HOW TO USE THE HISTOLOGY MANUAL

1. You are expected to recognize only the **structures highlighted** in the text and to know the morphological and theoretical description of them.

2. *Paragraphs in italics* help you to identify the structures. *Using italics*, we draw your attention to pathologic processes occurring frequently in a given histological localization, which stresses the marked importance of precise knowledge of the structures involved.

HOW TO STUDY THE HISTOLOGICAL SLIDES

1. Take the slide, and firstly study it with the naked eye (shape of the section, density of the tissue, colors etc.)

2. Place the slide into the microscope specimen holder **ALWAYS** with the **COVER GLASS UPRIGHT!!**

3. Always check the **WHOLE SURFACE** of the tissue section, starting with the lowest magnification followed by the medium magnification. Only change to the highest magnification when you have selected an area under the medium magnification objective lens.

4. **NEVER** start the identification of a slide with the highest magnification objective!!!

5. In the case when you are not able to identify the section, try to describe particular shapes of cells, number of cell layers, and staining properties of the cells. Check all of your statements in the microscope!

6. Do not try to illustrate all the structures seen in the round field of vision! Make a schematic drawing only of a part of the section which displays the most informative morphological details!!

7. **Besides the recognition of histological structures, students are expected to answer theoretical questions related to structures in question (gross anatomy, embryology, ultrastructure, biochemistry).**

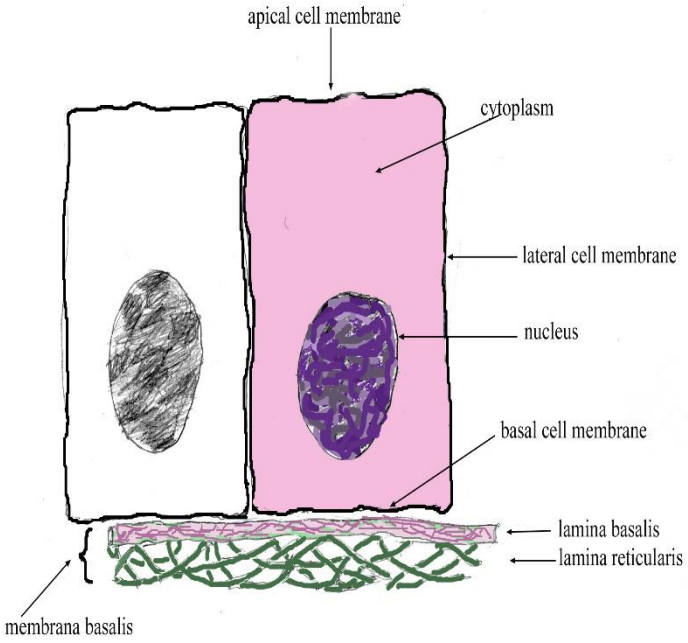
Your feedback on the Histology Manual is very important. Any of your suggestions to improve the effectiveness of it in learning Histology would be greatly appreciated! Please email me at:

nemeskeri.agnes@med.semmelweis-univ.hu

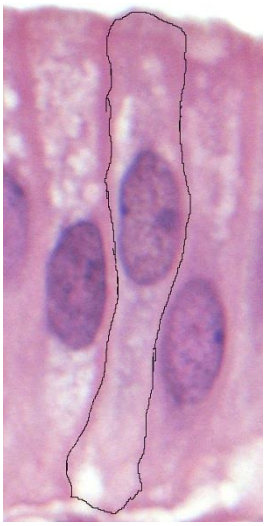
The cell

The cell is the morphological and functional unit of the body.

Schematic illustration of the cells

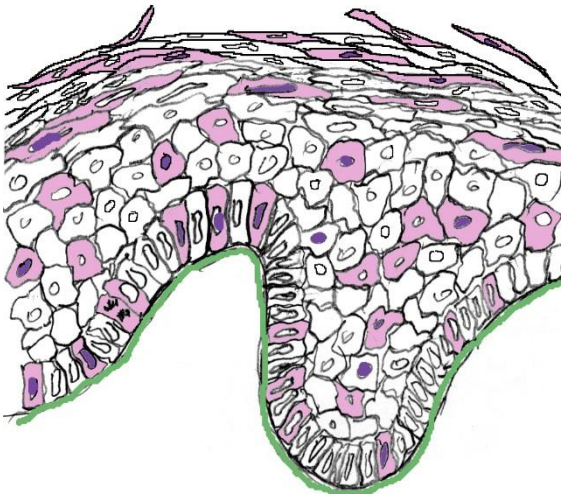


Cells in the microscope



Tissues

Tissue is an organized assembly of cells working together and subserving a particular function.



