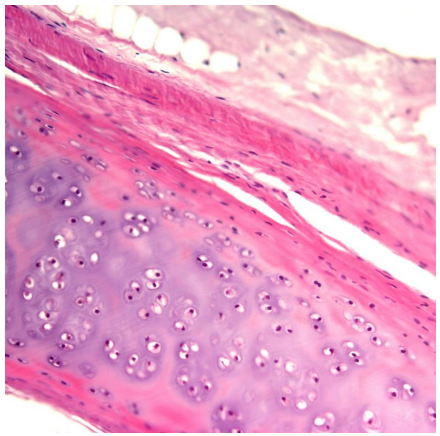
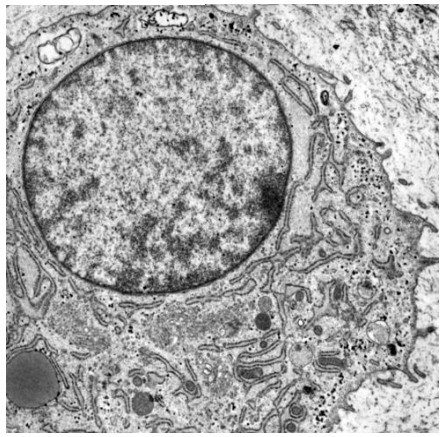
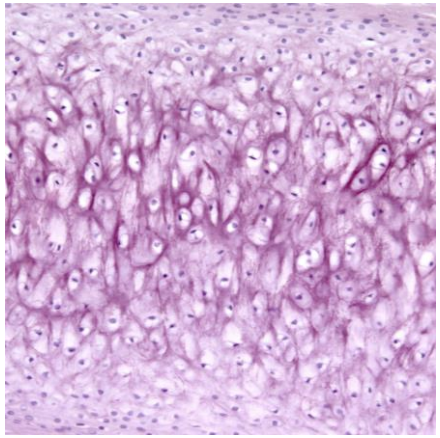
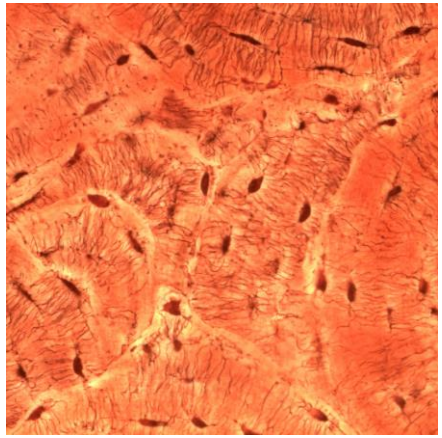
MUNI MED

Department of Histology and Embryology









CARTILAGE AND BONE

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Faculty of Medicine MU

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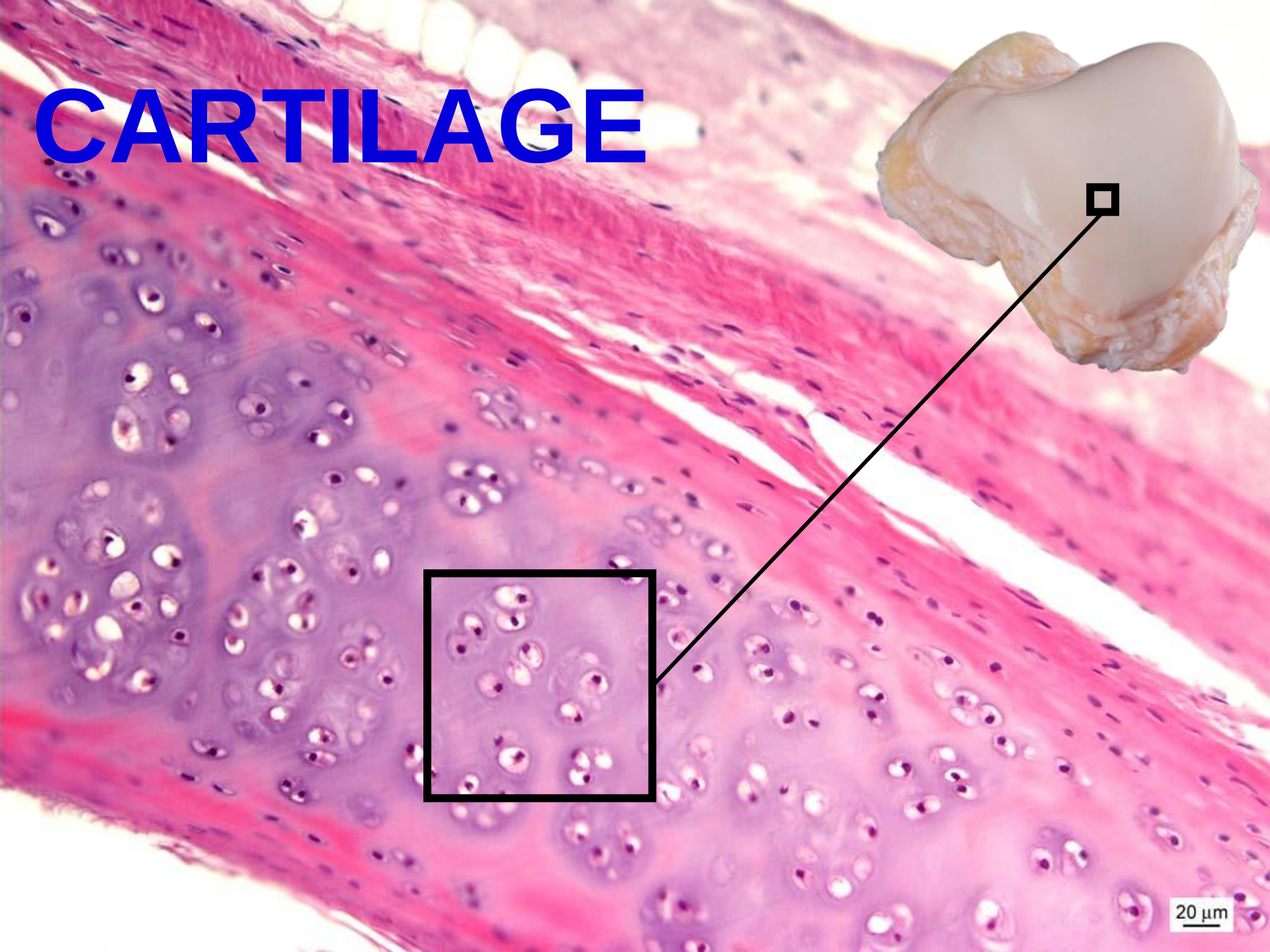
CONNECTIVE TISSUE

Consistency	Cells	Fibers	Amorphous ground substance
Soft	↑	↓	↑
Rigid	↓	↑	↓

CARTILAGE



CARTILAGE

General features:

- **specialized connective tissue** with continuous ECM
- flexible, mechanically resistant
- avascular, non-innervated
- support of soft tissues - trachea, larynx
- skeletal support - costal cartilages
- diarthrosis - joints
- bone growth

CARTILAGE – COMPOSITION AND STRUCTURE

- Perichondrium – connective tissue around cartilage (except joints)

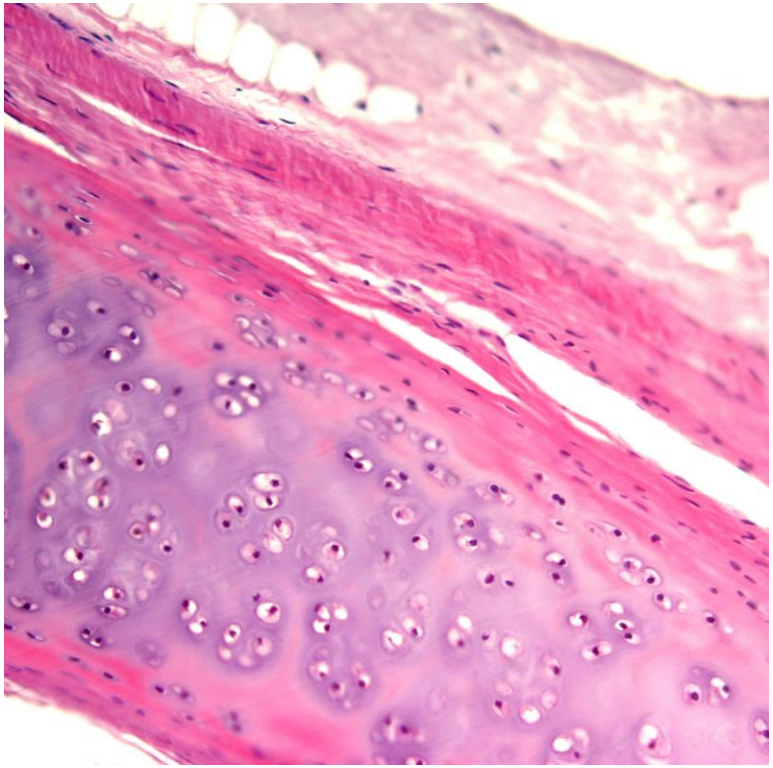
Nutrition
Growth

- Extracellular matrix – water, proteoglycans and collagen fibrils

Solid consistence
Pressure elasticity

- Cells of cartilage - chondroblasts, chondrocytes

Growth
ECM production



DISTRIBUTION

Hyaline

Elastic

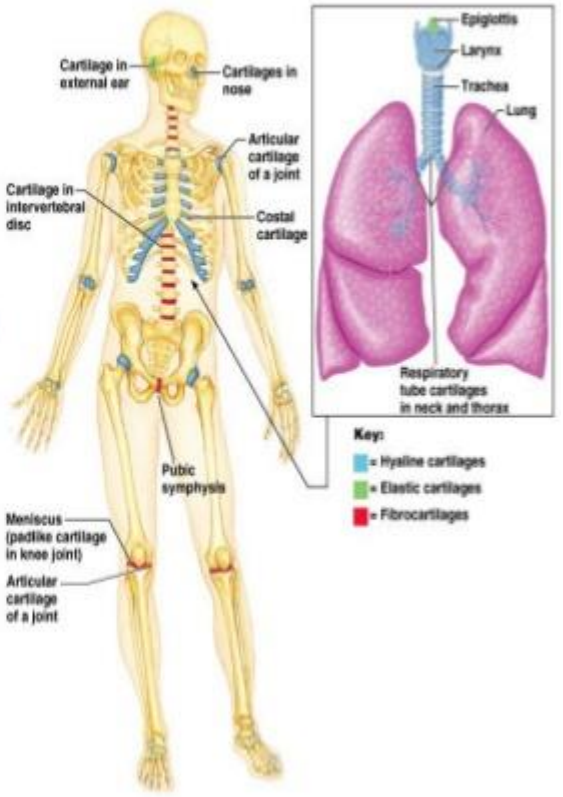
Fibrous

cartilage in adults

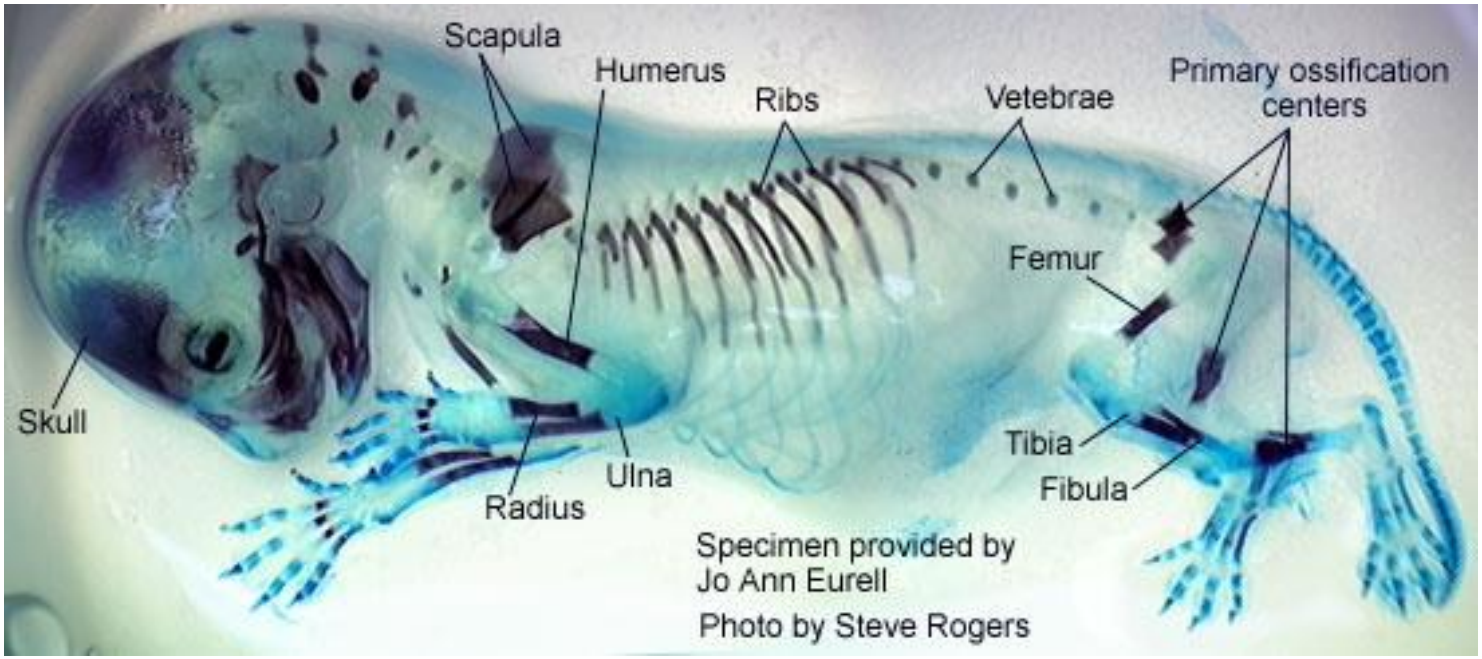
- Nose
- Joint surfaces
- Costal
- Larynx - voice box
- rings of trachea & bronch

- External ear
- Epiglottis
- Eustachian tube

- IVDs
- Pubic symphysis
- meniscus in knee joint



Hyaline



Alcian Blue&Alizarin Red

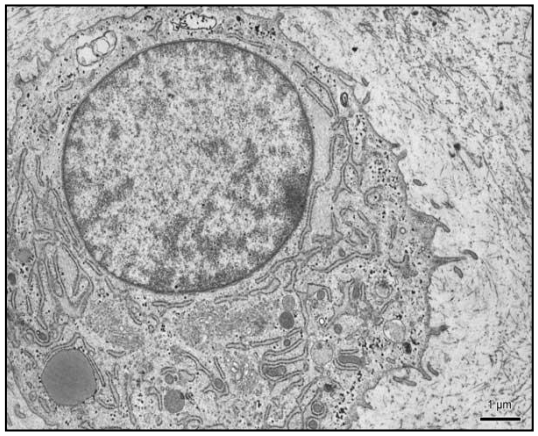
- most abundant
- temporary embryonal/fetal skeleton
- epiphyseal growth plate
- articulation (joints) respiratory passages

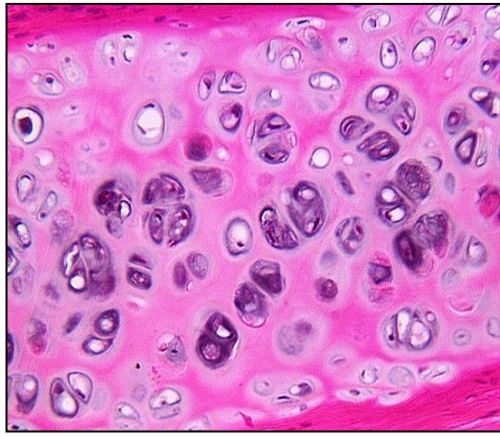
http://chickscope.beckman.uiuc.edu/explore/embryology/day14/dev2.html

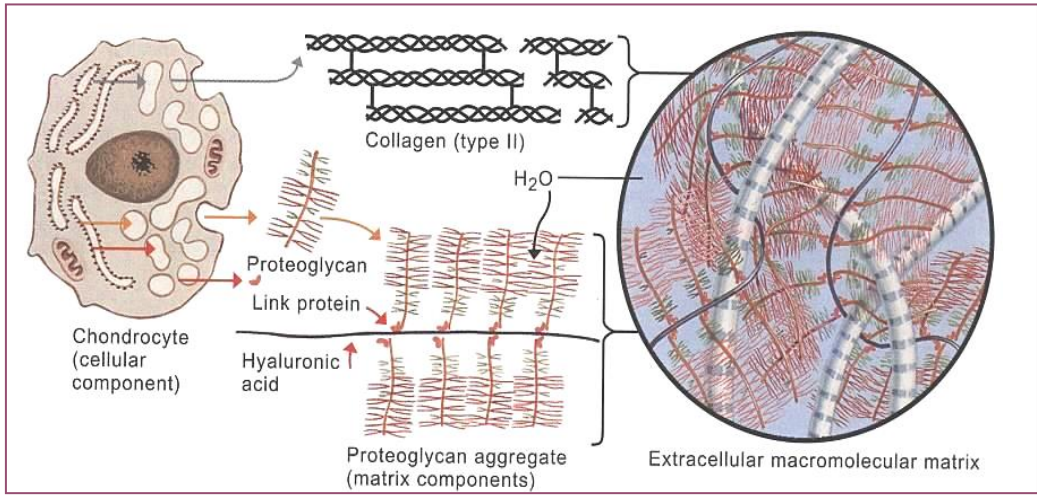
CELLS OF CARTILAGE

- **Chondroblasts and chondrocytes**

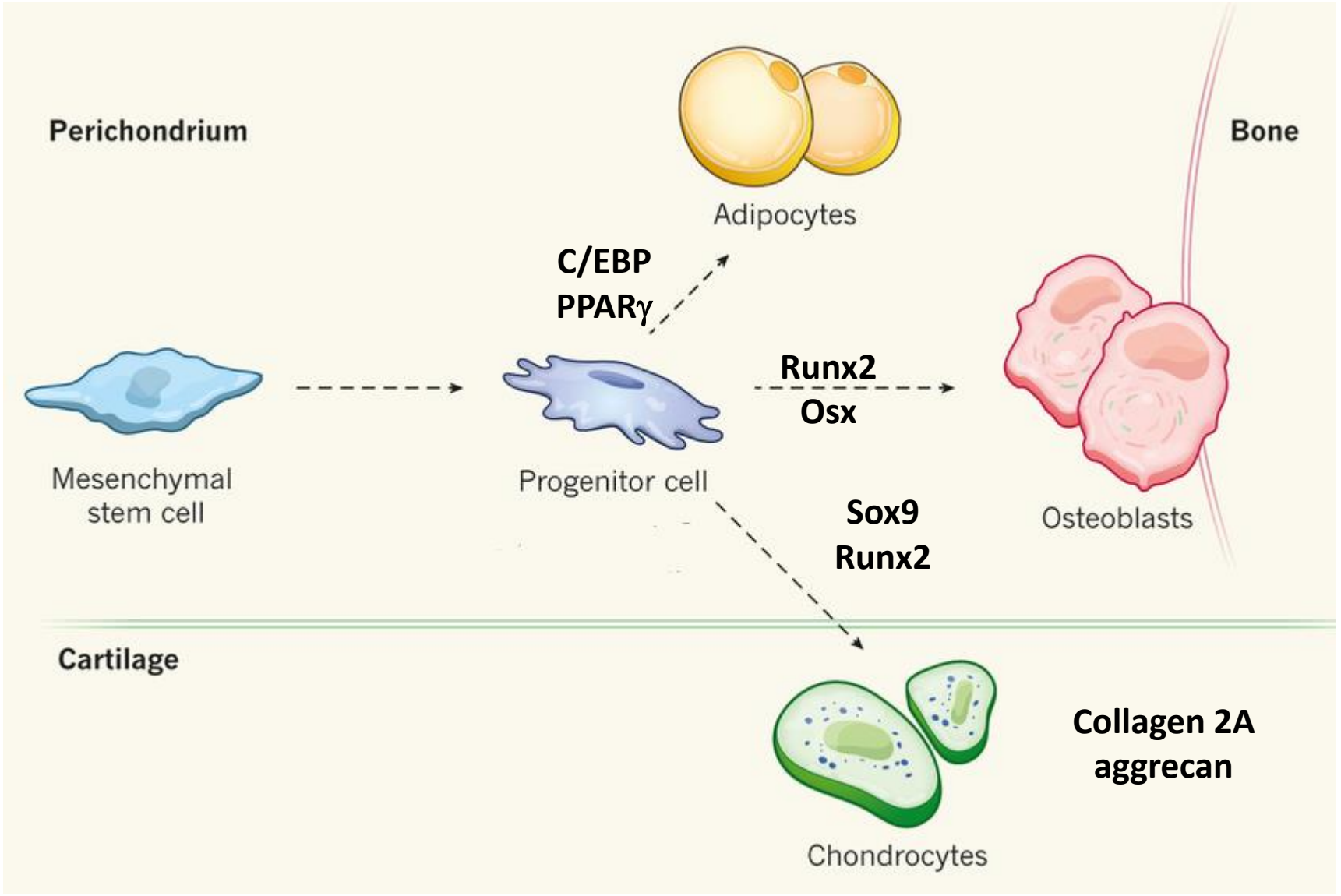
- mesenchymal origin
- typical ultrastructure of proteosynthetically active cells
- production of extracellular matrix
- interstitial proliferation
- isogenetic groups, lacunae



