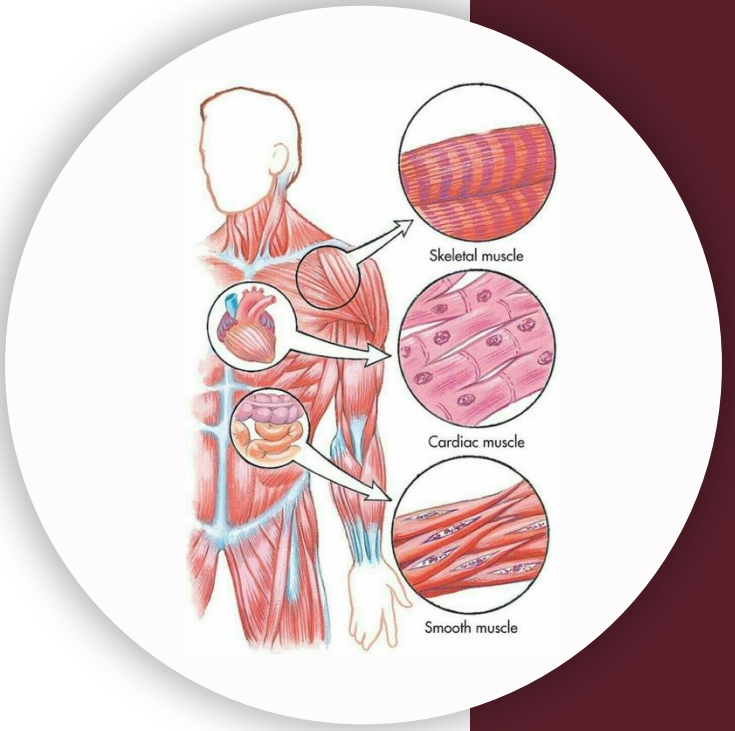
بسم الله الرحمن الرحيم

Histology - Lecture 7

Epithelium Images



Written by : Tala Alali
Heba Salah

Reviewed by : Rasha AlHamra

There are two main types of microscopes: **light microscopes** and **electron microscopes.**

1. Light Microscopes:

- The most common type is the bright-field microscope.
- It works by passing light through the specimen, creating an image based on light absorption.
- A camera captures the image, which is then displayed and stored on a computer.
- The image appears similar to what the human eye sees but with magnification and better detail.

2. Electron Microscopes:

- These include scanning (SEM) transmission (TEM) electron microscopes .

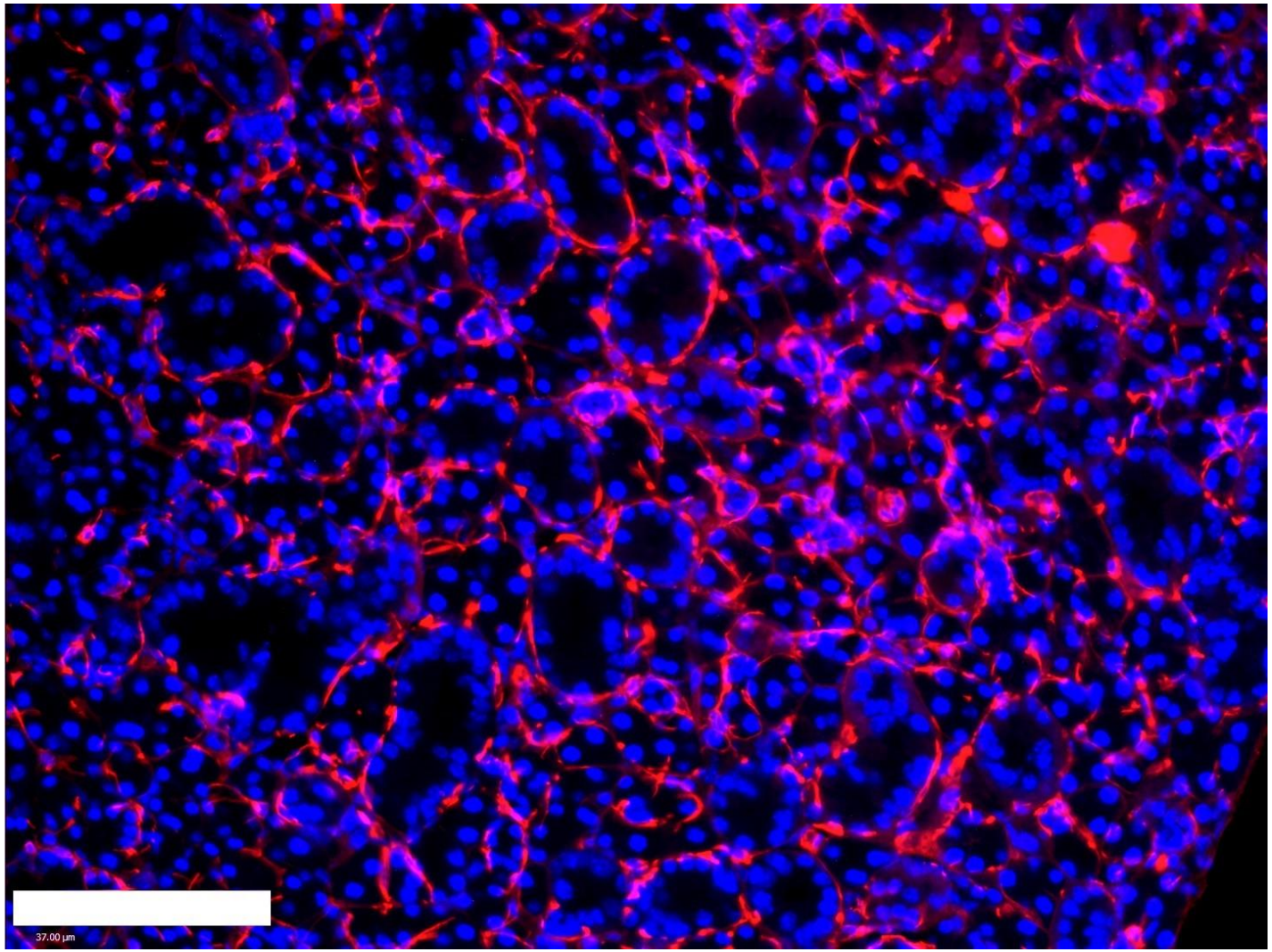
If we are targeting a specific molecule, such as the protein troponin in muscle cells, we use it to determine the amount of muscle cells in certain planted cells or to identify cells that are troponin-positive in the section we are working on.

When using immunostaining, we do not use hematoxylin and eosin stain, as they provide general features, such as epithelial tissue, muscle tissue—including its three types—and connective tissue.

The concept of immunostaining involves proteins and antibodies, allowing us to detect only the positive cells for a certain protein, rather than any other cells. Then, we simply count the positive cells.

There are actually two systems:
1. Immunofluorescence, which we recognize by its bright colors, like lamps—such as a red lamp or a blue lamp—while the rest appears black.
2. Immunohistochemistry, which is also part of immunostaining, meaning it involves antibodies and antigens.

Immunoflouresence Staining/Tissue

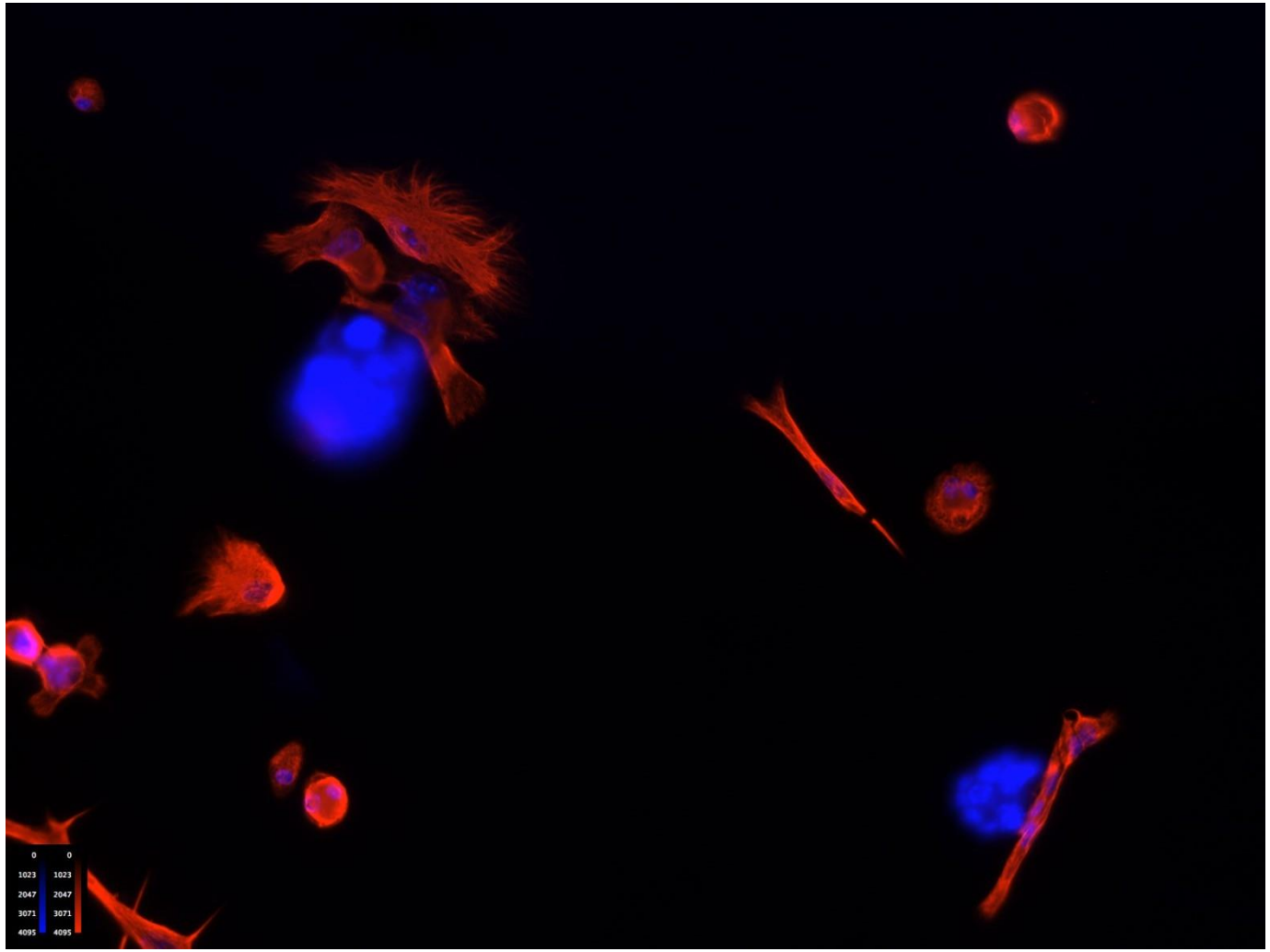


we can see how the cells behave when they are within a tissue. This is a tissue that was extracted from a patient or an animal, then processed, stained, and observed. These are cells observed within a natural tissue environment.

The red color marks the cells where the protein is found (positive staining), while the blue color only marks the nuclei of all cells (negative staining), regardless of whether they contain the target protein or not.

Note : (positive staining) and (negative staining) they describe whether a targeted protein is present or absent in the sample.

Immunoflouresence Staining/cultured cells



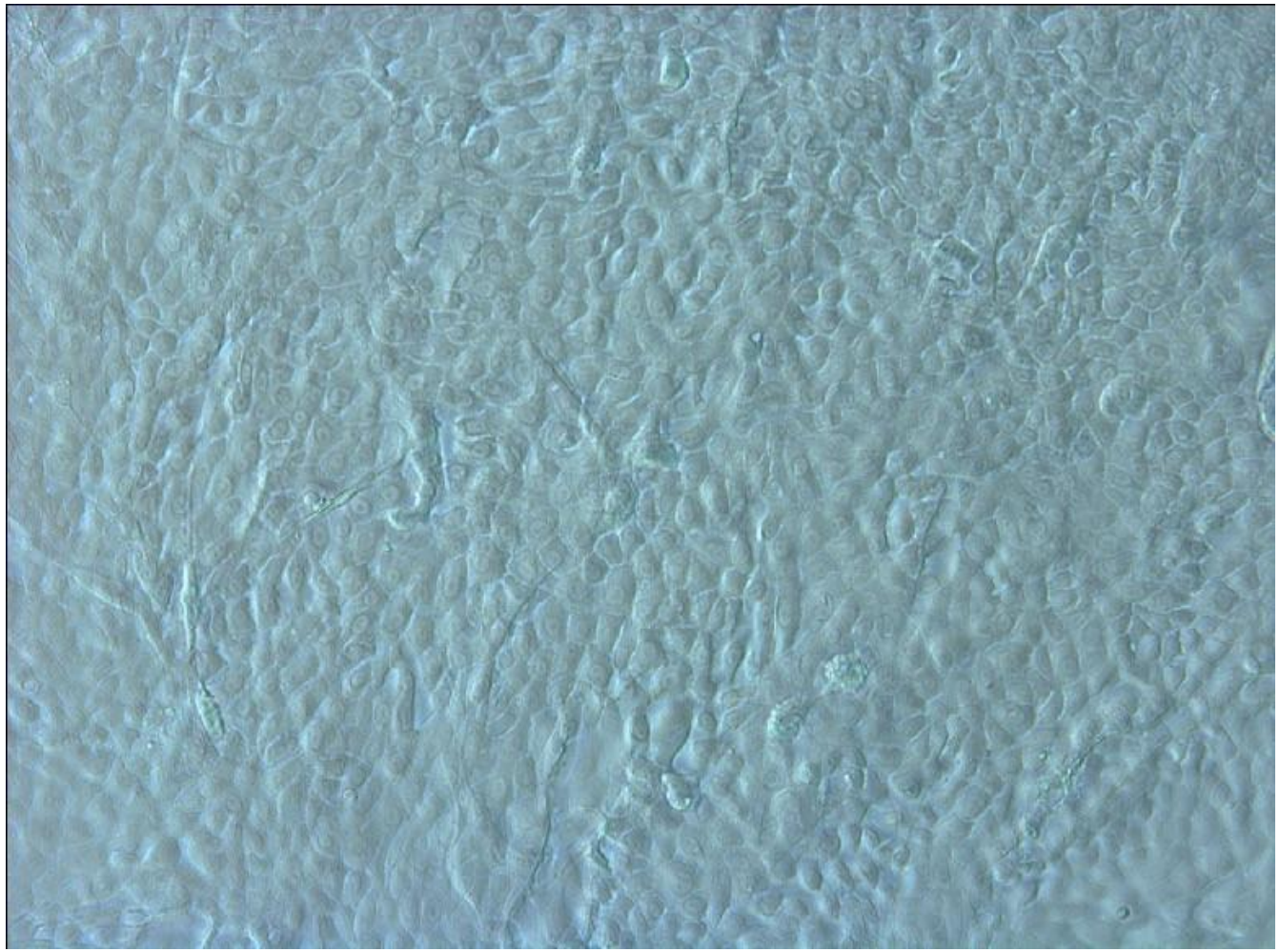
In this picture we see how the cells behave when they are alone.

In this type of microscopy, **a specific wavelength of light is used to illuminate the sample** after it has been prepared and immunostained by adding antibodies. Once the sample is covered, it is illuminated with a specific wavelength.

This molecule can be excited at a certain wavelength. **Different fluorophores have excitation at different wavelengths.** When the glass slide is illuminated with a specific wavelength, the fluorophore absorbs part of the light energy and emits another longer wavelength.

The red color marks the cells where the protein is found (positive staining), while the blue color only marks the nuclei of all cells (negative staining), regardless of whether they contain the target protein or not.

Phase-contrast Microscope



The planted cells appear here as scattered circles everywhere.

If we study living cells(planted cells) in the lab and need to track their growth or characteristics, we use phase contrast microscopy.

Why Phase Contrast?

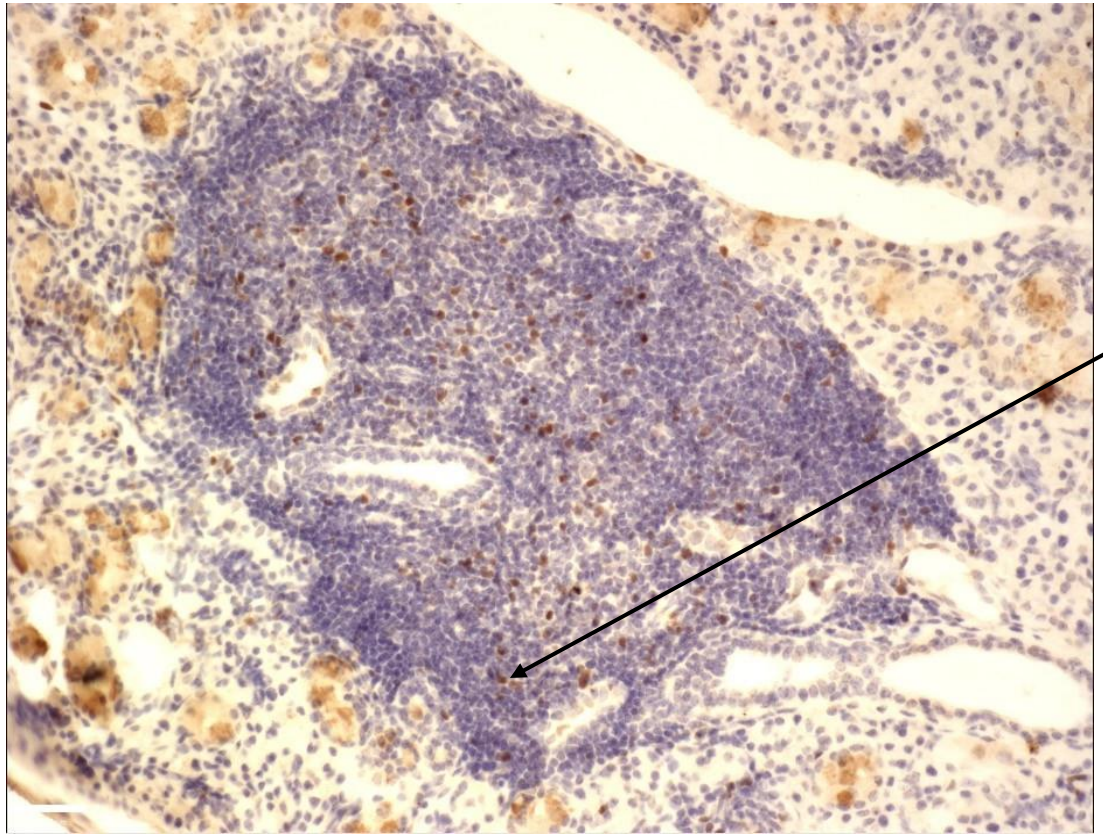
- **It modifies the optics to enhance contrast in unstained, living cells.**
- **It works by detecting differences in the refractive index between the cell and its surroundings.**
- **This allows us to see cells without staining, which is important because staining kills cells.**

Why Not Bright-Field?

