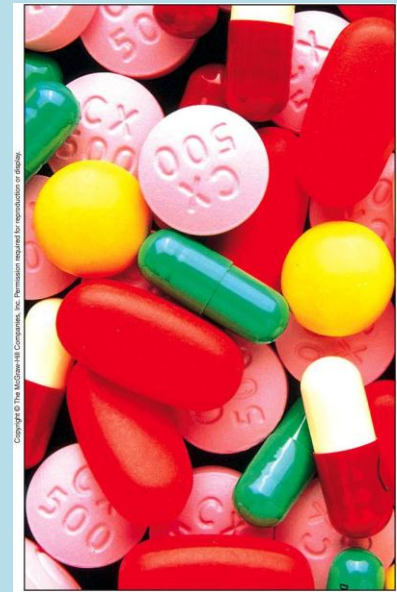
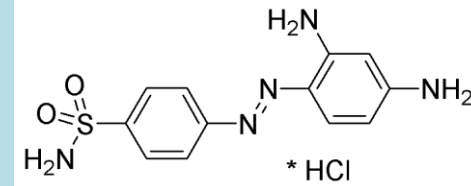
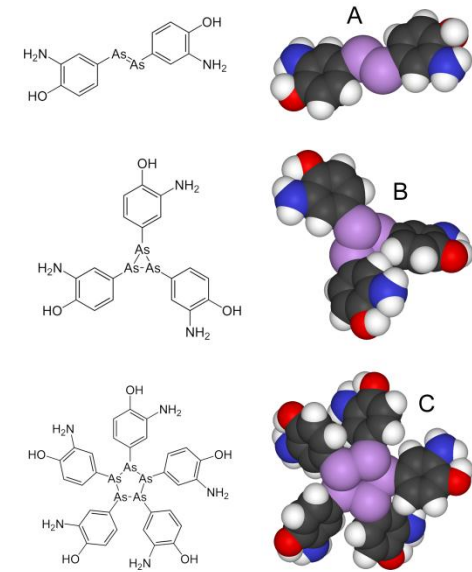
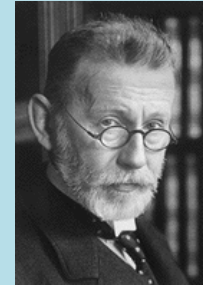


Preview: antimicrobial

- What's antibiotics? (history/development)
- How do the antibiotics work?
- What's the difference among antibacterial, antiviral and antifungal compounds?

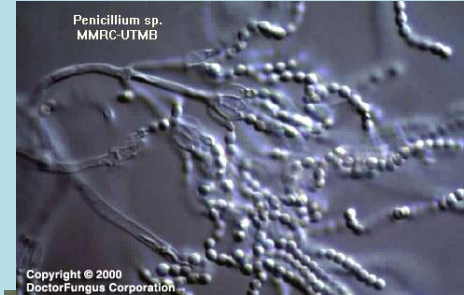
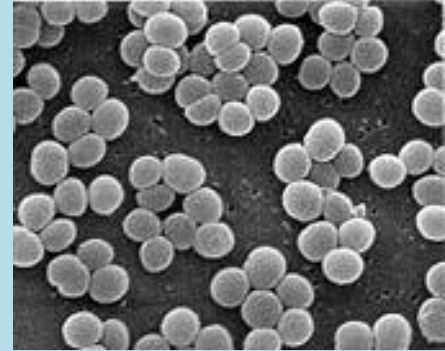
History and Development of Antimicrobial Drugs

- Discovery of antimicrobial drugs
 - Salvarsan first documented example of chemical successfully used as antimicrobial **1909**⁽⁶⁰⁶⁾
 - Discovered by **Paul Erlich** in for treatment of syphilis movie [The Magic Bullet](#),
 - Prontosil dye effective against streptococcal infections **1931**
 - No effect on *streptococcus* growing *in vitro*
- Enzymes in blood split prontosil into small sulfonamide molecules
 - Sulfonamide was the first sulfa drug
 - Acts as a competitive inhibitor to para-aminobenzoic acid (an essential nutrient for some [bacteria](#))



History and Development of Antimicrobial Drugs

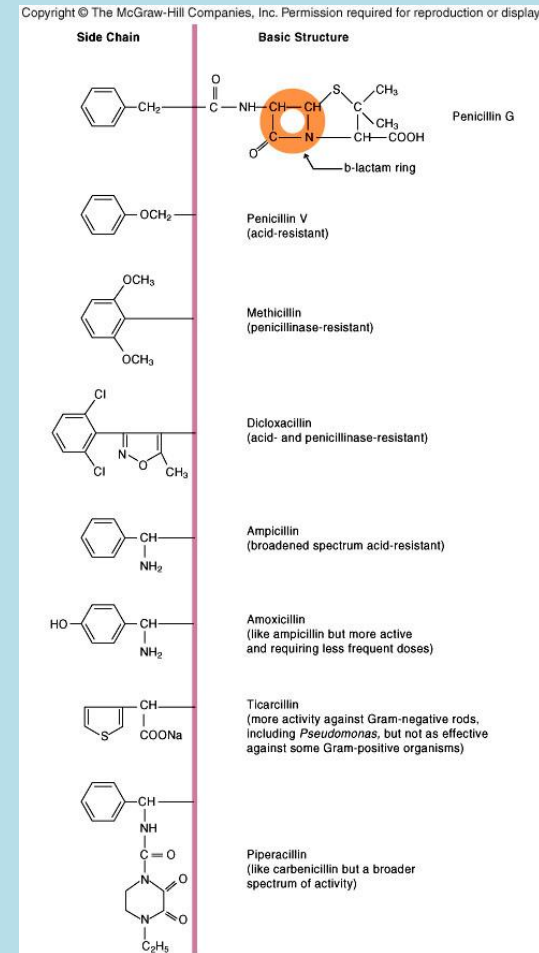
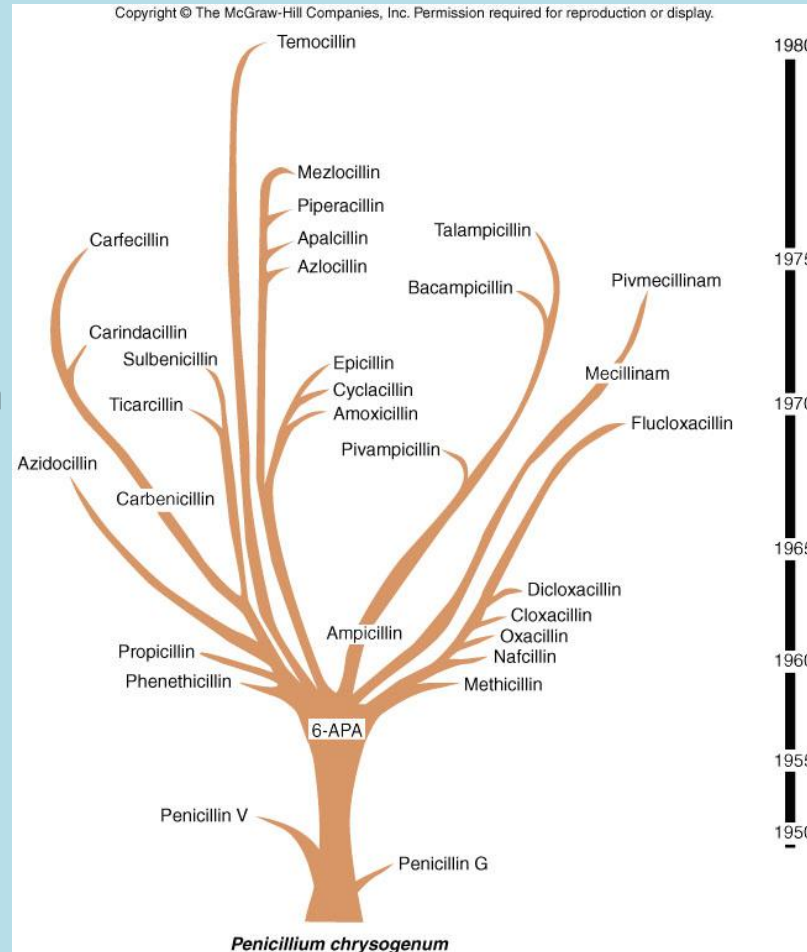
- Discovery of antibiotics
 - Alexander Fleming
 - Discovered penicillin while working with *Staphylococcus* 1928
 - Noticed there were no *Staph* colonies growing near a mold contaminant
 - The colonies appeared to be melting
 - Identified mold as *Penicillium* and was producing a bactericidal substance that was effective against a wide range of microbes
- Fleming unable to purify compound
 - Ernst Chain and Howard Florey successfully purified penicillin 1939