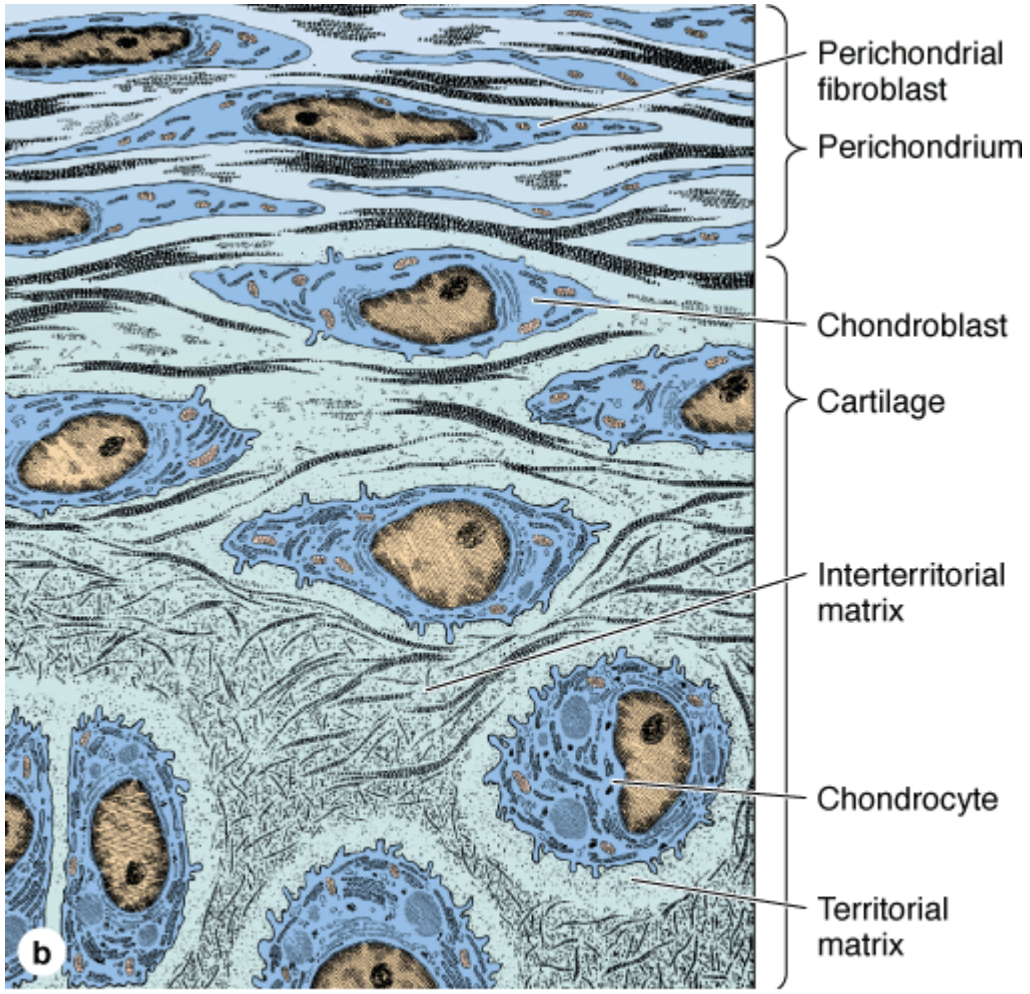


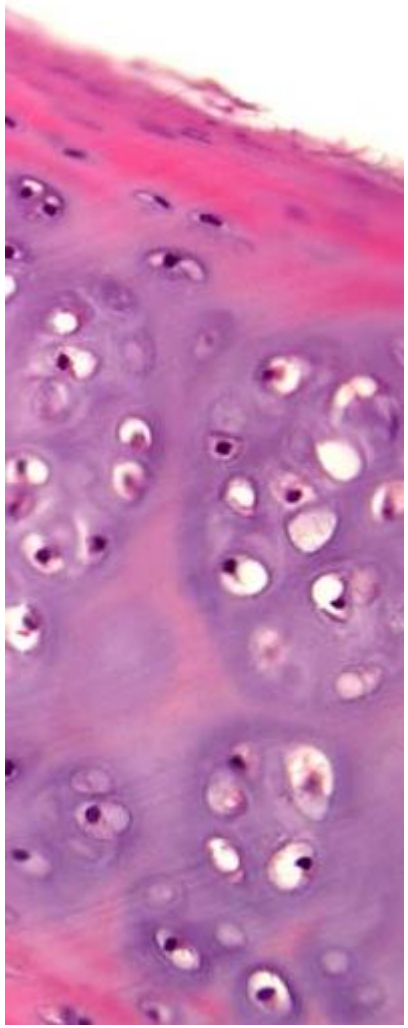


DIFFERENTIATION OF CHONDROBLASTS

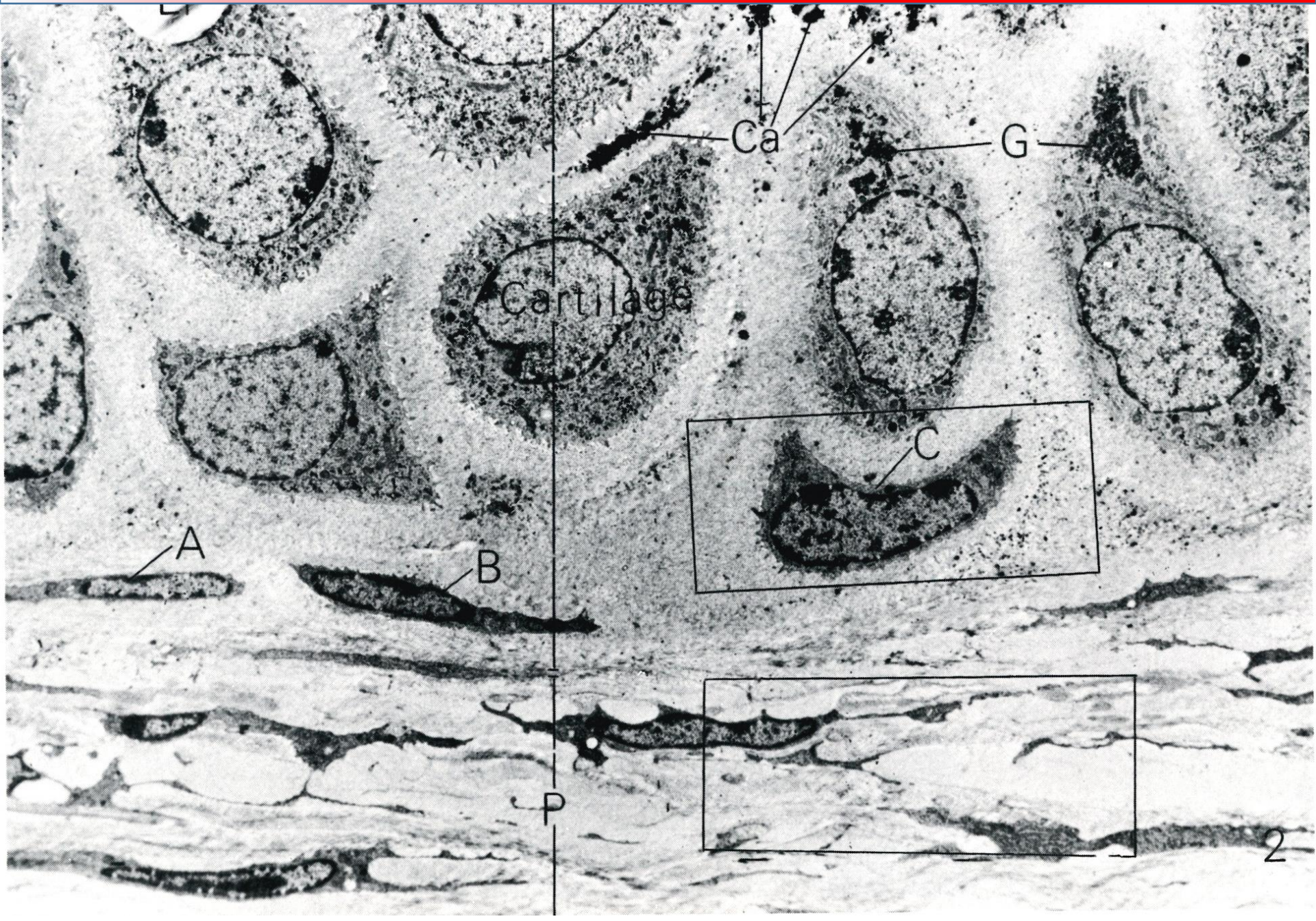


DIFFERENTIATION OF CHONDROBLASTS AND CHONDROCYTES

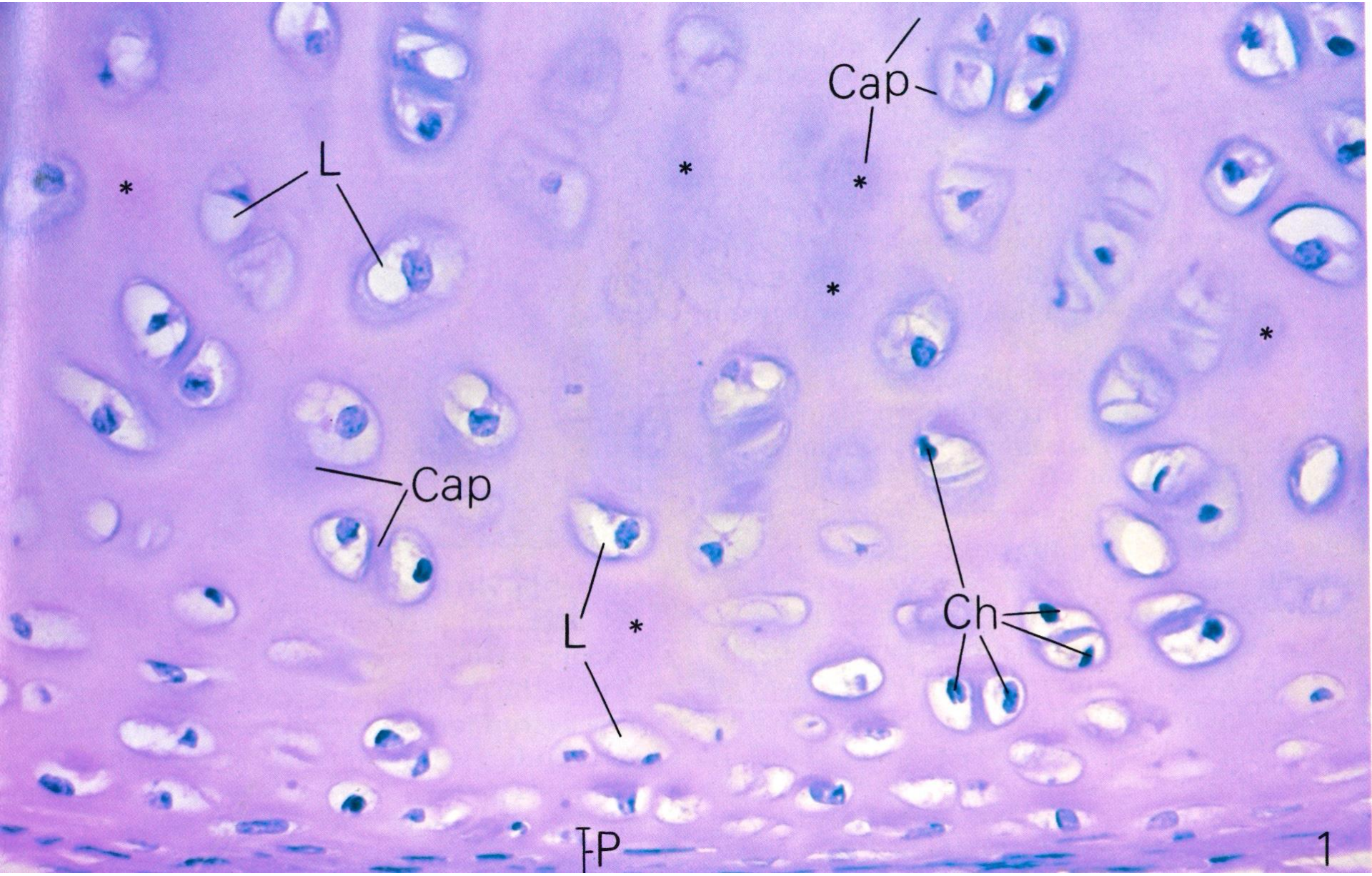




GROWTH OF HYALINE CARTILAGE

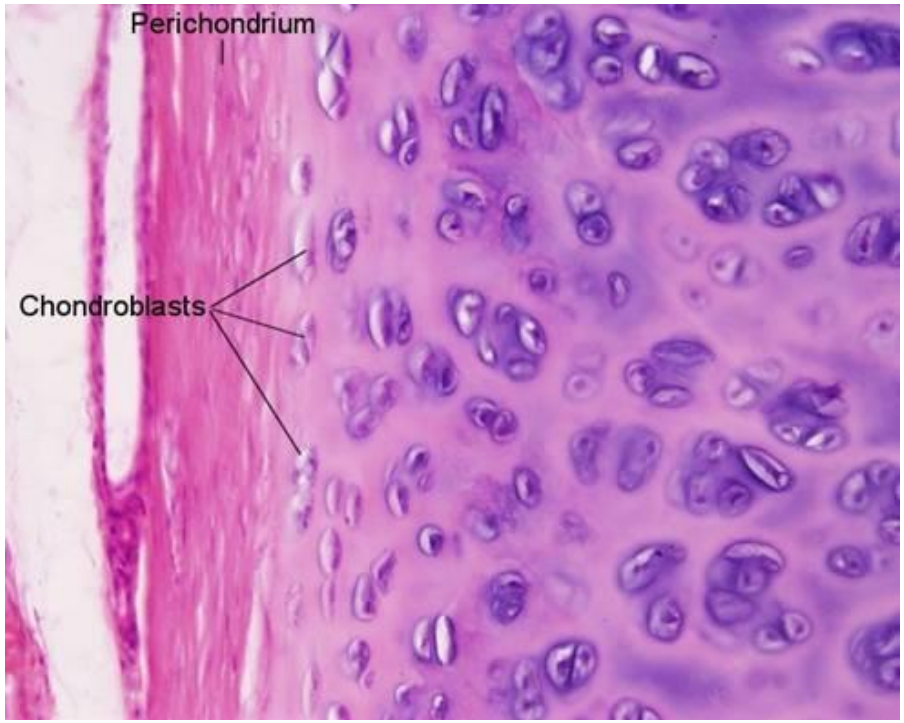


GROWTH OF HYALINE CARTILAGE

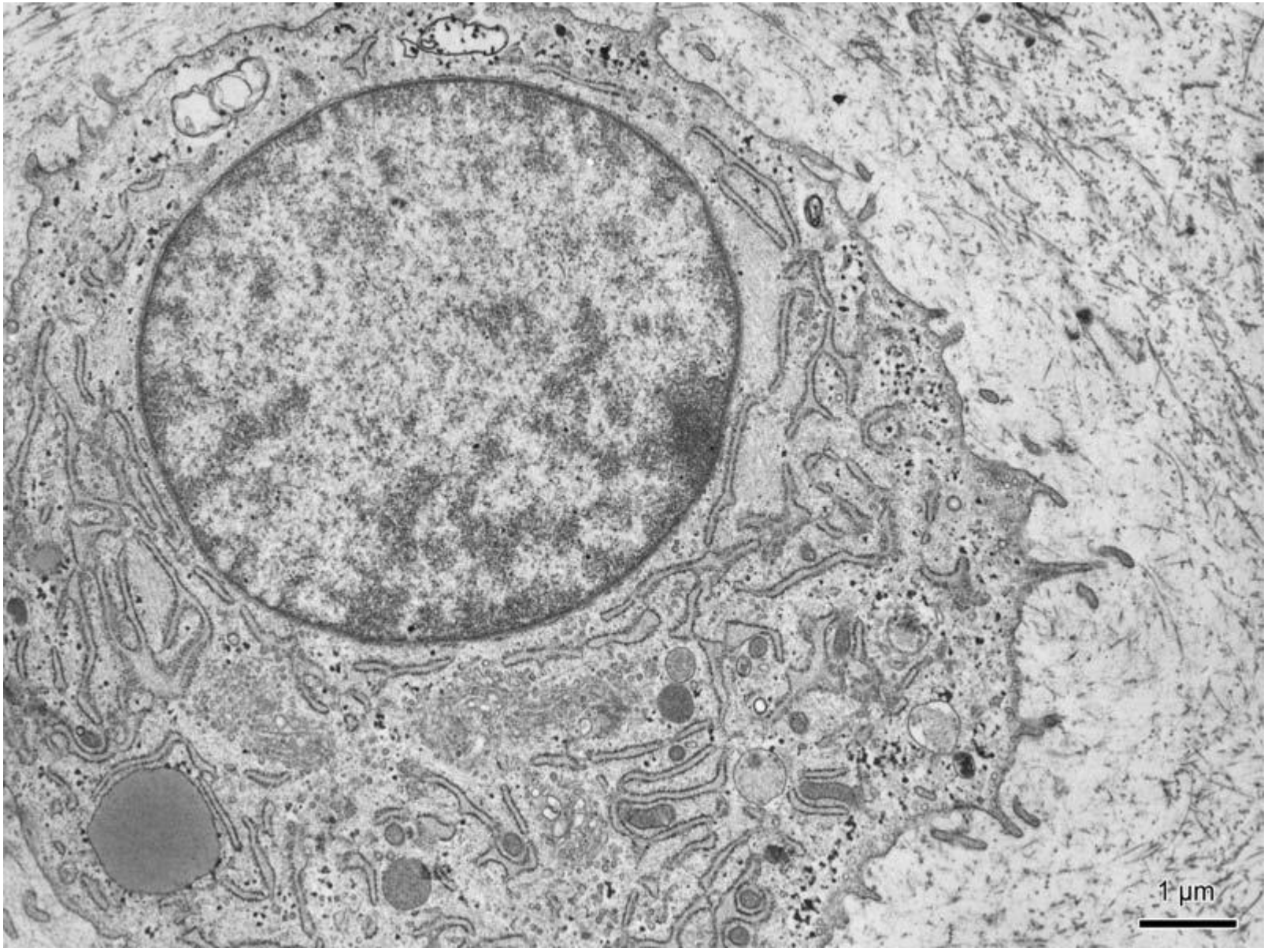


ULTRASTRUCTURE OF CHONDROCYTES

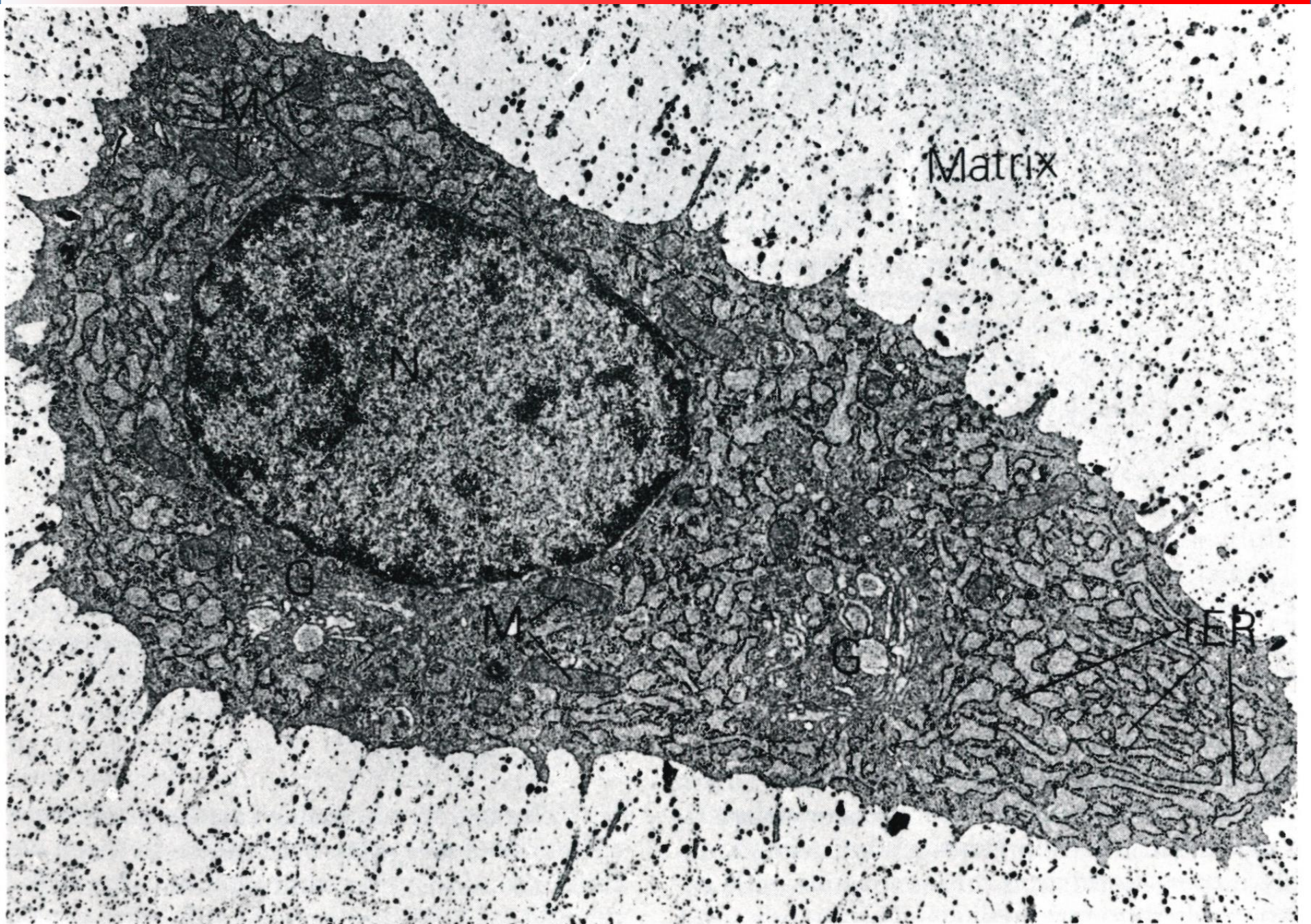
- oval → round cells
- rich in organelles, especially rER and GA
- glycogen granules (anaerobic metabolism)
- occasionally lipid droplets



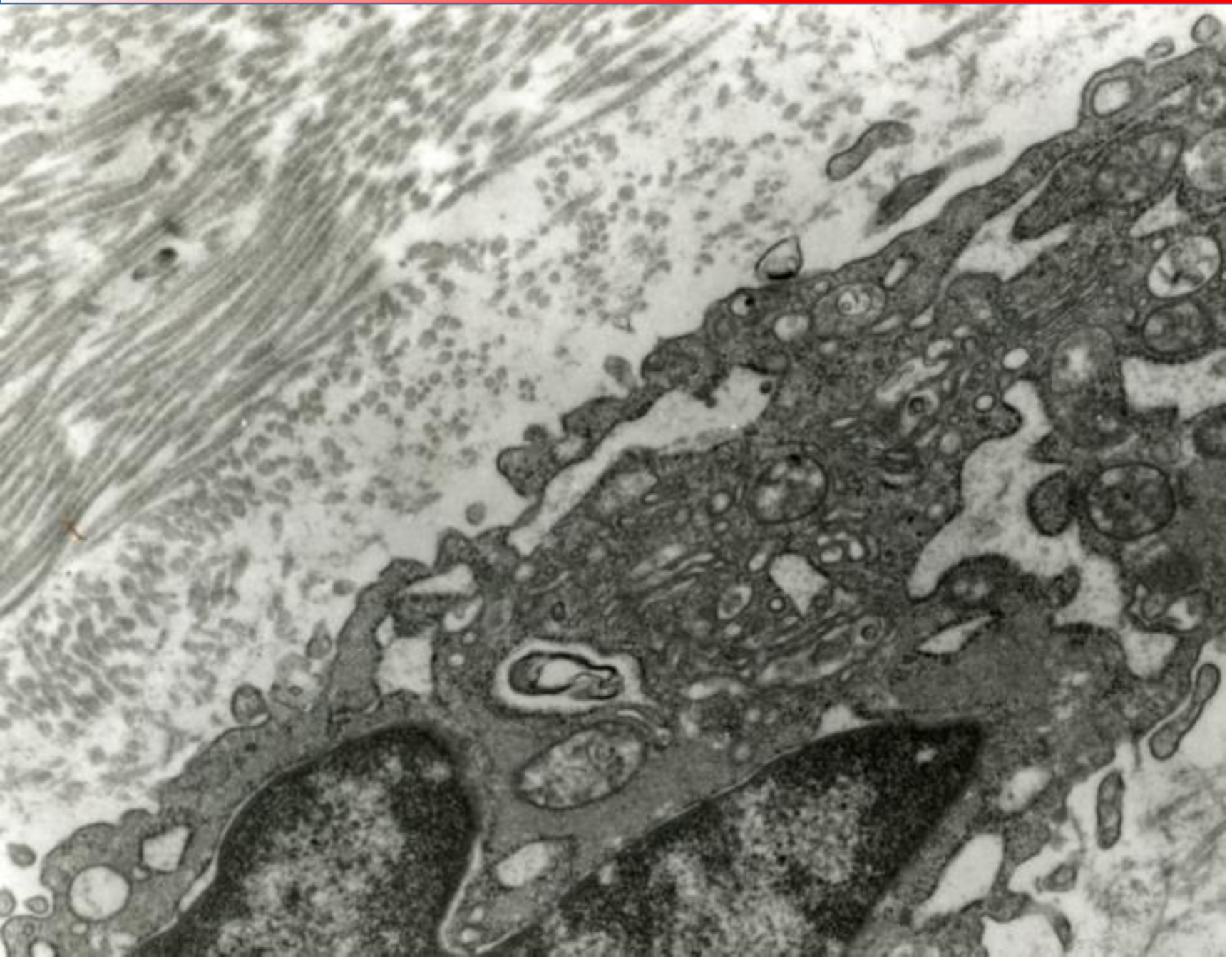
ULTRASTRUCTURE OF CHONDROCYTES



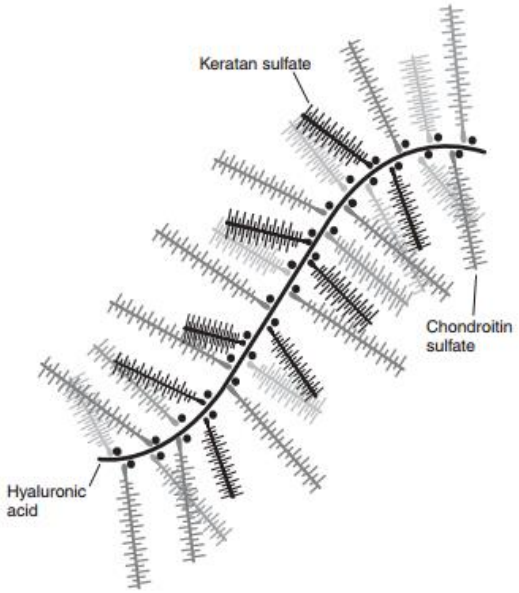
ULTRASTRUKTURA CHONDROCYTŮ



ULTRASTRUCTURE OF CHONDROBLASTS



- Extracelullar matrix



glycosaminoglycans

proteoglycans

fibers

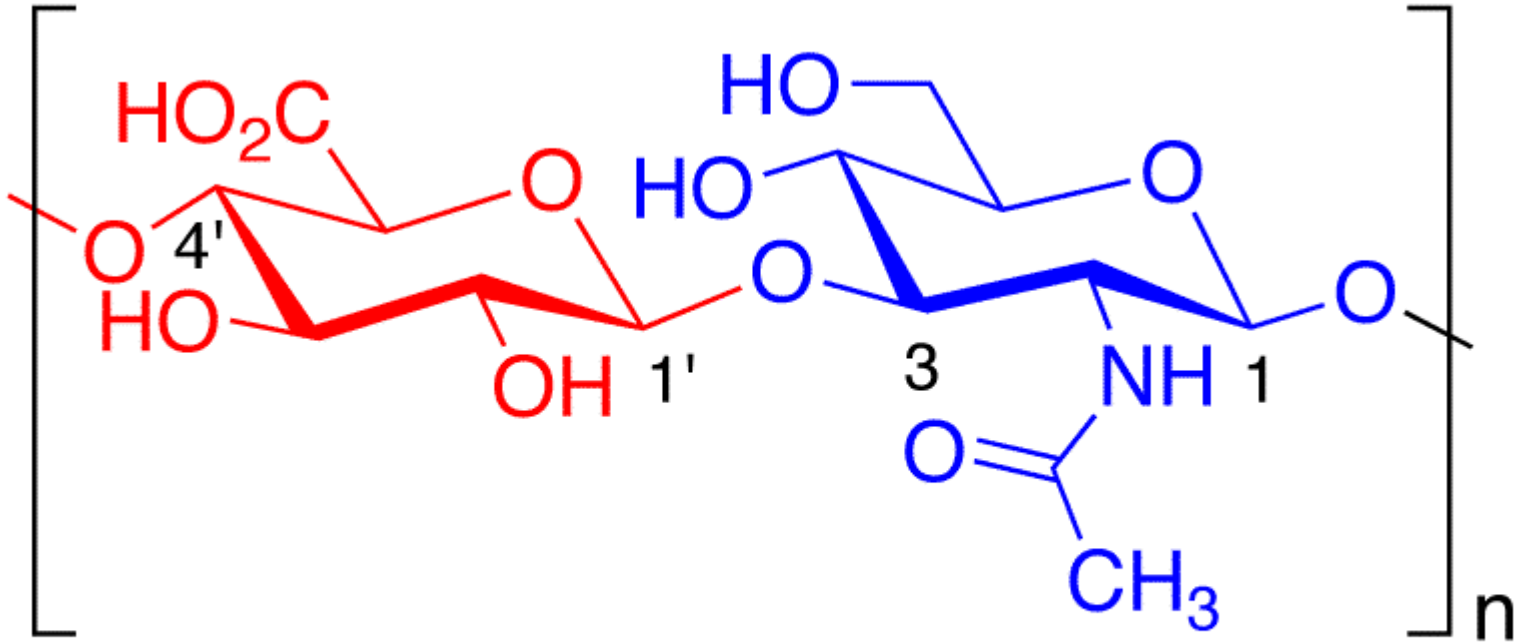
water

biomechanical properties

GLYCOSAMINOGLYCANS IN CARTILAGE

linear unbranched polysaccharides containing a repeating disaccharide unit:
1. N-acetylgalactosamine (GalNAc) or *N*-acetylglucosamine (GlcNAc)
2. uronic acid (glucuronate (GlcA)) or iduronate.

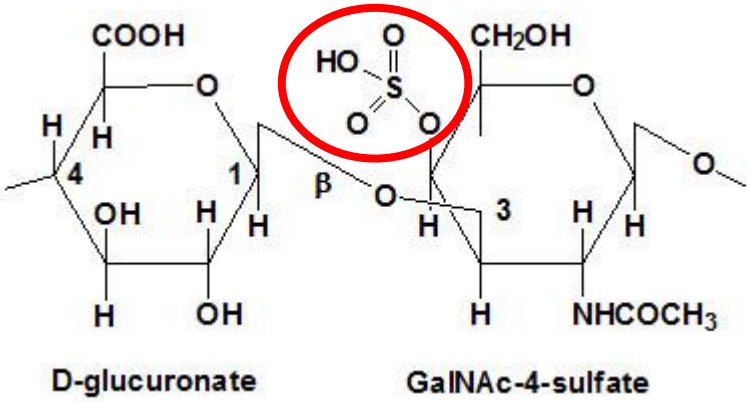
hyaluronic acid



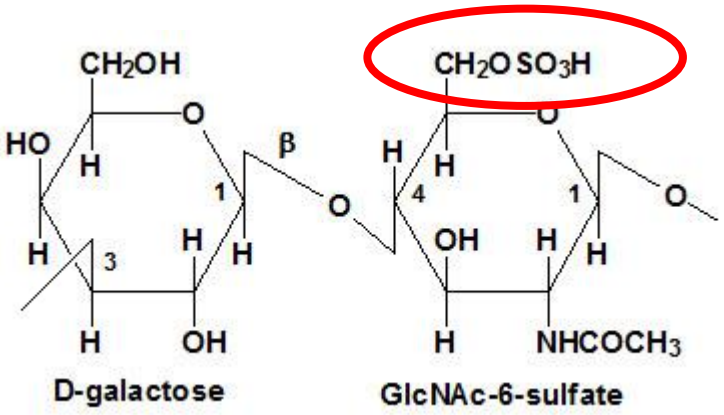
Glucuronic Acid *N-Acetyl-D-glucosamine*

GLYCOSAMINOGLYCANS IN CARTILAGE

Glycosaminoglycan	Localization
Hyaluronic acid	Umbilical cord, synovial fluid, fluid of corpus vitreum, cartilage
Chondroitinsulphate	Cartilage, bone, cornea, skin, notochord, aorta
Dermatansulphate	Skin, ligaments, adventitia of aorta
Heparansulphate	Aorta, lungs, liver, basal membranes
Keratansulphate	Iris, cartilage, nucleus pulposus, anulus fibrosus



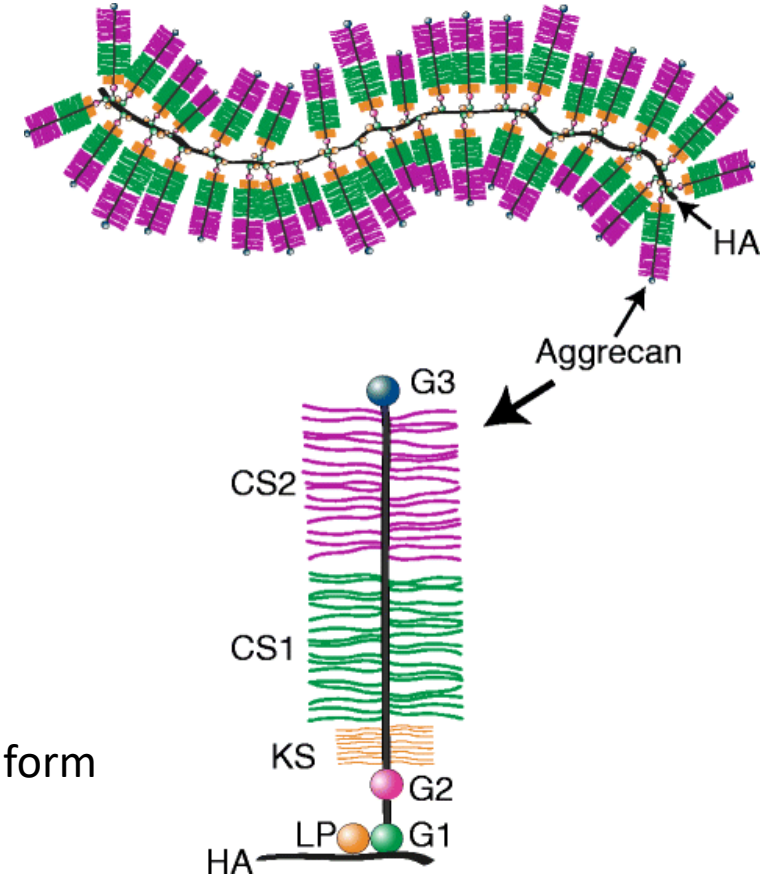
Chondroitinsulphate



Keratansulphate

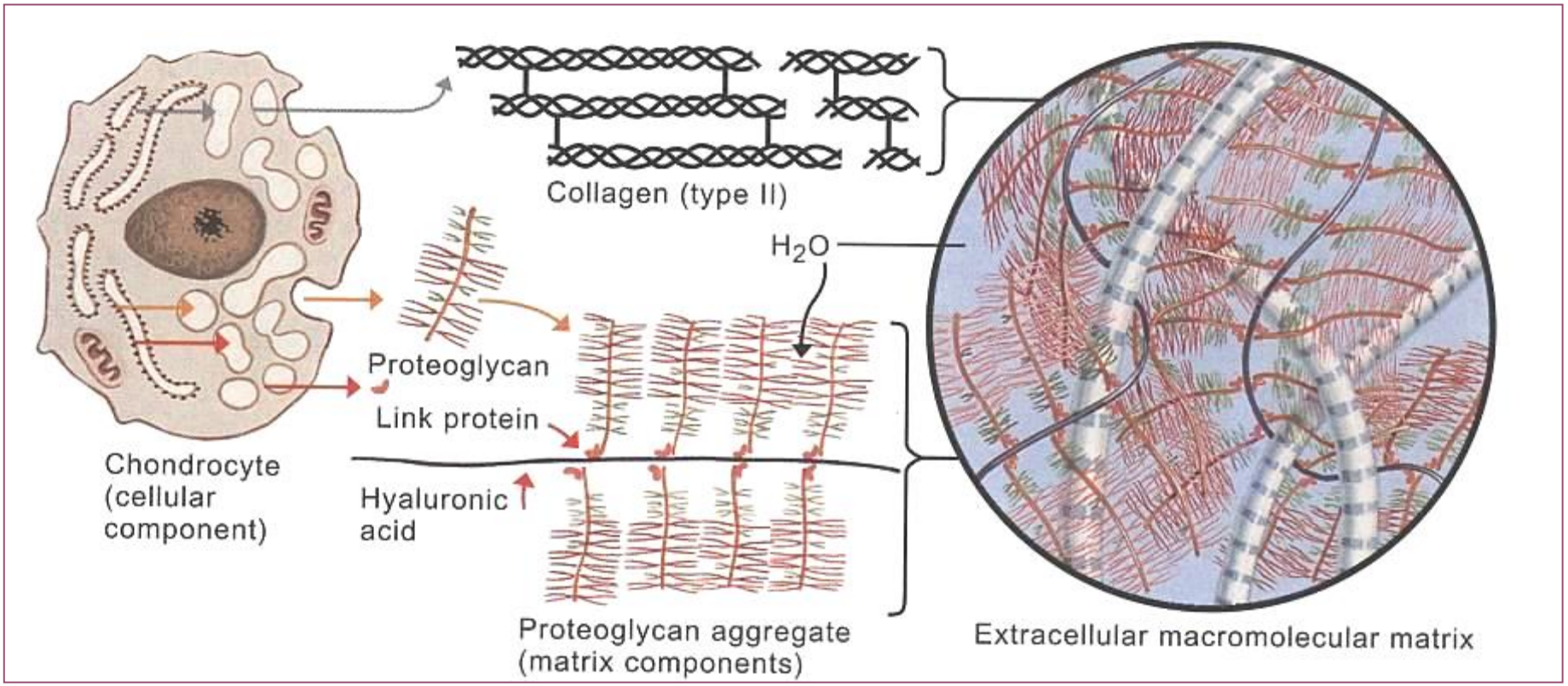
PROTEOGLYCANS AND FIBERS

- **proteoglycans**
- protein + dominant linear saccharide component
- proteoglycan aggregates
- water-binding – 80%, volume dependent of hydratation
  - **aggrecan (cartilage)**
  - syndekan
  - fibroglykan



