Interaction between Microbe and Host

Chapter 15
BIO 220

Portals of entry

• Mucous membranes in respiratory, GI, and genitourinary tracts and conjunctiva

• Skin

• Parenteral route
  – Microbe deposition directly into the tissues beneath the skin or into mucous membranes when these barriers are compromised

Most pathogens have a preferred portal of entry that is a prerequisite to their being able to cause disease

If they gain access to the host by another route, disease may not occur
  – *Salmonella typhi*, *streptococci*

Some organisms can initiate disease from more than one portal of entry
  – *Bacillus anthracis*, *Yersinia pestis*
Numbers of invading microbes

- The more microbes that gain access to the host, the increased likelihood of disease
- The virulence of a microbe can be expressed as the ID$_{50}$, which is the infectious dose for 50% of a sample population
- For *Bacillus anthracis* the ID$_{50}$ is
  - Skin, 10-50 endospores
  - Respiratory, 10,000-20,000 endospores
  - GI, 250,000 to 1,000,000 endospores

<table>
<thead>
<tr>
<th>Pathogen/Host</th>
<th>Disease</th>
<th>Incubation Period</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mucous Membranes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Genital tract</td>
<td>Gonorrhea</td>
<td>3-8 days</td>
</tr>
<tr>
<td>Oropharynx</td>
<td>Tetanus</td>
<td>3-60 days</td>
</tr>
<tr>
<td>Oropharynx</td>
<td>Neisseria gonorrhoeae</td>
<td>3-14 days</td>
</tr>
<tr>
<td>Hepatitis A virus type 1</td>
<td>Hepatitis A</td>
<td>4-28 days</td>
</tr>
<tr>
<td>Human immunodeficiency virus (HIV)</td>
<td>AIDS</td>
<td>12 years</td>
</tr>
<tr>
<td>Listeria monocytogenes</td>
<td>Listeriosis</td>
<td>2-5 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Skin or Parenteral Route</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cutaneous pathogens</td>
<td>Gangrene</td>
<td>1-5 days</td>
</tr>
<tr>
<td>Clostridium toxin</td>
<td>Tetanus</td>
<td>2-21 days</td>
</tr>
<tr>
<td>Bacillus cereus</td>
<td>Bacillus cereus</td>
<td>9-13 days</td>
</tr>
<tr>
<td>Marburg Virus</td>
<td>Marburg Virus</td>
<td>6-16 days</td>
</tr>
<tr>
<td>Bacillus anthracis</td>
<td>Anthrax</td>
<td>10 days - 3 weeks</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2 weeks</td>
</tr>
</tbody>
</table>

*All pathogens are bacteria, unless noted otherwise. For viruses, the viral species causing the disease is given.
*These pathogens can also cause disease after entering the body via the parenteral route. Marburg Virus is the only pathogen that can also cause disease after entering the body via the parenteral route.

Adherence

- The attachment between pathogen and host is accomplished by means of surface molecules on the pathogen called adhesins or ligands that bind to complementary receptors on the surface of host cells.
- Adhesins may be found on the microbe glycopalyx, or other structures like pili, fimbriae, and flagella
- *Streptococcus mutans* – glycopalyx
- *Actinomyces* – fimbriae

Numbers of invading microbes

- The potency of a toxin is often expressed as the LD$_{50}$, which is the lethal dose for 50% of a sample population
- In mice
  - LD$_{50}$ is 0.03 ng/kg for botulinum toxin
  - LD$_{50}$ is 250 ng/kg for Shiga toxin
  - LD$_{50}$ is 1350 ng/kg for staphylococcal enterotoxin
Adherence

- Adhesins are usually glycoproteins or lipoproteins
  - Strains within a species may have different adhesins
- Receptors on the host cells are usually sugars
  - Can vary between cell types

Adherence

- Microbes can come together in masses, cling to surfaces, and take in and share available nutrients in communities called biofilms

Adherence

- Enteropathogenic strains of *E. coli* have adhesins on fimbriae that adhere to receptors present on only certain cells in parts of the small intestine
How bacterial pathogens penetrate host defenses

- **Capsules**
  - Capsule formation increases the virulence of a species
  - Capsules resist phagocytosis by preventing attachment of the phagocyte to the microbe
  - Antibodies against the capsule will ultimately result in the destruction of the microbe

- **Streptococcus pneumoniae, Klebsiella pneumoniae, Haemophilus influenzae, Bacillus anthracis, Yersinia pestis**

