Antibiotics

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Definitions

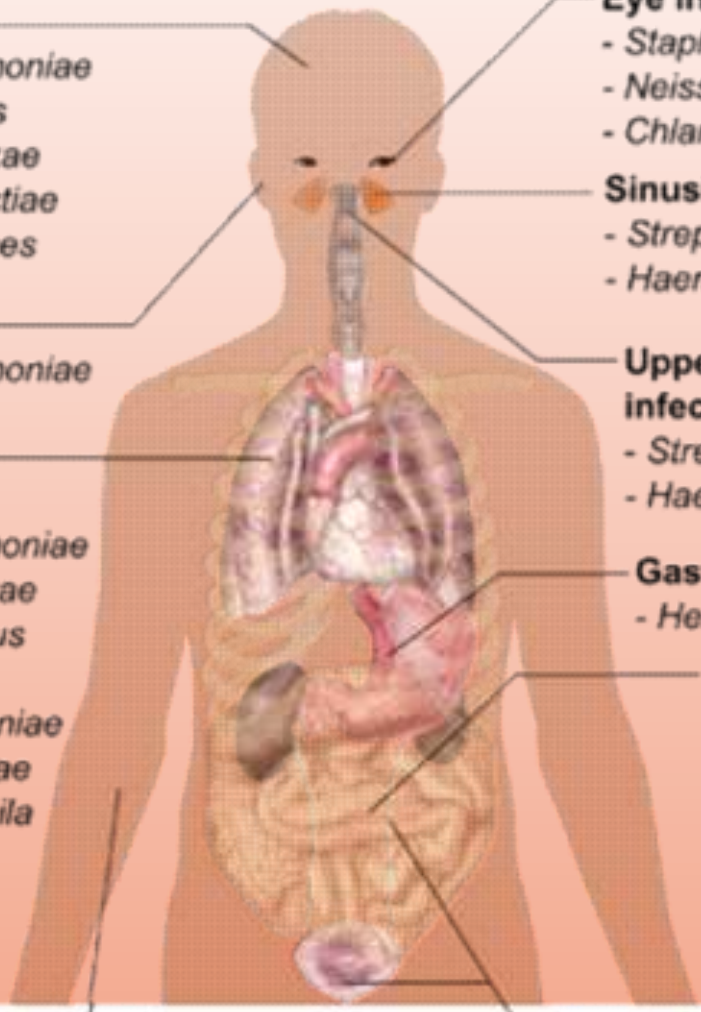
In 1942, Waksman proposed the definition of antibiotic

"An antibiotic or antibiotic substance is a substance produced by microorganisms, which has the capacity of inhibiting the growth and even of destroying other microorganisms."

Later definition was expanded

"Any substance produced by a living organism that is capable of inhibiting the growth or survival of one or more species of microorganisms in low concentrations."

Bacterial infections



Bacterial meningitis
- *Streptococcus pneumoniae*
- *Neisseria meningitidis*
- *Haemophilus influenzae*
- *Streptococcus agalactiae*
- *Listeria monocytogenes*

Otitis media
- *Streptococcus pneumoniae*

Pneumonia
Community-acquired:
- *Streptococcus pneumoniae*
- *Haemophilus influenzae*
- *Staphylococcus aureus*

Atypical:
- *Mycoplasma pneumoniae*
- *Chlamydia pneumoniae*
- *Legionella pneumophila*

Tuberculosis
- *Mycobacterium tuberculosis*

Eye infections
- *Staphylococcus aureus*
- *Neisseria gonorrhoeae*
- *Chlamydia trachomatis*

Sinusitis
- *Streptococcus pneumoniae*
- *Haemophilus influenzae*

Upper respiratory tract infection
- *Streptococcus pyogenes*
- *Haemophilus influenzae*

Gastritis
- *Helicobacter pylori*

Food poisoning
- *Campylobacter jejuni*
- *Salmonella*
- *Shigella*
- *Clostridium*
- *Staphylococcus aureus*
- *Escherichia coli*

Skin infections
- *Staphylococcus aureus*
- *Streptococcus pyogenes*
- *Pseudomonas aeruginosa*

Sexually transmitted diseases
- *Chlamydia trachomatis*
- *Neisseria gonorrhoeae*
- *Treponema pallidum*
- *Ureaplasma urealyticum*
- *Haemophilus ducreyi*

Urinary tract infections
- *Escherichia coli*
- Other Enterobacteriaceae
- *Staphylococcus saprophyticus*
- *Pseudomonas aeruginosa*

A substance is classified as an antibiotics, if the following conditions are meet.

1. **It is the product of metabolism.**
2. **It is a synthetic product, produced as a structurally similar with naturally occurring antibiotics.**
3. **It antagonizes the growth or survival of one or more species of micro-organisms.**
4. **It is effective in low concentration.**

Antibiotics classification

Antibiotics are usually classified based on their

1) Spectrum of activity,

2)Sources ,

3)Functions and

4)Structure

1) **Spectrum of Activity:**

Narrow spectrum (Highly selective for Gm+ve bacteria) : Penicillin G.

Broad Spectrum (Low selectivity or can affect wide variety of microbial species): Tetracyclins active against Gm+ve and Gm-Ve bacteria

2) **Sources:** Antibiotics have been isolated form three types of microorganism

Antibiotics from fungi

e.g. Penicillin form *Penicillium notatum* and *Penicillium chrysogenum*

Gresiofulvin from *Penicillium griseofulvum*

Antibiotics from actinomycetes

e.g. Sterptomycin from *Sterptomyces gresius*

Chloramphenicol from *Sterptomyces venezuelae*

Antibiotics from Bacteria (Gram positive Rods)

e.g. Bacitracin from *Bacillus subtillus*

Polymycins from *Bacillus polymyxa*

3) Mechanism of action:

Inhibition of bacterial cell wall synthesis:

penicillin, cephalosporin, cycloserine, vancomycine and bacitracin.

Interactions with the plasma membrane:

polymixin, amphotericin B, and nystatin.

Disruption of Protein synthesis:

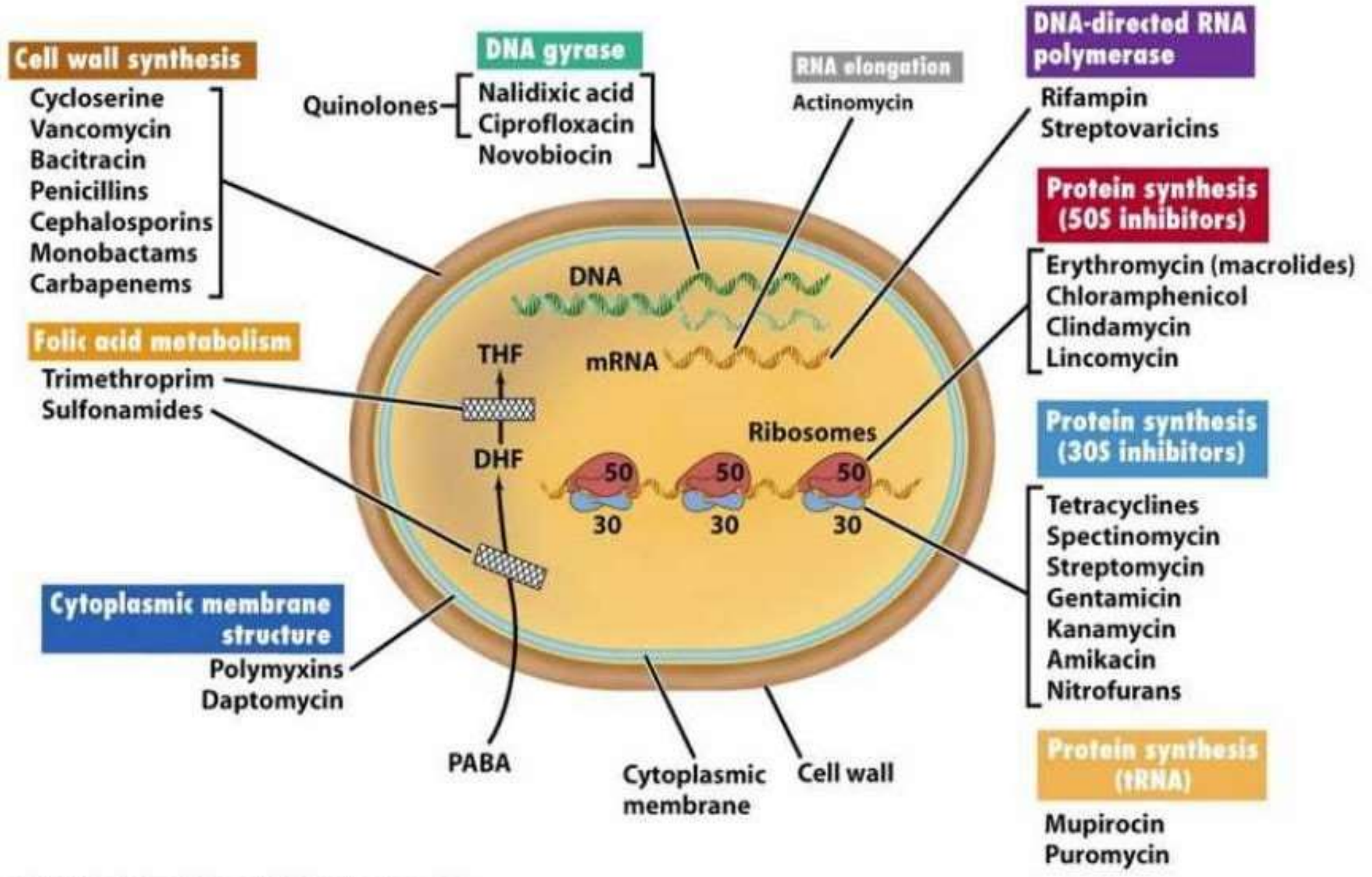
Rifamycins, Aminoglycosides, tetracyclins, and Chloramphenicol.

Inhibition of Nucleic acid synthesis:

Nalidixic acid, proflavin, griseofulvin

4) General Classification:

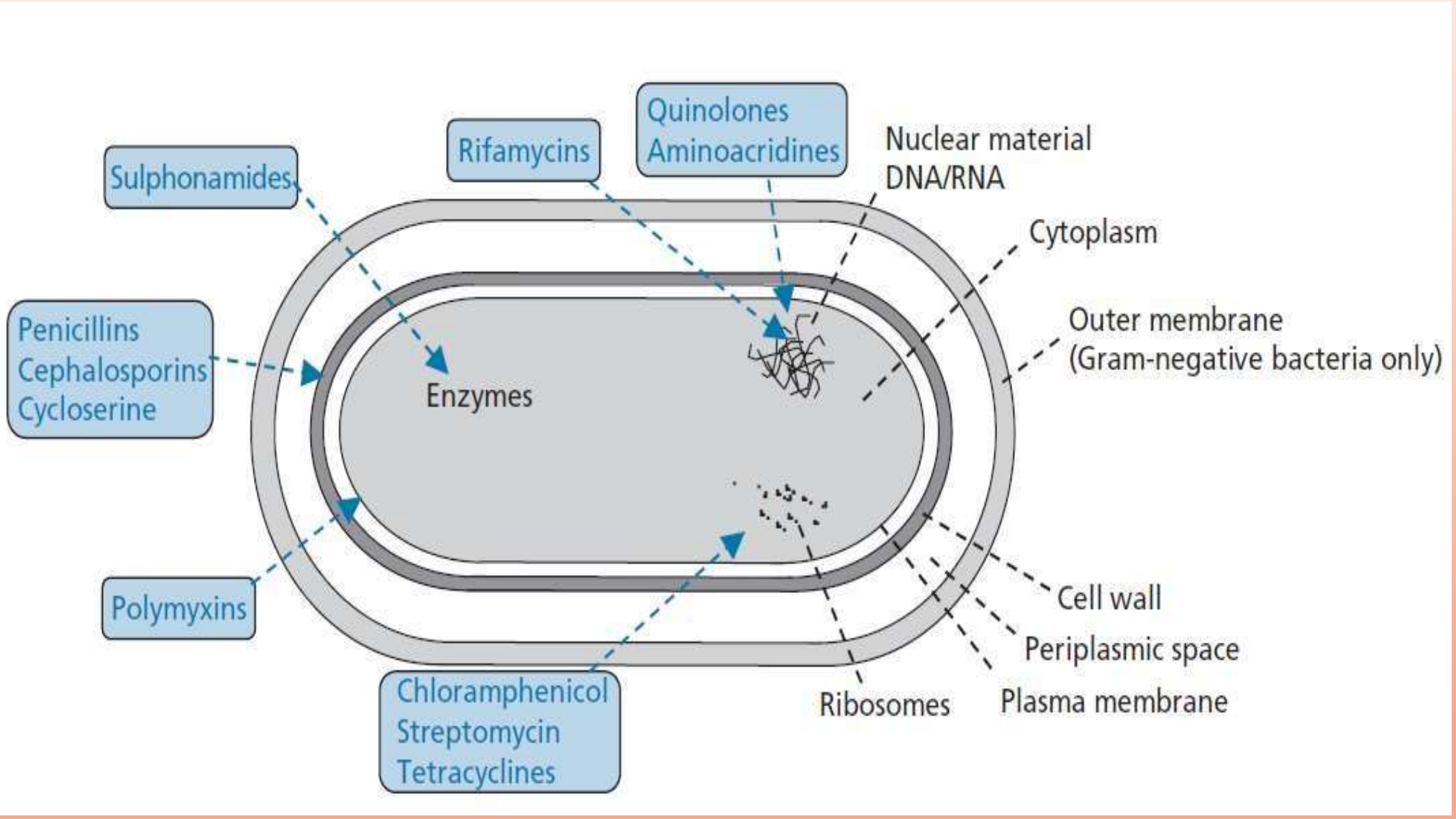
1. **β-Lactam antibiotics**: penicillins (e.g. amoxicillin), cephalosporins, carbapenems, monobactams, etc.
2. **Tetracyclines** : tetracycline
3. **Macrolide antibiotics**: erythromycin
4. **Aminoglycosides**: Gentamicin, Tobramycin, Amikacin
5. **Quinolones**: Ciprofloxacin (a fluoroquinolone)
6. **Cyclic peptides**: Vancomycin, Streptogramins, Polymyxins
7. **Lincosamides**: clindamycin
8. **Oxazolidinoes**: Linezolid (Zyvox)
9. **Sulfa antibiotics:** sulfisoxazole

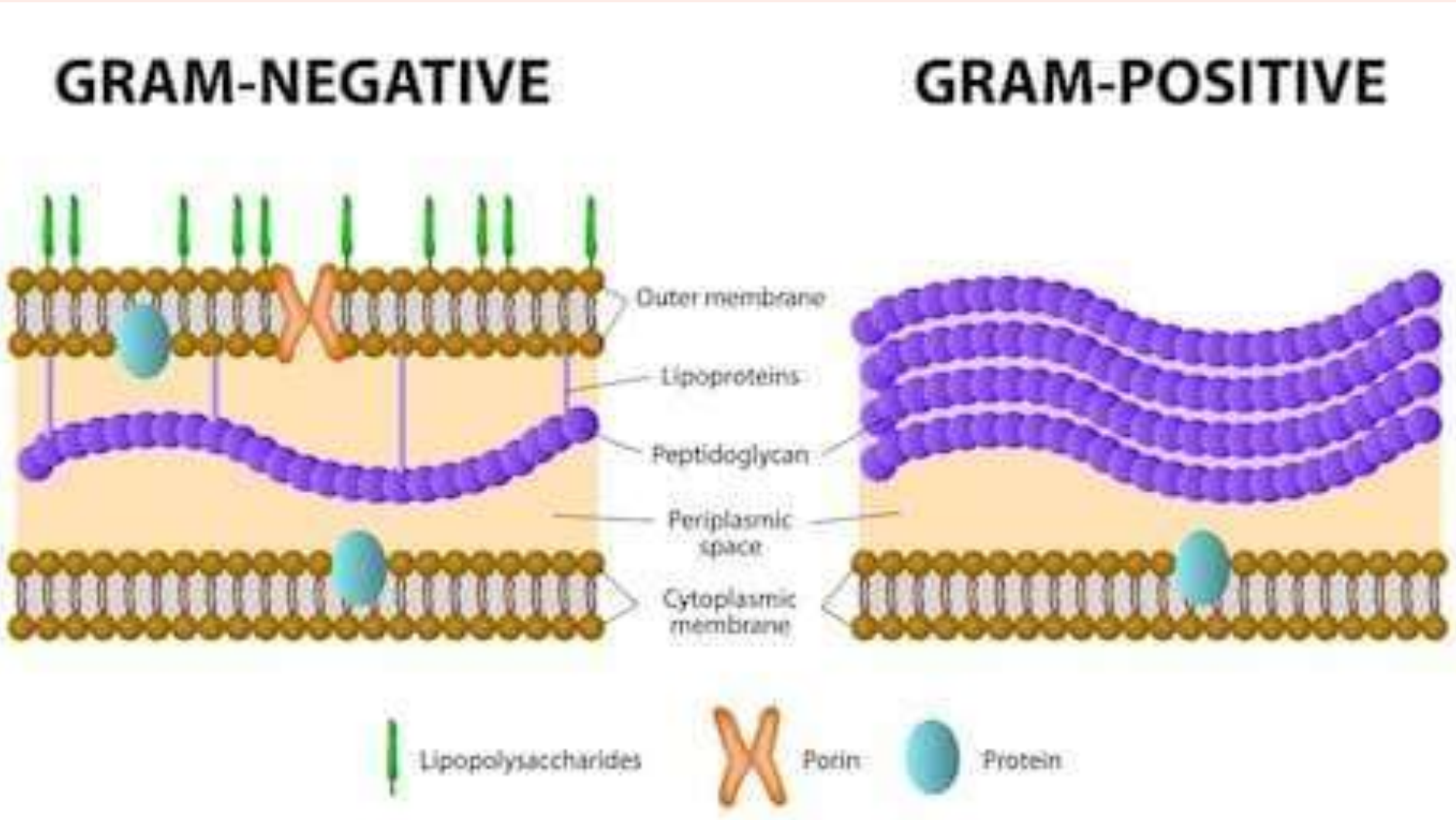

Cell wall synthesis
Cycloserine
Vancomycin
Bacitracin
Penicillins
Cephalosporins
Monobactams
Carbapenems
DNA gyrase
Quinolones
Nalidixic acid
Ciprofloxacin
Novobiocin
RNA elongation
Actinomycin
DNA-directed RNA polymerase
Rifampin
Streptovaricins
Protein synthesis (50S inhibitors)
Erythromycin (macrolides)
Chloramphenicol
Clindamycin
Lincomycin
DNA
mRNA
Folic acid metabolism
Trimethroprim
Sulfonamides
THF
DHF
Ribosomes
50
30
Protein synthesis (30S inhibitors)
Tetracyclines
Spectinomycin
Streptomycin
Gentamicin
Kanamycin
Amikacin
Nitrofurans
Cytoplasmic membrane structure
Polymyxins
Daptomycin
PABA
Cytoplasmic membrane
Cell wall
Protein synthesis (tRNA)
Mupirocin
Puromycin

Properties of antibiotics:

1. It must exhibit sufficient selective toxicity i.e. The agents should effective against pathogenic microorganisms but nontoxic to the host cells.
2. An antibiotic should be chemically stable enough to be isolated, processed, and stored for a reasonable length of time without deterioration of potency.
3. The rates of biotransformation and elimination of the antibiotic should be slow enough to allow a convenient dosing schedule yet rapid and complete enough to facilitate removal of the drug and its metabolites from the body soon after administration has been discontinued.

