Chapter 5

Digestion and Absorption

The Stomach

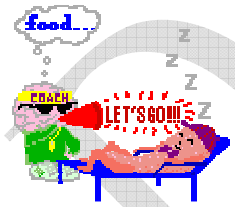
The stomach absorbs very few substances, although small amounts of certain lipid-soluble compounds can be taken up, including aspirin, other non-steroidal anti-infammatory drugs, and ethanol.

Notably, these substances are also well-recognized causes of gastric irritation and their use (especially overuse) is commonly associated with development of gastritis and gastric ulcers.

One Meal in the Life of the Stomach

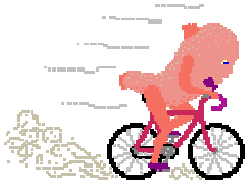
The stomach functions dynamically, in parallel with meals. Consider the stomach's most notable activity - secretion of acid. Acid is secreted in large quantities when the stomach is distended with food, which is useful because it facilitates the initial breakdown of proteins. However, once the meal has been liquefied and the stomach has emptied, acid secretion trickles to a stop and remains shut off during the interdigestive period. This shut-off in acid secretion is a good thing - otherwise excessive acid would damage the mucosa of the stomach and small intestine, as happens in certain disease states.

Gastric function is often classified into three phases in which secretory and motor activities are tightly coupled. Try identifying these phases in yourself or your loved ones around meal time:



Cephalic phase ("wake up call"): Seeing, smelling and anticipating food in perceived in the brain and the brain informs the stomach that it should prepare for receipt of a meal.

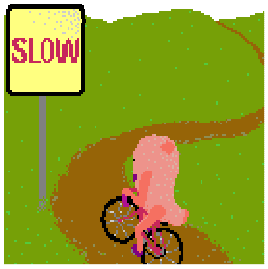
This communication is composed of parasympathetic stimuli transmitted through the vagus nerve to the enteric nervous system, resulting in release of acetylcholine in the vicinity of G cells and parietal cells. Binding of acetylcholine to its receptor on G cells induces secretion of the hormone gastrin, which, in concert with acetylcholine and histamine, stimulates parietal cells to secrete small amounts of acid. Additionally, a low level of gastric motility is induced. *In essense, the gastric motor is turned on and begins to idle.*



Gastric phase ("full steam ahead"): When a meal enters the stomach several additional factors come into play, foremost among them distension and mucosal irritation.

Distension excites stretch receptors and irritation activates chemoreceptors in the mucosa. These events are sensed by enteric neurons, which secrete additional acetylcholine, further

stimulating both G cells and parietal cells; gastrin from the G cells feeds back to the parietal cells, stimulating it even further. Additionally, activation of the enteric nervous system and release of gastrin cause vigorous smooth muscle contractions. The net result is that secretory and motor functions of the stomach are fully turned on - lots of acid and pepsinogen are secreted, pepsinogen is converted into pepsin and vigorous grinding and mixing contractions take place. However, there is a mechanism in place in the stomach to prevent excessive acid secretion - if lumenal pH drops low enough (less than about 2), motility and secretion are temporarily suspended.



Intestinal phase ("step on the brakes"): As food is liquefied in the stomach, it is emptied into the small intestine. Its seems to be important for the small intestine to be able to slow down gastric emptying, probably to allow it time to neutralize the acid and efficiently absorb incoming nutrients.

Hence, this phase of gastric function is dominated by the small intestine sending inhibitory signals to the stomach to slow secretion and motility. Two types of signals are used: nervous and endocrine. Distension of the small intestine, as well as chemical and osmotic irritation of the mucosa is transduced into gastric-inhibitory impulses in the enteric nervous system - this nervous pathway is called the enterogastric reflex. Secondly, enteric hormones such as cholecystokinin and secretin are released from cells in the small intestine and contribute to suppression of gastric activity.

Collectively, enteric hormones and the enterogastric reflex put a strong brake on gastric secretion and motility. As the ingesta in the small intestine is processed, these stimuli diminish, the damper on the stomach is released, and its secretory and motor activities resume.

To summarize, the brain alerts the stomach that it should expect arrival of a meal and the stomach comes out of its interdigestive quiescence and begins low level motor and secretory activity (cephalic phase). After a meal is consumed, the gastric motor and secretory activity is fully turned on (gastric phase). If the meal is at all substantial, the gastric phase is periodically suppressed by signals from the small intestine and, if gastric pH falls to very low levels, from the stomach itself. Eventually, the meal is fully liquefied and emptied, and the stomach falls back into a state of very low motor and secretory activity, where it remains until the next cephalic phase.

Secretion of Bile and the Role of Bile Acids In Digestion

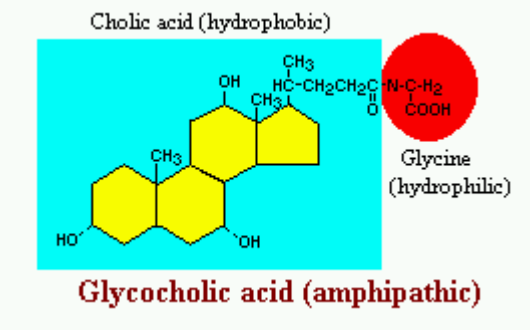
Bile is a complex fluid containing water, electrolytes and a battery of organic molecules including bile acids, cholesterol, phospholipids and bilirubin that flows through the biliary tract into the small intestine. **There are two fundamentally important functions of bile in all species:**

- Bile contains bile acids, which are critical for digestion and absorption of fats and fat-soluble vitamins in the small intestine.
- Many waste products, including bilirubin, are eliminated from the body by secretion into bile and elimination in feces.

Adult humans produce 400 to 800 ml of bile daily, and other animals proportionately similar amounts. The secretion of bile can be considered to occur in two stages:

- Initially, hepatocytes secrete bile into canaliculi, from which it flows into bile ducts. This hepatic bile contains large quantities of bile acids, cholesterol and other organic molecules.
- As bile flows through the bile ducts it is modified by addition of a watery, bicarbonate-rich secretion from ductal epithelial cells.

In species with a gallbladder (man and most domestic animals except horses and rats), further modification of bile occurs in that organ. **The gall bladder stores and concentrates bile during the fasting state.** Typically, bile is concentrated five-fold in the gall bladder by absorption of water and small electrolytes - virtually all of the the organic molecules are retained.



Secretion into bile is a major route for eliminating cholesterol. Free cholesterol is virtually insoluble in aqueous solutions, but in bile, it is made soluble by bile acids and lipids like lethicin. Gallstones, most of which are composed predominantly of cholesterol, result from processes that allow cholesterol to precipitate from solution in bile.

Role of Bile Acids in Fat Digestion and Absorption

Bile acids are derivatives of cholesterol synthesized in the hepatocyte. Cholesterol, ingested as part of the diet or derived from hepatic synthesis is converted into the bile acids cholic and chenodeoxycholic acids, which are then conjugated to an amino acid (glycine or taurine) to yield the conjugated form that is actively secreted into cannaliculi.

Bile acids are facial amphipathic, that is, they contain both hydrophobic (lipid soluble) and polar (hydrophilic) faces. The cholesterol-derived portion of a bile acid has one face that is hydrophobic (that with methyl groups) and one that is hydrophilic (that with the hydroxyl groups); the amino acid conjugate is polar and hydrophilic.

Their amphipathic nature enables bile acids to carry out two important functions:

- **Emulsification of lipid aggregates:** Bile acids have detergent action on particles of dietary fat which causes fat globules to break down or be emulsified into minute, microscopic droplets. Emulsification is not digestion per se, but is of importance because it greatly increases the surface area of fat, making it available for digestion by lipases, which cannot access the inside of lipid droplets.
- **Solubilization and transport of lipids in an aqueous environment:** Bile acids are lipid carriers and are able to solubilize many lipids by forming **micelles** - aggregates of lipids such as fatty acids, cholesterol and monoglycerides - that remain suspended

in water. Bile acids are also critical for transport and absorption of the fat-soluble vitamins.

Role of Bile Acids in Cholesterol Homeostasis

Hepatic synthesis of bile acids accounts for the majority of cholesterol breakdown in the body. In humans, roughly 500 mg of cholesterol are converted to bile acids and eliminated in bile every day. This route for elimination of excess cholesterol is probably important in all animals, but particularly in situations of massive cholesterol ingestion.

Interestingly, it has recently been demonstrated that bile acids participate in cholesterol metabolism by functioning as hormones that alter the transcription of the rate-limiting enzyme in cholesterol biosynthesis.

Enterohepatic Recirculation

Large amounts of bile acids are secreted into the intestine every day, but only relatively small quantities are lost from the body. This is because approximately 95% of the bile acids delivered to the duodenum are absorbed back into blood within the ileum.

Venous blood from the ileum goes straight into the portal vein, and hence through the sinusoids of the liver. Hepatocytes extract bile acids very efficiently from sinusoidal blood, and little escapes the healthy liver into systemic circulation. Bile acids are then transported across the hepatocytes to be resecreted into canaliculi. The net effect of this enterohepatic recirculation is that each bile salt molecule is reused about 20 times, often two or three times during a single digestive phase.

It should be noted that liver disease can dramatically alter this pattern of recirculation - for instance, sick hepatocytes have decreased ability to extract bile acids from portal blood and damage to the canalicular system can result in escape of bile acids into the systemic circulation. Assay of systemic levels of bile acids is used clinically as a sensitive indicator of hepatic disease.

Pattern and Control of Bile Secretion

The flow of bile is lowest during fasting, and a majority of that is diverted into the gallbladder for concentration. When chyme from an ingested meal enters the small intestine, acid and partially digested fats and proteins stimulate secretion of cholecystokinin and secretin. As discussed previously, these enteric hormones have important effects on pancreatic exocrine secretion. They are both also important for secretion and flow of bile:

- **Cholecystokinin**: The name of this hormone describes its effect on the biliary system - cholecysto = gallbladder and kinin = movement. The most potent stimulus for release of cholecystokinin is the presence of fat in the duodenum. Once released, it stimulates contractions of the gallbladder and common bile duct, resulting in delivery of bile into the gut.
- **Secretin**: This hormone is secreted in response to acid in the duodenum. Its effect on the biliary system is very similar to what was seen in the pancreas - it simulates biliary duct cells to secrete bicarbonate and water, which expands the volume of bile and increases its flow out into the intestine.

The processes of gallbladder filling and emptying described here can be visualized using an imaging technique called scintography. This procedure is utilized as a diagnostic aid in certain types of hepatobiliary disease.

