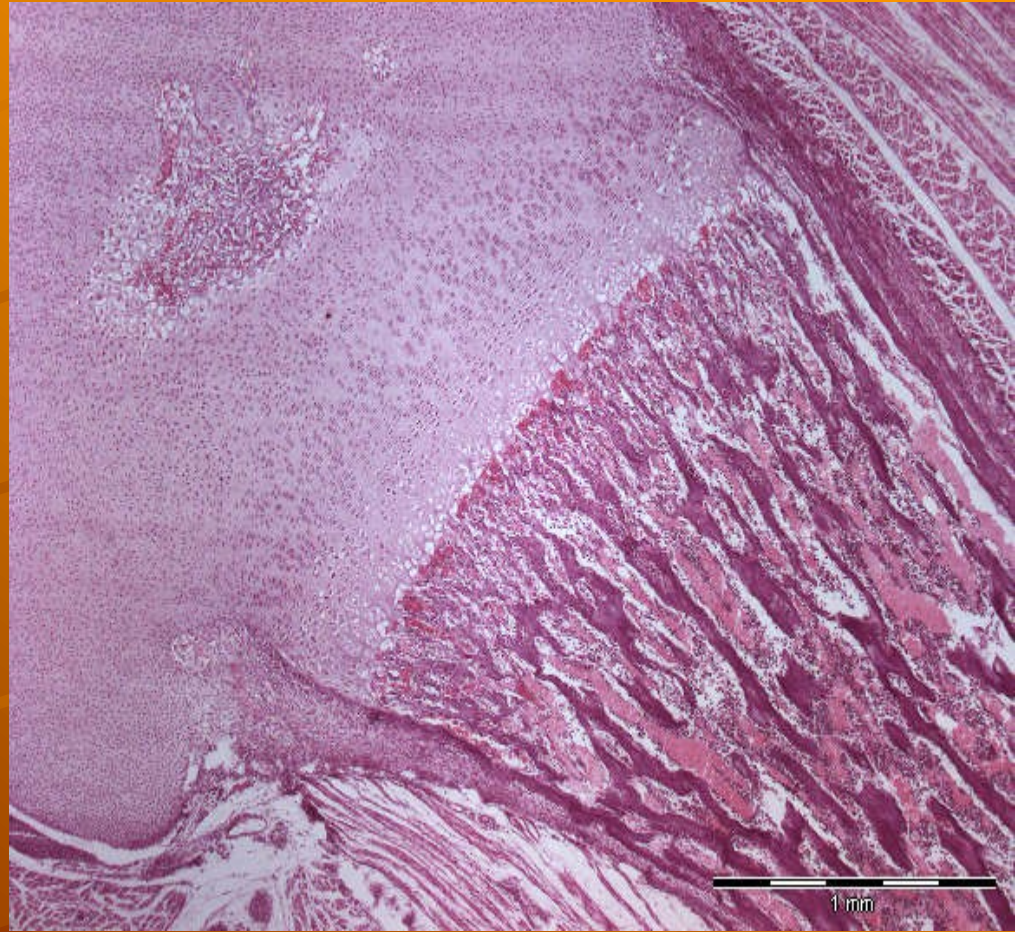
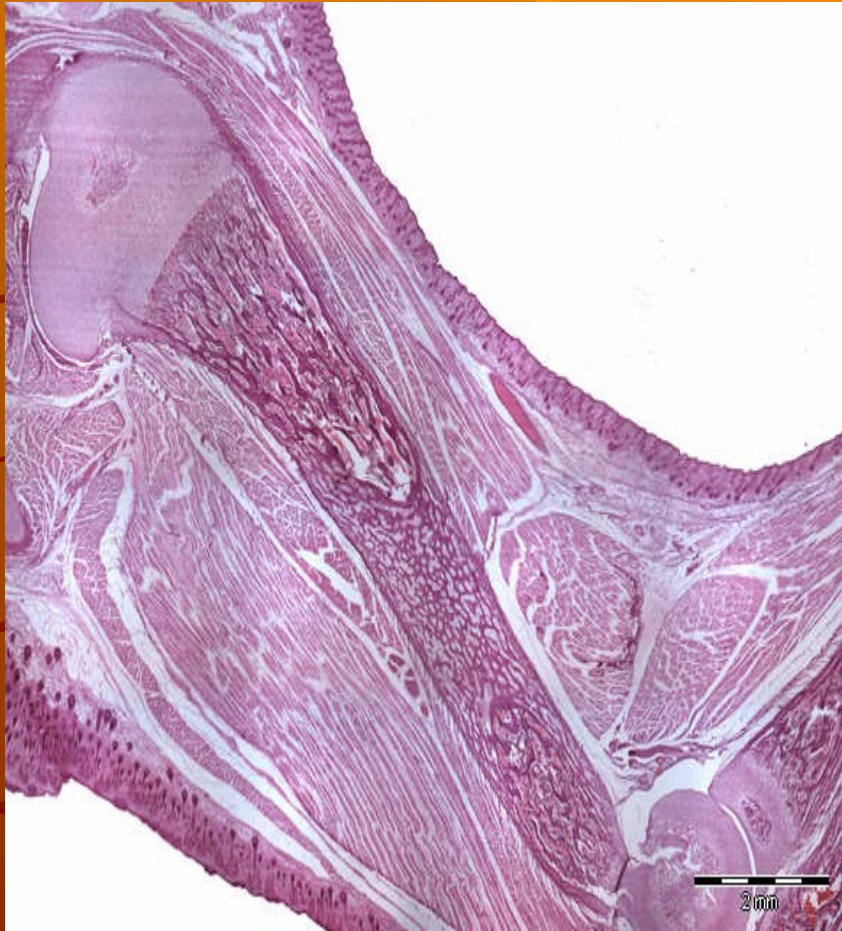
# Slide 69 Bone, femur

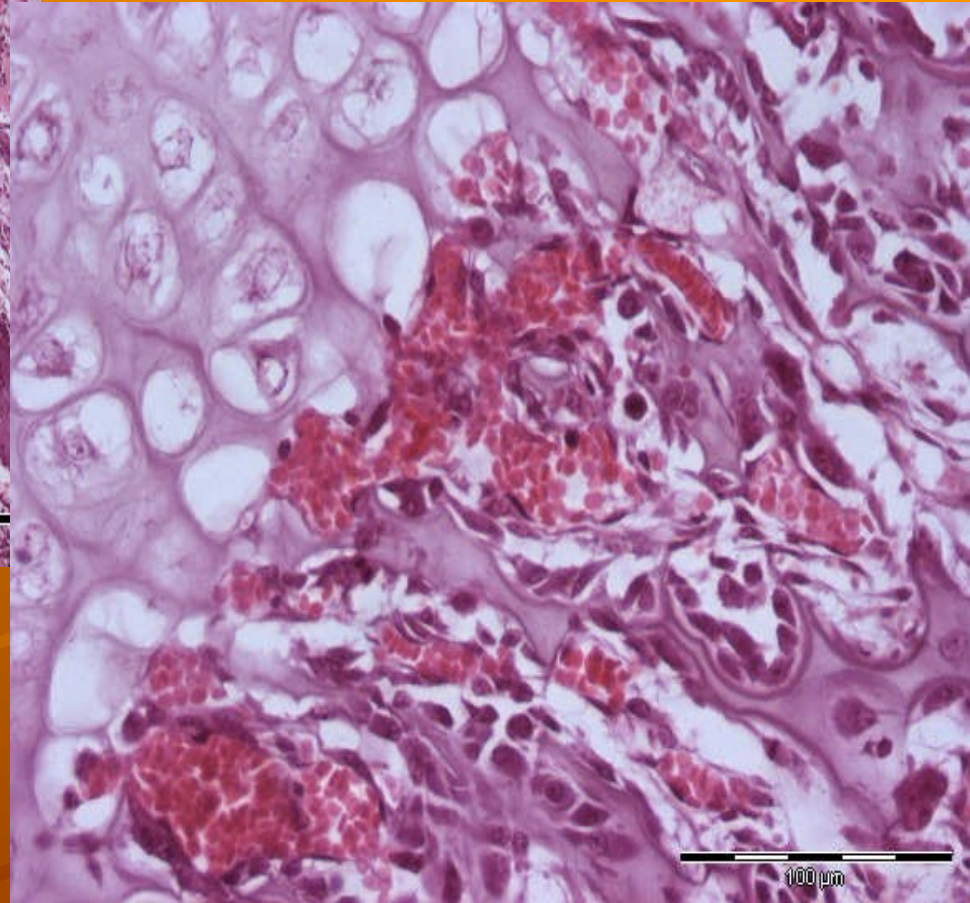
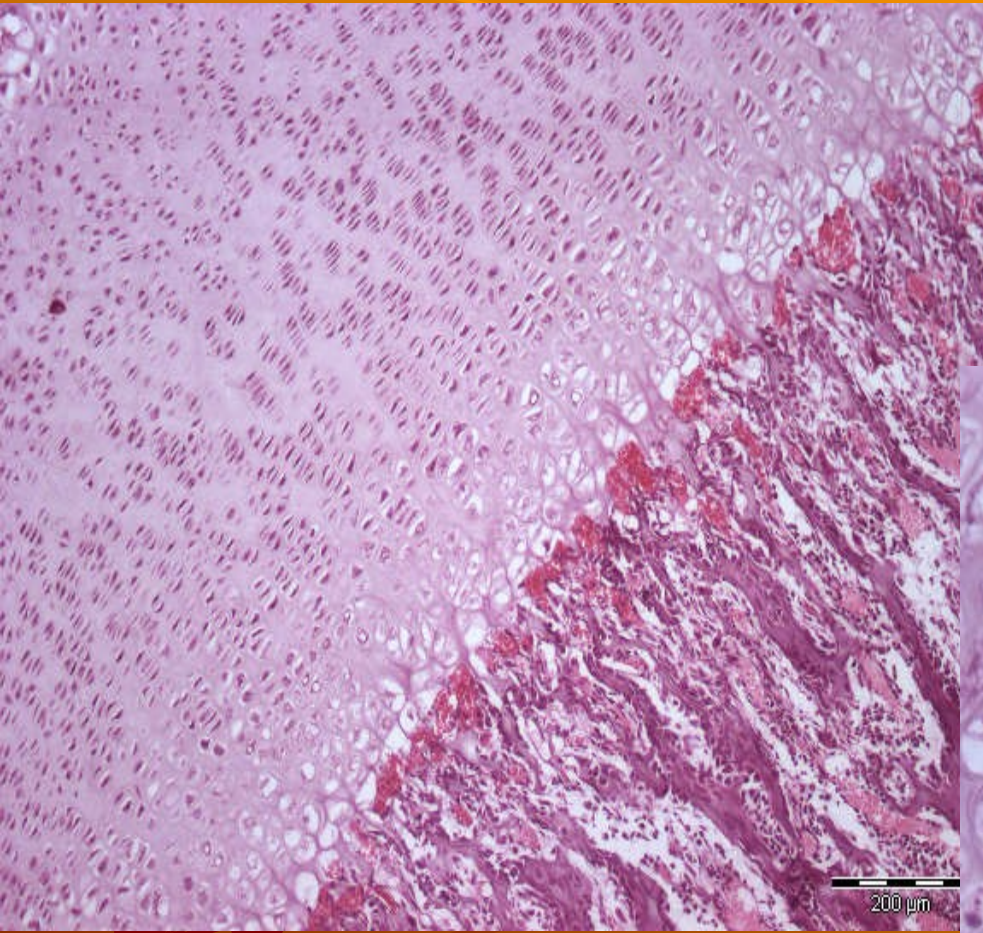
Epiphyseal plate

Hyaline cartilage

Endochondral bone formation







# Bone junctions - joints

✚ **SYNARTHROSES:** Poorly moveable (fibrous) or immobile joints.

✚ **Syndesmosis:** Bones connected by dense fibrous connective tissue, as in SKULL SUTURES

✚ **Synchondrosis:** Bones connected by cartilage, as in the PUBIC SYMPHYSIS.

