

Connective tissue proper

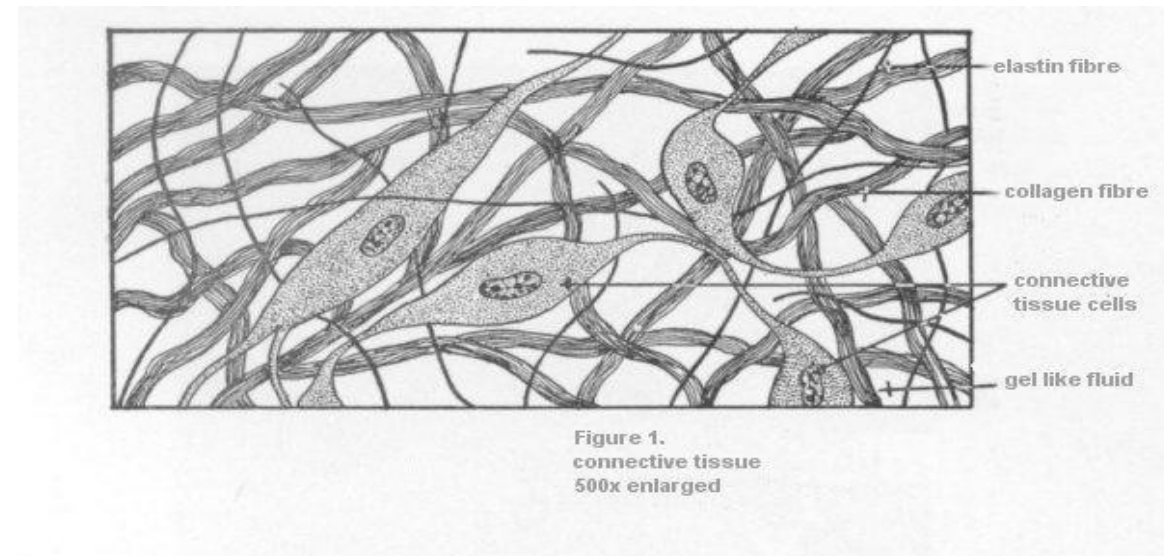
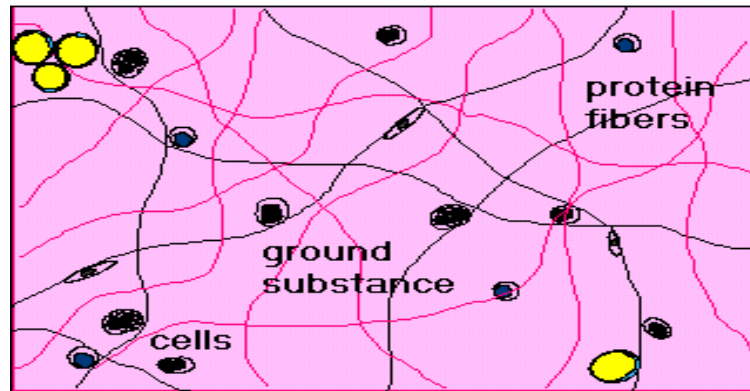
Cartilage

Connective tissue

- forms an extensive compartment in the body and can be considered as the "glue" that holds the body and organs together.
- 3 types of CT: **connective tissue proper** / **cartilage** / **bone**
- consistency is from soft gel-like (areolar CT) to hard (bone)
- originate from embryonic CT = mesenchyme (derivate of mesoderm)
- function of CT: supporting, nutritive (diffusion of nutritives from blood vessels)

Connective tissue

All types of CT consist of **cells** and **intercellular matrix** secreted by some of those cells. The intercellular matrix consists of **fibres** and **ground substance**



Connective tissue

Types of CT	Classification	Cells	intercellular	matrix
			Fibres	Ground substance
CT proper	Mesenchyme Mucose CT Loose CT Dense CT - regular - irregular Reticular CT Adipose CT - white fat - brown fat	<u>Fixed</u> : fibroblasts and fibrocytes, reticular cells, pigment cells, adipose cells, histiocytes, mast cells, plasma cells, <u>Free</u> : macrophages, leukocytes	collagen reticular elastic	
Cartilage	Hyaline Elastic Fibrocartilage	chondroblasts and chondrocytes	collagen elastic	
Bone	Fibrillar Lamellar - compact - spongy	osteoblasts, osteocytes	collagen	mineralized

Connective tissue

- **Ground substance:** occupies the space between the cells and fibres of CT
- colorless, transparent, homogenous substance of mucose consistency
- high water content
- in specially prepared sections, has an amorphous appearance. In routine preparations, it is lost during the fixation and dehydration process, and only cells and fibres can be seen.
- Consists of : glycosaminoglycans (hyaluronic acid, chondroitinsulfate, dermatansulfate, keratansulfate, heparansulfate)
 - proteoglycans – (aggrecan, syndecan, fibroglycan)
 - glycoproteins – (fibronectin, chondronectin, laminin, osteocalcin, osteonectin, osteopontin)

Connective tissue - fibers

- **Fibres:** There are three types of fibres secreted by connective tissue cells: **collagen fibres**, **reticular fibres**, and **elastic fibres**. The abundance of different types of fibres varies in different CTs. Each type of fibre is formed by proteins made of long peptide chains.
- Collagen fibres:
 - the most common type of fibres arranged into bundles
 - flexible fibres with a high tensile strength
 - in LM: wavy lines of variable width and indeterminate length.
 - in EM: fibres are seen to be made up of thread-like subunits called collagen fibrils – made of **protein COLLAGEN** (Several types of collagen have been identified- type I collagen in the dermis of the skin, bone, tendon, organ capsules and many other areas, type II in cartilage, type IV in the basal lamina (of basement membrane) of epithelia)

Connective tissue - fibers

- Reticular fibres:
 - are closely related to collagen fibres.
 - made up of **protein COLLAGEN** (type III collagen fibrils- fibres do not form bundles, and can be displayed with special silver preparations (impregnation))
 - fibres are arranged into network which provides a supporting net (the lymphnodes, the spleen). They are also found around adipocytes, small blood vessels, nerves and muscle cells.
 - are produced by fibroblasts, the reticular fibres that support the stroma of hemopoietic and lymphatic tissue are made by special cells called **reticular cells**.

Connective tissue - fibers

- Elastic fibres:

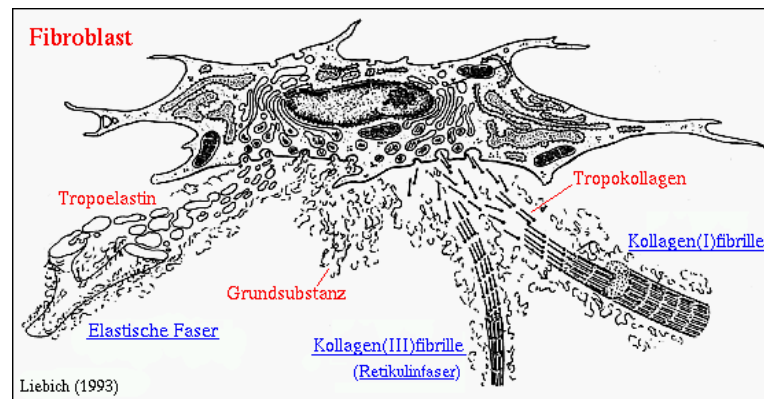
- thinner than collagen fibres and are arranged in a branching pattern to form a three dimensional network, they give tissue the ability to stretch and distension
- composed of two structural components:
 - protein ELASTIN** and **microfibrils** (microfibrils consist of a fibrillar glycoprotein)
 - are found in certain ligaments (elastic ligaments), some cartilage (auricle, epiglottis) and in large arteries (elastic arteries).
 - elastic fibres are produced by fibroblasts (in the case of elastic arteries, it is produced by the smooth muscle cells of the tunica media)
 - they do not stain very well with eosin and in routine preparations usually cannot be distinguished from collagen fibres. Elastic fibres are selectively stained with special dyes such as orcein and resorcin-fuchsin.

Connective tissue - cells

- many different kinds of cells can be found in CTs, some of the cells in CTs are **fixed** - they are permanent residents in the CT
 - other cells are **wandering (free)** - they are transient migrants who enter the CT from the blood in response to specific stimuli.

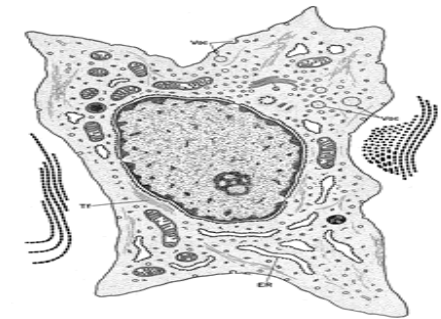
Fibroblasts:

- the principal cells of connective tissue.
- they produce precursors of all types of fibres (collagen, reticular, elastin) and ground substance.
- EM: rER, GA



Reticular cell:

- stellate cells
- production of collagen III of reticular fibres
- EM: rER, GA



Pigment cells: (melanocytes)

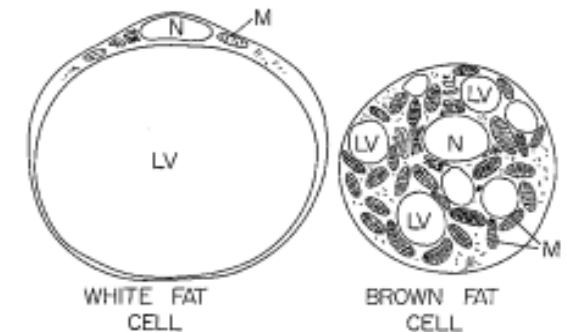
- production of pigment melanin
- cells originate from neuroectoderm

Adipose (fat) cells:

Also called adipocytes, these cells are specialized to store neutral fat.

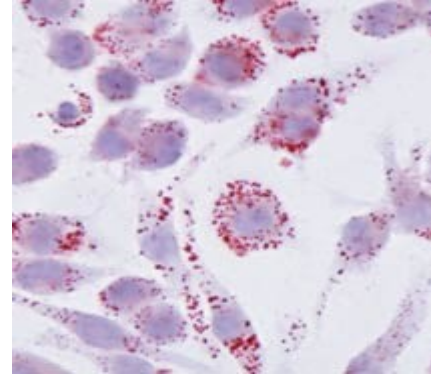
White fat – univacuolar cells (one large lipid droplet)

Brown fat – multivacuolar cells (several small lipid droplets)



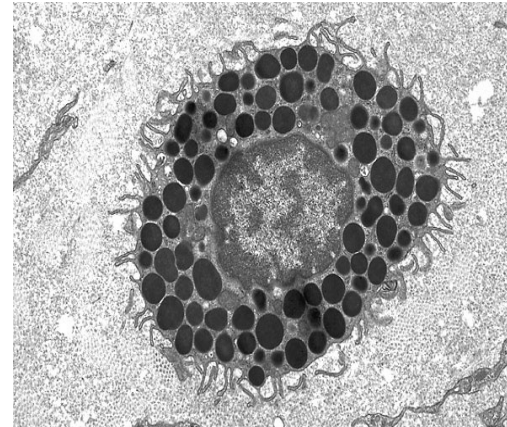
Histiocytes:

are phagocytic cells (fixed macrophages), after activation transform into migrating form – free macrophages
(detection of macrophages phagocytosing some dye→)



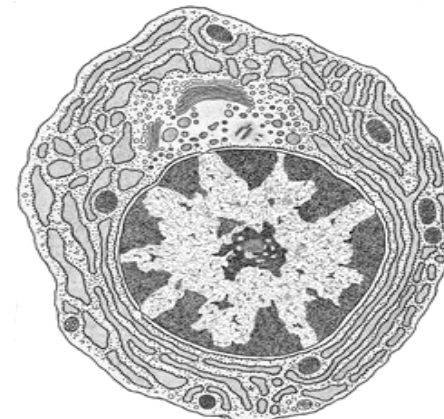
Mast cells:

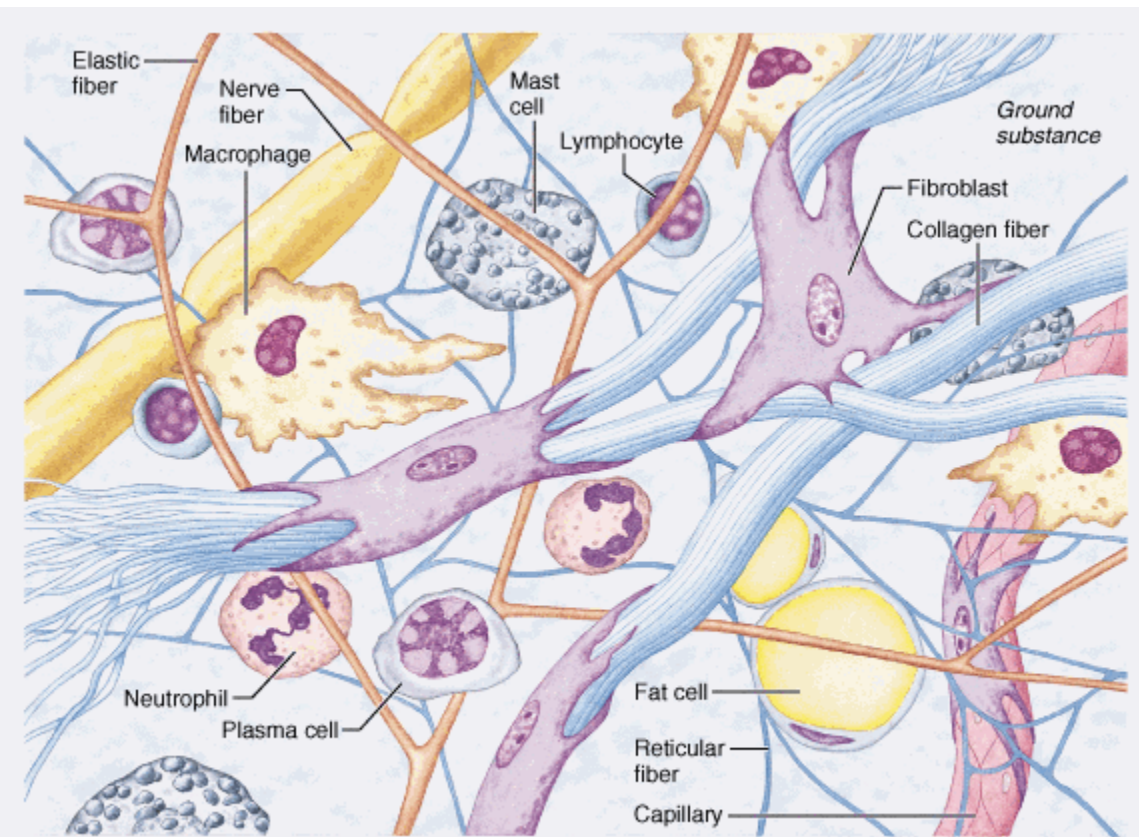
with granules containing histamine, heparin and anaphylactic factors.
When released in response to an antigen, they cause hypersensitivity reactions, allergy and anaphylaxis.



Plasma cell:

with voluminous cytoplasm and typical appearance of nucleus („clock“).
Plasma cells are derived from B-lymphocytes and produce antibodies against a specific antigen. They have a limited migratory ability and a short life.





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Wandering cells:

Neutrophils: white blood cells - phagocytes in the early stages of acute inflammation.

