

Co and Benjamin  
Laboratory Manual in Animal Histology  
2<sup>nd</sup> Ed

# **Laboratory Manual in Animal Histology (Biology 134)**



**Department of Biology  
College of Arts and Sciences  
University of the Philippines Manila**

**Elisa L. Co, Ph.D.  
Kimberly S. Beltran-Benjamin, M.Sc.**

## EXERCISE 10: THE DIGESTIVE SYSTEM

### Introduction

The digestive system is composed of the oral cavity, digestive tract, and the digestive glands. It is responsible for the digestion and absorption of food and elimination of indigestible materials.

### Objectives

At the end of the exercise, the students should be able to:

1. differentiate the different digestive organs based on their histological features;
2. identify the different cell types and correlate their structures w/ their functions; and,
3. identify the various cellular structures and their counterparts under electron microscope.

### I. Parts of the Digestive System

**A. Oral Cavity-** important in ingestion and preliminary digestion of food.

1. **Lip-** double folds of skin and muscles for the entry of food to the digestive tract.

- ❖ **Outer surface-** has a thin epidermis or mucosal epithelium that is keratinized. Many hair follicles and sebaceous glands are found just below the epidermis.

- ❖ **Transitional Zone** - also has a thin epidermis or mucosal epithelium that is keratinized. Many hair follicles and sebaceous glands are found just below the epidermis called the **vermillion border**. Curved region between the outer and inner lips characterized by the presence of folded mucosa w/c is thick and non-keratinized. Unique to this region is the **network of capillaries and branched venous plexus** in the dermal region. The abundant capillaries account for the reddish color of the lips.

A thick layer of skeletal muscles make up the core of the lips.

- ❖ **Inner Layer-** part facing the tooth and the gums. It has a non-keratinized stratified squamous epithelium w/c is intermediate in thickness between the outer and transitional regions.

- ❖ **Lamina propria** – thick layer of loose connective tissue and sero-mucous labial glands. Towards the lower part, in the furrow between the lip and gingival, the epithelium is changed into keratinized type. The gingival covers the alveolar bone of the maxilla and mandible.

2. **Tongue-** a highly muscular organ that aids in the mastication of food. It is divided into three parts:

- 1.1. **Apex-** conical tip of the tongue that is unattached.

- 1.2. **Body or Corpus** - middle part that constitute 2/3 of the entire tongue.

1.3. **Base or the root** - posterior third of the tongue leading to the esophagus.

**Dorsal surface** - irregular owing to the presence of small epithelial projections called **Lingual papillae**. The skeletal muscles are disposed into three planes and adhere to the mucuous membrane. Identify the different papillae.

- a) **Filiform** - cone-like elevations that curved posteriorly. Smallest and most numerous of all papillae. No taste buds are found in the slightly keratinized epithelium.
- b) **Fungiform** – Mushroom shape papillae with rounded tops and short stalks. These are interspersed among the filiform papillae. It has primary and secondary papillae on the anterior.
- c) **Vallate or Circumvallate** - largest type arranged in V-shaped row on the posterior third of the tongue. They have flat tops with tapering ends containing many taste buds. Narrow but deep furrows surround this papilla.
- d) **Foliate** - present in rabbits and absent in man. Column-like with very deep 2<sup>o</sup> papillae. They form a cluster on the margin of the tongue. Small as well as large papillae are found on their sides.

**Undersurface** - smooth surface covered with non-keratinized epithelium.

**Taste Buds** – light barrel shape structures composed of epithelial cells surrounding a central cavity.

- a. **Taste Pore** – a minute opening at the apex of the taste bud.
- b. **Taste Cells** – are spindle shaped cells with striated border.
  - ❖ **Sustentacular Cells** – more numerous and slightly dark with oval nucleus at the basal region.
  - ❖ **Olfactory Cells** – appears pale staining with round nucleus at the middle of taste bud.

3. **Tooth**- the hard calcified substance partly enclosed by alveolar bone. It consist of three parts:

- a. **Crown** – upper 1/3 which protrude from a bony socket. It is tapering and covered by the enamel.
- b. **Root** – lower 2/3 surrounded by mineralized cementum.
- c. **Cervix** – the neck part where the enamel and cementum meet.

