Epithelial Structure and Transport

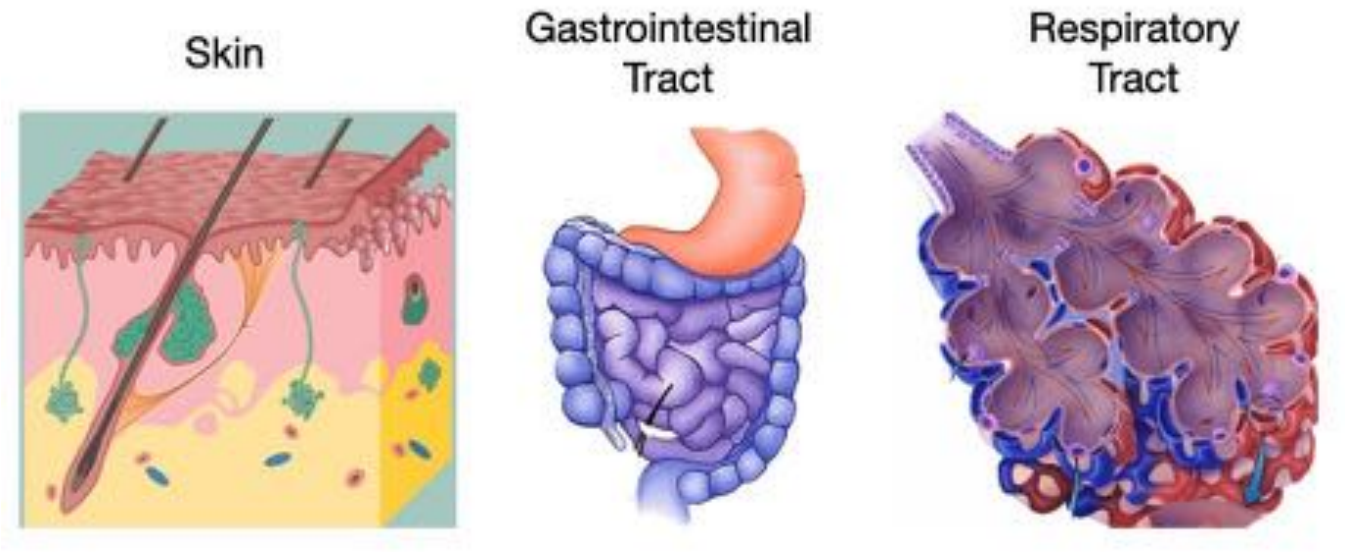
Scientific Foundations

Learning Objectives

1. Students should be able to describe how different types of epithelia (simple, stratified, squamous, columnar) facilitate different functions.
2. Students should be to list the junctional complexes between epithelial cells together and their primary functions in adhesion and permeability.
3. Students should be able to describe how the basement membrane regulates the activity of epithelia.
4. Students should be able to define the apical and basal surfaces of epithelial cells and describe how their unique compositions are maintained.
5. Students should be able to locate stem cells in the epithelia of intestine and trace their pathway of differentiation and development.

Barrier to Separate Compartments

An important structural feature of our bodies is the ability to separate two environments and create a functional interface between them. For example, our skin separates our bodies from the external environment but also interacts with the external environment through physical sensation and perspiration. Moreover, the external environment enters are bodies through the respiratory system and gastrointestinal tract. In both of these systems, we need a structure that interfaces with the external environment that offers both protection and the ability to take up components of the external environment that our bodies need to survive. We also need systems and structures that removes waste and excretes material from our bodies to the external environment.



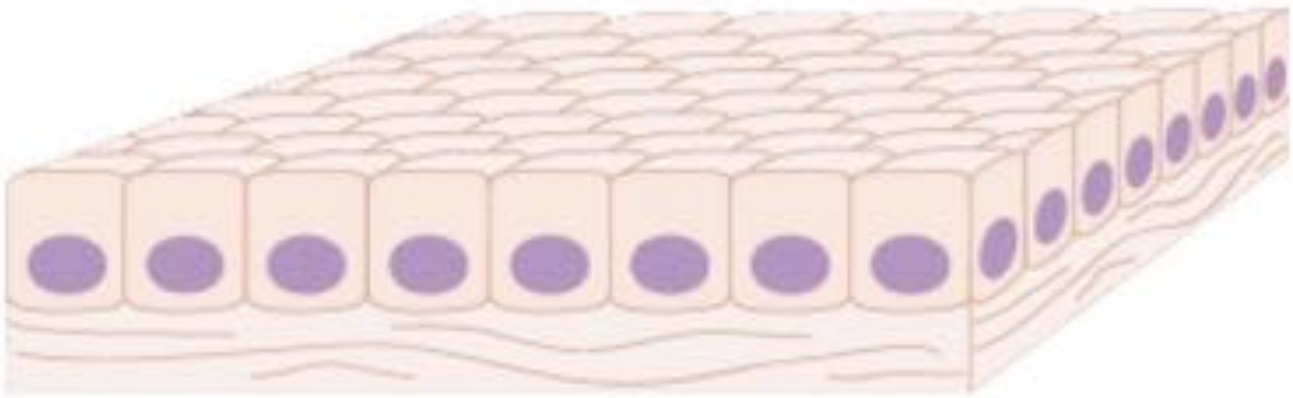
Epithelia form an interface between the human body and external environment.

In all of these systems the tissue that forms the interface between the external environment and our bodies is epithelium. Epithelium is the most abundant tissue and versions of it are found in most organs.

Epithelium is the most functionally diverse tissue. Versions of epithelia protect the body from mechanical force, chemical damage, electromagnetic radiation (UV light) and desiccation. Other epithelia exchange material with the external environment. For example, epithelia mediate gas exchange in our lungs, absorption of nutrients and excretion of waste in our gastrointestinal tract and controlled exchange of ions and water in our urinary system.

Epithelium Structure

The myriad of functions performed by epithelium require it do adopt different structures that facilitate those functions. Because epithelia often cover large surface areas (the internal surface area of your lungs is almost the same size as a tennis court), they form a sheet of cells. The lateral interaction between the cells and the interaction between the cells and the underlying structures in the body maintain the integrity of the epithelium. The structure of epithelia will differ based on the shape of their cells and the number of layers of cells.

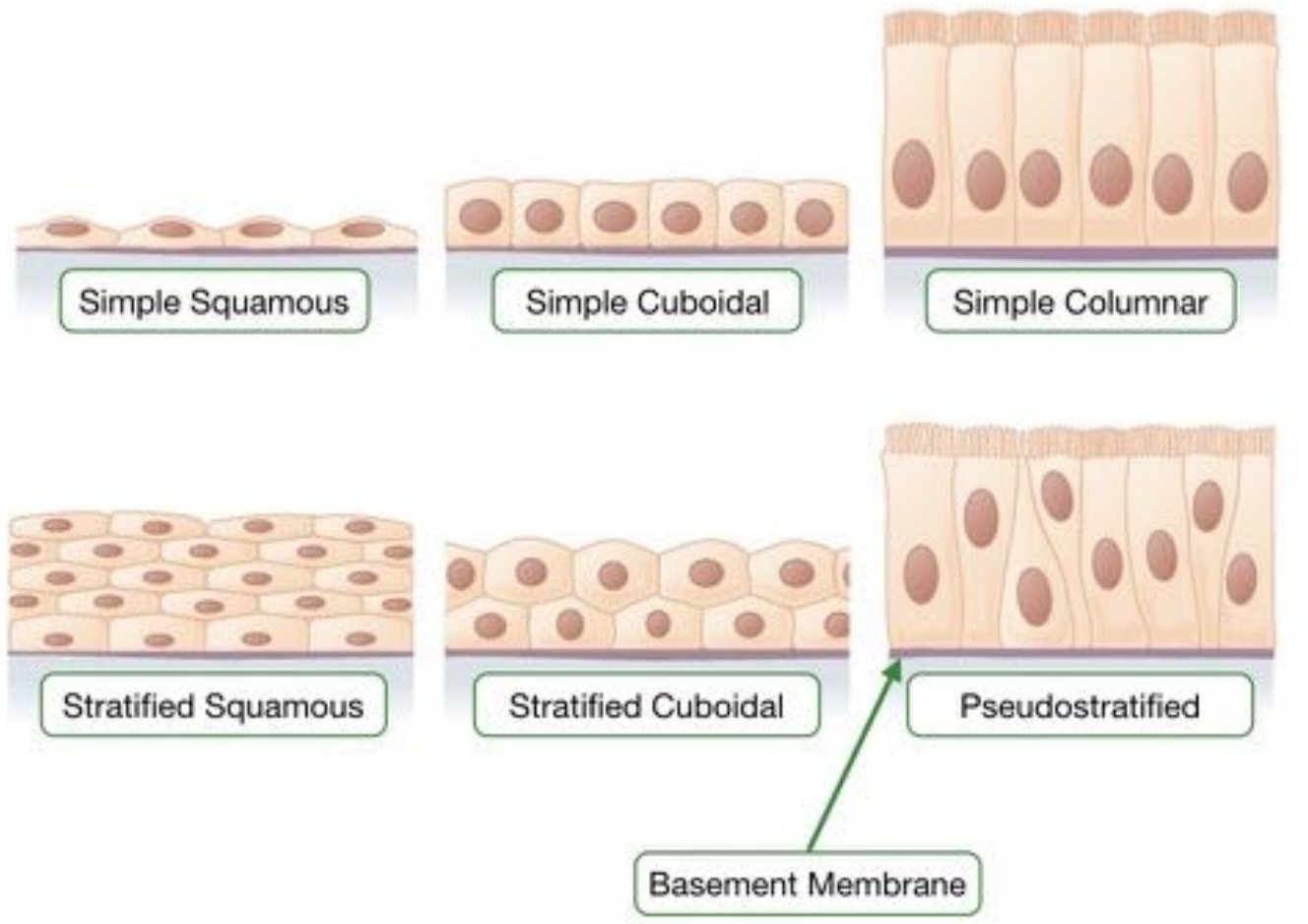


An epithelium is a sheet of cells that can cover a large surface area.

Epithelial Classification

An epithelium with a single layer of cells is called simple whereas an epithelium with two or more layers of cells is called stratified. If the most superficial (outer) layer of cells is flat, the epithelium is referred to as squamous. Epithelia where the outer layer of cells are about as tall as they are wide are called cuboidal. If the cells are taller than they are wide, then they are called columnar. Combining descriptors for the layers and shape of cells allows us to efficiently classify most epithelia.

For example, a simple squamous epithelium is one that has a single layer of flat cells. This type of epithelium is well-suited for exchange of gases because the thin cells reduce the distance that gases must diffuse between the external environment and the body. Epithelia with a single layer of cells that are taller are called simple cuboidal or simple columnar depending on the height of the cells. These types of epithelium are often involved in the exchange of nutrients and ions between the body and external environment. The larger volume of cytoplasm allows the cells to house more organelles which are needed to generate ATP or synthesize and deliver proteins to the cell membrane. An epithelium with multiple layers of cells where the most superficial layer is flat is called stratified squamous. Stratified squamous epithelium offer protection against mechanical and chemical insults and are more resistant to desiccation. This is the type of epithelium that composes are skin.



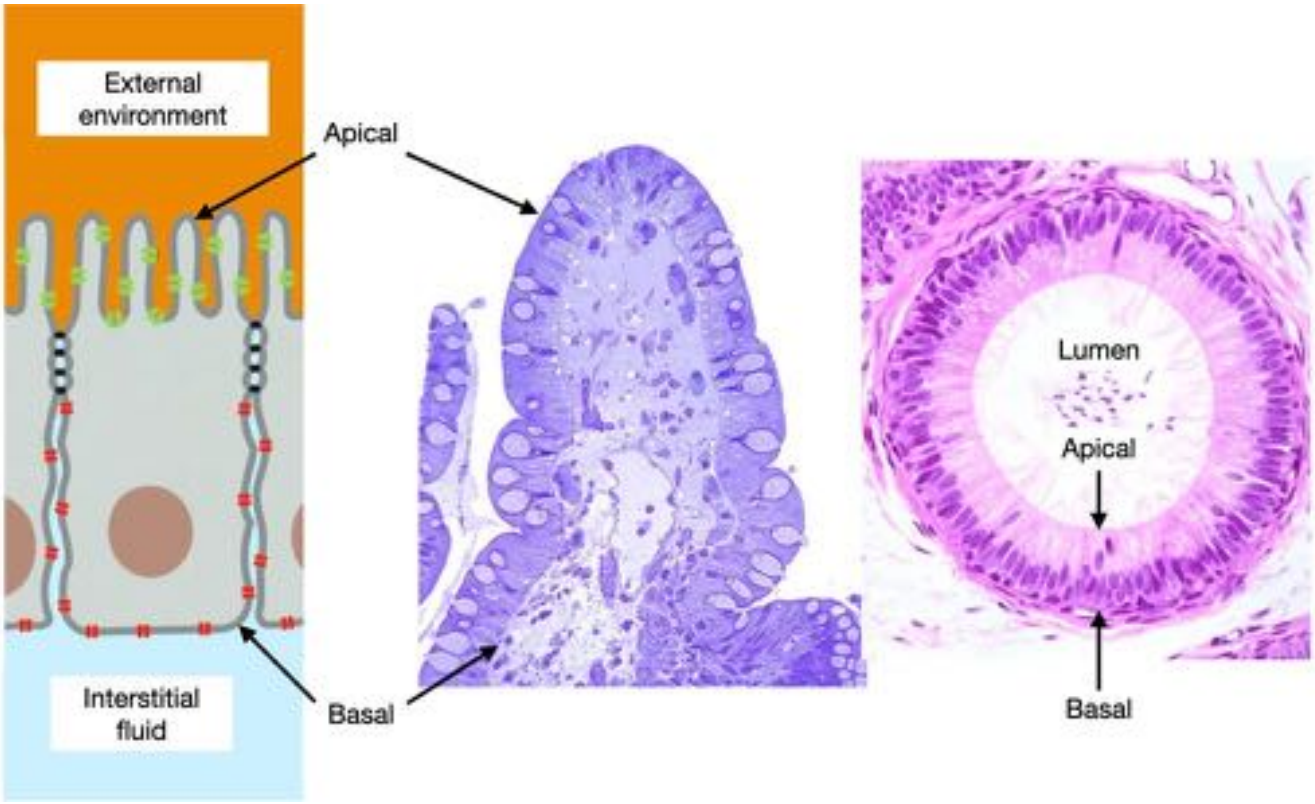
Epithelia are classified based on the shape of its cells and the number of layers of cells.

Some epithelia don't fit into the above classification scheme and so have different names that reflect their structure. Pseudostratified epithelia appear to have multiple layers of cells because the nuclei of the cells are at different levels giving the epithelium a layered appearance, but there is only one layer of cells. Another unique epithelium is transitional. Here, the epithelium can adopt different appearances depending on the state of the organ. Transitional epithelium lines the lumen of the urinary bladder which often has to store a large amount of urine. Transitional epithelium contains several layers of cells. When the bladder is filled the epithelium stretches and the number of layers of cells is reduced to accommodate a greater surface area. When bladder empties, the epithelium contracts and the layers of cells increase.

Epithelial Polarity

Because epithelium face two difference environments, the cells in most epithelia are polarized. That is the cell membrane on the side of the cell that faces the external

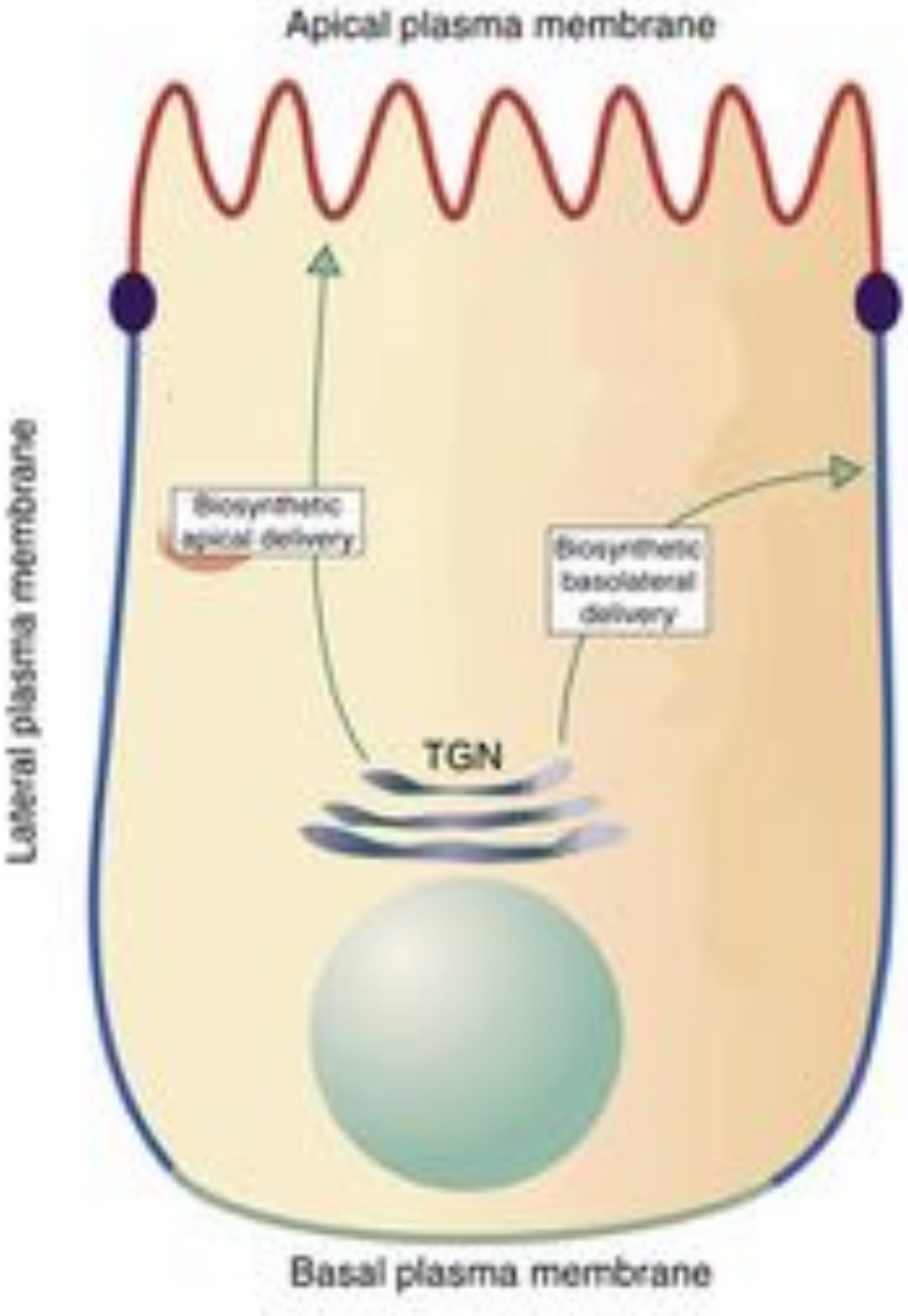
environment has a different biochemical composition that the cell membrane that faces the body. The side of the cell that faces the external environment or lumen of a tube is called apical and the side that faces the rest of the body is called basolateral (see below for why the lateral surface is included with the basal).