History and Development of Antimicrobial Drugs

Development of new generation of drugs

- In 1960s scientists alteration of drug structure gave them new properties
 - Penicillin G altered to created ampicillin
 - Broadened spectrum of antimicrobial killing



Features of Antimicrobial Drugs

- Selective toxicity
 - Antibiotics cause greater harm to microorganisms than to human host
 - Generally by interfering with biological structures or biochemical processes common to bacteria but not to humans
 - Toxicity of drug is expressed as therapeutic index
 - Lowest dose toxic to patient divided by dose typically used for treatment
 - High therapeutic index = less toxic to patient

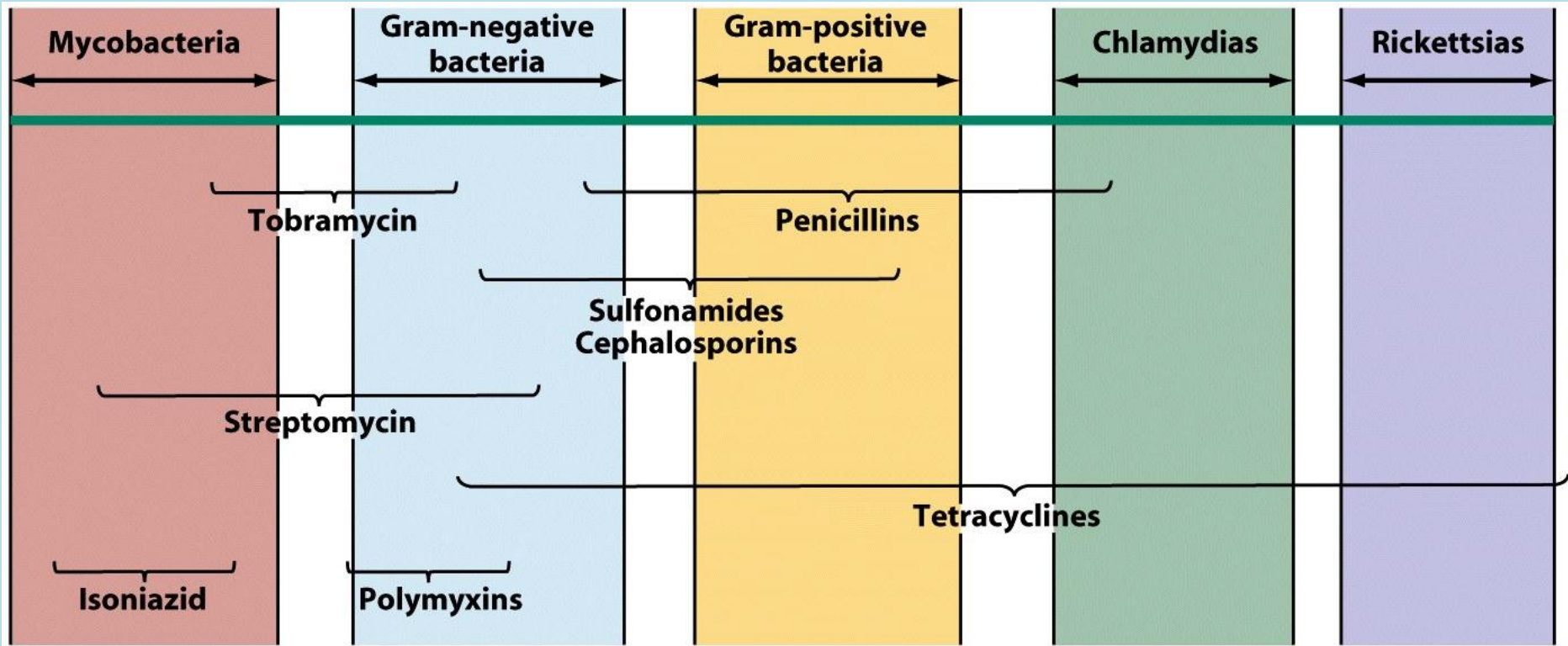
Features of Antimicrobial Drugs

- Antimicrobial action
 - Drugs may kill or inhibit bacterial growth
 - Inhibit = **bacteriostatic**
 - Kill = **bacteriocidal**
 - Bacteriostatic drugs rely on host immunity to eliminate pathogen
 - Bacteriocidal drugs are useful in situations when host defenses cannot be relied upon to control pathogen

Features of Antimicrobial Drugs

- **Spectrum** of activity
 - Antimicrobials vary with respect to range of organisms controlled
 - Narrow spectrum
 - Work on narrow range of organisms
 - » Gram positive only OR Gram negative only
 - Broad spectrum
 - Work on broad range of organisms
 - » Gram positive AND Gram negative
 - Disadvantage of broad spectrum is disruption of normal flora

Spectrum of Activity



Features of Antimicrobial Drugs



- Tissue **distribution**, **metabolism** and **excretion**
 - Drugs differ in how they are distributed, metabolized and excreted
 - Important factor for consideration when prescribing
 - Rate of elimination of drug from body expressed in **half-life**
 - Time it takes for the body to eliminate one half the original dose in serum
 - Half-life dictates frequency of dosage
 - Patients with liver or kidney damage tend to excrete drugs more slowly

Features of Antimicrobial Drugs

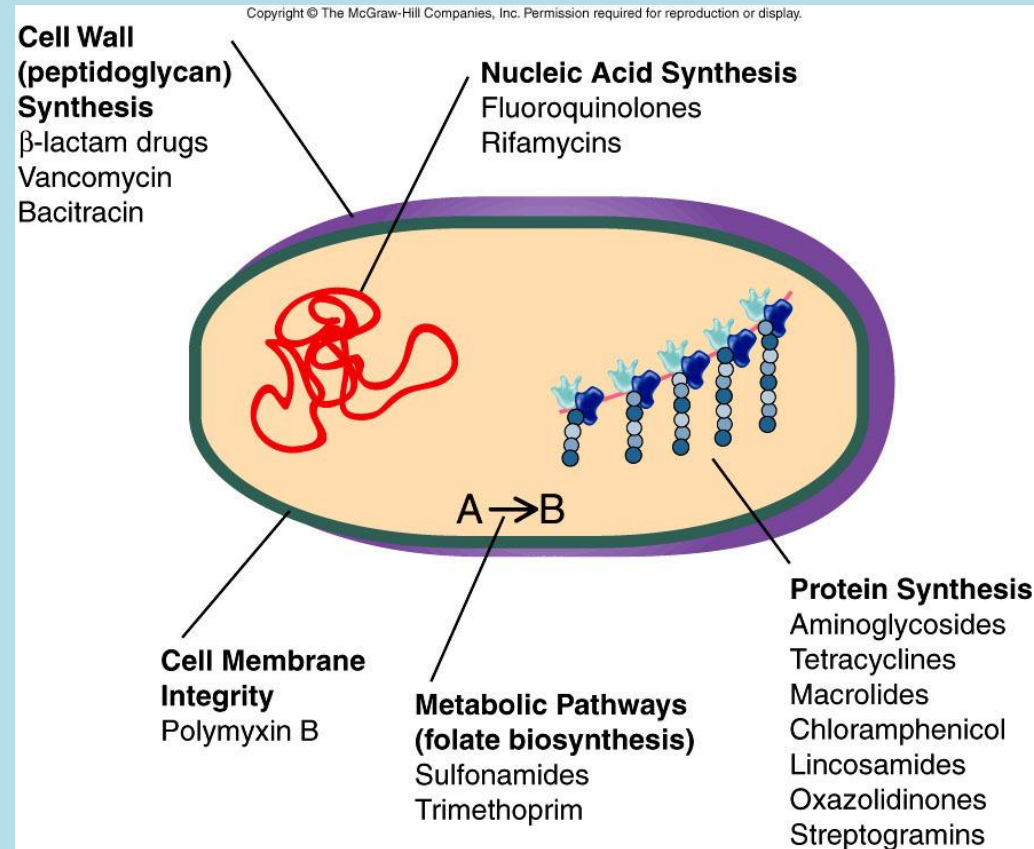
- Effects of combinations of antimicrobial drugs
 - Combination some times used to treat infections
 - When action of one drug enhances another, effect is synergistic $1+1>2$
 - When action of one drug interferes with another, effect is antagonistic $1+1<2$
 - When effect of combination is neither synergistic or antagonistic, effect said to be additive $1+1=2$

