Laboratory #1

This laboratory exercise will study cells, epithelia, glands and connective tissue. Since all organs are comprised of cells and tissues, we will use histologic slides of organs to study these features. However, there will be many features that will be studied in a later lab. Please focus on the elements described in the following protocol. Each slide contains a link to a description of features that you are to study.

The slides are located on the virtual histology link of the anatomy department website (also linked to from the course Canvas site).

The following descriptions will include some questions that may require you to recall some points from class. When you are done reviewing slides, please complete the pre-lab readiness quiz on the blackboard site. You can take this quiz as often as you want, but you must achieve a score of 80% or better to receive credit. Completion of this exercise will assure that you're ready to assist your group.

Examine each slide very briefly. Focus on the features that are discussed in the exercise below. Each of these slides will appear in a later discussion of the entire organ. Use the small inset of the entire block of tissue to orient yourself to areas of particular interest on the slide.

Cells

Slide 3

This slide of the liver should be examined at high power. Note the staining properties of the cells with eosinophilic cytoplasm and basophilic nuclei. Identify several different cell types based on staining. The majority of the cells are the hepatocytes and have rather euchromatic nuclei with large nucleoli. What is the functional significance of rather euchromatic nuclei? Why does heterochromatin appear to reside near the nuclear envelope? The cells with heterochromatic nuclei are different cell types. Note that these cells and nuclei are different both in shape and size. Note that it appears that you can see the membranes of some of these cells.
However, the cell membrane is below the resolution of the light microscope. Why does it appear that you can see membranes?

**Slide 91**

In this slide of the spinal cord smear focus on the thicker portions of the specimen. Note the cells with very blue (basophilic) cytoplasm. These are motor neurons. What are the characteristics of the nuclei of these cells? What do you think makes cytoplasm so blue?

**Slide 36**

On high-power view of the pancreas, notice that there are clumps of cells arranged in a more or less circular pattern, with dark blue cytoplasm around the edges and cytoplasm containing highly eosinophilic (red/pink) granules towards the center. These are actually alveolar glands, although it's hard to distinguish the lumen. These alveoli are separated from one another by light pink connective tissue septa. Try to identify some of the ducts and blood vessels within the connective tissue. This is best done by scanning at a slightly lower power. Note that the ducts are filled with eosinophilic material. These are lined by cuboidal cells. There are also small blood vessels containing blood cells. These are lined by flattened squamous cells called endothelium. What do you suppose is contained within the eosinophilic granules? Why are the bases of the cells so basophilic? What kind of gland comprises the majority of this organ?

**Slide 93**

This is a slide of a sympathetic ganglion. Most of it consists of nerve fibers with small dark nuclei of Schwann cells (which cover nerves). When scanning at low power, you'll see some dark blue cells. These are concentrated on the right side of the specimen and are ganglion neurons. Notice the color of the cytoplasm, the prominent nucleoli, and look for some neurons containing brownish granules. This is lipofuscin. What does lipofuscin represent? And why does it tend to accumulate in neurons?
Slide 83

The majority of this slide is dominated by cardiac muscle cells. Note that the cytoplasm is very eosinophilic because of the high concentration of proteins. Most proteins stain with eosin. At very high power, if you look carefully, you can see small brownish granules within many muscle cells. This is lipofuscin.

Slide 19

This is a classic slide of whitefish blastula, in which many mitotic figures can be seen. Try to identify as many stages of mitosis as possible. Many of the cells appear to have radiating lines within the cytoplasm. What are these?

Slide 21

This is a fibroblast tissue culture. Notice the star shaped appearance of most of the fibroblasts. Also notice that there are many smaller, denser and more round cells. If you look carefully, you can often see condensed chromatin in these. These are the cells undergoing mitosis.

**Epithelia & Glands**

Slide 26

This is a slide of kidney, stained with PAS stain and hematoxylin. If you look toward the top of the specimen, close to the apex of this triangular specimen you will find many examples of tubules, some of which have thick walls and some very thin walls. Focus on high power and you will notice that the thin-walled tubules have a single layer of squamous cells, while the thicker-walled tubules have cuboidal shaped cells. These tubules all seem to be outlined in purple. This is because the PAS stain shows the basement membranes that support these cells. Some of these thin-walled tubules are capillaries, while others are part of the kidney tubular system. It is hard to tell them apart, although some have red blood cells in them, and these are capillaries. If you look towards the bottom of the slide you will see more tubules lined by cuboidal epithelium with very dense basement membranes. Capillaries are present in this area as well.
Examine the lining of the jejunum. Note that the surface lining cells are columnar. Also, at high power, the surface of the cells appears fuzzy. This is due to microvilli. Occasionally within this lining there are clearer zones. These are actually cells called goblet cells. These are single cell glands that secrete mucus. If you move away from the lumen, there are round tubes that represent simple tubular glands (intestinal glands). The lining cells are still columnar. Occasionally, intermixed with these cells, you may find mitosing cells with condensed chromatin, and you can also identify some dark, apoptotic cells.