Beside different biochemical compositions, the apical and basolateral surface can generate different structures. Most of these are found on the apical surface. For example, some epithelial cells contain cilia on their apical surface. A cilium is a protrusion of the cell membrane that is filled with microtubules and powered by dynein motors to generate a wave-like movement in the cilium. Cilia allow epithelia cells to move material along their apical surface. Some epithelia have smaller protrusions of their apical cell membranes called microvilli. Microvilli are shorter the cilia and supported structurally by actin filaments. The main function of microvilli is to amplify the surface area of the apical surface and allow for greater rates of absorption or secretion.

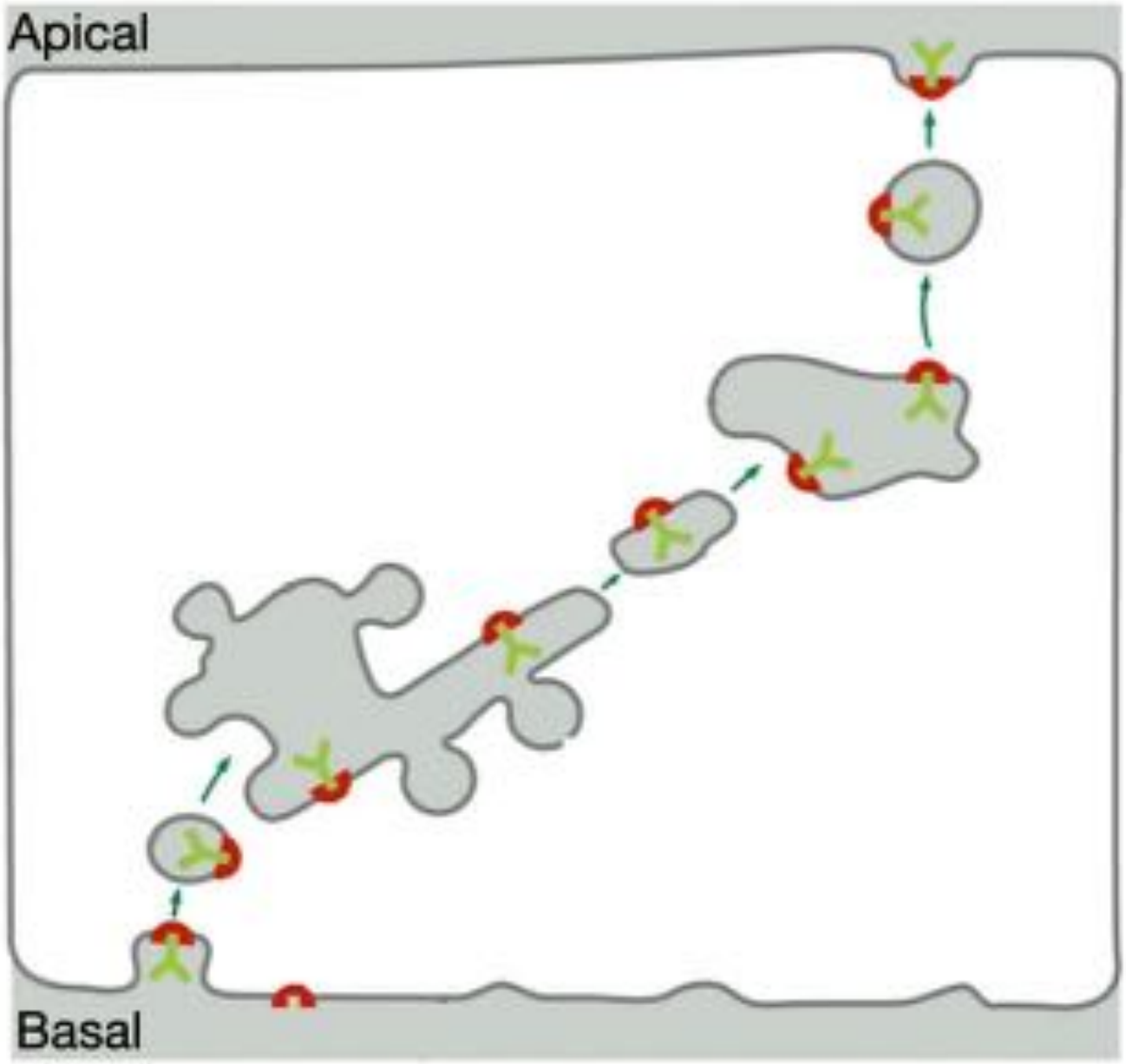
Because the biochemical compositions and functions of the apical and basolateral surfaces of epithelial cells differ, each needs a unique set of proteins. Epithelial cells need a way to sort apical and basolateral proteins in the secretory pathway and deliver them to the correct surface.

Proteins that are destined for apical or basal surface have signal sequences. These proteins are sorted in the trans-Golgi network and then delivered via vesicles to the apical or basal cell membrane. The signal sequence to deliver a protein to the basal surface is often found in the cytoplasmic tail of the protein. The signal sequence for apical proteins is less clear.



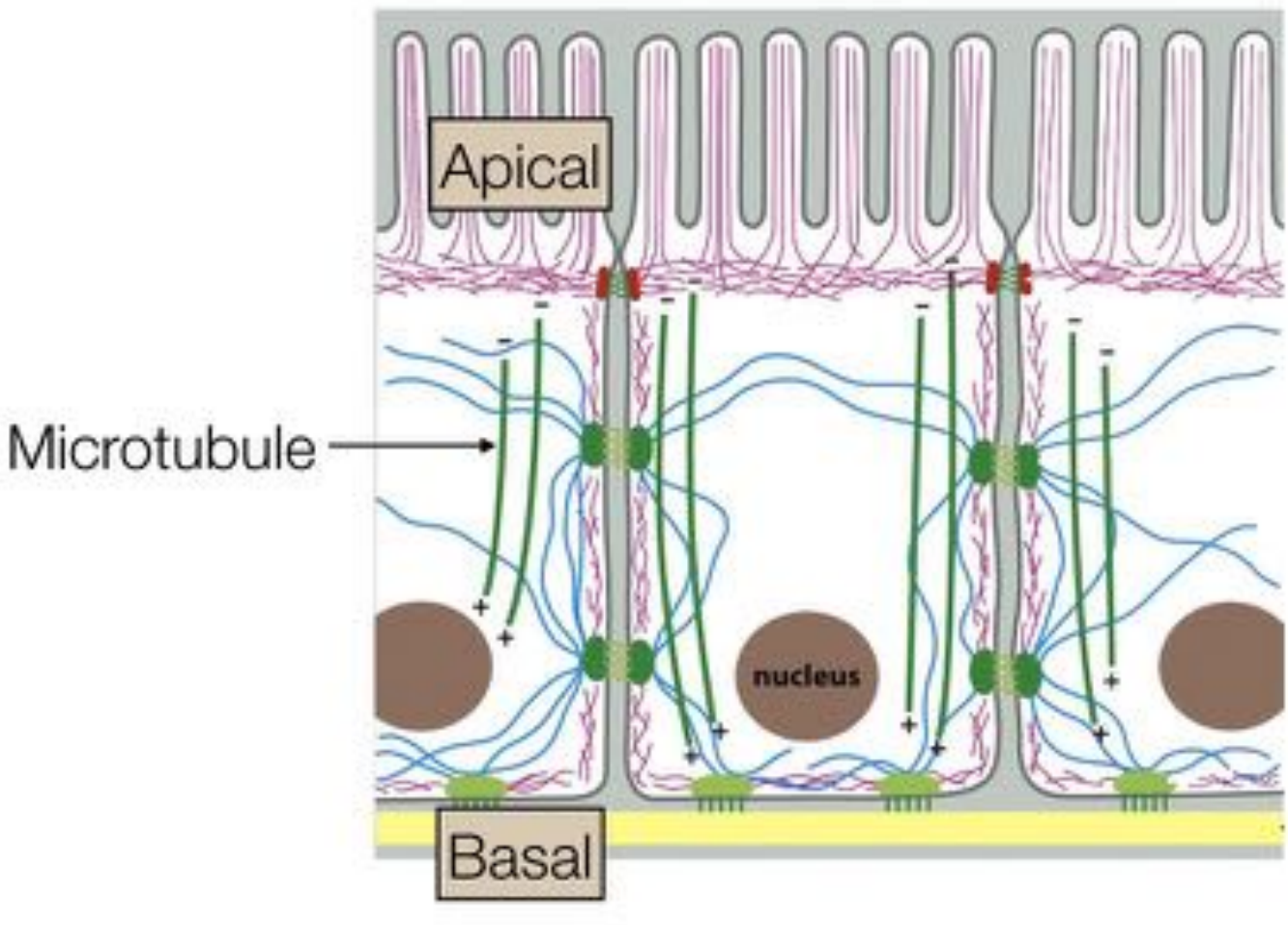
Proteins are sorted in the trans-Golgi network and delivered to apical or basolateral cell membrane.

Epithelial cells can also move protein and lipid from one side of the cell to the other by transcytosis. Transcytosis consist of endocytosis from on side of the cell, transport of the vesicles to the other side of the cell and then fusion of the vesicle with the cell membrane. Cells use transcytosis to transfer extracellular proteins from one side to the other. An example is the delivery of antibodies into the lumen of the intestine. B-cells, which produce antibodies, reside under the basal surface of the intestinal epithelial cells. The epithelial cells have a receptor on their basal surface that binds antibody. The receptor-antibody complexes are delivered the apical surface via transcytosis.



Transcytosis mediates transfer of protein across epithelial cells.

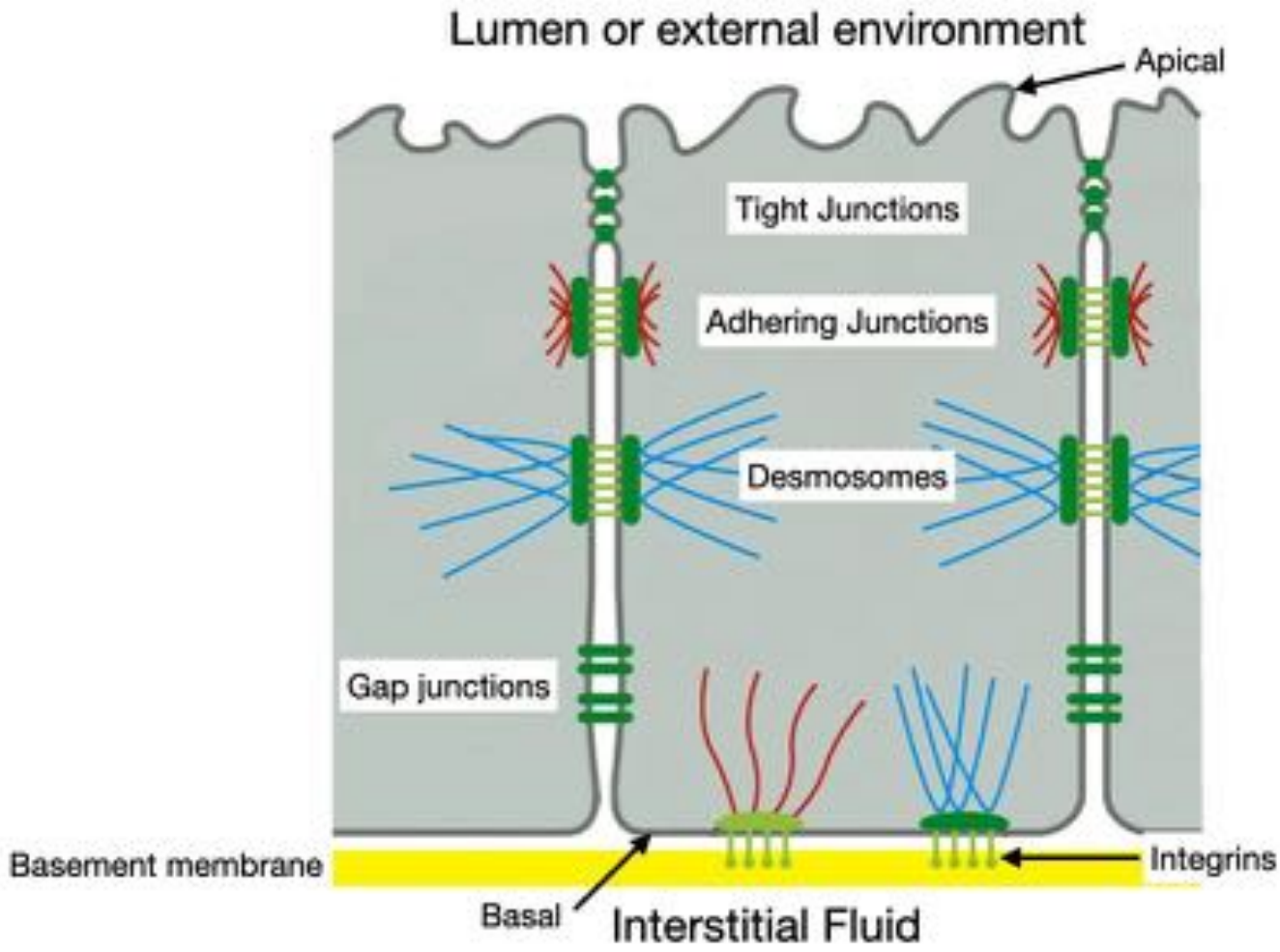
The cellular structures that create polarity in epithelia are still being identified. Microtubules appear to play a role as in many epithelial cells the minus ends of microtubules are found at apical side of the cell and their plus ends at basal side. This allows epithelial cells to use kinesins or dyneins to deliver material to their basal or apical side. Tight junctions (see below) are also important for maintaining the polarity of the cell membrane.



Minus ends of microtubules face the apical surface.

Adhesion

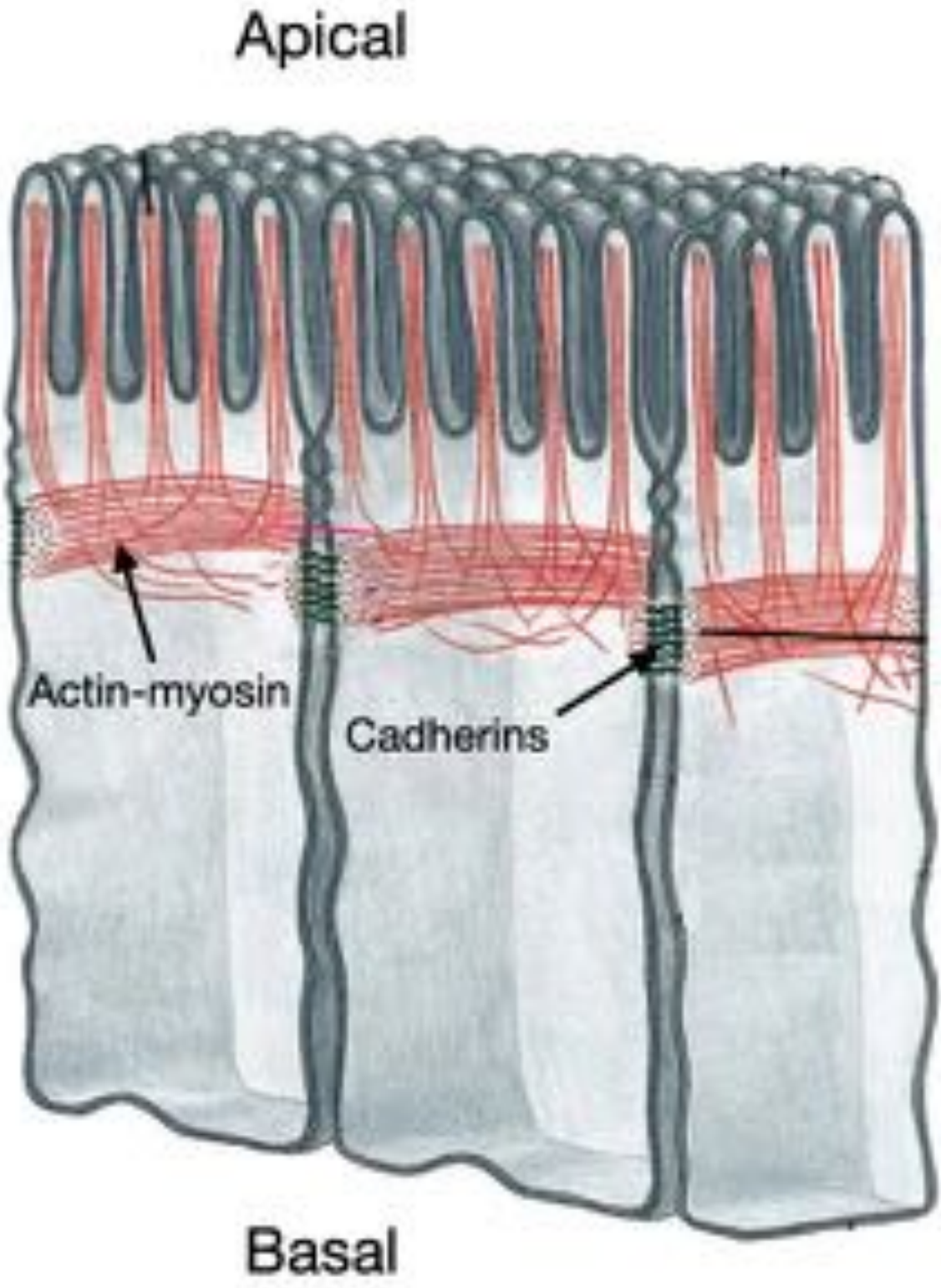
To maintain its structural integrity, epithelia depend upon adhesion between adjacent cells. Epithelial cells are held together by two junctional complexes with some epithelium using a third complex. All epithelia will have adhering junctions and tight junctions but only those epithelia in organs that face strong mechanical forces (e.g. skin) will have desmosomes. Desmosomes were described in the reading for Cells to Tissues so please refer to that reading for information about desmosomes.