Features of Antimicrobial Drugs

- Adverse effects
 - Allergic reactions
 - Allergies to penicillin
 - Allergies often life threatening
 - Toxic effects
 - Aplastic anemia
 - Body cannot make RBC or WBC
 - Suppression of normal flora
 - Antibiotic associated colitis
 - Toxic organisms given opportunity to establish themselves
 - Antimicrobial resistance
 - Microorganisms have innate or adaptive resistance to antibiotics

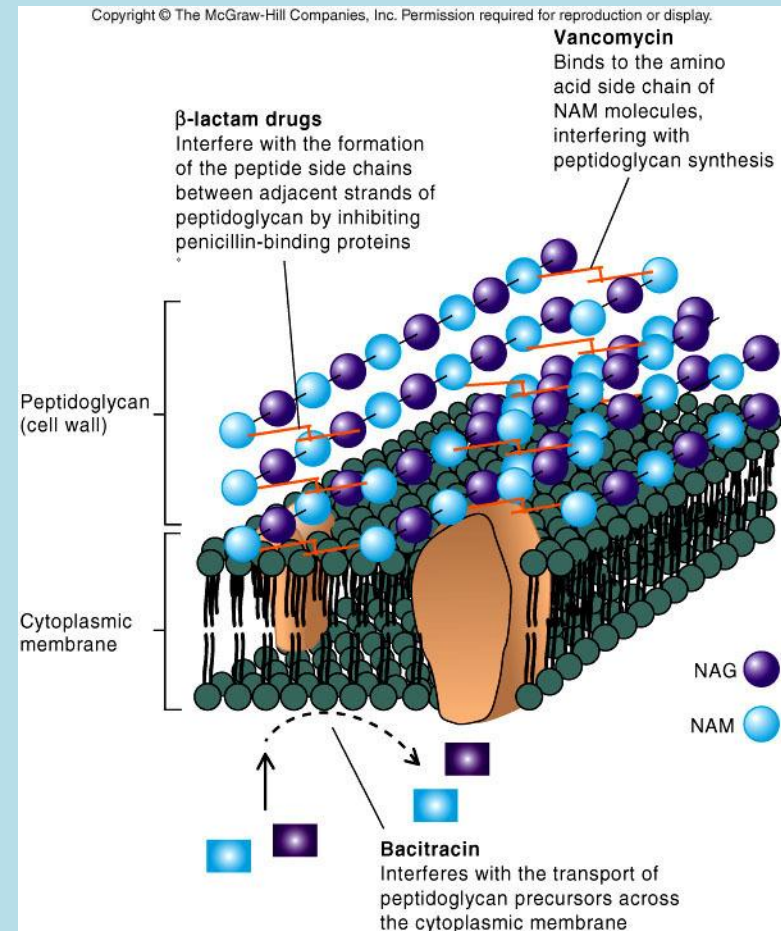
Mechanisms of Action of Antibacterial Drugs

- Mechanism of action include:
 - Inhibition of **cell wall** synthesis
 - Inhibition of **protein synthesis**
 - Inhibition of **nucleic acid synthesis**
 - Inhibition of **metabolic pathways**
 - Interference with cell **membrane** integrity
 - Interference with essential processes of *M. tuberculosis*



Mechanisms of Action of Antibacterial Drugs

- Inhibition of Cell wall synthesis
 - Bacteria cell wall unique in construction
 - Contains **peptidoglycan**
 - Antimicrobials that interfere with the synthesis of cell wall do not interfere with eukaryotic cell
 - Due to the lack of cell wall in animal cells and differences in cell wall in plant cells
 - These drugs have very high therapeutic index
 - Low toxicity with high effectiveness
 - Antimicrobials of this class include
 - **β lactam drugs**
 - **Vancomycin**
 - **Bacitracin**



Mechanisms of Action of Antibacterial Drugs

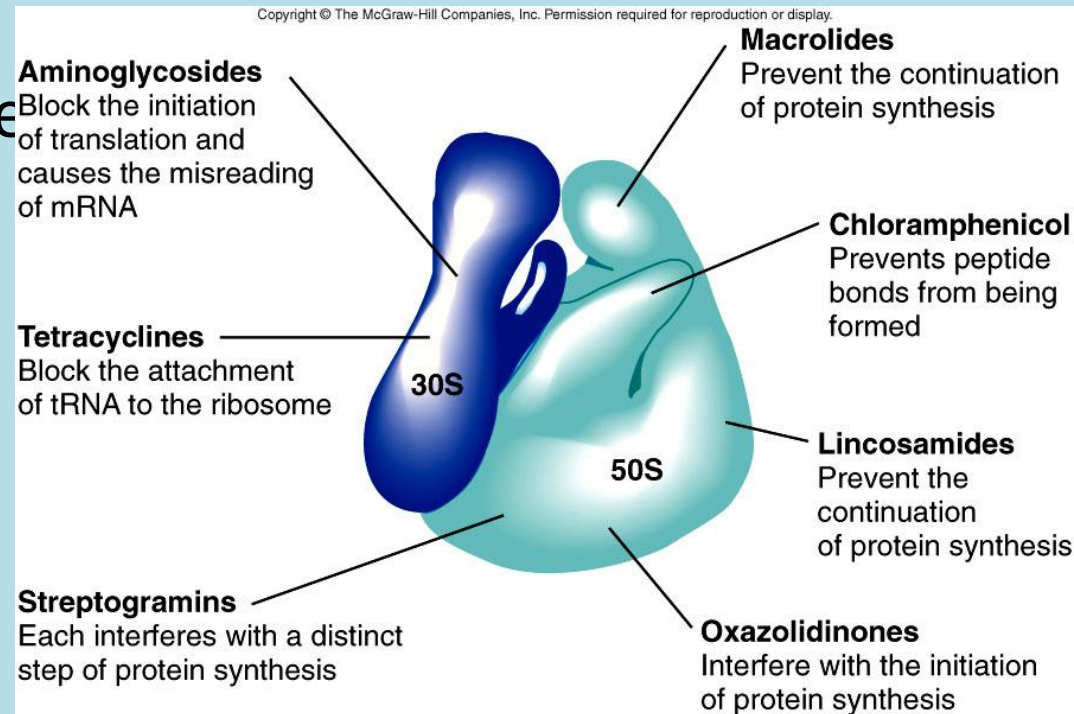
Inhibition of protein synthesis

– Structure of prokaryotic ribosome acts as target for many antimicrobials of this class

- Differences in prokaryotic and eukaryotic ribosomes responsible for selective toxicity

– Drugs of this class include

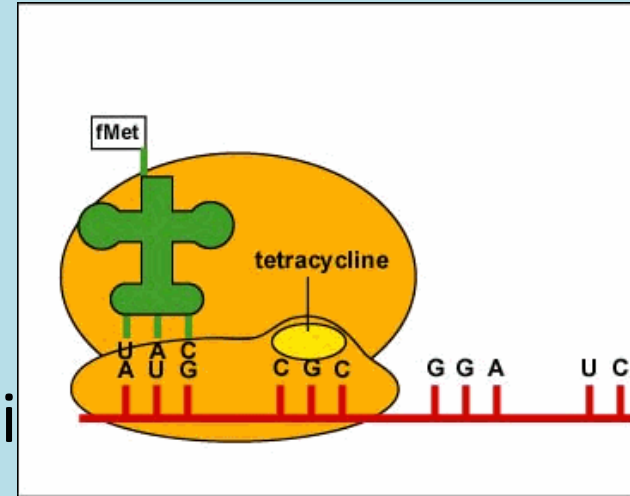
- Aminoglycosides
- Tetracyclins
- Macrolids
- Chloramphenicol
- Lincosamides
- Oxazolidinones
- Streptogramins



Mechanisms of Action of Antibacterial Drugs

- Aminoglycosides
 - Irreversibly binds to 30S ribosomal subunit
 - Causes distortion and malfunction of ribosome
 - Blocks initiation translation
 - Causes misreading of mRNA
 - Not effective against anaerobes, enterococci and streptococci
 - Often used in synergistic combination with β -lactam drugs
 - Allows aminoglycosides to enter cells that are often resistant
 - Examples of aminoglycosides include
 - Gentamicin, streptomycin and tobramycin
 - Side effects with extended use include
 - Nephrotoxicity
 - Ototoxicity

