

Histology

faculty of medicine - JU2015

Lecture9

Bone

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- Bone is a special CT that is composed of cells and ECM
- ECM is composed of fibers and ground substance
- the ECM is hard because of minerals, this makes bone solid
- Bone is also strong due to collagen fibers type I so it resists tensile forces
- Bones originate from mesenchyme
- **Functions:**

~Support

~Protection (skull protects brain, thoracic cage protects lungs and heart, vertebral canal protects spinal cord)

~Movement (skeletal muscles are inserted into bones by their tendons and when they contract, they transfer their movement into body movement)

~Mineral homeostasis (high amounts of Ca^{+2} and other minerals are stored in the bone matrix, so the bone acts as a reservoir for these minerals)

- 99% of our Ca^{+2} is stored inside the bones, if you have low Ca^{+2} level inside the blood, this will release Ca^{+2} from the bone in order to increase its level in the blood
- Bones maintain a constant level of Ca^{+2} in our blood

~Hematopoiesis (formation of blood cells which occurs in red bone marrow)

➤ **Bone Marrow:**

Type 1 : red bone marrow where Hematopoiesis occurs

Type 2 : yellow bone marrow which is adipose tissue (storage of fat)

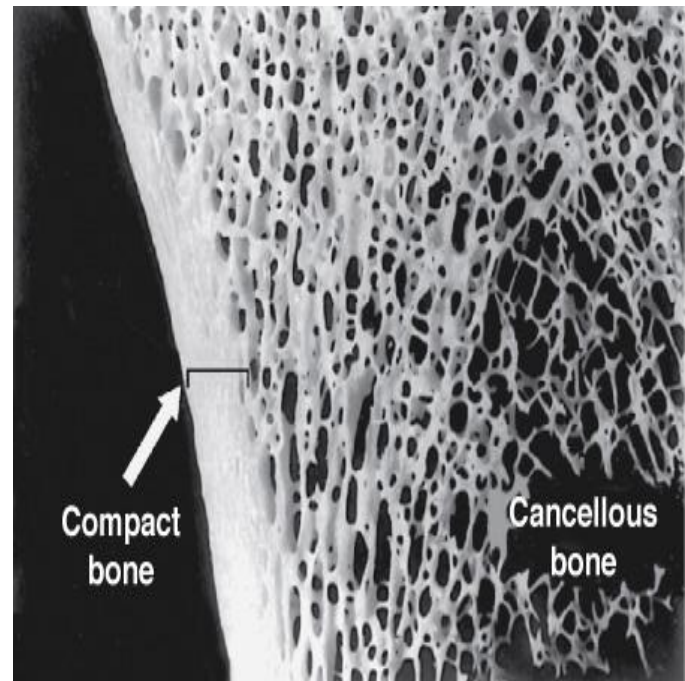
At the time of birth, the bone marrow is red bone marrow, with aging this is replaced by yellow bone marrow (white adipose tissue) except in certain places like the **sternum**

Anatomically , bones are divided according to their shape :

- a. Long bones: ex: radius and ulna, phalanges are also considered long bones as they have proximal end + shaft + distal end
- b. Short bones: ex: carpal bones, tarsal bones
- c. Irregular: ex: vertebrae
- d. Flat: ex: skull
- e. Sesamoid: ex: knee cap (patella)

Macroscopically , if you section the bone you will see two types of bone

- 1) Compact (dense): has no spaces. It forms the outer shell of bones (cortex), so it is also called cortical bone
- 2) Spongy (cancellous): With lots of spaces
Formed of bone spicules (pieces) called trabeculae, so it is also called trabecular bone