Epithelial cells are held together by several junctional complexes.

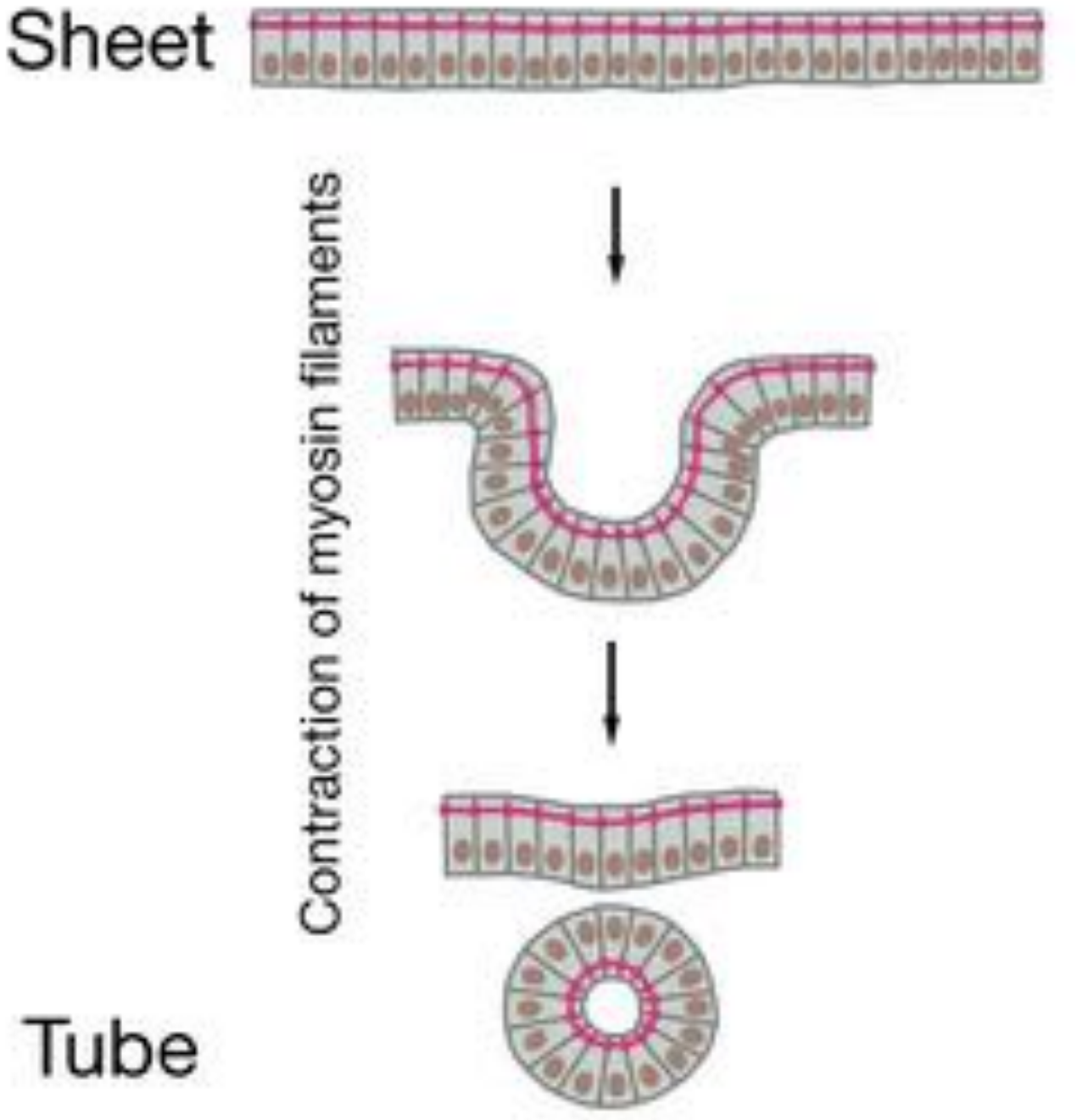
Adhering Junctions

The structure and molecular components of adhering junction was discussed in the reading on Cells to Tissues. Adhering junctions in epithelia are similar to cells in other tissues but have a characteristic arrangement and relationship with the internal cytoskeleton. Adhering junctions form a continuous belt-like structure around the circumference of epithelial cells. The adhering junctions are linked to bundles of actin filaments that also wrap around the cell. Myosin filaments can pull on the actin filaments to contract the cell.



Adhering junctions form a belt-like zone around epithelial cells.

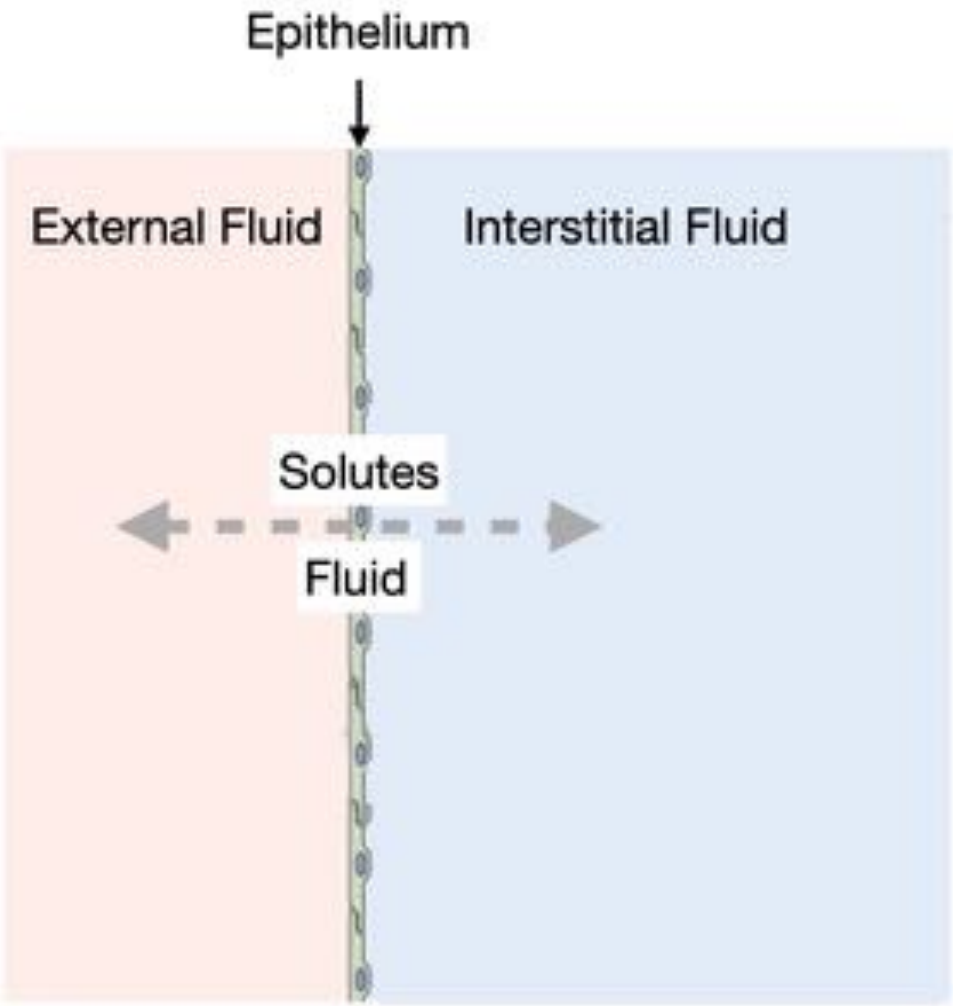
In cuboidal and columnar cells, the adhering junctions are often located closer to the apical surface, so that contraction causes the apical side of the cell to shrink. During development, this allows a sheet of epithelia cells to form a tube such as the gastrointestinal tract.



Contraction of myosin-actin filaments changes cells shape to generate different structures.

Epithelial Transport and Permeability

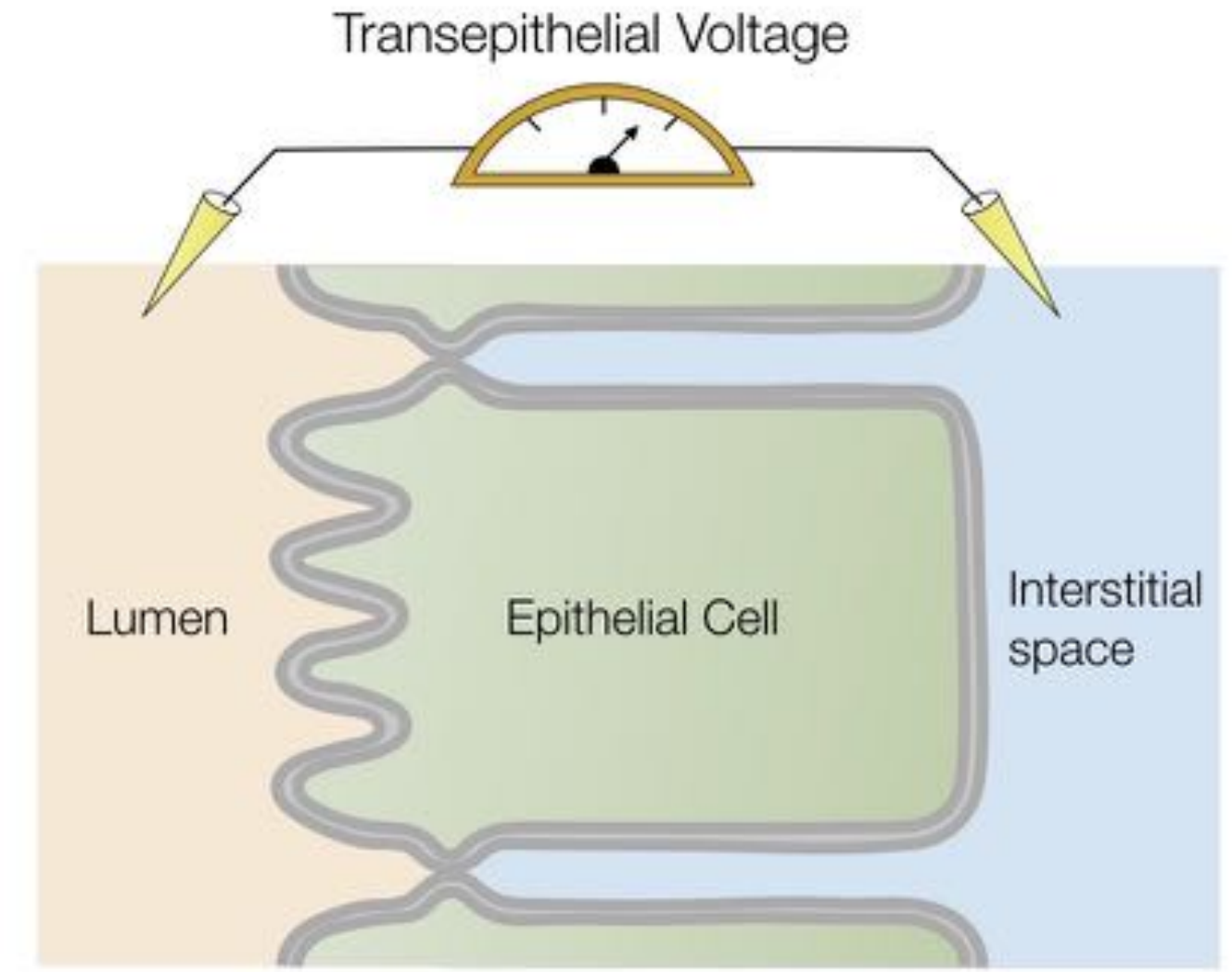
One of the most critical functions of some epithelia is to regulate the passage of solutes between two fluid compartments. For example, the epithelium in the intestine needs absorb nutrients form the lumen of the intestine and release them into the interstitial fluid. Likewise, the epithelium in the urinary system controls the amount of water and ions in urine by regulating the passage of these components between urine and interstitial fluid.



Epithelia control the passage of solutes and fluids between different fluid compartments.

Epithelia are often classified based on their leakiness or how easily solutes can pass across the epithelium. To determine the permeability of leakiness of an epithelium its electrical resistance can be measured with electrodes. An epithelium from an organ is isolated and electrodes are inserted into the lumen and interstitial fluid and connected to a voltmeter. A current is passed across the epithelium and the change in voltage determines the resistance

of the epithelium. Epithelia with higher resistance are considered restrictive whereas those with low resistance are considered leaky.

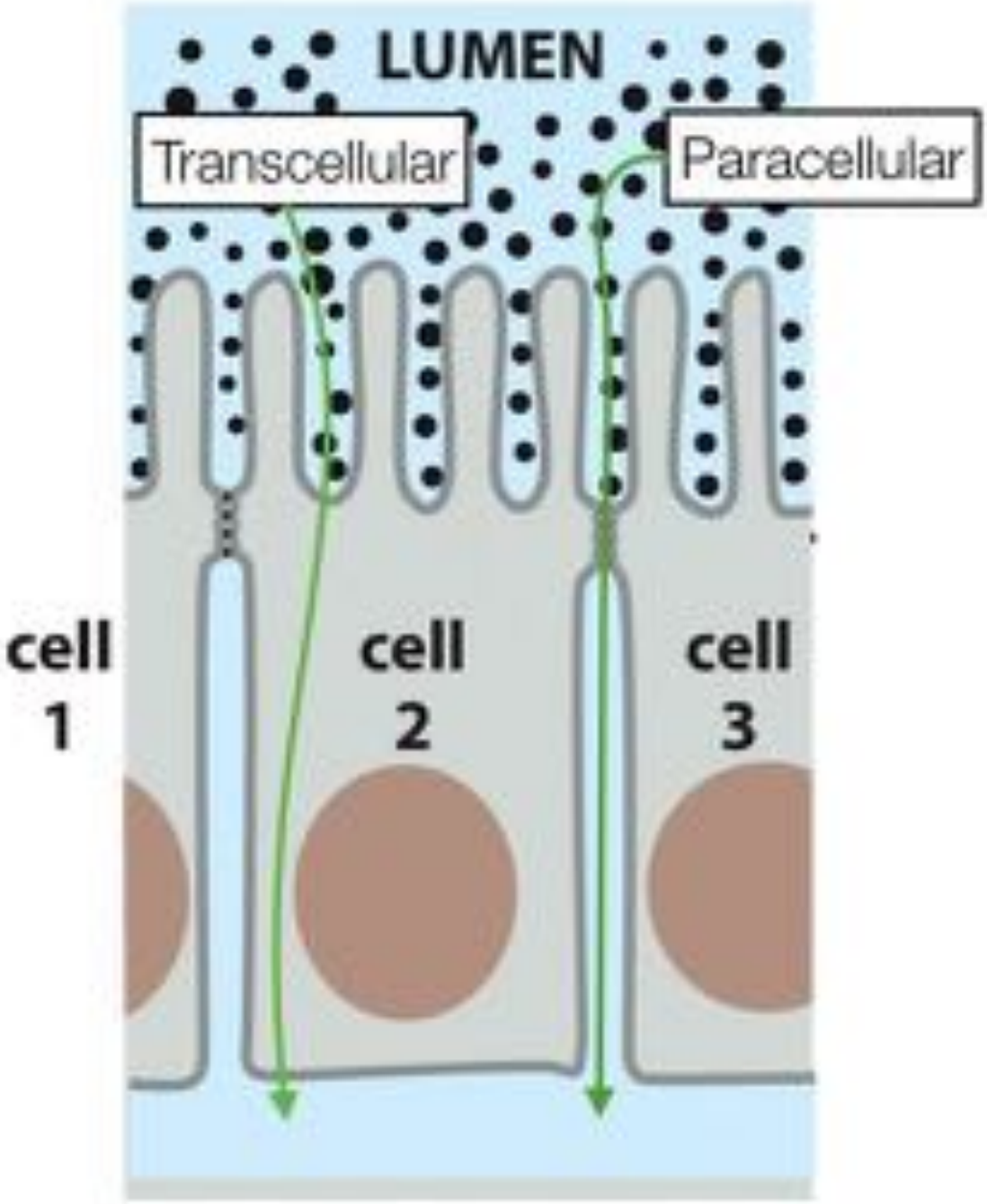


Voltage across an epithelium can be measured to determine the resistance of the epithelium.