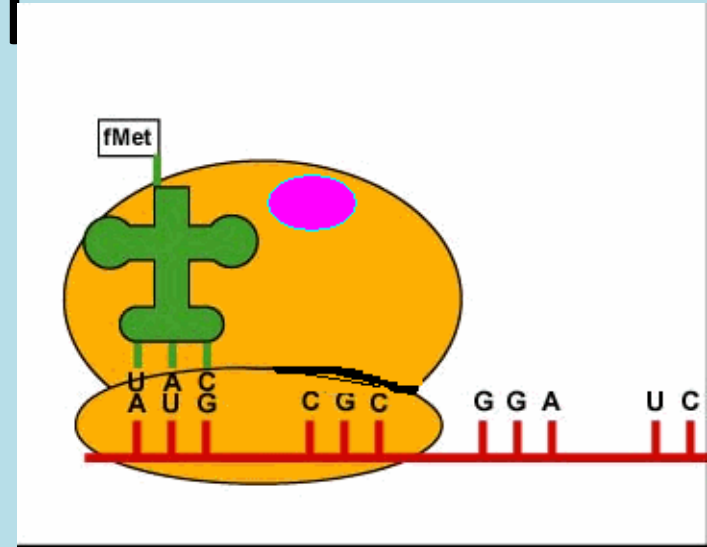
Mechanisms of Action of Antibacterial Drugs



- Tetracyclins

- Reversibly bind 30S ribosomal subunit
 - Blocks attachment of tRNA to ribosome
 - Prevents continuation of protein synthesis
- Effective against certain Gram (+) and Gram (-)
- Newer tetracyclines such as doxycycline have longer half-life
 - Allows for less frequent dosing
- Resistance due to decreased accumulation by bacterial cells
- Can cause discoloration of teeth if taken as young child

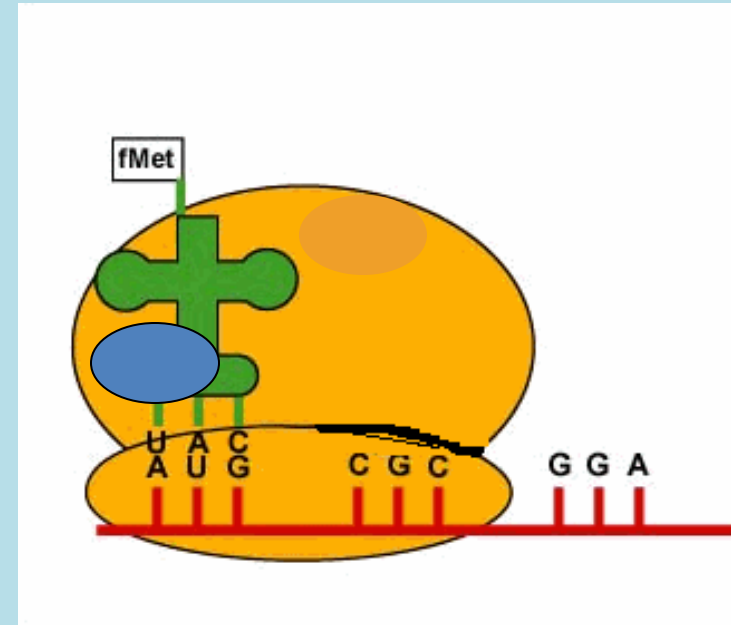
Mechanisms of Action of Antibacterial Drugs



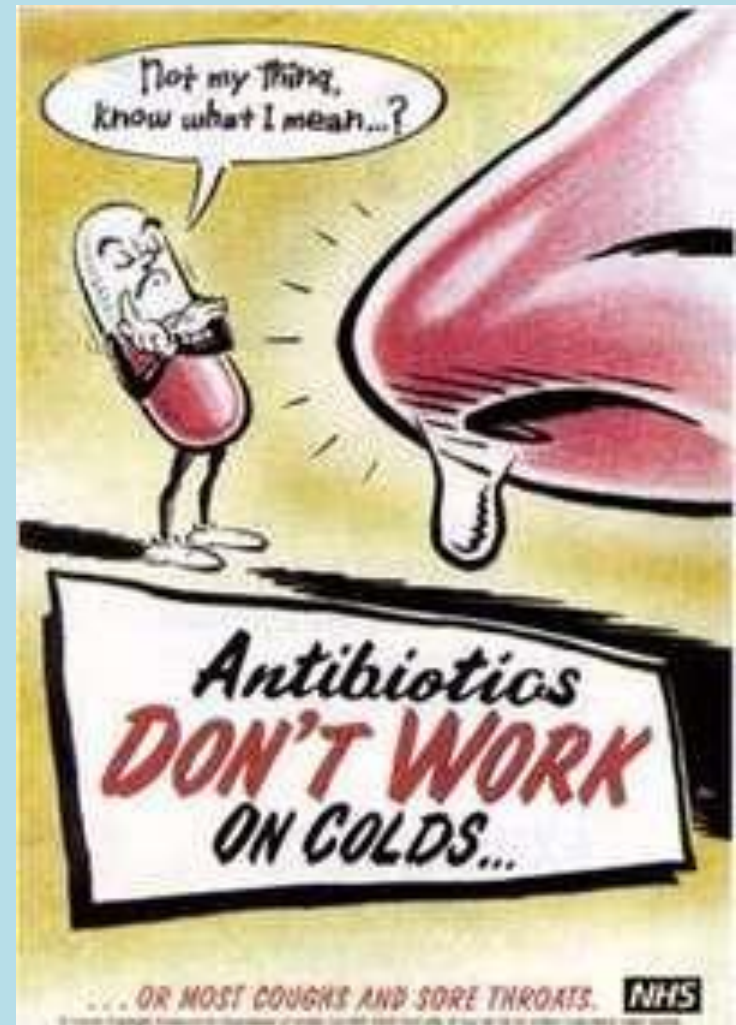
- Chloramphenicol
 - Binds to 50S ribosomal subunit
 - Prevents peptide bonds from forming and blocking proteins synthesis
 - Effective against a wide variety of organisms
 - Generally used as drug of last resort for life-threatening infections
 - Rare but lethal side effect is aplastic anemia

Mechanisms of Action of Antibacterial Drugs

- Oxazolidinones
 - New class of antimicrobials
 - Binds 50S ribosomal subunit
 - Interferes with initiation of proteins synthesis
 - Effective against variety of Gram (+) bacteria
 - Especially those resistant to β -lactams and vancomycin



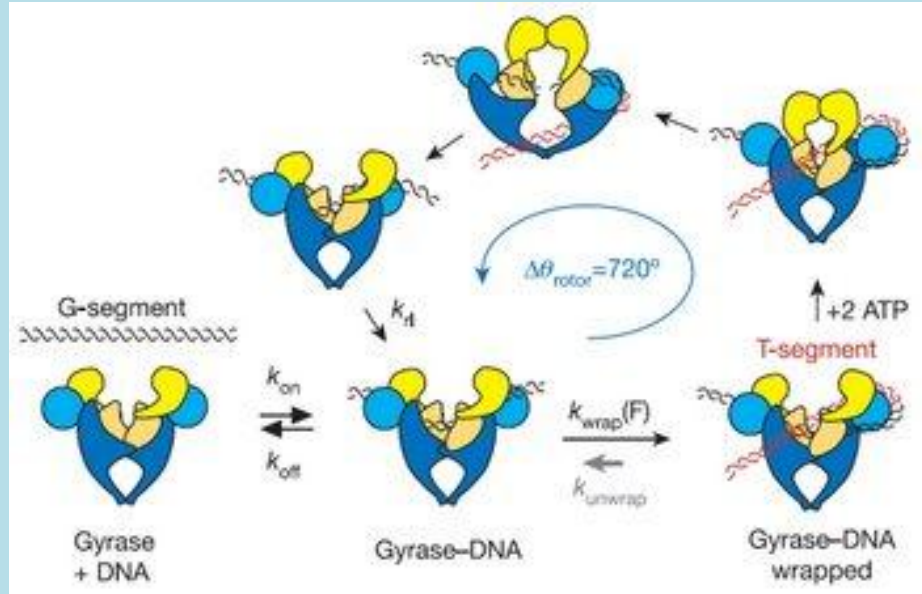
Quiz: Describe the cellular mechanisms that antibiotics kill bacteria, using a practical example.