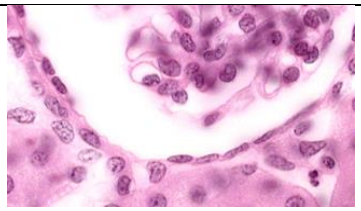
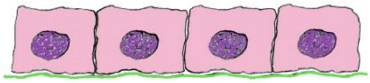
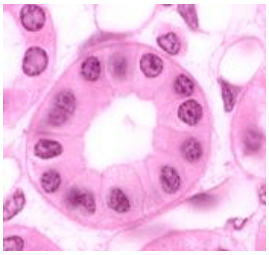
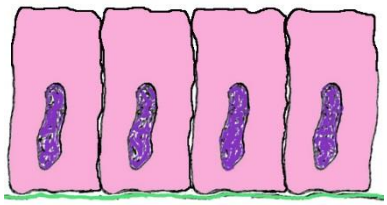
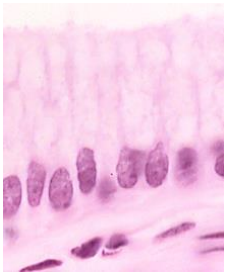
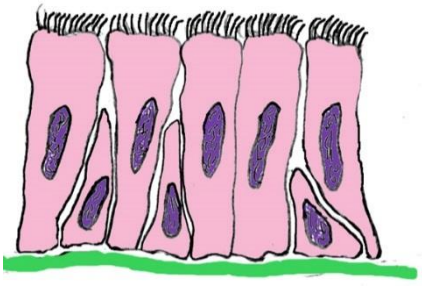
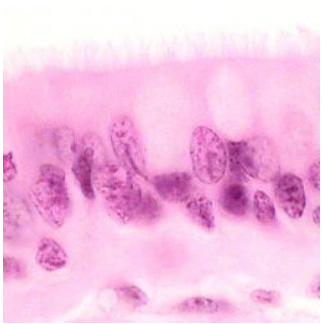
Basic tissues of the body: I. Epithelium

1) Covering epithelia
   a. Simple epithelia
   b. Stratified epithelia
2) Glandular epithelia

II. Connective tissue
III. Muscle tissue
IV. Nervous tissue

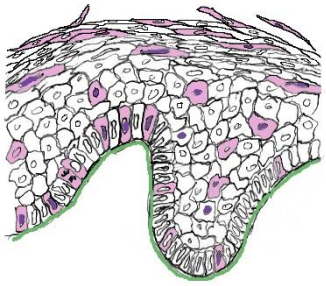
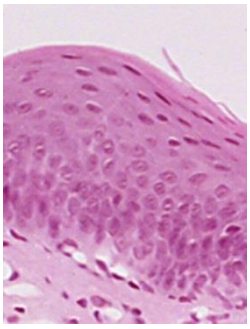
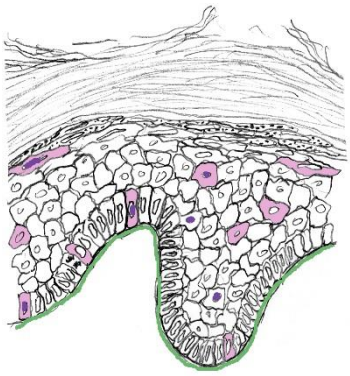
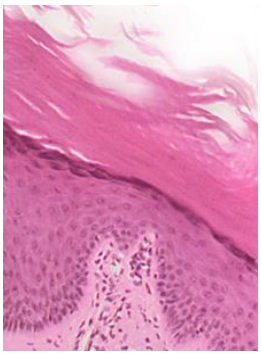
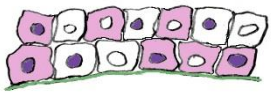
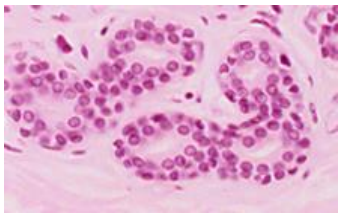
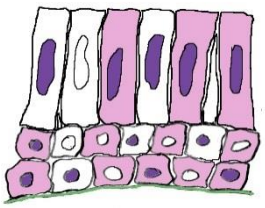
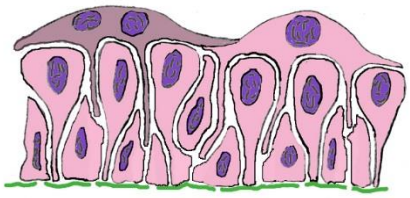
Simple Epithelial Tissues