Histologically compact and spongy bones are different

Long bone

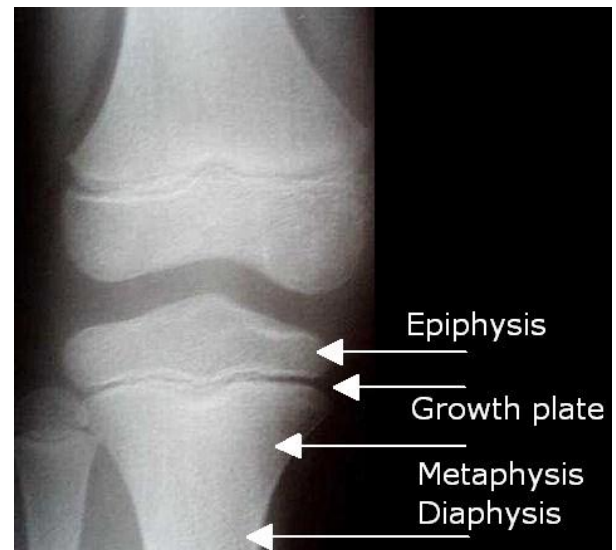
- Its body is called DIAPHYSIS and it is the constricted portion of the long bone.
 - Diaphysis is mainly composed of compact bone
 - At the center of the diaphysis we find a canal termed *marrow cavity* or *medullary canal*
 - Lining the medullary canal we find a thin layer of spongy bone
 - EPIPHYSIS is mainly composed of cancellous bone and covered by a thin layer of compact bone
- # Always and always spongy bone is covered by a layer of compact bone and it is impossible to find it exposed
- Articular surfaces of the joint are covered by a hyaline cartilage called articular cartilage which has no perichondrium and gets its nutrients from synovial fluid

- The spaces of the spongy bone are called marrow spaces or marrow cavities and are filled with bone marrow

Growth plate is composed of cartilage between Diaphysis and epiphysis

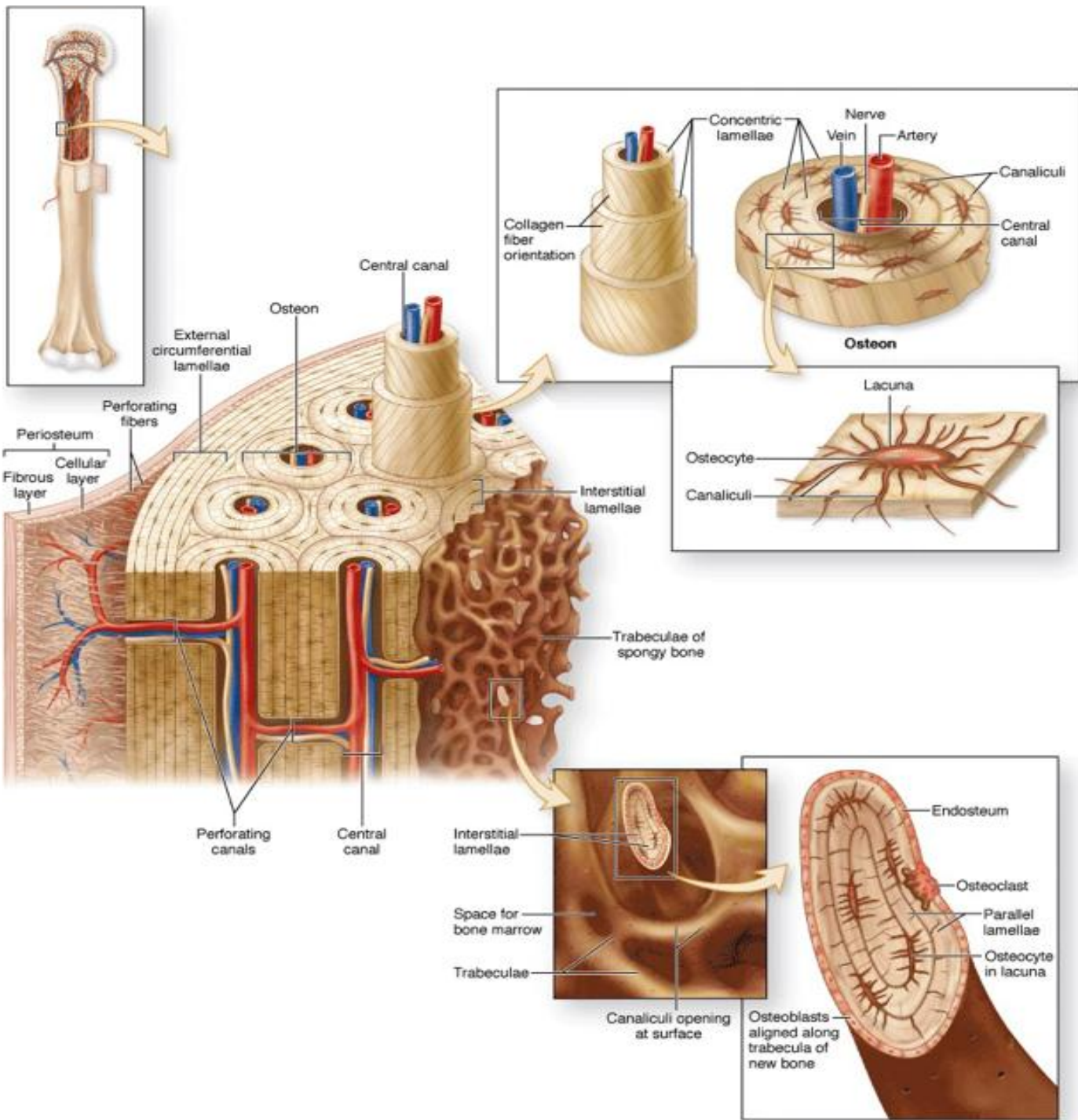
- In adults, when growth stops it becomes a line
- Cartilage is characterized by rapid mitotic activity causing fast growth
- Once the growth stops in adolescence due to lack of growth hormones, the cartilage plate becomes completely replaced by bone and becomes a line instead of the plate (epiphysial line)

