Drugs, Microbes, Host – The Elements of Chemotherapy

Chapter 12
<table>
<thead>
<tr>
<th>Characteristic</th>
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<tbody>
<tr>
<td>Selectively toxic to the microbe but nontoxic to host cells</td>
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<tr>
<td>Microbicidal rather than microbistatic</td>
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<td>Relatively soluble and functions even when highly diluted in body fluids</td>
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<td>Remains potent long enough to act and is not broken down or excreted prematurely</td>
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<td>Not subject to the development of antimicrobial resistance</td>
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<tr>
<td>Complements or assists the activities of the host’s defenses</td>
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<td>Remains active in tissues and body fluids</td>
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<td>Readily delivered to the site of infection</td>
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<td>Not excessive in cost</td>
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<td>Does not disrupt the host’s health by causing allergies or predisposing the host to other infections</td>
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<td>Term</td>
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<tr>
<td>Chemotherapeutic drug</td>
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<tr>
<td>Prophylaxis*</td>
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<td>Antimicrobial chemotherapy*</td>
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<tr>
<td>Antimicrobics</td>
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<tr>
<td>Antibiotics*</td>
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<tr>
<td>Semisynthetic drugs</td>
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<tr>
<td>Synthetic drugs</td>
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<tr>
<td>Narrow spectrum (limited spectrum)</td>
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<td>Broad spectrum (extended spectrum)</td>
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</table>
Origins of antimicrobial drugs

- Antibiotics are common metabolic products of aerobic spore-forming bacteria & fungi.
  - bacteria in genera *Streptomyces* & *Bacillus*
  - molds in genera *Penicillium* & *Cephalosporium*
- By inhibiting the other microbes in the same habitat, antibiotic producers have less competition for nutrients & space.
Streptomyces
Selectively toxic

• Drugs should kill or inhibit microbial cells without simultaneously damaging host tissues.

• As the characteristics of the infectious agent become more similar to the vertebrate host cell, complete selective toxicity becomes more difficult to achieve & more side effects are seen.
Targets of antimicrobial drugs

1. Inhibition of cell wall synthesis
2. Inhibition of nucleic acid synthesis, structure or function
3. Inhibition of protein synthesis
4. Disruption of cell membrane structure or function
5. Cell Membrane disruption
Drugs that act upon ribosome and block protein synthesis

Site of action: 50S subunit
- Chloramphenicol
- Erythromycin
- Clindamycin
- Oxalolidinones

Site of action: 30S subunit
- Aminoglycosides
- Tetracyclines
- Streptomycin
- Amikacin

Cell wall
- Block synthesis and repair
  - Penicillins
  - Cephalosporins
  - Vancomycin
  - Bacitracin
  - Monobactams
  - Fosfomycin
  - Cycloserine

DNA
- Inhibit replication and transcription
- Inhibit gyrase (unwinding enzymes)
- Quinolones (ciprofloxacin)
- Inhibit RNA polymerase
- Rifampin

Cell membrane
- Polymyxins

Cytoplasm
- Inhibit folic acid metabolism
  - Sulfonamides (sulfa drugs)
  - Trimethoprim

PABA → Folic acid
1. Drugs that affect the bacterial cell wall

• Most bacterial cell walls contain a rigid girdle of peptidoglycan.

• **Penicillin and cephalosporin** block synthesis of peptidoglycan, causing the cell wall to lyse.

• Penicillins do not penetrate the outer membrane and are less effective against gram-negative bacteria.

• Broad spectrum penicillins and cephalosporins can cross the cell walls of gram-negative bacteria.
1. Drugs that affect the bacterial cell wall
1. Drugs that affect the bacterial cell wall
2. Drugs that inhibit nucleic acid synthesis

- may block synthesis of nucleotides, inhibit replication, or stop transcription
- Sulfonamides and trimethoprim block enzymes required for tetrahydrofolate synthesis needed for DNA & RNA synthesis.

- **competitive inhibition** – drug competes with normal substrate for enzyme’s active site

- **synergistic effect** – an additive effect, achieved by multiple drugs working together, requiring a lower dose of each
2. Drugs that inhibit nucleic acid synthesis

(a) Normal metabolic pathway

- Sulfonamides inhibit enzyme
- Trimethoprim inhibits enzyme
- THFA is coenzyme required to synthesize

PABA → Dihydrofolate → Tetrahydrofolate (THFA)

Purines
Pyrimidines
Amino acids

(b) Normal folic acid synthesis

Initial folic acid molecule

(c) Inhibition of folic acid synthesis by sulfa drug

Active site
Pteridine synthetase
Folic acid synthesis cannot be completed
3. Drugs that block protein synthesis

- Ribosomes of eucaryotes differ in size and structure from procaryotes, so antimicrobics usually have a selective action against procaryotes. But they can also damage the eucaryotic mitochondria.