Resistance of epithelia ranges from about 6 ohms in the small intestine to around 70,000 ohms in the bladder. Thus, epithelia in the bladder is 10,000-fold more restrictive to the passage of ions than the epithelia in small intestine.

There are two pathways for solutes and water to cross an epithelium: through the cells (transcellular) or between the cells (paracellular). The transcellular pathway requires a set of channels and pumps in the apical and basolateral membranes to move solutes across an epithelial cell. The paracellular route is restricted by a set of proteins that form tight

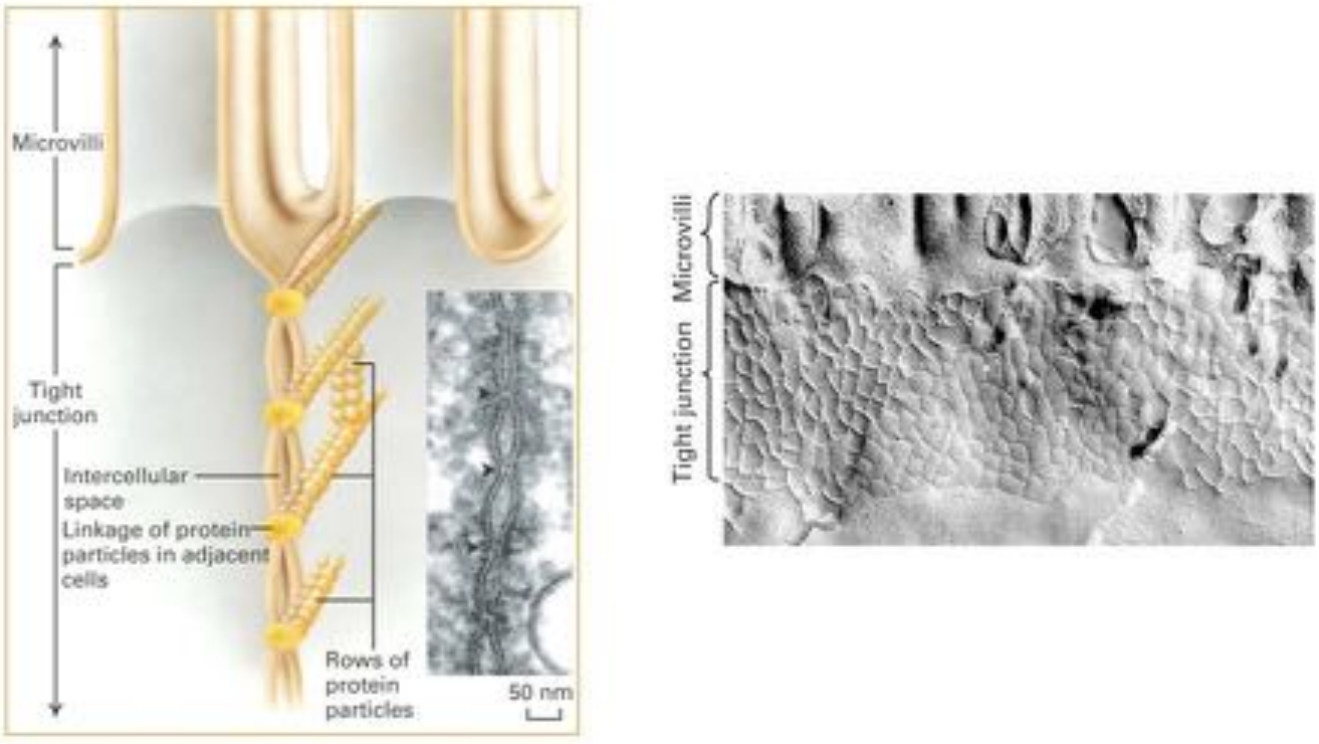
junctions. Depending on the type proteins in the tight junction, the paracellular pathway can be more or less permeable.



Transcellular and paracellular pathways allow passage of solutes and fluid.

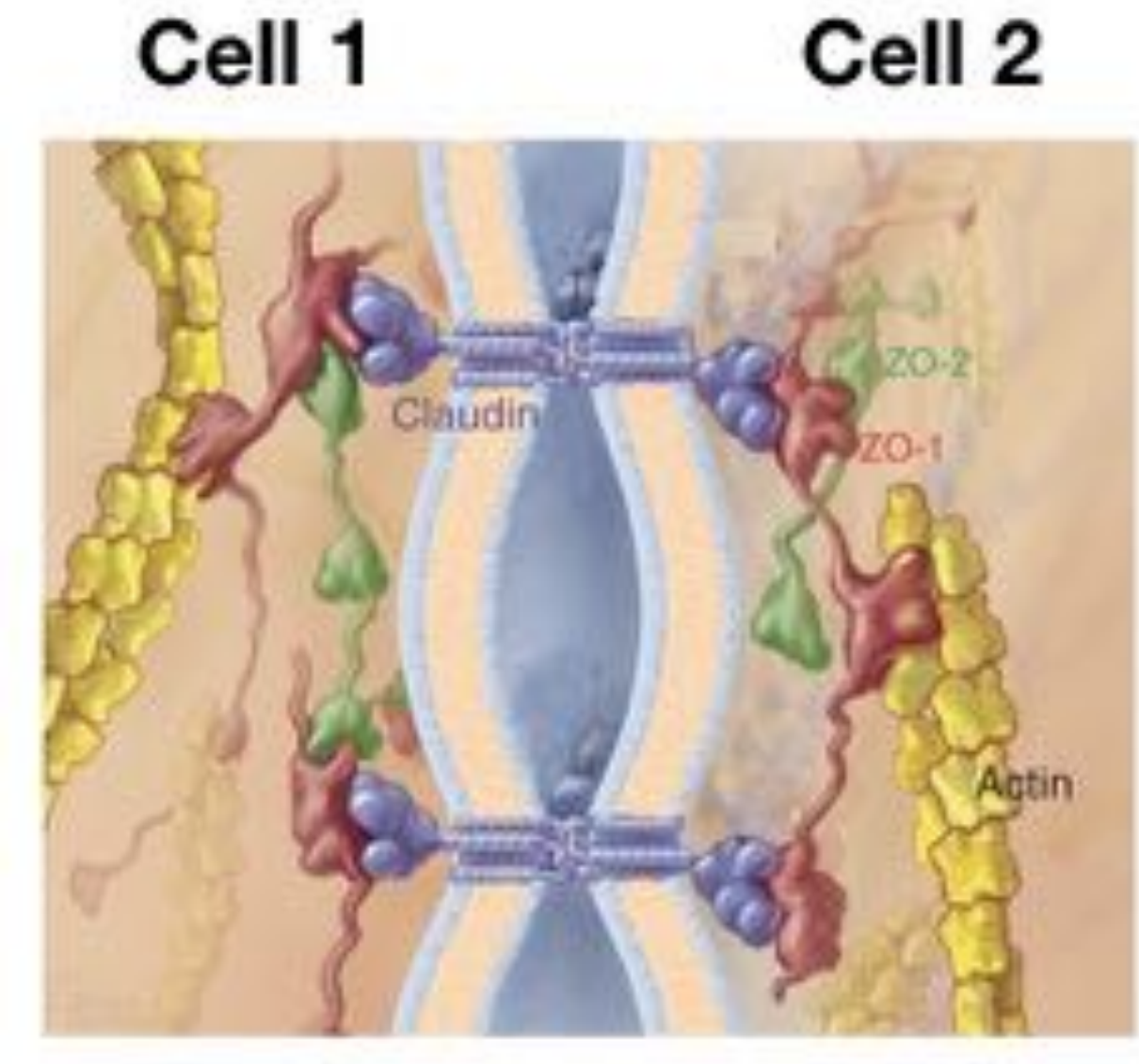
Tight Junctions

Tight junctions are similar to adhering junctions in that they encircle the entire circumference of cell. Electron micrographs of adjacent epithelial cells show how tight junctions bring the cell membranes of the two cells close together. Note how tight junction are composed of several strands of interactions between adjacent cells. Solutes that want to diffuse between epithelial cells must pass through this network of intercellular connections.



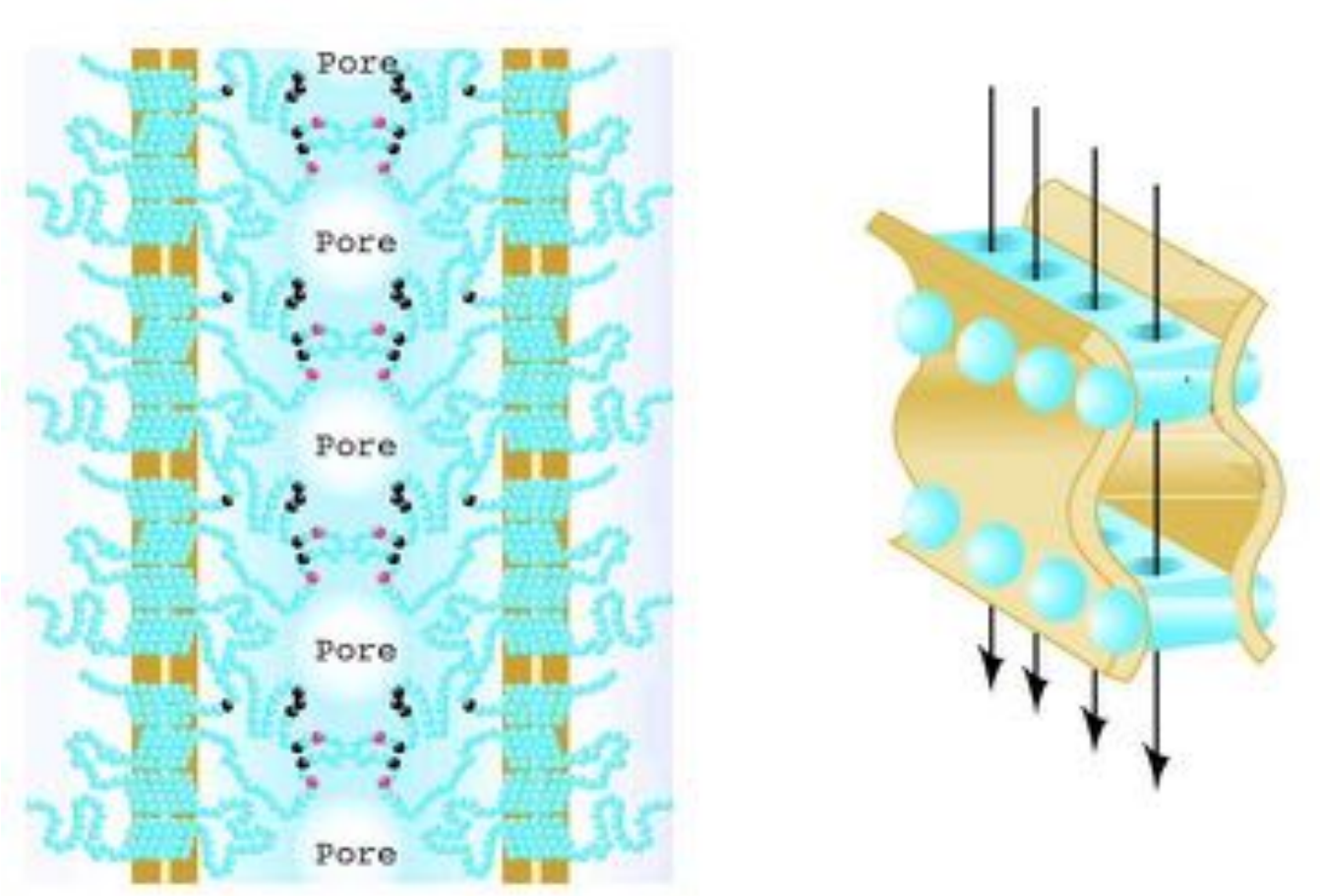
Tight junctions form a network of sealing strands that encircle epithelial cells.

Tight junctions contain more than 50 different types of proteins, but claudins are the proteins that primarily determine the permeability of the tight junction. Claudins contain four transmembrane domains and interact with claudins in adjacent cells. The intracellular domains of claudins are linked indirectly to actin filaments by a set of proteins called ZO proteins. The interaction with actin filaments help stabilize the claudins in the cell membrane.



Claudins are the primary component of tight junctions that determine permeability.

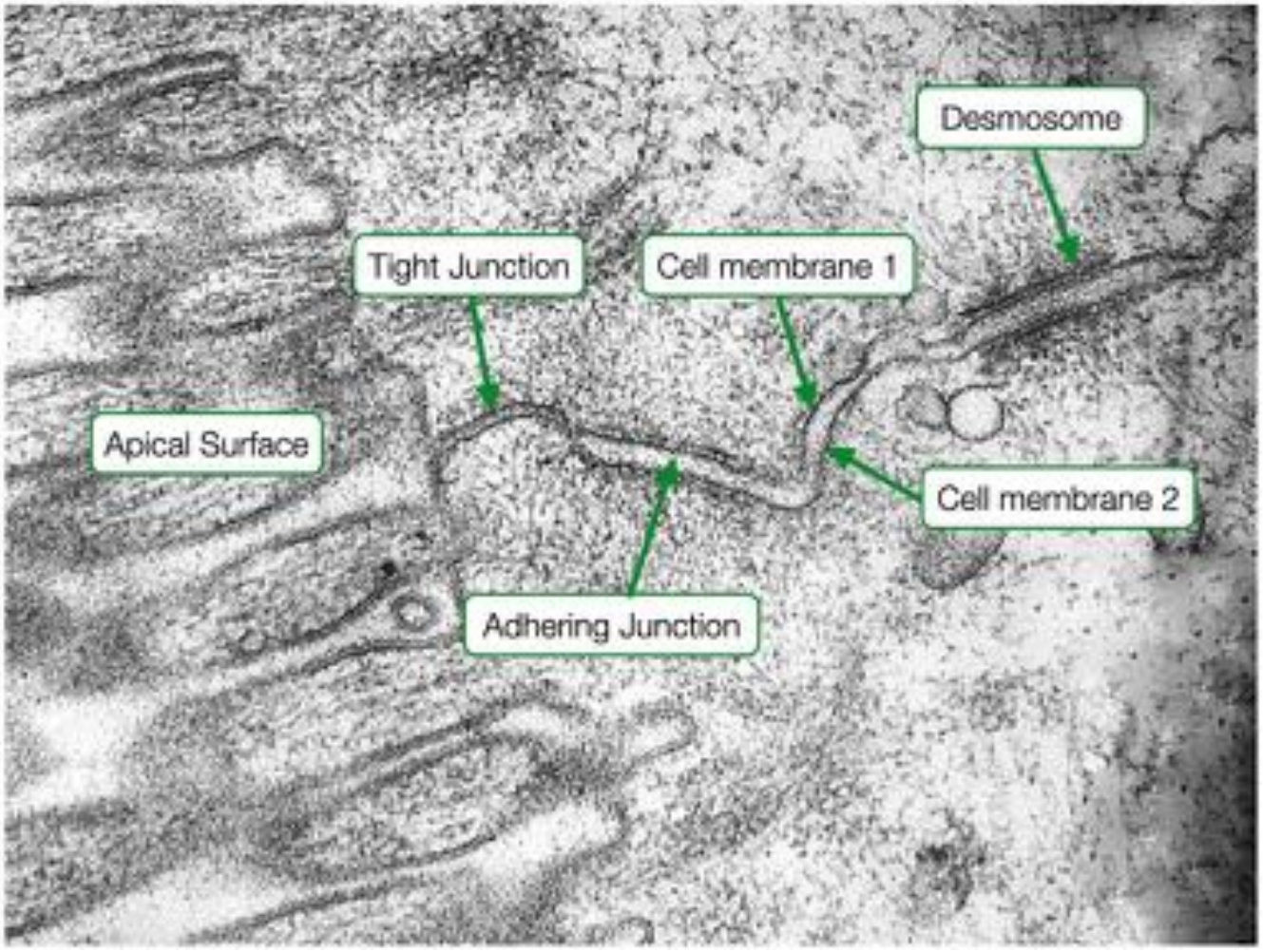
The interaction between claudins in adjacent cells creates pores that allow passage of ions and small molecules. To create epithelia with different permeabilities, epithelial cells can select from 24 different claudin genes to express. The proteins encoded by these genes have different permeability characteristics, including restricting passage of specific ions, that allow epithelia to be more or less permeable. Cells can also increase or decrease their permeability by the number of tight junction strands that encircle the cell membrane.



Interactions between claudins generates size restrictive pores.

