

DDD & MEDICINAL ORGANIC CHEMISTRY

BRIEF HISTORY

They used potions, herbal medicines in crude forms (not purified)

- Ancient people used natural products as drugs – however, many of the products were very toxic
 - Tartar emetic: banned in Paris on 1566
- 19th century- Extraction of pure substances from natural products
 - few of the compounds isolated proved to be satisfactory as therapeutic agents
 - majority discovered were too toxic
- The search to find less toxic substances than those based on natural sources resulted in the introduction of synthetic substances as drugs → LEADS

THE PRESENT

Finding the Lead

LEAD – compound w/ a significant pharmacologic activity (clinical value that must be desirable = therapeutic)

Choose a Disease

- *Market first, science follows*
- A huge investment has to be made in the research and development of a new drug
- Considerations:
 - Economic
 - Medical

Choose a Drug Target

- An understanding of which biomacromolecules are involved in a particular disease state is clearly important
- + Macromolecules that are targeted by drugs and those agents will modify the function of targets
- + Selecting target proteins that they could test using a certain compound of interest

Example: HTN (disease)

- contributory proteins that could participate to increase blood pressure:
- > receptors (could lead to increase in blood pressure) angiotensin receptors, vasopressin
- > co-transporters (membrane proteins that allow exchange of compounds) could lead to increase of blood volume
- > ion channels (could lead to constrictions of blood vessels/vasoconstriction)

Identify a Bioassay (testing method)

- The test should be simple, quick, and relevant, as there are usually a large number of compounds to be analyzed
 - Human testing is not possible at such an early stage
 - In vitro (w/o living organism) test first, then In vivo (w/ living organisms like animals and humans)
- > **to avoid risking a life**

In vitro tests

- Do not involve live animals
 - Specific tissues, cells or enzymes are used (being cultured on a laboratory)
- > **if the test becomes positive, it is now confident to proceed on In vivo test**

In vivo tests

- Tests on animals often involve inducing a clinical condition in the animal to produce observable symptoms
 - Often needed to check whether the drugs have desired pharmacological activity and also to monitor their pharmacokinetic properties
- > **simulate the effect of the drug inside the body of a living organism**

Find a Lead compound

- Lead compound is a compound which shows the desired pharmacological activity
 - The lead compound provides a start for the drug design and development process
- > **all positive results on the in vitro test, can be candidates**

Ways to discover a LEAD compound:

- Natural products screening (most common and oldest)
 - > everything that the environment can provide can be screened
 - Most biologically active natural products are secondary metabolites (products formed when the biological molecules like carbohydrates, lipids and proteins were metabolized) with quite complex structures and several chiral centers
 - The study of medicines derived from natural resources is known as pharmacognosy
 - Sources:
 - Plants

- Microorganisms
- marine life (algae)
- animals
- venoms and toxins

- Medical Folklore

→ Traditional/folkloric claims

>inspiration to test a compound from the “sabe sabe” or verifying if it’s true or not

- Screening Synthetic Compound Libraries

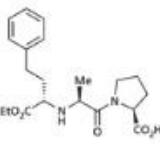
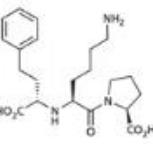
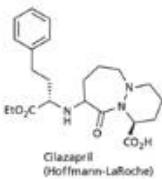
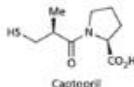
>saved compounds but not yet tested

- Pharmaceutical companies often screen their library of compounds whenever they study a new target
- Pharmaceutical companies often try to diversify their range of structures by purchasing novel compounds prepared by research group elsewhere

- Existing drugs

>network wars

- “Me too” and “Me better” drugs
 - many companies use established drugs from their competitors as lead compounds
 - AIM: to modify the structure sufficiently such that it avoids patent restrictions, retains activity and ideally has improved therapeutic properties
 - Example: Captopril



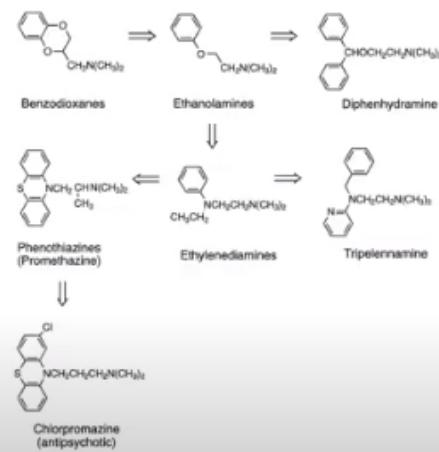
>Captopril – it has incompatibilities in the GIT, serves as the LEAD compound

>Ace-inhibitors: Cilazapril (Hoffmann-LaRoche), Lisinopril (Merck), Enalapril (Merck) – this are “better” than Captopril

→ Enhancing a side effect

>the side effect of a drug could serve as the company’s opportunity to make it a therapeutic application/clinical use of a drug

- Benzodioxanes (H-antagonist)
 - Diphenhydramine (antiallergy)
 - Tripelennamine (antiallergy)
- Promethazine (anesthetic agent)
- Chlorpromazine (tranquilizer)



>discovering agents that would act in the autonomic nervous system

>upon discovering benzodioxanes, they discovered that it could block Histamine-1 receptors and anti-allergy

>they improved the structure of benzodioxanes that resulted to extraction of ethanolamines (diphenhydramine-antiallergy) and ethylenediamines (tripelennamine-antiallergy)

>they further modified the structure of diphenhydramine and tripelennamine, and they discover that because of its anti-allergy effects it causes sedation

>they modified the structure of this anti-histamines and extracted promethazine (causes extensive sedation), its clinical use is anesthetic agent

>they modified again the structure of promethazine, until they discovered chlorpromazine which is a tranquilizer (pampakalma) that is antipsychotic agent

- Computer-aided Design (advance)

- Molecular modelling software programs can be used to study the binding site and to design molecules which will fit and bind to the site (de novo drug design)

>modify/enhance the effect of the drug by altering the binding ability of the drug to its target

- Serendipity

- Frequently, lead compounds are found as a result of serendipity (by chance)

Isolation & Purification

- If the lead compound (or active principle) is present in a mixture of compounds from a natural source or a combinatorial synthesis, it has to be isolated and purified

>it needs to be isolated and purified to be able to design the rightful structure for the lead compound

Structure Elucidation

- Structure determination is a relatively straightforward process and it is only when the natural product is obtained in minute quantities that a full synthesis is required to establish its structure

>identifying the structure of a compound to further modify it

- Analytical techniques:

- X-ray crystallography
 - requires a suitable crystal of the sample
- NMR spectroscopy
 - (most common because flexible) – it can be carried out on any sample, whether it be a solid, oil, or liquid

DRUG DESIGN

>raw materials are the lead

1. Identify Structure-activity relationships (SARs)

- Relationship of how structural features of the molecule contribute to, or take away from, the desired biologic activity

>aim is to identify the parts of the molecule that are active (retained) and not active (modified or remove)

- SAR studies are essential in drug optimization to find analogues with better activity and selectivity

2. Identify the Pharmacophore

The Pharmacophore

>functional groups that are required for the activity

>functional groups that goes to the target

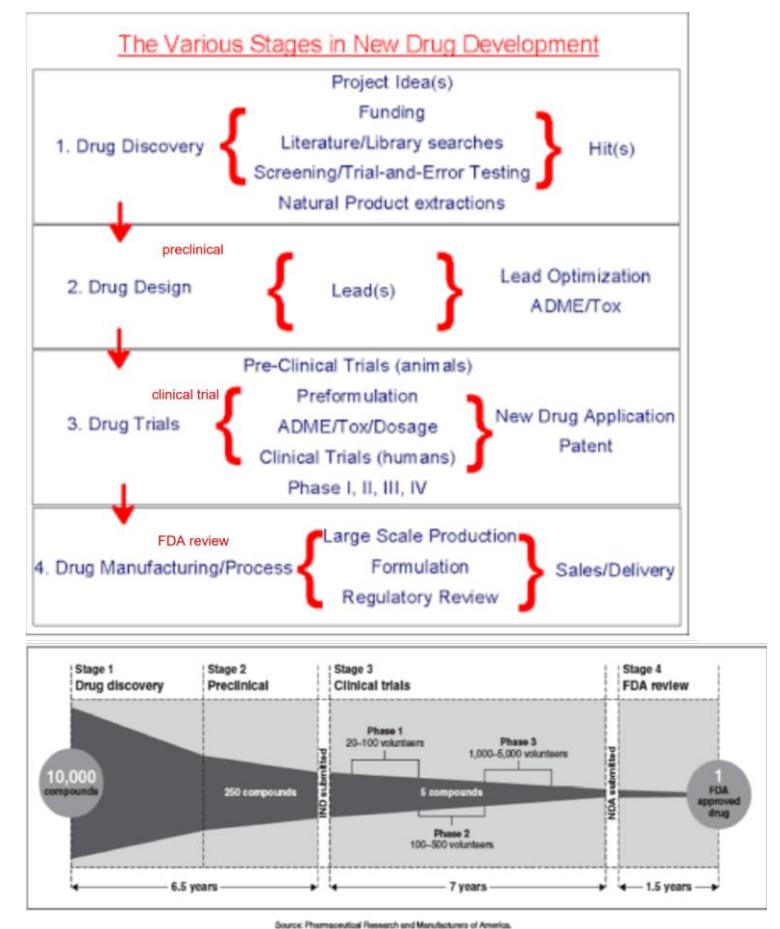
- Summarizes the important binding groups (active) that are required for activity, and their relative positions in space with respect to each other

3. Drug Optimization

- Pharmacodynamic optimization
- Pharmacokinetic optimization

>Enhance the intermolecular bonds between the target and the drugs to be able to make the drugs powerful and less toxic and improve the ADME profile of the drug to ensure that the drug will reach its target

DRUG DEVELOPMENT



Stage 1: Drug discovery

Stage 2: Preclinical

Stage 3: Clinical trials

Phase 1: Safety

Phase 2: Safety & Dosing

Phase 3: Safety & Efficacy

Phase 4: Post approval surveillance

Stage 4: FDA review

LEADS AND ANALOGUES: Desirable Properties

>desirable means is to have good profiles to be able to have efficient deliveries to the target system providing an effect

Bioavailability

- amount or fraction of the dose of a drug that is found in general circulation
 - For a compound to be suitable as a lead, it must be bioavailable

Lipinski's Rule of Fives

>guidelines to identify if a drug would be likely orally bioavailable or not

>multiples of five

- A molecular mass less than 500

- A calculated value of log P (partition coefficient) less than 5
- Less than ten hydrogen bond acceptor groups (e.g. -O- and -N-, etc.)
- Less than five hydrogen bond donor groups (e.g., NH and OH, etc.)

Note/Specification

- Any compound that fails to comply with two or more of the rules is unlikely to be bioavailable, that is, it is unlikely to be active
- It should be realized that Lipinski's and other similar rules are only guidelines

Solubility

> (aqueous sol'n to promote entry in the blood)

- It is desirable that leads and analogues have a balance between their water solubility and their lipophilicity
- Most drugs are administered either as aqueous or solid preparations, and so need to be water soluble in order to be transported through the body to its site of action

> conversion of drugs into its salt forms (HCl, SO₄, PO₄)

- The lipophilicity of a compound is often represented by the partition coefficient of that compound in a defined solvent system

$$P(k) = \frac{\text{concentration in the oil phase}}{\text{concentration in the water phase}}$$

> in vitro test (use of glassware) oil phase (octanol)

> ↑P = more drugs will go with the oil (lipophilic)

> ↓P = hydrophilic

Biopharmaceutical Classification system

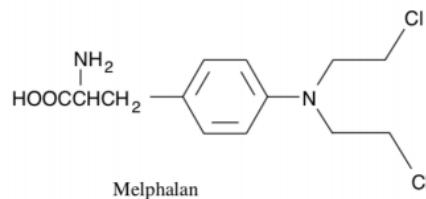
- Developed to optimize the development of an oral dosage from taking into consideration two rate-limiting factors, drug permeability and drug dissolution, the latter which is related to drug solubility
- Divided into four classes

<u>Class</u>	<u>Solubility</u>	<u>Permeability</u>
I	↑	↑
II	↓	↑
III	↑	↓
IV	↓	↓

- Class I
 - are well absorbed and are affected by a limited set of interaction that alter drug absorption
 - Examples: Acetaminophen and Metoprolol
- Class II
 - dissolution rate limits (slowest step) drug absorption
 - Absorption of this class of drugs is often enhanced in proportion to the fat content of the co-administered meal
 - Examples: Griseofulvin and Digoxin
- Class III
 - permeation is the rate-limiting step in the absorption from IR dosage forms
 - Examples: Acyclovir and Chloramphenicol
- Class IV
 - undesirable for good oral drug absorption
 - it takes large doses to facilitate the sufficient amount of drug that would be available in the SC
 - Examples: Furosemide and Paclitaxel

Structure

- Determines its ability to bind to receptors and other target sites
- Binding is the formation, either temporary or permanent of chemical bonds between the drug or analogue with the receptor
- Melphalan, which is used to treat cancer, owes its action to the strong covalent bonds it forms with DNA



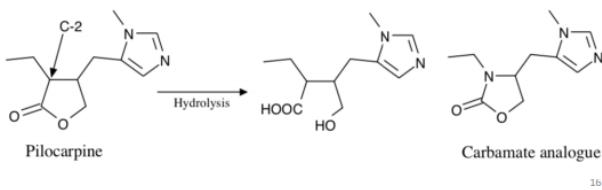
Stability

Stability after Administration (inside the body)

- for a drug to be effective, it must be stable long enough after administration for sufficient quantities of its to reach its target site
- Three strategies are commonly used for improving a drug *in situ* stability, namely:
 1. Modifying its structure
 2. Administering the drug as a more stable prodrug
 3. Using a suitable dosage form

Structure modification

- The main method of increasing drug stability in the biological system is to prepare a more stable analogue with the same pharmacological activity



Example: Pilocarpine (anti-glaucoma- eye condition) thru instillation

>FG- lactone (cyclic ester-can be hydrolyzed)
 >when it is instilled in the eyes, it will undergo hydrolysis and it becomes Inactive already
 >the activity of pilocarpine can only last for 3 hours
 >in the lactone ring at the carbon 2 it was replaced by nitrogen that became carbamate that is resistant to hydrolysis

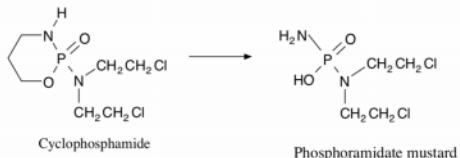
Complex Formation

>make a complex by adding carbohydrate myoti

- Complexing with hydroxypropyl- β -cyclodextrin (oligosaccharide w/ 6-8 glucose units) is used to improve both the stability and solubility of thalidomide, which is used to inhibit rejection of bone marrow transplants in the treatment of leukemia

Prodrugs (initially active but due to metabolism it become active)

- For example, cyclophosphamide (anticancer), which is used to treat a number of carcinomas and lymphomas, is metabolized in the liver to the corresponding phosphoramide mustard (alkylating agent), the active form of the drug



Suitable Dosage Form

- Use of EC tablet (disintegrate in the small intestine) – for drugs that can undergo extensive hydrolysis due to acidity of the stomach

Shelf-life (storage)

>determine a period of time if a drug is considered effective or not

- Time taken for a drug's pharmacological activity to decline to an unacceptable level
- No universal specification

- However, 10% (reduction) decomposition is often taken as an acceptable limit provided that the decomposition products are not toxic

Factors that deteriorate shelf-life

1. Microbial degradation and adverse chemical interaction and reactions

- Can be avoided by preparing the dosage form in the appropriate manner
- Storage under sterile conditions (maintain especially parenterals)
- Use of antimicrobial excipients

2. Adverse chemical interactions between the components of a dosage form

- Can be avoided by the use of suitable excipients (inert in character)

3. Decomposition by chemical reaction

- Usually brought about by heat, light, atmospheric oxidation, hydrolysis by atmospheric moisture and racemization
- May be minimized by correct storage with the use of refrigerators (heat resistant), light-proof containers (light resistant), air-tight lids (oxidation prone) and the appropriate excipients (prevent unwanted chemical reactions)

DRUG METABOLISM

Introduction

Drugs – important substances responsible for pharmacological actions

>drugs effect should not be perpetual (hindi pang habang buhay, not for long term)

>drugs should be excreted for it not to produce a toxic effect

>drugs should undergo metabolism (pre-requisite) so it can be excreted

- Xenobiotics require to undergo biotransformation (metabolism) for them to be excreted as hydrophilic metabolites (drugs are ionize, polar)

> Biotransformation – converting/transforming the drugs into hydrophilic, ionized and polar

→ Enzymes (biological catalyst responsible for different chemical rxn. In the body (hastening)) – hepatic and extrahepatic

>enzymes are highly concentrated in the hepatic tissues

>major organ for metabolism – liver

>tissues or other organs capable of metabolism (extrahepatic enzyme)

Importance

- Understanding biotransformation and bioactivation of xenobiotics is essential in assessing drug safety
- Pharmacologic and toxicologic activities of a drug are consequences of its metabolism

>pharmacologic – thru metabolism it can identify if the drugs are already inactive that is ready for excretion or to know if the drug is activated (prodrug)

>toxicologic – goal of metabolism is to make the drug non-toxic

Example: acetaminophen (well known in causing hepatotoxicity) – its toxic metabolite causes liver damage

Important note

- Hydrophilic drugs → paracellular transport (applicable for hydrophilic drugs)

>drugs enter through the presence of cellular pores (seen)

>gap junctions serve as the way so that hydrophilic drugs could enter

*there are drugs that do not need to undergo drug metabolism because it is already hydrophilic its route of transport is via paracellular transport

>lipophilic – cell membrane via diffusion

- Resistant to metabolizing enzymes
- Excreted largely “unchanged” (no shortcut)

Factors affecting drug metabolism

>drug metabolism is not absolute (not same fate)

Genetic factors

- Depends on racial and ethnic characteristics

>genes dictate certain characteristics and synthesis of enzymes

>because of genetic factors there is variation of activities of enzymes

- Pharmacogenomics the study of heritable traits affecting patient response to drug treatment

Example: metabolism of ETHANOL

>Ethanol (alcohol) when enters to the body it has metabolic fate

>Ethanol (primary alcohol) → Oxidation

1st oxi. rxn: $ETOH \xrightarrow{\text{dehydrogenase}} \text{acetaldehyde}$

=toxic in the body especially when accumulated

>can cause N & V, tachycardia, and redness

2nd oxi. rxn: $\text{acetaldehyde} \xrightarrow{\text{dehydrogenase}} \text{acetic acid}$

=non-toxic, readily excreted

>aldehyde dehydrogenase – poorly expressed (mababa) in Chinese and Japanese = accumulation of acetaldehyde =alcohol intolerance

Physiologic factors

- Age

>in elderly, there is a decline in drug metabolism related to decrease in hepatic blood flow, glomerular filtration rate, hepatic microsomal enzyme activity, plasma protein binding and body mass resulting in profound changes in drug plasma concentrations and renal clearance

>there are metabolizing enzymes in the placenta helping to protect the fetus

>fetus and newborns are deficient in some microsomal enzymes, particularly glucuronide conjugation, which can lead to jaundice in newborns

- Pregnancy different approach because they are more sensitive
- Hormones varies because hormones of a man differ from the hormones of a woman
- Gender

>hormonal differences which can lead to different levels of inducible oxidizing enzymes

Examples:

>side-chain oxidation of propranolol is 50% faster in males than in females

>drugs that are cleared by oxidative metabolism are more rapid in males than in female (chlordiazepoxide and lidocaine)

>diazepam, prednisolone, caffeine and acetaminophen are metabolized slightly faster by women than by men

- Changes in intestinal flora -it has metabolizing enzyme or metabolic activity
- Disease there are disease that can enhance or reduce activity of enzyme depending on the situation
- Nutritional status

Pharmacodynamic and Pharmacokinetic factors

>dynamic (effects/molecular actions by drugs), kinetics (ADME)

- Dose, frequency and route of administration, plus tissue distribution and protein binding of a drug, affect its metabolism

>dose = ↑ dose, greater extent of metabolism

>frequency = ↑ frequent, greater frequency of metabolism (more intake, more requirement for excretion)

>route = less or more (IV-less extent of metabolism)

- Example: conjugation of drugs with glutathione

>since there is a limited amount of glutathione available for conjugation, the dose and frequency of dosing of drugs conjugated by glutathione can saturate the system and lead to alternative paths of metabolism

Environmental factors

- Interaction with environmental substances
>ingested or inhaled environmental substances can compete for metabolizing enzymes, induce or inhibit enzymes, or even poison enzymes (carbon monoxide and pesticides)

Drug-Drug, Drug-Food, and Drug-Supplement interactions

>interactions that could alter the metabolism (change/modify the potential activities of drugs)

- a) Enzyme Induction – enhance rate of metabolism (happens because of “suspect” enzyme inducers)
- b) Enzyme Inhibition – reduce rate of metabolism (happens because of “suspect” enzyme inhibitors)

Metabolism Inhibitors	Example
Macrolide antibiotics	Erythromycin
Imidazole antifungals	Ketoconazole
Quinolone antimicrobial	Ciprofloxacin
Additional drugs	Cimetidine Omeprazole Ranitidine

Metabolism Inducers	Example
Tuberculosis drugs	Rifampin, Isoniazid
Anti-seizure drugs	Phenobarbital, Phenytion
Environmental toxins	Polycyclic aromatic hydrocarbons
Additional drugs	Prednisone Ethanol

Example: Drug interaction – drug interacting w/ other components (suspect- cause of interaction, victim-affected)

- >**suspect – precipitant drug**
- >**victim – object drug**

Drug-Drug Interactions

Object: WARFARIN – can cause bleeding

Suspects:

Rifampin	Cimetidine
>inducer >stimulate the enzyme that would metabolize (inactivate) warfarin >enhance metabolism of warfarin >more IA = less effect =clot (thrombosis)	>inhibitor >reduce enzymic action >reduce metabolism of warfarin >enhance of effect (extended) >extensive bleeding

>drug-drug and drug-food/herbal interaction are a common clinical problem

>CYP450 isozymes are the enzymes most affected
>inhibition of metabolizing enzymes can lead to prolonged drug action and possible toxic overdose

Drug-Food Interactions

Examples of GFJ-Drug Interactions

- GFJ → Statins rhabdomyolysis
- GFJ → Dihydropyridines increased vasodilation
- GFJ → Repaglinide hypoglycemia
- GFJ → Amiodarone increased toxicity
- GFJ → Clopidogrel modulated antiplatelet activity

>induction of metabolizing enzymes decreases duration of action and causes ineffectual drug therapy

>most drug-drug interaction are metabolism-based, that is, competition between two drugs for the enzyme-active-site

>Some drug-drug interactions are mechanism-based in that the inhibitory effect of the drug only occurs after an activation step producing an active inhibitor

>Grapefruit juice can significantly increase the oral bioavailability of drugs that are metabolized by CYP3A4 in the intestinal tract. In addition, compounds present in GFJ can inhibit intestinal P-glycoprotein (P-gp), which can also enhance drug bioavailability

Drug-Dietary Supplement Interactions

>the present interest and widespread use of herbal remedies creates the possibility of their interaction with prescription drugs

>Some of the more common dietary supplement that cause interactions include St. John’s Wort, Echinacea, Ginko Biloba, and Kava

>some dietary supplements are hepatotoxic: Chaparral, Comfrey, Germander, Skullcap, Pennyroyal and Valerian Root

Metabolic Pathways

>metabolism/detoxication makes the drug inactive meaning, it is not capable of inducing its pharmacological action

- Drugs undergo enzymic transformations that result frequently in the loss of pharmacologic activity
- The term “detoxification” (make IA & nontoxic) describes the result of such metabolic changes
- Exception → Bioactivation (because of chemical conversion) Examples:
 - Acetanilide (undergoes hydroxylation) → Acetaminophen (more active analgesic/antipyretic agent)
 - Parathion (route: inhalation) undergoes desulfurization (desulfurized) → Paraoxon (highly toxic that can cause extensive parasympathetic toxicity/deadly)
- The liver (\uparrow concentration of enzymes) is the primary site of drug metabolism
- Although extrahepatic metabolism can also happen:
 - Nervous tissue – nerves/neurons that possess metabolizing enzymes

>monoamine oxidase (found in nerve tissues) – responsible for metabolism of biogenic amines & catecholamines like Norepinephrine, epinephrine & dopamine that would make them IA

>acetylcholinesterase that can cause metabolism of acetylcholine

→ GIT

Example: Peripheral DOPA decarboxylase – enzyme that is responsible for metabolism of Levodopa → Dopamine

Parkinsonism (CNS disease characterized by low level of Dopamine in the brain)

Goal: increase dopamine levels in the brain

Initial thinking: administer dopamine by mouth

>the problem is if it's administered by mouth, dopamine is not permeable in the 3B

Remedy: instead of administering dopamine, it is converted to Levodopa (permeable in 3B)

>when it passes the 3B it will meet the enzyme DOPA decarboxylase so when it enters the brain it will be converted to dopamine

>Levodopa (administered by mouth it will go through GIT) when it is intake, it will meet

peripheral DOPA decarboxylase to convert it to pre-maturely dopamine

>in GIT it will be dopamine meaning it cannot pass thru 3B and it will not go to the brain

Solution: **Levodopa Combined w/ Carbidopa**

>Carbidopa is a peripheral DOPA decarboxylase inhibitor (it will protect levodopa for it not to be dopamine)

>Carbidopa cannot enter the 3B, only levodopa can enter the brain that will become dopamine

→ Kidneys

Example: Renal dehydropeptidase responsible for metabolism of Imipenem (antibacterial)

>decrease antibacterial activity of Imipenem that can enhance toxicity

>problem is renal dehydropeptidase

>Imipenem is combined with Cilastatin which is an inhibitor of renal dehydropeptidase

→ Lungs and Plasma

>you can see cytochromes

Categories of Drug Metabolism

1. Phase 1 Reactions
2. Phase 2 reactions

PHASE 1 REACTIONS (FUNCTIONALIZATION)

- Include oxidation (dominant), hydroxylation, reduction and hydrolysis
- Major goals: to enhance the polarity/water solubility of the substrate (drug intended to metabolize)
- Introduction of a new functional group into the substrate molecule
 - Modification of an existing functional group
 - Exposition of an existing functional group for phase 2 transfer reactions

1. Oxidation
2. Reduction
3. Hydrolysis

OXIDATION (most common)

>oxidation happens because of enzymes (capable of oxidation can vary depending on its source or properties)

- Mechanism of Oxidation
 - Cytochrome (CYP)-mediated
 - Non-CYP mediated

Cytochrome (CYP) mediated

>cytochrome is the enzyme that undergoes oxidation

>majority of oxidative reactions are caused by cytochrome that is why it is dominant

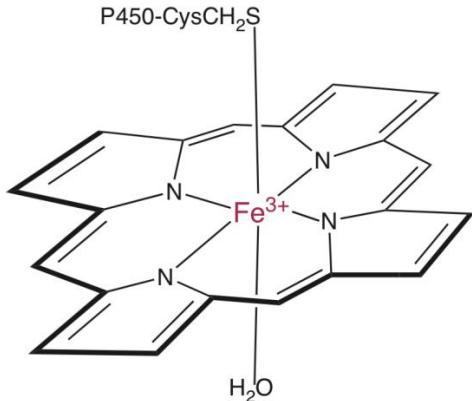
>50% drugs undergo CYP mediation

- Catalyzed by a cytochrome P450 monooxygenase enzyme system (CYP450)
 - Group of membrane-bound monooxygenases found in the smooth ER of the liver and other extrahepatic tissues
 - Mixed function oxidase or microsomal hydroxylase

CYP450

- Provide Multicomponent electron-transport system (lead to oxidation of substrate), responsible for the oxidative metabolism of a variety of endogenous substrates and exogenous compounds
- Most important function: to “activate” molecular oxygen (dioxygen)
- The introduction of a hydroxyl group into the hydrophobic substrate molecule provides a site for subsequent conjugation with hydrophilic compounds (Phase 2)*
- Components of CYP450:
 - Heme protein (P450) (most important)
 - Iron protoporphyrin → central to the functioning of this unique superfamily
>most important central component in the functioning as a whole of CYP450
 - Flavoprotein: NADPH-P450 reductase
 - Contains both flavin mononucleotide (FMN) and flavin dinucleotide (FAD)
 - Phospholipid: Phosphatidylcholine
 - Facilitates the transfer of electrons from NADPH-P450 reductase to P450
>flavoprotein and phospholipid is for electron transfer

Heme protein (P450)



>Heme is an example of protein containing central iron ring structure attached w/ protoporphyrin (structure that shield the iron)
>the state of iron is 3+ (Ferric state for the substrate to bind to it)

>there are 6 coordinated bonds attached with the iron

>1-4 attached w/ nitrogen that are designated for the attachment of iron to the ring

>5th coordinated bond (proximal bond) – attached w/ amino acid residue cysteine, important for oxenoid intermediate formation

>6th coordinated bond (distal bond) depends on the state of iron ($\text{Fe}^{3+} = \text{H}_2\text{O}$) ($\text{Fe}^{2+} = \text{O}_2$ or CO)

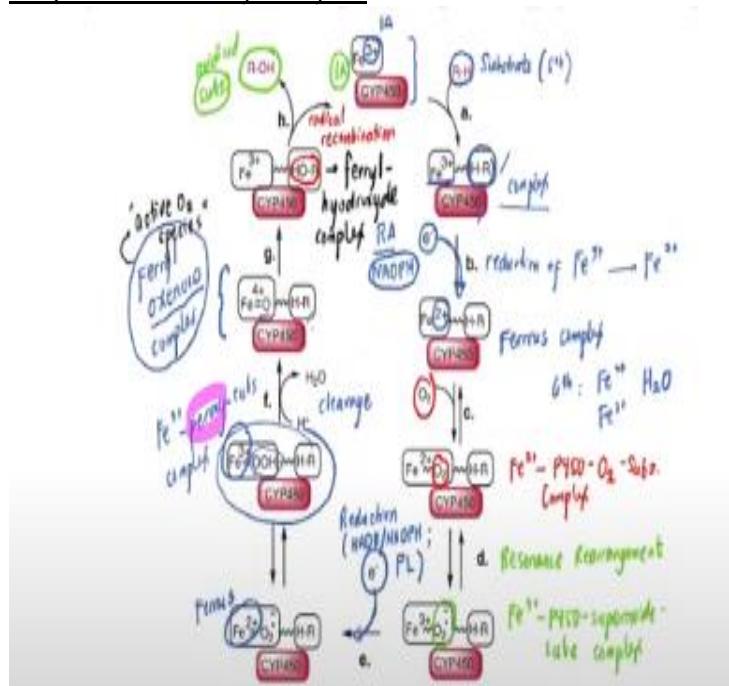
- P450 is important due to its vital role in oxygen activation and substrate binding (5th)
 - >make the ferric state at the center to become ferrous*
- The active site of P450 consists of a hydrophobic substrate-binding domain in which is embedded an iron protoporphyrin (heme) prosthetic group

Requirements

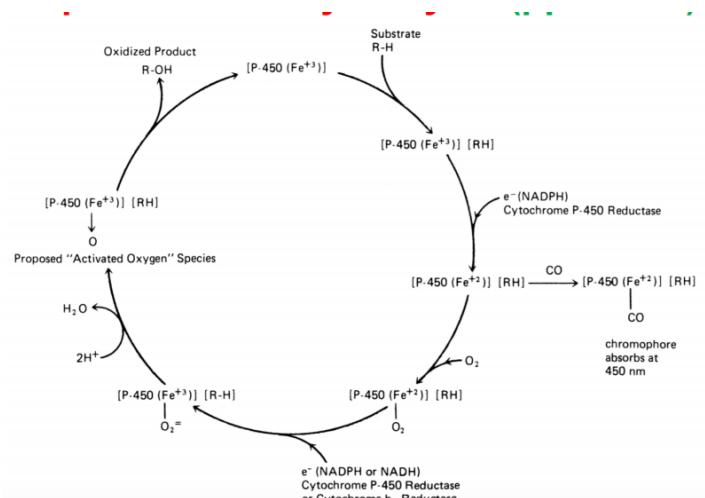
- A **heme protein**: CYP450 is in resting state when bound to Fe^{3+} (oxidized) and an active state when bound to Fe^{2+} (reduced form)
- NADH or NADPH: **Cofactors** of CYP450 reductase of CYP450 b5 reductase (nonheme flavoprotein) to provide an electron to the S-CYP450 complex yielding a peroxide dianion intermediate
- A **hydrogen atom** present on the drug molecule that can be abstracted
- Molecular oxygen** (O_2)

$\text{R-H (drug)} + \text{O}_2 + 2 \text{ electrons (NADH/NADPH)} + 2 \text{ hydrogens} \rightarrow (\text{via cytochrome}) \text{ R-OH (hydroxylated)} + \text{H}_2\text{O}$

Steps in the Catalytic Cycle



- >Heme protein is in the ferric state (inactive)
- >attachment of R-H (substrate) to the heme at 5th coordinated bond
- (a) Ferric CYP450 substrate complex
- (b) reduction of iron making it ferrous that needed reducing agent that would provide electron
the reducing agent is NADPH/NADH
there is electron transfer to the complex making it Ferrous
- (c) oxygenation step (addition of oxygen) Ferrous state -open for oxygen
- >**Ferrous P450 oxygen substrate complex**
- (d) involves resonance rearrangement that would convert ferrous to ferric
- >**Ferric P450 superoxide substrate complex**
- (e) electron enters, reduction reaction (NADH/NADPH/Phospholipid)
reduce state of iron into ferrous
- >consider fast reaction process (short), it will undergo rearrangement right away, it will go back to its ferric state
- >but superoxide has been modified it became peroxy component
- >**Ferric P450 peroxy substrate complex**
- (f) involved heterocyclic cleavage that causes conversion of ferric peroxy into ferryl oxynoid complex (Fe⁴⁺ and oxygen)
- >**ferryl oxenoid complex is observed at the 5th coordinated bond which is considered as "active oxygen species"**
- >when it is in ferryl oxenoid complex (causes hydroxylation) that would cause the formation of ferryl hydroxide complex (g)
- (h) due to radical recombination, the P450 it becomes IA again, and will detached and the substrate (oxidized) will be separated



Classifications of P450: Multigene Family

Nomenclature of P450s:

1. Root symbol CYP
 2. Followed by an Arabic numeral designating the family member (CYP1, CYP2, CYP3)
 3. A letter denoting the subfamily (CYP1A, CYP2C, CYP2E)
 4. And another Arabic numeral representing the individual gene (CYP1A2)
- There are about 17 P450 isoforms
 - **CYP3A and CYP2C families**
 - Most involved in the metabolism of clinically relevant drugs
 - CYP3A4 (dominant) → metabolized 2/3 of all drugs
 - **CYP1A2**
 - Predominantly involved in the bioactivation of environmental substances
 - **oxidizes to become active and sometime toxic**
 - **CYP1A1/2** isoforms have planar binding sites and metabolize only aromatic planar compounds
 - **CYP2D6** exhibits high affinities for specific apoprotein interactions (hydrogen bonds, ion-polar formation) for specific substrates such as lipophilic amines

Oxidative Reaction by CYP450 Isoforms

Aliphatic and Alicyclic Hydroxylation

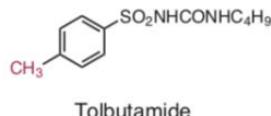
- The principal metabolic pathway of the methyl group is oxidation
- >**aliphatic (HC straight chain)**
- >**alicyclic (ring structure but not aromatic)**
- >**product of methyl is hydroxymethyl and could lead to carboxylic acids**

Methyl → Hydroxymethyl derivative → RCOOH

- Some methyl groups are oxidized only to the hydroxymethyl derivative without further oxidation to the acid
- Structures with several equivalent methyl groups:
 - Only one methyl group is oxidized
 - Aromatic methyl groups → the **para** methyl is the most vulnerable because it is less sterically hindered
- >**para (opposite)**
- >**meta (alternate)**
- >**ortho (adjacent)**

Aromatic methyl oxidation

1. Aromatic Methyl Oxidation



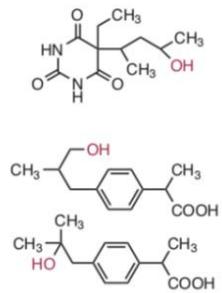
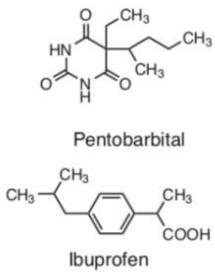
>aromatic methyl group (oxidation at the para methyl position)

>active myo (functional group)

>product is hydroxylated methyl group

Alkyl side chain oxidation

2. Alkyl Side Chain Oxidation



>methyl group of the straight chain

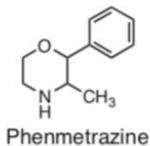
>oxidation at straight chain

>2 possible fates

>hydroxylation at the terminal (dulo) or penultimate (2nd to the last)

Heterocyclic Ring Oxidation

3. Heterocyclic Ring Oxidation



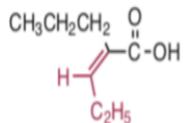
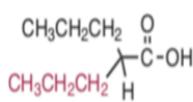
>the targeted substrate contains a ring structure but its heterocyclic (other atoms than C)

>methylene (FG) = carbon with only 2 hydrogen atoms (least hindered, adjacent to the hetero atom) being oxidized

>product is carbonyl (C=O) that is methylene group

Dehydration

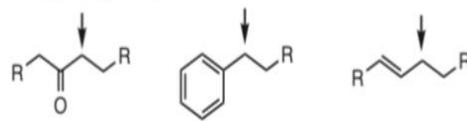
4. Dehydration



>could happen if the substrate is alkane (paraffin) that via dehydration becomes alkene (=, olefin)

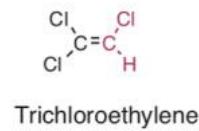
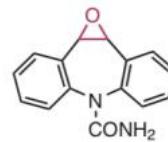
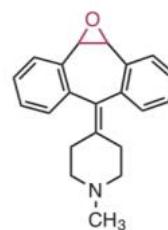
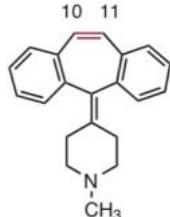
Privileged hydroxylation positions: effect of activating factors

5. Privileged Hydroxylation Positions: Effect of α -Activating Factors



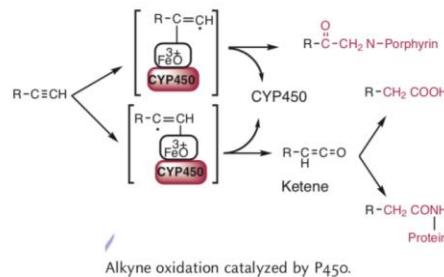
>alpha carbon adjacent to given FG is the one that undergoes hydroxylation

Alkene hydroxylation



>substrate is double bound the fates are
>formation of epoxide (alkene portion is part of the ring) or carbonyl (not part of the ring)
>epoxide (cyclic ether ROR)

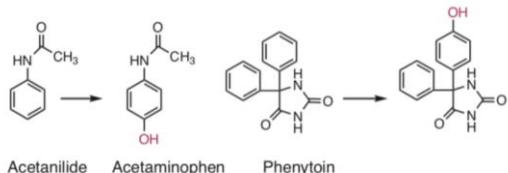
Alkyne hydroxylation



Alkyne oxidation catalyzed by P450.

