Infectious Diseases
# Infectious Diseases
## Student Workbooks

### Unit 1 Why should we care about infectious disease?
- Lesson 1: 3
- Lesson 2: 9
- Lesson 3: 17
- Lesson 4: 25
- Lesson 5: 32

### Unit 2 What does it mean to have an infectious disease?
- Lesson 1: 41
- Lesson 2: 45
- Lesson 3: 51
- Lesson 4: 57
- Lesson 5: 61
- Lesson 6: 66
- Lesson 7: 71

### Unit 3: When does a microbe become a pathogen?
- Lesson 1: 74
- Lesson 2: 77
- Lesson 3: 87
- Lesson 4: 90

### Unit 4: How do pathogens make us sick?
- Lesson 1: 93
- Lesson 2: 99
- Lesson 3: 112
- Lesson 4: 116
- Lesson 5: 122

### Unit 5: How do we get better?
- Lesson 1: 125
- Lesson 2: 125
- Lesson 3: 130
- Lesson 4: 134
- Lesson 5: 138
Lesson 1.1 Workbook

What is an infectious disease and why do we care?

Why should we care about infectious disease?
In this lesson, we will discuss what makes a disease infectious. We will use what happened with H1N1 in 2009 to illustrate the potential impact infectious diseases can have. You will be asked to question how we reacted to the 2009 H1N1 epidemic. Did we overreact?

What is an infectious disease?

An infectious disease is a clinically evident illness (meaning that it produces characteristic symptoms or medical signs) resulting from the presence of microbial agents. Because these agents can cause disease they are termed pathogenic. Pathogenic microbes include viruses, bacteria, fungi, protozoa, and multicellular parasites. Infectious diseases are also called communicable diseases or transmissible diseases because they have the potential to be transmitted from one organism to another. Unlike toxins that can also cause disease, only infectious agents can also replicate.

Transmission of an infectious disease can occur through different routes that may or may not require actual physical contact between infected individuals. Examples of such routes are liquids, food, body fluids, contaminated objects, airborne inhalation, or through vectors. An example of a vector is a mosquito.

The term infectivity describes the ability of a pathogenic microbial agent to enter, survive, and multiply in the host. The term infectiousness of a disease describes the ease with which the pathogenic microbe is transmitted to other hosts.
**LES S O N M A T E R I A L S**

**How we can receive and transmit infections:**

**Through the mouth:**
Microbes can enter the digestive system through the mouth in either food or water, but in order to cause disease they must overcome a powerful host defense - the acid in the stomach. The stomach’s effectiveness in killing microbes depends on the kind and amount of the food that the microbe is contaminating. Certain foods, like fats, can protect the microbes from being killed by acid. Some bacteria and yeasts are resistant even after maximal exposure to stomach acid. They can escape the stomach alive, although with reduced numbers. Microbes that survive the stomach’s acid barrier enter the small intestine. There they meet the enzymes of the pancreatic juice and bile salts that can digest them. They also encounter the strong sweeping force of peristalsis. Few microbes survive or avoid being washed away. The very few pathogenic microbes that can survive the upper reaches of the small intestine use attachment mechanisms that allow them to adhere to the lining of the small intestine. For example, Cholera and its milder relative travelers’ diarrhea produce powerful toxins that affect the epithelia lining the intestine; they are therefore able to cause disease even without crossing the lining and entering the inside of the body.

**Through the nose:**
Microbes that are inhaled in aerosol droplets or dust particles must overcome a series of aerodynamic and hydrodynamic obstacles to enter the respiratory system. First they must navigate complex structures in the nose, the pharynx, and the larynx. Then, the microbes that do arrive in the lower reaches of the respiratory tree face the powerful upward-sweeping action of cilia in the lungs. Just as we saw in the gut, microbes need to stick to the lining of the lungs if they are to survive. Persons in whom the ciliary ‘elevator’ is impaired (e.g., heavy smokers) are more likely to succumb to these kinds of infections.

**Cuts and wounds:**
Penetration through cuts and wounds is common, and often goes unnoticed because it frequently does not cause symptoms of disease. For example, brushing one’s teeth or even defecating can cause minute abrasions to the epithelial membranes that allow small numbers of bacteria to enter in the bloodstream. However the filtering mechanisms of the lymph system usually remove them rapidly.

**Insect Bites:**
Insect bites can allow viruses (viral encephalitis, yellow fever) as well as bacteria (plague, typhus), protozoa (malaria, sleeping sickness) and worms (river blindness, elephantiasis) to enter the skin. A particularly unsavory example of an insect bite allowing microbial transmission through the skin is that of the so-called reduviid bugs, which defecate as they are biting. Parasites in the insect’s feces can penetrate the skin when the person scratches the bite area.

**Definitions of Terms**

- **Peristalsis** - relaxation and contraction of muscles in the small intestine that propels food through the digestive tract

- **Traveler’s diarrhea** - common to international travelers, it is a relatively mild diarrhea that can be caused by any number of pathogens, that is most often transmitted by poor sanitation.

- **Epithelia** – cells lining the inside of the body cavity. They are often ciliated.

- **Cilia** - finger-like protrusions in the intestines that help propel substances, particularly mucus.
Microbes that have penetrated into the body can gain a foothold and cause serious disease if internal tissues are also damaged or if the immune system defense mechanisms are disrupted, for instance due to immune suppression. An example is infection of heart valves by strep bacteria. Strep can invade heart valves that have been damaged by a previous disease, usually rheumatic fever. In the pre-antibiotic era the ensuing swelling of the heart was often fatal.

**Other modes of transmission:**

Microbes can also be actively carried into tissues by cells of the immune system that lie in the body cavity, which is actually outside the body itself. Immune cells in the alveoli of the lungs, known as dust cells, can ‘eat’ or engulf infectious agents by phagocytosis. Most of the time, the microbe-containing dust cells are then carried upward by the ciliary ‘elevator’, but occasionally the infected cells can enter the body, carrying their load of microbes into deeper locations. This cell-mediated entry can occur at other mucous membranes as well. For example, it is thought that HIV, the virus that causes AIDS, may be sexually transmitted when virus-laden immune cells in the semen penetrate the genital epithelial lining.

**Blood transfusions and organ transplants:**

Yet another way for organisms to gain access to deeper tissue is through blood transfusions or organ transplants. Of all the infectious agents that can be acquired through blood transfusions, none causes greater concern than HIV. However, many others, such as hepatitis C virus (HCV) can also be transmitted in this way. It is therefore critical to screen blood in blood banks. Organ transplants can also transmit infections – when corneas that were infected with the prion that causes Creutzfeldt-Jakob disease (the human form of Mad Cow Disease) were transplanted, they caused disease in the transplant recipients.

**Inoculum size:**

Whether or not microbes that have penetrated the skin or mucous membranes of the body cavity will cause disease depends in part on the number of the infectious agents i.e. the inoculum size. It usually takes many infectious agents to overcome host defenses, so an encounter with a small number of organisms is unlikely to result in an infection. In contrast, even bacteria that are normally harmless can overcome the normal defenses of the skin and cause infection if they are present in large enough numbers. An example is Pseudomonas bacteria infection from a contaminated hot tub, which often contains as many as 100 million Pseudomonas bacteria per milliliter. If large numbers of microbes gain access to deeper tissues infections are almost inevitable. Patients with open wounds must be treated carefully using sterile technique. For example, before making an incision in the skin, a surgeon will prepare the area with disinfectant to reduce the potential inoculum size of the skin bacteria that could invade the surgical wound.
DEFINITIONS OF TERMS

Pandemic - an epidemic that is spreading through human populations across a large region.

Cyanosis - Blue or purple coloration of the skin due to tissues being low on oxygen.

Mortality - a measure of the number of deaths from the disease in a given population.

H1N1 - a subtype of influenza A virus.

Epidemic - an outbreak of disease greater than would otherwise be expected at a particular time and place.

WHO - World Health Organization.

LESSON MATERIALS

Spanish Flu pandemic 1918

The influenza pandemic of 1918-1919 killed more people than the Great War, known today as World War I (WWI) - somewhere between 20 and 40 million people. It has been cited as the most devastating epidemic in recorded world history. More people died of influenza in that single year than in the four years of the Black Death Bubonic Plague from 1347 to 1351. Known as “Spanish Flu” or “La Grippe“ the influenza of 1918-1919 was a global disaster.