- **Bright-field microscopy requires staining, as unstained cells are nearly invisible.**
- **Since staining kills cells, phase contrast is better for live-cell imaging.**

Immunohistochemical Staining/Tissue

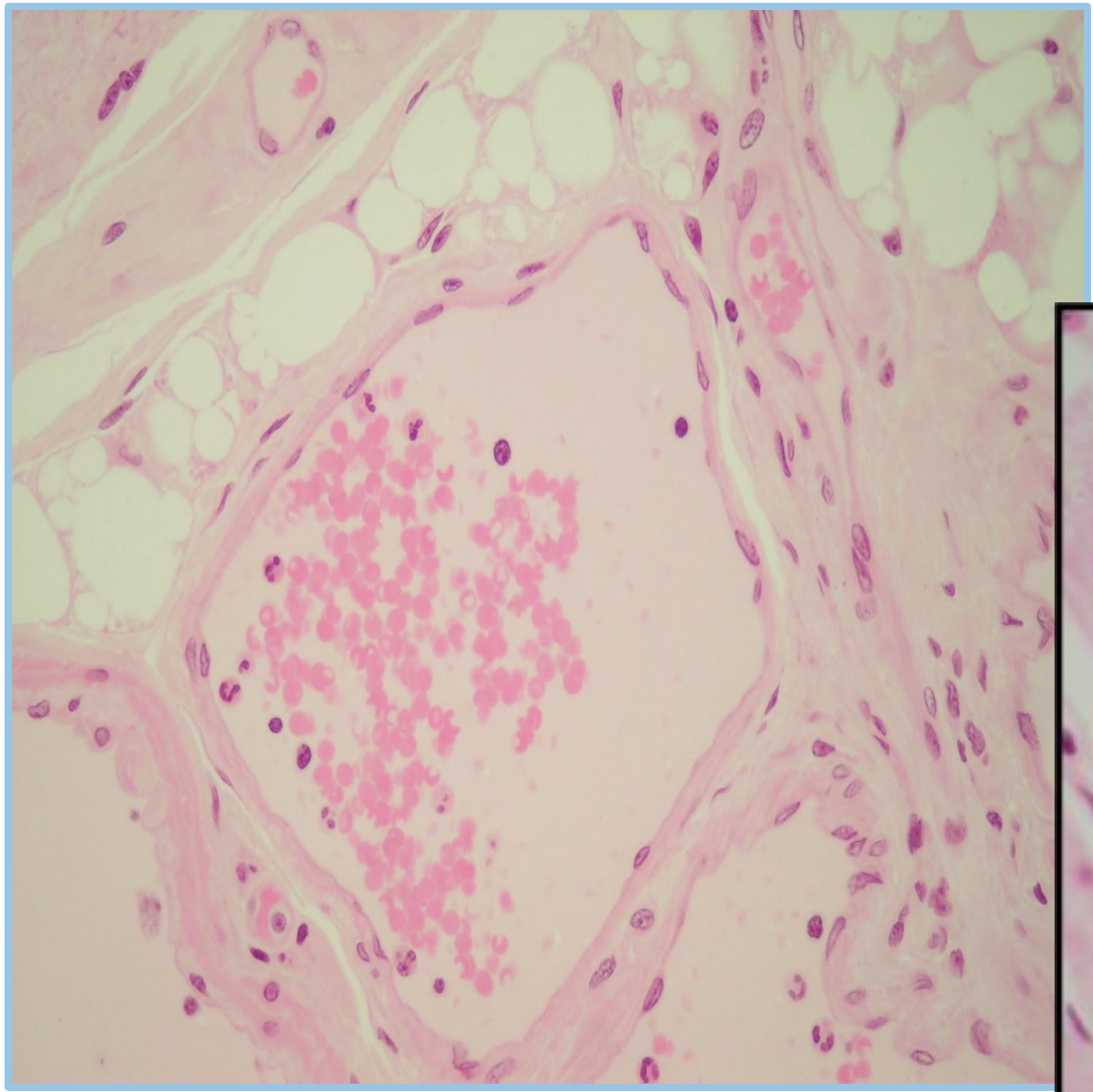
We use this technique if we only have a bright field microscope. However, if we have a more sophisticated light microscope, such as what is called immunofluorescence microscopy, we can use a different approach.

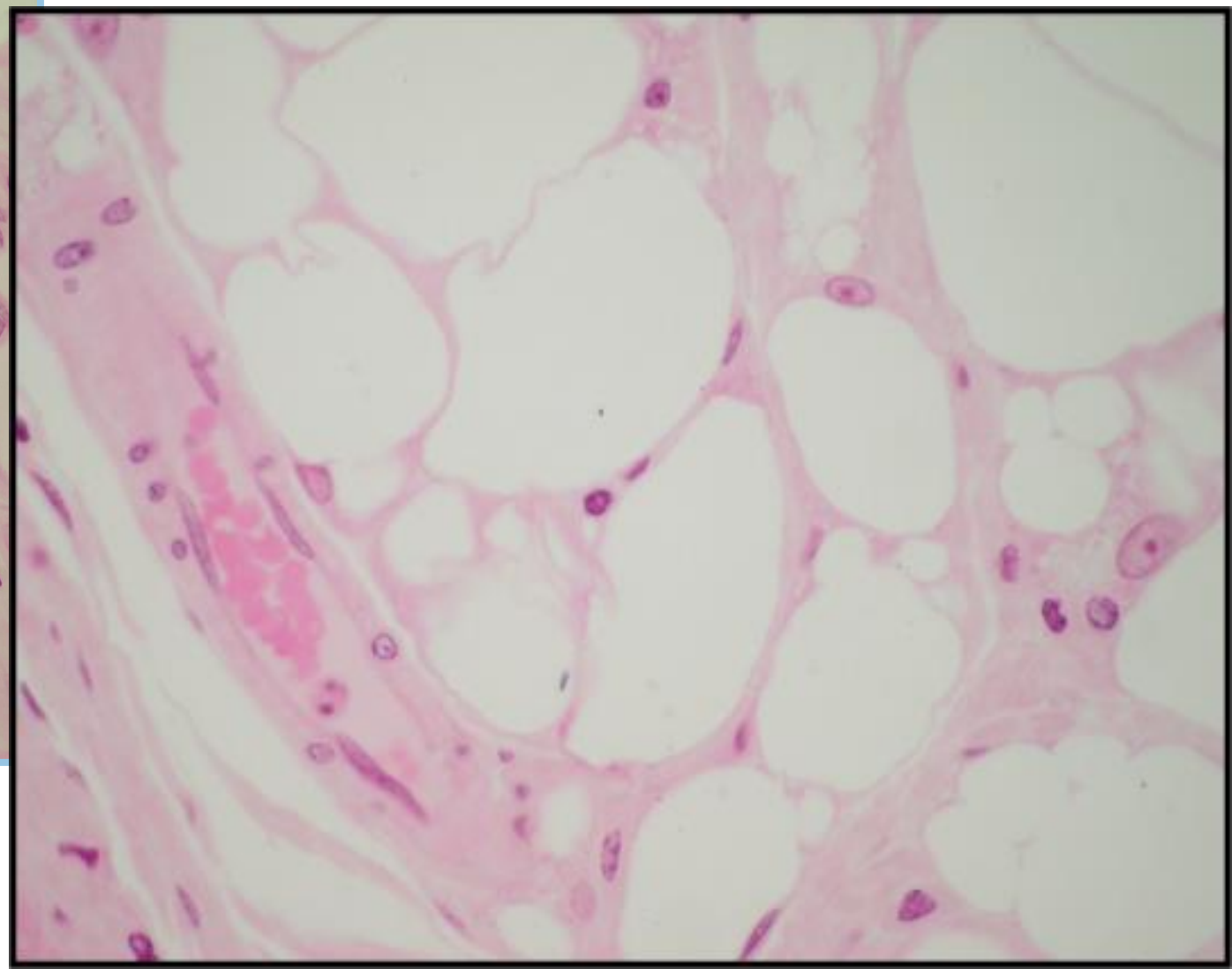


We acquired this image using bright field. During staining, there is a binding between antibodies and antigens. Capable antibodies are used when an antibody is loaded with an enzyme. Then, we add a substrate for a specific enzyme. These substrates are usually oxidatives, which leads to a reaction that produces color.
In this picture, we used only hematoxylin without eosin. As you can see, there is no pinkish color. This is a nuclear staining technique used to highlight distinctive cells. Also, you can observe that many cells are clustered together. They appear this way because they are small cells with compact nuclei.

As you see, some cells are brown in color. These are the only positive cells for the marker I am looking for. This is the concept of immunostaining, as I search for these cells to count them. These cells already have the antigen. Then, I put an antibody that is loaded with an enzyme. After that, I add a substrate. The reaction gives color.
This is how I get a brown color. But the bluish cells don't have antigens. The antibody doesn't bind to them, so the enzyme doesn't work, even if I add a substrate.