- **collagen fibrils**
- col II + col IX/XI
- thin fibrils (15-20 nm → no striation) that do not form fibers like col I
- interconnected with perichondrium
- elastic fibers

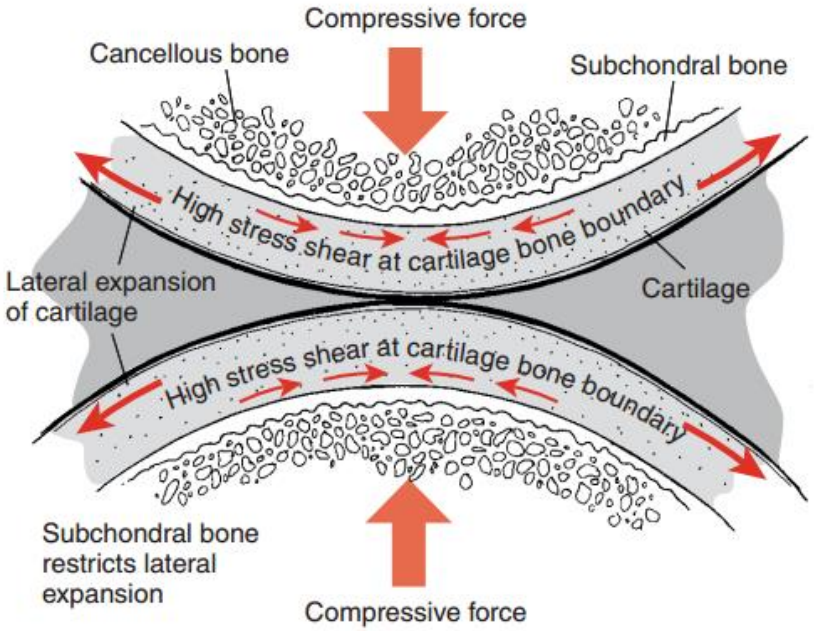
TISSUE ARCHITECTURE OF CARTILAGE ECM



transduction of biochemical and biomechanical signals

TISSUE ARCHITECTURE OF CARTILAGE ECM

- **pressure elasticity**
- proteoglycans – polyanionic (COO^-, SO_4^{II-})
- expansion prevented by collagen fibrils
- repulsion forces



- **biphasic model of cartilage** → ECM composition
- proteoglycans, collagen, cells, and lipids constitute the solid phase of the mixture
- interstitial fluid that is free to move through the matrix fluid phase)
- under impact loads, fluid flows through the framework, until the cartilage start to behave as a single-phase, incompressible, elastic solid - the fluid does not flow
- after load release, fluid returns
- nutritive aspect

TISSUE ARCHITECTURE OF CARTILAGE ECM

- **synovial cartilage**

I. tangential (superficial) zone

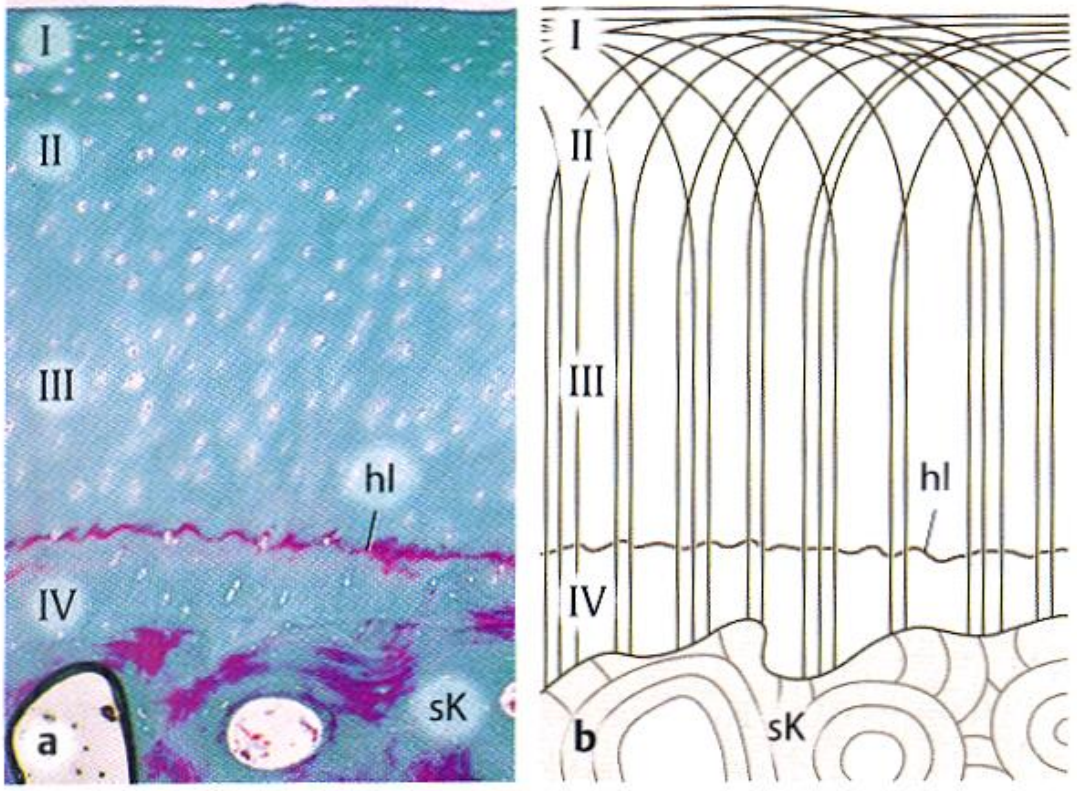
II. transitional zone

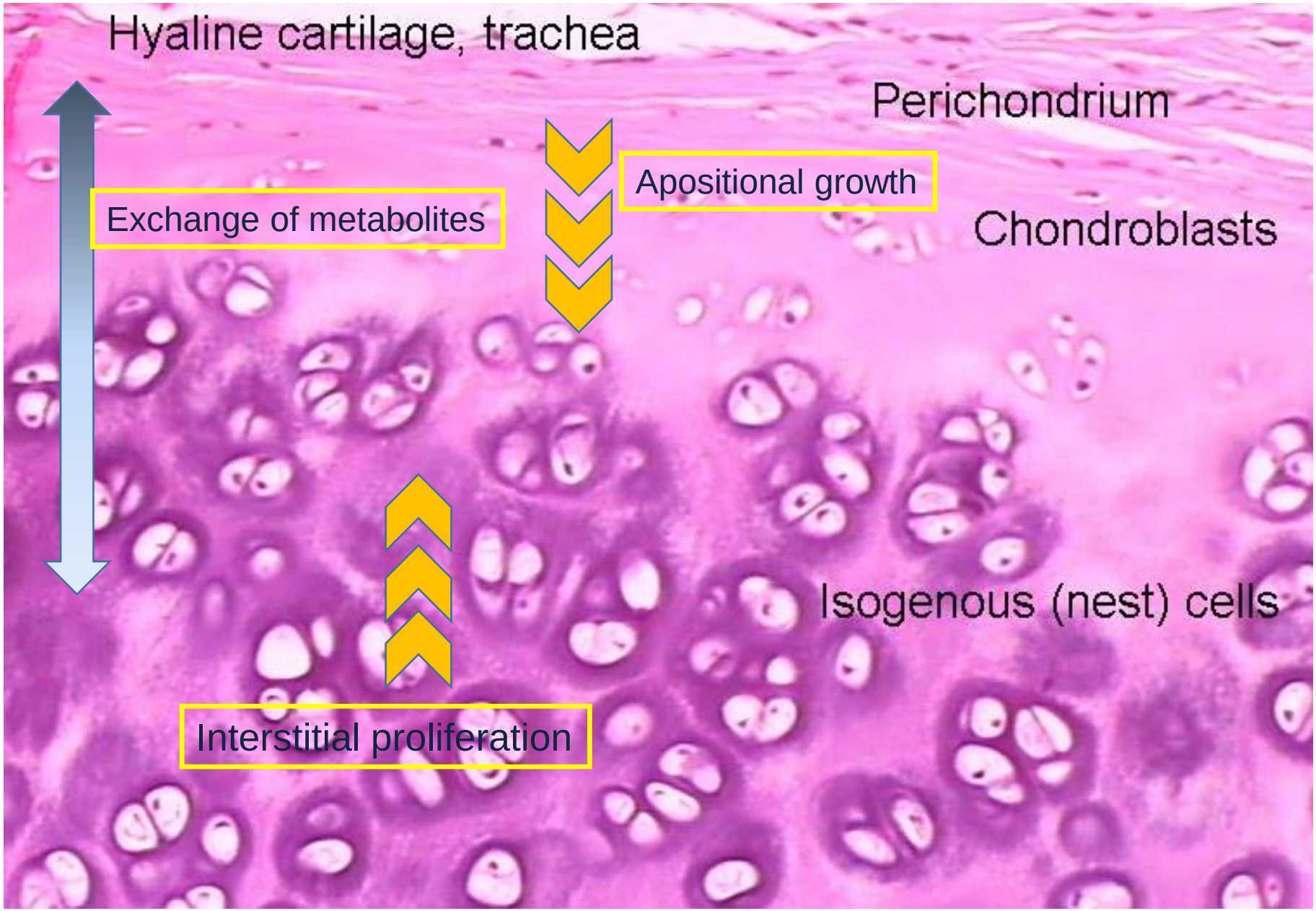
III. radial (deep) zone

tide mark

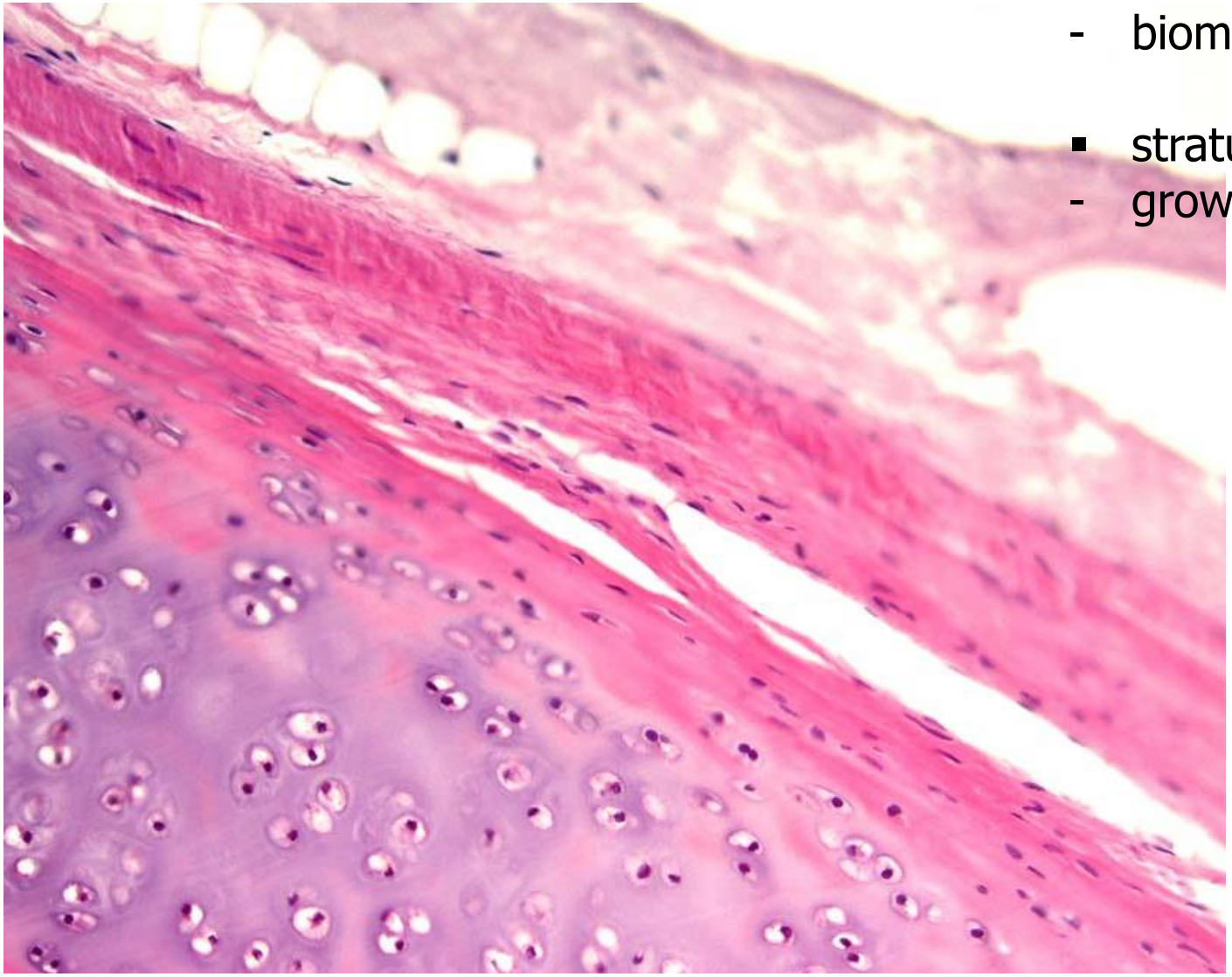
I. mineralized cartilage zone

subchondral bone




Hyaline cartilage, trachea
Perichondrium
Apositional growth
Exchange of metabolites
Chondroblasts
Isogenous (nest) cells
Interstitial proliferation

PERICHONDRIUM

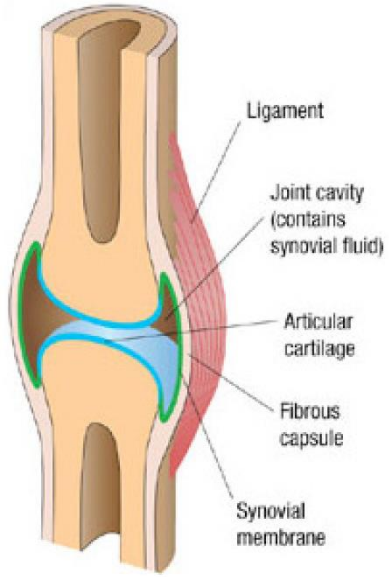


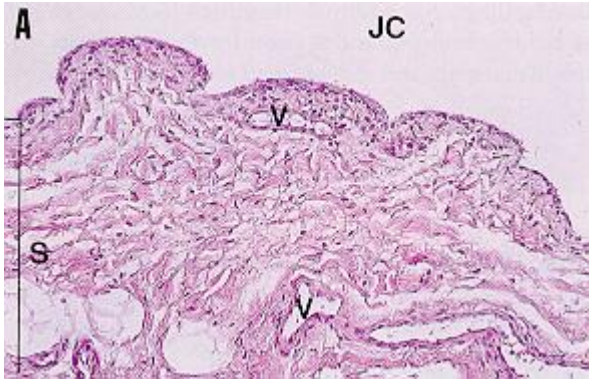
- stratum fibrosum
  - biomechanics

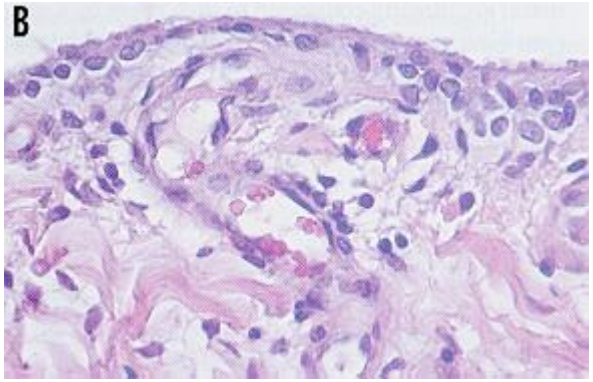
- stratum chondrogenicum
  - growth

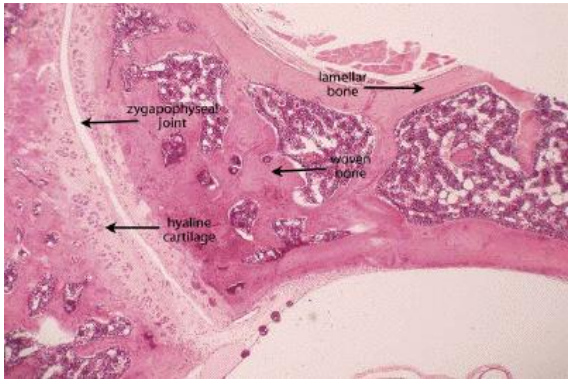
SYNOVIUM

- *membrana fibrosa*
- dense collagen c.t.
- *membrana synovialis*
- intima, subintima
- folds extending to the joint cavity
- numerous blood and lymphatic vessels, nerves
- discontinuous cell layers (synovialocytes)
- basal membrane and intercellular junctions absent - **not an epithelium:** mesenchymal (c.t.) origin
- synovial fluid rich in hyaluronans
- *bursae synoviales, vaginae tendineum*









http://www2.indstate.edu/thcme/mmmoga/histology/slide35.html

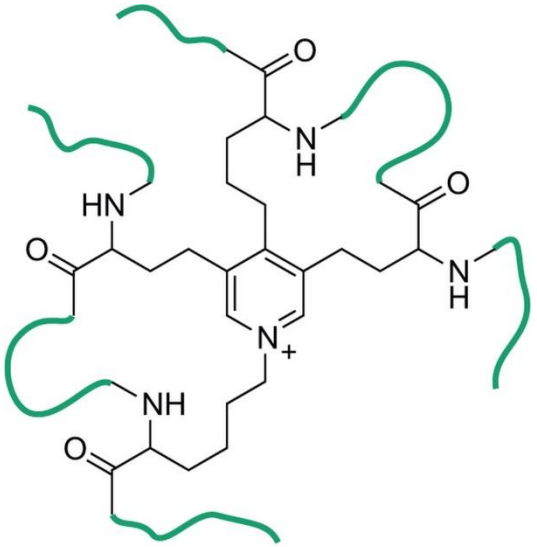
https://www.dartmouth.edu/~anatomy/Histo/lab_2/bone/DMS090/53.gif

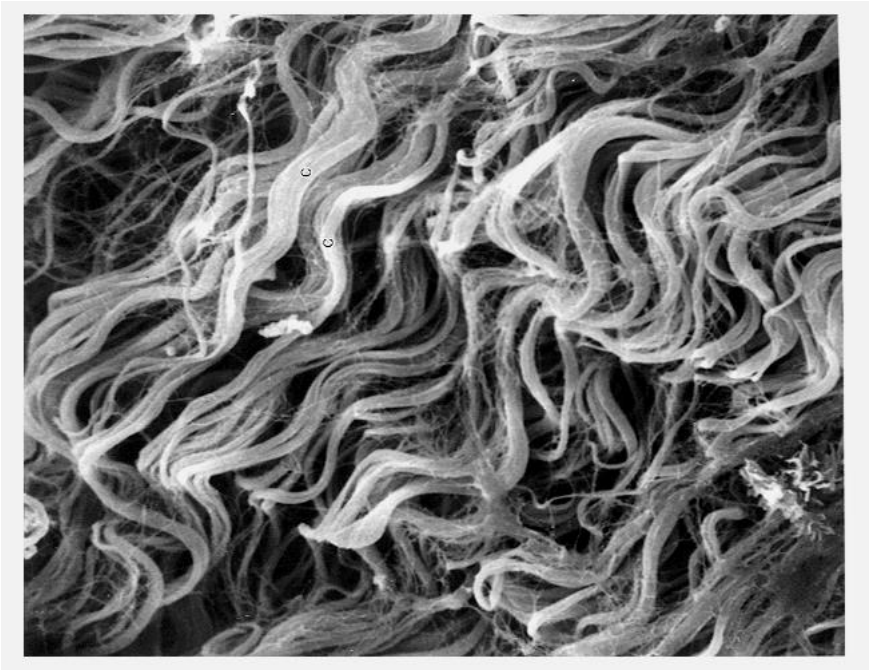
ELASTIC CARTILAGE

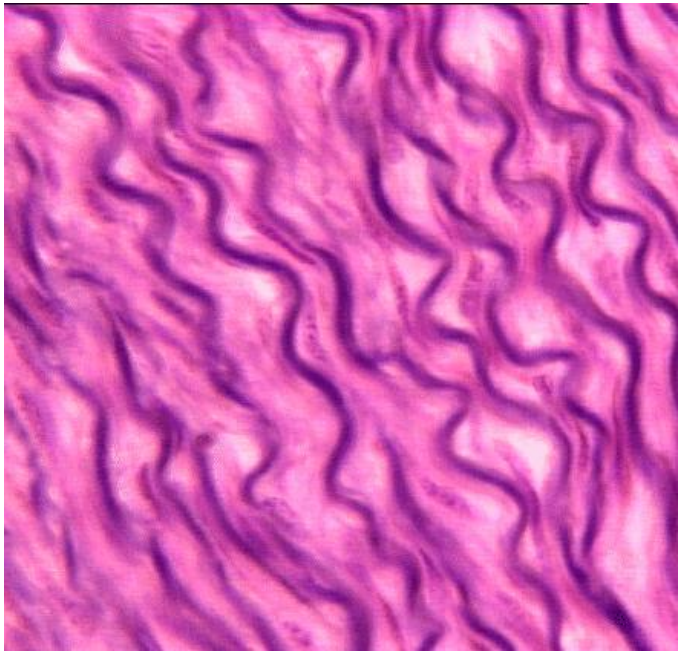
- acidophilic elastic fibers dispersed in matrix
- no isogenetic groups
- auricula, meatus, larynx, epiglottis

ELASTIC FIBERS

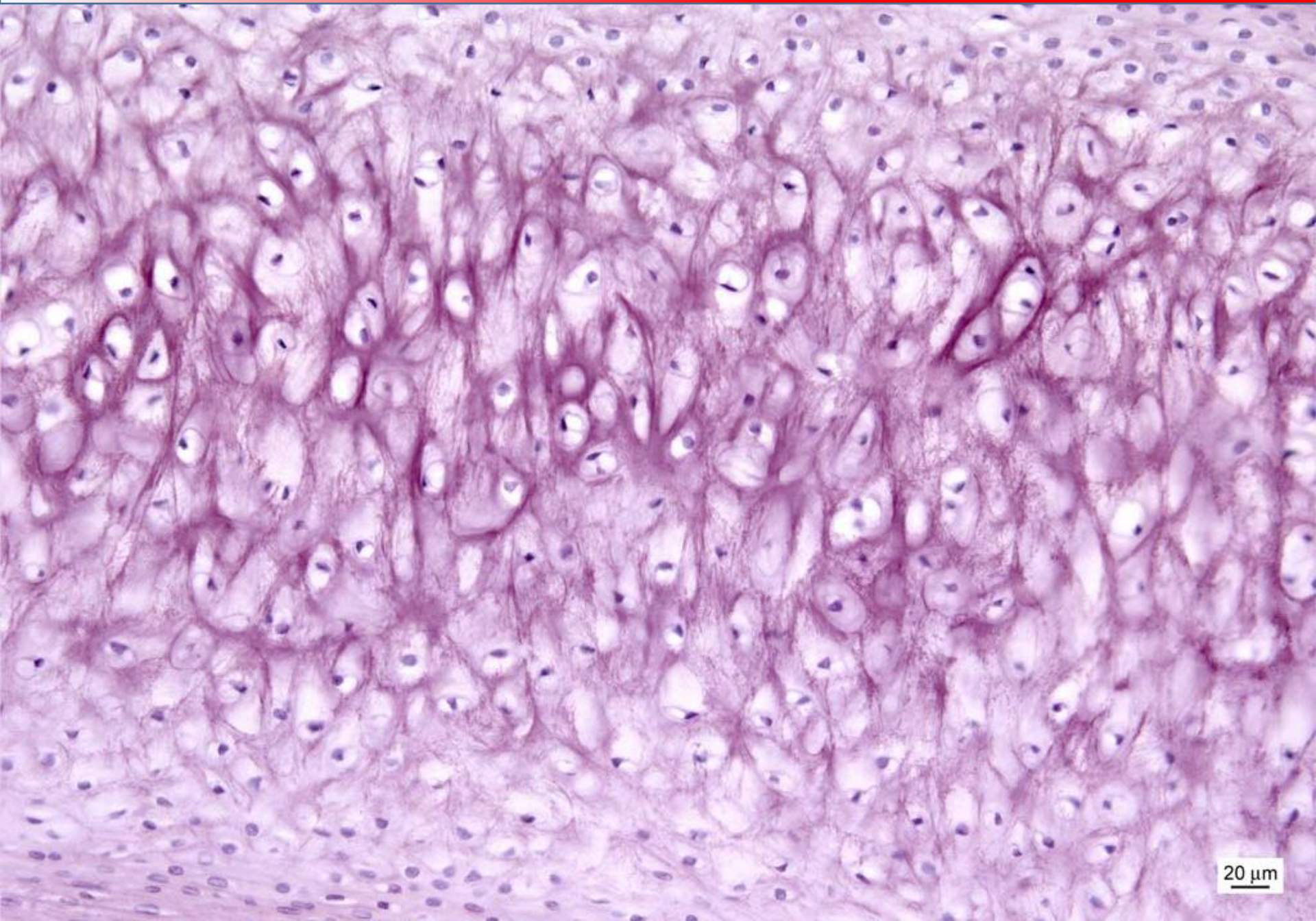
- less abundant than collagen
- polymer – tropoelastin
- minimal tensile resistance, loss of elasticity if overstretched
- reduction of hysteresis = allow return back to original state after mechanic change





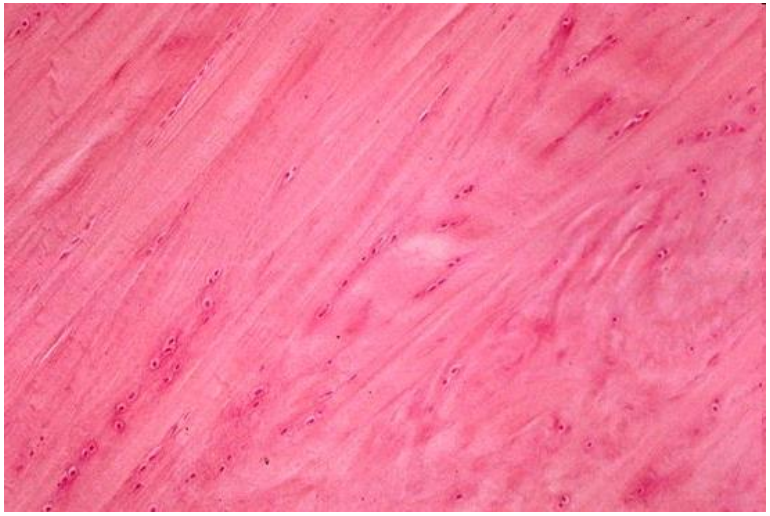


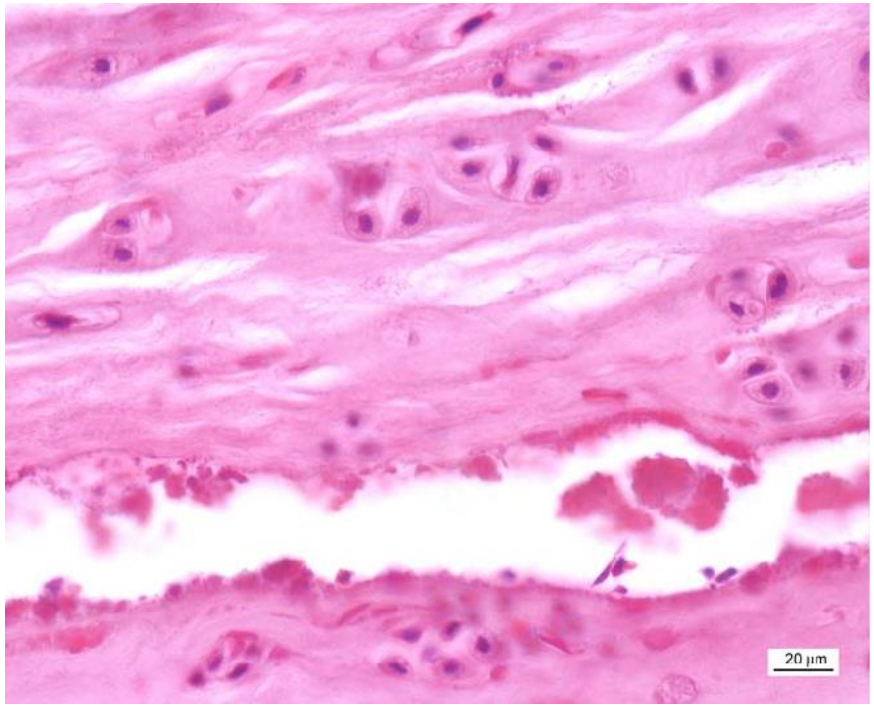
ELASTIC CARTILAGE

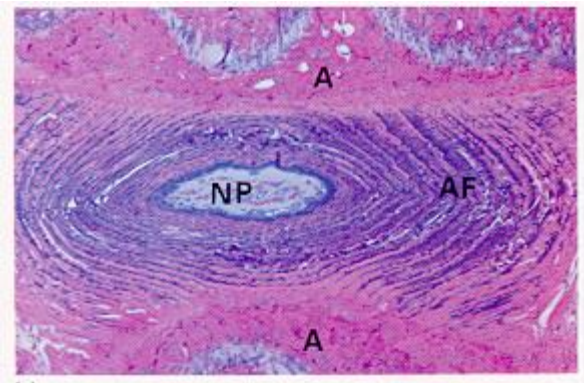


FIBROCARTILAGE

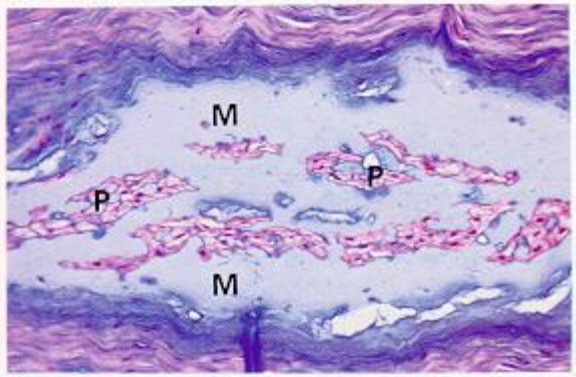
- fibrous compound dominant – collagen I and II – mechanical durability
- minimum of amorphous matrix-fibers visible
- intervertebral discs, symphysis pubis, articular discs, meniscus