In the fall of 1918 the Great War in Europe was winding down and peace was on the horizon. The Americans had joined in the fight, bringing the Allies closer to victory against the Germans. Deep within the trenches the soldiers lived through some of the most brutal conditions imaginable. Then, in pockets across the globe, something erupted that seemed as benign as the common cold. The influenza of that season, however, was far more than a cold. In the two years that the scourge ravaged the earth, a fifth of the world’s population was infected. The flu was most deadly for people ages 20 to 40. This pattern of morbidity was unusual — influenza is usually a killer of the elderly and the very young. It infected 28% of all Americans and an estimated 675,000 Americans died of influenza during the pandemic, ten times as many as in the World War I. Of the U.S. soldiers who died in Europe, half of them fell to the influenza virus and not to the enemy.

The death rate for 15 to 34-year-olds from influenza and the resulting pneumonia was 20 times higher in 1918 than in previous years, reducing the average life span in the US by 10 years. People were struck with illness on the street and died rapid deaths. In Boston stories are told of people getting on the T and then falling out dead when the doors opened at the next stop. One anecdote told of four women playing bridge together late into the night. Overnight, three of them died from influenza. One physician wrote that patients with seemingly ordinary influenza would rapidly “develop the most viscous type of pneumonia that has ever been seen” and later when cyanosis appeared in the patients, “it is simply a struggle for air until they suffocate.”

Most of humanity felt the effects of Spanish flu as it spread following the path of its human carriers, along trade routes and shipping lines. The Great War, with its mass movements of men in armies and aboard ships, probably aided its rapid diffusion. The origins of the deadly flu disease were unknown but widely speculated upon. Some of the allies thought of the pandemic as a biological warfare tool used by the Germans. A national campaign began using the ready rhetoric of war to fight the new enemy of microscopic proportions.
H1N1 2009

The 2009 flu pandemic was a global outbreak of a new strain of the same virus that caused the 1918 pandemic - H1N1. H1N1 is often called “swine flu”. This is a mischaracterization: H1N1 cannot be spread by eating pork or pork products. Like other influenza viruses, it needs person to person transmission usually after sneezing or coughing. Symptoms usually last 4–6 days. The 2009 H1N1 virus, like the 1918 strain infects young people as well as older adults and the very young. Even persons that were previously very healthy may develop pneumonia or breathing difficulties within 3–6 days after initial onset of flu symptoms.

When the outbreak began in the state of Veracruz, Mexico, the Mexican government closed most of Mexico City’s public and private facilities in an attempt to contain the spread of the virus. However it continued to spread globally and clinics in some areas were overwhelmed by infected people. In June, the World Health Organization (WHO) and US Centers for Disease Control (CDC) stopped counting cases and declared the epidemic to be a pandemic.

Infections began to taper off in November 2009 and by May 2010 the number of cases was in steep decline. In August 2010, the WHO announced the H1N1 pandemic had ended. According to the latest WHO statistics, the virus has killed more than 18,000 people since it appeared in April 2009, approximately 4% of the annual influenza deaths. The number seems low but the WHO states that total mortality from the new H1N1 strain (including unconfirmed deaths) is “questionably higher” than would be expected.

On the other hand, research released in September 2010 disclosed that children with the pandemic flu were less likely to develop complications than those sick with seasonal flu strains, contradicting early reports on the severity of the pandemic. Critics claimed the WHO had exaggerated the danger, spreading “fear and confusion” rather than “immediate information.” The WHO has begun an investigation to determine whether it had “frightened people unnecessarily.”

Figure 1.1.4: Investigating a suspected case of H1N1 on an airplane in 2009.

DEFINITIONS OF TERMS

H1N1 - a subtype of influenza A virus

Epidemic - an outbreak of disease greater than would otherwise be expected at a particular time and place

Mortality - a measure of the number of deaths from the disease in a given population

WHO – World health Organization.

For a complete list of defined terms, see the Glossary.

LESSON MATERIALS

H1N1 - The Swine Flu

How is the Swine Flu similar to the Spanish Flu?

How is it different?
STUDENT RESPONSES

Have you changed your mind about whether the response to H1N1 was an overreaction?

Why or why not?

In a paragraph identify three globally important infectious diseases: 1) one bacterial, 2) one viral, and 3) one parasitic. Identify your sources here.
How infectious diseases have molded history - including ours

Epidemics such as the Spanish flu we saw in lesson one, have impacted us far beyond the number of people they infected and killed. Before we examine the current and potential future impact of infectious diseases, we’re going to look at how ancient plagues affected history.

How infectious diseases have molded history

People began to theorize about infections long before microbes were identified: As far back as the first century BC the Roman Marcus Terentius Varro wrote a book called ‘On Agriculture’ in which he warned against locating a homestead near swamps. Others also spoke of infections, for example the Islamic physician Abu Ali ibn Sina (also known by his Roman name of Avicenna), wrote ‘The Canon of Medicine’ in 1020, in which he stated that ‘bodily secretion is contaminated by foul foreign earthly bodies before being infected’. He also hypothesized that tuberculosis (which did not have this name yet) might be an infectious disease. Even so most people only accepted that microbes existed once they had actually seen them with microscopes. Before that, a common response to a devastating infectious disease was: Is this the work of the devil?
DEFINITIONS OF TERMS

Bubonic plague- is a severe systemic infection caused by the bacteria Yersinia pestis

Crimea - a peninsula to the south of the Ukraine.

Virulence - a pathogen’s ability to infect and cause disease.

For a complete list of defined terms, see the Glossary.

LESSON MATERIALS

Plagues of the past changed lives and shaped

The Antonine Plague in 165 -180 B.C. killed about one-third of the population and weakened the expansion of the Roman empire

The plague was either smallpox or measles. It led to a dramatic decrease in the population of Rome, weakening the army and slowing the expansion of the empire. We might all be speaking Latin today if Rome had not been weakened by this epidemic! It was brought back to the Roman Empire by troops returning from campaigns in the Near East, claimed the lives of two Roman emperors — Lucius Verus, and Marcus Aurelius Antoninus, and decimated the Roman army. It caused up to 2,000 deaths a day at Rome, one quarter of those infected. Total deaths have been estimated at five million, as much as one-third of the population in some areas. The Romans’ defense of the eastern territories was hampered when large numbers of troops succumbed to the disease. Many towns and villages in the Italian peninsula and the European provinces lost all their inhabitants. As the disease swept north to the Rhine, it also infected Germanic and Gallic peoples outside the Empire’s borders. For a number of years, these northern groups had pressed south in search of more lands to sustain their growing populations. With their ranks thinned by the epidemic, Roman armies were now unable to push the tribes back. The plague caused drastic effects throughout the Roman Empire, particularly on literature and art. The ancient world never recovered.

One of the deadliest plagues: The Black Death killed 30-60% of Europe’s population.

One of the deadliest pandemics the world had ever experienced is the Black Death. This bubonic plague outbreak started in Central Asia and reached Crimea in 1346. The Black Death is estimated to have killed 30% to 60% of Europe’s population, reducing the world’s population from an estimated 450 million to between 350 and 375 million in 1400. The Black Death returned every generation with varying virulence and mortalities until the 1700s. During this period, more than 100 plague epidemics swept across Europe. People knew that the Black Death was transmitted by rats, but they couldn’t do anything about it because they didn’t understand that rats aren’t the actual cause or infectious agent. The disease is actually transmitted by a bacterium (Yersinia pestis) that infects the fleas the rats carry.

In fact the common belief at the time was that the plague was spread by birds. Birds’ beak-shaped face-masks like the one you see on the left top of the page were worn by Plague Doctors during the Black Death and acted like a primitive gas mask. It was also thought that the mask would draw the disease away from the sufferer. Along the same lines the red glass eye pieces were thought to help protect the plague doctor from evil influences. The beak usually contained strongly aromatic herbs and spices to combat the terrible stench of unburied corpses and fluids from ill plague patients.

Fig 1.2.1: Medieval woodcut of the Antonine plague.
LESSON MATERIALS

Plagues in the Americas played an important role in overthrowing the Native Americans

Have you ever wondered how the Native Americans were overthrown by settlers that were far from home? Yes, technologies like guns certainly helped, but so did losing your population to infectious disease. Before the Europeans arrived, the Americas had been largely isolated from the infectious disease epidemics that spread throughout Europe. The first large-scale contacts between Europeans and native people of the American continents brought overwhelming pandemics of measles and smallpox, as well as other Eurasian diseases. These diseases from Europe spread rapidly among native peoples and led to a drastic drop in population and the collapse of indigenous American cultures.