Biliary Excretion of Waste Products: Elimination of Bilirubin

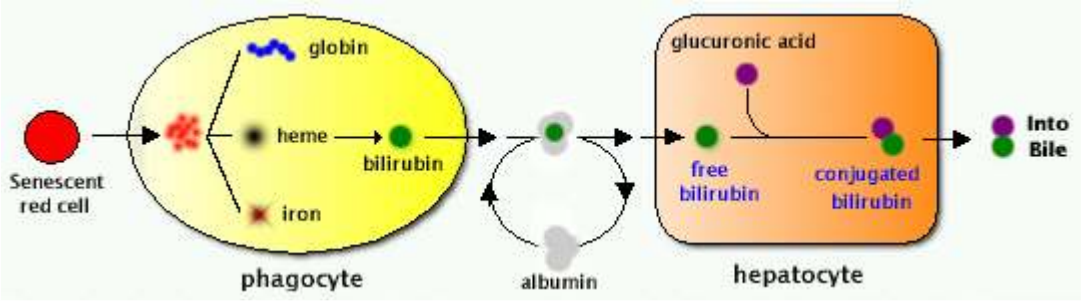
The liver is well known to metabolize and excrete into bile many compounds and toxins, thus eliminating them (usually) from the body. Examples can be found among both endogenous molecules (steroid hormones, calcium) and exogenous compounds (many antibiotics and metabolities of drugs). A substantial number of these compounds are reabsorbed in the small intestine and ultimately eliminated by the kidney.

One of the most important and clinically relevant examples of waste elimination via bile is that of bilirubin. Additionally, the mechanisms involved in elimination of bilirubin are similar to those used for elimination of many drugs and toxins.

Bilirubin is a useless and toxic breakdown product of hemoglobin, which also means that it is generated in large quantities. In the time it takes you to read this sentence aloud, roughly 20 million of your red blood cells have died and roughly 5 quintillion (5×10^{15}) molecules of hemoglobin are in need of disposal.

Dead, damaged and senescent red blood cells are picked up by phagocytic cells throughout the body (including Kuppfer cells in the liver) and digested. The iron is precious and is efficiently recycled. The globin chains are protein and are catabolized and their components reused. However, hemoglobin also contains a porphyrin called heme that cannot be recycled and must be eliminated. **Elimination of heme is accomplished in a series of steps:**

- Within the phagocytic cells, heme is converted through a series of steps into free bilirubin, which is released into plasma where it is carried around bound to albumin, itself a secretory product of the liver.
- Free bilirubin is stripped off albumin and absorbed by - you guessed it - hepatocytes. Within hepatocytes, free bilirubin is conjugated to either glucuronic acid or sulfate - it is then called conjugated bilirubin.
- Conjugated bilirubin is secreted into the bile canaliculus as part of bile and thus delivered to the small intestine. Bacteria in the intestinal lumen metabolize bilirubin to a series of other compounds which are ultimately eliminated either in feces or, after reabsortion, in urine. The major metabolite of bilirubin in feces is sterobilin, which gives feces their characteristic brown color.



If excessive quantities of either free or conjugated bilirubin accumulate in extracellular fluid, a yellow discoloration of the skin, sclera and mucous membranes is observed - this condition is called **icterus or jaundice**. Determining whether the excessive bilirubin is free or conjugated can aid in diagnosing the cause of the problem.

Metabolic Functions of the Live

Hepatocytes are metabolic superachievers in the body. They play critical roles in synthesizing molecules that are utilized elsewhere to support homeostasis, in converting molecules of one type to another, and in regulating energy balances. *If you have taken a course in biochemistry, you probably spent most of that class studying metabolic pathways of the liver.* At the risk of damning by faint praise, the major metabolic functions of the liver can be summarized into several major categories:

Carbohydrate Metabolism

It is critical for all animals to maintain concentrations of glucose in blood within a narrow, normal range. Maintainance of normal blood glucose levels over both short (hours) and long (days to weeks) periods of time is one particularly important function of the liver.

Hepatocytes house many different metabolic pathways and employ dozens of enzymes that are alternatively turned on or off depending on whether blood levels of glucose are rising or falling out of the normal range. Two important examples of these abilities are:

- Excess glucose entering the blood after a meal is rapidly taken up by the liver and sequestered as the large polymer, glycogen (a process called **glycogenesis**). Later, when blood concentrations of glucose begin to decline, the liver activates other pathways which lead to depolymerization of glycogen (**glycogenolysis**) and export of glucose back into the blood for transport to all other tissues.
- When hepatic glycogen reserves become exhaused, as occurs when an animal has not eaten for several hours, do the hepatocytes give up? No! They recognize the problem and activate additional groups of enzymes that begin synthesizing glucose out of such things as amino acids and non-hexose carbohydrates (**gluconeogenesis**). The ability of the liver to synthesize this "new" glucose is of monumental importance to carnivores, which, at least in the wild, have diets virtually devoid of starch.

Fat Metabolism

Few aspects of lipid metabolism are unique to the liver, but many are carried out predominantly by the liver. Major examples of the role of the liver in fat metabolism include:

- The liver is extremely active in oxidizing triglycerides to produce energy. The liver breaks down many more fatty acids that the hepatocytes need, and exports large quantities of acetoacetate into blood where it can be picked up and readily metabolized by other tissues.
- A bulk of the lipoproteins are synthesized in the liver.
- The liver is the major site for converting excess carbohydrates and proteins into fatty

acids and triglyceride, which are then exported and stored in adipose tissue.
- The liver synthesizes large quantities of cholesterol and phospholipids. Some of this is packaged with lipoproteins and made available to the rest of the body. The remainder is excreted in bile as cholesterol or after conversion to bile acids.

Protein Metabolism

The most critical aspects of protein metabolism that occur in the liver are:

- Deamination and transamination of amino acids, followed by conversion of the non-nitrogenous part of those molecules to glucose or lipids. Several of the enzymes used in these pathways (for example, alanine and aspartate aminotransferases) are commonly assayed in serum to assess liver damage.
- Removal of ammonia from the body by synthesis of urea. Ammonia is very toxic and if not rapidly and efficiently removed from the circulation, will result in central nervous system disease. A frequent cause of such hepatic encephalopathy in dogs and cats are malformations of the blood supply to the liver called portosystemic shunts.
- Synthesis of non-essential amino acids.
- Hepatocytes are responsible for synthesis of most of the plasma proteins. Albumin, the major plasma protein, is synthesized almost exclusively by the liver. Also, the liver synthesizes many of the clotting factors necessary for blood coagulation.

The Pancreas:

As chyme floods into the small intestine from the stomach, two things must happen:

- *acid must be quickly and efficiently neutralized* to prevent damage to the duodenal mucosa
- *macromolecular nutrients - proteins, fats and starch - must be broken down much further* before their constitutents can be absorbed through the mucosa into blood

The pancreas plays a vital role in accomplishing both of these objectives, so vital in fact that insufficient exocrine secretion by the pancreas leads to starvation, even if the animal is consuming adequate quantities of high quality food.

In addition to its role as an exocrine organ, the *pancreas is also an endocrine organ* and the major hormones it secretes - insulin and glucagon - play a vital role in carbohydrate and lipid metabolism. They are, for example, absolutely necessary for maintaining normal blood concentrations of glucose.

Gross and Microscopic Anatomy of the Pancreas

The pancreas is a elongated organ, light tan or pinkish in color, that lies in close proximity to the duodenum. It is covered with a very thin connective tissue capsule which extends inward as septa, partitioning the gland into lobules. The image below shows a canine pancreas in relation to the stomach and duodenumThe bulk of the pancreas is composed of *pancreatic exocrine cells* and their associated ducts. Embedded within this exocrine tissue are roughly one million small clusters of cells called the *Islets of Langerhans*, which are the endocrine cells of the pancreas and secrete insulin, glucagon and several other hormones. In the

histologic image of an equine pancreas seen below, a single islet is seen in the middle as a large, pale-staining cluster of cells. All of the surrounding tissue is exocrine. Pancreatic exocrine cells are arranged in grape-like clusters called acini (a single one is an acinus). The exocrine cells themselves are packed with membrane-bound secretory granules which contain digestive enzymes that are exocytosed into the lumen of the acinus. From there these secretions flow into larger and larger, intralobular ducts, which eventually coalesce into the main pancreatic duct which drains directly into the duodenum.

The lumen of an acinus communicates directly with intralobular ducts, which coalesce into interlobular ducts and then into the major pancreatic duct. Epithelial cells of the the intralobular ducts actually project "back" into the lumen of the acinus, where they are called centroacinar cells. The anatomy of the main pancreatic duct varies among species. In some animals, two ducts enter the duodenum rather than a single duct. In some species, the main pancreatic duct fuses with the common bile duct just before its entry into the duodenum.

Additional information on microscopic anatomy of the pancrease can be found in the section on Histology of the Pancreas.

Histology of the Pancreas

he structure of the pancreas is dominated by the fact that it is a dual function organ with both exocrine and endocrine cell types. The vast bulk of the pancreas is composed of exocrine tissue, and secretions from those cells flow into a series ducts for ultimate delivery into the duodenum. Scattered within the exocrine tissue are clumps of cells that secrete hormones into blood.

The pancreas is surrounded by a very thin connective tissue capsule that invaginates into the gland to form septae, which serve as scaffolding for large blood vessels. Further, these septae divide the pancreas into distinctive lobules**, as can clearly be seen in the image of mouse pancreas below (H&E).**

The Acinus

The exocrine pancreas is classified as a compound tubuloacinous gland. **The cells that synthesize and secrete digestive enzymes are arranged in grape-like clusters called acini**, very similar to what is seen in salivary glands. In standard histologic sections, most acini are cut obliquely, making it difficult to discern their characteristic shape. In the image of equine pancreas below, one fairly-good cross section through an acinus is circled; note the wedge-shapped cells arranged around a small lumen:

Pancreatic Ducts

Digestive enzymes from acinar cells ultimately are delivered into the duodenum. Secretions from acini flow out of the pancreas through a tree-like series of ducts. Duct cells secrete a watery, bicarbonate-rich fluid which flush the enzymes through the ducts and play a pivotal role in neutralizing acid within the small intestine. Pancreatic ducts are classified into four types which are discussed here beginning with the terminal branches which extend into acini.

Intercalated ducts receive secretions from acini. They have flattened cuboidal epithelium that extends up into the lumen of the acinus to form what are called**centroacinar cells**.

Intralobular ducts have a classical cuboidal epithelium and, as the name implies, are seen within lobules. They receive secretions from intercalated ducts.