This is also a view of the small intestine, although the lining here is on the lower part of the slide. This is stained with PAS, and it shows the goblet cells in purple. These goblet cells secrete their purple mucus to coat the surface of the microvilli of the small intestine. Note that the lining cells continue to be simple columnar, as they are throughout much of the gastrointestinal tract.

This is a whole mount slide of mesentery. It is stained with silver stain, which stains reticular fibers and also cell boundaries. On high power you will note faint round nuclei in the center of an area surrounded by a dark polygonal line. These look much like irregular paving stones that are fitted neatly together. These
represent simple squamous cells covering the surface of the mesentery. The fainter, oval, bluish structures are the nuclei of the cells. These cells fit together almost like groups of fried eggs cooked in a large pan. Their borders meet and interdigitate. As you know these are held together by junctional complexes.

**Slide 53**

This is also a mesentery, although it is thicker and stained with a stain that will color elastic fibers dark blue and collagen fibers red. This is a good example of areolar connective tissue. Most of the small dark nuclei are fibroblasts. In some places you can see the nuclei of squamous cells. The elastic fibers are thin and dark blue while the collagen fibers are thicker and eosinophilic. There are some large cells with dark granules called mast cells. Blood vessels can be seen as dark thick lines with many nuclei running through this loose connective tissue.

**Slide 80**

This is a slide of the uterus. The left side of the slide shows the lining of the uterus (endometrium). The surface is lined by columnar cells. You'll also notice that there are many irregular lumens in the tissue below this surface. These are branched tubular glands. These are lined mostly by cuboidal to low columnar cells.

**Slide 39**

Examine the lining epithelium of this section of a uterine tube. The lumen of the tube is on the lower portion of the slide. This is a simple columnar epithelium, although in places it looks pseudostratified. There are 2 types of cells, one lighter and one darker. Notice that at high power many of the cells show a prominent fuzzy border on the lumen side. These are cilia. These are more prominent than microvilli and are motile.

**Slide 29**

This is a section of trachea. The lumen would be on the lower part of the slide. Examine this at high power to see classic pseudostratified columnar epithelium with cilia. Notice that in a pseudostratified epithelium there appears to be several layers of nuclei even though all cells touch the basement membrane.
Slide 40

The epididymis is a coiled tube. Examine the lining of some of the tubes that are cut in cross-section. Notice the pseudostratified epithelium with significant projections from the apical surface of the epithelial cells. These are stereocilia, although they cannot be distinguished from cilia by light microscopy.

Slide 32

Examine the epithelium lining the lumen of the esophagus. This is a stratified squamous epithelium that is non-keratinized. Note that the deeper cells (the dark blue cells at the base of the epithelium) are cuboidal. Also note that the surface cells retain their nuclei. Therefore, they can be distinguished from keratinized squamous epithelium.

Slide 33

This is a section of thick skin. The epithelium is at the bottom of the slide. At high power, note that the cells lose their nuclei towards the surface. These are keratinized squamous cells. The cells below that appear in several layers that become flatter as they approach the keratinized part of the epithelium.

Now focus on the dark blue areas closer to the top of the slide. These are surrounded by large white cells that are fat cells. These appear empty because lipid is removed in histological processing. Focusing on the dark blue region, notice that there are tubes of cells with lumens. The cells themselves appear cuboidal. These coiled tubular glands are sweat glands. One would have to reconstruct a gland from multiple sections to prove that this was a single, coiled tube. Closer to the epithelium you may find slightly lighter staining ducts from the sweat glands.