From the longitudinal section of the tooth, identify the following structures starting from the center to the outside.

- a. **Pulp Cavity** – central space filled with connective tissue and mesenchyme cells. Towards the basal region, it tapers to and form a tiny orifice called **apical foramina**-that allows exit of blood vessels and nerves.

- b. **Dentin** – appear dark, thick layer of extremely hard material- mainly calcium salts and hydroxyapatite crystals (HAC) and collagen type I, GAGs and phospholipids.
- c. **Odontoblast** – layer of columnar cells immediately below the dentin. It appears as thin line outer to the pulp cavity. They secrete dentin. Just outer to this layer is the **predentin** which is the immature or unmineralized dentin. It appears very light.
- d. **Enamel** – very dark due to its intense staining. It is synthesized by tall cells called ameloblasts which are devoid of cytoplasmic processes.
- e. **Periodontal ligament** – thick bundles of collagen fibers that attached the tooth to the alveolar bone. **What is the other name of this layer?**
- f. **Alveolar bone** – appear as double fingers with a central space occupied by the tooth.
- g. **Gingiva** – mucous membrane outer to the alveolar bone. It is a layer of stratified squamous epithelium.

## II. Histological Architecture of the Digestive Tract

The tubular organs of the digestive tract share a common histological organization but are modified in accordance to their specific functions. In general, the walls of the digestive tract are divided into four layers, namely:

1. **Mucosa-** – innermost layer surrounding the lumen or cavity. It is composed of epithelial membrane supported by a highly cellular layer of connective tissue called **lamina propria**. A thin layer of smooth muscles, the muscularis mucosa, surrounds the lamina propria.
2. **Submucosa** - composed of areolar connective tissue containing many blood vessels, nerves and lymphoid tissue. A small group of autonomic ganglion cells called Meissner's Plexus is found in this layer. The cytoplasm of the ganglion cells is light-staining with centrally located large nuclei.
3. **Muscularis Externa** - thick layer of smooth muscles subdivided into inner circular and outer longitudinal layers. Scattered between these two sublayers, several autonomic ganglia can be observed. These are the Myenteric or Auerbach's Plexus. They appear in small cluster of cells that are light and large.
4. **Serosa or Adventitia** – the outermost layer of cells and connective tissue. **Adventitia** – outermost layer of the visceral organ. When the organ is found outside the coelom, there is a thick fibrous connective tissue with adipose tissue and blood vessels. In some parts of the alimentary tract, only a thin layer of mesothelium surrounds the organ and this is called the serosa. **Serosa** - is composed of an outer layer of simple squamous cells. It is found in organs within the peritoneal cavity.

### III. Digestive Organs

1. **Esophagus** – short narrow tube for the passage of food to the stomach.
  - a. **Mucosa** – thick non-keratinized stratified squamous epithelium with distinctly basophilic basal layer that appear wavelike. Hence, it forms ridges that partly enclosed the lamina propria.
    - ❖ **Lamina propria** - is less cellular with fine areolar connective tissue.
    - ❖ **Muscularis mucosa** – thick layer of smooth muscles that are disposed in bundles. Their nuclei are round and dark.
  - b. **Submucosa** – dense connective tissue containing blood vessels and nerves. Mucosal glands are found in the upper half of the esophagus
  - c. **Muscularis Externa** – variable in composition according to location.
    - ❖ **Upper third** – has striated muscles which allow voluntary control of the passage of food. This is the unique feature of this organ.
    - ❖ **Middle third** – has striated and smooth muscles.
    - ❖ **Lower third** – has smooth muscles only.
  - d. **Adventitia** – thin layer of loose connective tissue which merges with the surrounding connective tissue.
  