It is important to note that diseases were passed in both directions. Syphilis was carried back from the Americas and swept through the European population, decimating large numbers there. Both are examples of how populations that haven’t encountered a ‘foreign’ infectious disease may be particularly susceptible to it at first.

Plagues were used as agents of biological warfare

The British are thought to have had the first idea to use smallpox as a bioterrorism agent by giving smallpox infected blankets to the indigenous population. On at least one occasion a high-ranking European considered infecting the Indians with smallpox as a tactic of war. Lord Jeffrey Amherst, commander of British forces in North America during the French and Indian War (1756-'63) discussed sending infected blankets to hostile tribes.

According to historian Francis Parkman, Amherst first raised the possibility of giving the Indians infected blankets in this letter to Colonel Henry Bouquet, who would lead reinforcements to Fort Pitt. We don't know if Bouquet actually put the plan into effect. We do know that a supply of smallpox-infected blankets was available, since the disease had broken out at Fort Pitt some weeks previously. We also know that the following spring smallpox was reported among the Indians in the vicinity. The smallpox epidemic between 1775 and 1782 raged across much of North America, killing more than 130,000 people. Tens of thousands of people died throughout Mexico from smallpox beginning in 1779. Smallpox then swept through the Pueblos of New Mexico beginning in 1780. It caused loss of Aztec and Inca populations and death of military and social leaders, contributing to the downfall of both empires and the subjugation of American peoples to Europeans.

Figure 1.2.2: Did the British knowingly give infected blankets to the Native Americans?
“P.S. I will try to inoculate the Indians by means of Blankets that may fall in their hands, taking care however not to get the disease myself. As it is pity to oppose good men against them, I wish we could make use of the Spaniard's Method, and hunt them with English Dogs. Supported by Rangers, and some Light Horse, who would I think effectively extirpate or remove that Vermine.”
Colonel Henry Bouquet, letter to Lord Jeffrey Amherst, Commander of British Forces July 13th 1763
Plagues still impact our lives and shape history:

Now we will turn our attention to infectious diseases that greatly impact societies today. Keep in mind that even with modern medicine and infrastructures like clean water and toilets it is hard to control many pathogens. And areas with endemic infectious diseases are often crippled by the burden of illness.

HIV causes crippling mortality and morbidity.

The largest recent pandemic is due to HIV (Human Immunodeficiency Virus). Over 30 million people worldwide are living with HIV/AIDS (Acquired Immune Deficiency Syndrome). The number of AIDS cases in the United States and Europe rose steadily through the mid-1990s and then plateaued as a result of increasingly effective efforts at prevention and drug treatments. In the developing world the disease has continued to spread. Africa has just over 12% of the world’s population but more than 60% of the AIDS cases worldwide.

From its discovery in 1981 to 2006, AIDS has killed more than 25 million people. AIDS is caused by HIV, which is a retrovirus. HIV infects about 0.6% of the world’s population. In 2009, an estimated 2.6 million people were newly infected and AIDS claimed an estimated 1.8 million lives, down from a global peak of 2.1 million in 2004. Approximately 260,000 of those were children. The disproportionate number of AIDS deaths occurring in Sub-Saharan Africa retards economic growth and exacerbates the burden of poverty. A 2005 study that estimated 90 million HIV infections in Africa also estimated a minimum of 18 million of orphans. The WHO considers HIV infection to be a global pandemic.

Infection with HIV causes the immune system to progressively fail. Once HIV virus has been transmitted it infects vital immune system cells such as helper T cells that normally protect the body against infection. HIV infection either kills the T cells directly, or targets them to be killed by other immune system cells that are surveying the body to eliminate infections. When helper T cell numbers decline below a critical level, they cannot protect the body against other infections allowing life-threatening opportunistic infections and cancers to thrive. This stage of infection is called AIDS.

For a complete list of defined terms, see the Glossary.

What causes AIDS and how long does it take to get after the initial infection with the infectious agent?

How might the AIDS epidemic in Africa impact society? (Think about mortality and morbidity)

One reason for the spread of HIV in developing countries is poor education. How do you think education could be used to stop the spread of HIV?
**Prevention and management:**

Most people infected with HIV-1 who are untreated eventually develop AIDS. They mostly die from infections or cancers that result from failure of the immune system. HIV progresses to AIDS at a variable rate that is affected by viral, host, and environmental factors. Most untreated HIV infections progress to AIDS within 10 years, but some will progress much sooner, and some will take much longer. Treatment with antiretroviral drugs increases the life expectancy of people infected with HIV and reduces both the mortality and the morbidity of HIV infection. As of 2005, the average survival time of someone with AIDS who receives antiretroviral therapy was estimated to be more than 5 years. Without antiretroviral therapy, someone who has AIDS typically dies within a year.

Although antiretroviral medication is still not available everywhere and does not actually cure the disease, expanded treatment programs have helped to turn the tide of new infections and AIDS deaths in many parts of the world. Intensified awareness and preventive measures, as well as the natural course of the epidemic, have also played a role. Nevertheless, complacency about HIV continues to contribute to risk for sexual transmission.

**Tuberculosis infects about one-third of the worlds population**

Tuberculosis (TB) is another common, and in many cases lethal, infectious disease caused by the tubercle bacillus *M. tuberculosis*. Tuberculosis usually attacks the lungs but can also affect other parts of the body. It is spread through the air when people who have an active infection cough, sneeze, or otherwise transmit their saliva through the air. Most infections in humans result in an asymptomatic, latent infection, and in about one in ten latent infection eventually progresses to active disease, which, if left untreated, kills more than 50% of its victims. The classic symptoms are a chronic cough with blood-tinged sputum, fever, night sweats, and weight loss (the last giving rise to the formerly prevalent colloquial term “consumption”).

Roughly a third of the world’s population has been infected with *M. tuberculosis*, and new infections occur at a rate of one per second. In 2007, an estimated 13.7 million people had active TB disease, with 9.3 million new cases and 1.8 million deaths; the annual incidence rate varied from 363 per 100,000 in Africa to 32 per 100,000 in the Americas. Tuberculosis is the world’s greatest infectious killer of women of reproductive age and the leading cause of death among people with HIV/AIDS. The proportion of people who become sick with tuberculosis each year is stable or falling worldwide but, because of population growth, the absolute number of new cases is still increasing. In 2007 there were an estimated 13.7 million chronic active cases, 9.3 million new cases, and 1.8 million deaths, mostly in developing countries.
LESSON MATERIALS

Malaria is arguably the highest impact infectious disease of our times.

Malaria is a mosquito-borne infectious disease caused by a parasitic protozoan discovered in 1880 by Charles Louis Alphonse Laveran. While working in the military hospital in Constantine, Algeria, he observed the parasites in a blood smear taken from a patient who had just died of malaria. The disease results when the malaria parasites multiply within red blood cells causing them to burst. The classic symptom of malaria is a cycle in which sudden chills are followed by fever and sweating lasting four to six hours. The cycle repeats every two to three days as new red blood cells mature, become infected and then burst.

Malaria is widespread in tropical and subtropical regions, including much of Sub-Saharan Africa, Asia and the Americas. It is prevalent in these regions because the high rainfall and consistent high temperatures along with stagnant waters in which their larvae mature provide mosquitoes with the environment needed for continuous breeding.

Each year, there are more than 225 million cases of malaria, killing around 781,000 people according to the WHO’s 2010 World Malaria Report, 2.23% of the total number of deaths worldwide. Malaria is commonly associated with poverty and can indeed be a cause of poverty and a major hindrance to economic development. Ninety percent of malaria-related deaths occur in sub-Saharan Africa, the majority young children.

Malaria transmission can be reduced by preventing mosquito bites. Prevention can occur by using inexpensive ($5) mosquito nets and insect repellents, or by mosquito-control measures such as spraying insecticide inside houses and draining standing water where mosquitoes lay their eggs. Malaria can be treated with multiple drugs, but the malaria protozoa have developed resistance to many of the historically effectives antimalarials, most notably Quinine, which was the most effective and popular drug for many years.

Figure 1.2.5: Malaria is transmitted by the Anopheles mosquito.
List 4-5 behaviors we have developed to avoid infectious disease transmission and explain what they are for.