Next, look at the lighter staining region just beneath the epithelium. This is called the dermis and consists of dense irregular connective tissue. At high power notice the eosinophilic collagen fibers and the very small, dark fibroblast nuclei in between these fibers. This is the classic appearance of dense irregular connective tissue.
Slide 58

This is a slide of scalp. The surface epithelium is on the top of the slide. Notice that the epithelium is much thinner than on the last slide, and there appear to be only a few layers of keratinized cells at the surface. Also notice that there are many inward extensions from the surface, which represent hair follicles. Some of these hair follicles contain shafts of brownish hair. The hair follicle also has multiple layers of cells. Toward the middle of the slide there are lighter staining glands adjacent to the hair follicles. At high power you may notice that the cells in these glands appear foamy, and those toward the center appeared to be decomposing. These are sebaceous glands and the secretion is holocrine (that is, the entire cell becomes the secretion).

Slide 154

This slide shows a section through the ureter. The lining is urothelium (transitional epithelium). Note the bulging cells near the surface, and that cells almost appear cuboidal. If this were stretched out, however, the epithelium appears almost squamous.

Slide 41

In the clearer areas in between gland tissue of the salivary gland, there are ducts lined with an unusual type of epithelium: stratified cuboidal or stratified columnar. Now focus on the gland tissue, which is the darker staining portions. Most of these glands have very dark blue staining cells arranged in an acinar pattern. These are serous glands and serous acini. Particularly in the lower left of the
specimen, there are lighter staining cells around the acini. These are mucous acini. Occasionally you can find a mucus acinus with a cap of dark blue serous cells. This is called a serous demilune.

**Slide 41B**

Try to find the same features in this submandibular salivary gland. This gland contains more mucus acini.

**Slide 77**

First look at the epithelium of the tongue, which is on the upper part of this section. Notice that it is stratified squamous epithelium. Is it keratinized? Next notice the glands that are interspersed amongst muscle of the tongue. Towards the right of the specimen they are mostly serous glands, towards the left there are many mucous glands.

**Connective tissue**

**Slide 51**

This section through a fetal pig head has extensive mesenchymal connective tissue. Look for the lighter staining regions of the specimen. This is very loose connective tissue, and fibers are not prominent. Most of the cells are destined to become fibroblasts.

**Slide 52**

This is a section of an umbilical cord, which contains a very unusual connective tissue called Wharton's jelly. There is a large amount of mucoid tissue in between the thin fibers and fibroblasts within this tissue. This is carbohydrate rich.
Slide 54

This is a trichrome stain that stains collagen green. It is a section through the cervix. At higher power notice the small, flattened, dark nuclei between the irregular fibers. These are fibroblasts. Particularly on the right side of the specimen you can see some smaller red cells amongst the fibers. These are smooth muscle cells. You may also notice the lower left portion of the specimen lined with stratified squamous, non-keratinized epithelium.

Slide 55

This is a longitudinal section through a tendon. At high power, notice that all of the collagen fibers are slightly wavy and lined up, and fibroblast nuclei are sandwiched in between them. This provides strength in one direction. Consider what this would look like in cross-section.

Slide 56

This is a slide through the ear is stained with a stain that stains elastic fibers dark purple. At the center of the specimen there is an area of elastic cartilage. On either side of this is connective tissue with many elastic fibers. You can see the dark blue fibers interspersed with pink, collagen fibers. What is the function of elastic fibers in this area?

Slide 57

This is a slide of a lymph node. This organ contains many lymphocytes and blood vessels with an internal framework of reticular fibers. Many organs have a stroma comprised of reticular fibers. In this case a silver stain is used to highlight the reticular fibers. Go to high power and you will notice multiple black stained fibers in between and around cords of cells and blood vessels. Without special staining this reticular network is very difficult to visualize because the fibers are so thin.
Slide 90

Many of the slides we've seen contain white fat. White fat cells appear as empty circles with a very flattened nucleus on the side. This slide shows the appearance of brown fat on either side of the vertebra. At high power you will notice that these cells appear to be full of small, clear vesicles (see the picture for location).

Slide 60

This animal had been injected with non-toxic black particles. These are taken up by phagocytic cells, here the Kupffer cells in the liver. At high power, you'll see many cells with black granules within them. These phagocytic cells, which are components of the macrophage monocyte system, appear in many tissues. However, they are often difficult to identify without doing some special staining. This slide is a good indication of just how many there are.

Electron Microscopy

Finally, review the electron micrographs that are found on the virtual histology site. There are three short modules: one on organelles; one on cytoskeleton; and one on epithelium (and epithelial specializations). Each module has a series of micrographs, and each has a short caption pointing out what you should look for on the picture (usually labeled). You will not be expected to identify everything on these slides, but you should recognize classic examples of the features that are demonstrated.