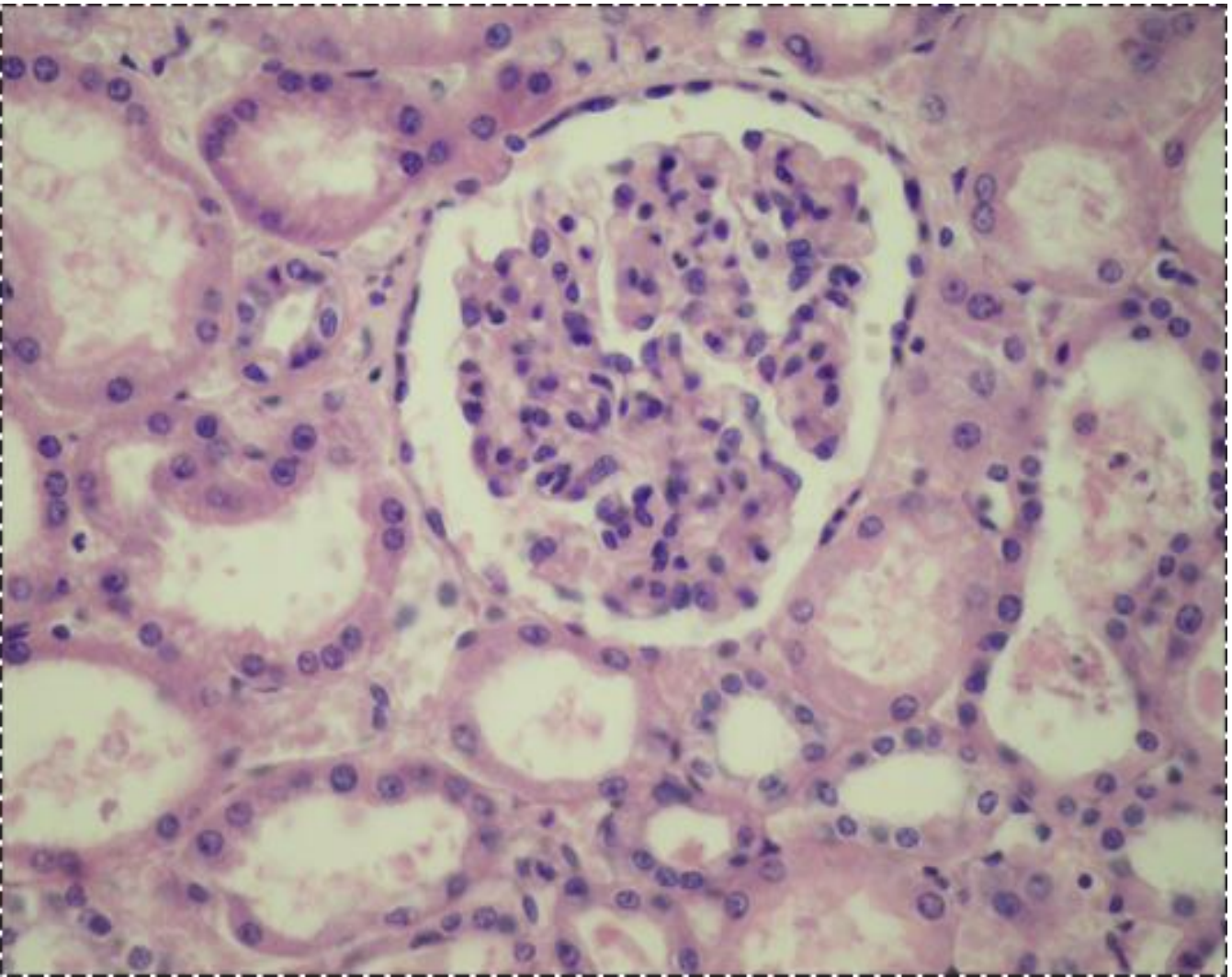
ENDOTHELIUM

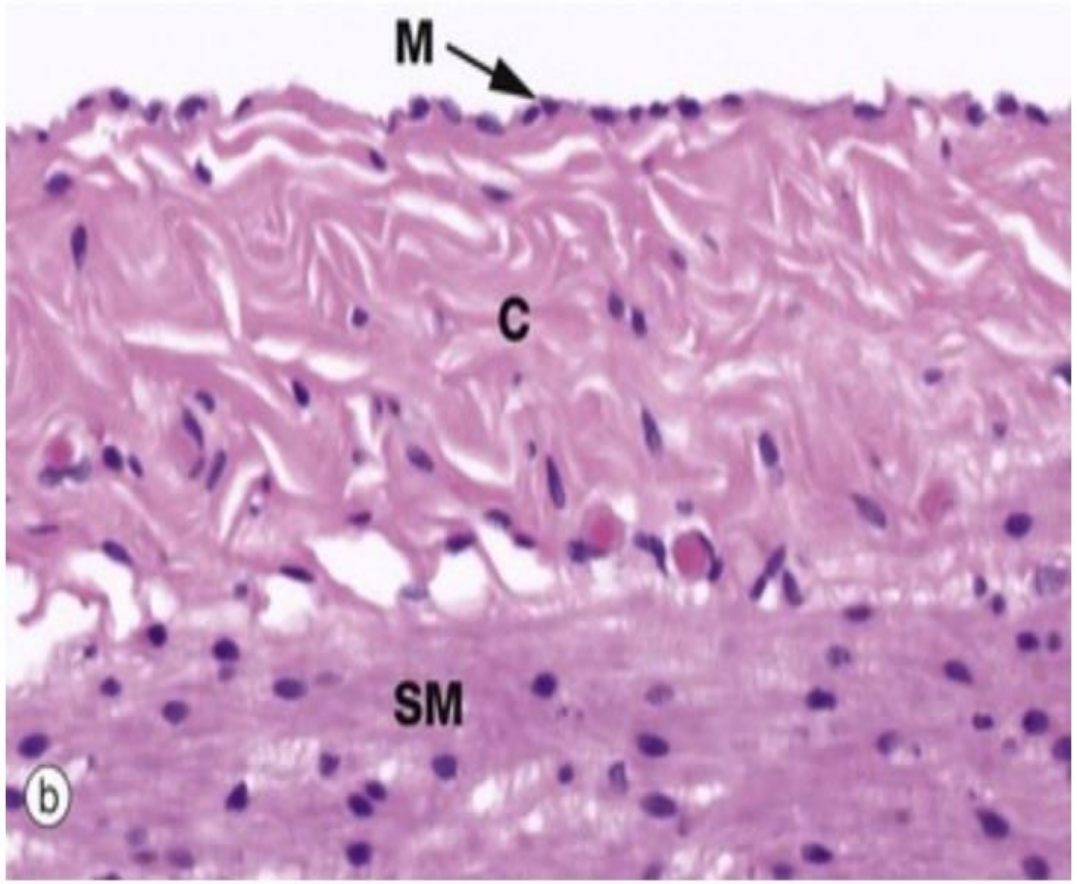




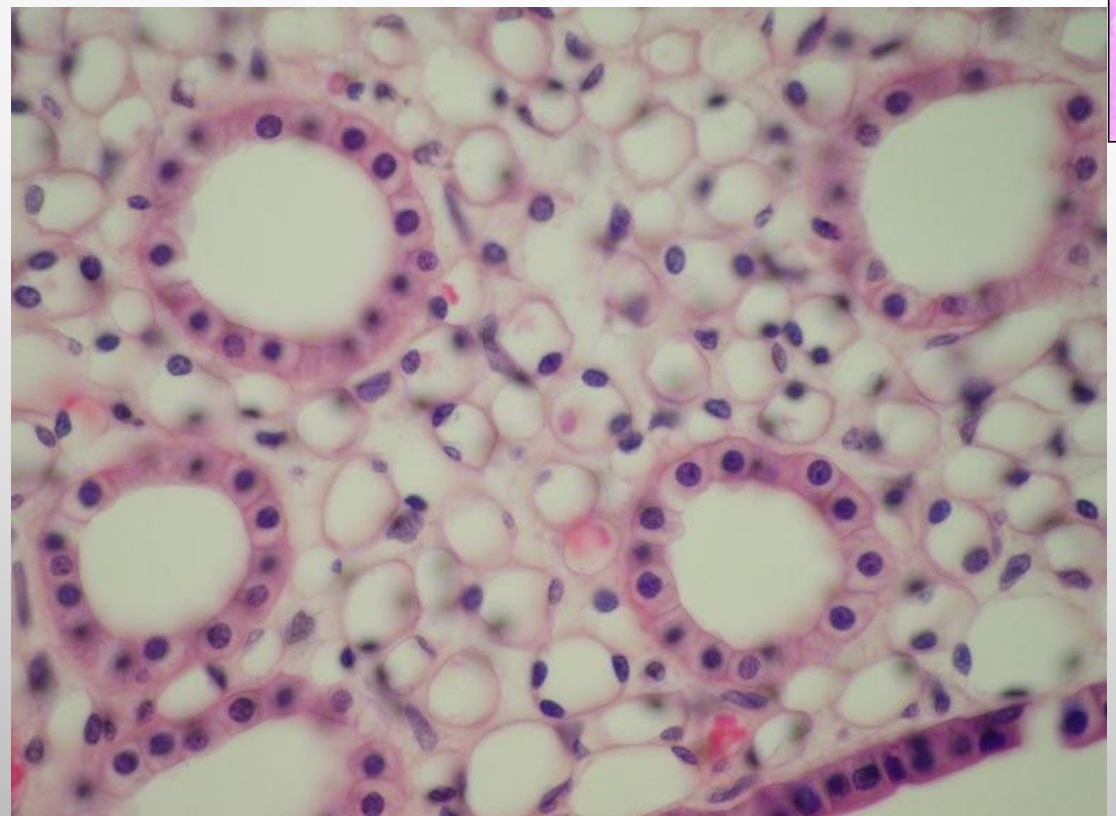
Slides 8-13 were explained in Lecture 6

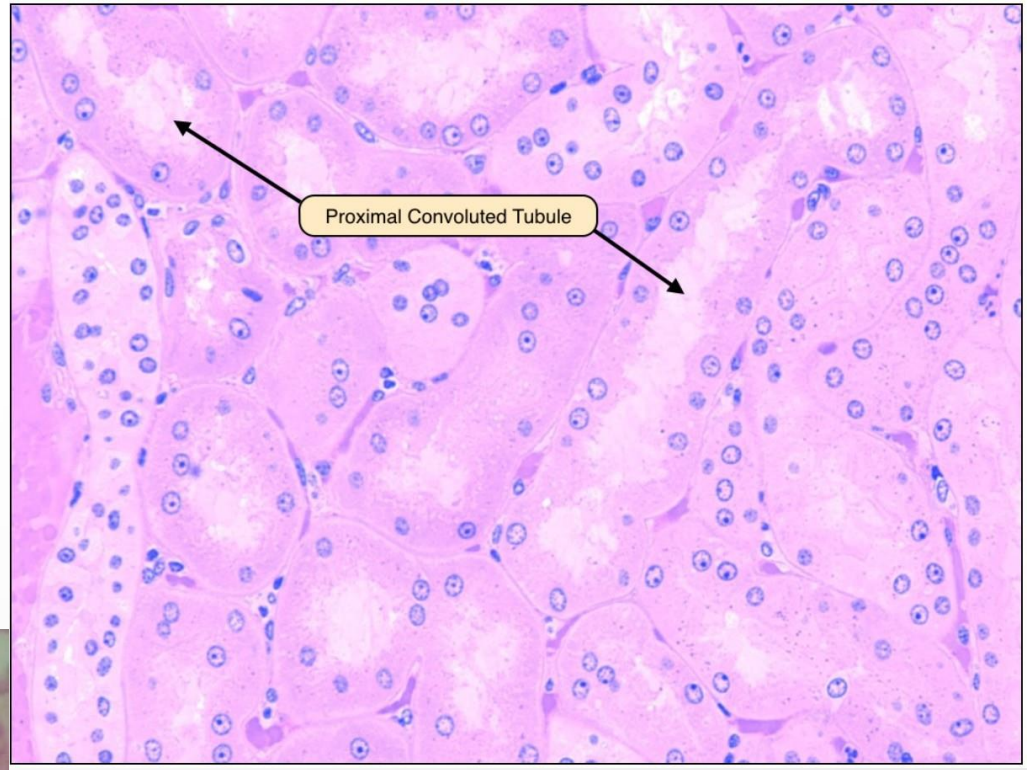
SIMPLE SQUAMOUS EPITHELIUM



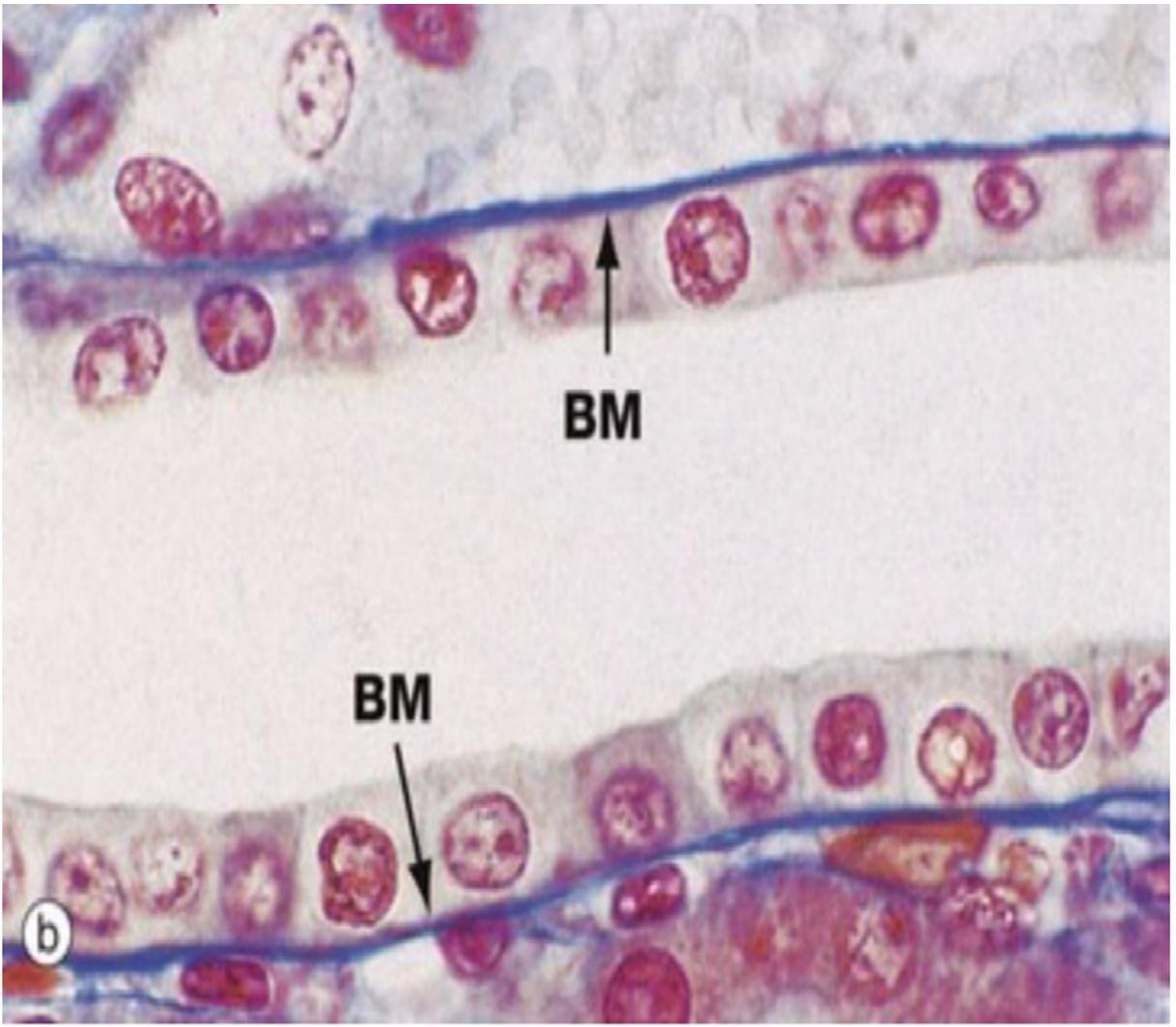


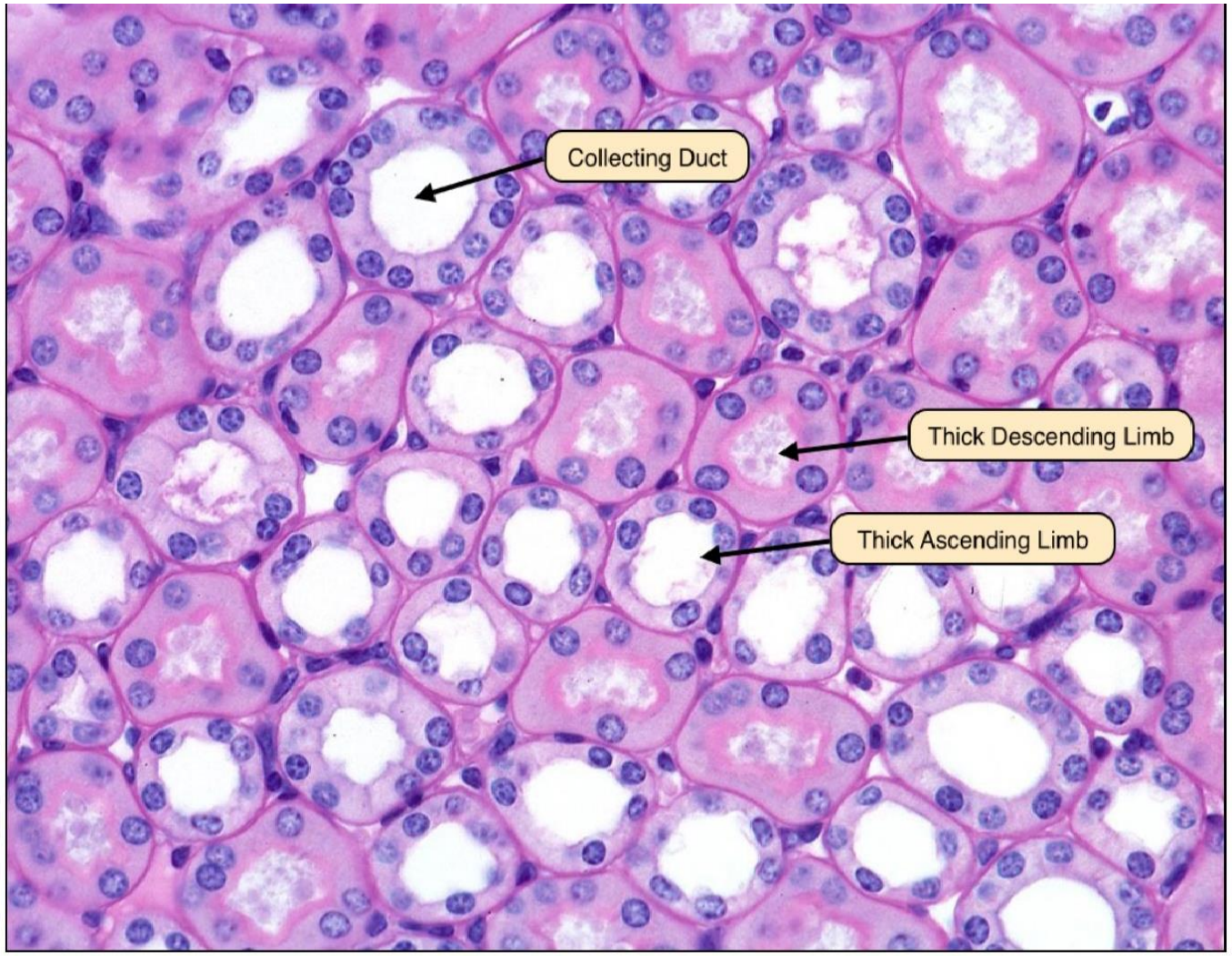
SIMPLE CUBOIDAL EPITHELIUM



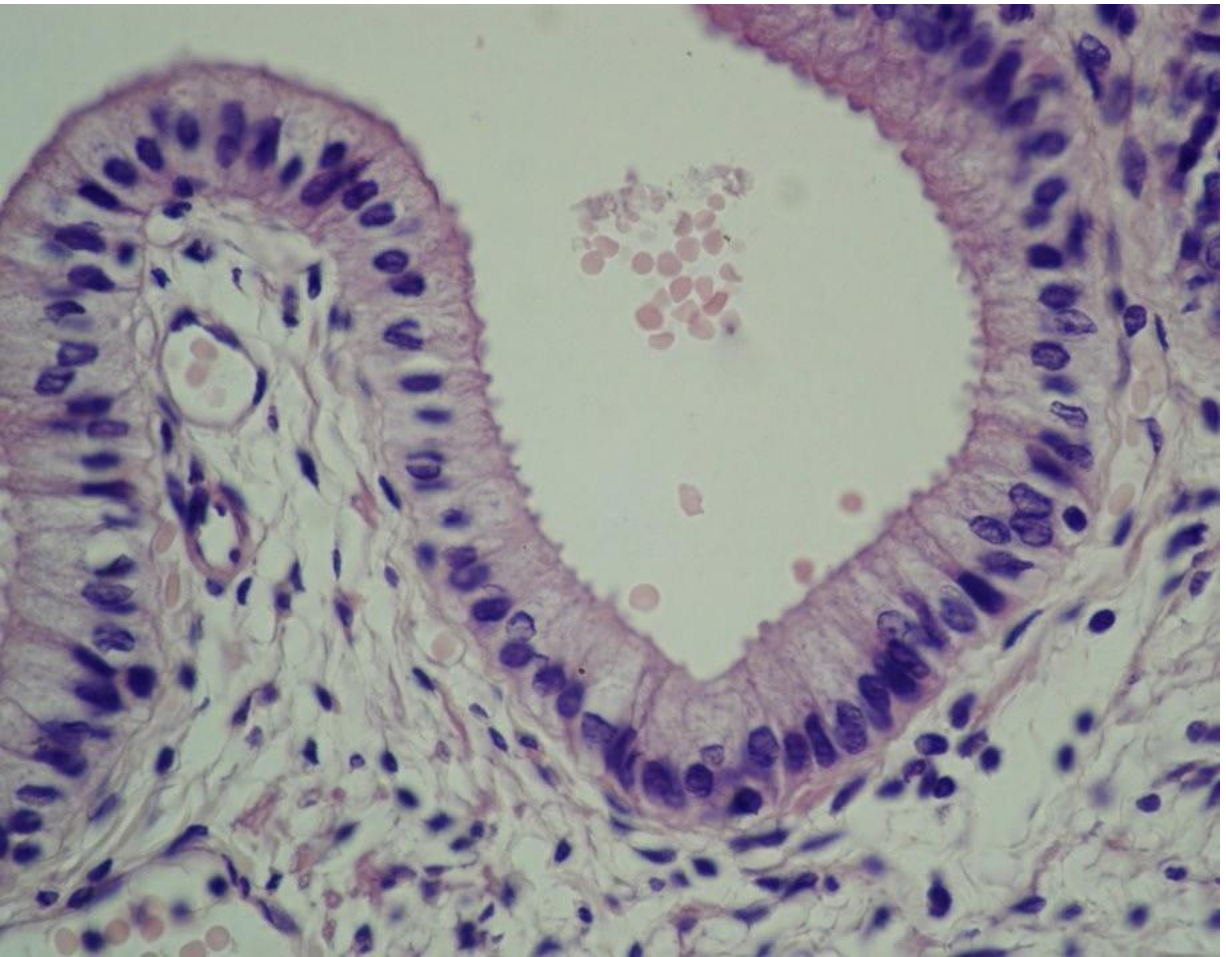


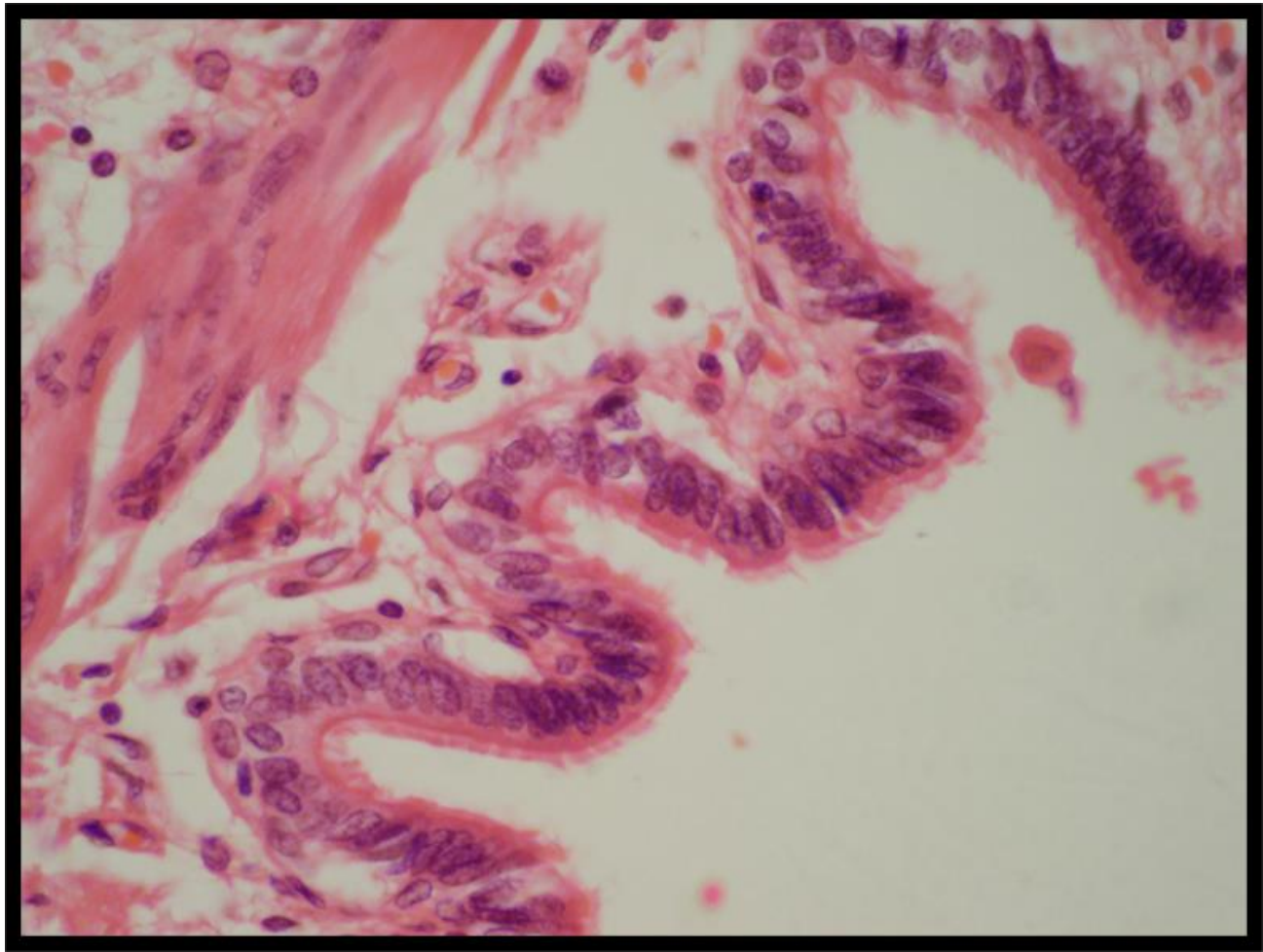
SIMPLE CUBOIDAL EPITHELIUM





SIMPLE COLUMNAR EPITHELIUM

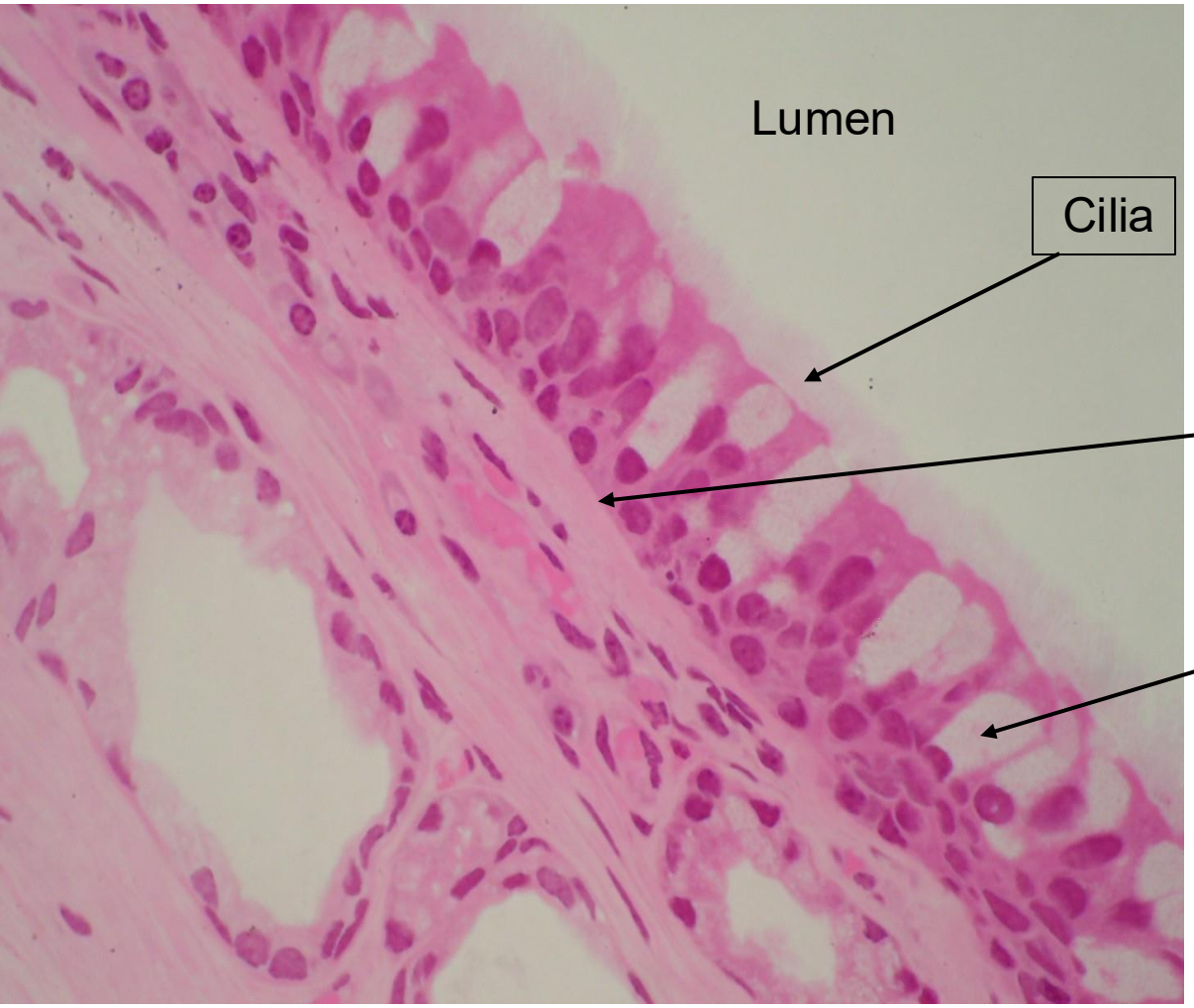




SIMPLE COLUMNAR EPITHELIUM

PSEUDOSTRATIFIED COLUMNAR EPITHELIUM

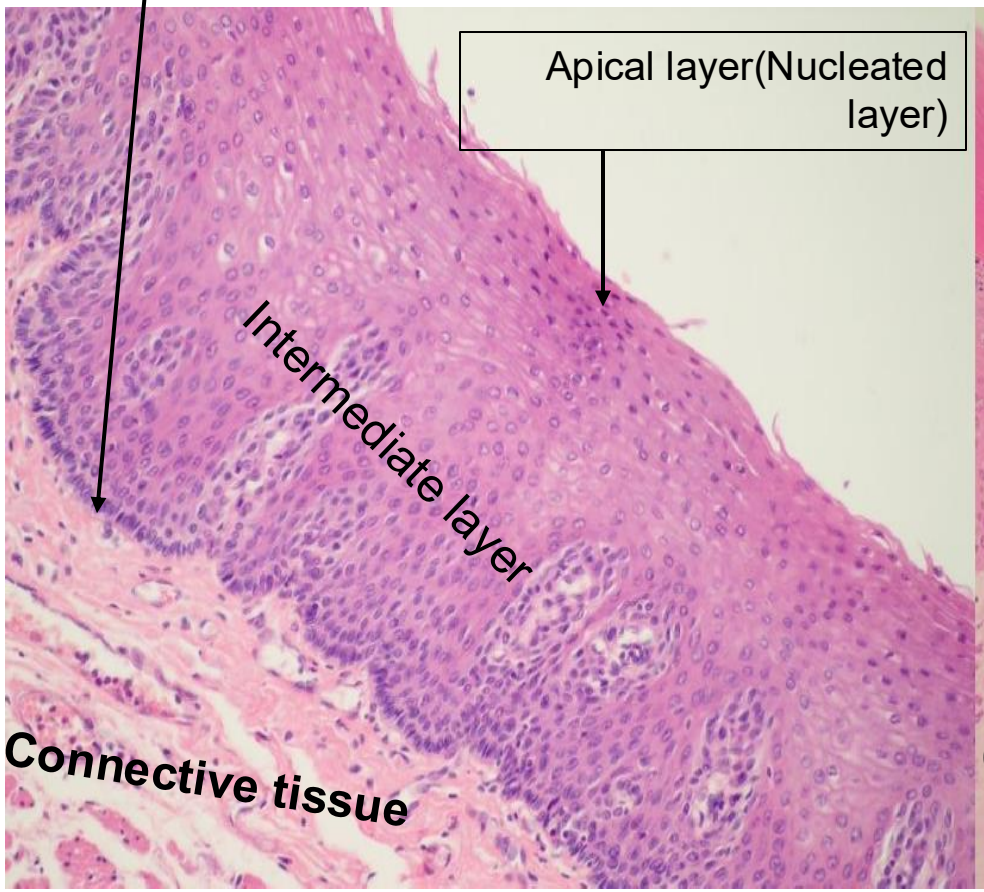
Stain: H&E (as we can't see specific details in the nuclei, and the pinkish areas appear throughout, with darker and lighter regions depending on the cell type. However, we cannot observe fine details).
Microscope: Bright field light microscope

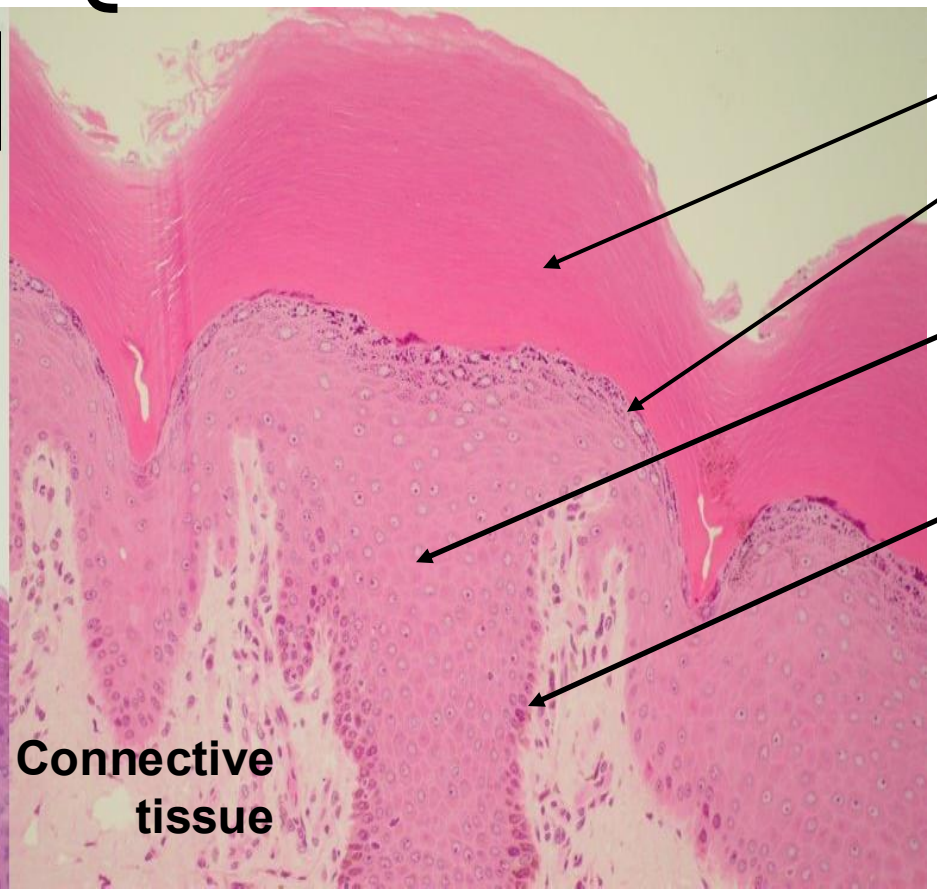


This type gives a false impression of being stratified. It consists of basal cells and apical cells. The basal cells are shorter, and their nuclei are positioned closer to the basement membrane. Most of these epithelial tissues are found in certain ducts, with apical cells attaching to the basement membrane while also reaching the surface, often alongside goblet cells. This type of epithelium is present in the male genital tract and the respiratory tract, where it exhibits apical modifications such as cilia or stereocilia.

The difference between epithelial tissue and connective tissue is that epithelial tissue consists of tightly packed cells arranged side by side, forming a sheet. In contrast, connective tissue consists of cells scattered in different directions without a specific shape.

STRATIFIED SQUAMOUS





The basal cell layer is at the interface with the connective tissue, and it is the only layer that attaches to the basement membrane.