2. **Stomach** – a large chamber for the storage and partial digestion of food.
  - a. **Mucosa** – simple columnar epithelium surrounding the lumen of the organ. It is generally called gastric glands but named according to their location: cardiac, fundic or corpus and pyloric. The surface of the mucosa form temporary longitudinal folds when the organ is empty. These folds are called rugae.
    - ❖ **Gastric glands** – simple branched tubular secretory structures composed of mucosal epithelium. This is divided into three parts:
      1. **Gastric pits or foveolae** – narrow cavities formed by the deep infoldings of the upper third of the mucosa. These are actually the ducts through which the secretions are released.
      2. **Neck** – the short and narrow point at which the glands bifurcate and rebranch. The branches of the glands are seen as straight or spherical bodies with small lumens. The cells are distinguished by virtue of their differential staining.
      3. **Body** – lower third of the mucosa. It is long and branching part of the gastric glands. The glands are separated by thin almost virtually indistinct lamina propria. The nuclei of the cells in the lamina propria are the only ones visible as oval dark structures. Small lymphatic nodules are scattered among the coiled glands.

The gastric glands are classified into three types based primarily on their locations:

1. **Cardiac glands** – found between the esophagus and the stomach. This is also called the gastro-esophageal junction. The glands are short, usually coiled with dilated lumen. The gastric pits are shallow.
2. **Fundic or body** – note that the glands here are longer and with shallow pits. They are tightly packed containing different cell types.
3. **Pyloric** – they are shorter and more coiled. However, the pits are very deep or longer than the other two types of glands.

The cells of the gastric mucosa as observed in the fundic glands are:

1. **Mucous Surface Cells (MSC)** – most superficial cells made of simple columnar epithelium. They appear light and disposed in a wave-like fashion. Each cell has a light oval cytoplasm and a basal nucleus.
  2. **Mucous Neck Cells (MNC)** – a cluster of cells on the basal parts of the duct. The cells are quite bigger than the MSC with many larger mucus droplets. Their nuclei are also oval and basal in location. They may also occur singly between parietal cells.
  3. **Parietal or Oxyphil Cells** – acidophilic cells often found in the neck region. They form long array of cells of interspersed among the chief cells and MNC. They are large, round or pyramidal cells with round nuclei. They secrete HCl and intrinsic factor.
  4. **Chief or Zymogen Cells** – basophilic cells prevalent in the basal part of the gland. Their nuclei are round and basal in location. In contrast to parietal cells, the nuclei are smaller and basal in position. They produce pepsinogen.
  5. **Enteroendocrine or Argentaffin Cells** – mostly found in the pylorus between the basement membrane and chief cells. They produce gastrin to release HCl secretions.
- b. **Submucosa** – contain the usual collagen and elastic fibers.
- c. **Muscularis Externa** – very thick layer of smooth muscles divided into three layers:
- ❖ **Inner Oblique** – exclusively found in the cardiac and fundic regions.
  - ❖ **Middle Circular** – much thicken to form sphincters in the cardiac and pyloric regions.
  - ❖ **Outer Longitudinal** – bundles of nerves called Auerbach's plexus are located between the circular and longitudinal muscle layer.
- d. **Serosa** – thin layer of loose connective tissue and mesothelium continuous with the parietal peritoneum of the mesogastrum.

**3. Small Intestine** – long (6 meters) tubular organ but highly coiled with narrow lumen. It is subdivided into three regions: duodenum, jejunum, and ileum where complete digestion and absorption of food occur. Such functions are reflected in its histological features.

View the entire organ under LPO and note the variation in each layer.

**a. Mucosa** – simple columnar epithelium which forms finger-like projections called **villi** on the surface of the lumen. It has a central core of lamina propria. These villi are most numerous and tallest in the duodenum and become fewer in the lower regions. The shape of the villi also varies from conical to flattened leaf-shaped, thus, greatly increasing the surface area of the organ. The cells of the mucosa are the following:

❖ **Simple Columnar Epithelium** – tall cells with dark oval nucleus. The apical surface of the cells have **striated border** appearing as parallel thin lines on the surface of each cells. Note the very dense line below the striated border. **What is it called?**

❖ **Goblet Cells** – cells are shaped like cones of ice cream. Find the nuclei on their tapered posterior end. They are fewer and squeezed between the more numerous columnar cells.

❖ **Paneth Cells** – occur in clusters on the basal part of the glands. They are readily identified by their large acidophilic granules on the apical cytoplasm. The circular profiles of intestinal glands are embedded in the lamina propria and consist chiefly of Paneth cells.

❖ **Enteroendocrine Cells** – they are found between basement membrane and nuclei of the columnar cells. They occur singly but widely scattered. They synthesize a variety of substances to control the functions of the intestines and other digestive organs.