>triple bonds present in the substrate
>undergo hydroxylation that depends on the reference carbon
>dulo (terminal), lower carbon number (alkenyl),
>terminal = formation of intermediate called ketene
>first p: carboxylic acid; second p: protein adduct
>alkenyl = product is ketoheme adduct

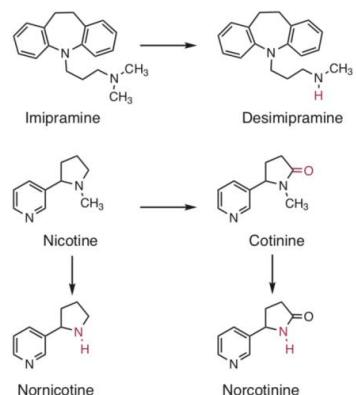
Aromatic hydroxylation



- >carbon in the benzene ring undergoes hydroxylation
- >the product being formed is a phenol (derivative of benzene that has OH group) OH group is attached at the para position

N-Dealkylation

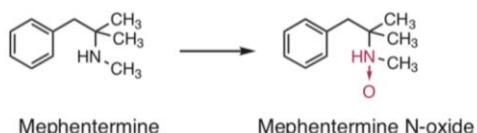
Dealkylation:



- >removal of methyl group attached to nitrogen
- >substrate are amines (RNH_2)
- >if the amine is tertiary the product is secondary amine
- >if the amine is secondary the product is primary
- >if the substrate is amine it is resistant to N-dealkylation
- >pag nor- removal of methyl group

N-oxidation

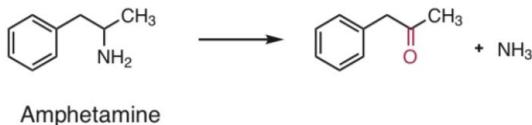
N-Oxidation



- >the amine may become N-oxide (addition of oxygen in the amine)

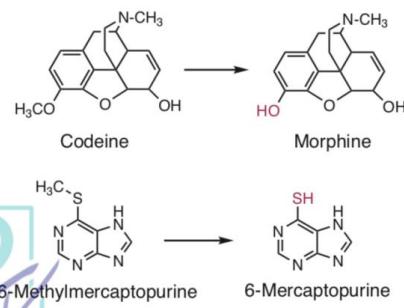
Deamination

Deamination:



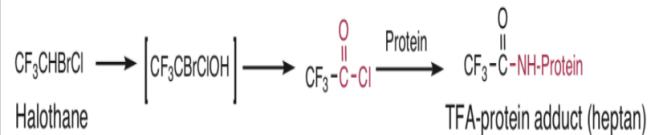
- >removing amino group
- >product is carbonyl and formation of ammonia

O- and S-dealkylations



- >occurs in ethers (ROR) and thioether (ROS)
- >product can be alcohol, phenol and carbonyl
- >removal of methyl to form ether, or thioether

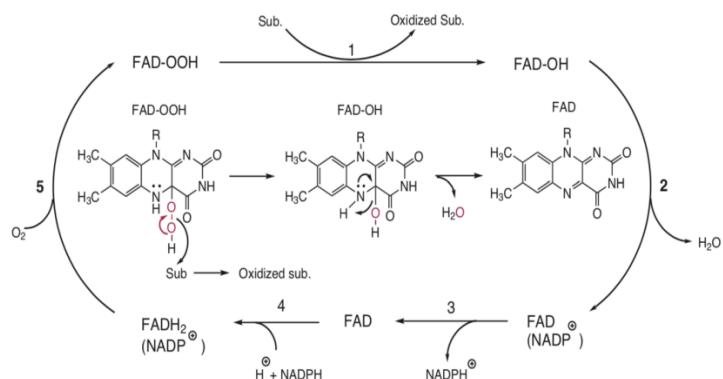
Oxidative dehydrohalogenation



- >Halogenated hydrocarbons (substrate) once they undergo oxidation, they become carbonyl halides and after that they may interact with physiological proteins producing immune response which are toxic

- >halogenated hydrocarbons contain halogens
- >carbonyl halides (halogen and carbonyl)
- >compounds that interact with proteins resulting to immune response is called hapten

Oxidations catalyzed by Flavin Monooxygenases (FMOs)



1. oxidation of substrate
>flavin is already IA
>2-5 goal is regeneration of enzyme
2. release of water
3. release of NADPH
4. reduction of FAD
5. oxidation of FADH2

Peroxidases and other Monooxygenases

- Closely related to CYP450
- Difference in coordinating ligands: His in POX
- Example: COX- cyclooxygenase

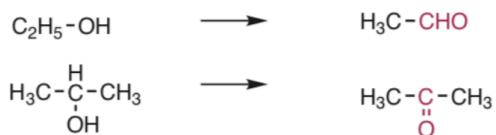
Non-CYP mediated

>the enzymes capable of oxidation are not members of CYP family

Oxidation of alcohol

Alcohol dehydrogenases

- alcohol to aldehyde/ketone



>depends on type of alcohol

>if the substrate is primary alcohol, the product is aldehyde

>if the substrate is secondary alcohol, the product is ketone

>tertiary alcohol is resistant from alcohol dehydrogenase

Aldehyde dehydrogenases

- aldehydes to carboxylic acids



>aldehydes are the substrate the product is carboxylic acids

Xanthine oxidases and Xanthine dehydrogenases

- xanthine oxidoreductases



>responsible for oxidation of purines

>hypoxanthine is being oxidized with the enzyme xanthine oxidase that results in the formation of uric acid

>too much hypoxanthine, uric acid will increase too
>too much uric acid (joint) can cause gout (inflammation/pain)

>allopurinol blocks xanthine oxidase



>6-mercaptopurine is an agent that is used as anti-cancer (very potent)

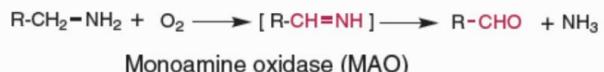
>to inactivate the substrate it oxidizes via XO resulting in inactive 6-mercaptopurine that is ready for excretion

>6-mercaptopurine is prepared in the form of prodrug: Azathioprine

>drug interaction between azathioprine and allopurinol (risky)

Monoamine oxidases

- Oxidative deamination of amines

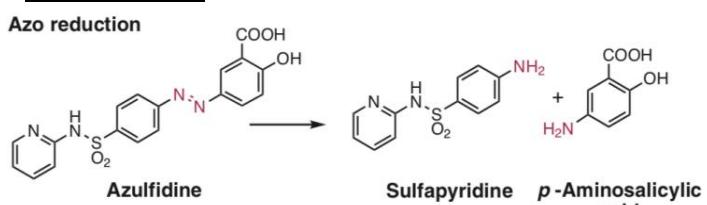


>MAO is the enzyme responsible for oxidation

REDUCTION

>less common

- Azo reduction

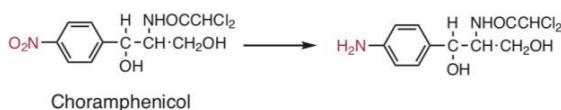


>azo functional group

>product: aromatic primary amines

- Nitro reduction

Nitro reduction

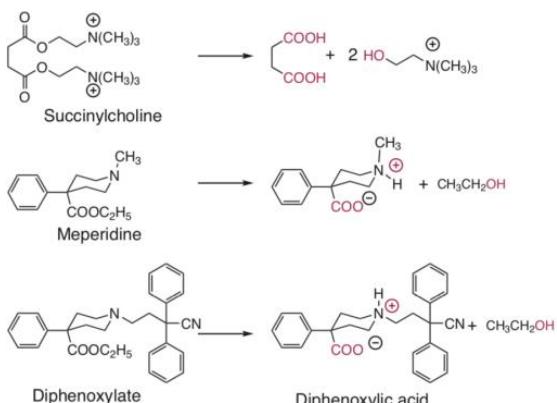


>nitro functional group

>product: aromatic primary amines

HYDROLYSIS

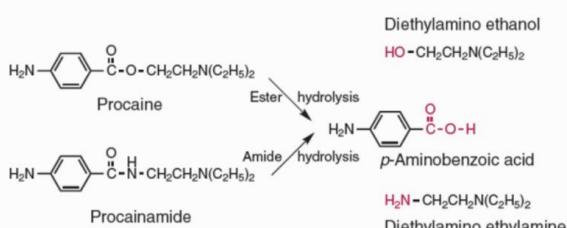
>break down of bonds because of water (hydrolytic enzymes)



Affected:

>ester substrate, product is carboxylic acid and alcohol

>amide substrate, product is carboxylic acid and amine



PHASE 2 REACTIONS (CONJUGATION) (add only)

- Enzymatic syntheses whereby a functional group is masked by the addition of a new group
→ Acetyl, sulfate, glucuronic acid, or certain amino acids
= increases the polarity of the drug or xenobiotic
- Most substances undergo both phase 1 and phase 2 reactions sequentially
- Addition of ionic hydrophilic moiety
→ Glucuronic acid, sulfate ester, glycine
- Excreted in the urine or bile
- Can be preceded by Ph 1, but for some with FG available for Ph 2, conjugation can be their fate

Products of Phase 2 Metabolism

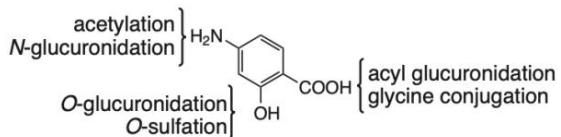
- Detoxification** (make it ready for excretion)
 - Hydrophilic products → excretion
- More active products** (A → more active)
 - Morphine → morphine-6-glucuronide (more effective analgesic)
 - Minoxidil → minoxidil sulfate (antihypertensive)

3) Formation of toxic products

- Carcinogenesis, allergy, tissue damage

Sequential conjugation

p-aminosalicylic acid



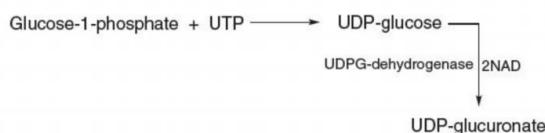
>simultaneously (sabay-sabay)

GLUCURONIDATION

- Most common route in adults
- Depends on the available supply of the ff:
 - Glucuronic acid in the liver (intended to bind to the drug)
 - Functional groups (present in the drugs) forming glucuronide conjugates
- >phenol
- >alcohol
- >Carboxylic acid
- >amine

Glucuronic acid (didikit sa gamut)

Synthesis: (liver)



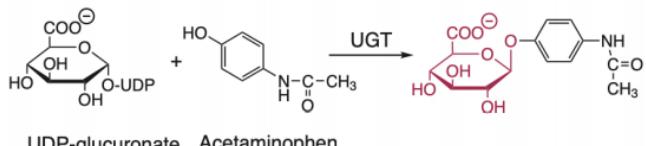
>glucose-1-phosphate and UTP (raw materials) undergo reactions forming the compound UDPGA (chemical name for glucuronic acid) this will form conjugate with xenobiotic

Mechanism:

- Xenobiotic + UDP-glucuronic acid (UDPGA)
 - Glucuronic acid in the liver
 - Functional groups forming glucuronide conjugates

O-Glucuronidation

O - Glucuronidation:

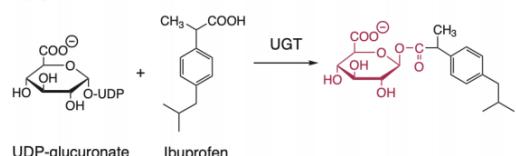


>substrate: phenol and alcohol

>product: ether-o-glucuronide

Acyl-glucuronidation

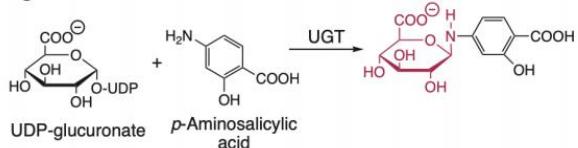
Acylglucuronidation:



>substrate: aromatic and aliphatic carboxylic acids
>product: ester glucuronides

N-glucuronidation

N-glucuronidation:



>substrate: aromatic amines
>product: N-glucuronides

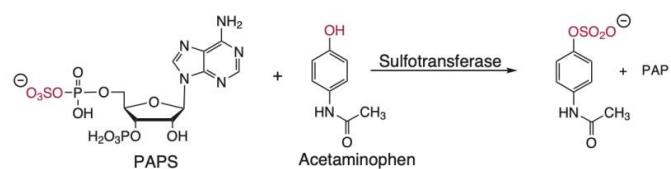
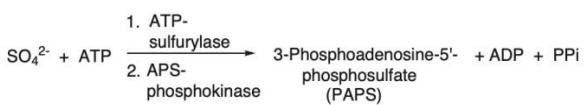
SULFONATION

- Important in the metabolism of steroids, catecholamines, thyroxine, bile acids, phenolics
- Product: sulfate ester \rightarrow totally ionized (ready for excretion)
- Dominant in neonates

Mechanism:

- Xenobiotic + PAPS \rightarrow R-O-SO₃H
- "participants"- required for sulfation to occur
- PAPS (hydrophilic myeoti)- provides FG to the xenobiotics
- contains sulfonic group (SO₃-) transferred to the xenobiotic that would lead to the metabolite (sulfate ester)
- >2 major compounds required (ATP and Inorganic sulfate)
- \rightarrow ↑PAPS, enhance sulfation

PAPS:

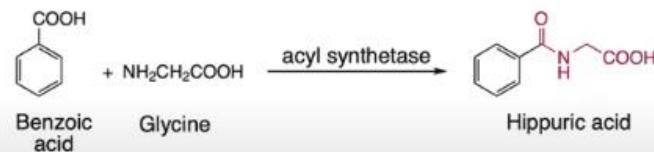


AMINO ACID CONJUGATION

- Important metabolism of drugs containing carboxylic acids

- Most common amino acid: Glycine (simplest amino acid)
- Product: Ionic conjugates with aromatic, arylalipathic and heterocyclic amino acids
 \rightarrow low in newborns

Glycine conjugation:

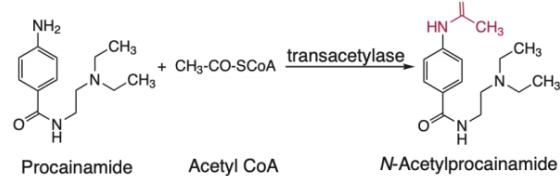


>contains CA and glycine that via acyl synthetase will form Hippuric acid

ACETYLATION

- Involves transfer of acetyl CoA to 1° aliphatic and aromatic amines, amino acids, hydrazines or sulfonamide groups

Acetylation:

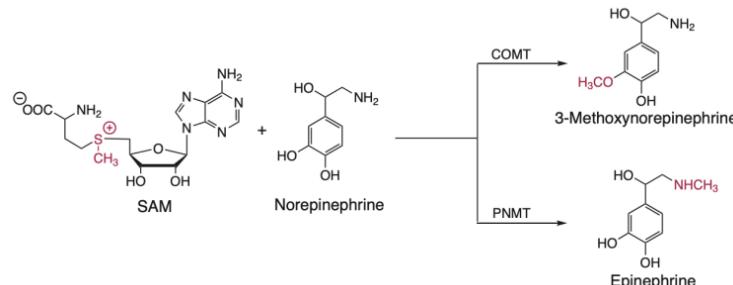


Polymorphism (genes that living organism have)

- Slow (poorly expressed, Egyptians, Mediterranean Jews) vs Fast acetylators (high enzymic action, Eskimos, Orientals)
 - Slow acetylators are more prone to drug-induced toxicities
 - Hydralazine, Isoniazid, Procainamide, Sulfonamides (HIPS)
- >HIPS- slow to activate (toxic)

METHYLATION (addition of methyl)

- Greater significance in the metabolism of endogenous compounds (naturally present in the body like catecholamines: NE, EPI, DA) than xenobiotics
- May form metabolites that are more active and lipophilic (unique)



- >SAM (Radical S-adenosyl methionine) myo
- >1st outcome: O-methylation (methyl group will be added at the meta position) meta-hydroxy position
- >COMT Catechol-O-Methyl transferases
- >2nd outcome: N-methylation (formation of amine)
- >PNMT Phenyl ethanolamine N-methyl transferases

Glutathione Conjugation and Mercapturic Acid synthesis

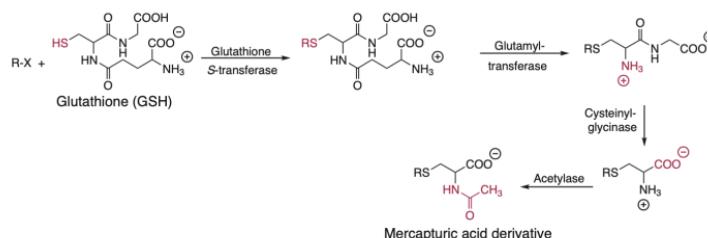
- Mercapturic acids are S-derivatives of NAC from glutathione
- The MAS is a protective mechanism against xenobiotic-induced hepatotoxicity or carcinogenicity, which are electrophiles

>xenobiotics that are electrophiles that could form covalent bonds with macromolecules like proteins and nucleic acids that could lead to toxicity

>protein and nucleic acids are neutrophiles

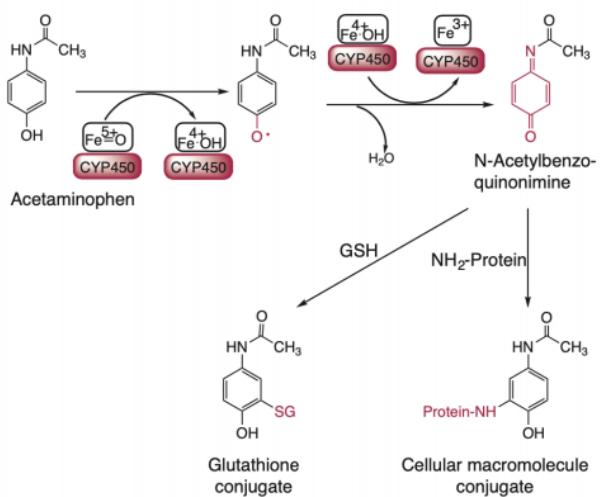
>protective mechanism to be able to prevent attachment of xenobiotic electrophiles to neutrophiles

>protection by diverting the xenobiotics



>glutathione could act as shield to divert the attention of the substrate

>action of glutathione s-transferase can increase thiol ionization in glutathione = enhancement of nucleophilicity, formation of glutathione adducts



LOCAL ANTI-INFECTIVES AND PRESERVATIVES

Overview

- Joseph Lister introduced antiseptic principles in surgery
 - >the main goal of antisepsis is to lower the incidence of post- surgical infection.
 - Phenol or AKA Carbolic Acid
- Paul Ehrlich's Concept of Selective Toxicity
 - >(Property of certain chemicals to kill one type of organism without harming another)
 - main tenet of antimicrobial chemotherapy.
 - >The concept of selective toxicity gives us idea the importance of the safety of medication, because if drugs or anti-infectives would target the harmful organisms only, that would protect the living cells in the body. (Kill those that shall be killed, and protect those that shall be protected)
- Local Agents → Iodine and Phenol
- The only systemic chemotherapeutic agents before Ehrlich were Cinchona for Malaria and Ipecac for Amebic Dysentery

>Successful Antiseptic contains these elements, from group 2B and also 5A of periodic table:

- Heavy Metals (Hg, As, Sb)
 - limited by their toxicity
- Dyes (Gentian violet, methylene blue) – Effective agent but is quite toxic.
- Discovery of Sulfonamides, other synthetic antimicrobials and antibiotics

Anti-Infectives

>Definition of terms regarding anti- infectives, agents may be classified according to different schemes, such as: type of chemical compound, biological property, therapeutic indication.

- Agents used to counteract or prevent infection
 - Local anti-infectives
 - Antifungals
 - Preservatives
 - Anthelmintics
 - Antibacterial
 - Antiprotzoa
 - Antimalarials
 - Antineoplastics
 - Antivirals

LOCAL ANTI-INFECTIVES

>This is also known as germicides – applied to the certain area affected by microorganism. (Locally Administered – topical, orally “for oral cavity only”)

- Antiseptics
 - Living tissue
- Disinfectants
 - Inanimate/nonliving tissue

>Not all goals are attained.

>Similarity is they will inhibit/kill growth of organisms. (basta threat such as pathogens)

>Difference is where you apply/put them, if it is applied in living tissue, it is antiseptic (such as on hands), Disinfectants are applied in inanimate/non-living things (Example: alcohols on objects are applied)

>In searching for local anti- infectives, usually the scientist/medicinal chemists have goals → This goal is called Ideal Agents.

Ideal Antiseptic

- exert a rapid and sustained (be it a broad spectrum or narrow spectrum agent) lethal action (kill) against microorganisms
- have a low surface tension
 - >there is easier spreadability on the parts if it has a low surface tension.
- retain activity in the presence of body fluids
 - >there are local injuries that is associated with secretion of certain fluid (those could serve obstruction for drugs)
 - >goal is despite of certain body fluids; the antiseptic should be able to reach its goal.
- non-irritating to tissues
- non-allergenic
- lacks systemic toxicity when applied to skin and mucous membrane
 - >if it produces toxicity its purpose will be defeated, that must have local effect only.
- should not interfere with healing

Ideal Disinfectant

- exerts a rapidly lethal action against all potentially pathogenic microorganisms and spores
- has good penetrating properties into organic matter
 - >to be able to reach the deeper parts of the object, to kill those.
- shares compatibility with organic compounds (particularly soaps)
- not inactivated by living tissue

- Non-corrosive
- Aesthetically pleasing (non-staining and odorless)

Definitions and Standards for Removing Microorganisms

- Antisepsis – Application of an agent to living tissue for the purpose of preventing infection.
- Decontamination – Destruction or marked reduction in the number or activity of microorganisms.
- Disinfection – Chemical or Physical treatment that destroys most vegetative microbes or viruses, but not spores, in or on inanimate surfaces.
- Sanitization – Reduction of microbial load on an inanimate surface to a level considered acceptable for public health purposes.
- Sterilization – A process intended to kill or remove all types of microorganisms, including spores, and usually including viruses with an acceptably low probability of survival.
- Pasteurization – A process that kills non-sporulating microorganisms by heat water or steam at 65 C to 100 C.

Handwashing

- Is the most important means of preventing transmission of infectious agents.

>That is why in every instruction worldwide, is to always wash your hands. (use soap or warm water)

>After washing hands, always use towel/tissue, not air dryer.

Alcohols and Related Compounds

- >Related compounds to alcohol is aldehydes.
- >Alcohol compounds are containing OH groups
- >Aldehydes contains RCOH (carbonyl compounds)
- >Alcohols and Aldehydes had been used as disinfectants for many years.

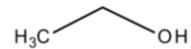
>The most common alcohol is, Ethyl alcohol and Isopropyl Alcohol.

- The antibacterial potencies of primary alcohols (tested against S. aureus) increase with MW up to C8.
 - >Antibacterial potency of alcohol can be observed in the number of carbons – the more carbon atom, the greater the activity.
 - >Maximum of 8 carbons because the more carbon present the greater the Vander Waals forces of attraction – if this force is higher, greater penetration of the alcohol on the microbial membrane.

- Branching Decreases antibacterial potency
- Primary (Greatest activity) > Secondary > Tertiary (Least Activity)
- 2-propanol Isopropyl Alcohol (secondary alcohol) is used commercially instead of n-propyl alcohol (Primary Alcohol).

>The reason why isopropyl is more used compared to n-propyl alcohol is because it is less expensive.
- Isopropyl alcohol is slightly more active than ethyl alcohol but both alcohols are not effective against spores.
- MOA: Protein and Carbohydrate Denaturation

>why microorganisms are being killed is by protein and carbohydrate denaturation (make proteins non-functional)



Alcohol

- ethyl alcohol, wine spirit, hydrated oxide of ethyl
- Commercial ethanol (95% v/v)

>this concentration forms azeotrope with water and distills at 78.2 C.
- Historical: Product of fermentation of grain
- Synthetic: Hydration of ethylene

>thru sulfuric acid (H_2SO_4) catalyzed hydration of ethylene.
- Greater than or equal 160 proof/concentration (does not alcoholic beverages, like whiskey, rum, gin)

Denatured Alcohol

- ethanol that is unfit for use in intoxicating beverages by addition of other substances

>because there are additional substances that could lead potential toxic if accumulated.

 - Completely Denatured
 - w/ methanol (wood alcohol) and benzene

>when taken inside the body, it could kill an individual.
 - Specially Denatured
 - Ethanol treated w/ other substance for specialized use
 - Ex. Tincture of iodine, methanol in mouthwashes and plant extracts

>not given internally

Uses of Alcohol

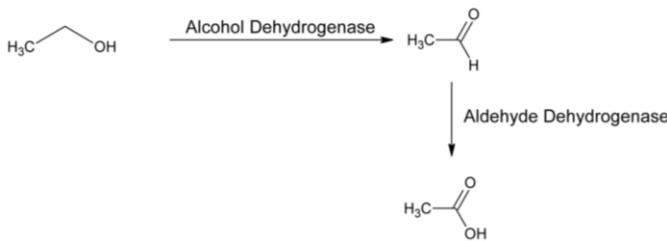
>Can be used for both external and internal use (majority of alcohol is for external).

- Antiseptic
 - Preservative
 - Mild Counterirritant
 - Solvent
 - Mild local anesthetic
 - Mild sedative
 - Carminative
- external
- internal

>Rubbing Alcohol – Usually used as astringent, rubefacient, and Mild Local Anesthetic – refrigerant feeling (cool feeling)

>It is also used internally such as for: Mild Pain killer, mild sedative, Weak vasodilator, Carminative.

Metabolism of Alcohol



- Acetaldehyde
→ N, V, flushing
- Antidote: Disulfiram

>Alcohol is further oxidized by Alcohol Dehydrogenase into an acetaldehyde and with the help of the enzyme aldehyde dehydrogenase it oxidizes into an acetic acid (accumulation of acetic acid is toxic in the body)

>Disulfiram (Aldehyde Dehydrogenase Inhibitor) is used to stop the accumulation of acetic acid

Application of Alcohols in Pharmacy

- Used in Preparations of the ff:
 - Spirits (Alcohol Only)
 - Tinctures (Hydroalcoholic)
 - Fluidextracts (Alcohol as cosolvent)

Different Concentrations of Alcohol

- 70% = most common (globally) (commercially used)
>70% is a rigid concentration as bactericidal.
- 60 – 95% = NO significant difference (as long as in this range, the activity is the same)
- <60% = effective but requires longer contact time
>long rubbing like 40%.
- >70%: used safely for preoperative sterilization of the skin

Dehydrated Ethanol

- Absolute ethanol (NLT 99% w/w)
 - >**Injected for local relief of pain in carcinomas or neuralgias.**
 - >**Must not be ingested because of presence of benzene (carcinogen).**
 - Prepared by azeotropic distillation of ethanol-benzene mixture

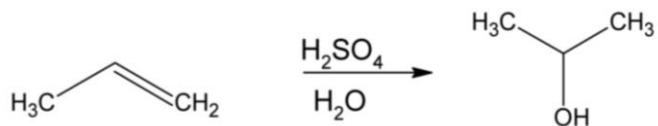
>Not considered as antiseptic or disinfectant.

>The reason why ethanol (Disinfectant and Antiseptic) is not formulated as 100% is because to be an effective protein denaturation, there must be presence of water.

- Efficient removal of water
- Very high affinity for water
- Stored in tightly sealed containers
- USES: Chemical reagent; solvent

Isopropyl Alcohol

- 2-Propanol – secondary alcohol.
- Prepared by hydration of propylene
 - >**Prepared commercially by sulfuric acid catalyze hydration of propylene.**
- Disinfectant
- Bactericidal at 50% to 95%
- 40% Isopropyl alcohol = 60% ethanol
- Used in gauze pads



Ethylene Oxide (gas form)

- Sterilize temperature-sensitive (heat labile compounds) equipment and pharmaceuticals
- Explosive in air (3 – 80%)
 - Remedy: Add carbon dioxide CARBOXIDE = 10% E.O. + 90% CO2
- MOA: Alkylating Agent
 - >**carcinogenic because it is alkylating, promoting DNA mutation.**

Aldehyde (Formaldehyde) $\text{H}_2\text{C}=\text{O}$

- IUPAC name: Methanal
- Germicidal action is slow but powerful
- Non-specific alkylation of nucleophilic functional groups in proteins and nucleic acids (DNA mutation)

>Used as embalming fluid – used because due to its activity it can fix tissues (fixation) – the problem is its prone to polymerization, and once polymerized, the product if paraformaldehyde (it cannot fix tissues), in order to fix the polymerization, the formaldehyde is converted to formalin.

Formalin (Aldehyde – Formaldehyde)

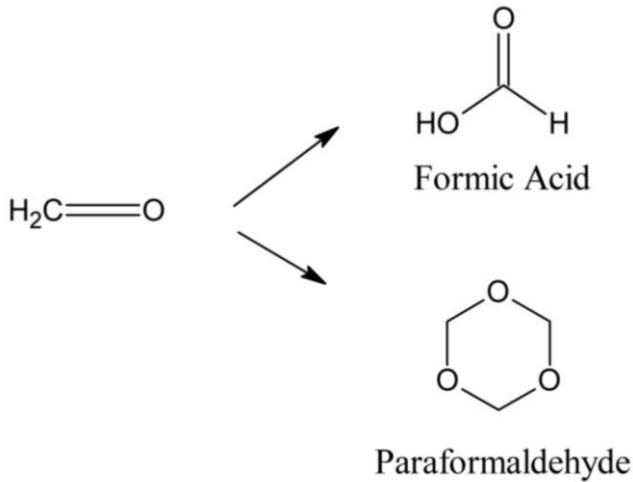
>Formalin is different from formaldehyde, because formalin has 2 compositions – formaldehyde + Methanol (retards polymerization)

→ NLT 37% w/v formaldehyde, with methanol (retards polymerization)

- Polymerization → Paraformaldehyde
- Oxidation → FORMIC ACID (methanoic acid)
- Irritating to mucous membrane, contact dermatitis, carcinogenic

>When the formaldehyde oxidizes

>Formic Acid – can cause coagulation necrosis (in the optic nerve of the brain – resulting to blindness)

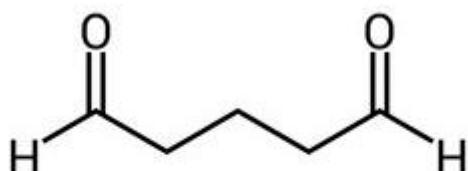


Aldehyde (Glutaraldehyde)

>Pentanedral

>Aldehyde with 2 aldehydes and 5 carbons.

- Cidex, a 5-Carbon dialdehyde
- Used as sterilant for equipment and instrument
- Stabilized in Alkaline Solution

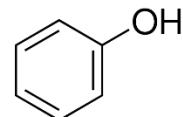


Glutaraldehyde



Phenols and their Derivatives

- Phenol, USP, remains the standard to which the activity of most germicidal substances is compared.



>Serves as the standard of comparison as a germicide (used by Joseph Lister).

- Phenolic Coefficient

>Used to be able to assess the activity of the other derivatives of phenol, this is used.

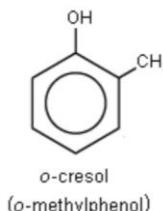
→ the ratio of a dilution of a given test disinfectant to the dilution of phenol that is required to kill a strain of salmonella typhi.

>At time and also temperature conditions.

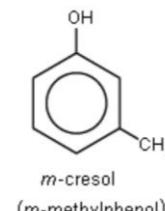
>The phenol coefficient of phenol = 1

>If the PC of derivative of phenol is greater than 1, then it is stronger than phenol (more potent).

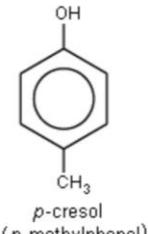
- Substitution with alkyl, aryl, and halogen (especially in the para position) groups INCREASES bactericidal activity.
- Straight-chain (has greater activity) > Branched (lesser activity – because can alter the forces of attraction)



o-cresol
(*o*-methylphenol)



m-cresol
(*m*-methylphenol)



p-cresol
(*p*-methylphenol)

>Para position has the strongest activity.

- MOA:

→ At low concentration → Protein Denaturation
→ At high concentration → membrane lysis

Phenol

>Prototype – solid

- Carbolic acid, pale pink crystalline with “medicinal odor”

>General protoplasmic poison (caustic)

>Also used as anesthetics.

>Must be diluted to avoid physio destruction and dermatitis.

- Phenolated calamine lotion - Antipruritic
- Almost obsolete – antiseptic and disinfectant.

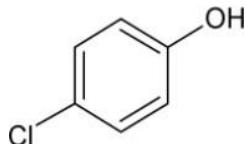
Liquified Phenol

- Phenol with 10% water
- Can be measured and transferred easily
- Not miscible with lipophilic ointment bases.

>The benefits of making phenol in a liquid form: easier of transferring, convenient for adding in pharmaceutical preparation as well.

p-Chlorophenol

- used in combination with camphor in liquid petrolatum as an external antiseptic and anti-irritant.



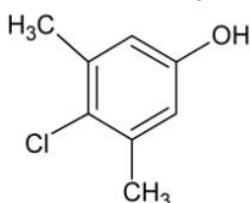
>Phenol Coefficient = 4

>Greater activity since para is the position of chlorine.

p-Chloro-m-Xylenol

>There are branching meaning greater activity.

- Metasep, 2% shampoo
- Treatment of Tinea (ringworm) infections
 - >athlete's foot, tinea pedis, jock itch, and grubis.



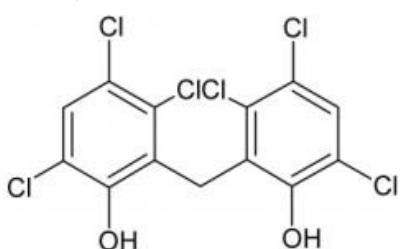
Hexachlorophene

- Gamophen; Surgicon; pHisoHex (marketed under this brand name)

>Can penetrate onto the skin and enter the sebaceous gland, elicit a prolong antiseptic even at low concentration.

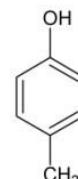
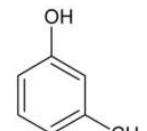
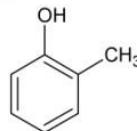
>Prescribed – Rx because high risk of toxicity.

- Has greater potency due to:
 - 2 phenol rings (a biphenol/bisphenol)
 - Chlorines (increasing the activity)
- 2-3% in soaps, creams, lotions, shampoo



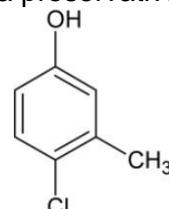
Cresol

- A mixture of three isomeric methylphenols
 - >substituted at every position (ortho, meta, para)
- Obtained from coal tar, inexpensive but has unpleasant odor – antiseptic



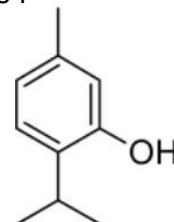
Chlorocresol

- 4-chloro-3-methylphenol
- Useful only as a preservative.



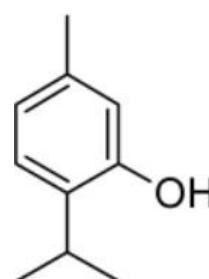
Thymol

- Isopropyl m-cresol
- From oil of Thymus Vulgaris (Thyme)
- Has mild fungicidal properties
 - >used in tinea as well.
- Used in dusting powders for tinea infections



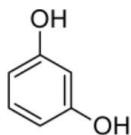
Eugenol

- 4-allyl-2-methoxyphenol
 - >Allyl means carbon chain containing adjacent carbon that has double bond
- From clove oil (*Syzygium aromaticum*)
- Local anesthetic and antiseptic
- Toothache drops and mouthwashes



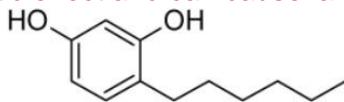
Resorcinol

- M-dihydroxybenzene (resorcin)
- Problem: Light-sensitive and oxidizes readily
 - >must be stored in tight and light resistant containers.
 - >**Phenolic Coefficient = 0.4 (Weak)**
- Antiseptic and Keratolytic (causes stratum corneum to slough)
 - >**the advantage of sloughing, the parts affected by microorganism is open**
 - >Sometimes, resorcinol is combined with other fungal agent.