✚ **Synostosis:** bones connected by bone tissue, as in the PELVIS

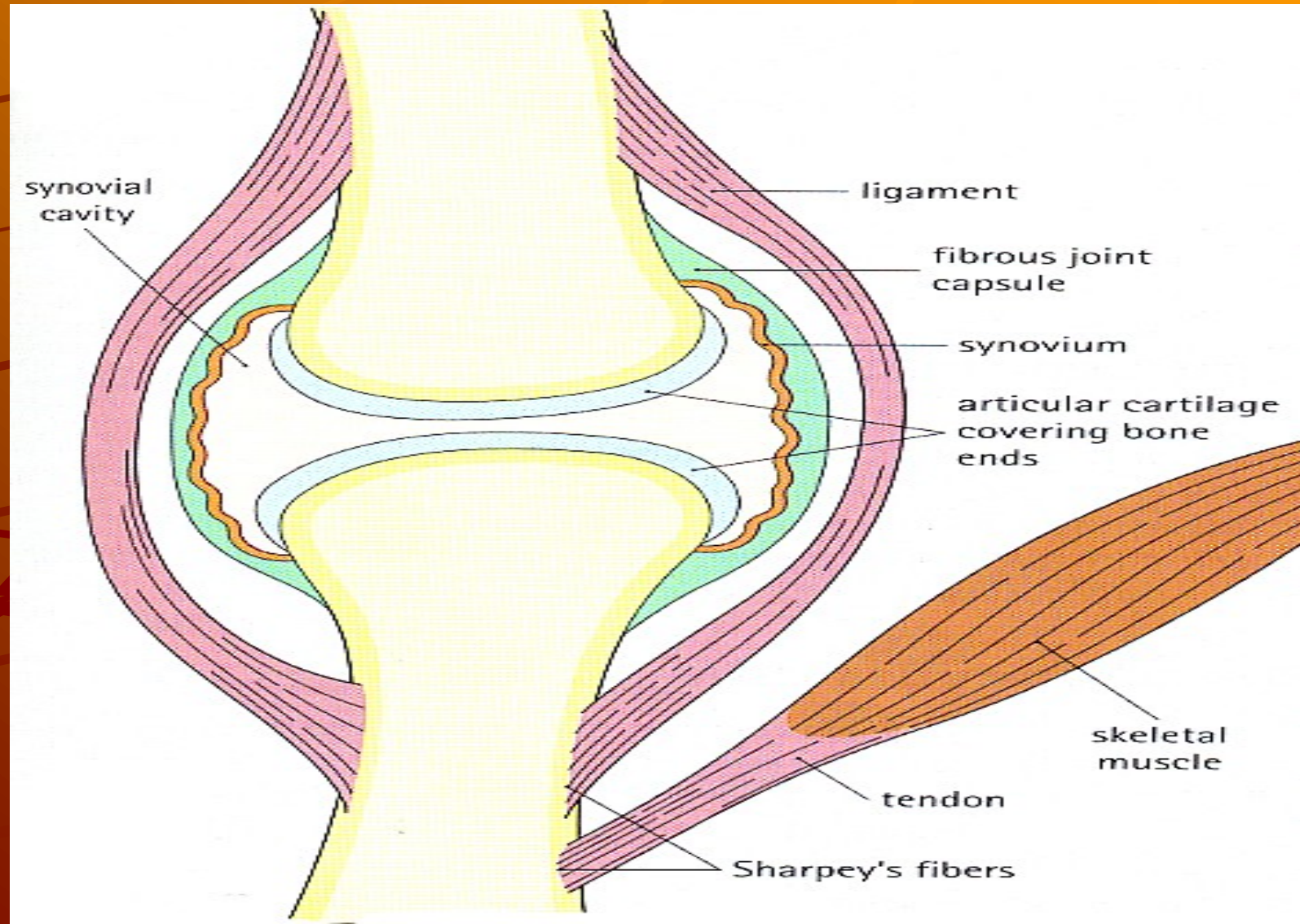
– **DIARTHROSES:** Movable joints

✚ **Articular Cartilage** made of hyaline cartilage, without perichondrium, covers the moving bone-ends.

✚ **Joint Capsule** is continuous with the periosteum.

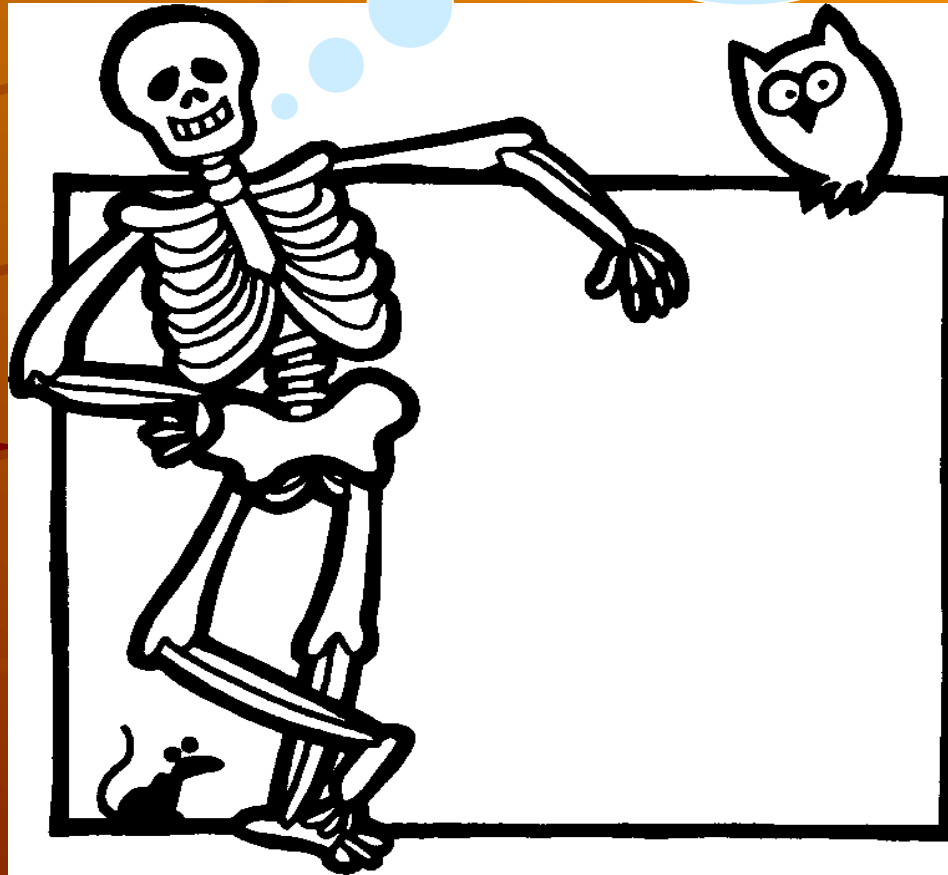
✚ **Synovial Membrane** lines the joint capsule. It secretes **synovial fluid** into the joint space.