❖ **Intestinal Glands or Crpyts of Lieberkuhn** – simple tubular glands which directly opens to the villi.

**b. Lamina Propria** – this occupy the central part of the villi where it appears as large irregular spaces surrounded by cellular connective tissue. The spaces are actually the blind-end capillaries called the **lacteals** that allow fat absorption in the small intestine. In between the glands, the lamina propria is viewed as mass of nuclei.

**c. Submucosa** – this forms a circular or semilunar folds called **Plicae Circularis** or **Valves of Kerckring**. These are permanent folds with villi on their surface and most numerous and tallest in the jejunum. In the duodenum, the submucosa has a great number of glands called the Brunner's glands that are branched tubular or tubulo-alveolar. In the ileum, the lamina propria and submucosa have a high

concentration of lymph nodules as the **Peyer's Patches**. These are visible as dark circular masses of cells in the submucosa.

A few clusters of **Myenteric Plexus** composed of parasympathetic cells are seen as pale elongated mass of cells with prominent nuclei. They are few and scattered throughout the submucosa. **What are their functions?**

- d. **Muscularis Externa** – similar histologic features as the other digestive organs.
- e. **Serosa** – thin mesothelium and fibrous connective tissue.

**4. Large Intestine** – also tubular and coiled organ but with larger lumen. This part of the gastro-intestinal tract is responsible for the reabsorption of fluid and formation of fecal material for elimination from the body. This is subdivided into several parts:

- a. **Caecum** – a small sac-like pouch near the junction of the ileocecal valve.
- b. **Vermiform Appendix** – tiny worm-like extension near the ileum.
- c. **Colon** – tubular part disposed in an ascending, transverse and descending parts terminating in a sigmoid.

**Appendix** – there are no villi or Paneth cells but most distinct in having abundant lymphocytes aggregated into **lymph nodules** in the submucosa. Has fewer and shorter intestinal glands and small lumen. It has a high concentration of goblet cells among the columnar epithelial cells. The Lamina propria contains much lymphocyte, plasma cells and macrophages.

**Colon and Caecum** – peculiar feature is the presence of :

1. **Taenia Coli** - aggregates of 3 thick bundles of longitudinal muscles. It is viewed as semilunar mass of cells outer to the thin circular muscle layer.
2. **Haustra** - sac-like appearance of the walls of colon and cecum resulting from tonic contraction of taenia coli.
3. **Appendices epiploica** - large globular mass of adipose tissue hanging down from the adventitia.
4. **Columns of Morgagni** – closely packed longitudinal folds of the mucosal epithelium.
5. **Rectum** – short, dilated part continuous with the colon.
6. **Anus or External orifice** – histologically, it has the same layers as the small intestine but for the absence of villi and Paneth cells.

**Anal Canal** – mucosa is changed into stratified squamous epithelium.

1. **Muscularis Externa** – the inner circular smooth muscle layer is thickened



to form the **Internal Anal Sphincter**. The outer circular band of skeletal muscles form the **External Anal Sphincter**.

**2. Hemorrhoidal Plexus** – large veins in the anus susceptible to varicosities.

**IV. The Accessory Digestive Organs** – responsible for the synthesis, storage and release of the digestive enzymes for the digestion of food.

**A. Salivary Glands**– exocrine glands in the oral cavity which secrete saliva for lubrication and digestion of food. They are compound tubulo-acinar glands and classified into two types: major and minor salivary glands. The major salivary glands are paired sets of glands: parotid, sublingual and submandibular or submaxillary.