-the cells are arranged in one layer
- all the cells are sitting on the basal membrane-

Classification	Schematic illustration	Under the microscope
Simple squamous epithelium	 flattened cells – flattened nuclei	
Simple cuboidal epithelium	 cuboidal cells have cubic shape - the height is approximately of the same size as the width – nuclei are round	
Simple columnar epithelium	 the height of the columnar cells is greater than the width nuclei are vertically elongated	
Pseudostratified epithelium ciliated	 all the cells touch the basal membrane – some of them do not reach the lumen – apical membrane specialization (moving cilia - kinocilia)	 **Nemeskéri**

Stratified Epithelial Tissues

- cells are arranged in two or more cell layers -
- shape of cells varies from layer to layer –

Classification	Schematic illustration	Under the microscope
Stratified squamous epithelium nonkeratinized	squamous cells form the superficial layer 	
Stratified squamous epithelium keratinized		
Stratified cuboidal epithelium	cuboidal cells form the superficial layer 	
Stratified columnar epithelium	columnar cells form the superficial layer 	
Uroepithelium or urothelium	special umbrella shape of cells facing the lumen 	

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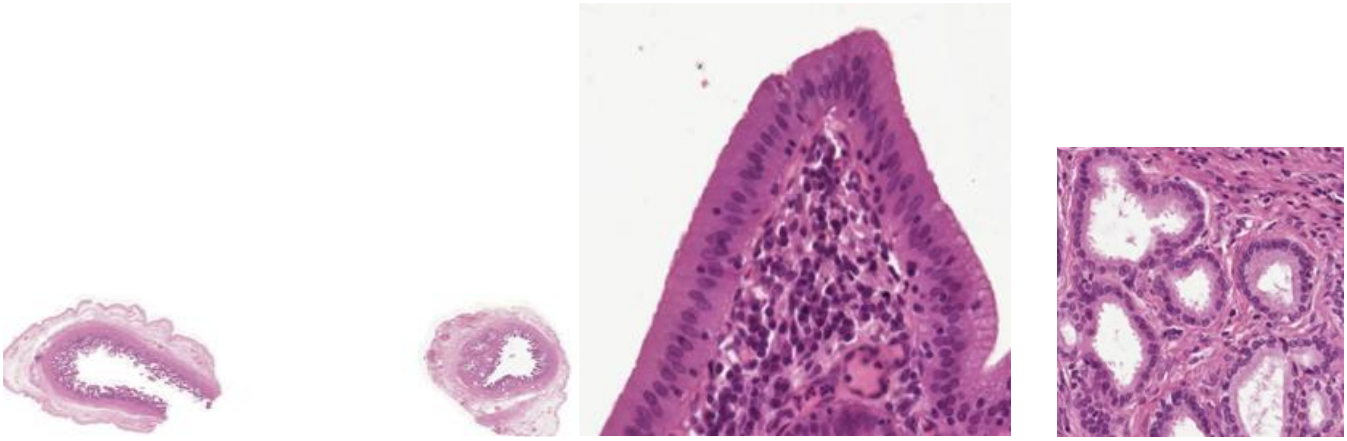
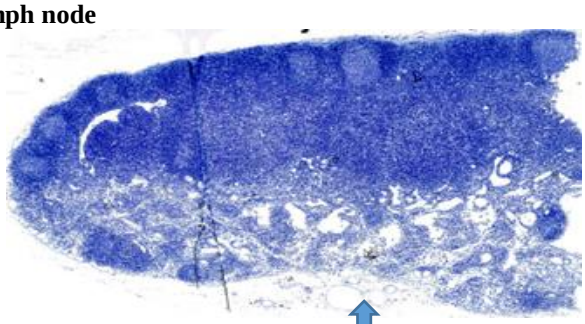
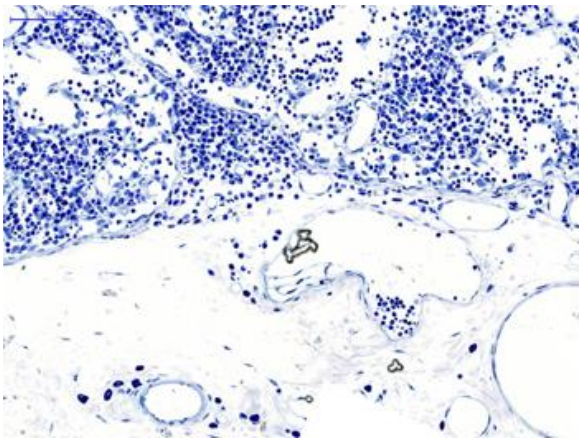
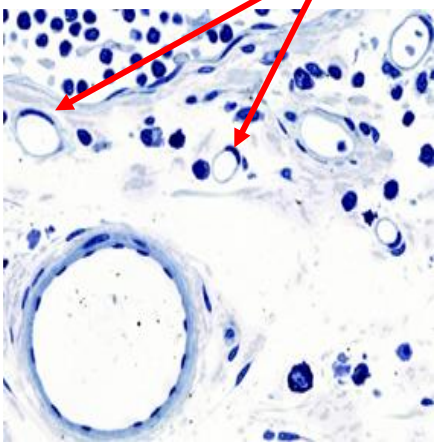
3. Gall bladder – fundus - neck