Interlobular ducts are found between lobules, within the connective tissue septae. They vary considerably in size. The smaller forms have a cuboidal epithelium, while a columnar epithelium lines the larger ducts. Intralobular ducts transmit secretions from intralobular ducts to the major pancreatic duct.

The main pancreatic duct received secretion from interlobular ducts and penetrates through the wall of the duodenum. In some species, including man, the pancreatic duct joins the bile duct prior to entering the intestine.

Exocrine Secretions of the Pancreas

Pancreatic juice is composed of two secretory products critical to proper digestion: digestive enzymes and bicarbonate. The enzymes are synthesized and secreted from the exocrine acinar cells, whereas bicarbonate is secreted from the epithelial cells lining small pancreatic ducts.

Digestive Enzymes

The pancreas secretes a magnificent battery of enzymes that collectively have the capacity to reduce virtually all digestible macromolecules into forms that are capable of, or nearly capable of being absorbed. Three major groups of enzymes are critical to efficient digestion:

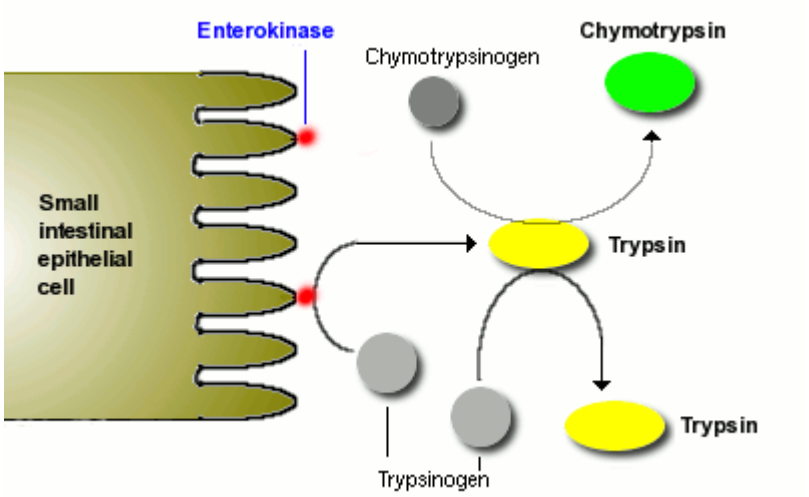
1. Proteases

Digestion of proteins is initiated by pepsin in the stomach, but the bulk of protein digestion is due to the pancreatic proteases. Several proteases are synthesized in the pancreas and secreted into the lumen of the small intestine. The two major pancreatic proteases are **trypsin** and**chymotrypsin**, which are synthesized and packaged into secretory vesicles as an the inactive proenzymes trypsinogen and chymotrypsinogen.

As you might anticipate, proteases are rather dangerous enzymes to have in cells, and packaging of an inactive precursor is a way for the cells to safely handle these enzymes. The secretory vesicles also contain a trypsin inhibitor which serves as an additional safeguard should some of the trypsinogen be activated to trypsin; following exocytosis this inhibitor is diluted out and becomes ineffective - the pin is out of the grenade.

Once trypsinogen and chymotrypsinogen are released into the lumen of the small intestine, they must be converted into their active forms in order to digest proteins. Trypsinogen is activated by the enzyme **enterokinase**, which is embedded in the intestinal mucosa.

Once trypsin is formed it activates chymotrypsinogen, as well as additional molecules of trypsinogen. The net result is a rather explosive appearance of active protease once the pancreatic secretions reach the small intestine.

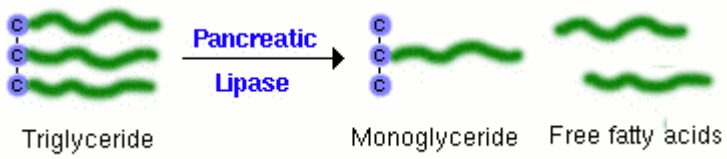


Trypsin and chymotrypsin digest proteins into peptides and peptides into smaller peptides, but they cannot digest proteins and peptides to single amino acids. Some of the other proteases from the pancreas, for instance carboxypeptidase, have that ability, but the final digestion of peptides into amino acids is largely the effect of peptidases on the surface of small intestinal epithelial cells. More on this later.

2. Pancreatic Lipase

A major component of dietary fat is triglyceride, or neutral lipid. A triglyceride molecule cannot be directly absorbed across the intestinal mucosa. Rather, it must first be digested into a 2-monoglyceride and two free fatty acids. The enzyme that performs this hydrolysis is pancreatic lipase, which is delivered into the lumen of the gut as a constituent of pancreatic juice.

Sufficient quantities of bile salts must also be present in the lumen of the intestine in order for lipase to efficiently digest dietary triglyceride and for the resulting fatty acids and monoglyceride to be absorbed. This means that normal digestion and absorption of dietary fat is critically dependent on secretions from both the pancreas and liver.



Pancreatic lipase has recently been in the limelight as a target for management of obesity. The drug orlistat (Xenical) is a pancreatic lipase inhibitor that interferes with digestion of triglyceride and thereby reduces absorption of dietary fat. Clinical trials support the contention that inhibiting lipase can lead to significant reductions in body weight in some patients.

3. Amylase

The major dietary carbohydrate for many species is starch, a storage form of glucose in plants. Amylase (technically alpha-amylase) is the enzyme that hydrolyses starch to maltose (a glucose-glucose disaccharide), as well as the trisaccharide maltotriose and small branchpoints fragments called limit dextrins. The major source of amylase in all species is pancreatic secretions, although amylase is also present in saliva of some animals, including humans.

Other Pancreatic Enzymes

In addition to the proteases, lipase and amylase, the pancreas produces a host of other digestive enzymes, including ribonuclease, deoxyribonuclease, gelatinase and elastase.

Bicarbonate and Water

Epithelial cells in pancreatic ducts are the source of the bicarbonate and water secreted by the pancreas. Bicarbonate is a base and critical to neutralizing the acid coming into the small intestine from the stomach. The mechanism underlying bicarbonate secretion is essentially the same as for acid secretion parietal cells and is dependent on the enzymecarbonic anhydrase. In pancreatic duct cells, the bicarbonate is secreted into the lumen of the duct and hence into pancreatic juice.

Control of Pancreatic Exocrine Secretion

As you might expect, secretion from the exocrine pancreas is regulated by both neural and endocrine controls. During interdigestive periods, very little secretion takes place, but as food enters the stomach and, a little later, chyme flows into the small intestine, pancreatic secretion is strongly stimulated.

Like the stomach, the pancreas is innervated by the vagus nerve, which applies a low level stimulus to secretion in response to anticipation of a meal. However, the most important stimuli for pancreatic secretion comes from three hormones secreted by theenteric endocrine system:

- Cholecystokinin: This hormone is synthesized and secreted by enteric endocrine cells located in the duodenum. Its secretion is strongly stimulated by the presence of partially digested proteins and fats in the small intestine. As chyme floods into the small intestine, cholecystokinin is released into blood and binds to receptors on pancreatic acinar cells, ordering them to secrete large quantities of digestive enzymes.
- Secretin: This hormone is also a product of endocrinocytes located in the epithelium of the proximal small intestine. Secretin is secreted (!) in response to acid in the duodenum, which of course occurs when acid-laden chyme from the stomach flows through the pylorus. The predominant effect of secretin on the pancreas is to stimulate duct cells to secrete water and bicarbonate. As soon as this occurs, the enyzmes secreted by the acinar cells are flushed out of the pancreas, through the pancreatic duct into the duodenum.
- Gastrin: This hormone, which is very similar to cholecystokinin, is secreted in large amounts by the stomach in response to gastric distention and irritation. In addition to stimulating acid secretion by the parietal cell, gastrin stimulates pancreatic acinar cells to secrete digestive enzymes.

Stop and think about this for a minute - control of pancreatic secretion makes perfect sense. Pancreatic secretions contain enzymes which are needed to digest proteins, starch and triglyceride. When these substances enter stomach, and especially the small intestine, they stimulate release of gastrin and cholecystokinin, which in turn stimulate secretion of the enzymes of destruction.

Pancreatic secretions are also the major mechanism for neutralizing gastric acid in the small intestine. When acid enters the small gut, it stimulates secretin to be released, and the effect of this hormone is to stimulate secretion of lots of bicarbonate. As proteins and fats are digested and absorbed, and acid is neutralized, the stimuli for cholecystokinin and secretin secretion disappear and pancreatic secretion falls off.

The Small Intestine

he small intestine is the portal for absorption of virtually all nutrients into blood. Accomplishing this transport entails breaking down large supramolecular aggregates into small molecules that can be transported across the epithelium. An exception to this statement is seen in herbivores, where large amounts of short chain fatty acids are absorbed at other sites.

By the time ingesta reaches the small intestine, foodstuffs have been mechanically broken down and reduced to a liquid by mastication and grinding in the stomach. Once within the small intestine, these macromolecular aggregates are exposed to pancreatic enzymesand bile, which enables digestion to molecules capable or almost capable of being absorbed. The final stages of digestion occur on the surface of the small intestinal epithelium.

The net effect of passage through the small intestine is absorption of most of the water and electrolytes (sodium, chloride, potassium) and essentially all dietary organic molecules (including glucose, amino acids and fatty acids). Through these activities, the small intestine not only provides nutrients to the body, but plays a critical role in water and acid-base balance.

Gross and Microscopic Anatomy of the Small Intestine

The small intestine is the longest section of the digestive tube and consists of three segments forming a passage from the pylorus to the large intestine:

- **Duodenum:** a short section that receives secretions from the pancreas and liver via the **pancreatic and common bile ducts**.
- **Jejunum:** considered to be roughly 40% of the small gut in man, but closer to 90% in animals.
- **Ileum** empties into the large intestine; considered to be about 60% of the intestine in man, but veterinary anatomists usually refer to it as being only the short terminal section of the small intestine.

In most animals, the length of the small intestine is roughly 3.5 times body length - your small intestine, or that of a large dog, is about 6 meters in length. Although precise boundaries

between these three segments of bowel are not observed grossly or microscopically, there are histologic differences among duodenum, jejunum and ileum.