#### Common Histological Features:

- 1. Capsule** – connective tissue rich in collagen fibers covering the glands. This may extend into the parenchyma of the glands and is called the septum.
- 2. Acini** – secretory units composed of cells seen as oval or spherical bodies. There are two types of acini:
  - 2.1. Serous** – basophilic structures whose cells are triangular in shape, the apex which surround a small lumen. The nuclei are round and located near the base of the cell.
  - 2.2. Mucous** – pale-staining bodies with cuboidal or columnar cells around a large lumen. The nuclei are oval and basal in position.
  - 2.3. Serous Demilune-** resembles a cap or D-shaped on the ends of mucous acini. They have identical histologic features as seen in serous acini devoid in lumen.
- 3. Myoepithelial Cells** – stellate cells with long cytoplasmic processes. Also called the **Basket cells**. They are found between the BM and the secretory cells. They are thought to facilitate the release of secretion products of the acini.
- 4. Duct System** – branching system of tubes for the release of the secretion of salivary gland. This is analogous to the branches of the tree. Identify the following parts:
  - a. Intercalated Ducts** – short tubes with cuboidal cells in direct contact with the secretory part of the gland.
  - b. Striated Ducts** – larger tubes formed by the fusion of several intercalated ducts. These appear as oval structures with dilated lumen. They are lined with columnar epithelium with radial striations on its basal cytoplasm. The intercalated ducts and striated ducts are also called intralobular ducts by virtue of their location in the gland.
  - c. Interlobular D** – main duct into which the striated ducts drain their contents. They are lined with stratified cuboidal with columnar epithelial in the distal part.

**d. Primary duct** – formed by the fusion of interlobular ducts which then empties into the oral cavity. The names vary according to the organ.

### Diagnostic Features of the Major Salivary Glands

- 1. Parotid Gland** – characterized by the presence of serous acini only. Have long interlobular ducts and few striated ducts. They secrete the enzyme amylase and IgA.
- 2. Sublingual Gland** – contain mostly mucous acini. Serous cells are also present as demilunes in association with mucous acini.
- 3. Submaxillary or Submandibular Gland** – has mixed serous and mucous acini but more predominant serous acini. They have few serous demilunes which secrete lysosomes and lactoferrin.

**B. Pancreas** – small elongated organ ventral to the stomach. It is a dual organ, with exocrine and endocrine portions secreting digestive enzymes and hormones, respectively.

#### Histological Features:

**1. Exocrine Pancreas** – a compound tubular-alveolar gland subdivided into several lobes by connective tissue septa. Each lobe is likewise subdivided into indistinct lobules.

**a. Acini** – functional units of the exocrine pancreas consist of several pyramidal cells that are clustered around a lumen. The apical cytoplasm of the cells appears lighter with many zymogen granules while the basal cytoplasm appears more basophilic. The nuclei are round and euchromatic.

**b. Centroacinar Cells** - one or two pale cells located on the lumen of the gland and directly connected to the intercalated ducts. These cells are found exclusively in the pancreas

**c. Islets of Langerhans** – a round mass of light-stained cells randomly distributed among the exocrine acini. The different cell types of the endocrine cells are difficult to distinguish with ordinary H and E preparation. However, they are classified into four types: alpha, beta, D and EC cells. **What hormones do they synthesize?**

**2. Ducts** – a round or tubular structures which convey the pancreatic enzymes to the duodenum.

**a. Intralobular or Intercalated Duct** – smallest branch of the main duct lined by squamous or cuboidal epithelium.

**b. Interlobular** – seen as elongated duct in the connective tissue septa. It is composed of simple columnar epithelium.

**c. Pancreatic Duct** – main duct lined with simple columnar and with few goblet cells near duodenum.

**C. Liver** – largest exocrine gland composed of 3 or 4 lobes. Under LPO, study a transverse section of the liver and identify the following:

1. **Glisson's Capsule** – a thick fibrous connective tissue surrounding the organ.
2. **Porta Hepatis** – central hilus or fissure for the entry of vessels and nerves. It is also the exit point of the ducts.
3. **Portal Canal** – connective tissue septum between the lobules that supports the smaller tributaries of blood vessels and ducts. Examine the components of portal canal under HPO.
  - ❖ **Portal Vein** – largest structure with a collapsed lumen. It has a typical vein structure.
  - ❖ **Hepatic Artery** – round and medium-sized structure. Has typical arterial structure. This conveys blood to the central vein.
  - ❖ **Hepatic Duct** – smallest among the three structures and lined with simple cuboidal or columnar cells.

These three structures are collectively known as the **Portal Triad**. They represent the corners of the classical lobule.

**4. Lobule** – considered as the functional unit of the liver. Subdivisions of the liver may appear hexagonal or triangular in transverse sections. If you have observed a hexagonal profile of the lobule, this is the so-called *classical lobule*.