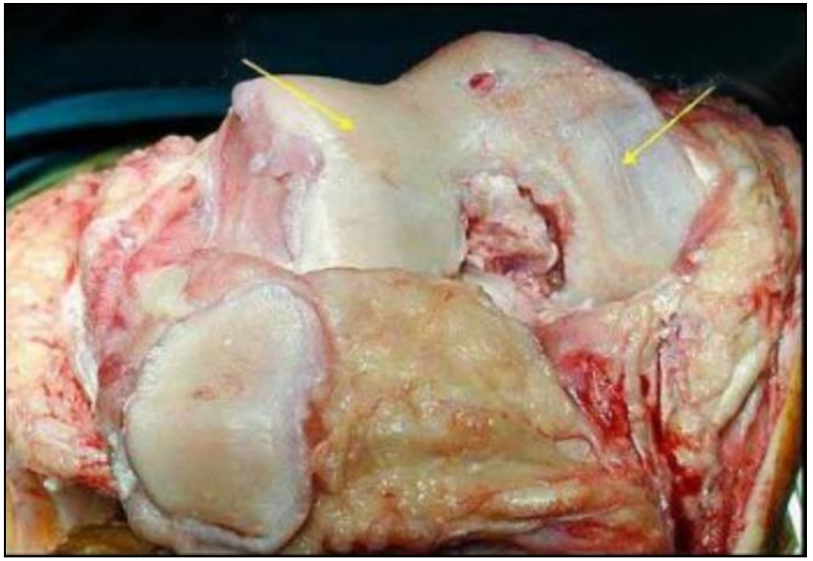
(a)



(b)

CLINICAL CORRELATION

- Cartilage – no innervation, no vascularization – no spontaneous regeneration
- No migration of chondrocytes to site of damage
- Initiation of other degenerative events leading to cartilage erosion (arthritis)



Therapy:

- joint mobility
- restoration of biochemical and biophysical parameters of cartilage
- prevention of further damage
- removal of damaged tissue, autologous transplantation
- MSCs on biocompatible scaffolds (still only experimental)

Further reading:

Stem Cell Research 44 (2020) 101738

Contents lists available at ScienceDirect

Stem Cell Research

journal homepage: www.elsevier.com/locate/scr

Human mesenchymal stem cell therapy for cartilage repair: Review on isolation, expansion, and constructs

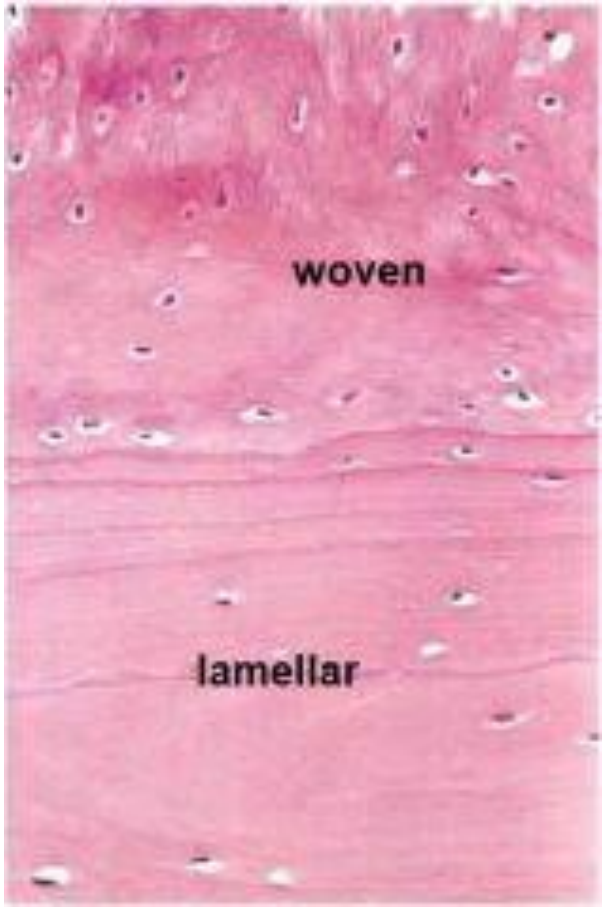
Alan T.L. Lam*, Shaul Reuveny, Steve Kah-Weng Oh

*Bioprocessing Technology Institute, A*STAR (Agency for Science, Technology and Research), Singapore 138668, Singapore*


BONE
20 µm

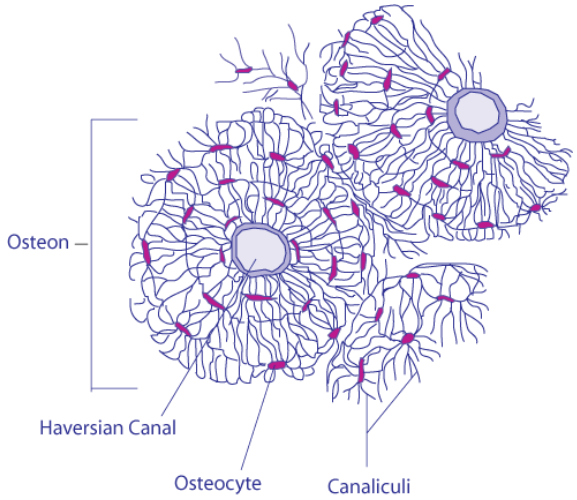
HISTOLOGICAL CLASSIFICATION OF BONE TISSUE

- **Primary (woven, fibrous)**



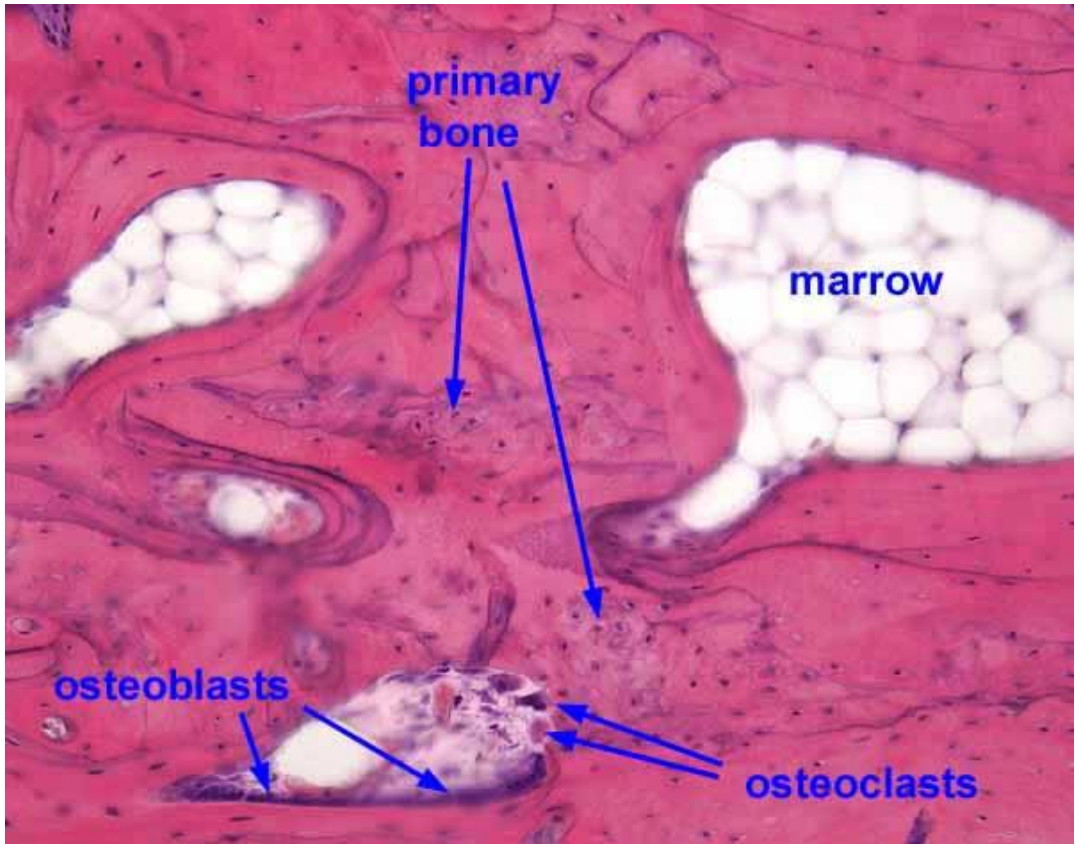
- **Secondary (lamellar)**

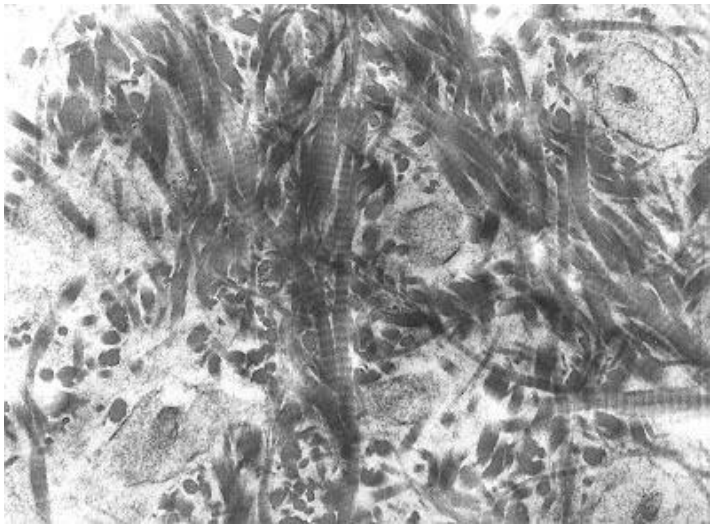
- Lamellae – collagen fibers in concentric layers (3-7μm) around a canal with capillaries = Haversian system (osteon)



PRIMARY (WOVEN) BONE

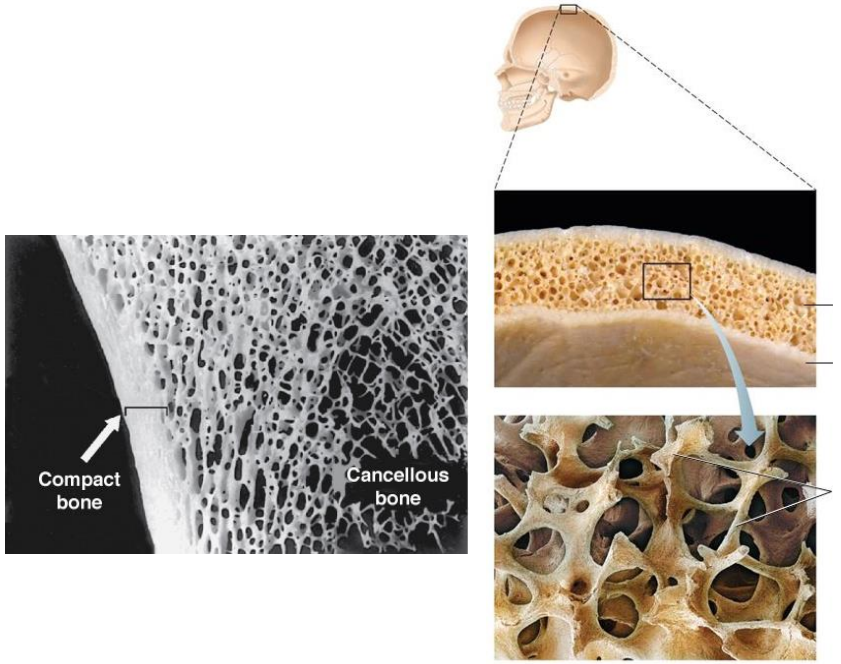
- Temporary, growth and regeneration of bones, collagen fibrils woven
- Replaced by secondary bone
- Remains only in some parts of body - sutures of skull, *tuberositas ossium*, tooth cementum

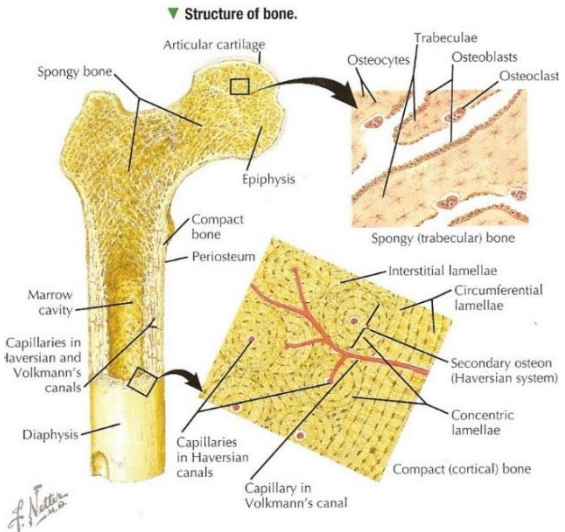


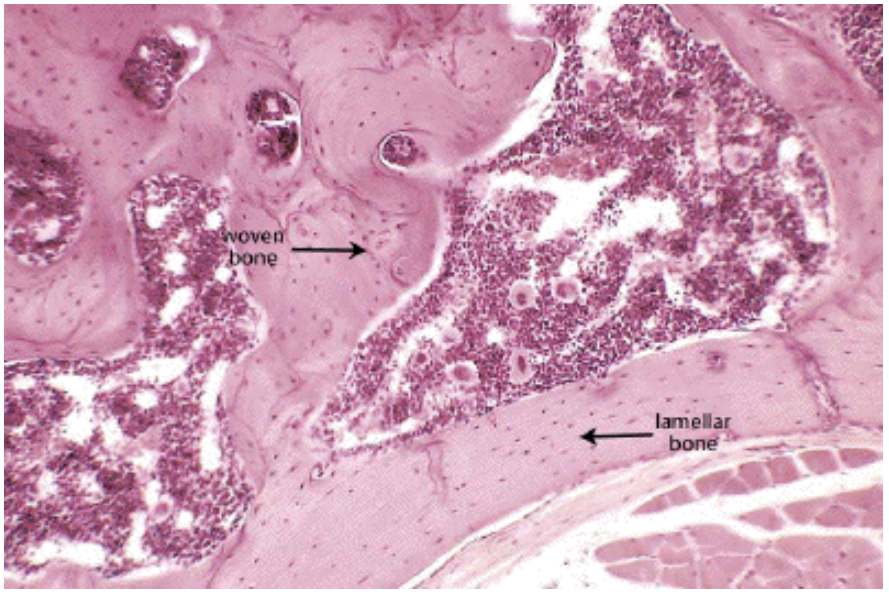


SECONDARY (LAMELLAR) BONE

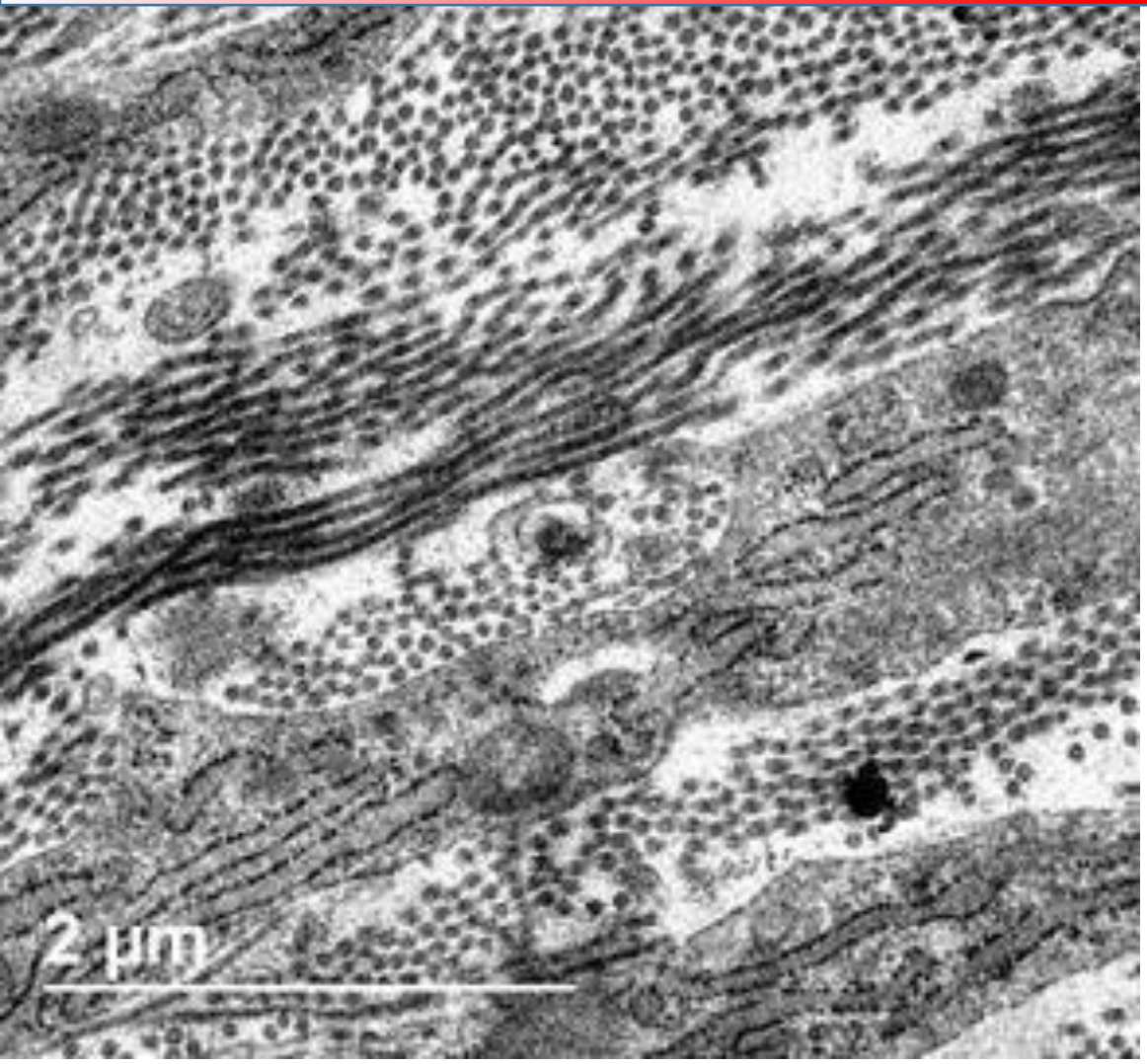
- Lamellae – collagen fibers in concentric layers (3-7μm) around a canal with capillaries = Haversian system (osteon)
- **Spongy (trabecular)**
  -Trabeculae, similar to compact
  -Epiphyses of long bones, short bones, middle layer of flat bones of the skull (*diploe*)
- **Compact**
  - Outer and inner coat lamellae typical Haversian systems
  - Volkmann's canals
  - Interstitial canals