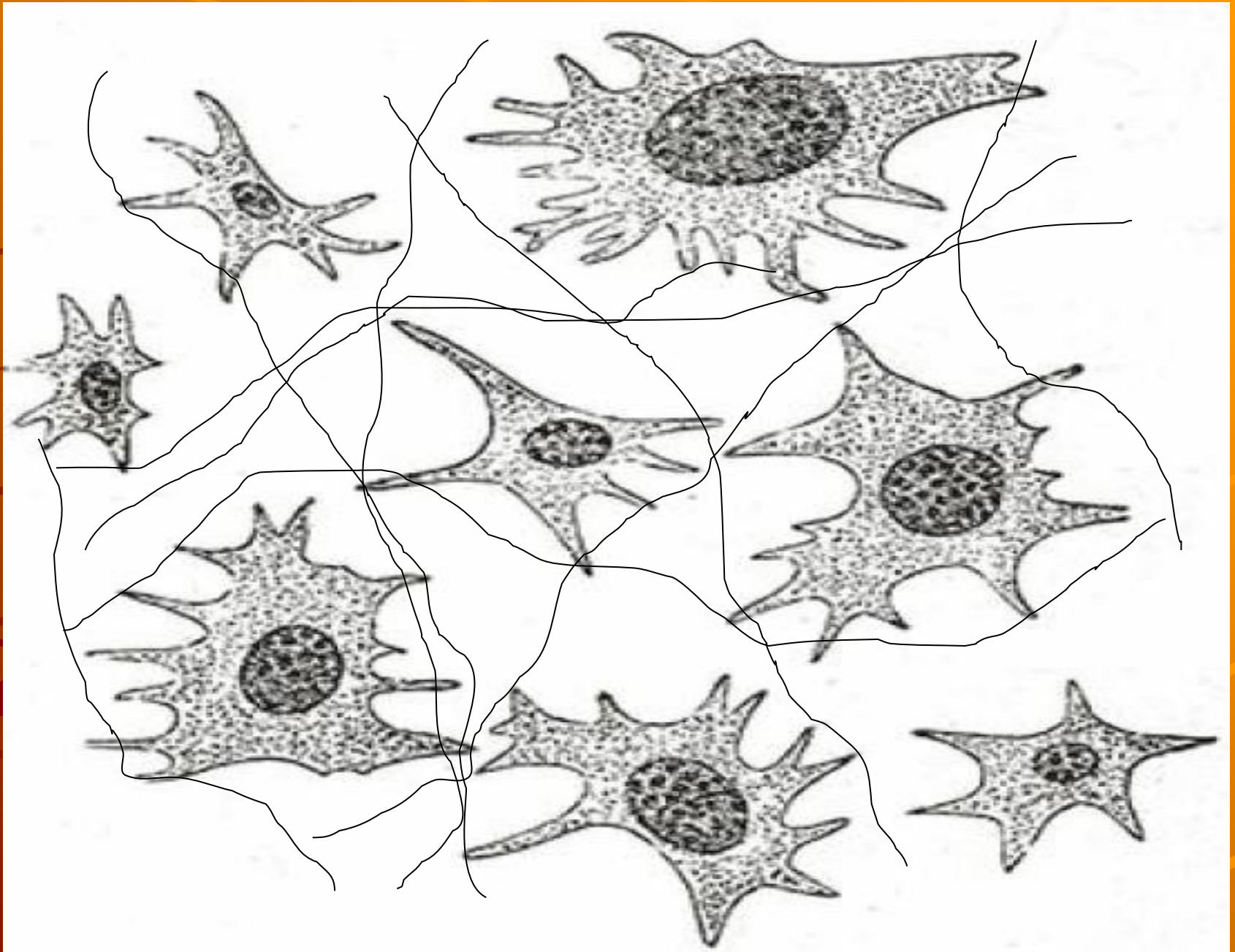
# Diarthrosis





***Thanks for  
attention***







A

B

C

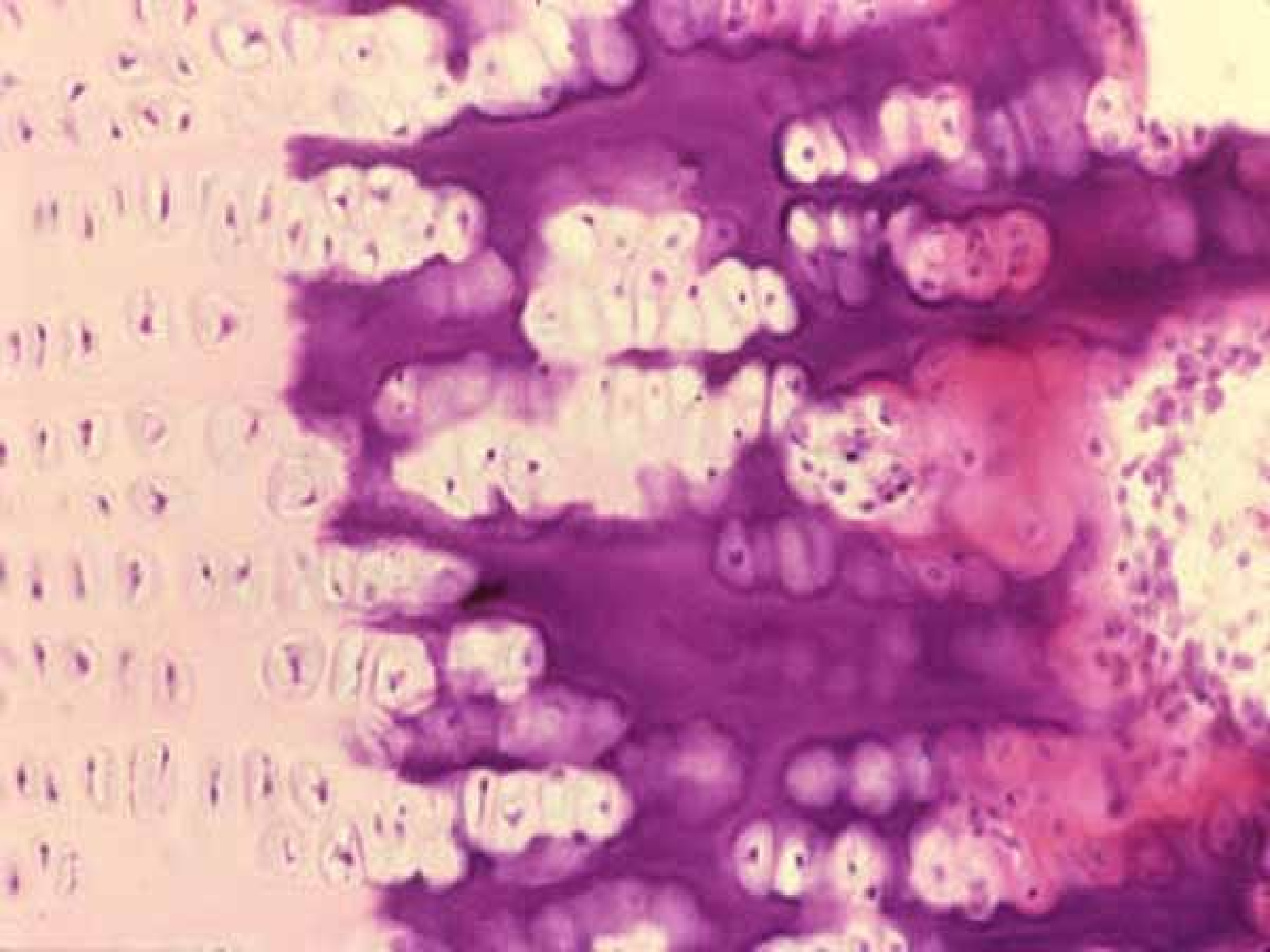
D

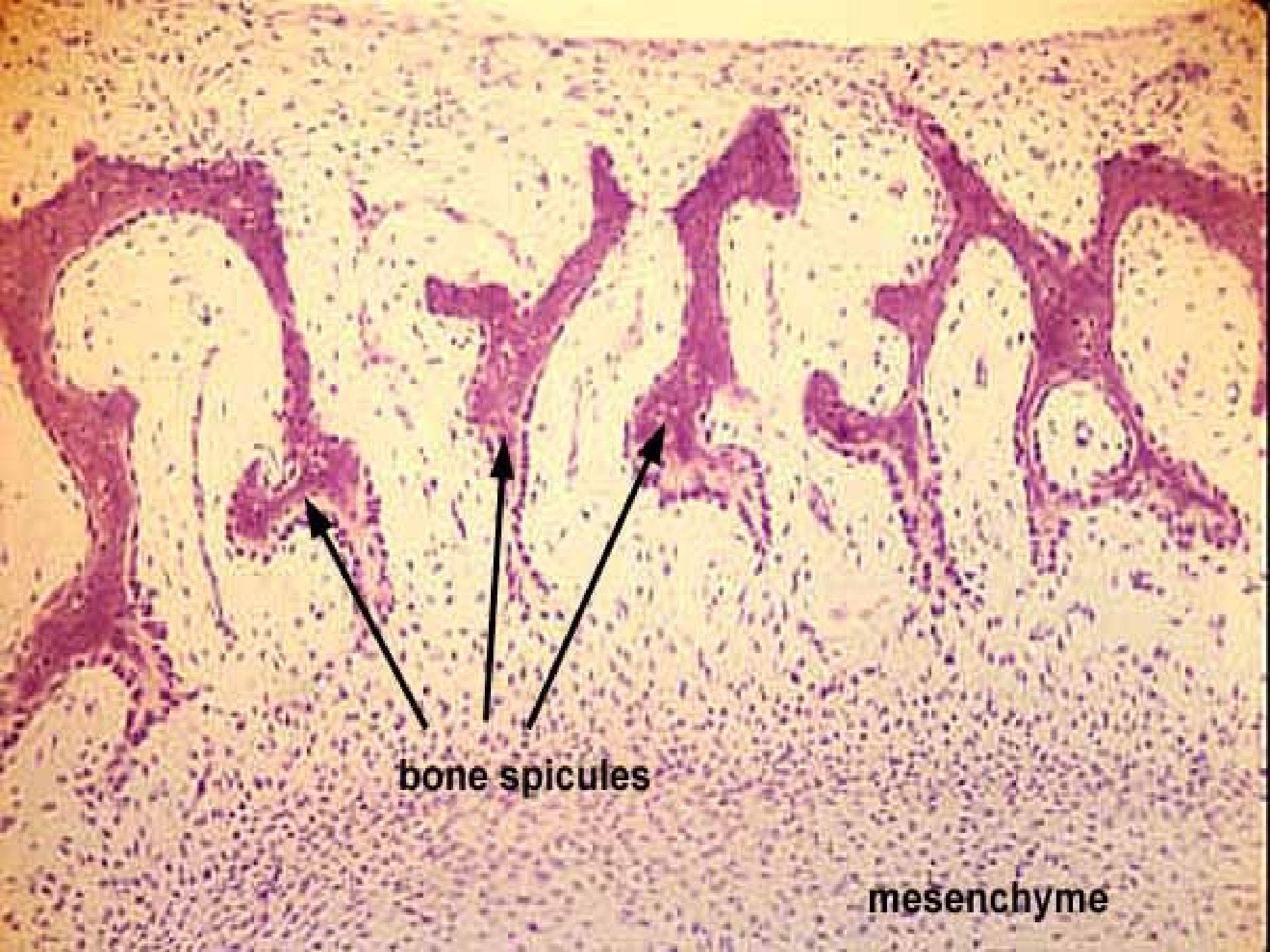
E

cartilage

bone

bone marrow





**bone spicules**

**mesenchyme**

# (Intra)membranózní osifikace



Mezenchym + vaskularizace: **mezenchymocyty** ⇒

**osteoprogenitorní bb** ⇒

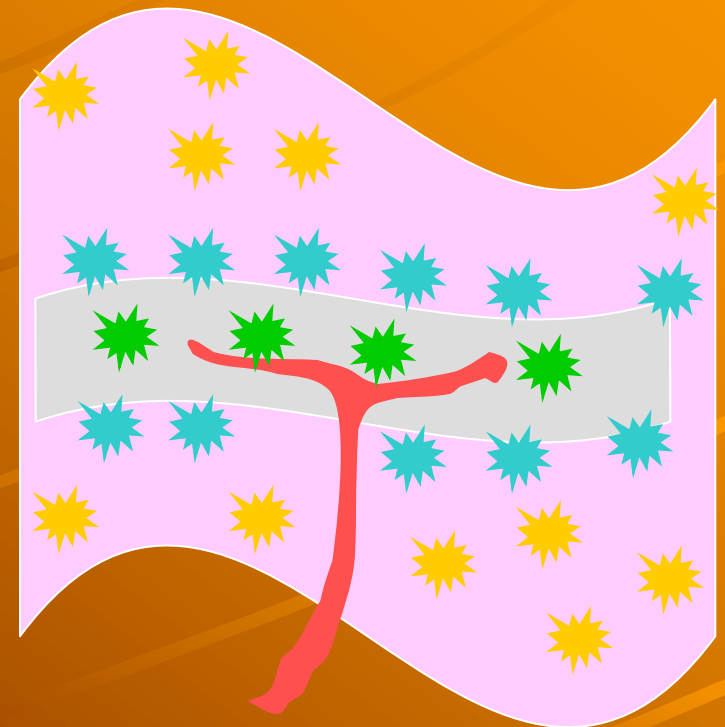
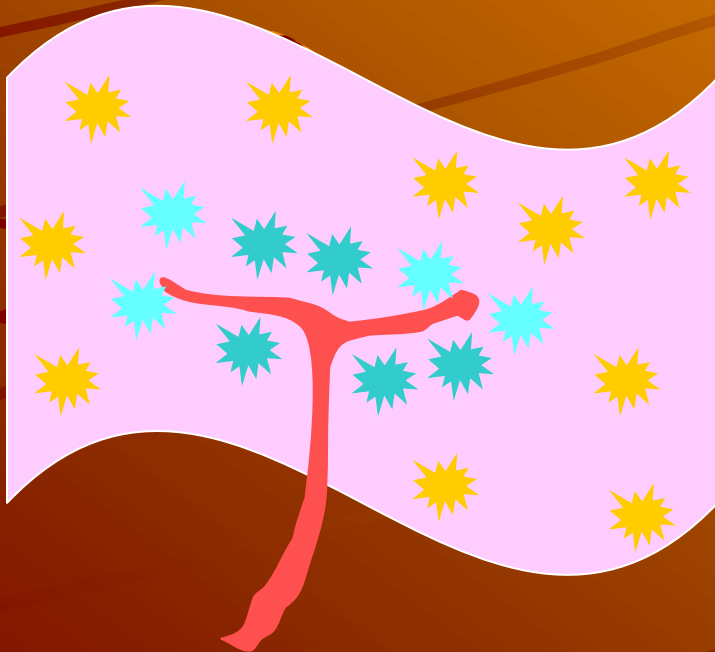
**osteoblasty** :