Mechanisms of Action of Antibacterial Drugs

Fluoroquinolones

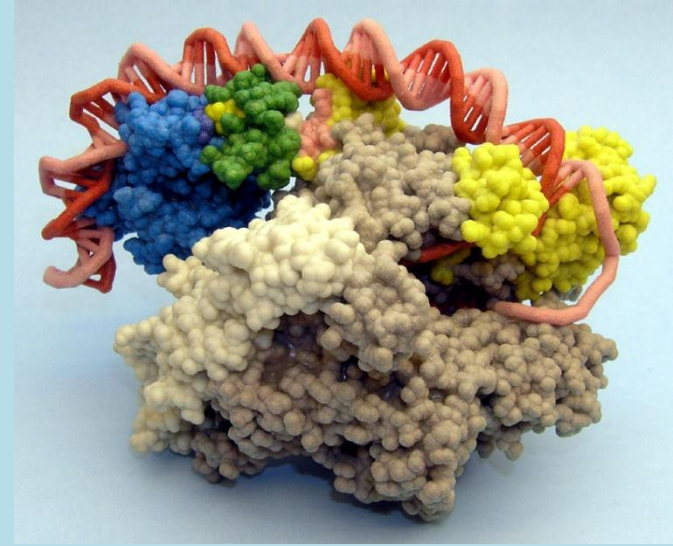
- Inhibit action of topoisomerase DNA gyrase
 - Topoisomerase maintains supercoiling of DNA
- Effective against Gram (+) and Gram (-)
- Examples include
 - Ciprofloxacin and ofloxacin
- Resistance due to alteration of DNA gyrase



Mechanisms of Action of Antibacterial Drugs

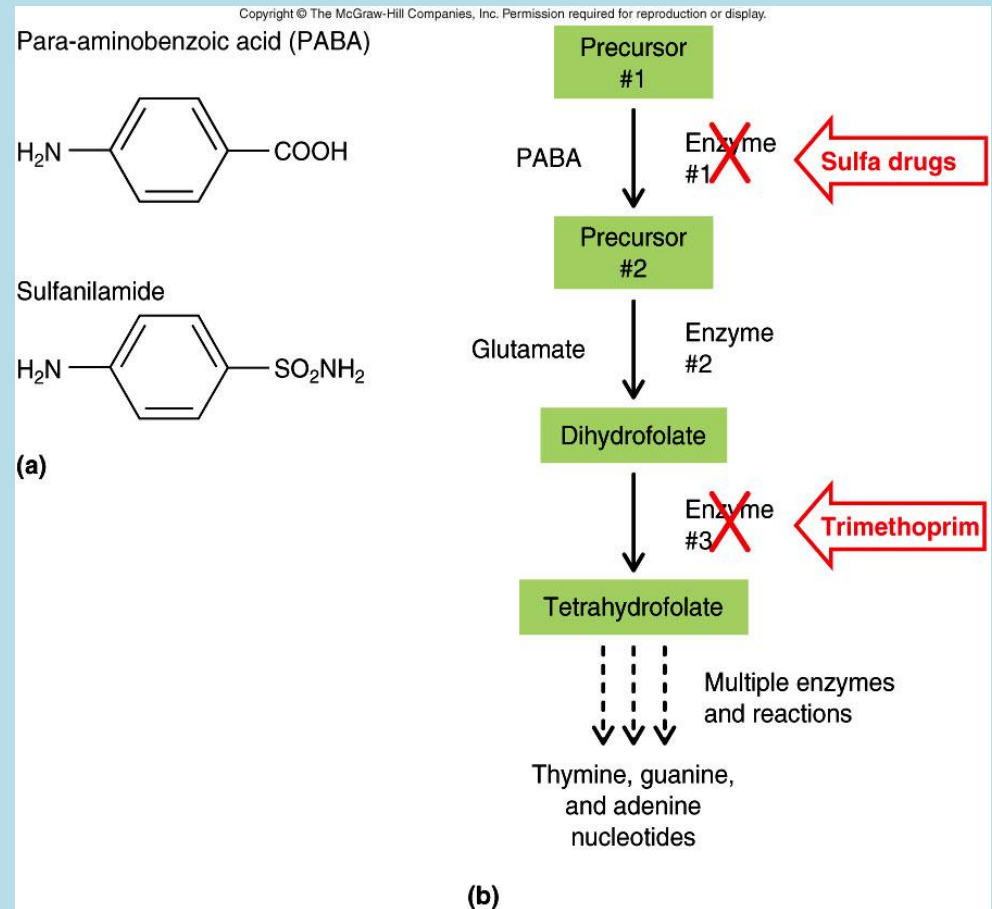
- Rifamycins

- Block prokaryotic RNA polymerase
 - Block initiation of transcription
- Rifampin most widely used rifamycins
- Effective against many Gram (+) and some Gram (-) as well as members of genus *Mycobacterium*
- Primarily used to treat tuberculosis and Hansen's disease as well as preventing meningitis after exposure to *N. meningitidis*
- Resistance due to mutation coding RNA polymerase
 - Resistance develops rapidly



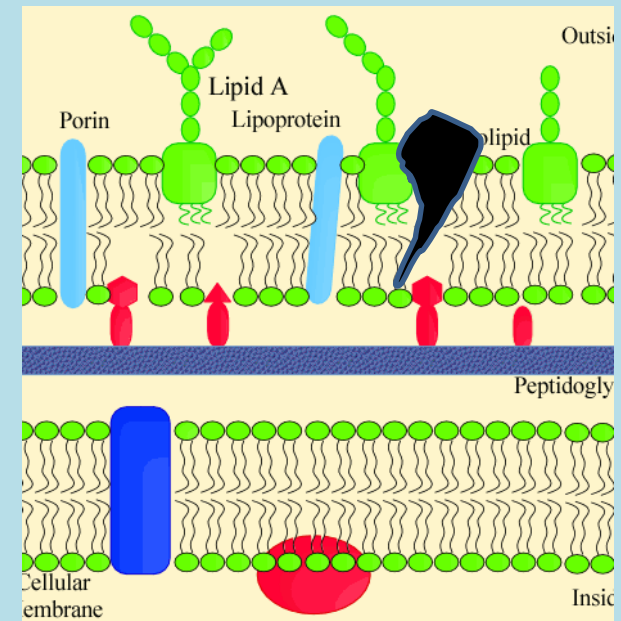
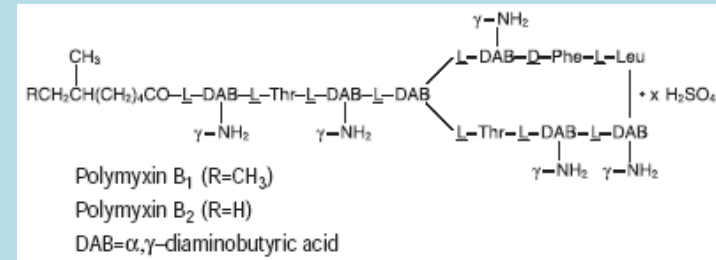
Mechanisms of Action of Antibacterial Drugs

- Inhibition of metabolic pathways
 - Relatively few
 - Most useful are folate inhibitors
 - Mode of actions to inhibit the production of folic acid
 - Antimicrobials in this class include
 - Sulfonamides
 - Trimethoprim



Mechanisms of Action of Antibacterial Drugs

- Interference with cell membrane integrity
 - Few damage cell membrane
 - Polymyxin B most common
 - Common ingredient in first-aid skin ointments
 - Binds membrane of Gram (-) cells
 - Alters permeability
 - Leads to leakage of cell and cell death
 - Also bind eukaryotic cells but to lesser extent
 - Limits use to topical application