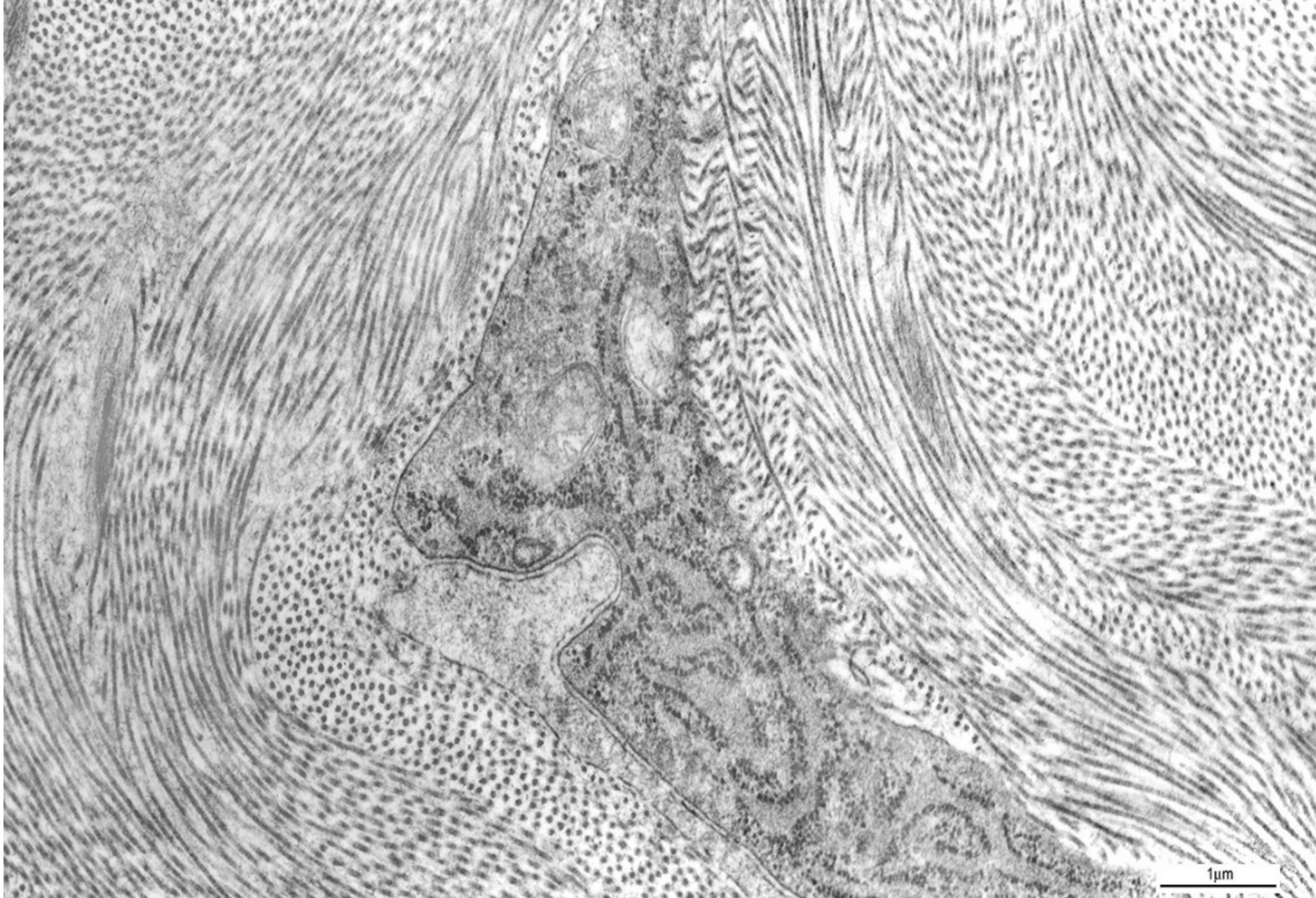
Eosinophils: white blood cells that - in the lamina propria of the GI tract, and at sites of allergic reaction and parasitic infection.

Basophils: white blood cells - similar to mast cells in having vasoactive agents released in response to an allergen.

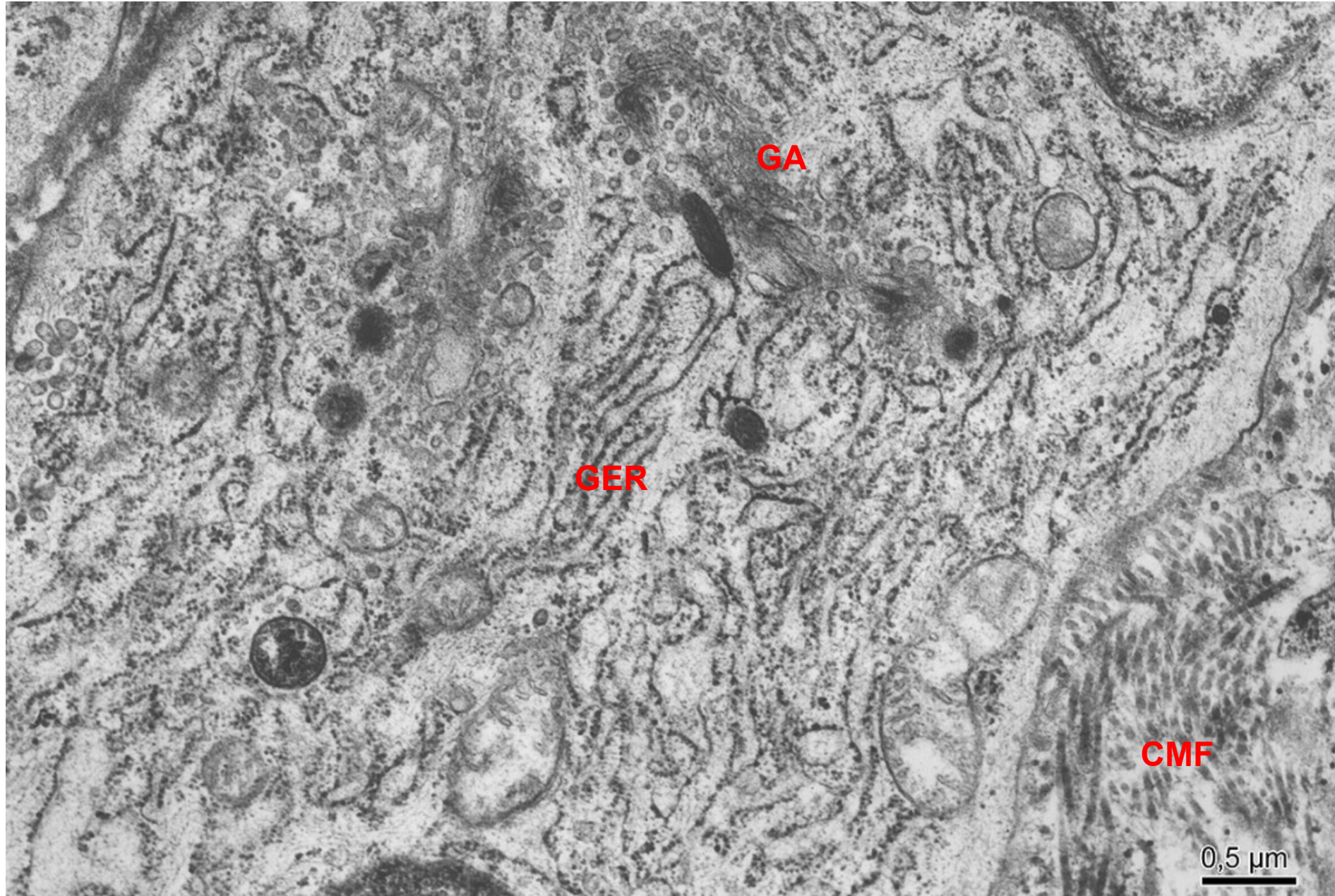
Lymphocytes: cells responsible for immune responses that circulate in the blood. Normally, only small numbers are found in the CTs throughout the body. The number increases dramatically at certain sites of tissue inflammation. They are also very numerous in the **lamina propria** of the respiratory and gastrointestinal tracts, where they are involved in immunosurveillance. The lamina propria is a layer of loose CT lying immediately beneath the epithelium.

Monocytes: white blood cells - will give rise to all the phagocytes of the mononuclear phagocytic system.

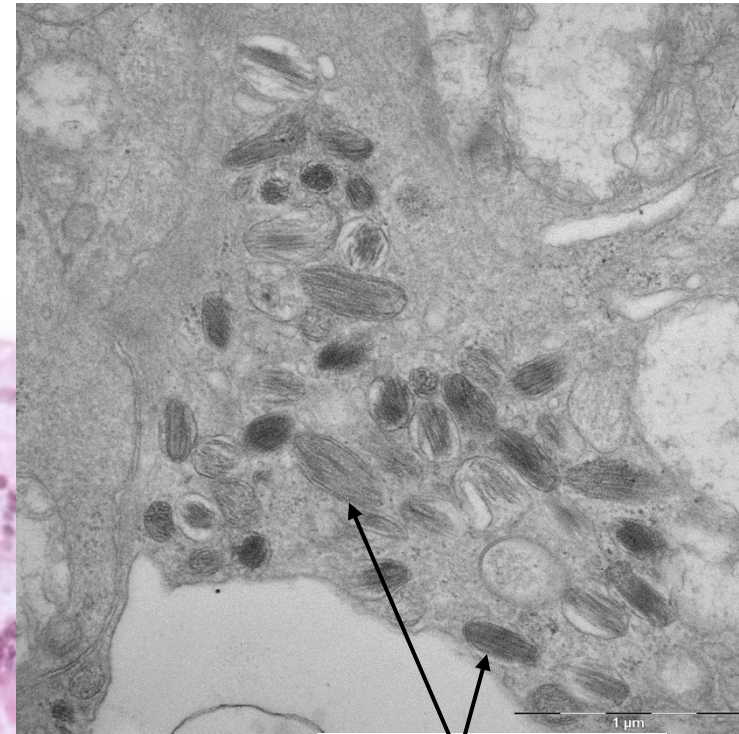
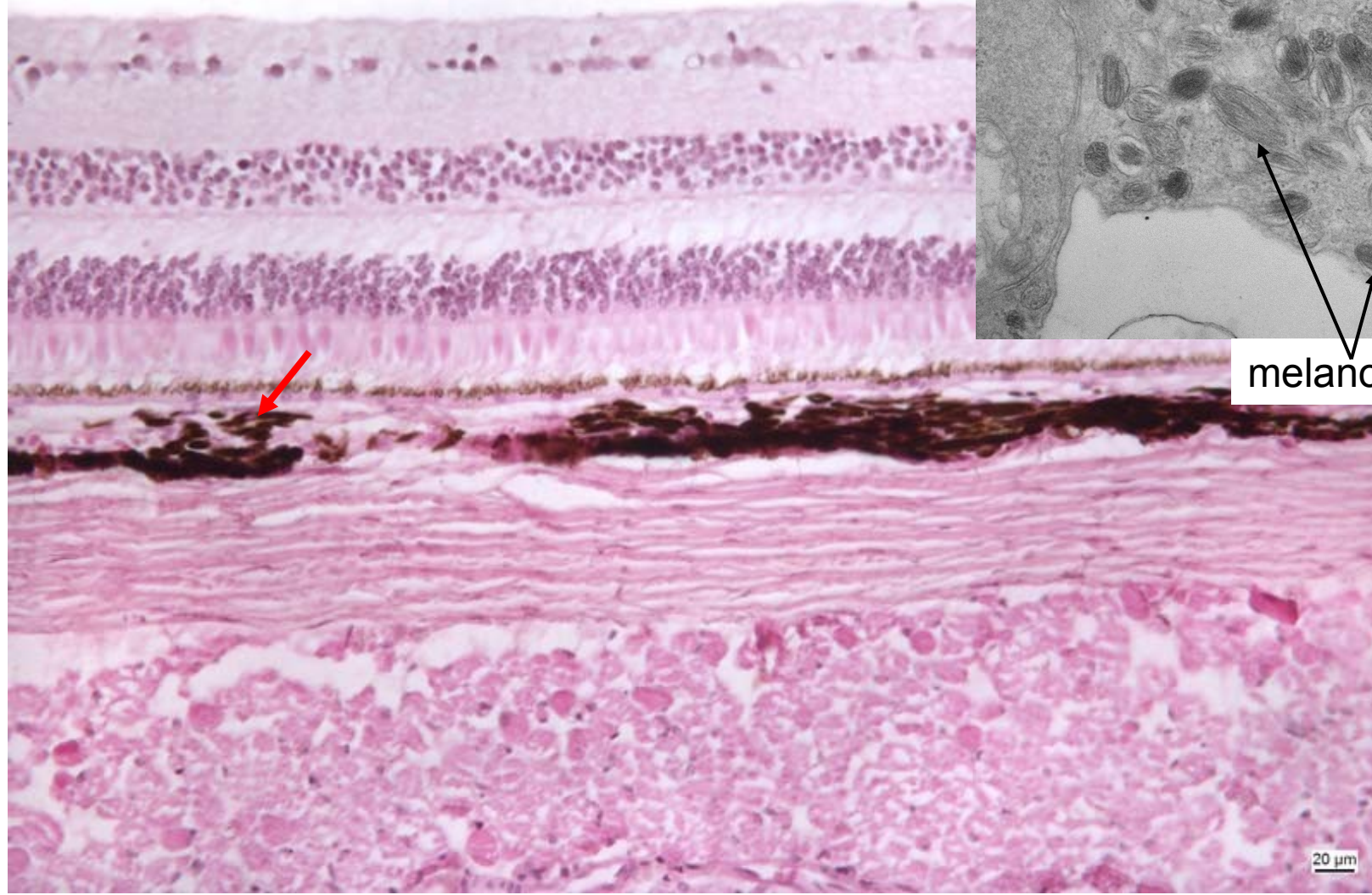
Fibroblast + collagen fibrils