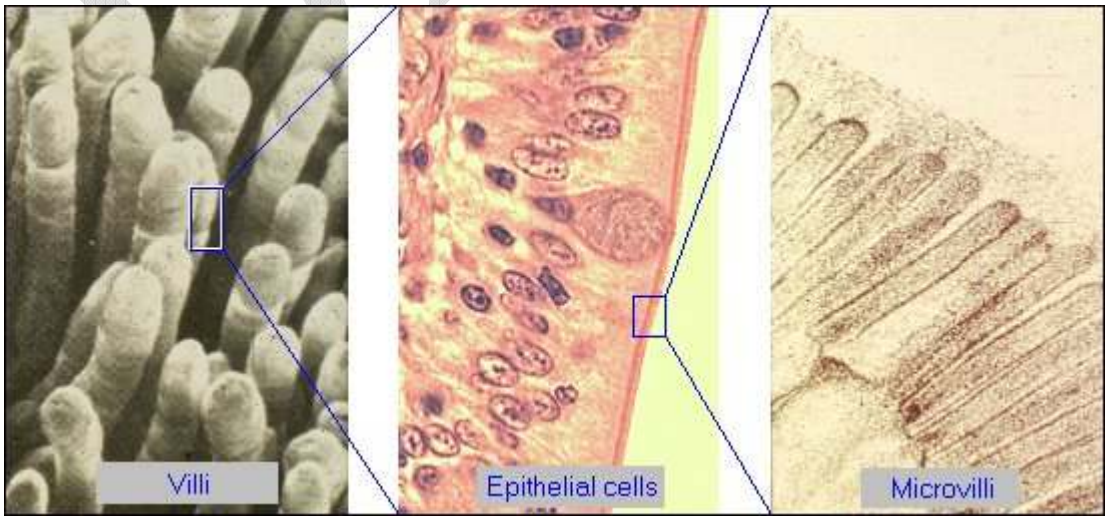
A bulk of the small intestine is suspended from the body wall by an extension of the peritoneum called the mesentery. As seen in the image to the right, blood vessels to and from the intestine lie between the two sheets of the mesentery. Lymphatic vessels are also present, but are not easy to discern grossly in normal specimens.

It is within the small intestine that the final stages of enzymatic digestion occur, liberating small molecules capable of being absorbed. The small intestine is also the sole site in the digestive tube for absorption of amino acids and monosaccharides. Most lipids are also absorbed in this organ. All of this absorption and much of the enzymatic digestion takes place on the surface of small intestinal epithelial cells, and to accomodate these processes, a huge mucosal surface area is required.

If the small intestine is viewed as a simple pipe, its lumenal surface area would be on the order of one half of a square meter. But in reality, the absorptive surface area of the small intestine is roughly 250 square meters - the size of a tennis court! How is this possible? At first glance, the structure of the small intestine is similar to other regions of the digestive tube, but the small intestine incorporates three features which account for its huge absorptive surface area:

- **Mucosal folds:** the inner surface of the small intestine is not flat, but thrown into circular folds, which not only increase surface area, but aid in mixing the ingesta by acting as baffles.
- **Villi:** the mucosa forms multitudes of projections which protrude into the lumen and are covered with epithelial cells.
- **Microvilli:** the lumenal plasma membrane of absorptive epithelial cells is studded with densely-packed microvilli.

The panels below depict the bulk of this surface area expansion, showing villi, epithelial cells that cover the villi and the microvilli of the epithelial cells. Note in the middle panel, a light micrograph, that the microvilli are visible and look something like a brush. For this reason, the microvillus border of intestinal epithelial cells is referred to as the **"brush border"**.



Most of the discussion on following pages focuses on enterocytes, the epithelial cells which mature into absorptive epithelial cells that cover the villi. These are the cells that take up and deliver into blood virtually all nutrients from the diet. However, two other major cell types populate the small intestinal epithelium:

- **Enteroendocrine cells** which, as part of the enteric endocrine system sense the lumenal environment and secrete hormones such as cholecystokinin and gastrin into blood.
- **Goblet cells**, which secrete a lubricating mucus into the intestinal lumen.

Villi, Crypts and the Life Cycle of Small Intestinal Enterocytes

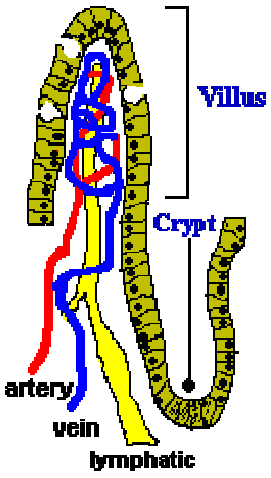
If examined closely, the lumenal surface of the small intestine appears similar to velvet due to its being covered by millions of small projections called villi which extend about 1 mm into the lumen. Villi are only the most obvious feature of the mucosa which houses a dynamic, self-renewing population of epithelial cells that includes secretory cells, endocrine cells and the mature absorptive epithelial cells which take up nutrients from the lumen and transport them into blood, fulfilling the basic function of the digestive system. Understanding how the small intestine functions requires looking at the structure of the mucosa in more detail.

Epithelial Cell Dynamics

The mucosa of small intestinal mucosa is arranged into two fundamental structures:

Villi are projections into the lumen covered predominantly with mature, absorptive enterocytes, along with occasional mucus-secreting goblet cells. These cells live only for a few days, die and are shed into the lumen to become part of the ingesta to be digested and absorbed. That's right, we're all really cannibals.

Crypts (of Lieberkuhn) are moat-like invaginations of the epithelium around the villi, and are lined largely with younger epithelial cells which are involved primarily in secretion. Toward the base of the crypts are stem cells, which continually divide and provide the source of all the epithelial cells in the crypts and on the villi.



The system described above is really quite elegant. Stem cells in the crypts divide to form daughter cells. One daughter cell from each stem cell division is retained as a stem cell. The other becomes committed to differentiate along one of four pathways to become an enterocyte, enteroendocrine cell, goblet cell or Paneth cell. Cells in the enterocyte lineage divide several more times as they migrate up the crypts, and as they migrate onto the villi,

differentiate further into the mature absorptive cells that express all the transport proteins and enzymes characteristic of those cells. To put it another way, enterocytes are born at the bottom of the crypts, pass through childhood migrating up the walls of the crypts, then settle down briefly to enjoy an absorptive adulthood on the villi.

Inside the Villus

Virtally all nurtients, including all amino acids and sugars, enter the body across the epithelium covering small intestinal villi. As shown in the diagram above, each villus contains a **capillary bed** and a blunt-ended lymphatic vessel referred to as the **"central lacteal"**.

After crossing the epithelium, most of these molecules diffuse into a capillary network inside the villus, and hence into systemic blood. Some molecules, fats in particular, are transported not into capillaries, but rather into the lymphatic vessel, which drains from the intestine and rapidly flows into blood via the thoracic duct.

Details of the transport of major nutrients into the capillaries or lymphatics in the villi are presented in subsequent sections.

Small Intestinal Motility

Coordinated contractions of smooth muscle participate in several ways to facilitate digestion and absorption in the small intestine:

- foodstuffs are mixed with digestive enzymes from the pancreas and bile salts from the biliary system
- nutrient molecules in the lumen are constantly dispersed, allowing them to contact the epithelium where enzymatic digestion is completed and absorption occurs
- chyme is moved down the digestive tube, making way for the next load and also eliminating undigestable, perhaps toxic substances

In most animals, the small intestine cycles through two states, each of which is associated with distinctive patterns of motility:

- **Following a meal**, when the lumen of the small intestine contains chyme, two types of motility predominate: *segmentation contractions* chop, mix and roll the chyme and *peristalsis* slowly propels it toward the large intestine.
- **The interdigestive period** is seen between meals, when the lumen is largely devoid of contents. During such times, so-called housekeeping contractions propagate from the stomach through the entire small intestine, sweeping it clear of debris. This complex pattern of motility is also known as the migrating motor complex and is the cause of "growling".

Motility in the small intestine, as in all parts of the digestive tube, is controlled predominantly by excitatatory and inhibitory signals from the enteric nervous system. These local nervous signals are however modulated by inputs from the central nervous system, and a number of gastrointestinal hormones appear to affect intestinal motility to some degree.

Overview of Transport Across the Intestinal Epithelium

There are two routes for transport of molecules and ions across the epithelium of the gut:

- Across the plasma membrane of the epithelial cells (**transcellular route**)
- Across tight junctions between epithelial cells (**paracellular route**)

Some molecules, water for instance, are transported by both routes. In contrast, the tight junctions are impermeable to large organic molecules from the diet (e.g. amino acids and glucose). Those types of molecules are transported exclusively by the transcellular route, and only because the plasma membrane of the absorptive enterocytes is equipped with transporter molecules that facilitate entry into and out of the cells.

It is important to recognize that the epithelium of the gut is not a monotonous sheet of functionally identical cells. Additionally, tight junctions linking epithelial cells vary considerably in permiability along the gastrointestinal tract. As ingesta travels through the intestine, it is sequentially exposed to regions having epithelia with very different characteristics. This diversity in function results from differences in phenotype of the enterocytes - that is, the number and type of transporter molecules they express in their plasma membrane and the structure of the tight junctions they form. Even within a given segment there are major differences in the type of transport that occurs - for example, cells in the crypts transport very differently than cells on the tips of villi.

Within the intestine, there is a proximal to distal gradient in osmotic permiability. As you proceed down the tube, the effective pore size through the epithelium decreases. This means that the duodenum is much more "leaky" to water than the ileum and the ileum more leaky than the colon. Do not interpret this to mean that as you go down the tube, the ability to absorb water decreases! It means that water flows across the epithelium more "freely" in the proximal compared to distal gut because the effective pore size is larger. The distal intestine actually can absorb water better than the proximal gut.

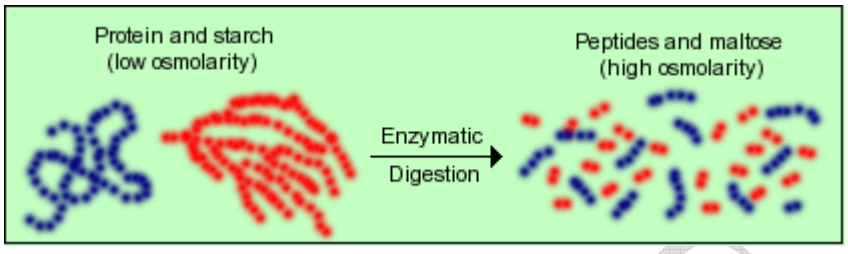
The observed differences in permiability to water across the epithelium is due almost entirely to differences in conductivity across the paracellular path - the takehome message is that tight junctions vary considerably in "tightness" along the length of the gut.

Secretion in the Small Intestine

Large quantities of water are secreted into the lumen of the small intestine during the digestive process. Almost all of this water is also reabsorbed in the small intestine. Regardless of whether it is being secreted or absorbed, water flows across the mucosa in response to osmotic gradients. In the case of secretion, two distinct processes establish an osmotic gradient that pulls water into the lumen of the intestine:

1. **Increases in luminal osmotic pressure resulting from influx and digestion of foodstuffs:** The chyme that floods into the intestine from the stomach typically is not terribly hyperosmotic, but as its macromolecular components are digested, osmolarlity of that solution increases dramatically.