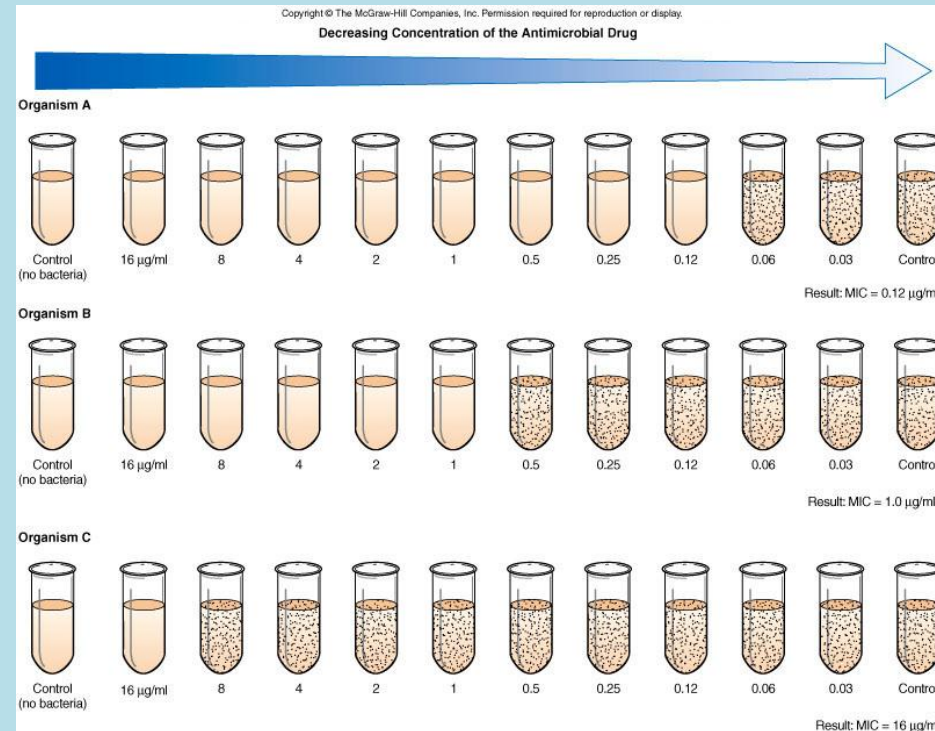


Mechanisms of Action of Antibacterial Drugs

- Inference with processes essential to *Mycobacterium tuberculosis*
 - Limited range of antimicrobials used in treatment of infections caused by *M. tuberculosis*
 - Due to numerous factors including chronic nature of disease, slow growth and waxy lipid in cell wall
 - Waxy cell wall due to mycolic acid is impervious to many drugs
 - Five medications termed first-line drugs preferentially because of effectiveness with low toxicity
 - First-line drugs include rifampin, streptomycin and
 - Isoniazid → inhibits synthesis of mycolic acid
 - Ethambutol → inhibits enzymes for cell wall synthesis
 - Pyrazinamide → mechanism of action unknown

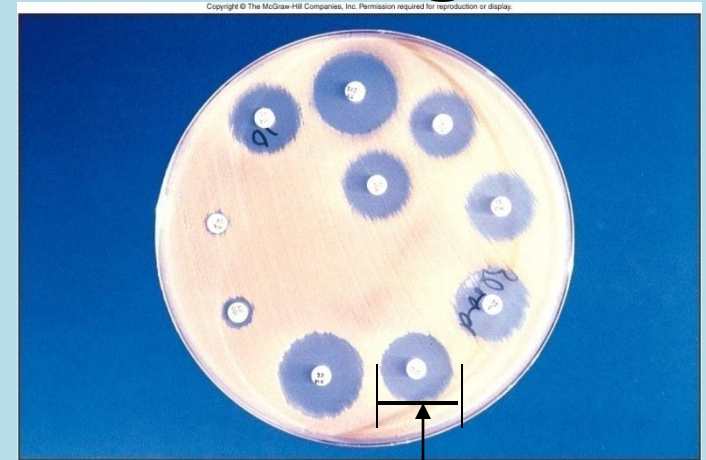
Determining Susceptibility of Bacterial to Antimicrobial Drug

- *MIC* is determined by producing serial dilutions with decreasing concentrations of test drug
- Known concentrations of organism is added to each test tube
- Tubes are incubated and examined for growth
 - Growth determined by turbidity of growth medium
- Lowest concentration to prevent growth is MIC



Determining Susceptibility of Bacterial to Antimicrobial Drug

- Conventional disc diffusion method
 - Kirby-Bauer test disc diffusion routinely used to qualitatively determine susceptibility
 - Standard concentration of strain uniformly spread of standard media
 - Discs impregnated with specific concentration of antibiotic placed on



zone of inhibition

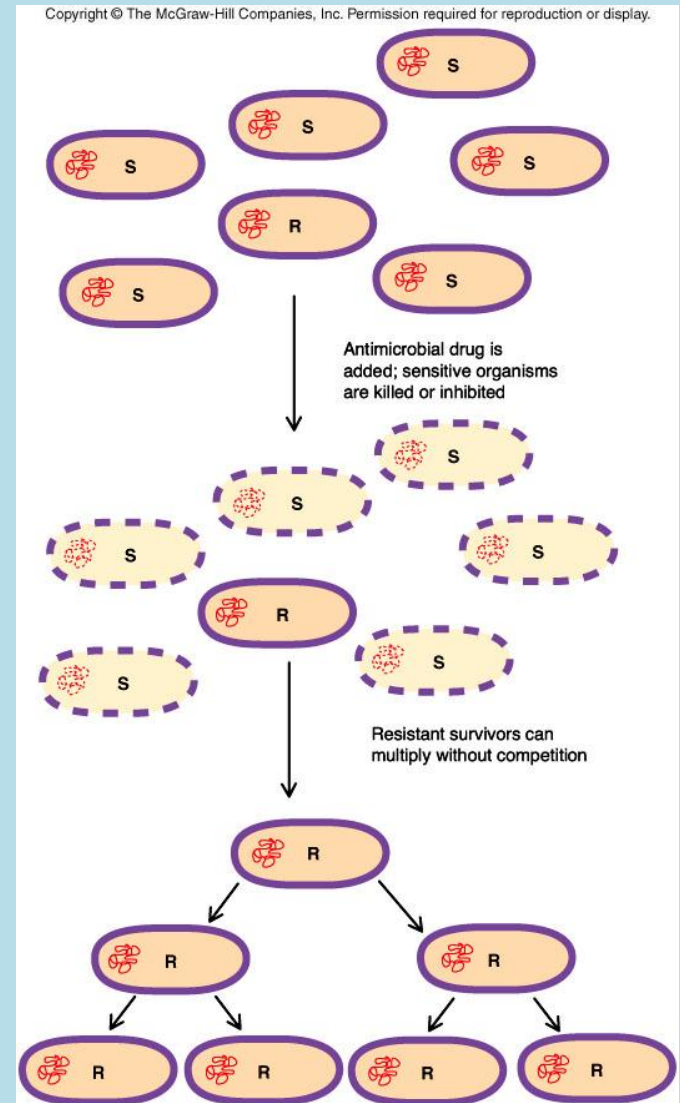
- Clear zone of inhibition around disc reflects susceptibility
 - Based on size of zone organism can be described as susceptible or resistant

Mechanisms of Drug Resistance

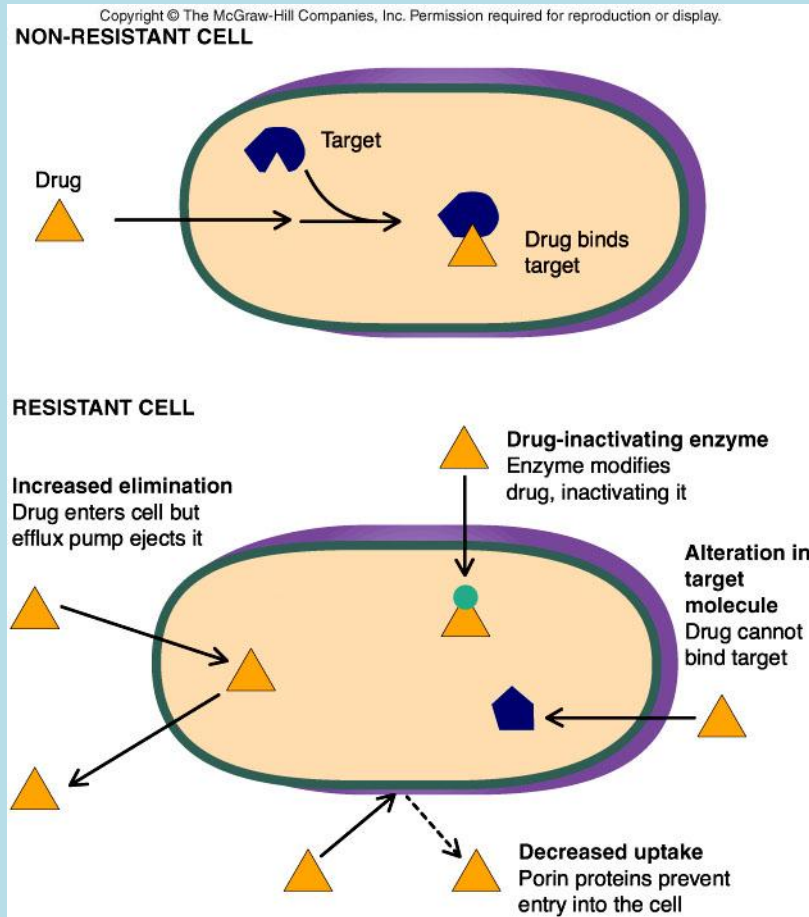
- Mutations in Target molecules
- Alterations in membrane permeability
- Enzyme development
- Enzyme Activity Changes
- Alterations in Anabolic Pathways