Bacterial Cell




GRAM-NEGATIVE
GRAM-POSITIVE
Outer membrane
Lipoproteins
Peptidoglycan
Periplasmic space
Cytoplasmic membrane
Lipopolysaccharides
Porin
Protein

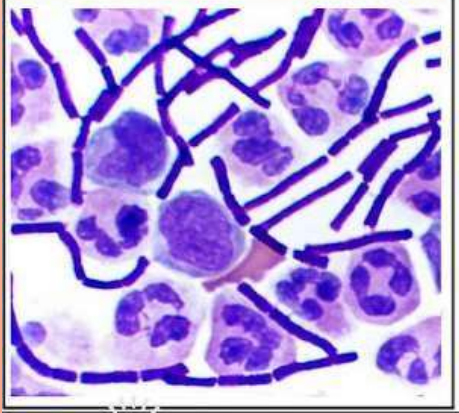
Difference between Gm +ve and Gm -ve

Character	Gram positive	Gram negative
Number of layers	One	Two
Thickness	Thick(20-80nm)	Thin (8-10 nm)
Outermembrane	Absent	Present
Periplasmic space	Present in some	Present in all
Chemical composition	Petidoglycan,Teichoic acid and lipotechoic acid	Lipopolysaccharide,lipoproteins and peptodoglycan
Porins proteins	Absent	Present
Lipid	Less	More
Peptidoglycan	More	Less
Permeablilty of molecules	More penetrable	Less penetrable
Resistance to molecules	Less	More

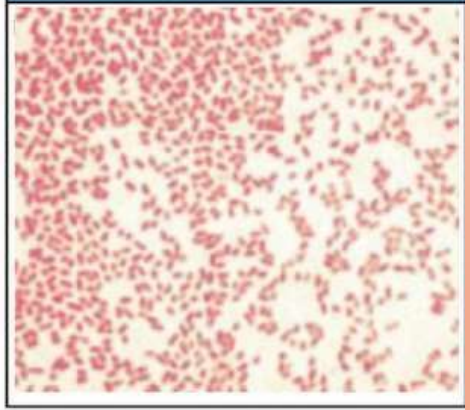
GRAM POSITIVE BACTERIA

- **Cocci**
  - Aerobe or facultative anaerobe → *Staphylococcus*, *Streptococcus*, *Enterococcus*
  - Anaerobe → *Peptostreptococcus*
- **Rods**
  - Aerobe or facultative anaerobe → *Corynebacterium*, *Listeria*, *Lactobacillus*, *Nocardia*, *Mycobacterium*, *Bacillus*
  - Anaerobe → *Actinomyces*, *Clostridium*

Gram-positive bacteria



Gram-negative bacteria



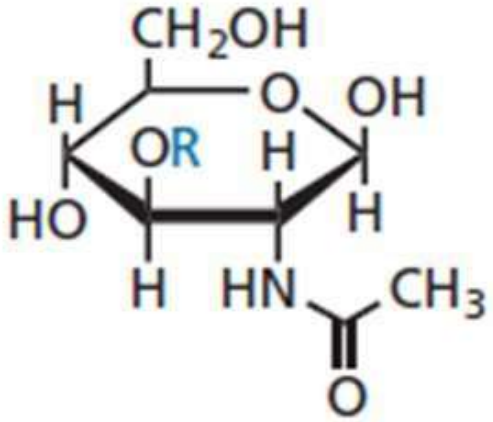
GRAM NEGATIVE BACTERIA

- **Cocci**
  - Aerobe → *Neisseria*
- **Rods**
  - Aerobe → *Pseudomonas*
  - Facultative anaerobe → *Salmonella*, *Shigella*, *Klebsiella*, *Proteus*, *Escherichia*, *Yersinia*, *Bordetella*, *Haemophilus*, *Brucella*, *Pasteurella*, *Vibrio*
  - Anaerobe → *Bacteroides*, *Fusobacterium*, *Prevotella*
  - Microaerophile → *Campylobacter*
- **Spirochaetes**
  - Aerobe → *Leptospira*
  - Anaerobe → *Borrelia*, *Treponema*

Structure of Peptidoglycan layer:

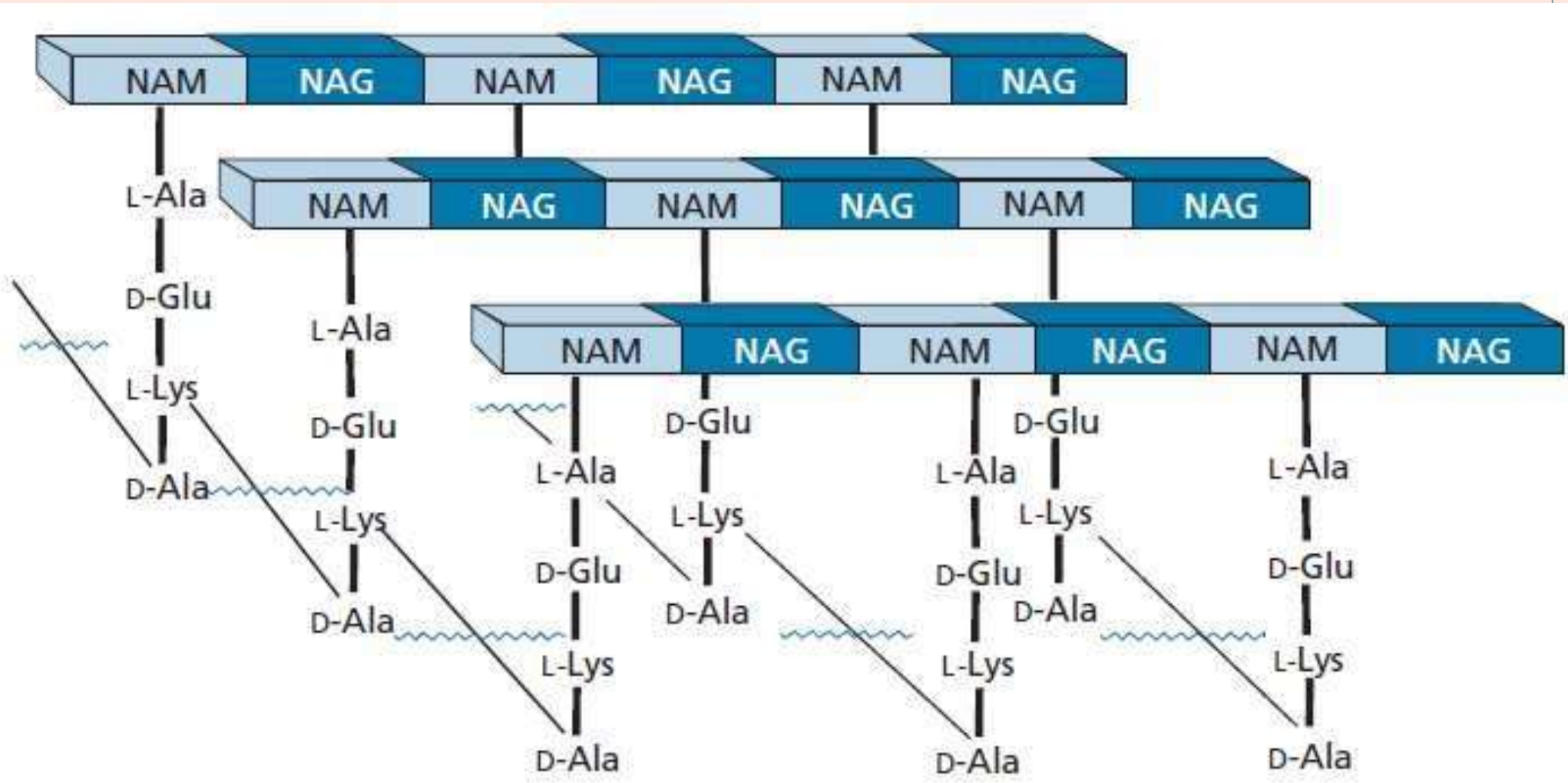
Made up by- Peptide and Sugar units

Two types of sugar units in parallel series- N-acetylmuramic acid(NAM) and N-acetylglucaosamine (NAG)

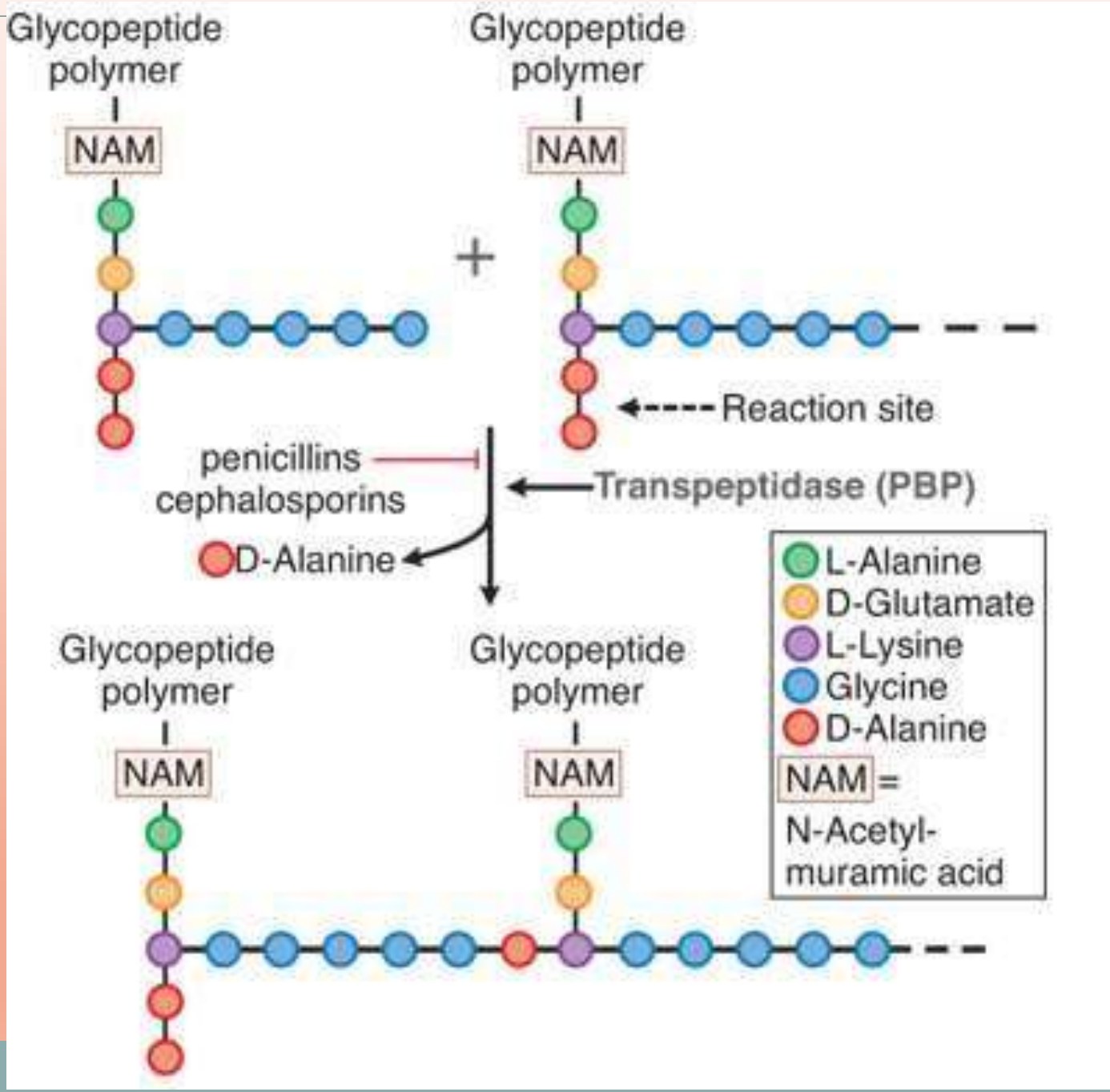


R = H, *N*-acetylglucosamine (NAG);
R = $CHMeCO_2H$, *N*-acetylmuramic acid (NAM).

Peptide chains are bound to the NAM sugars and it is interesting to note the presence of d-amino acids in these chains.



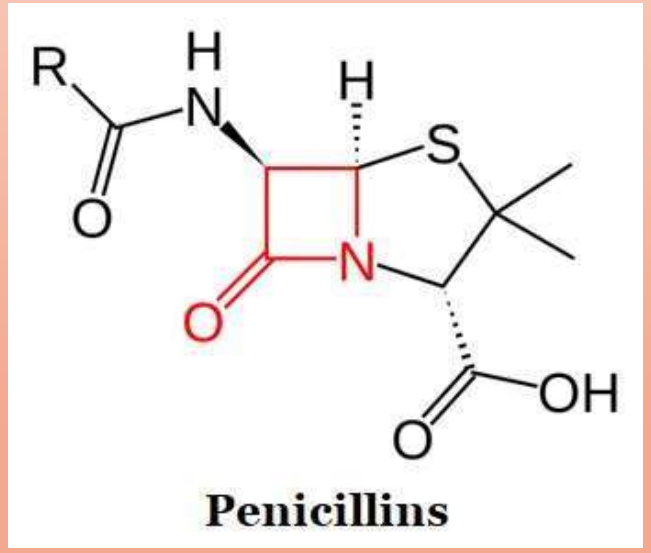
Peptidoglycan structure of bacterial cell walls.


Glycopeptide polymer
NAM
Glycopeptide polymer
NAM
Reaction site
penicillins
cephalosporins
Transpeptidase (PBP)
D-Alanine
L-Alanine
D-Glutamate
L-Lysine
Glycine
D-Alanine
NAM =
N-Acetyl-muramic acid
Glycopeptide polymer
NAM
Glycopeptide polymer
NAM

β-lactam antibiotics and Its SAR

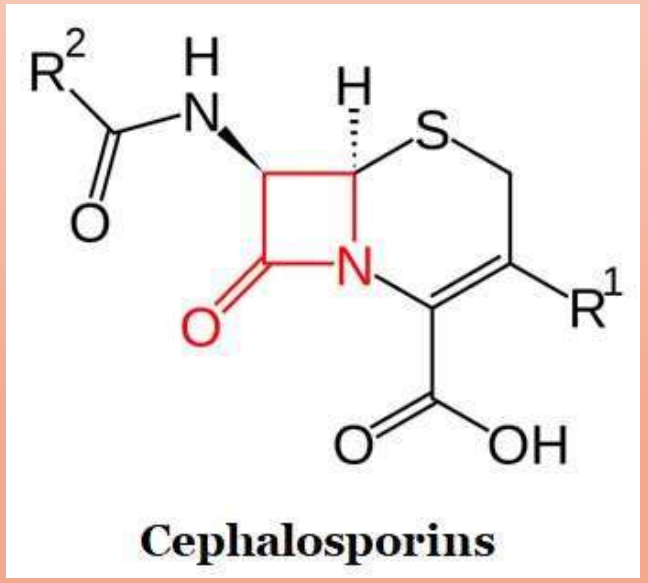
Antibiotics that possess the β-lactam (a four-membered cyclic amide) ring structure

It is named as such because the nitrogen atom is attached to the β-carbon atom relative to the carbonyl. The simplest β-lactam possible is 2-azetidinone.



Penicillins

2-azetidinone



Cephalosporins

β-lactams are classified according to their core ring structures.

β-lactams fused to saturated five-membered rings:

β-lactams containing thiazolidine rings are named penams.

β-lactams containing pyrrolidine rings are named carbapenams.

β-lactams fused to oxazolidine rings are named oxapenams or clavams.

β-lactams fused to unsaturated five-membered rings:

β-lactams containing 2,3-dihydrothiazole rings are named penems.

β-lactams containing 2,3-dihydro-1H-pyrrole rings are named carbapenems.

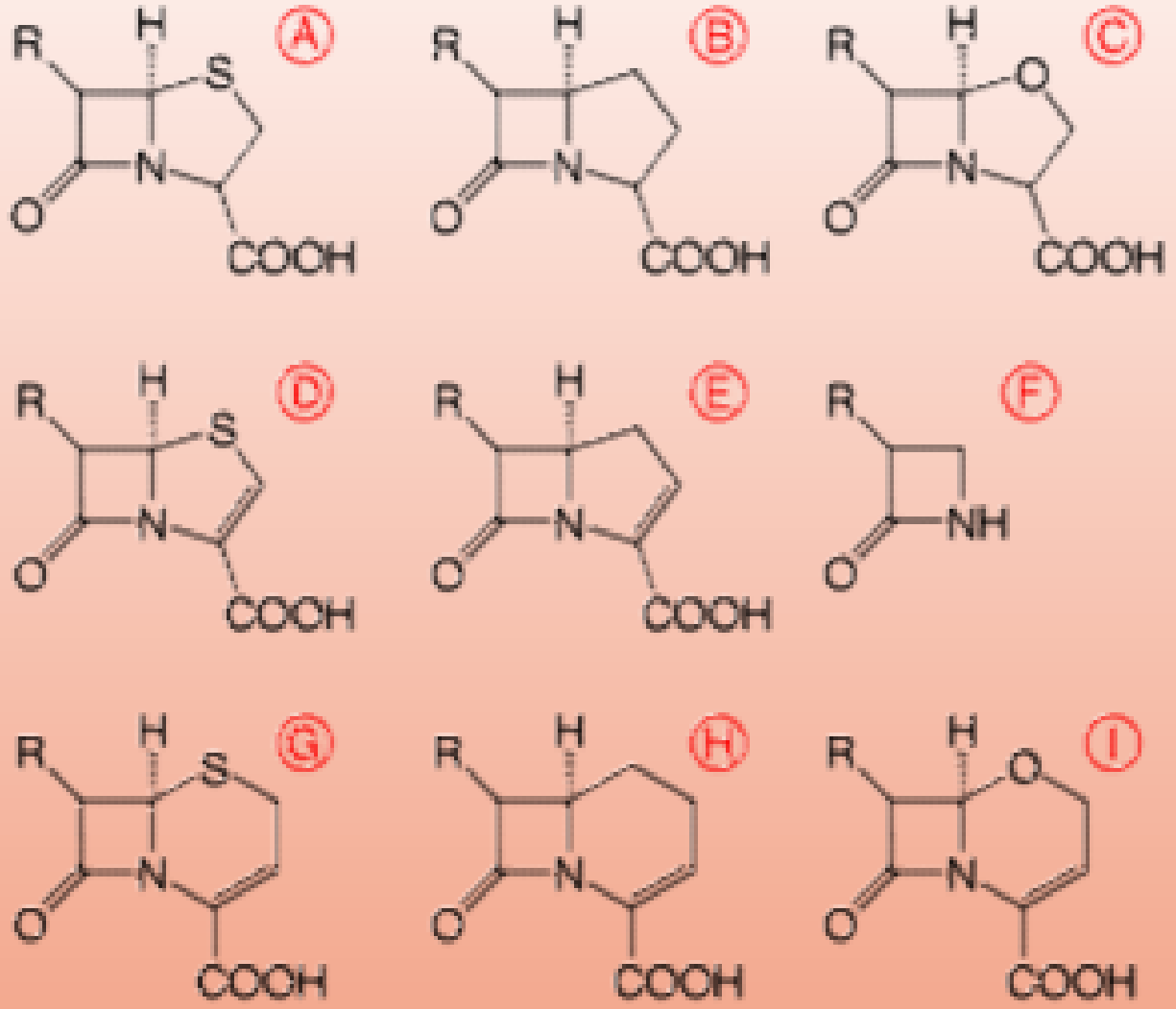
β-lactams fused to unsaturated six-membered rings:

β-lactams containing 3,6-dihydro-2H-1,3-thiazine rings are named cephems.

β-lactams containing 1,2,3,4-tetrahydropyridine rings are named carbacephems.

β-lactams containing 3,6-dihydro-2H-1,3-oxazine rings are named oxacephems.

β-lactams not fused to any other ring are named monobactams.



The β-lactam core structures.

(A) A penam.

(B) A carbapenam.

(C) An oxapenam.

(D) A penem.

(E) A carbapenem.

(F) A monobactam.

(G)A cephem.

(H)A carbacephem.

(I) An oxacephem.

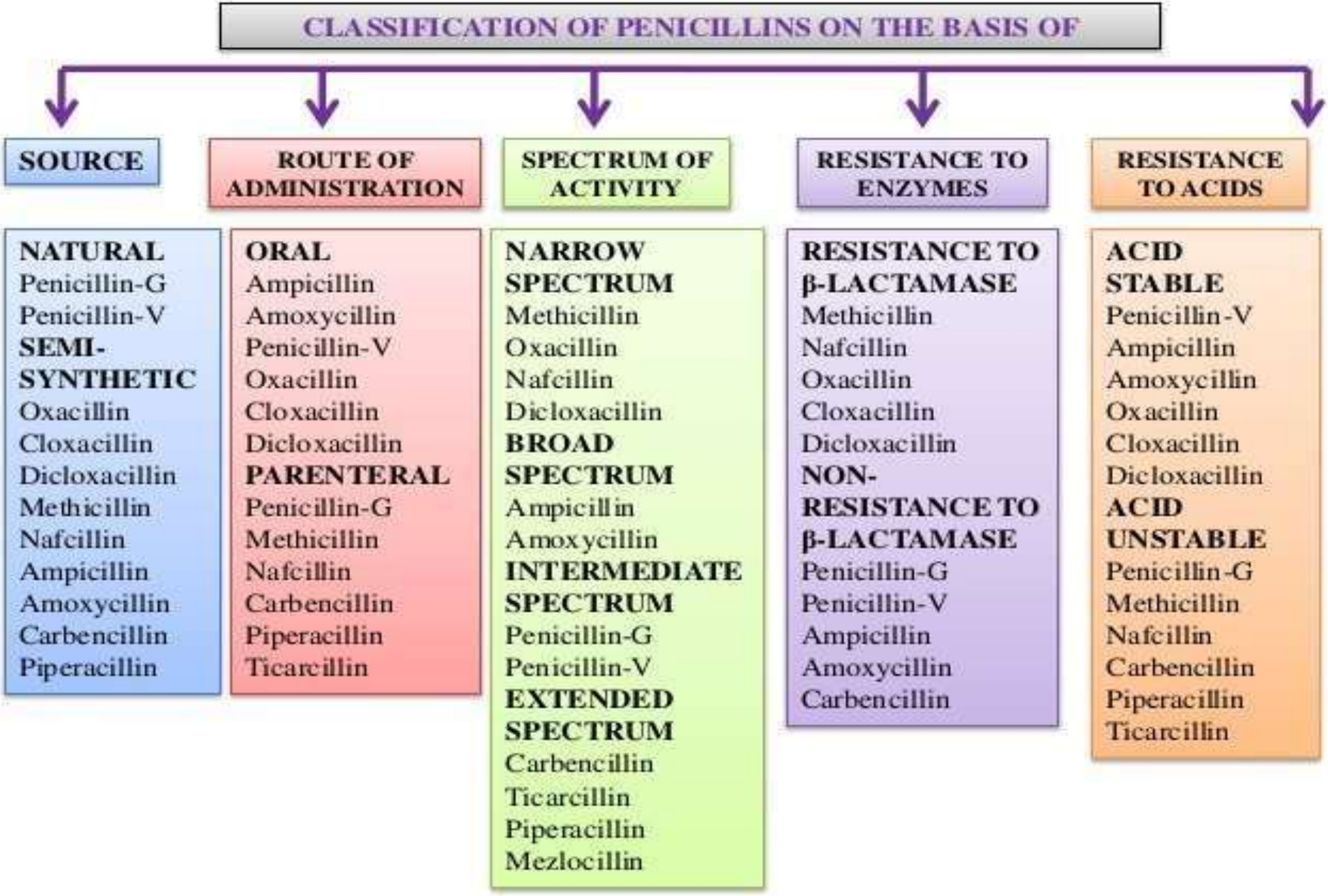
Penicillins

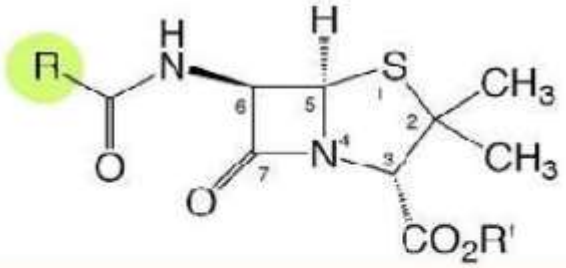
HISTORY

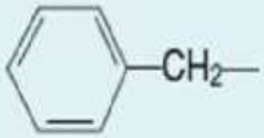
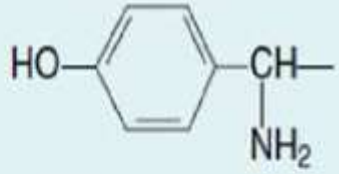
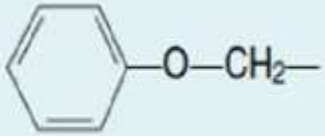
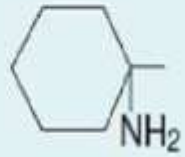
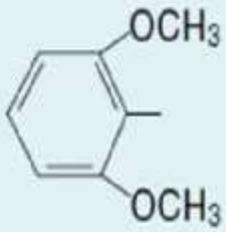
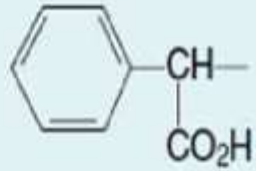
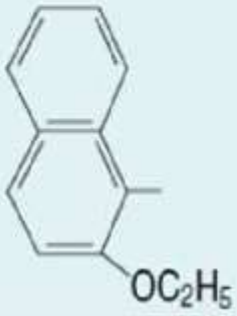
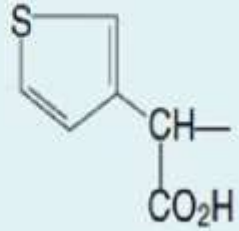
Penicillin, the world's first antibiotic, was discovered by British scientist Alexander Fleming in 1928 on accident.

