Department of Histology and Embryology
Faculty of Medicine, PU, Olomouc, CZ

Histology practical No. 6

Topics:

1- STRUCTURAL AND FUNCTIONAL CHARACTERISTICS OF THE CARTILAGE AND BONE TISSUES (Pre-lab rev.ppt).

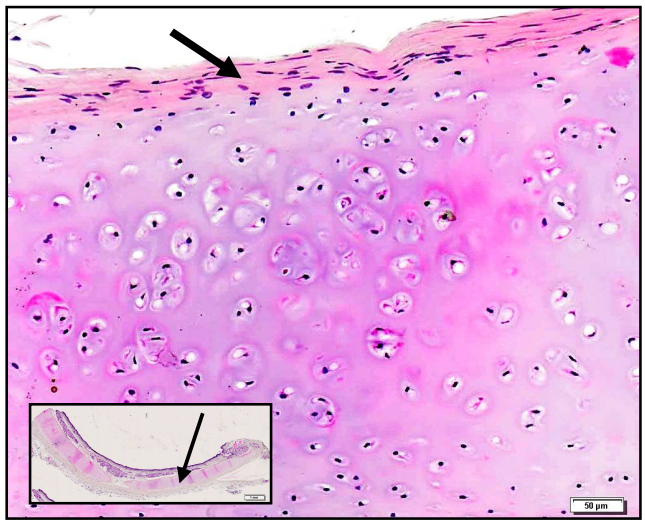
2- RECOGNITION OF HYALINE, ELASTIC AND FIBROCARTILAGE IN STAINED HISTOLOGY AND PC-MONITORED SLIDES OR PRINTED IMAGES.

3- RECOGNITION OF COMPACT AND CANCELLOUS BONE IN STAINED HISTOLOGY AND PC-MONITORED SLIDES OR PRINTED IMAGES.

4- UNDERSTANDING THE PROCESS OF ENDOCHONDRAL AND INTRAMEMBRANOUS OSSIFICATION AND STAGES OF BONE DEVELOPMENT. IDENTIFICATION OF THE OSSIFICATIONS IN STAINED HISTOLOGY AND PC-MONITORED SLIDES OR PRINTED IMAGES.

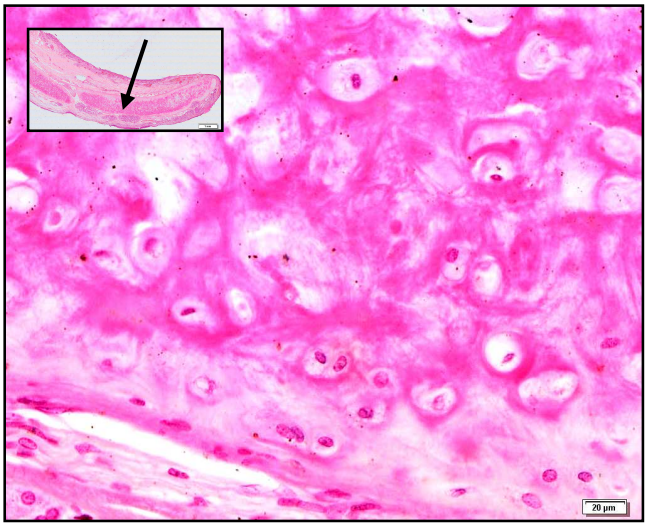
Slides:

1. Trachea (trachea, sl.n. 34) t.s., H&E stain.



In transverse section of the trachea, find the middle layer of the wall supported with a strip of a **hyaline cartilage**. Note the abundance of chondrocytes located in lacunae either solitary or in small groups (isogenous groups). The amorphous **matrix** is basophilic. Identify the **perichondrium** (arrow) located on two sides of the hyaline cartilage. Can you find any blood vessels in the matrix of this cartilage?