- **Cell wall components**
  - *Streptococcus pyogenes* – produces a heat-resistant and acid-resistant protein called the **M protein**, which mediates attachment to the host and helps microbe resist phagocytosis
  - *Neisseria gonorrhoeae* – uses fimbriae and an outer membrane protein called **Opa** to attach to and gain entry to host cells
  - *Mycobacterium* – mycolic acids also resist phagocytosis

- **Extracellular enzymes**
  - Coagulases – coagulate fibrinogen in blood, which may protect the bacterium from phagocytosis and isolate it from other host defenses
    - *Staphylococci*
  - Kinases – break down fibrin (blood clots)
    - *Streptococcus pyogenes* – fibrinolysin (streptokinase)
    - *Staphylococcus aureus* – staphylokinase
### How bacterial pathogens penetrate host defenses

- Extracellular enzymes
  - Hyaluronidase – hydrolyzes hyaluronic acid, a type of polysaccharide that holds together certain cells in the body, especially in connective tissue which can then allow the microbe to spread
    - *Clostridium*
  - Collagenase
    - *Clostridium*
  - IgA proteases
    - *Neisseria*

### How bacterial pathogens penetrate host defenses

- Antigenic variation
  - Some pathogens can vary their surface antigens, which may not interact with host-produced antibodies
    - *Neisseria gonorrhoeae* has several versions of the Opa-encoding gene
    - *Influenza*
    - *Trypanosoma brucei gambiense*

### How bacterial pathogens penetrate host defenses

- Penetration into the host cell cytoskeleton
  - When adhesins of microbe bind receptors on host cells, a series of events is triggered that allows entry of the microbe into the host cell
  - Microbe entry is facilitated by the host cell cytoskeleton
    - *Salmonella* and *E.coli* produce *invasins* that rearrange host cell actin filaments
    - *S. typhimurium* causes membrane ruffling
    - *Shigella* and *Listeria* use actin filaments and cadherins

### How bacterial pathogens damage host cells
Using the host’s nutrients

- Some pathogens secrete proteins called siderophores, which bind iron in the human body
- Siderophores can take iron from endogenous iron-transport proteins like lactoferrin, transferrin, ferritin, and hemoglobin
- Iron is transported into microbe to support vegetative growth and reproduction

Using the host’s nutrients

- Some pathogens have receptors that bind iron-transport proteins, resulting in both iron and the binding protein entering the microbial cell
- Some pathogens release toxins when iron levels are low, resulting in the death of host cells and subsequent release of iron

Direct damage

- As pathogens metabolize and reproduce, they ultimately cause the host cell to rupture
- When the host cell ruptures, pathogens are released to spread to other tissues
- Some pathogens promote their uptake and release from host cells using other strategies — E. coli, Shigella, Salmonella, N. gonorrhoeae

Toxin production

- Toxins — poisonous substances produced by certain microbes
- Toxigenicity — capacity of microbes to produce toxins
- Toxemia — presence of toxins in the blood
- Intoxications — caused by the presence of toxins, not microbial growth
Exotoxins

- Produced inside some bacteria and later secreted by the bacterium into the surrounding medium or released following lysis
- Exotoxins are proteins, usually enzymes which can be harmful even in low concentrations
- Can be produced by both gram (+) and (-) cells
- Genes for most exotoxins are carried on bacterial plasmids or phages

**Fig. 15.4**

**Exotoxins**

- Soluble in bodily fluids, so can easily diffuse into the blood and subsequently spread throughout the body
- Work by destroying particular parts of the host’s cells or by inhibiting certain metabolic functions
- Diseases caused by bacteria that produce exotoxins are often caused by minute amounts of exotoxin, not by bacteria themselves (exotoxins produce symptoms)

**Endotoxins**

- The body produces antibodies called antitoxins that provide immunity to exotoxins
- When exotoxins are inactivated (toxoids), they no longer cause disease but can still stimulate the body to produce antitoxins
- Toxoids are sometimes used in vaccines
  - Diphtheria and tetanus
How are exotoxins named?