Practice Calculation: Percent Error
When scientists are trying to find out how close their calculation is from the ideal, they calculate the percent error.

\[
\text{Percent Error} = \frac{(\text{Actual} - \text{Estimate})}{\text{Actual}} \times 100\%.
\]

For example: If I guess that there are 350 jelly beans in a jar, and there are actually 379, what is my percent error?

Answer: \(\frac{(379 - 350)}{379} \times 100\% = 7.65\%\) This means that this guess was 7.65% inaccurate.

What is the percent error if you guess that there are 466 jelly beans in the jar, and there are actually 379?
STUDENT RESPONSES

Remember to identify your sources.
Bacterial structures

Before we can discuss processes used to identify infectious diseases, details about specific diseases, and immune responses, we need to become familiar with the structures of bacteria, viruses, and immune barriers. In this lesson we will focus on the structures of bacteria that directly relate to infectious diseases. As we will see here and in future lessons, these structures are often precisely adapted in microbes that cause disease.

The Bacterial Envelope

When bacteria attempt to grow in, or colonize, an organism, the normal immune system responds to the foreign invader immediately. The immune system recognizes the surface of the bacteria called its envelope. The bacterial envelope is composed of a capsule, plasma membrane and cell wall. Because the immune system is so vigilant, pathogenic bacteria must try to stay one step ahead of the immune system by continually modifying their surface components.

The Capsule is camouflage and protection from the environment

The capsule is not essential to bacterial life and not all bacteria have one, but those that do have an advantage in infecting their hosts. The capsule is composed of a slimy layer of sugars and lipids. It plays two roles: First, it is durable and so can protect the bacterium from physical stresses such as osmotic challenges. Second, it camouflages the bacteria from the immune system. It can do this because the capsule consists of the same sugars that are found on the surface of the infected host's cells. The immune system therefore doesn't recognize the bacterium as a foreign interloper and doesn't target it for death. Examples of bacteria that use a capsule to evade immune system recognition are the Streptococci that cause both strep throat and 'flesh eating' disease, the Pneumococci that cause pneumonia and the Meningococci that cause meningitis.
The Cell Wall protects the bacteria from environmental stresses
In contrast to the capsule, all bacteria have some kind of cell wall. It is located internal to the capsule (if there is one) and external to the plasma membrane. The cell wall reinforces the cell membrane and protects it from environmental stress. If we think about the stresses bacteria face in their natural environment, the reasons they need a cell wall become clear. For example, intestinal bacteria, such as E. coli, are constantly exposed to bile salts that have detergent-like properties able to dissolve an unprotected cell membrane. The cell wall plays key roles in bacterial infections, which will be addresses by groups 2 and 3.

As in eukaryotic cells, the Plasma Membrane separates the cell from the environment
The bacterial plasma membrane lies internal to the cell wall. It is similar in many respects to eukaryotic plasma membranes, but it also plays specific roles in bacterial infection. For example, the bacterium Streptococcus exports toxins across its plasma membrane. These toxins give rise to sore throat.

The Acid-Fast solution to membrane protection: slow growing but very tough and well camouflaged
Acid-fast bacteria have cell walls that contain large amounts of waxes. This protective cover makes them impervious to many chemicals and able to avoid being killed by immune cells. The cost of this protection, is that they grow very slowly, probably because they cannot take up nutrients very rapidly. For example, the tubercle bacillus divides only once every 24 hours. Only a few pathogenic bacteria are acid fast. One notable example is the tubercle bacillus that causes tuberculosis. Its acid-fast coating means it can wall itself off from the immune system when it infects the lungs. Because of this, and because it divides very slowly, the infection can persist for a long time.

Figure 1.3.1: The bacterial envelope consists of the capsule (if there is one), the cell wall, and the plasma membrane.
Lesson Materials

The Gram-positive solution to membrane protection

All bacteria protect their cell membranes with a thick exterior cell wall that plays a critical role in maintaining shape and rigidity. In Gram-positive bacteria the wall is made of a polymer composed of sugars and amino acids - called murein. Bacteria are the only organisms to have murein. The murein on the surface of a Gram-positive bacterium can absorb a purple dye - the Gram stain – hence the term ‘Gram-positive’. This is used to identify Gram positive bacteria in an infection.

How does the murein work?

Murein is composed of sugar chains that are cross-linked to one another in an organization resembling a chain link fence. Layers of murein are wrapped around the length and width of the bacterium to form a sac. Depending on the shape of the murein sac, Gram positive bacteria may resemble rods (bacilli) like Bacillus anthracis, the bacterium that causes anthrax. Other Gram-positive bacteria may resemble spheres (coccis) like the Staphylococcus aureus that causes MRSA.

Why is murein useful?

The thick dense layer of murein allows bacteria to survive in environments where the osmotic pressure (pressure on the membrane) is high. This allows the bacteria to live in solutions that have a low or high salt concentration. However if the murein coat is breached the bacteria will burst.

Gram positive cell wall components can cause illness.

Gram-bacterial cell walls contain other unique polymers such as the lipid (fat) molecule LPA. Both murein and LPA are involved in how the immune system recognizes an infection has occurred. Taking the Gram-positive bacterium Staphylococcus aureus as an example: the staphylococcus bacterium produces a slimy capsule, but in its case the capsule cannot camouflage it from the immune system. Instead, cells of the immune system can recognize both the murein and LPA on the S. Aureus cell wall. The immune system then initiates a response to the invading bacteria that gives rise to typical symptoms of bacterial infection such as fever.

For a complete list of defined terms, see the Glossary.
The Gram-negative solution to membrane protection

Gram-negative bacteria have adopted a radically different solution to the problem of how to protect their plasma membranes. Their cell wall also has a murein component for rigidity, but it is far less prominent than in the Gram-positive cell wall. Instead, Gram-negative bacteria build a second membrane external to the murein wall. This outer membrane is just like other typical biological membranes, except it contains molecules called phospholipids that are unique to Gram-negative bacteria. Because of this cell membrane Gram negative bacteria fail to absorb the purple Gram dye, instead they remain pale pink.

The unique phospholipids of Gram-negative bacteria can cause symptoms of disease.

The major phospholipid found in the Gram-negative outer membrane is LPS. Like the LPA found in Gram-positive bacteria, LPS is critically important for infection and stimulates a strong immune response. LPS is very potent - even small amounts of LPS in the bloodstream will cause the host to become severely ill. The LPS is different in each Gram-negative species. As a consequence each different species of Gram-negative bacteria stimulates the immune system to produce different specific antibodies against it. These antibodies can then be used to determine which Gram-negative bacterium is present in an infection.

The Gram-negative cell wall can inhibit antibiotics.

The inner membrane of Gram-negative bacteria is surrounded by the murein layer for strength together with a gel-like solution of enzymes. These enzymes play important roles in disease. One enzyme in particular - β-lactamase - can inactivate certain types of antibiotics like penicillins and cephalosporins that interfere with the synthesis of the bacterial cell wall. For this reason Gram-negative bacteria are somewhat more resistant to antibiotics than Gram positives. Examples of the many disease-causing bacteria with Gram-negative cell walls are the Escherichia coli (E.coli) that cause intestinal diarrhea (hamburger disease) and Haemophilus influenzae (H. influenzae) that causes flu-like symptoms.

The complex architecture of the Gram-negative cell wall must work very well because in nature (but not necessarily in the human body) Gram-negatives outnumber Gram-positives!

**Figure 1.3.3:** Gram negative bacteria have an additional outer membrane external to the murein cell wall. The outer membrane also contains LPS. The red shapes in the lipid bilayer are membrane proteins.
Flagella - How bacteria get around

Many successful pathogens are actively motile, which helps them spread in both the environment and the body. This motility is largely produced by long helical flagella. Depending on the species, bacteria may have one or several flagella.

Why bacteria need to move.

Flagella allow bacteria to move toward substances that attract them – such as nutrients and away from those that they want to avoid such as the cellular predators of the immune system. This process of movement is called chemotaxis. Swimming allows bacteria to chemotax toward or away from a stimulus.

How flagella help bacteria move.

Flagella are attached to the surface of one end of the bacterium. When they spin around counterclockwise, the movement will push the bacteria forward in a straight line like a propeller. If the bacterium has several flagella, they will form a bundle. When all the flagella in a bundle spin counterclockwise the bacteria will also be propelled in a straight line. However, if any of the flagella begin to rotate clockwise, their movement becomes erratic causing the bacteria to tumble randomly. These two types of motion, swimming and tumbling, both occur during chemotaxis. In the absence of a stimulus bacteria alternate between swimming and tumbling and move randomly, but when an attractive stimulus, like nutrients, or a negative stimulus, like a white blood cell, appears, the flagella begin to rotate counterclockwise so that swimming predominates. Then the bacteria can move towards the stimulus or away from it.