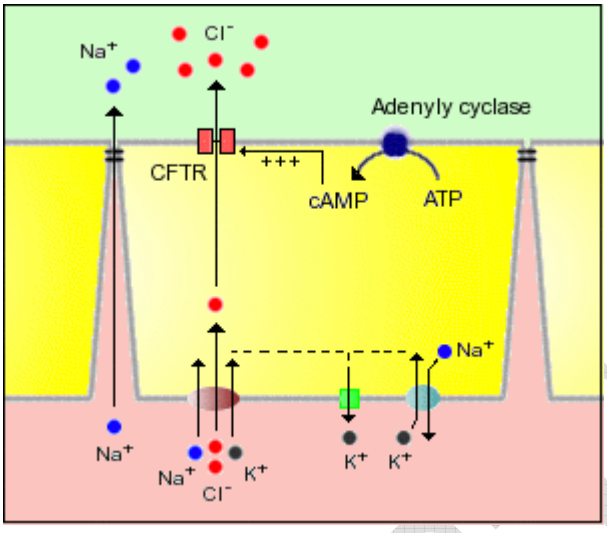
Starch, for example, is a huge molecule that contributes only a small amount to osmotic pressure, but as it is digested, thousands of molecules of maltose are generated, each of which is as osmotically active as the original starch molecule.



Thus, as digestion proceeds lumenal osmolarity increases dramatically and water is pulled into the lumen. Then, as the osmotically active molecules (maltose, glucose, amino acids) are absorbed, osmolarity of the intestinal contents decreases and water can be absorbed.

2. **Crypt cells actively secrete electrolytes, leading to water secretion:** The apical or lumenal membrane of crypt epithelial cells contain a ion channel of immense medical significance - a *cyclic AMP-dependent chloride channel known also as the cystic fibrosis transmembrane conductance regulator or CFTR.* Mutations in the gene for this ion channel result in the disease cystic fibrosis. This channel is responsible for secretion of water by the following steps:

- Chloride ions enter the crypt epithelial cell by cotransport with sodium and potassium; sodium is pumped back out via sodium pumps, and potassium is exported via a number of channels.
- Activation of adenylyl cyclase by a number of so-called secretagogues leads to generation of cyclic AMP.
- Elevated intracellular concentrations of cAMP in crypt cells activate the CFTR, resulting in secretion of chloride ions into the lumen.
- Accumulation of negatively-charged chloride anions in the crypt creates an electric potential that attracts sodium, pulling it into the lumen, apparently across tight junctions - the net result is secretion of NaCl.
- Secretion of NaCl into the crypt creates an osmotic gradient across the tight junction and water is drawn into the lumen.



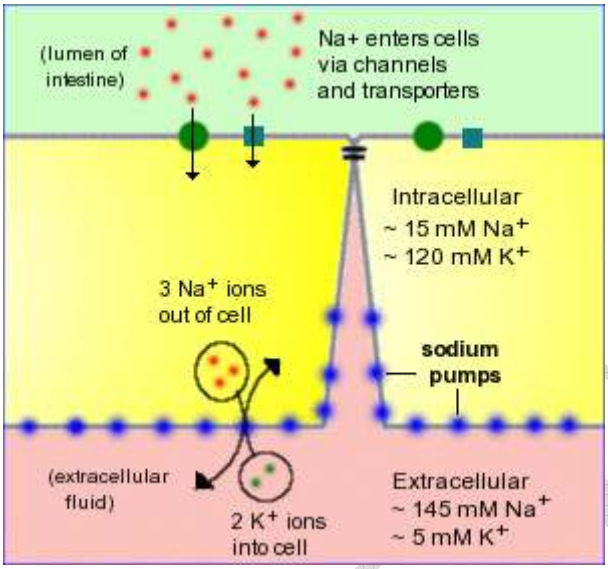
Abnormal activation of the cAMP-dependent chloride channel (CFTR) in crypt cells has resulted in the deaths of millions upon millions of people. Several types of bacteria produce toxins that strongly, often permanently, activate the adenylate cyclase in crypt enterocytes. This leads to elevated levels of cAMP, causing the chloride channels to essentially become stuck in the "open" position". The result is massive secretion of water that is manifest as severe diarrhea. Cholera toxin, produced by cholera bacteria, is the best known example of this phenomenon, but several other bacteria produce toxins that act similarly.

Absorption in the Small Intestine: General Mechanisms

Virtually all nutrients from the diet are absorbed into blood across the mucosa of the small intestine. In addition, the intestine absorbs water and electrolytes, thus playing a critical role in maintenance of body water and acid-base balance.

It's probably fair to say that *the single most important process that takes place in the small gut to make such absorption possible is establishment of an electrochemical gradient of sodium across the epithelial cell boundary of the lumen.* This is a critical concept and actually quite interesting. Also, as we will see, understanding this process has undeniably resulted in the saving of millions of lives.

To remain viable, all cells are required to maintain a low intracellular concentration of sodium. In polarized epithelial cells like enterocytes, low intracellular sodium is maintained by a large number of Na^+/K^+ ATPases - so-called sodium pumps - embedded in the basolateral membrane. These pumps export 3 sodium ions from the cell in exchange for 2 potassium ions, thus establishing a gradient of both charge and sodium concentration across the basolateral membrane.



In rats, as a model of all mammals, there are about 150,000 sodium pumps per small intestinal enterocyte which collectively allow each cell to transport about 4.5 billion sodium ions out of each cell per minute (J Membr Biol 53:119-128, 1980). Pretty impressive! This flow and accumulation of sodium is ultimately responsible for absorption of water, amino acids and carbohydrates.

Aside from the electrochemical gradient of sodium just discussed, several other concepts are required to understand absorption in the small intestine. Also, dietary sources of protein, carbohydrate and fat must all undergo the final stages of chemical digestion just prior to absorption of, for example, amino acids, glucose and fatty acids.

At this point, its easiest to talk separately about absorption of each of the major food groups, recognizing that all of these processes take place simultaneously.

- Water and electrolytes
- Carbohydrates, after digestion to monosaccharides
- Proteins, after digestion to small peptides and amino acids
- Neutral fat, after digestion to monoglyceride and free fatty acids

Absorption of Water and Electrolytes

The small intestine must absorb massive quantities of water. A normal person or animal of similar size takes in roughly 1 to 2 liters of dietary fluid every day. On top of that, another 6 to 7 liters of fluid is received by the small intestine daily as secretions from salivary glands, stomach, pancreas, liver and the small intestine itself.

By the time the ingesta enters the large intestine, approximately 80% of this fluid has been absorbed. Net movement of water across cell membranes always occurs byosmosis, and *the fundamental concept needed to understand absorption in the small gut is that there is a tight coupling between water and solute absorption.* Another way of saying this is that absorption of water is absolutely dependent on absorption of solutes, particularly sodium:

- Sodium is absorbed into the cell by several mechanisms, but chief among them is by cotransport with glucose and amino acids - this means that efficient sodium absorption is dependent on absorption of these organic solutes.
- Absorbed sodium is rapidly exported from the cell via sodium pumps - when a lot of sodium is entering the cell, a lot of sodium is pumped out of the cell, which establishes a high osmolarity in the small intercellular spaces between adjacent enterocytes.
- Water diffuses in response to the osmotic gradient established by sodium - in this case into the intercellular space. It seems that the bulk of the water absorption is transcellular, but some also diffuses through the tight junctions.
- Water, as well as sodium, then diffuses into capillary blood within the villus.

Examine the animation above and consider the osmotic gradient between the lumen and the intercellular space (inside the villus). As sodium (green balls) is rapidly pumped out of the cell, it achieves very high concentration in the narrow space between enterocytes. The osmotic gradient is thus formed across apical cell membranes and their connecting junctional complexes. The arrow that appears denotes movement of water across the epithelium.

Water is thus absorbed into the intercellular space by diffusion down an osmotic gradient. However, looking at the process as a whole, transport of water from lumen to blood is often against an osmotic gradient - this is important because it means that the intestine can absorb water into blood even when the osmolarity in the lumen is higher than osmolarity of blood.

This ability is best explained by the "three compartment model" for absorption of water and, like many aspects of gut permeability, varies along the length of the gut. The proximal small intestine functions as a highly permeable mixing segment, and absorption of water is basically isotonic. That is, water is not absorbed until the ingesta has been diluted out to just above the osmolarity of blood. The ileum and especially the colon are able to absorb water against an osmotic gradient of several hundred milliosmols.

Absorption of Monosaccharides

Simple sugars are far and away the predominant carbohydrate absorbed in the digestive tract, and in many animals the most important source of energy. Monosaccharides, however, are only rarely found in normal diets. Rather, they are derived by enzymatic digestion of more complex carbohydrates within the digestive tube.

Particularly important dietary carbohydrates include starch and disaccharides such as lactose and sucrose. None of these molecules can be absorbed for the simple reason that they cannot cross cell membranes unaided and, unlike the situation for monosaccharides, there are no transporters to carry them across.

This section will focus on understanding the processes involved in assimilation of three

important carbohydrates: starch, lactose and sucrose. The key concepts involved in all three cases are that:

- the final enzymatic digestion that liberates monosaccharides is conducted by enzymes that are tethered in the lumenal plasma membrane of absorptive enterocytes (so-called *"brush border hydrolyases"*).
- glucose generated by digestion of starch or lactose is absorbed in the small intestine only by *cotransport with sodium*, a fact that has exceptionally important implications in medicine.

Brush Border Hydrolases Generate Monosaccharides

Polysaccharides and disaccharides must be digested to monosaccharides prior to absorption and the key players in these processes are the brush border hydrolases, which include maltase, lactase and sucrase. Dietary lactose and sucrose are "ready" for digestion by their respective brush border enzymes. Starch, as discussed previously, is first digested to maltose by amylase in pancreatic secretions and, in some species, saliva.

Dietary lactose and sucrose, and maltose derived from digestion of starch, diffuse in the small intestinal lumen and come in contact with the surface of absorptive epithelial cells covering the villi where they engage with brush border hydrolases:

- maltase cleaves maltose into two molecules of glucose
- lactase cleaves lactose into a glucose and a galactose
- sucrase cleaves sucrose into a glucose and a fructose

At long last, we're ready to actually absorb these monosaccharides. Glucose and galactose are taken into the enterocyte by cotransport with sodium using the same transporter. Fructose enters the cell from the intestinal lumen via facilitated diffusion through another transporter.