- Aminoglycosides (streptomycin, gentamicin) insert on sites on the 30S subunit and cause misreading of mRNA.

- Tetracyclines block attachment of tRNA on the A acceptor site and stop further synthesis.
3. Drugs that block protein synthesis

*FIGURE 12.5*
Sites of inhibition on the procaryotic ribosome and major antibiotics that act on these sites. All have the general effect of blocking protein synthesis. Blockage actions are indicated by X.
4. Drugs that disrupt cell membrane function

• A cell with a damaged membrane dies from disruption in metabolism or lysis.

• These drugs have specificity for a particular microbial group, based on differences in types of lipids in their cell membranes.

• **Polymyxins** interact with phospholipids and cause leakage, particularly in gram-negative bacteria

• **Amphotericin B** and nystatin form complexes with sterols on fungal membranes which causes leakage.
4. Drugs that disrupt cell membrane function
Survey of major antimicrobial drug groups

• Antibacterial drugs
  – Antibiotics
  – Synthetic drugs

• Antifungal drugs

• Antiparasitic drugs

• Antiviral drugs

About 260 different antimicrobial drugs are classified in 20 drug families.
Antibacterial antibiotics

- Penicillins
- Cephalosporins
- Other beta-lactam antibiotics
- Aminoglycosides
- Tetracycline antibiotics
- Chloramphenicol
- Other *Streptomyces* antibiotics
- The *Bacillus* antibiotics
- New classes
Penicillins

• Large diverse group of compounds
• Could be synthesized in the laboratory
• more economical to obtain natural penicillin through microbial fermentation and modify it to semi-synthetic forms
• *Penicillium chrysogenum* – major source
• All consist of 3 parts
  – thiazolidine ring
  – beta-lactam ring
  – variable side chain dictates microbial activity
Penicillins
Penicillins Video

- Penicillins G and V most important natural forms
- Penicillin is the drug of choice for gram-positive cocci (streptococci) and some gram-negative bacteria (meningococci and syphilis spirochete)
- Semisynthetic penicillins – ampicillin, carbenicillin & amoxicillin have broader spectra – gram negative enterics rods
- Penicillinase-resistant – methicillin, nafcillin, cloxacillin
- Primary problems – allergies and resistant strains of bacteria
YOU KNOW, CLAUDIA, WHEN THEY MADE YOU, THEY BROKE THE MOLD.
Cephalosporins

- Account for majority of all antibiotics administered
- Isolated from *Cephalosporium acremonium* mold
- Beta-lactam ring that can be altered
- Relatively broad-spectrum, resistant to most penicillinases, & cause fewer allergic reactions
- Some are given orally, many must be administered parenterally
Cephalosporins

<table>
<thead>
<tr>
<th>R Group 1</th>
<th>Basic Nucleus</th>
<th>R Group 2</th>
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<tbody>
<tr>
<td><img src="image1" alt="Chemical Structure" /></td>
<td><img src="image2" alt="Chemical Structure" /></td>
<td>Cephalothin (first generation)</td>
</tr>
<tr>
<td><img src="image3" alt="Chemical Structure" /></td>
<td><img src="image4" alt="Chemical Structure" /></td>
<td>Cefotiam (second generation)</td>
</tr>
<tr>
<td><img src="image5" alt="Chemical Structure" /></td>
<td><img src="image6" alt="Chemical Structure" /></td>
<td>Moxalactam (third generation)</td>
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</table>
Cephalosporins

• 3 generations exist

• **First generation** – cephalothin, cefazolin – most effective against gram-positive cocci

• **Second generation** – cefaclor, cefonacid – more effective against gram-negative bacteria

• **Third generation** – cephalexin, cefotaxime – broad-spectrum activity against enteric bacteria with beta-lactamases

• Ceftriaxone – new semisynthetic broad-spectrum drug for treating wide variety of infections
Aminoglycosides

• products of various species of soil actinomycetes in genera *Streptomyces* & *Micromonospora*

• Broad-spectrum, inhibit protein synthesis, especially useful against aerobic gram-negative rods & certain gram-positive bacteria
  
  – *Streptomycin* – bubonic plague, tularemia, TB
  
  – *Gentamicin* – less toxic, used against gram-negative rods

  – Newer – *Tobramycin* & *amikacin* gram-negative bacteria
Tetracycline antibiotics

- Broad-spectrum, block protein synthesis
- Doxycycline & minocycline – oral drugs taken for STDs, Rocky Mountain spotted fever, Lyme disease, typhus, acne & protozoa
Chloramphenicol