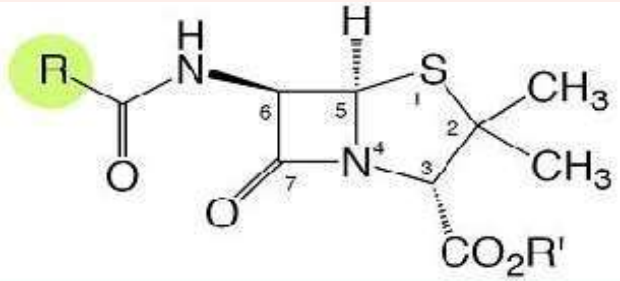
- In 1928 Alexander Fleming discovered the compound produced by the **fungus**.
- The fungus was called **Penicillium notatum**.
- The isolated compound he called Penicillin.
- Fleming noted a fungus growing on his bacterial plates had killed off the surrounding bacteria.

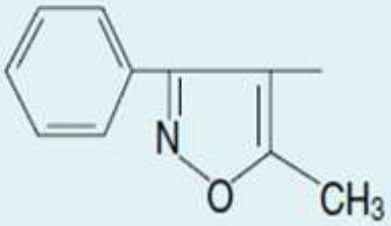
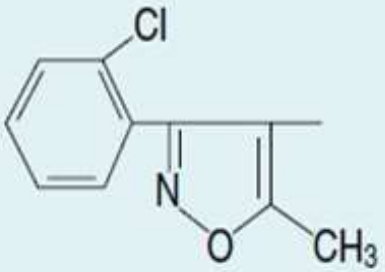
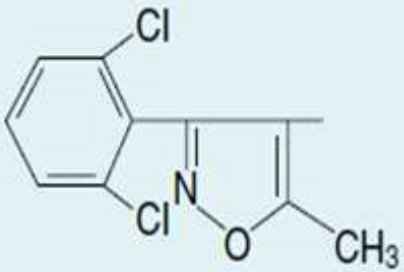
Fleming, Florey and Chain received a Nobel prize in **1945** for medicine for their work on penicillin.

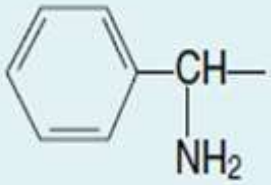

CLASSIFICATION OF PENICILLINS ON THE BASIS OF
SOURCE
NATURAL
Penicillin-G
Penicillin-V
SEMI-SYNTHETIC
Oxacillin
Cloxacillin
Dicloxacillin
Methicillin
Nafcillin
Ampicillin
Amoxycillin
Carbencillin
Piperacillin
ROUTE OF ADMINISTRATION
ORAL
Ampicillin
Amoxycillin
Penicillin-V
Oxacillin
Cloxacillin
Dicloxacillin
PARENTERAL
Penicillin-G
Methicillin
Nafcillin
Carbencillin
Piperacillin
Ticarcillin
SPECTRUM OF ACTIVITY
NARROW SPECTRUM
Methicillin
Oxacillin
Nafcillin
Dicloxacillin
BROAD SPECTRUM
Ampicillin
Amoxycillin
INTERMEDIATE SPECTRUM
Penicillin-G
Penicillin-V
EXTENDED SPECTRUM
Carbencillin
Ticarcillin
Piperacillin
Mezlocillin
RESISTANCE TO ENZYMES
RESISTANCE TO β-LACTAMASE
Methicillin
Nafcillin
Oxacillin
Cloxacillin
Dicloxacillin
NON-RESISTANCE TO β-LACTAMASE
Penicillin-G
Penicillin-V
Ampicillin
Amoxycillin
Carbencillin
RESISTANCE TO ACIDS
ACID STABLE
Penicillin-V
Ampicillin
Amoxycillin
Oxacillin
Cloxacillin
Dicloxacillin
ACID UNSTABLE
Penicillin-G
Methicillin
Nafcillin
Carbencillin
Piperacillin
Ticarcillin



Generic Name	Chemical Name	R Group	Generic Name	Chemical Name	R Group
Penicillin G	Benzylpenicillin		Amoxicillin	D-α-Amino-*p*-hydroxybenzylpenicillin	
Penicillin V	Phenoxymethylpenicillin		Cyclacillin	1-Aminocyclohexyl-penicillin	
Methicillin	2,6-Dimethoxyphenyl-penicillin		Carbenicillin	α-Carboxybenzyl-penicillin	
Nafcillin	2-Ethoxy-1-naphthyl-penicillin		Ticarcillin	α-Carboxy-3-thienyl-penicillin	



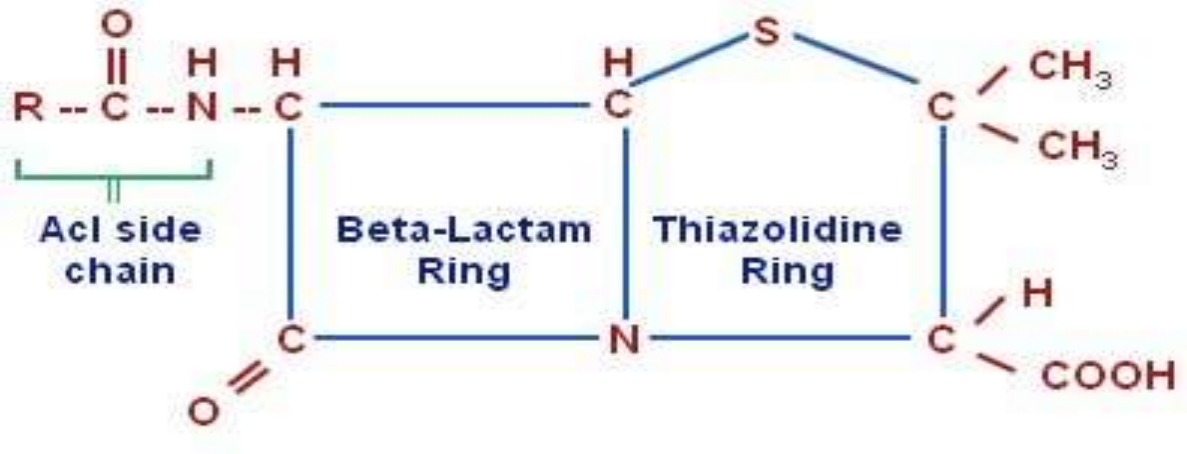
Generic Name	Chemical Name	R Group
Oxacillin	5-Methyl-3-phenyl-4-isoxazolylpenicillin	
Cloxacillin	5-Methyl-3-(2-chlorophenyl)-4-isoxazolylpenicillin	
Dicloxacillin	5-Methyl-3-(2,6-dichlorophenyl)-4-isoxazolylpenicillin	

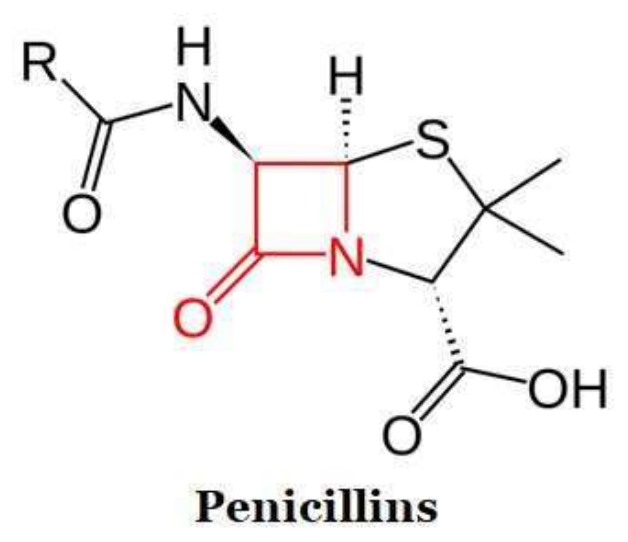
Generic Name	Chemical Name	R Group
Ampicillin	D-α-Aminobenzyl-penicillin	

Chemistry: Penicillin nucleus contains a highly unstable looking bicyclic system consists of

- •Thiazolidine ring (Ring A)-
  - •Sulphur containing with COOH (Carboxyl group),
- •Beta lactam ring (Ring B) – **(Broken by Beta-lactamase)**
  - •Side chain is attached at position – 6- (NHCOR)
- •Side chains attached through amide linkage. **(Broken by Amidase)**

Penicillins generally are designated according to the Chemical Abstracts system as 5-acylamino-2,2-dimethylpenam-3-carboxylic acids





The acyl side chain **(R)** varies, depending on the components of the fermentation medium. For example,

A fermentation medium of corn steep liquor contains high levels of phenylacetic acid ($PhCH_2CO_2H$) and gives **benzylpenicillin (penicillin-G ; R = benzyl).** A fermentation medium containing phenoxyacetic acid ($PhOCH_2CO_2H$) gives **phenoxymethylpenicillin (penicillin-V ; R = $PhOCH_2$)**

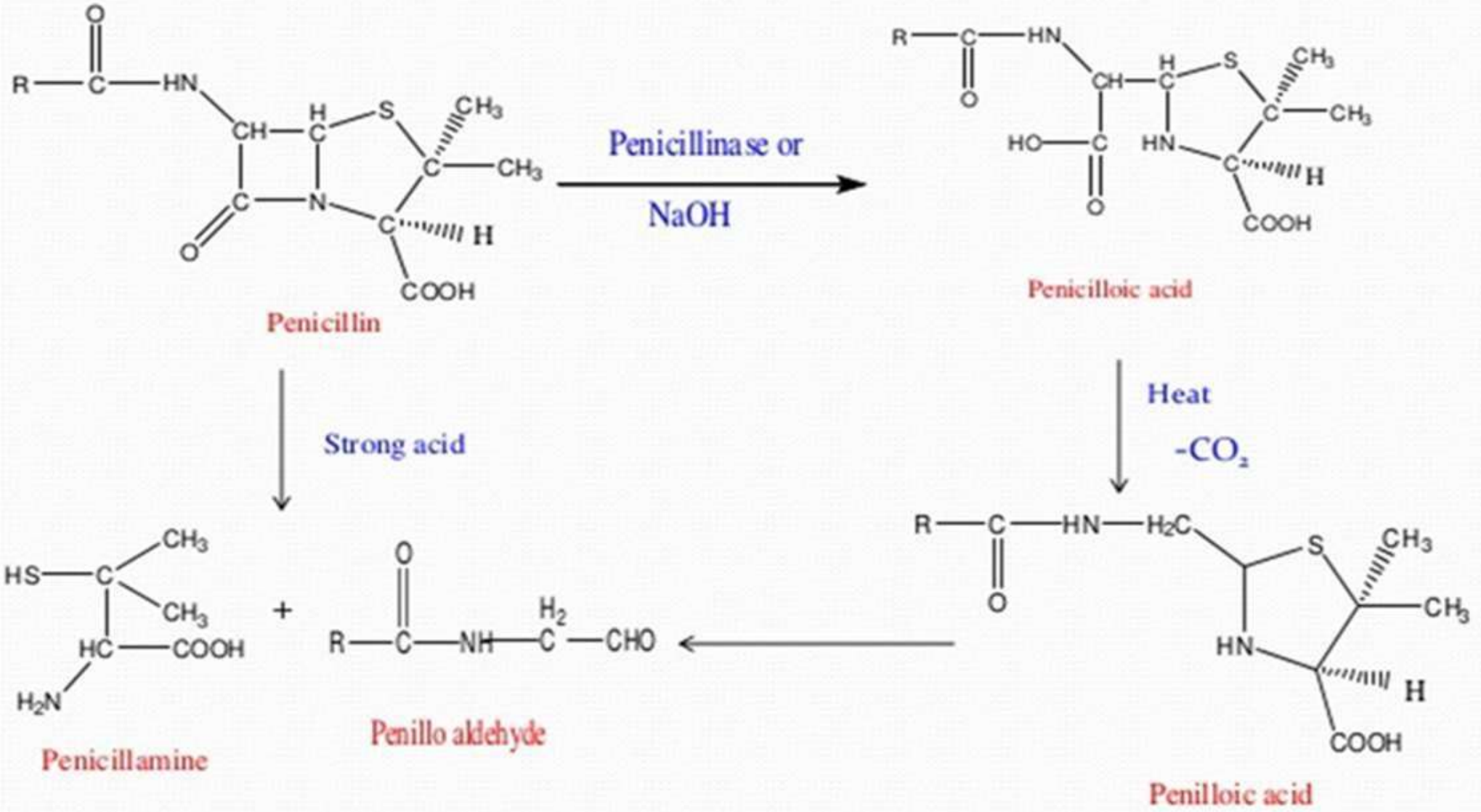
Properties:

The early penicillin were yellow-brown or red amorphous powder, that was so unstable that **refrigeration was required to maintain potency**, even for short period of time. For stability purpose, penicillin is converted into salt form, which is crystalline white in nature and store for years without refrigeration, only required thing is that it **should be protected from moisture**. The **sodium and potassium salts** of most of the penicillins are **water soluble and readily absorbed** when given by injection or orally, but less stable.

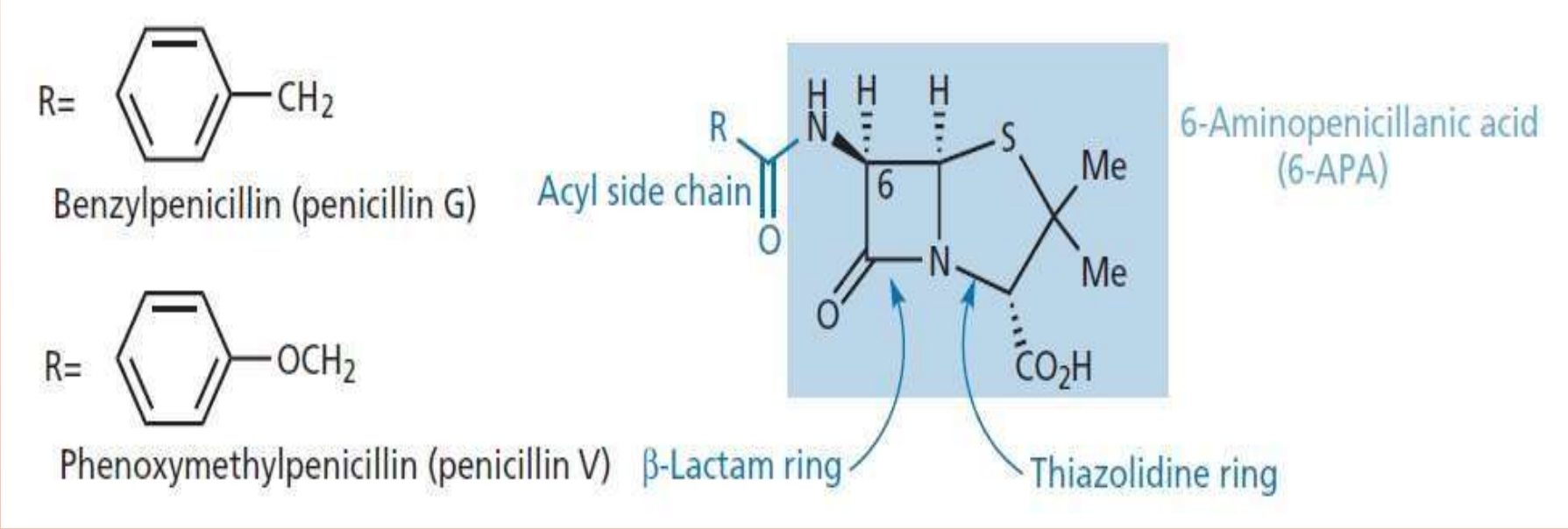
Chemical Degradation/ Instability of Penicillins

- Most unstable bond in penicillin molecule is β-lactam amide bond
- Bond cleaves slowly in water
- It breaks rapidly in alkaline solution to produce penicilloic acid , which readily decarboxylates to form penilloic acid.
- Penicilloic acid have anti-bacterial activity but acts as antigenic determinant (antigenic determinant a site on the surface of an antigen molecule to which a single antibody molecu le binds)
- In acidic solution, hydrolysis of penicillin is complex
- Hydrolysis of β-lactam amide bond involves participation of side chain amide oxygen
- As rate of reaction depends on nature of R- group
- Acidic degradation end products are penicillamine, penilloic acid, and penilloaldehyde

Chemical degradation:

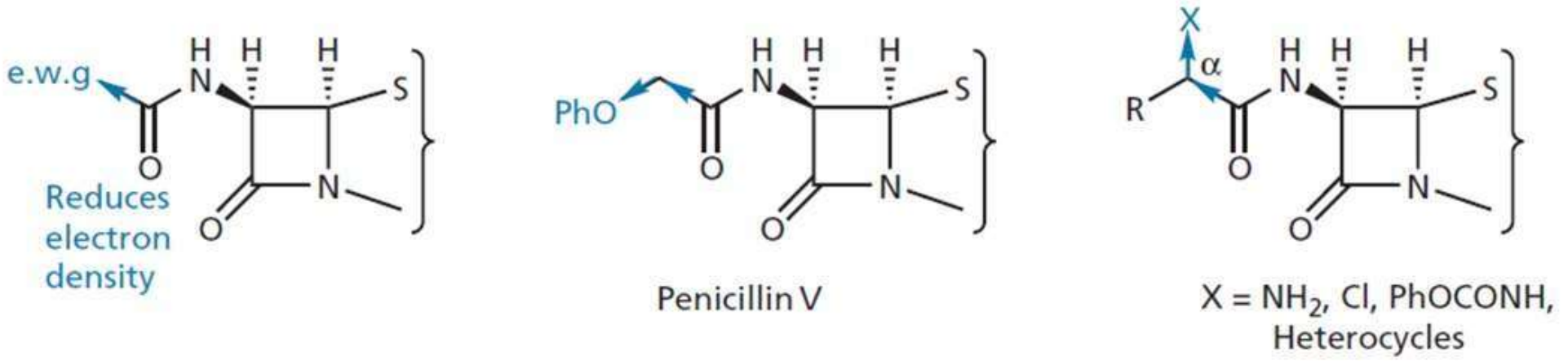


Penicillin G and Penicillin V



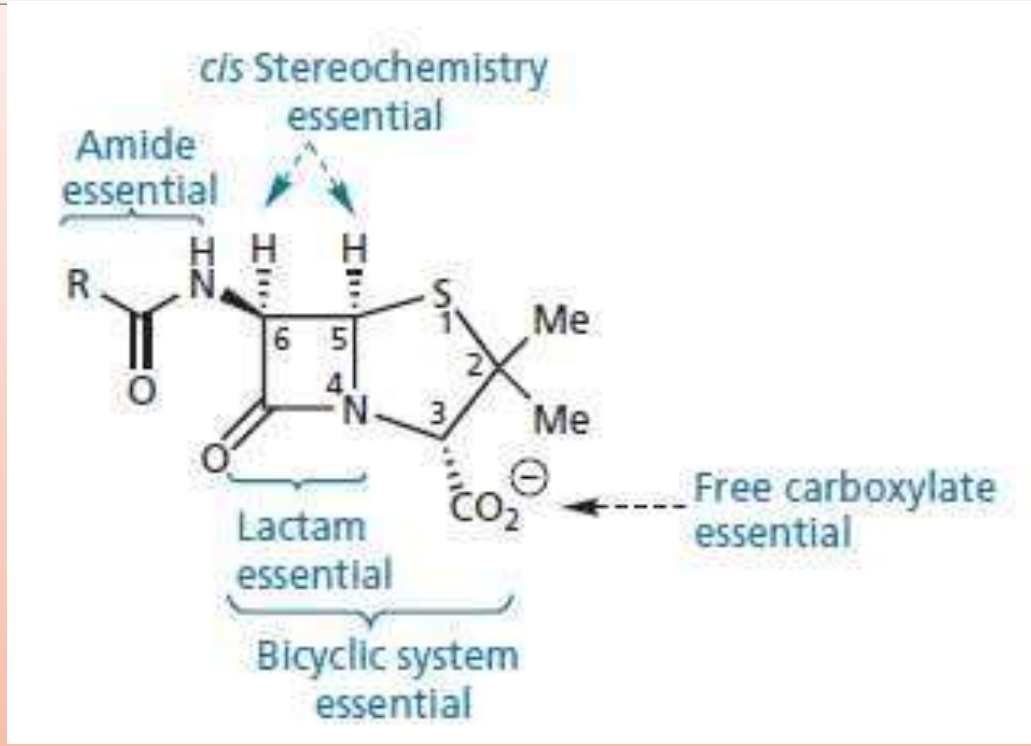
- Active against wide range of bacterial infections
- Lacks serious side effects (*hypersensitivity* reactions including urticaria, fever, joint pains, rashes, angioedema, anaphylaxis, serum sickness-like reaction) for most patients.
- Drawbacks: It can not be taken orally, has narrow spectrum of activity

Phenoxymethylpenicillin (penicillin V) has an electronegative oxygen on the acyl side chain with the electron withdrawing effect required. **Penicillin V has better acid stability than penicillin G** and is stable enough to survive the acid in the stomach, so it can be given orally; i.e. Penicillin V is acid resistant penicillin



Reduction of neighbouring group participation with an electron-withdrawing group (e.w.g.).