Absorption of Glucose and Other Monosaccharides: Transport Across the Intestinal Epithelium

Absorption of glucose entails transport from the intestinal lumen, across the epithelium and into blood. The transporter that carries glucose and galactose into the enterocyte is the sodium-dependent hexose transporter, known more formally as SGLUT-1. As the name indicates, this molecule transports both glucose and sodium ion into the cell and in fact, will not transport either alone.

The essence of transport by the sodium-dependent hexose transporter involves a series of conformational changes induced by binding and release of sodium and glucose, and can be summarized as follows:

1. the transporter is initially oriented facing into the lumen - at this point it is capable of binding sodium, but not glucose
2. sodium binds, inducing a conformational change that opens the glucose-binding pocket
3. glucose binds and the transporter reorients in the membrane such that the pockets holding sodium and glucose are moved inside the cell

4. sodium dissociates into the cytoplasm, causing glucose binding to destabilize
5. glucose dissociates into the cytoplasm and the unloaded transporter reorients back to its original, outward-facing position

The animation seen below depicts digestion of maltose and entry of the resulting glucose, along with sodium, into the enterocyte (actually, two sodium ions are transported for each glucose). Despite the simplicity of the diagram, you should easily be able to identify the sodium-dependent hexose transporter and "watch" its conformational changes. Also, imagine the corresponding process involving lactose and sucrose assimilation.

Fructose is not co-transported with sodium. Rather it enters the enterocyte by another hexose transporter (GLUT5).

Once inside the enterocyte, glucose and sodium must be exported from the cell into blood. We've seen previously how sodium is rapidly shuttled out in exchange for potassium by the battery of sodium pumps on the basolateral membrane, and how that process maintains the electrochemical gradient across the epithelium. The energy stored in this gradient is actually what is driving glucose entry through the sodium-dependent hexose transporter described above. Recall also how the massive transport of sodium out of the cell establishes the osmotic gradient responsible for absorption of water.

Glucose, galactose and fructose are tranported out of the enterocyte through another hexose transporter (called GLUT-2) in the basolateral membrane. These monosaccharides then diffuse "down" a concentration gradient into capillary blood within the villus.

Absorption of Amino Acids and Peptides

Dietary proteins are, with very few exceptions, not absorbed. Rather, they must be digested into amino acids or di- and tripeptides first. In previous sections, we've seen two sources secrete proteolytic enzymes into the lumen of the digestive tube:

- the stomach secretes pepsinogen, which is converted to the active protease pepsin by the action of acid.
- the pancreas secretes a group of potent proteases, chief among them trypsin, chymotrypsin and carboxypeptidases.

Through the action of these gastric and pancreatic proteases, dietary proteins are hydrolyzed within the lumen of the small intestine predominantly into medium and small peptides (oligopeptides).

The brush border of the small intestine is equipped with a family of peptidases. Like lactase and maltase, these peptidases are integral membrane proteins rather than soluble enzymes. They function to further the hydrolysis of lumenal peptides, converting them to free amino acids and very small peptides. These endproducts of digestion, formed on the surface of the enterocyte, are ready for absorption.

Absorption of Amino Acids

The mechanism by which amino acids are absorbed is conceptually identical to that of monosaccharides. The lumenal plasma membrane of the absorptive cell bears at least four **sodium-dependent amino acid transporters** - one each for acidic, basic, neutral and amino acids. These transporters bind amino acids only after binding sodium. The fully loaded transporter then undergoes a conformational change that dumps sodium and the amino acid into the cytoplasm, followed by its reorientation back to the original form.

Thus, absorption of amino acids is also absolutely dependent on the electrochemical gradient of sodium across the epithelium. Further, absorption of amino acids, like that of monosaccharides, contributes to generating the osmotic gradient that drives water absorption.

The basolateral membrane of the enterocyte contains additional transporters which export amino acids from the cell into blood. These are not dependent on sodium gradients.

Absorption of Peptides

There is virtually no absorption of peptides longer than four amino acids. However, there is abundant absorption of di- and tripeptides in the small intestine. These small peptides are absorbed into the small intestinal epithelial cell by cotransport with H^+ ions via a transporter called PepT1.

Once inside the enterocyte, the vast bulk of absorbed di- and tripeptides are digested into amino acids by cytoplasmic peptidases and exported from the cell into blood. Only a very small number of these small peptides enter blood intact.

Absorption of Intact Proteins

As emphasized, absorption of intact proteins occurs only in a few circumstances. In the first place, very few proteins get through the gauntlet of soluble and membrane-bound proteases intact. Second, "normal" enterocytes do not have transporters to carry proteins across the plasma membrane and they certainly cannot permeate tight junctions.

One important exception to these general statements is that for a very few days after birth, neonates have the ability to absorb intact proteins. This ability, which is rapidly lost, is of immense importance because it allows the newborn animal to acquire passive immunity by absorbing immunoglobulins in colostral milk.

In constrast to humans and rodents, there is no significant transfer of antibodies across the placenta in many animals (cattle, sheep, horses and pigs to name a few), and the young are born without circulating antibodies. If fed colostrum during the first day or so after birth, they absorb large quantities of immunoglobulins and acquire a temporary immune system that provides protection until they generate their own immune responses.

The small intestine rapidly loses the capacity to absorb intact proteins - a process called closure - and consequently, animals that do not receive colostrum within the first few days after birth will likely die due to opportunistic infections.

Absorption of Lipids

The bulk of dietary lipid is neutral fat or triglyceride, composed of a glycerol backbone with each carbon linked to a fatty acid. Foodstuffs typically also contain phospholipids, sterols like cholesterol and many minor lipids, including fat-soluble vitamins. Finally, small intestinal contents contain lipids from sloughed epithelial cells and considerable cholesterol delivered in bile.

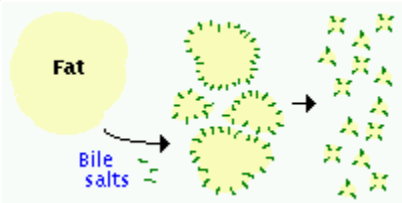
In order for the triglyceride to be absorbed, two processes must occur:

- Large aggregates of dietary triglyceride, which are virtually insoluble in an aqueous environment, must be broken down physically and held in suspension - a process called emulsification.
- Triglyceride molecules must be enzymatically digested to yield monoglyceride and fatty acids, both of which can efficiently diffuse or be transported into the enterocyte

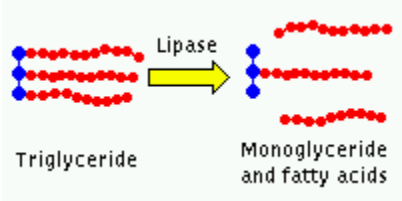
The key players in these two transformations are *bile acids* and *pancreatic lipase*, both of which are mixed with chyme and act in the lumen of the small intestine. Bile acids are also necessary to solubilize other lipids, including cholesterol.

Emulsification, Hydrolysis and Micelle Formation

Bile acids play their first critical role in lipid assimilation by promoting emulsification. As derivatives of cholesterol, bile acids have both hydrophilic and hydrophobic domains (i.e. they are amphipathic). On exposure to a large aggregate of triglyceride, the hydrophobic portions of bile acids intercalate into the lipid, with the hydrophilic domains remaining at the surface. Such coating with bile acids aids in breakdown of large aggregates or droplets into smaller and smaller droplets.



Hydrolysis of triglyceride into monoglyceride and free fatty acids is accomplished predominantly by pancreatic lipase. The activity of this enzyme is to clip the fatty acids at positions 1 and 3 of the triglyceride, leaving two free fatty acids and a 2-monoglyceride.

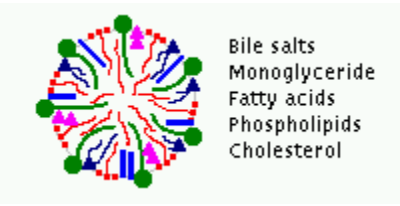


Lipase is a water-soluble enzyme, and with a little imagination, it's easy to understand why emulsification is a necessary prelude to its efficient activity. Shortly after a meal, lipase is present within the small intestine in rather huge quantities, but can act only on the surface of triglyeride droplets. For a given volume of lipid, the smaller the droplet size, the greater the surface area, which means more lipase molecules can get to work.

The drug orlistat (Xenical) that is promoted for treatment of obesity works by inhibiting pancreatic lipase, thereby

reducing the digestion and absorption of fat in the small intestine.

As monoglycerides and fatty acids are liberated through the action of lipase, they retain their association with bile acids and complex with other lipids to form structures called**micelles**. Micelles are essentially small aggregates (4-8 nm in diameter) of mixed lipids and bile acids suspended within the ingesta. As the ingesta is mixed, micelles bump into the brush border of small intestinal enterocytes, and the lipids, including monoglyceride and fatty acids, are taken up into the epithelial cells.



Absorption and Transport into Blood

The major products of lipid digestion - fatty acids and 2-monoglycerides - enter the enterocyte by simple diffusion across the plasma membrane. A considerable fraction of the fatty acids also enter the enterocyte via a specific fatty acid transporter protein in the membrane.

Lipids are transported from the enterocyte into blood by a mechanism distinctly different from what we've seen for monosaccharides and amino acids.

Once inside the enterocyte, fatty acids and monoglyceride are transported into the endoplasmic reticulum, where they are used to synthesize triglyeride. Beginning in the endoplasmic reticulum and continuing in the Golgi, triglyceride is packaged with cholesterol, lipoproteins and other lipids into particles called**chylomicrons**. *Remember where this is occurring - in the absorptive enterocyte of the small intestine.*

Chylomicrons are extruded from the Golgi into exocytotic vesicles, which are transported to the basolateral aspect of the enterocyte. The vesicles fuse with the plasma membrane and undergo exocytosis, dumping the chylomicrons into the space outside the cells.

Because chylomicrons are particles, virtually all steps in this pathway can be visualized using an electron microscope, as the montage of images to the right demonstrates.

Transport of lipids into the circulation is also different from what occurs with sugars and amino acids. Instead of being absorbed directly into capillary blood, chylomicrons are transported first into the lymphatic vessel that penetrates into each villus. Chylomicron-rich lymph then drains into the system lymphatic system, which rapidly flows into blood. Blood-borne chylomicrons are rapidly disassembled and their constitutent lipids utilized throughout the body.