Hexylresorcinol

- Sensation of numbness to the tongue
 - >**because of local anesthetic effect.**
- Antiseptic (bactericidal, fungicidal)
- Local anesthetic
- Formulated in throat lozenges
 - >**there are studies saying that it is not okay to use it, because when administered as lozenge, administered via mouth, it will not carry out anesthetic effect and can cause laryngitis.**



>Strepsils Max

Oxidizing Agents

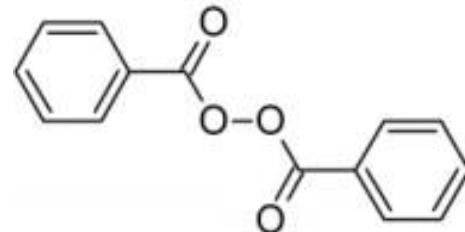
- MOA: Active liberation of oxygen in the tissues
- Effective against Anaerobic Bacteria.
- Bubbles dislodge debris
 - >**effective if they are bubbling.**
 - >**The effectiveness of oxidizing agent is somewhat limited by their generally poor penetrability to infected tissue and organic matter and also transient effect.**
- Examples: Hydrogen peroxide and potassium permanganate
- Limitation:
 - Poor Penetrability
 - Transient effect

Carbamide Peroxide

- Stable complex of urea and hydrogen peroxide
 - >**when mixed with water, hydrogen peroxide is being liberated.**
- Add water → H₂O₂
- Treatment of oral ulcerations or in dental care

Hydrous Benzoyl Peroxide

- In pure powder form → Explosive
- + 30% water to make it safer to handle
- Uses: Keratolytic and keratogenic
 - >**This is being compounded at 5% and 10% concentrations to be able to it to become keratolytic agent.**
- Induces proliferation of epithelial cells
- Treatment of Acne



Halogen-containing Compounds

Iodine-Containing Compounds

>**Also called as Iodophores**

- Elemental Iodine is the oldest germicide still in use today
 - >**very effective germicide.**
- MOA: Iodination of aromatic residues → inactivation of proteins
- Strong Iodine solutions – 5% iodine in water → Hyperthyroidism
- Iodine solutions – 5% iodine in water → Treatment of infections (cuts)
- Iodine Tincture – 2% iodine in 50% alcohol → For minor wounds
- KI or NaI is added to:
 - increase solubility of iodine
 - reduce its volatility

>**When Solubilizing agent is added to iodine, it becomes iodophores.**

Povidone Iodine (Betadine)

- Iodophors – active complexes of iodine with reduced volatility and irritating property
- Example:
 - Povidone iodine
 - iodine + PVP (polyvinylpyrrolidone)
 - >**non-ionic surfactant that would aid solubility of iodine.**
 - non-toxic,
 - non-irritating,
 - non-staining,
 - non-volatile

Chlorine-Containing

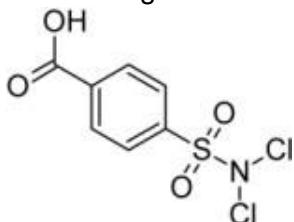
- Used in water disinfection
- Example: NaOCl and Ca(OCl)2
- MOA:
 - (1) Chlorination of amides
 - (2) Oxidation of –SH in proteins
>which will render them ineffective
- Optimal effect at pH 7.0

Halazone

>p-Dichlorosulfamoylbenzoic Acid

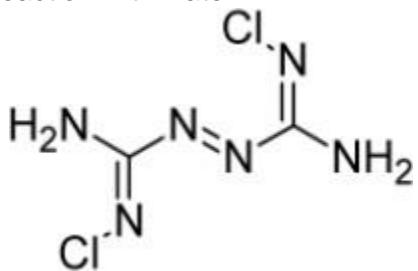
>Sulfamoyl – atoms adjacent to nitrogen is oxygen but instead, chlorine is seen.

- Problem: Photosensitive – dark place, and light resistant.
- Very soluble in alkaline solution
- Disinfection of drinking water



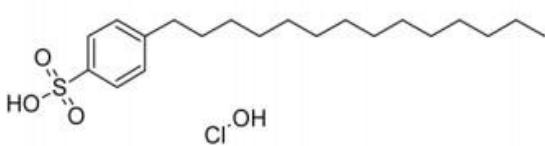
Chloroazodin

- Water-insoluble, unstable in light or heat
- Wound Dressing, for lavage and irrigation
>(Glyceryl Triacetate Solution)
- Antiseptic effect is: Long lasting effect due to slow reaction with water



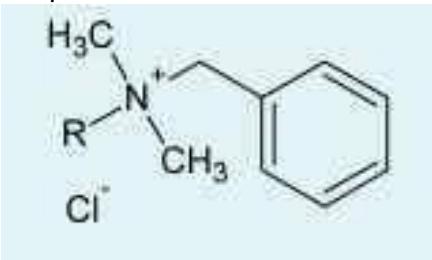
Oxychlorosene Sodium

- Slowly release HOCl (Hypochlorous acid) when exposed in solution
- Rapid -cidal effect against most microorganism, including gram positive and gram negative, molds, yeasts, and microphones.
- Treat localized infections
- Remove necrotic tissues
- Counteract odoriferous discharge



Cationic Surfactants

- Quaternary Ammonium Compounds
>always ionized when exposed in water, exhibit surface active properties.
- Has surface-active properties due to Polar head and non-polar tail



>Exert bactericidal action against broad spectrum of gram positive and negative – also active against fungi and also protozoa.

- Resistant to spores
- MOA: Dissolution of cell membrane → Destabilization → Lysis (cause immediate death)
- Advantages:
 - Highly water-soluble
 - Non-toxic
 - Stable
 - Non-staining
 - Non-corrosive
 - Good penetration
 - Germicides
- Disadvantages:
 - Inactivated by soaps and anionic detergent
>that is why traces of soap must be remove in the skin and other surfaces before they are applied
 - Tissues debris, blood, serum reduce activity, puss
 - Adsorbed on glass, talc, and also kaolin
>to reduce or prevent their action
 - Slower action than iodine

>Cannot be re-used (equipment that has cationic surfactants cannot be re-used) – disinfecting purposes

>it would serve as reservoir for infectious microorganism particularly pseudomonas and enterobacter species.

Benzalkonium Chloride

>The new active ingredient of gamot na pula (disinfectant before applying manicure in the nails) – old API is Merthiolate.

>Merthiolate was changed because it contains mercury which is toxic.

- Antiseptic for skin and mucous membrane at concentration 1:750 to 1:20000

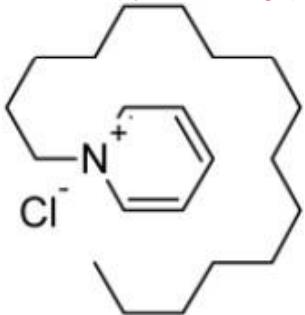
Methylbenzethonium Chloride

- Tx of diaper rash (C. Albicans) → NH₃

Benzethonium Chloride

Cetylpyridinium Chloride

- Q. Nitrogen is replaced with aromatic PYRIDINE RING
- Most active in a series of ALKYL PYRIDINIUM COMPOUNDS
>There is aromatic pyridine ring (quaternary)



Chlorhexidine Gluconate

- Most active of a series of antibacterial biguanides
- Technically NOT a quaternary ammonium compounds
- Similar properties to cationic surfactants

Dyes

>Were used extensively as an anti-infectives before the discovery of sulfonamides and antibiotics – however few are being used nowadays (Commonly used are the following example)

- Colorless leucobase in alkaline condition
- Cationic dyes are effective for Gram + bacteria, and also on fungi

>Gram – bacteria are resistant.

Gentian Violet

>Chemically called as hexamethyl-p-rosaniline or methyl rosaniline chloride.

- Crystal violet, methyl violet
- Vaginal suppositories for yeast infections
- Orally as anthelmintic

Basic Fuchsin

- A component of carbol-fuschin solution (Castellani's Paint)
- Anti-fungal, Topical – tx of ringworm (athlete's foot)

Methylene Blue

- Treatment of cystitis and urethritis
>problem in urinary tract
- Bacteriostatic
>inhibiting growth
- Blue green urine and stool as its adverse effects.

Mercurial

>Effective bacteriostatic agents.

3 types of Mercurial Agent:

- (1) Elemental (Incorporated in ointment bases, used in local infections, and also syphilis)
- (2) Inorganic Mercury
- (3) Organic Mercurial

Inorganic Mercuric Salts

- Used as antiseptics or Disinfectants
 - Mercuric chloride (Corrosive Sublimate)
 - Mercurous chloride (Calomel)
 - Ammoniated mercury
 - Mercuric oxide
- Limitations
 - low water solubility
 - irritating
 - hypersensitivity reactions (No longer used nowadays).
- Organomercurials (Commonly used)

>Could be used as antiseptics and also preservatives, and also diuretics

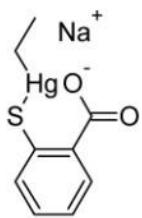
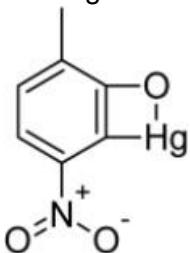
- Non-Ionizable (Hg-C) – one carbon bond.
- Ionizable (Hg bonded to O, N, S)
- More lipid soluble, bonded hetero atom
- MOA: Covalent bonding with SH in proteins
>rendering them non-functional.
- Reversed by (Antidote when poisoned with mercury):
 - Cysteine
 - >high level exposure to mercury, Penicillamine is used for low level.
 - Dimercaprol (BAL) British Anti-Lewisite
 - >Most useful antidote is Sodium Formaldehyde Sulfoxinate

>Mercurials are toxic that its risk could be outweighed by benefits, that is why they are not used most of the time

- Bacteriostatic EFFECT
- Inactivated by serum
- Not very effective on spores

Nitromersol (Methapen)&Thimerosal (Merthiolate)

- Non-irritating to mucous membrane
- Non-staining

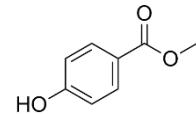


>OH is being released after hydrolysis that could easily undergo conjugation reaction, thus easily excreted.

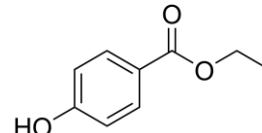
>Once this is being esterified with certain alkyl groups we can have parabens.

Different Parabens and their structures

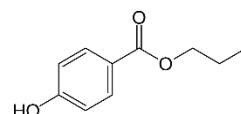
- Methyl paraben



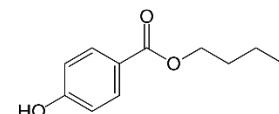
- Ethyl paraben



- Propyl paraben

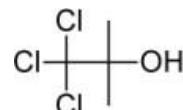


- Butyl paraben



Miscellaneous Preservatives

Chlorobutanol



- Bacteriostatic
- Unstable in heat, pH>7
- Used in ophthalmic preparations

Benzyl Alcohol

- Unesterified form in Oil of jasmin,
- Esterified form in gum benzoin, storax resin, Peru balsam, tolu balsam
- Commonly used in injectable vials (1-4%)
- Local anesthetic

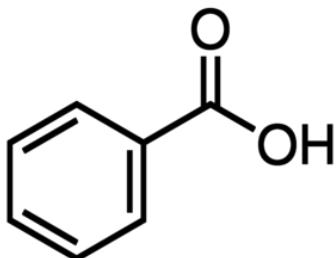
>Component of BWFI (Bacteriostatic water for injection) – which is a type of water

>It can cause gasping syndrome in neonate, that is why BWFI not used for neonates

Phenylethyl Alcohol

- Orange oil, rose oil, pineapple oil
- Used in perfumery

Benzoic Acid



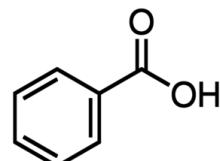
- Gum benzoin, Peru balsam, tolu balsam

- White crystalline solid

- Slightly soluble in water

- Antiseptic

- Preservative at low pH



Sodium Benzoate

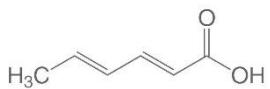
- Preservative in acidic pH (low pH)
→ benzoic acid is released

Sodium Propionate

- Most effective at low pH

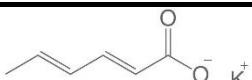
Sorbic Acid

- 2,4-Hexadienoic Acid
- Used in syrups, elixirs, ointments and lotions



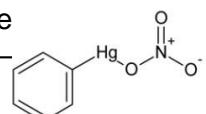
Potassium Sorbate

- Same as sorbic acid but more water soluble

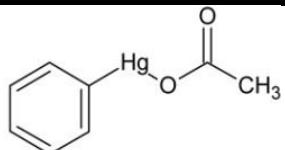


PhenylMercuric Nitrate

- Efficacy reduced in the presence of serum preservative



Phenyl Mercuric Acetate



ANTIFUNGAL AGENTS

Overview of Fungi Key Concepts

- The fungal kingdom (Mycenae Kingdom) includes yeasts, molds, rusts, and mushrooms.
- Most fungi are saprophytic, some are opportunistic, some are obligate parasites (dermatophytes)

>Saprophytic – they utilize decaying organic matter as their sources of energies (Decaying woods, or in soil)

>Opportunistic – usually majority of fungi do not cause disease, some are beneficial to us, but there are cases that fungi become pathogen (causes diseases), for patients with weak immune system (immunocompromise), they are vulnerable to diseases/infection caused by fungi.

>Patients with weak immune system – (1) Those patients with existing infection (HIV/AIDS), potential ultimate cause are going chemotherapy, >(2) Patients going chemotherapy because they are taking drugs that weaken their immune system

>Obligate Parasites – for their survival they need host (cells or organism), relative to our body an example parasite is dermatophytes.

- The field of science that focuses on the study of fungi is mycology

>Common classes of fungi with corresponding diseases/infection that they can elicit in body

Dermatophytes

>As the term suggest, dermat-, related in the integumentary system.

>Includes the fungi such as – Trichophyton, Epidermophyton, Microsporum.

>They are obligate parasites because they need to house in the skin because the nutrient that they need is keratin.

- fungi causing infections of skin, hair, and nails.
- Dermatophytes obtain nutrients by attacking the cross-linked structural protein keratin.
- Dermatophytic infections is also known as Dermatophytosis (Plural – Dermatophytoses) – another related term based on dermatophytosis, AKA Tinea Infection.

Molds

- Most common is from Aspergillus Species.
>Aspergillosis is caused by aspergillus species.
- Route
→ Inhalational (Target organ of aspergillus is lungs, lung infection),

- Ingestion (Target organ is liver, causing liver cancer)

>Aflatoxin – came from mushroom that came from aspergillus.

Yeasts

- most common fungus that is yeast that can cause fungal infection is Candida Albicans.
- Candida Albicans is part of the normal flora (oropharynx, gastrointestinal tract, vagina, and surrounding skin)
 - Principal cause of vaginal yeast infections
 - Oral yeast infections (thrush) – singaw.

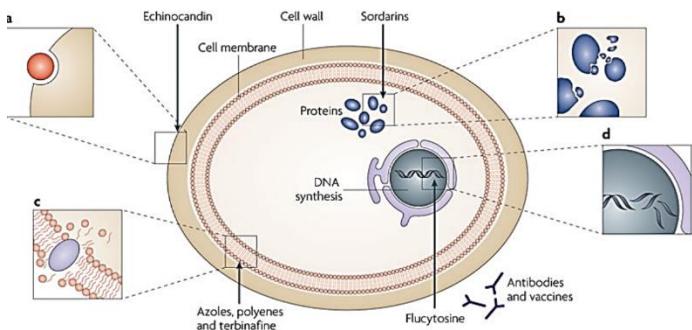
>Even though Candida Albicans do not cause disease, in some individual who is immunocompromise, CA could become pathogenic or disease causing – example: for females it can cause vaginal yeast infection if the patient is immunocompromised.

Summary

- The reason why there is majority of fungal agents as topical is because dermatophytes is the most common fungal infection causing.
- There are fungal microorganisms that can attack our internal organs, that is why we have systemically administered, particularly to immunocompromised patients.

Biochemical Targets for Antifungal Chemotherapy

>Drug target is important since it will determine if the action is toxic or therapeutic of antifungal agents.



Cell Membrane of Fungi

- The Cell Membrane is very important in every cell (Provides partition from one cell to another), and it also facilitates different nutrients.
- If the cell membrane of the fungi is targeted, we tend to kill somehow the fungi, that is why the antifungal effect if effective.
- Numerous of commercially available antifungal targets cell membrane of fungi.

Cell Wall of Fungi

- Cell wall is also a potential target, some antifungal drugs targets cell wall.

DNA Synthesis

- DNA Synthesis is the pre-requisite in the formation of mRNA and eventually it is essential in the synthesis of protein.
- In short, if the DNA production is targeted, the protein synthesis will be inhibited in the fungi, no proteins, fungi will die.

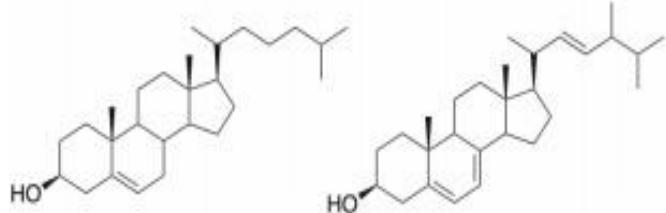
Mitosis

- Mitotic Division (Cell Division) are blocked so the multiplying or synthesis for cloning of fungi is therefore inhibited as well.

Intermediary Metabolism

- In the synthesis of certain compounds or substances in the fungal cell.

Cell Membrane of Fungi



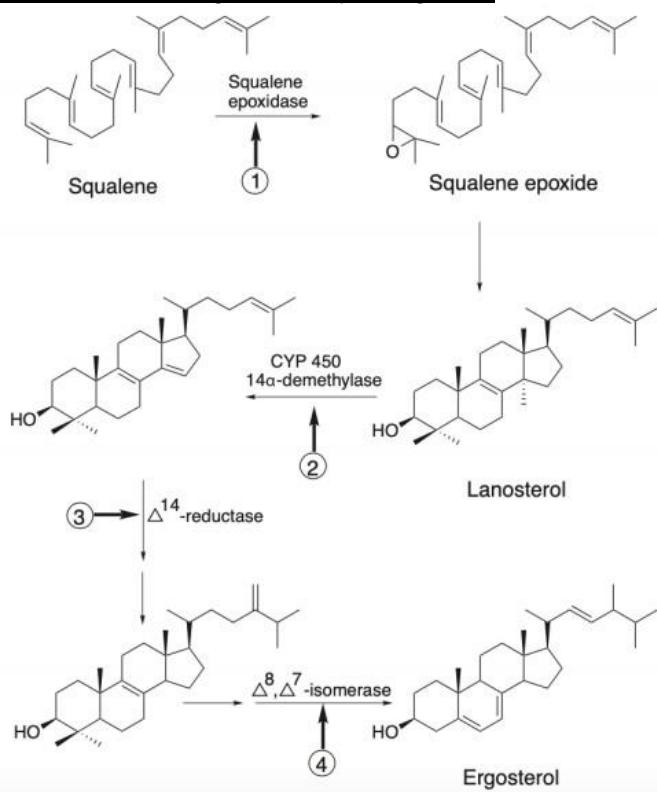
Cholesterol

Ergosterol

- The challenge on targeting the cell membrane (before) is quite risky, because mammalian cells also have a cell membrane, so selective toxicity can be compromised – In short, the antifungal drugs can target our cells and also the cells of fungi.
- As the deeper studies arises, it was realized that there is different components in the cell membrane of fungi from mammalian cells.
- Importance of Sterols (Steroidal Substances) in the cell membrane – A certain sterols structure has a 4 cyclic ring structure, sterols are important because they (1) provide stability of cell membrane, and also (2) transport of substances because of stability of the transport proteins, (3) Aids also on activities of enzymes in facilitating different chemical reactions.
- So, if the sterol is targeted, the fungi can be killed, and according to studies, the sterols of animal cell and fungi are different.

- In fungi cell, Ergosterol is present while in the Mammalian cell, Cholesterol is the present sterol – this is a big thing to be able to establish selective toxicity
- why? Because when searching for an antifungal drug, we will develop antifungal agent and that should target ergosterol only.
- The difference in the side chain of the ring B, major difference is the presence of double bond and also the methyl group in the side chain.
- In creating antifungal drug, ergosterol should be targeted.

Production of Ergosterol by Fungi Cell



- The reason why it is important to know how ergosterol is being synthesized, is because there are numerous agents can target this pathway
- There are 2 important reaction focused on this synthesis of Ergosterol, because these 2 reactions will be targeted by drugs: (1) via the enzyme Squalene Epoxidase – is responsible for the epoxidation of squalene (not yet cyclic hydrocarbons – not yet no structure yet), epoxidation of squalene is very important for the sterol to be synthesized (results in squalene epoxide).
- The squalene epoxide eventually will turn into Lanosterol (First sterol produced), when lanosterol is produced, demethylation is the next step, with the help of the enzyme CYP 450 14d-

demethylase (Oxidative enzyme classified under CYP 450), the 14th carbon (below the long side chain) is removed.

- The 14d-demethylase is also known as CYP51
- Lanosterol will undergo oxidative reactions that would eventually lead in the formation of ergosterol.
- Summary: Two important step is
 - (1) The conversion of squalene-to-squalene epoxide (epoxidation) eventually leading to formation of lanosterol,
 - (2) Demethylation by CYP51 to lanosterol (Oxidation) leading to formation after series of Oxidation process into Ergosterol.

ANTIFUNGAL AGENTS

Topical Antifungals

- Superficial or Topical Antifungal agents are the ones that can be used for fungal affection affecting the skin.
- Since it is for the skin, it is used for tinea infections.

Topical Agents for Dermatophytes

- Directly applied to affected surface areas of the skin

>however, one of the most challenging of the routes of administration of antifungals are in the skin – because when applied in skin, the skin serves as a barrier itself that is why the drug will have a hard time to permeate or to be absorbed.

>In order to surpass the effect of being less permeable in the skin, the antifungal agent is combined with other drugs to enhance the penetration of antifungal which is called **Keratolytic agents**
- Skin: formidable barrier of the skin
 - Topical agents work best if an adjuvant (Keratolytic agents) is added.

Fatty Acids

>Fatty acid is a carboxylic acid wherein the RCOOH the R group is composed of Long chain Hydrocarbons (LC HCs the portion of fatty acids that causes its lipophilicity).

>Fatty Acids MOA: Easily permeate in the cell membrane that would eventually enhance the fluidity of cell membrane, thus causing destruction of the cell membrane

Sebum (Endogenous) (on the skin)

- acidic, fatty substance in and on the skin and functions as a natural antifungal agent

>Why is fatty acids used in the antifungal agent, it is because our skin is also lipophilic, that is why oil based will be spread in our skin for easier absorption or relatively faster absorption.

Propionic Acid

- Clear, Corrosive liquid with a characteristic odor.

Zinc Propionate

- salt form, used as fungicide, particularly on adhesive tape (to prolong contact time), thus greater antifungal activity

Sodium Caprylate

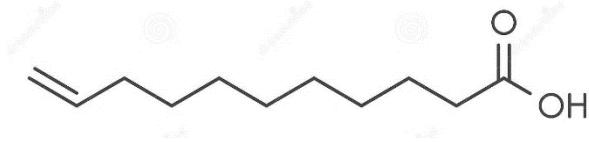
- Prepared from caprylic acid, which is a component of coconut and palm oils.

Zinc Caprylate

- Highly unstable to moisture (Reminder: The skin should be dry when applied)

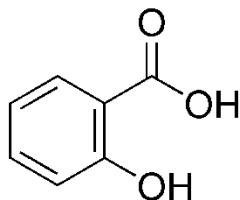
Undecylenic Acid

- Obtained from castor oil (made up of fatty acids) and traditionally used for athlete's foot (Tinea Infection). There is the presence of long chain fatty acids.



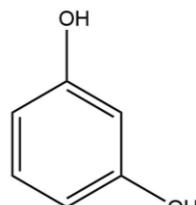
Salicylic Acid

- used externally in ointments and solutions for its antifungal and keratolytic properties.
- >(These is its two effects) – can be used alone as antifungal topical. There are instances where salicylic acid is combined with other drugs – because of its keratolytic effect. (can also be used in anti-acne drugs)



Resorcinol

- m-Hydroxyphenol
- with antiseptic and keratolytic activity.



Benzoic Acid

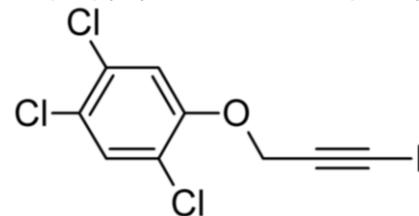
- Possesses antifungal effects but it cannot penetrate the outer layer of the skin in infected areas. ad
→ mixed with keratolytic agent
- It is used in Whitfield's Ointment, USP
→ Benzoic Acid 6%, Salicylic Acid 6%, in a petrolatum base because it is a hydrocarbon base – employ the distribution of correct mixed because they are both lipophilic

Phenols and Their Derivatives

- Clioquinol and Halopropgin (still official in the USP)
- Interfere with cell membrane integrity and function

Halopropgin

- 3-Iodo-2-propynyl-2,4,5-trichlorophenyl ether



- high cure rate, and is expensive.

Clioquinol (Vioform ®)

- initially used as a substitute for iodoform.

Ciclopirox Olamine

- AOC (Agent of Choice) for cutaneous candidiasis, tinea corporis, tinea cruris, tinea pedis, and tinea versicolor.
- 2nd-line agent for the treatment of onychomycosis (ringworm of the nails).

Flucytosine

- A nucleoside antifungal
- metabolized to 5- Fluorouracil (ACTIVE) which blocks DNA synthesis

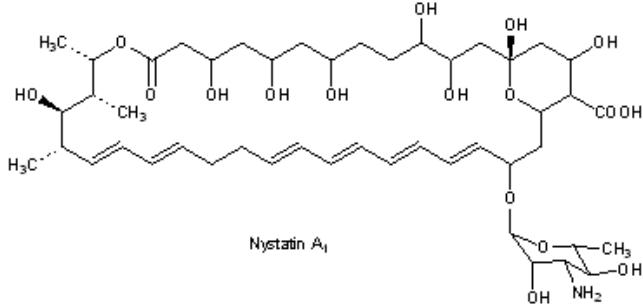
Antifungals Antibiotics

- Like dissolves like (applicable in anti-acne drugs)
– there is a higher chance of getting acne when you washed your face using water because sebum is lipophilic and water is hydrophilic, wash first with lipophilic agent.
- Antibiotics – came from microorganism that are used to kill microbes.
- Two specific example of Antifungal Antibiotics – Polyenes and Griseofulvin.

Polyenes

>Called poly-enes because of multiple double bond carbon to carbon seen in the structure.

- Isolated from soil bacteria (Streptomyces)
- Conjugated -ene system of C=C in large macrocyclic lactone rings
 - Natamycin (26- membered)
 - Amphotericin B and nystatin (38-membered)



>Nystatin Structure – All polyenes have similar structure that has a lactone ring and multiple double bond carbon.

>The reason behind the membered number is the lactone ring of the natamycin and amphotericin B and also nystatin has 26 and 38 carbon atoms respectively.

>There are 2 major regions observed in the structure of polyenes (Hydrophilic and Lipophilic) - (1) Hydrophilic Regions (because of FG alcohol), (2) Presence of Carboxyl Group (RCOOH) (Hydrophilic Group), (3) Sugar Portion – Mycosamine is the reason of being hydrophilic as well.

>Mycosamine is glycosidically linked with the lactone ring structure – glycosidically linked is based on glycoside structure (Glycoside is sugar ether) and ROR connects the lactone to glycosamine. (First R is the sugar portion and the second R is the non-sugar portion). – glycane portion is the sugar portion.

>Lipophilic region is the hydrocarbons (especially C=C).

>This lipophilic region is responsible for the (1) antifungal (targets cell membrane) activity of polyenes.

>(2) The higher the number of double bonds, the greater the antifungal effect, the lesser the toxicity. (high DB=high AF=low Toxicity).

>The reason why it is not toxic in humans because the higher the number of double bond is the more higher chance of targeting ergosterol rather than cholesterol.

Polyenes Mechanism of Action

>Polyene targets cell membrane.

>Polyenes are like “barena” wherein they force their way in the cell membrane and then it goes there, causes: (Refer to the 3 dots located below)

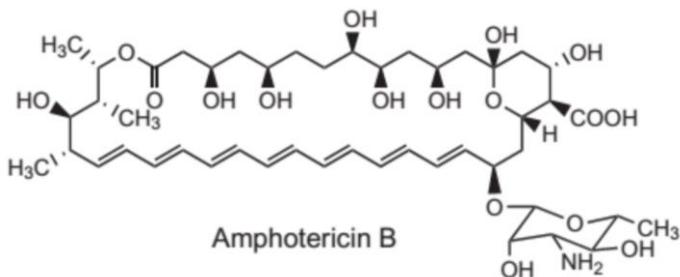
- Act as “false membrane components” and bind to ERGOSTEROL (essential sterol found in the cell membrane of fungi), causing
 - membrane disruption (is important for exchange of compounds)
 - cessation of membrane enzyme activity
 - loss of cellular components (K⁺ leakage)
- Highly potent, broad-spectrum of actions (It is still toxic)
- Effective against yeasts, molds and dermatophytes (Polyenes has a systemically administered and topically)
- NOT ACTIVE against bacteria, rickettsia or viruses (FUNGI ONLY)
- Use for systemic infections (powerful agents) but limited by
 - toxicities (highly)
 - low water solubility (despite the presence of hydrophilic region)
 - poor chemical stability (limited nalang ang application ng polyenes)
- AMPHOTERICIN B – only useful polyene for systemic infections, because of the structure of Amphotericin B (relatively safe compared to other polyenes), but is still toxic.

Amphotericin B

- isolated from Streptomyces nodosus
- Compound B is more active than A – It has the greater activity among A and B amphotericin.
 - Carries the highest number of double bonds compared to the two other polyenes.
 - The reason why it is called amphotericin is because it is an amphoteric substance.
 - Acts as a false membrane which enters the cell membrane of fungi causing disruption.
- MOA: interact with ergosterol forming a TRANSMEMBRANE CHANNEL
 - Also affects cholesterol(NON-SELECTIVE) – toxic by itself
- An amphoteric substance
- Indicate for life-threatening fungal infections (indented for systemic infection)
 - Amphotericin is hard to store and in chemically incompatible, hard to prepare, to be able to enhance its stability, Parenteral: Stabilized with Sodium Deoxycholate.

- It cannot enter the Blood Brain Barrier, even though it is lipophilic it cannot enter BBB even though it is lipophilic, the route of administration is changed instead of IV, it is for CNS infections: Route is Intrathecal
- Toxicity: Nephrotoxicity (nearly 80%) – major toxic effect is damage in the kidney, nearly 80% attributed to kidney damage effect.
- Pain at site of injection and thrombophlebitis → never administer IM – if administered parenterally (like IV), it can result to thrombophlebitis.

Structure of Amphotericin

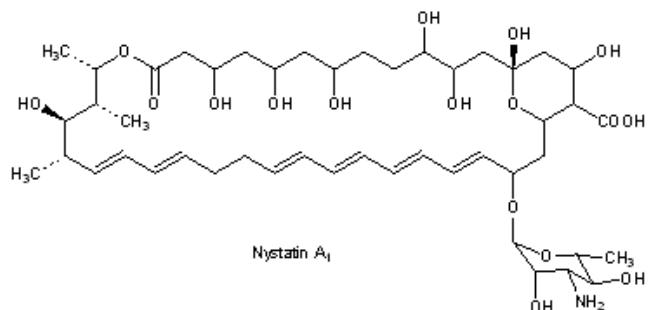


- The effect matters in the lipophilic region.
- It is a heptaene (7 double bonds).
- Amphoteric substance – it means the drug can act as an acid or base, there are functional groups of amphotericin that can act as donor or acceptor – carboxyl groups (acid) donate, while the amino group (base) in mycosamine sugar is the acceptor.

Nystatin

- isolated from *Streptomyces noursei*
- Aglycone portion, which is called nystatinolide.
- Used only as topical agent – limited for dermatophytosis, because much more toxic than amphotericin B.
- Treatment of cutaneous candidiasis
- Treatment of local and GI monilial infections caused by *C. albicans*

>nystatin can be taken by mouth but is not absorbed systemically, if taken by mouth, it is concentrated in the GIT not in the intestine.



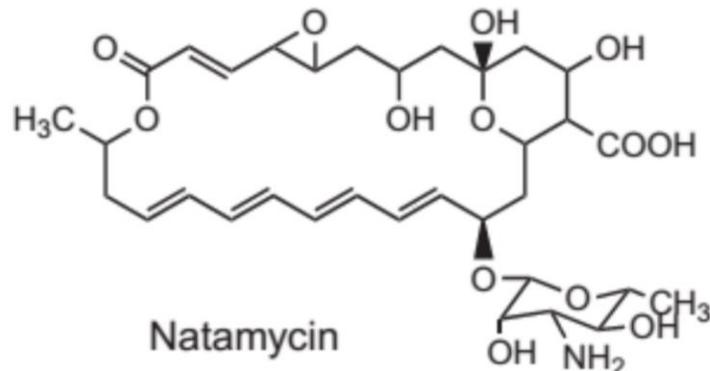
>Less double bond (6 bonds - hexene) compared to amphotericin B, meaning more toxic.

>The double bond is not continuous compared to amphotericin B – divided into 2 double bonds and 4 double bonds (divided in diene and tetraene, that is separated by 2 methylene groups)

Natamycin

>It has 4 double bonds (focus on the lipophilic region) (tetraene)

- Isolated from *Streptomyces natalensis*



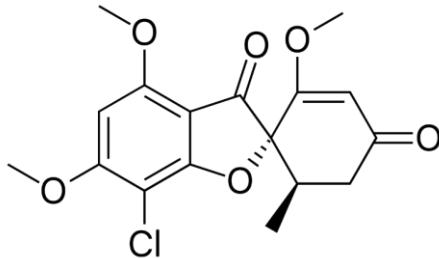
- MOA: - Static and cidal at the same concentration (whether high or low concentration),
 - It is fungistatic because it promotes potassium ion leakage from the cell membrane, fungicidal because it promotes fungal cell lysis.
 - Highly toxic since it has 4 ene bonds.

>Difference of natamycin in amphotericin B and also nystatin, the Amphotericin B and nystatin are dose dependent, meaning different meaning in low and high concentration (For low, static effect and for high, cidal effect).

Griseofulvin

>Is a certain drug, no class.

- isolated from *Penicillium griseofulvum*
- Treatment of refractory (meaning not curable by topical antifungal) ringworm infections of the body, hair, nails and feet caused by TEM
- Dermatophytic infections, known as tinea, are caused by various species of 3 genera: *Trichophyton*, *Microsporum*, and *Epidermophyton*



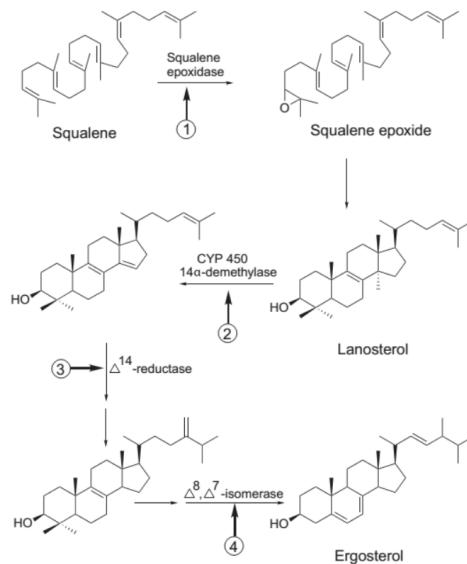
>The structure is named as Spiro compound (which is quite rare)

- MOA: mitotic spindle poison (targets mitosis or cell division)
 - >stops only the multiplication and not literally kill it, that is why it is only fungistatic.
→ Fungistatic
- Not effective against bacteria and *Pityrosporum orbiculare* (tinea versicolor)
- Poor oral bioavailability
→ taken with high fat meal
- Targets refractory cases, because when using topical in antifungal, it only targets the superficial part (not the part of the skin that creates keratin) and that cannot be reached by antifungal topical. Systemic administration (PO) is used for that deeper part, and once it enters systemically, it enters in the capillary beds in the precursor cell which produces keratin, and when that keratin arises again in the superficial layer, it is now treated with griseofulvin
- Infections treated with Griseofulvin:
 - Tinea capitis – hair and scalp
 - Tinea pedis - feet
 - Tinea manuum – hands
 - Tinea cruris – groin (jock itch)
 - Tinea unguium – fingernails

Allylamines and related compounds

Allylamines

- interfere with early step of ergosterol biosynthesis
 - >**Inhibitors of Squalene Epoxidase Enzyme**
→ epoxidation of SQUALENE, catalyzed by squalene epoxidase
- Cholesterol biosynthesis is not affected
- There are 2 possible effects that could happen if squalene epoxidase is blocked (leading to cell membrane destruction) – (1) Squalene levels rises (accumulation of squalene in fungi cell which is toxic in fungi cell), (2) Low production of Lanosterol (when lanosterol depresses meaning the sterol which is important in fungi cell membrane will decrease)

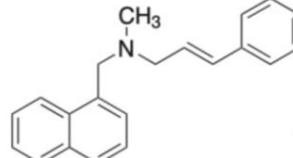


Examples of Allylamines

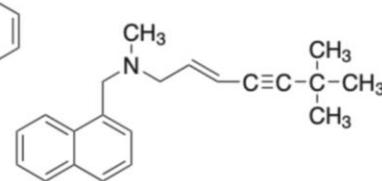
Naftifine – first discovered (topical) – prototype allylamine but is limited for topical administration because if administered via mouth it undergoes extensive first pass effect

Terbinafine – more potent than naftifine

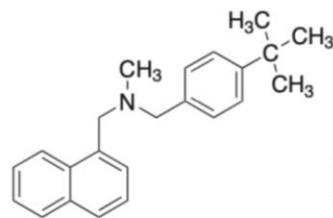
Butenafine, Tolnaftate – not an allylamine but has similar MOA



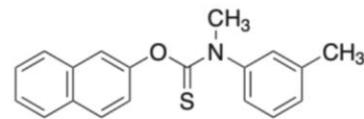
Naftifine



Terbinafine



Butenafine



Tolnaftate

>chemically speaking, base on the structure, they are not allylamine, but they are both squalene epoxidase inhibitors.