Fibroblast – synthetically active cell



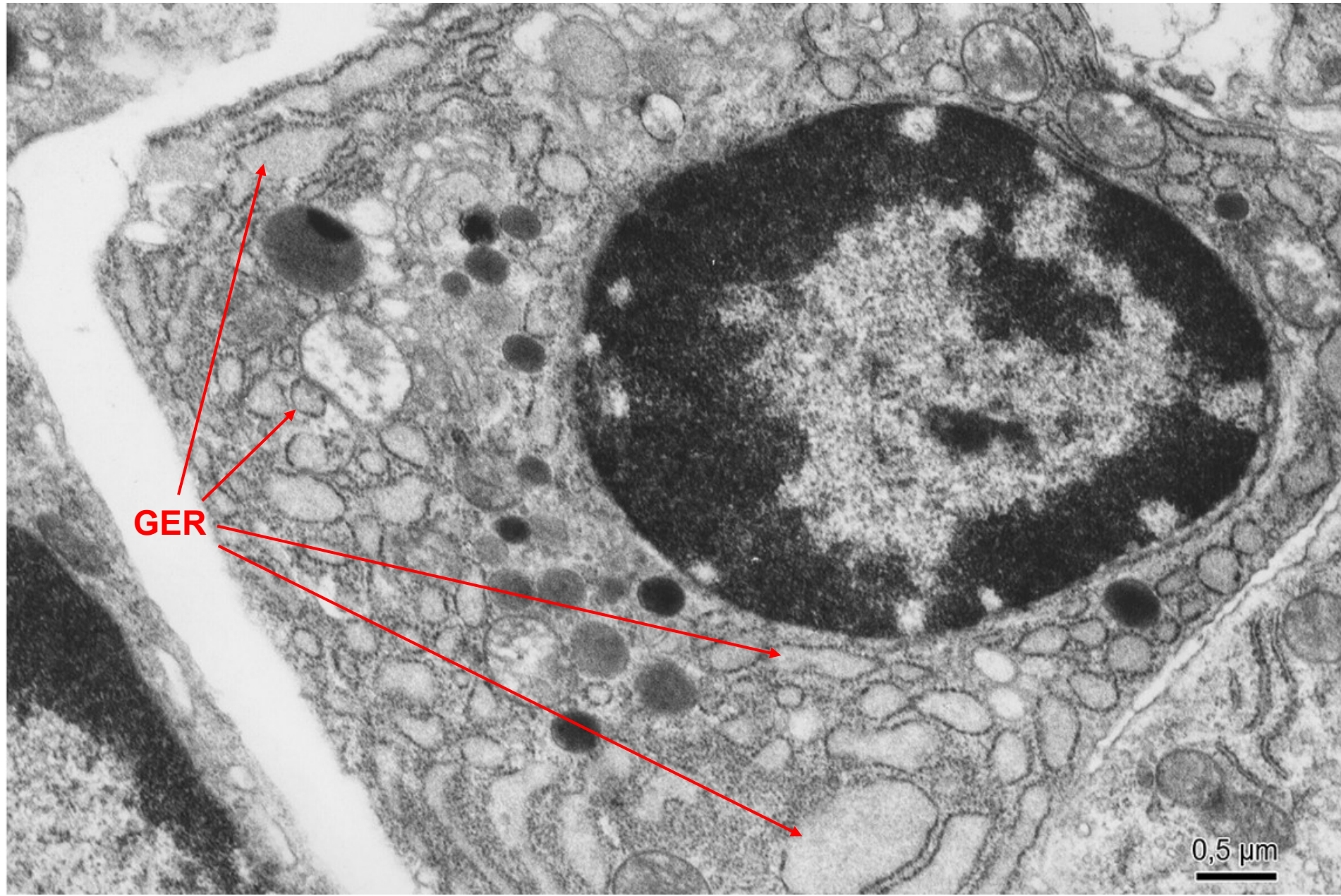
Pigment cells



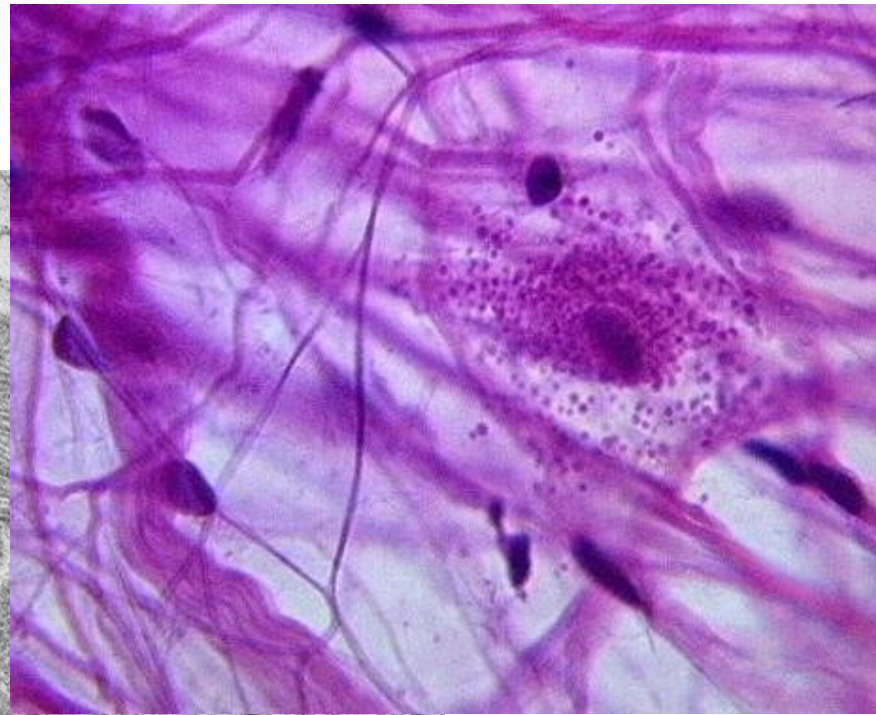
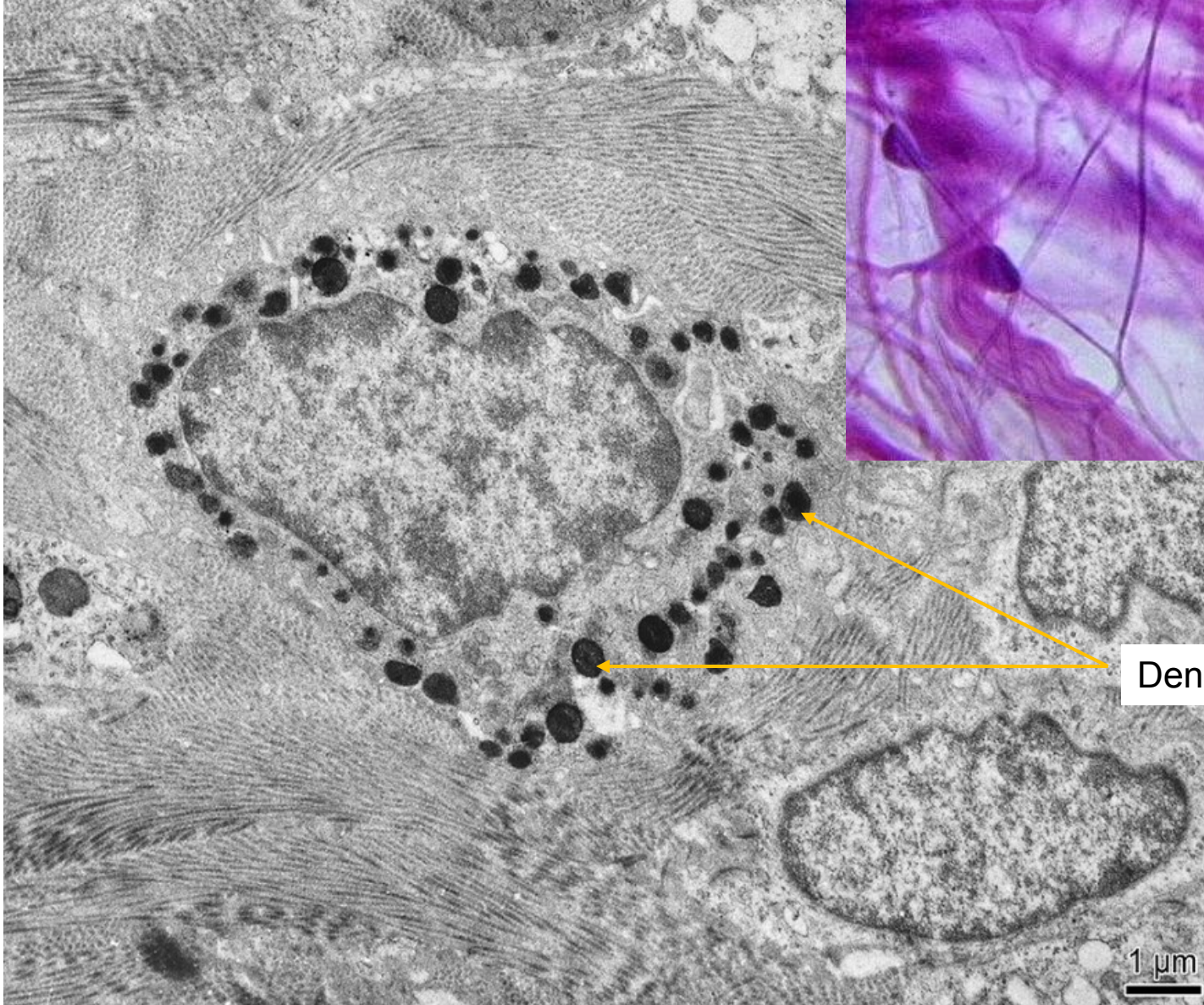
melanosomes

20 μm

Plasma cell



Mast cell



<http://www.gwc.maricopa.edu>

Dense secretory granules

Connective tissue proper

- Classification of connective tissues:

CTs are classified on the basis of types of cells, fibres and ground substance, and on the organization of fibres.

- Loose
- Dense (regular, irregular)
- Elastic
- Reticular
- Adipose
- Mucose
- Mesenchyme

Connective tissue proper

- Loose (areolar) CT:
 - cellular type of CT – all types of cells
 - abundant ground substance and thin and relatively sparse fibres (mainly collagenous)
 - viscous gel-like consistency
 - important for the diffusion of oxygen and nutrients, phagocytosis; connective function
 - examples of occurrence: **beneath epithelia as lamina propria, submucosa** – distinct layer of the wall of hollow tubular organs

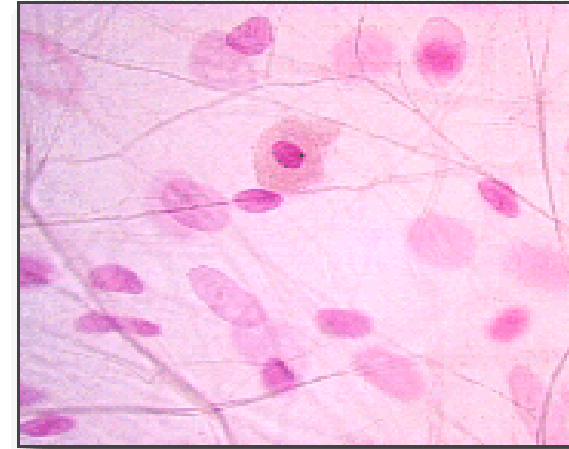
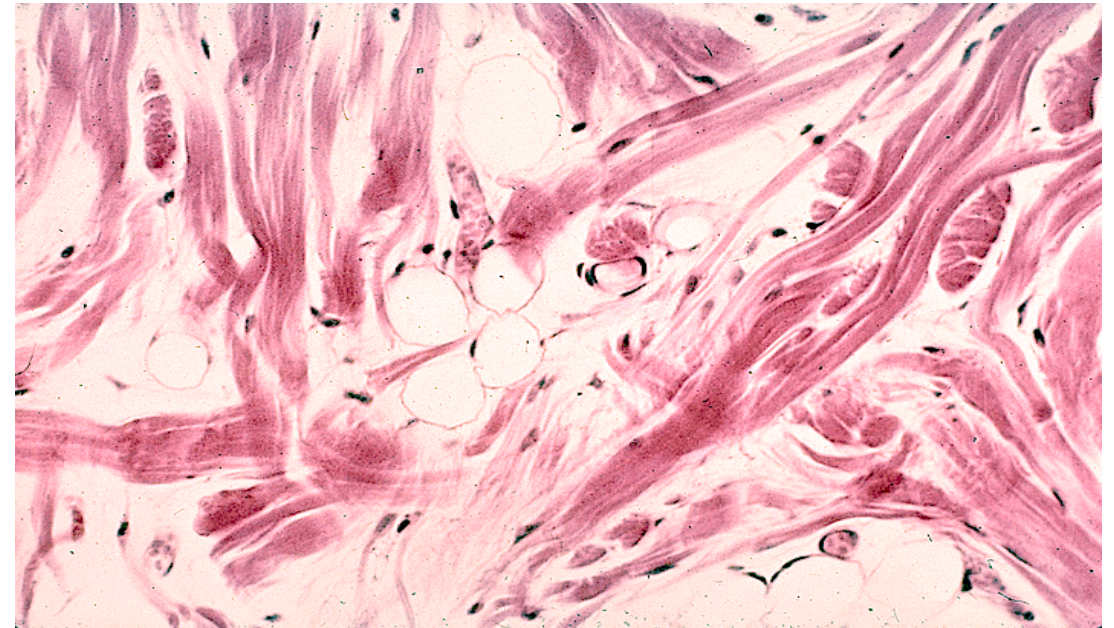
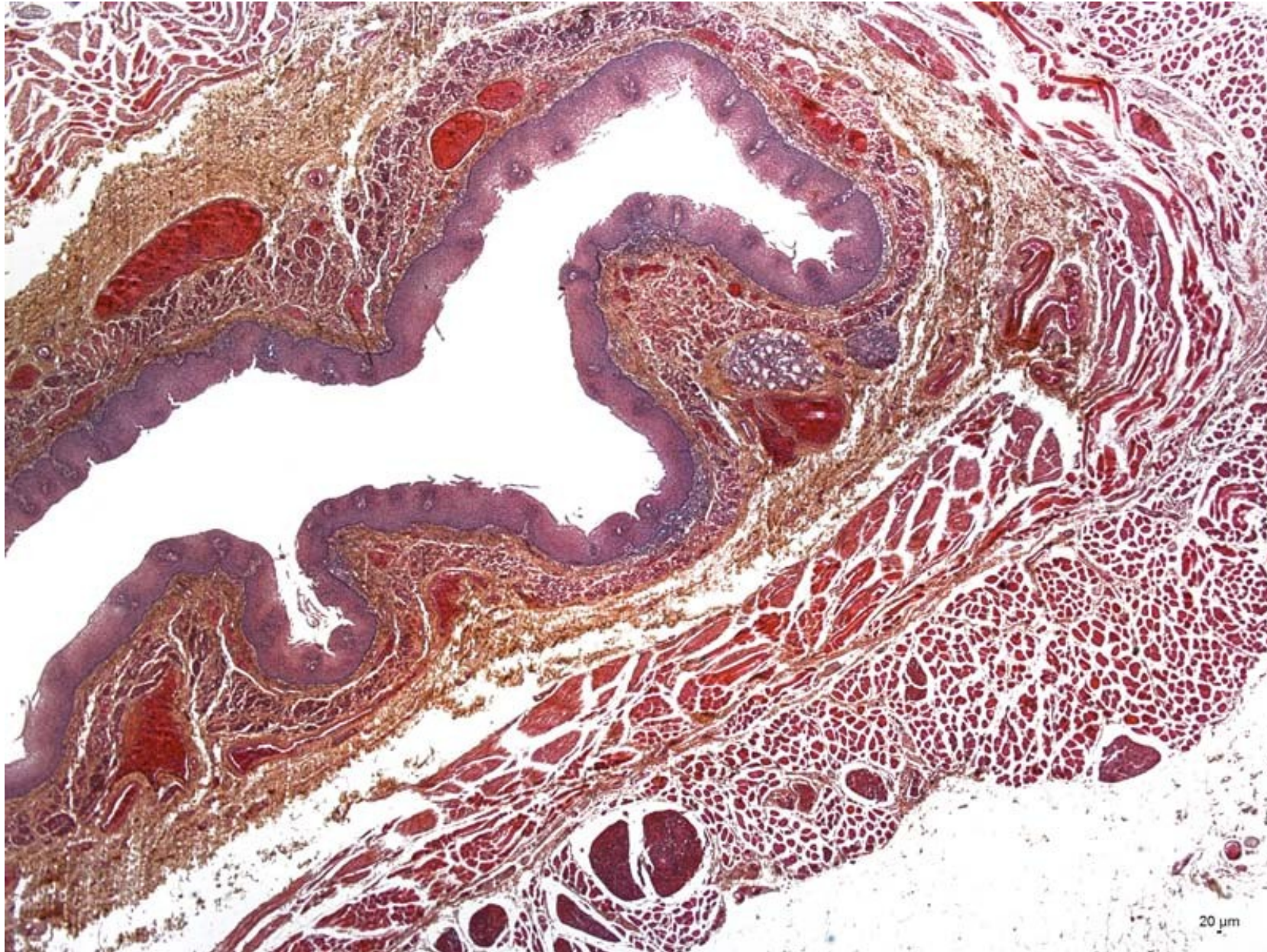


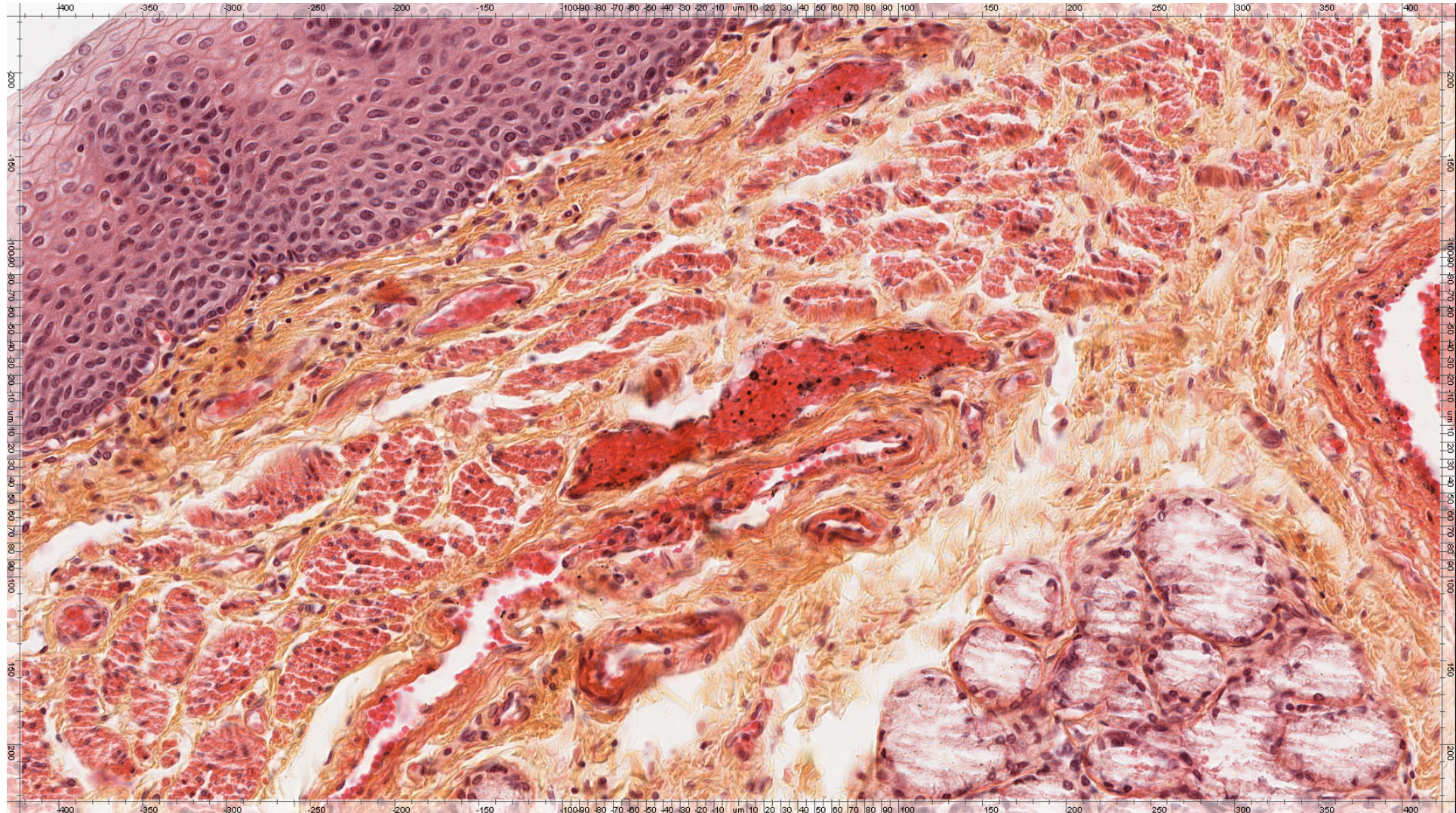
Figure 1



Loose connective tissue - Esophagus



Loose connective tissue – mucosa, submucosa



Connective tissue proper

- Dense regular CT:
- collagenous fibres packed in dense regular arrays, between which lie rows of cells.
- examples of occurrence: in tendons, ligaments, (some also contain large amounts of elastic fibres and are called elastic ligaments), and aponeuroses.