Arrangement of Junctional Complexes

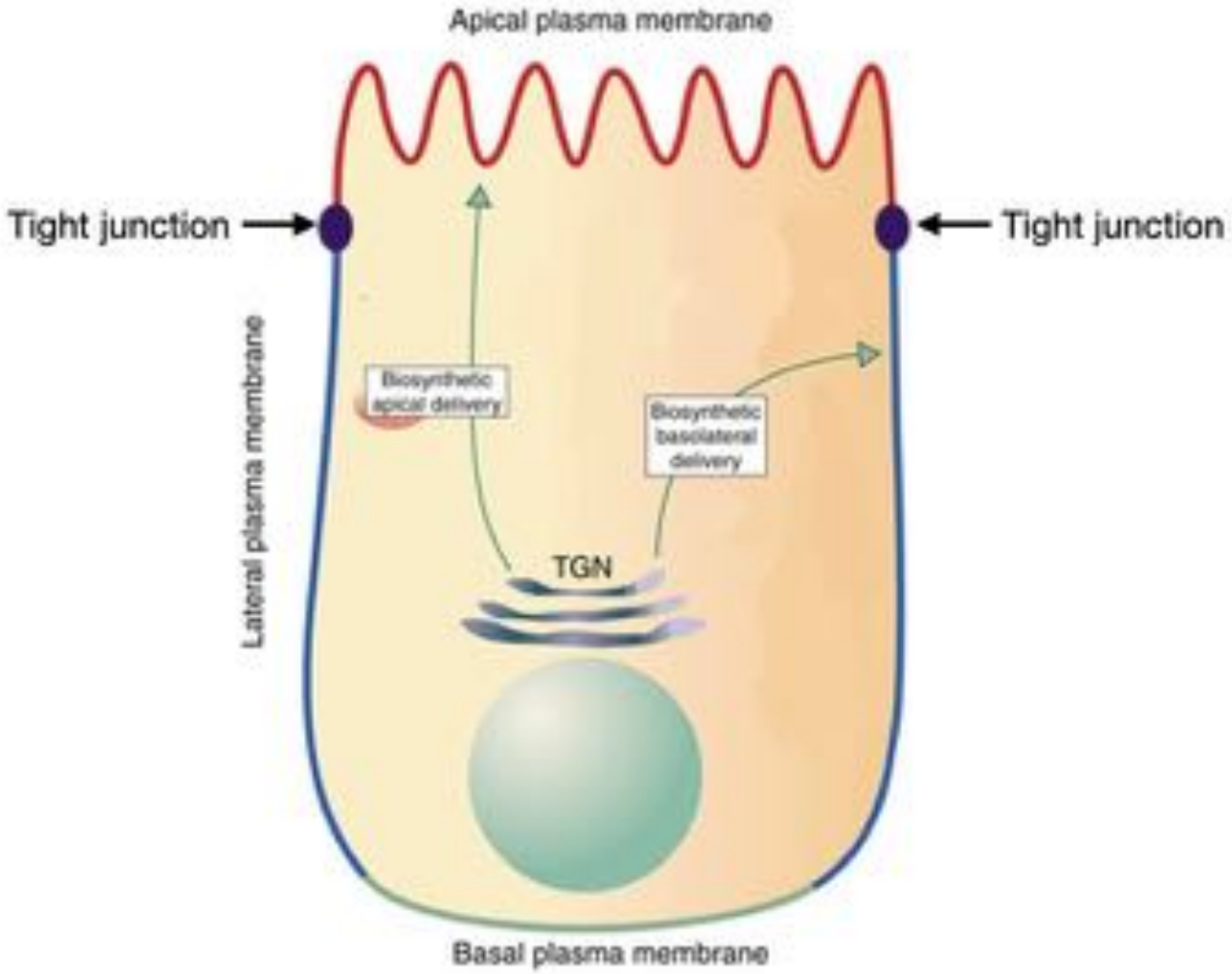
The junctional complexes between epithelial cells can be seen in electron micrographs. The tight junctions are closest to the apical surface, followed by the adhering junctions and desmosomes, if present. Note the relative locations of each junction and the distance between the cell membranes of the two adjacent cells.



Tight junctions are located close to the apical surface.

Tight Junctions and Cell Polarity

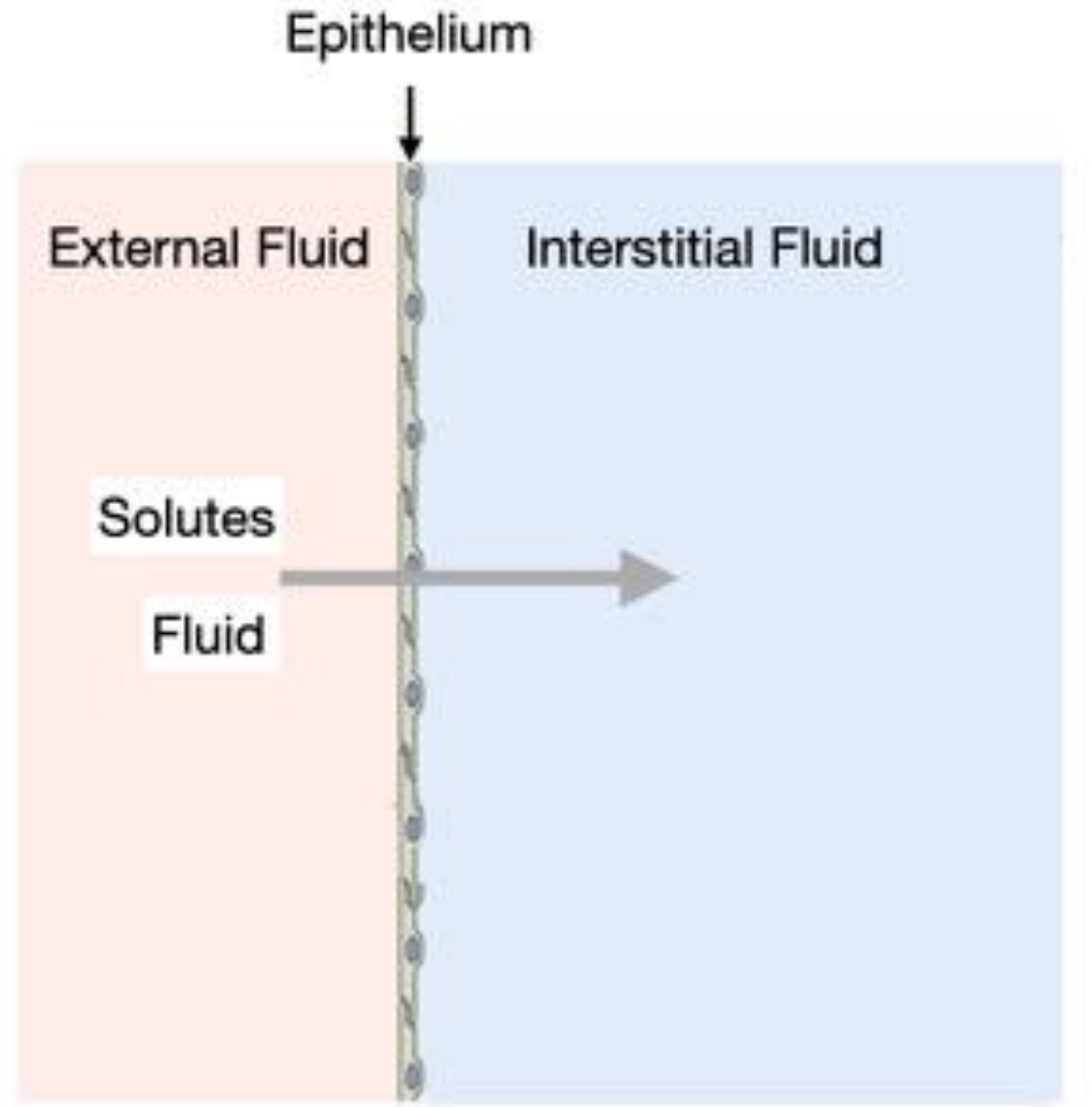
Tight junctions also play an important role in cell polarity. Recall that the secretory pathway in epithelial cells can deliver specific proteins to the apical or basolateral surface, but the cell membrane is one continuous lipid bilayer. Lateral diffusion within the bilayer would soon mix apical and basolateral proteins. To prevent his mixing due to diffusion, tight junctions create a diffusion barrier in the cell membrane that inhibits the diffusion of membrane proteins. Thus, tight junctions define the border between apical and basolateral. Because tight junctions are located very close to the apical surface, the basal surface (base of the cell) and lateral surfaces have a similar composition and is the reason we refer to the surface opposite to apical as basolateral.



Tight junctions restrict the diffusion of proteins in the cell membrane to maintain cell polarity.

Transport Across Epithelia

Epithelia must often allow specific ions or molecule to pass from one fluid compartment to another in a process called vectorial transport. The transcellular pathway can generate transport of specific solutes as epithelial cells can express channels that allow diffusion of specific ions or molecules across their cell membranes. Importantly, these channels need to be localized to either the apical or basolateral cell membrane to allow vectorial movement of ions or molecules.



Vectorial transport is the movement of specific solutes or water from one compartment to another.

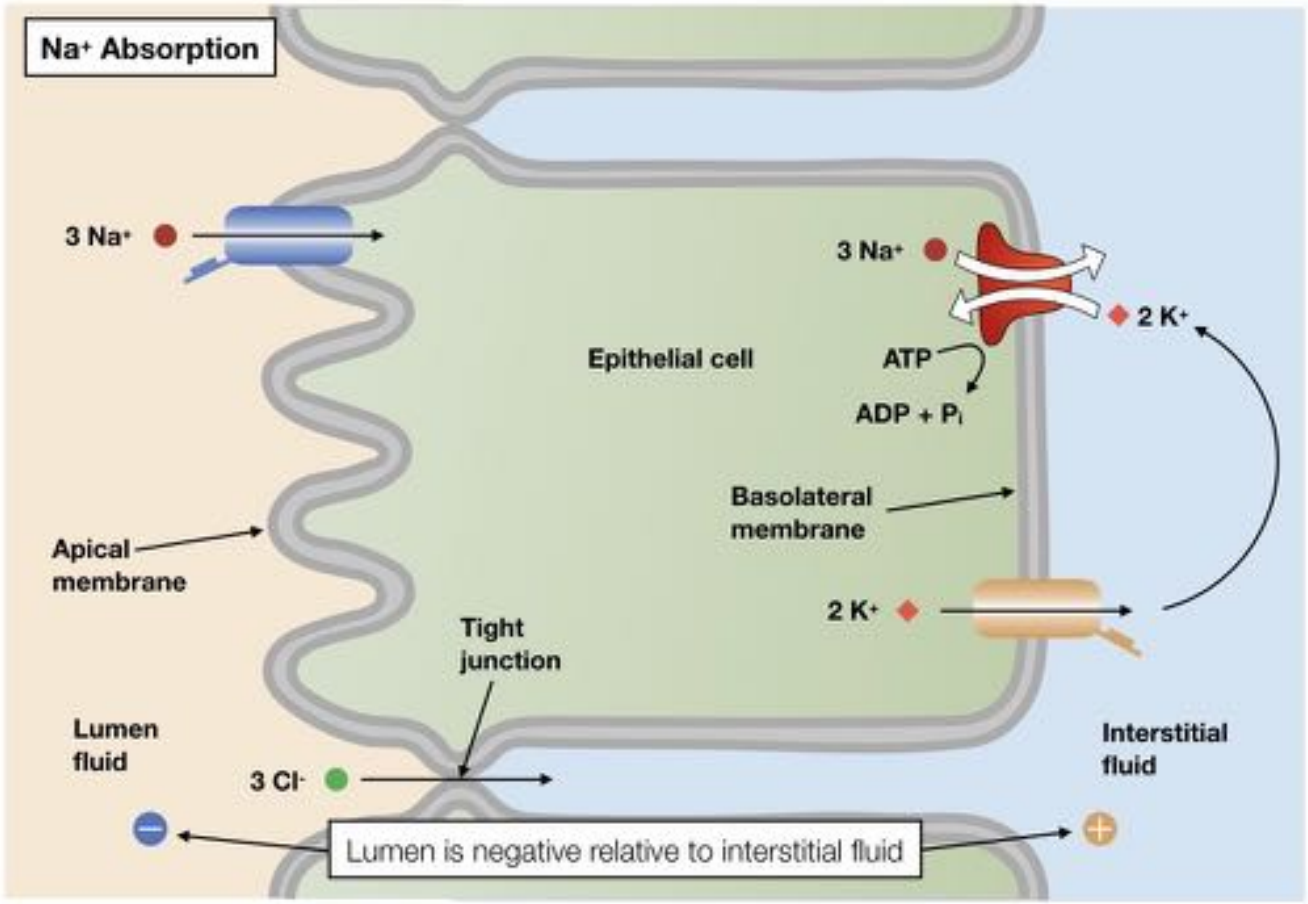
Another determinant of directional transport across an epithelium is its transepithelial voltage. If the voltage in the lumen is more negative relative to interstitial fluid, it can draw positive ions into the lumen and push negative ions into interstitial fluid. A positive voltage in the lumen relative to interstitial fluid would draw negative ions into the lumen and push

positive ions into the interstitial fluid. As described above, we can use electrodes to measure the transepithelial voltage.

Like most cells, epithelial cells express the sodium-potassium pump that extrudes from the cytoplasm 3 sodium atoms for every 2 potassium atoms in takes in. In almost every epithelium, the Na-K pump is localized to the basolateral surface. Below we describe the methods epithelia use to move certain ions and small molecules. For the purposes of the discussion, let's imagine that we have an epithelium that lines inner surface of a tube (e.g. gastrointestinal tract or a tube in the urinary tract) and we want either to absorb solutes from the fluid in the lumen of the tube into the interstitial fluid or we want to secrete solutes from the interstitial fluid into the fluid in the lumen of the tube. Remember that the apical surface of the epithelial cells faces the fluid in the lumen whereas the basolateral surface faces the interstitial fluid.

Absorption of Sodium

Some cells absorb sodium from a fluid in the lumen of a tube. The Na-K pump creates an electrochemical gradient for sodium across the apical membrane by pumping sodium out of the cell along the basolateral cell membrane. This gradient provides the driving force for sodium to diffuse from the fluid in the lumen across the apical cell membrane into the cytoplasm. To facilitate the diffusion of sodium into the cell, epithelial cells express a sodium channel (epithelial sodium channel or ENaC) in their apical cell membrane. The sodium that enters the cells is then pumped into the interstitial fluid by the Na-K pump. The potassium that is pumped into the cell by the Na-K pump can diffuse back into the interstitial fluid through a potassium channel localized to the basolateral surface.



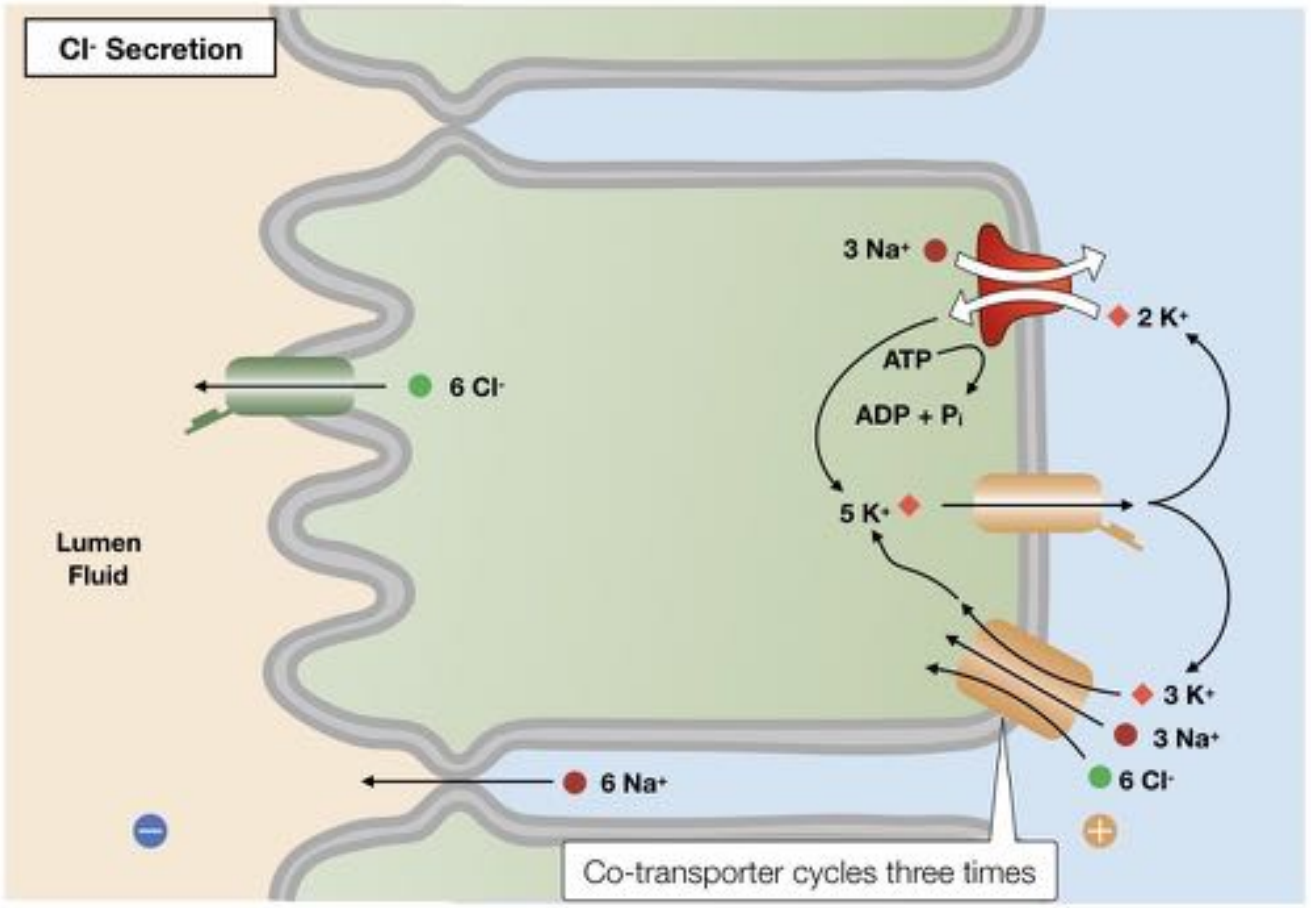
Sodium chloride absorption is mediated by sodium channels in the apical cell membrane.