>Allylic carbon – carbon bonded to another carbon which interm bonded with double bonded carbon (C Allylic carbon – C = C)

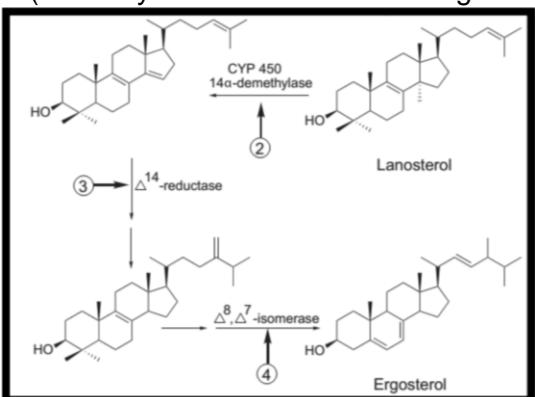
>Allylic carbon has a amino group on its side that is why it is called allylamine.

Azole Antifungals

- Largest class of antifungals – because the number of examples is greatest compared to other commercially available.
- Synthetic antifungal agents
- Effective against fungi that cause infections of the skin and mucous membranes such as Trichophyton, Epidermophyton, Microsporum
- MOA – it has an effect on the ergosterol synthesis
 - high conc (micromolar) – cidal
 - low conc (nanomolar) – static
 - cell membrane damage
 - loss of cell components

At low conc (targets the fungal cell membrane)

→ Inhibition of lanosterol 14 α -demethylase (demethylation of lanosterol to ergosterol)



>Azole antifungal blocks the CYP51 resulting to cell membrane disruption.

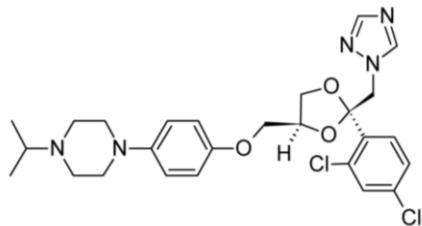
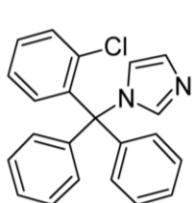
At high conc (TOXIC in human/animal)

- also inhibit mammalian enzyme
- SELECTIVE TOXICITY (Toxic in high concentration and therapeutic in high concentration)

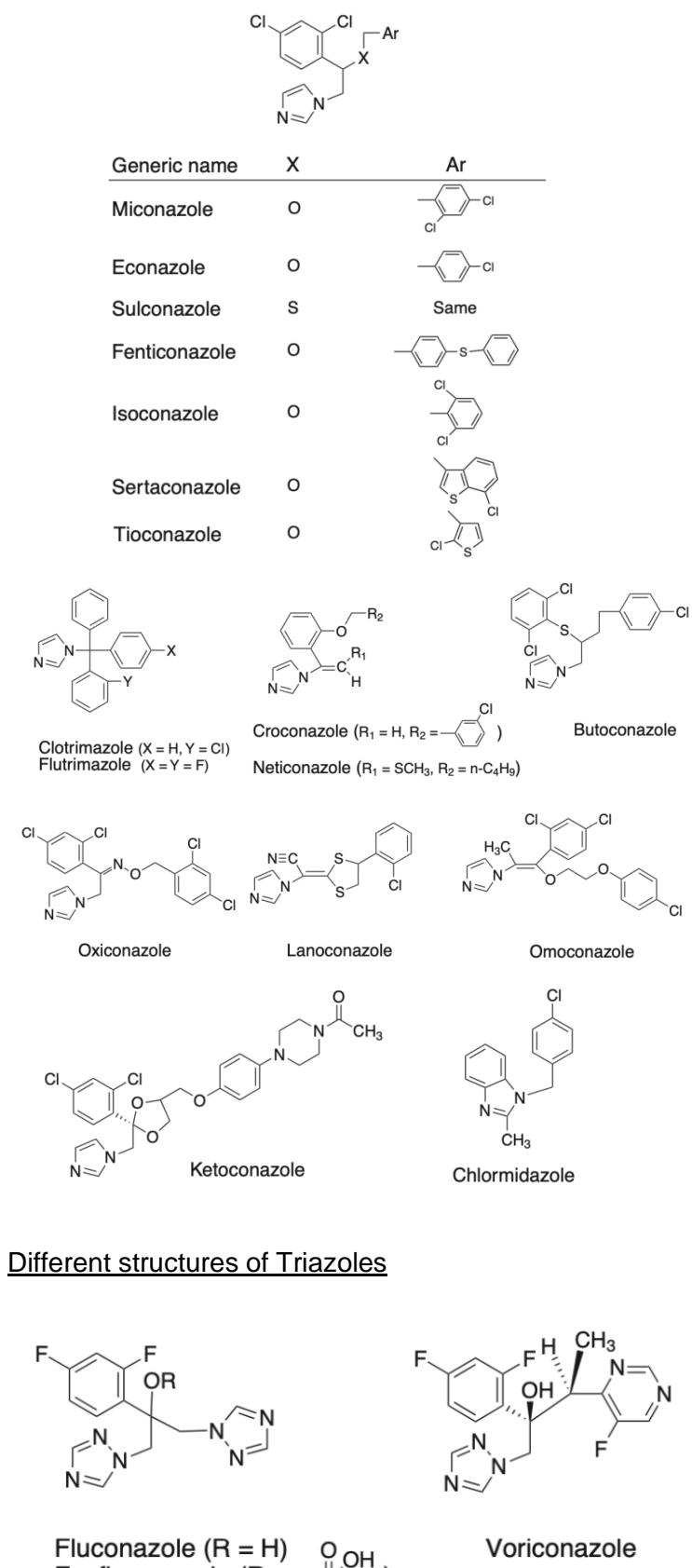
Azole structure

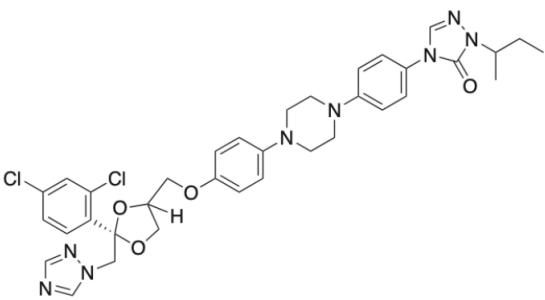
>Azole – 5 membered heterocyclic ring structure containing nitrogen atom

- Presence of five-membered aromatic ring containing 2 or 3 N atoms
- >Those that contain 2 nitrogen on the heterocyclic compound are called – imidazole
- >Those that contain 3 nitrogen on the heterocyclic compound are called – triazoles

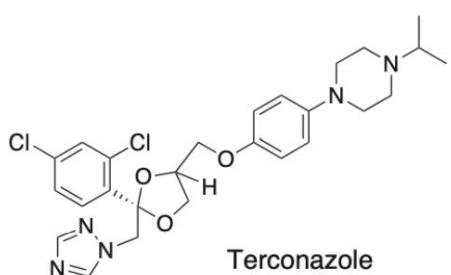


Specific structures of Imidazoles

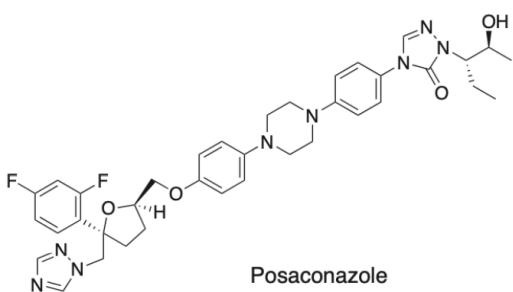




Itraconazole



Terconazole



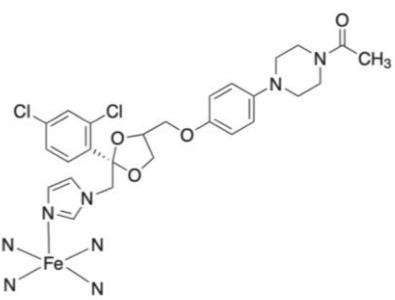
Posaconazole

Examples of Azole Antifungals

1. **Clotrimazole** – not suitable for systemic infection
2. **Tioconazole**
3. **Miconazole**
4. **Ketoconazole** – hepatotoxic, good oral bioavailability at low pH (acidic - intended to be absorbed in the stomach) – not suitable for systemic infection. It has significant drug interaction with antacids preparation (bases meaning they make the stomach acid which reduces the bioavailability of ketoconazole)
5. **Terconazole**
6. **Itraconazole** – more effective than ketoconazole, not hepatotoxic
7. **Fluconazole** – oral absorption is not affected by pH (Solution to ketoconazole)

Physicochemical & Pharmacokinetic Properties

- Topical imidazoles tend to be rapidly metabolized if used systemically, increasing toxicity, and as a result, are not used systemically.
- Topical products are available as creams, powders, gels, solutions, suppositories, lotions, ointments.
- Systemic products are available as tablets, capsules, and powders for oral suspension, injectable solutions.
- Systemic azoles (e.g., ketoconazole – imidazole, and the triazoles: fluconazole, voriconazole, itraconazole, posaconazole, and isavuconazonium sulfate are slowly metabolized).
- Isavuconazonium sulfate is unique among the azoles in that it is a prodrug, which undergoes hydrolysis to isavuconazole the active drug (see metabolism below).
- Isavuconazonium sulfate is water soluble and can be used as such for IV administration (also used orally).
- Other azole parenteral preparations require cyclodextrin for solubility. Cyclodextrin has been associated with nephrotoxicity.
- Ketoconazole and itraconazole require low stomach pH for oral absorption and, as a result, drug–drug interactions associated with administration of an antacid should be anticipated.
- Ketoconazole's action is significantly reduced in the presence of phenytoin, carbamazepine, and rifampin.



14α-Demethylase heme

Structure Activity Relationship (SAR) of Azole Antifungals

>How will Azole bind on the CYP51 (14 alpha-d)
 >Because of the presence of their nitrogen atoms (in heme) that is the reason why they will bind with azole antifungals

- N-3 of Imidazole and N-4 of Triazole
 → Bind to the heme iron of CYP450 = prevents oxidation of steroid substrates
 >N-3 of Imidazole and N-4 of Triazole will directly bind to the oxygen leaving no oxygen remaining in the heme protein (oxygen is important in oxidation)

- Systemic products have good bioavailability (~90%) with high protein binding (~90% with the exception of voriconazole – 56% and fluconazole ~12%). binding (~90% with the exception of voriconazole – 56% and fluconazole ~12%).
- Azoles are substrates and/or inhibitors of P-glycoprotein (P-gp) effecting their own excretion and excretion of P-gp substrates.

Metabolism of Azole

- The systemic azoles are inhibitors and substrates for various CYP450 isoenzymes , although for posaconazole and isavuconazonium sulfate the effects of CYP450 isoforms are minimal.
- Ketoconazole, itraconazole, and voriconazole are extensively metabolized by CYP450 isoforms
- Posaconazole is metabolized to its glucuronide via uridine glucuronyltransferase (UTG), while isavuconazonium sulfate undergoes butylcholinesterase catalyzed hydrolysis to isavuconazole the active drug

Clinical Application

- The topical azoles are generally effective in the treatment of tinea corporis, pedis, cruris, versicolor, and cutaneous candidiasis.
- The newest azoles luliconazole and efinaconazole can effectively penetrate the nail plate to treat onychomycosis (also known as tinea unguis) caused by *Trichophyton rubrum* and *Mentagrophytes*, *Epidermophyton*, and *Microsporum* nail infections.
- Published literature indicates that luliconazole, as a 10% solution, is effective in the treatment of onychomycosis of the nail, although presently not FDA approved for this use.
- Vaginal candidiasis is treatable with the imidazole antifungals available in vaginal creams, suppositories, and tablets.
- Oropharyngeal candidiasis is treatable with clotrimazole troches.
- Systemic azoles are effective against most of the human fungal infections including candida, cryptococcosis, coccidioidomycosis, blastomycosis, sporotrichosis, aspergillosis. Isavuconazole has been granted orphan drug approval for invasive candidiasis, aspergillosis, and mucormycosis.

Adverse Effects of Azole

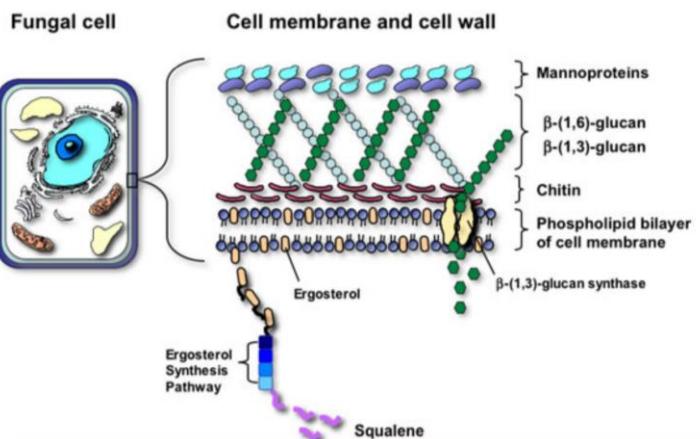
- **Systemic Azoles:** The most common adverse effects involve the GI track and include nausea, vomiting, diarrhea. Rash has been reported with some azoles. Several azoles should be limited during pregnancy (Category C) or contraindicated (Category D – voriconazole). Isavuconazonium sulfate should not be used during pregnancy and serious hepatic reactions also have been reported for this drug

Morpholines

- Amorolfine (Europe and in Asia, not available in US)
 - Inhibits 14-reductase and Delta 8, Delta 7-isomerase

Echinocandins

- large cyclic peptides linked to a long fatty acid from *Glaera Lozoyensis*
- Examples: Caspofungin, Micafungin, Anidulafungin
 - MOA: Inhibit beta-1,3-glucan synthase = inhibit fungal cell wall synthesis



>The importance of beta-1,3- glucan synthase is in the production of beta-1,3-glucan.

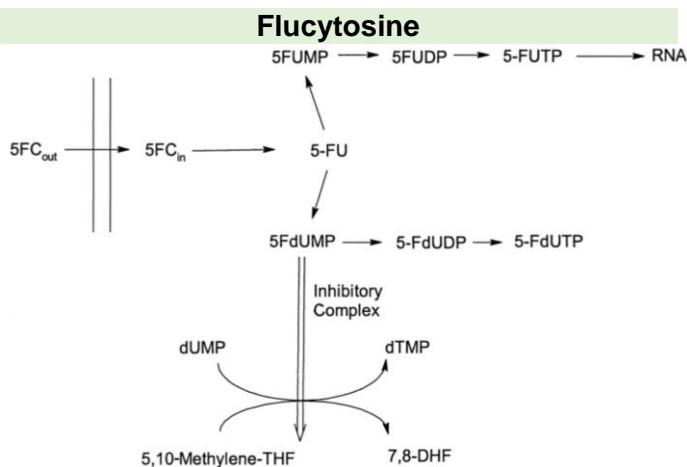
>beta-1,3-glucan is an important component of fungi cell wall.

>When cell wall is destroyed, cell lysis could happen.

Aureobasidins

- There are Aureobasidin A and B.
- A cyclic depsipeptide produced by *Aureobasidium Pullulan*.
- MOA: act as a tight binding noncompetitive inhibitor of the enzyme inositol phosphorylceramide synthase.

- >Blocking this enzyme is important in production of sphingolipid (important part of cell membrane)
- IPC synthase: essential for fungal sphingolipid biosynthesis



- Is an example of prodrug – when it undergoes in the fungal cell, it undergoes metabolism to be activated. Its active form has the ability to facilitate counterfeit mechanism.
- So its molecular action is an example of counterfeit mechanism “fake substance” that would be utilized for the certain metabolic process.
- 5 Flucytosine which is found outside the fungal cell, and say that it entered the cell, this will undergo activation leading to 5-Fluorouracil (which is the active metabolite of flucytosine) – 2 fates.
 - The upper fate is all about the disruption of RNA production/synthesis.
 - Disruption of DNA synthesis in the bottom fate.
- Since Fluorouracil it resembles the structure of uracil (nitrogenous base) – it can join the RNA synthesis (but fake), resulting to formation of faulty RNA (abnormal production of RNA=abnormal protein production=fungistatic effect).
- 5 FU inhibits the enzyme thymidylate synthetase which would lead to inhibition of pyrimidine base synthesis. (Leading to a faulty DNA synthesis)
- Resistance if very common – to be able to delay it, it is combined with amphotericin B (enhance penetration to enter the fungal cell)
- Delayed when combined with Amphotericin B and/or by lowering dose administered

ANTIBACTERIAL ANTIBIOTICS

Introduction

- >bacterial infections have been deemed as health threats even in centuries earlier
- >humans learned to use crude preparations (from natural sources like plants, microorganisms and other natural related products for the treatment of certain infections)

Historical Overview

- Molded curd of soybean to treat boils and carbuncles (in Chinese folk medicine) (500-600BC)
 - >problem before: not scientifically established but it was assumed before that this was effective because of the antibiotic substances present
- 1877- Pasteur and Joubert
 - Anthrax bacilli were killed when grown in culture in the presence of certain bacteria
 - >because of the presence of other bacteria/microorganism, the anthrax bacilli were killed
 - >there were numerous scientists/microbiologists supported that realization/conclusion
- Vuillemin's concept of ANTIBIOSIS
 - >anti – against; bio - life = against life
 - >ANTIBIOSIS – biological concept of survival of the fittest
 - (one organism has to be destroyed/killed for the other organism to win)
 - (one organism destroys another to preserve itself)
 - >other discoveries that had led in the concept of antibiosis were observed:
 - 1909 – Erlich's discovery of Arsphenamine
 - >he was able to apply the concept of chemotherapeutic index (related to selective toxicity)
 - >selective toxicity – the threat is the only target not the normal (safety)
 - 1929 – Fleming's discovery of penicillin
 - >related to the concept of antibiosis
 - >discovery of penicillin by accident (serendipity)
 - >the growth of the bacteria stops because of the growth of the fungus (survival of the fittest)
 - 1935 – Domagk's discovery of sulfa drugs
 - 1938 – Florey and Chain introduced penicillin in therapy
 - Waksman defined “An antibiotic or antibiotic substance is a substance produced by microorganisms (bacteria/fungi), which has the capacity of inhibiting the growth and even of destroying/kill other microorganisms”

>1942 – Waksman was able to propose his cited definition of antibiotic

>inhibiting the growth means that the microorganism has a -static (bacteriostatic/fungistatic) activity depending on the microorganism being targeted by the antibiotic

>destroying means it has -cidal action

>Waksman definition has loophole because other scientists think that his definition is rigid (coverage is focus on the natural sources)

>the problem with natural sources is, it is not sustainable especially if the source is hard to produce or cannot be easily seen

>other scientists said that they must include synthetic sources so that kapag napatunayan na effective yung isang microorganism, ieextract dun kung ano ba yung compound precursor/metabolite na efficacious at gagawan sya ng synthetic precursor or synthetic equivalent

Expanded definition of Antibiotics:

1. It is a product of metabolism (although it may be duplicated or even have been anticipated by chemical synthesis)

>it is important that the organism/metabolite coming from the organism must be sustainable (pang matagalang)

>by identifying the product of metabolism, makukuha yung gagamitin and it will be produced synthetically

2. It is a synthetic product produced as a structural analog of a naturally occurring antibiotic

>after discovering from a natural source, the potentially active/bioactive compound from the microorganism, pwede syang gawan ng synthetic equivalent that can be useful in the long run

3. It antagonizes the growth or survival of one or more species of microorganisms

>inspired by Waksman definition

>growth = static

>survival = -cidal

4. It is effective in low concentrations

>very important to prove this

>to be able to prove that the given antibiotic is effective at low concentration, it must be test in terms of its potency

>the lower the amount, the more potent the potential compound/drug is (\downarrow dose/conc. = significant activity of the given antibiotic-maganda ang result)

>in experiment, pag mataas ang value ng concentration needed to kill a certain pathogen, hindi maganda

>it is related to in vitro test called IC50 (median inhibitory concentration) that is the concentration needed to inhibit the growth and survival of the half of given population (certain strain of the microorganism)

>mababang value mas maganda ang result (mas potent)

>part of the journey of drug discovery is to have mission and vision, and part of the set-up of vision is to have the idea characteristics of an antibiotic

An Ideal Antibiotic

>ideal means gustong makuha pero never makukuha, meron lang parts na makukuha merong hindi

- Selectively toxic

>only those threats are the target

>Paul Erlich's was able to connect the concept of selective toxicity to chemotherapy or chemotherapeutic agents (drugs/pharmacological agents that can kill/inhibit growth of foreign cells in the body)

>foreign cells (bacteria, viruses, fungi, mutated cells) are cells that normally should not be seen in the body otherwise diseases could occur

>chemotherapeutic agents are related to antibiotics

>as long as the antibiotics are pasok sa 4 na given drugs it is called antibiotic

>it is somehow related to anti-infectives (targets the threats)

>the difference is about mutated cells (cancer cells)

>cancer cells are produced in the body but considered foreign because it causes diseases or result to complications

>chemo agents targets cancer cells while anti-infectives targets only yung mga tiga labas

>chemotherapeutic agents included mutated cells that is a foreign cell produced inside the body

>anti-infectives are limited only to foreign in the first place (hindi nagagawa sa katawan)

- Chemically stable

>can be stored for a long time (hindi magastos sa part ng manufacturer)

- Rate of elimination is slow enough for dosing schedule, but rapid and complete enough for removal

>in pharmacokinetics elimination is recombination of metabolism and excretion, so these processes must be slow enough to allow convenient dosing regimen

>when there is convenient dosing regimen, higher compliance, and there is higher rate of success in the therapy

>if the patient is not compliant in antibiotic therapy that could lead to antimicrobial resistance making the drug ineffective already the next time you take it
>it is important na para maging compliant sya, matagal sa katawan ang gamut pero rapid and complete enough for removal of the drug and its metabolites from the body soon after the administration has been discontinued
>when the dosing is stop, mabilis din maaalis pero pag continuous matagal mawawala

Commercial Production

>appreciate the importance of sustainability
>is the effective antibiotic sustainable?
>in order for it to be sustainable, there is commercial production for it to be sell for a long run

1. Preparation of pure culture

>this includes preparation of the culture of the desired organism for use in inoculation of fermentation medium

2. Fermentation

>facilitates fermentation to be able to get the formation of the potential source of antibiotic

3. Isolation

>isolate the antibiotic itself

4. Purification

>in purification, this is important to ensure that the one being extracted is really the one that has the activity

5. Assays

>assays would tell us if the objectives are being attained and to be able to prove that the one that was isolated is really an antibiotic

>potency assay (related to IC₅₀), sterility testing (to ensure that the one that was purified does not contain impurities), absence of pyrogen (fever-causing compounds/substance, this must be absent sa na-isolate and na purify)

6. Formulation

>formulation of suitable dosage form

Review of Bacteria

>understanding on bacterial characteristics will give an idea on the pharmacological actions of drugs
>the success of the antibacterial agents owes much to the fact that they can act selectively against bacterial cells rather than animal cells and by understanding that the clinically available drugs target only bacteria, this concludes that they are selectively toxic
>because bacterial and animal cells differ both in their structure and biosynthetic pathways

Differences between Bacterial and Animal Cells

- The bacterial cell does not have a defined nucleus, whereas the animal cell does;
- Animal cells contain a variety of structures called organelles, whereas the bacterial cell is relatively simple;
- The biochemistry of a bacterial cell differs significantly from that of an animal cell; and

>Example: there are vitamins that bacterial cells need to survive and for them to have a source of that vitamins, they have to produce it on themselves (sila lang gumagawa ng sarili nilang kailangan)

>in animal cells, they can get it from the food (effortless)

>in biochemistry of bacterial cells there are more enzymes that bacterial cells possess needed for the production of certain compounds for survival rather than in animal cell

>if there are more enzymes that will make compounds necessary for survival of the bacteria, the drugs can target many

>and by targeting those enzymes necessary for the survival of the bacteria particularly by targeting it, that would cause inhibition of bacterial growth so there is antibiotic effect

>so it is very important to know the enzymes that the bacteria have that the animals doesn't have dahil yun ang hahanapan ng gamot para maging selectively toxic sya

- The bacterial cell has a cell membrane and a cell wall, whereas the animal cell has only a cell membrane

>cell wall is important in bacteria because it protects the bacteria against lysis whenever there are changes in the osmotic pressure, and other related environmental changes

>so kapag nasira ang cell wall ng isang bacterium, that can cause lysis of the bacterium that eventually causing death

>by knowing the importance of the cell wall in the bacteria, we can think that to be able to kill the bacteria, we can target the cell wall

>and by targeting such, there is lysis

>there are clinically important antibacterials that has a mechanism of inhibiting the production of cell wall for the bacteria to die

>to choose a drug that is selectively toxic

Mechanisms of Antibacterial Action

Mechanism of action

- The MOA of antibiotics varies
- Understanding the MOA is the basis for future development of antibacterial antibiotics

1. Inhibition of cell wall synthesis

>by blocking the production of cell wall that causes lysis (cell death) so that can be a potential target
>by targeting cell wall, this makes us conclude that the given agent displays selective toxicity (because cell wall is only present in the bacteria) (toxic sa pathogen, sa human cells hindi = safe)

2. Disruption of protein synthesis

>nina-alter ng isang given antibiotic drug ang translation process (producing polypeptides with certain function)
>so, by altering polypeptide formation/translation process, mapipigalan ang growth ng microorganism or the bacteria

3. Inhibition of nucleic acid synthesis

>nucleic acids (DNA and RNA)

>the goal of such medication is to block the production of DNA and RNA,
>by blocking DNA production, we are altering replication of DNA and when RNA, transcription
>Pag na block ang DNA replication, the RNA production will be affected too, in which the mRNA will be stop
>if the production of mRNA is inhibited, damay na din ang translation

4. Inhibition of cell metabolism

>related to antimetabolites, in which they target the enzymes that are exclusively present in the bacteria but not in the animal cell
>display selective toxicity because they only target those in bacteria

5. Interactions with the plasma membrane

>some antibacterial agents interact with the plasma membrane of the bacterial cells to affect membrane permeability that is also fatal to the cell of the bacteria

>so with these mechanisms, we could think of that antibacterial agents could either be -cidal or -static
>examples: inhibitors of cell wall automatic patay (cidal due to lysis)

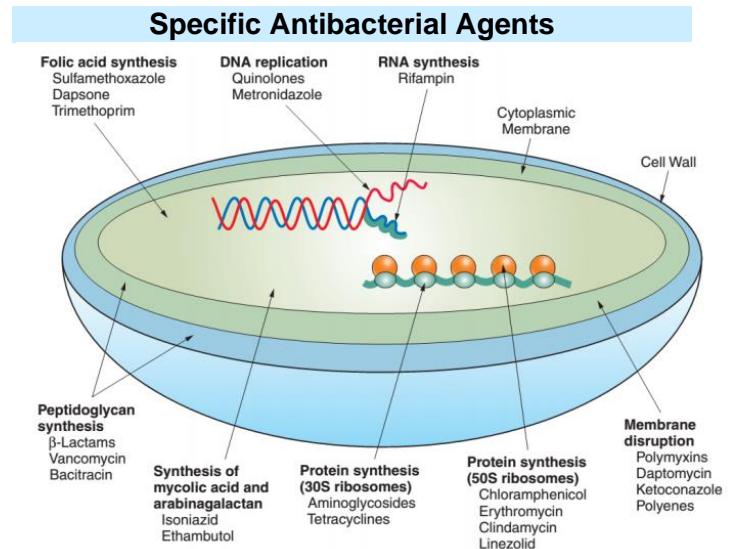
>in protein synthesis it depends, majority of the protein inhibitors is just -static agents except aminoglycosides which are -cidal agents

>nucleic acid inhibitors are -cidals

>antimetabolites are usually -static

>alternants of plasma membrane activity are -cidals

Antimicrobial drugs can be either bacteriostatic (inhibit growth) or bactericidal (kill)

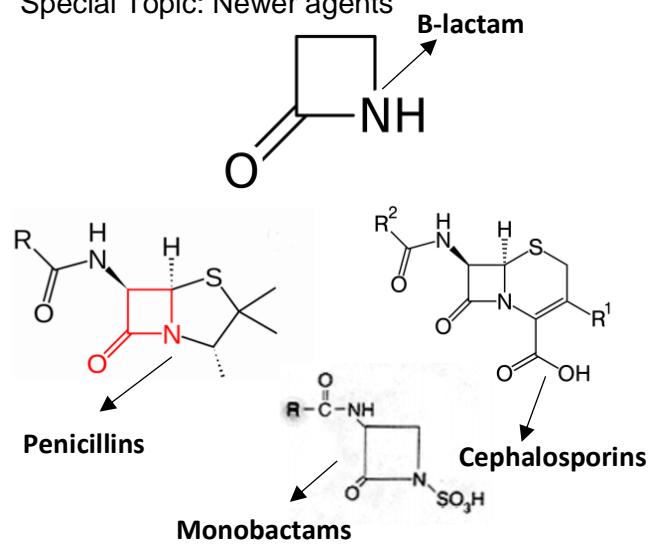


>there are variety of examples of antibacterials and each example there is a corresponding target in the bacteria, its either in the structure or in the process. Such targeting would cause a significant effect

Chemical Classification

>related on the structures of the commercially available antibacterials

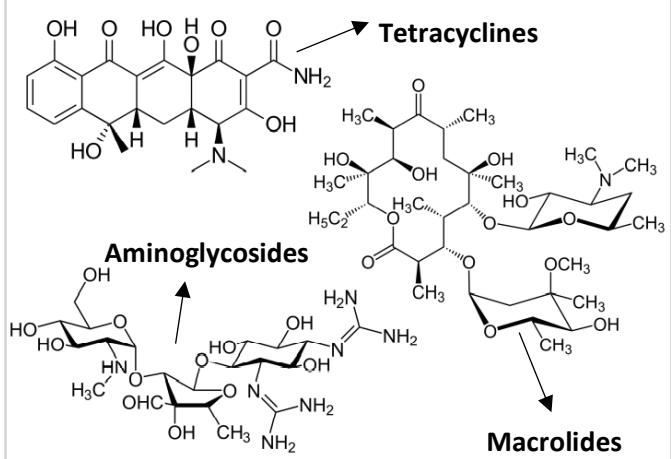
- Penicillins, cephalosporins (B-lactam ring)
- >collectively, these drugs are called B-lactam antibiotics
- Tetracyclines (4-annulated rings)
- Aminoglycosides (amino sugar)
- Macrolides (large lactone ring)
- Bacitracins, polymyxins (polypeptides)
- Special Topic: Newer agents



>those B-lactam antibiotics, its structure has common ring found in them that is B-lactam (lactam is a cyclic amide)

>this is important to know because by considering the amide structure of B-lactam antibiotics, we can trace in the ring kung nasan ba ang amide

>it is called monobactams because there is only one cyclic ring structure found that is solely the β -lactam
>penicillin (5-membered ring) and cephalosporins (6-membered ring) are bicyclic



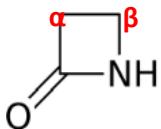
>tetracyclines have 4-annulated rings
>aminoglycosides (amino ethers) are agents that contains amino sugars that are linked by glycosidic bonds (ether structure ROR)
>macrolides have common large lactone (cyclic ester $RC=OOR$) ring structure
>epoxide (cyclic ether), lactam (cyclic amide), lactone (cyclic ester)

Inhibitors of Cell Wall Synthesis

>-cids because they can cause lysis

1. β -lactam antibiotics

- Penicillins
- Carbapenems (under P.)
- Cephalosporins
- Monobactams



>it is a secondary amine because it has adjacent carbon

>it is important to see substituent/side chain that is attached on the Carbonyl

>the first carbon adjacent to the carbonyl is the alpha carbon and the succeeding carbons next to alpha carbons are Greek alphabet
>Beta carbon which is attached with the amine group that's why it is β -lactam

The Bacterial Cell Wall

- Absent from mammalian cells
 - *Potential for selective chemotherapy
- Functions:
 - 1) Semipermeable barrier from substances
 - >goes inside or outside of the bacteria
 - 2) Barrier from osmotic pressure changes
 - >when osmotic pressure changes from the outside environment, that could be fatal to

the bacterial cell because that can cause shrinkage or lysis

>it can be prevented because they have cell wall

3) Prevent digestion by host enzymes

>by knowing that β -lactam antibiotics can inhibit cell production these (3 functions) will be oppose by them and automatically there will be lysis effect to the bacterial cell causing immediate death/fatality
>we can also conclude that by using drugs that targets the cell wall, that drugs display selective toxicity because the one that being targeted by the drug is a structure that is absent in mammalian cells (safe)

>there are 2 groupings of bacteria base on gram staining which are the gram positive and the gram negative

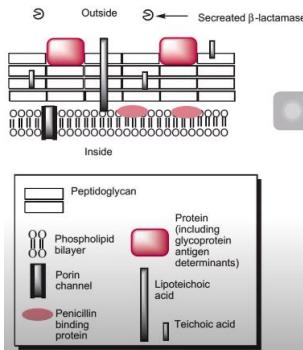
Gram Positive Cell Wall

>significant structure present in the gram positive

- Carbohydrates and Proteins - antigen determinants and adherence

>on the very outside layer or very outside part of the cell, is a set of characteristic carbohydrates and proteins that altogether serve as antigenic determinants and adherence

- Peptidoglycan - spongy layer of alternating sugars NAG (N-acetylglucosamine) and NAM (N-acetylmuramic acid)



>next or second layer barrier

>gel-forming layer

>represented by the rectangle shape

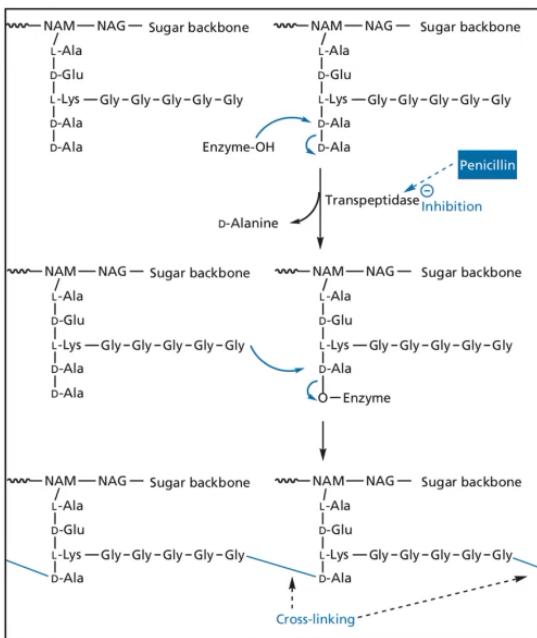
>NAG and NAM is being linked together by glycosidic bond (1,4 β -glycosidic bond)

>peptidoglycan layer has alternating sugars of NAG and NAM that is link by 1,4 β -glycosidic bonds

>peptidoglycan layer will not be stable/strong if it only has alternating sugars of NAG and NAM, kailangan ang mga layers ay mag kapit-kapit sa isa't isa (cross-linking)

>how will they have a cross link?

>because cross linking will make the peptidoglycan stronger and cross linking will be the one that will be block by β -lactams



>NAM has residue/moiety that is lactic acid and, in LA portion, it has amide bond where a pentapeptide (5 amino acyl residue) is being attached
 >5 amino acyl residue are Alanine, Glutamic acid, Lysine, and Dipeptide comprising both of alanine
 >in the lysine residue there are 5 pentaglycine (glycyl residues)

Goal: pentaglycine that is bound in the lysine should attached to other pentapeptide so that when it is attached already there will be cross linking
 >for it to be attached, it needed an enzyme (PBPs-Penicillin-Binding Proteins)

>PBPs has two functions/activities

1. it has carboxy peptidase action
2. transpeptidase action

>when PBPs arrive on the site of production of peptidoglycan (cross-linking site):

>the first step is, the terminal D-alanine has to be cleaved off (mawala) and for it to be remove it needed an enzyme particularly carboxy peptidase action

>that's why when the carboxy peptidase goes to the pentapeptide the terminal d-alanine will be remove and the enzyme itself will be attached

>and when the enzyme has attached, that facilitates movement of pentaglycine chain that is link with the lysine residue

>so when the pentaglycine go to the tetrapeptide chain, it will be attached to d-alanine, and when it is attached already that causes removal of the enzyme bound and the responsible for the removal is the transpeptidase activity of PBPs

>the pentaglycine that is attached on the lysine earlier, it is now attached on the 4th amino acyl

residue that is d-alanine and the enzyme was already remove

>the result is, it has now a bridging on each peptide chain

>bridging is responsible for the cross-linking making the peptidoglycan stronger

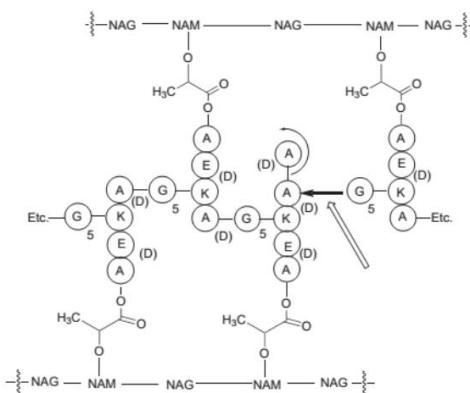


FIGURE 33.10 Schematic of cell wall cross-linking. Pentaglycyl group replaces terminal d-alanine.

>alternating sugar of NAG and NAM

>and on the NAM, it has lactic acid residue

>and in the LA portion it has amide bond from pentapeptide

>and to facilitate attachment of the pentaglycine chain, kailangan ma cleaved off muna ang terminal alanine via carboxy peptidase

>and kapag Nawala na ang papalit ay enzyme and didikit na ang pentaglycine residue kaya meron nang cross-linking

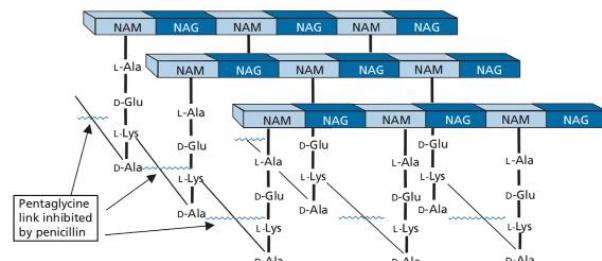


FIGURE 19.13 Peptidoglycan structure of bacterial cell walls.