- ❖ **Central Vein** – literally, a large vein at the center of the lobule. It is seen as round space with thin walls, surrounding it are the hepatocytes disposed radially.
- ❖ **Hepatic Plates** – linear array of hepatocytes that seem to radiate from the central vein. They are interconnected with each other like cords, thus, also called the **hepatic cords**. Under HPO, examine closely a single hepatocyte. Note that the cell is cuboidal or polygonal. It has a round central nucleus with euchromatic nucleus. It is not uncommon to find some cells that are binucleated.
- ❖ **Sinusoids** – irregular spaces between the hepatic plates. The presence of these structures account for the overall netlike appearance of the lobule.

The thin walls are really composed of attenuated cytoplasm of endothelial cells. Their nuclei may protrude or flattened against the hepatic plates.

- ❖ **Kupffer Cells** – phagocytic cells that are variable in shape and with elongated nucleus. They are usually lying within the sinusoids.

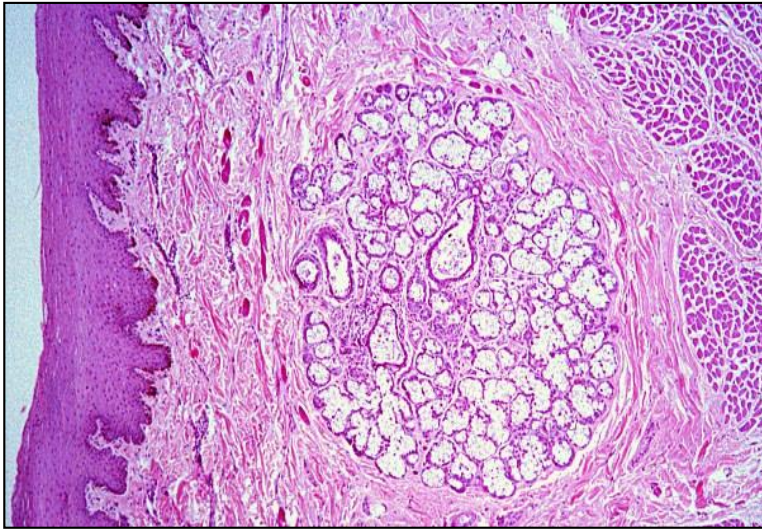
**Bile Canaliculi** – often viewed as thin dark line between two cells. In reality, they are minute channels between adjacent hepatocytes. They collect bile from the cells and convey it to the hepatic duct.

**D. Gall Bladder** – a small, green, pear-shaped organ for storage of bile. It has the same histologic feature as the other digestive organs except that it lacks submucosal region.

1. **Mucosa** – composed only of one cell type, one layer of extremely tall columnar cells with prominent striated border. The mucosal folds are highly variable in height, shape, and thickness.
2. **Lamina Propria** – broader and thicker than in other digestive organs.
3. **Muscularis Externa** – has thin inner longitudinal layer and another circular muscle layer.
4. **Adventitia** – thick layer consisting of connective tissue with blood vessels, nerves and many adipose tissues. A thin mesothelium surrounds this layer.