- Type of host cell that is attacked
  - Neurotoxins, cardiotoxins, leukotoxins, enterotoxins, hepatotoxins, cytotoxins

- Named for the disease with which they are associated
  - Diphtheria toxin, tetanus toxin, botulinum toxin, Vibrio enterotoxin

Types of exotoxins (A-B exotoxins)

- Most exotoxins are of this type

- Consist of two parts (A and B) which are polypeptides

- A is the active (enzymatic) component

- B is the binding component

Exotoxins (Membrane-disrupting toxins)

- Cause lysis of host cells by disrupting PM by forming protein channels or disrupting the phospholipid bilayer
  - *S. aureus*, forms protein channels
  - *C. perfringens*, disrupts phospholipids

- Cause death of host cells (esp. phagocytes) and aid the escape of bacteria from phagosomes into the host cytoplasm
Exotoxins (Membrane-disrupting toxins)

- **Leukocidins** – membrane-disrupting toxins that kill phagocytic leukocytes
  - Form protein channels
  - Mostly produced by *Staphylococcus* and *Streptococcus*

- **Hemolysins** – destroy erythrocytes
  - *Staphylococcus* and *Streptococcus*
  - *Streptolysins* are hemolysins produced by . . .
    - Can cause lysis of other body cells as well

**Types of exotoxins (Superantigens)**

- Antigens (bacterial proteins) that provoke a very intense immune response
- Superantigens stimulate the proliferation of T cells, which then release enormous amounts of cytokines
- Cytokines regulate immune responses and mediate cell-to-cell communication
- Symptoms include fever, nausea, diarrhea, etc.
- *Staphylococcus* toxins that cause food poisoning and toxic shock syndrome

**Genotoxins**

- Produced by some gram (-) bacteria including *Haemophilus ducreyi* and *Helicobacter*
- Causes damage to cellular DNA, resulting in mutations and potentially cancer

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**Table 15.2 Diseases Caused by Exotoxins**

<table>
<thead>
<tr>
<th>Disease</th>
<th>Bacteria</th>
<th>Type of Exotoxin</th>
<th>Mechanism</th>
</tr>
</thead>
<tbody>
<tr>
<td>Botulism</td>
<td>Clostridium botulinum</td>
<td>A-B</td>
<td>Botulinum protein is a neurotoxin that paralyzes nerves by blocking acetylcholine release.</td>
</tr>
<tr>
<td>Tetanus</td>
<td>Clostridium tetani</td>
<td>A-B</td>
<td>Tetanospasmin is an exotoxin that induces muscle spasms by blocking the release of inhibitory neurotransmitters.</td>
</tr>
<tr>
<td>Diphtheria</td>
<td>Corynebacterium diphtheriae</td>
<td>A-B</td>
<td>Diphtheria toxin is a hemolysin that destroys red blood cells and causes tissue damage.</td>
</tr>
<tr>
<td>Staphylococcal sore throat</td>
<td><em>Staphylococcus aureus</em></td>
<td>A-B</td>
<td>Staphylococcal sore throat is caused by a coagulase-positive strain that produces a beta hemolysin.</td>
</tr>
<tr>
<td>Cholera</td>
<td>Vibrio cholera</td>
<td>A-B</td>
<td>Cholera toxin is an exotoxin that acts on the small intestine and causes severe diarrhea.</td>
</tr>
<tr>
<td>Tonsillar abscess</td>
<td><em>Streptococcus pyogenes</em></td>
<td>A-B</td>
<td>Strepococcal abscess is caused by a group A streptococcal strain that produces a strengthening protein.</td>
</tr>
<tr>
<td>Parotitis</td>
<td><em>Streptococcus pneumoniae</em></td>
<td>A-B</td>
<td>Parotitis is caused by a strain of <em>Streptococcus pneumoniae</em> that produces a hemolysin.</td>
</tr>
<tr>
<td>Gas gangrene</td>
<td><em>Clostridium perfringens</em> and other species of Clostridium</td>
<td>A-B</td>
<td>Gas gangrene is caused by clostridial strains that produce a variety of exotoxins that cause tissue damage.</td>
</tr>
<tr>
<td>Anthrax</td>
<td><em>Bacillus anthracis</em></td>
<td>A-B</td>
<td>Anthrax is caused by <em>Bacillus anthracis</em> that produces a lethal exotoxin.</td>
</tr>
<tr>
<td>Helicobacter</td>
<td><em>Helicobacter pylori</em></td>
<td>A-B</td>
<td><em>Helicobacter pylori</em> is a gram-negative bacteria that causes ulcers and peptic ulcers.</td>
</tr>
</tbody>
</table>

**Notes**

- *Helicobacter pylori* is a gram-negative bacteria that causes ulcers and peptic ulcers.
- *Clostridium perfringens* is a gram-negative bacteria that causes gas gangrene.
- *Streptococcus pneumoniae* is a gram-positive bacteria that causes pneumonia.
- *Staphylococcus aureus* is a gram-positive bacteria that causes skin infections and food poisoning.