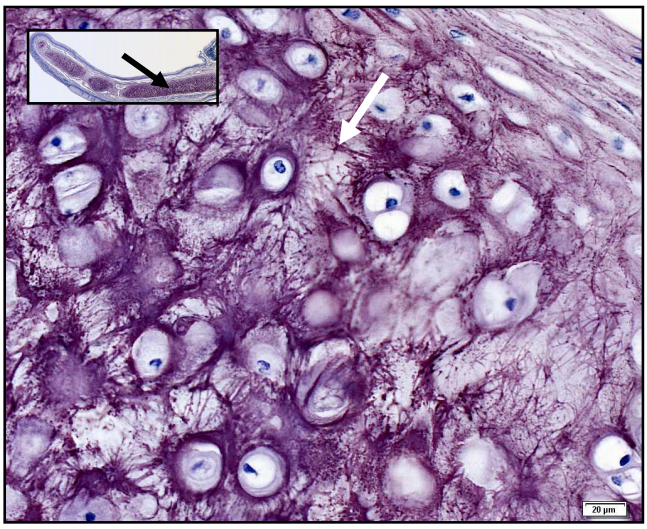
2. Elastic cartilage (chrupavka elastická, sl.n. 33A), t.s., H&E stain.



A plate of the **elastic cartilage** is located in the middle part of epiglottis. Numerous, mostly solitary chondrocytes are crowded in the matrix that shows some traces of **fibrillar** structures. The elastic component of the **matrix** is not clearly visible due to the non-specific H&E staining. Find the perichondrium at the surface of the cartilage. What is the function of the perichondrium? In your spare time, you can revise and locate the lining epithelium and some glands in the surrounding tissue. What kind of secretion is produced by these glands?

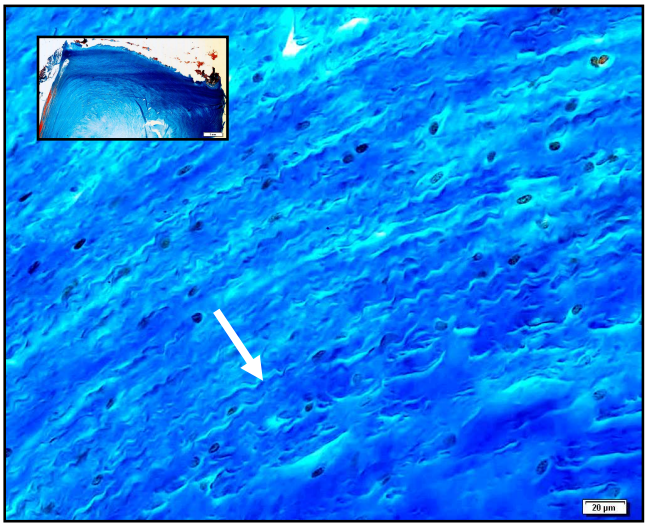
3. Epiglottis (epiglottis, orcein, sl.n. 33), t.s., Orcein stain.

This slide is similar to the slide n. 33A, but this one is specifically stained for elastic fibers. The centrally-located **elastic cartilaginous plate** is fragmented into several pieces. The special staining for elastin shows abundance of elastic fibers (purple lines) in the cartilaginous matrix (white arrow). They embrace the lacunae and run across the plate from one surface to the other. Note the epithelia around and identify the adipose tissue in this section. Why is this organ supported with the elastic cartilage?



