LABORATORY 16
LOWER GI TRACT

OBJECTIVES:

At the end of this lab, you should be able to:

1. describe the structural variations that allow you to distinguish between:
   - small intestine and large intestine
   - the parts of the small intestine - duodenum, jejunum and ileum
2. identify and explain the function(s) of intestinal epithelial cell types including:
   - absorptive cells (enterocytes)
   - goblet cells
   - Paneth cells
   - enteroendocrine cells
3. describe where plicae circulares, villi & microvilli are found, and what layers are involved in forming each structure
4. identify the location of the epithelial stem cells in small intestine and large intestine
5. explain the structure and function of a lacteal and find an example of one in a section
6. identify the submucosal plexus and the myenteric plexus. Look for ganglion cells in each. Explain the function of the enteric nervous system.
7. describe the structural features you could use to distinguish small and large intestine from esophagus or stomach
8. identify the following components of the GALT:
   - diffuse lymphocytic infiltration
   - individual nodules
   - groups of nodules including those of the appendix, Peyer’s patches and tonsils

LABORATORY:

Please study the following slides in your set:

A. SMALL INTESTINE
   Slide 16 (HU Box): Stomach and Duodenum, I.s.
   Slide 62A: Pyloro-Duodenal Junction, or
   Slide 63: Stomach and Duodenum, LS at Junction

Identify the abrupt epithelial transition between pyloric stomach and duodenum. Note that both organs are lined by a simple columnar epithelium. However, in the stomach essentially all the surface cells are of one type (surface mucous cells), while in the duodenum and the rest of the intestine the surface epithelium contains two major cell types, goblet cells and intestinal absorptive cells.
Identify the pyloric sphincter present at the pyloro-duodenal junction. What type of muscle makes up the sphincter? (Answer: Smooth muscle) Is it longitudinally or circularly arranged? (Answer: All sphincters must be circularly arranged in the wall of the organ or else they would not be able to act as sphincters.) Compare this region to the cardio-esophageal junction where there is a physiological sphincter but not an anatomical one.

Slide 60 (HU Box): Duodenum, c.s.
Identify the villi. Their presence clearly characterizes this section as small intestine. The presence of Brunner's glands in the submucosa clearly identifies it as duodenum. Another clue that this is duodenum comes from examining the organ visible next to it. What organ is this? (Answer: The pancreas. The head of the pancreas is directly surrounded on three sides by the duodenum.) Identify the intestinal absorptive cells and goblet cells, which are the major cells types in the epithelium covering the villi. Scattered enteroendocrine cells may also be present but again are difficult to identify. These same cell types (at varying stages of differentiation) are present in the intestinal glands (crypts of Lieberkühn). These are simple tubular glands formed by the invagination of the surface epithelium into the lamina propria. By scanning electron microscopy they would look like holes scattered between the bases of the villi. The stem cell population in the small and large intestine is located near the base of the intestinal glands. See if you can identify any mitotic figures in the crypts in this or any other slide of small intestine. Often the nuclei of dividing cells are located closer to the lumen of the crypt than the nuclei of other cells, as if they stepped toward the lumen during mitosis and then stepped back into line when they were finished dividing.

Slide 61 (HU Box): Jejunum, or
Slide 64 or 64A: Jejunum
Jejunum is usually identified by what it does not have. It is the part of the small intestine that normally contains "no" Brunner's glands (Brunner's glands are typical of duodenum) and "no" Peyer's patches (Peyer's patches are typical of ileum). In some versions of these slides Paneth cells with large eosinophilic cytoplasmic granules can be seen at the base of the intestinal glands. They are particularly numerous in jejunum and ileum.

Slide 62 (HU Box): Ileum, c.s., and
Slide 65 (Trichrome) or 65A: Ileum (H&E)
Most, but not all, sections of ileum contain several lymphoid nodules that are part of a Peyer's patch. Usually these tend to be located on the abmesenteric border of the ileum (the side opposite the point where the mesentery attaches to the intestinal wall, but on some of our slides this is not the case. Notice that the normal pattern of intestinal villi tends to be somewhat altered in the region covering a lymphoid nodule in Peyer's patches. Specifically there tend to be no villi immediately overlying the nodules of a Peyer's patch. Instead there is a dome epithelium. What cell type is present on the villi but largely absent from the dome epithelium? (Answer: Goblet cell) A unique cell type located in the dome epithelium is the M cell. M cells transport antigens across the epithelium & release it near lymphocytes and antigen-presenting cells (APCs). Many lymphocytes and APCs often lie within a deep pocket created by an infolding of the M cell's basal plasmalemma. M cells themselves are often difficult to see by LM, but by
looking for lymphocyte nuclei that appear to be clustered within the epithelium you can deduce where an M cell must be present.