In terms of their presence in the body, stratified squamous epithelium is the most common type of epithelial tissue, followed by stratified cuboidal epithelium, and then stratified columnar epithelium. For stratified columnar epithelium, its thickness must be significant in order to be clearly visible.

Stratified squamous non-Keratinized

- is found in the oral cavity, as well as in the gastrointestinal (GI) tract, specifically in the anal canal, and also in the vagina in females. This epithelium is subjected to mechanical stress, which is why it provides appropriate protection for these areas.

Stratified squamous keratinized

- is found in the epidermis of the skin, which lies above the dermis.

The Stratum Corneum is the outermost layer of the epidermis in stratified squamous keratinized epithelium. It consists of dead, unnucleated cells filled with keratin, forming a strong protective barrier against mechanical stress and dehydration

Notes related to the images above

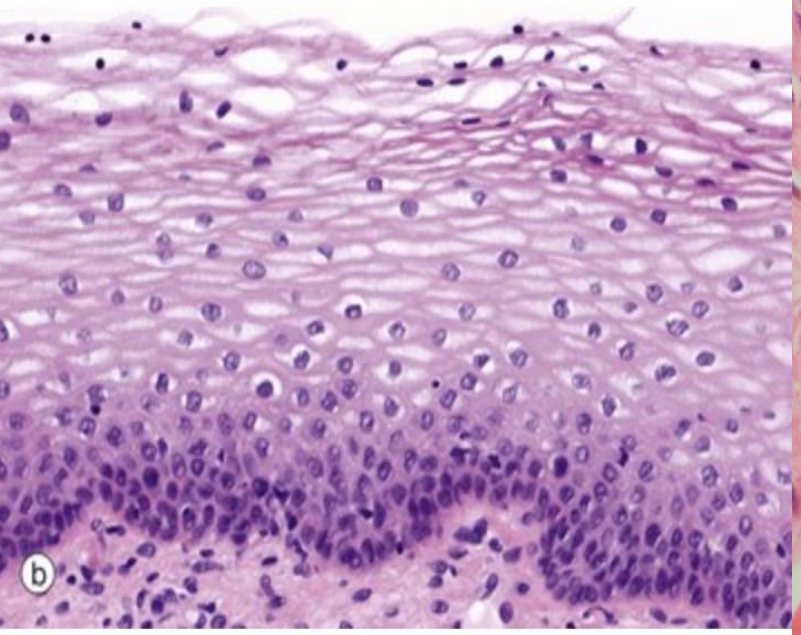
-Stratified squamous epithelium provides maximum protection. Even if some layers are lost, it is still protective because the loss does not quickly expose the underlying connective tissue. In contrast, simple squamous epithelium, or any simple epithelium, does not provide the same level of protection. Losing a layer in simple epithelium can lead to immediate exposure of underlying tissues, potentially resulting in bleeding or damage.

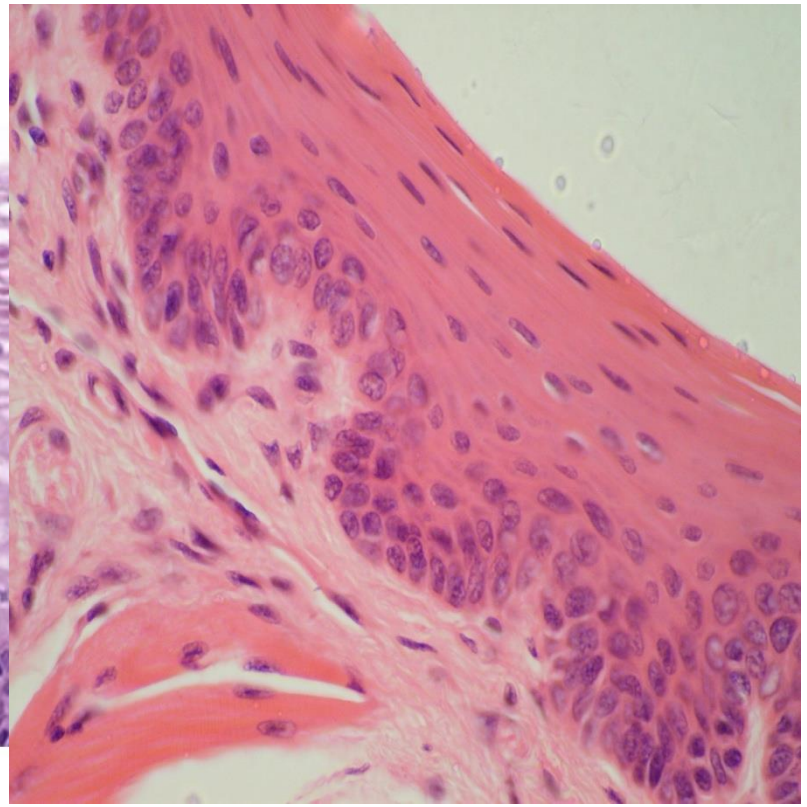
-In Stratified Squamous non-Keratinized Epithelium, the uppermost surface develops into what is known as superficial living cells, while the keratinized layer is referred to as superficial dead cells. This is because the cells spend their entire life synthesizing keratin and accumulating it within their cytoplasm up to a certain threshold. Eventually, their nuclei become condensed and lose their function, leading to the formation of a fully keratinized, unnucleated protective layer.