The movement of sodium from apical side of the epithelium into the interstitial fluid generates a slight potential across the epithelium. The fluid in the lumen becomes slightly electronegative (- 3 mV) relative to the interstitial fluid. The electronegativity provides a driving force for chloride from the lumen into the interstitial fluid. Chloride passes through tight junctions, which must contain a claudin that is permeable to chloride, to get from the lumen to the interstitial fluid.

Sodium Chloride Secretion

Epithelia can also move sodium chloride from interstitial fluid into the fluid on their apical surface. The epithelial cells express a Na-K-Cl co-transporter in their basolateral membrane. The co-transporter allows passage of Na, K and Cl from interstitial fluid into the cytoplasm. The movement of K and Cl into the cell is driven by the strong electrochemical gradient for Na created by the Na-K pump. Chloride that enters the cell can diffuse into the apical fluid through chloride channels (e.g. CFTR) in the apical cell membrane. The net movement of

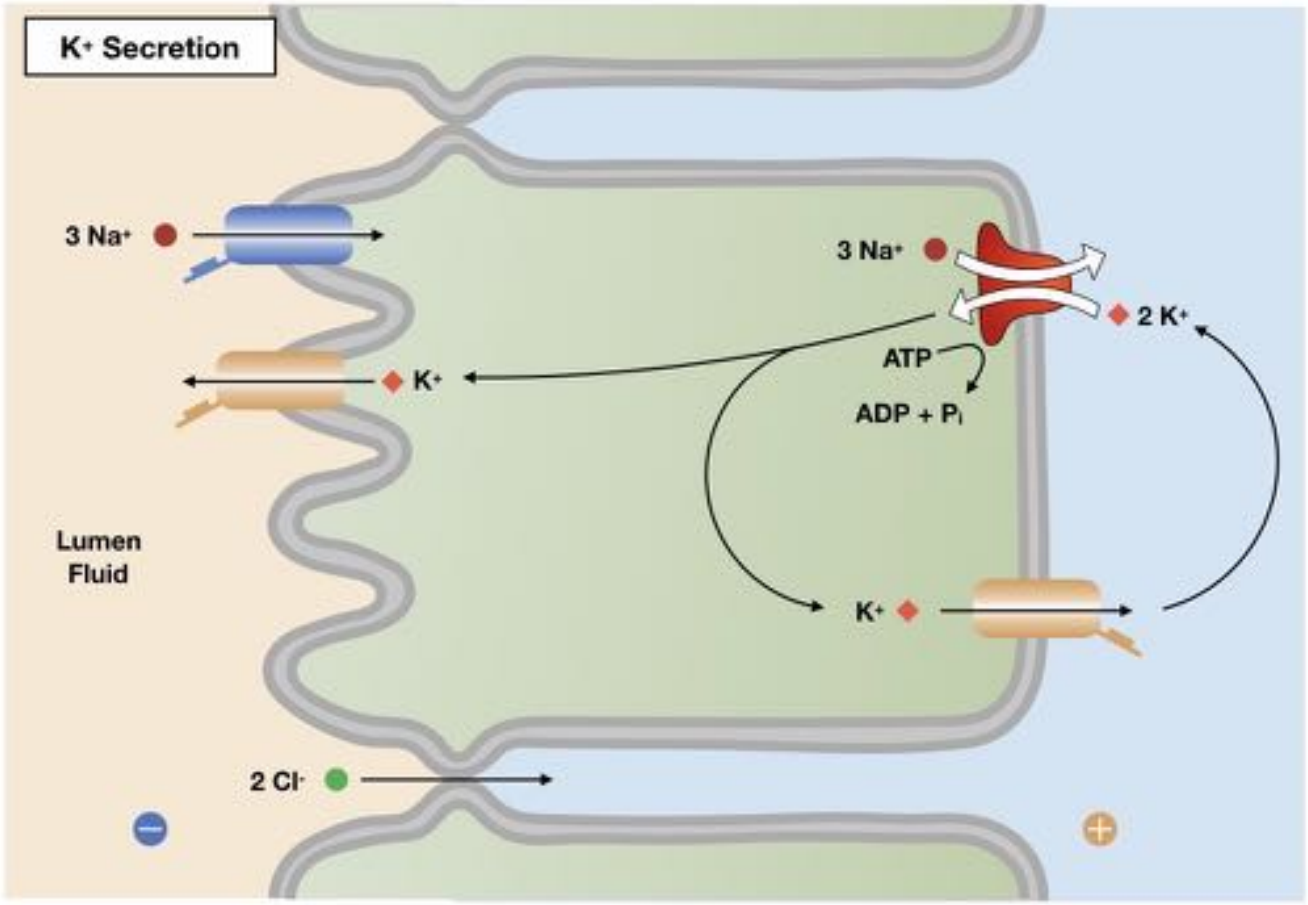
chloride from interstitial fluid to apical fluid makes the apical fluid electronegative relative to interstitial fluid. This draws sodium from interstitial fluid into the apical fluid with the sodium passing through the tight junctions.



Sodium chloride secretion is mediated by chloride channels in the apical cell membrane.

Potassium Secretion

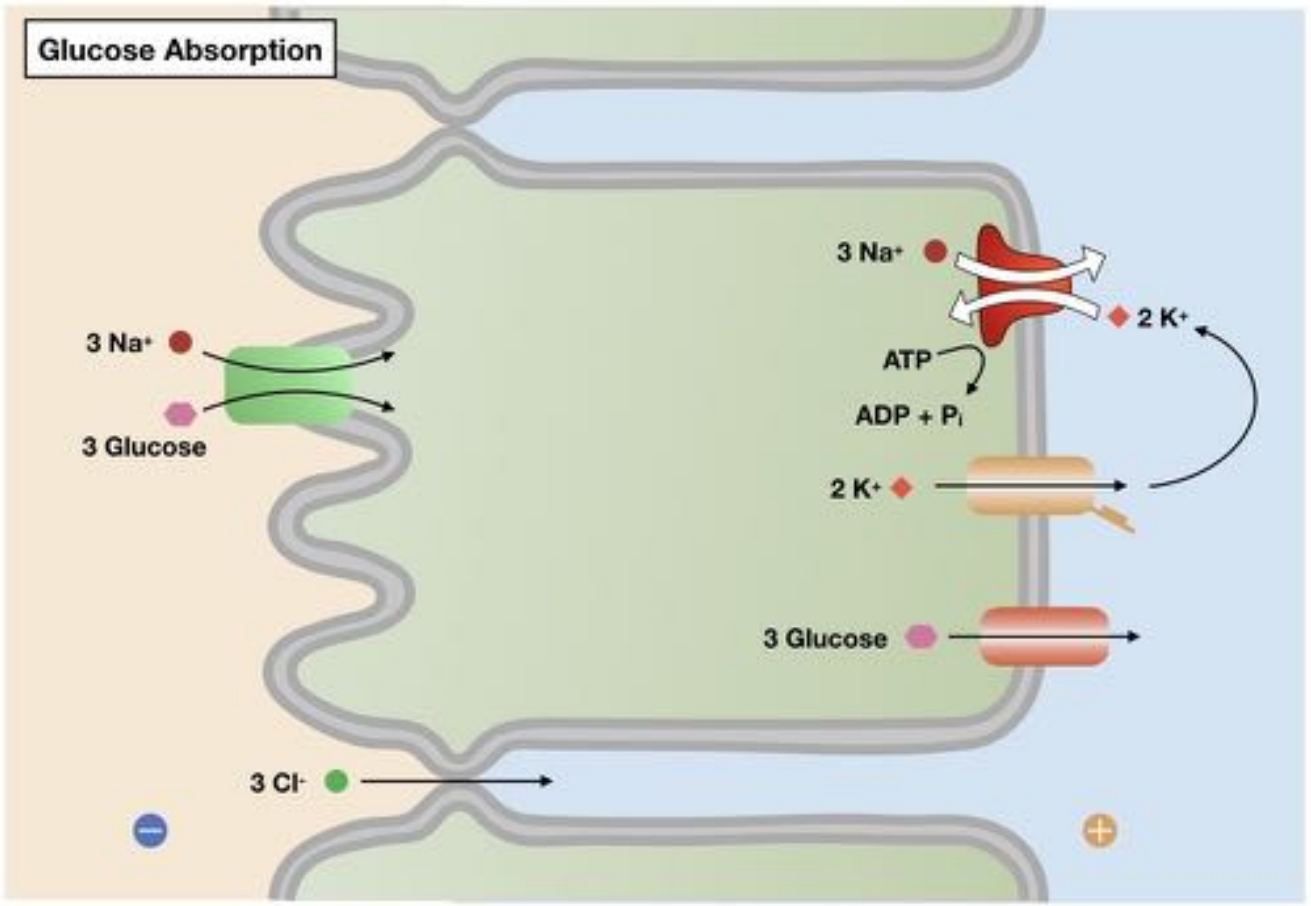
Potassium secretion from interstitial fluid into the lumen requires a slight modification to the model of sodium absorption. Cells localize potassium channels to the apical cell membrane so that potassium pumped into the cell by the Na-K pump can diffuse from the cytoplasm into the fluid of the lumen. Note that potassium secretion requires not only the Na-K pump on the basolateral surface but also ENaC on the apical cell membrane to maintain intracellular sodium concentration for the Na-K pump.



Potassium channels in the apical membrane mediate secretion of potassium.

Glucose Absorption

Besides ions, epithelia can also move small molecules from one fluid compartment to another. A prime example is the absorption of glucose in the small intestine. Here, the epithelial cells express Na-glucose co-transporter (SGLT) in their apical cell membrane. The Na-K pump in the basolateral membrane generates a strong Na electrochemical gradient into the cell that allows SGLT to move glucose against a concentration gradient. Glucose that enters the cell passively diffuses down its concentration gradient through a glucose channel in the basolateral membrane.



Water Movement Across Epithelia

Water follows the osmotic gradient across epithelia so an epithelium that secretes NaCl into the apical fluid will also secrete water and one the absorbs NaCl will also absorb water. Water can pass across an epithelium either by diffusion through the lipid bilayer, diffusion through water channels (aquaporins) or diffusion through certain types of tight junctions. The rate at which water flows across an epithelium via osmosis depends upon the number of aquaporins in the apical and basolateral membrane and the permeability of the tight junctions to water. The ability to regulate the rate of water flow allows an epithelium to absorb hyperosmotic and hypo-osmotic fluids.

Absorption of Hyperosmotic Fluid

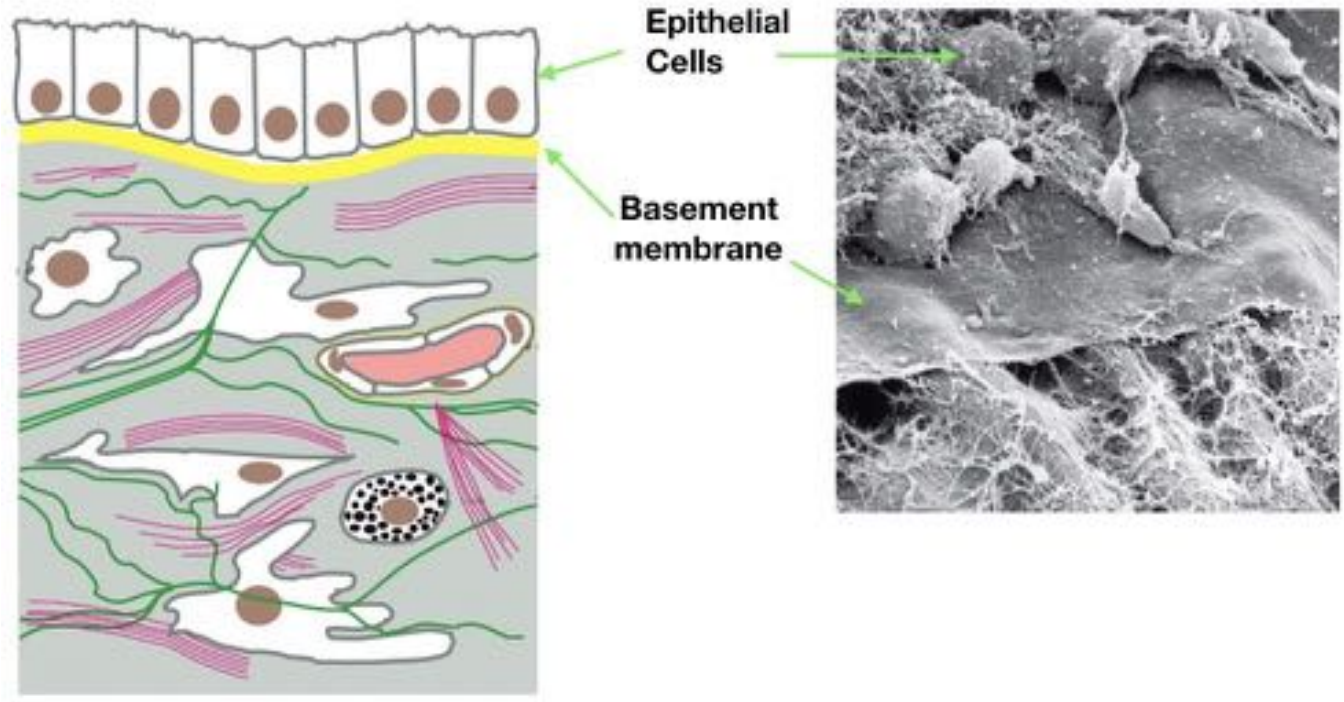
An epithelium can absorb a hyperosmotic fluid if the rate of salt absorption is greater than the rate of water absorption. An epithelium that is highly permeable to sodium and chloride but less permeable to water will create a hyperosmotic interstitial fluid and a dilute fluid in the lumen. This type of epithelium is used by the kidney to generate dilute urine.

Absorption of Hypo-osmotic Fluid

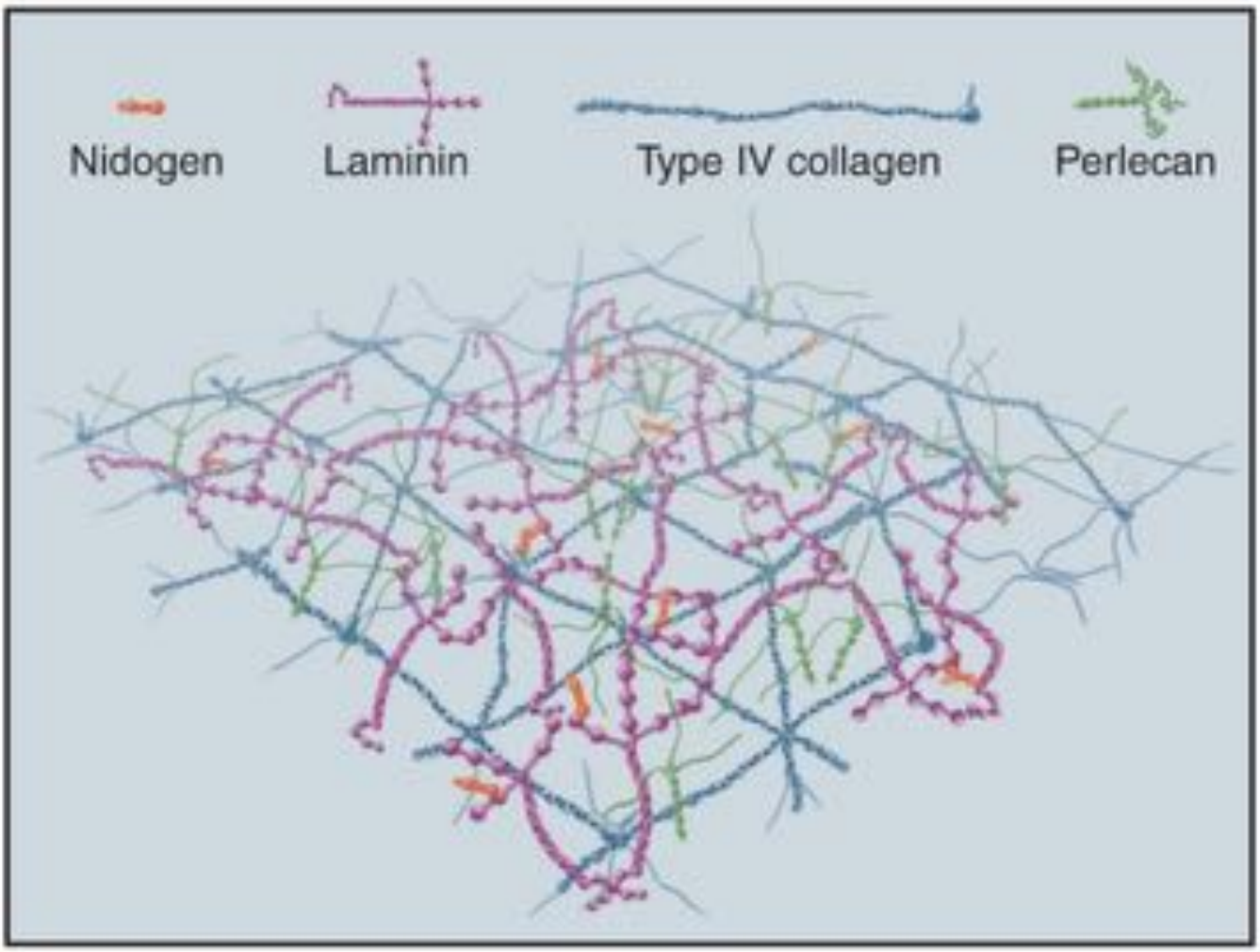
An epithelium can absorb a hypo-osmotic fluid if the osmolality of the interstitial fluid is higher than the fluid on apical side of the epithelium. This condition exists in certain regions of the urinary system. It also helps if the epithelium has a relatively high permeability to water by having a large number of aquaporin channels in its apical and basolateral membranes.