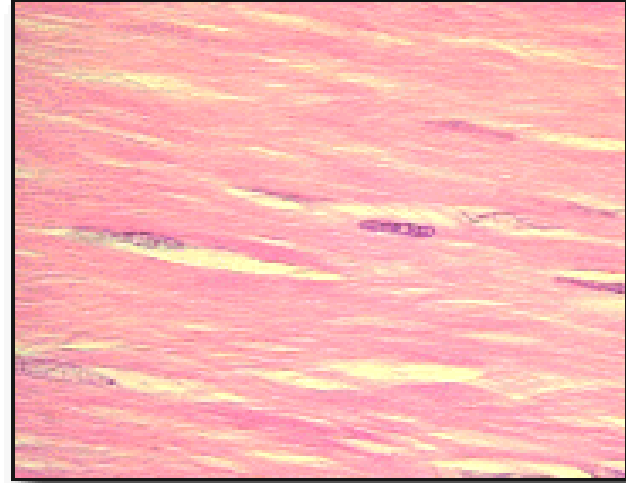
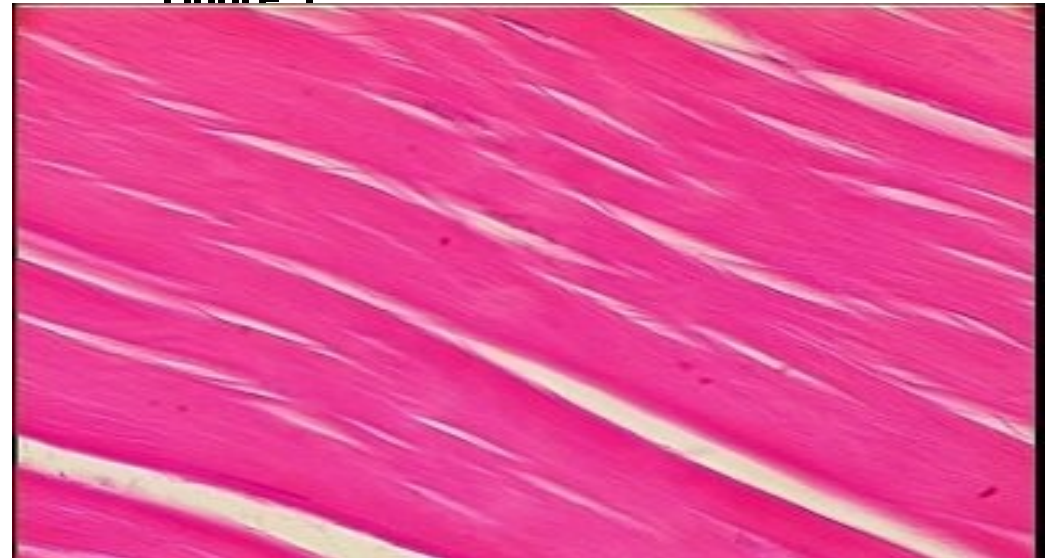
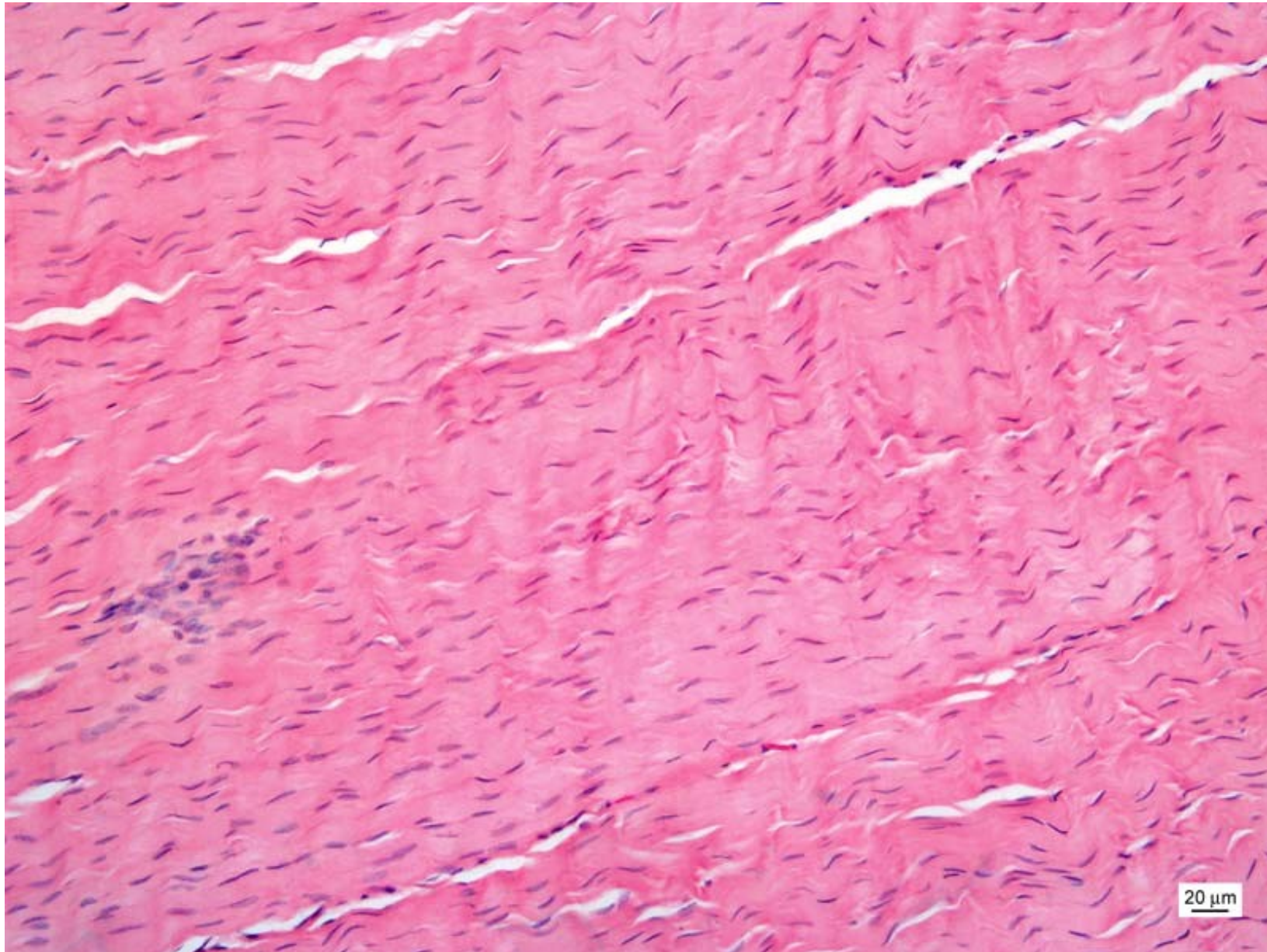


Figure 3

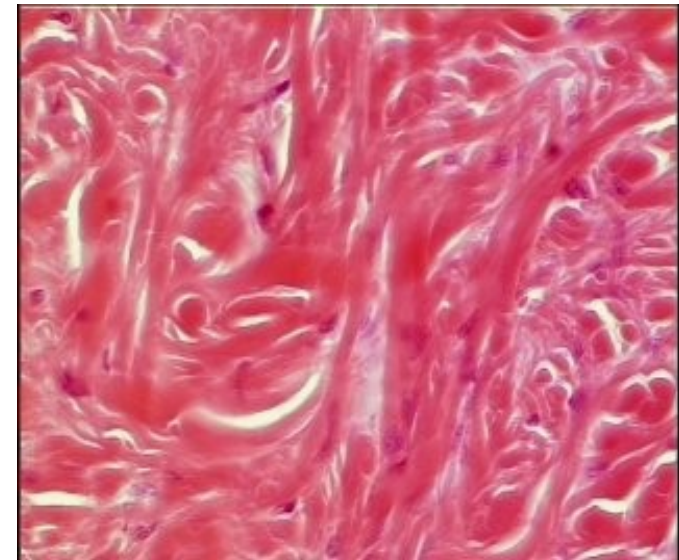


Dense connective tissue regularly arranged - Tendon

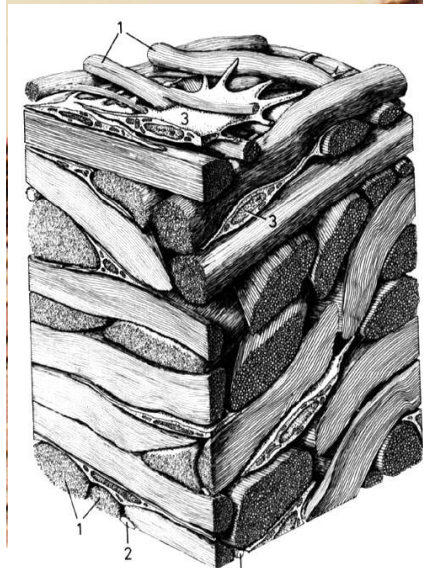
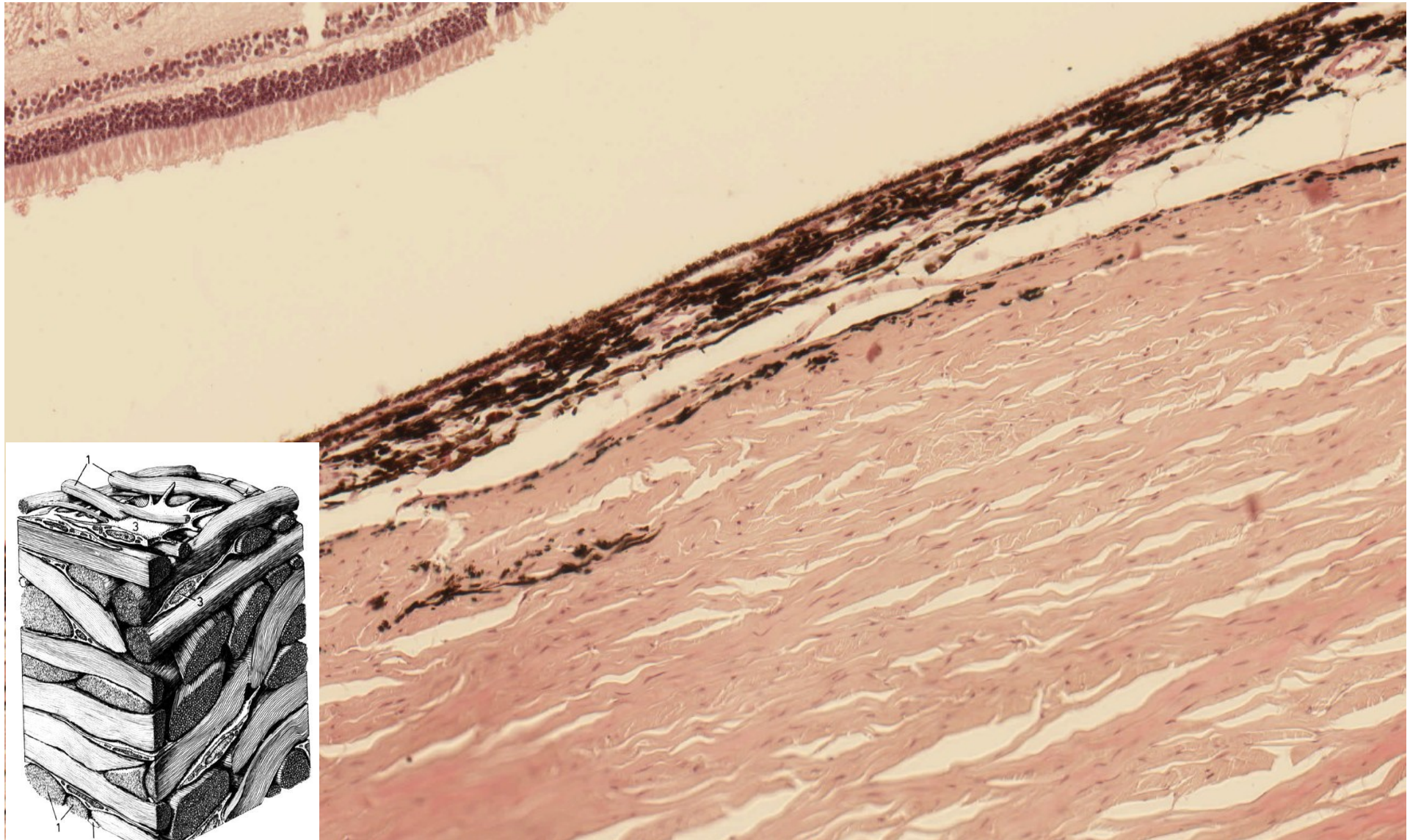


Connective tissue proper

- Dense irregular CT:
- collagenous fibres form a bundles running in various directions (hence irregular)
- fibroblasts are usually the only cell type present
- little ground substance is present.
- examples of occurrence: is found on the outside of many organs as fibrous capsule, in the dermis of the skin

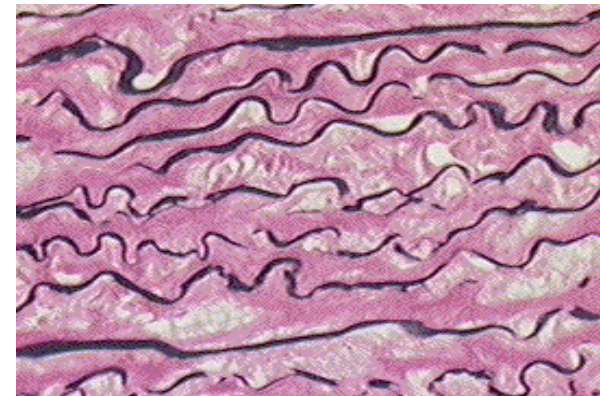
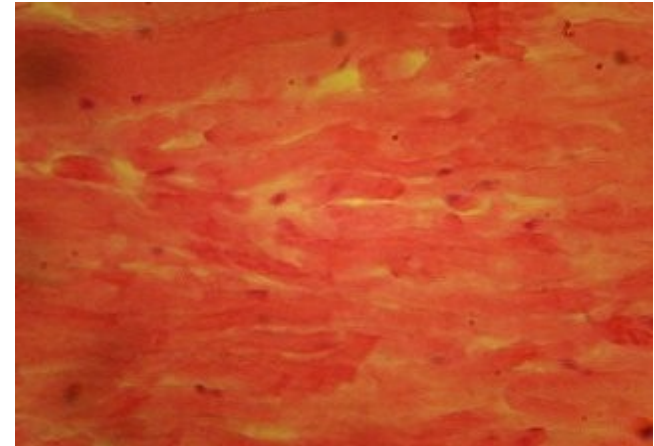


Dense connective tissue irregularly arranged - Sclera



Connective tissue proper

- Elastic CT:
- bundles of elastic fibres (elastin causes yellow colour of CT)
- cells and ground substance are scarce
- mechanical functions
- examples of occurrence: **ligamenta flava (spinal column), vocal ligaments**
- **elastic fibres are present as elastic membranes in the wall of blood vessels (*selective staining* →)**