-There are two types of skin: thick skin, found on the palms of the hands and soles of the feet, and thin skin, found in areas like the wrists. The primary difference between these types is the thickness of the keratinized layer, known as the stratum corneum (the outermost layer of the epidermis), which is significantly thicker in thick skin.

Some sources discuss keratinization in the oral cavity, specifically on the tongue, but it is not as thick as shown in the right image above.

STRATIFIED SQUAMOUS



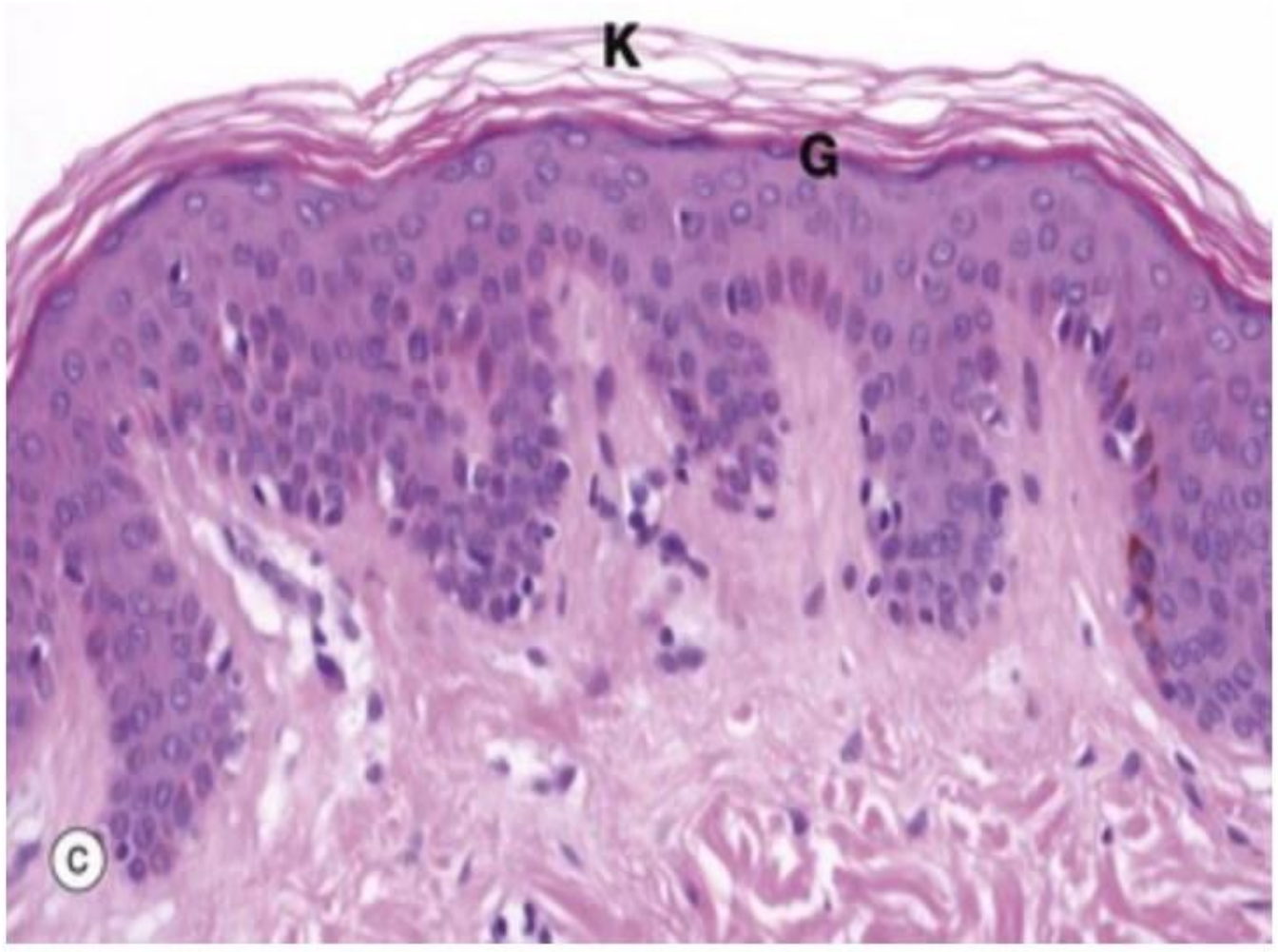


The cornea has stratified squamous epithelium, not simple squamous epithelium. If it had simple squamous epithelium, this would imply that the layers are very thin and would easily be rubbed off, which could cause damage like bleeding. In reality, the cornea has stratified squamous epithelium, but it is thinner compared to other areas, allowing light to pass through effectively for vision. If the cornea were thicker, it would impede the passage of light, making vision more difficult.

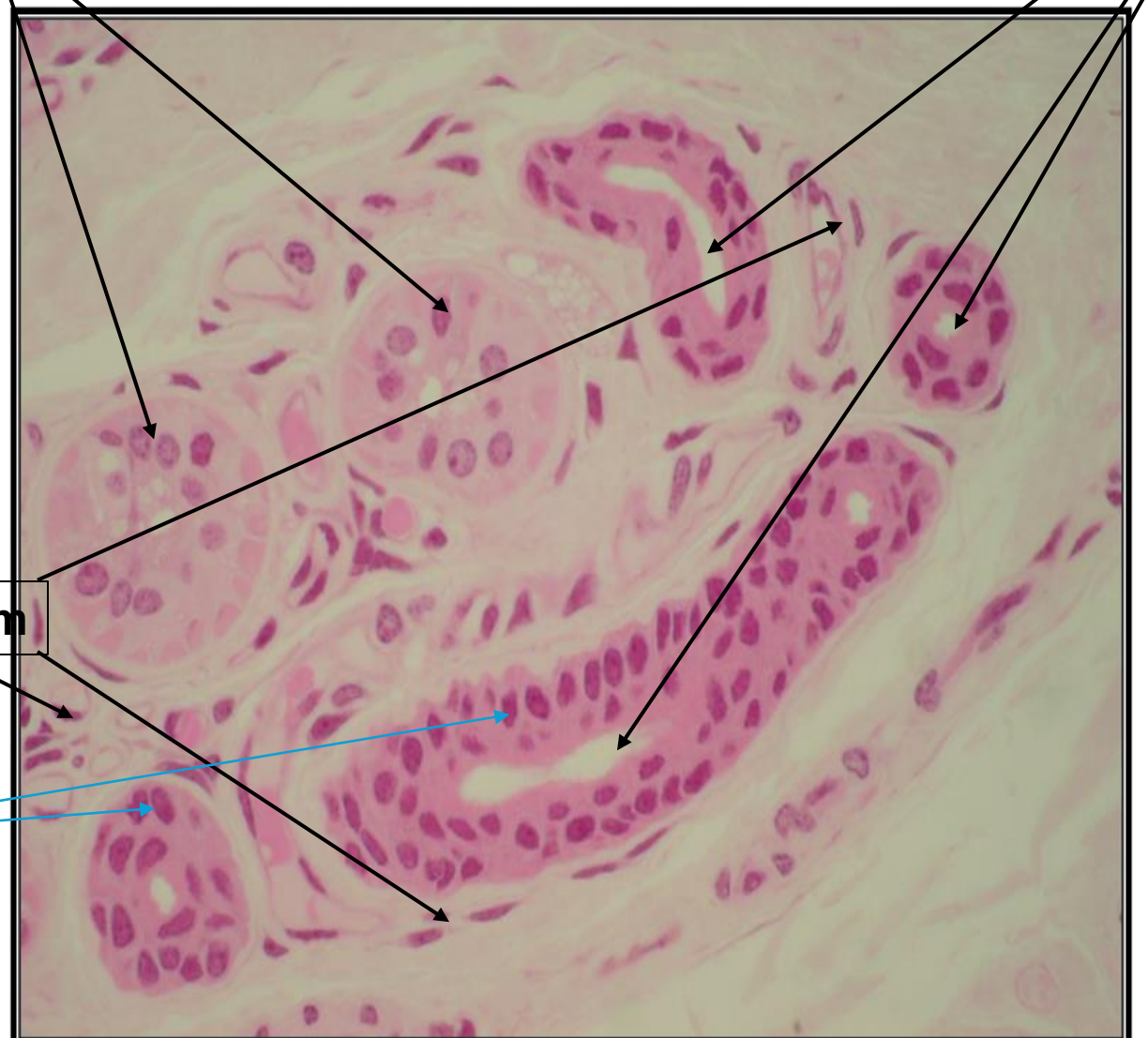
There are two images, and both belong to stratified squamous epithelium that is non-keratinized. These images represent stratified squamous non-keratinized epithelium because the upper layer in both contains nuclei, indicating that they are living cells. It is true that there might be some differences between these two layers in terms of thickness and the number of layers. However, as is known, stratified squamous epithelium varies in terms of the number of layers. For example, the layers in the anal canal are not the same as those in the oral cavity.

The connective tissue in the cornea is unique because it consists of specialized collagen fibers arranged in a precise, orderly pattern, allowing transparency and providing structural support.

KERATINIZED-STRATIFIED SQUAMOUS



This is keratinized stratified squamous epithelium, as we can see that there are no nuclei in the upper layer, indicating that these are dead cells. Additionally, the number of keratin layers is few, which makes it a type of thin skin.

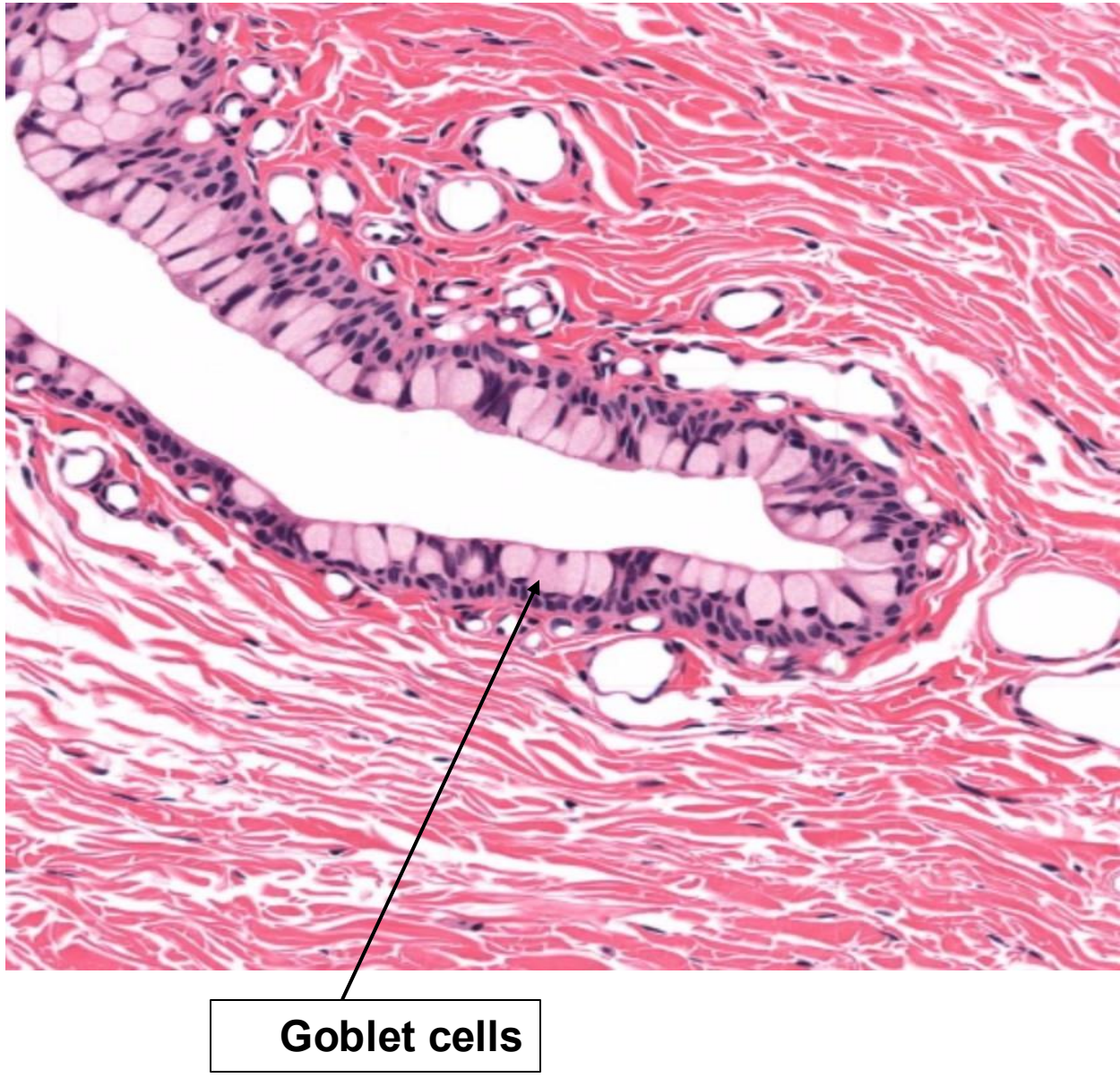


Cuboidal cells cannot all be classified uniformly because there are different types, including low cuboidal, medium cuboidal, and high cuboidal cells. In high cuboidal cells, the cytoplasm is equal in height to the nucleus, whereas in low cuboidal cells, the cytoplasm is much thinner compared to the height of the nucleus.

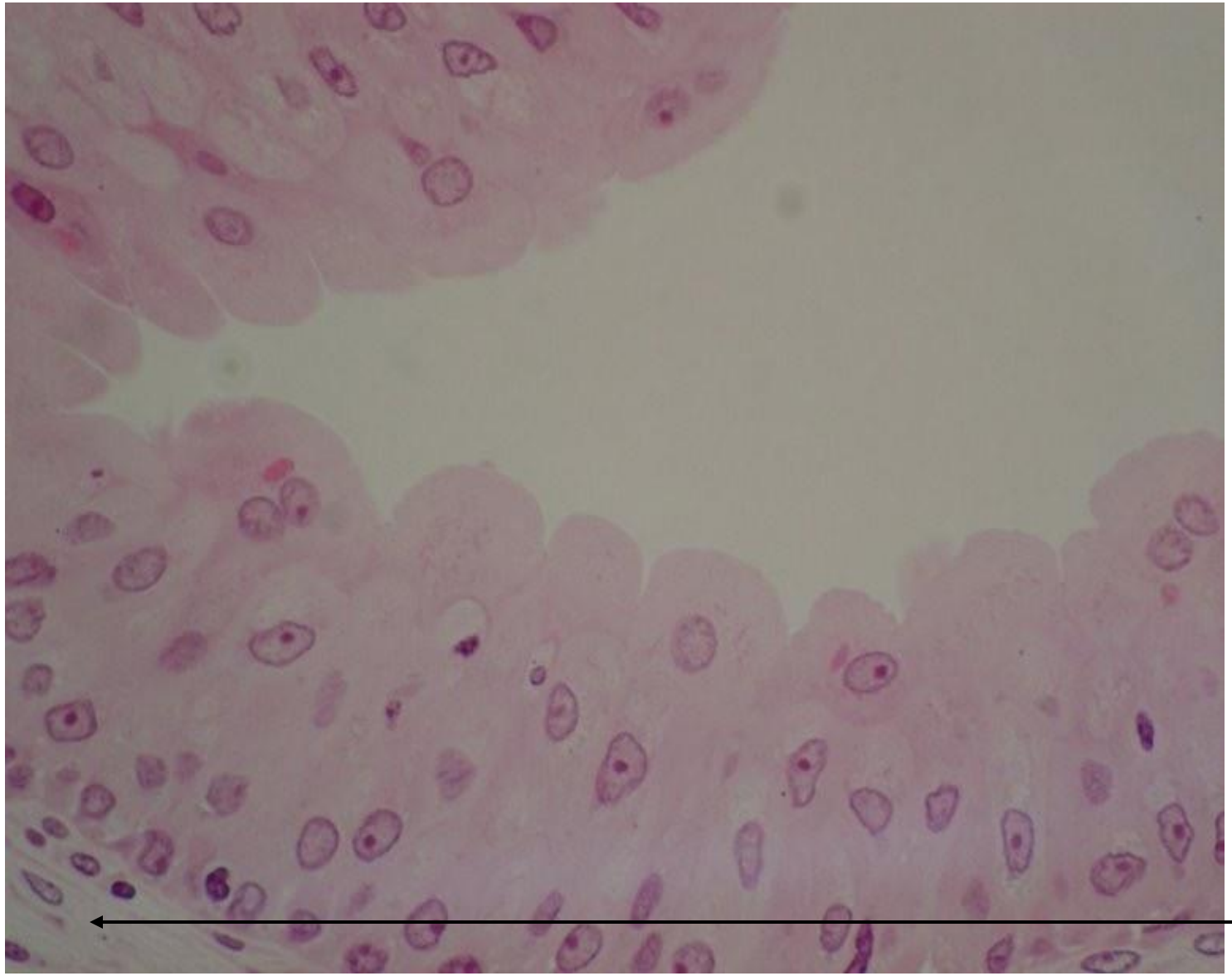
Typically, cuboidal cells can show apical modifications, such as cilia or usually microvilli , depending on the location. For example, in the female genital tract or in other specialized regions, cuboidal cells may have cilia to aid in the movement of substances.