- Teichoic and teichouronic acid – traverses peptidoglycan layer

>tumatakos sa peptidoglycan layer

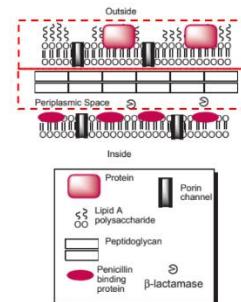
- Cell membrane – important proteins float (PBPs – enzymes necessary in cell wall formation and remodeling)

Gram Negative Cell Wall

>the cell wall is more complex and more lipoidal

- More complex with additional outer lipid membrane

>the outer layer contains complex lipopolysaccharides that encode antigenic responses that can



cause septic shock and provide zero type and influence morphology

- Lipid A – encodes antigenic response (septic shock)
- Porins – selective

>serves as channels for the movement of hydrophilic compounds

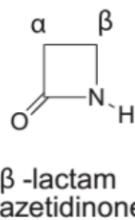
>usually, thicker ang peptidoglycan ng gram positive kaysa gram negative

Beta-Lactam Antibiotics

>they can inhibit cell wall production because they target/inhibit the PBPs (carboxypeptidase and transpeptidase) and by inhibiting it there is no cross linking in the peptidoglycan layer thus, peptidoglycan will destroy

>destruction of peptidoglycan layer can cause -cidal effect in the bacteria

- Possess β -lactam ring (4-membered-cyclic amide)



>contemporary name: Azetidinone

- Penicillin G (benzylpenicillin) and Penicillin V (phenoxy methylpenicillin) → agents of choice

- Potent, rapid bactericidal action

>target cell wall causing lysis

- Low toxicity

- Inhibits synthesis of Peptidoglycan – provides strength and rigidity to the cell wall

1) D-TRANSPEPTIDASE

→ Cross-linking of peptidoglycan

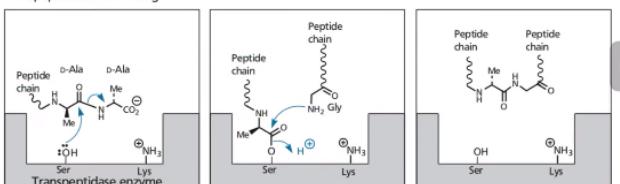
>thru this enzyme, there us attachment of the pentaglycine chain to the lysine residue

2) D-ALANINE CARBOXYPEPTIDASE

→ Hydrolysis of D-alanine-D-alanine terminal peptide bonds

>MOA of penicillin (β -lactam antibiotics):

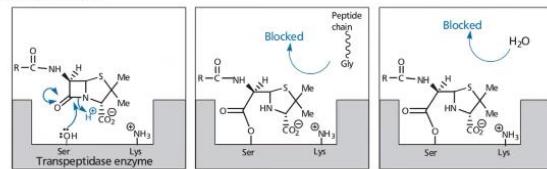
(a) Transpeptidase cross-linking



>in transpeptidase cross-linking, it happens because of transpeptidase enzyme can cause binding of the pentaglycine residue on the tetrapeptide

>then there is cross-linking of 5 glycine units bound in the lysine then alanine

(b) Penicillin inhibition



>Penicillins can inhibit transpeptidase action because these are the ones that would bind to the enzyme

>and when it binds, hindi na makakarating ang peptide chain comprising of glycine residues and that's why wala nang bridging sa pagitan ng glycine at tetrapeptide so walang cross-linking

Penicillin-Binding Proteins (PBPs)

- PBPs 1a and 1b
 - Cell elongation
 - Inh → spheroplast formation → cell lysis
- PBP 2
 - Maintains rod shape of bacilli
 - Inh → ovoid/round forms → delayed lysis
- PBP 3
 - Septum formation
 - Inh → filamentous form → cannot separate
- PBP 4 to 6
 - Carboxylpeptidase
 - Inh → NOT lethal to bacterium

Specific examples:

- Penicillin G → PBP 3
- 1st Gen Cephalosporins → PBP 1a
- Amdinocillin → PBP 2
- Others → PBPs 1, 2, 3

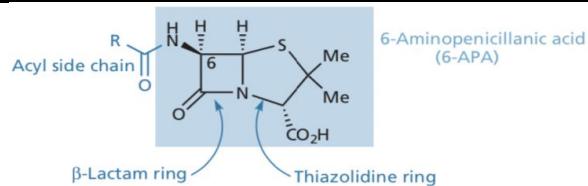
THE PENICILLINS

>came from natural sources

>acidic drugs

- Commercial production from *P. notatum* and *P. chrysogenum*
- Fermentation derived: Pen G, Pen V and 6-APA (6-aminopenicillanic acid)
- Sodium and potassium salts are crystalline, hydroscopic and water soluble

Structure of Penicillin



>general structure

>**B-lactam ring** which is a cyclic amide ring structure

>important to recognize the structure of penicillin is the ring that is attached with the β -lactam and that ring is called **thiazolidine ring**

>thiazolidine ring is a 5-membered ring structure
>in the thiazolidine ring there are substituents that can be found which are **carboxylic acid substituent** and **dimethyl substituents**

>all together, if the β -lactam ring is attached already with the thiazolidine ring together with the side chains it is called **6-Aminopenicillanic acid (6-APA)**

>6-APA is the core nucleus of all penicillins
>you can tell if a given drug is a penicillin if it has 6-APA that is a combination of β -lactam ring and thiazolidine ring and its substituents
>acyl side chain is not part of 6-APA
>the acyl side chain contains the variable side chain represented by the R and by having the variable side chain by adding certain substituents dyan maba branch out ang iba't-ibang examples ng penicillins

Nomenclature

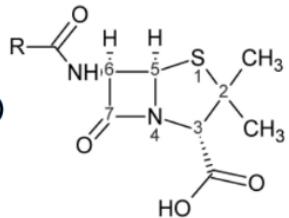
>it is important to have numberings because it has side chains

>in nomenclature it has two ways which are CAS (chemical abstracts system) and USP

- CAS**

>when referring to thiazolidine ring, the first atom is for the Sulfur atom, and the 4th is for Nitrogen

>the substituents that bound in the thiazolidine ring is can be seen in the 2nd carbon and the CA is) on the 3rd carbon



>the acyl amino is attached on the 6th member

- Sulfur atom (1); Nitrogen atom (4)
- Definitive but too complex
- 6-acylamino-2,2,-dimethyl-3-carboxylic acid

Example:

Benzylpenicillin Sodium

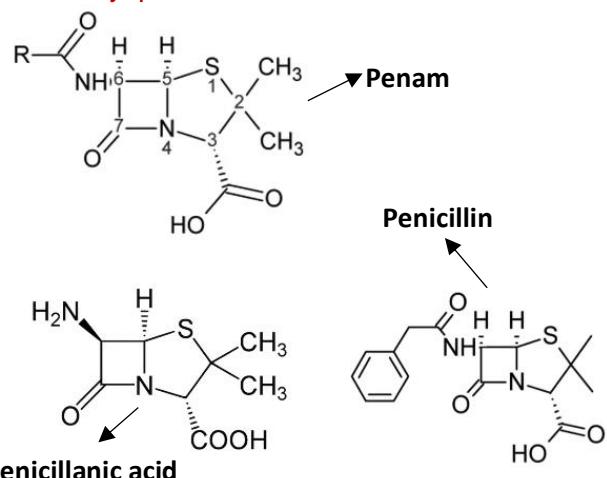
- Monosodium (2S,5R,6R)-3,3-dimethyl-7-oxo-6-(2-phenylacetamido)-4-thia-1-azabicyclo [3.2.0] heptane-2-carboxylate

- USP**

- Nitrogen atom (1); R
- Sulfur atom (4)
- 4-thia-1-azabicycloheptone

Simplified forms of Nomenclature:

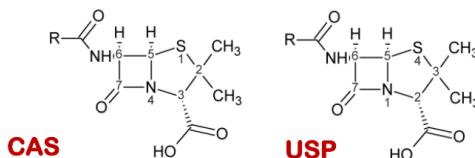
- “**penam**” – bicyclic system w/o substituents
>B & T only
>6-acylamino-2,2-dimethyl penam-3- carboxylic acid
- “**penicillanic acid**” – ring w/ substituents
> β -lactam and substituents but acyl amino is not included
>6-amino penicillanic acid
>it is 6-amino because the R-group in the penicillanic is hydrogen only
- “**penicillin**” + R-chain
>whole 6-APA plus the variable side chain
>benzyl penicillin



Stereochemistry

>detection of chiral carbons

>chiral is the carbon atom surrounded by other different atoms



CAS

- >for the C3, surrounding it is a nitrogen, carbonyl carbon, carbon atom and hydrogen
- >for the C5, surrounding it is a sulfur, nitrogen, hydrogen and carbon
- >for the C6, surrounding it is a hydrogen, carbon, carbonyl and amino

USP

- >same explanation with CAS, numbering is the only difference

- Three chiral centers
 - **CAS:** C3, C5, C6 **USP:** C2, C5, C6

Biosynthesis of Penicillins

>there are numerous penicillins which are considered semi-synthetic and in the production of

semi-synthetic penicillins it needed precursors and in the formation of 6-APA it needed two amino acids which are L-cysteine and L-valine

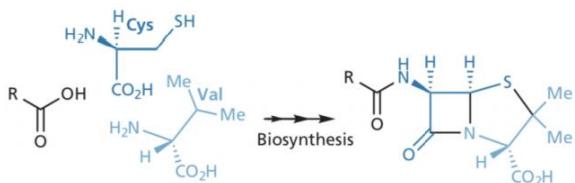


FIGURE 19.11 The biosynthetic precursors of penicillin.

- 6-APA derived from two amino acids:
 - L-cysteine and L-valine

>cysteine is a thiol containing (sulfur containing amino acid) in which it is called thiomethyl
 >valine is a branch chain amino acid containing isopropyl side chain
 >thru biosynthesis of penicillins, mabubuo ang β -lactam ring because of the structure from cysteine
 >and the portion of thiazolidine ring that is the sulfur from cysteine
 >and the valine is a isopropyl, it will form the other portion of thiazolidine ring containing 2 methyl side chains

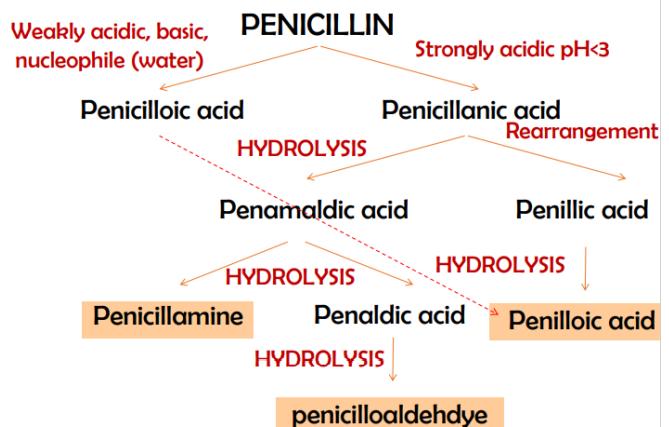
Chemical Instabilities

- Most penicillins are acids
- >acids containing pKa value ranging from 2.5 to 3.0
- >concludes that free acids are not suitable for parenteral administration
- >to address this issue, we must convert penicillins into its salt form and to achieve that goal we have to react penicillins with metals:
 - Na and K salts – highly water-soluble
 - Benzathine, procaine – poorly water soluble (depot forms for longer duration)
- >could be used for repository forms which would provide prolonged actions
- Hygroscopic
- >they have to be stored in sealed containers

Chemical Degradation

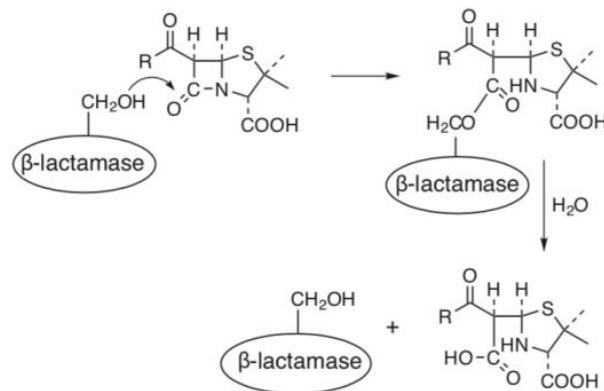
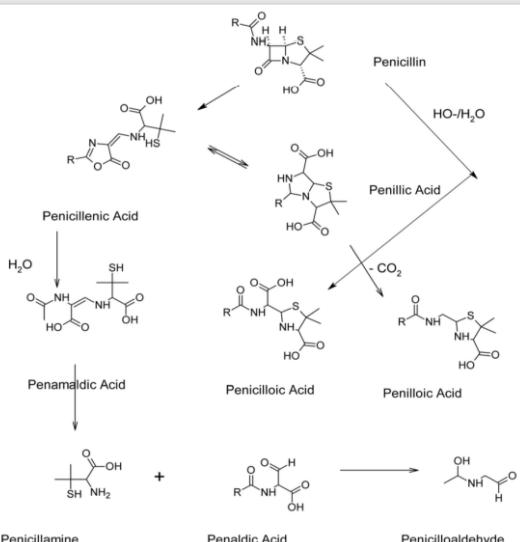
>this has clinical correlations

- Hydrolysis – main cause of deterioration
- >breaking down of chemical bond due to water (H_2O) or other hydrolytic enzymes
- >it is the main cause because of its susceptible β -lactam ring
- β -lactam amide bond – most unstable bond
- >amide can be hydrolyzed forming carboxylic acid and amine
- >it is important because when the β -lactam ring is broken, that would render the penicillin inactive already (not efficacious bacterial agents)



>the important factor that could degrade/hydrolyze penicillin is pH

Fate of penicillin if exposed to varying pH:
 >if penicillin is exposed in a **strongly acidic solution** ($pH<3$), that causes formation of penicillanic acid
 >penicillanic acid has a distinct structure called oxazolone ring structure
 >oxazolone ring structure is prone to hydrolysis forming penamaldic acid and then that the penamaldic acid containing enamine structure (combination of carbonyl and secondary amine)
 >the carbonyl could vary depending if its aldehyde or ketone
 >due to the presence of enamine structure to penamaldic acid it can also be hydrolyzed and hydrolysis of penamaldic acid would result to formation of penicillamine and penaldoic acid in which the latter would further be hydrolyzed into penilloaldehyde
 >penicillamine and penilloaldehyde are major metabolites (major compounds produced after hydrolysis)
 >in penicillanic acid, it could also undergo rearrangement process forming penillic acid
 >thru formation of penillic acid this will undergo hydrolysis leading to formation of penilloic acid
 >in penilloic acid (major metabolite) formation, this involves decarboxylation process
 >if it's exposed in **weak acid** in the presence of nucleophile, the product is penicilloic acid and thru decarboxylation process that causes formation of penilloic acid



- Drug interactions

>when penicillin undergo chemical degradation, it could have certain drug interactions particularly with aminoglycosides

>in practice doctors usually combined aminoglycosides with penicillins to enhance bactericidal effects like for examples use of:

>gentamicin and carbenicillin but these have to administer separately (don't mix)

>because if it administers together, that can cause destruction of the b-lactam ring present in the structure of penicillin rendering it inactive that can cause:

- Immune responses

>the metabolite of penicillin when undergo degradation particularly the penicilloic acid it could act as a hapten

>hence the presence of penicilloic acid in the body will react with physiologic proteins forming penicilloyl proteins and this protein is antigenic causing allergic reaction

>in other words, the immune responses when there is a metabolite, hydrolytic product ang penicillin that could serve as a trigger for the allergy to happen

Bacterial Resistance

>bacteria are capable of evolution making them protected against the action of penicillins particularly species of gram-negative bacilli are the ones that are naturally resistant to the action of penicillin

>2 recognized mechanisms related to penicillin:

- **Penicillinases** – bacterial enzymes that inactivate penicillins

→ **B-LACTAMASES** – hydrolytic opening of ring (*Staphylococcus aureus*)

>the ones that can destroy b-lactam rings or the amide cyclic ring that is present in penicillin making the drug ineffective

→ **ACYLASES** – hydrolyze acylamino side chain (Gm-bacteria, decreased permeability due to outer membrane and porins)

Clinical Correlations

>there is an impact on medicine if penicillin will undergo chemical degradation

- Beta lactamases

>hydrolytic enzymes that are being synthesized by bacteria as part of their evolution

>can destroy b-lactam rings rendering penicillins inactive that's why it is a problem when the drug is sensitive to b-lactamase, hydrolysis brought by b-lactamase will render the drug ineffectual already

>the ones that hydrolyzes acylamino side chain where the variable side chain is observed

- Decreased permeability due to OUTER MEMBRANE – additional barrier **PORINS**

>outer membrane is an additional barrier on top of the peptidoglycan layer and on top of the cell membrane present in gram-negative bacteria

- Altered permeability = Altered PBP binding
 - *Neisseria gonorrhoeae*
 - *Methicillin-Resistant S. aureus (MRSA)*

>altered permeability signifies that lesser tendency for the drug (penicillin) to target the PBP that's why the enzyme will not be blocked

>it can be resolved by targeting **porins**

>porins are the ones that allow entry of hydrophilic molecules/compounds

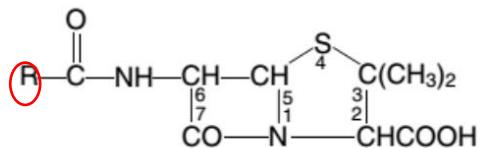
What are the solutions to these bacterial resistance problems?

>both are related in designing the drug by having their structure activity relationships

Penicillinase-Resistant Penicillins

>designed to resist the action of the hydrolytic enzyme penicillinases like β -lactamases

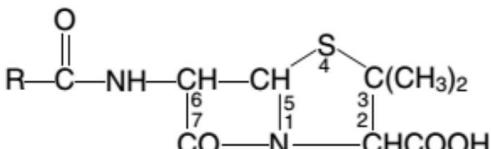
- The availability of 6-APA made possible synthesis of other semi-synthetic penicillins modified at the ACYLMAMINO SIDE CHAIN



>modified the side chain R

- Increasing STERIC HINDRANCE in the α -carbon of acyl group increases resistance
 - STERIC HINDRANCE – prevention of interaction due to spatial structure

>enhance steric hindrance at the α -carbon of acyl group



> α -carbon, this is the carbon adjacent to the carbonyl group that is part of acylamino side chain
>when the α -carbon has steric hindrance, it is penicillinase resistant

>how we can achieve that? It can be achieved by having the α -carbon either part of aromatic ring (α -

carbon is member of aromatic ring) or heteroaromatic system

>if phenyl, it is a substituent of benzene ring

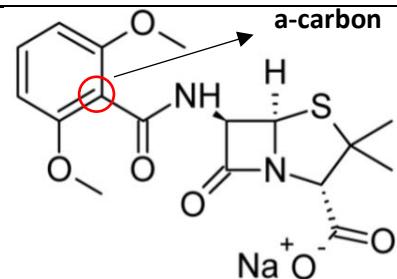
>if naphthyl, substituent of naphthalene (2 aromatic rings)

>if isoxazoyl, derived from isoxazole ring structure

- α -acyl carbon can be part of an aromatic (phenyl), naphthyl) or heteroaromatic (4-isoxazoyl) system

Examples:

Methicillin

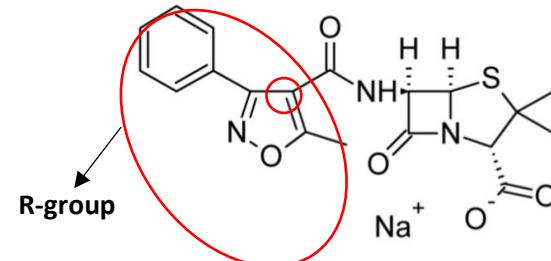


>1st step is to prove that it is a penicillin by checking the presence of 6-APA

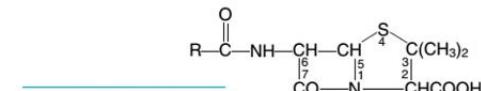
> α -carbon is part of the aromatic ring in which it can enhance steric hindrance

>methicillin is penicillinase-resistant

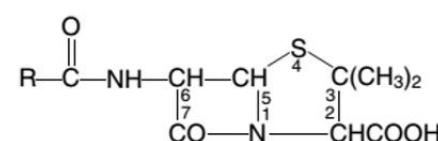
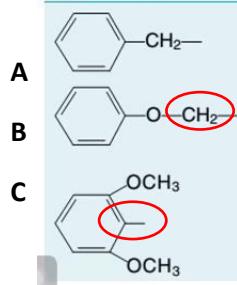
Oxacillin



>the α -carbon is a member of heteroatom ring system and the heteroatom is isoxazole so it means it increases steric hindrance rendering the drug resistant to penicillinases like β -lactamases



R Group



>which among the R-group would provide increased steric hindrance thus making the drug penicillinase resistant?

>**C** is the answer because the α -carbon is part of the ring and it would provide increase steric hindrance

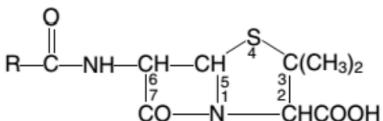
Note:

>another consideration in improving steric hindrance:

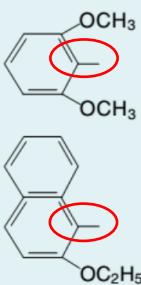
- Substitution at the **ortho position** of a phenyl ring or 2-position of a naphthyl system

>in this case there is enhance steric hindrance if there is ortho (katabi ng α -carbon) substitution in the ring

→ INCREASES STERIC HINDRANCE → INCREASED RESISTANCE



Methicillin 2,6-Dimethoxyphenyl-penicillin



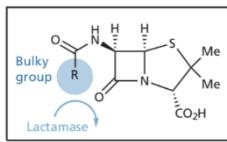
Nafticillin 2-Ethoxy-1-naphthyl-penicillin

>**Methicillin**: α -carbon is part of the ring which is a phenyl substituent and there is a methoxy on the 2nd and 6th carbon

>**Nafticillin**: ethoxy substituent at the ortho position

2. Bulkier substituents in heteroaromatic (isoxazoyl) rings confer effective β -lactamase resistance

>bulkier substituents can only be added in heteroaromatic ring structure present on the R-chain

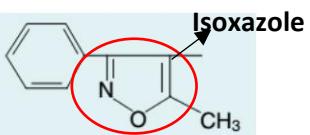


>penicillins with isoxazole ring

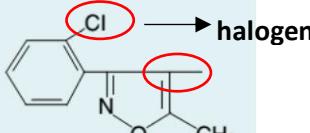
>usually, the bulkier substituents that is used is methyl which is also ortho position

Examples:

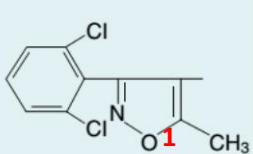
Oxacillin 5-Methyl-3-phenyl-4-isoxazolylpenicillin



Cloxacillin 5-Methyl-3-(2-chlorophenyl)-4-isoxazolylpenicillin



Dicloxacillin 5-Methyl-3-(2,6-dichlorophenyl)-4-isoxazolylpenicillin



>**Oxacillin, cloxacillin and dicloxacillin**: R-group containing isoxazole ring (heteroatom ring structure because it contains atom other than carbon)

> α -carbons are part of the actual ring structure that's why it is still conform to increase steric hindrance

>1-oxygen, 2-nitrogen, 3-carbon, 4- α -carbon

>in the 5th carbon there is a methyl constituent that can be seen

>by adding methyl group on the ortho position of the α -carbon, that confers increase bulkiness thus increasing steric hindrance, penicillinase resistant

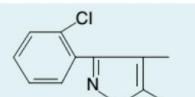
>cloxacillin and dicloxacillin are halo benzylpenicillins because they have halogens in the phenyl ring

- Isoxazoyl penicillins with electronegative substituent in the 3-phenyl group are also acid-resistant

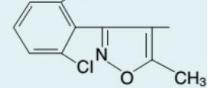
>isoxazoyl has isoxazole ring that is heteroatom and also have bulky substituent methyl that's why it is penicillin resistant

>cloxacillin and dicloxacillin has heteroatom Chlorine that is electronegative atom

Cloxacillin 5-Methyl-3-(2-chlorophenyl)-4-isoxazolylpenicillin



Dicloxacillin 5-Methyl-3-(2,6-dichlorophenyl)-4-isoxazolylpenicillin



>if its acid-resistant there will be no forming of other penicillins (penicillanic acid, etc.) that's why it can be taken by mouth because it can resist the acidity in the stomach due to hydrochloric acid

- Increasing the bulkiness leads to significantly less activity

>leads to penicillinase resistant effect however less activity compared to Pen G but protected against β -lactamases

- Steric factor does not necessarily confer acid stability

>involves in electronegative atom

Parenteral

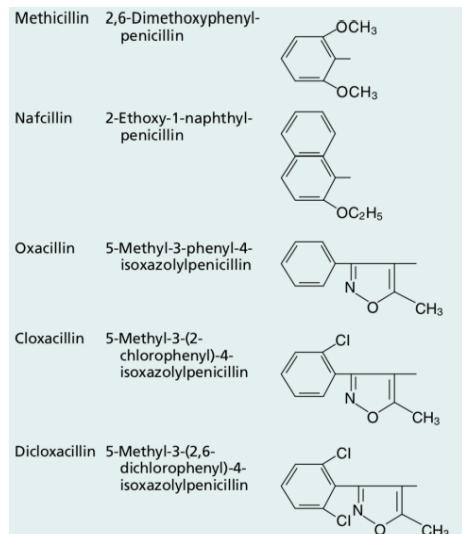
- Methicillin (part of benzene ring)
- Nafticillin (part of naphthalene)

>its parenteral because they are not acid stable, they can only provide penicillinase-resistant because they have increase steric hindrance and ortho substitution of methoxy and ethoxy group

Oral

- Oxacillin
- Cloxacillin
- Dicloxacillin

>because of presence of electronegative atom
 >penicillinase-resistant penicillins are narrow spectrum



Extended-Spectrum Penicillins

>in which these agents can still enter the peptidoglycan layer of gram-negative bacteria despite of having the outer membrane

>broad spectrum

>address the second problem in bacterial resistance and that is the formation of outer membrane

>the problem in the outer membrane is, it is an extra shield of the bacteria in gram-negative that's why it is hard for other penicillins to enter (ineffective)

Goal: makatawid ang penicillin sa loob ng gram-negative papuntang peptidoglycan kahit may outer membrane

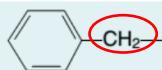
>the opportunity of extended-spectrum is daanan ang porins (channels that allow entry of hydrophilic molecules)

>to make the penicillins extended-spectrum, penicillins must be hydrophilic as well

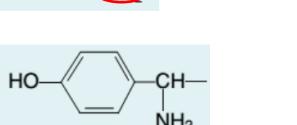
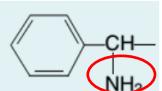
1. Introduction of ionized or polar group into a-position of benzyl carbon of Pen G (precursor) confers activity to Gm (-) bacilli

- Alpha-amino
- Alpha-hydroxy
- Acidic (carboxylic acid) substituent

Penicillin G Benzylpenicillin



Ampicillin D- α -Aminobenzyl-penicillin



Amoxicillin D- α -Amino-p-hydroxybenzylpenicillin HO-

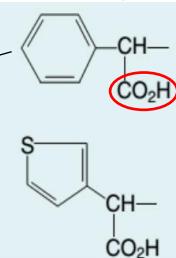
>the precursor/inspiration is modification of the carbon adjacent to the benzene ring
 >ampicillin, amoxicillin and bacampicillin are extended-spectrum because they can pass thru porins because they are hydrophilic already, they are aminopenicillins

>bacampicillin is the prodrug of ampicillin

- a-hydroxy substitution yields "extended-spectrum" penicillins but are less active than Pen G
- incorporation of acidic (CA) substituents at the a-benzyl carbon also imparts extended spectrum

Carbenicillin α -Carboxybenzyl-penicillin
Carboxypenicillin

Ticarcillin α -Carboxy-3-thienyl-penicillin



>addition of carboxylic acid at the a-benzyl carbon

- Carbenicillins, and other penicillins, exert SYNHERGISTIC bactericidal action with aminoglycosides

>carbenicillin is usually combined with aminoglycoside like gentamicin

>carbenicillin is a broad spectrum (safe) and gentamicin is toxic requiring the usage of limited dose

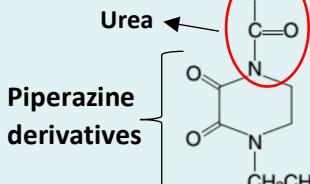
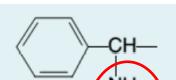
>to combine gentamicin is should be lesser dose

- however, chemical incompatibility requires these drugs to be administered separately

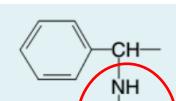
2. A series of a-acylureido-substituted penicillins exhibit greater activity against Gm- than carbenicillin

>addition of acylureido (presence of urea in the a-carbon) substitution

Piperacillin α -(4-Ethyl-2,3-dioxo-1-piperazinylcarbonylamino)benzylpenicillin



Mezlocillin α -(1-Methanesulfonyl-2-oxoimidazolidino-carbonylamino)benzylpenicillin



>piperacillin, mezlocillin and azlocillin are ureidopenicillins

>extended-spectrum are hydrophilic/polar to permit entry of the drug thru porins to bypass the outer membrane

Oral

- Ampicillin prodrug of Bacampicillin
- Amoxicillin

Parenteral

- Carbenicillin
- Ticarcillin
- Piperacillin
- Mezlocillin

Penicillinase-resistant

→ NARROW SPECTRUM

Broad/extended spectrum

→ PENICILLINASE-SENSITIVE

>they are combined w/ other drugs

Penicillin	β -Lactamase Resistance (<i>S. aureus</i>)	Spectrum of Activity
Benzylpenicillin	No	Intermediate
Penicillin V	No	Intermediate
Methicillin	Yes	Narrow
Nafcillin	Yes	Narrow
Oxacillin	Yes	Narrow
Cloxacillin	Yes	Narrow
Dicloxacillin	Yes	Narrow
Ampicillin	No	Broad
Amoxicillin	No	Broad
Carbenicillin	No	Extended
Ticarcillin	No	Extended
Mezlocillin	No	Extended
Piperacillin	No	Extended

REVIEW:

>SAR addresses two problems which are involved to resistance

1. formation of penicillinases like β -lactamases

2. formation of outer membrane

>in order to solve this, in β -lactamase the steric hindrance must increase for it to have greater resistance against penicillinases and to achieve it we must focus on the α -carbon that should be part of the aromatic ring structure (whether single or multiple) or heteroaromatic ring structure

>or in the aromatic there is a substitution of halogen/ortho positioning

>in the heteroatom, addition of bulky substituent like methyl

>in the outer membrane the goal is to make it polar and to make it polar, we have to focus on the α -carbon adjacent to the benzyl substituent of Pen G (amino, carboxyl, urea)

> β -lactamase resistant – narrow spectrum

>polar – broad spectrum

Specific examples of penicillin:

>there are different ways on how to classify agents under penicillins

1. natural/synthetic
2. resistant/sensitive to penicillinases (penicillin can be narrow or broad spectrum)
3. based on route of administration

Biosynthetic Penicillins

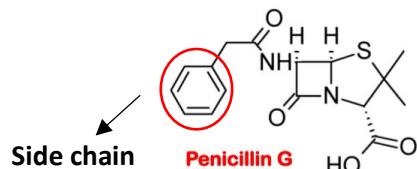
>these are the agents that were derived from the fungus *penicillium chrysogenum*

- 6-APA – core nucleus of all penicillins
- Pen G – parenteral (benzyl penicillin)

>parenteral because it is labile/sensitive to acidic environment and its goal is to bypass the stomach

- Pen V – oral (phenoxy methyl penicillin)

>orally administered because on its R-group/side chain there is presence of electron withdrawing group



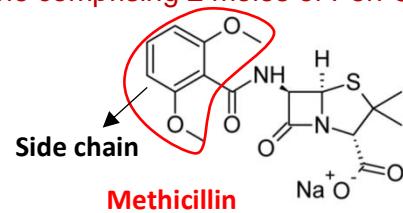
>**Pen G:** it is called benzyl penicillin because of the benzyl that serves as the side chain

>remains to be the agent of choice in the tx of numerous bacterial infections

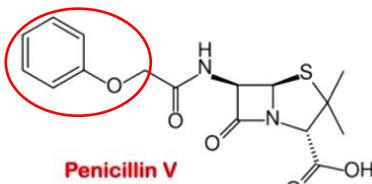
>penicillin is an acidic drug, to make it water-soluble it has to be converted into salt forms and the available salt forms for penicillin is K, Ca, Na

>its advantage of kinetic is it has too fast ADME that requires greater frequency of administration. The remedy to prolong the ADME of Pen G is that to make amine salt

>and when it is amine salt it would provide repository/depot effect (Pen G procaine-1st widely used amine salt of Pen G, Pen G benzathine-salt of diamine comprising 2 moles of Pen G)



>**Methicillin:** it is called phenoxy methyl penicillin because in the R-group/side chain it has benzene ring in which there is ortho substitution/positioning of methoxy group



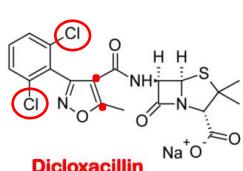
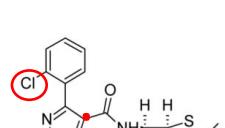
>Pen V: resistant against gastric juice thus acid stable and that can be attributed to electro withdrawing group of it
 >has phenoxy methyl substituent and electron withdrawing group that is why it is acid-stable

Penicillinase-Resistant, Narrow Spectrum, Oral Penicillins

>they are called penicillin resistant because they provide steric hindrance/shield
 >they are orally administered because they have enhanced acid stability because of ortho-halogenation at the 3rd atom of the isoxazoyl ring

isoxazoyl

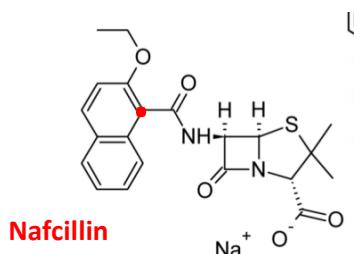
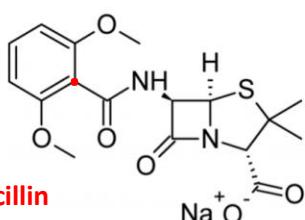
- Oxacillin
- Cloxacillin
- Dicloxacillin



Penicillinase-Resistant, Narrow Spectrum, Parenteral

>they are parenteral because there is no electron withdrawing group present in the R-chain making them susceptible to acids

- Methicillin
- Nafcillin



>difference is single aromatic only in methicillin and in the nafcillin there is 2 aromatics (naphthalene)
 >there is ortho substitution in which in the methicillin-methoxy and in nafcillin-ethoxy

Penicillinase-Sensitive, Broad Spectrum, Oral Penicillins

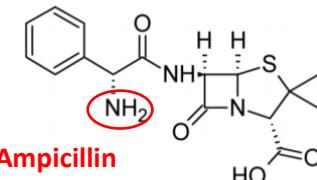
>they are broad spectrum because they are polar permitting the entry thru the porins of gram-negative by passing the outer membrane

>they are penicillinase-sensitive because they have no steric shields present that's why they are vulnerable to the action of β -lactamase that could render them ineffective

Aminopen

- Ampicillin
- Amoxicillin
- Bacampicillin

>aminopenicillins because on the a-carbon they possess amino functional group making them polar



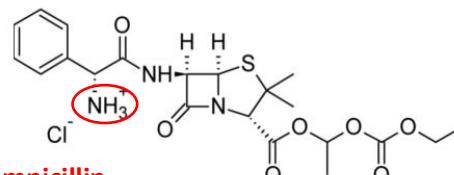
>agent that is usually combined with probenecid in the tx of gonorrhea (STD tulo)

>the reason for its combination is because the probenecid is a uricosuric agent (process of excretion is renal), if they were given together, they will compete which drug would be excreted and will be finally removed

>between the two, probenecid will be permitted to be excreted, and the ampicillin will prevent/inhibits the active tubular secretion hence ampicillin will be reabsorbed

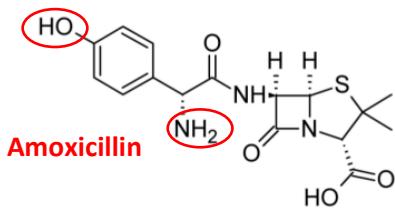
>reabsorption of ampicillin will render it more effective

>Ampicillin + Probenecid = enhance ampicillin's effect because of reabsorption



>prodrug of ampicillin

>bacampicillin itself has no antibacterial activity/property but if taken, this agent undergoes hydrolysis and thru it the active form ampicillin will be derived



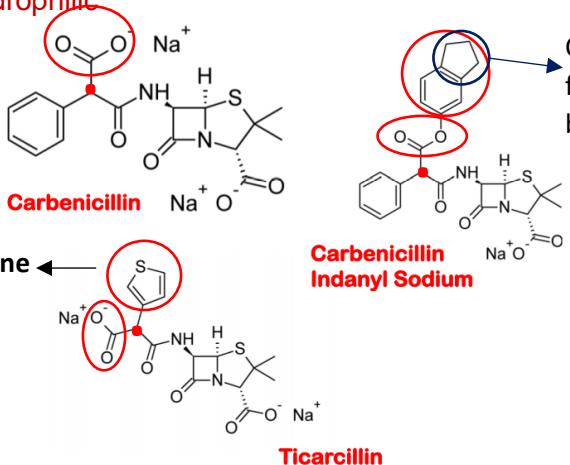
>have hydroxyl group at the para position of the benzene ring that's why it is called p-hydroxy analog of ampicillin

Penicillinase-Sensitive, Extended Spectrum, Parenteral

>not stable to acids

- Carbenicillin
 - Ticarcillin
 - Piperacillin
 - Mezlocillin
 - Azlocillin
- } Carboxy
- } Ureidopen

>carboxypenicillins because on the α -carbon there is carboxyl group and in the ureido there is urea FG
 >they are extended spectrum because they are hydrophilic



>**carbenicillin:** has low toxicity and its limited adverse effect is allergy due to existence of penicillionic proteins, it permits large doses of administering