Basement Membrane

All epithelial cells are attached on their basal surface to a basement membrane. The basement membrane provides some mechanical support as it tethers together a sheet of epithelial cells. It also supports the growth and survival of the epithelia as it controls the access of epithelia to nutrients, ions, proteins and oxygen. Epithelia lack their own blood supply and rely on the capillaries in the underlying tissues. All the nutrients from the blood must cross the basement membrane to reach the epithelial cells. The basement membrane also regulates the growth and division of epithelial cells.

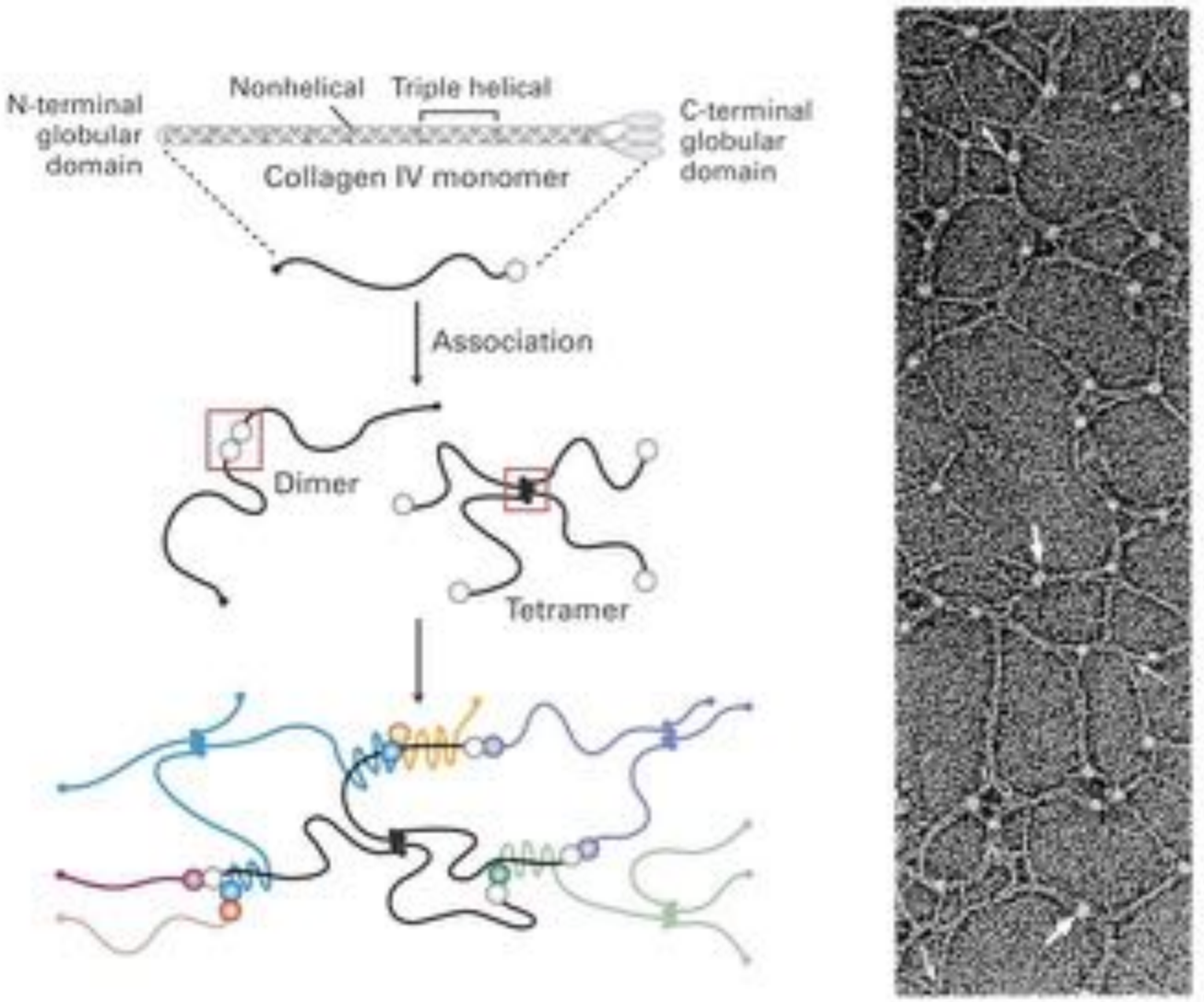


The basement membrane is composed of the interlocking networks of protein fibers: a type IV collagen network and an laminin network. The two protein networks are linked to each other by two proteins: nidogen and perlecan.



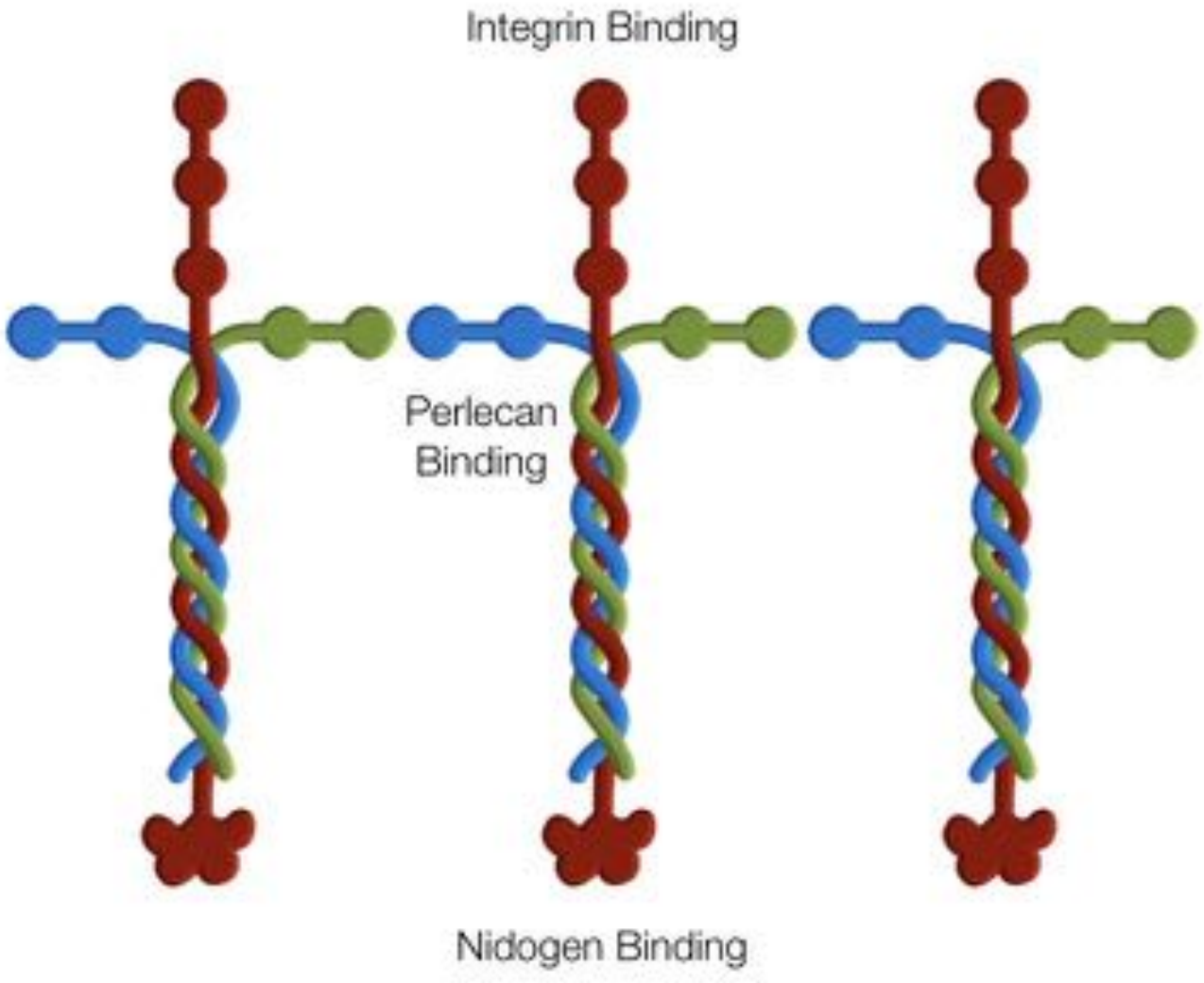
The basement membrane is a meshwork of interconnected fibers.

Type IV collagen is similar in structure to its fiber-forming family members in that it contains trimers that wrap around one another into a helical structure. However, these trimers do not assemble into parallel, crosslinked arrays to form fibrils. Instead, type IV collagen retains domains at its N and C-termini (remember that these are removed in fibril-forming collagens). These domains interact to assemble type IV collagen into branching networks as seen in this EM image of type IV collagen. These branching networks of collagen are what give the basal lamina it sheet-like structure.



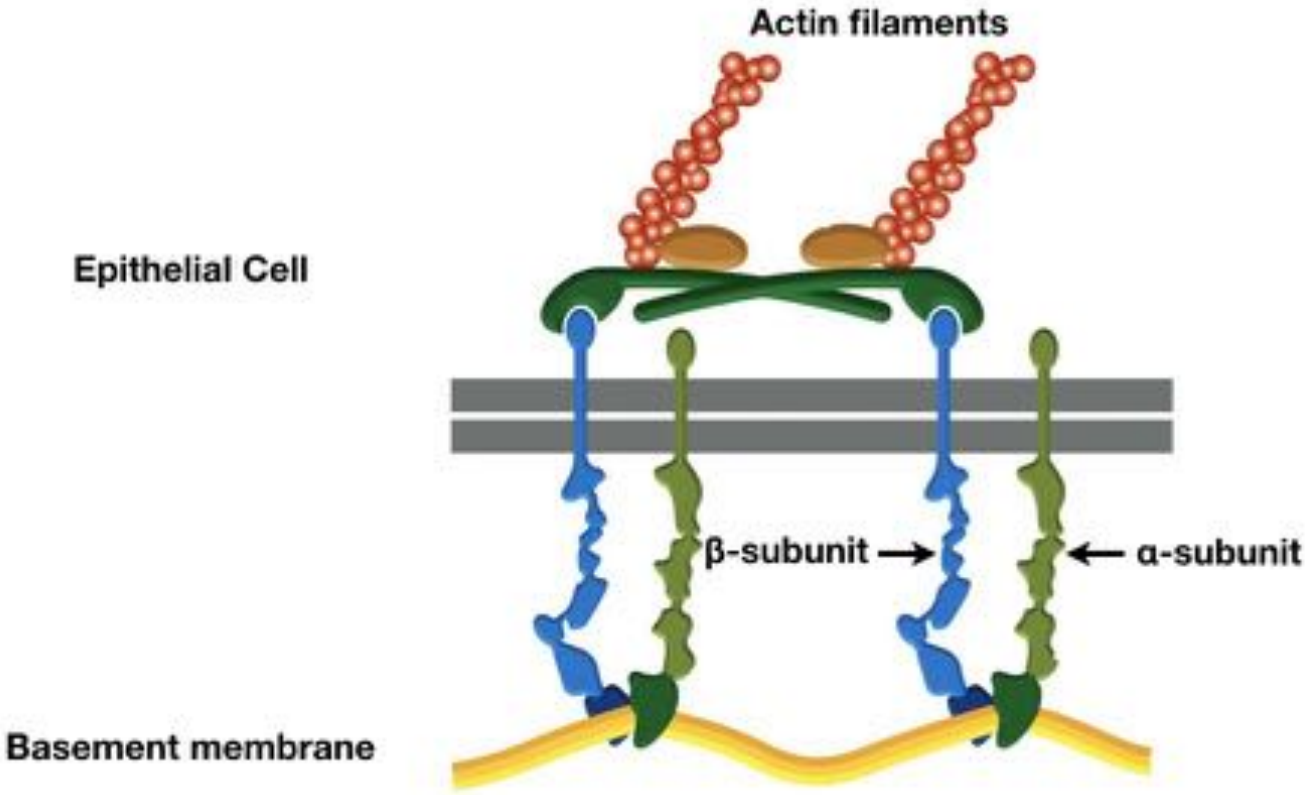
Type IV collagen is the main structural component of the basement membrane.

Laminin functions as a molecular glue in the basement membrane as it contains domains that interact with different proteins. One domain interacts with itself to allow laminin to form a network. Other domains bind to cells and interact with perlecan and nidogen to the laminin network with the collagen network. Laminin consists of a trimer of alpha, β and gamma subunits.



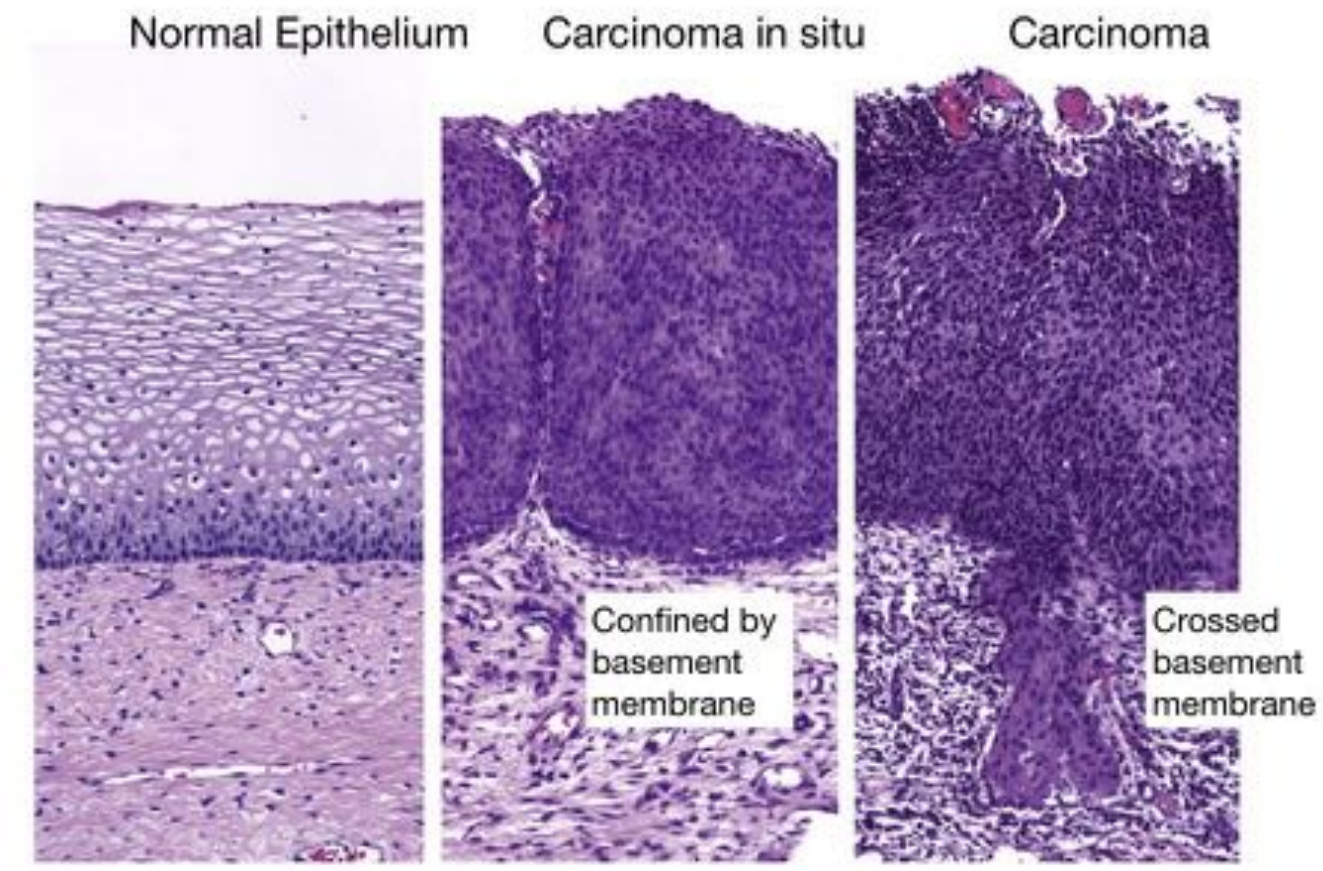
Laminins form network and organize components of the basement membrane.

Epithelial cells attach to their basement membrane via integrins. Integrins in the cell membrane associate with components of the basement membrane, mostly laminin and fibronectin. Inside the cell, integrins are linked to actin filaments in most epithelial cells, but in epithelium subjected to strong mechanical forces integrins are linked to intermediate filaments.



Integrins in epithelial cells bind laminin and fibronectin in basement membrane.

One medically important function of the basement membrane is that it keeps carcinomas (cancers of epithelial origin) from gaining access to the lymph and blood vessels and spreading to other parts of the body. Because epithelia lack blood vessels and lymph vessels, cancerous cells must cross the basement membrane into the underlying tissue to enter the blood or lymph system. Carcinomas that have not crossed the basement membrane are often referred to as carcinomas in situ. Carcinomas that have crossed the basement membrane have a high probability of becoming metastatic.