STRATIFIED COLUMNAR

This type of epithelium, stratified columnar epithelium, covers the underlying structures beneath the eyelids as well as the sclera. One of the best locations to study stratified columnar epithelium is the conjunctiva, as it is a well-known site for this type of tissue. When examining stratified columnar epithelium, the basal cells are usually polyhedral or cuboidal in shape, while the apical cells are columnar. Additionally, goblet cells are found in between, which secrete mucus. These goblet cells typically have a basally located nucleus.



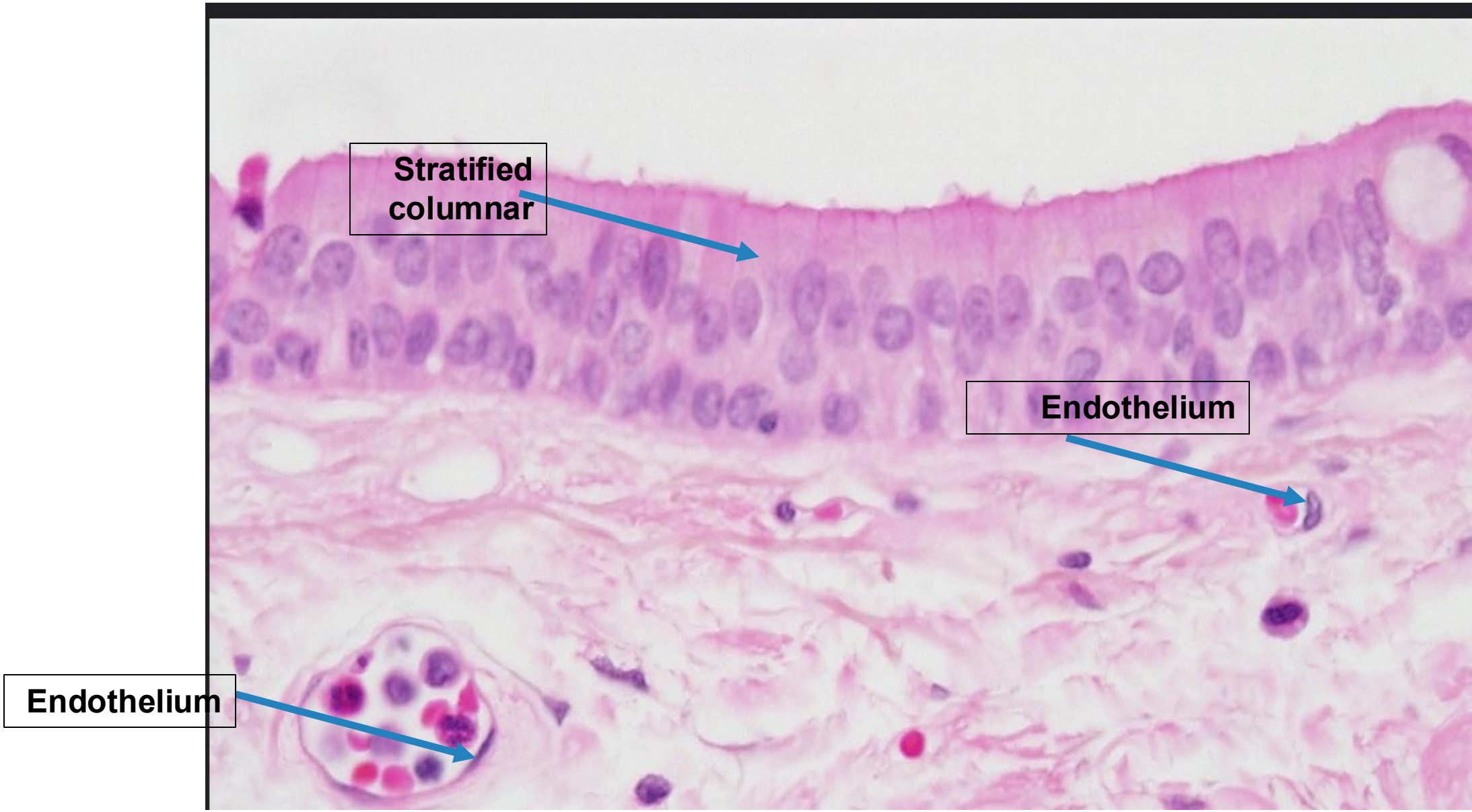
TRANSITIONAL EPITHELIUM

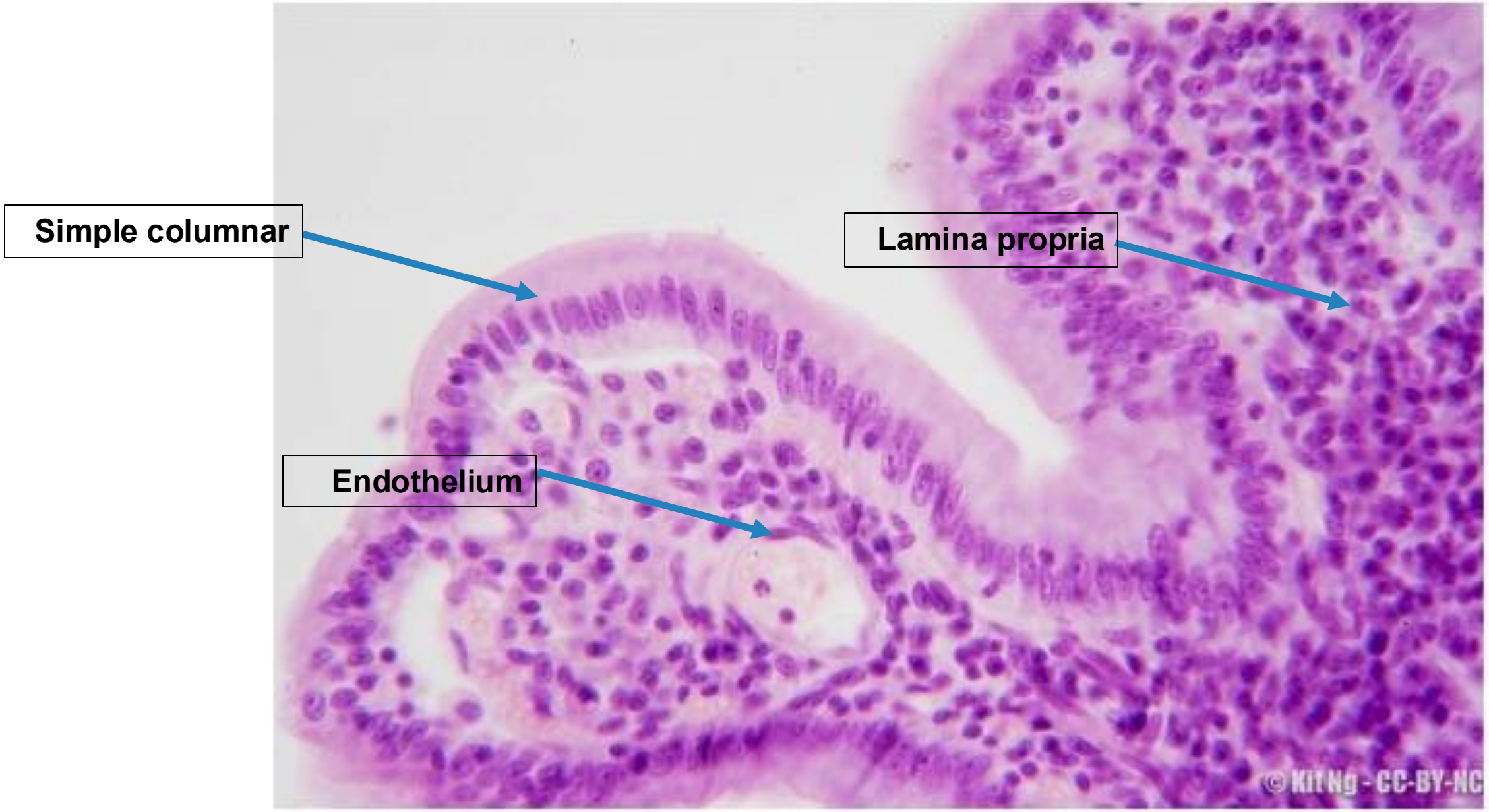


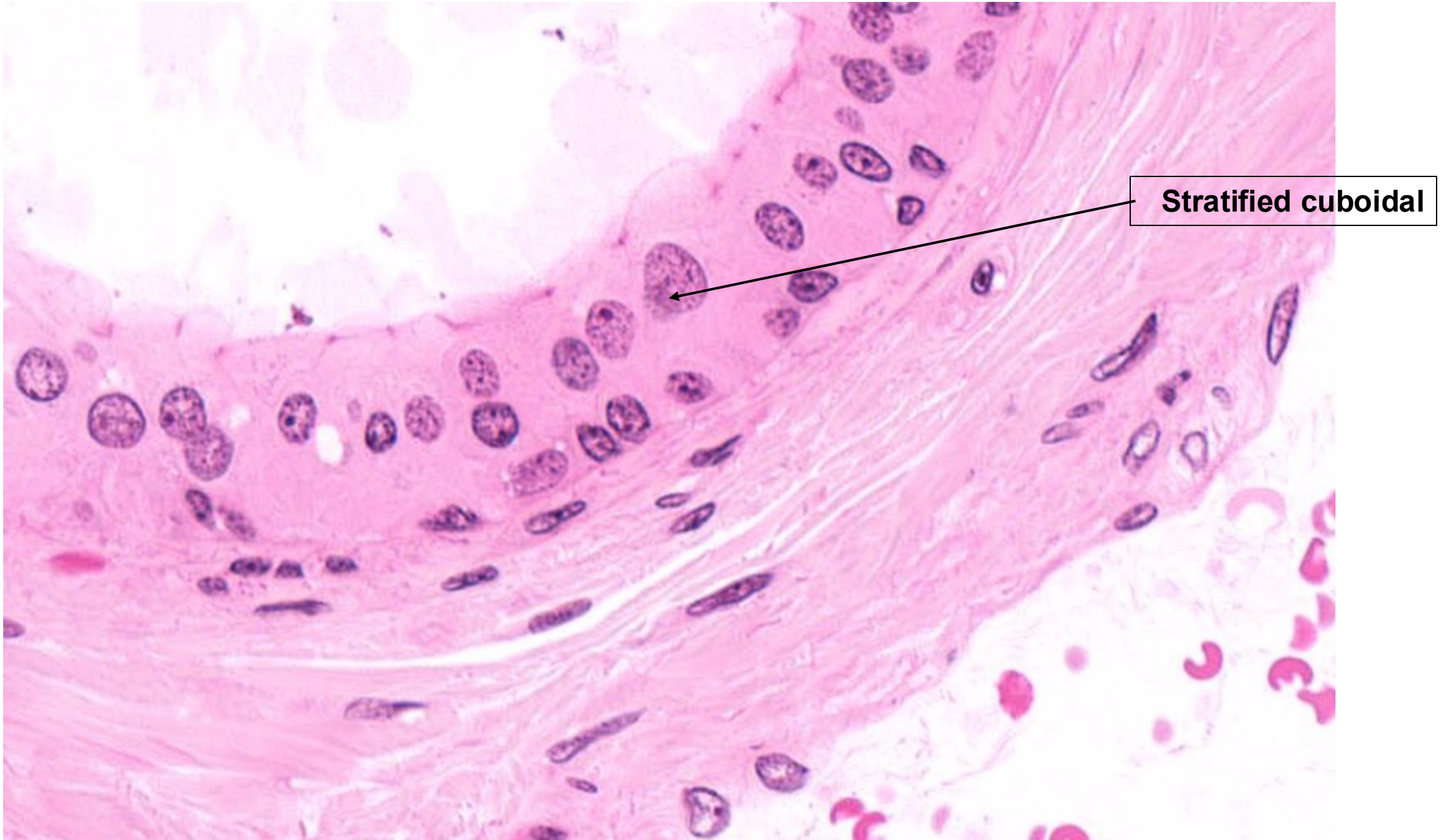
To identify transitional epithelium, we need to consider two key factors. First, it often shows binucleation, meaning some of the superficial cells may have two nuclei, though this is not always the case. Second, the shape of the superficial cells is dome-shaped or umbrella-like in a relaxed state.
For example, in this image, the cells appear to be in a relaxed state, as they maintain their dome shape. If they were not in this state, the cells would appear flattened to accommodate stretching.

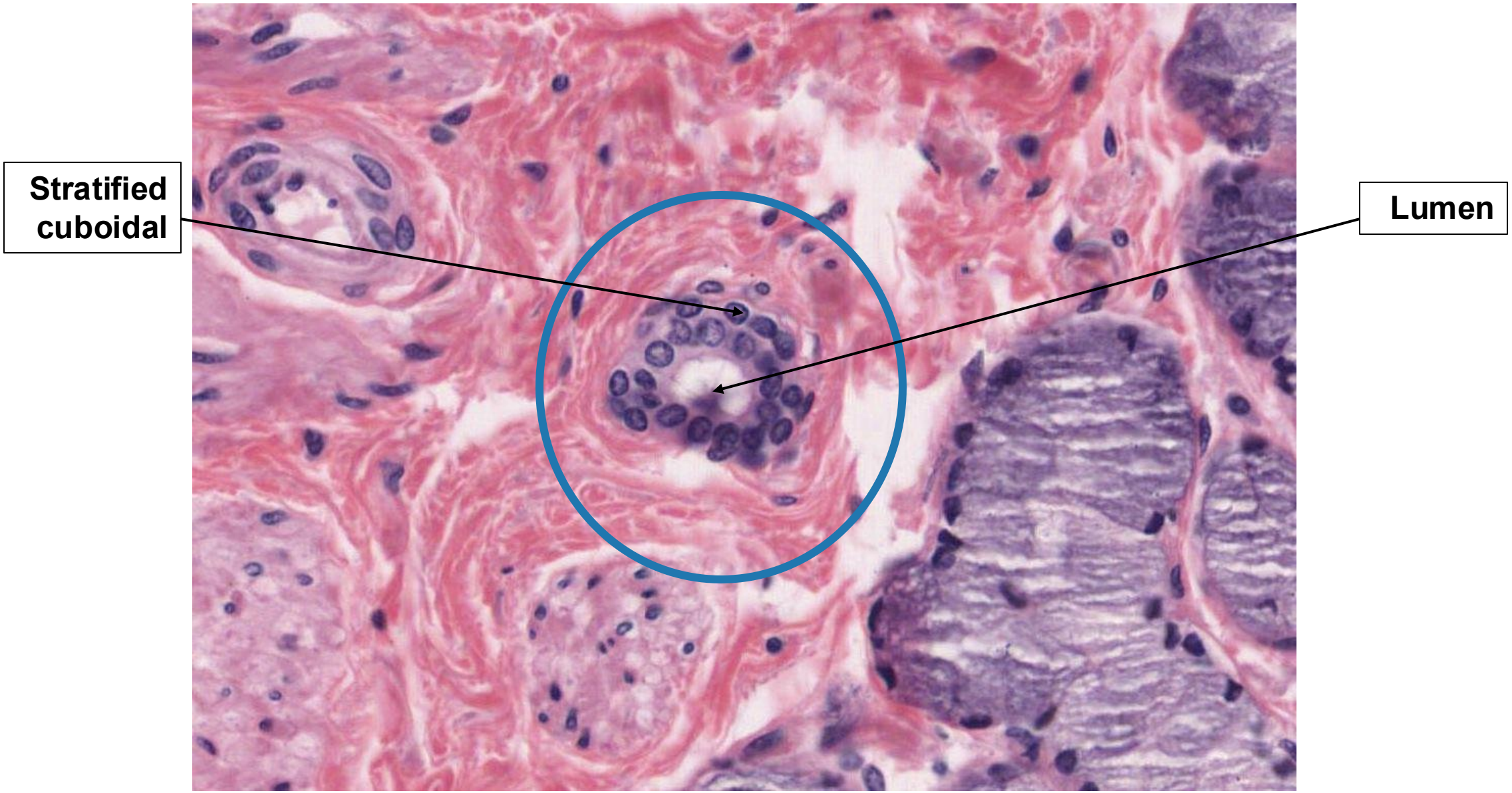
This is the lamina propria, which is a type of loose connective tissue located beneath the epithelial tissue in the gastrointestinal (GI) tract, respiratory tract, and urogenital tract.