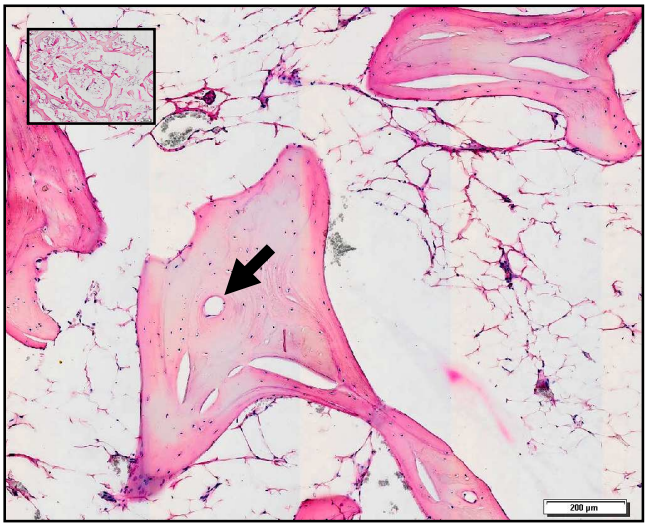
4. Fibrocartilage (chrupavka vazivová, sl.n. 76), section, AZAN stain.

This is a section of an intervertebral disc that contains the **fibrocartilage** in its superficial, more densely-stained portion (*annulus fibrosus*). Due to the special staining method, the bundles of **collagen fibers Collagen** are in blue color, and the elongated nuclei of solitary **chondrocytes** are orange-stained. Note the prevalence of collagen bundles arranged in overlapping layers (arrow), and the small number of chondrocytes sandwiched between them, sometimes like in rows. The central, pale-looking portion of the section (*nucleus pulposus*) contains rounded cells embedded in a **viscous matrix**. What type of collagen is present in the superficial layer?



5. Cancellous bone (spongiosní kost, sl.n. 77) sect., decalcified, H&E stain.

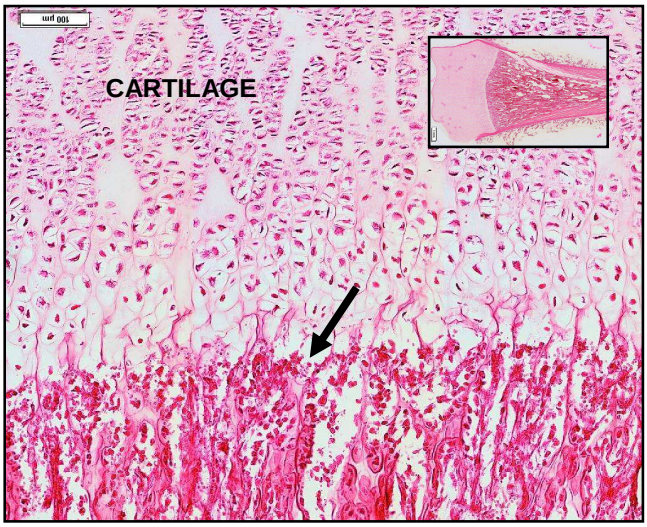
In this bit of **cancellous (spongy) bone** from epiphysis of a long bone, observe the pink-stained, branching **trabeculi** of bone matrix with solitary **osteocytes** embedded inside it. Note the lamellar background of the trabeculi that contain some complete and incomplete **Haversian systems** (arrow). Due to the destructive effect of the decalcification process, the osteoblasts and osteoclasts are difficult to observe at the surfaces of bone trabeculi. The intertrabecular spaces contain some adipose tissue and blood vessels (*white bone marrow*).



6. Compact bone (kost kompaktní, sl.n. 78) sect., decalcified, H&E stain.

This transverse section of diaphysis of a long bone contains a layer of **compact bone** on one side, and a transition to the **cancellous bone** on the opposite side. Study the compact bone in details as it shows many transverse sections of complete **Haversian systems - osteons** (arrow). Due to the higher thickness of this section, you have to focus through the specimen repeatedly to get a clear picture. Observe the osteocytes in their **lacunae** with radiating **canaliculi**, find the **Haversian canals**, **Volkmann's canals**, **interstitial lamelli** and **cementing lines**. Draw a simple diagram of the osteon. The periosteum may not be shown in some slides due to the absence of the complete surface of the bone sample.