produkce

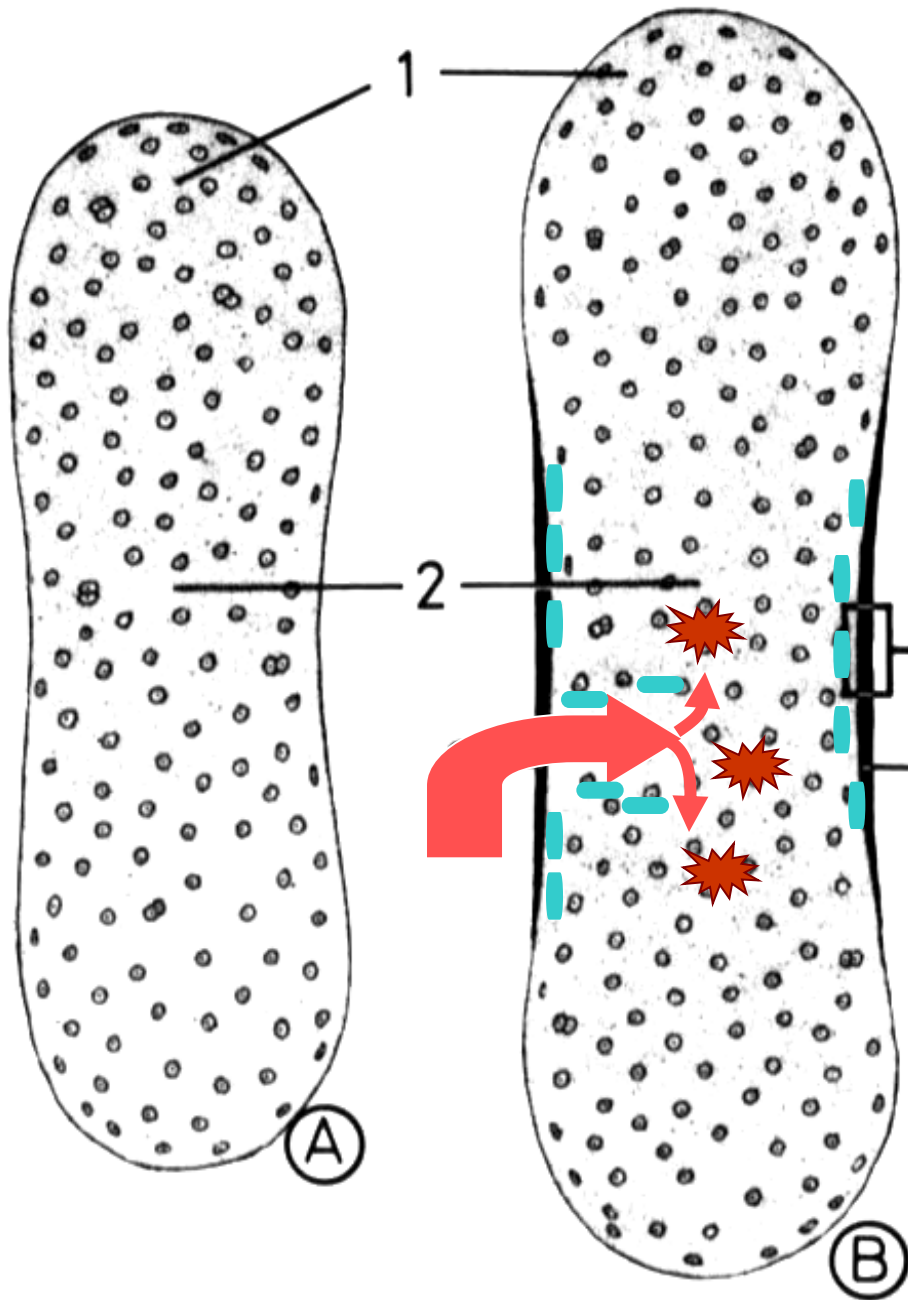
**osteoidu**

Osteoblasty + osteoid ⇔ mineralizace: osteoblasty ⇒



**osteocyty**

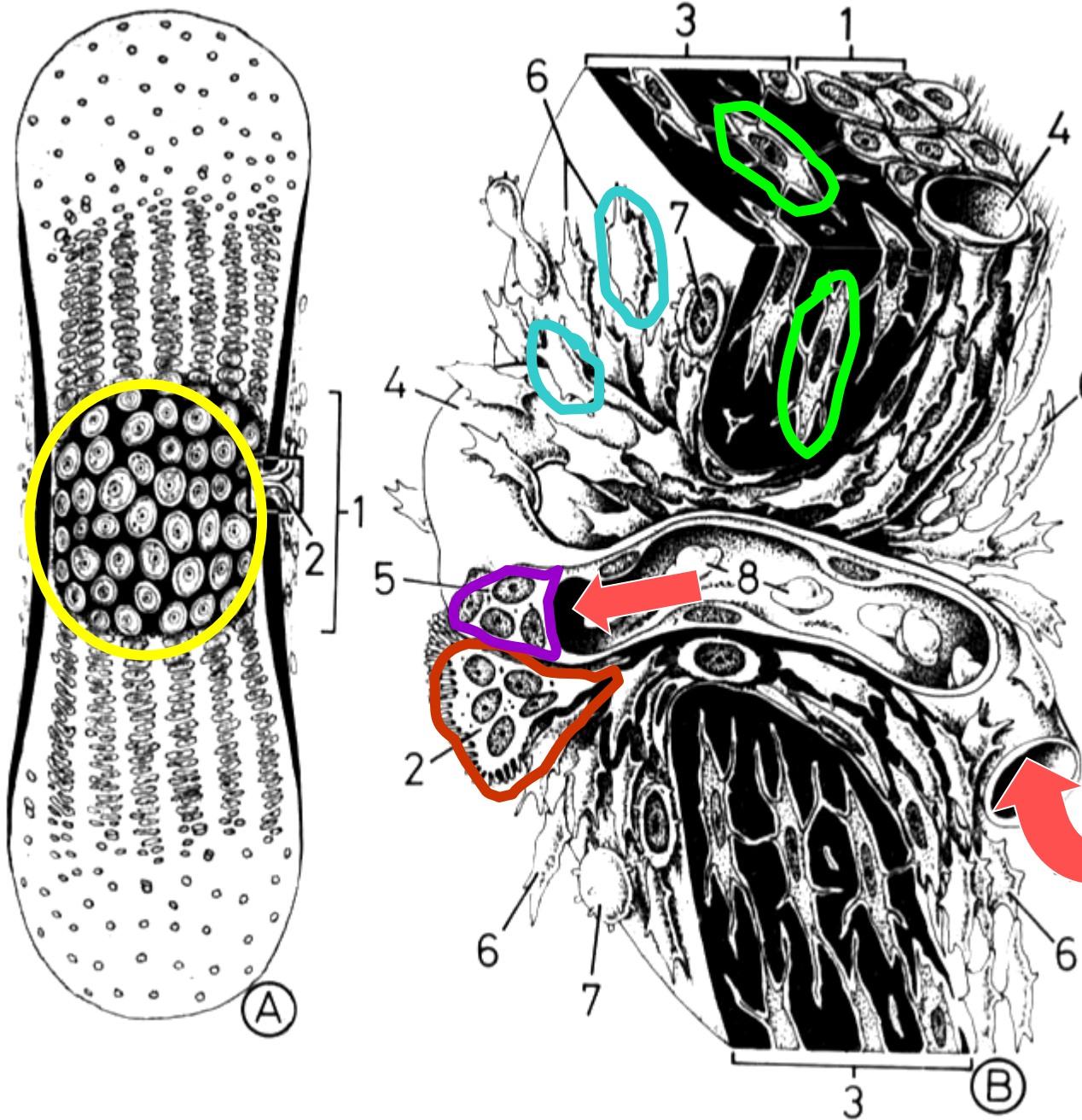


# Enchondrální osifikace (chondrogenní)



A: chrupavčitý model kosti:  
1- epifýza, 2, střed diafýzy

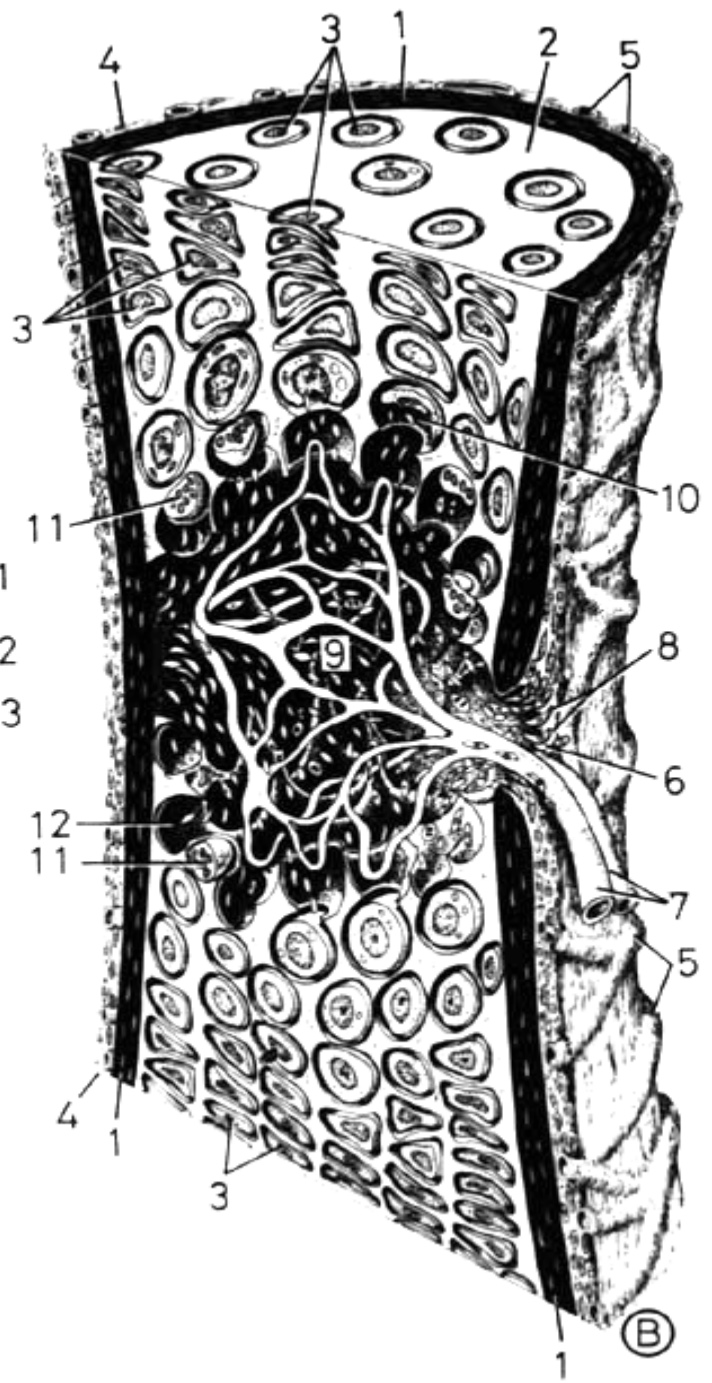
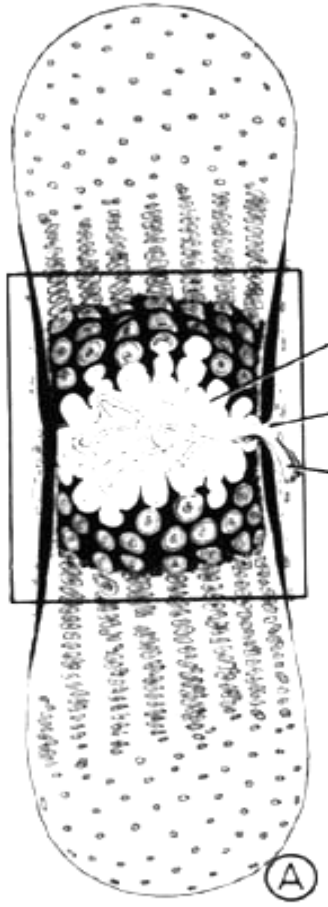
B: periostální manžeta  
(z perichondria)  
invaze krevních cév s osteoklasty  
(na čele ) a osteoprogenitorní-  
mi bb. (podél cév )

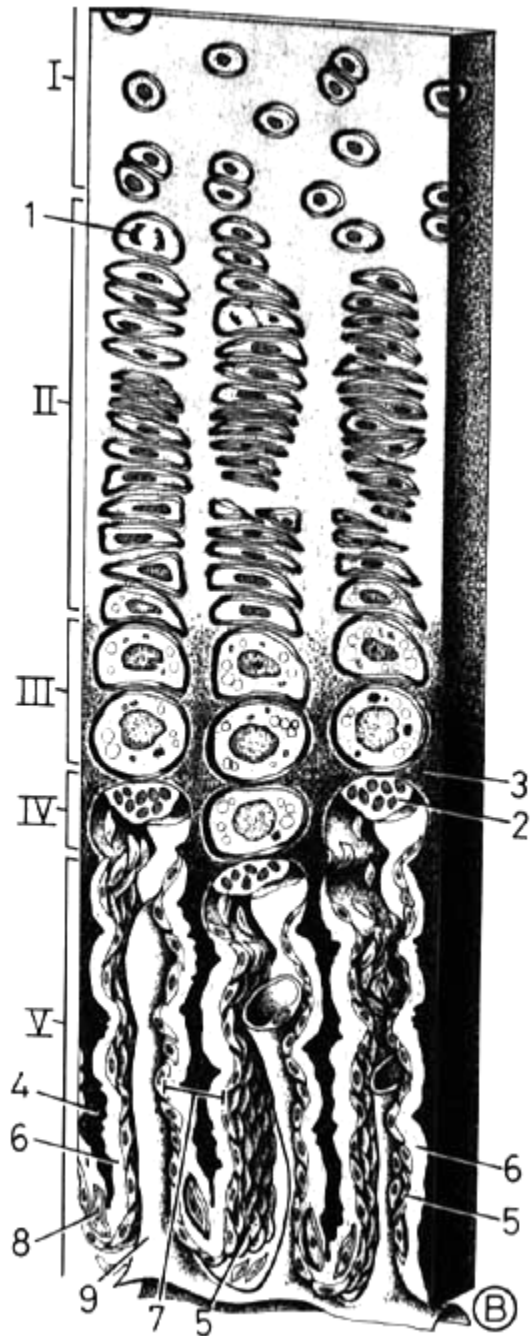
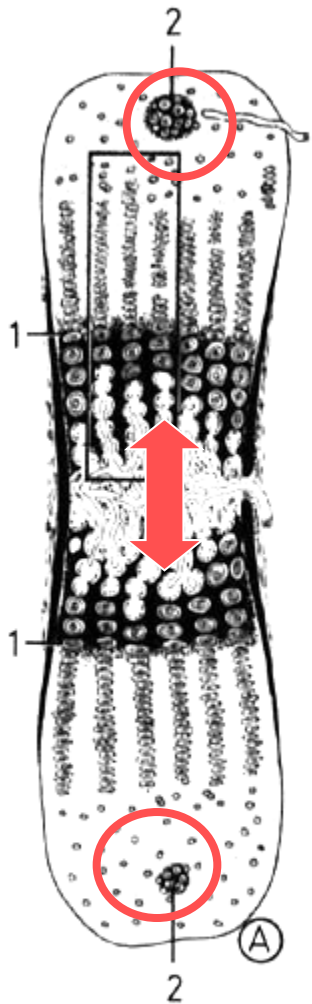


A: hypertrofie a  
kalcifikace  
chrupavky

B: detail (A)  
1 - periost  
2 - **osteoklasty**  
3 - **osteocyty**  
v kostěné manžetě  
pod periostem  
5 - **monocyty**  
6 - **osteoprogen. bb.**