• Isolated from *Streptomyces venezuelae*
• **Potent broad-spectrum** drug with unique nitrobenzene structure
• Blocks peptide bond formation
• No longer derived from natural source
• **Very toxic**, restricted uses, can cause irreversible damage to bone marrow
• Typhoid fever, brain abscesses, rickettsial & chlamydial infections
Other *Streptomyces* antibiotics

- **Erythromycin** – macrolide, large lactone ring with sugars
- **Broad-spectrum**, fairly low toxicity
- Attaches to ribosome
- Taken orally for Mycoplasma pneumonia, legionellosis, Chlamydia, pertussis, diptheria and as a prophylactic prior to intestinal surgery
- For **penicillin-resistant** – gonococci, syphilis, acne
- Newer semi-synthetic **macrolides** – clarithromycin, azithromycin
Other *Streptomyces* antibiotics

- **Clindamycin** – *broad-spectrum*, serious abdominal anaerobic infections
- **Vancomycin** – *narrow-spectrum*, effective against penicillin & methicillin resistant staphylococcal infections; very toxic, hard to administer
- **Rifampin** – *limited spectrum*, cannot pass through many cell membranes, used to treat gram-positive bacteria, TB, leprosy
Miscellaneous antibacterial drugs

• Isoniazid – used with rifampicin to treat TB
• Oxazolidinones - new class of antibacterial drugs inhibit initiation of protein synthesis (50S ribosome)
  – Linezolid – MRSA, VRE
• Quinolones (Fluoroquinolones) – broad-spectrum, potent
  – norfloxacin, ciprofloxacin – UTI, STD, GI, osteomyletitis, respiratory & soft tissue infections
  – sparofloxacin, levofloxacin – pneumonia, bronchitis, sinusitis
Antifungal drugs

• Macrolide polyene
  – **Amphotericin B** – mimic lipids, most versatile & effective, topical & systemic treatments
  – **Nystatin** – topical treatment

• **Griseofulvin** – stubborn cases of dermatophyte infections, nephrotoxic

• Synthetic azoles – **broad-spectrum**; **ketoconazole, clotrimazole, miconazole**

• **Flucytosine** – analog of cytosine; cutaneous mycoses or in combination with amphotericin B for systemic mycoses
Antiparasitic drugs

• Antimalarial drugs – quinine, chloroquinine, primaquine, mefloquine
• Antiprotozoan drugs - Metronidazole (Flagyl), quinicrine, sulfonamides, tetracyclines
• Antihelminthic drugs – immobilize, disintegrate, or inhibit metabolism
  – mebendazole, thiabendazole- broad-spectrum – inhibit function of microtubules, interferes with glucose utilization & disables them
  – pyrantel, piperazine- paralyze muscles
  – niclosamide – destroys scolex
Antiviral drugs

• Block penetration into host cell
• Block transcription or translation
  – Nucleotide analogs
    • **Acyclovir** – herpesviruses
    • **Ribavirin** - a guanine analog – RSV, hemorrhagic fevers
    • **AZT** – thymine analog - HIV
• Prevent maturation of viral particles
  – Protease inhibitors – HIV
• **Interferon** - HCV
Until Bob entered her life, Lisa never even knew the meaning of the phrase “acute multiple-drug resistant infection with transgenic Staphylococcus aureus.”

Funny now to think back... Somehow all these little things change when you date a careless microbiologist.
Mechanisms drug resistance

• Drug inactivation – penicillinases
• Decreased permeability to drug or increased elimination of drug from cell
• Change in metabolic patterns
• Change in drug receptors
Mechanisms drug resistance

(a) Drug inactivation

Drug inactivation is the process by which drugs are rendered inactive. For example, penicillin can be inactivated by penicillinase, an enzyme that cleaves a portion of the molecule and renders it inactive.

(b) Decreased permeability

Decreased permeability is the process by which drugs cannot enter into the cell. The receptor that transports the drug is altered, so that the drug cannot enter the cell.

(c) Activation of drug pumps

Activation of drug pumps is the process by which specialized membrane proteins are activated and continually pump the drug out of the cell.

(d) Use of alternate metabolic pathway

The drug has blocked the usual metabolic pathway, so the microbe circumvents it by using an alternate, unblocked pathway that achieves the required outcome.
Selection for drug resistance

(a) Population of microbial cells

(b) Sensitive cells eliminated by drug; resistant mutants survive

(c) Most cells are now resistant
Side effects of drugs

1. Toxicity to organs
2. Allergic responses
3. Suppression and alteration of microflora
"Would you like that to be a steak with a broad-spectrum antibiotic, or one with a variety of therapeutic proteins?"