7. Ossification (ossifikace, sl.n. 79) l.s., decalcified, H&E stain.



This is a very large section of diaphysis and of adjacent part of epiphysis of fetal bone that shows both, the **endochondral ossification** in the region of epihyseal plate (**cartilage**), and **intramembranous ossification** at the surface bone collar (**subperiostal** ossification). Before you begin to study details, check out the two parts of this section and locate the **epihyseal end** of the bone. This end contains a piece of a **hyaline cartilage** with **chondrocytes** that change gradually when shifted towards the zone of ossification. Identify the zones of proliferation, maturation, hypertrophy and calcification of chondrocytes. Find the zone of **erosion** (arrow) and note that the chondrocytes disappear there, leaving behind just the thin septa of the cartilaginous matrix. Observe many osteoblasts adhering to the surface of cartilaginous septa. Now, shift your section further more in the direction towards the **diaphysis** and observe the thicker septa that are covered with a thin layer of primary bone. Note, that some of the osteoblasts there became entrapped within the newly produced bone matrix and have already the shape of osteocytes. Some of these osteocytes contain brown-black precipitations that make them easy to observe.

The **intramembranous ossification** creates the superficial bone collar that can be observed at the surface of the diaphyseal part of this section. Find the superficial layer of the connective tissue that makes the primitive periosteum and observe the cells transformed into osteoblasts that adhere to the surface of the bone trabeculi. In this part of the newly-produced bone, you can also observe **osteocytes** and **osteoclasts**. Answer these three questions: 1. What is the function of chondrocytes in the ossification of epiphyseal plate? 2. Where the osteoblasts originate from, and how they get into this ossification region? 3. How you describe the origin and morphology of osteoclasts?

8. <u>Watch the DVD-video on ossification process and development of long bones.</u>
(DVD-video, English lang. subset, 9 min.)

9. <u>In your selfstudy, revise the ultrastructure of osteocyte, osteoblast and osteoclast.</u>
(Use the recommended textbook, atlas with CD-ROM and WWW)