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**References**

- *Bacterial Pathogenesis: A Molecular Approach* by Stephen B. Goodsell
- *Medical Microbiology* by John A. Perfect
- *Principles of Infectious Diseases* by Anthony S. Fauci

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*11/13/2016*
Endotoxins

- Produced only by gram (-) bacteria
- Lipid A of LPS is the endotoxin

Endotoxins

- Released during bacterial multiplication and when cells die and cell walls lyse
- Antibiotics can lyse cell walls, causing the release of the toxins and a worsening of symptoms
- Endotoxins stimulate macrophages to release cytokines in high concentrations, which are toxic at these concentrations
- Chills, fever, weakness, aches, shock, death, miscarriage

Endotoxins activate blood-clotting proteins, causing the formation of small blood clots which can obstruct capillaries potentially resulting in tissue death

Disseminated intravascular coagulation (DIC)

Endotoxins and the pyrogenic response

Bacterial cell death caused by lysis or antibiotics can also produce fever. Medications like aspirin and acetaminophen reduce fever by blocking prostaglandin formation.
Endotoxins

- **Shock** – life-threatening decrease in bp
- **Septic shock** – shock caused by bacteria
- **Gram (-) bacteria cause endotoxic shock**
  - Phagocytosis of these bacteria causes macrophages to release tumor necrosis factor (TNF)
    - Damages capillaries, which allows fluid loss
  - *Haemophilus influenza* type B is CSF causes the release of IL-1 and TNF, which affects the integrity of the blood-brain-barrier
    - *Lets phagocytes and more bacteria in to nervous system*

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Endotoxins

- **Endotoxins do not promote the formation of effective antitoxins against the CHO component of an endotoxin**

- **Antibodies that are produced do not counter the effect of the toxin, and sometimes make things worse**

  - *Salmonella typhi, Proteus, N. meningitidis*

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**Limulus amebocyte lysate (LAL) assay**

- This is a test to identify the presence of endotoxins in drugs, medical devices, and body fluids
- **Blood of *Limulus polyphemus* contains WBCs called amebocytes, which have large amounts of a protein that causes clotting.**
- **In the presence of endotoxins, amebocytes lyse, liberate their clotting protein and cause a gel-clot (precipitate)**

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### Table 15.3. Criteria and Endotoxins

<table>
<thead>
<tr>
<th>Property</th>
<th>Endotoxins</th>
<th>Endotoxins</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Bacterial Source</strong></td>
<td>Mostly from gram-negative bacteria</td>
<td>Gram-negative bacteria</td>
</tr>
<tr>
<td><strong>Relation to Microorganism</strong></td>
<td>Metabolite product of growing cell</td>
<td>Metabolite product of growing cell</td>
</tr>
<tr>
<td><strong>Chemistry</strong></td>
<td>Protein, usually with two parts (A-B)</td>
<td>Lipid protein (10-18% of total membrane lipid)</td>
</tr>
<tr>
<td><strong>Pharmacology/Effect on Cells</strong></td>
<td>Splits the mucopolysaccharide core of lipopolysaccharide and affects cell viability, membrane integrity, and protein synthesis</td>
<td>Splits the mucopolysaccharide core of lipopolysaccharide and affects cell viability, membrane integrity, and protein synthesis</td>
</tr>
<tr>
<td><strong>Heat Stability</strong></td>
<td>Stable to 60°C for 1 hour</td>
<td>Stable to 100°C for 1 hour</td>
</tr>
<tr>
<td><strong>Tissue/Ability to Cause Damage</strong></td>
<td>High</td>
<td>Low</td>
</tr>
<tr>
<td><strong>Peroxide Producing</strong></td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td><strong>Immunochemistry (Relation to Antibodies)</strong></td>
<td>Can be immobilized and preserved on glass slides and reacted with antibodies</td>
<td>Not easily immobilized by antibodies, therefore, effective need to be made to membrane-bound form</td>
</tr>
<tr>
<td><strong>Lethal Dose</strong></td>
<td>Small</td>
<td>Very large</td>
</tr>
<tr>
<td><strong>Representative Diseases</strong></td>
<td>Gram-negative, staphylococcal, salmonella, typhoid fever, septicemia, meningitis</td>
<td>Taphoid fever, staphylococcal, salmonella, meningitis</td>
</tr>
</tbody>
</table>
Plasmids