Elastic membranes - Aorta



Connective tissue proper

- Reticular CT:
- consists of reticular cells and reticular fibres
- serves as supporting network for free cells (lymphocytes or hematopoietic cells)
- examples of occurrence: **some lymph organs, bone marrow**

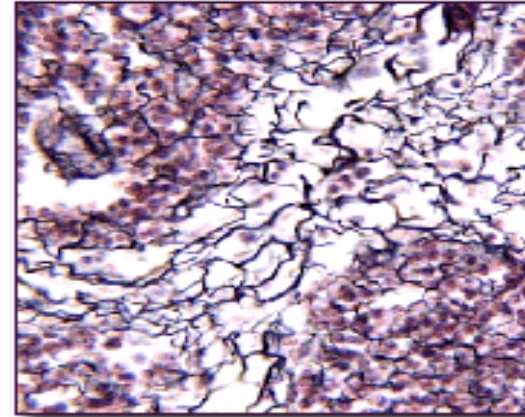
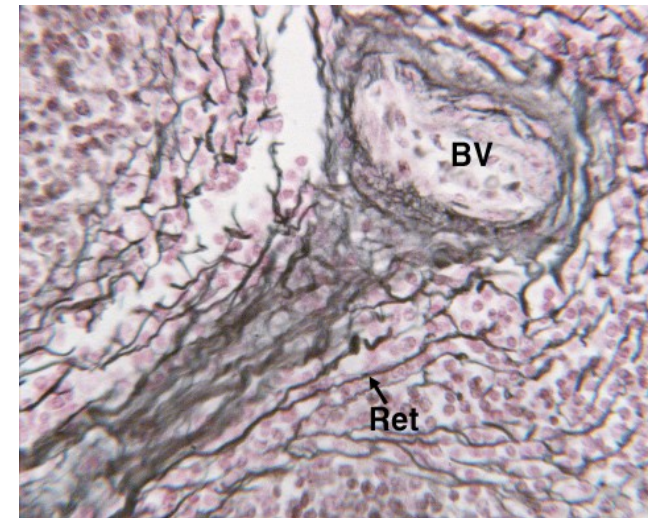
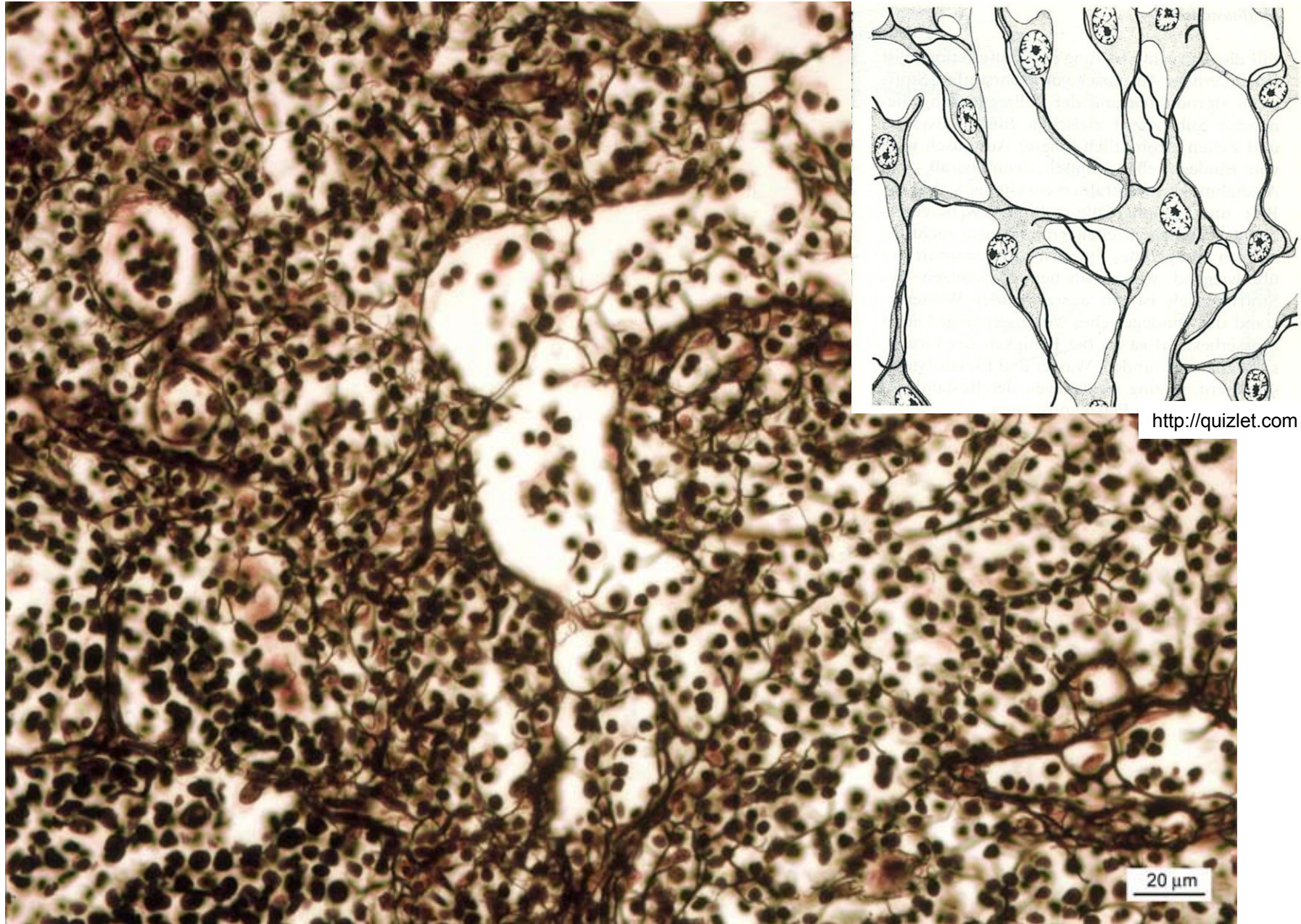


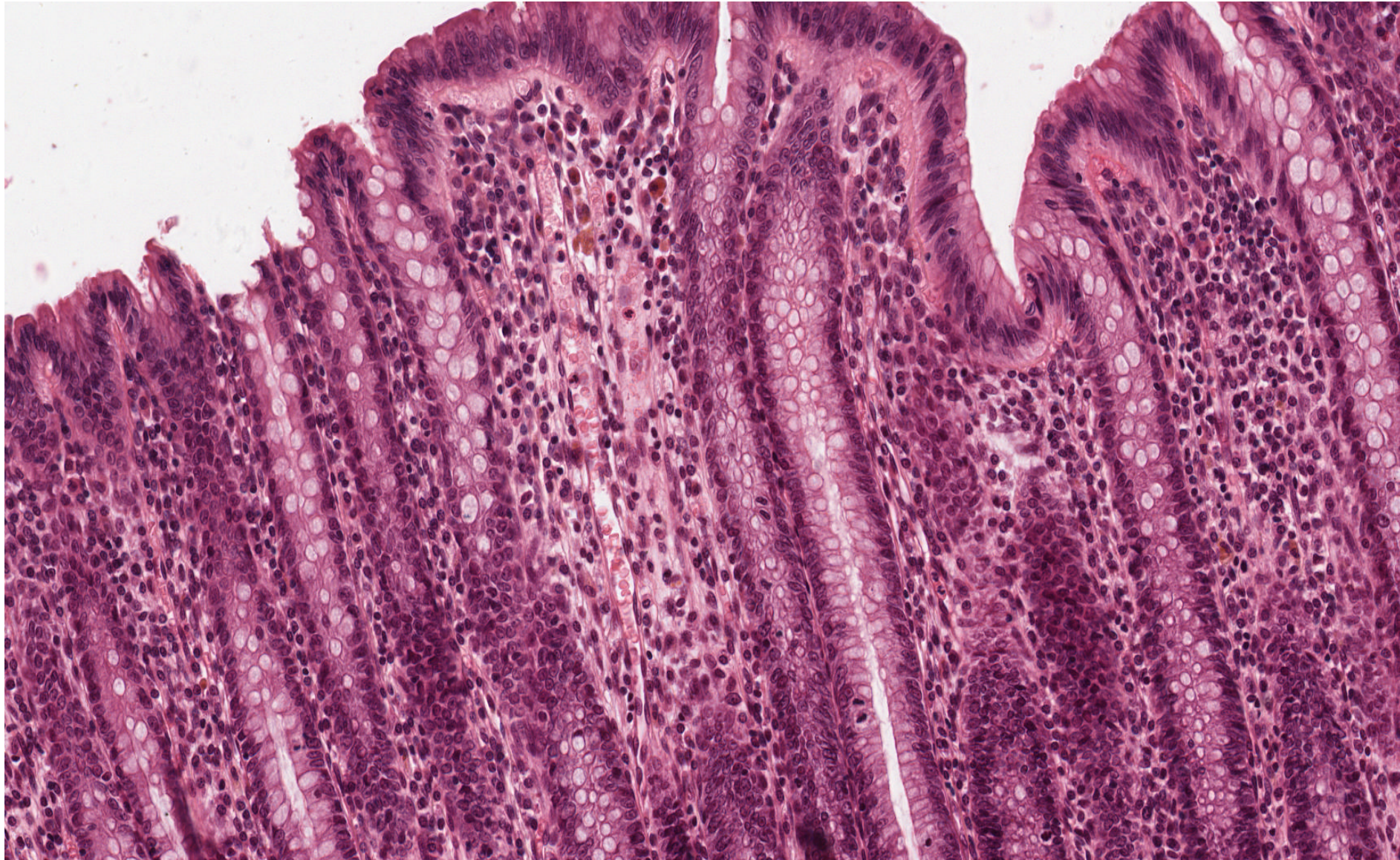
Figure 16



Reticular tissue – Lien (impregnation)



Reticular connective tissue - intestinum crassum (HE)



Connective tissue proper

- Adipose CT:
- adipocytes are specialized to store fat, are found throughout loose connective tissue
- adipocytes are the predominant cell present, the tissue is called adipose tissue
- in **white or unilocular** fat, adipocytes contain a single, large lipid droplet surrounded by a thin layer of cytoplasm. The lipid mass compresses the nucleus to an eccentric position, producing a "signet ring" appearance.
- A different kind of adipose tissue is known as **brown or multilocular** fat, contains fat droplets of varying sizes. The cells are smaller than those of white fat, with an eccentric round nucleus. Brown fat has a very limited distribution in adult humans, but is found in many animals. In hibernating animals, the oxidation of brown fat warms the blood flowing through it during arousal from hibernation. Human newborns, whose large surface to volume ratio can result in heat loss, also have a lot of brown fat. Most of it disappears during the first decade of life.

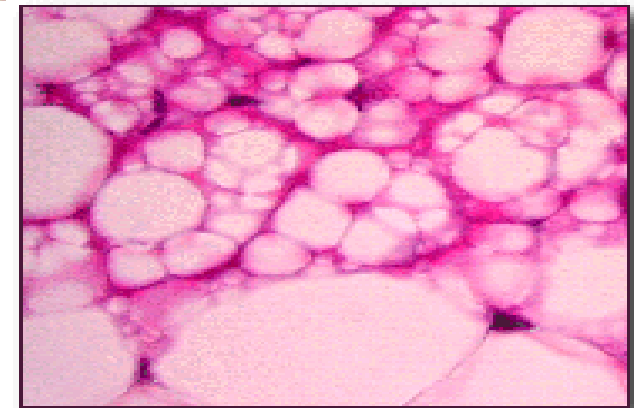
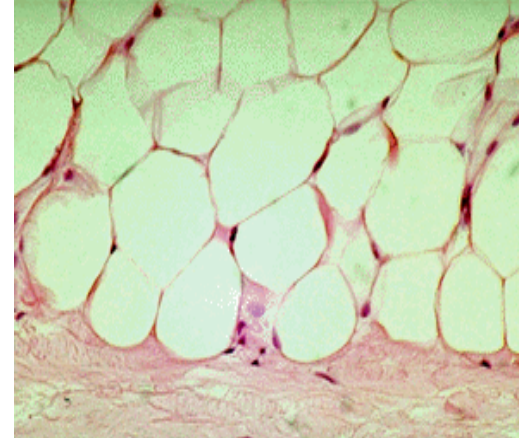
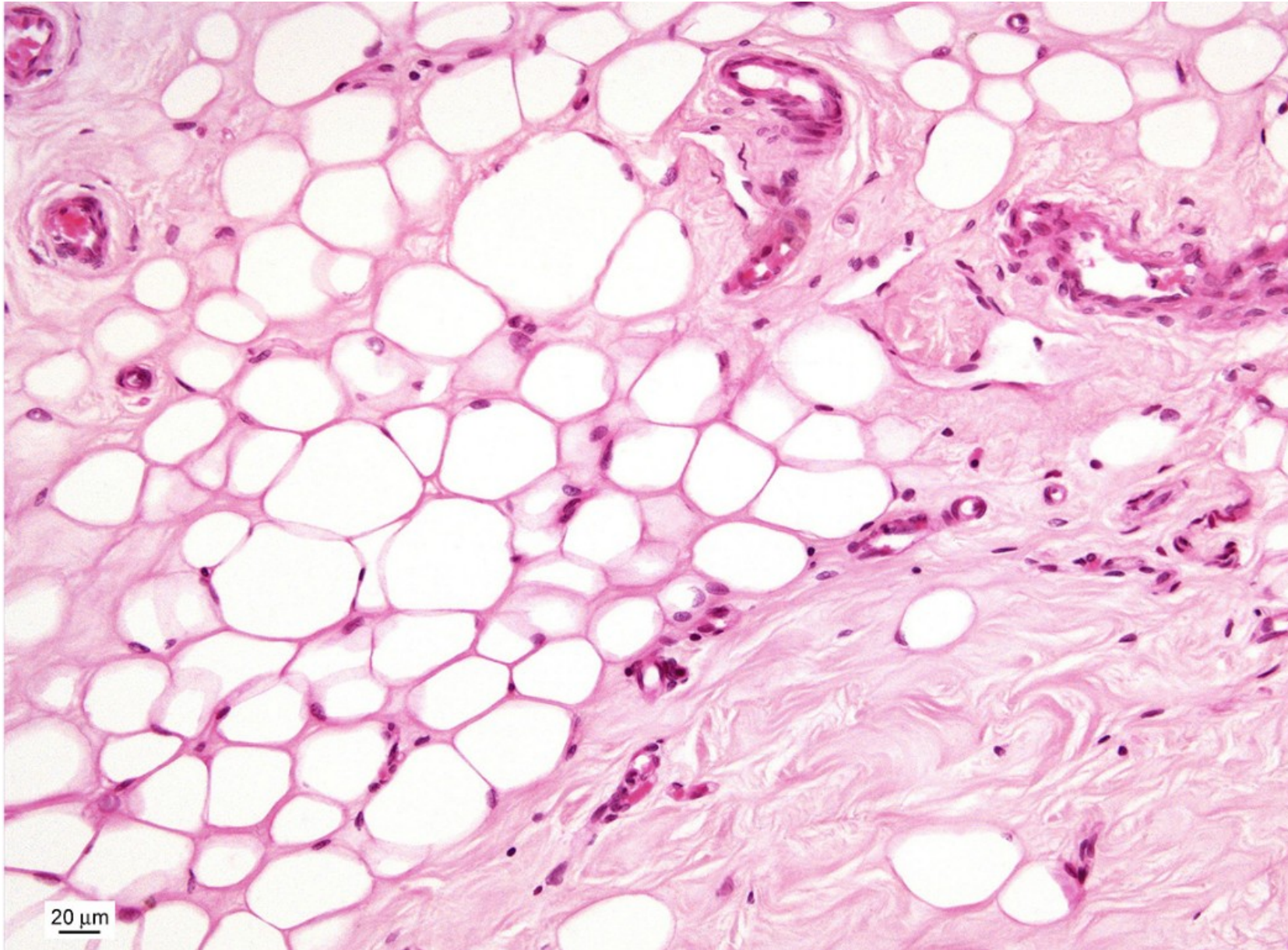