Metaphysis is the area between epiphysis and diaphysis where the diaphysis starts to flare outward and becomes epiphysis then you start to see spongy bone instead of medullary canal



- Proximal part of metaphysis is the epiphysial plate
Some text books consider the epiphysial plate part of metaphysis

Section of the Shaft



- The medullary canal has bone marrow
- The wall is compact bone and the bone is covered by a membrane which is analogous to the perichondrium. It is called **PERIOSTEUM**

Periosteum

- ❖ Outer layer is fibrous

- ❖ The inner layer is cellular contains OSTEOPROGENITOR cells that are able to differentiate to osteoblasts
- ❖ Growth of bone can occur from the outer layer (periphery) because the inner layer of periosteum has osteoprogenitor cells
- ❖ Collagen type I fibers connect the periosteum with the bone matrix
- ❖ Collagen type I fibers in the periosteum merge with collagen type I inside the bone tissue
- ❖ These fibers are called SHARPEY's fibers which function to anchor the periosteum to the bone tissue itself
- ❖ In order to operate on the bone, periosteum must be removed first by cutting the sharpey's fibers

Histological Structure of the Compact Bone:

- Bone is covered externally by periosteum
- OSTEON is the basic functional unit of the compact bone
- It is also called HAVERSIAN system
- Osteon is a tall column of compact bone
- The longitudinal axis of osteon is parallel to longitudinal axis of shaft of long bone
- We find space in the middle of the osteon, this tiny canal contains blood vessels and nerve supply- central canal
- These central canals lie parallel to each other and have connection perpendicularly: VOLKMANN's canal
- Bone tissue is highly vascularized and richly innervated



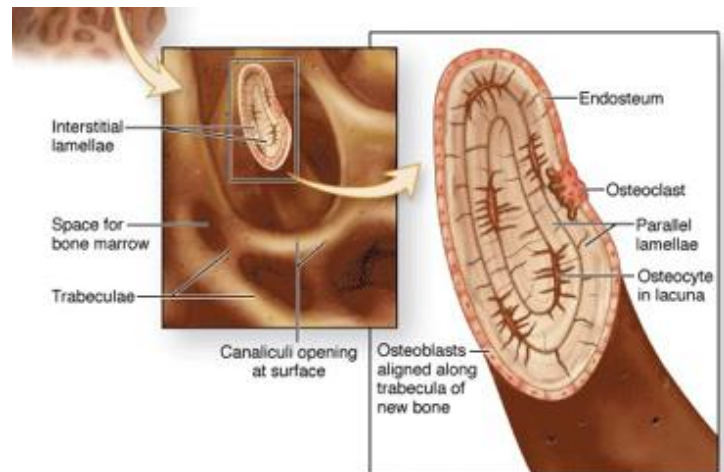
Osteons#

- *Concentric rings called lamellae surrounding central canal. Between these lamellae you can see cells which are called OSTEOCYTES
- * Lamellae are the hard bone matrix. (collagen I and minerals)
- * Collagen I are arranged (in a highly regular manner) parallel to each other in each lamella but at right angle to the nearby lamella which gives strength to the bone
- *Minerals are deposited around the collagen type I fibers and that's why the bone is hard