Zóna normál. hyalinní chrupavky

Zóna proliferující chrupavky

Zóna hypertrofické chrupavky

Zóna kalcifikující chrupavky

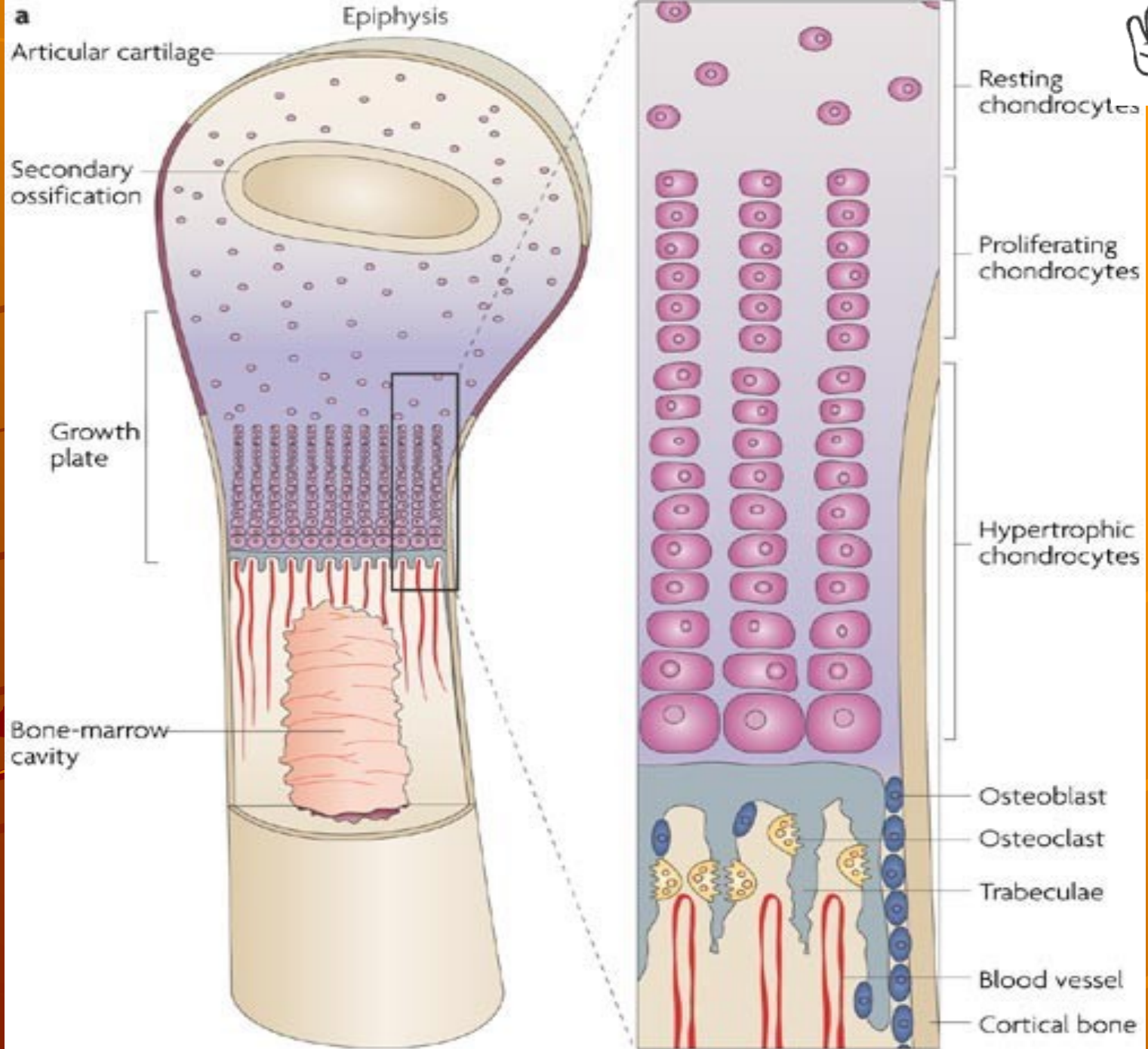
2 – osteoklasty

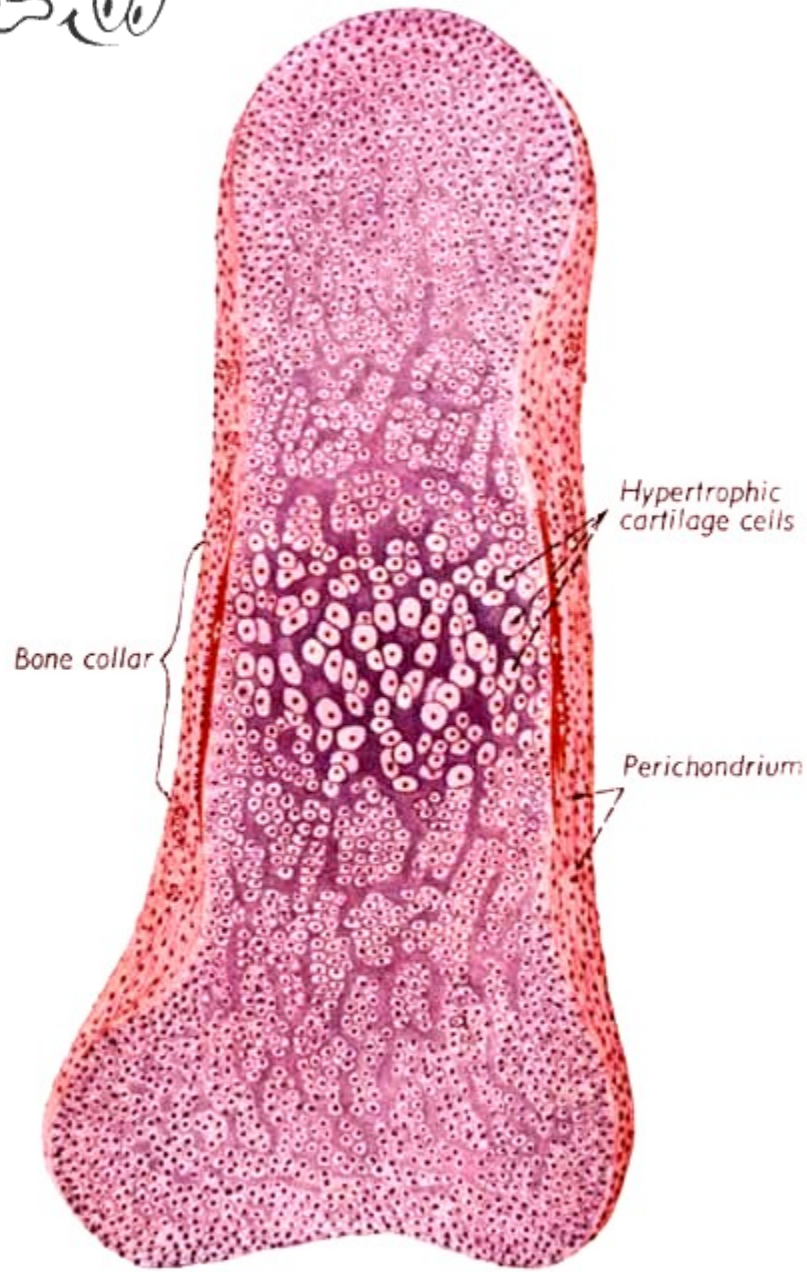
4 – primární kost s osteocyty

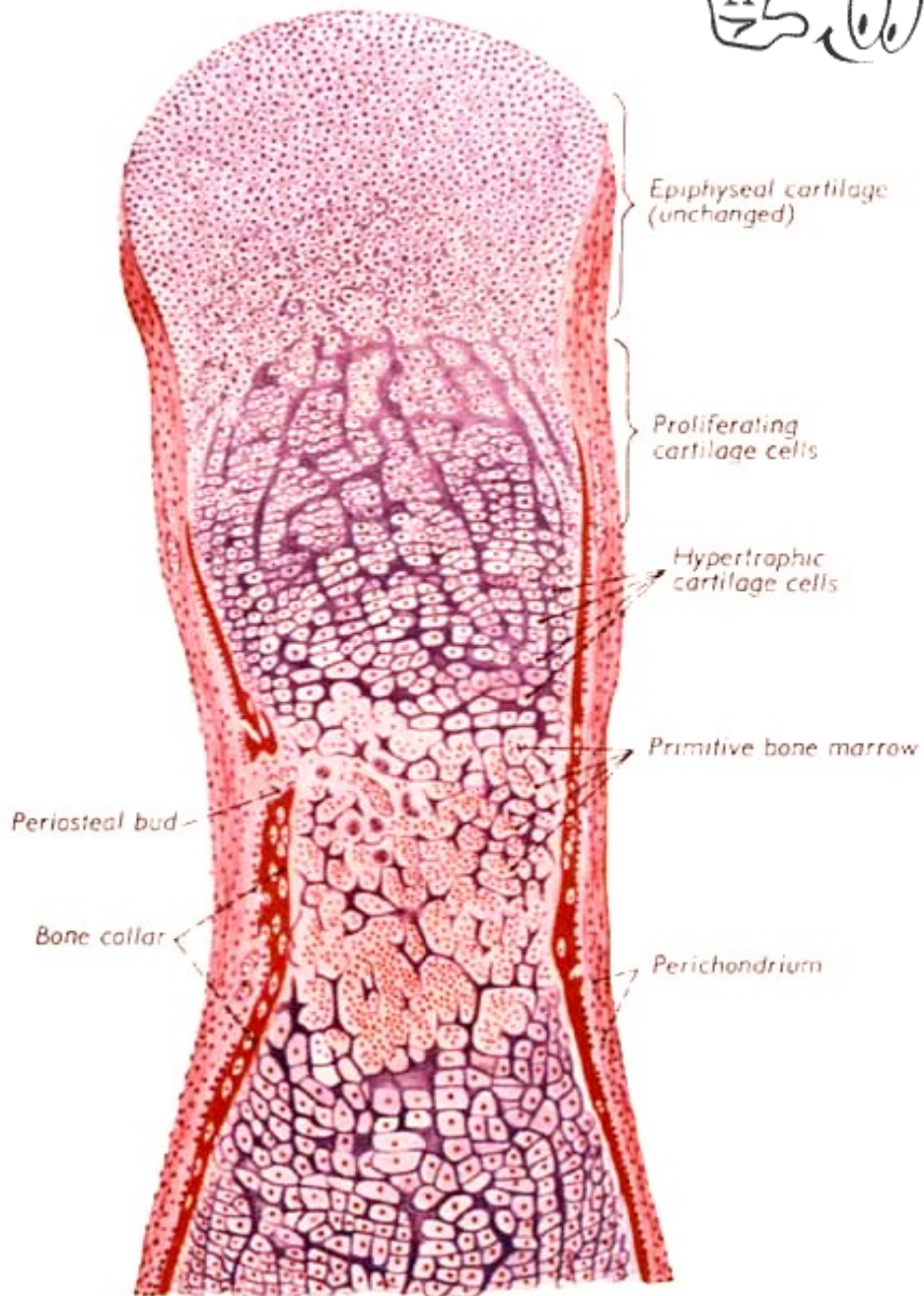
5 – osteoblasty

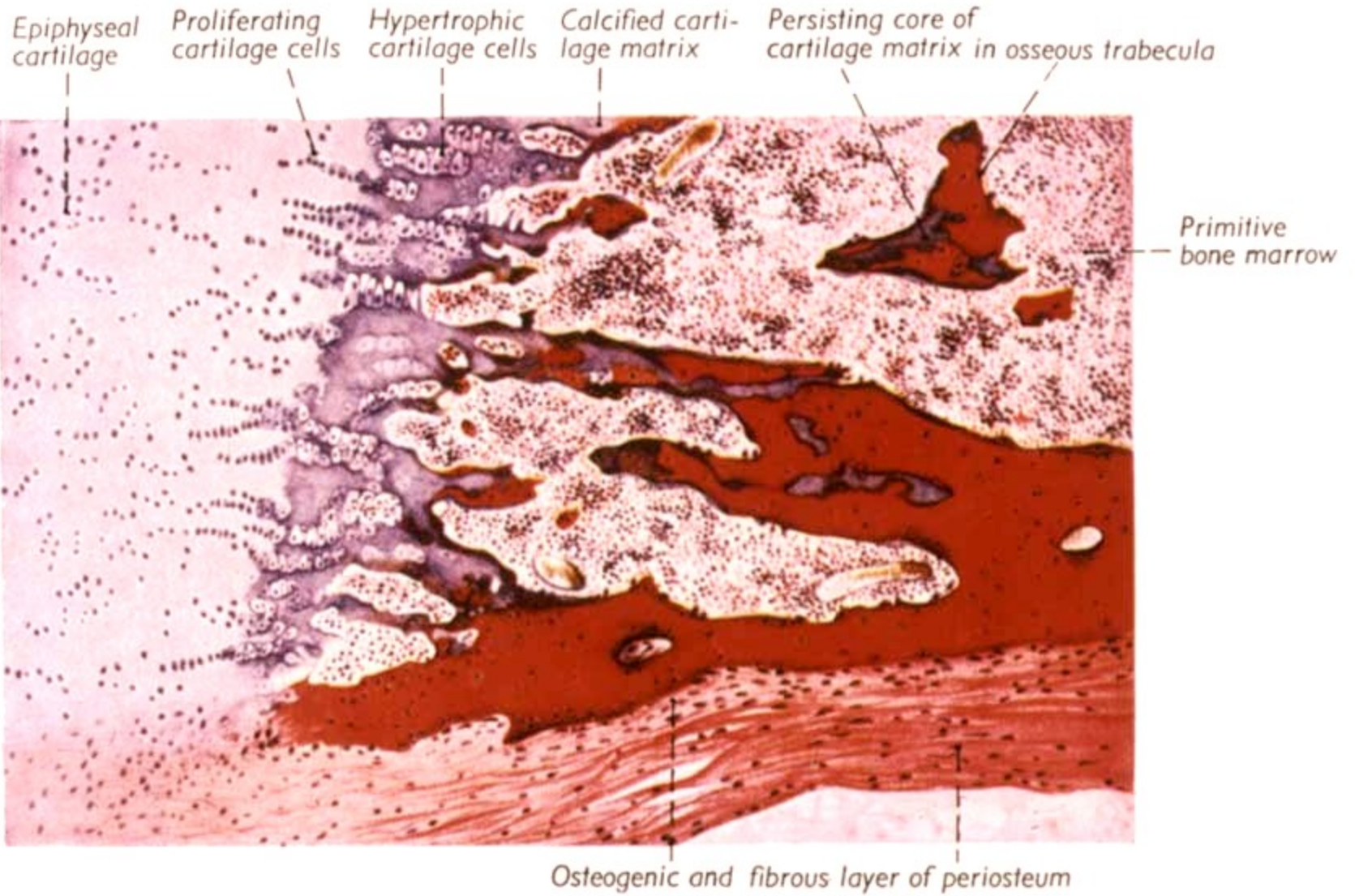
6 – osteoid (pod osteoblasty)

9 – kapiláry









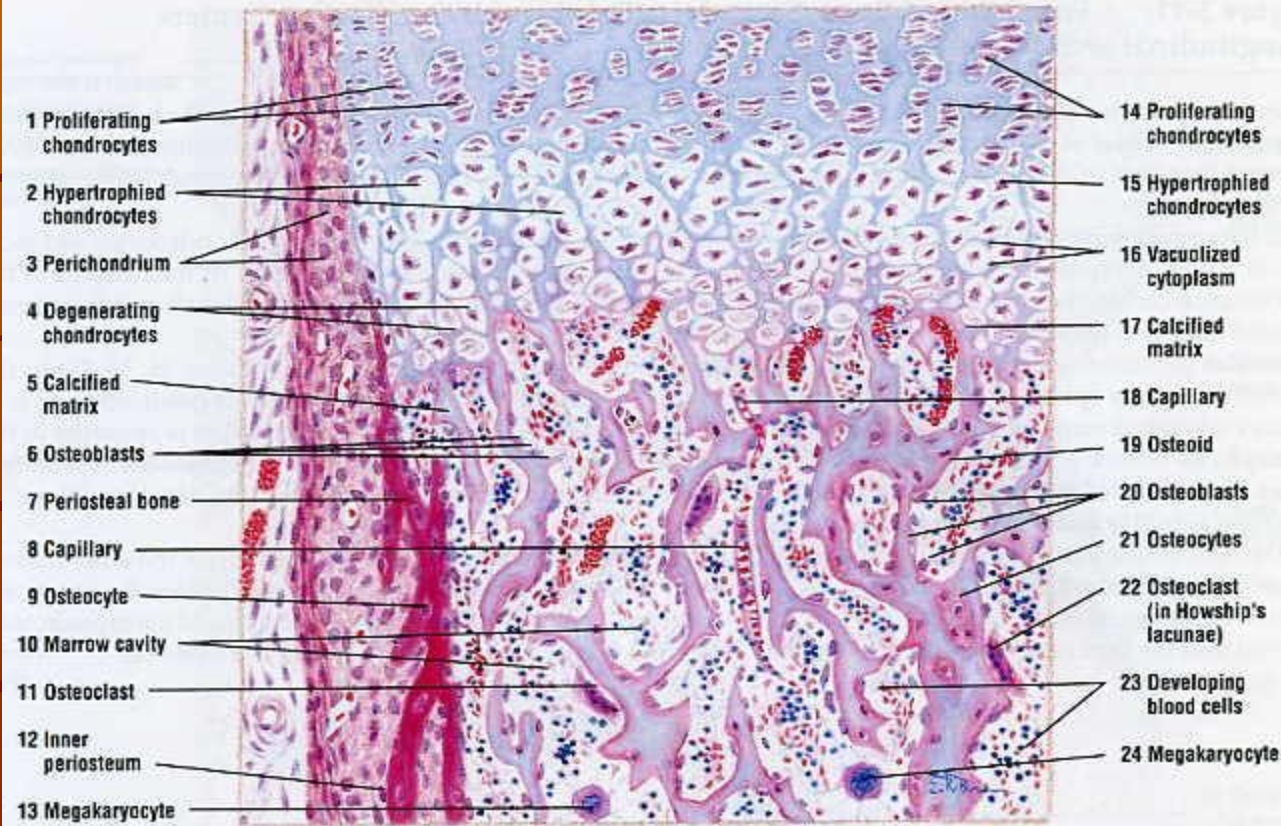
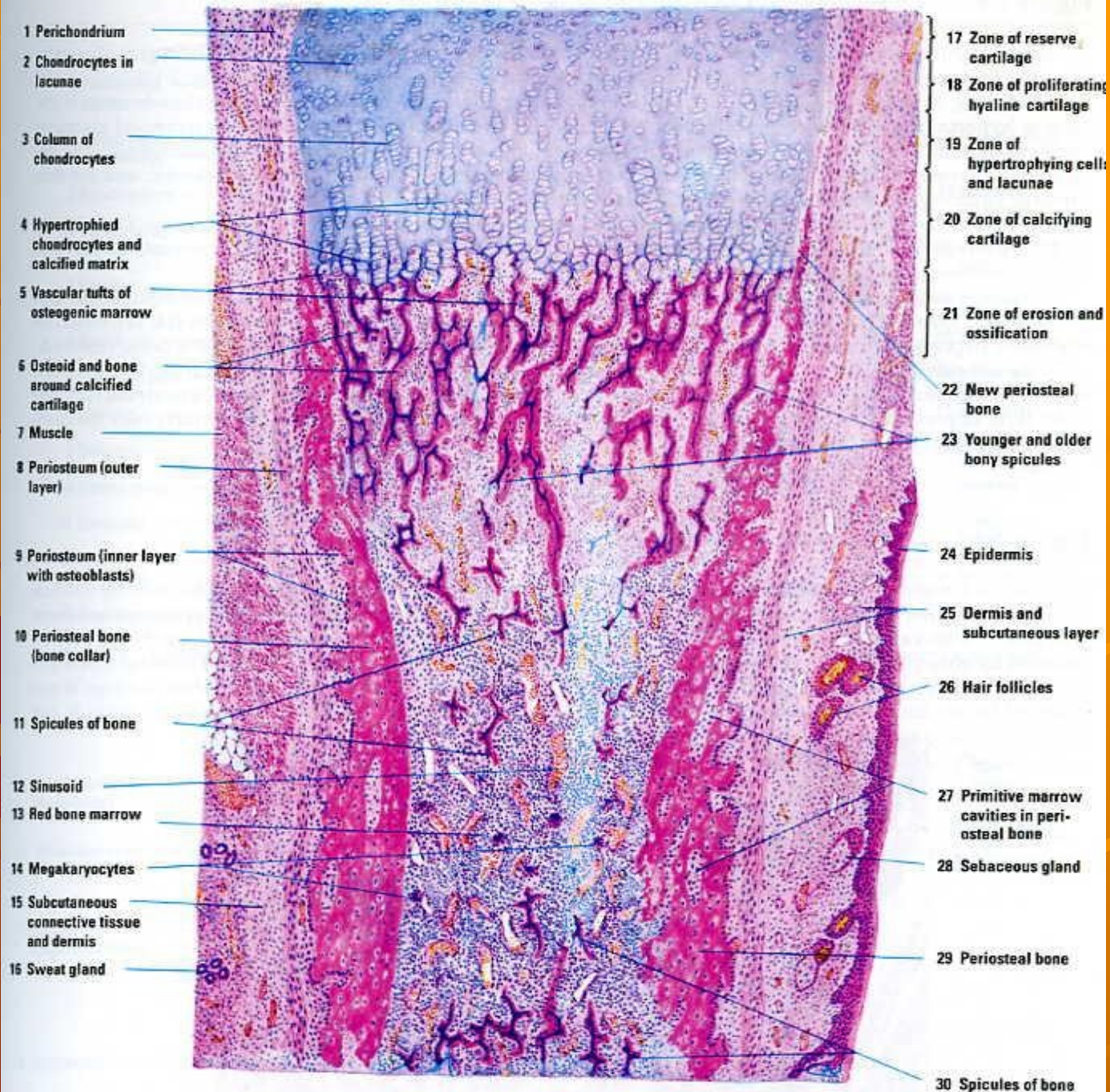