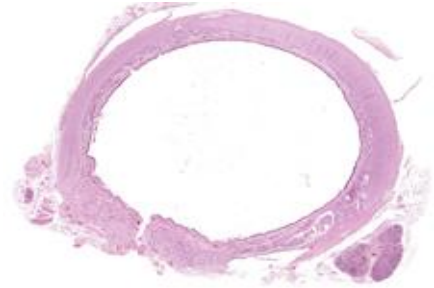
40. Lymph node

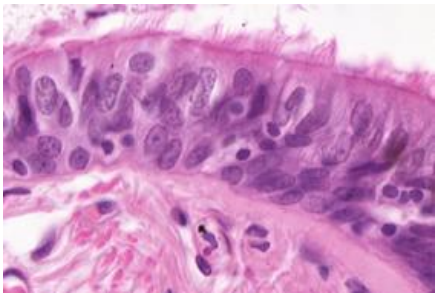






57. Trachea





Epithelial tissue
Unilaminar (simple) covering epithelia

Slides (N°):

3. Gall bladder – fundus + neck (2 sections) – human - HE staining
40. Lymph node – rat – toluidine blue staining
57. Trachea – human – HE staining

N° 3. Simple columnar epithelium + Simple cuboidal epithelium – gall bladder – fundus + neck (2 sections) – human - HE staining

This section was prepared from the entire wall of the gall bladder. On the luminal side, irregular folds of the mucosa are found. A **very regular simple columnar epithelium** lines these folds. At high magnification, the simple columnar epithelium is seen to consist of tall cells; that is they are taller than their diameter. The **vertically elongated nuclei** are usually located towards the base of the cells, their cytoplasm is stained eosinophilic. When the plane of section is not parallel with the axis of cells but oblique or transverse, the nuclei show round shape. The underlying supporting basement membrane is not visible.

On either of sections find the epithelial surface, at low power magnification. At high power, observe the columnar epithelium. Try to imagine the cylindrical 3–D shape of the columnar cells and when they are sectioned horizontally the cell profiles show polygonal shapes and the nuclei are not elongated but round!