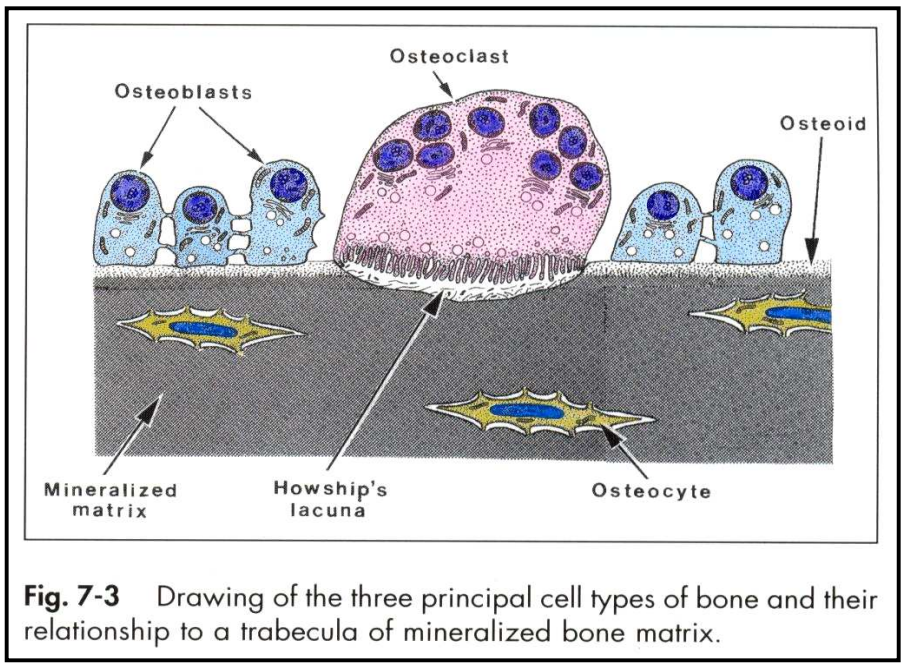
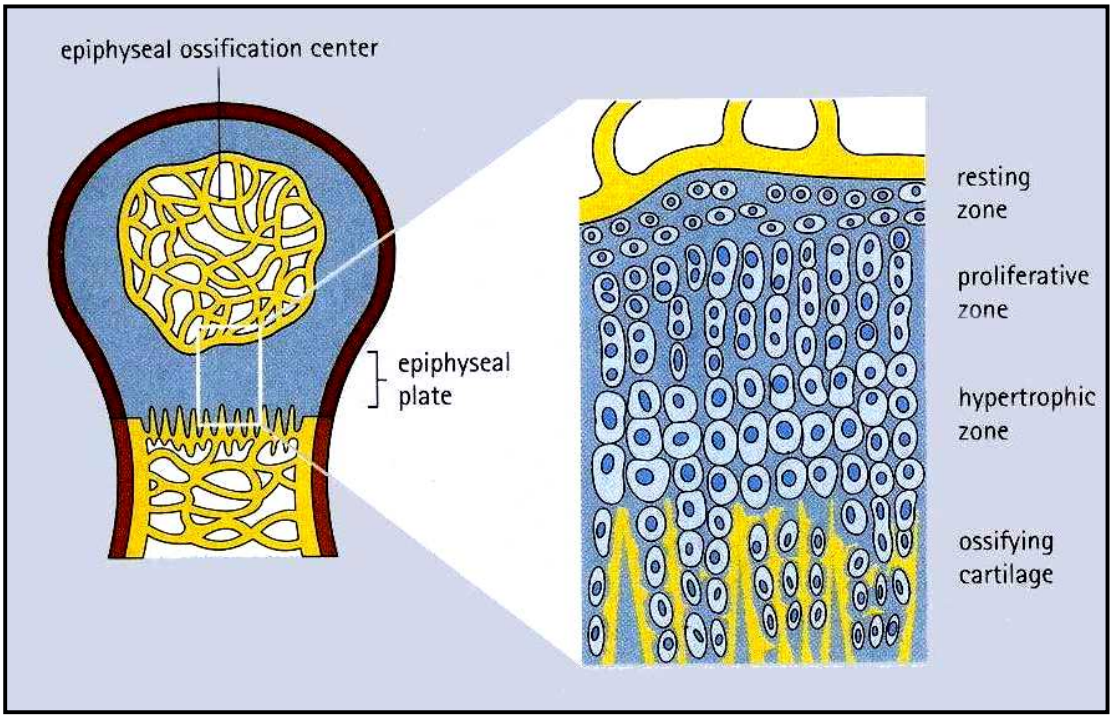
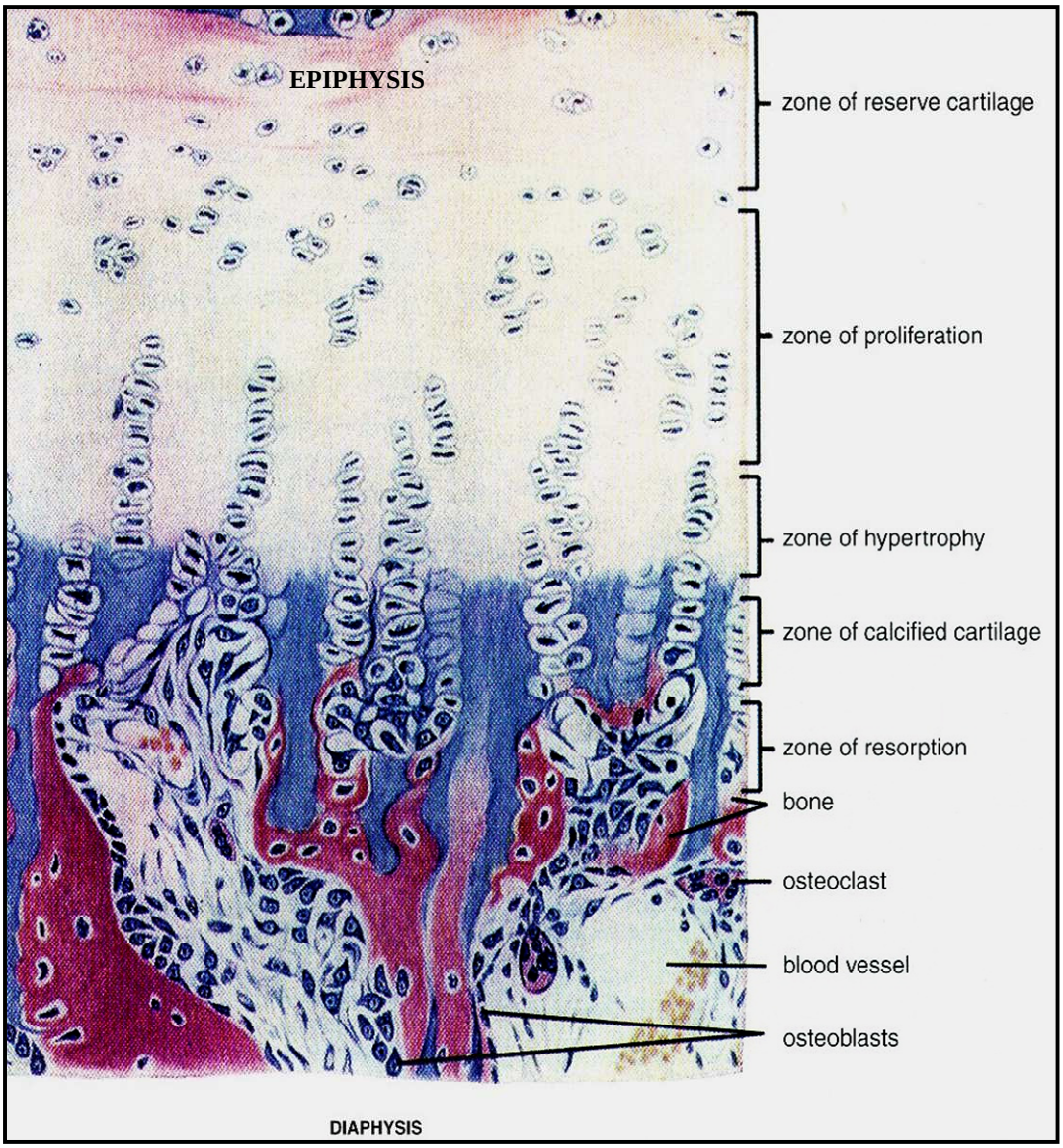


Fig. 7-3 Drawing of the three principal cell types of bone and their relationship to a trabecula of mineralized bone matrix.

From: Fawcett D.W., Jensh R.P.: Concise Histology, Chapman&Hill, New York, 1997

10. <u>For better understanding, compare the following images with the slide n. 79.</u>





From: Ross MH et al.: Histology. A text and atlas. 4th ed., Lippincott Wiliams &Wilkins, Philadelphia, 2003

Websites:

http://www.histology-world.com/keyfeatures/hyaline1.htm
http://www.youtube.com/watch?v=PVoz28_n0uU&feature=related
http://www.youtube.com/watch?v=PVoz28_n0uU&feature=related
http://www.youtube.com/watch?v=imDyTwvgNaQ&feature=related
http://www.youtube.com/watch?v=bTP2hAG0wcM&feature=related
http://www.youtube.com/results?search_type=&search_query=Shotgun+Histology+bone&aq=f
http://visualhistology.net/Visual_Histology_Atlas/VHA_Chpt5_Cartilage.html
http://www.med.uc.edu/medware2/microanatomy/car2p04.htm
http://www.medicine.uiowa.edu/anatomy/dental/genhisto/GHWIN/index.html