>effective against strains that are resistant to ampicillin

>usually combined w/ gentamicin and its effect is bactericidal but it should not be administered literally together because gentamicin can inactivate carbenicillin thru hydrolysis

>gentamicin is toxic so it must be administered in low doses only

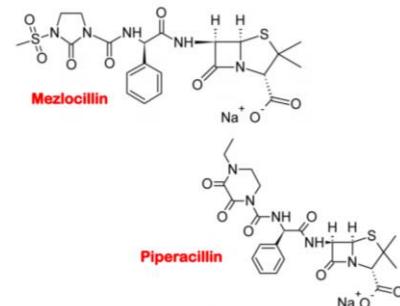
>**carbenicillin indanyl sodium:** form of carbenicillin that is orally active because of the presence of indanyl substituent

>indanyl substituent is a cyclopentane fused w/ benzene ring

>**ticarcillin:** thiophene can be seen in the R-group

>similar to carbenicillin but it has advantages:

1. better pharmacokinetics, (high serum levels, and longer duration of action)
2. greater potency against gram-negative bacteria such as pseudomonas aeruginosa and Bacteroides fragilis



>more active against klebsiella, pseudomonas, haemophilus, anaerobic bacteria

>piperacillin contains piperazine derivatives

>mezlocillin contains 2-oxoimidazolidine

B-LACTAMASE INHIBITORS

>the ones that would target b-lactamase enzyme

>b-lactamases are the enzymes that can hydrolyze the b-lactam structure which will render penicillin inactive/ineffective

- B-lactam combination therapy

>combined w/ other b-lactam antibiotics to be able to enhance the bactericidal effect of these b-lactams

- Referred to as "suicide substrates"

>serve as substrates for the enzyme that they inactivate

>b-lactamase inhibitors have affinity to the activity of b-lactamases

>sila ang tatargetin nung enzyme, sila ang ma hahydrolyze (b-lactam nila ang ma hahydrolyze) after that, the enzyme cannot target the actual b-lactam antibiotic anymore

- Knowles described two classes

CLASS I	CLASS II
Have heteroatom leaving group at position 1	Do NOT have heteroatom leaving group at position 1
Ex. Clavulanic acid, sulbactam, tazobactam	Ex. Carbapenems
Cause prolonged inactivation (extended-spectrum)	Cause transient inhibition

>the difference can be seen at position 1 of bicyclic ring

>**Class I:** they have heteroatom living group at position 1 like presence of S and O heteroatom

>have insignificant antibacterial activity

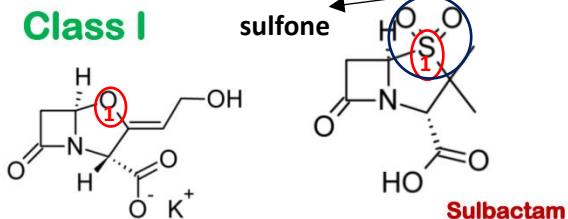
>usually combined w/ other powerful bactericidal b-lactams

>**Class II:** no heteroatom present

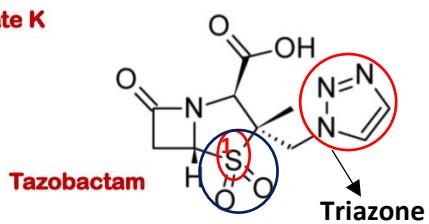
>thienamycin, imipenem, meropenem

>have significant antibacterial activity

Class I



Clavulanate K



>presence of heteroatom

>**clavulanic acid:** was isolated from *streptomyces clavuligeris* and it has weak antibacterial activity

>very effective inhibitor of b-lactamase

>acid stable that's why it permits oral administration but not limited to oral administration

>prepare usually w/ combination of penicillins especially those penicillinase-sensitive

Example: Amox + CA = Co-Amoxiclav (Augmentin) by mouth

Ticarcillin + CA = Timentin (IV)

>**Sulbactam:** has sulfone structure

>usually combined w/ ampicillin = Sultamicillin (Unasyn)

>effective against *Acinetobacter baumannii*

>**tazobactam:** more potent than sulbactam and contains sulfone and has presence of trizone

>usually combine w/ piperacillin = Piptaz

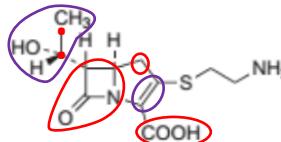
B-lactam Combination

- Amoxicillin + Clavulanic acid
- Ticarcillin + Clavulanic acid
- Ampicillin + Sulbactam
- Carbenicillin + Sulbactam
- Piperacillin +Tazobactam
→ broadest spectrum

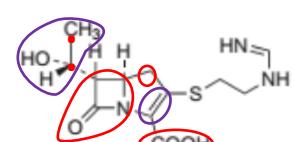
>if a penicillinase-sensitive penicillin is combined with b-lactamase inhibitor, the drug interaction is potentiation which has a mathematical equation ($1 + 0 = >1$)

>1 has bactericidal action and the other one has insignificant/nearly w/o antibacterial activity but if combined greater effect (beneficial effect)

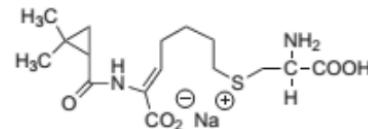
Class II: CARBAPENEMS



Thienamycin



Imipenem



Cilastatin sodium

>at position 1 there is no heteroatom

>they possess antibacterial activity

Similarities and difference based on structures Carbapenems vs. Penicillins:

>they both have b-lactam rings that's why it has antibacterial activity

>on the 3rd position it has carboxyl group

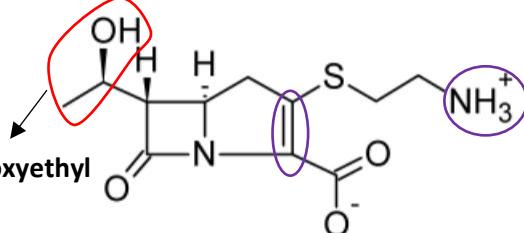
>on the ring structure containing 5-membered atom, in carbapenems there is a presence of double bond between carbon 2 and 3

>in penicillin there is acyl group while in carbapenems there is 1-hydroxyethyl substituent

Thienamycin

>has broad spectrum of antibacterial activity and resistant against b-lactamases

- First isolated from *S. cattleya*
- C2=C3 increases reactivity to ring opening
- Resistant to b-lactamases due to a-1-hydroxyethyl side chain



>disadvantage: chemically unstable in both acid and basic solutions due to the presence of double bond that could render the drug susceptible to chemical instability and presence of primary amino group

- More susceptible to hydrolysis by renal dehydropeptidase-I (DHP-1)

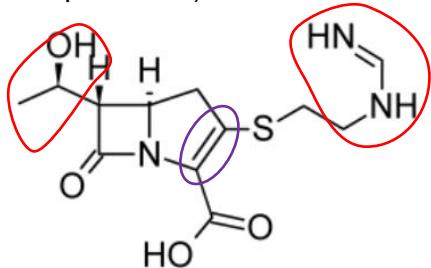
>could render carbapenem ineffective

- Shorter half-life in vivo

>requires more frequent administration

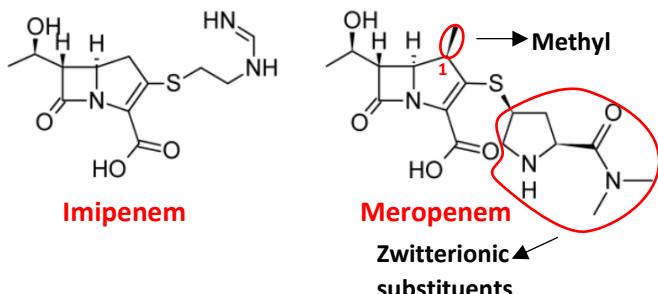
Imipenem

- Most successful derivative of thienamycin
- Not orally active
- +CILASTATIN → inhibitor of renal DHP-1 (provides protection)



>the primary amino group in thienamycin was converted to a non-nucleophilic basic function
 >presence of 1-hydroxyl makes it resistant to b-lactamases but it is now orally active
 >there is a presence of double bond that's why it is sensitive/prone to renal dehydropeptidases
 >the role of cilastatin is to block renal DHP-1 thus enhancing/prolonging the action of imipenem

(2nd gen) Newer Carbapenem: MEROPENEM (adv)



- Methyl group at C1 provides resistance to renal DHP-I
- Substitution at the 2-position affects spectrum
 >they modified the structure of imipenem by adding a methyl group at position 1 which will have the resistant against DHP-I therefore meropenem will no longer need to be combined with cilastatin
 >on the 2nd position there is a presence of zwitterionic substituent (combination of proton donor and acceptor group that's why the charge is zero)
 >if there is zwitterionic substituent at the 2nd position it can promote entry thru porins that can enhance/expand spectrum of activity
 >not active orally

Biapenem

- Newer 2nd -gen carbapenem

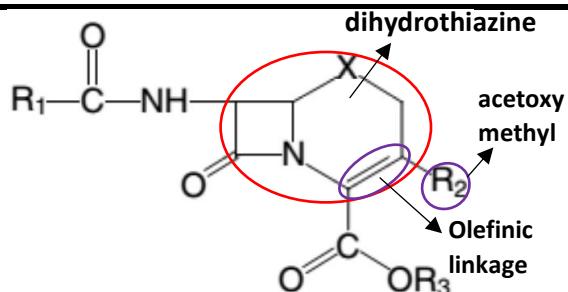
Doripenem

- Doribax

CEPHALOSPORINS

- 1945: Giuseppe Brotzu discovery of the fungus *Cephalosporium acremonium* having antibacterial properties
 >currently known as *acremonium chrysogenum* inhibited the growth of wide variety of gram-positive and gram-negative bacteria
- 1948: Abraham and Newton isolation of Cephalosporin C
 >from oxford university
 >cephalosporin C is significant because although it is not very potent (1/1000) as compared to Pen G but it has greater resistance against acid hydrolysis and b-lactamase enzyme and less likely to cause allergic reaction
 >there were modifications done in cephalosporin C to be able to get the major nucleus/product that is considered to be biologically important
- Cephalosporin C – first cephalosporin
- Core nucleus: 7-aminocephalosporanic acid (7-ACA)
- B-lactam ring + dihydrothiazine = cefem/cephem

Nomenclature



- >has bicyclic ring system containing the b-lactam ring and dihydrothiazine (6-membered ring)
- >consequence of bigger ring: less strain, less reactive and potent
- >attributable to the olefinic linkage (C2-C3)
- >presence of acetoxy methyl can reduce the activity
- >olefinic linkage and acetoxy methyl can modulate hydrolysis

Semi-synthetic Derivatives

- >the precursors in production of 7-ACA are amino acids valine and cysteine
- >delay tolerance
- Improvements in preparations:
 1. Increases acid stability
 >could be observed in the substitution of R₁, R₃
 2. Better oral absorption
 3. Broadened spectrum
 4. Increased activity
 5. Decreased allergenicity

6. Increased tolerance

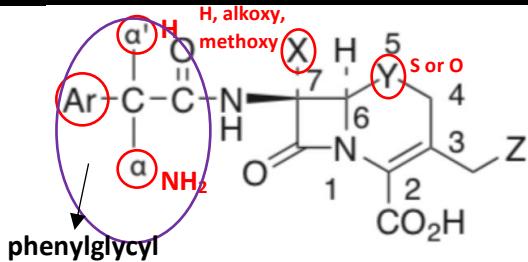
>mechanism of resistance by bacteria in which there are antibiotics that are first are -cidal and because of tolerance it becomes -static

Chemical degradation

Hydrolysis

- 3-acetoxymethyl: Parenteral cephalosporins
- >**has ester (hydrolysable) substituent**
- Beta-lactam: the reactive functionality common to all cephalosporins
- Cephalosporanic acids

Structure-Activity Relationship



C3 & C7 - b-lactam cleavage (hydrolysis)

1. in vitro stability of the drug being designed
2. antibacterial activity

3. stability of the agent against b-lactamases

>**Ar** represents benzene ring as a substituent which is a phenyl substitution it has implications regarding acid stability of certain cephalosporins

>if the alpha substitution is an amino group, where in the alpha prime is substituted as hydrogen that confers acid stability

>if there is phenyl, amino and hydrogen these confers acid stability hence it can be given by mouth

>these substitutions is called phenylglycyl substitution

>**variable X:** substitution on position 7 is essential on antimicrobial activity, when this variable is being replaced by hydrogen, this is essential for antibacterial activity

>enhance the antibacterial activity by replacing hydrogen by another substituent alkoxy (OR) that results to enhanced antibacterial activity held by cephalosporin

>there is additional activity of cephalosporin if in the 7th position there is methoxy that will provide stability against b-lactamases

>**Variable Y:** represents the heteroatom that could be S or O

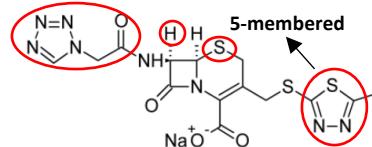
>if sulfur is the substituted heteroatom in the y variable, there is greater antibacterial activity than oxygen

>if oxygen, it is more stable against b-lactamase than those cephalosporins that has sulfur

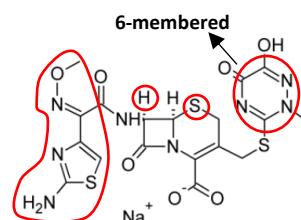
>**variable Z:** if its 5-membered hetero cycle it has greater antibacterial activity than those who have 5-membered

Examples:

Cefazolin



Ceftriaxone



>there is no aromatic ring structure and phenylglycyl on both examples therefore it is not administered by mouth

>it has S substituted atom making it better antibacterial and hydrogen can be seen on the 7th position

>for the Z variable cefazolin has greater activity since it has 5-membered ring

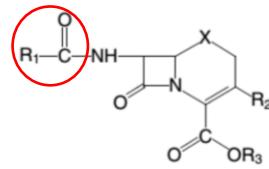
>one major determinant/basis in classifying cephalosporins is based on the route of administration

Oral Cephalosporins

>this can be achieved/produced if there is substitution on the 7th position of the 7-ACA core nucleus

- Oral activity conferred by **phenylglycyl substituent**

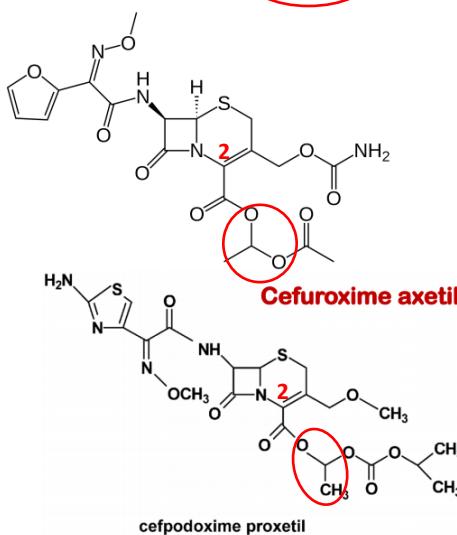
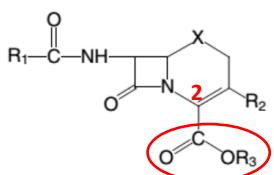
>if there is phenylglycyl substitution it can achieve acid resistance hence facilitating oral absorption



- Oral activity conferred by **esterification of 2-carboxylic acid group**

>on the 2nd position of the dihydrothiazine ring of the 7-ACA, originally it is Carboxylic group but when it is converted to its ester form (esterify), that also confers oral activity

→ Ex. Cefuroxime axetil and cefpodoxime proxetil



R₃ represents the 2nd position that is ginagawang ester yung carboxylic acid

X represents the 5th position of the dihydrothiazine that is heteroatom (S or O)

>all examples are in the form of phenylglycyl substituents having amino group, hydrogen and benzene ring that's why they confer acid-stability

Modified R1

- Cephalexin: oral inactivation that it undergoes is thru acid hydrolysis of b-lactam
- Cephradine: only cephalosporins that can be given by mouth or parenteral
- Cefadroxil: p-hydroxy phenylglycyl substituents
- Cefachlor: has almost the same structure w/ cephalexin only that instead of methyl it was replaced by chlorine atom, usually used in the tx of life-threatening infections caused by haemophilus influenzae that is resistant to ampicillin
- Cefprozil: has similar structure w/ cefadroxil but the difference is on the 2nd variable at the 3rd position, instead of methyl there is double bond
- Loracarbef: the X variable is not a heteroatom, it is already methylene and because of this, this is said to be chemically stable in the plasma

Modified R3 (carbon position 2)

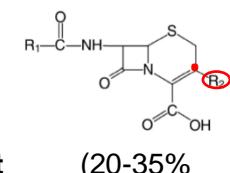
- Cefuroxime axetil: it has 1-acetoxyethyl ester form of cefuroxime which is an oral prodrug
- >to activate it to cefuroxime, it has to undergo hydrolysis
- Cefpodoxime proxetil: isopropyl carbonylethyl ester of cefpodoxime
- Cefixime: is not an ester prodrug but it has good oral absorption

>it lacks an amino group that is essential to make the drug stable

PARENTERAL CEPHALOSPORINS

>susceptible to hydrolytic activity by renal and hepatic esterase's

- Hydrolysis of the ester function is due to the presence of **3-acetoxyethyl substituent** (20-35% inactivation)



>presence of an ester group at the 3rd position makes this group hydrolysable but it does not seriously compromise the in vivo effectiveness of parenterally administered cephalosporins

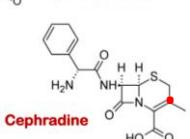
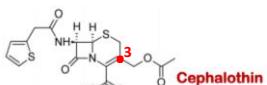
>esterified forms of cefuroxime and cefpodoxime
>on the 2nd position, instead of carboxylic acid substituent it became ester substituent

Specific examples of cephalosporins that orally administered:

Generic Name	R ₁	R ₂	R ₃	X
Cephalexin		-CH ₃	-H	-S-
Cephradine		-CH ₃	-H	-S-
Cefadroxil		-CH ₃	-H	-S-
Cefachlor		-Cl	-H	-S-
Cefprozil		-CH=CHCH ₃	-H	-S-
Loracarbef		-Cl	-H	-CH ₂ -
Cefuroxime axetil		-CH ₂ OCONH ₂		-S-
Cefpodoxime proxetil		-CH ₂ OCH ₃		-S-
Cefixime		-C=CH ₂	-H	-S-

R₁ represents the substitution on the left side usually on the phenylglycyl substituents at position 7

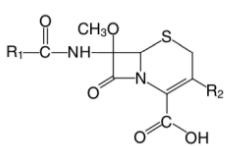
R₂ represents the 3rd position of the dihydrothiazine ring



>cephalothin is ester containing at the 3rd position that's why it is parenterally administered

>cephradine can be parenteral and oral because it is resistant to hydrolysis by renal and hepatic esterases

Generic Name	R ₁	R ₂
Cephalothin		
Cephapirin		
Generic Name	R ₁	R ₂
Cefazolin		
Cefamandole		
Cefonicid		
Ceforanide		
Cefuroxime		
Cefotaxime		
Ceftizoxime		
Ceftriaxone		
Ceftazidime		
Cefoperazone		



Parenteral Cephamycins

>cephamycins are cephalosporins containing methoxy substituents at 7th position therefore, there is increased resistance against b-lactamases

Generic Name	R ₁	R ₂
Cefoxitin		
Cefotetan		
Cefmetazole		

Spectrum of Activity

- Broad spectrum
- >effective against gram-positive
- More resistant to b-lactamase
- Uniquely potent against *Klebsiella* spp.

Adverse Reaction

- Allergic and hypersensitivity (less likely observed compared to penicillin)
→ CROSS ALLERGINICITY
- Patients that are allergic to penicillin should not be treated with cephalosporins

Methylthiotetrazole (MTT) (substituent at 3rd position)

→ clotting difficulties and alcohol intolerance

Examples: Cefamandole, Cefotetan, Cefmetazole, Moxalactam, and Cefoperazone

>there are clotting difficulties because these agents can inhibit synthesis of vitamin K dependent clotting factors (X, IX, VII, II) (prone to bleeding)

>they have significant drug interaction with warfarin

>they cause alcohol intolerance due to accumulation of acetaldehyde it is called disulfiram-like reaction

>disulfiram is a drug that inhibits aldehyde dehydrogenase enzyme and by inhibiting it that concludes that there is increased in acetaldehyde in patients taking disulfiram

Classification

- Based roughly on their time of discovery and antimicrobial properties
- Progression associated with
 - broadening Gm- spectrum
 - some reduction with Gm+, and
 - resistance to b-lactamases
- 1st gen Gm+
- Not effective in MRSA
- 2nd Gen Gm+, H. influenzae
- 3rd Gen Gm-, nosocomial resistant (hospital acquired)
- 4th Gen Gm-, Enterobacteria

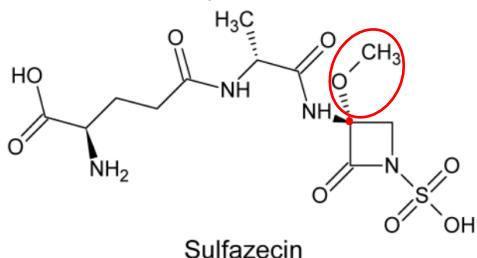
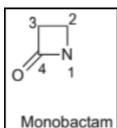
MONOBACTAMS

>drugs containing only β -lactam ring

- Extensive SAR studies on sulfazecin

Methoxy group at C3

→ stable at β -lactamase but low activity and chemical stability



>weak agent that's why there is a modification

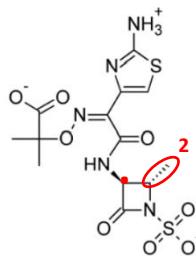
Aztreonam

- Totally synthetic parenteral antibiotic
- Methyl group at C2 provides stability to β -lactamases and activity to Gm-
- Exclusive for Gm – (PBP3)

Parts that were modified:

>methyl substitution at 2nd position was proven advantageous because it confers stability against β -lactamases and Gm- bacteria

>there is no methoxy group at the 3rd position



Protein Synthesis Inhibitors

>some of the most successful antibiotic families exert their lethal actions by inhibiting ribosomal mediated protein biosynthesis observed in bacteria

Ribosomes

>serves as the sites of protein synthesis

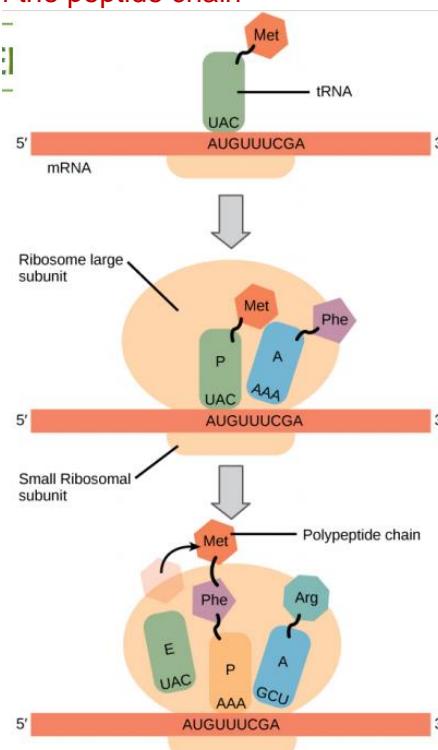
- Ribosomes are large complexes of protein and ribosomal RNA (rRNA), in which rRNA predominates
- Two subunits (one large and one small)
 - Relative sizes are given in terms of their sedimentation coefficients, or S (Svedberg) values
 - Prokaryotes: 50S and 30S (bacteria)
 - Eukaryotes: 60S and 40S (animal cell)
- Small ribosomal subunit (30S)
 - Determines the accuracy of translation by insuring correct base-pairing between the mRNA codon and the tRNA anticodon

>codon is found in the mRNA strand and the anticodon is being carried by tRNA in which a specific amino acid is being attached to it

>to ensure complementary we need the ribosomal subunit

- Large ribosomal subunit
 - Catalyzes formation of the peptide bonds that link amino acid residues in a protein

>formation of peptide bonds are the ones that connects amino acids residues, responsible for the growth of the peptide chain



Tigemonam

- Similar spectrum of activity with aztreonam but orally active

>mRNA strand that contains different codons, and the tRNA that pairs the anticodon with the corresponding amino acid that it possesses

>to ensure complementarity we need the small ribosomal subunit found below

>to facilitate growth/lengthening of the peptide chain, there must be an attachment of large ribosomal subunit

Inhibitors of Protein Synthesis

30s ribosomal unit

1. Aminoglycosides
2. Tetracyclines

50s ribosomal unit

1. Macrolides
2. Lincosamides
3. Chloramphenicol
4. Oxazolidinone
5. Streptogramins

AMINOGLYCOSIDES

>amino functional group and presence of sugar (hexose 6C, pentose 5C) that has ether functional group

>antibacterial agents that were isolated from streptomyces species

- Streptomycin – 1st AG used in therapy
 - Poorly absorbed orally (<1%)
 - Usually administered IM
 - Potent, broad spectrum
- >requires minimal dose to exemplify the effect, cover numerous strains of microorganism
- Ototoxicity (ears) and nephrotoxicity (kidneys) – common SE

Biopharmaceutics

- Strongly basic compounds

>due to presence of amino functional group

>bases are proton (H⁺) acceptors = they tend to be ionized (positively charge = cations)

→ Polycations at physiologic pH

>to be able to facilitate water dissolution for this agent, they are being prepared as sulfate salts

- Available as sulfate (acidic in nature) salt
- Very soluble in water

Pharmacokinetics (ADME)

- Absorption – poorly absorbed PO

>recommended to be administered parenterally via IM

>if taken by mouth, tendency its bacterial action is concentrated to GIT

>if the bacterial infection is “localized” (GIT is only affected) and if ever the strain is sensitive to aminoglycosides it could be given by mouth

- Distribution – distribute well into most body fluids
 - Except CNS, bones, fatty or connective tissues
- Metabolism – not metabolized in vivo
- Excretion – concentrate in the kidneys for excretion
 - >thru glomerular filtration process
 - >it can cause side effects in the kidneys which is nephrotoxicity

Mechanism of Action

General: impair proof-reading function of the ribosome

>there is misreading of codon expressed by mRNA causing abnormal protein being produced

>it is the only protein synthesis inhibitors that are bactericidal except spectinomycin because It does not cause misreading of codons

>depends on the dose

- Less than toxic doses (beneficial/therapeutic dose)
 - >targets the ribosomal subunit of bacteria
 - Bind to 16S ribosomal DNA portion of the 30S ribosomal subparticle
 - >because of targeting the 30S, there is a conformational change in the peptidyl A site found in the ribosome upon aminoglycoside binding that leads to mistranslation of RNA template so there is expression of non-sense proteins and because of it, it could destroy the semi-permeability of the cell membrane causing death (bactericidal)
 - Conformational change in the A site
 - Mistranslation = non-sense protein
- Increasing concentration
 - Protein synthesis ceases altogether
 - >there is no production of protein at all
- At even higher concentrations
 - Eukaryotic protein synthesis inhibition
 - >toxic
 - >dose matters

Spectrum of Activity

>the greatest usefulness of AG lies in the tx of serious systemic infections caused by:

- Reserved for use against **Aerobic Gm- bacilli**
- Streptomycin – tx of TB, brucellosis, tularemia, and *Yersinia* infections
- Paromomycin – tx of amebic dysentery

- Spectinomycin – tx of gonorrhea

>although AG have broad spectrum of action, there are also other strains that are less sensitive to AG
Examples of microorganisms that are less sensitive to AG thus limiting the application of these drugs:

- Gm- and Gm+ Coccis
- >to be able to exert more powerful effect they are being combined with another bactericidal drugs (Penicillins)
- Synergistic with B-lactam antibiotics
 - Carbenicillin + Gentamicin
 - Pen G + Streptomycin
 - Increased penetration of AG

*Chemically incompatible

Chemistry

1) Pharmacophore

>the one that would be responsible on the antibacterial effect

>active group of AG

- 1, 3-diaminoinositol moiety

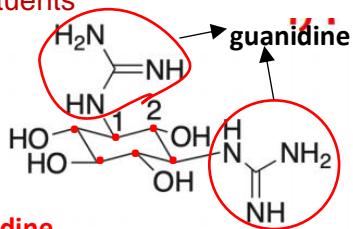
>inositol is cyclohexane hexol (all carbons have attached OH group)

>inositol of AG has some variations; this is where the pharmacophore of AG was derived

>when looking for the pharmacophoric group in the structure of AG, look first on the cyclohexane then check the substituents if it's derived from 1,3-diaminoinositol moiety

- 1,3-diaminocyclohexane

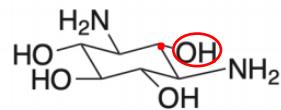
>with some modifications on the substituents found in the inositol moiety, we can come up with different variations depending on the attached chains/substituents



Streptidine

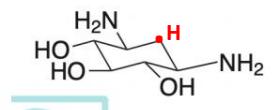
N^1,N^3 -bis(aminoimino-methyl)streptamine

>there is modifications/substitution to other OH groups wherein on the 1st and 3rd carbon there is guanidine group



Streptamine

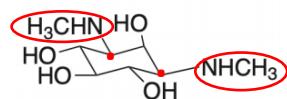
>on the 2nd carbon there is a presence of OH group



2-Deoxystreptamine

>on the 2nd carbon there is no OH, it is replaced by hydrogen only

Dimethyl amino (1,3)



Spectinamine

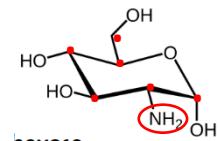
>modification at the 1st and 3rd carbon on the cyclohexane group

>it is replaced by methylamino that's why it is dimethyl amino

- Deoxystreptamine – kanamycin, neomycin, gentamycin, tobramycin
- Streptidine - streptomycin

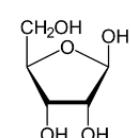
2) Amino sugars

- All have at least one aminohexose (sugar portion)



>amino hexose is a 6-carbon sugar that has attached amino group

- Pentose without amino group (RIBOSE)

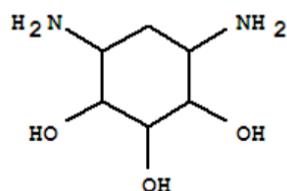


>5-carbon sugar

- Streptomycin, neomycin, paromomycin

Note:

Several of the alcoholic functions of the 1,3-diaminoinositol are substituted through glycosidic bonds with characteristic amino sugars to form pseudo-oligosaccharides

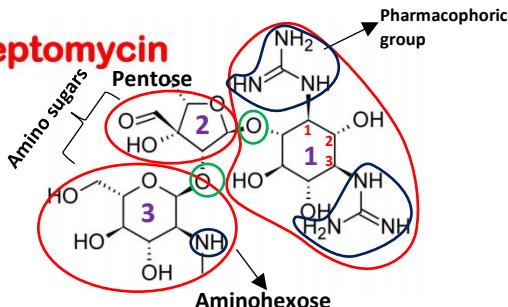


>the pharmacophore of AG is inositol moiety that has OH group that would form a bond with the sugar moiety, tendency because of the attachment

between the sugar and the pharmacophore it will form an ether linkage resulting to AG

Examples:

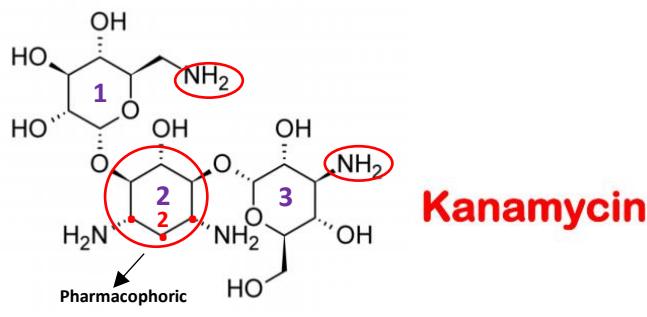
Streptomycin



Guide: search for the pharmacophore (not hetero cycle only carbon) group and the amino sugar
 >on the 1st and 3rd carbon there is guanidine substitution that's why the pharmacophoric portion of streptomycin is streptidine

>the 1st ring is the pharmacophoric group, the 2nd and 3rd ring is the amino sugars in which the 3rd ring is the aminohexose which has 6-carbon and amino and the 2nd ring is the pentose where it only has 5-carbon and no amino group

>between the pharmacophore and amino sugar, there is a glycosidic bond (ROR/ether)



Kanamycin

>the 1st and 3rd carbon has amino group
 >on the 2nd carbon only H that's why it is deoxystreptamine
 >on the 1st and 3rd ring there is no pentose, it is both aminohexose

Bacterial Resistance

>bacterial resistance is very common on AG that would render the drugs less effective

>because of bacterial resistances that would prevents ribosomal binding of AG

- Due to bacterial elaboration of R-factor mediated enzymes = metabolism of aminoglycosides

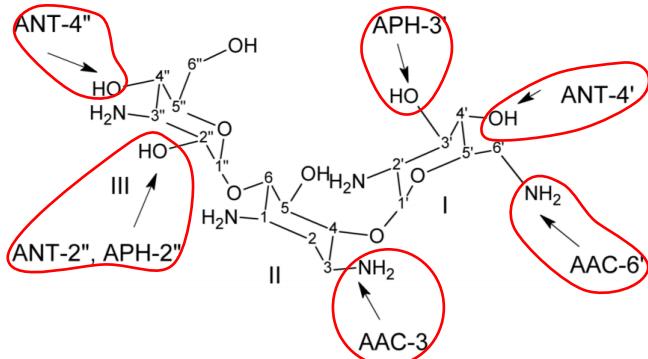
>the mechanism of bacterial resistance is synthesis of enzymes that would metabolize AG

- Aminoglycoside acetylase (AAC)
 - >can promote N-acetylation
- Aminoglycoside phosphorylase (APH)
 - >it can O-phosphorylate a certain FG present in the structure of AG

→ Aminoglycoside nucleotide transferase (ANT)

>it can O-adenylylate

>it is important to recognize the target groups of the enzymes because thru med chem, we can modify that group for it to be resistant to the enzyme action



>1st ring is an example of amino sugar enzymes that can target ring I:

>acetylase, this could target the 6 prime amino group rendering the drug less effective or ineffective
 >transferase enzyme, this could target the 4 prime OH

>phosphorylase enzyme that could target the 3 prime OH

>on ring II: the acetylase will target the amino group on the 3rd carbon

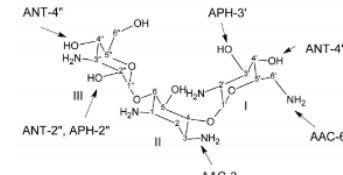
>on the ring III: the nucleotide transferase and phosphorylase will target the OH group at the 2nd carbon, and another nucleotide transferase will target the OH group at the 4th carbon

SARs

Ring I

>amino sugar that is also responsible for the broad spectrum of action held by AG

- Broad-spectrum activity
- Target for inactivating enzymes



Modifications that can be done:

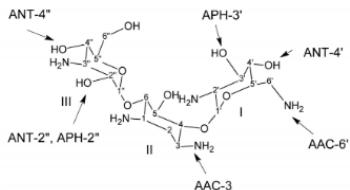
>addition of methyl group at the 6th carbon or 6th prime amino positions does not appreciably lower the antibacterial activity but confers resistance against the enzyme (acetylase)

>removal of OH group is applicable without compromising the antibacterial potency on the 3rd carbon (by removing the OH group it confers resistance w/o compromising the antibacterial effect)

Ring II

>pharmacophore

- Few modifications are possible without appreciable loss of activity



Modifications

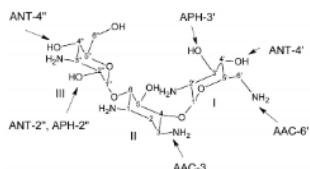
>the amino group at carbon 1, there is an example of AG that modified the amino group which is Kanamycin A

>in kanamycin A, the 1 amino group was acylated

Ring III

>slightly not changed

- Functional groups are less sensitive to structural changes than the two rings



Bacterial sources of AG

Micromonospora (-Mycin)

- Gentamicin (*M. purpurea*)
- Netilmicin (*M. inyoensis*)
- Sisomicin

Streptomyces (-Mycin)

- Streptomycin A (*S. griseus*)
- Neomycin (*S. fradiae*) – most useful for GI
- Paromomycin (*S. rimosus*)
- Kanamycin (*S. kanamyceticus*)
- Amikacin – acylation product of kanamycin
- Tobramycin (*S. tenebrarius*)
- Spectinomycin (*S. spectabilis*)

Products (as sulfates)

- Streptomycin
- Neomycin
- Paromomycin
- Kanamycin
- Amikacin
- Gentamicin
- Tobramycin
- Netilmicin
- Sisomicin
- Spectinomycin

TETRACYCLINES

>obtained from fermentation procedures

>static agents only

- From *Streptomyces* spp.