Note that there is no submucosa in this organ. ***What might take its place?***

Illustrations



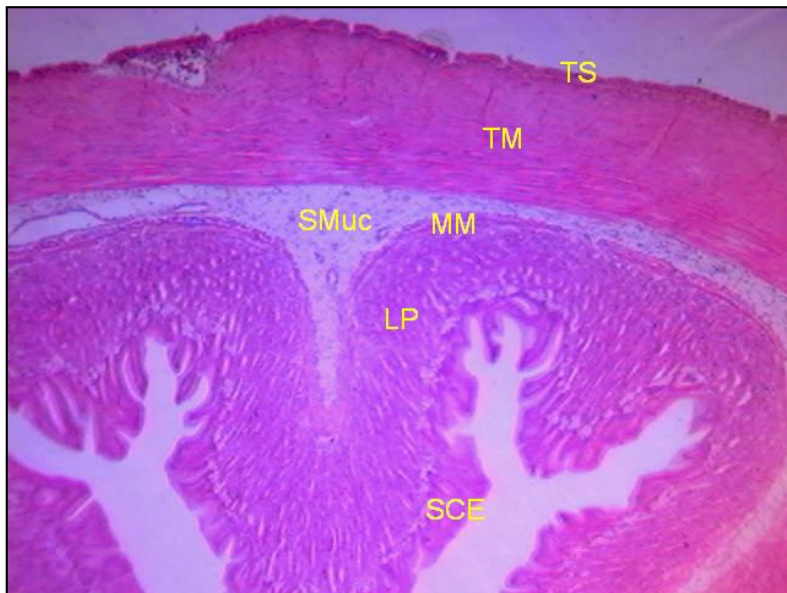
Lip



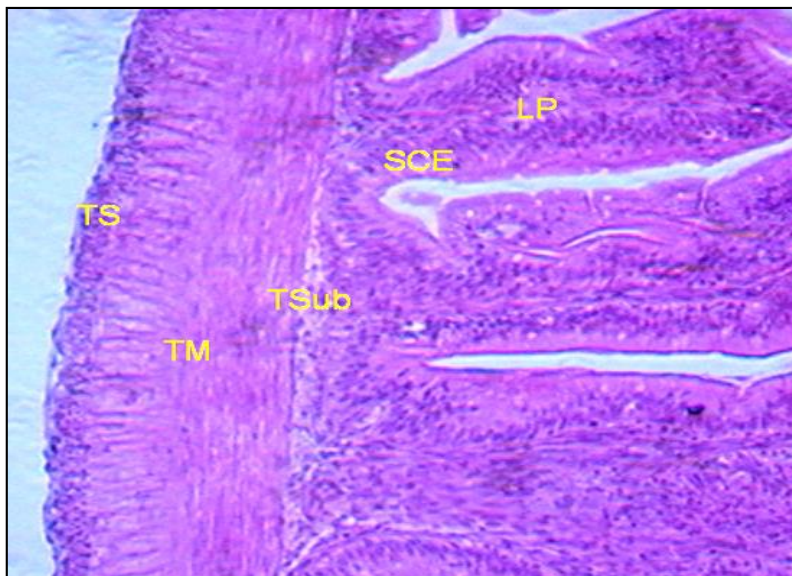
Tooth



Esophagus



Stomach



Small intestine



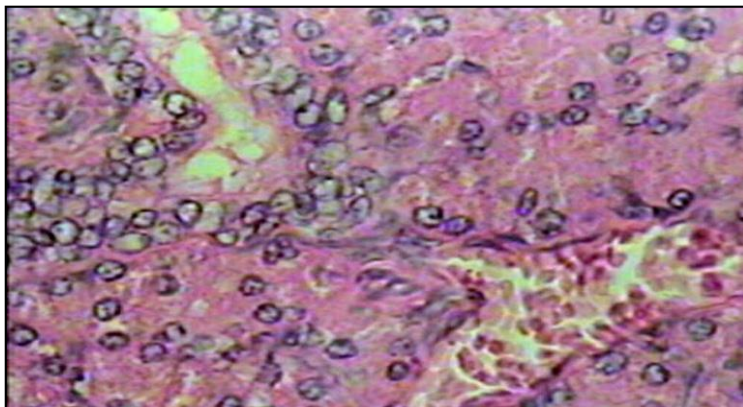
Large intestine



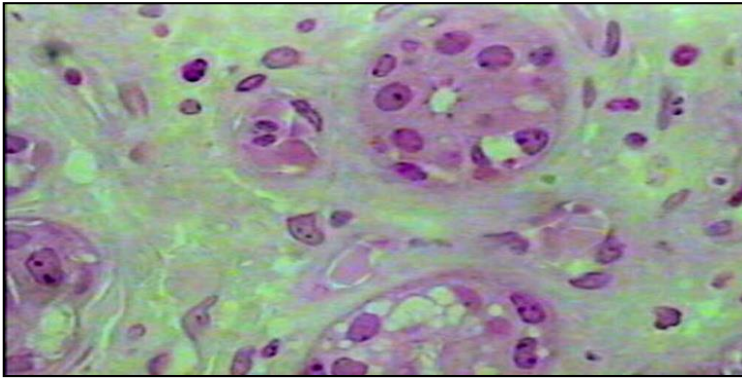
Appendix



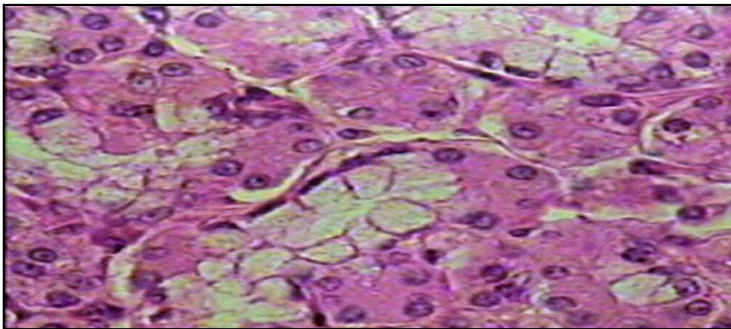
Rectum



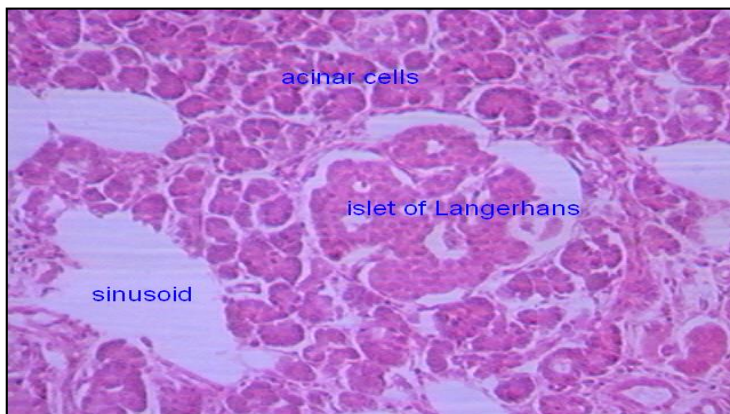
Parotid gland