How flagella impact disease.

Flagella play a direct role in disease: The protein that makes up flagella (named flagellin or the bacterial H antigen) can be recognized by the immune system, so bacteria have to continually modify it in an attempt to camouflage themselves. For example, the Salmonella species that causes food poisoning (Salmonella Typhimurium) has two different kinds of H antigen - one H antigen stimulates an immune system response and one does not. When S. typhimurium is outside of the body it expresses the H antigen that can stimulate immune responses, but as soon as it infects its host, it switches to the H-antigen that cannot stimulate the immune system. In this way it is able to infect its host and avoid detection.
**Pili - How bacteria stick around**

Some sites in the host are very inhospitable to infection. For instance, the nasal cavity is continually being cleansed by sneezing, while swallowing washes contents of the mouth down into the harsh acid environment of the stomach. Bacteria that need to survive these challenges use special structures—pili—to attach to cells of the host or other bacteria.

**Pili are used to stick to surfaces.**

Like flagella, pili also protrude from the cell wall, but unlike flagella they are used to attach to specific surfaces and not for movement. Pili are shorter than flagella and often distributed in large numbers over the entire surface of the bacteria. During attachment, the tip of the pilus attaches to cells or other bacteria, in a process that resembles the use of grappling hooks or Velcro. In addition to these “common pili,” some bacteria also use sex pili to link donor (male) and recipient (female) cells before transferring DNA between them.

**How pili impact disease.**

Like flagella, pili are often recognized by the immune system, so many bacteria also continually vary the types of pili they produce. This enables them to keep one step ahead of the immune system. For example, Gonococci that cause gonorrhea have a large number of genes that code for different pili proteins. However, they only make a few at any given time. The immune system rapidly makes antibodies against that pilus protein and destroys the bacterium that is producing it. As a result, there will be a rapid selection in favor of any gonococci bacteria that are making pili that cannot be recognized by the antibodies. In this quick-change scenario, the bacteria keep one step ahead of the immune system. This is why attempts to immunize against gonococci using a vaccine that stimulates production of only one kind of antibody have failed so far.

**Spores - How bacteria stay alive**

When environmental stresses become too harsh simply sticking to a surface is not enough. Some bacteria wait out harsh stresses such as starvation, desiccation, heat and radiation until more favorable conditions appear. They do this by forming a spore by condensing its cell within a protective coat. Each bacterium usually makes only one spore.

**How spores impact disease.**

Some particularly important pathogenic bacteria form spores. The *Clostridium* species that cause tetanus and botulism are key examples. Tetanus spores can lie dormant in the soil for many years before becoming reactivated in the presence of a skin wound, while botulinum spores are resistant to heat and can survive food canning processes. They can cause food poisoning when the food is eaten.
DEFINITIONS OF TERMS

**Nucleoid** - a region within prokaryotes that contains the nuclear material, such as the DNA. It is not enclosed by a membrane.

**Plasmid** - a DNA molecule that is separate from and can replicate independently from the chromosomal DNA.

For a complete list of defined terms, see the *Glossary*.

**LESSON MATERIALS**

**Nucleoid and plasmid - what about their genes?**

Bacterial genes are in the nucleoid.

Bacteria do not have a nucleus like eukaryotic cells. Instead there is a DNA-rich area in the cytoplasm called the **nucleoid**. Also unlike eukaryotic cells that store DNA in many thousands of genes organized in several structures called chromosomes, bacteria nucleoids contain genes that are usually in only one chromosome. Because no nuclear membrane separates DNA transcription from protein synthesis, both processes can occur at the same time and as a result bacteria can divide very rapidly. This adaptation makes bacterial growth and replication extremely efficient, and so bacteria can quickly take advantage of a favorable environment. Bacterial genes obviously play a key role in their ability to cause disease because they determine whether they are Gram positive or negative, have multiple types of flagellae or pili and whether they can form spores.

Bacterial genes are also found in plasmids.

Unlike eukaryotic cells whose genes are confined to chromosomes, bacteria also have small circular pieces of DNA called **plasmids**. Although these plasmids are not critical to the life of the bacterium itself, they play important functions in disease. For example, some plasmids contain genes that produce toxins, such as those secreted by *Streptococcus* species that cause food poisoning. In fact, the two most potent toxins known to man - tetanus and botulinum toxin - are produced from plasmids that are found in different Clostridium species.

Plasmids can also contain genes that make the bacteria resistant to specific antibiotics. The advantage of plasmid DNA, rather than chromosomal DNA, is that plasmids are relatively easy to transfer between bacteria, using their **sex pili**. This exchange of advantageous genes is of great benefit to the bacteria but causes a public health nightmare. It is easy to imagine how bacteria swapping drug resistance genes can rapidly become problematic. *Vibrio cholera*, which causes cholera, routinely swaps genes via plasmids. Recently two species – one of which was highly infectious and one of which displayed significant drug resistance were found to have combined to make a single highly infectious species that was resistant to many more antibiotics. This new cholera species is now producing epidemics throughout the third world.
### STUDENT RESPONSES

<table>
<thead>
<tr>
<th>Bacterial structure</th>
<th>Description</th>
<th>How is it advantageous?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Capsule</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gram-positive cell wall</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gram-negative cell wall</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plasma membrane</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Murein</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LPA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LPS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flagella</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pili</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spore</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nucleoid</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plasmid</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
In this lesson we will focus on the structures of viruses that directly relate to infectious diseases. As with bacterial structures, viral structures are precisely adapted, in this case for replication. You may also notice that many of the structures are quite similar those that you saw in the last lesson, in fact you may be able to predict the function of the viral structures by extrapolating on what you know about bacteria structure and function.

**Just how small are viruses?**

As we mentioned before, microbes come in a variety of flavors. The larger microbes, such as fungi and many parasites, are eukaryotic, which means they have a membrane-bound nucleus. Bacteria are prokaryotic, which, as we saw before, means they lack a membrane-bound nucleus. Viruses are neither prokaryotic nor eukaryotic, in fact, they are not even alive! They are simply a collection of genetic material and proteins that are able replicate with the help of the cell they infect. Viruses are much, much smaller that bacteria and this impacts every aspect of their structure, how they replicate and how they infect. Their small size also impacts how we are able to detect them, because they are too small to see under a normal microscope.
How big are viruses compared with other pathogens?

The microbial world generally exists below the limit of what the unaided human eye can see. Sizes can vary considerably within each category. Eukaryotes are the biggest, followed by prokaryotes, and then by viruses. Viruses can vary in volume by a thousand fold. The largest viruses fall within the lower size range of prokaryotes - similar to the size of small bacteria such as *chlamydiae* that cause chlamydia infections. Only the largest viruses such as *smallpox virus* are visible with the light microscope, and then only barely.

Because the differences in microbe sizes are so large we need more than one unit of measurement. Most bacteria are on the order of 1 micron in diameter. A micron, or a micrometer, is one millionth of a meter. To give you an idea of how small this is, bacteria are so small that when they are in suspension in a fluid like urine, it only becomes cloudy only after there are between one million to ten million bacteria per milliliter. This means that even when water looks completely clear, it can contain millions of bacteria. Viruses are even smaller, at about one one-hundredth of the size of a bacterium. To give you a sense of scale, if a virus were the size of an orange, a bacterium would be about the size of a sofa.

Practice Calculation: Conversion

Conversion between units is important for every aspect of science. Fortunately in this course we are only concerned with a few conversions that relate to microbe size. So let’s practice:

**How big is a micron?**

1 millimeter$= 1 \times 10^{-3}$ meters or 0.001 meters
1 micron $= 1 \times 10^{-6}$ meters, or 0.000001 meters
1 nanometer $= 1 \times 10^{-9}$ meters or 0.000000001 meters

**For example: How many microns are in a millimeter?**

**Answer:**

1 micron $= (0.000001$ meters)
1 millimeter $= (0.001$ meters)

A millimeter is 1000 times bigger than a micron so there are 1000 microns in a millimeter.
Viral structure: It's all about delivering the genome to the host cell.