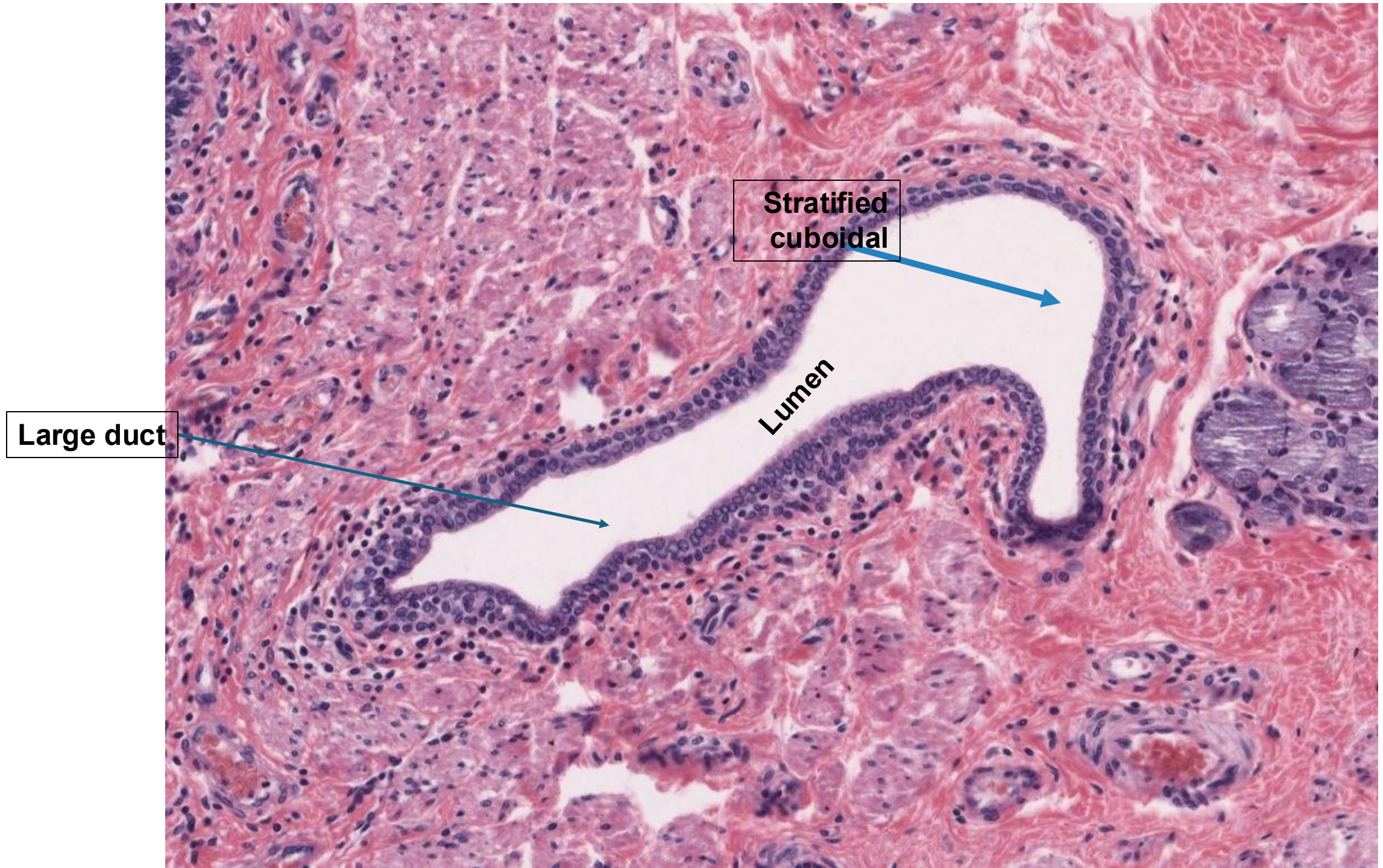
IDENTIFY

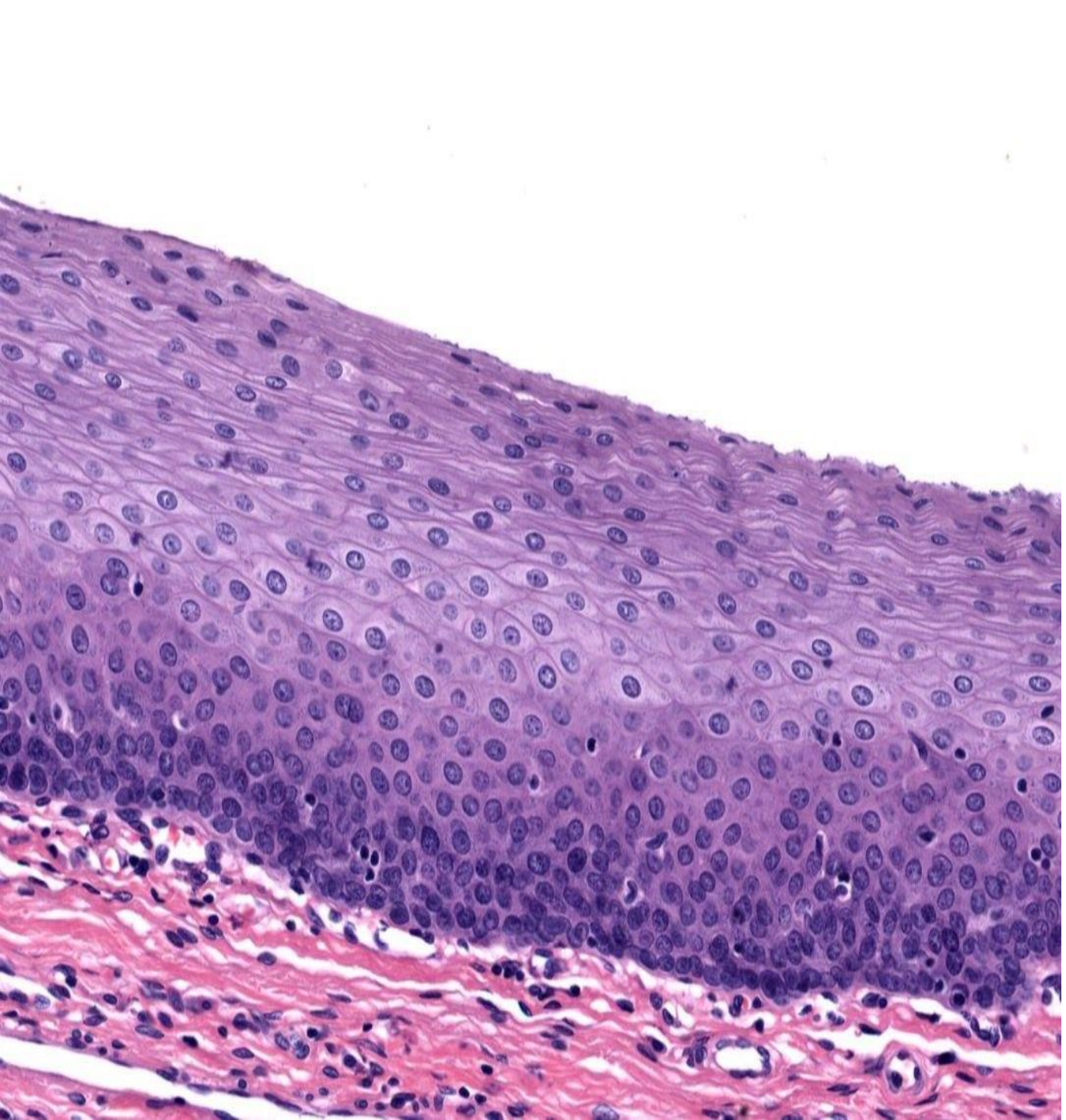

Stratified columnar
Endothelium
Endothelium


Simple columnar
Lamina propria
Endothelium
© Kit Ng - CC-BY-NC

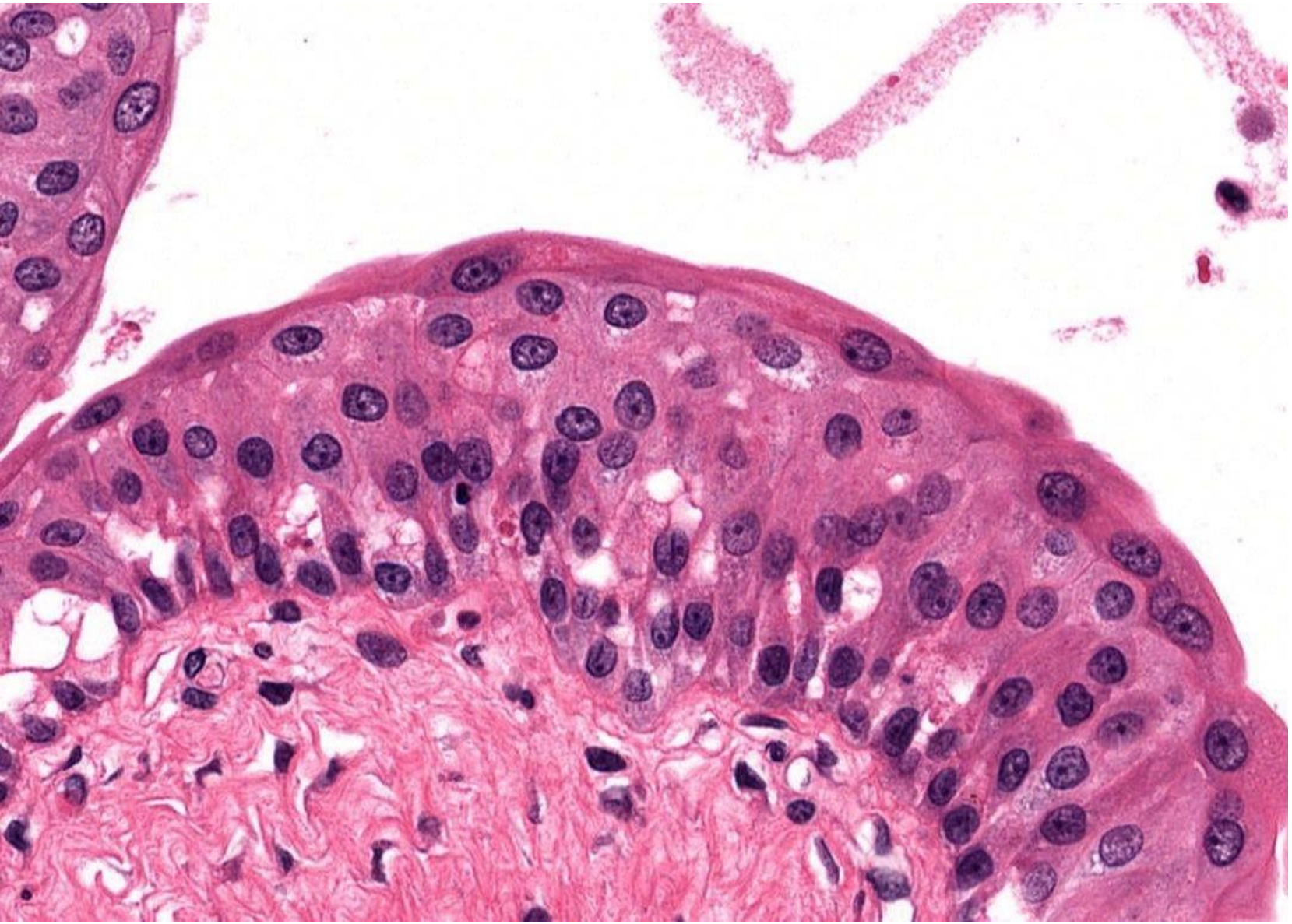

Stratified cuboidal


Stratified cuboidal
Lumen

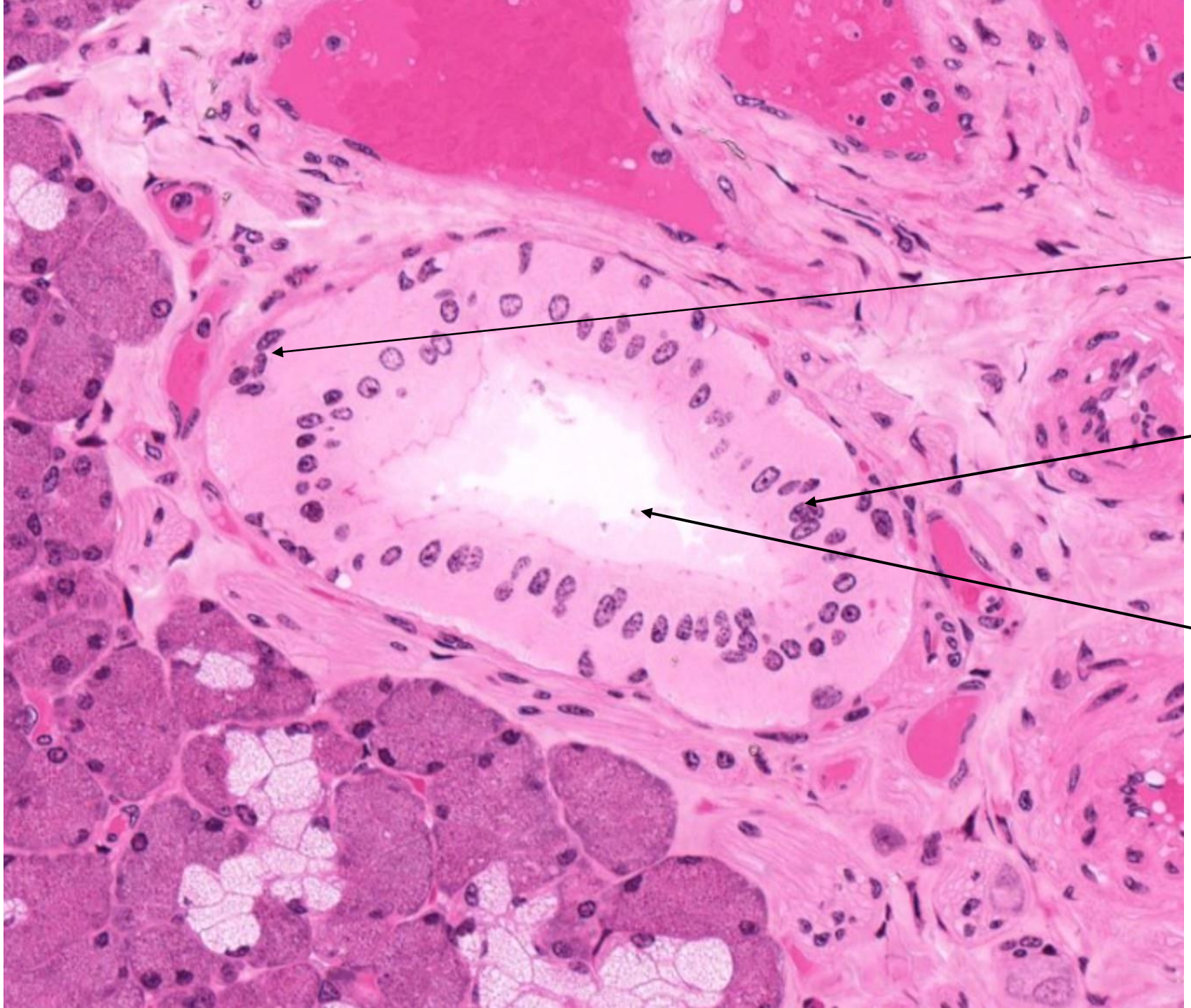

Stratified cuboidal
Lumen
Large duct



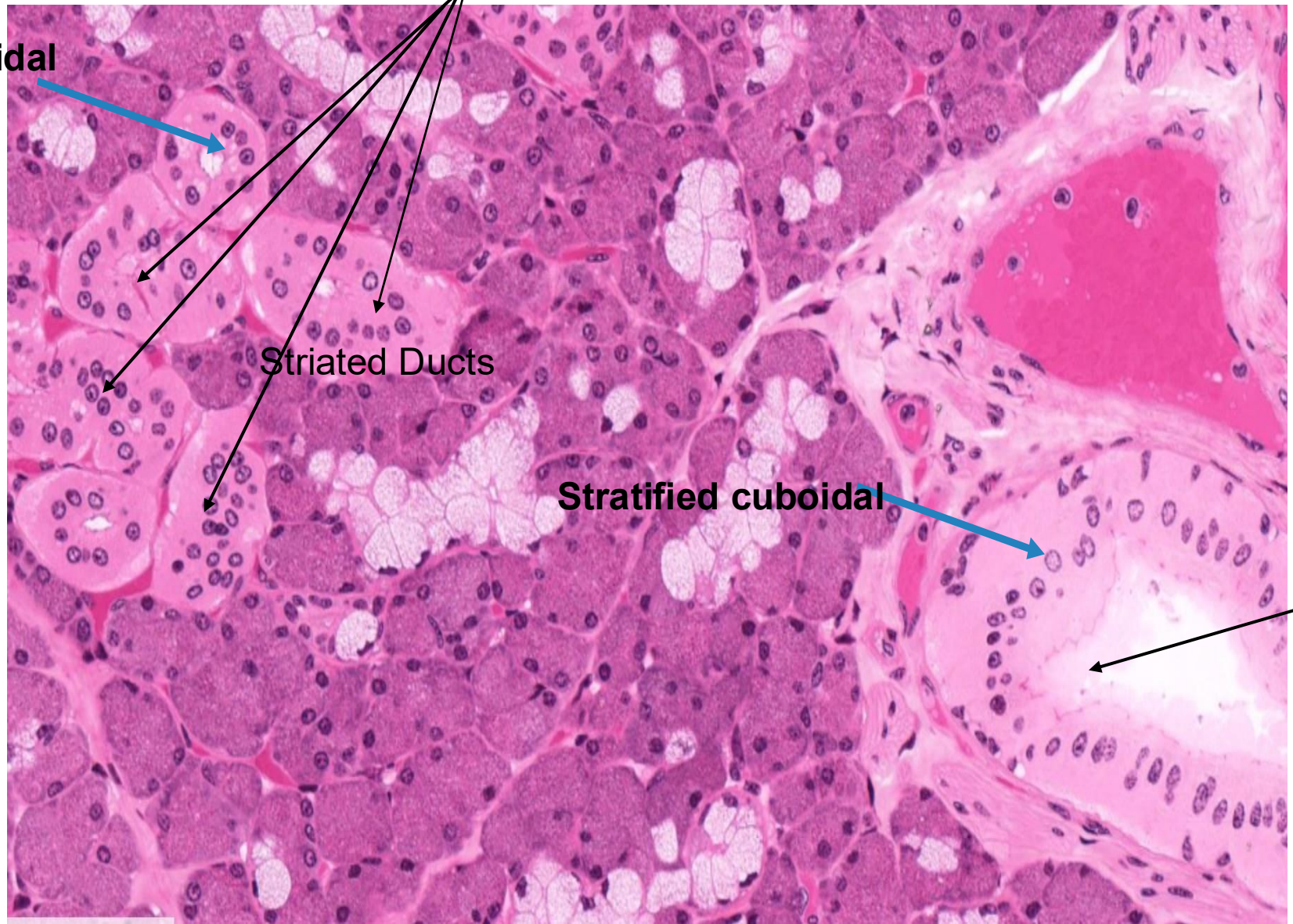
This is Stratified Squamous <u>Non-Keratinized</u> Epithelium because the superficial cells are nucleated, meaning all the cells are living cells.