Compare the relative abundance of goblet cells vs. absorptive cells in the ileum vs. duodenum. You should find that goblet cells increase in frequency as you move from duodenum through small and large intestine, reflecting the greater need for lubrication as the fecal mass becomes more solid.

Look for plicae circulares (see Wheater, Fig. 14.16b, p.262). These are present in most of the duodenum, well-developed in the jejunum, and present in the proximal half of the ileum. A plica circularis includes mucosa and submucosa, and differs from a ruga in the stomach in that plica is a permanent structure, while a ruga is temporary. In addition, a plica has many villi on its surface. A ruga, being part of the stomach, has no villi. What makes up the core of a villus (see Wheater, Figs. 14.19, p. 265)? (Answer: Only lamina propria – no submucosa)

B. LARGE INTESTINE

Slide 63 (HU Box): Colon, Human, or
Slide 67 or 67A: Colon, Human

Note that the large intestine has no villi. It also has no permanent folds such as the plicae circulares of the small intestine, although temporary folds of the mucosa and submucosa can occur. Identify the intestinal glands. In both small and large intestine these are simple tubular glands lined by a simple epithelium with goblet cells and absorptive cells. In the colon Paneth cells are rare or absent. Goblet cells are even more abundant than in small intestine, sometimes making it difficult to find the intestinal absorptive cells interspersed among them.

Examine the muscularis externa, especially its outer longitudinal layer. Around the full circumference of the colon this layer has three thickened regions called the teniae coli. Between the teniae, the outer longitudinal layer is extremely thin or absent. Decide whether your section shows the teniae coli or the attenuated region of the outer longitudinal layer.

Re-examine a section of cardiac stomach (Slide 15HU, 57HU, or 60A). Note that you can distinguish between colon and cardiac stomach based on the types of cells in the surface epithelium, the number and orientation of the layers in the muscularis externa, whether the glands open into pits or directly onto the surface, and whether the glands are straight or coiled.

On any slide of small or large intestine (or stomach) identify the ganglia and nerves of the enteric nervous system. They are found in the submucosa (submucosal or Meissner’s plexus) and between the two layers of the muscularis externa (myenteric or Auerbach’s plexus.) Recall that these ganglia contain both sensory and motor neurons, and that they can produce coordinated peristalsis even if the normal input from the autonomic nervous system is cut.

Slide 64 (HU Box): Appendix, c.s., or
Slide 66 or 66A: Appendix, c.s

Notice that the structure of the appendix closely resembles that of the colon except that appendix has no teniae coli; instead the 3 teniae meet at the appendix and unite to form a uniformly thick outer longitudinal layer of the muscularis externa.

The appendix normally contains many lymphoid nodules extending from lamina propria into submucosa. You may confuse these with Peyer’s patches in the ileum, but
in the appendix the nodules tend to be more uniformly distributed around the entire circumference of the gut tube. Also, recall that the basic structure of ileum and appendix differ. Ileum is a part of small intestine and therefore has villi whereas the appendix resembles colon and has no villi. Which class of antibody is produced by most of the plasma cells in the gut? (Answer: IgA)

Slide 17 (HU Box): Recto-Anal Junction, I.s.

Study the epithelial transition between rectum and anal canal (see Wheater, Fig. 14-32, p. 273). The rectum is lined by a simple columnar epithelium similar to that of the colon and has typical straight unbranched intestinal glands. In the anal canal the epithelium changes from a simple epithelium to a stratified epithelium, ending up as a minimally keratinized stratified squamous epithelium in the lower part of the canal. The anal canal also has branched tubular glands that open via ducts onto the anal surface. Follow the anal canal and look for another epithelial transition to the maximally keratinized stratified squamous epithelium of the perianal skin. In the transition zone between anal canal and the perianal region you may see apocrine glands with their characteristically wide lumen.

Compare recto-anal junction with previously studied transition zones:
- Gastro-esophageal junction (Slide 15HU, or 60A)
- Gastro-duodenal junction (Slide 16HU, 62A or 63)

Be able to distinguish one transition zone from another.