- Plasmids, specifically R factors, carry resistance genes
- In addition, plasmids may also carry genes for virulence factors
  - Tetanus neurotoxin
  - Staphylococcal enterotoxin D
  - Dextranucrase, involved in tooth decay

Lysogeny

- One outcome of lysogeny is that the host bacterial cell and its progeny may exhibit new properties encoded by the bacteriophage DNA (lysogenic conversion)
- Bacteriophage genes code for diphtheria toxin, staphylococcal enterotoxin A and pyrogenic toxin, botulinum neurotoxin, Shiga toxin

Pathogenic properties of viruses

Viral mechanisms for evading host defenses

- Think back to Chapter 13 . . .
Cytopathic effects of viruses

- These are the visible effects of viral infection
- **Cytopathic effects** (CPEs) that result in cell death are called **cytocidal effects**
- Viral infection usually kills host cell
  - Accumulation of virus particles
  - Effects of viral proteins on host cell PM
  - Inhibition of host cell nucleic acid or protein synthesis
- CPEs that only cause cell damage but not cell death are called **noncytocidal effects**

Cytopathic effects

- Some viruses irreversibly stop mitosis
- Viruses stimulate the release of host cell lysosomal enzymes
- Viruses can induce the formation of inclusion bodies
  - Can be used as a diagnostic tool
- **Syncytium formation**
  - Fusion of adjacent infected cells produces a giant, multi-nucleated cell

Cytopathic effects cont.

- Viral infections can induce antigenic changes on the surface of infected cells
- Some viruses can induce chromosomal changes in the host cell
- Certain viruses can transform cells, resulting in unregulated cell growth
- Interferons are released by virus-infected cells to help protect neighboring cells
  - Promote virus-infected cell apoptosis and inhibit viral replication in neighboring cells
Pathogenic properties of fungi

- **Trichothecenes**
  - Toxin that inhibits protein synthesis in eukaryotic cells
  - Headache, chills, nausea, vomiting, visual
  - *Fusarium, Stachybotrys* on grains and wallpaper
- **Some fungi secrete proteases**
  - *Candida albicans, Trichophyton*
- **Cryptococcus neoformans** produces a capsule
- **Toxin production**
  - *Claviceps purpurea, Aspergillus flavus*
  - Mycotoxin production (phalloidin, amanitin)

### Table 15.4 Cytopathic Effects of Selected Viruses

<table>
<thead>
<tr>
<th>Virus (Genus)</th>
<th>Cytopathic Effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>Poliovirus (Enterovirus)</td>
<td>Cytocidal (cell death)</td>
</tr>
<tr>
<td>Genital warts virus (Papillomavirus)</td>
<td>Acidophilic inclusion bodies in nucleus</td>
</tr>
<tr>
<td>Adenovirus (Hepadnavirus)</td>
<td>Basophilic inclusion bodies in nucleus</td>
</tr>
<tr>
<td>Lyssavirus</td>
<td>Acidophilic inclusion bodies in cytoplasm</td>
</tr>
<tr>
<td>CMV (Cytomegalovirus)</td>
<td>Acidophilic inclusion bodies in nucleus and cytoplasm</td>
</tr>
<tr>
<td>Measles virus (Morbillivirus)</td>
<td>Cell fusion</td>
</tr>
<tr>
<td>Polyomavirus</td>
<td>Transformation</td>
</tr>
<tr>
<td>HIV (Lentivirus)</td>
<td>Destruction of T cells</td>
</tr>
</tbody>
</table>

Pathogenic properties of protozoa

- **Antigenic variation**
  - *Giardia*
  - *Trypanosoma*
  - When first enters the bloodstream the flagellate displays a specific antigen which triggers the production of antibodies
  - Within a few weeks the microbe stops displaying the original antigen and displays a different one
  - This is repeated each time the body’s immune system is successful in suppressing the microbe

Trypanosomes evade the immune system
Portals of exit

- Portals of exit relate to the infected part of the body, with microbes tending to use the same portal for entry and exit.
- Portals of exit include the respiratory, GI, and genitourinary tracts, skin, wounds, biting insects.