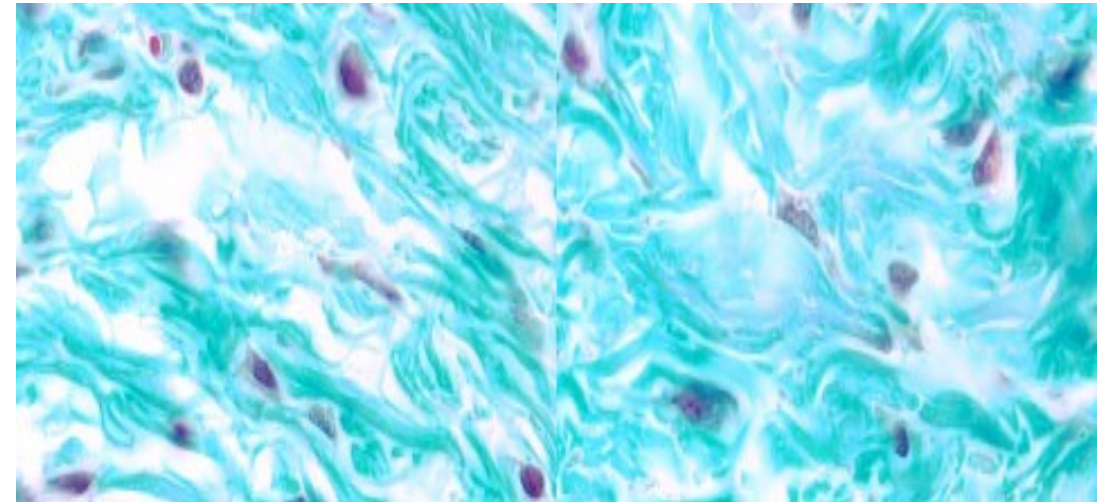
Figure 6

Adipocytes

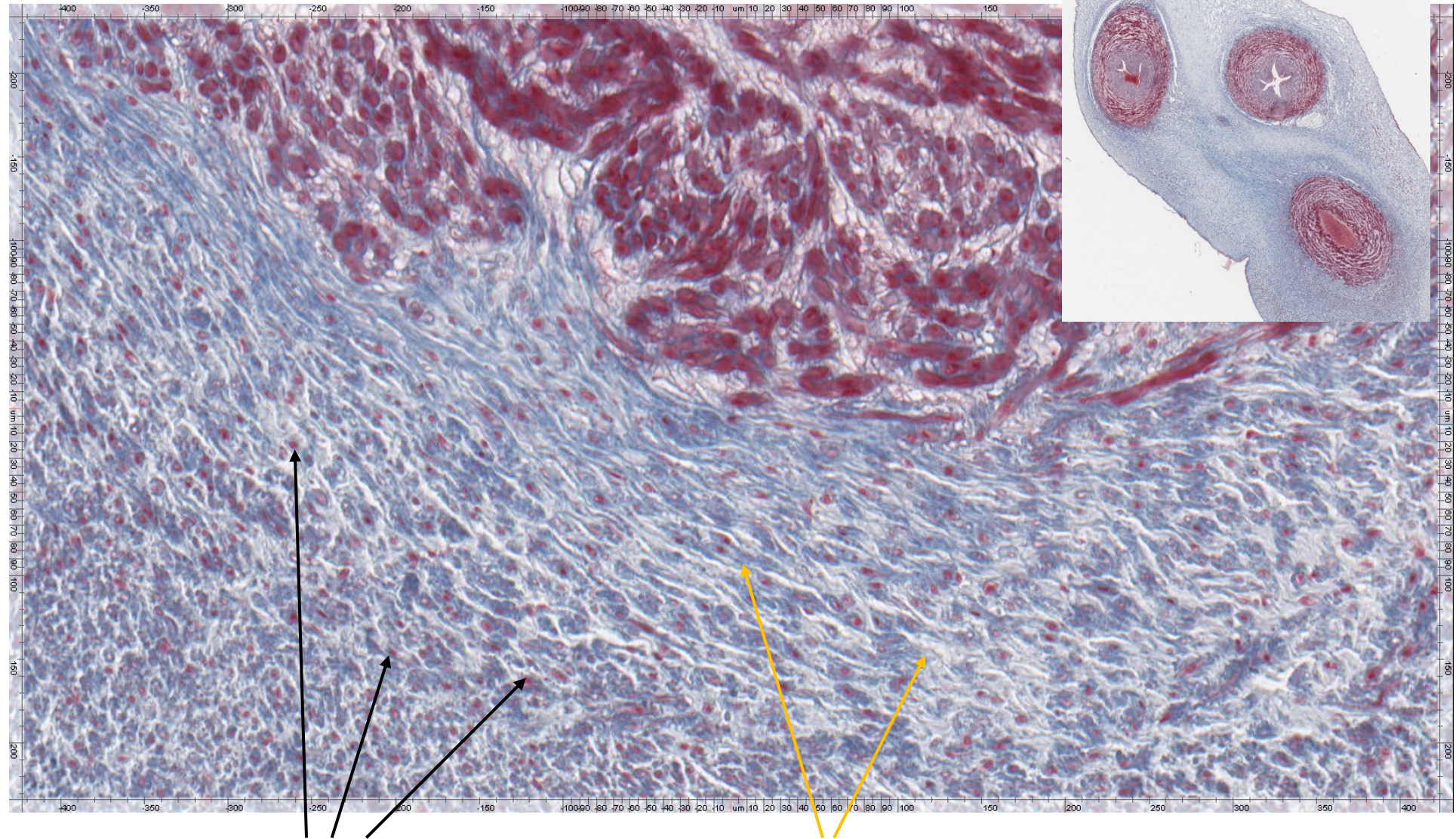


Connective tissue proper

- Mucose CT = Wharton's jelly
- present in the umbilical cord (in adults is found in the iris of the eye and in dental pulp of deciduous teeth.)
- the ground substance is more viscous or jelly-like than in mesenchyme, fibroblasts are the predominant cell type, and the number of fibres increases with age.



Wharton's jelly – Funiculus umbilicalis



Nuclei of fibrocytes - **violet**

Collagen fibers - **blue**

Connective tissue proper

- Mesenchyme:

- contains fairly uniform appearing, small spindle-shaped cells whose processes extend and contact those of other cells to form a three dimensional cellular network, ground substance fills the intercellular spaces, fibres are present, but are very fine and sparse.

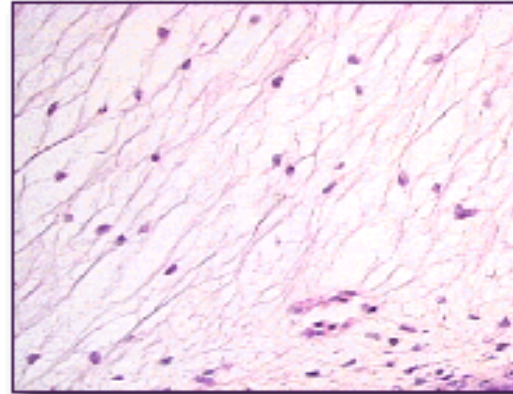
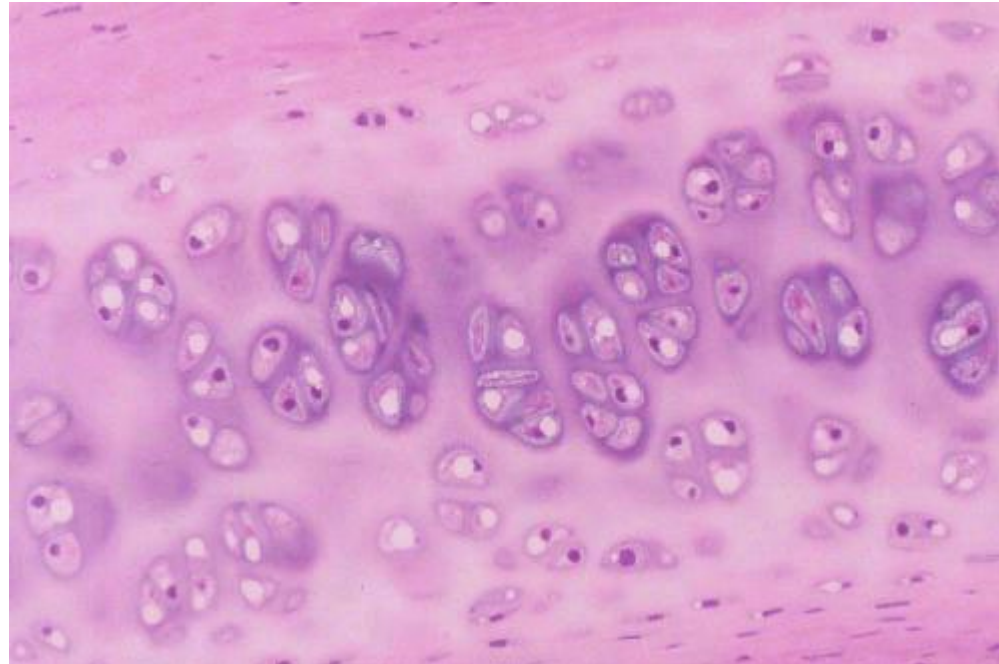


Figure 17

Cartilage

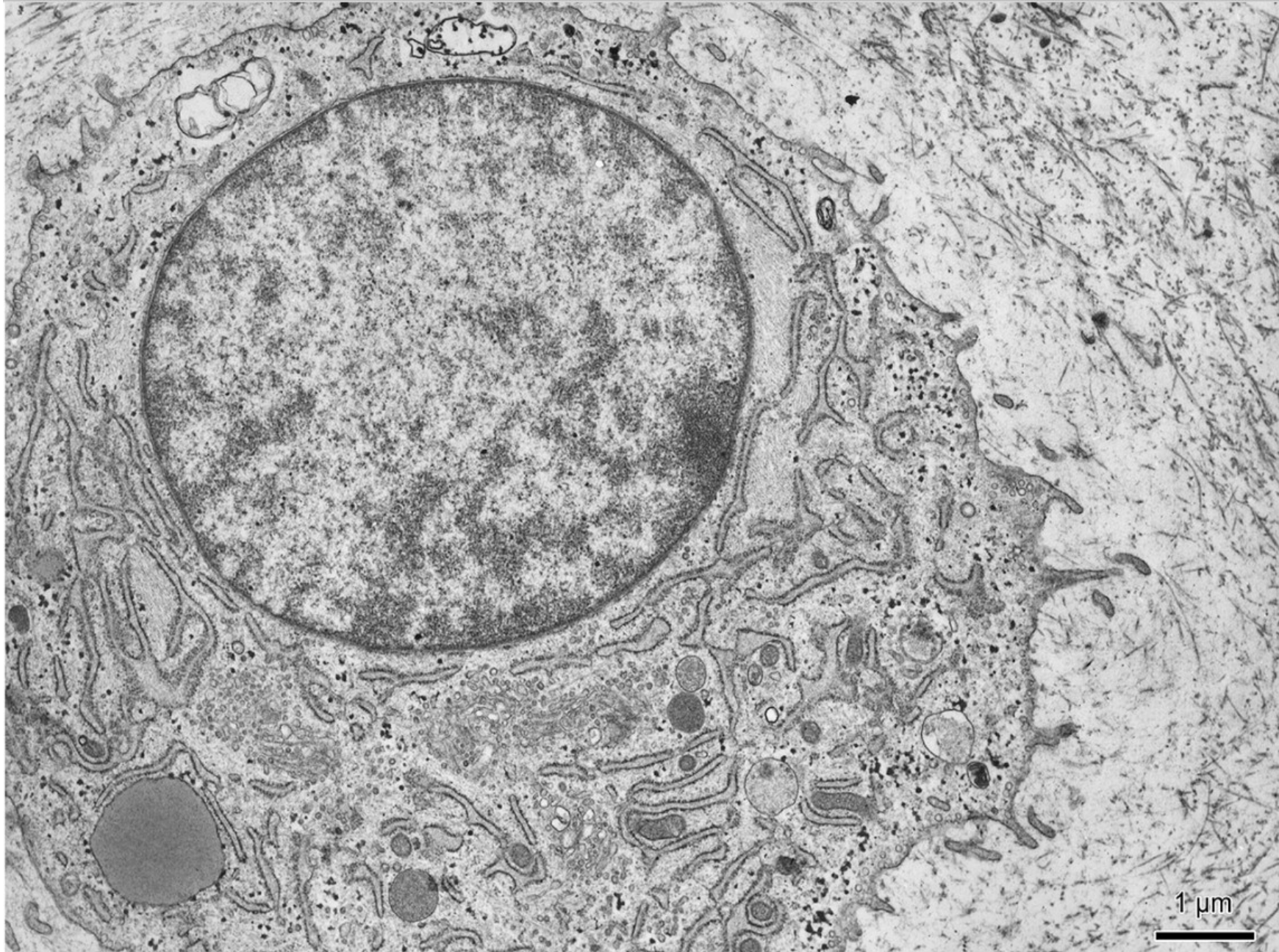
- type of connective tissue whose cells, called **chondrocytes**, secrete extracellular substances of ground mater and proteins of collagen or elastic fibres



Cartilage

- Consists of :
- **chondrocytes (and chondroblasts)** in lacunae
- extracellular matrix.
 - **MATRIX:** Consists of FIBERS + GROUND SUBSTANCE
 - **ground substance:** Consists of **glycosaminoglycans** - **hyaluronic acid, keratan sulfate, chondroitin sulfate.**
 - basophilia of matrix is due to the glycosaminoglycans
 - **AVASCULAR:** All cartilage does not contain blood vessels
 - nutrients are received via passive diffusion from blood vessels in the perichondrium.
 - **PERICHONDRIUM:** Dense connective tissue (capsule) around the cartilage.
 - is not found on the articular surfaces of long bones and on the surface of fibrocartilage.
 - contains blood vessels which provides nutrients to the cartilage.

Chondrocyte



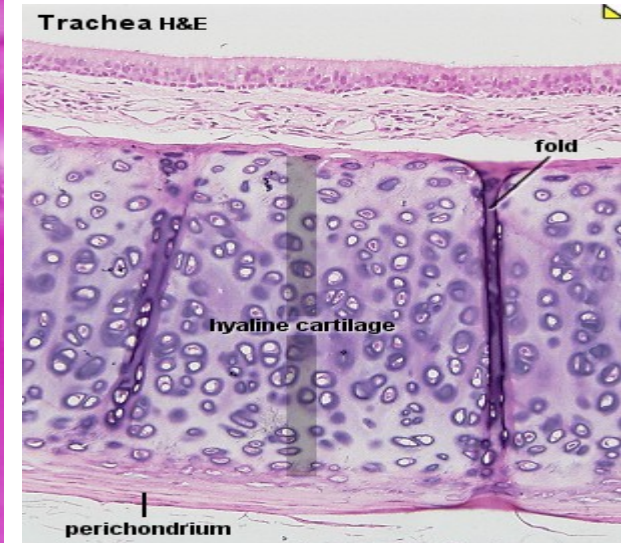
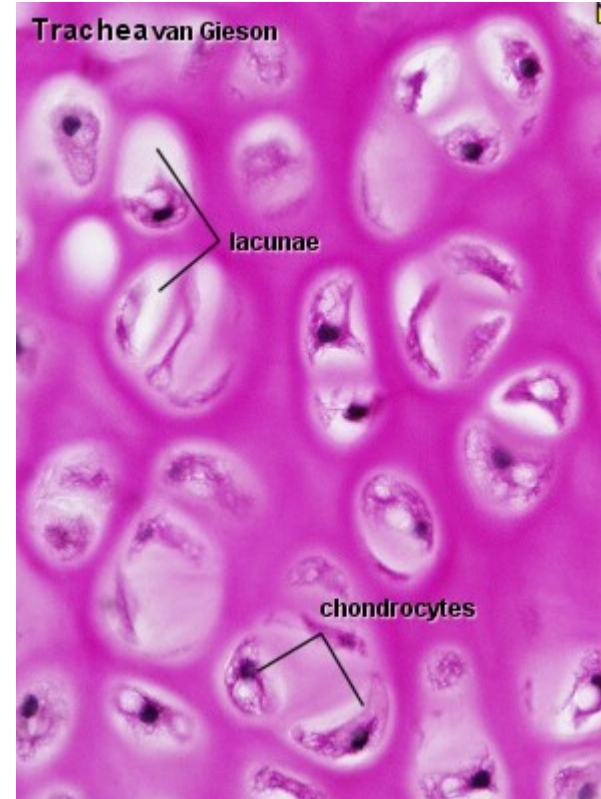
Cartilage