>further chemical modifications had resulted in the formulation of different derivatives related to the parent structure

MOA:

- Bind to 30s ribosomal subunit → prevent aminoacyl tRNA binding → stops protein chain formation

>tRNA is the RNA type that pairs/carries specific amino acid that is equivalent to the codon from mRNA

>when the action of tRNA is prevented, that would inhibit peptide chain elongation, the tendency is it would prevent the protein synthesis

Spectrum of Activity

- Tetracycline have the broadest spectrum of activity of any known antibacterial agents
- Active against gram positive and gram-negative bacteria, *Spirochetes*, *Mycoplasma*, *Rickettsiae* and *Chlamydiae*
- Not effective against life-threatening infections

>not effective against septicemia, endocarditis and meningitis

- May include superinfections caused by *Candida albicans* (yeast)

>because of its effectiveness against natural intestinal bacterial flora, the tendency, they can eventually inhibit the growth of those bacterial cells

>there will be sensitivity or susceptible of intestine against some opportunistic infection

Adverse Effects

- Phototoxicity

>in the skin

- Tooth staining

>teeth mottling

- Kidney damage → azotemia (high blood urea nitrogen levels and increase serum creatinine levels)

>patients taking tetracyclines (long period) these laboratory measurements are being conducted to check if the drug causes nephrotoxicity

- N & V, diarrhea

>gastrointestinal mediated toxicity

- CNS effects

- Thrombophlebitis

- Significant antianabolic effect

Contraindications

>situations wherein use of tetracyclines must be avoided by a patient

- Children

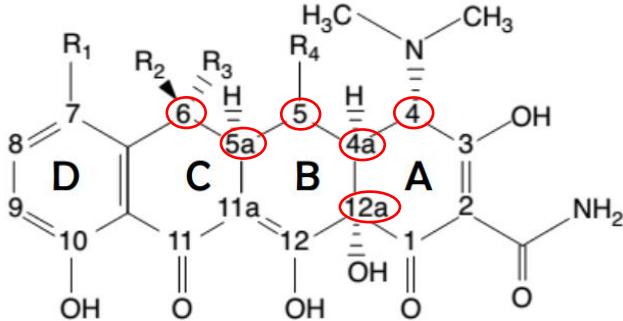
Drug Interaction (Pharmacokinetic)

- Complexation with metals
- >polyvalent metals (divalent and trivalent)
- Alteration in drug absorption
- >tetra means 4, cyclines means cyclic ring structure
- >tetracyclines are derivatives of:

Octahydroronaphthacene

>hydrocarbon system

→ (four annulated 6-membered rings)



>there are numerous carbons that are considered to be chiral

>there are 6 potentially chiral centers

>it depends on the variable side chain that is attached on the chiral carbons

- At position 4, 4a, 5, 5a, 6, 12a – chiral carbons
- Oxytetracycline and doxycycline → 6 chiral carbons

>only tetracyclines that have 6 chiral carbons

- Others → 5 chiral carbons (lack chirality at C5)

>at carbon 5 the substituted is OH group, it has 5-hydroxyl substituent making it a chiral carbon

TABLE 8.6 Structures of Tetracyclines

	R ₁	R ₂	R ₃	R ₄
Tetracycline	H	OH	CH ₃	H
Chlortetracycline	Cl	OH	CH ₃	H
Oxytetracycline	H	OH	CH ₃	OH
Demeclocycline	Cl	OH	H	H
Methacycline	H	CH ₂	H	OH
Doxycycline	H	CH ₃	H	OH
Mimocycline	N(CH ₃) ₂	H	H	H

>there are variables side chains that are allowed to be a substitute depending on the specific examples

>there are 4 potentials substitution depending on what carbon

Chemical properties

• Amphoteric

- could act as either acids or bases (proton donor or proton acceptor) depending on the situation
- >depending on what environment it will be exposed
- 1. neutral solution (neutral pH)

>zwitterions (provide proton transfer to the ionizable groups)

2. acidic pH

>this could be achieved by protonation of the enol group found at carbon 2 of TC

>enol group will accept proton

>enol has OH group with double bond

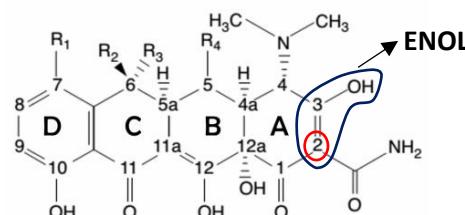
>HCl provide the proton to the enol group = HCl salts

3. basic pH

>TC will donate proton

>usually metals but not stable (not suitable preparation)

- Commercially available: HCl salts



Chemical properties: CHELATION

>formation of chelates

>when the chelates are formed it is non-absorbed compounds that's why the drug cannot enter the bloodstream/SI

>a drug that undergoes chelation will be less effective (reduction of efficacy)

- Form stable complexes with polyvalent metals (Fe²⁺, Al³⁺, Mg²⁺)

>this happens because the acidic functions of TC serves as proton donors to be accepted by the metals

>provides chemical and clinical implications:

- Incompatibility in solution preparation
- Interferes with blood levels on oral administration
- Drug interaction

>w/ antacids, FeSO₄ = TC will be less effective
→ Teeth discoloration

>teeth are made up of calcium

>teeth mottling for children

Chemical Properties: EPIMERIZATION

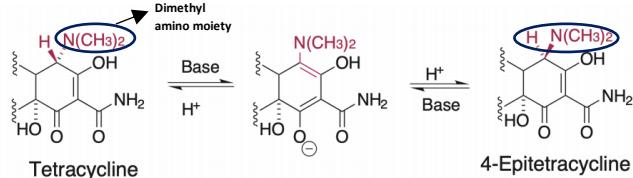
>process wherein a stereo center (chiral carbon) can exist in forms different in absolute configuration

>product: formation of epimers (like twins but have single point of difference in their configuration)

>100% identical but have 1 difference in the rearrangement of atom

>the susceptible in epimerization is 1 chiral carbon only which is carbon 4 (dimethyl amino moiety)

- Occurs at intermediate pH: C4 = 4-EPISETETRACYCLINE



>the dimethyl amino moiety at active TC is arranged away from you while the Hydrogen is towards
 >if its exposed at intermediate pH, there could be epimerization at carbon 4 wherein there is rearrangement of orientation of dimethyl amino and hydrogen such that the product would become hydrogen being at the back while dimethyl amino moiety is found in front

>4-Epitetracycline is an inactive form of TC
 >when TC undergoes epimerization, it becomes inactive

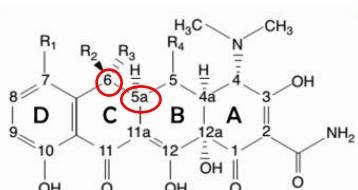
Chemical properties: DEHYDRATION

- By Strong Acids:

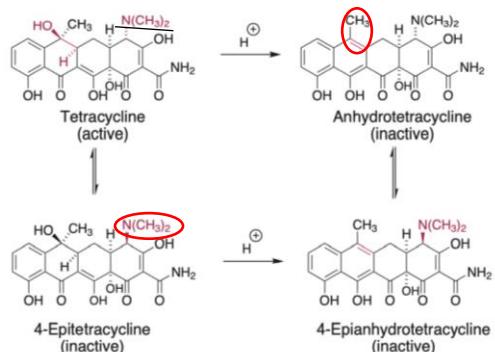
>groups/carbon susceptible to the attack:

- 6-hydroxyl group & 5a-hydrogen
- naphthalene group in

ANHYDROTETRACYCLINES



>thru the attack of strong acid, that causes formation of naphthalene derivative that is called anhydrotetracyclines which is also inactive
 >if there is a formation of anhydrotetracyclines, (TC is yellow in color originally) it became deeper yellow



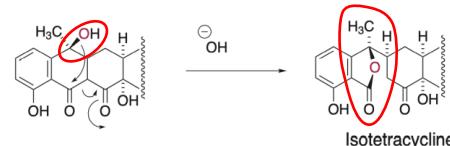
>tetracycline is active, in the carbon 4 there is dimethyl amino at the back and via epimerization it goes to the front that became 4-epitetracycline which is inactive

>when exposed to strong acid which is proton donation from the environment, the affected is carbon 6 and 5a that causes formation of naphthalene derivative called anhydrotetracycline that is inactive

- By Strong Bases (pH 8.5):

- Susceptible portions: 6-hydroxyl group & ketone group at C11
- lactone ring in ISOTETRACYCLINES (inactive)

>proof that there is isotetraacyclines is there are formed lactone ring



Chemical properties: PHOTOTOXICITY

>target the skin especially those C-7-chlorine substitution

- Tetracyclines with C-7-chlorine absorb light in the visible region
 - Causes sunburn
- or redness called erythema

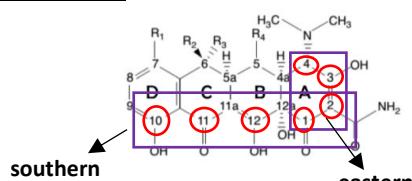
Structure Activity Relationships

>there is non-modifiable portion and modifiable portion

>non-modifiable portion, these are the parts of the TC that should not be modified or substituted because it could lessen or remove all the activity

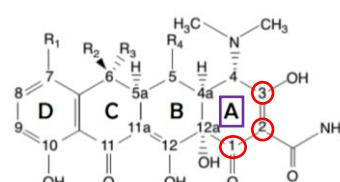
>modifiable portion, it can have substitution to improve the features/properties of TC

Non-modifiable



- Carbons 1,2,3,4,10,11,12 cannot be violated
 - HYDROPHILIC FACES

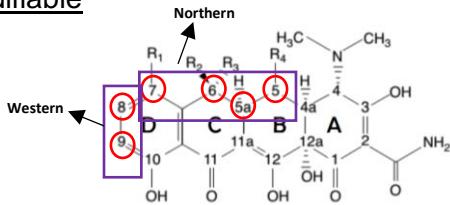
>southern and eastern faces of TC



- Ring A can be modified slightly keeping C1, C2, C3 intact
- Amide at C2 is removed → LOSS OF ACTIVITY

>do not violate the hydrophilic faces

Modifiable



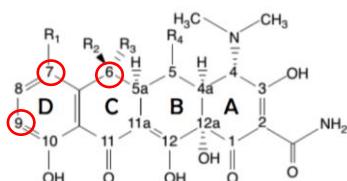
- Carbons 5,5a,6,7,8,9 can be modified
→ HYDROPHOBIC FACES

>northern and western faces of TC

>majority/all of the potential substitutions of R-group can be seen in the hydrophobic faces

>R1 at carbon 7, R2 & R3 at carbon 6, R4 at carbon 5

>the purpose of modifying is to improve the activity of TC



>Well established substitution for greater features of TC:

- Substitution at C7 and C9 → acid stability
 - The most fruitful site for modification → C6
- >either the absence of OH or presence of OH
- >absence of OH in C6 is better because if the C6 is deoxy in nature that would enhance antibacterial activity of TC, greater kinetic profile (inhibited of dehydration), greater lipophilicity by the drug (high partition coefficient)
- Polar substitution at C5 and C6 → increases water solubility
- >presence of OH, that could lower partition coefficient (greater water solubility)

Specific examples:

- Tetracycline
- Rolitetracycline
- Chlortetracycline
- Oxytetracycline
- Methacycline
- Demeclocycline
- Meclocycline
- Doxycycline
- Minocycline

MACROLIDES

- Isolated from *Actinomycetes spp.*
- PICROMYCIN – first identified
- Erythromycin and Oleandomycin – commonly used
- Spiramycin, josamycin, rosamicin – inferior to erythromycin
- Clarithromycin and Azithromycin – superior pharmacokinetic properties to erythromycin
- Chemically unstable in acid (stomach) – ENTERI-COATED TABLETS

Mechanism of Action

- Binds selectively to 50s ribosomal unit to prevent translocation step in bacterial protein synthesis (BACTERIOSTATIC)

>50s ribosomal subunit is responsible for the elongation of growing peptide chain and one of the processes needed to facilitate elongation of peptide chain is to provide translocation process wherein the large and small ribosomal subunit will slide/move for them to go to other codons that is not translated yet
>if translocation process is inhibited, protein synthesis is stop that's why this agent is effective antibiotics

>compare to AG, macrolide is only bacteriostatic

- Does not bind to mammalian ribosomes
- >selectively toxic

Therapeutic Application

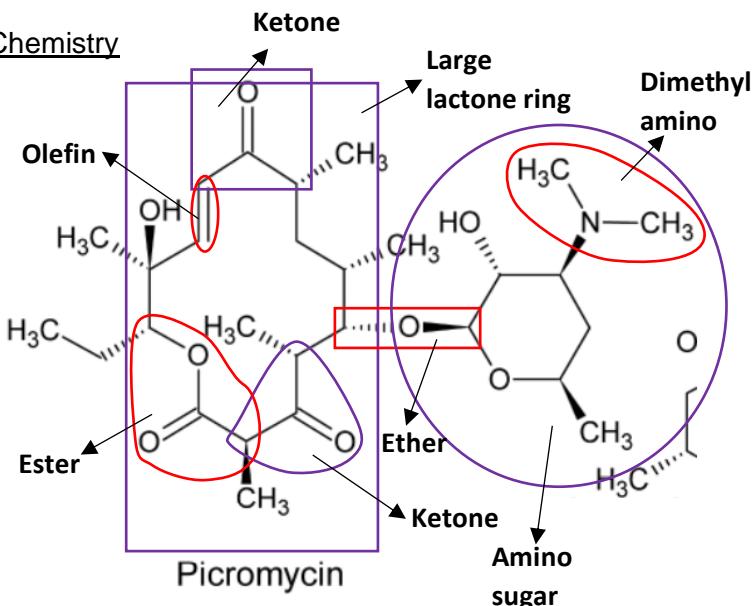
>tx of infections caused by bacterial strains that are penicillin resistant

- Active against bacterial strains that are resistant to penicillins
 - G+ bacteria and some G- bacteria, especially *Neisseria spp.*
 - Useful against *Treponema pallidum*
 - Effective against *Mycoplasma*, *Chlamydia*, *Campylobacter* and *Legionella spp.*

Attributes:

- Among the safest antibiotics in common use
- >because they are selective in targeting bacterial protein synthesis process
- Lower and upper respiratory tract
 - Narrow spectrum

Chemistry

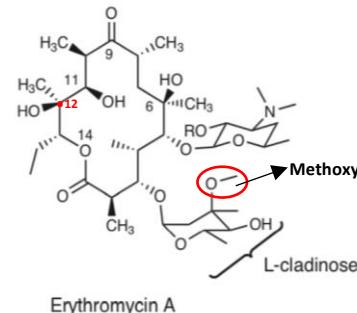


>Macro (large) lide – derived from the characteristic macro ring structure

1. Large lactone ring (cyclic ester (RC=OOR)))
- >there is characteristic number of atoms found in the large lactone ring
- >it can vary from 12, 14, 16 carbon atoms
2. Ketone groups (RCOR)
3. Glycosidically linked amino sugar (desosamine/glycone portion)
- >if there is presence of sugar, it can be linked to the lactone ring via glycosidic bond
- >ether FG serves as bridge between the glycone portion and aglycone (lactone ring)
- >distinct: presence of olefinic group/unsaturated group to the lactone ring that can be conjugated/adjacent to the ketone group
- >in desosamine portion there is presence of dimethyl amino group that renders macrolides basic ($\text{pK}_a = 6\text{-}9$)

Specific Agents

- Erythromycin (*S. erythraeus*)
 - **Erythromycin A:** commercial product
 - >characterized by having an OH group in the C12 of the lactone ring
 - **Erythromycin B:** more acid stable, less active than A
 - >have Hydrogen atom only at carbon 12 of lactone ring
 - **Erythromycin C:** equally active with A, but minimal percent yield during fermentation
 - >medyo hindi sustainable kasi maliit ang nasysynthesize
 - In the cladinose moiety
 - >A possesses methoxy group (OCH_3)
 - >C atom hydrogen only



>at carbon 12 there is presence of OH group, in the cladinose moiety there is presence of methoxy

- Erythromycin A
 - Active against URTI, soft tissue infections, and venereal diseases
 - Very bitter: formulation of EC and DR dosage forms
 - Extremely unstable at a pH of 4 or below (acidic)

>apply in the stomach

>can cause abdominal problems or stomach problems

- Chemical modification of Erythromycin Goals:

>to fill the gap

- 1) To increase water solubility or lipid solubility for parenteral dosage forms
- 2) Increase acid stability for improved oral absorption

- Derivative types (Erythromycin):

>Formulation:

- a) Acid salts of the dimethylamino group of desosamine moiety

Example of erythromycin salt that are acid salts:

>erythromycin glucoheptonate

>erythromycin lactobionate

>erythromycin stearate

- b) Esters of the 2'-hydroxyl group of desosamine

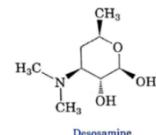
Examples

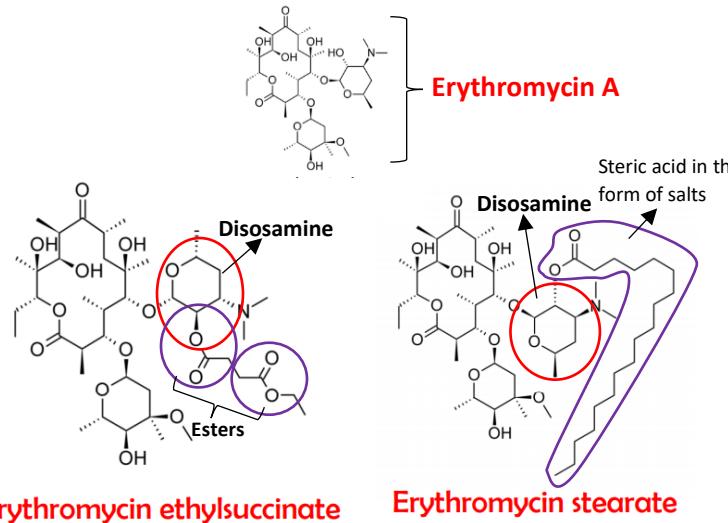
>erythromycin ethylsuccinate

>erythromycin propionate

>erythromycin estolate

>desosamine is the sugar portion of macrolides, the modifications can be seen here





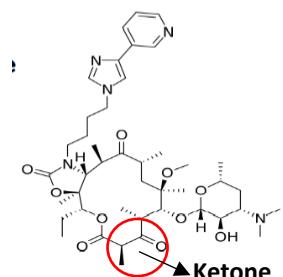
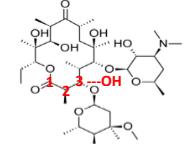
- Administered at least 1 hour before or 2 hours after a meal
- Food decreases absorption

- **Dirithromycin** – prodrug (EC)
- **Troleandomycin** – triacetyl derivative of Oleandomycin (2 sugars and a 14-membered lactone ring)

- **Ketolides**

- Oxidation of 3 position from alcohol to ketone
- Active against erythromycin-resistant microorganisms

- **Telithromycin (Ketek)**



LINCOMYCINS/LINCOBACAMIDES

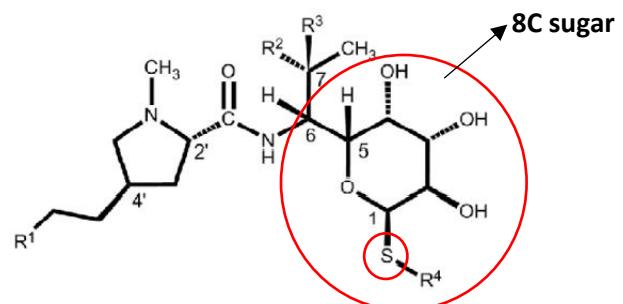
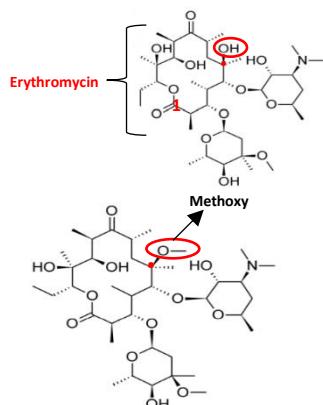
- Sulfur-containing antibiotics isolated from *STREPTOMYCES LINCOLNENSIS*

>there is sulfur atom in the structure

- Lincomycin: most actively and medically useful compounds obtained from fermentation
- 8C sugar (O-thio-lincosamide)
- Weakly basic, as HCl salts

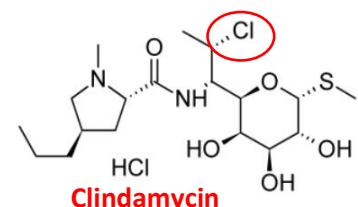
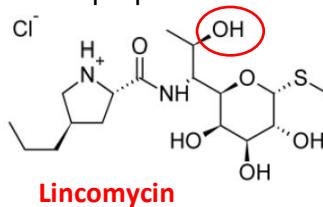
>formulate into HCl salts to permit water solubility

- Binds to 50s ribosomal unit (MOA)



- **Lincomycin**

- **Clindamycin** (more commonly used)
 - 7-chloro-7deoxy derivative of lincomycin
 - Has greater potency and pharmacokinetic properties



>**stearate**: in the disosamine portion there is a formation of acid salt or steric acid in the form of salt

>long hydrocarbon chain

>**ethylsuccinate**: in the disosamine sugar at the 2nd hydroxyl group, from being an OH it became esters

- **Erythromycin**

- Stearate, ethylsuccinate, propionate salts (PO)
- Ethylsuccinate (IM)
- Glucoheptonate and lactobionate salts (IV)
- Adverse effects: T/E: Abdominal cramps, epigastric distress, diarrhea

- **Clarithromycin**

- 6-methyl ether of erythromycin

>there is methylation at 6th hydroxyl group of lactone ring to enhance acid stability

- Part of cocktail in tx of gastric ulcer due to *H. influenzae*

- **Azithromycin**

>unique: its lactone ring is called azalide

- Contains an azalide structure

>azalide is nitrogen containing and 15-membered ring structure

Modification:

- Removal of 9-keto group coupled with incorporation of a tertiary amine nitrogen into the macrolide ring

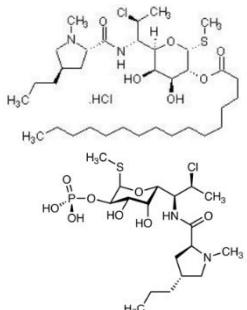
>the nitrogen is a tertiary amine that confers acid stability

- Broader spectrum, first choice

Clindamycin

- For URTI, skin and tissue infections and Bacteriodes infections
- Salts: palmitate HCl, phosphate
- T/E: GI Toxicity

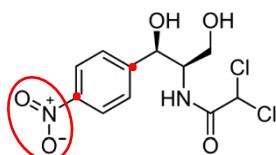
Adverse effects: diarrhea, pseudomembranous colitis (cause by over growth of clindamycin resistance strain of *Clostridium difficile*)
Tx: vancomycin, metronidazole



Unclassified Antibiotics

CHLORAMPHENICOL

- Isolated from *Streptomyces venezuelae*
- Contains p-nitroacetophenone
- Metabolism: 3-O-glucuronide (metabolic product that is ready for excretion)
- Inhibits 50s of the ribosome (-static) (MOA)
- Salts: palmitate, sodium succinate
- T/E: Aplastic anemia, gray baby syndrome

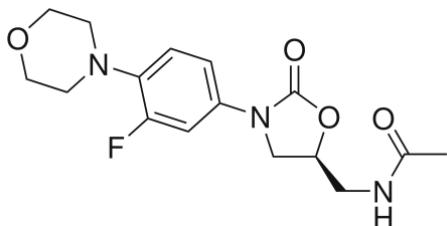


Oxazolidinone: LINEZOLID

- Bacteriostatic agent
- Newer synthetic class of antibacterials
- Inhibits initiation of translation by binding to the 50s subparticle = inhibits formation of functional initiation complex (N-formylmethionyl-tRNA)

>inhibiting the action of 50s ribosomal subunit thus blocking the functional initiation complex of translation process

>by inhibiting the action of the large ribosomal subunit, tendency there is no provider of peptide bond that's why the elongation of growing peptide chain is inhibited also



- Effective against VREF (Vancomycin Resistant Enterococcus Faecium), MRSA (Methicillin-resistant *Staphylococcus aureus*), CAP (Community acquired pneumonia) and nosocomial pneumonia (hospital acquired)
- T/E: Blood dyscrasias

- D/I: MAOIs (Monoamine oxidase inhibitors), SSRIs (Selective Serotonin Re-uptake inhibitors)

>linezolid is a MAO inhibitor

>if taken with another MAOIs or SSRIs, that can cause an increase levels of biogenic amines particularly increase of serotonin levels abnormally speaking causing serotonin syndrome (example of hyperthermic disorder)

STREPTOGRAMINS

- Quinupristin (Streptogramin B) & Dalfopristin (Streptogramin A) at 30:70 ratio via (IV)
- >the 2 drugs are bacteriostatic when administered individually but if combined they act synergistically (-cidal)
- Semisynthetic derivatives from *Streptomyces pristinaspis*
- MOA: bind to 50s ribosomal subunit
 - Quinupristin – binds at the same site as macrolides (similar effect)
 - Dalfopristin – binds to a site near that of Quinupristin

Inhibitors of Nucleic Acid Synthesis

QUINOLONES

>comprised of series of synthetic antibacterial drugs

- Patterned after nalidixic acid

>used in the tx of urinary tract infections

- Classified as

1. Quinolones (norfloxacin, ciprofloxacin)

>common commercially used

2. Naphthyridines (nalidixic acid)

3. Cinnolines (cinoxacin)

- Good oral absorption

- High urinary concentrations

- Activity against G-urinary pathogens

>which are usually causative agents to UTI

Mechanism of Action

>they have specific enzyme that is targeted to affect nucleic acid synthesis

- Inhibit DNA gyrase (topoisomerase II)

>DNA gyrase is a name applicable to bacteria and in humans it is equivalent to topoisomerase II

>topoisomerase II is an important enzyme during DNA synthesis, it is responsible in relieving strain or super coiling during unwinding of the double stranded DNA

>one of the important process for the DNA process to happen is it needed to unwind/unzip for it to be single stranded that would be used as templates

>associated with the unzipping is the distal portion of double stranded DNA (nagbuhohol-buhohol)

→ Promote supercoiling

>by inhibiting the enzyme topoisomerase II, there will be no enzyme anymore that is responsible in preventing supercoiling hence quinolones will promote supercoiling to the double stranded

>if its supercoiled, greater strain would inhibit ultimately the succeeding steps in DNA replication process

- Inhibits DNA Replication (-cidal)

>no more genes available to produce RNA, proteins that eventually cell will die

- Phototoxicity (side effect)

>affects the skin when exposed to natural sunlight causing sunburn or erythema

SAR:

- 1,4-dihydro-4-oxo-pyridinecarboxylic acid moiety is essential for antibacterial activity

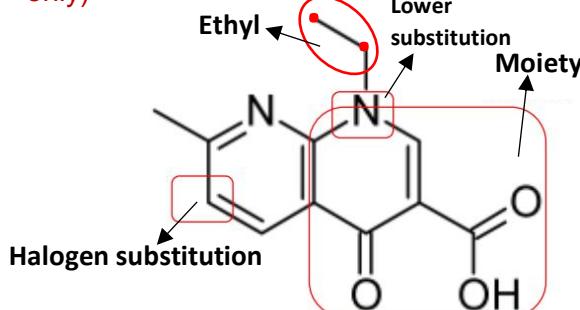
Modifications to intensify the antibacterial actions:

- Fluorine substitution at C6 → increase activity

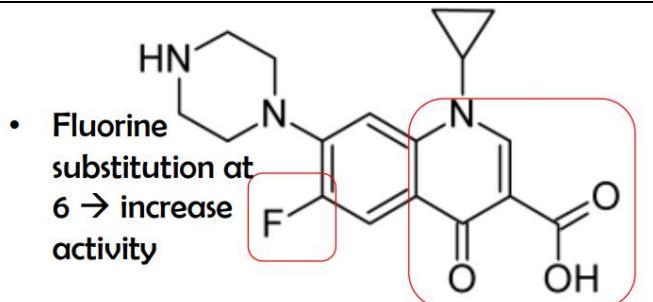
>fluoroquinolones, fluorine substituted

- Lower # of C/alkyl group at 1 → increase activity

>substitute or put an atom at position 1 like methyl group or ethyl or cyclopropyl (maximum of 3 carbons only)

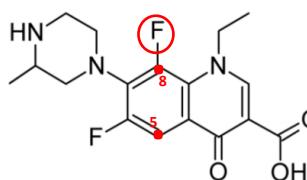


Ciprofloxacin

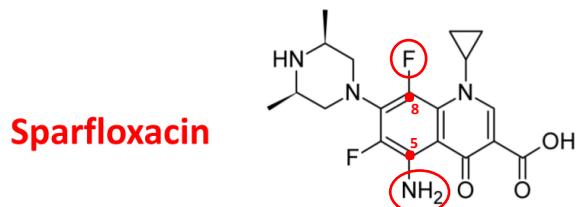


- 1,4-dihydro-4-oxo-3-pyridinecarboxylic acid

>at the first position, there is substituted cyclopropyl making it a potent quinolone



Lomefloxacin



Sparfloxacin

>potential phototoxicity that can be induced by using some examples of quinolones

>it can be detected if there is high incidence of phototoxicity at position 8

>there is high incidence of phototoxicity if there is halogen substitution at position 8

>way of decreasing or counter-balancing the incidence of phototoxicity is thru amino or methoxy substitution at position 5

>greater phototoxicity if seen in lomefloxacin while in sparfloxacin less incidence

>lomefloxacin is the most phototoxic quinolone while the sparfloxacin is the least phototoxic

Classification

- First Class → targets Gm (-) bacteria → lacks systemic activity
 1. Nalidixic acid (Negram)
 2. Oxolinic acid
 3. Cinoxacin (Cinobac) – more absorbed
- Second class → targets Gm (-) + Pseudomonas
 1. Norfloxacin (Noroxin)
 2. Enoxacin (Penetrex)
 3. Ciprofloxacin (Ciprobay)
 - best distribution into the different parts of the body specifically in the renal system
 4. Ofloxacin (Inoflox)
 - more bioavailable
- Third class → targets Gm (+) bacteria
 1. Lomefloxacin (Maxaquin)
 - Longer half-life, once daily
 2. Sparfloxacin
 - Less phototoxicity
 3. Levofloxacin
 - Isomer of ofloxacin
- Fourth class
 - Travofloxacin

ANTIMETABOLITES

>the ones that inhibit the normal metabolic process particularly those processes that are related to DNA synthesis

>agents that interfere the metabolism that are needed in DNA synthesis

Antibacterial Sulfonamides

- Synthetic antimicrobial agents
- First effective chemotherapeutic agents
- Comparatively classified as minor drugs today
- Relatively cheap

History

Mietzsch & Klarer
synthesized azo dyes

G. Domagk evaluated
Prontosil rubrum (dye)

→ Inactive in vitro, but
active in vivo

Trefouel & Bovet showed animal-treated urine
was bioactive in vitro.

p-aminobenzenesulfonamide
(sulfanilamide)

Foerster treated an infant suffering
staphylococcal septicemia (puerperal sepsis).

properties that's why greater attention was drawn to different dyes to extract more potential antimicrobial agents

>this path was continued, wherein he evaluates the dye prontosil rubrum and when he checked its antimicrobial, it is inactive in vitro but active in vivo (mice)

>another significant research was done, wherein they extracted the urine of drug treated rats

>the metabolite in the urine was the one they tested in vitro and they realized that the metabolite extracted from the urine is active in vitro and they called the active metabolite as sulfanilamide

>they realized that the drug can be effective once metabolized and that started the concept of prodrugs

Modern Era of Chemotherapy

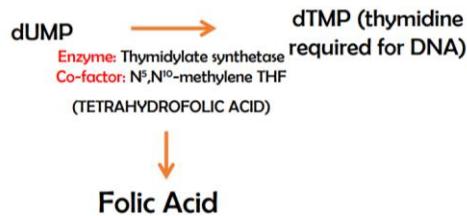
- Discovery of in vitro and in vivo antibacterial properties of sulfanilamides
 - Era of Sulfa Drugs (1935)
- Introduced the concept of prodrug
 - based on sulfanilamides journey

Mechanism of Action

>very important to recognize one of the important nucleotides in DNA

>in our DNA, there is one nucleotide that is essential in the synthesis is called deoxythymidine monophosphate

>for the dTMP to be produced, it came from a precursor deoxy uridine monophosphate



>for the dUMP to become dTMP it should be metabolized/convert using an enzyme

>the enzyme needed for the production of dTMP is called thymidylate synthetase

>thymidylate synthetase would be activated, provided that we have a co-factor (alay) called tetrahydrofolic acid (THF)

>THF is produced because in humans we have dietary source of folic acid

>bacteria cannot assimilate folic acid (bacteria cannot absorb/utilize) folic acid)

>there were 2 scientists, they were able to synthesize azo dyes containing sulfonamides functional group that have potential antimicrobial

Source:

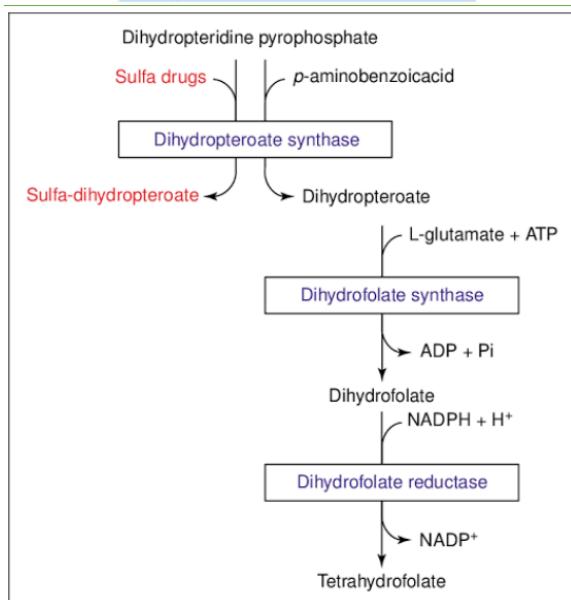
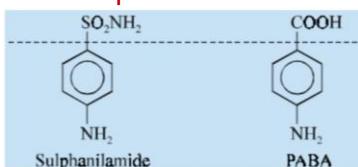
Folate coenzymes (humans)

→ FOLIC ACID

Folate enzymes (bacteria)

→ PABA + pteridine phosphate

>bacteria needed to produce THF



> the dihydropteridine pyrophosphate and PABA via the enzyme dihydropteroate synthase that causes formation of the intermediate product dihydropteroate

>the processes will continue until such time it will form tetrahydrofolate

>tetrahydrofolate is formed because dihydropteroate will be targeted/converted into dihydrofolate via the enzyme dihydrofolate synthase
>then dihydrofolate will become tetrahydrofolate via the enzyme dihydrofolate reductase

>then when it is tetrahydrofolate in bacteria, there will be utilized thymidylate synthetase for the bacteria to produced dTMP

>MOA: inhibit dihydropteroate synthase

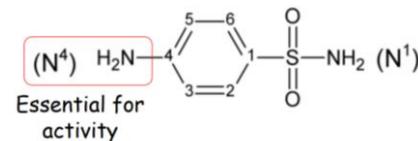
>so, no dihydropteroate, no tetrahydrofolate, no alay, no enzyme, no dTMP, no DNA synthesis

>most sulfonamide are aniline substituted

>methyl group is substituted by aniline

Generic Name		
		<chem>R1-SO2N-R2</chem>
Sulfamethoxazole	H	<chem>Nc1ccc(cc1)S(=O)(=O)N(C)C</chem>
Silver Sulfadiazine	Ag ⁺	<chem>Cc1ccncn1</chem>
Sulfisoxazole	H	<chem>Cc1cc(C)c(O)cn1</chem>
Sulfacetamide	H	<chem>CC(=O)c1cc(C)on1</chem>

Sulfanilamide



>very important for the activity of sulfonamides is the p-amino group that is from aniline

Side effects

- Crystalluria
- Rashes
- Anemia
- Nausea
- Kernicterus

Specific Examples:

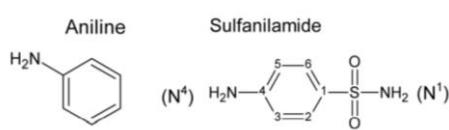
- Sulfomethiazole
- Sulfisoxazole
- Sulfamethazine
- Sulfacetamide
- Sulfachloropyridazine
- Sulfapyridine (1st drug in tx of pneumonia)
- Sulfamethoxazole
- Sulfadiazine

Combination/Mixed

- Trisulfapyrimidines
 1. Sulfadiazine
 2. Sulfamerazine
 3. Sulfamethazine
- Sulfadoxine + Pyrimethamine (Fansidar)
 - Tx of P. falcifarum malaria

Topical Sulfonamides

- Sulfacetamide – ophthalmic
- Sulfisoxazole
- Triple Sulfa (Femguard)
 - Tx of H. vaginitis



Non-absorbable

For Burn Therapy

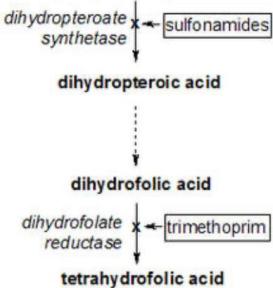
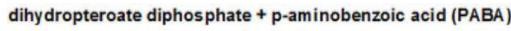
- Mafenide
- Silver sulfadiazine (Silvadene)
- Sulfasalazine

Trimethoprim

- Dihydrofolate reductase inhibitor

>important enzyme to produce tetrahydrofolate
 >usually combined w/ another sulfonamide which is sulfamethoxazole to provide synergistic effect (1+1=3, sum is more than 2)
 >when drugs are combined with the same activity, they can further enhance the certain effect

Co-trimoxazole (TMP + SMX) – increase antibacterial activity



Dapsone

- A sulfone
- DOC for leprosy

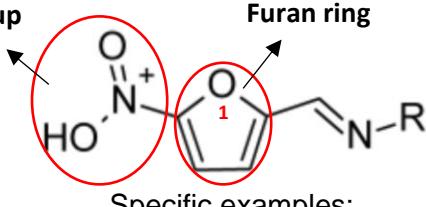
NITROFURANS

- First nitroheterocyclic compounds introduced

Mechanism of Action

- Reduction of nitro group forming free radicals → damage to ribosomal synthesis
- Antimicrobial activity is present only when the nitro group is in the 5-position

Nitro group



- Nitrofurazone
- Furazolidone
- Nitrofurantoin
- Nifurtimox
 - Antiprotozoal agent to treat trypanosomiasis and leishmaniasis
- Metronidazole
 - Amoebicide

METHENAMINE

>weak base

- Activity of hexamethylenetetramine
 - Liberation of formaldehyde
 - optimized with acidifying agent (combined)
 - *Ammonium chloride
 - *Sodium biphosphate
- Used as a urinary antiseptic for chronic UTI

Alteration of Cell Membrane

- Polymyxin
 - Polymyxin B Sulfate
 - *Bacillus polymyxa*
- Colistin
 - *Aerobacillus colistinus*
- Daptomycin

Anti-TB Drugs

- **Rifampicin/Rifampin**
 - Most active agent in clinical use
 - Inhibits RNA polymerase (important in transcription process)
 - Hepatotoxicity, orange to reddish brown urine
- **Isoniazid**
 - Inhibit Mycolic acid synthesis
 - *Mycolic acid – important components of the cell walls of *mycobacteria*
 - Isonicotinyl hydrazide (INH) – chemical name
 - Peripheral neuritis (side effect)
 - *Admin of pyridoxine Vit B6
 - >deficiency of vit b6
- **Pyrazinamide**
 - Pyrazinecarboxamide (PZA)
 - First-line status in short-term TB treatment regimens