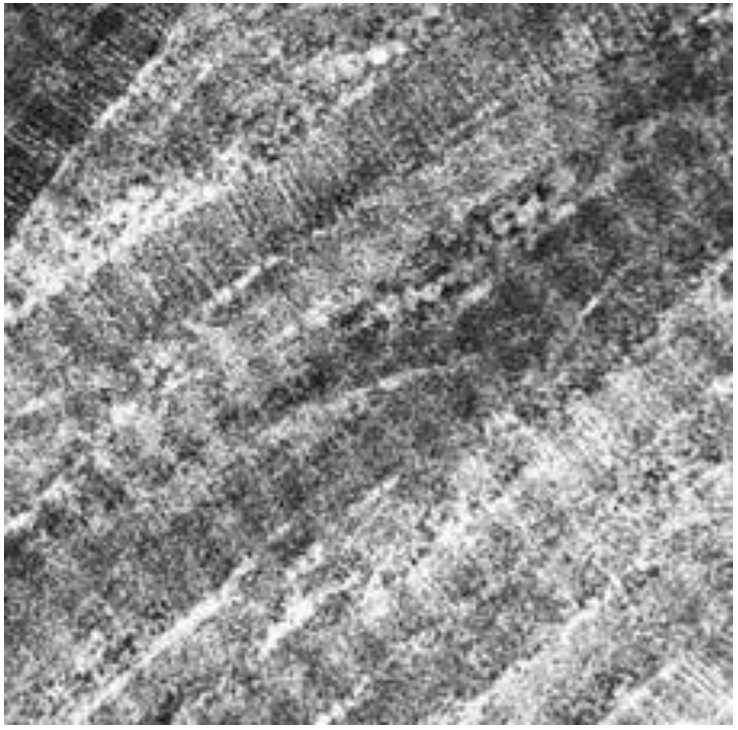




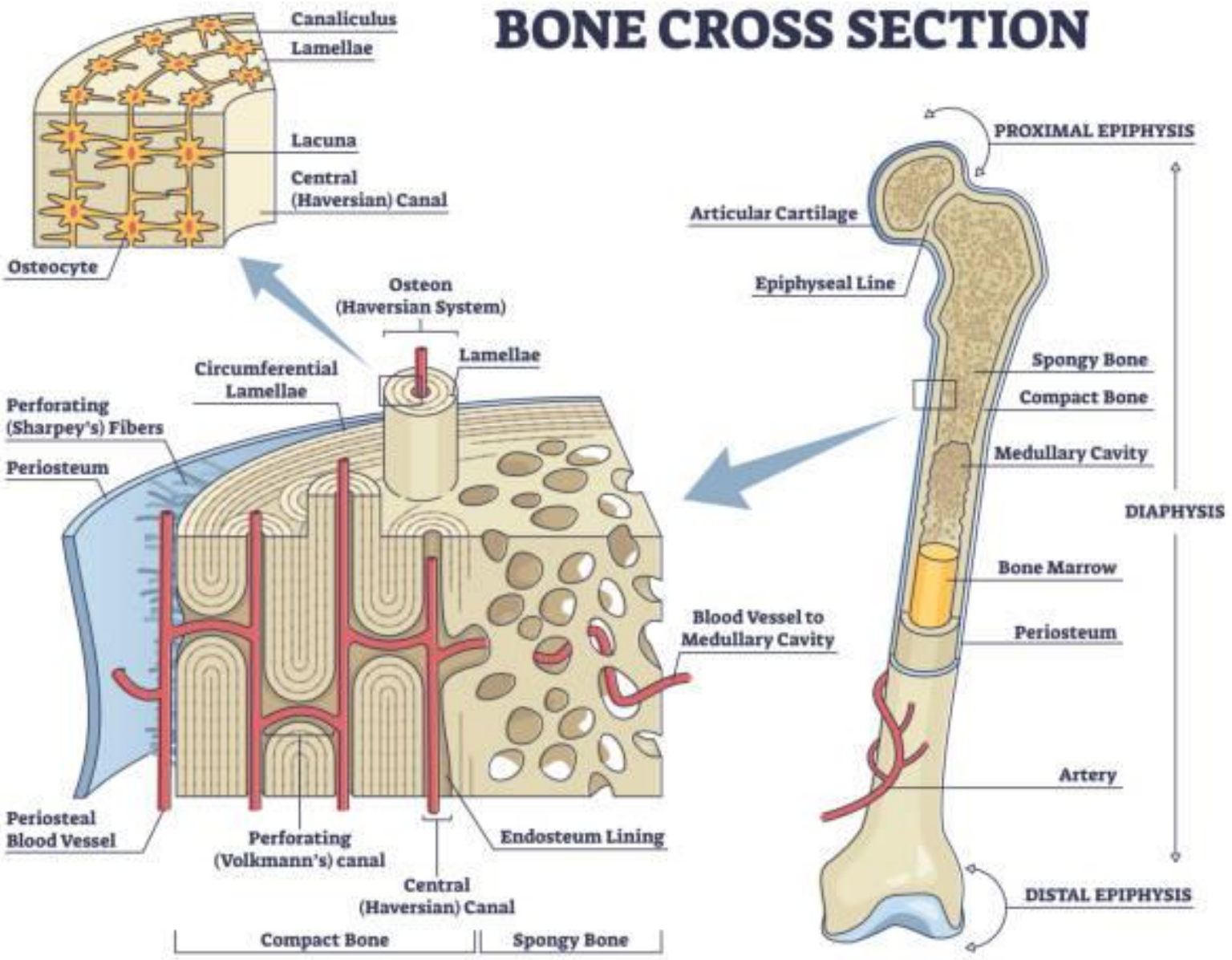


SECONDARY (LAMELLAR) BONE

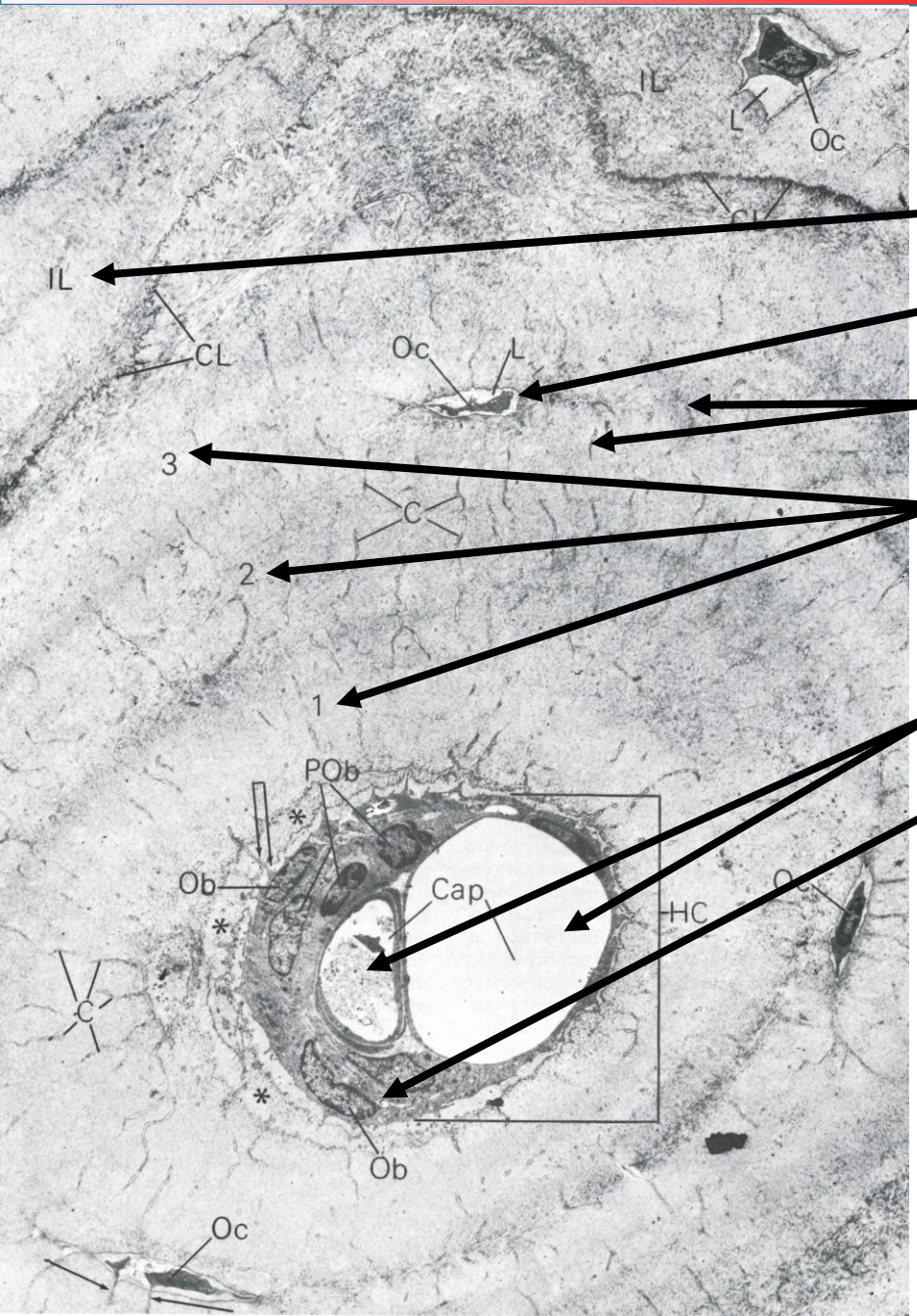




SECONDARY (LAMELLAR) BONE



SECONDARY (LAMELLAR) BONE



Interstitial lamella

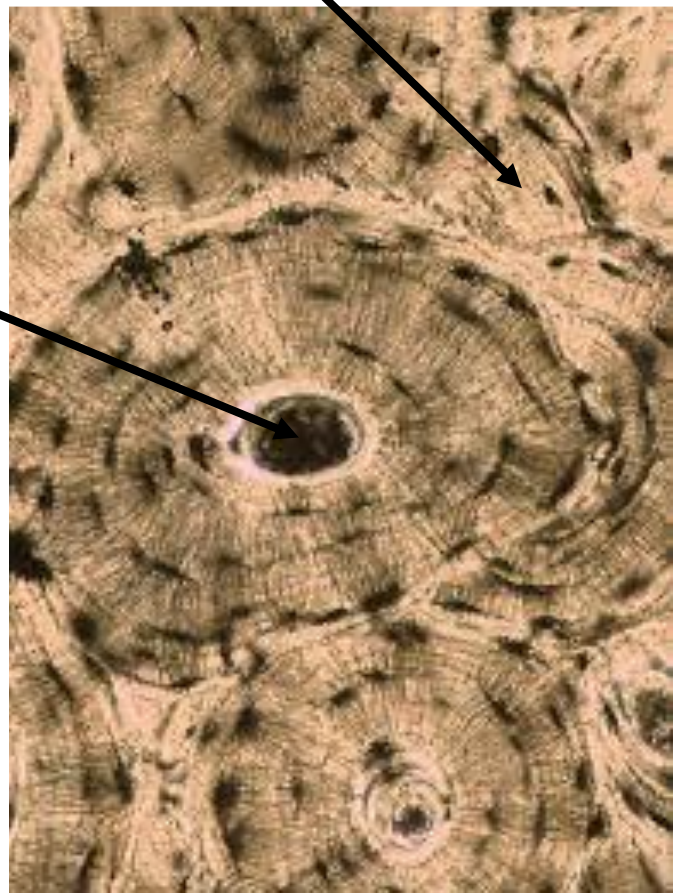
Osteocyte in lacune

Canaliculi ossium

Lamellae

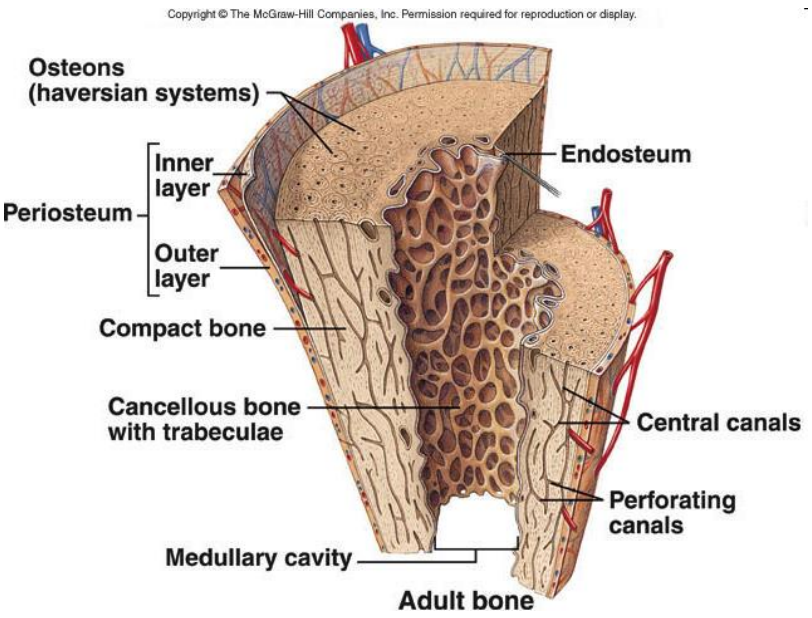
Capillary

Osteoblasts

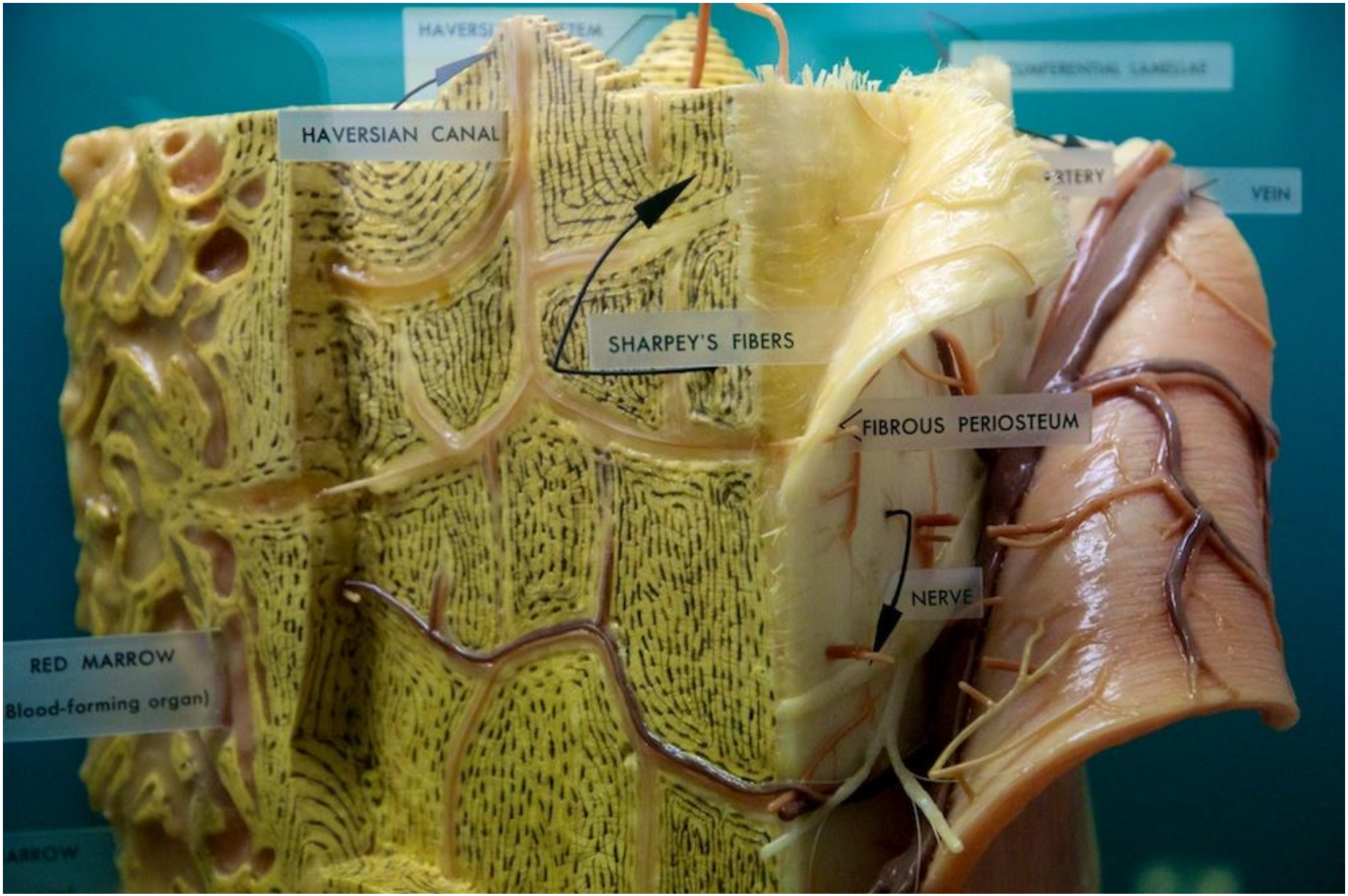


BONE SURFACES

- **Outer surface**
- Synovial joint – hyaline cartilage
- **Periosteum (periost)** – membrane – dense CT, inner layer (osteoblasts) and outer layer (fibrous CT)
- Inactive bone - fibrous CT in periost dominant
- Collagen fibers – parallel to the bone surface
- Sharpey's fibers fix periost to the bone

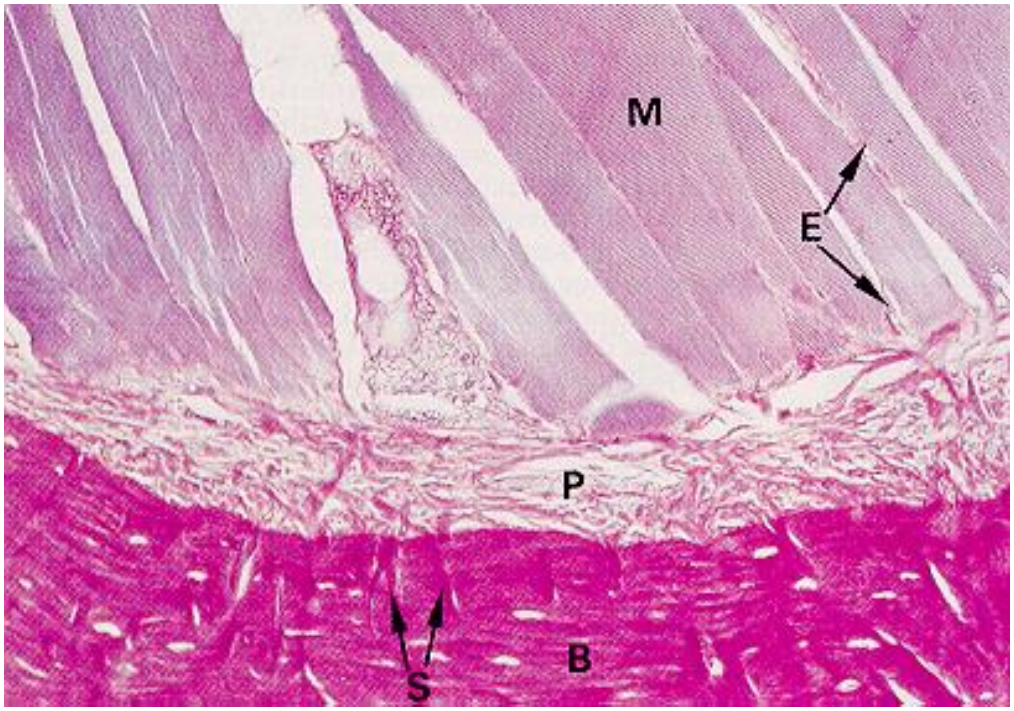


BONE SURFACES

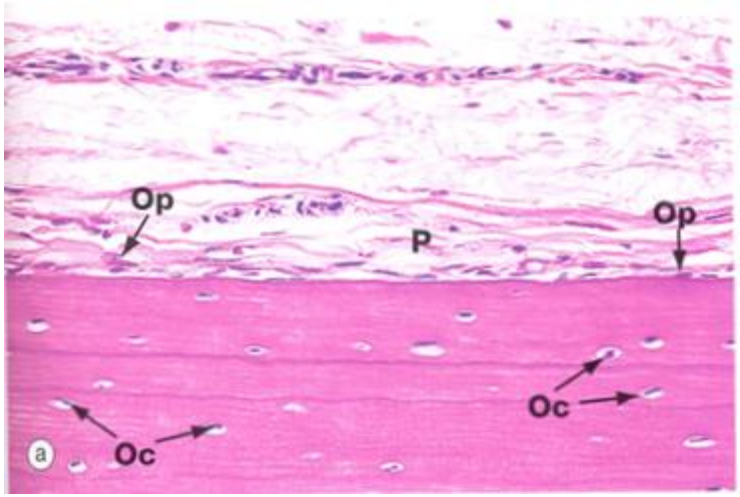


National Museum of Natural History NY, USA

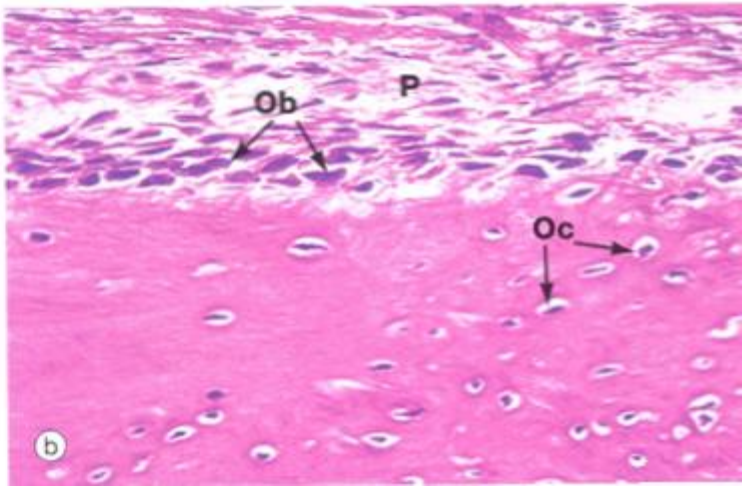
BONE SURFACES



Inactive

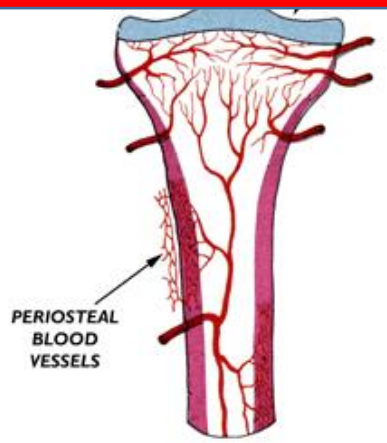


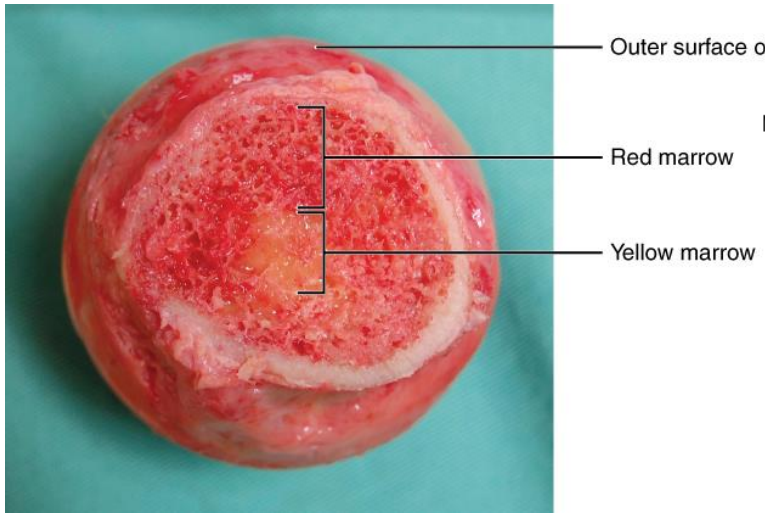
Active

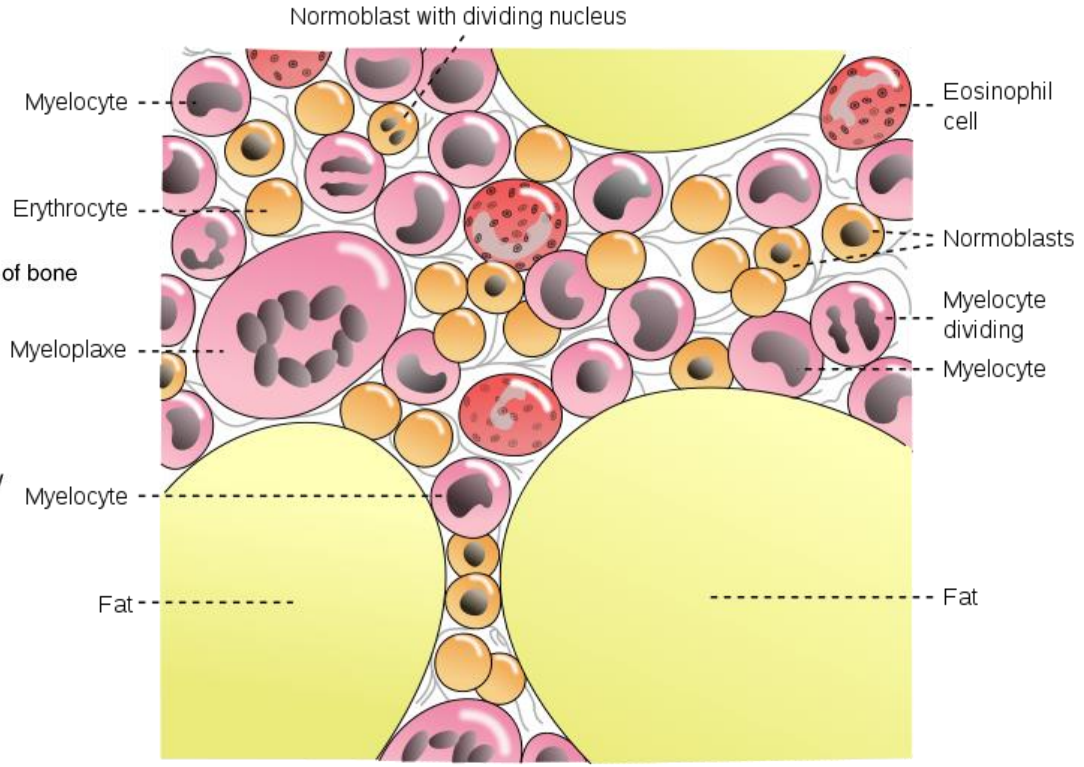


BONE SURFACES

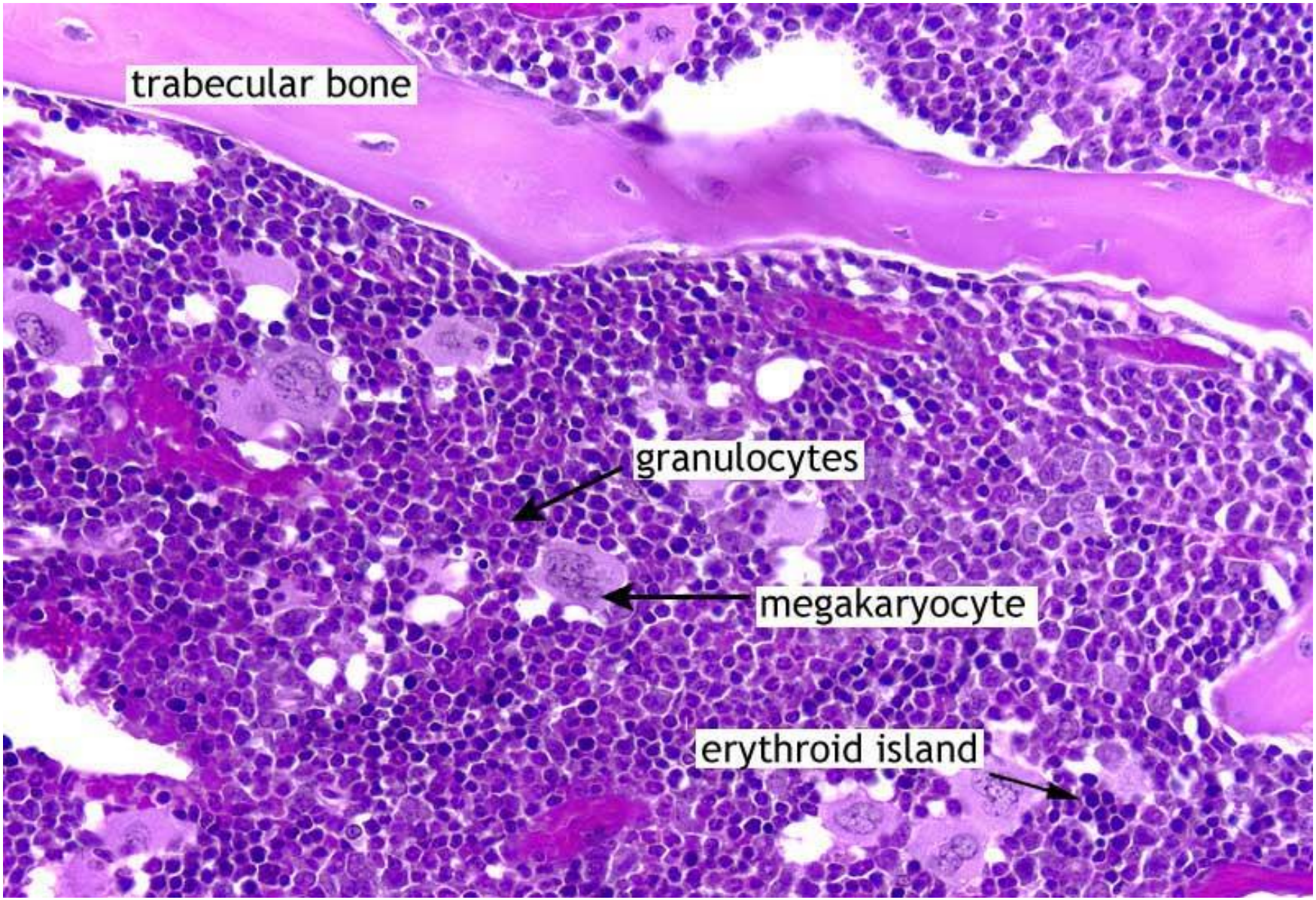
- **Inner surface** – lining of cavities
- medullar cavity
- endosteum (endost) – single cell lining – bone remodeling
- red bone marrow – hematopoiesis
- yellow and gray bone marrow – adipocytes or CT
- rich vascularization
- hematopoietic niche





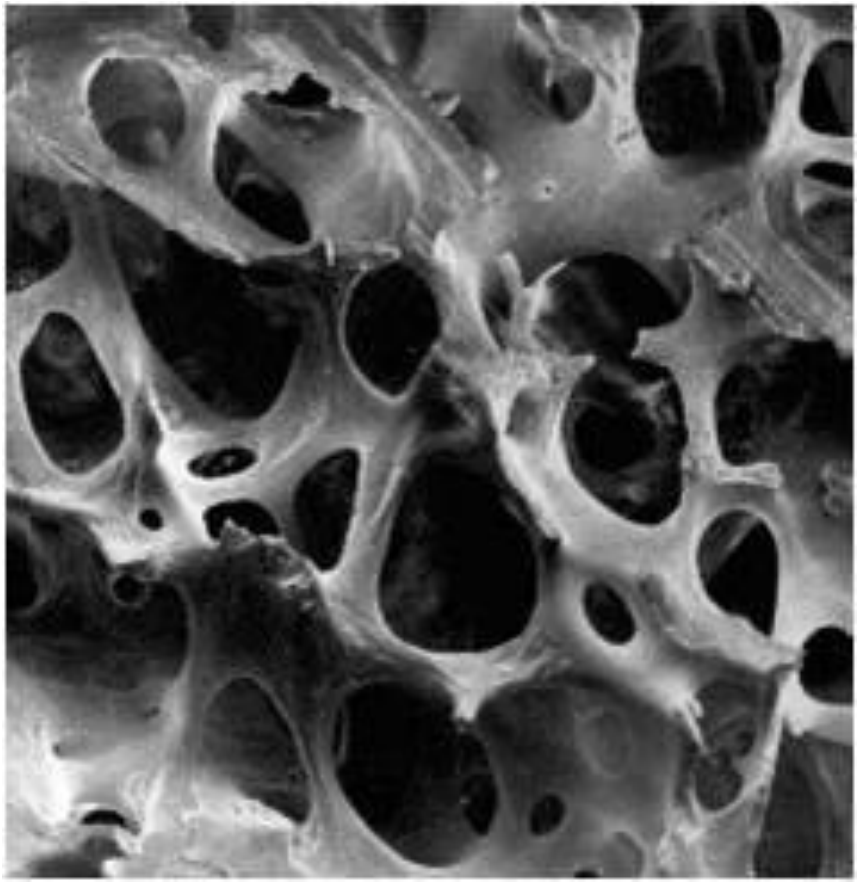


ENDOSTEAL SURFACE OF COMPACT BONE

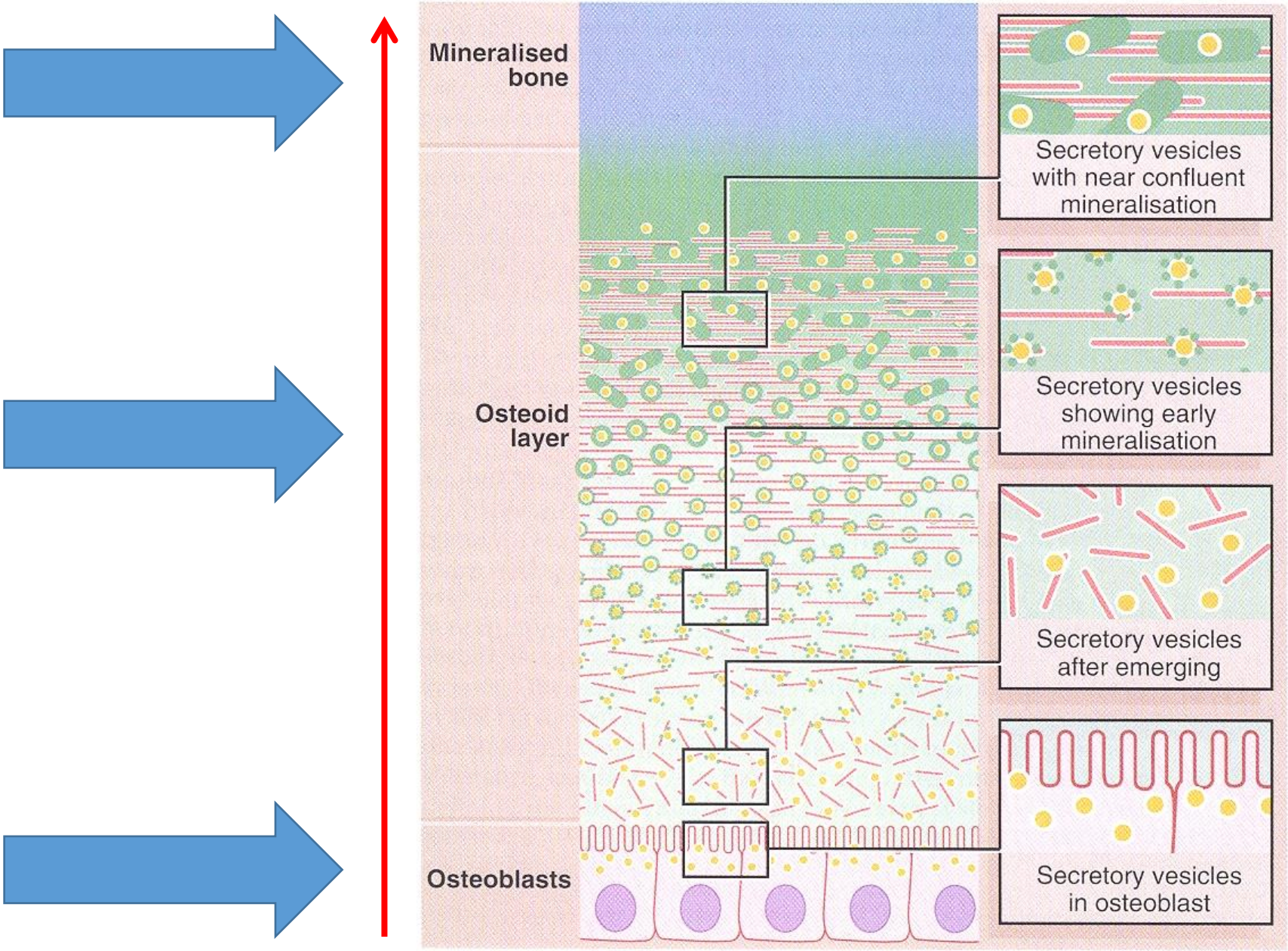


BONE MATRIX

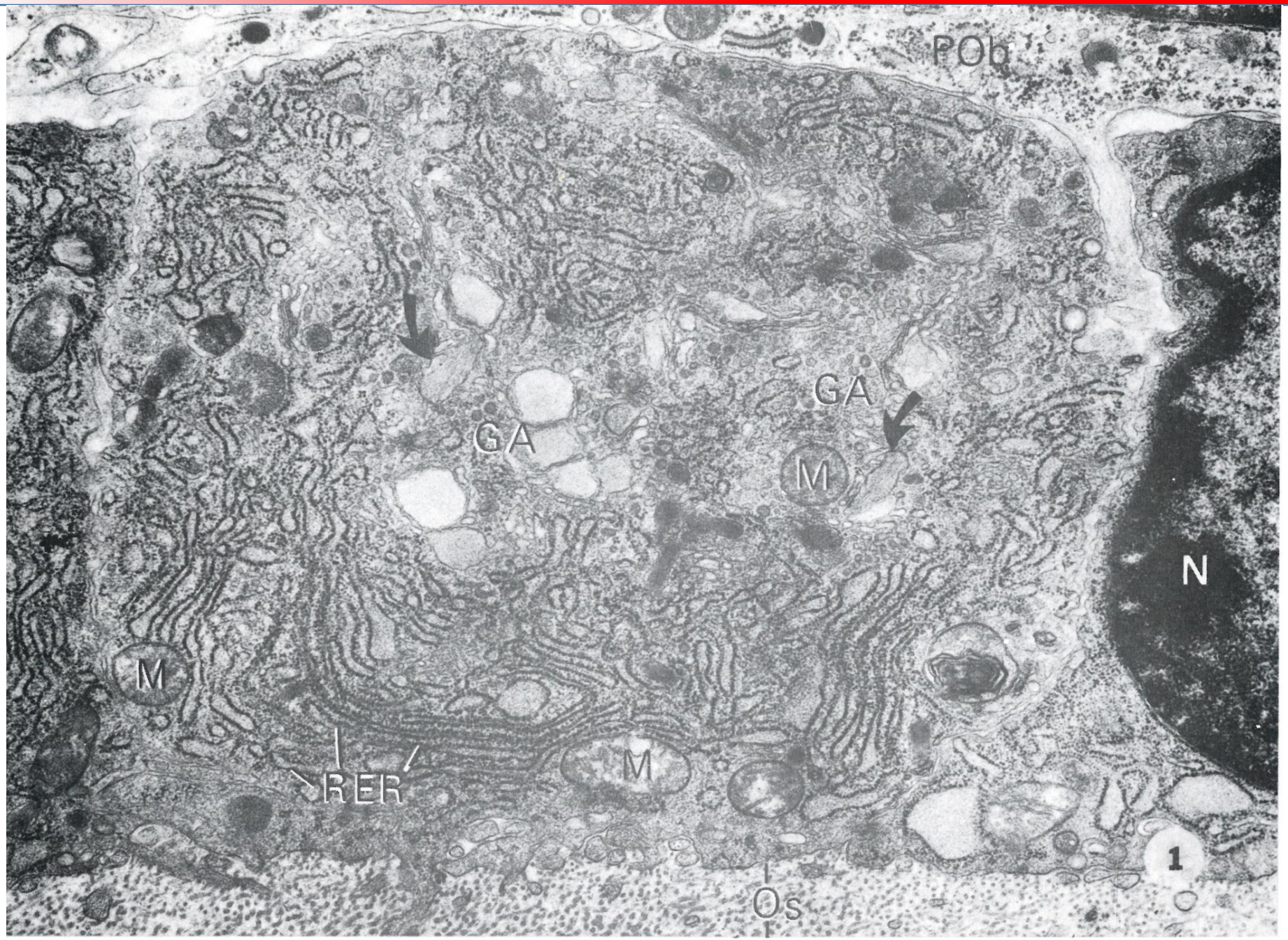
- 60% mineral compound, 24% organic compound 12% H_20, 4% fat
- crystals – calcium phosphate, hydroxyapatite