- Each osteocyte has processes in order to communicate with the neighboring cells and with blood vessels in the central canal. diffusion is impossible due to matrix hardness
- The areas where the processes pass through are called CANALICULI (tiny canals)
- Spaces where we find cells are called lacunae as in cartilage
- Volkmann's canals connect central canals of different osteons with the blood vessels of the periosteum and endosteum
- ** Any internal cavity inside the bone is lined by a membrane called ENDOSTEUM
As in : medullary canal, volkmann's canal , central canal
- The Lamellae exactly located under periosteum are called outer circumferential lamellae. The lamellae deep to the endosteum are inner circumferential lamellae and the lamellae between the osteons are called interstitial lamellae

Section of Spongy Bone:

- We can see lamellae but no central canals
- It is also composed of lamellae which are not arranged into osteons
- Spaces where we find cells are called lacunae
- Lamellae are parallel to each other rather than forming concentric lamellae
- Structure of trabecular bone :
 1. Irregularly shaped pieces of bone
 2. Spaces of spongy bone are filled with bone marrow (marrow cavities) and lined by endosteum



- ** Endosteum is composed of only single layer (cellular layer containing osteoprogenitor cells)
- REM: Bone tissue itself contains only osteocytes
- ~ Periosteum and endosteum both contain osteoprogenitor cells, so bone can grow in width

^^Structure of Flat Bones ^^

ex: skull

- Mainly composed of spongy bone covered by a thin layer of compact bone
- Area of spongy bone is named **diploë**
- Spongy bone is porous and lightly weighted

- **If the bones are composed only of compact bone we won't be able to move; this is so heavy
- Spongy bone is less stronger than compact bone

Content of the Bone Tissue:

- Cells and matrix (Organic and inorganic matters)
- Around 70% of ECM is minerals
- Organic part (30%): Mainly collagen type I, water, proteoglycans , glycoproteins