Now back to viral structures. When you are reading about viral structures it is important to keep in mind that viruses require their host cells to supply everything they need to replicate with the exception of a few key viral proteins and the viral **genome**. For this reason viruses can be thought of simply as vessels that transport genetic information. Because the virus uses so many of the host cell proteins to replicate, it does not need to code for these genes because the host cell does. Hence viral **genomes** are much smaller than either bacterial or eukaryotic genomes. Viral genomes can be either **DNA** or **RNA**, as we will discuss later.

Viral genomes are packaged in two structures:
1. The protein coat (capsid)
2. The envelope

**The capsid protects the genome:**
All viruses have capsids that protect their genomes. The capsid can be a single or double layer, and fits into one of three symmetrical patterns: **icosahedral**, **helical or complex** (as shown above left). The capsid is composed of proteins, without the lipids that are in envelopes and other membranes. Some of the capsid proteins are not found in the host and are required for viral replication. For naked or non-enveloped viruses the capsid is the only protection the genome has from the environment.

**The envelope is made of host cell lipids/proteins and viral proteins:**
Only some viruses have an envelope. For those that do, it is vital for their function. The envelope is made of viral proteins that help viruses enter host cells, plus lipids and proteins ‘stolen’ from the cell membrane of the host cell. The inner surface of the envelope has virus-specific proteins that contacts the capsid. These proteins stabilize the interactions between the lipids in the envelope and the capsid. As we will see later, they also determine where the virus will assemble in the host cell.

**The genome**
- Is inside the capsid.
- Can be DNA or RNA.
- Codes for viral proteins.

**Figure 1.4.1:** The viral genome sits inside the capsid.

---

**DEFINITION OF TERMS**

*Genome* - the genetic material a cell (or a virus) uses to replicate itself.

*Icosahedral* - a regular polygon with 20 identical equilateral triangular faces (top picture).

*Helical* – having the shape of a helix or spiral (middle picture).

*Complex* - having a structure that is neither purely helical nor icosahedral, possibly with additional structures (bottom picture).

For a complete list of defined terms, see the Glossary.

---

**Workbook**

**Lesson 1.4**
The four stages of the viral life cycle

The rest of this reading describes viral structures in the context of how they function. To simplify we can divide the viral life cycle into four steps:

1) attachment, 2) entry, 3) replication, 4) exit.

We will focus on viral replication later. While you are reading keep in mind that, like bacterial structures, each viral structure performs its own unique key function.

The virus has one goal – to make more virus. To reproduce, viruses need to enter their host cell where they make more virus and then leave. First, the virus attaches to the cell it is going to infect, then the virus enters the cell and uncoats its genome so that it can begin to replicate. Viruses replicate their genomes and make the structural proteins they need to assemble a new virus particle. Once the virus has made all its parts it assembles new virions that exit into the environment to infect other cells.

1) Attachment usually requires a specific receptor.

Both naked and enveloped viruses need to attach to a host cell before they can enter it. To do this all viruses have receptors on their surface that act like ‘keys’, which interact with select receptors on the host cell surface that act like ‘locks’. For example, HIV is an enveloped virus that uses the receptor protein gp120 to attach to a receptor on the host cell surface called CD4. Like other viruses, HIV can only infect cells that have the right host receptor. This means that different viruses have affinity for different cell types. In the case of HIV, only cells of the immune system like helper T cells and dendritic cells have the CD4 receptor.

All viruses have affinity for particular host cell targets, called tropism. Consider influenza virus. Influenza virus has a receptor in its envelope called hemagglutinin (the H in H1N1). Hemagglutinin binds to sialic acid sugars that are on epithelial cells in the respiratory tract – it is tropic for those cells. Different variants of influenza have different ‘H’ receptors that attach to different sialic acid sugars. These different sialic acid sugars are located on different cells in the respiratory tract. This means that different variants of H1N1 infect different sites in the respiratory tract.

Why would a virus need to enter a cell in order to reproduce?

Could an HIV virus infect any cell? Why or why not?

What do ‘H’ and ‘N’ stand for in H1N1?

What does Hemagglutinin bind to on host cells? How might binding of Hemagglutinin impact how severe the disease is?
2) Entry depends on whether the virus is enveloped or naked.

In order to replicate all viruses must cross the plasma membrane and get inside the host cell. As we will see, enveloped and naked viruses enter cells differently.

**Enveloped Viruses fuse with the host membrane.**

The most direct way for enveloped viruses to deliver their capsid to the host cell cytoplasm is to fuse their own envelope membrane with the membrane of the host. Because both viral envelopes and host cell membranes are composed of similar host cell membrane lipids, fusion happens easily, like two oil droplets combining in water. After the membranes have fused the virus can then spill its capsid, which contains its genome, into the cytoplasm. HIV is an example of a virus that fuses its envelope with the T cell membranes like this.

A less direct route enveloped viruses can take is via endocytosis, using the same mechanism the host cell uses to take up molecules from the extracellular space. A virus particle that hijacks endocytosis to enter the host cell ends up in a vesicle, called an endosome. Once in the cell inside the endosome the virus gets its capsid into the cytoplasm by fusing with the endosome membrane - just like the other virus did when it fused with the plasma membrane. Influenza is an example of a virus that enters the cell by endocytosis and then fuses its membrane with the endosome membrane. Once it is in the cytoplasm the virus can begin to replicate.

**Naked viruses cross the host membrane**

Naked viruses aren't coated in a lipid envelope, so they can't fuse with membranes. Instead, naked viruses either inject their genomes into the host cell cytoplasm through the plasma membrane, or are taken up through endocytosis. Either way the virus needs to get its capsid through host cell membranes into the cytoplasm. Some very small naked viruses like Poliovirus can ‘feed’ their genomes through tiny pores present in the endosome membrane. Other naked viruses release enzymes that punch holes in the host cell endosome membrane before injecting their genome and other viral proteins into the host cell cytoplasm. Again, once the virus has its genome inside the host cell it is able to replicate.

**DEFINITIONS OF TERMS**

**Cytoplasm** - the inner material of the cell that is enclosed by the plasma membrane and surrounding the nuclear membrane (when present).

**Endocytosis** - the process by which cells absorb small molecules by engulfing them. It differs from phagocytosis, which is used to internalize large particles like bacteria, in the size of particle ingested.

**Extracellular** - outside of the cell.

**Endosome** - a membrane bound vesicle that is produced when a cell engulfs material from the extracellular space.

**Workbook**

**Lesson 1.4**

**LESSON MATERIALS**

Describe how enveloped viruses get their genomes into the host cell cytoplasm. Make sure you understand how endocytosis works.

Describe how naked viruses get their genomes into host cell cytoplasm.

Why would a naked virus in an endosome use enzymes for?
3) Exit also depends on whether the virus is enveloped or naked.

Once replication has occurred, the virus must leave the host cell so it can carry on its cycles of infection and replication. Again viruses have two different exit strategies depending on whether they are enveloped or naked.

Enveloped viruses bud off of host cells:
Enveloped viruses leave the cell in distinct stages. First, the envelope proteins collect in the host cell membrane close to where the genome is making the capsid proteins. Then the genome and capsid proteins travel together to the site on the host cell membrane where the envelope is located, and attach to the envelope proteins on the inside of the membrane. Next, the fully formed capsid containing the genome buds out of the host cell. As it does so it gathers the virus envelope proteins and ‘stolen’ host membrane surrounding the capsid and forming a complete new virus particle or virion. This is true of both HIV and influenza. Whether or not the budding off of virus particles will cause the host cell to die depends on how fast the virus is replicating; if a virus is replicating slowly the cell may survive, but quickly budding viruses will probably cause the host cell to die.

Naked viruses lyse the host cell:
Naked viruses employ a rather more straightforward exit strategy than enveloped viruses. Quite simply, the virus replicates rapidly, and the huge numbers of virus particles that accumulate in the cytoplasm burst the cell open by force. This process of rupturing a cell is termed lysis. It is an important phenomenon and we will use the term routinely to describe when a cell is broken open. An example of a naked virus that exits by budding is human papilloma virus or HPV, the virus that causes genital warts, and that is linked to forms of cervical cancer.

What might be some advantages to budding slowly?

What might be some advantages to budding quickly?

What will happen to the host cells if they are lysed?

Figure 1.4.6: Enveloped viruses release by budding. Naked viruses lyse the host cell.
The exit strategy must also prevent viruses re-entering into the same infected cell:

An important part of exiting the host cell successfully is not re-infecting the same cell. Think about this - if a virus keeps re-infecting the same host cell it will not be able to spread and eventually the host cell too will die. Without being able to replicate the virus will eventually disappear. In fact, mutations that prevent influenza from detaching from their host cell result in disabled viruses that are not virulent.