When large numbers of chylomicrons are being absorbed, the lymph draining from the small intestine appears milky and the lymphatics are easy to see. In the image below, of abdominal contents from a coyote, the fine white lines (arrows) are intestinal lymphatics packed with chylomicrons. That lymph passes through mesenteric lymph nodes (LN) and then into larger lymphatics

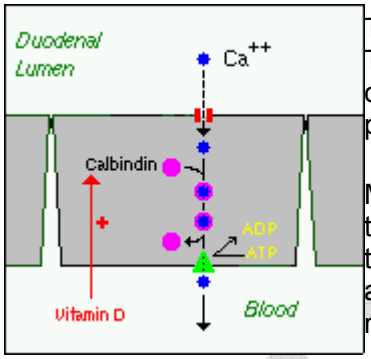
Another lipid of importance that is absorbed in the small intestine is cholesterol. Cholesterol homeostatis results from a balance of cholestrol synthesis, absorption of dietary cholesterol,

and elimination of cholesterol by excretion in bile. Years ago it was shown that cholesterol, but not plant sterols, is readily absorbed in the intestine. More recently, a specific transport protein (NPC1L1) has been identified that ferries cholesterol from the intestinal lumen into the enterocyte. From there, a bulk of the cholesterol is esterified, incorporated into chylomicrons and shuttled into blood by the mechanisms described above.

If you are interested in confirming for yourself at least some of the processes described above, you should perform the following experiment:

- Consume a cup of rich cream or a sack of fast-food French fries.
- Do something productive like studying for about 30 minutes.
- Draw a blood sample from yourself (a capillary tube is enough) - use an anticoagulant to prevent clotting.
- Centrifuge the blood sample to separate cells and plasma.

Absorption of Minerals and Metals



The vast bulk of mineral absorption occurs in the small intestine. The best-studied mechanisms of absorption are clearly for calcium and iron, deficiencies of which are significant health problems throughout the world.

Minerals are clearly required for health, but most also are quite toxic when present at higher than normal concentrations. Thus, there is a physiologic challenge of supporting efficient but limited absorption. In many cases intestinal absorption is a key regulatory step in mineral homeostasis.

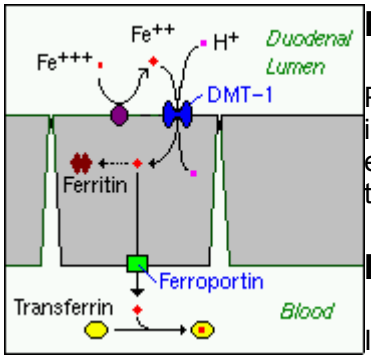
Calcium

Calcium is absorbed from the intestinal luman by two distinct mechanims, and their relative magnitude of importance is determined by the amount of free calcium available for absorption:

1. *Active, transcellular absorption* occurs only in the duodenum when calcium intake is low. This process involves import of calcium into the enterocyte, transport across the cell, and export into extracellular fluid and blood. Calcium enters the intestinal epithelial cells through voltage-insensitive (TRP) channels and is pumped out of the cell via a calcium-ATPase.

The rate limiting step in transcellular calcium absorption is transport across the epithelial cell, which is greatly enhanced by the carrier protein calbindin, the synthesis of which is totally dependent on vitamin D.

2. *Passive, paracellular absorption* occurs in the jejunum and ileum, and, to a much lesser extent, in the colon when dietary calcium levels are moderate or high. In this case, ionized calcium diffuses through tight junctions into the basolateral spaces around enterocytes, and hence into blood. When calcium availability is high, this pathway responsible for the bulk of calcium absorption, due to the very short time available for active transport in the duodenum.



Phosphorus

Phosphorus is predominantly absorbed as inorganic phosphate in the upper small intestine. Phosphate is transported into the epithelial cells by contransport with sodium, and expression of this (or these) transporters is enhanced by vitamin D.

Iron

Iron homeostasis is regulated at the level of intestinal absorption, and it is important that adequate but not excessive quantities of iron be absorbed from the diet. Inadequate absorption can lead to iron-deficiency disorders such as anemia. On the other hand, excessive iron is toxic because mammals do not have a physiologic pathway for its elimination.

Iron is absorbed by villus enterocytes in the proximal duodenum. Efficient absorption requires an acidic environment, and antacids or other conditions that interfere with gastric acid secretion can interfere with iron absorption.

Ferric iron (Fe+++) in the duodenal lumen is reduced to its ferrous form through the action of a brush border ferrireductase. Iron is the cotransported with a proton into the enterocyte via the divalent metal transporter DMT-1. This transporter is not specific for iron, and also transports many divalent metal ions.

Once inside the enterocyte, iron follows one of two major pathways. Which path is taken depends on a complex programming of the cell based on both dietary and systemic iron loads:

- *Iron abundance states*: iron within the enterocyte is trapped by incorporation intoferritin and hence, not transported into blood. When the enterocyte dies and is shed, this iron is lost.
- *Iron limiting states*: iron is exported out of the enterocyte via a transporter (ferroportin) located in the basolateral membrane. It then binds to the iron-carrier transferrin for transport throughout the body.

Iron in the form of heme, from ingestion of hemoglobin or myoglobin, is also readily absorbed. In this case, it appears that intact heme is take up by the small intestinal enterocyte by endocytosis. Once inside the enterocyte, iron is liberated and essentially follows the same pathway for export as absorbed inorganic iron. Some heme may be transported intact into the circulation.

Copper

There appear to be two processes responsible for copper absorption - a rapid, low capacity system and a slower, high capacity system, which may be similar to the two processes seen with calcium absorption. Many of the molecular details of copper absorption remain to be elucidated. Inactivating mutations in the gene encoding an intracellular copper ATPase have been shown responsible for the failure of intestinal copper absorption in Menkes disease.

A number of dietary factors have been shown to influence copper absorption. For example, excessive dietary intake of either zinc or molybdenum can induce secondary copper deficiency states.

Zinc

Zinc homeostasis is largely regulated by its uptake and loss through the small intestine. Although a number of zinc transporters and binding proteins have been identified in villus epithelial cells, a detailed picture of the molecules involved in zinc absorption is not yet in hand.

Intestinal excretion of zinc occurs via shedding of epithelial cells and in pancreatic and biliary secretions.

A number of nutritional factors have been identified that modulate zinc absorption. Certain animal proteins in the diet enhance zinc absorption. Phytates from dietary plant material (including cereal grains, corn, rice) chelate zinc and inhibit its absorption. Subsistance on phytate-rich diets is thought responsible for a considerable fraction of human zinc deficiencies.

The Large Intestine

The large intestine is the last attraction in digestive tube and the location of the terminal phases of digestion. In comparison to other regions of the tube, there are huge differences among species in the relative size and complexity of the large intestine. Nonetheless, in all species it functions in three processes:

- **Recovery of water and electrolytes from ingesta**: By the time ingesta reaches the terminal ileum, roughly 90% of its water has been absorbed, but considerable water and electrolytes like sodium and chloride remain and must be recovered by absorption in the large gut.
- **Formation and storage of feces**: As ingesta is moved through the large intestine, it is dehydrated, mixed with bacteria and mucus, and formed into feces.
  The *craftsmanship* (for want of a better term) with which this is carried out varies among species.
- **Microbial fermentation**: The large intestine of all species teems with microbial life. Those microbes produce enzymes capable of digesting many of molecules that to vertebrates are indigestible, cellulose being a premier example. The extent and benefit of fermentation also varies greatly among species.

Gross and Microscopic Anatomy of the Large Intestine

The large intestine is that part of the digestive tube between the terminal ileum and anus. Depending on the species, ingesta from the small intestine enters the large intestine through either the ileocecal or ileocolic valve. Within the large intestine, three major segments are recognized:

- the **cecum** is a blind-ended pouch that in humans carries a worm-like extension called the vermiform **appendix**.
- the **colon** constitutes the majority of the length of the large intestine and is subclassified into ascending, transverse and descending segments.
- the **rectum** is the short, terminal segment of the digestive tube, continuous with the anal canal.

The variation in relative dimension of the large intestine is largely correlated with diet. In herbivores like horses and rabbits which depend largely on microbial fermentation, the large intestine is very large and complex. Omnivores like pigs and humans have a substantial large intestine, but nothing like that seen in herbivores. Finally, carnivores such as dogs and cats have a simple and small large intestine.

There are many similarities in the histologic structure of the mucosa in large and small intestine. The most obvious difference is that the mucosa of the large intestine is devoid of villi. It has numerous crypts which extend deeply and open onto a flat lumenal surface. The stem cells which support rapid and continuous renewal of the epithelium are located either at the bottom or midway down the crypts. These cells divide to populate the cryptal and surface epithelium.

Mucus-secreting goblet cells are also much more abundant in the colonic epithelium than in the small gut.

Absorption, Secretion and Formation of Feces in the Large Intestine

To a first approximation, absorption and secretion in the colon is straighforward:

- **Absorption:** water, sodium ions and chloride ions
- **Secretion:** bicarbonate ions and mucus

Water, as always, is absorbed in response to an osmotic gradient. The mechanism responsible for generating this osmotic pressure is essentially identical to what was seen in the small intestine - sodium ions are transported from the lumen across the epithelium by virtue of the epithelial cells having very active sodium pumps on their basolateral membranes and a means of absorbing sodium through their lumenal membranes. The colonic epithelium is actually more efficient at absorbing water than the small intestine and sodium absorption in the colon is enhanced by the hormone aldosterone.

Chloride is absorbed by exchange with bicarbonate. The resulting secretion of bicarbonate ions into the lumen aids in neutralization of the acids generated by microbial fermentation in the large gut.

Goblet cells are abundant in the colonic epithelium, and secrete mucus in response to tactile stimuli from lumenal contents, as well as parasympathetic stimuli from pelvic nerves.Mucus is an important lubricant that protects the epithelium, and also serves to bind the dehydrated ingesta to form feces.

Normal feces are roughly 75% water and 25% solids. The bulk of fecal solids are bacteria

and undigested organic matter and fiber. The characteristic brown color of feces are due to stercobilin and urobinin, both of which are produced by bacterial degradation of bilirubin. Fecal odor results from gases produced by bacterial metabolism, including skatole, mercaptans, and hydrogen sulfide.

Large Intestinal Motility

Three prominent patterns of motility are observed the colon:

- **Segmentation contractions** which chop and mix the ingesta, presenting it to the mucosa where absorption occurs. These contractions are quite prominent in some species, forming sacculations in the colon known as *hausta*.
- **Antiperistaltic contractions propagate toward the ileum**, which serves to retard the movement of ingesta through the colon, allowing additional opportunity for absorption of water and electrolytes. Peristaltic contractions, in addition to influx from the small intestine, facilitate movement of ingesta through the colon.
- **Mass movements** constitute a type of motility not seen elsewhere in the digestive tube. Known also as giant migrating contractions, this pattern of motility is like a very intense and prolonged peristaltic contraction which strips an area of large intestine clear of contents.