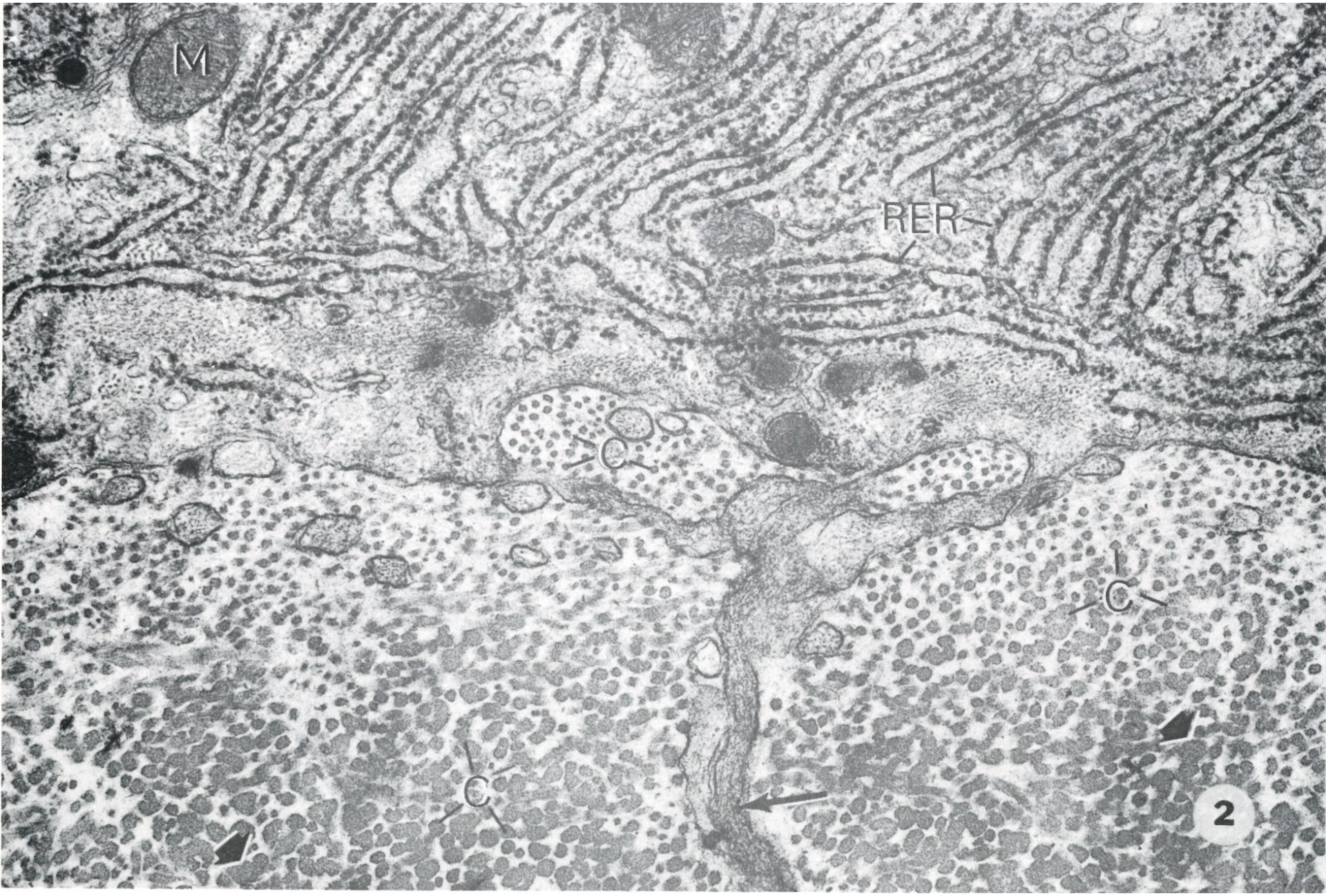
BONE MATRIX


Mineralised bone
Osteoid layer
Osteoblasts
Secretory vesicles with near confluent mineralisation
Secretory vesicles showing early mineralisation
Secretory vesicles after emerging
Secretory vesicles in osteoblast

BONE MATRIX



BONE MATRIX



BONE CELLS – OSTEOBLASTS, OSTEOCYTES, OSTEOCLASTSY

Production and degradation of bone ECM

- collagen (I)
- non-collagenous proteins
- proteoglycans/glycoproteins,
- mineralized matrix

Architecture of mineralized tissue and bone morphology

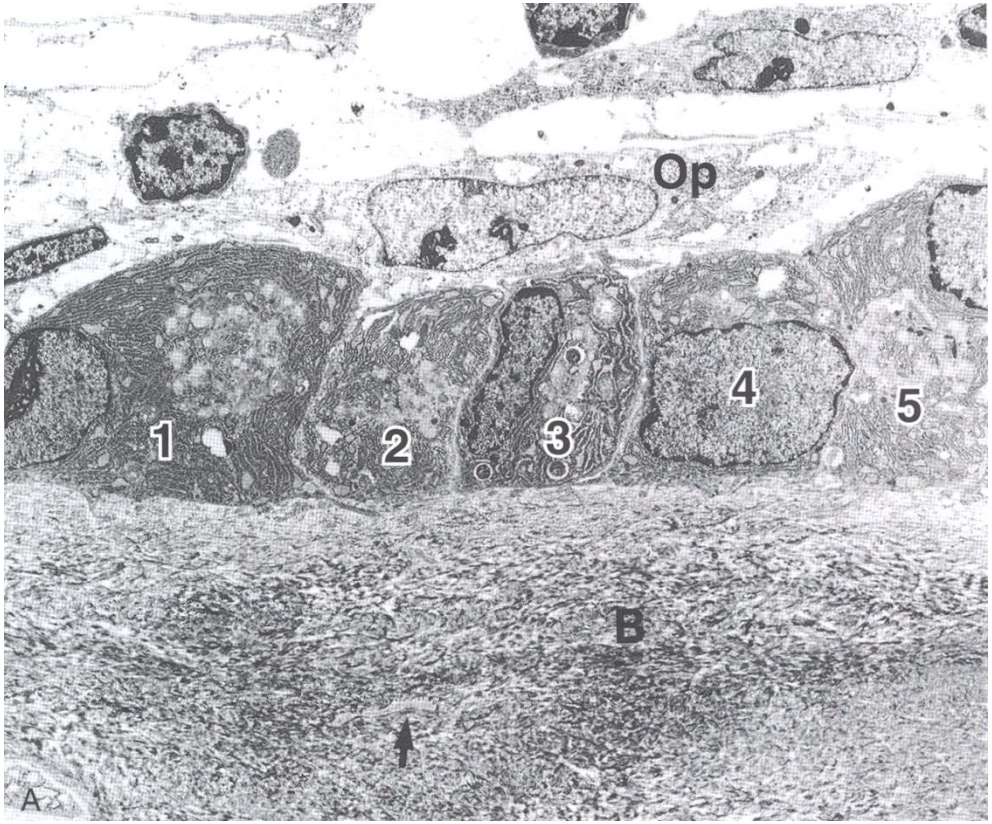
Phosphate and calcium metabolism

Bone marrow and hematopoiesis

10.13140/RG.2.2.20595.32802

BONE CELLS - OSTEOPROGENITORS

- Inner celllayer of perios, Haversian cannals and endost
- origin in embryonic mesenchyme
- mitotic divisions, diferentiaation to osteoblasts

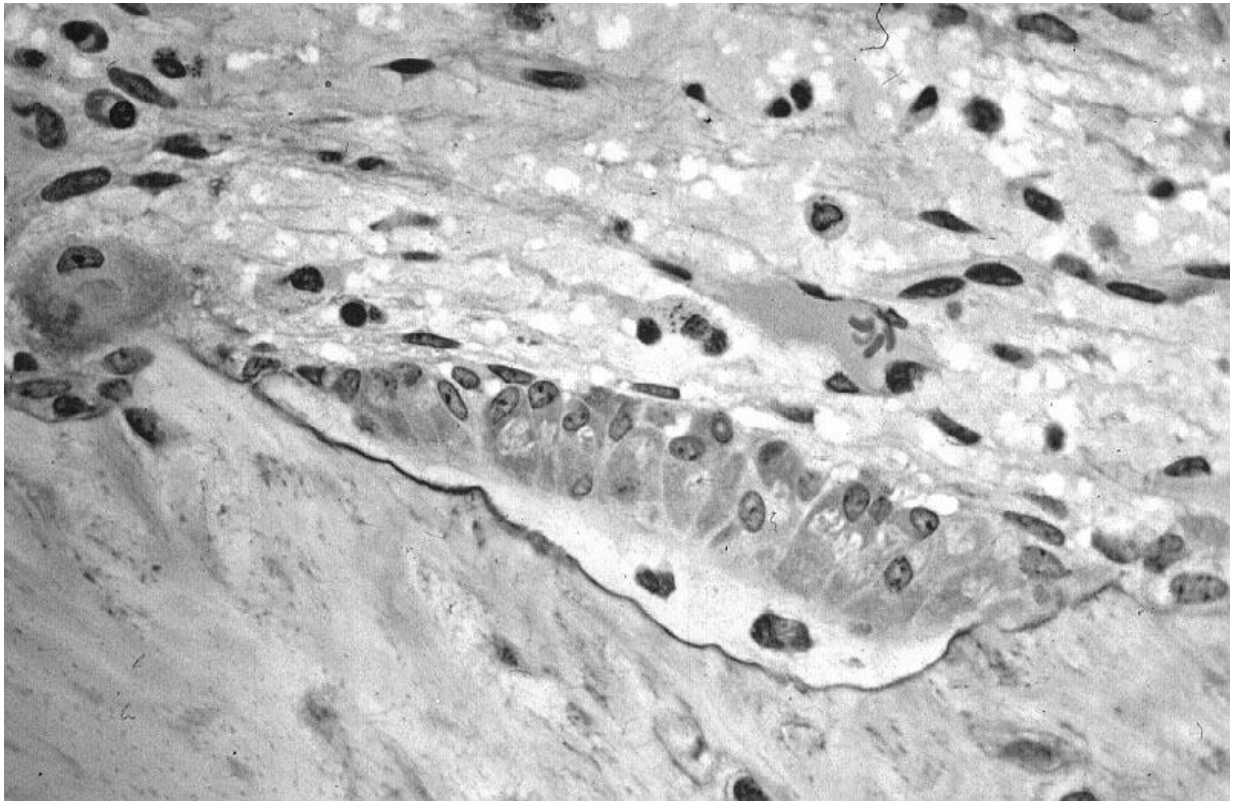


BONE CELLS - OSTEOBLASTS

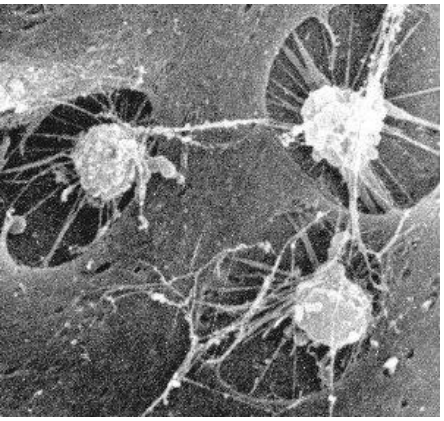
BONE CELLS – OSTEOBLASTS

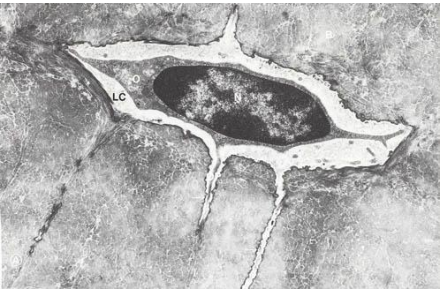
- lining bone surface
- produce ECM – collagen (I) and noncollagenous proteoglycans, glycoproteins
- basophilic cytoplasm, rER, well developed Golgi Apparatus
- euchromatin nucleus

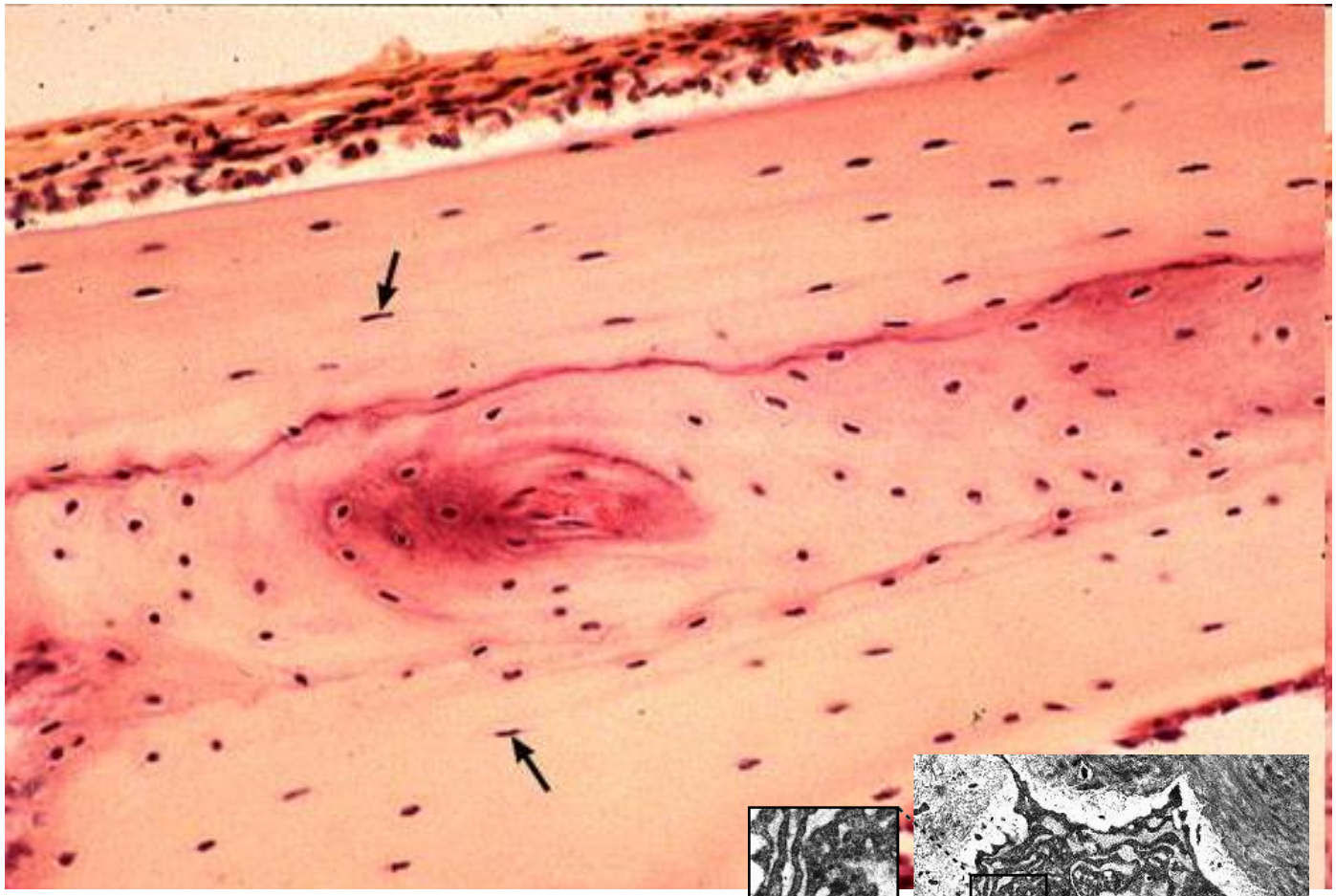
- **osteocytes** embedded in matrix
- *canalliculi ossium*



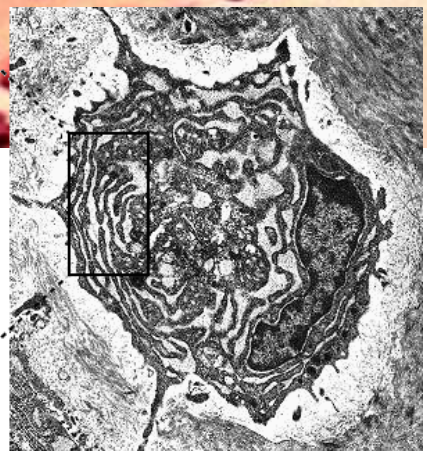
BONE CELLS – OSTEOCYTES



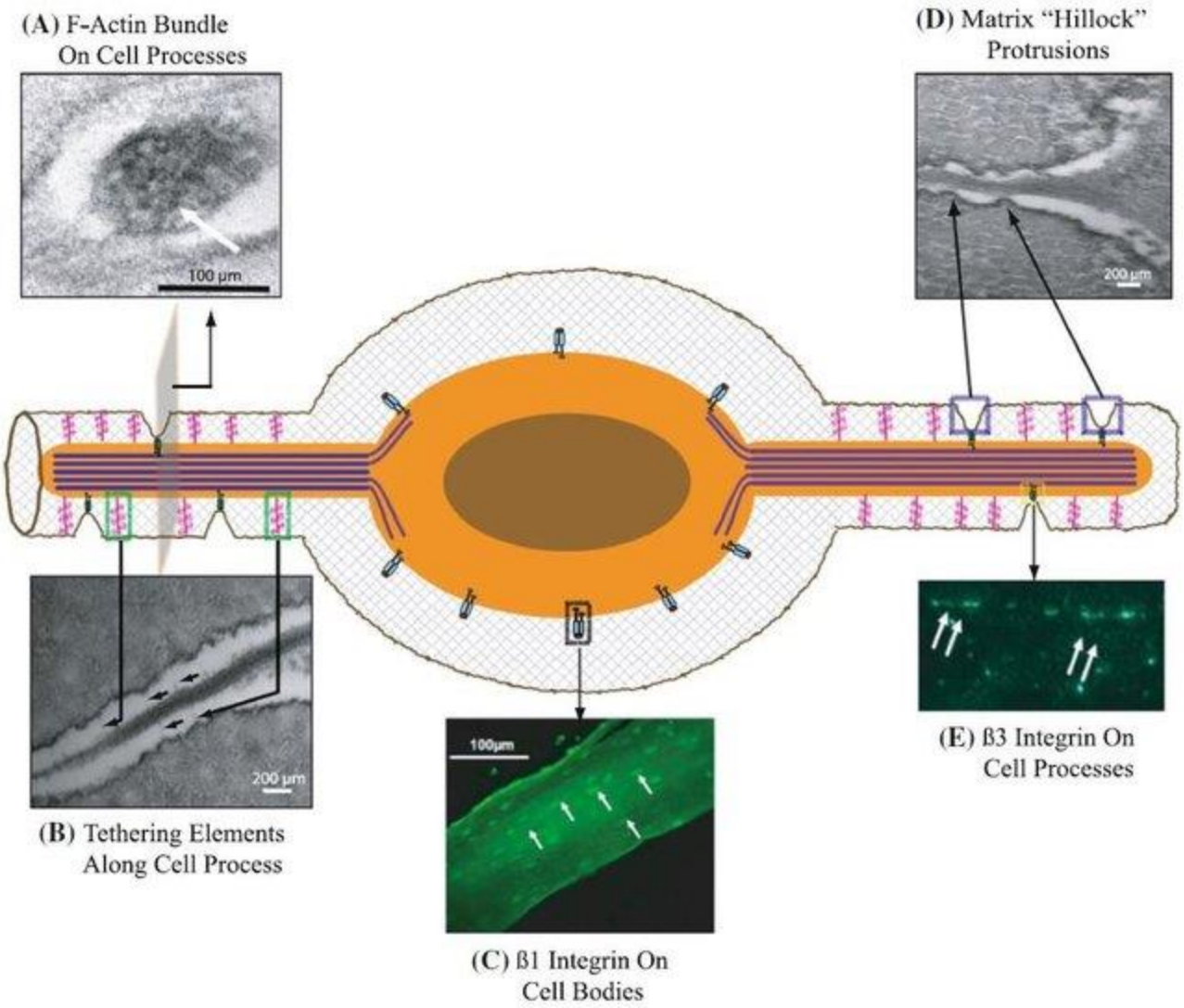




RER
-rough
endoplasmic
reticulum



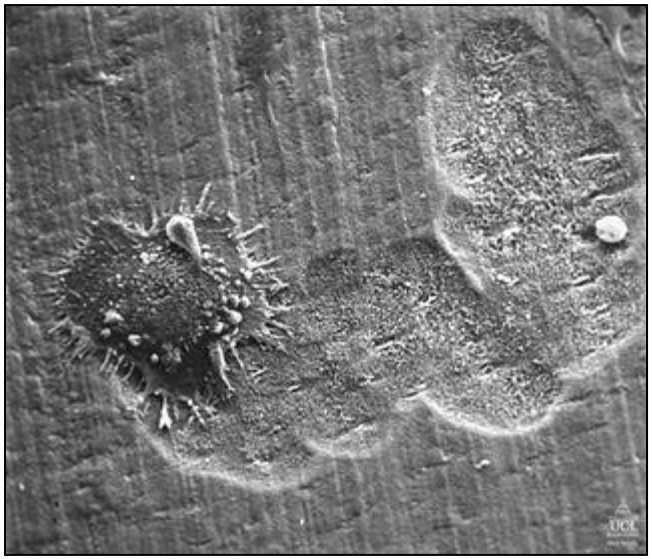
BONE CELLS - OSTEOCYTES

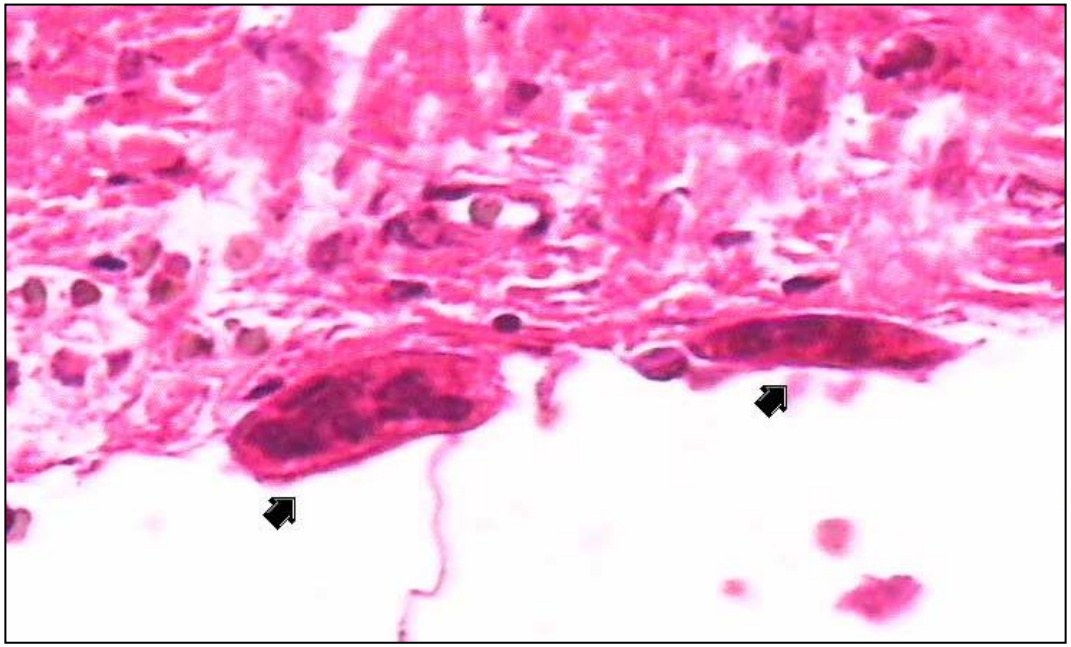


10.13140/RG.2.2.20595.32802

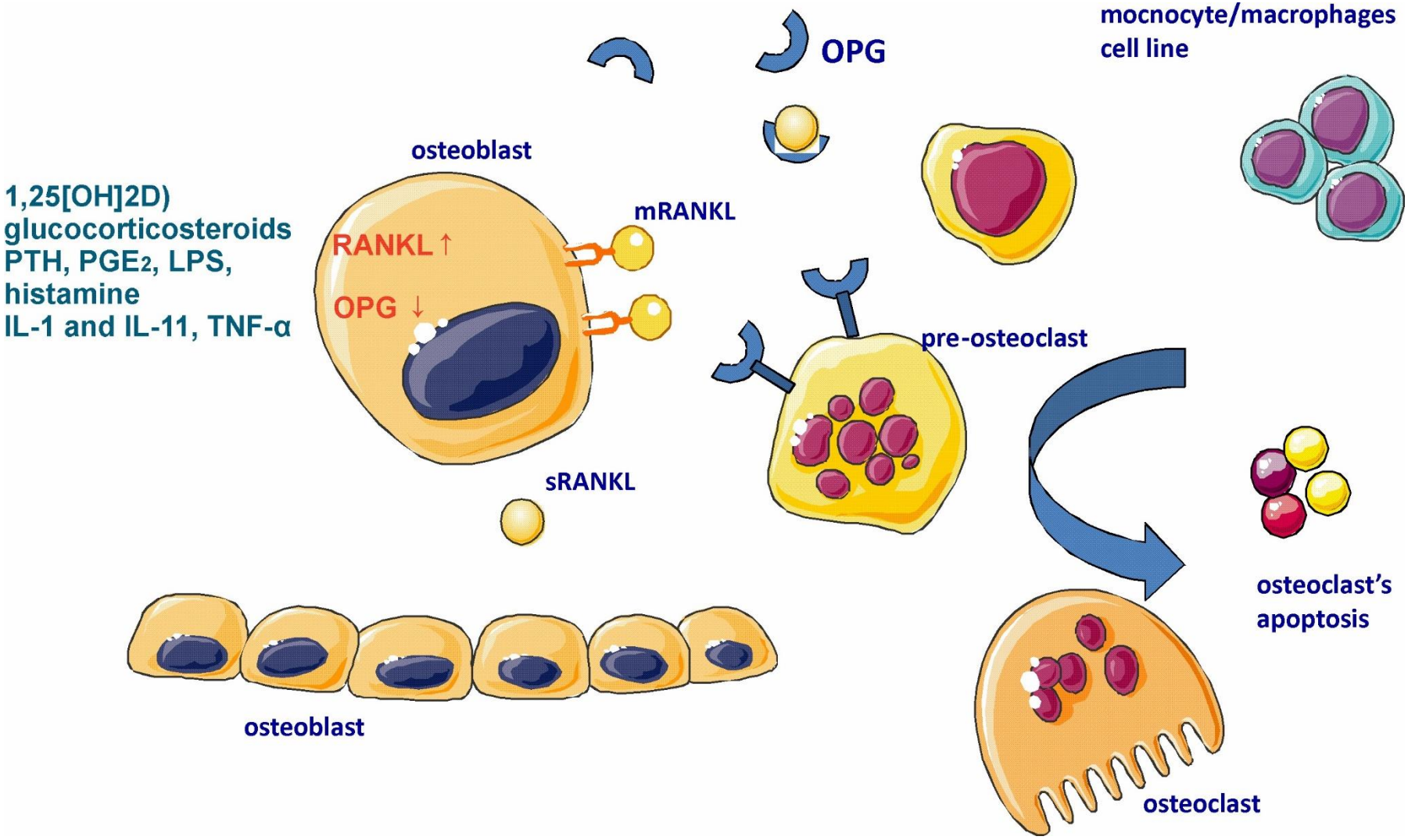
BONE CELLS – OSTEOCLASTS

- multinuclear, formed by fusion of mononuclear macrophages
- bone matrix resorption