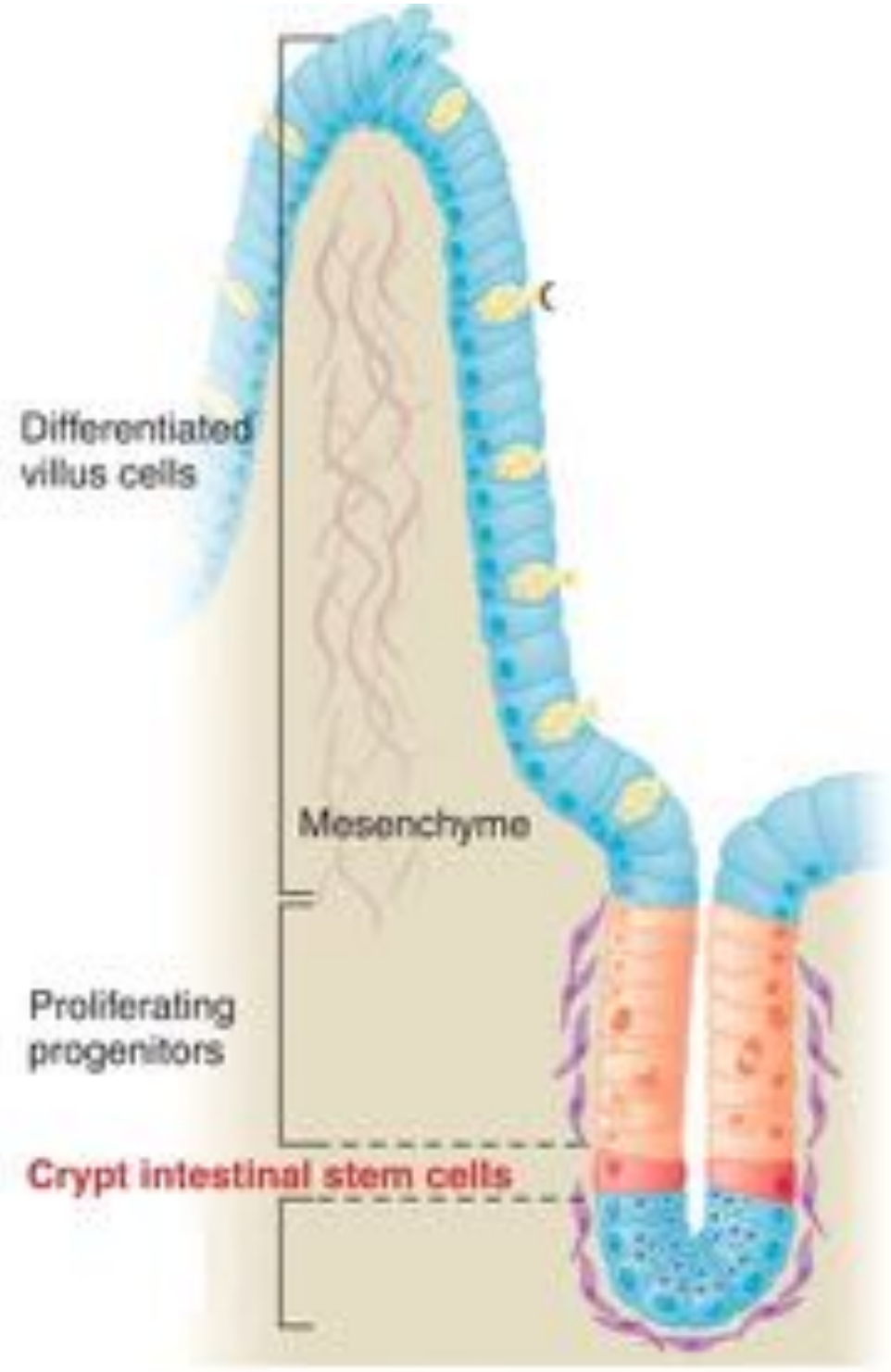
Basement membrane restricts invasion of malignant cells.

Renewal

Many epithelia face external the environment and lose cells due to mechanical strain and the natural aging of cells. In particular, the epithelia of the skin and intestine have high rates of cell loss. The epithelia of a typical villus in the small intestine turns over every 3 to 5 days, and the epidermis is completely replaced over 1000 times over the lifetime of an individual. To replace cells, epithelia need a pool of stem cells that proliferate and differentiate into a specific type of epithelial cells. Because the skin and intestine require many a steady supply of new cells, both epithelia have high rates of cell division and are prone to developing cancers.

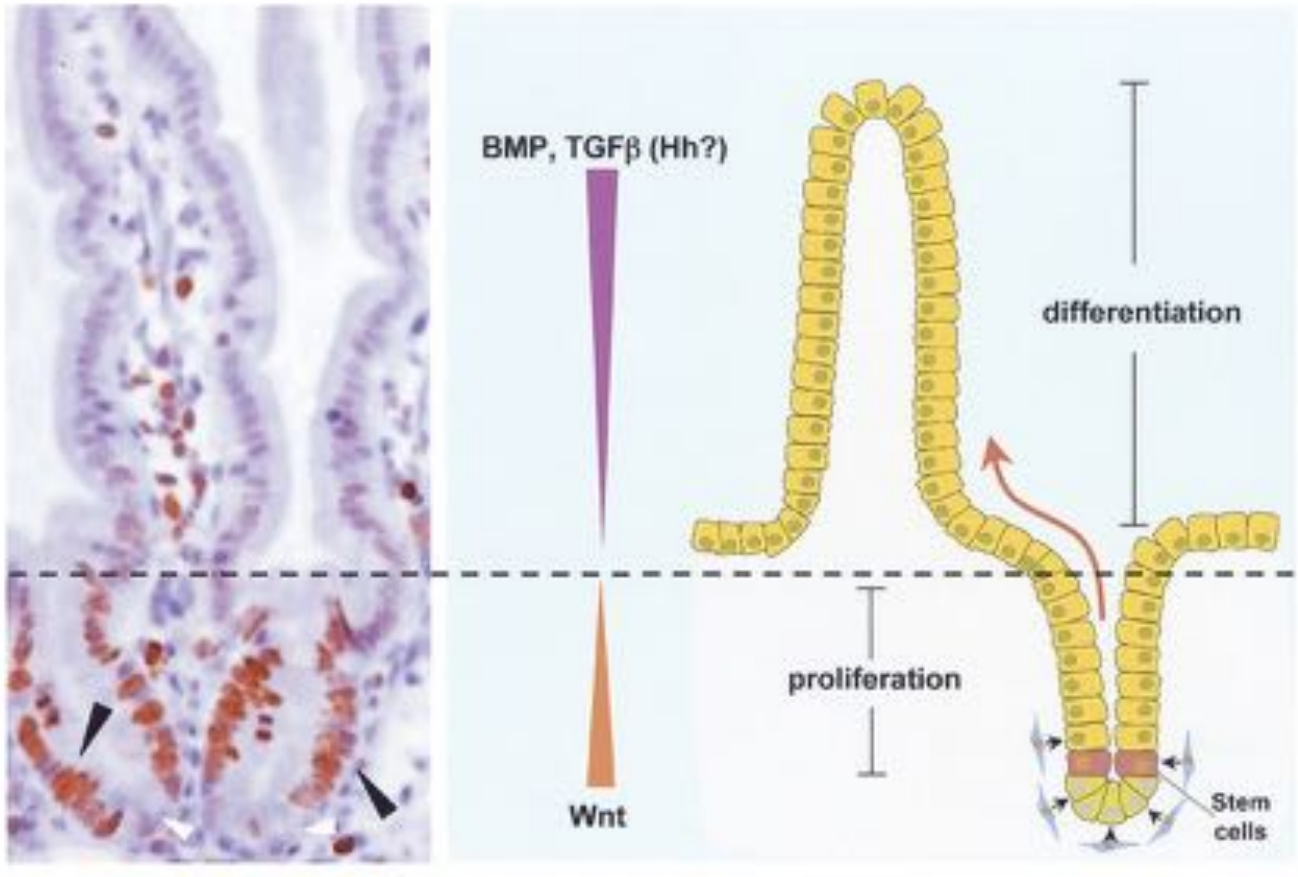
In many epithelia, the stems cells reside in discrete locations called niches. Non-epithelial cells around the niches produce growth factors and mitogens that regulate the division and differentiation of the stem cells. The epithelium of the small intestine is a single layer of cells. The stem cells reside in one domain of this layer which is located at the base or crypts of the

villi. These cells give rise to faster dividing cells that differentiate into intestinal cells as they migrate up the villi.



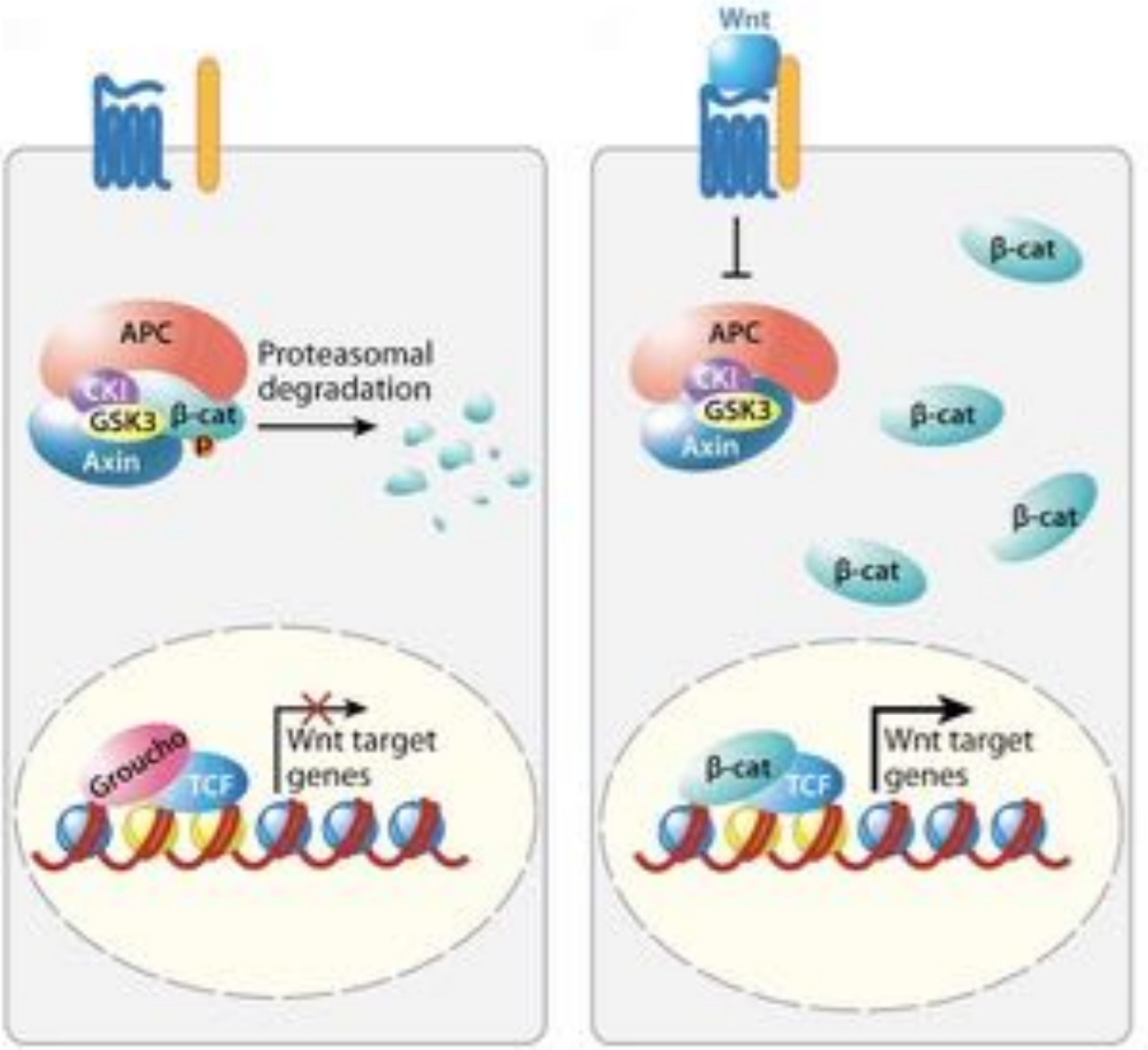
Intestinal epithelial stem cells resides in niche at the base of the epithelium.

The cell division of the stem cells and differentiated cells in the intestine is regulated by different signaling molecules that are produced by cells in the extracellular matrix. Wnt stimulates cell proliferation in the crypts of intestinal epithelium, whereas other signaling molecules (BMP, TGF-β) inhibit cell proliferation further up the intestinal epithelium. The concentration of these signaling molecules along the epithelium determines whether cells are stimulated to divide.



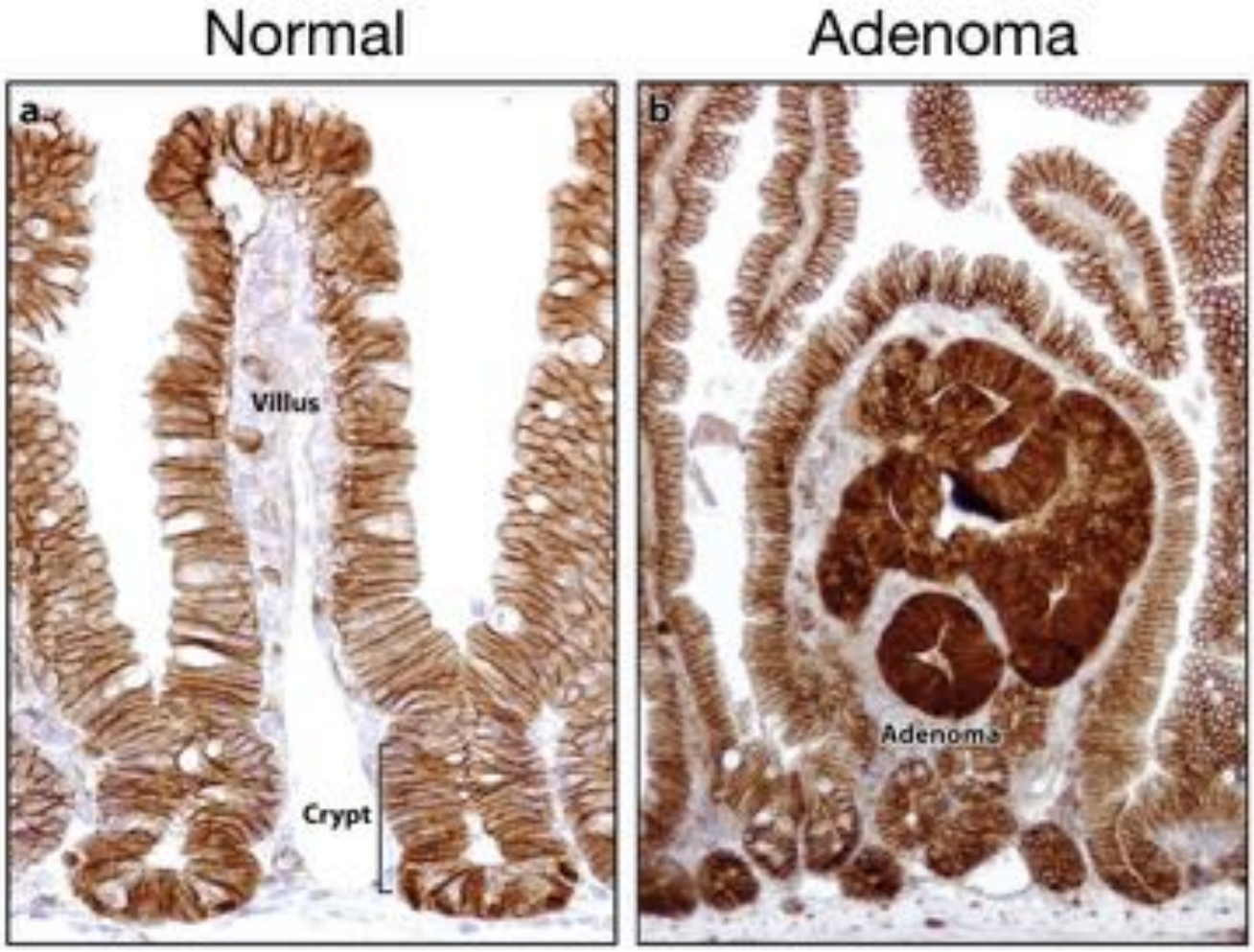
Growth factors regulate the cell division of cells in different regions of the epithelium

A key regulator of cell division in intestinal epithelial cells is APC (adenomatous polyposis coli) which is part of a complex that keeps the amount of cytosolic β-catenin low. APC, along with other proteins, phosphorylates β-catenin targeting it for degradation. Consequently, β-catenin cannot activate transcription of genes that encode proteins that trigger cell division. Stem cells in the intestine reside in an area with a high concentration of Wnt. Wnt activates a signaling pathway that inhibits the activity of the APC complex resulting in elevated cytosolic levels of β-catenin. Cytosolic β-catenin enters the nucleus and turns on expression of genes that stimulate entry into the cell cycle.



Wnt increases concentration of β-catenin allowing cells to grow and divide.

By inhibiting the activity of β-catenin, APC functions as a tumor suppressor and mutations in APC can lead to colon cancer. In normal color (left), β-catenin (brown stain) localizes to the cell membrane because it is associated with cadherins and cytosolic β-catenin is low due to its degradation driven by APC. In APC mutants, the amount of cytosolic β-catenin increases leading to increased cell division and the development of adenomas. Adenomas are a precursor to colon cancer.



Mutated APC leads to elevated β-catenin in nucleus and cell proliferation.