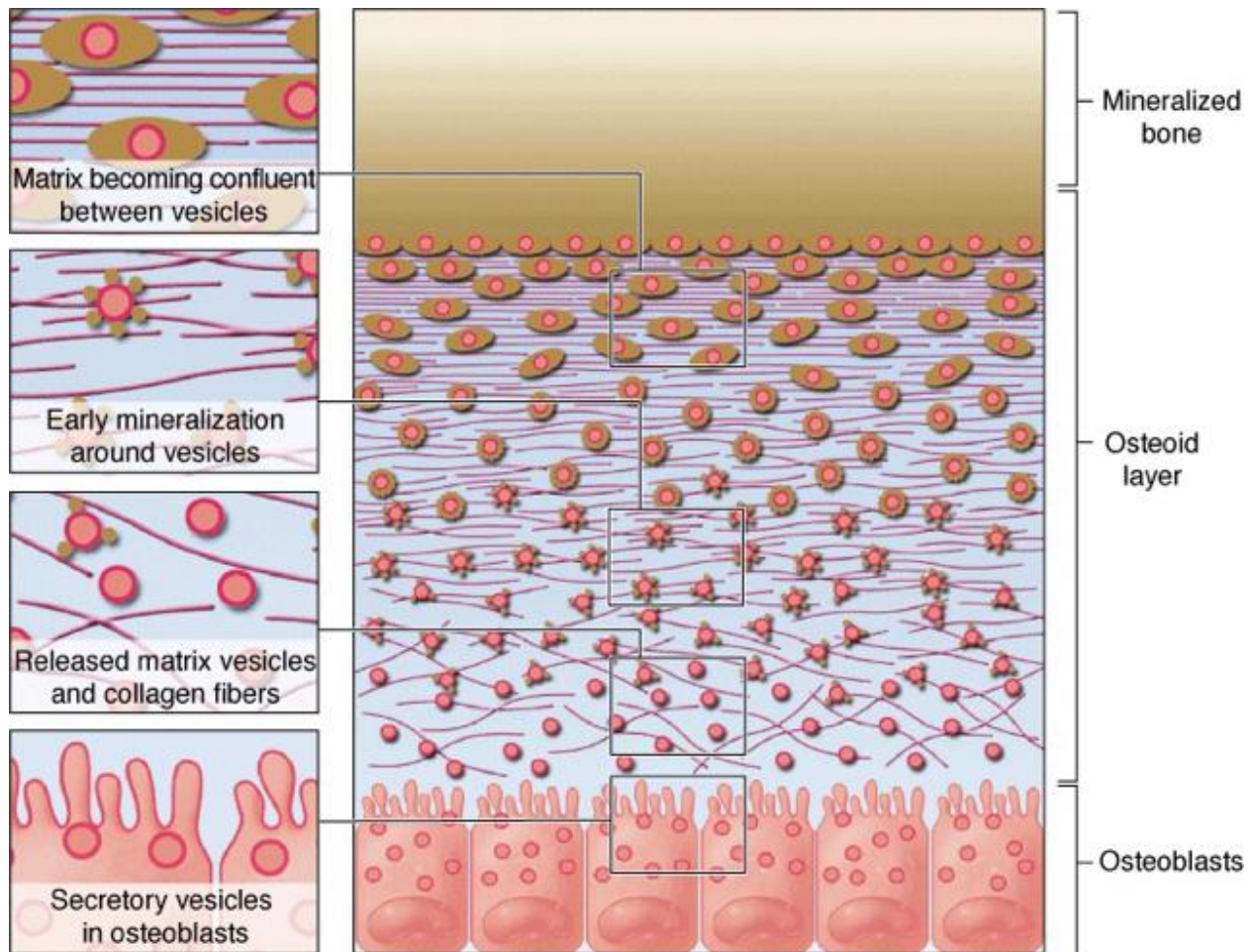
Osteoprogenitor cells

Mesenchymal unspecialized stem cells in periosteum and endosteum that can undergo mitosis to form osteoblasts

Osteoblasts(building cells)

- On periosteum and endosteum (bone surfaces)
- In order to build up bone, osteoblasts need hard surface acting as a scaffold
- Inner layer of periosteum contains osteoprogenitor cells which differentiate into osteoblasts.
- Osteoblasts sit on the periosteum and they look like simple cuboidal or columnar epithelium.
- These cells build up bone matrix at the direction of the old bone between themselves and the old bone. Then when they mineralize their matrix , they become encased within spaces or chambers called lacunae
- Osteoblasts synthesize the organic components of ECM (collagen type I)
- Bone matrix synthesized by osteoblasts is called osteoid and it is the first material secreted from the osteoblasts before the mineralization
- Osteoblasts are also involved in calcification of their matrix
- These cells secrete vesicles containing enzymes including alkaline phosphatase which takes the phosphate group from certain macromolecules and increases its concentration locally
- They also secrete a glycoprotein called osteocalcin which has high affinity for Ca^{+2} , so it increases the concentration of Ca^{+2} inside osteoid
- The presence of high concentrations of Ca^{+2} and phosphate will cause deposition of minerals around these vesicles and form hydroxy apatite crystals