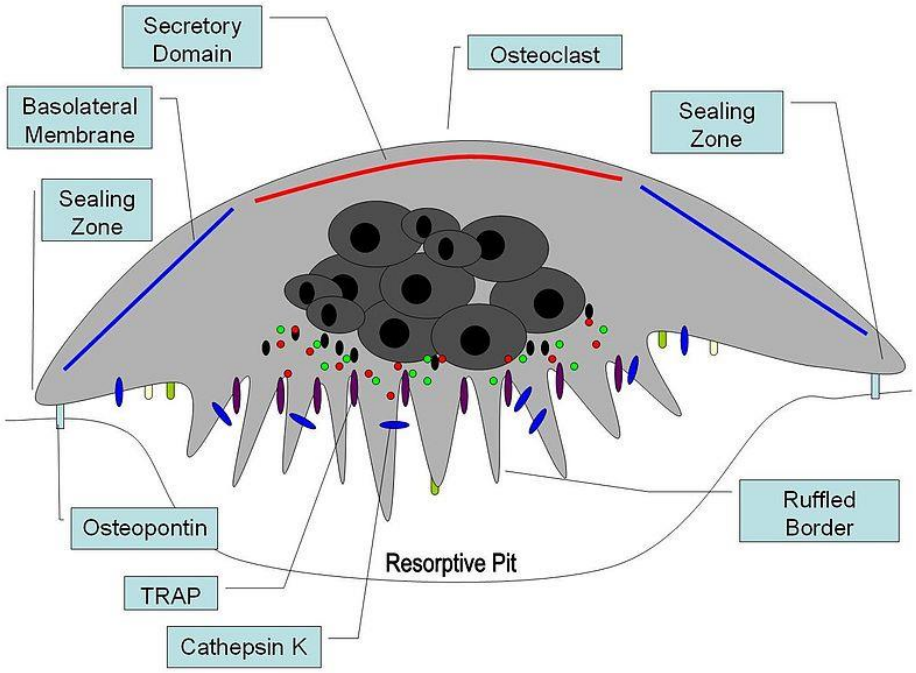


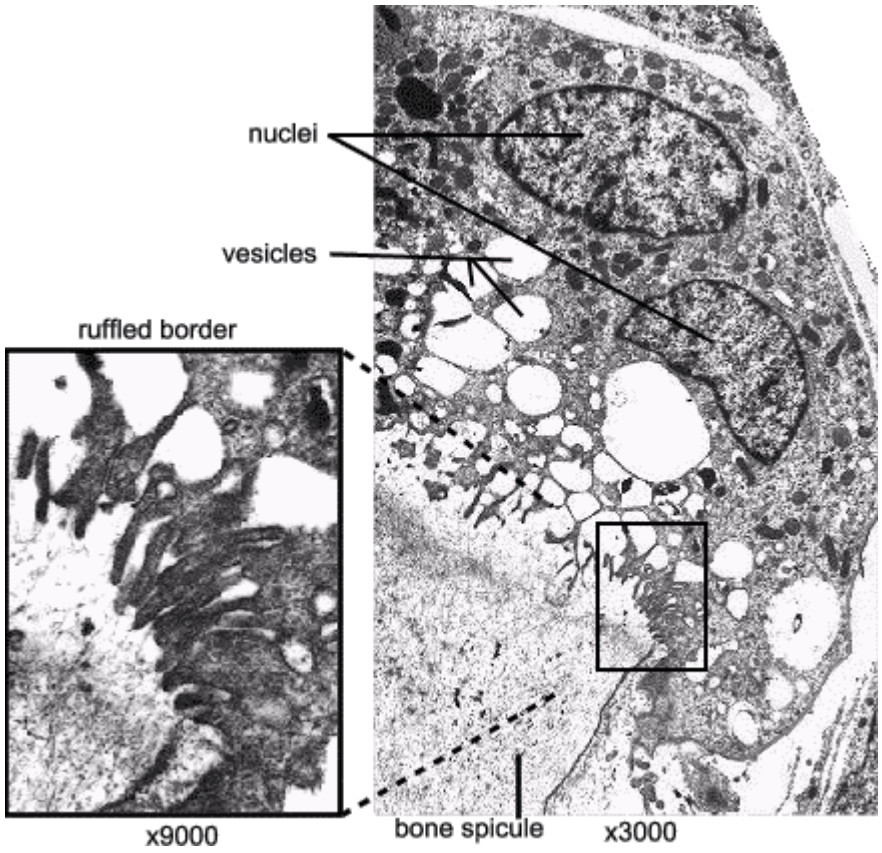
BONE CELLS – OSTEOCLASTS



BONE CELLS – OSTEOCLASTS

- complex architecture
- enzymes degrading organic matrix
- HCl

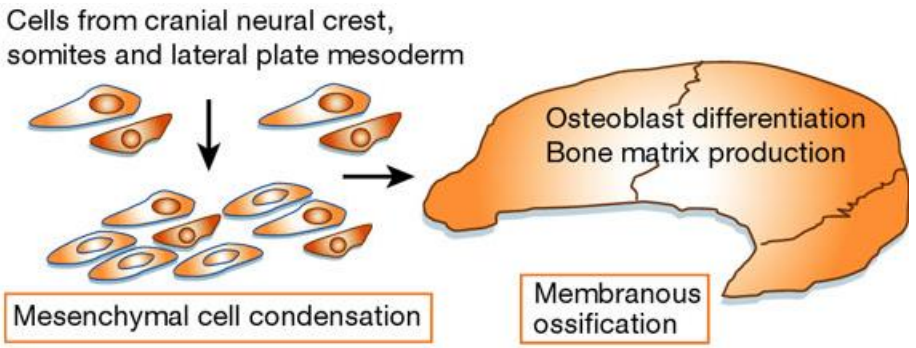




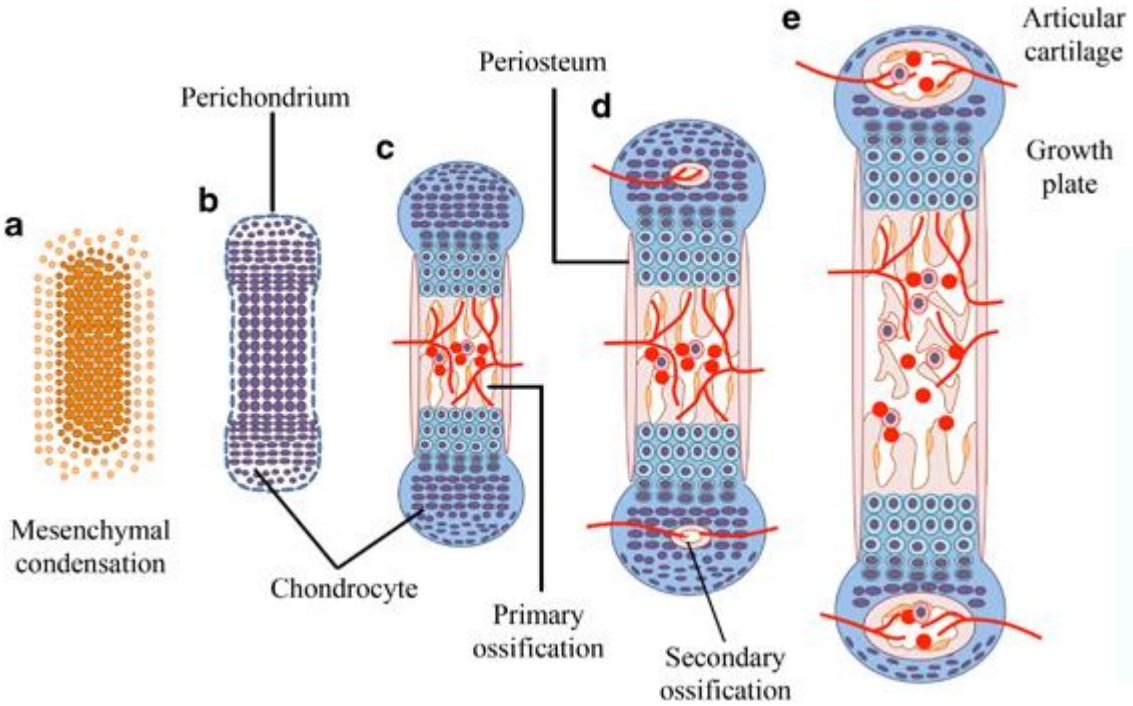
BONE CELLS - OSTEOCLASTS

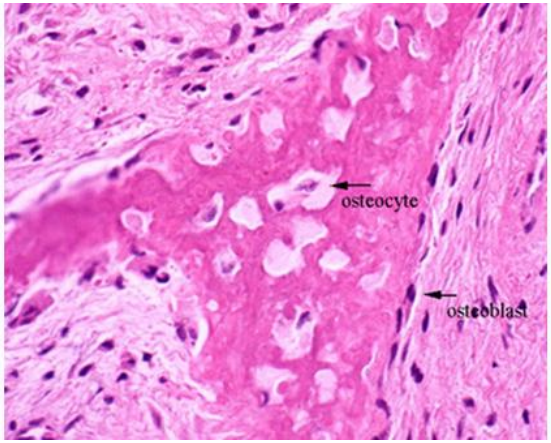
BONE OSSIFICATION

- Intramembraneous

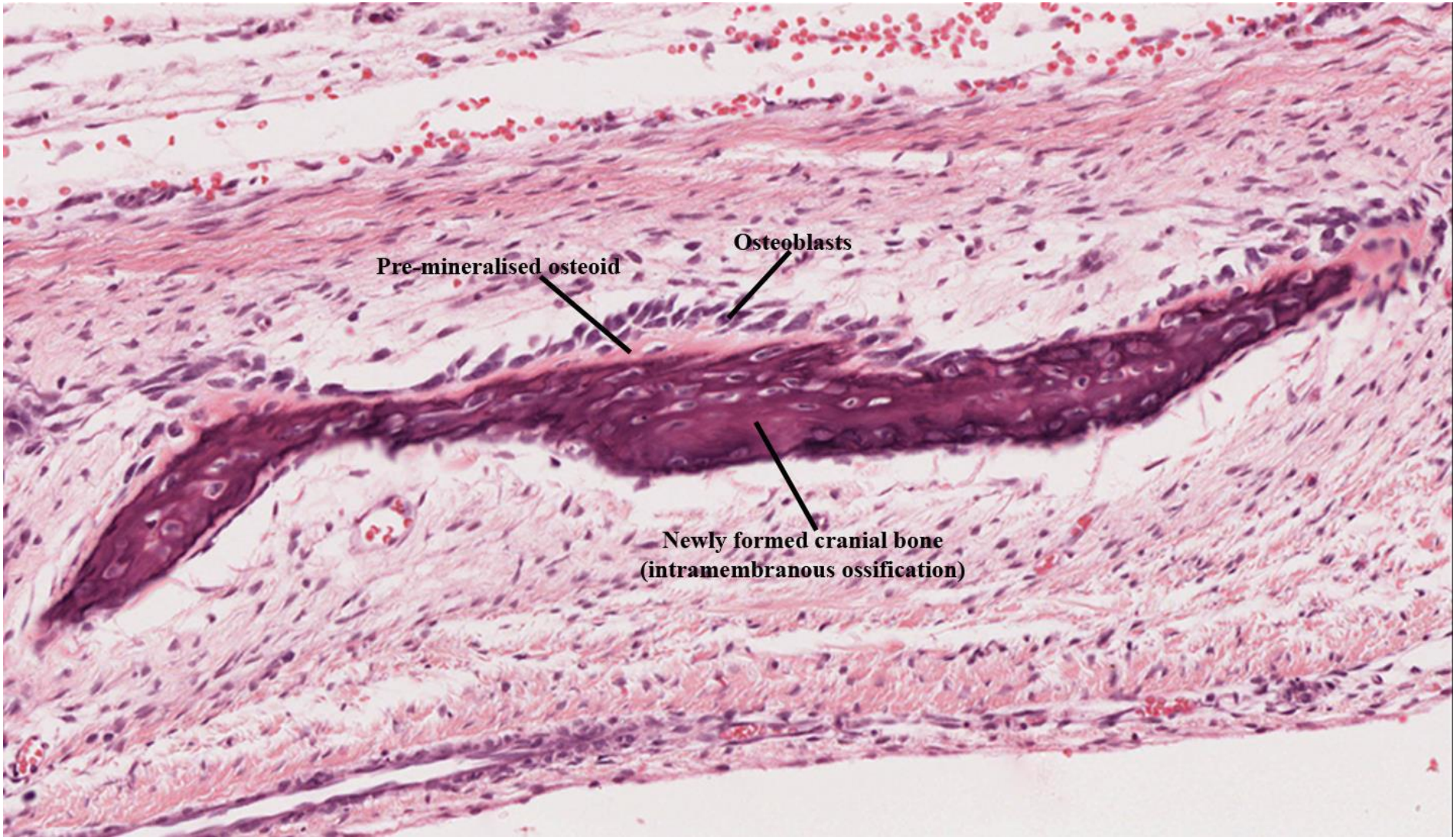


- Endochondral

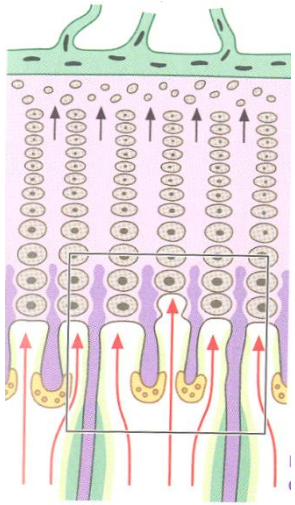


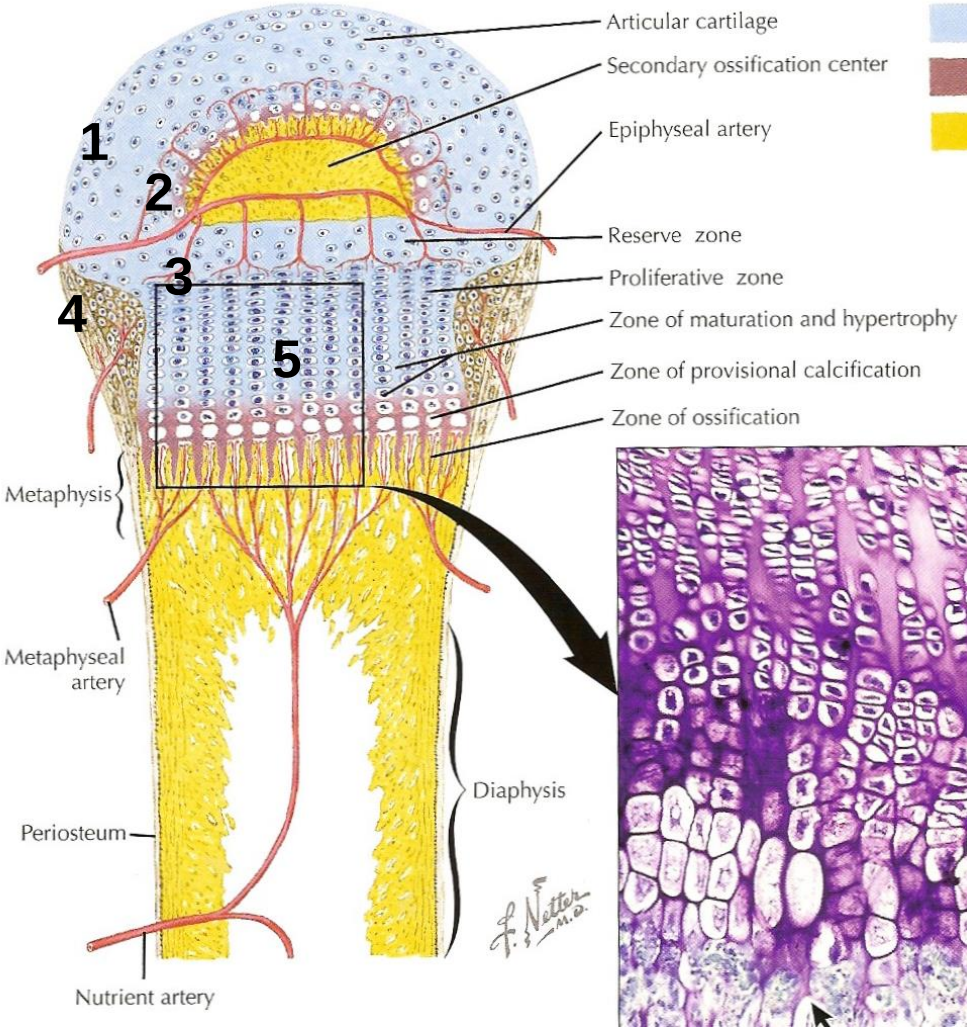


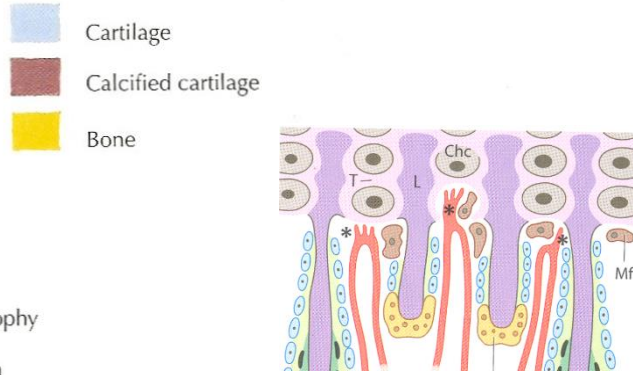
INTRAMEMBRANEOUS OSSIFICATION

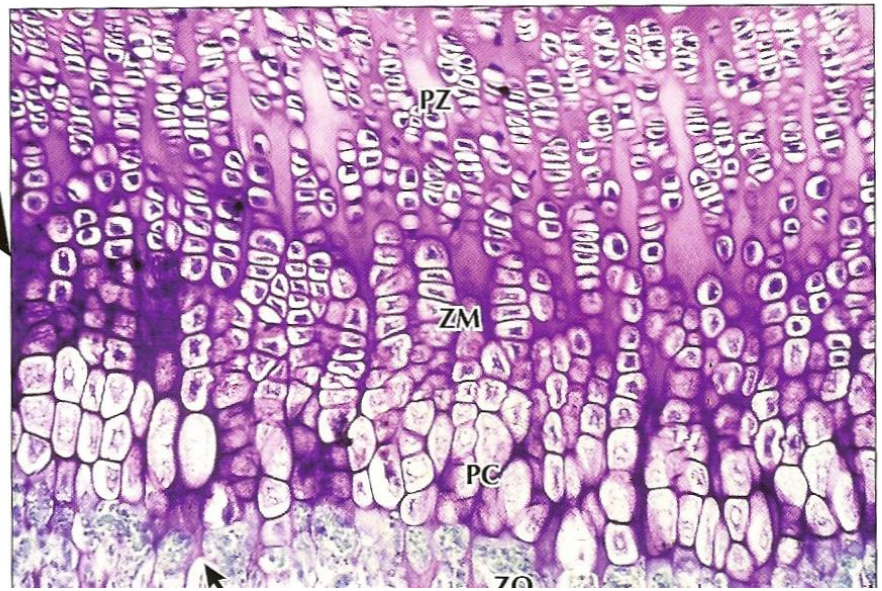


ENDOCHONDRAL OSSIFICATION









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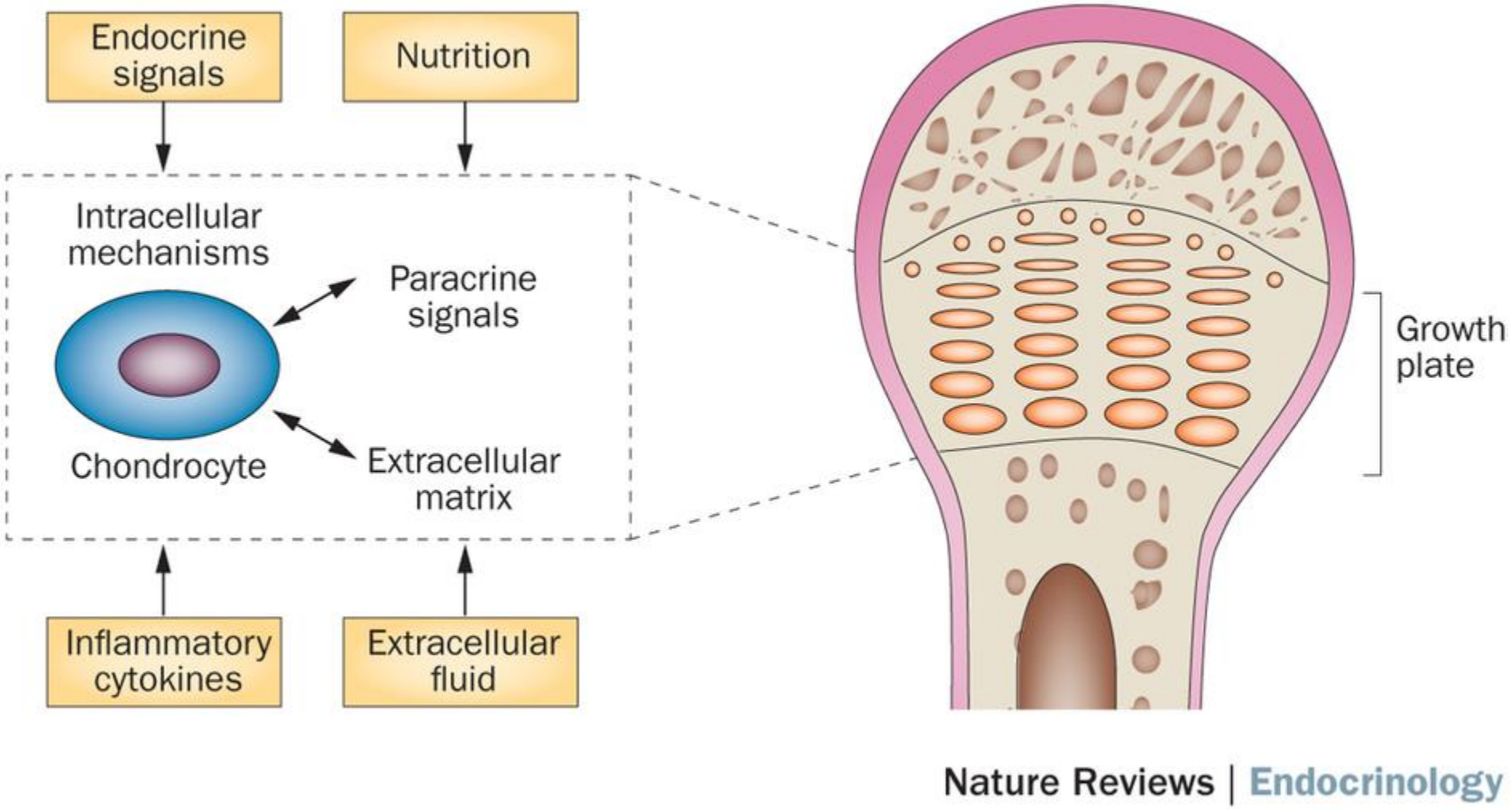

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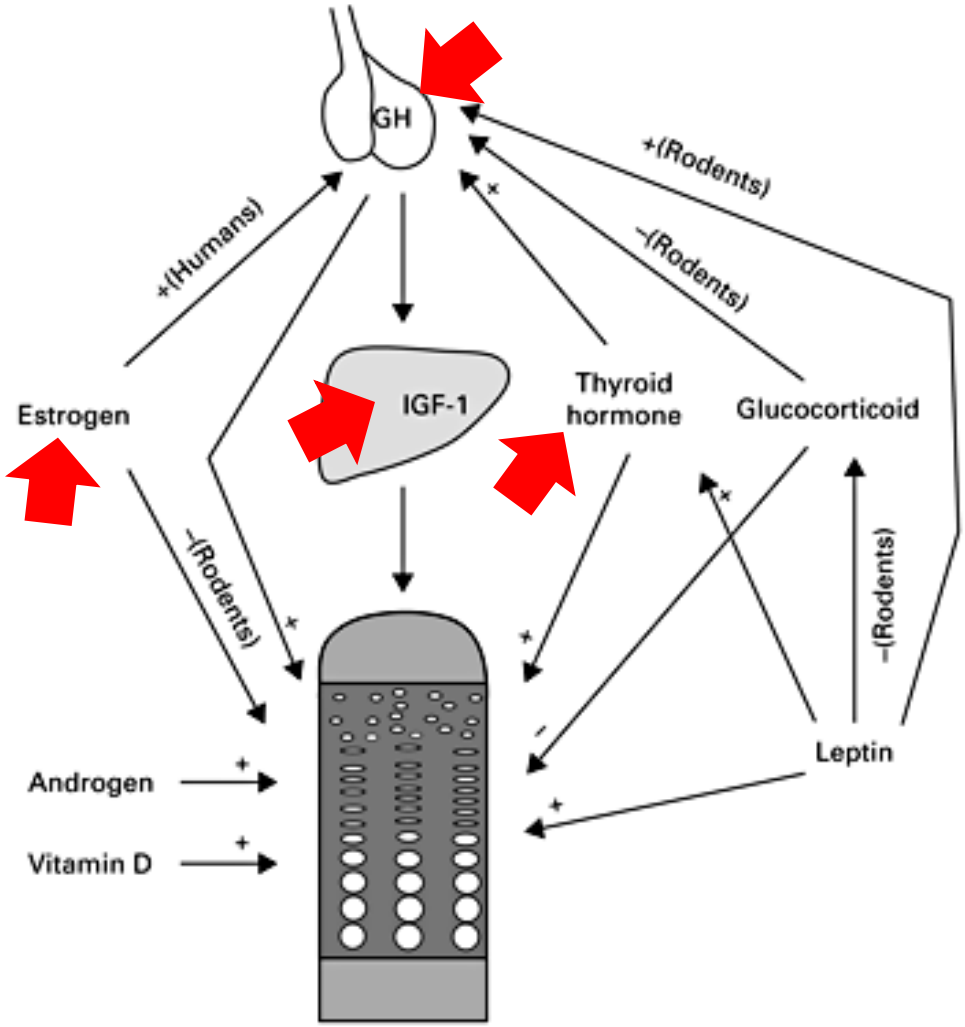
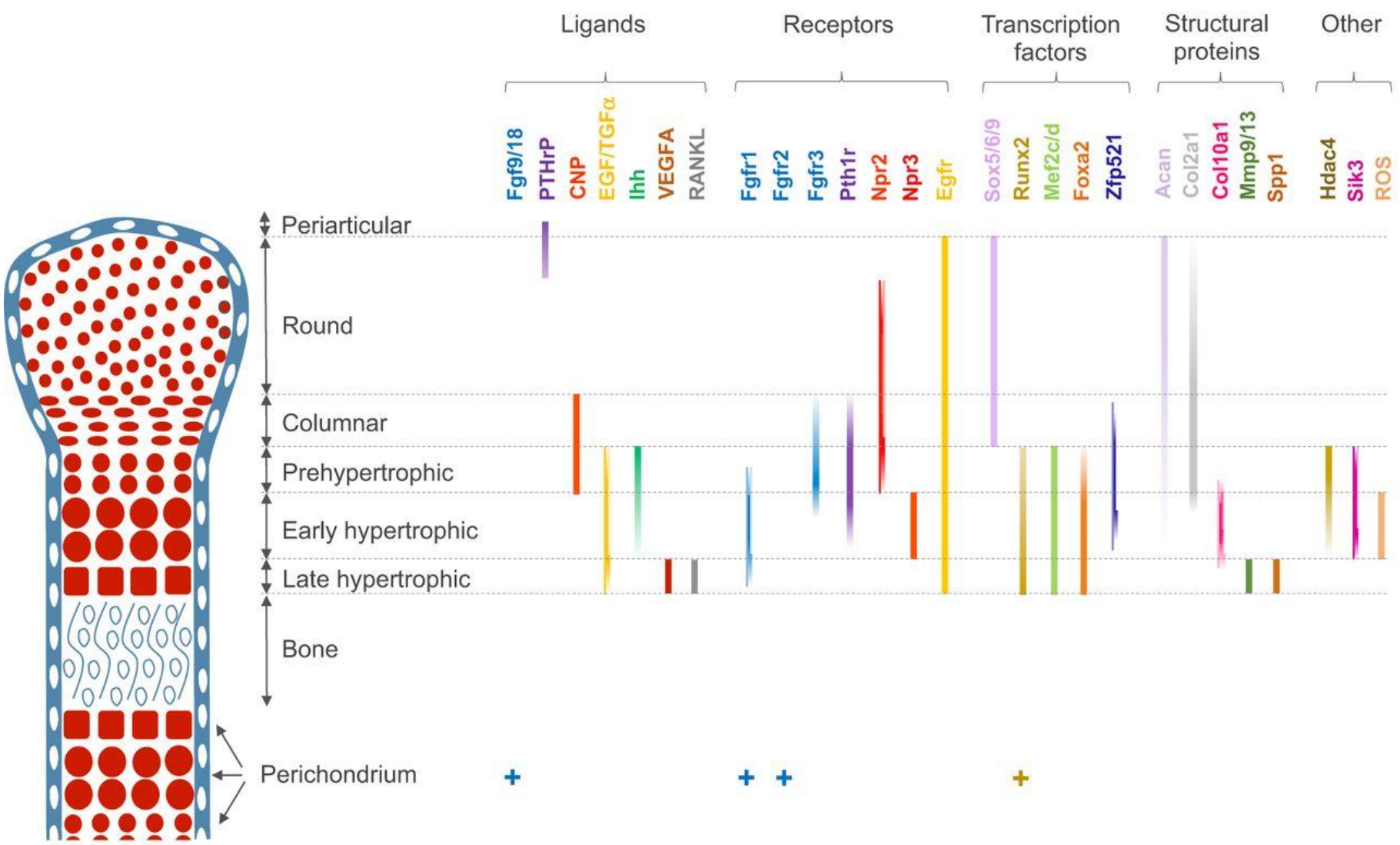


	Local effects on the growth plate
GH	Proliferation of resting zone chondrocytes Stimulates local IGF-1 expression
IGF-1	Increases proliferation of resting and proliferative chondrocytes Increases hypertrophic cell size
Glucocorticoid	Inhibits chondrocyte proliferation Delays growth plate senescence Induces chondrocyte apoptosis
Thyroid hormone	Permissive for proliferation and differentiation
Estrogen	Inhibits proliferation in the proliferative zone Accelerates growth plate senescence
Androgen	Stimulates proliferation, matrix production Increases IGF-1 expression
Vitamin D	Permissive for normal differentiation and apoptosis of hypertrophic chondrocytes
Leptin	Stimulates proliferation and differentiation