SAR of penicillin antibiotics



- β-lactam ring is essential for antibacterial activity.
- The free carboxylic acid is essential. This is usually ionized and penicillins are administered as sodium or potassium salts. The carboxylate ion binds to the charged nitrogen of a lysine residue in the binding site.
- the bicyclic system is important. This confers further strain on the β-lactam ring—the greater the strain, the greater the activity, but the greater the instability of the molecule to other factors;

- the acylamino side chain is essential;
- sulphur is usual but not essential;
- the stereochemistry of the bicyclic ring with respect to the acylamino side chain is important.
- Substituents attached to the penicillin nucleus have great effects on stability of penicillins and spectrum of activity

1) If side chain R group is an electron-withdrawing group

It decreases the electron density on the side chain carbonyl and protects the penicillins, in part, from acid degradation.

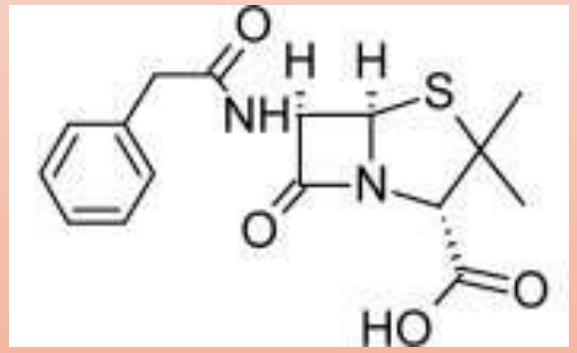
Such compounds can be given orally. **(% abs of intact drug)**

Benzyl penicillin	**15-30**
Pen V	**60-73**
Ampicillin	**30-50**
Amoxicillin	**75-90**

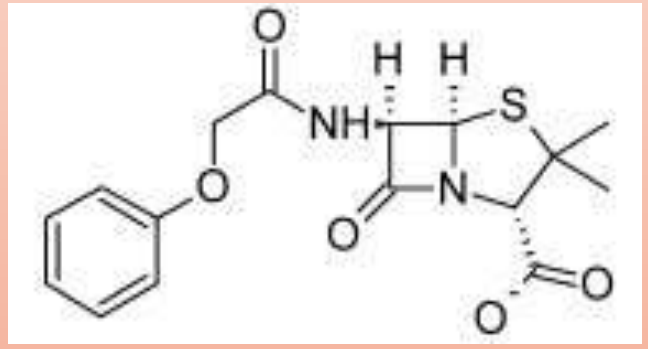
2) More lipophillic the side chain of penicillin, more is the serum protein binding Benzyl penicillin (45-68 %), Pen V (75-89 %),

Ampicillin & Amoxicillin (25-30 %),

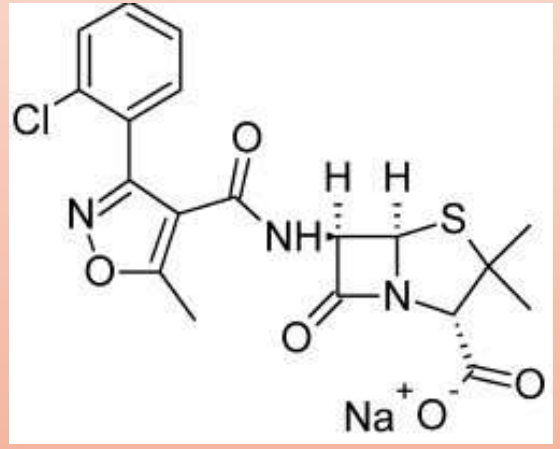
Oxacillin & Cloxacillin (> 90%)



Benzyl penicillin



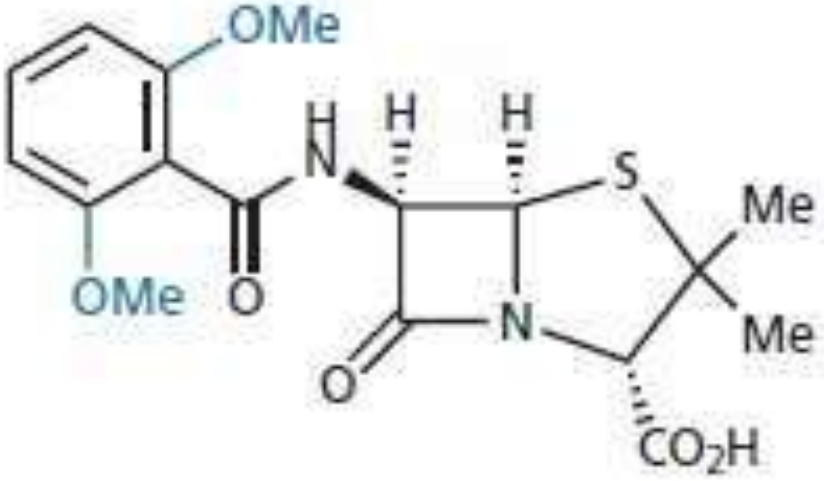
Pen V



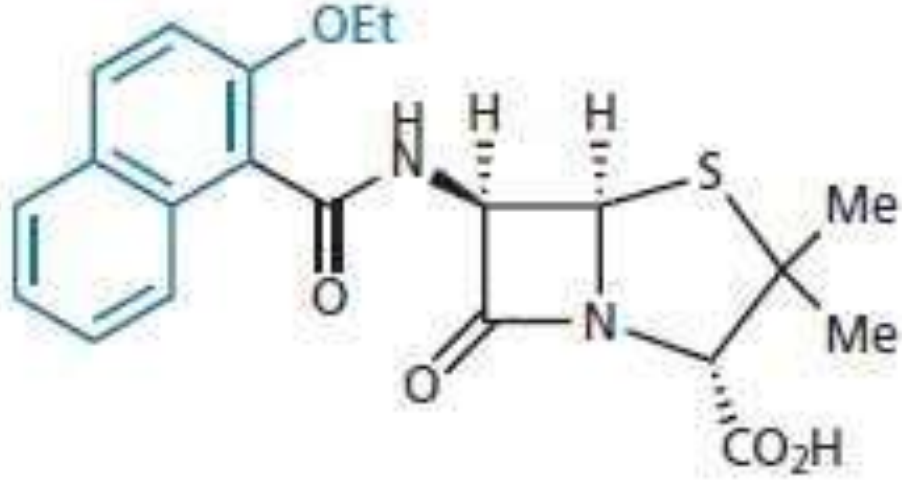
Cloxacillin

3) Bulky groups in the side chain provides stability against β- lactamase

The strategy of steric shields was used successfully to block penicillin from accessing the **penicillinase or β-lactamase** active site by placing a bulky group on the side chain e.g. Methicillin, Nafcillin, Oxacillin, Cloxacillin



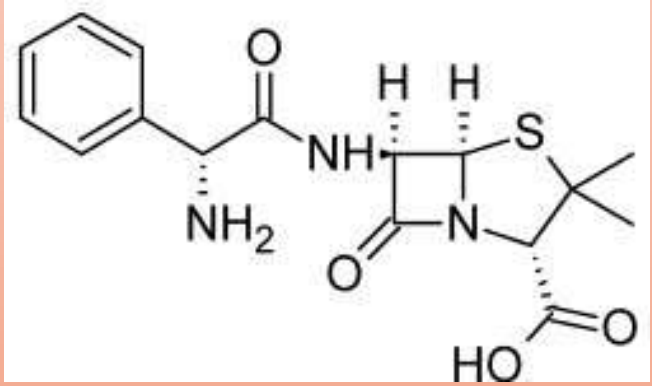
Methicillin



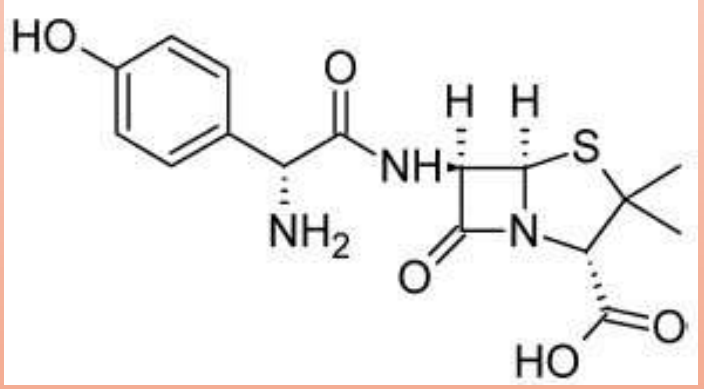
Nafcillin

4) Introduction of an ionized or polar group into the α-position to carbonyl group on the side chain increases the activity against gram-negative bacilli

- Hydrophilic penicillins (Ampicillin, Amoxicillin) penetrates Gram-negative bacteria more readily through porin channels of cell membrane
- D-isomer is 2 to 8 times more effective than L-isomer.



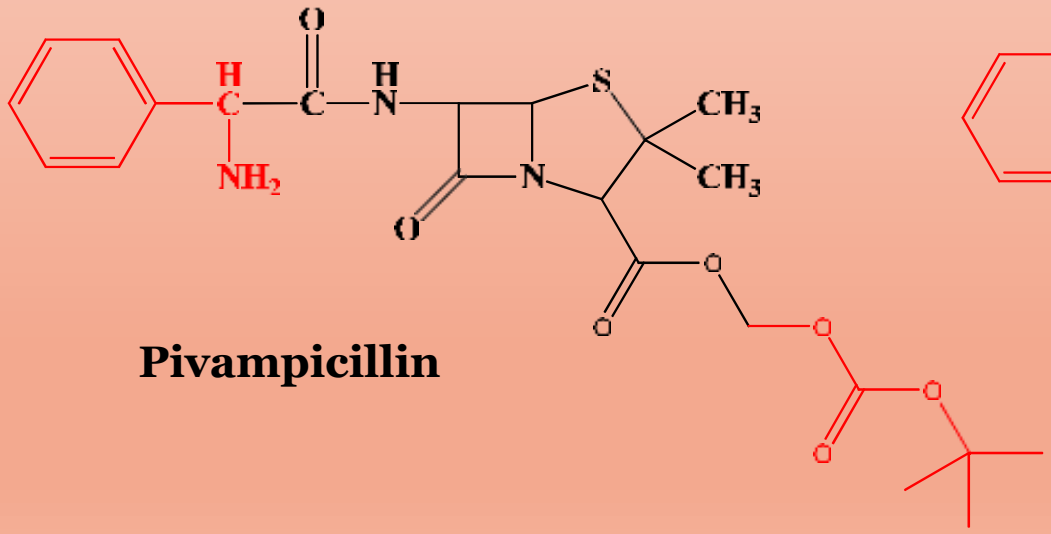
Ampicillin



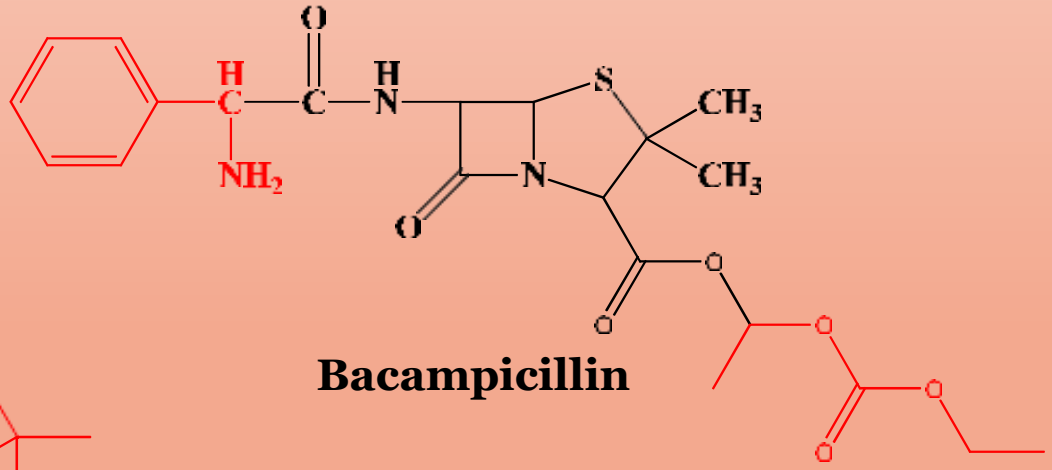
Amoxicillin

5) **The presence of the electron withdrawing amino group on both broad spectrum antibiotics ampicillin and amoxicillin increases acid stability.**

No Steric sheild present on ampicillin and amoxicillin so both are sensitive to β-lactamase enzyme. Ampicillin and amoxicillin are poorly absorbed by the gut due to ionization of both the amino and carboxylic groups. Modification of carboxylic acid of ampicillin to esters were made to solve this problem. **Bacampicillin and Pivampicillin** are examples of **ester prodrug of ampicillin**.



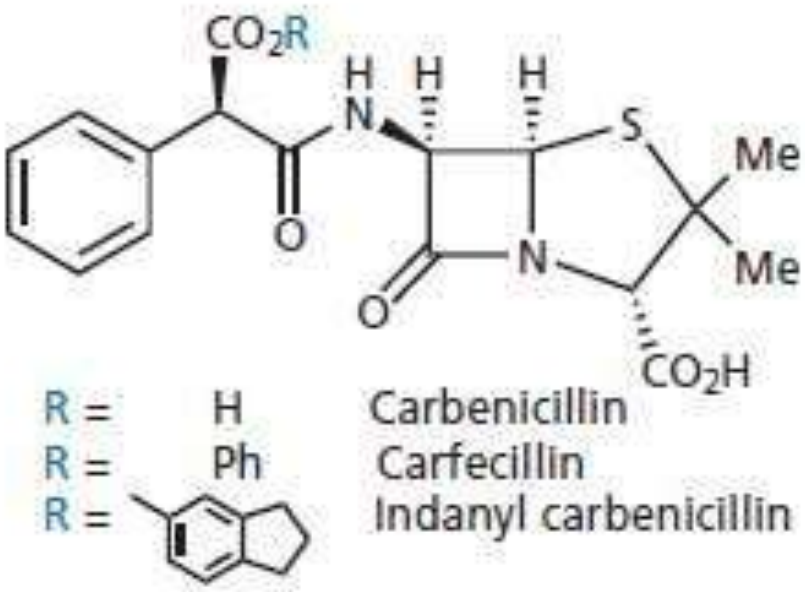
Pivampicillin

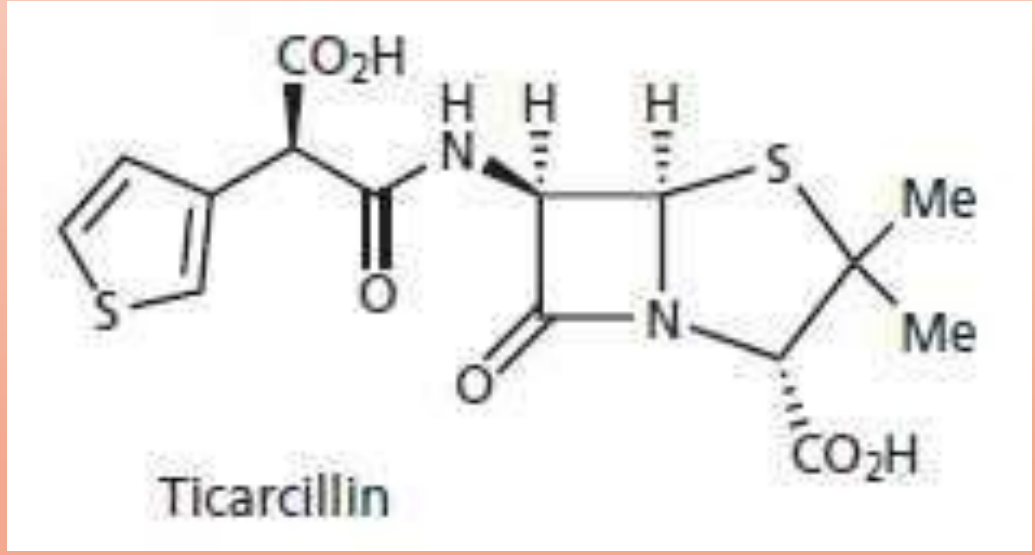


Bacampicillin

6) **Incorporation of acidic substitution at α-position to carbonyl carbon: shows broad spectrum of activity due to hydrophilic carboxyl group on the side chain. This leads to improved absorption through gut wall.**

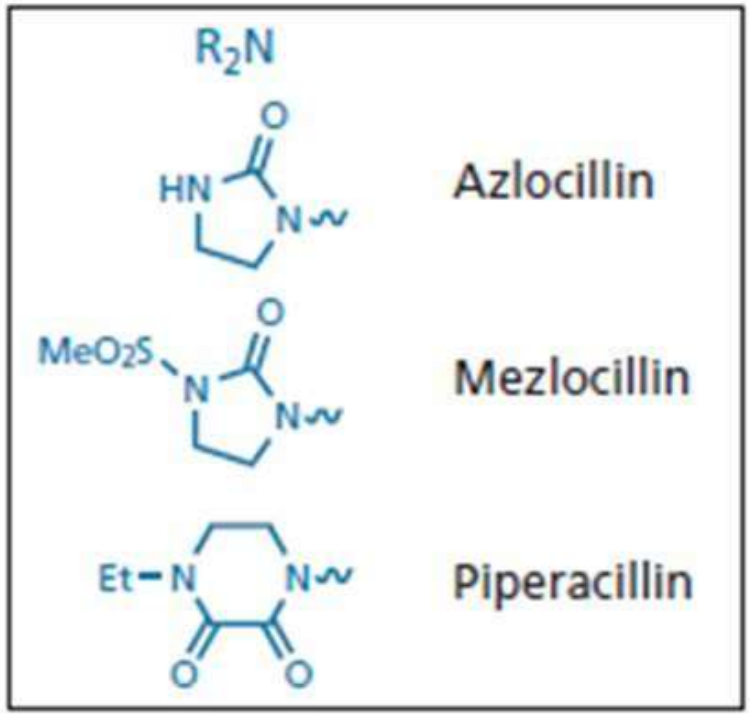
α-carboxybenzyl penicillin (carbenicillin) is active against ampicillin resistant organisms

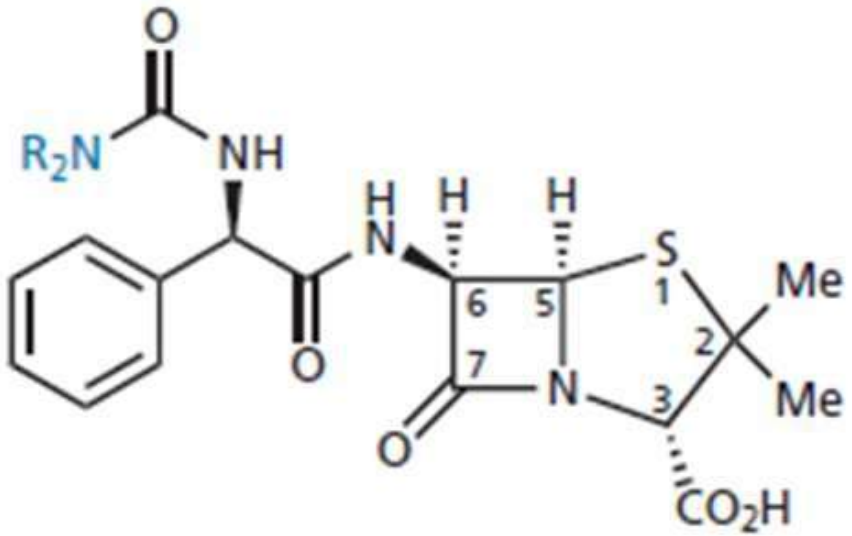




8) Ureidopenicillins newest class of broad spectrum penicllins and have a urea functional group at α-position. It exhibits greater activity against certain Gram-negative bacilli than carbenicillin

- e.g. Azlocillin, mezlocillin and piperacillin



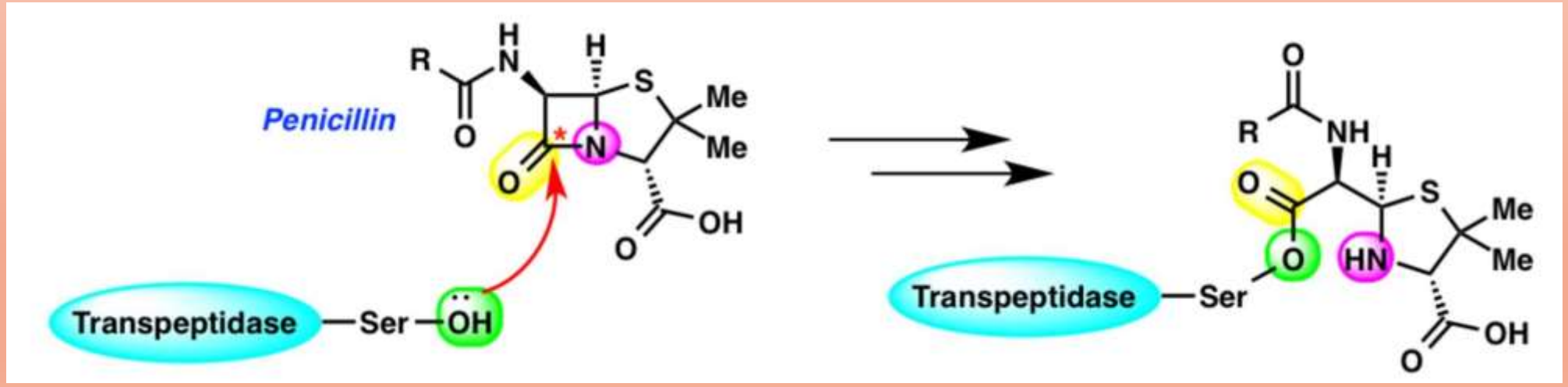


Ureidopenicillins

Mechanism of action of penicillin

Penicillin interferes with the synthesis of peptidoglycane, major component of the cell wall that protects bacteria from osmotic lysis.