Fig. 3-9 Endochondral Ossification: Zone of Ossification. Stain: hematoxylin-eosin. Medium magnification.

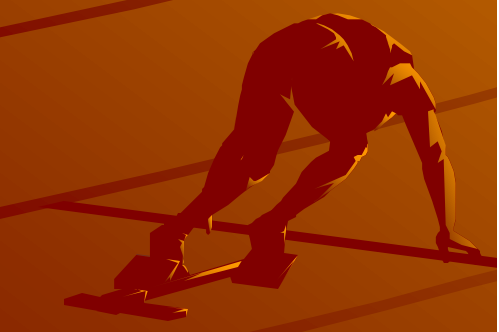
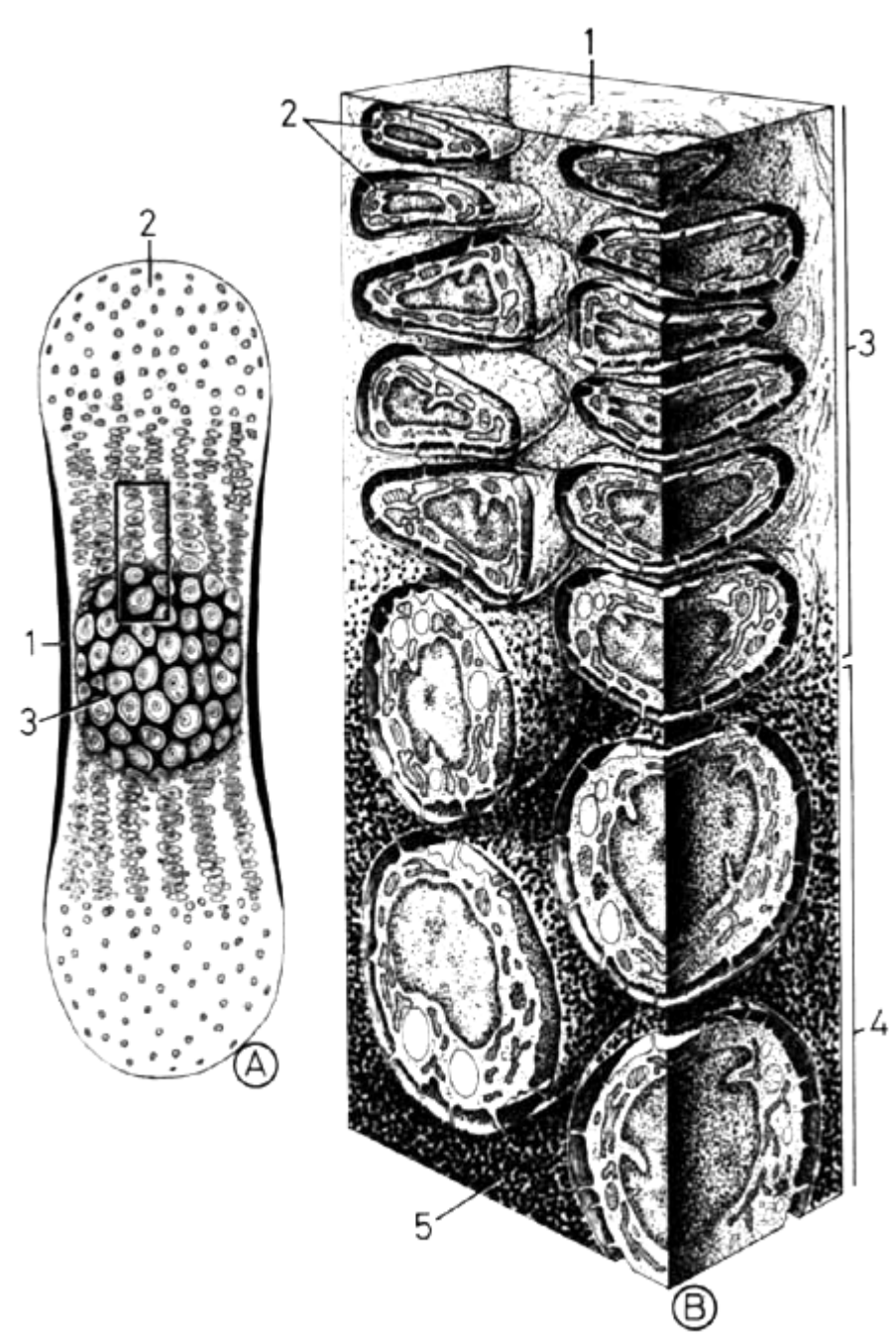
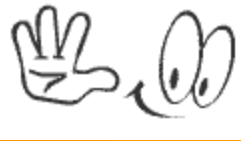
الواقع والحياة





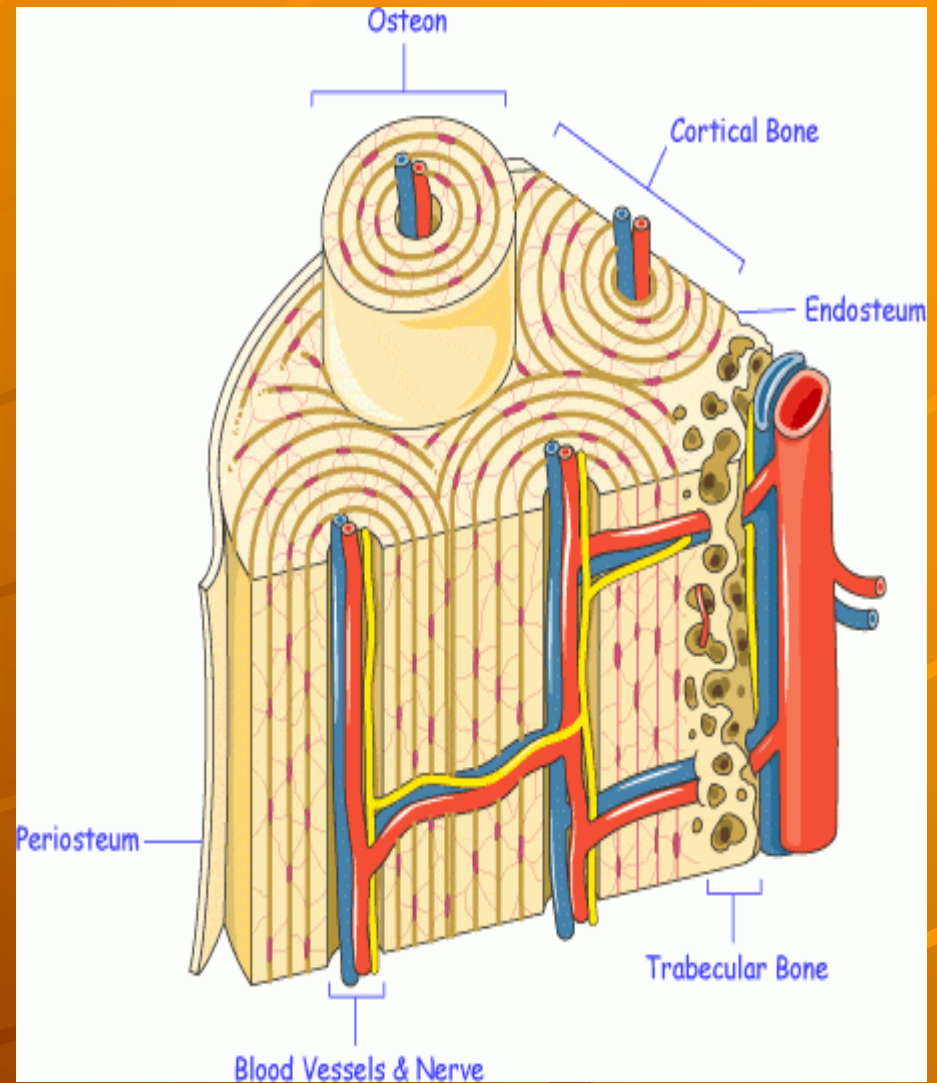
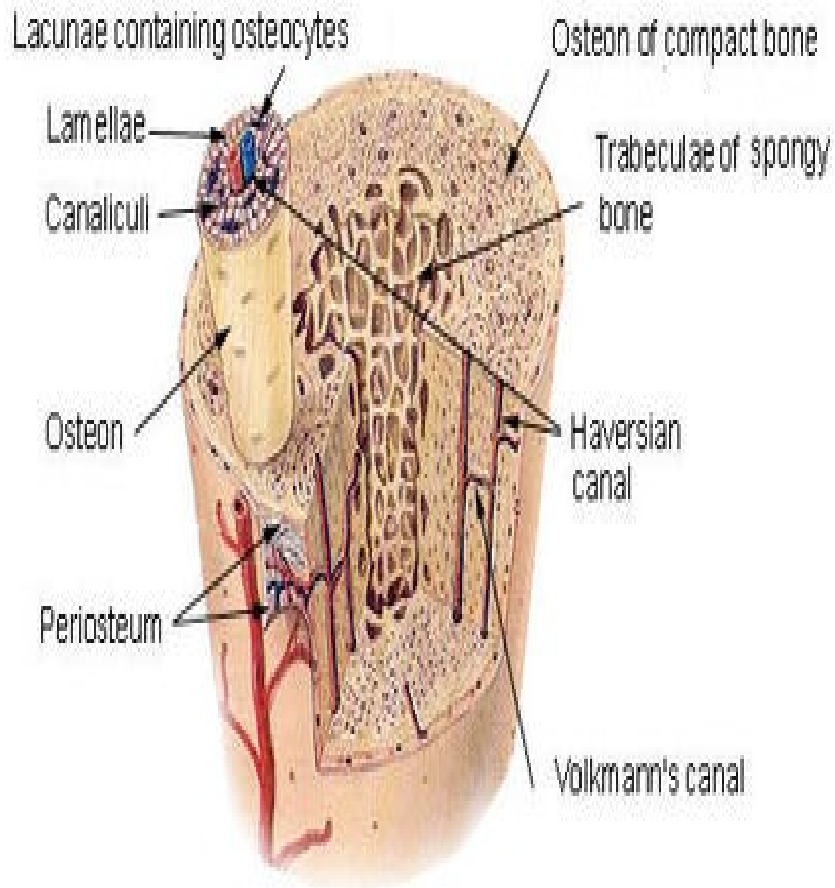
**Fig. 3-8 Endochondral Ossification: Developing Long Bone (panoramic view, longitudinal section).** Stain: hematoxylin-eosin. Low magnification.