The section of the neck is a complete ring. At the bases of the mucosal folds, ovoid structures can be seen. Some of these structures are lined by a **single layer of cuboidal cells** (secretory acini of mucus producing glands and excretory ducts, opening into the space between the folds). These cells are approximately square in vertical section. The shape of cell nuclei is rounded or flattened.

In the section of the neck look for structures displaying rounded or ovoid empty areas (alveoli, acini), which are lined by cuboidal or columnar cells. Compare the two cell types.

40. Endothelium, simple squamous epithelial cells – lymph node – endothelial cells at the hilum - rat – toluidine blue staining – semi-thin section

A **single layer of squamous cells called endothelium** lines the interior of blood and lymphatic vessels. The cytoplasm of these cells is so flattened that it is usually not visible with light microscope. Endothelial cells can be recognized by their dark stained **flattened nuclei** which bulge into the lumen. The basement membrane supporting the endothelial cells is also thin; hence, it cannot be distinguished from the thin and similarly stained cytoplasm.

The blood and lymphatic vessels are round or ovoid mainly empty structures, Dark, flattened nuclei of endothelial cells can be recognized lining the internal surface of the very thin wall.

57. Pseudostratified ciliated columnar epithelium – trachea – human - HE staining

The trachea is lined by **pseudostratified columnar epithelium**. This epithelium seems to be multilayered since the nuclei lie at different levels when it is vertically sectioned. In fact, it is a simple columnar epithelium, all cells contact the basal membrane, but not all extend to the surface. Some cells are shorter, they form a basal layer, and this gives a pseudostratified appearance. Among them, dividing cells are often seen; these basal cells replace the dead epithelial cells. The apical surface of columnar cells bears **kinocilia**. This epithelium is found in the respiratory tract. Among the columnar epithelial cells unicellular glands, the goblet cells can be observed. An **unusually thick basement membrane** can be observed.

The trachea is a tubular organ. The slide usually is a portion of the cross section, but there might be a whole circular section also. The lining epithelium can be found on the inner, concave surface of the section. Observe the lining epithelium at high power, and make a drawing.

Stratified covering epithelia

Slides N°:

8. Urinary bladder – monkey - HE
5. Esophagus – upper third + middle third (2 sections) – human – HE
6. Sole (skin) – human - HE
7. Penis – human – Verhoeff's elastic stain

.

N° 8. Urothelium (transitional epithelium) – urinary bladder – monkey - HE

The lumen of urinary bladder is lined by **stratified epithelium** composed of cells whose nuclei are seen in 3–6 layers. The morphology of surface cells and numbers of layers changes, according to the degree of distension or contraction of the urinary bladder. In stretching, the superficial cells become flattened and the number of layers decreases, while in non-distended state the superficial cells are large dome shaped. The cells of the superficial layer are called: **"umbrella cells"**, they are often binucleated. The cells beneath the surface cells are smaller and have pear shape (they are often referred to as **"pear shaped cells"**). The basal layer is composed of cuboidal or columnar cells and these cells are the smallest.

The section is from a portion of the wall of the urinary bladder. In the tissue of the bladder the inlet of ureter is also present. Make a drawing about the layers of the epithelium, and try to define whether the organ was in a stretched or in a non-distended state. Compare this epithelium with the stratified squamous non-keratinized epithelium; they are frequently confused.

Stratified epithelia are classified according to the shape of cells lying on the surface of a given stratified epithelium (Except the urothelium).

N° 5. Stratified squamous non-keratinized epithelium – esophagus, lining epithelium – upper third + middle third (2 sections) – human – HE

This organ is a thick walled muscular tube and the contraction of its muscular component results in the stellate shape of its lumen. The epithelium lining of the lumen is an unusually broad **non-keratinized stratified squamous epithelium**.
Three layers can be readily distinguished in the non-keratinized stratified epithelium.

I. **Stratum basale**: it is composed of a layer of columnar or cuboidal cells supported by a basal membrane. These cells assure the continuous renewal of desquamated surface cells.
II. **Stratum spinosum**: it contains many layers of large polygonal cells.
III. **Stratum planocellulare**: the most superficial layers of flattened cells, that are nucleated, but they show the signs of progressive degeneration. The nuclei are pyknotic (the dark stained nuclei show the signs of shrinkage).