Since viruses are designed to attach to and then enter a cell they are coated in receptors, like the Hemagglutinin receptor we discussed previously, that bind very specifically to their host cells. How do they prevent themselves from remaining attached to the host cell and reinfecting the same cell, rather than moving off to infect new targets? Even though not all enveloped viruses kill their host cells, a host cell that has been hijacked by a virus will no longer be healthy, so re-infection of the cell would be unproductive for the virus.

The Influenza virus solves the problem of detachment very neatly by carrying another protein in its envelope, the enzyme **Neuraminidase** (the N in H1N1). Neuraminidase is able to cut the host cell sialic acid, destroying it. This lowers the number of sialic acid molecules on the host cell, thereby preventing any newly budded virus particles from using them to either remain attached, or to reattach to the host. Conversely, inhibiting Neuraminidase function will prevent viruses being released from the host cell. This in turn will reduce the number of infective viruses that are circulating (the viral load). The drugs Tamiflu and Relenza target Neuraminidase and inhibit it, thereby reducing the number of infective virus particles and the viral load.

**Why doesn’t H1N1 re-inflect the same cell?**

**Would a naked virus have the same re-infection problem that a enveloped virus has? Why or why not?**

**How do anti-influeza medications like Tamiflu work?**

For a complete list of defined terms, see the **Glossary**.
**LESSON 1.5 WORKBOOK**

**So why aren’t we always sick? Our body’s defenses.**

We are covered in bacteria and exposed to viruses all the time but we rarely get sick – why not? This lesson will focus on immune barriers that separate and protect us from pathogenic microbes in the environment. Again, it is important to pay attention to how the immune barriers function because if they become damaged or pathogens adapt to get around them they will lose their protective function.

**Our natural defenses protect us from pathogens**

We’ve now discussed several examples of how microbes can come into contact with humans to cause disease, but contact is not enough. The microbe must also evade our body’s natural defenses and possibly outcompete our residential microbes (*commensal flora*) that are happily coexisting with us.

First, let’s define the **sterile** and non-sterile, inside and outside of the body. Although this might seem like common sense, you may be surprised. Many of the locations that you may think of as being inside the body are actually considered to be on the outside. For example, the outside of the body includes not only the surface of the skin but any location that contacts the environment. Some outside locations, like the mouth, digestive tract and the lower genital tract are usually highly populated with microbes and are therefore non-sterile. In contrast, other outside locations are not populated with microbes and are therefore kept sterile. For example, the conjunctiva of the eyes, the lower respiratory tract and the upper genital tract.

It is critically important to keep the inside of the body sterile. The inside is therefore anything beneath the surface layers of the body, such as the brain, circulatory system, and the peritoneal cavity. As we will see in this lesson microbes are kept at bay in sterile areas by a series of protective barriers, which can be physical, molecular, or cellular. Surprisingly, our coexisting commensal microbes themselves also constitute a 2nd line of defense against infection.

---

**DEFINITIONS OF TERMS**

**Commensal flora** - the microbes that normally inhabit our bodies and co-exist with us without causing disease.

**Sterile** - free of biological contaminants, including bacteria, viruses, fungi, and parasites.

**Commensalism** - a symbiotic relationship in which one organism derives benefit and the other is unharmed.
**LESSON MATERIALS**

**The 1st line of defense: Chemical and physical barriers**

Epithelial cells separate the sterile inside of the body from the environment.

Epithelial cells are the major physical barrier between the outside and inside of our bodies. The epithelium is constructed very much like a wall with very different top and bottom surfaces, and tight connections at the side. The top *apical side* always faces the outside world - whether this world is the non-sterile surface of your skin, the contents of your intestines or the sterile depths of your lungs.

In contrast, the bottom, *basal side* always faces the inside of the body and it is critical to keep this world sterile. To restrict microbes to the correct (apical) side of the epithelia and away from the interior, the epithelial cells are normally sealed together tightly to prevent microbes from sliding between them.

As we will see next, different locations require highly specialized types of epithelium. For example, the non-sterile mouth, pharynx, esophagus, and lower urinary tract, all of which are heavily colonized with commensal microbes, have several layers of epithelia that are covered with a coat of mucus that provides a chemical barrier against infection. On the other hand, the epithelium of the non-sterile upper respiratory tract has a mechanical barrier - it is ciliated and the *cilia* brush particles out of the lungs, as we have seen before. In contrast the epithelia of the lower respiratory tract (the alveoli) are sterile and without a barrier. They form a delicate single layer, specialized mainly to exchange gases.

**DEFINITIONS OF TERMS**

**Conjunctiva** - a clear mucous membrane that covers the whites of the eyes and lines the eyelids.

**Peritoneal cavity** - the space in between the organs in the abdominal cavity and the abdominal wall.

**Commensalism** - a symbiotic relationship in which one organism derives benefit and the other is unharmed.

**Ciliated** - having cilia, or mobile finger-like organelles.

For a complete list of defined terms, see the Glossary.

**Workbook**

Lesson 1.5

---

Which locations in Figure 1.5.1 are considered to be part of the inside or outside of the body?

**Inside**

___________________________________

___________________________________

___________________________________

**Outside**

___________________________________

___________________________________

___________________________________

Which locations are considered sterile and non-sterile?

**Sterile**

___________________________________

___________________________________

___________________________________

**Non-sterile**

___________________________________

___________________________________

___________________________________

What surface of epithelial cells faces the A) outside environment and B) inside environment?

**A)**

___________________________________

**B)**

___________________________________

What prevents microbes from passing between the epithelial walls?

___________________________________

___________________________________

---

**Figure 1.5.1:** Interior areas of the body not connected to the outside are sterile, interior areas of the body connected to the outside

**Figure 1.5.2:** Three columnar epithelial cells standing side by side.
The skin is a thick and durable type of epithelium. The skin is a specialized epithelium that is a highly effective physical barrier. It has three layers: the epidermis, dermis, and hypodermis. Most of the cells of the epidermis are dead. They are continually shed as newer skin forms below. The epidermis is enriched with a protein keratin that is impermeable to most microbes and very resistant to mechanical stress. Because of this, the dermis and the hypodermis that are beneath the epidermis can normally be kept sterile.

What if epithelia that are structured to prevent passage of microorganisms becomes damaged, for instance by abrasion? As a second line of defense, the epithelia of the skin and mucous membranes contain chemical and molecular antimicrobial factors on their surfaces. Some of these factors are always present, while others only appear if a microbial breach appears.

Chemical barriers on epithelial surfaces restrict colonization

Some epithelial membranes are covered with a layer of mucus. Mucus, as you may know, is a viscous gel. It entraps particles and prevents them from reaching the apical membrane of the epithelium. Once entrapped, particles like viruses and bacteria can be swept away by the cilia on the epithelial cells. The mucus and particles are then moved out of the body or are swallowed. In the lungs, the cilia prevent microbes from reaching the sterile depths of the alveoli in lower lungs. Mucus can increase during times of infection and is important for preventing infection. Consider the coughing reflex that normally prevents material in your mouth or pharynx being getting into your lungs. The coughing reflex is less productive in smokers because smoking paralyzes the cilia, thereby preventing them from sweeping debris and microbes out of the lung. Since the cilia can’t function, aspirates are more likely to enter the lungs. Even a small amount of a virulent strain of pneumococcus inhaled into the lungs may be sufficient to cause severe disease. This is why smokers are very susceptible to respiratory infectious like bronchitis.

Why is the epidermis so effective at keeping microbes out of the body?

Chemical and molecular barriers:

Why do smokers have a high incidence of respiratory infections?

For a complete list of defined terms, see the Glossary.
Molecular barriers on epithelial surfaces restrict colonization

The skin and mucus membranes also host several specific molecules that work to prevent microbial invasion. Many of them are always present, while others are only made when high numbers of bacteria appear. For instance, the enzyme lysozyme is present in large amounts in tears, sweat, saliva, and in the blood. Lysozyme acts mainly to digest the cell wall of Gram-positive bacteria. Remember, Gram-negative cell walls are shielded by an outer membrane, so lysozyme only works on Gram-negative bacteria when their outer membrane is damaged.