>usual side effect: hepatotoxicity (greatest among the risk), hyperuricemia

- **Ethambutol**

- EMB
- Inhibits arabinogalactan synthesis

>important process/step in mycobacterial structure

- Active only against dividing mycobacteria
- Not recommended for use alone

>side effect: optic neuritis

- **Rifamycins** – *Streptomyces mediterranei*
- **P-Aminosalicylic Acid (PAS)**
- **Clofazimine** – basic red dye against *M. leprae*
- **Cycloserine**
 - Inhibits cell wall synthesis

- Recommended to patient who fail to respond other TB agents

Polypeptides

- Describe as cyclic peptides
- Most powerful bactericidal antibiotics characterized by presence of sever-CONH linkages
- S/E: neurotoxicity and nephrotoxicity

Vancomycin: from *Strep. orientalis* DOC for Pseudomembranous colitis

- Inhibits cell wall synthesis by binding to D-ala-D-ala terminus
- Resistance: replacement of terminal D-ala with d-lactate
- ADR: “red-man” syndrome: flushing due to histamine release caused by rapid infusion

Teicoplanin: *Actinoplanes teichomyceticus*

- MOA: inhibitor of cell wall synthesis affecting peptidoglycan layer

Bacitracin: from *Bacillus subtilis* enhance activity with zinc, topical use only (nephrotoxic)

Polymyxin B: *Bacillus polymyxa*

Colistin: *Aerobacillus colistinus*

Colismethane sodium

Gramicidin: *Bacillus brevis*

ANTIVIRAL AGENTS

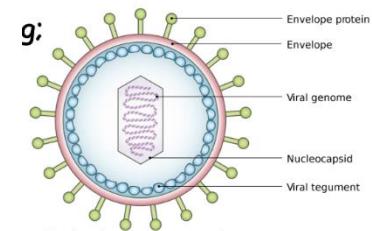
Introduction

Virus

- Smallest of the human infectious agents
 - A nucleic acid strand with associated proteins
- >DNA or RNA
- Arguably non-living
- >they cannot move on their own power
- >they are deemed as parasites because they utilize the host biochemical machinery for their replication purpose
- Relies on host's biochemical machinery

Viral Structure

- Simple, consisting;
 - a. Nucleic acid
- >genetic material than can be either DNA or RNA
- b. Capsid
- >structure comprised of both the nucleic acid plus the protein
- c. Lipid envelope

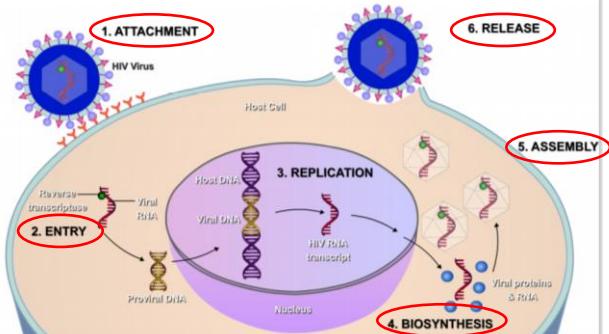


A complete viral particle is termed as **VIRION**

Virus Families

- | | |
|--------------------|----------------------|
| • Papillomaviridae | Warts |
| • Herpesviridae | Chickenpox, Shingles |
- >1ST onset (cp), recurrence will result to shingles especially for patients who are immunocompromise
- | | |
|-------------------|-------------------|
| • Poxviridae | Smallpox |
| • Togaviridae | Measles (Rubeola) |
| • Paramyxoviridae | Mumps |
| • Flaviviridae | Dengue Fever |
| • Retroviridae | AIDS (HIV) |

Viral Life Cycle



>it will start with the **attachment** of the virus to the cells surface receptors and this would cause entry of the virus

>upon **entry** of the virus, it would release its genetic material (DNA or RNA), and that would result in

biochemical events resulting in the formation of new DNA/RNA viral particles

>these biochemical events could be **replication** and then biosynthesis that would result again to a new viral particle

>and then after that is the **assembly** and finally **release**

- Attachment (1)

- - Intact virion binds to host cell through adsorption to a specific receptor site

- - >the receptor site being possess by the host's cell

- - >so, when the viral particle is attached on the receptor site, that would be the beginning step for the virions to enter inside the host

- - Example: HIV binds to CD4
 - >the HIV, to be able to provide attachment to the host's cell, this usually interacts with the receptor in the cell membrane called the CD4
 - >that's why it is important determinant in checking for the HIV in the patient is the measurement of CD4 because it is the attachment site of HIV
 - >once attached, there is already an opportunity for the virus/virion to enter the cell

- Penetration (2)

- - The virus gains entry into the cell via;

- - >after adsorption into the cell surface

- - Receptor-mediated endocytosis
 - >wherein after binding of the virion on the cell surface receptor, that would facilitate endocytosis

- - Fusion

- - Direct penetration

- - >thru penetration virus could now be available inside the cytoplasm of the host's cell

- Uncoating (3)

>from the virion, the genetic material of a virion will be release

- - Results to either naked nucleic acid or in the nucleocapsid form

- - >reveal of genetic material

- Eclipse Period (4-7)

- - >biochemical event

- - Time that the virus utilizes host resources to replicate and produce viral proteins

- - >certain entities/biomolecules from the host's cell is being used by the virus to

produce more, examples is different enzymes needed for gene expression that virus used from the host's cell

>we can classify the host cells whether they will be susceptible to the virus or not

>those cells that are susceptible, they can support viral reproduction are called:

→ **Permissive** cells → **PRODUCTIVE INFECTION**

>result to production of more viruses or more matured virion

- Can support viral reproduction

→ **Non-permissive** cells → **ABORTIVE INFECTION**

- Unable to support reproduction

>resistant to eclipse period

>central dogma is important in viruses because these are the processes wherein the genetic material of the virus would be replicated and reproduce

Example; Central dogma of molecular biology of viruses

>the nucleic acid composition the DNA of the virus was already exposed after uncoating

>the viral DNA would undergo DNA replication that results in the formation of new viral DNA

>replication is being attained because of certain enzymes and the important enzyme for replication is DNA polymerase enzyme

>although DNA polymerase is required for replication of viral DNA, this is not yet synthesized

>to be able to facilitate replication of the viral DNA, the DNA polymerase must first be produced/synthesize because there is no available DNA polymerase so, the viral DNA can't be replicated

How to make DNA polymerase that would be utilize/use to reproduce new viral DNA?

>the viral DNA must undergo early viral (viral DNA) gene expression (the portion of the DNA would be converted into mRNA and into protein)

>to make DNA polymerase, the viral DNA must be expressed first (make a mRNA from it and protein)

>the viral DNA would undergo gene expression first

>the viral gene (DNA) would undergo mRNA synthesis (transcription) and then from it, it would be protein via translation

>to be able for transcription to happen, it needed enzymes and other proteins and in translation, it also needed enzymes and other proteins

>why does early gene expression happen even if it uses enzymes and proteins?

>it is because the enzymes and proteins the that the virus used is from the host

>when the protein is already formed from the early gene expression, the one protein that will be make is DNA polymerase

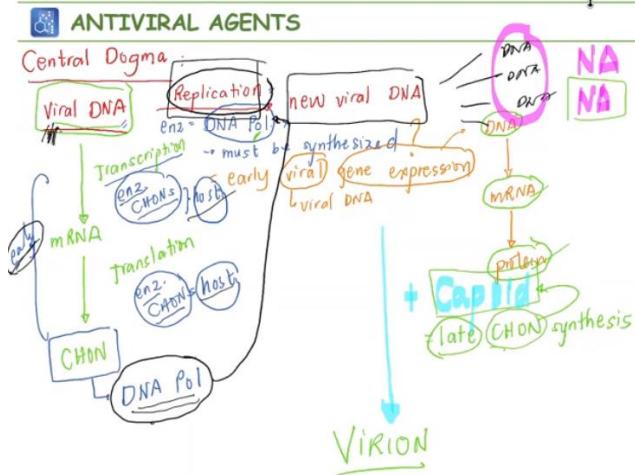
>because of the early gene expression, we tend to produce DNA polymerase that would be utilize for viral DNA replication that's why there is new viral DNA

>same process will continue for the DNA to produce a lot

>even if there are many viral DNA, it is needed to be encapsulate/covered by capsid

>for it to have additional capsid, it must undergo late protein synthesis

>so, when it is already produced, there will be a new virion that is ready to invade another host' cell



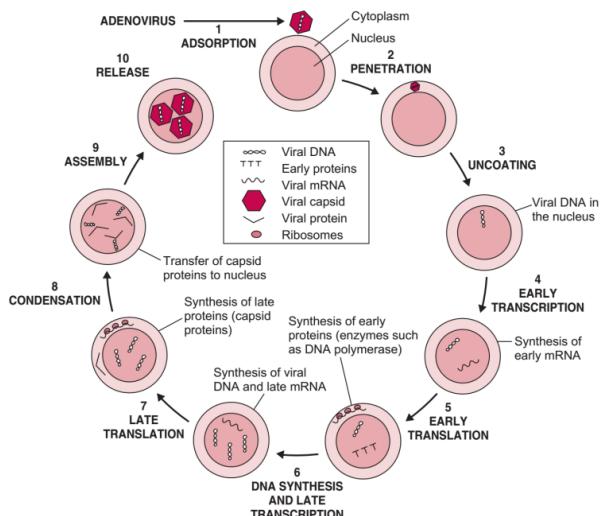
Condensation (8)

- Transfer of capsid proteins to nucleus
>mature virion that is ready to invade other host's cells

Assembly (9)

Release (Cellular Egress) (10)

- Through exocytosis or budding



Viral Diseases

HIV (Human Immunodeficiency Virus)

- HIV1 → cause of AIDS
- No successful drugs proven yet
>there are available drugs that could delay the progression
- Ability of virus to mutate → Resistance
>there are different chemical pathways that they can make/facilitate
- Contains
 - RNA genome
 - Reverse Transcriptase (RT)
 - Viral protease
 - Integrase

>usually in the commercially available antiviral agents, these drugs can target RT, protease and integrase

Reverse transcriptase

>this is an enzyme being possess by certain family of virus and those are retroviruses

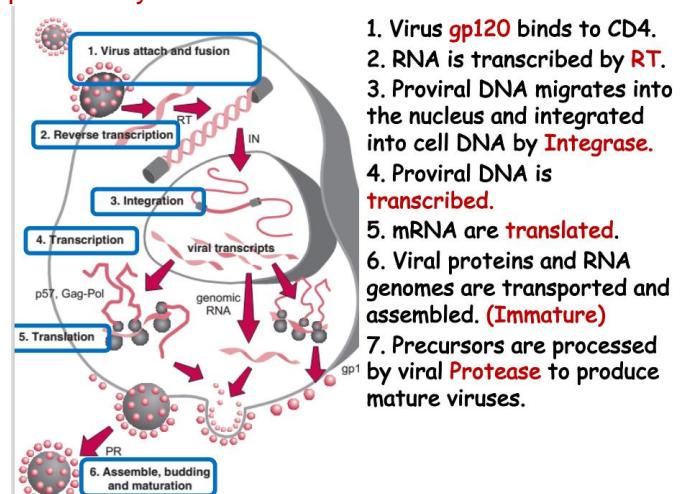
>the retroviruses are specialized viruses because they contain RT (RNA dependent DNA polymerase)

>RNA dependent means, the template that is used is RNA and the enzyme DNA polymerase that causes formation of DNA as its complementary strand

>the template is RNA and the product is DNA

>RNA → DNA (deviation of central dogma because the DNA will be RNA via transcription)

>if its reverse transcriptase inhibitors, it means they inhibit the formation of proviral DNA of retroviruses particularly HIV



- As viral replication continues, T-lymphocytes decrease → Compromised Immune System

>the immune system of an individual will be incompetent that's why common to HIV patients is the:

- OPPORTUNISTIC INFECTIONS

>cause by yeasts or fungi

- RT makes mistakes in reading viral RNA → MUTATION → change in surface structure
→ RESISTANCE

>solution is combination therapy

Herpesvirus

- Establish lifelong persistent infections with periodic reactivation
- Herpes Simplex Virus (HSV)

HSV1 – oral cold sores HSV2 – genital herpes

- Varicella virus – chicken pox

>normally when a patient recovered from chicken pox it will not reoccur, unless the patient is immunocompromise

- Herpes Zoster virus – shingles (reoccur)

>infection that has neuropathic pain (numbness)

- Cytomegalovirus (CMV) – mononucleosis-like infection

Hepatitis

- Hepatitis A Virus (HAV) – infectious hepatitis
→ Onset 24h, complete recovery occurs
- Hepatitis B Virus (HBV) – serum hepatitis
→ Chronic infections
- Hepatitis C Virus (HCV) – posttransfusion hepatitis
- Hepatitis E Virus (HEV) – transmitted enterically (feces contaminated water)

Influenza

- Respiratory illnesses (colds and flu)
- Genetic mutation on viral surfaces
- Influenza A – cause majority of epidemics
- Influenza B
- Influenza C – stable, only mild illness
- Influence virion structure
- Lipid envelope that contains:
 - HEMAGGLUTININ (HA) – for binding
 - NEURAMINIDASE (NA) – enhance adsorption (and penetration of the virus to the host cell)
>when there is penetration, that could result to uncoating and eclipse period will happen resulting to more reproduction of influenza virus
 - Important for the spread of infection
 - Determinant of antigenicity and host immunity

Tumor Viruses

>viruses that can cause cancer

- Cervical cancer (caused by HPV) and liver cancer
→ caused by papilloma virus
- Viruses
 - (1) Introduce “transforming gene” OR
 - (2) Alter gene expression

CELLULAR TRANSFORMATION leads to change in growth control → TUMOR

MOA: Viral Chemotherapy

Potential mechanisms of antiviral agents

- Disrupt attachment, penetration, uncoating
- Inhibit virus-associated enzymes → Replication
>blocking the enzyme DNA polymerase

- Inhibit transcription

>inhibit mRNA synthesis that would eventually lead to formation of proteins

- Inhibit translation
- Interfere with assembly

>the final packaging of the matured virion particles comprising of nucleic acids and capsid

- Prohibit the release

Need for New Antiviral Agents

- Viral infections are not curable after the virus invades the host and begins to replicate
- Vaccines are effective but r prevention only
- Vaccines provide immunity against specific viruses

Viral Chemotherapy

- Target some viral processes NOT present in the host cell (selectively toxic)
- Early agents were toxic or with limited spectrum
- Viral infection may not appear until REPLICATION is complete

An Ideal Antiviral Agent

- Has broad-spectrum activity
- Completely inhibit viral replication
>because by targeting viral replication, the viral life cycle will be arrest altogether
- Effective against mutant strains
>to prevent resistance
- Reach the target without affecting the host or its immune system
>selectively toxic

Agents Inhibiting Virus Attachment, Penetration, Uncoating and Early Viral Replication

Amantadine

- MOA: Inhibits penetration and blocks uncoating

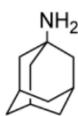
>distinct: is its structure, this has an unusual cage tricyclic amine structure

- USE: effective for prevention and treatment of all strains of Influenza A

>not for Influenza B

>anti-Parkinson drug that can cause anti-parkinsonian effects

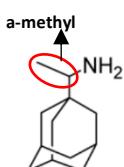
- CNS & GI side effects – HA, confusion, N&V



Amantadine

Rimantadine

>there is an α -methyl addition in the structure that's why it is α -methyl derivative of amantadine



Rimantadine

- MOA: Blocks uncoating but does not inhibit penetration

- USE: Similar with amantadine but more effective and has fewer CNS side effects

Interferons

- Produced by almost all cells response to viral challenge

- Type I**
- α – secreted by leukocytes
 - β – secreted by fibroblasts
- >used in the management of multiple sclerosis

- Type II**
- γ – immune interferon (NK cells, T cells)
- >self-produced, naturally occurring

- MOA: Bind to specific surface of virus-infected cells
- Inhibit viral penetration, uncoating, synthesis of mRNA, **translation** (most pronounced effect), assembly and release

>usually administered via IM or SQ

NEURAMINIDASE INHIBITORS

>enzyme that is found in the envelope of influenza virus

- HEMAGGLUTININ (HA) – glycoprotein enzyme important for viral binding to host cells via terminal sialic acid residue
 - NEURAMINIDASE (NA) – enzyme that facilitates cleavage of sialic bond, thereby enhancing adsorption to cell surface receptors
- >enhance penetration

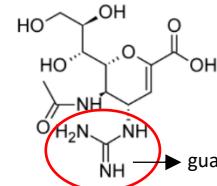
>by blocking neuraminidase, we can inhibit adsorption and penetration

Zanamivir

>distinct in the structure is the presence of guanidino group at position 4

>effective in Influenza A and B

- Effective via nasal, intraperitoneal and IV routes but INACTIVE orally
- More rapidly resolve influenza symptoms and improve recovery



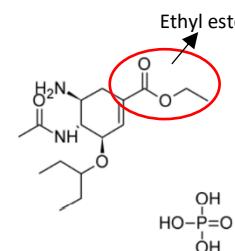
guanidino

Oseltamivir

- First orally administered NA inhibitor for Influenza A & B (as phosphate salt)

>the structure is prodrug because at first it has an ethyl ester FG that would undergo hydrolysis

>once hydrolyze, that causes formation of the active oseltamivir molecule



Ethyl ester

OH

HO-P(OH)2

ENTRY (FUSION) INHIBITORS

>fusion is one of the processes to penetrate the virion inside the host cell

Enfuvirtide

- First drug approved for clinical use
 - Used in combination with other antiretrovirals
- >often use for HIV

Maraviroc

- CCR5 antagonist

>type of chemokine receptor which is important in the entry/penetration of HIV

- Active orally but has low bioavailability

Agent Interfering with Viral Nucleic Acid Replication

>nucleic acid replication means that the original template DNA would serve as a template to synthesize complementary DNA strand and the important enzyme needed is the DNA polymerase

NUCLEIC ACIDS Examples are DNA, RNA

are made up of

Nucleotides

composed of

Phosphate groups

Nucleosides

composed of

Pentose sugar

Nitrogenous bases

- >nucleotides are the functional units of NA
- >It is nucleotides if it has presence of phosphate group, presence of nucleosides
- >nucleosides has 2 composition which are pentose sugar (monosaccharide) and nitrogenous base

Nucleotide structure

A. Pentose (five-carbon sugar)

- 1) Ribose
- 2) Deoxyribose

>can be known at the 2' carbon

>if ribose there is presence of oxygen and if deoxyribose there is no oxygen

B. Nitrogenous Bases

- 1) Purine – Adenine, Guanine (PURGA)
 - 2) Pyrimidine – Cytosine, Uracil, Thymine (PYCAT)
- >in DNA thymine is present while uracil is absent
>for RNA uracil is present while thymine is absent

C. Phosphate

BASE	NUCLEOSIDE	NUCLEOTIDE
Adenine	Adenosine Deoxyadenosine	Adenosine monophosphate
Guanine	Guanosine Deoxyguanosine	Guanosine monophosphate
Cytosine	Cytidine Deoxycytidine	Cytidine monophosphate
Uracil	Uridine	Uridine monophosphate
Thymine	Thymidine	Thymidine monophosphate

>in nucleotide there must be addition of phosphate and naming is based on the number of phosphates

Recall:

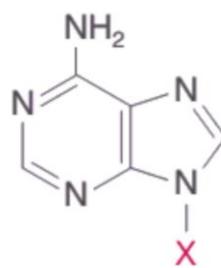
- 3 components of nucleotide
 - NB + Pentose + Phosphate
- Base + Pentose = NUCLEOSIDE
- Base + Pentose + Phosphate = NUCLEOTIDE

Classify whether a Nitrogenous base, Nucleoside, or Nucleotide

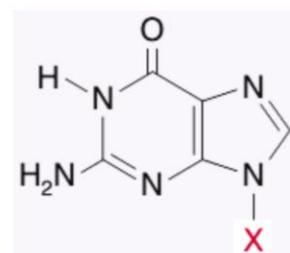
- Adenine
 - Nitrogenous base
- Guanosine monophosphate (GMP)
 - Nucleotide
- Cytidine
 - Nucleoside
- ATP
 - Nucleotide

Purines: (2 rings)

ADENINE

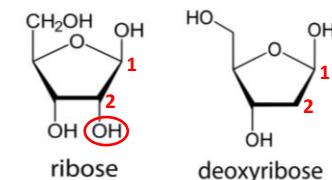
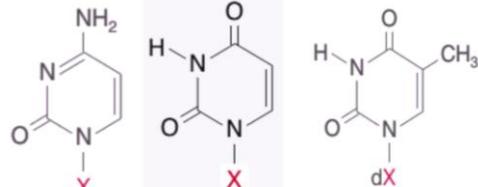


GUANINE

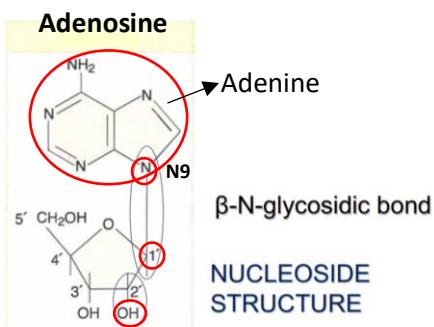


Pyrimidine (single ring)

Cytosine Uracil Thymine



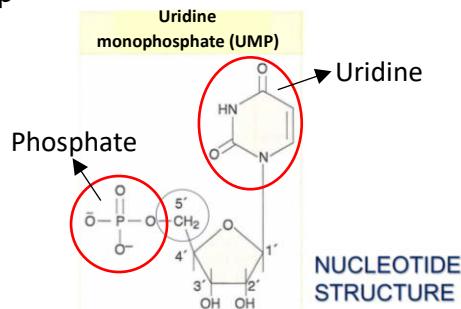
Nucleoside = Monosaccharide + Nitrogenous base



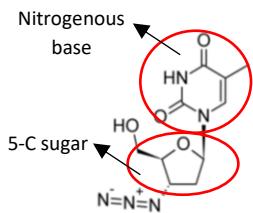
>it will have linkage between the 1st prime carbon at monosaccharide and at if the nitrogenous base is purine at Nitrogen 9

>this is Adenosine because it has OH (ribose) and NB + sugar

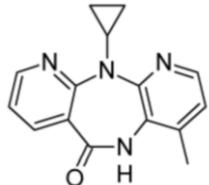
Nucleotide: because of additional of phosphate group



- >phosphate group will form linkage at the 5' carbon of the sugar
- >glycosidic bond will be seen at N1
- >the example drugs that can interfere viral nucleic acid replication they are analogs of these structures (kamukha)



Nucleoside analog



Non-nucleoside analog

General MOA:

>agents that can inhibit viral DNA replication process and those than can interfere the DNA polymerase enzyme, they are not directly blockers of DNA polymerase but rather they facilitate **counterfeit mechanism**

>also known as **drug (antiviral agent) incorporation process**

>gamot ay sumasali

>antiviral that are DNA replication inhibitors

>analog of natural purine and pyrimidine bases

>the drugs resemble the structure of natural purines and pyrimidines (if it resembles it can join in the normal process)

>analogs can join the DNA replication process but since they are fake, they can cause formation of faulty of DNA that would lead to viral death

>they are called antimetabolites

CONVENTIONAL NUCLEOSIDE ANALOGS

>they are nucleoside with some modifications to become analogs (NB + sugar)

Iodoxyuridine

- Nucleoside analog of THYMIDINE

>there is presence of the halogen iodine found at the 5' position

- Must first be phosphorylated to inhibit DNA polymerase

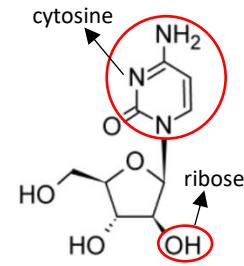
>to become nucleotide

>but since this is fake it can join the process but can result to faulty production of DNA

- FALSE PAIRING SYSTEM
 - Faulty viral proteins and defective viruses
 - DOC: tx of HSV keratitis

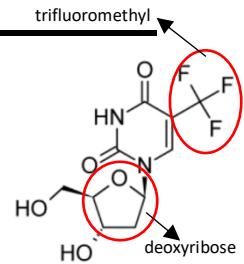
Cytarabine

- Nucleoside analog of CYTIDINE
- Anticancer agent for Burkitt lymphoma



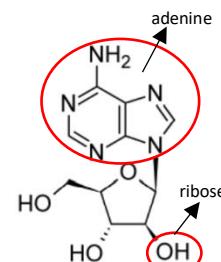
Trifluorothymidine

- Nucleoside analog of THYMIDINE
- >there is replacement of trifluoromethyl at the 5th position of pyrimidine ring
- aka TFT or TRIFLURIDINE
- Tx of HSV1 and HSV2 keratitis



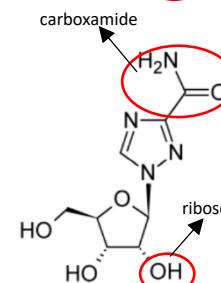
Vidarabine

- Nucleoside analog of ADENOSINE
- Obtained from *Streptomyces antibioticus*
- Alternative to Idoxuridine for HSV keratitis



Ribavirin

- Nucleoside analog of GUANOSINE
- >there is presence of carboxamide moiety
- >there is further modification that's why the guanosine structure cannot be seen
- Tx of Respiratory Syncytial Virus (RSV) in young children



ACYCLIC NUCLEOSIDE ANALOGS

>open chain

>there is a component of nucleoside that is not ring in nature which is the sugar portion

>the sugar attached to the NB are not ring

Acyclovir

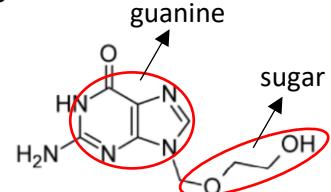
- 9-[2-(hydroxyethoxy) methyl]-9H-guanine

>guanine analog

- Most effective among acyclic analogs
- Limited to herpesviruses

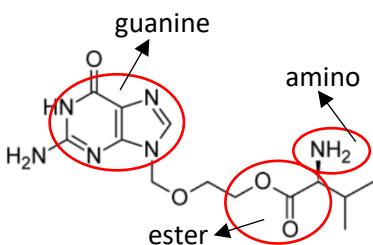
>chicken pox

- Via topical, IV
- BN: Zovirax



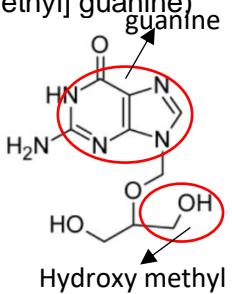
Valacyclovir

- Amino acid ester prodrug of acyclovir
- >addition of amino acid valine (HC acid, aliphatic, lipophilic)
- Metabolized in intestine/liver → ACYCLOVIR
- High GI absorption
- BN: Valtrex
- Tx of VZV



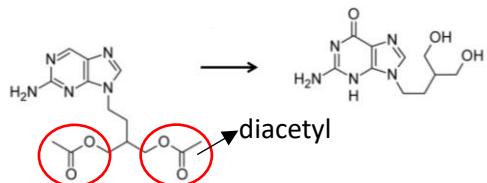
Ganciclovir

- 9-[(1,3-dihydroxy-2-propoxy) methyl] guanine
- >analog of acyclovir with an additional hydroxymethyl group in the acyclic side chain
- Active against CMV (Cytomegalovirus)
- Toxicity: MYELOSUPPRESSION
- BN: Cytovene



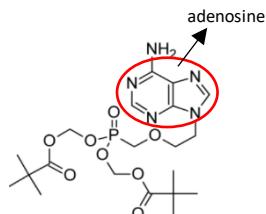
Famciclovir & Penciclovir

- Famciclovir is a diacetyl prodrug of Penciclovir
- >thru hydrolysis
- Tx of recurrent herpes labialis



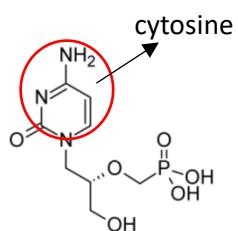
Adefovir

- Orally active prodrug indicated for chronic HBV



Cidofovir

- Tc of CMV retinitis in AIDS patients
- Dose-limiting toxicity: RENAL IMPAIRMENT
- >dose dependent

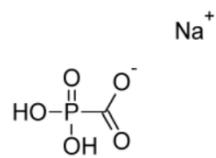


NON-NUCLEOSIDE ANALOGS

>there is no nucleoside structure (No NB and monosaccharide)

Foscarnet Sodium

- Does not require bioactivation
- 2nd line Tx of CMV retinitis in AIDS patients
- Causes NEPHROTOXICITY
- Ligand for metal ions



Anti-retroviral (Anti-HIV) agents including Protease Inhibitors

NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS (NRTI)

>the structure of these agents are nucleoside analogs (presence of M and NB)

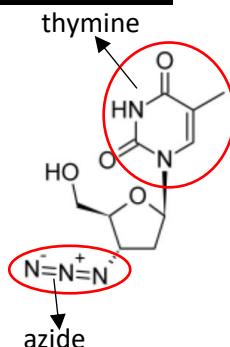
>reverse transcriptase which is also known as RNA dependent DNA polymerase

>this enzyme is observed in retroviruses like HIV and thru this enzyme they utilize/use RNA as template strand to be able to produce complementary DNA

>by inhibiting this enzyme, we tend to prevent formation of proviral DNA that's why there will be an antiviral effect, tend to inhibit the reproduction of HIV

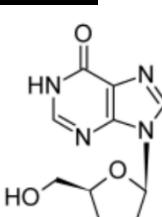
Zidovudine (AZT)

- Nucleoside analog of THYMIDINE
- 1st anti-HIV drug:
For Management of symptomatic HIV in adults (AIDS and ARC)
- >presence of azide FG
- >also known as azidothymidine
- ARC → AIDS-Related Complex
- Toxicity to Bone marrow
- BN: Retrovir



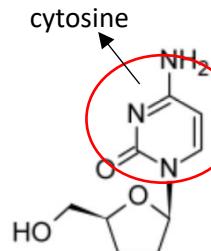
Didanosine (ddI)

- Nucleoside analog of DEOXYINOSINE
- For advanced HIV infection
- SE: Painful Peripheral neuropathy & pancreatitis
- BN: Videx



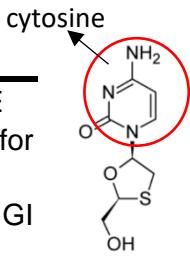
Zalcitabine (ddC)

- Nucleoside analog of CYTIDINE
- Alternative to AZT
- SE: Painful Peripheral neuropathy & pancreatitis



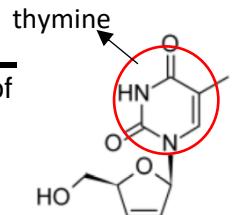
Lamivudine (3TC)

- Nucleoside analog of CYTIDINE
- Used in combination with AZT for progressive HIV infections
- SE: Peripheral neuropathy and GI side effects



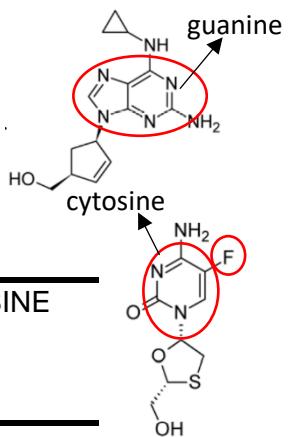
Stavudine (D4T)

- Nucleoside analog of THYMIDINE
- For advance HIV infections
- SE: Peripheral neuropathy



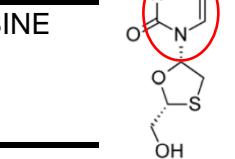
Abacavir

- Nucleoside analog of GUANOSINE
- Penetrates BBB
- >can act in the brain
- TRIZIVIR = Abacavir + AZT



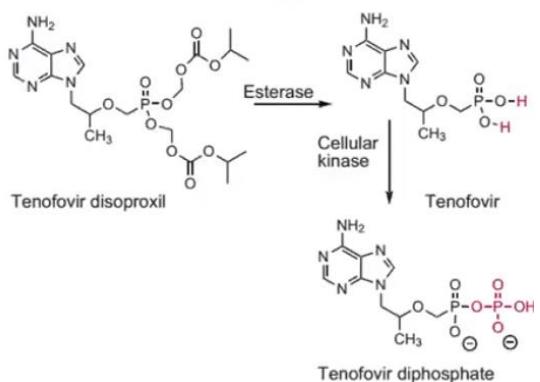
Emtricitabine

- Nucleoside analog of CYTOSINE
- Orally active NRTI



Tenofovir Disoproxil

- A prodrug
- >being converted thru hydrolysis via the enzyme esterase and its active form is Tenofovir
- >bioavailability can be improved w/ food
- Nucleoside analog of ADENOSINE
- TRUVADA = Tenofovir Disoproxil + Emtricitabine
- ATRIPLA = T + E + Efavirenz



NON-NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS (NNRTI)

>there is no nucleoside analog

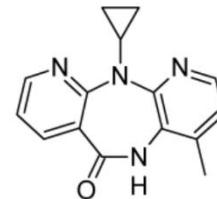
- Do not require BIOACTIVATION
- NON-COMPETITIVE INHIBITORS OF RT
- High therapeutic indices
- Do not inhibit mammalian DNA polymerases

>not use for monotherapy because if they are used as monotherapy the incidence of resistance is very high

- Synergistic with NRTI
- Rapid emergence of resistance

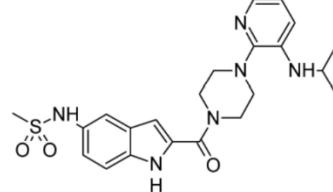
Nevirapine

- Dipyridodiazepinone derivative
- Readily cross the placenta and found in breast milk
- Combined with AZD and ddI
- BN: Viramune



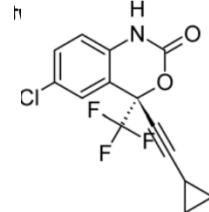
Delavirdine

- Rapidly absorbed orally
- Combined with AZD, ddI and ddC
- BN: Rescriptor



Efavirenz

- Must be used with at least two antiretroviral agents
- ATRIPLA = AZT + 3TC + Efavirenz



HIV PROTEASE INHIBITORS (PI)

>protease enzyme is an important enzyme to facilitate assembly and maturation of the newly produced virion particles

>inhibit the assembly of newly synthesized virion particles that's why there is an antiviral effect

- HIV protease is an enzyme that cleaves gag-pro polypeptides to yield active enzymes that function in the MATURATION and propagation of new virus
- HIV PI → HMW peptide-like structures
- HIV PI causes DYSLIPIDEMIA

>elevated blood cholesterol and triglycerides

→ "BUFFALO" HUMP, facial atrophy, breast enlargement

- Also causes HYPERGLYCEMIA

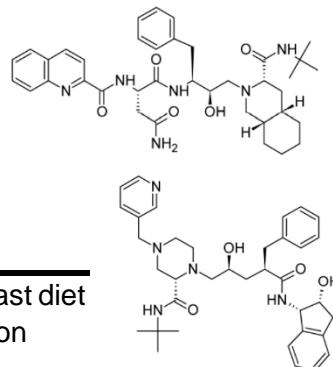
>blood sugar levels must be monitored

- LOW Bioavailability
- Short elimination half-life due to HYDROLYSIS or hepatic metabolism

Generic Name	Trade Name	Generic Name	Trade Name
Saquinavir	Invirase	Lopinavir/ritonavir	Kaletra
	Fortovase	Atazanavir	Reyataz
Ritonavir	Norvir	Fosamprenavir	Lexiva
Indinavir	Crixivan	Tipranavir	Aptivus
Nelfinavir	Viracept	Darunavir	Prezista
Amprenavir	Agenerase		

Saquinavir

- First PI approved by FDA
- Absorption is increased with fatty meals
- Synergistic w/ AZT and ddC



Indinavir

- Administered with a high fat meal to enhance absorption

Ritonavir, Amprenavir, Nelfinavir

- Inhibit CYP3A4
- Must be used with at least two antiretroviral agents

Lopinavir

- PI approved in combination with RITONAVIR (no activity but an enzyme inhibitor of CYP3A4)
- Used for patients with HIV who are not responsive to other treatments
- First to be approved for patients <6 months of age

Atazanavir

- Always used in combination with other RTI

Fosamprenavir

- A prodrug of Amprenavir

Tipranavir

- Unique among other PIs (not a peptidomimetic)
- Indicated for patient who have developed resistance to other drugs including PIs
- Administered with booster doses of RITONAVIR
- Potent but had severe side effects

HIV INTEGRASE INHIBITORS

- HIV integrase facilitates the insertion of viral complementary DNA into the host genome thru integrase enzyme, this could cause entry of the virus inside the nucleus of the host cell
- by inhibiting integrase, it will prevent the entry of the viral genome going to the host genome inside the nucleus and by blocking it there is antiviral effect

Raltegravir

- First (and thus far only) FDA-approved HIV Integrase inhibitor for resistant-HIV infections

Elvitegravir

- Currently in Phase III clinical trial

COMBINATION ANTIVIRAL THERAPY

Highly Active Antiretroviral Therapy (HAART)

- Approach using the SYNERGISTIC EFFECT of 3-4 combination of drugs
- NRTI or NNRTI + PI
- Foscarnet + Ganciclovir → HSV1 & 2
- TFT/Acyclovir + Interferon → herpetic keratitis
- AZT causes bone marrow toxicity but overcome when combined with
 - Foscarnet
 - ddC
 - ddI
- AZT + alpha interferon
 - delayed emergence of AZT-resistant HIV strains

Advantages of HAART

- Therapeutic antiviral effect
- Decrease toxicity
- Low incidence of drug-resistant infections
 - Delays progression of HIV infection and;
 - Prolongs the life of AIDS patients

INVESTIGATIONAL ANTIVIRAL AGENTS

Short Interfering RNA (siRNA)

- MOA: forms RNA-silencing complex blocking genes expression
- Problems:
 1. Poor stability
 2. Inefficient uptake
 3. Non-selective