It does this by reacting with and irreversibly inactivating an enzyme called transpeptidase. As a result cell wall synthesis halted and most bacteria undergo cell death, as the fragile inner membrane burst due to osmotic pressure.



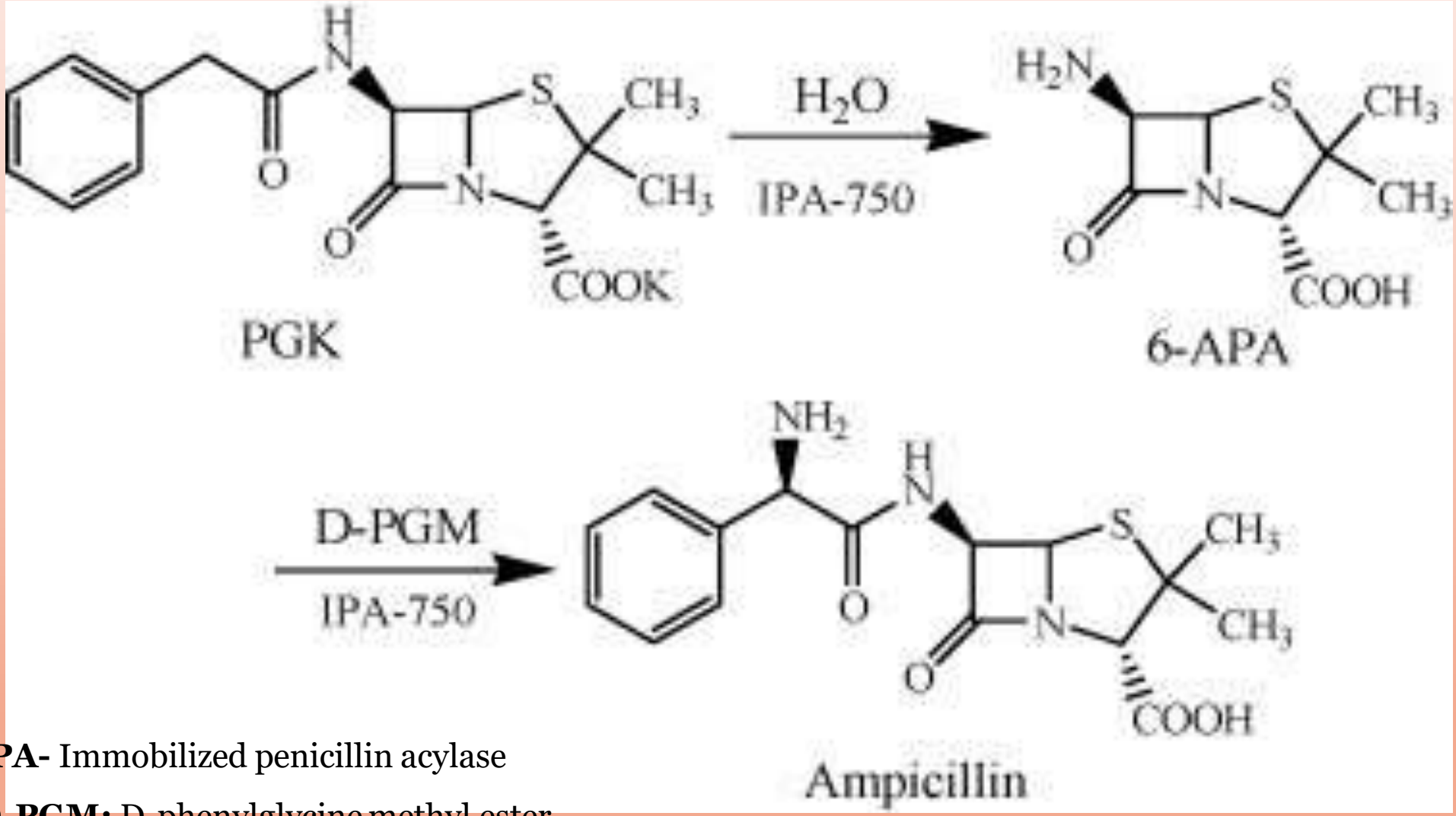
Resistance to Penicillin

Bacterial strains vary in resistant to penicillin.

Some species like sterptococci are quiet susceptible, *Pseudomonas aeruginosa* is particularly resistant, *S. aureus* are initially vulnerable, but acquire resistance when they are exposed to penicillin over a period of time. Several reasons for this variations:

- Physical barriers:
- Presence of β-lactamase
- High levels of transpeptidase enzymes produced in gram-negative
- Affinity of the transpeptidase enzyme to penicillin
- Transport back across the outer membrane of Gram-negative bacteria
- Mutations and genetic transfers

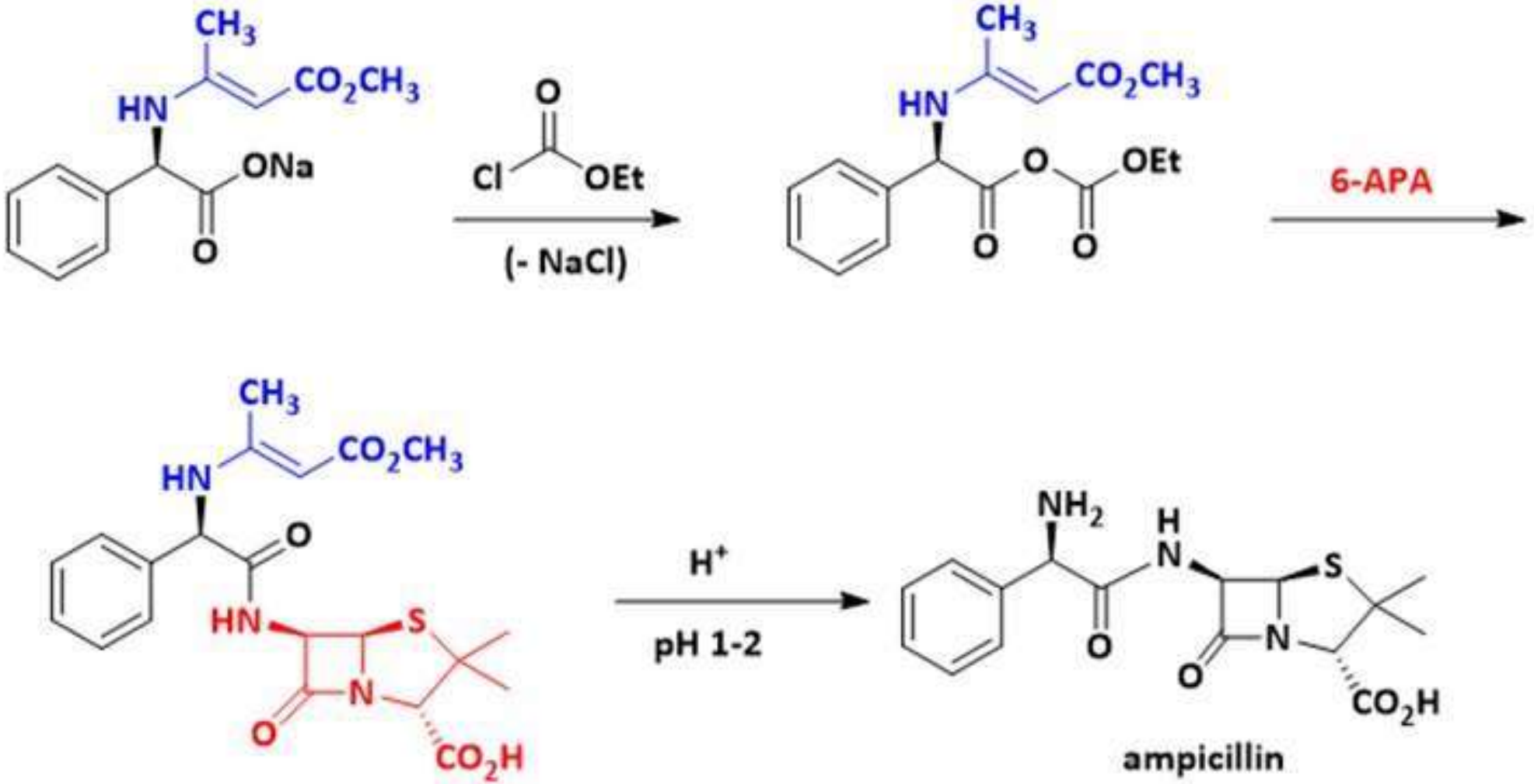
Synthesis of Ampicillin



IPA- Immobilized penicillin acylase

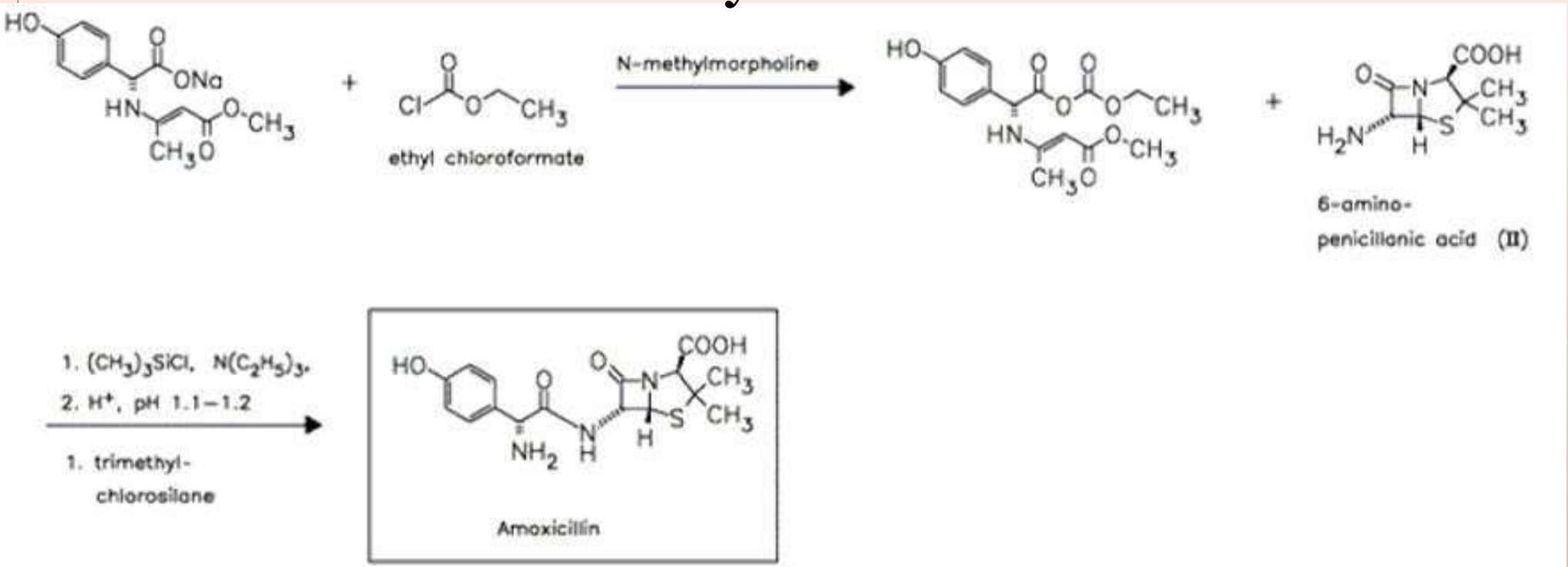
D-PGM: D-phenylglycine methyl ester

Synthesis of Ampicillin

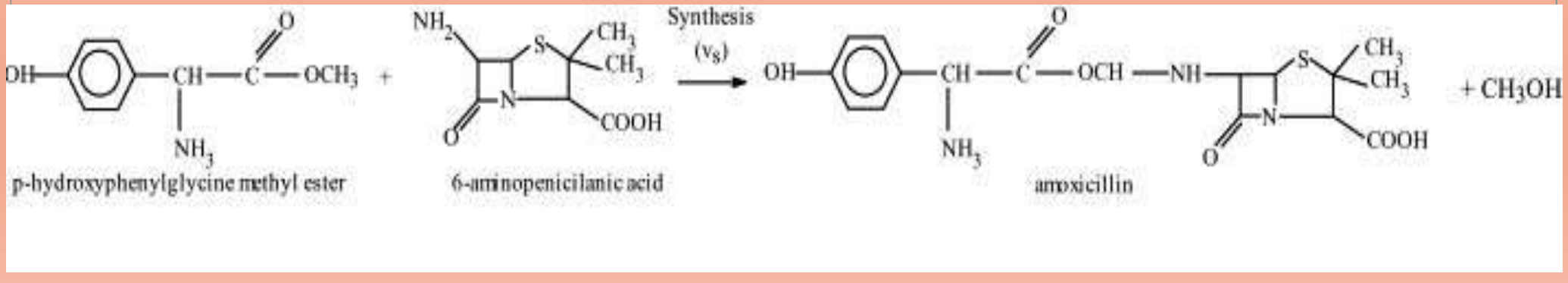


ampicillin

Chemical Synthesis of Amoxicillin



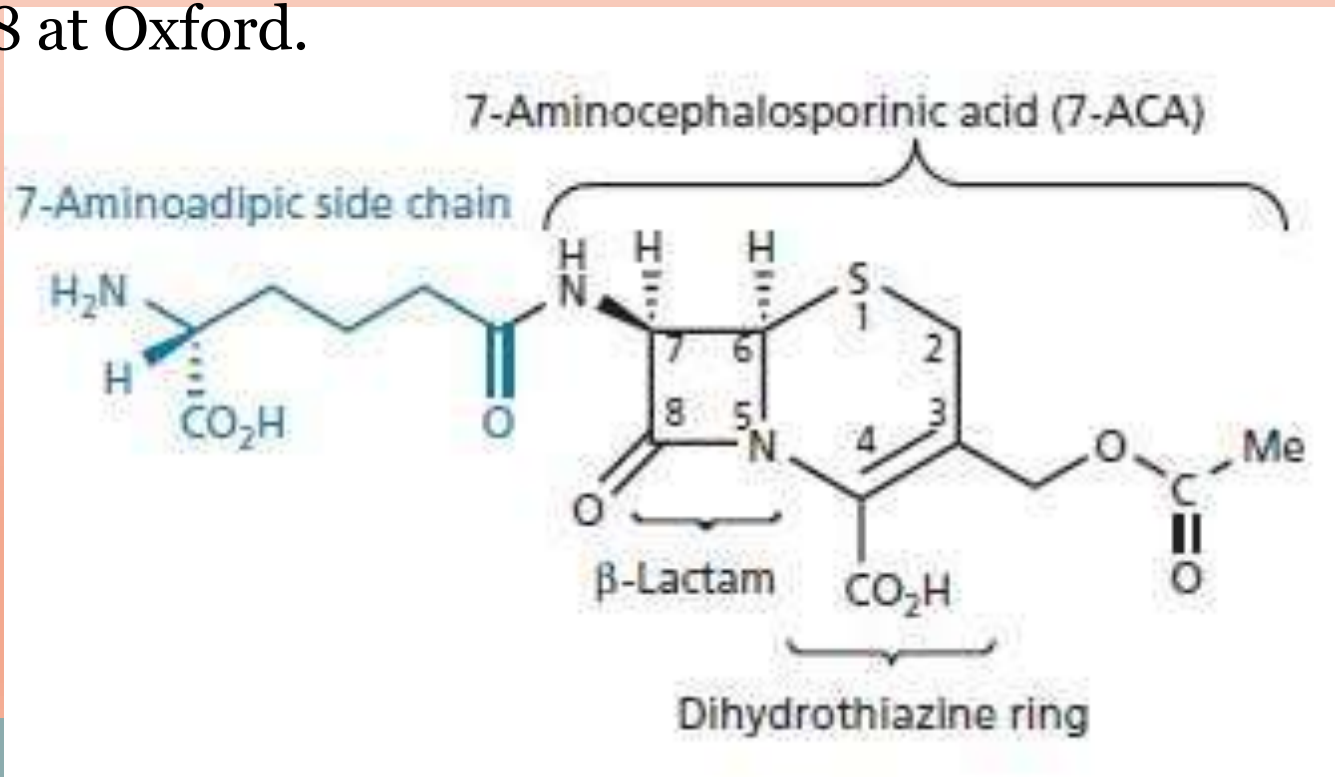
Enzymatic Synthesis of Amoxicillin



Cephalosoprins

Second major group of β-lactam antibiotics.

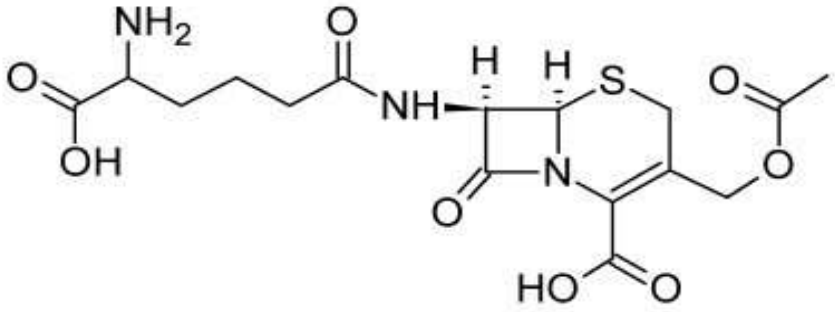
The first cephalosporin (**cephalosporin C) was derived from a fungus *Cephalosporium acremonium* obtained** in the mid 1940s from sewer waters on the island of Sardinia, and antibacterial activity studied in 1948 at Oxford.



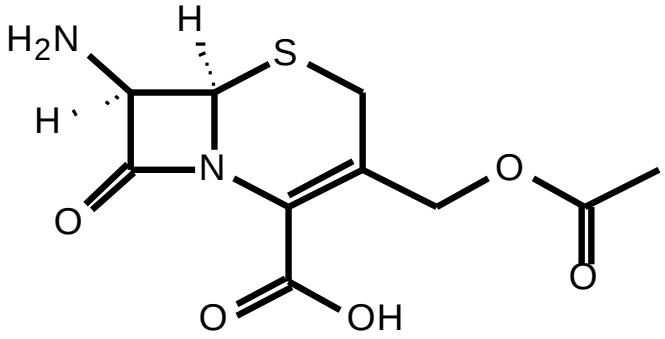
- By addition of different side chains at position **7** of β-lactam ring **(altering spectrum of activity)** and at position **3** of dihydrothiazine ring **(affecting pharmacokinetics)**, a large number of semisynthetic compounds have been produced.
- In-built advantage of cephalosporin-C is its greater resistance to acid hydrolysis and β–lactamase enzyme.