And this is how mineralization of the osteoid occurs



**This process is life-time process

Osteocytes

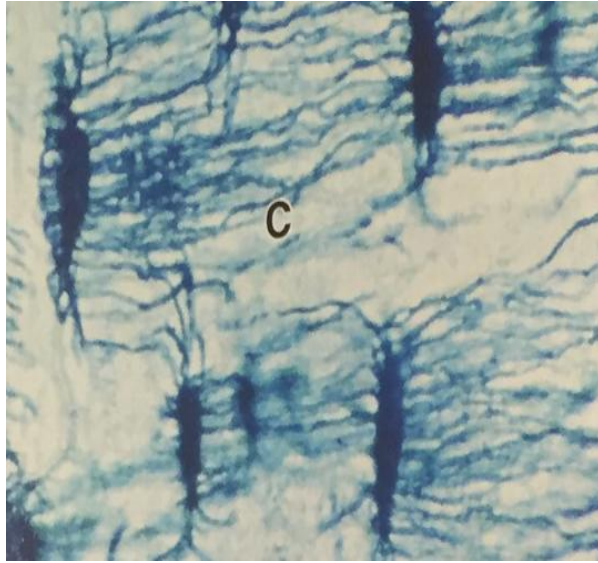
- Situated within lacunae

Characteristics

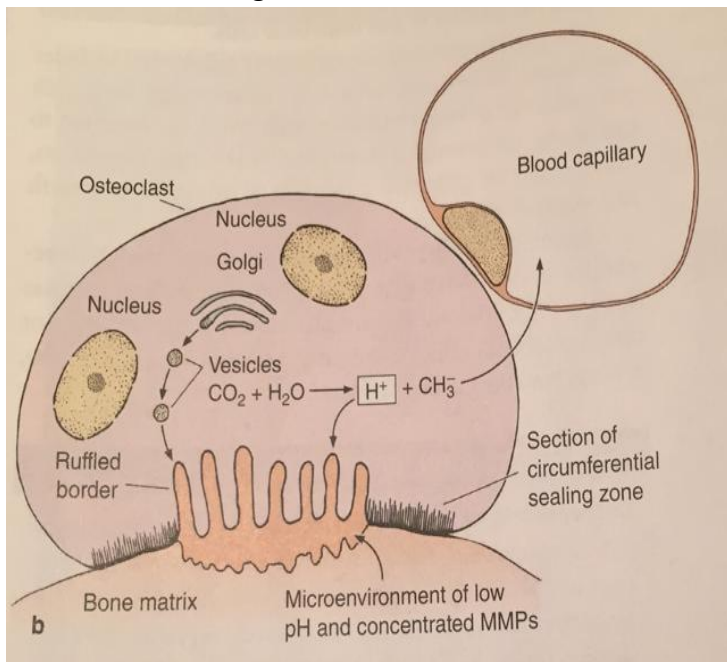
- Inactive cells
- With few organelles
- Small in size

^^The one and only cell that is seen within the tissue not on surface





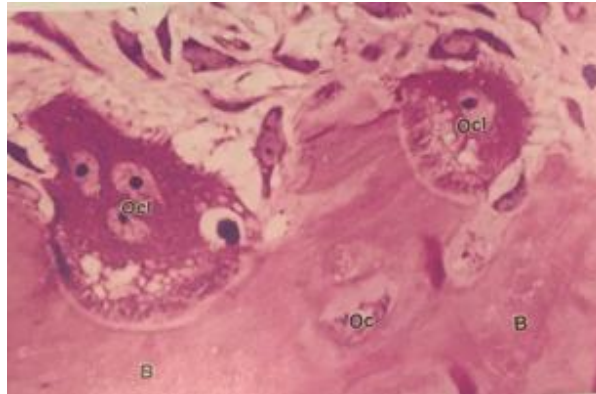
Osteoclasts Cutting cells



- ✓ Resorb ECM of the bone
- ✓ They are found in periosteum and endosteum (surfaces of bone)

- ✓ They create their own lacunae on the bone surface, called HOWSHIP lacunae (resorption cavities)
- ✓ large multi nuclear cells which can contain up to 50 nuclei