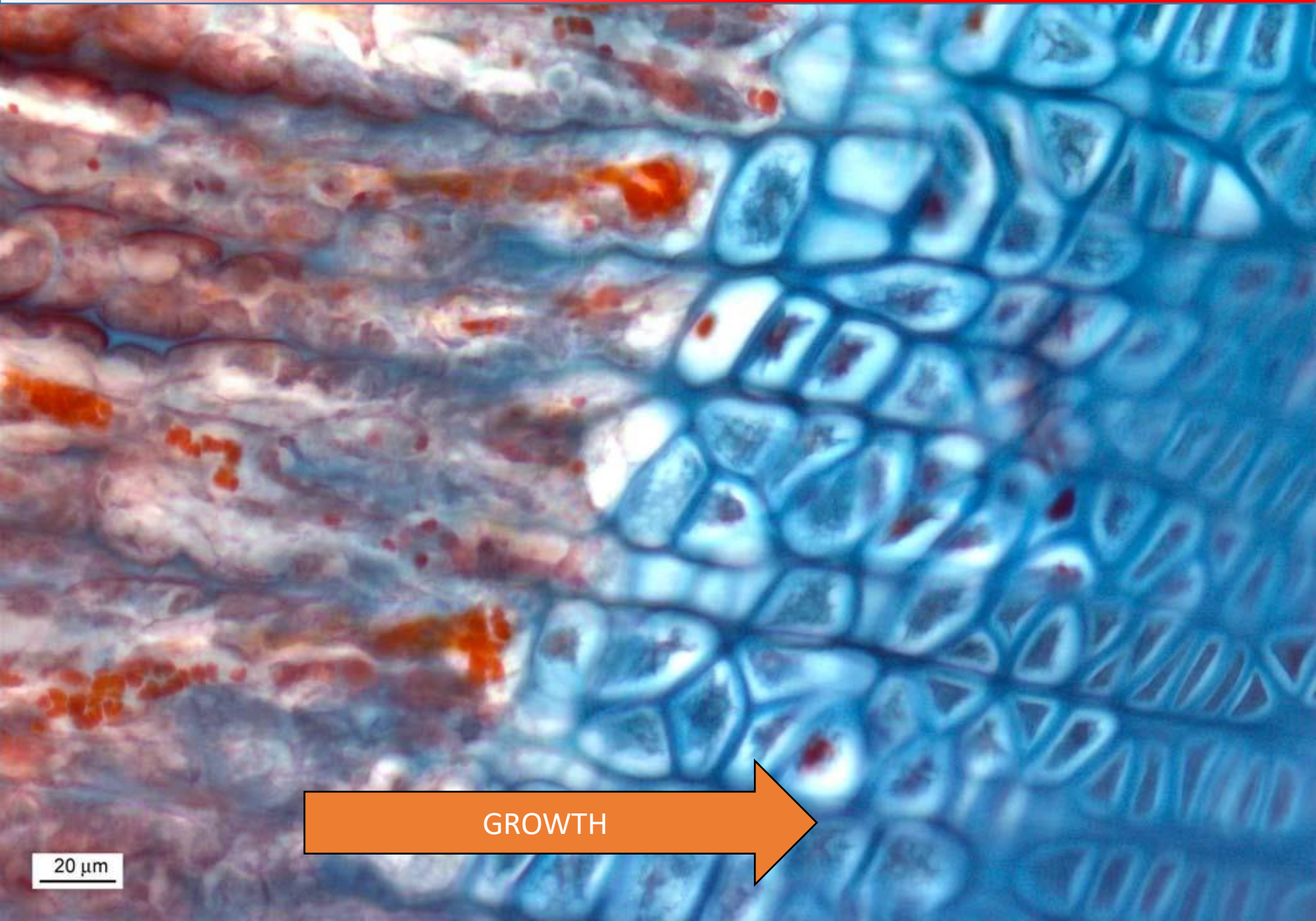
https://doi.org/10.1159/000088791

ENDOCHONDRAL OSSIFICATION

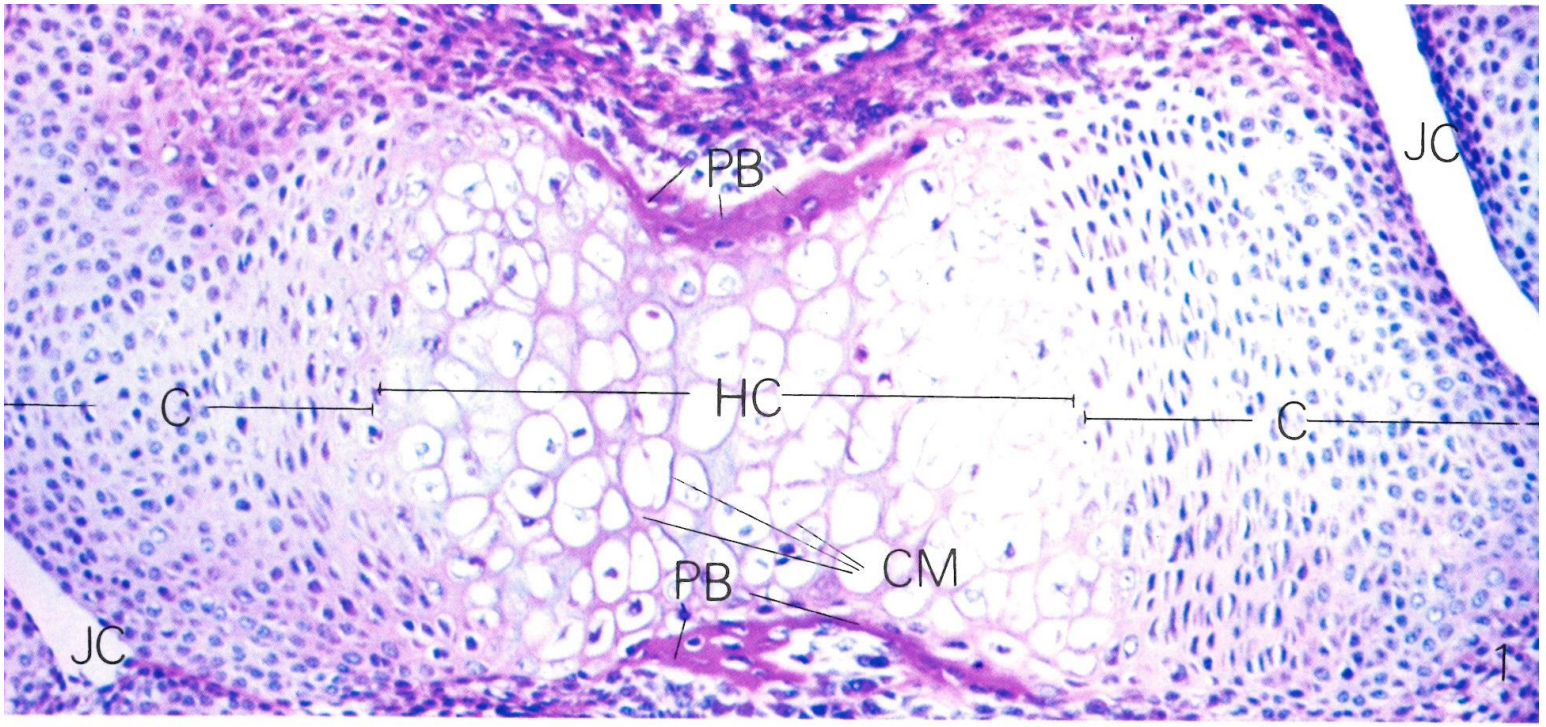


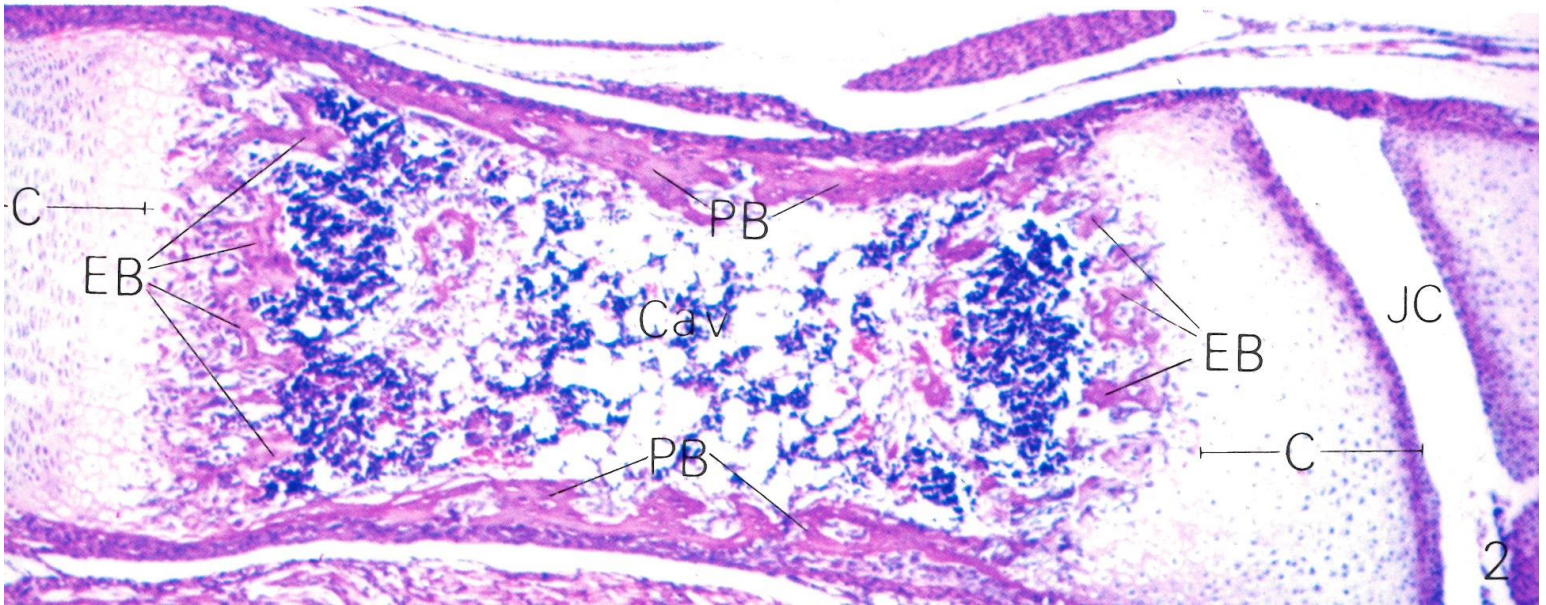
GROWTH PLATE

GROWTH PLATE

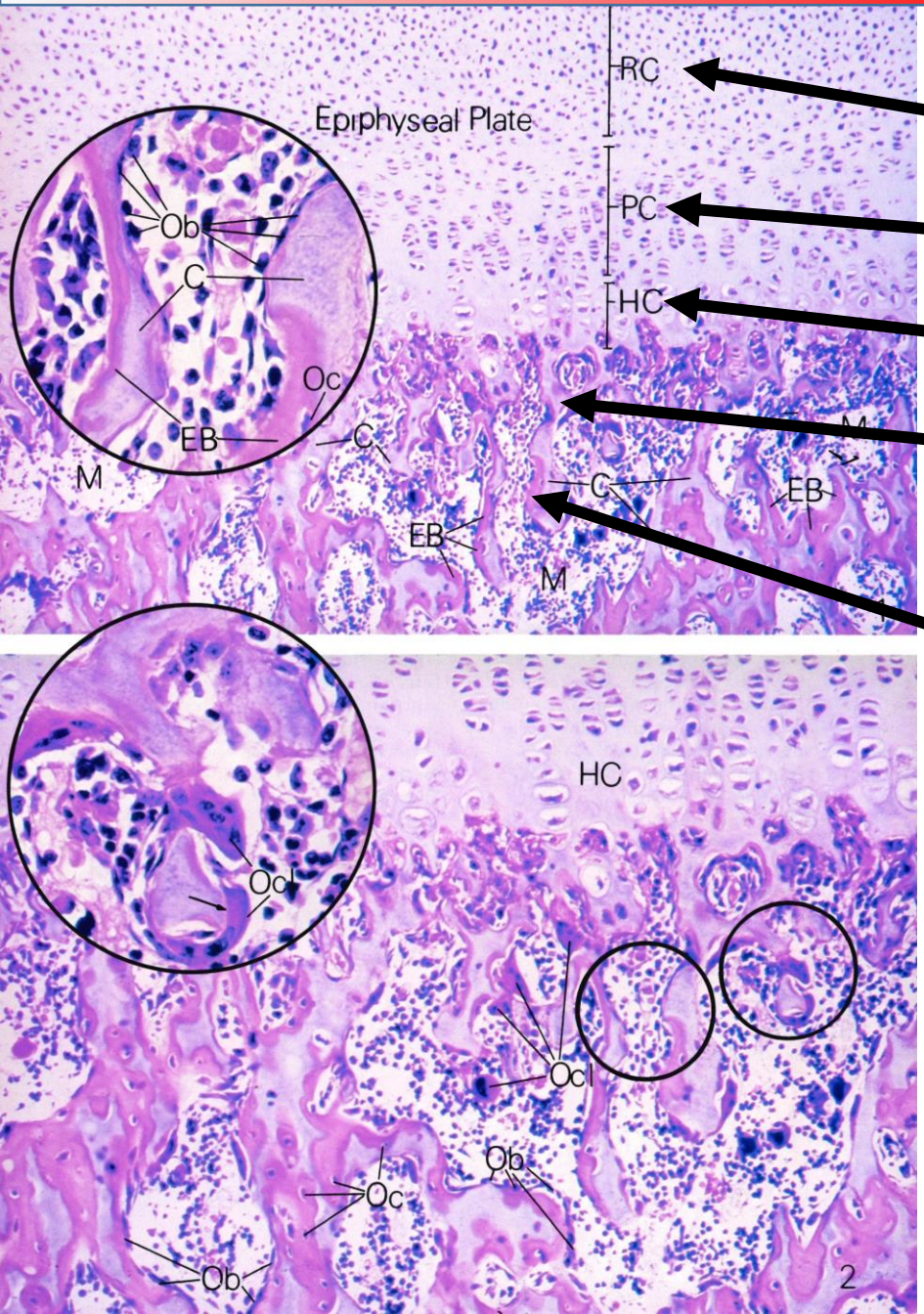


GROWTH PLATE





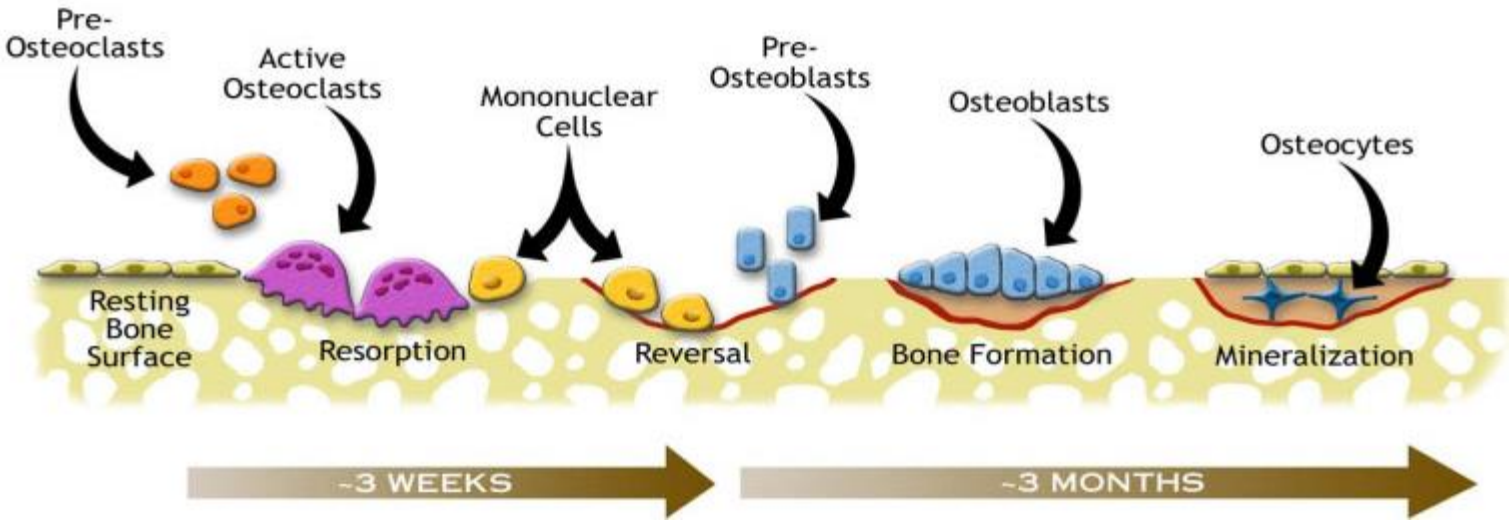
GROWTH PLATE

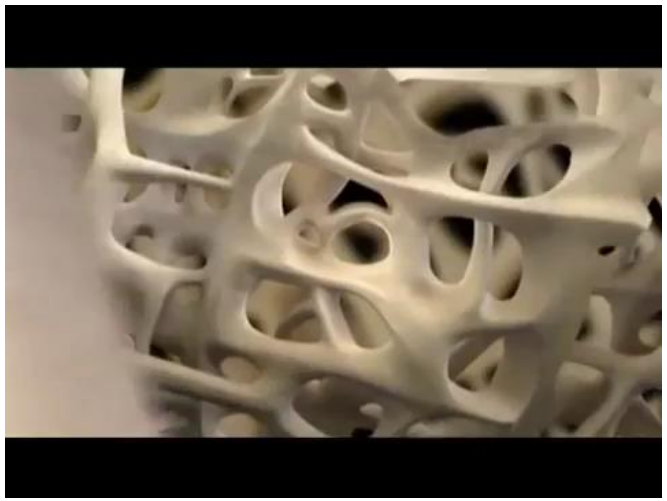


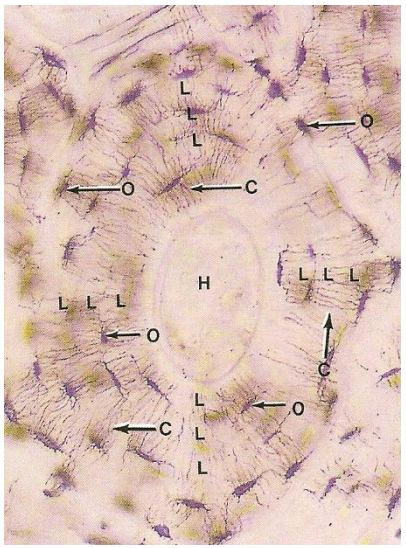
- Reserve zone
- Zone of proliferation
- Zone of hypertrophy
- Zone of calcification
- Line of erosion
- Zone of ossification

Ossification in growth plate is a 3D phenomenon

BONE REMODELLING

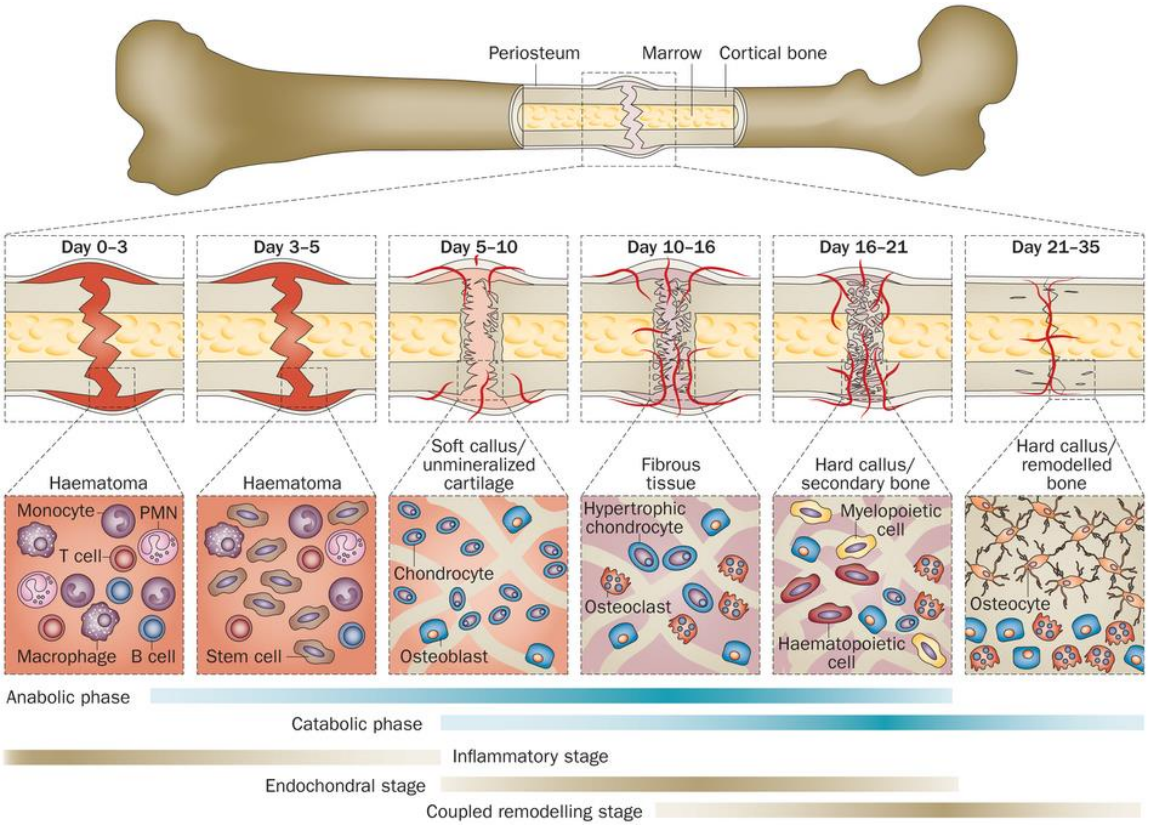






http://ns.umich.edu/Releases/2005/Feb05/img/bone.jpg

CLINICAL CORRELATIONS – FRACTURE HEALING



Reactive phase

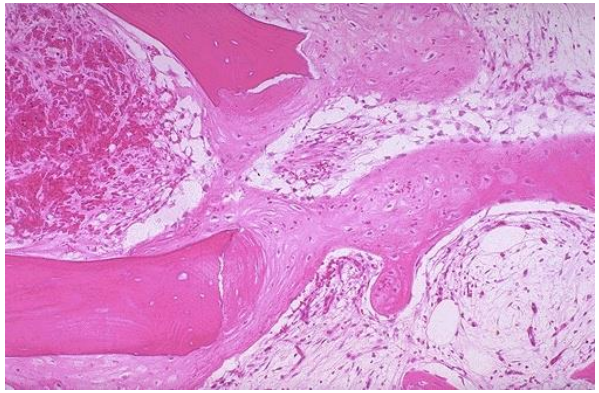
- fracture and inflammatory phase
- granulation tissue formation

Reparative phase

- cartilage *callus* formation
- lamellar bone deposition

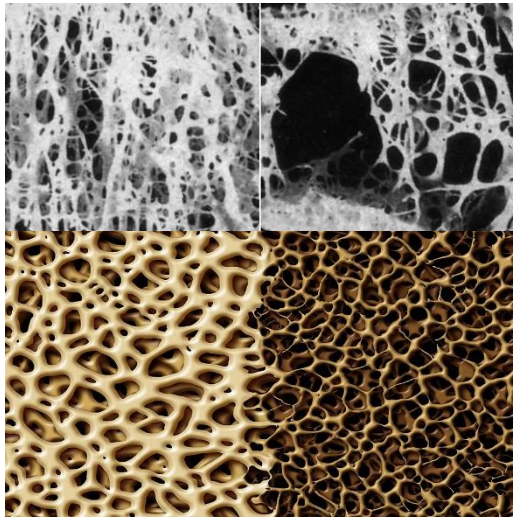
Remodeling phase

- remodeling to original bone shape



CLINICAL CORRELATIONS – DISBALANCE OF BONE HOMEOSTASIS

- **OSTEOPOROSIS**

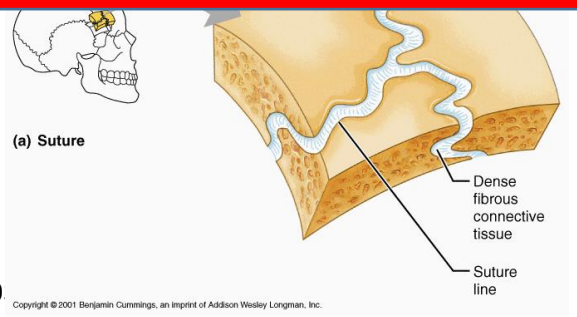


- **REVMATOID ARTHRITIS**

- **OSTEOPETROSIS**

- **PAGET DISEASE**

JOINTS

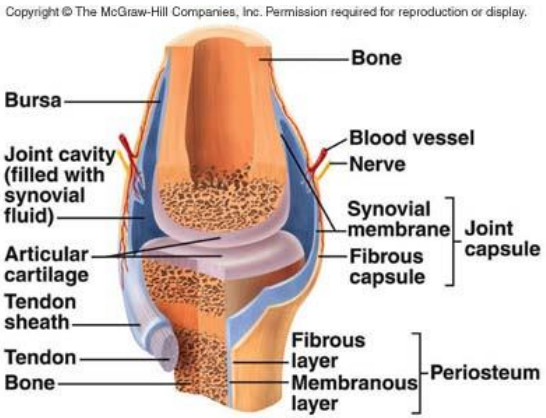


Synarthrosis

- joint by intercalated tissue (catilage, bone or c.t.)
  - **Synostoses** – joint by bone tissue – os coxae, os sacrum
  - **Synchondrosis** – joint by hyaline cartilage – development of synosto
  - **Symphysis** – joint by fibrocartilage– os pubis, intervertebral discs
  - **Syndesmosis** – dense collage regular c.t. – sutures of skull, gomphosis

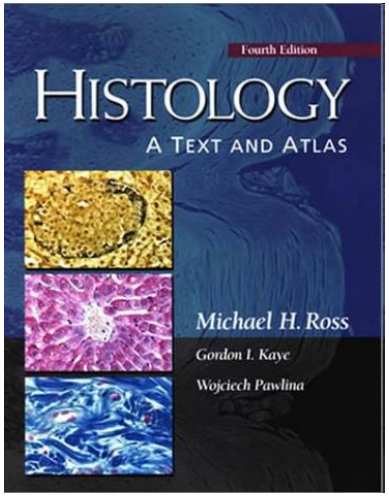
Diarthrosis

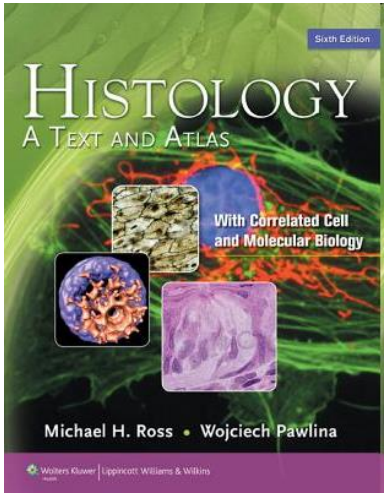
- synovial joint
  - hyaline cartilage without perichondrium
  - cartilage calcification in site of attachment to the bone
  - joint capsule
    - *Stratum fibrosum*
    - *Stratum synoviale*

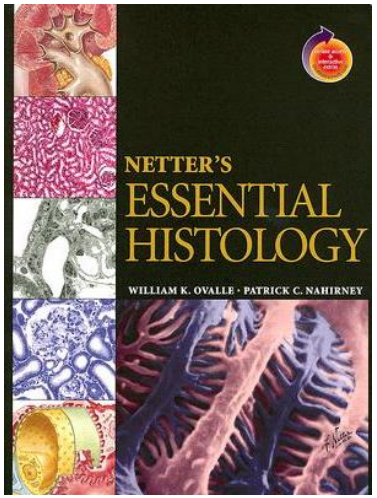


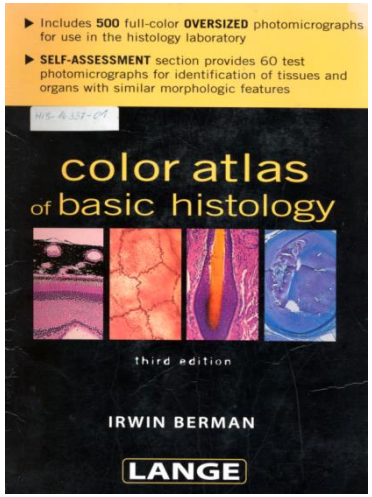
  - meniscus – fibrocartialge, avascular, without inervation
  - tendons – dense collagen regular c.t., elastic fibers
  - bursae – like joint capsule

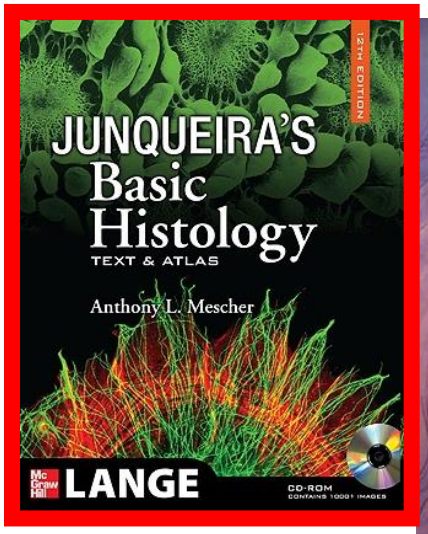
FURTHER STUDY

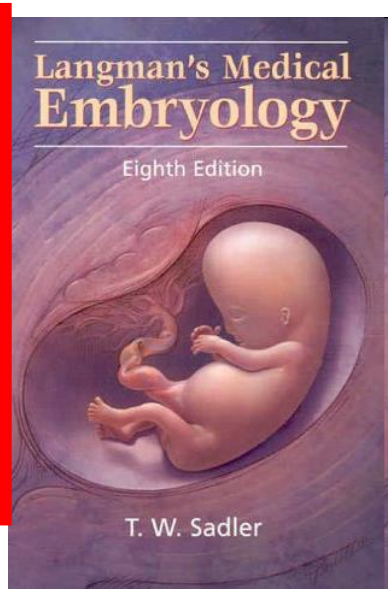


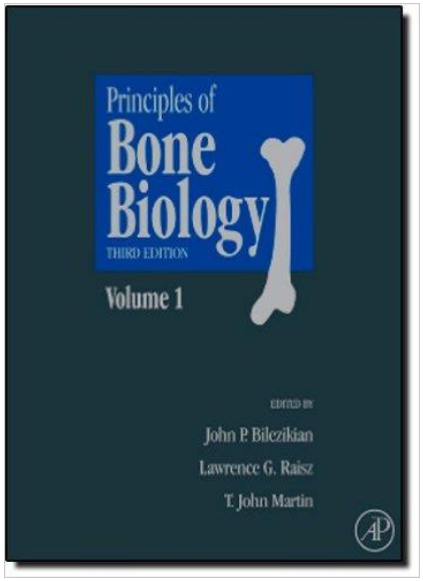


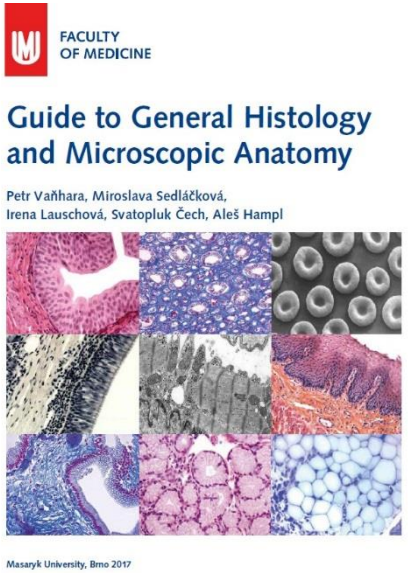












http://www.histology.med.muni.cz/

Thank you for attention