CEPHALOSPORIN C

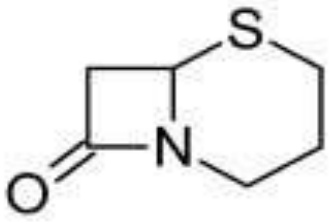
- It is a true cephalosporin and is a derivative of **7-aminocephalosporanic acid**. The latter served as a **lead nucleus** for the development of totally new series of compounds, **cephalosporins.**



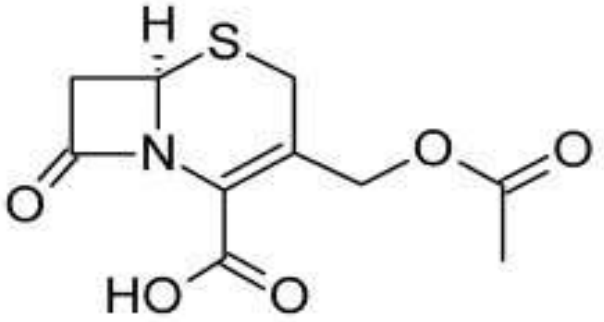
Cephalosporin C



7-Aminocephalosporanic acid (7-ACA)



Cepham



Cephalosporanic Acid

CEPHALOSPORIN CLASSIFICATION

- Cephalosporin are antimicrobial drugs, Acts by inhibiting bacterial cell wall synthesis
- Classified into 5 generations

1st GENERATION	2nd GENERATION	3rd GENERATION	4th GENERATION	5th GENERATION
• CEFAZOLIN • CEFALOTHIN • CEFALORIDINE • CEFADROXIL • CEPHALEXIN • CEPHRADINE	• CEFOXITIN • CEFPROZIL • CEFOTITAN • CEFMETAZOLE • CEFACLOR • CEFUROXIME	• CEFEXIME • CEFTAZIDIME • CEFOTAXIME • CEFTIZOXIME • CEFPODOXIME • CEFTRIAXONE • CEFOPERAZONE • CEFTIBUTEN • CEFDINIR • MOXALACTAM	• CEFEPIME • CEFPIROME	• CEFTOBIPROLE • CEFTAROLINE

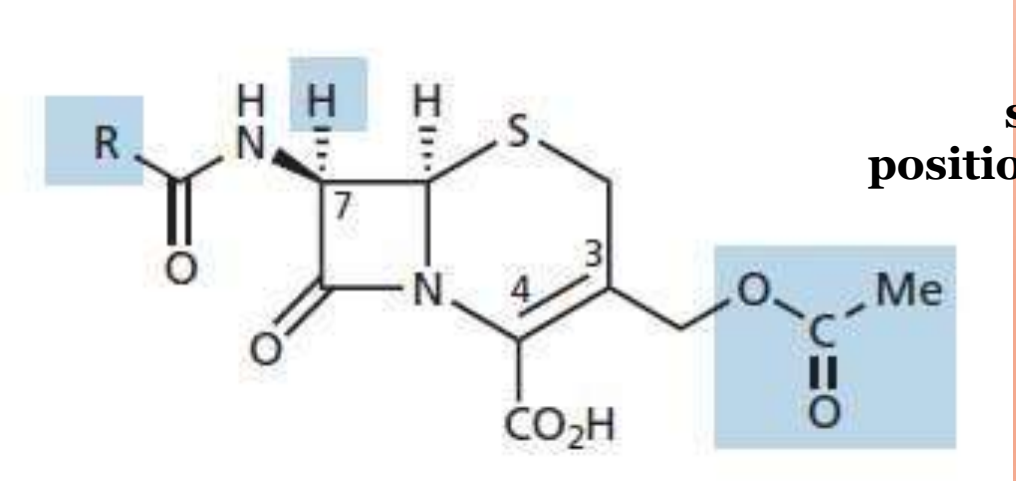
- Drugs that have 'FA' or 'PHA/PHRA' in their name are 1st generation except Cefaclor which is 2nd generation drug
- Drugs that end with 'IME' or 'ONE' or 'TEN' are 3rd generation except Cefuroxime which is 2nd generation drug
- Drugs that have 'PI' in their name are 4th generation
- Drugs that have 'ROL' in their name are 5th generation

SAR of Cephalopsorin C

Many analogues of cephalosporins have been made which demonstrate the SAR as follows-

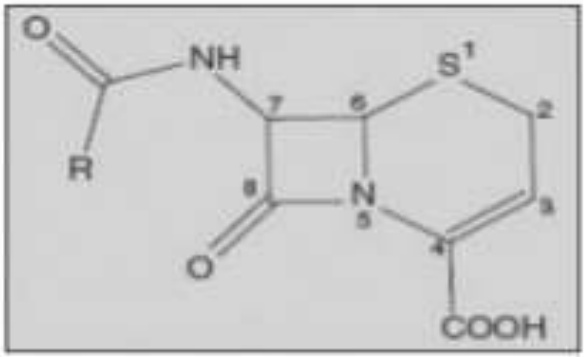
- β-lactam ring within the bicyclic system is essential.
- Free carboxylic group is needed at position 4.
- Acylamino group side chain at position 7

Molecular changes in cephalosporin were made to improve Stability, Antibacterial activity & stability towards β-lactamases



shading indicates positions which can be varied

SAR of cephalosporins



1. 7-Acylamino substituents:

a) Acylation of amino group generally increases the potency against gram-positive bacteria, but it is accompanied by a decrease in gram-negative potency.

b) High antibacterial activity is observed only when the new acyl groups are derived from carboxylic acids for gram-positive bacteria.

c) Substituents on the aromatic ring that increases lipophilicity provide higher gram-positive activity and generally lower gram-negative activity.

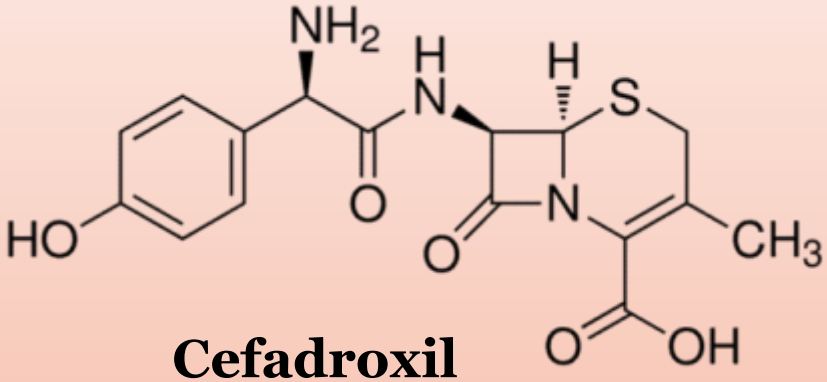
d) The phenyl ring in the side-chain can be replaced with other heterocycles with improved spectrum of activity and pharmacokinetic properties, and these include thiophene, tetrazole, furan, pyridine, and aminothiazoles

2. C-3 substituents: The nature of C-3 substituents influences pharmacokinetic and pharmacological properties as well as antibacterial activity. Modification at C-3 position has been made to reduce the degradation (lactone of desacetyl cephalosporin) of cephalosporins.

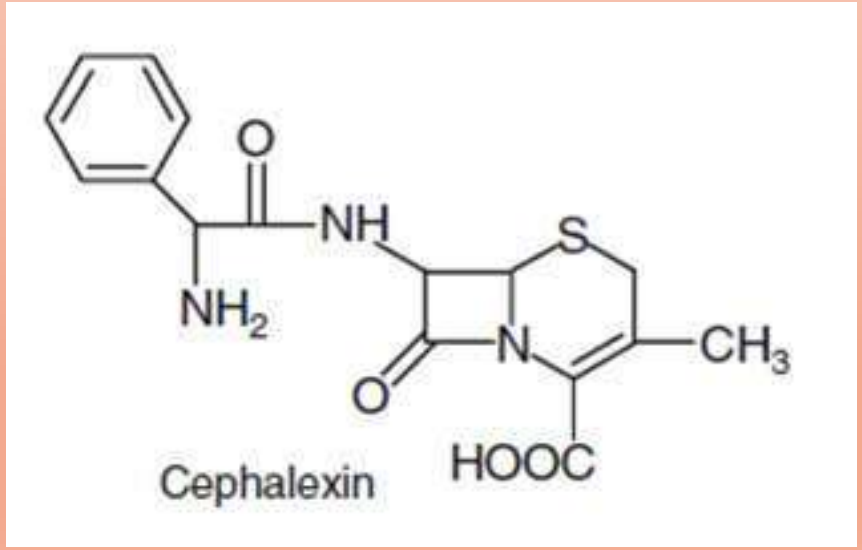
3. Pyridine and imidazole-replaced acetoxy groups show improved activity against *P. aeruginosa*. Displacement of acetoxy group by azide ion yields derivatives with relatively low gram-negative activity.
4. Displacement with aromatic thiols of 3-acetoxy group results in an enhancement of activity against gram-negative bacteria with improved pharmacokinetic properties.
5. Replacement of acetoxy group at C-3 position with $-CH_2Cl$ has resulted in orally active compounds.
6. Oxidation of ring sulphur to sulphoxide or sulphone greatly diminishes or destroys the antibacterial activity.
7. Replacement of sulphur with oxygen leads to oxacepam (latamoxef) with increased antibacterial activity, because of its enhanced acylating power.
8. Similarly, replacement of sulphur with methylene group (loracarbef) has greater chemical stability and a longer half-life.
9. The carboxyl group of position-4 has been converted into ester prodrugs to increase bioavailability of cephalosporins, and these can be given orally as well. Examples include cefuroxime axetil and cefodoxime proxetil
10. Olefinic linkage at C 3-4 is essential for antibacterial activity. Isomerization of the double bond to 2-3 position leads to great losses in antibacterial activity.