In the histological section we can see 5 – 10 nuclei

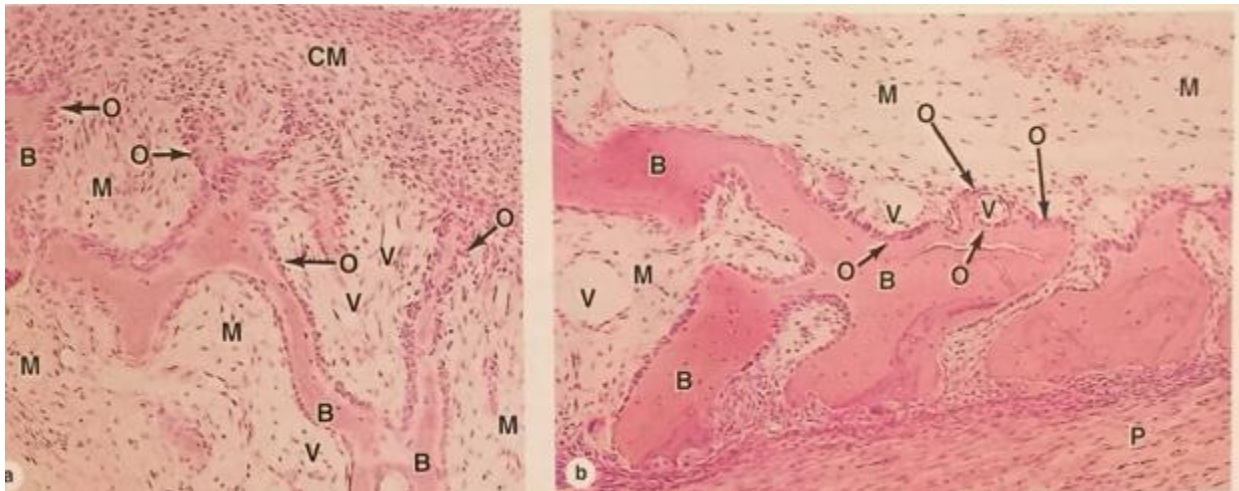


- ✓ They belong to Mononuclear Phagocytes System, remember this system is composed of Macrophage and macrophage like cells situated in different sites of the body
- ✓ Originate from bone marrow (fusion of monocytes)
- ✓ In order for these cells to cut through bone, they should be firmly adherent to the bone surface (through actin filaments)
- ✓ Plasma membrane of these cells (osteoclasts) is folded in order to increase surface area for contact and secretion. This is called **ruffled border**
- ✓ Clear zone : free of organelles
- ✓ Cells have lysosomes containing hydrolytic enzymes including collagenase
- ✓ They release enzymes to ECM
- ✓ The ruffled border contains high concentration of proton pumps
- ✓ $[H^+]$ will increase in ECM resulting in an acidic environment. The low pH causes demineralization of the bone matrix.
- ✓ The dissolved minerals pass through the cell (by transcytosis) to enter the blood vessels, thus increasing the calcium level in the blood

How to Prepare Bone for Histological View ?

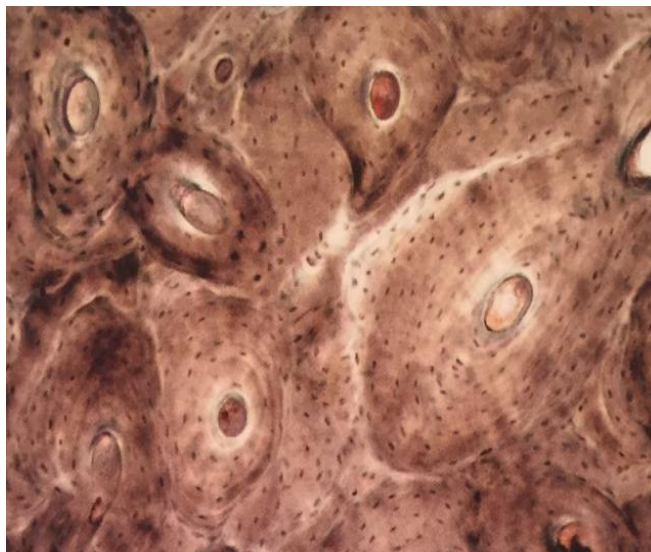
1- Decalcification:

- ✓ is to remove Ca^{+2} and minerals from the bone tissue using decalcifying agent , so fibers, cells and ground substance will remain (as the tendon) , after that you can do fixation, mounting and staining by H&E
- ✓ This doesn't preserve the morphology of the osteons
- ✓ CANALICULAI can't be seen



2- Grinding

- ✓ Is to grind the bone so you will get bony chips and without fixation or staining you can view the bone under the microscope
- ✓ Spaces of the bone (lacuna, central canal, canaliculi) are filled with dusts
- ✓ Overall morphology of the bone is preserved



- ✓ Notes:
- ✓ Osteoblasts build and osteoclasts cut in a process called ***bone remodeling*** which occurs throughout our life
- ✓ Tuberosity , tubercle , ridge and other features on the bone surface are due to bone deposition in the site of the pull of the muscle
- ✓ Both the compact and spongy bones are lamellar bones