Resistance to Antimicrobial Drugs

- Mechanisms of **resistance**
 - Drug inactivating **enzymes**
 - Some organisms produce enzymes that chemically modify drug
 - Penicillinase breaks β -lactam ring of penicillin antibiotics
 - Alteration of **target molecule**
 - Minor structural changes in antibiotic target can prevent binding
 - Changes in ribosomal RNA prevent macrolids from binding to ribosomal subunits



Determining Susceptibility of Bacterial to Antimicrobial Drug



Mechanisms of resistance

- Decreased uptake of the drug
 - Alterations in porin proteins decrease permeability of cells
 - Prevents certain drugs from entering
- Increased elimination of the drug
 - Some organisms produce efflux pumps
 - Increases overall capacity of organism to eliminate drug
 - » Enables organism to resist higher concentrations of drug
 - » Tetracycline resistance

Determining Susceptibility of Bacterial to Antimicrobial Drug

- Spontaneous mutation
- Acquisition of new genes through gene transfer
 - Most common mechanism of transfer is through conjugation
 - Transfer of R plasmid
 - Plasmid often carries several different resistance genes
 - Each gene mediating resistance to a specific antibiotic
 - » Organism acquires resistance to several different drugs simultaneously

Determining Susceptibility of Bacterial to Antimicrobial Drug

- *Staphylococcus aureus*
 - Common cause of **nosocomial infections**
 - Becoming increasingly resistant
 - In past 50 years most strains acquired resistance to penicillin
 - Due to acquisition of penicillinase genes
 - Until recently most infections could be treated with methicillin (penicillinase resistant penicillin)
 - Many strains have become resistant
 - » MRSA → methicillin resistant *Staphylococcus aureus*
 - MRSA many of these strains still susceptible to vancomycin
 - Some hospitals identified VISA
 - » VISA → vancomycin intermediate *Staphylococcus aureus*

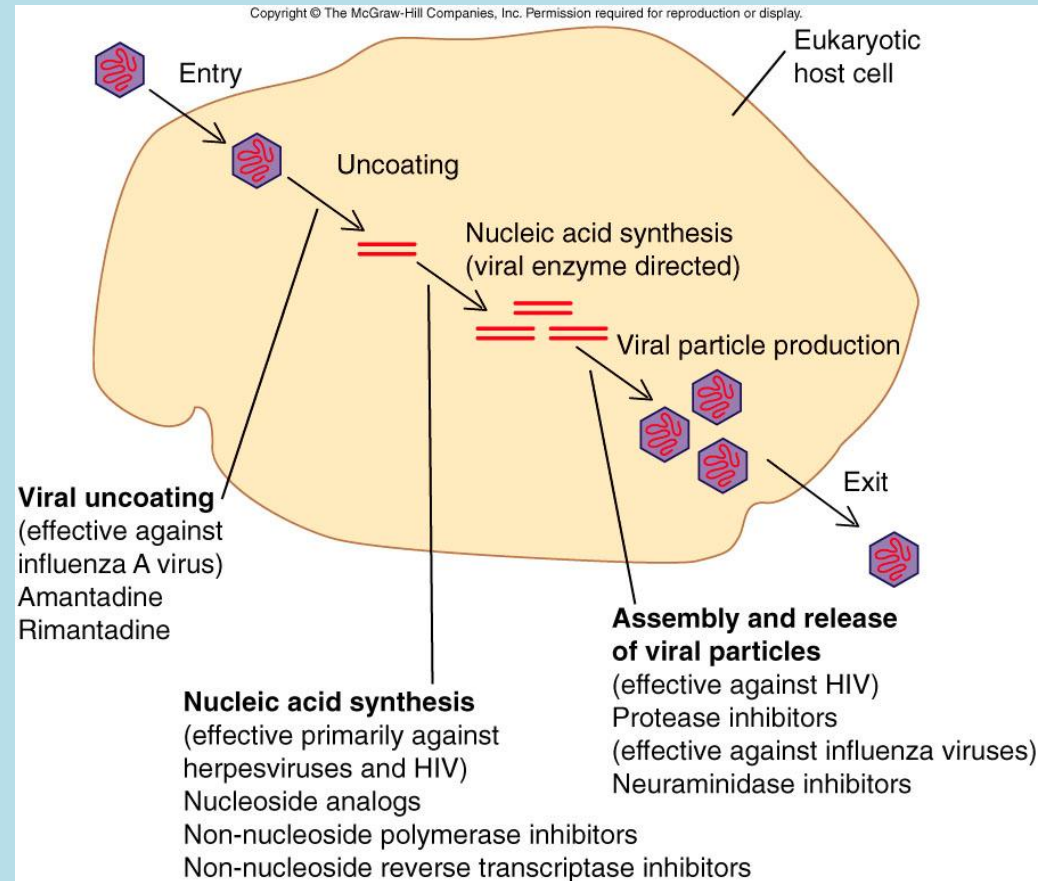
Mechanisms of Action of Antiviral Drugs

- Available antiviral drugs effective specific type of virus

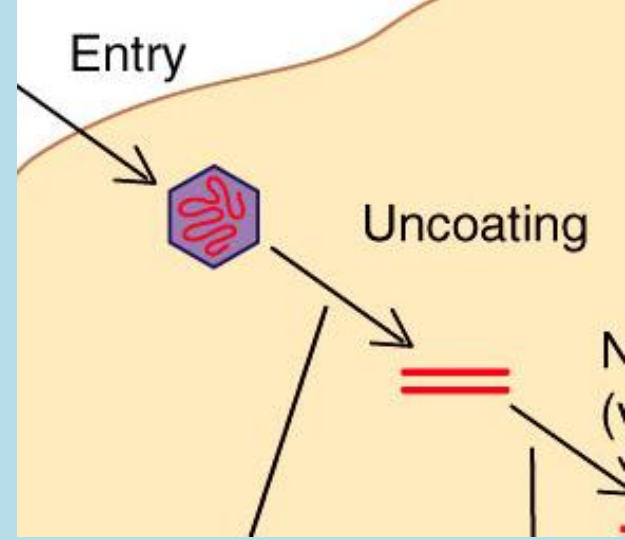
- None eliminate latent virus

- Targets include

- Viral uncoating
 - Nucleoside analogs
 - Non-nucleoside polymerase inhibitors
 - Non-nucleoside reverse transcriptase inhibitors
 - Protease inhibitors



Mechanisms of Action of Antiviral Drugs

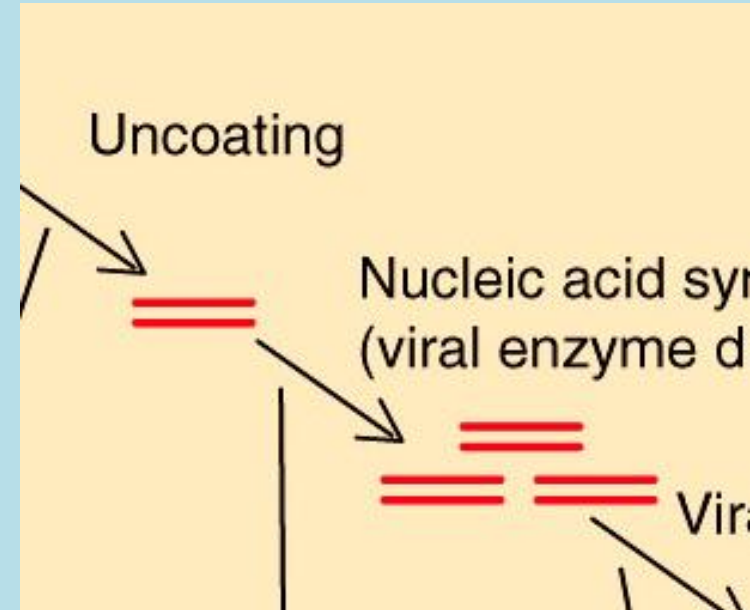


- Viral uncoating
 - Drugs include amantadine and rimantadine
 - Similar in chemical structure and mechanism of action
 - Mode of action is blocking uncoating of influenza virus after it enters cell
 - Prevents severity and duration of disease
 - Resistance develops frequently and may limit effectiveness of drug