First generation Cephalosporin

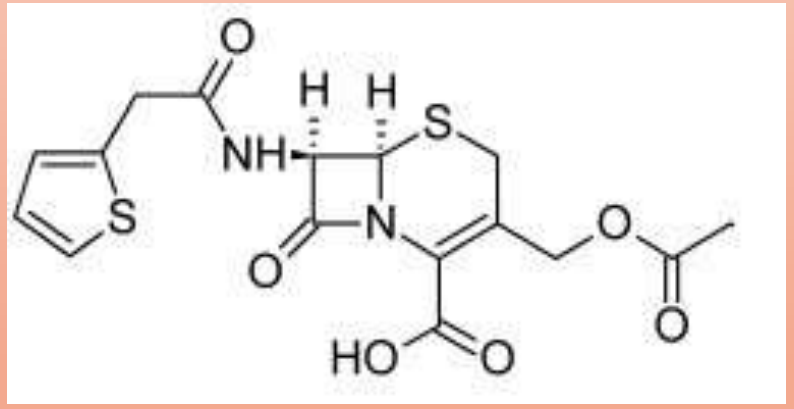
Cefazolin



Cefadroxil

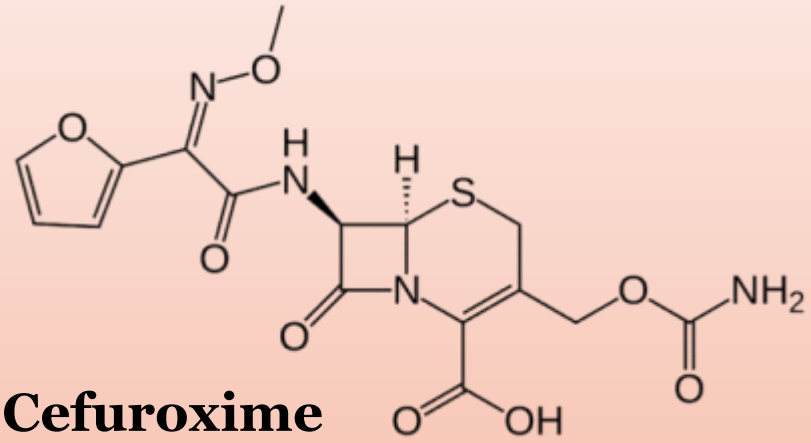


Cephalexin

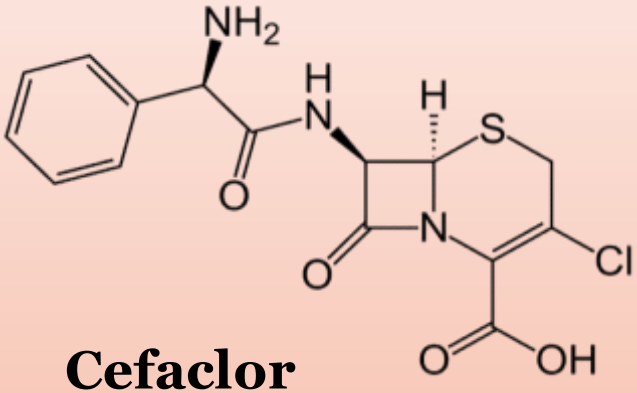


Cephalothin

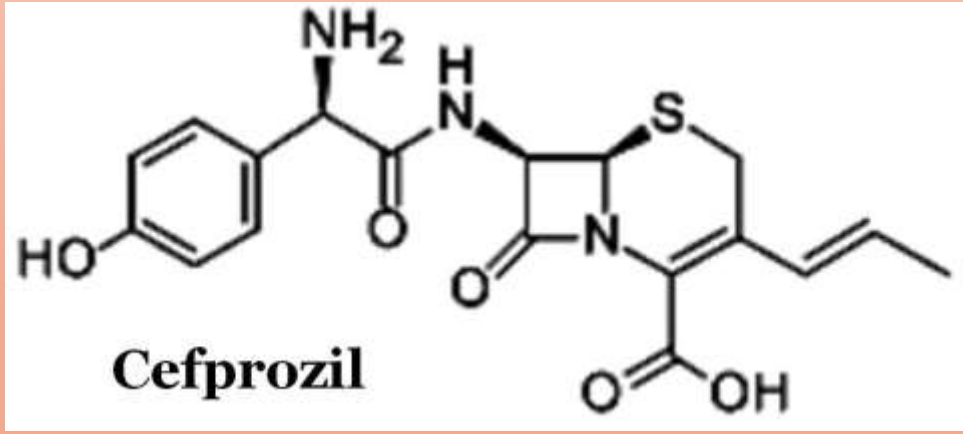
Second generation



Cefuroxime

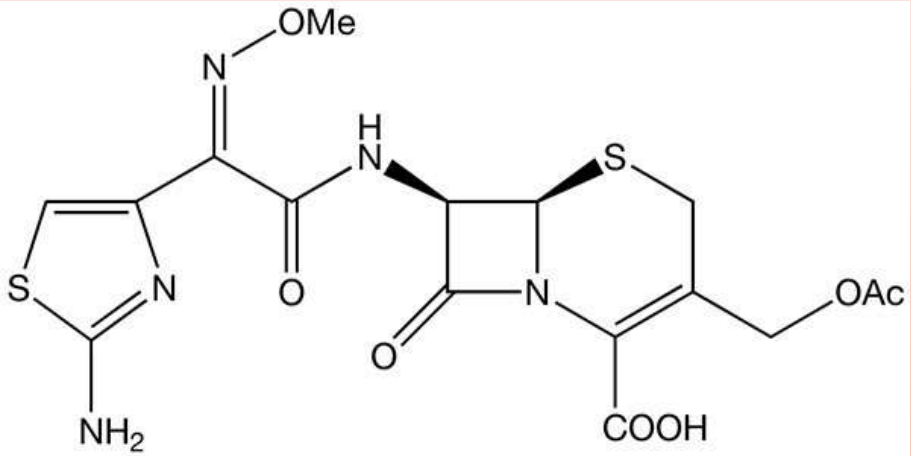


Cefaclor

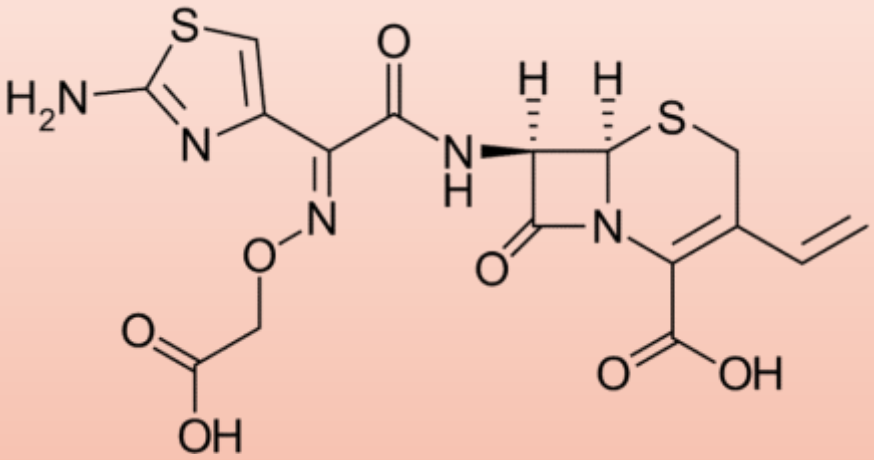


Cefprozil

Third generation

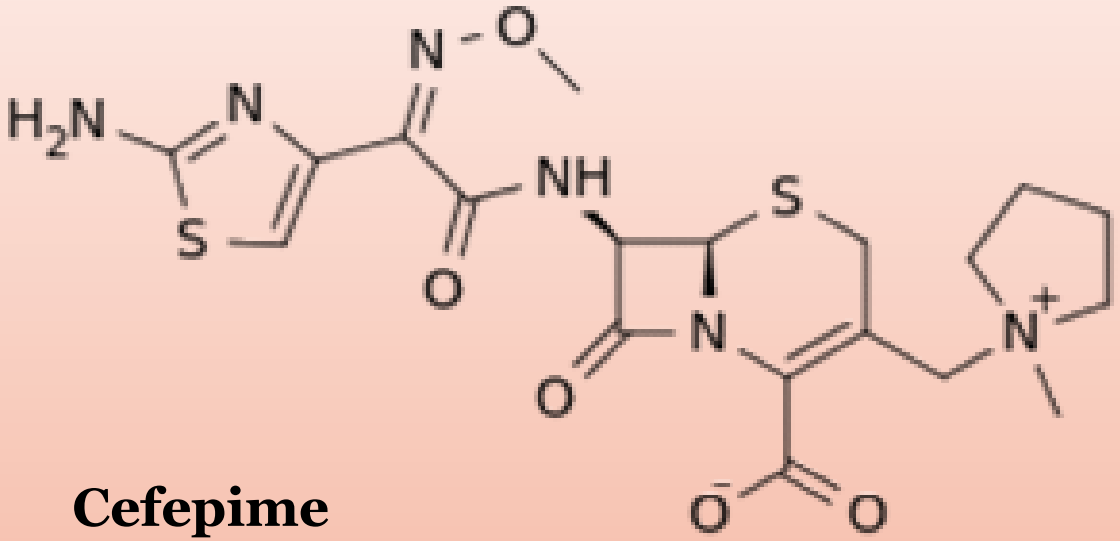


Cefotaxime

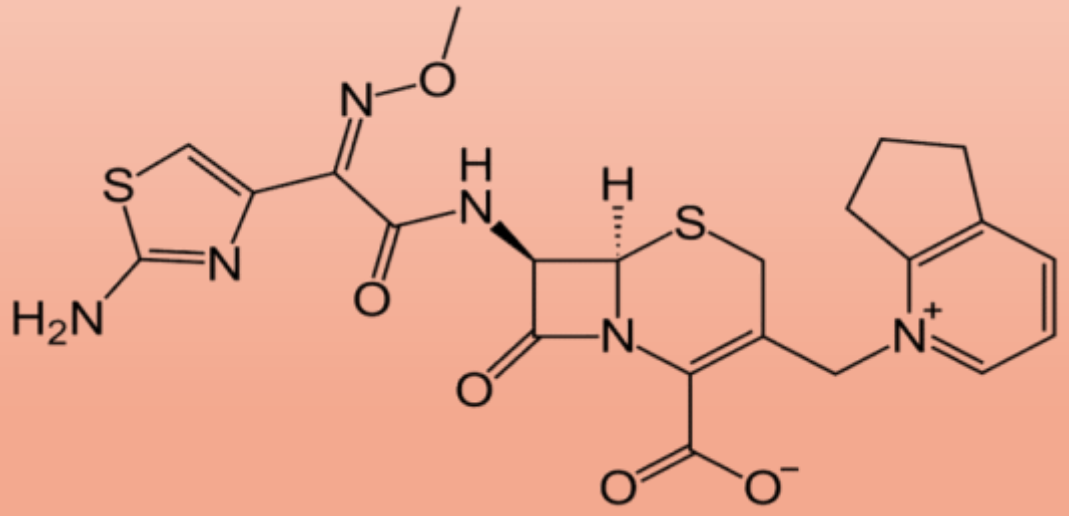


Cefixime

Fourth Generation



Cefepime



Cefpirome

Major classes of protein synthesis–inhibiting antibacterials

Chloramphenicol, macrolides, and lincosamides

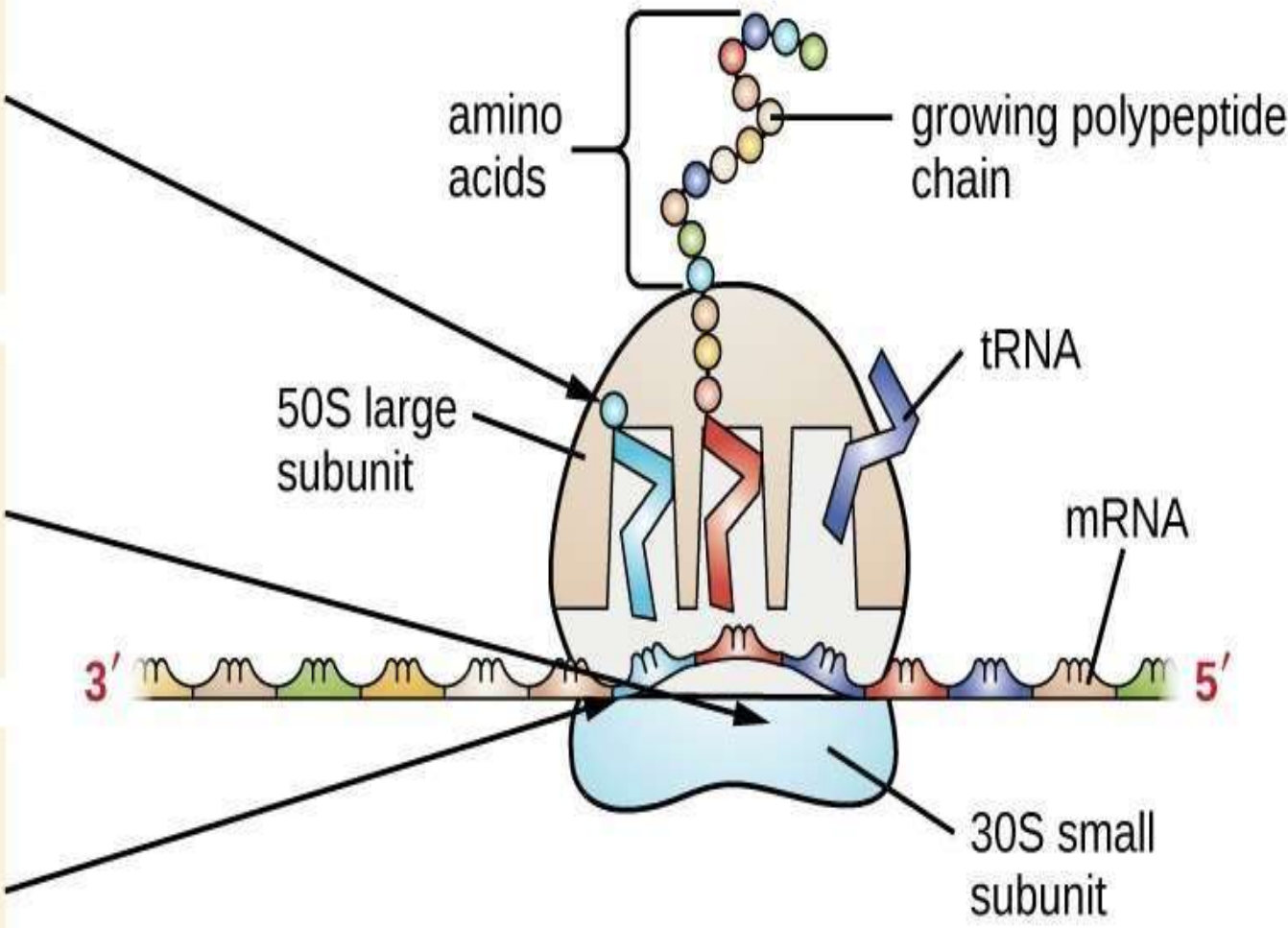
- Bind to the 50S ribosomal subunit
- Prevent peptide bond formation
- Stop protein synthesis

Aminoglycosides

- Bind to the 30S ribosomal subunit
- Impair proofreading, resulting in production of faulty proteins

Tetracyclines

- Bind to the 30S ribosomal subunit
- Block the binding of tRNAs, thereby inhibiting protein synthesis

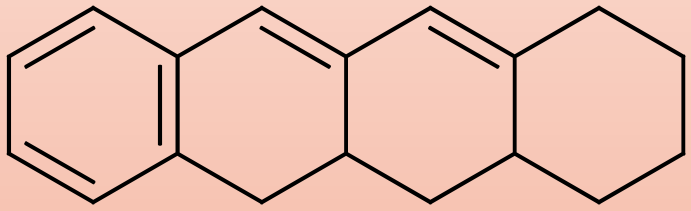


Tetracyclins

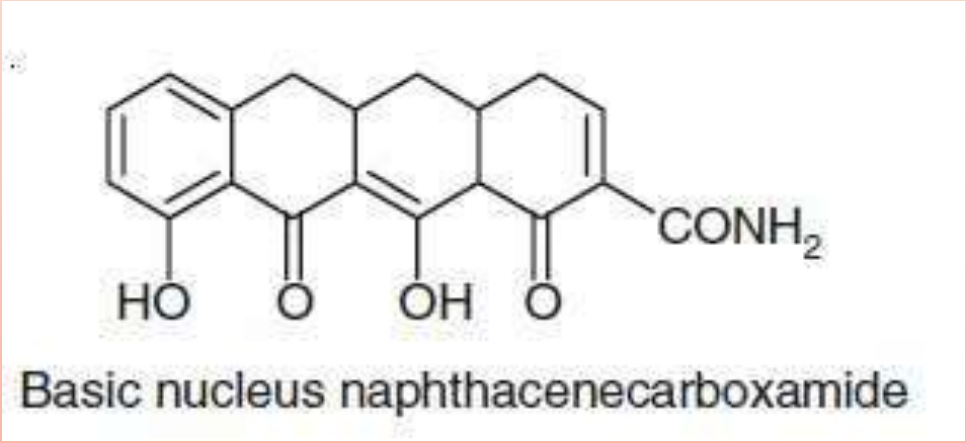
- Amongst most important **broad-spectrum antibiotics.**
- Nine such compounds, rolitetracycline, oxytetracycline, chlortetracycline, demeclocycline, meclocycline, methacycline, doxycycline, and minocycline- have been introduced into medical use.
- Tetracyclines are obtained by fermentation procedures from ***Streptomyces*** *spp. or by chemical transformations* of the natural products.
- Other choices & high incidence of resistance decreased their medical importance.
- Still recommended for use against rickettsia, chlamydia, mycoplasm, anthrax, plague & helicobacter organisms.

Structures of tetracyclines

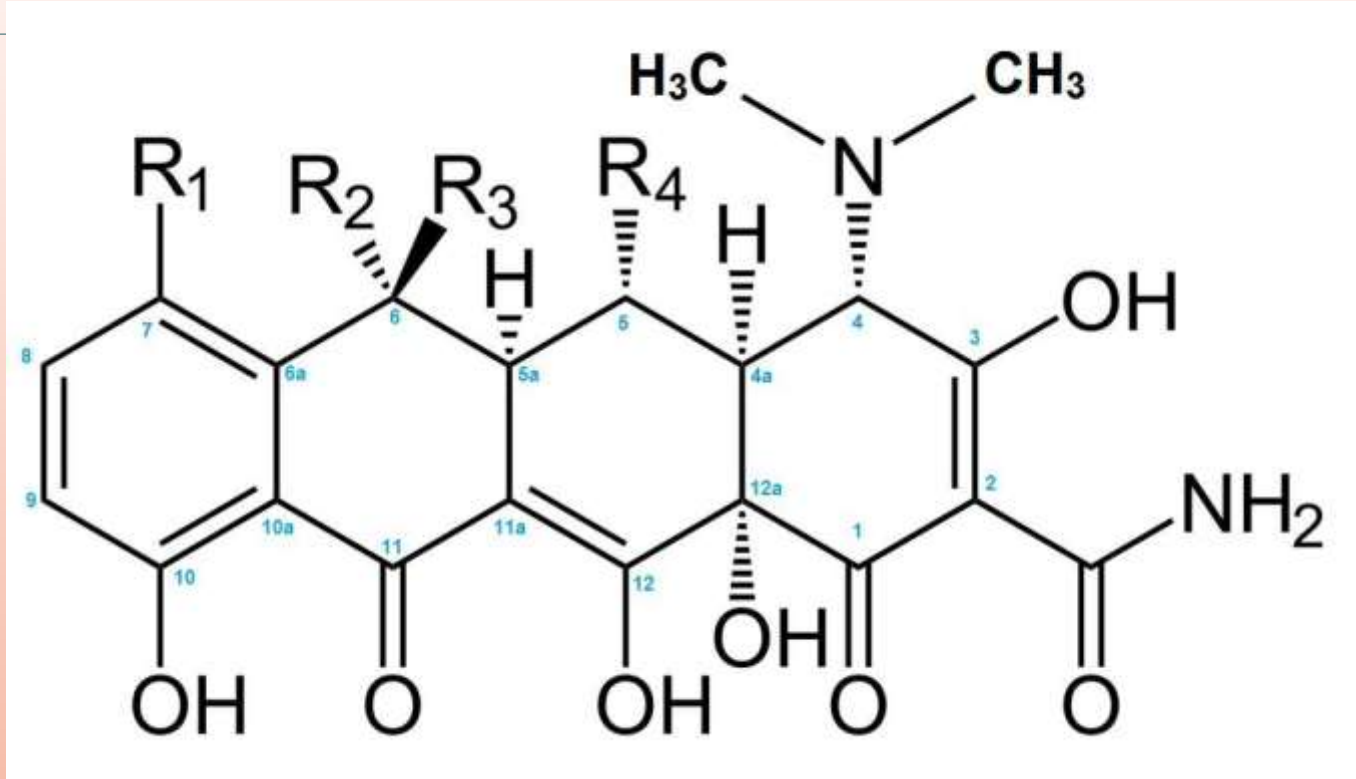
- They are considered as derivatives of the **octahydronaphthacene** and basic nucleus is **naphthacene carboxamide.**



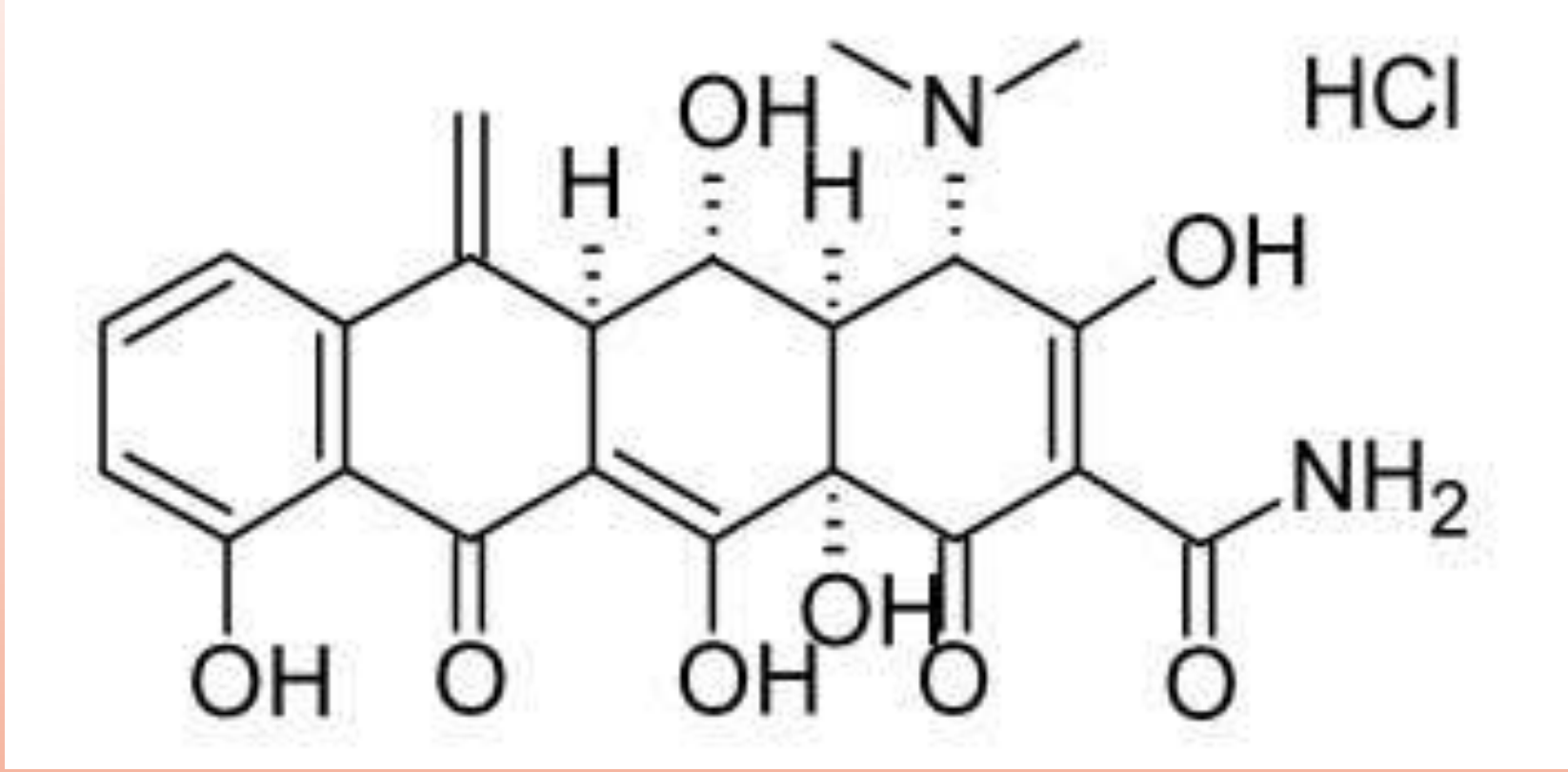
octahydronaphthacene



Basic nucleus naphthacenecarboxamide

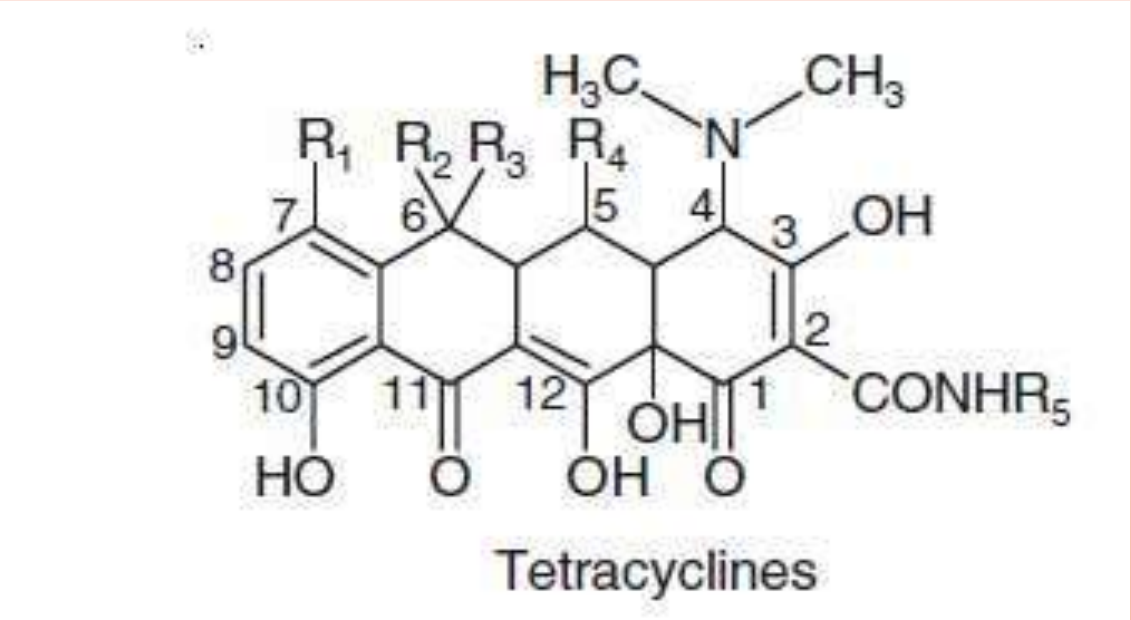


	R_1	R_2	R_3	R_4
Tetracycline	H	OH	CH_3	H
Chlortetracycline	Cl	OH	CH_3	H
Oxytetracycline	H	OH	CH_3	OH
Demeclocycline	Cl	OH	H	H
Methacycline	H	CH_2		OH
Doxycycline	H	CH_3	H	OH
Minocycline	$N(CH_3)_2$	H	H	H



Methacyclin

Structure of Activity Relationship

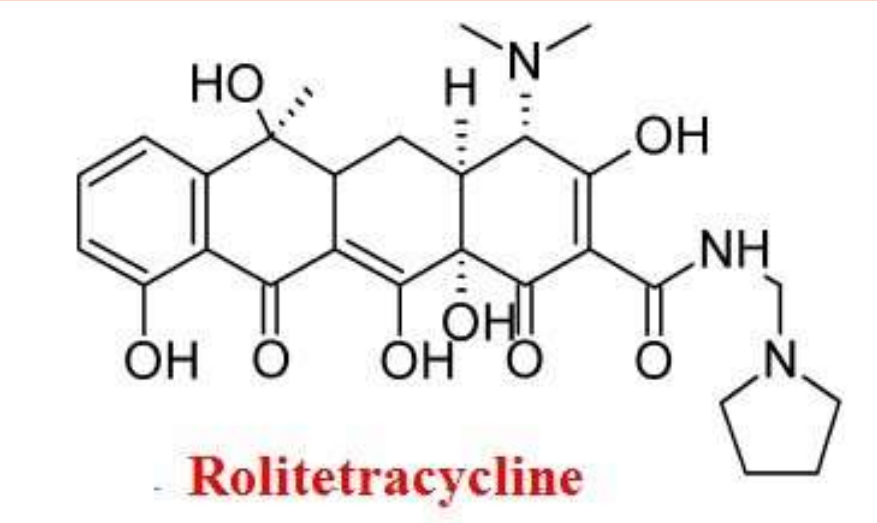


Tetracyclines

1. A tetracycline backbone skeleton is essential for activity.
2. The carbon system present at carbons 1 to 3 must be intact for good activity.
3. Replacement of amide at C-2 with other functions such as aldehyde, nitrile reduces or abolishes activity.
4. Monoalkylation of the amide nitrogen reduces activity.

5. Epitetracyclines are very much less active than the neutral isomers.
6. Removal of 4-dimethylamino group reduces activity. Activity is largely retained in the primary and secondary amines but rapidly diminishes in the higher alkylamines.
7. Alkylaton of 11a also leads to inactive compounds
8. Dehyrogenation to form a double bond between C5a and C11a markdly decreases activity.
9. Substituent's at positions 5,6,7,8,9 can be modified with varying degrees of impurity resulting in retention and in some cases improvement of antibiotic activity.
10. A 5-hydroxy group as in Oxytetracyclin and doxycycline may influence pharmacokinetic properties but does not change antimicrobial activity as compared to 5-deoxy compounds.
11. Strongly electron withrawing groups e.g. Chloro and nitro and strongly electron donation groups e.g. Dimethylamino enhance the activity.
12. The effect of introducing substitutents at C-8 has not been studied because this position cannot be substituted.

13. Thiatetracyclines contain sulfur atom at C-6. A recent derivative thiacycline is found to be more active than minocycline against tetracycline-resistant bacteria.
14. Tetracyclines have low solubility in water which may be overcomed by aminoalkylation at carboxamido group. The clinically effective mannich bases are rolitetracycline (pyrrolidinomethyl-tetracycline)



Rolitetracycline

15. Semisynthetic analogs have also been obtained in an attempt to achieve advances in chemotherapy. Methacycline, doxycycline and minocycline are some results of such deliveries.

Mechanism of action of Tetracyclins

- Tetracyclines are specific **inhibitors of bacterial protein synthesis.**
- **They bind to the 30S ribosomal subunit and, thereby, prevent the binding of aminoacyl tRNA (charged tRNA) to the mRNA–ribosome complex.**
- Both the binding of aminoacyl tRNA and the binding of tetracyclines at the ribosomal binding site require magnesium ions.