Mucus also contains antibodies (more about antibodies in detail later) and antimicrobial peptides like the defensins, that are made by all vertebrates and invertebrates. Humans have more than 30 defensin genes. Defensins, like most other antimicrobial peptides, are positively charged. This means they can bind to the negatively charged cell walls of bacteria and fungi. They then induce pores to be made in the microbial membranes, killing them. As you might expect, our own membranes are resistant to the actions of defenses.

If microbes do manage to cross the skin and mucous membranes they will encounter even more powerful molecular and cellular immune defenses in the underlying tissues.

The Other Kind of Host Defense: Our Normal Flora

Throughout our history we have coexisted and co-evolved with the microbial world. During that time our bodies have designated some spaces as safe to share with microbes. As a result a healthy human body normally contains thousands of species of bacteria and a smaller number of viruses, fungi, and protozoa, chiefly located in moist areas of the skin such as the groin and between the toes, the upper part of the respiratory tract, the mouth and large intestine, the lower parts of the urethra and the vagina. In these sites the number of microbes varies widely but the densest possible packing (about a million per milliliter) is found in the pockets around the teeth and in normal feces, which is about one-third bacteria by weight. The so-called commensal organisms or normal flora are extremely complex. For example, the intestinal flora of just one person has about 400 distinct species of bacteria. Most commensal flora coexist with humans without causing harm.

To put this in perspective consider the following: our bodies are composed of ten times as many bacteria as human cells. In this respect we are an ecosystem.
Any newly invading microbe with plans to colonize the human body must not only resist host mechanisms that could dislodge or kill them, they must also compete with the normal flora. This is especially true in sites like the intestine, where normal flora takes up all the free space. Colonization by commensal bacteria takes place within days after birth, when bacteria are transferred from mother to child. The intestinal flora of an individual is usually remarkably constant, and colonization by a new species is infrequent. The normal flora can resist challenges from newly ingested bacteria. They do this by producing substances like antibiotics or lethal proteins called bacteriocins that inhibit the newcomers from growing.

Not surprisingly, competition between microbes does not play a big role at sites that normally carry a sparse load of microbes, most of which are in transit (such as the lower part of the respiratory tract, the stomach, the urinary bladder and the uterus). In fact, if a pathogenic microbe lodges here, alarm bells will sound and our body will immediately respond to eliminate it.

Normal Flora in Disease – opportunistic infections
Under the right circumstances even commensal microbes can cause disease. In almost all cases, this happens when the normal flora find themselves in the normally sterile sites of the body. For example, *Staphylococcus epidermidis* is a common commensal organism on the skin. However, it can form a biofilm which attaches tightly to the plastic surfaces of intravenous catheters causing severe bloodstream infections.

This type of infection is called opportunistic because a failure of the immune barriers presents an ‘opportunity’ for otherwise harmless commensal bacteria to be pathogenic. However, not all members of the normal flora have the potential to become pathogens. For example, if the intestinal wall breaks (perhaps as a result of a ruptured appendix) and releases intestinal bacteria into the peritoneal cavity, the resulting infection is caused by only a few of the many bacterial species that normally live in the intestines.
What happens if microbes gain access to sterile sites of the body?

Molecular barriers in the blood and immune cells

Chemical barriers like the complement system in the blood restrict microbial survival

The blood also contains chemical barriers that are always present and ready to respond to an infection. These barriers are called the complement system. Complement can be thought of as toxins produced by the body that will kill any cell that does not display an appropriate antidote protein. Fortunately all of our self-cells do display this antidote protein, however microbes do not. Thus, when microbes are exposed to complement they are killed.

The complement system is made up of more than 35 proteins that can lyse microbes. Like lysozyme, complement punches holes in bacteria or enveloped viruses by inserting donut-shaped protein complexes into their membranes. These pores allow water to enter the microbes and raises the internal osmotic pressure, eventually causing them to burst - lysis. Complement proteins can also mark microbes so they can be detected by and phagocytosed by immune cells, like painting a target.

Not surprisingly, microbes have evolved countermeasures to evade all these attacks. One interesting example of how a microbe defends itself against complement is shown by herpes simplex virus (HSV): One of the surface proteins of HSV is an ‘antidote protein’ that prevents complement-mediated lysis of the virus.

Innate immune cells are the first cells to respond to an infection.

Innate immune cells have a defined set of receptors on their surfaces that can detect microbes. However, unlike cells of the adaptive immune system they do not learn to respond to specific microbes.

Neutrophils

All epithelial surfaces are possible crossover sites for microbe entry into the sterile parts of the body. The large intestine in particular has a huge microbial population just one epithelial cell away from the host’s sterile interior. So, immune cells like neutrophils constantly patrol the basal surface of the epithelium to remove the rare microbe that manages to pass through the intact epithelium. Neutrophils respond to invaders quickly by chemical attack, phagocytosis, and then by sounding the alarm to alert other cells using cytokines.
Macrophages and Monocytes

Macrophages and monocytes come into play after the neutrophils have attacked. In fact the neutrophils play a role in recruiting the macrophages by releasing cytokines. Macrophages, like neutrophils, phagocytose invading microbes. In many ways macrophages are professional eaters because they are excellent at phagocytosis. They also contribute to the inflammatory response by calling other immune cells to help. For example, macrophages recognize foreign invaders and in turn help to activate B and T cells.

Monocytes and macrophages also turn their eating skills to mop up the dead host cells and debris left from fighting infections and normal cell death.

Adaptive immune cells are comprised of B and T cells and they are the last line of defense

If the innate cells cannot clear the infection on their own they call in the adaptive immune cells (B and T cells). Unlike innate cells, adaptive immune cells learn to respond to a select pathogen following an infection. In this way, they use their special receptors that are now specific to a particular microbe to protect against future infections by the same pathogen.

There are two types of adaptive immune cells, B and T cells, that are named according to the organs in which they develop; B cells develop in the bone marrow, and T cells develop in the thymus. Each B and T cell has one type of receptor that recognizes a very specific target. These are called the B cell receptor (BCR) and the T cell receptor (TCR) respectively. You may already know that BCRs are antibodies. We will discuss how B and T cells learn to recognize a specific infection in Unit 5. For now, think of them as the last line of defense.
Inflammation is an important part of the immune response

Have you ever felt inflammation? What does it feel like? Do you know what causes the heat, pain, and swelling?

Immune cells can cause inflammation in response to an infection or when tissue is damaged. The purpose of inflammation is to amplify the body’s defenses by increasing the blood supply to the site of infection. The increased blood supply will in turn bring more antimicrobial proteins and immune cells to fight the infection.

Early signs of inflammation are felt at the site of the infection/injury and include pain and/or swelling and a sense of heat and throbbing. As a result, the inflamed site may appear red, shiny, hot, and painful to the touch. The extent of the inflammation is usually proportional to the need the tissue has to control the infection. For example, external tissues (skin and mucus membranes) are constantly in contact with microorganisms and they can usually control them with a constant supply of antimicrobial peptides like the defensins we talked about before. Therefore, they will only become inflamed when exceptionally high numbers of pathogenic microbes are present because if they continually responded to the low numbers of microbes that are normally present, the inflammation would be constant and this could be damaging. In contrast, if microbes gain access to normally sterile areas like those under the skin, they need to be eliminated immediately and this may require a rapid response with robust inflammation.

Inflammation can also be felt systemically as fever, weakness, and disorientation. Essentially, systemic inflammation causes the symptoms we experience when we feel ‘sick’. Although unpleasant, it is an essential part of the immune response and is usually self-limiting; therefore, patients with infections should generally not be treated with anti-inflammatory agents. However uncontrolled inflammation, or inflammation that is not caused by infection may cause serious tissue damage and/or death. This type of uncontrolled inflammation is often seen in diseases like arthritis.

The Immune response

In reality, immune responses vary in their effectiveness against different types of pathogens. Which responses will be effective in providing protection is determined by the physical nature of the pathogen as well as its “lifestyle.” As we will see as we move forward, pathogens use various strategies to enter the body and stay hidden while they replicate.
### STUDENT RESPONSES

<table>
<thead>
<tr>
<th>Barrier</th>
<th>Description</th>
<th>How does it protect the host?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epithelial cells</td>
<td></td>
<td></td>
</tr>
<tr>
<td>The skin</td>
<td></td>
<td></td>
</tr>
<tr>
<td>The stomach</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chemical barriers</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Molecular barriers</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal flora</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Macrophages</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neutrophils</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phagocytes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B and T cells</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Remember to identify your sources.