Make a drawing about the whole slide at low power magnification, and about the epithelium at high power. On either of sections look for an intact part of the epithelium!

N° 6. Stratified squamous keratinized epithelium – thick skin, skin of the sole – human - HE

This section has been prepared from the **thick skin** of the sole. The external layer of skin is composed of a **stratified squamous keratinized epithelium** known as epidermis.
Five histological layers of the epidermis can be identified:

I. **Stratum basale or germinativum**: the basally situated cells are cuboidal or columnar in shape. Mitotic figures are frequently seen in this layer. The cells of germinal layer are anchored to the basement membrane (hardly seen) that separates the epithelium from the underlying connective tissue layer.
II. **Stratum spinosum or polygonale**: the newly formed cells gradually move towards the surface, meanwhile their shape changes to polyhedral. The stratum spinosum is several cells thick. A **spiny appearance of cell surfaces** can be observed which is due to **desmosomes** and to the effect of fixatives. At the desmosomal contacts, the adjacent cells are tightly bound to each other, but between these strong contact points, the cytoplasm and cell membranes are shrunk in fixatives.
III. **Stratum granulosum or granular layer**: As the cells of the stratum spinosum pass more superficially, they begin to flatten, and their staining becomes strongly basophilic. At higher magnification, dense **basophilic granules** can be observed in the cytoplasm of the flattened cells. These coarse, irregular granules contain basophilic proteins called **keratohyalin**. Stratum granulosum consists of 1–5 layers of granular cells. The cells dye as the mature protein, the keratin gradually fills them.
IV. **Stratum lucidum**: above the nucleated cells of the stratum granulosum, there is an abrupt change: the highly **flattened cells lose their nuclei** and form a compressed homogeneous eosinophilic layer. This layer can be found only in very thick skin.
V. **Stratum corneum**: this layer is extremely thick in thick skin; this gives the main difference between the thick and thin skin. Towards the surface, the desmosomes and organelles of the dead flat cells completely disintegrate that result in **desquamation of keratinized cells**.

Make a sketch about the whole section, and afterwards draw the epithelium. Pay attention to the ratios between the thicknesses of the different layers.

N° 6. Stratified cuboidal epithelium – thick skin, skin of the sole, excretory duct of the merocrine sweat gland – human - H&E

Merocrine sweat glands are coiled tubular glands composed of a secretory part situated deep in the skin, and a duct that leads to the skin surface. The epithelium lining the excretory ducts is composed of **stratified cuboidal epithelium**. Two layers of small cuboidal cells can be observed around the duct lumen. Excretory duct leaving the compact coil of the gland shows a light spiral course through the dermis (connective tissue layer under the epidermis). Look for this segment of excretory duct or the rounded ovoid cross sections of the duct inside the compact coil of the gland where the darker staining of the duct cells can help to identify them.

N° 7. Stratified columnar epithelium – penis, lining epithelium of the urethra – human – Verhoeff's elastic stain

On the transverse section of the penis, sections of three cylindrical structures are seen. These are erectile tissues of the paired corpora cavernosa penis and in the midline the corpus spongiosum penis. The latter one surrounds the penile urethra, which is lined by **stratified columnar epithelium**. Look for the lumen of the penile urethra with naked eye and then focus on the lining epithelium. The epithelium is wavy, and usually consists of two–four layers. **The cells in the superficial layer have columnar shape**. The basal layer composed of columnar or high cuboidal cells.

Glandular epithelia

Slides (N°):

99. Ileum – human – HE
52. Submandibular gland – human – HE
39. Eye lid – human - HE
11. Hairy skin – human - HE

N° 99. Unicellular gland – goblet cell – ileum – human – HE

The luminal surface of the small intestines is lined by simple columnar epithelium. Mucus secreting modified epithelial cells called **goblet cells** are scattered amongst the columnar enterocytes. Distended apical part of these unicellular glands contains **mucoid granules**, while the condensed, often "Y" –shaped nucleus occupies the narrowing stem. The routine H&E staining method stains poorly the mucigen granules, therefore the widened apical cytoplasm seems to be empty.