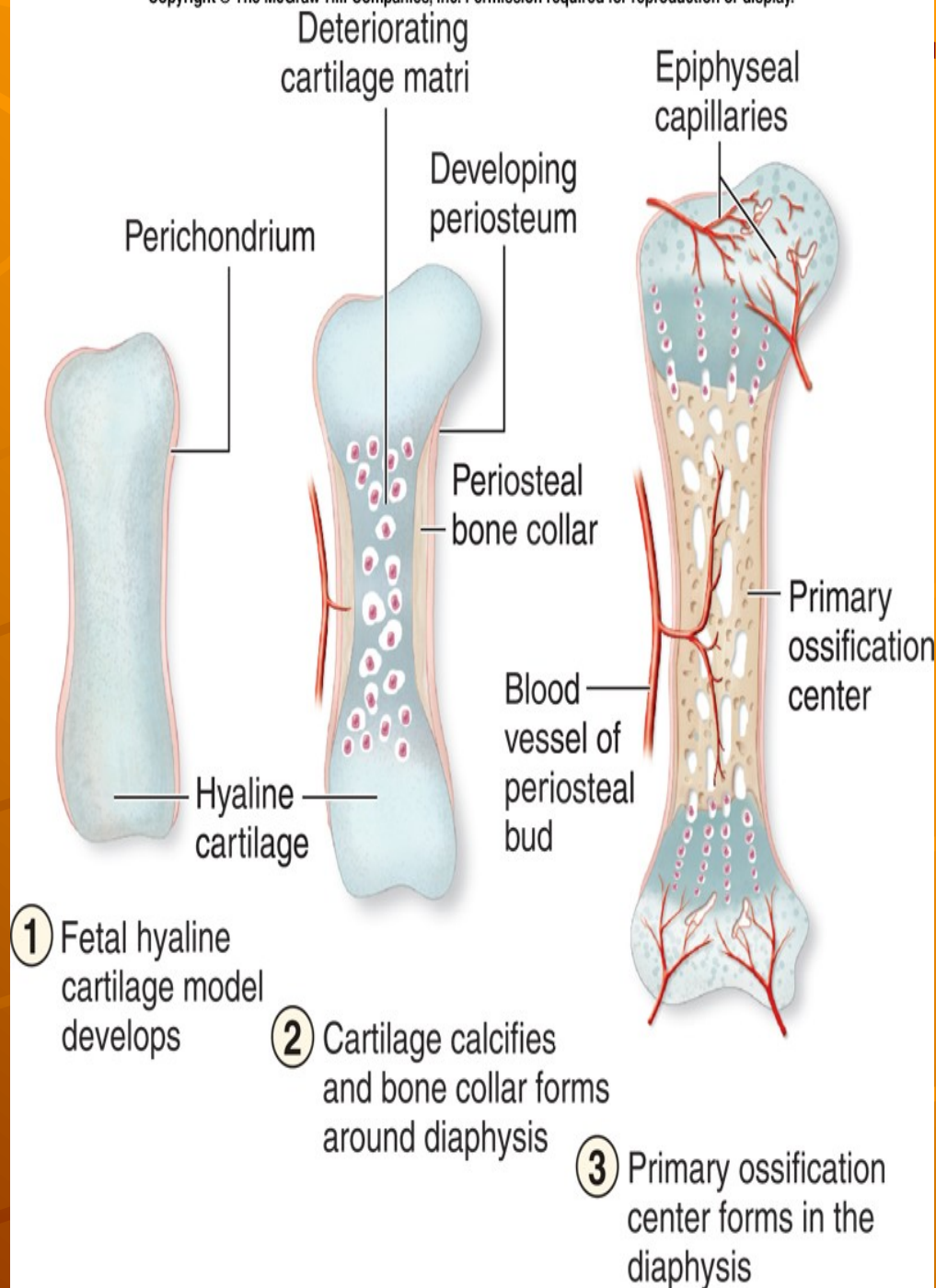
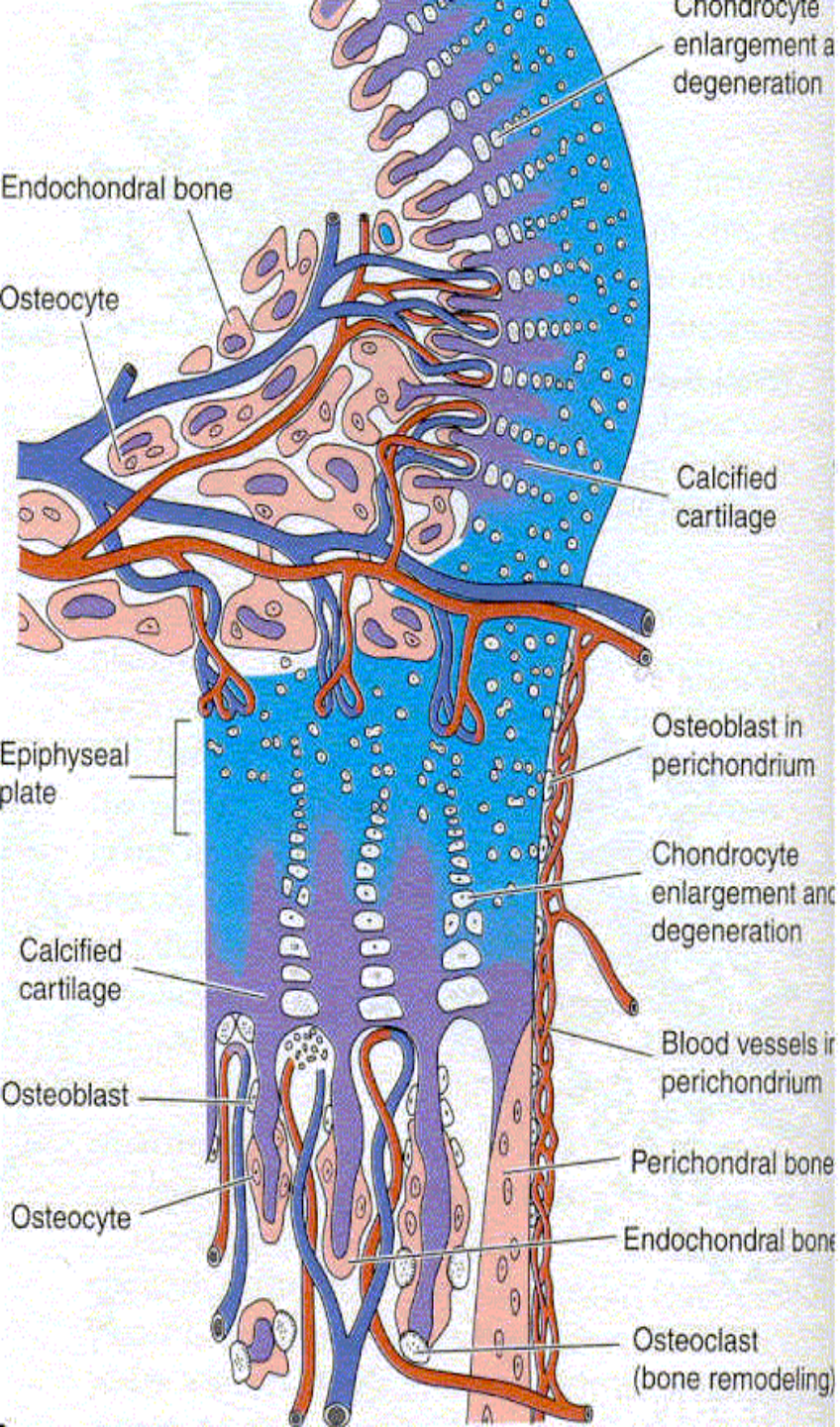


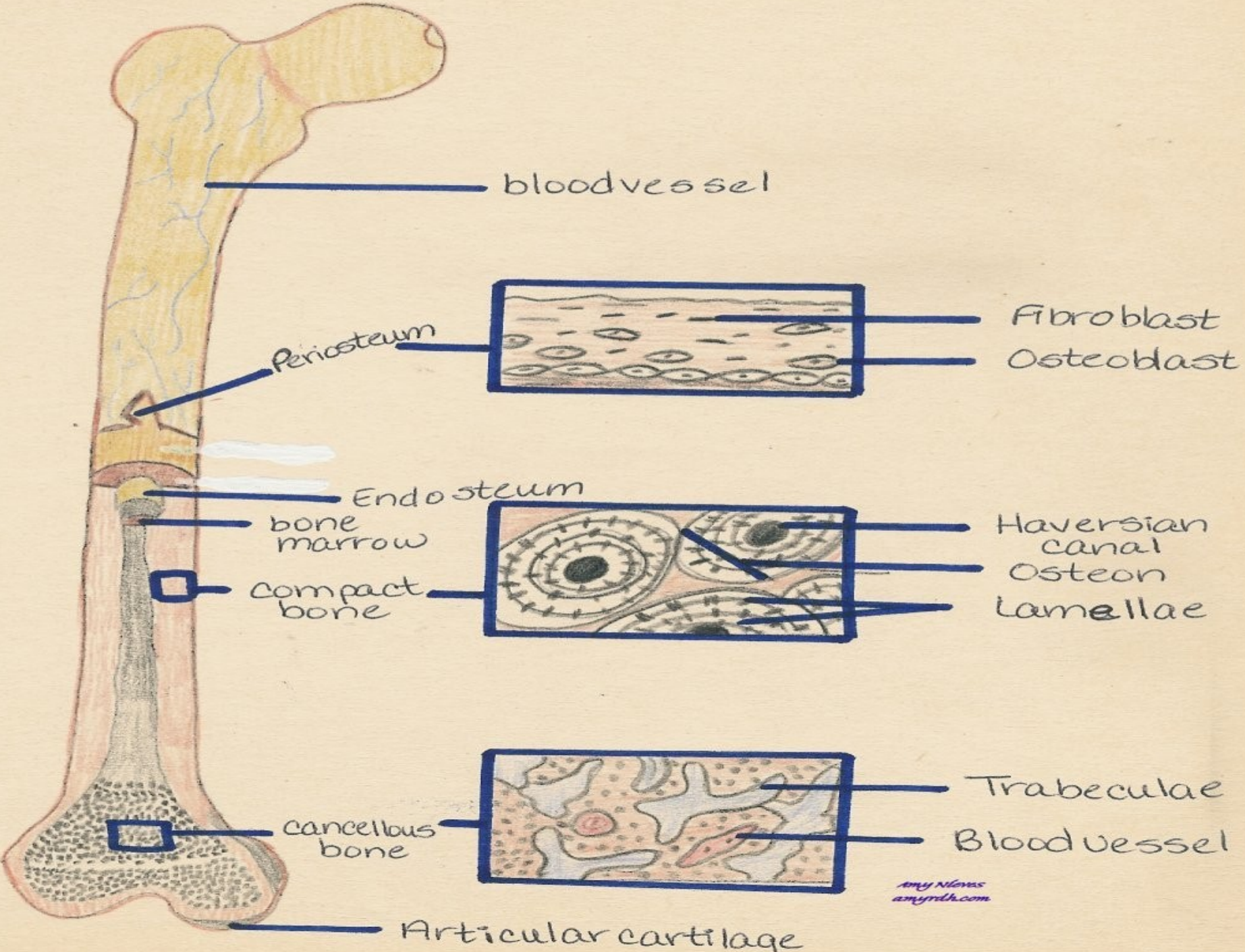


# Lamellar bone

## Compact Bone & Spongy (Cancellous Bone)







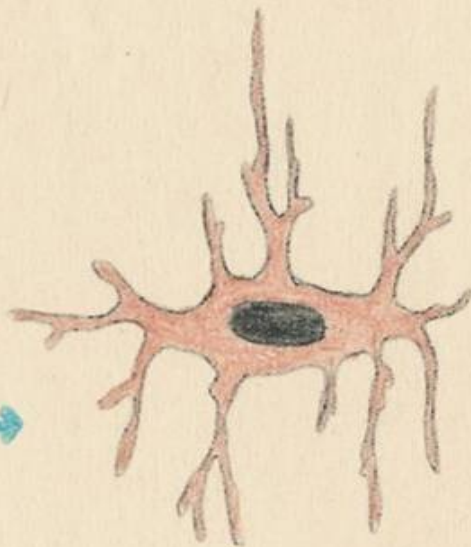
# Types of Cells in Bone Tissue



Osteoprogenitor cell  
- cell produces bone cell.



Osteoblast  
- cell forms bone matrix.

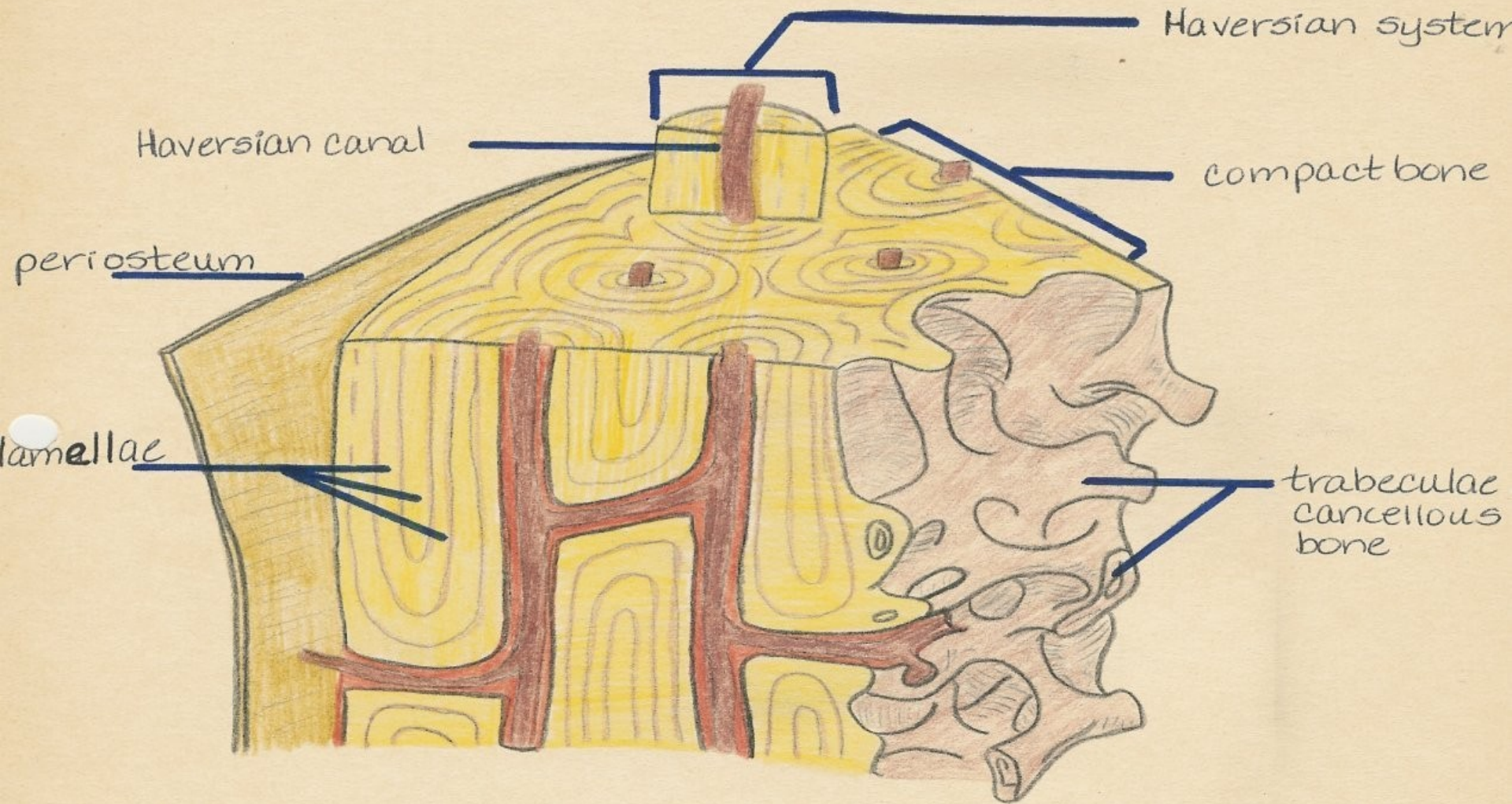


Osteocyte  
- mature bone cell.