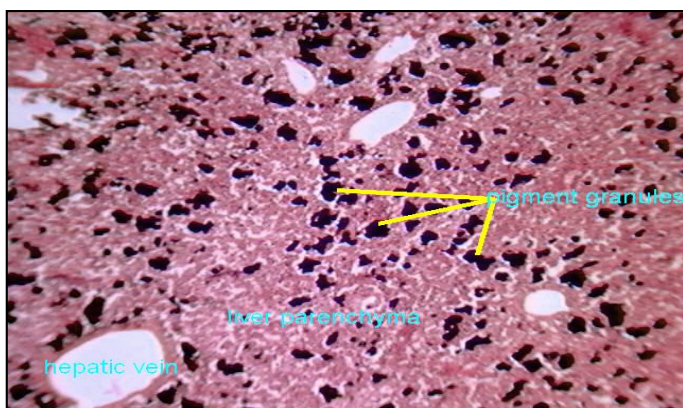
Submandibular gland



Sublingual gland

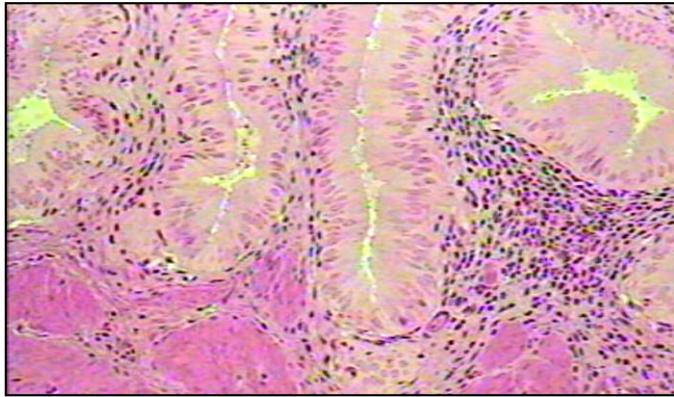


Pancreas



Liver





Gall bladder

### Guide Questions

1. Distinguish the following structures as observed under the light microscope.  
Plica circularis and rugae

---

---

Villi and microvilli

---

---

Peyer's patches and Meissner's plexus

---

---

---

2. Draw and label the following cells:

a. Odontoblast

b. Ameloblast

c. Parietal cell

d. Paneth's cell