In periods between meals, the colon is generally quiescent. Following a meal, colonic motility increases significantly, due to signals propagated through the enteric nervous system - the so called *gastrocolic and duodenocolic reflexes*, manifestation of enteric nervous system control. In humans, the signal seems to be stimulated almost exclusively by the presence of fat in the proximal small intestine. Additionally, distension of the colon is a primary stimulator of contractions.

Several times each day, mass movements push feces into the rectum, which is usually empty. The gastrocolic reflex mentioned above is a stimulus for this. Distension of the rectum stimulates the defecation reflex. This is largely a spinal reflex mediated via the pelvic nerves, and results in reflex relaxation of the internal anal sphincter followed by voluntary relaxation of the external anal sphincter and defecation.

In humans and "house-trained" animals, defecation can be prevented by voluntary constriction of the external sphincter. When this happens, the rectum soon relaxes and the internal sphincter again contracts, a state which persists until another bolus of feces is forced into the rectum.

Microbial Fermentation

Fermentation is the enzymatic decomposition and utililization of foodstuffs, particularly carbohydrates, by microbes. Fermentation takes place in the large bowel of all animals, but there are major differences in its contribution to the nutrition of different species. In carnivores like dogs and cats, and even in omnivores like humans, fermentation generates very few calories. In herbivores, however, fermentation is a way of life.

The large intestine does not produce its own digestive enzymes, but contains huge numbers of bacteria which have the enzymes to digest and utilize many substrates. In all animals, two

processes are attributed to the microbial flora of the large intestine:

- Digestion of carbohydrates not digested in the small intestine
- Synthesis of vitamin K and certain B vitamins

Cellulose is common constituent in the diet of many animals, including man, but no mammalian cell is known to produce a cellulase. Several species of bacteria in the large bowel synthesize cellulases and digest cellulose. Importantly, the major end products of microbial digestion of cellulose and other carbohydrates are volatile fatty acids, lactic acid, methane, hydrogen and carbon dioxide. Fermentation is thus the major source ofintestinal gas. Volatile fatty acids (acetic, proprionic and butyric acids) generated from fermentation can be absorbed by diffusion in the colon.

You obtain a few calories from eating a salad. A rabbit, on the other hand, has a relatively huge fermentation vat (cecum), and obtains much of its energy from the plants it consumes.

Synthesis of vitamin K by colonic bacteria provides a valuable supplement to dietary sources and makes clinical vitamin K deficiency rare. Similarly, formation of B vitamins by the microbial flora in the large intestine is useful to many animals. They are not absorbed in the large intestine, but are present in feces. The behavior of coprophagy or eating feces seen particularly in rodents, rabbits and other animals is thought to be a behavioral adaption to recovery of these valuable resources.

A more comprehensive description of fermentation is presented in the section ondigestive physiology of herbivores.

Intestinal Gas Production

A considerable amount of gas is present in the gastrointestinal contents of all animals, and much of this is eliminated through the anus as *flatus*. Complaints of excessive gastrointestinal gas production in people and their pets are common. What we know about intestinal gas production and disposition has largely be gathered from studies with humans.

Five gases constitute greater than 99% of the gases passed as flatus: N_2, O_2, CO_2, $H_2$and methane. None of these gases has an odor, and the characteristic odor of feces is due to very small quantities of a few other gases, including hydrogen sulfide, scatols and indoles. There is considerable individual variation in the contribution of each of these gases to total gas, but nitrogen typically predominates. Volume of gas elaborated also varies widely. In normal adult humans, the rate of excretion of gas per rectum ranges between 200 and 2000 ml per day. Ingestion of certain foods, beans being the classical example, is widely recognized to increase the rate of gas production.

There are three principal sources of the five major intestinal gases:

1. **Air swallowing is the major source of gas in the stomach.** Several milliliters of air are swallowed with every bolus of food or saliva. Most of this seems to be eructated and, apparently, very little passes into the duodenum.

2. **Intraluminal generation of gases results from two major processes;**

First, in the proximal intestine, the interaction of hydrogen and bicarbonate ions (principally from gastric and pancreatic secretions) leads to generation of CO_2. The amount of gas generated by this pathway is not great, because the lumenal contents do not contain carbonic anhydrase and the dissociation of H_2CO_3 is thus quite slow. Additionally, most of the CO_2 produced in this way is absorbed into blood.

The second and much more productive source of gas is fermentation by colonic bacteria. Microbes appear to be the sole source of all of the hydrogen and methane produced in the intestine. Fermentable substrates that escape digestion or absorption in the small intestine are often prime substrates for bacteria in the large intestine. A variety of fruits and vegetables contain polysaccharides that are not digested in the small intestine and lead to voluminous gas production by microbes. Indeed, the primary medical treatment for excessive gas production is dietary manipulation to eliminate foodstuffs that the individual cannot digest and absorb.

3. **Gases readily diffuse across the mucosa.** The direction of diffusion is dictated by the partial pressure of that gas in blood versus luminal contents. For methane and hydrogen, diffusion is always out of the lumen into blood. Nitrogen and CO_2 diffuse in either direction, depending on specific conditions within the individual.

Intestinal gases are a frequent cause of minor, occasionally major, social embarrassment, but can they ever be of truly dangerous? Both H_2 and CH_4 are combusible and potentially explosive. In human hospitals, there have been many explosions in the colon triggered by use of electrocautery performed through a proctosigmoidoscope. Many of these cases occurred when mannitol, a fermentable carbohydrate, was used as a purgitive to cleanse the colon. Use of non-fermentable cleansing agents has virtually eliminated this kind of accident.

Pathophysiology of Diarrhea

Diarrhea is an increase in the volume of stool or frequency of defecation. It is one of the most common clinical signs of gastrointestinal disease, but also can reflect primary disorders outside of the digestive system. Certainly, disorders affecting either the small or large bowel can lead to diarrhea.

For many people, diarrhea represents an occasional inconvenience or annoyance, yet at least 2 million people in the world, mostly children, die from the consequences of diarrhea each year.

There are numerous causes of diarrhea, but in almost all cases, this disorder is a manifestation of one of the four basic mechanisms described below. It is also common for more than one of the four mechanisms to be involved in the pathogenesis of a given case.

Osmotic Diarrhea

Absorption of water in the intestines is dependent on adequate absorption of solutes. If excessive amounts of solutes are retained in the intestinal lumen, water will not be absorbed and diarrhea will result. Osmotic diarrhea typically results from one of two situations:

- *Ingestion of a poorly absorbed substrate:* The offending molecule is usually a carbohydrate or divalent ion. Common examples include mannitol or sorbitol, epson

salt ($MgSO_4$) and some antacids ($MgOH_2$).

- *Malabsorption:* Inability to absorb certain carbohydrates is the most common deficit in this category of diarrhea, but it can result virtually any type of malabsorption. A common example of malabsorption, afflicting many adults humans and pets islactose intolerance resulting from a deficiency in the brush border enzyme lactase. In such cases, a moderate quantity of lactose is consumed (usually as milk), but the intestinal epithelium is deficient in lactase, and lactose cannot be effectively hydrolyzed into glucose and galactose for absorption. The osmotically-active lactose is retained in the intestinal lumen, where it "holds" water. To add insult to injury, the unabsorbed lactose passes into the large intestine where it is fermented by colonic bacteria, resulting in production of excessive gas.

A distinguishing feature of osmotic diarrhea is that it stops after the patient is fasted or stops consuming the poorly absorbed solute.

Secretory Diarrhea

Large volumes of water are normally secreted into the small intestinal lumen, but a large majority of this water is efficienty absorbed before reaching the large intestine. Diarrhea occurs when secretion of water into the intestinal lumen exceeds absorption.

Many millions of people have died of the secretory diarrhea associated with cholera. The responsible organism, *Vibrio cholerae*, produces cholera toxin, which strongly activates adenylyl cyclase, causing a prolonged increase in intracellular concentration of cyclic AMP within crypt enterocytes. This change results in prolonged opening of the chloride channels that are instrumental in secretion of water from the crypts, allowing uncontrolled secretion of water. Additionally, cholera toxin affects the enteric nervous system, resulting in an independent stimulus of secretion.

Exposure to toxins from several other types of bacteria (e.g. *E. coli* heat-labile toxin) induce the same series of steps and massive secretory diarrhea that is often lethal unless the person or animal is aggressively treated to maintain hydration.

In addition to bacterial toxins, a large number of other agents can induce secretory diarrhea by turning on the intestinal secretory machinery, including:

- some laxatives
- hormones secreted by certain types of tumors (e.g. vasoactive intestinal peptide)
- a broad range of drugs (e.g. some types of asthma medications, antidepressants, cardiac drugs)
- certain metals, organic toxins, and plant products (e.g. arsenic, insecticides, mushroom toxins, caffeine)

In most cases, secretory diarrheas will not resolve during a 2-3 day fast.

Inflammatory and Infectious Diarrhea

The epithelium of the digestive tube is protected from insult by a number of mechanisms constituting thegastrointestinal barrier, but like many barriers, it can be breached. Disruption

of diarrhea in all species. Destruction of the epithelium results not only in exudation of serum and blood into the lumen but often is associated with widespread destruction of absorptive epithelium. In such cases, absorption of water occurs very inefficiently and diarrhea results. Examples of pathogens frequently associated with infectious diarrhea include:

- Bacteria: *Salmonella, E. coli, Campylobacter*
- Viruses: rotaviruses, coronaviruses, parvoviruses (canine and feline), norovirus
- Protozoa: coccidia species, *Cryptosporium*, *Giardia*

The immune response to inflammatory conditions in the bowel contributes substantively to development of diarrhea. Activation of white blood cells leads them to secrete inflammatory mediators and cytokines which can stimulate secretion, in effect imposing a secretory component on top of an inflammatory diarrhea. Reactive oxygen species from leukocytes can damage or kill intestinal epithelial cells, which are replaced with immature cells that typically are deficient in the brush border enyzmes and transporters necessary for absorption of nutrients and water. In this way, components of an osmotic (malabsorption) diarrhea are added to the problem.

Diarrhea Associated with Deranged Motility

In order for nutrients and water to be efficiently absorbed, the intestinal contents must be adequately exposed to the mucosal epithelium and retained long enough to allow absorption. Disorders in motility than accelerate transit time could decrease absorption, resulting in diarrhea even if the absorptive process per se was proceeding properly.

Alterations in intestinal motility (usually increased propulsion) are observed in many types of diarrhea. What is not usally clear, and very difficult to demonstrate, is whether primary alterations in motility are actually the cause of diarrhea or simply an effect.