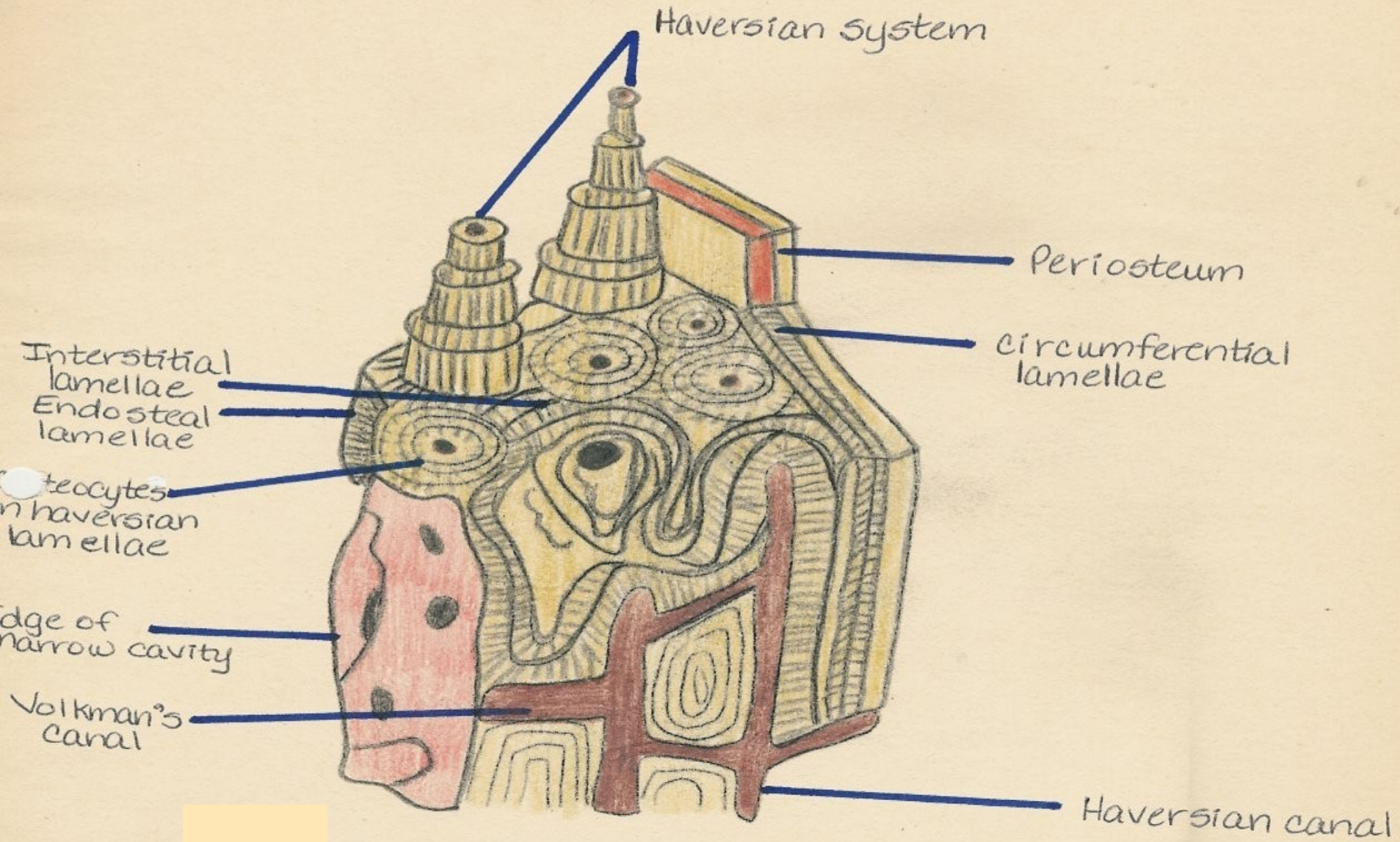


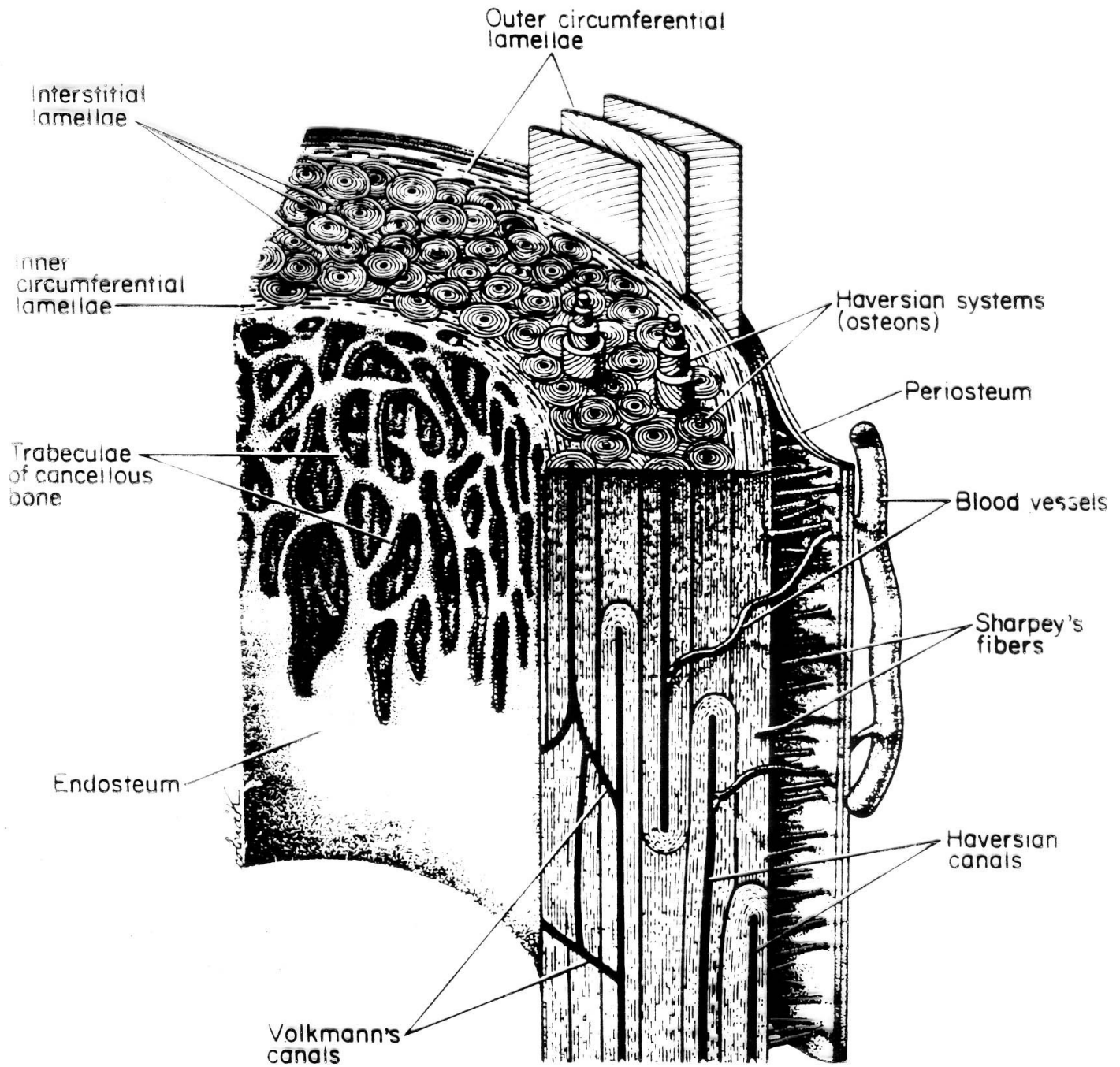
Osteoclast  
- cell dissolves bone.

# Compact Bone



# Bone





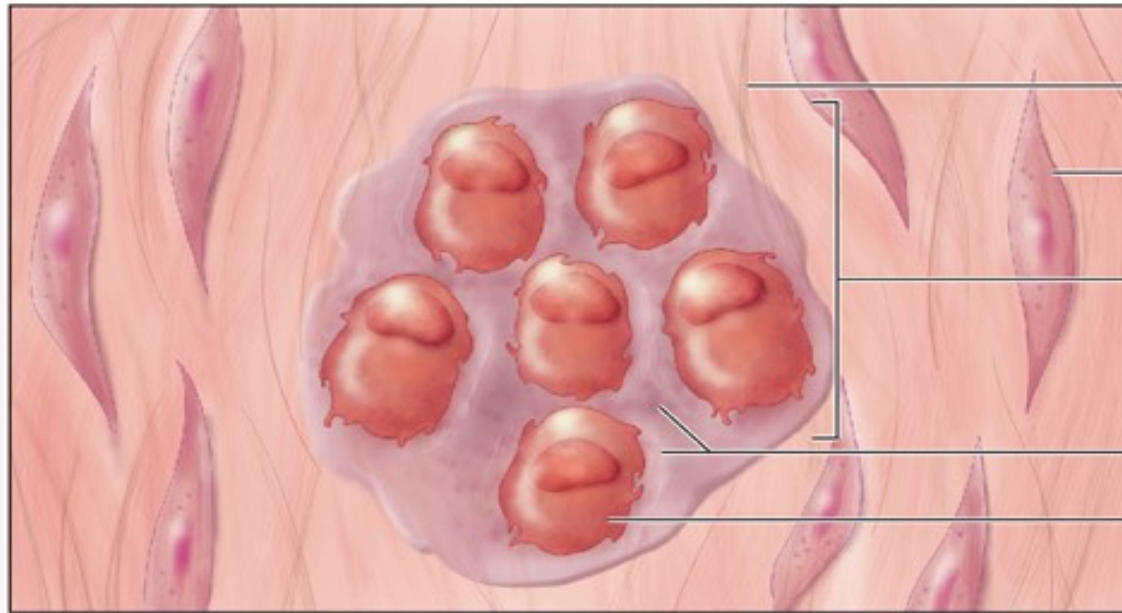


# Ossification – bone formation

- PERIOSTEUM contains **blood vessels** and **osteoprogenitor cells**  osteogenic potential
- Osteoprogenitor cells  **osteoblasts** (production of intercellular matrix **OSTEOID**)
- Osteoid + minerals (**from blood**) = ossein
- Osteoblasts entrapped in ossein  **osteocytes**,  
osteocytes + ossein = BONE
- The result of **primary ossification** is woven bone
- The result of **secondary ossification** is compact bone (osteoclasts are involved in destruction of woven bone)

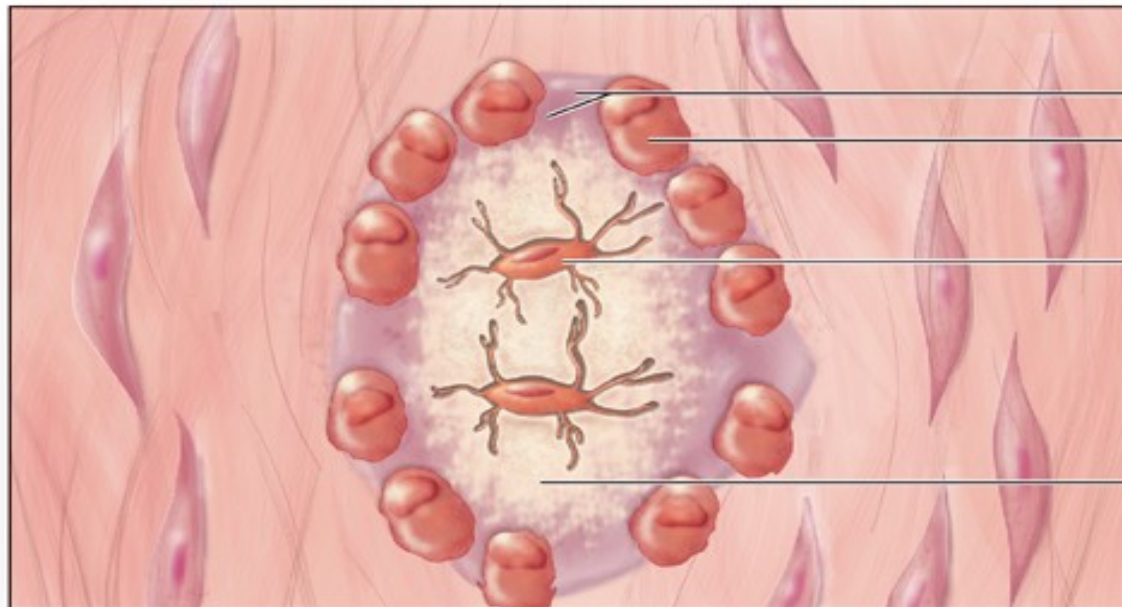
### Intramembranous Ossification

- ① Ossification centers form within thickened regions of mesenchyme



Collagen fiber  
Mesenchymal cell  
Ossification center  
Osteoid  
Osteoblast

- ② Bone matrix (osteoid) undergoes calcification.



Osteoid  
Osteoblast  
Osteocyte  
Newly calcified bone matrix