- **CHONDROCYTES (and chondroblasts)**: are located within little caves called **lacunae**, which they fill during life, during tissue preparation, chondrocytes shrink and frequently fall out, and lacuna appear only partially filled or empty.
 - they have lots of ER and secrete the extracellular matrix components .
 - produce **matrix components**: 1) glycosaminoglycans - hyaluronic acid, keratan sulfate, chondroitin sulfate, 2) chondronectin (increases adhesiveness of chondrocytes to matrix) .
 - produce **proteins of fibers** (collagen II or elastin). Collagen fibers are not distinguishable in slides as their refractive index is almost identical to that of the ground substance.
- **DEVELOPMENT: Mesenchymal** cells aggregate to start formation. The Perichondrium is formed.
 - **INTERSTITIAL GROWTH**: Formation of isogenous groups leads to an expansion of the cartilage from within the cartilage (isogenous groups are mitotic clusters of chondrocytes in mature cartilage, formed by interstitial growth during cartilage development)
 - **APPOSITIONAL GROWTH**: Adding cells from the Perichondrium. Layers from the perichondrium are added from the outside perimeter.
- **CARTILAGE REGENERATION**: Regeneration is limited because cartilage is avascular. (some appositional regeneration can occur from chondroblasts around the periphery)

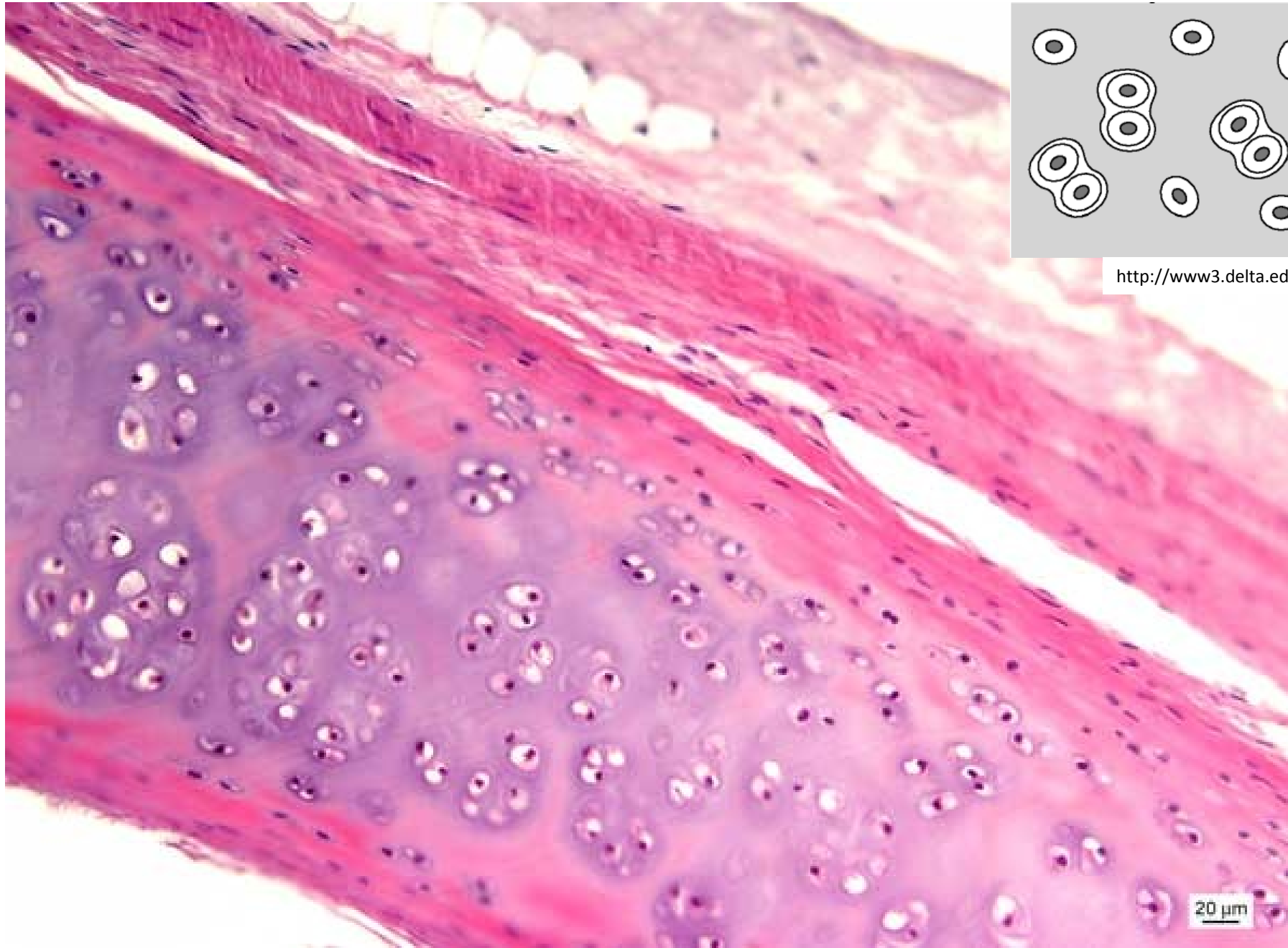
Cartilage - types

HYALINE CARTILAGE: most common, basic type

- FIBERS: **Collagen Type II** (Cartilaginous Collagen)
- Chondrocytes form isogenic groups, ground substance around them shows an intensive basophilia and so forms basophilic capsule – isogenic group + basophilic capsule = chondron or territorium, ground substance among them = interterritorium
 - DISTRIBUTION:
 - Nasal cartilage
 - Trachea, bronchi, large cartilages of larynx
 - Articular ends of bones



Hyaline cartilage - trachea (HE)

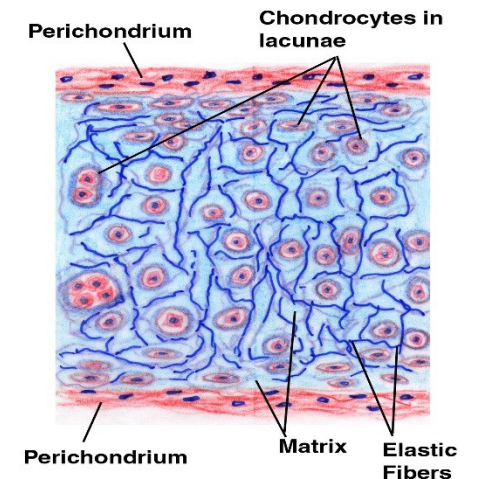
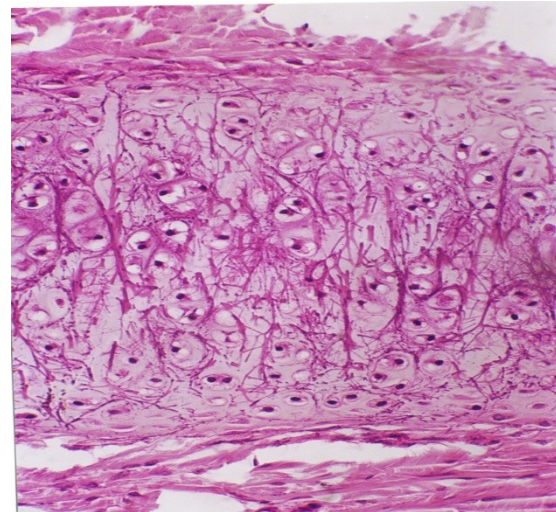


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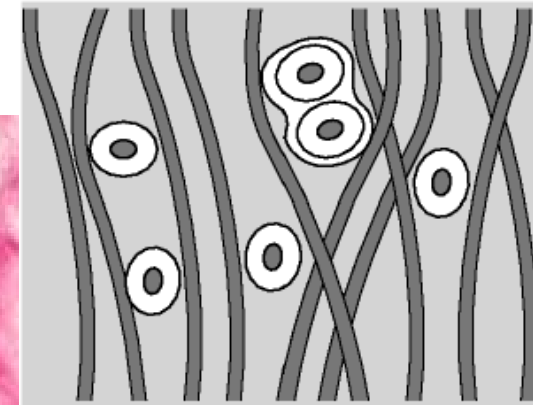
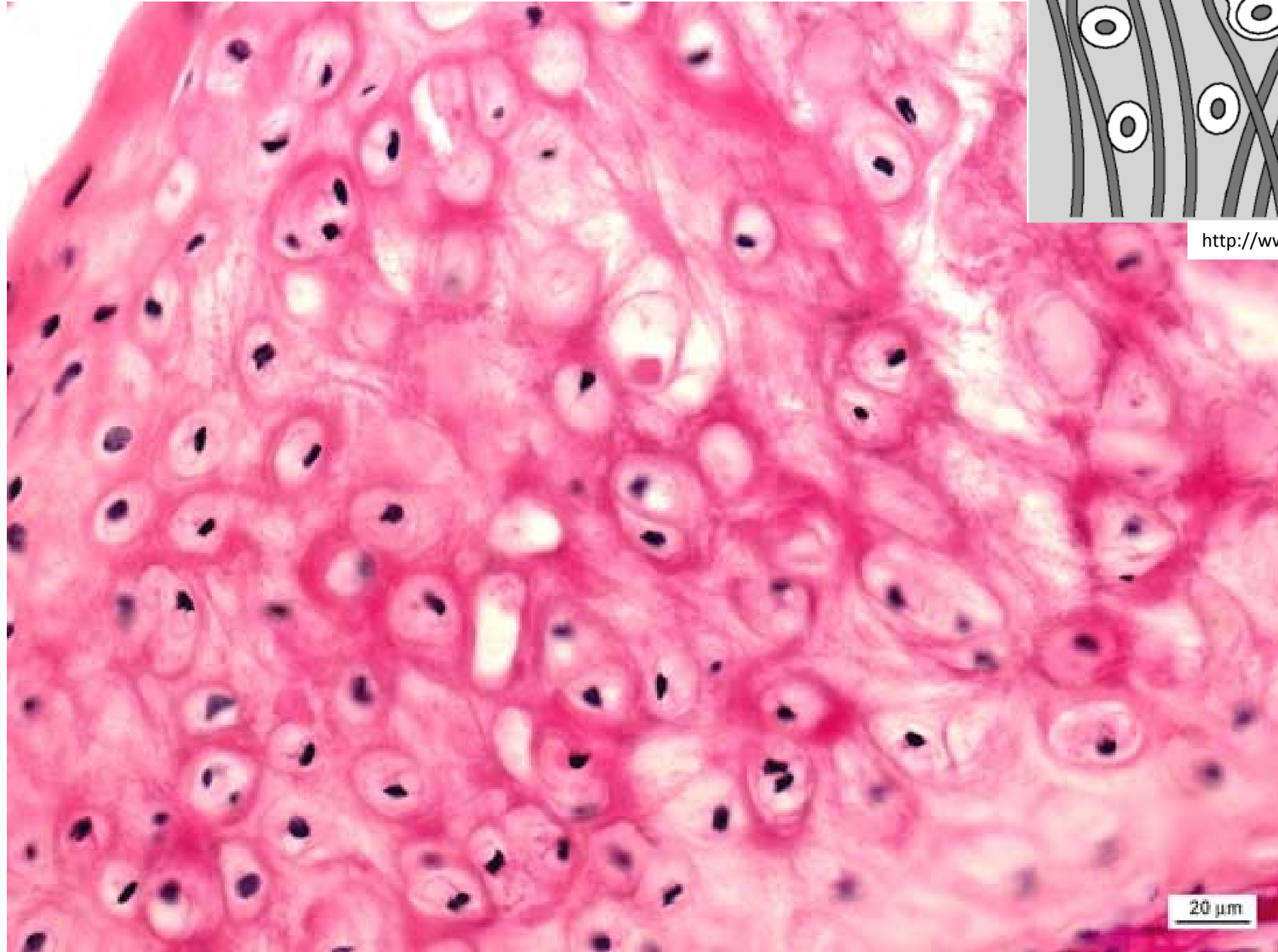
Cartilage - types

ELASTIC CARTILAGE:

- FIBERS: **Elastins** (proteins of elastic fibers)
- Chondrocytes (in lacunae) are dispersed difusely, are not arranged in isogenic groups.
- MICROSCOPIC APPEARANCE: Under low light, when you focus in and out, you can see refraction of the elastic fibers under the microscope. Elastic fibers can be visualized by staining with orcein or resorcin fuchsin.
- DISTRIBUTION:
 - Auditory tube + auricle of ear
 - Epiglottis
 - Small laryngeal cartilages



Elastic cartilage – auricula (HE)



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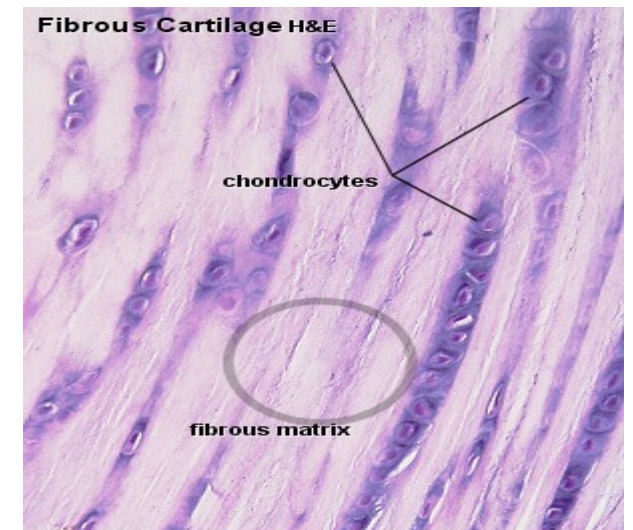
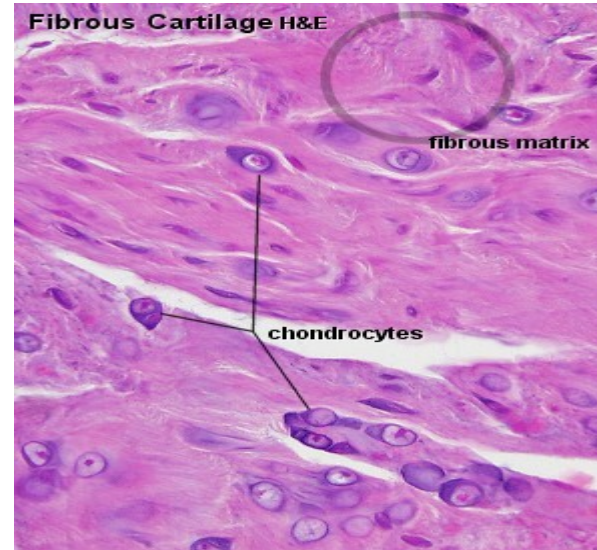
20 μ m

Elastic cartilage (fuchsin)

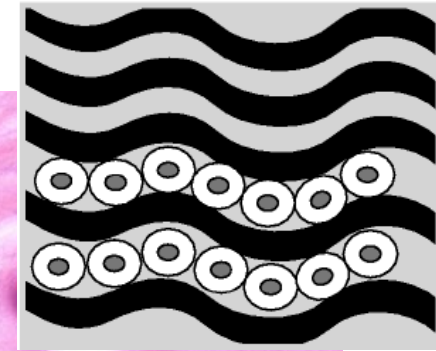
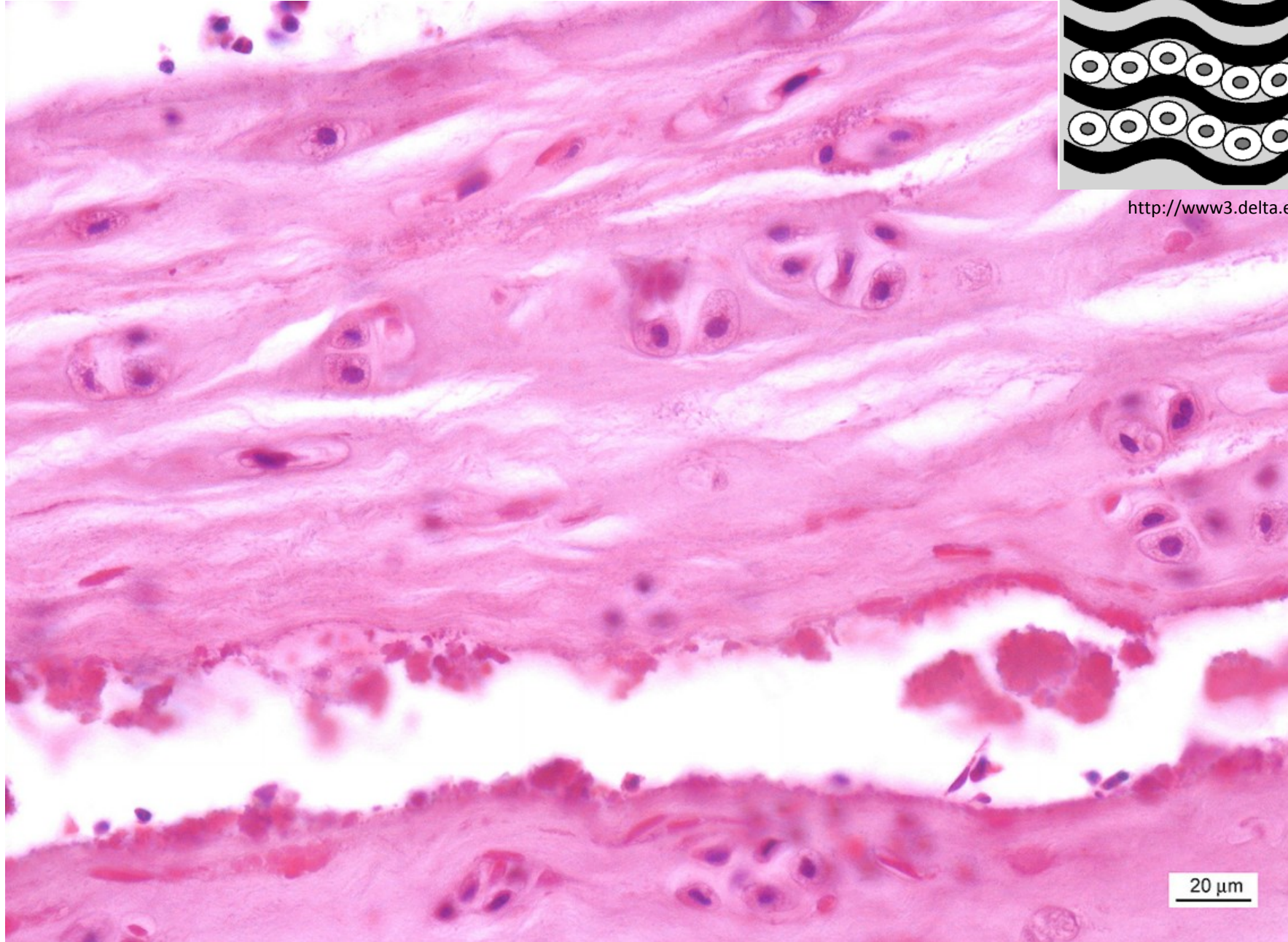