N° 52. Mucous and serous glandular cells – submandibular gland – branched tubuloalveolar gland – human - HE

Exocrine glands have a secretory portion, which contains the cells producing and releasing the secretory product; and the ducts, which transport the product to a surface. The **serous cells** are protein–secreting cells. The main bulk of the gland is composed of **serous acini** with minute central lumina. The secretory cells are triangular in shape; their rounded nuclei are basally located in the **darkly stained basal cytoplasm**. The intense **basophilia** of the basal cytoplasm results from local accumulation of **rough endoplasmic reticulum**. The apices of the cells are acidophilic stained due to the **secretory granules (zymogen granules)** in the luminal part of the cells.

Mucous secretory cells are characterized by the presence of glycoprotein containing granules known as **mucigen granules**. Mucous acini display very **light staining** with H&E. The dark stained nuclei are usually flattened (not always!) and lie against the basement membrane. Several mucous secretory units exhibit a cup of serous cells called **serous demilune (demilune of Gianuzzi).** These mixed sero–mucous secretory units show above mentioned characteristic staining differences. The lumen of mucous acini is wider than that of the serous acini.

Duct system

The initial segment of excretory duct, the intercalated duct (low cuboidal epithelium) drains into larger striated ducts (high cuboidal or columnar epithelium). Striations are seen in the basal part of cuboidal or columnar duct cells, which result from the deep infoldings of the basal plasma membrane containing several mitochondria. Between the lobuli of the gland large interlobular duct can be observed, lined by stratified cuboidal or stratified columnar epithelium**.**

According to the mode of secretion three groups of glands are classified:

a) merocrine type of secretion – secretory granules are released without loss of cytoplasm and cell membrane – all of the above described glands are of merocrine type

b) apocrine type of secretion - detachment of apical cytoplasm (containing secretory granules) with its cell membrane – apocrine sweat gland

c) holocrine type of secretion - degeneration, disintegration of the whole epithelial cells – sebaceous gland

N° 39. Apocrine gland – modified apocrine sweat gland (Moll) - coiled tubular gland – eyelid – HE

The **wide secretory portions** are lined by **eosinophilic stained secretory cells** that are low **cuboidal to columnar** in shape depending on the phase of secretory process. The heights of cells can be different in the same cut profile of the tubule. The morphology of the glandular cells can be studied at higher magnification. Numerous granules fill the apical cytoplasm of the eosinophilic stained epithelial cells and **bleb–like protrusions** can be observed on their luminal cell surface**. Detached cytoplasmic protrusions** are seen in the lumen that means the loss of a certain amount of apical cytoplasm and cell membrane. This **apocrine** type of secretion explains the variable cell size in the same transection. Recent studies revealed that the secretory process of apocrine glands includes a) **merocrine** secretion of granules, b) detachment of apical cytoplasm with its cell membrane (**apocrine** secretion), and c) degeneration, disintegration of the whole epithelial cells (**holocrine** secretion).

Merocrine sweat glands are coiled tubular glands composed of a secretory part situated in the deep part of the dermis, and a duct that leads to the skin surface. In the secretory segment is pale stained, the cells are pyramidal shaped. The epithelium lining the excretory ducts is composed of **stratified cuboidal epithelium**.

At low power magnification look for wide lumina near to lid margin, thereafter study the morphology of the glandular cells with high power magnification.

N° 11. Holocrine gland – sebaceous gland – branched acinar gland – hairy skin – HE

Sebaceous glands are widely distributed in the dermis of the skin. The hair follicles are always associated with sebaceous glands; however, in some locations sebaceous glands appear without hair follicle (Meibomian gland in the eyelid, the prepuce, in the lip at the corner of the mouth, the nipple and areola). These glands are of **simple branched acinar** type; their acini are opening into a short duct which enters the pilosebaceous canal. At the periphery of acini undifferentiated small basal cells are dividing and move away from the basal lamina. Their cytoplasm enlarges, and lipid droplets start to fill it (lipid solves out, therefore staining becomes pale), meanwhile the nuclei gradually become pyknotic (apoptosis). In the center of sebaceous acini, the large rounded cells degenerate and disintegrate. The mixture of oily secretory product and cell debris of necrotic cells form the final composition of the sebum. During this process the whole cell is transformed into secretory material, hence this process can be referred to as **holocrine secretion**.