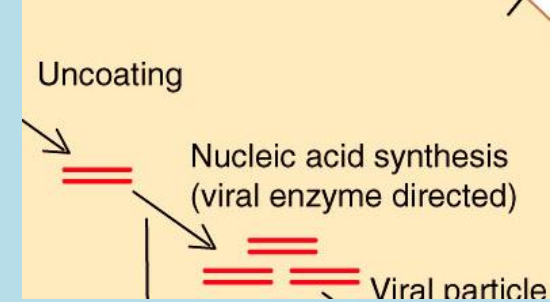
Mechanisms of Action of Antiviral Drugs

- Nucleoside analogs

- Similar in structure to nucleoside
- Analogs phosphorylated and become nucleotide analogs
- Incorporation of analog results in termination of growing nucleotide chain
- Examples of nucleoside analogs
 - Zidovudine (AZT)
 - Didanosine (ddI)
 - Lamivudine (3TC)



Mechanisms of Action of Antiviral Drugs



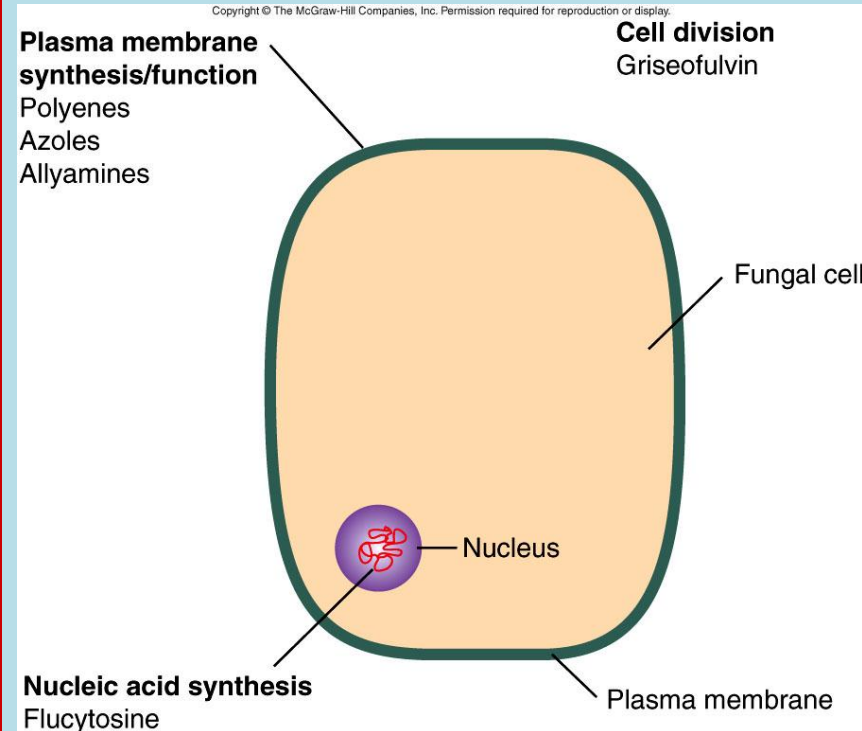
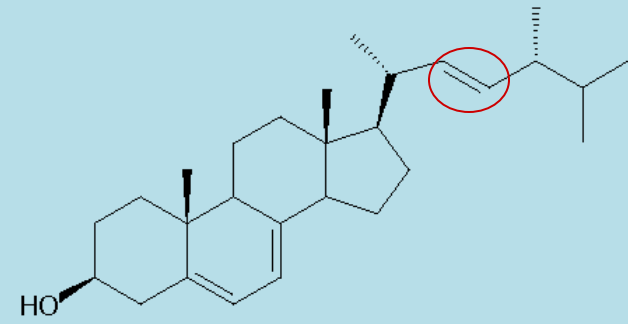
- Non-nucleoside polymerase inhibitor
 - Inhibit activation of viral polymerases by binding to site other than nucleotide binding site
 - Example = foscarnet and acyclovir
 - Used to treat CMV and HSV
- Non-nucleoside reverse transcriptase inhibitor
 - Inhibits activity of reverse transcriptase by binding to site other than nucleotide binding site
 - Example = nevirapine, delavirdine, efavirenz
 - Used to in combination to treat HIV

Mechanisms of Action of Antiviral Drugs

- Protease inhibitor
 - Inhibit HIV encoded enzyme protease
 - Enzyme essential for production of viral particles
 - Examples = indinavir and ritonavir
 - Used in treatment of HIV
- Neuraminidase inhibitor
 - Inhibit neuraminidase enzyme of influenza
 - Enzyme essential for release of virus
 - Examples = zanamivir and oseltamivir
 - Zanamivir administered via inhalation
 - Oseltamivir administered orally

Antifungal Drugs

- Target for most antifungal medications is **ergosterol**
 - In plasma membrane
- Drugs targeting ergosterol include
 - Polyenes
 - Azoles
 - Allylamines
- Other targets
 - Cell wall synthesis
 - Cell division
 - Nucleic acid synthesis



Mechanisms of Action of Antifungal Drugs

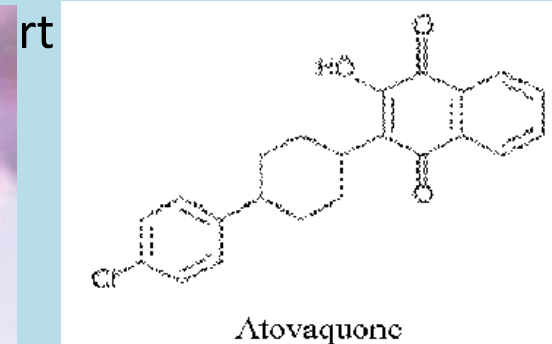
- Polyenes
 - Produced by *Streptomyces*
 - Disrupts fungal membrane
 - Causes leakage of cytoplasmic contents leading to cell death
 - Polyenes very toxic to humans
 - Limited to use in life-threatening disease
 - Amphotericin B effective against systemic infection
 - » Causes severe side effects
 - Nystatin used topically due to toxicity
- Azoles
 - Chemically synthesized drugs
 - Includes two classes
 - Imidazoles and triazoles
 - Both inhibit synthesis of ergosterol
 - Triazoles including fluconazole and itraconazole increased in use for systemic infections
 - Imidazoles like miconazole used in topical creams and ointments
- Allylamines
 - Inhibit pathway of ergosterol synthesis
 - Administered topically
 - Used for treatment of dermatophyte infections

Mechanisms of Action of Antifungal Drugs

- Cell wall synthesis
 - Echinocandins
 - Family of agents that interfere with synthesis component of fungal cell wall
- Cell division
 - Griseofulvin
 - Exact mechanism unknown
 - Appears to interfere with action of tubulin
 - Selective toxicity may be due to increased uptake by fungal cells
 - Used to treat skin and nail infections
- Nucleic acid synthesis
 - Flucytosine
 - Synthetic derivative of cytosine
 - Flucytosine converted to 5-fluorouracil
 - Inhibits enzymes required for nucleic acid synthesis
 - Often combined with amphotericin

Mechanisms of Action of Antiprotozoans and Anthelmintics

- Many antiparasitic drugs most likely interfere with biosynthetic pathways of protozoan parasites or neuromuscular function of worms
- Example of parasitic drugs includes
 - Malarone
 - Synergistic combination of atovaquone and proguanil HCl
- Used to treat malaria
 - Interferes with mitochondrial folate synthesis



Review: antimicrobial

- What's antibiotics? (history/development)

Inhibit = bacteriostatic

Kill = bacteriocidal

- How do the antibiotics work?

Target on pathogen specific molecules
to stop its colonization/activity

- What's the difference among antibacterial, antiviral and antifungal compounds?

Target on different targets, different
processes