Cartilage - types

- **FIBROCARTILAGE:** Found in places where high stress occurs.
 - **DISTRIBUTION:** It never occurs alone. It is closely associated with either dense connective tissue or with hyaline cartilage.
 - Intervertebral Disks
 - Articular Disks
 - Pubic Symphysis
 - **STRUCTURE:** It has **no perichondrium**
 - **GROWTH:** It grows more like connective tissue (i.e. interstitial growth) due to the absence of a perichondrium.
 - **FIBERS: Collagen Type I**
 - Chondrocytes are not numerous, they are flattened and arranged in rows due to press of thick bundles of collagen fibers.



Fibrocartilage – intervertebral disc (HE)



<http://www3.delta.edu/>

20 μm

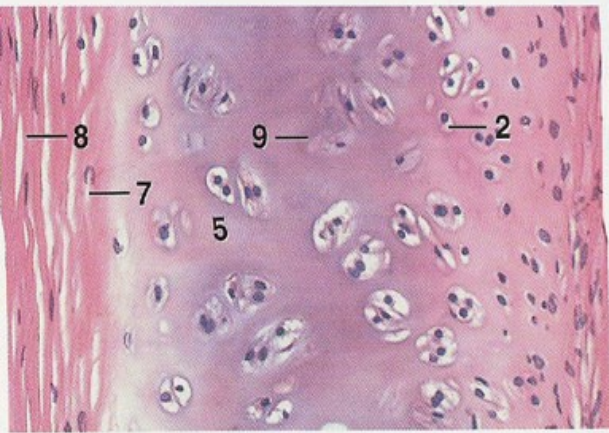


Figure 4.1 × 125

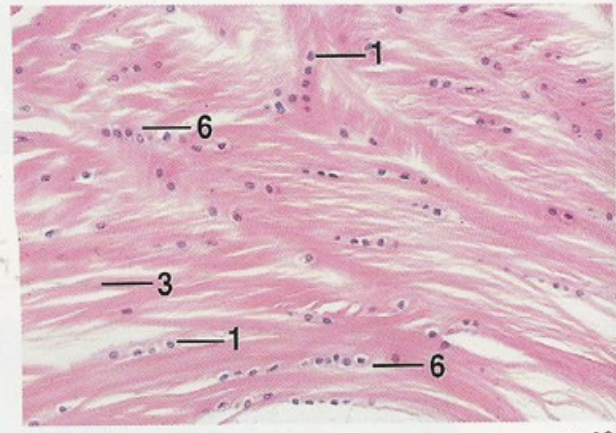


Figure 4.5 × 125

KEY	
1. Chondrocyte	6. Matrix
2. Chondrocyte in lacuna	7. Perichondrium, chondrogenic
3. Collagenous fiber	8. Perichondrium, fibrous
4. Elastic fiber	9. Territorial matrix
5. Interterritorial matrix	

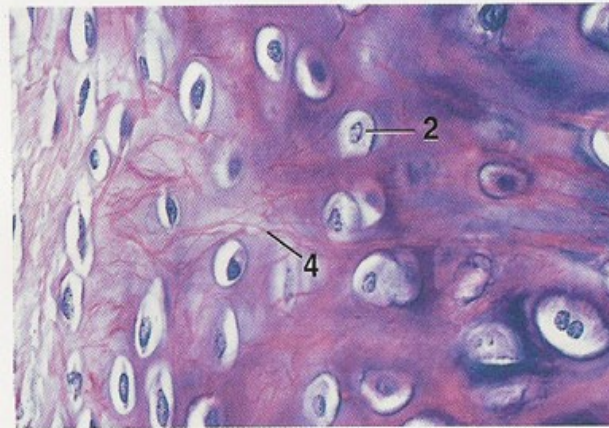


Figure 4.2 × 250

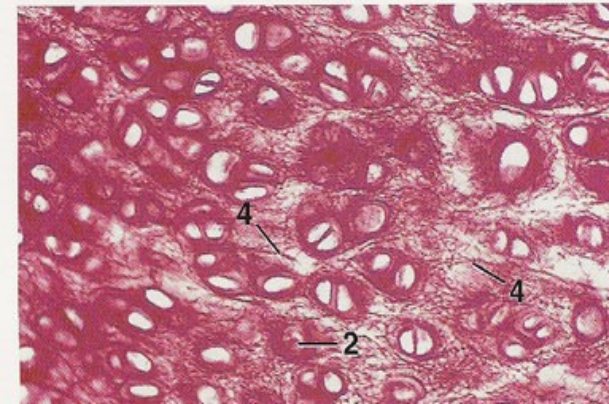
Figure 4.1. Hyaline Cartilage, Trachea, Cow. The perichondrium consists of an outer fibrous and an inner chondrogenic layer. Isogenous groups and single chondrocytes are scattered throughout the matrix.

Figure 4.2. Elastic Cartilage, Epiglottis, Dog. Pink elastic fibers can be seen throughout the cartilage matrix.

Figure 4.3. Elastic Cartilage, Wattle, Pig (Orcein). The elastic fibers are stained red with orcein.

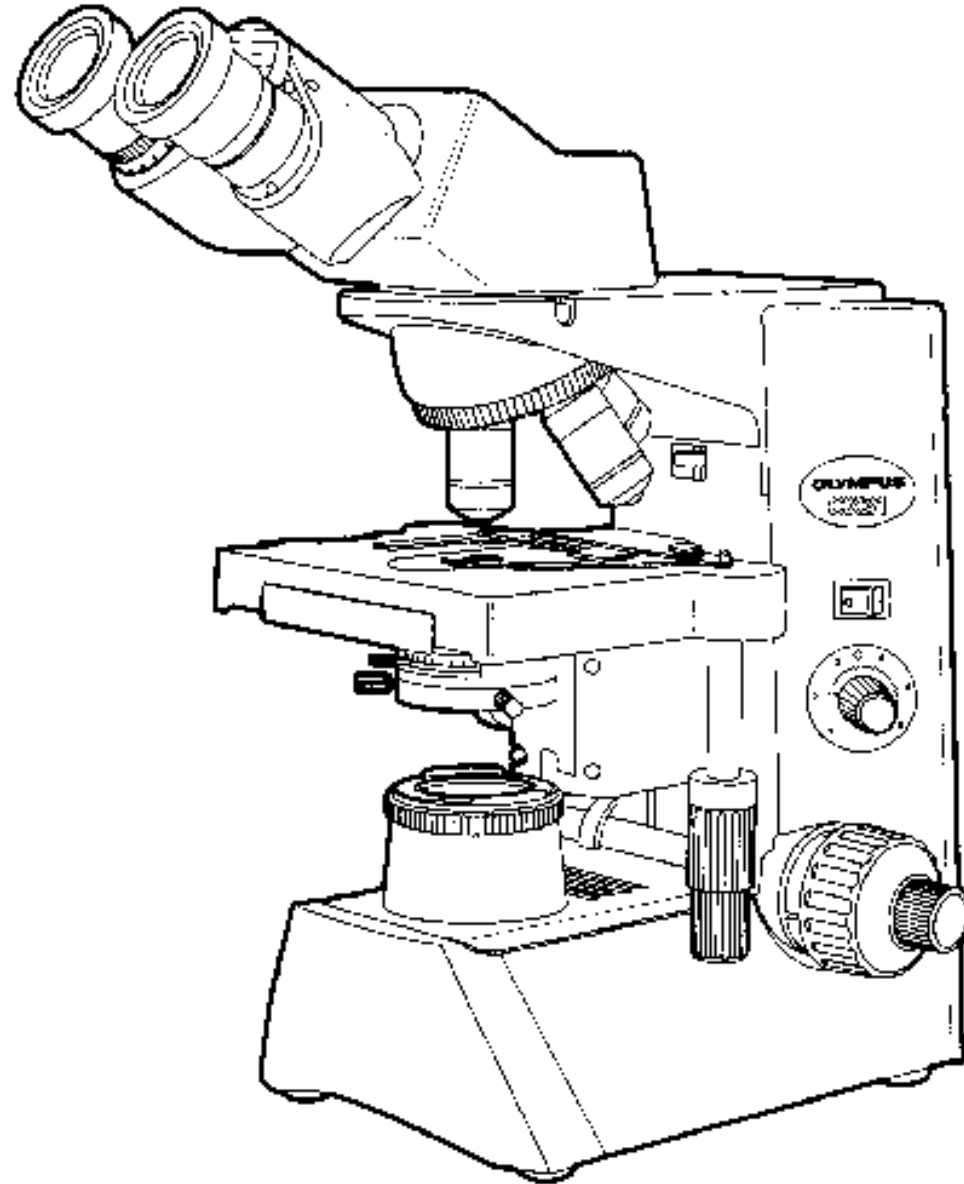
Figure 4.4. Fibrocartilage, Intervertebral Disc, Horse. Chondrocytes are arranged in rows and framed by a hazy rim of pale blue matrix. Collagenous fibers are visible between rows of chondrocytes.

Figure 4.5. Fibrocartilage, Claw, Chicken. Rows of chondrocytes are randomly scattered among collagenous fibers. Pale blue matrix is visible around some chondrocytes.



Light microscope

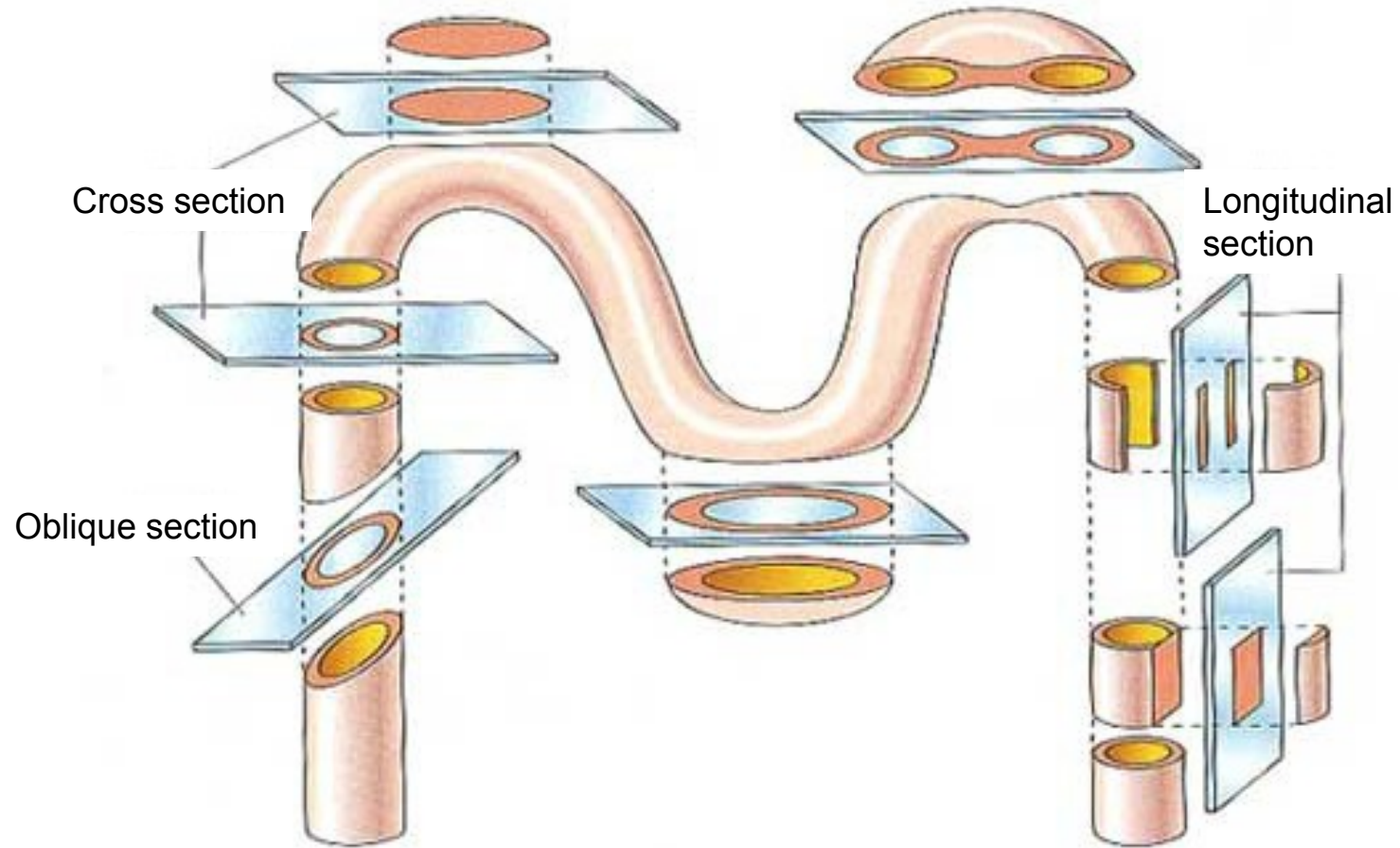
- Eyepieces
- Objective lens
- Stage with specimen holder
- On/Off
- Light control
- Condenser and Iris aperture
- Stage controls
- Coarse focus
- Fine focus
- Light source with diaphragm



Instructions

- Turn on the light.
- **Start with the 4x objective.**
- Put the slide on the stage – cover glass must be above
- Look through the scope and focus. Use the coarse focus knob at first, until the image is more or less in focus; then switch to the fine focus.
- Adjust the light. Not too bright, not too dim.
- Adjust the oculars.
- Switch to the 10x objective. A slight adjustment with the fine focus knob should get it just right. If you lose the focus and can't see your specimen at all, go back to the 4x and start again.
- Switch to the 40x objective if you want to see more detail. **Don't use the 100x!**
- When you want to look at a new slide, switch back to the 4x before changing slides.
- **Only one slide is out of the box at the moment!** Do not remove more!
- When you're done with the scope, switch to the 4x and turn the light all the way down before turning it off.
- At the end of lesson, the box with slides is checked in your presence before you leave your place

Various shapes on different sections of tubular structures



Connective tissue proper:

Slides:

Wharton's jelly (99. Funiculus umbilicalis)

Loose (areolar) connective tissue (11. Esophagus)

Dense connective tissue - irregular (89. Posterior part of the eye)

Reticular tissue (68. Lien – impregnation)

Elastic membranes (62. Aorta)

Atlas EM:

Fibroblast (42)

Plasma cell (43)

Mast cell (44)

Collagen fibers (42, 44)