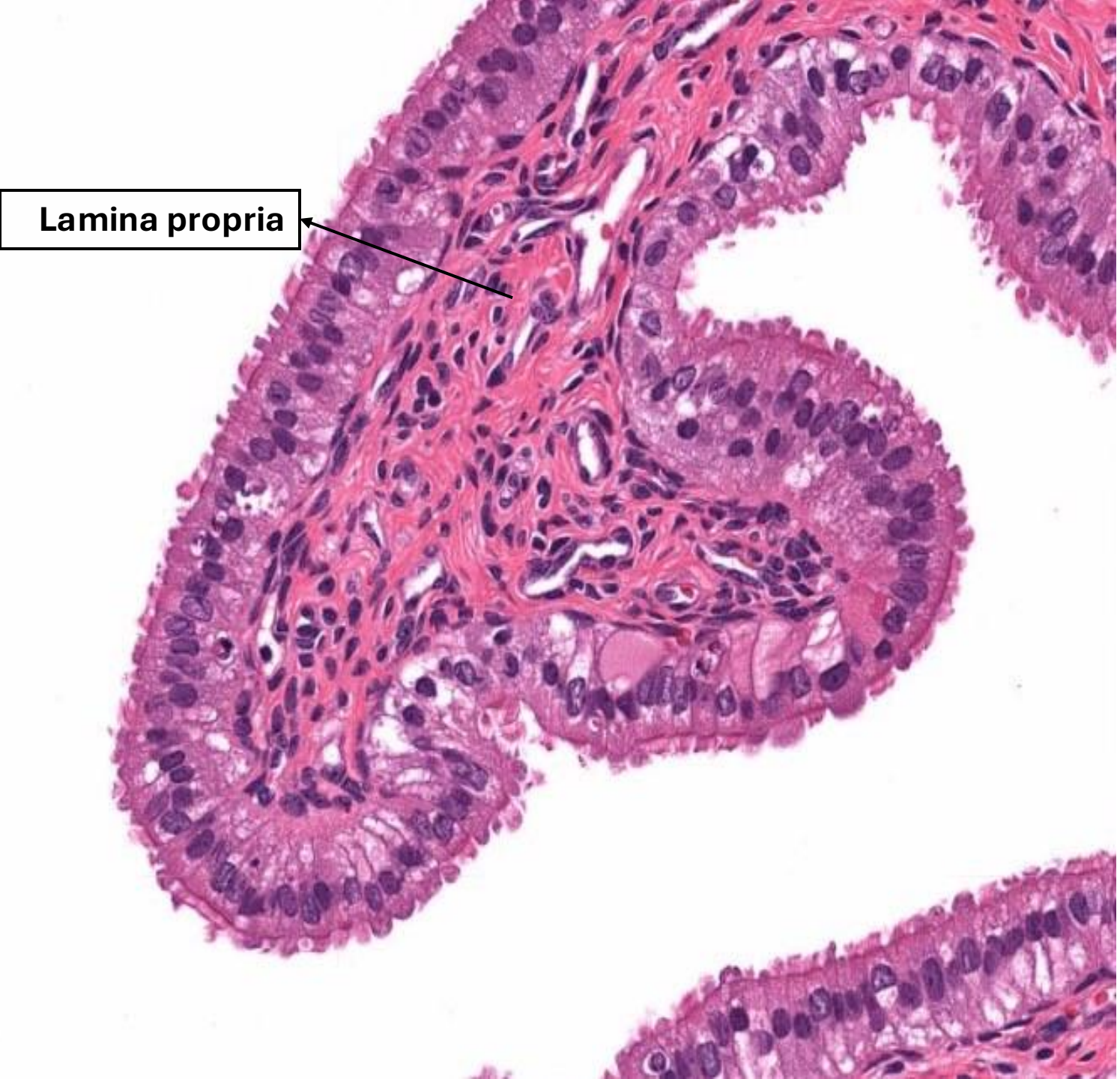
This is Transitional Epithelium, as we can see that the superficial cells have an umbrella-like or dome shape. Additionally, some of these cells contain two nuclei, a characteristic known as binucleation.


Basal cells
Stratified cuboidal
Large duct

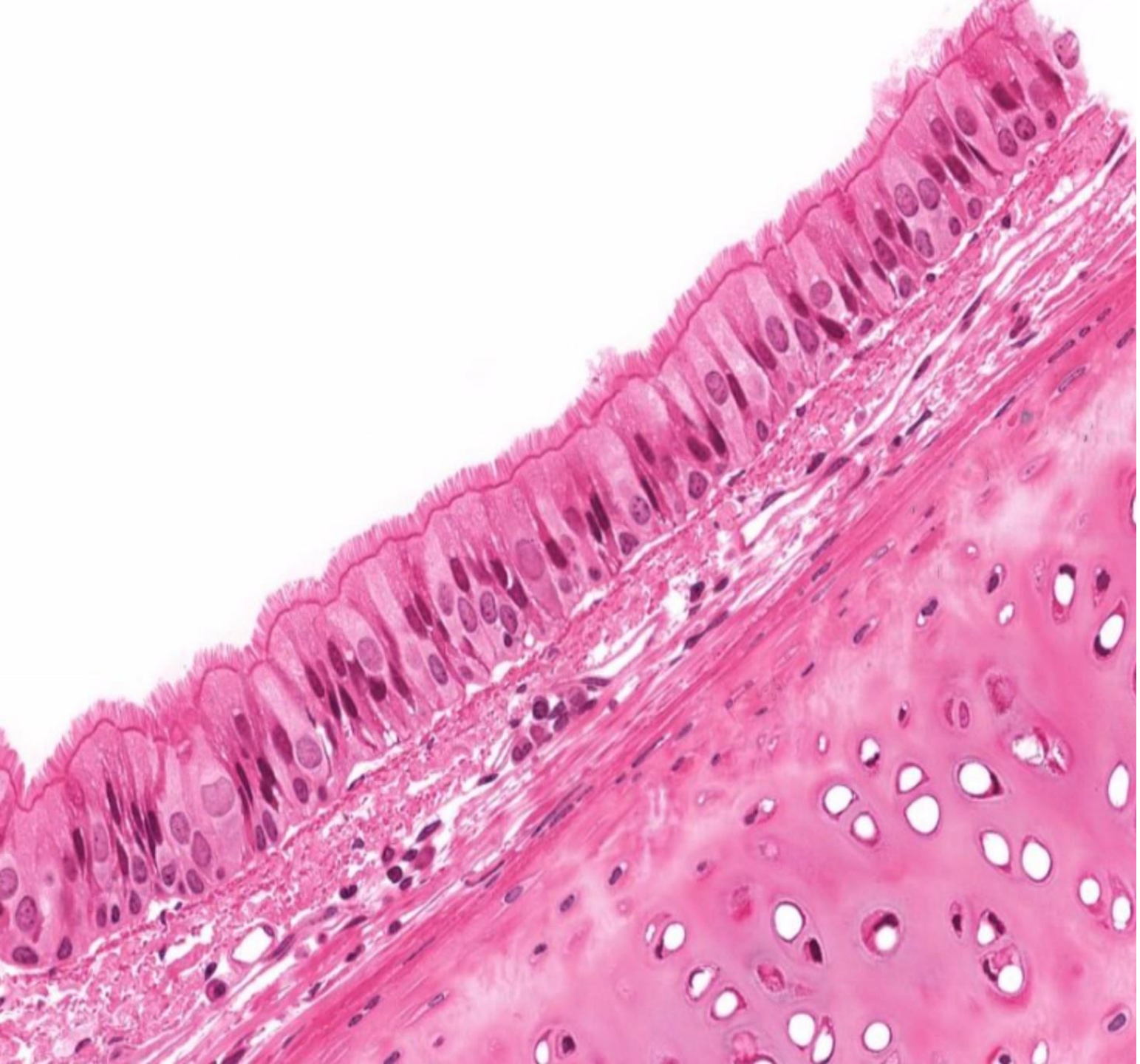
This is image was taken from mandibular gland



Small ducts have a thinner lining , while larger ducts have a thicker lining

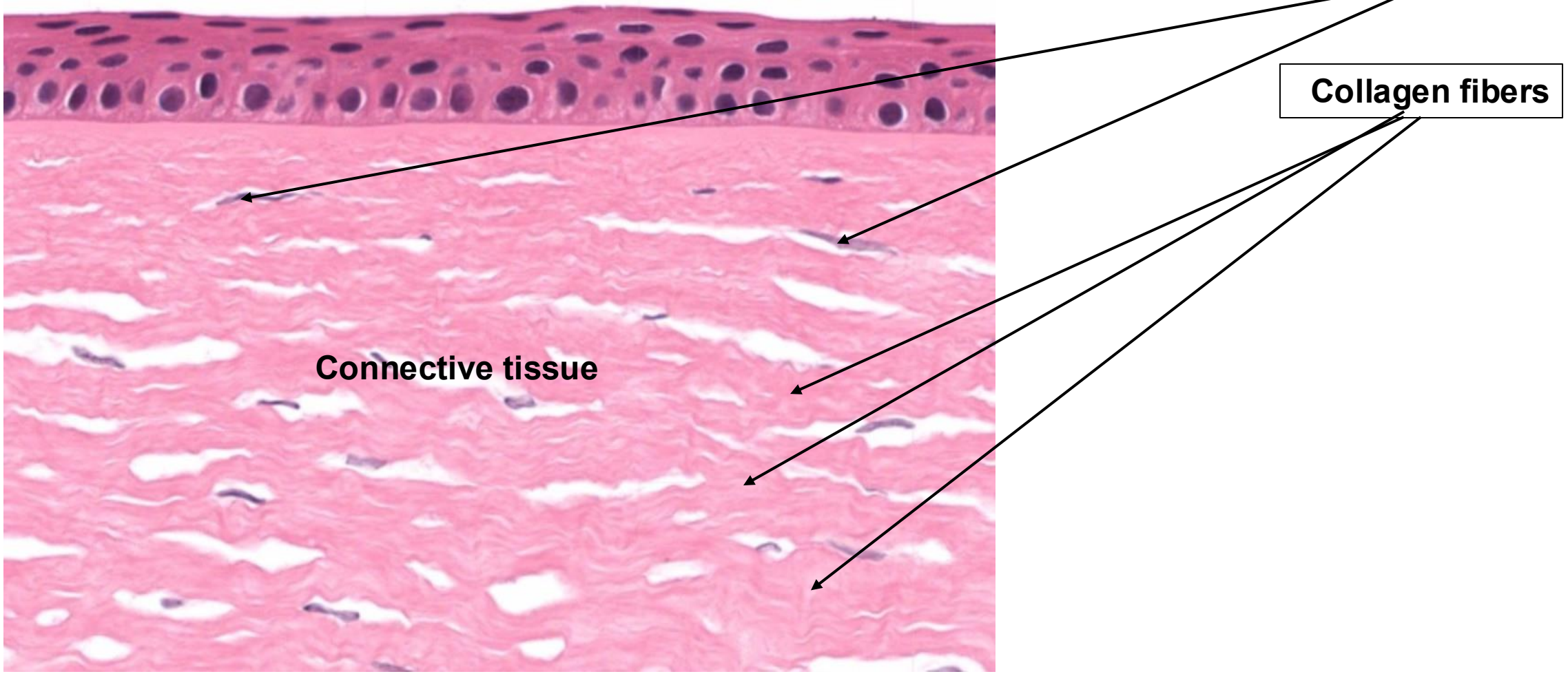


*This tissue is *simple columnar epithelium, and we have identified that it contains cilia, not microvilli, because cilia are longer than microvilli. If there were microvilli, it would be difficult to see the spaces between them, while with cilia, the spaces between them are visible because of their length. It cannot be considered stereocilia because they are not branched. Additionally, this image was taken from the female genital tract.*

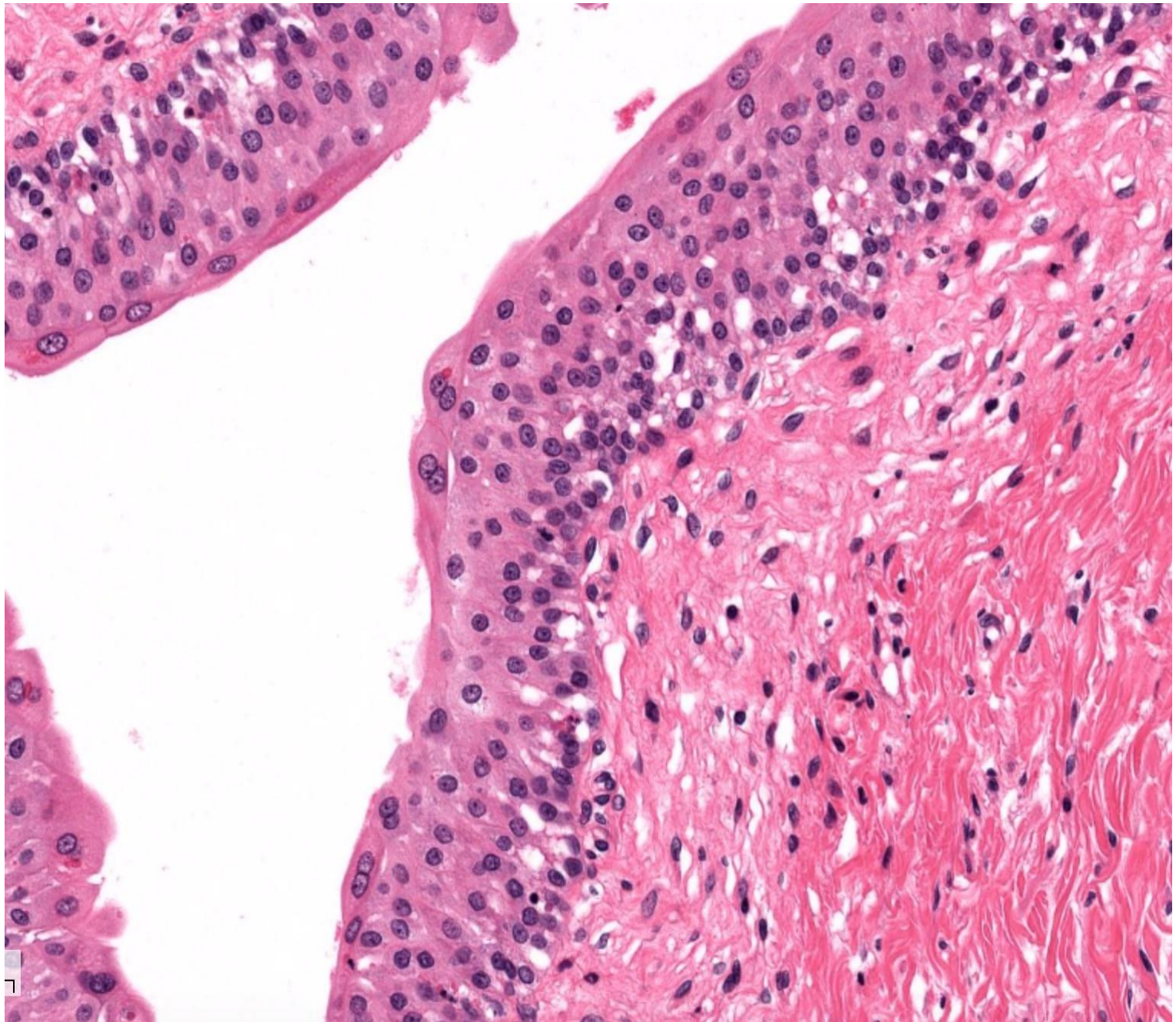


Pseudostratified columnar epithelium is referred to as respiratory epithelium when it is ciliated and contains goblet cells. In this case, it is specifically called ciliated pseudostratified columnar epithelium with goblet cells, as shown in this image.

This represents Stratified Squamous Non-Keratinized Epithelium, which covers the cornea.



This is Transitional Epithelium.



For any feedback, scan the code or click on it.

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1	Slide 4 A wrong sentence	These are planted or grown cell	These are cells observed within a natural tissue environment.
	Slide 16 The type of epithelium	Keratinized epithelium	Non-keratinized epithelium
	Slide 32 The type of epithelium	Pseudostratified	Simple columnar
V1 → V2			