C. ELECTRON MICROSCOPY (RHODIN)

Identify the following:

1. In the small intestine distinguish between plicae circulares (Figs. 29-1 to 29-3), villi (Figs. 29-1 to 29-5), and microvilli (Figs. 29-5 to 29-7).

2. Study the structure of a villus (Fig. 29-5). Identify the simple columnar epithelium composed of intestinal absorptive cells, goblet cells and occasional enteroendocrine cells. The core of the villus is the loose cellular connective tissue of the lamina propria. It contains blood vessels, lacteals, and smooth muscle cells arranged parallel to the long axis of the villus.

3. Study the ultrastructure of the following cell types:
   - Intestinal absorptive cells (Figs. 29-5 & 29-6)
   - Goblet cells (Figs. 29-5 & 29-32).
   - Paneth cells (Figs. 29-12 & 29-14). The unusually large secretory granules of these cells contain lysozyme and α-defensins among other mediators. These granules would be highly eosinophilic by LM. Paneth cells are characteristically located in groups near the bottom of the intestinal glands of the small intestine.
   - Enteroendocrine cells (Fig. 29-16 & #14 in Fig. 29-5). Don’t worry about distinguishing the different types of enteroendocrine cells from one another.

4. Identify the inner circular and outer longitudinal layers of the muscularis externa and the neurons of the myenteric plexus, which lies between the two layers (Fig. 29-41).
5. In the large intestine identify a teniae coli (Figs. 29-28 & 29-29).
6. At the ano-rectal junction (Figs. 29-37) find the internal anal sphincter (smooth muscle) and notice how it represents a thickening of the inner circular layer of the muscularis externa. Circularly arranged muscle is what you would expect to find in a sphincter. Identify the external anal sphincter. This is skeletal muscle and is voluntary; the internal sphincter is involuntary.

LABORATORY 16 CHECKLIST
LOWER GI TRACT

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<th>LIGHT MICROSCOPY</th>
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<td>gastro-duodenal junction</td>
<td>plica circularis vs. villus</td>
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<td>duodenum vs. jejunum vs. ileum</td>
<td>teniae coli</td>
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<td>goblet cell</td>
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<td>intestinal absorptive cell</td>
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<td>villus</td>
<td>muscularis externa</td>
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<td>intestinal gland (crypt of Lieberkühn)</td>
<td>serosa/adventitia</td>
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<td>Brunner’s glands</td>
<td>myenteric vs. submucosal plexus</td>
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<td>Paneth cell</td>
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<td>location of epithelial stem cells in small vs. large intestine</td>
<td>recto-anal junction</td>
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<td>Peyer’s patch</td>
<td>perianal apocrine glands</td>
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<th>ELECTRON MICROGRAPHS</th>
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<td>villus vs. microvillus</td>
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**NOTE:** These checklists include MOST of the structures that you should be able to identify. Exams may include structures not on these lists.
FOCUS QUESTIONS
LAB 16: LOWER GI TRACT

See whether you can answer the following questions. The correct answers are posted on the course website (http://neurobio.drexelmed.edu/education/ifm/microanatomy) under “Lab Focus Questions”.

1. Epithelial turnover is quite rapid in the GI tract. This fact is clinically significant since the rapidly dividing epithelial stem cells are often damaged by the treatments used to kill rapidly dividing cancer cells. This can lead to major problems with nutrition and fluid balance in cancer patients. Where are the epithelial stem cells located in the following organs: esophagus, stomach, small intestine and large intestine?

2. Describe three structural modifications that increase the surface area in the small intestine. Why is this important?

3. Describe the process by which IgA is transported across the intestinal epithelium into the lumen.

4. How does the presence of antibody in the intestinal lumen protect the body against invasion by that antigen?

5. Antimicrobial activity in the GI tract depends on a number of other mediators in addition to secretory IgA. How does the Paneth cell contribute to mucosal immunity?

6. What role do M cells play in the mucosal immune system? Where are they found?

7. Colon and cardiac stomach both lack villi and contain many mucus-producing cells. How can you distinguish between them morphologically?

8. What morphological characteristics allow you to distinguish duodenum from ileum and jejunum?

9. At two of the epithelial junctions in the GI tract a minimally keratinized stratified squamous epithelium abruptly changes to simple columnar or vice versa. This occurs at the gastroesophageal junction and at the rectoanal junction. Given this similarity, how can you distinguish these two junctions from one another?

10. At the gastroduodenal junction, how can you distinguish the two organs from one another since both are lined by simple columnar epithelium?