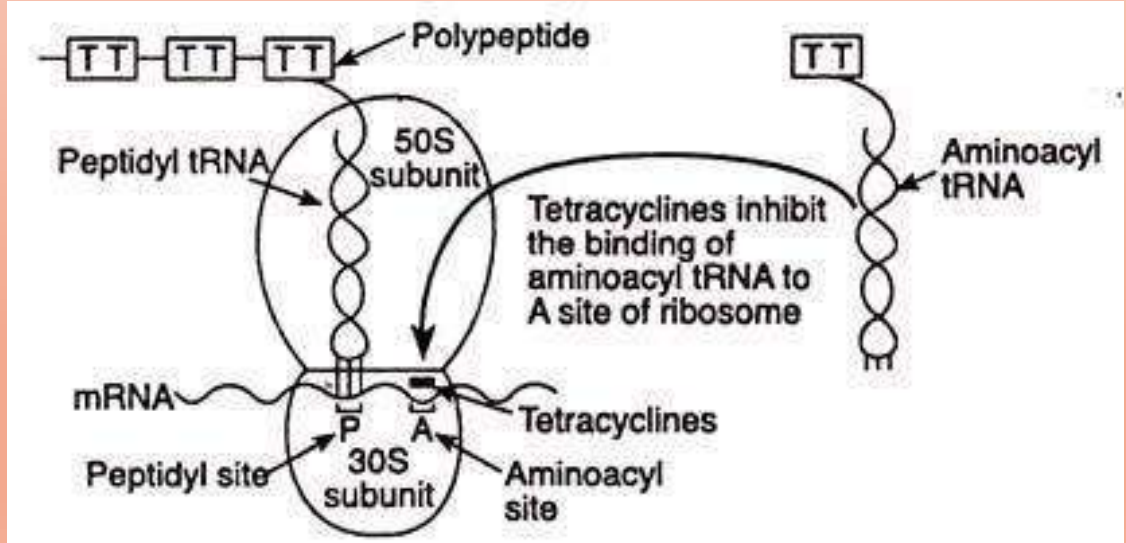
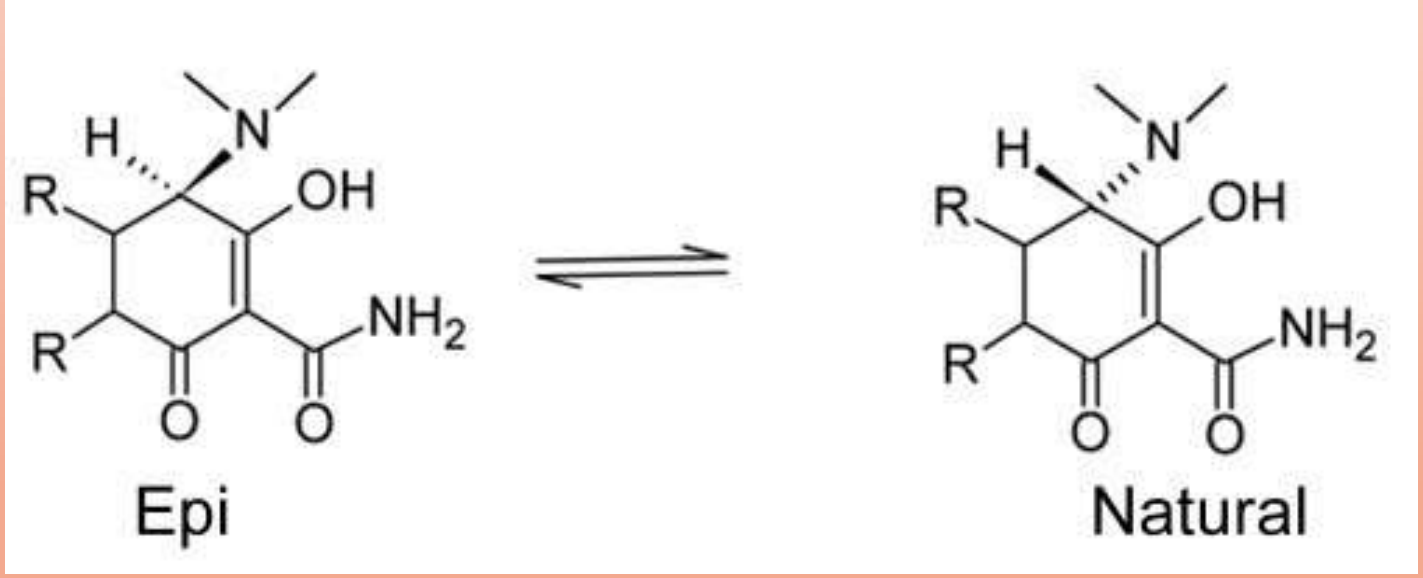


FIG. 45.15. Schematic representation of mechanism of action of tetracycline antibiotics.

Chemical Instability of tetracyclines

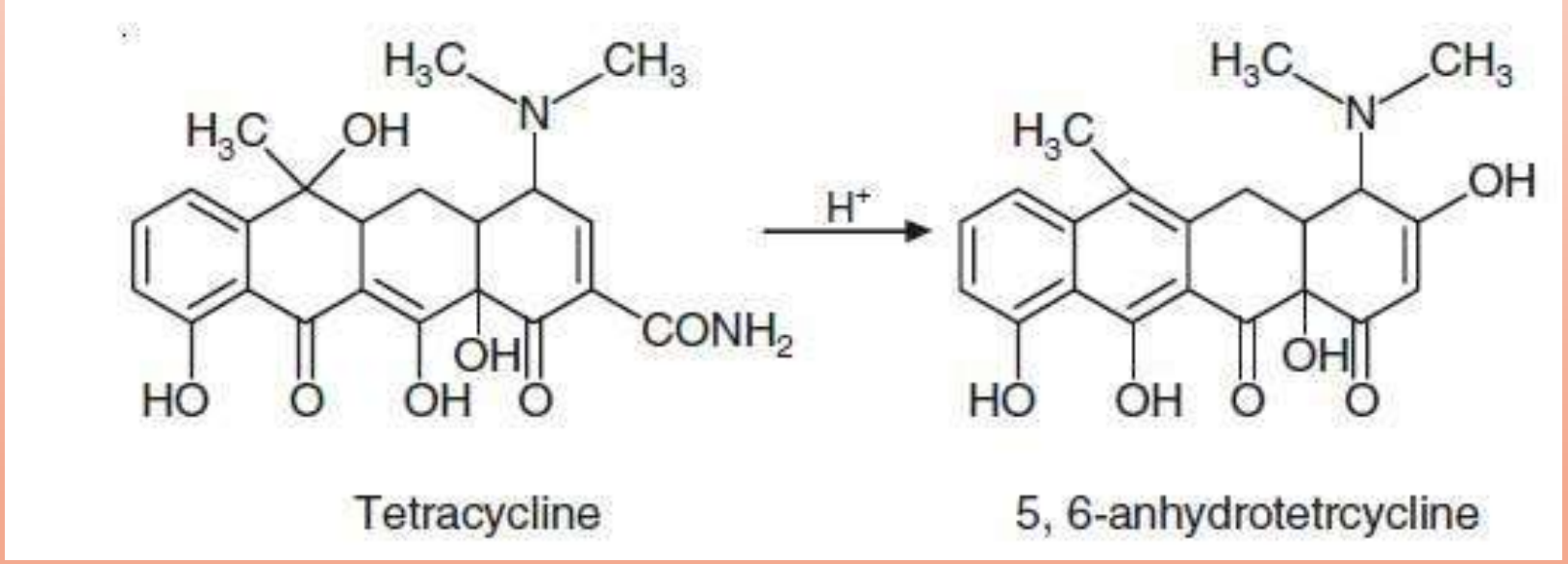
1) Epimerization:

- **All the tetracyclines undergo epimerization at carbon-4 in solution between pH 2 and 6**
- 4-epitetracyclines exhibit much less activity than the “natural” isomers.



2) Dehydration by acid:

Strong acid attacks the tetracyclines at hydroxyl group C-6 atom, causing the loss of activity through modification of the C-ring that results in the formation of inactive tetracyclines like anhydrotetracyclines.

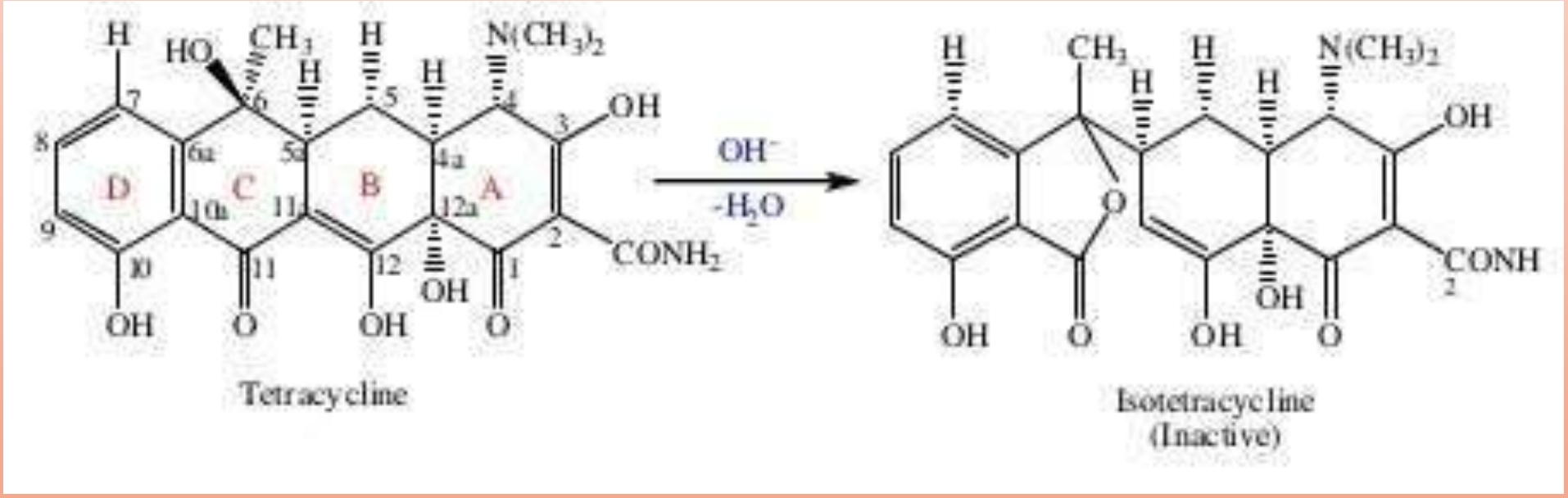


Tetracycline

5, 6-anhydrotetrcycline

3) Dehydration by base:

On the other hand; bases promote reaction (at or above pH 8.5) between 6-hydroxyl group and the ketone group at the 11-position that causes the formation of the lactone ring which is inactive and named as isotetracyclines.



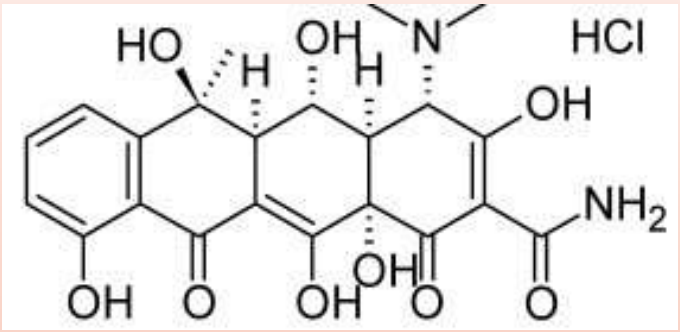
4) Chelation:

- This insolubility is not only inconvenient for the preparation of solutions but also interferes with blood levels on oral administration.
- The tetracycline's are incompatible with co-administered, multivalent ion-rich antacids and with hematinics and concomitant consumption of daily products rich in calcium ion also is contraindicated.

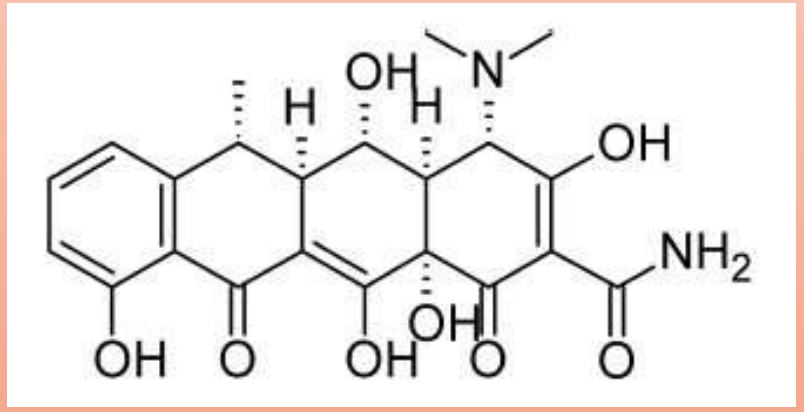
Contraindications of Tetracycline Antibiotics :

- Can stain developing teeth (even when taken by the mother during pregnancy)
- Inactivated by Ca2+ ion, not to be taken with milk, yogurt, and other dairy products.
- Skin photosensitivity; exposure to the Sun or intense light is not recommended
- Drug-induced lupus, and hepatitis
- Can induce microvesicular fatty liver.
- May interfere with methotrexate by displacing it from the various protein binding sites

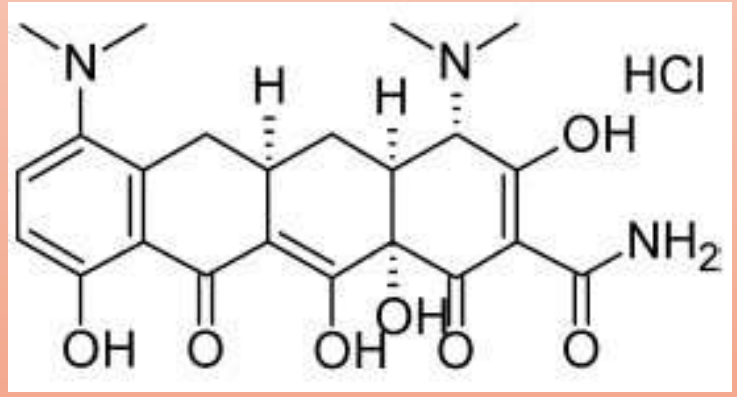
Tetracycline



Oxytetracycline



Doxycycline



Minocycline

Aminoglycoside antibiotics (Aminocylitol antibiotics)

The aminoglycoside as so named because they are composed of aminosugars linked by glycosidic linkages. Most aminoglycosides are prepared by natural fermentation from various species of ***streptomyces*** except Gentamicin and Amikacin**.**

The aminoglycosides consist of two or more amino sugars joined in glycoside linkage to a highly substituted 1, 3-diaminocyclohexane (aminocyclitol) centrally placed ring. Thus:

a.In kanamycin and gentamicin families, two amino sugars are attached to **2-deoxystreptamine.**

b. In streptomycin, two amino sugars are attached to **streptidine.**

c.In neomycin family, there are three amino sugars attached to **2-deoxystreptamine**

SAR of **Aminoglycoside Antibiotics**

The aminoglycoside antibiotics contain two important structural features:

a. Amino sugar portion
b. Centrally placed hexose ring either 2-deoxystreptamine or streptidine

<u>SAR of First Amino Sugar Portion</u>

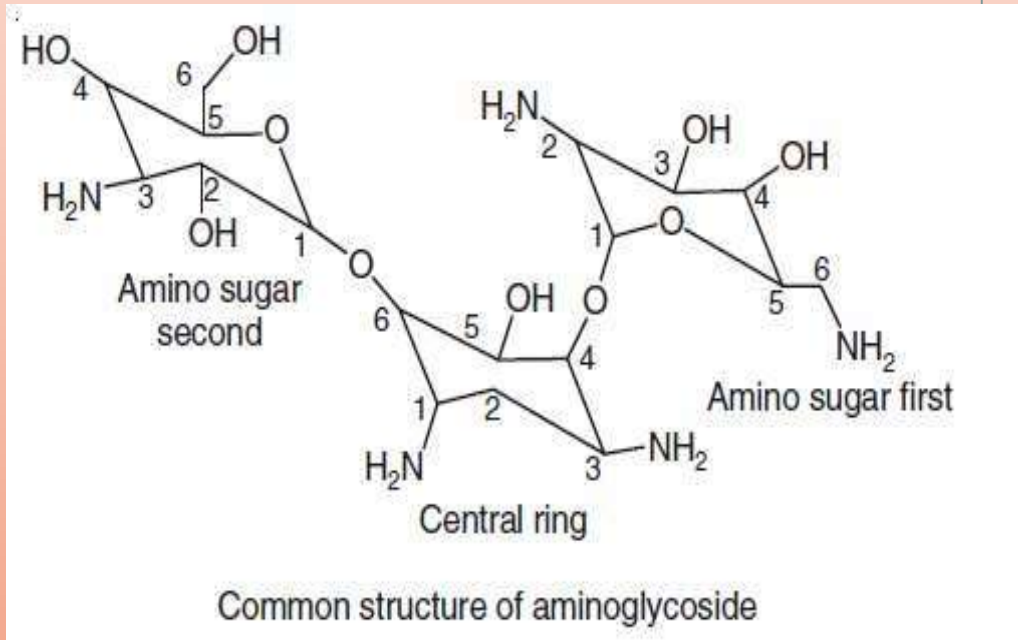
a.The amino functions at C-6 and C-2 serve as a major target sites for bacterial inactivating enzymes.

b.Methylation at C-6 positions does not decrease the activity; instead increases enzyme resistance.

c.Removal of 3-hydroxyl or 4-hydroxyl or both groups does not affect the activity.

d.This ring is essential for characteristic broad-spectrum antibacterial activity.

This ring is mainly responsible for inactivating the bacterial enzyme.



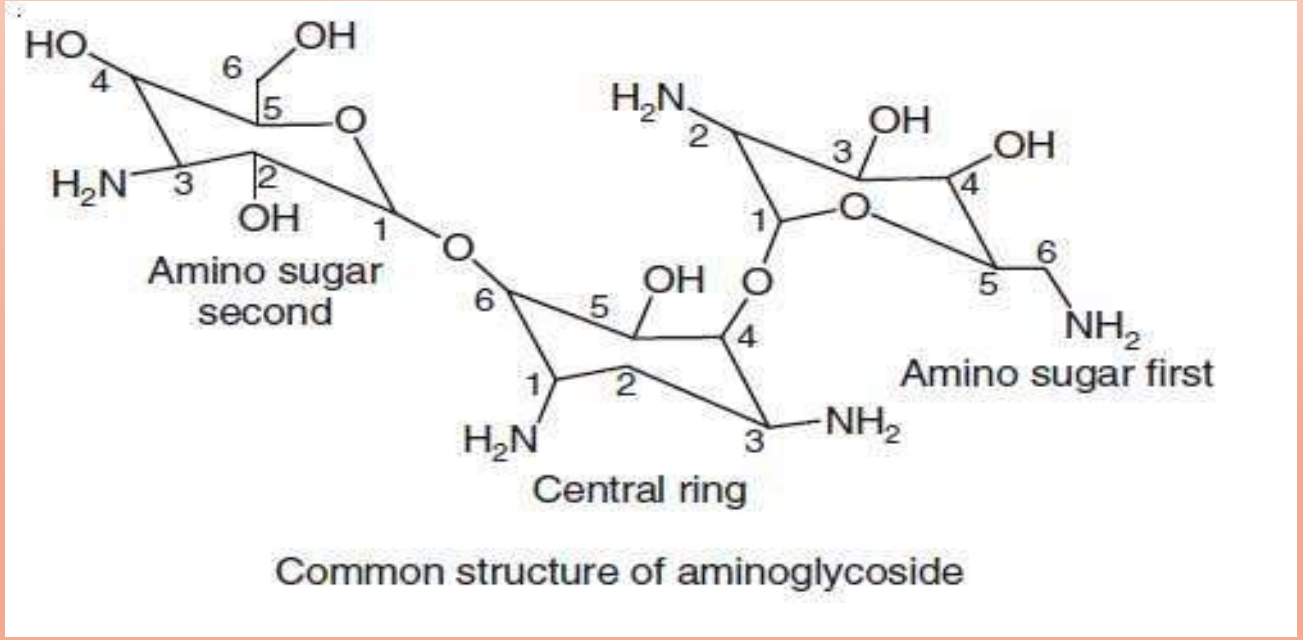
Common structure of aminoglycoside

SAR of Second Amino Sugar Portion

a. Here functional changes are less sensitive as compared to that of first amino sugar.

b. Only replacement of 2-OH by amino may increase the activity.

c. Similarly replacement at C-3 by secondary and tertiary amine for primary amine increases the antibacterial activity.



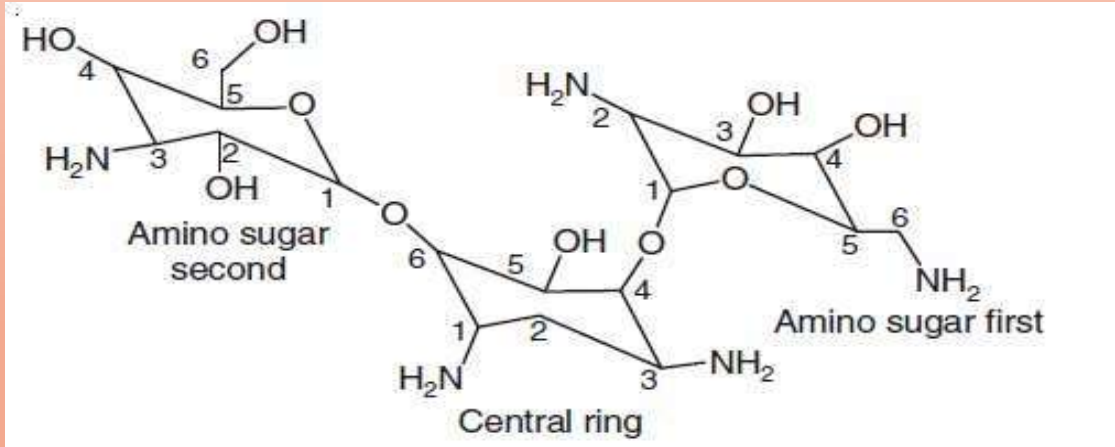
Common structure of aminoglycoside

SAR of Centrally Placed Hexose Ring

a. Various modifications at C-1 amino group have been tested. The acylation (e.g. amikacin) and ethylation (e.g. 1- N-ethylsisomicin) though not increases the activity helps to retain the antibacterial potency.

b. In sisomicin series, 2-hydroxylation and 5-deoxygenation results in increased inhibition of bacterial inactivating enzyme systems.

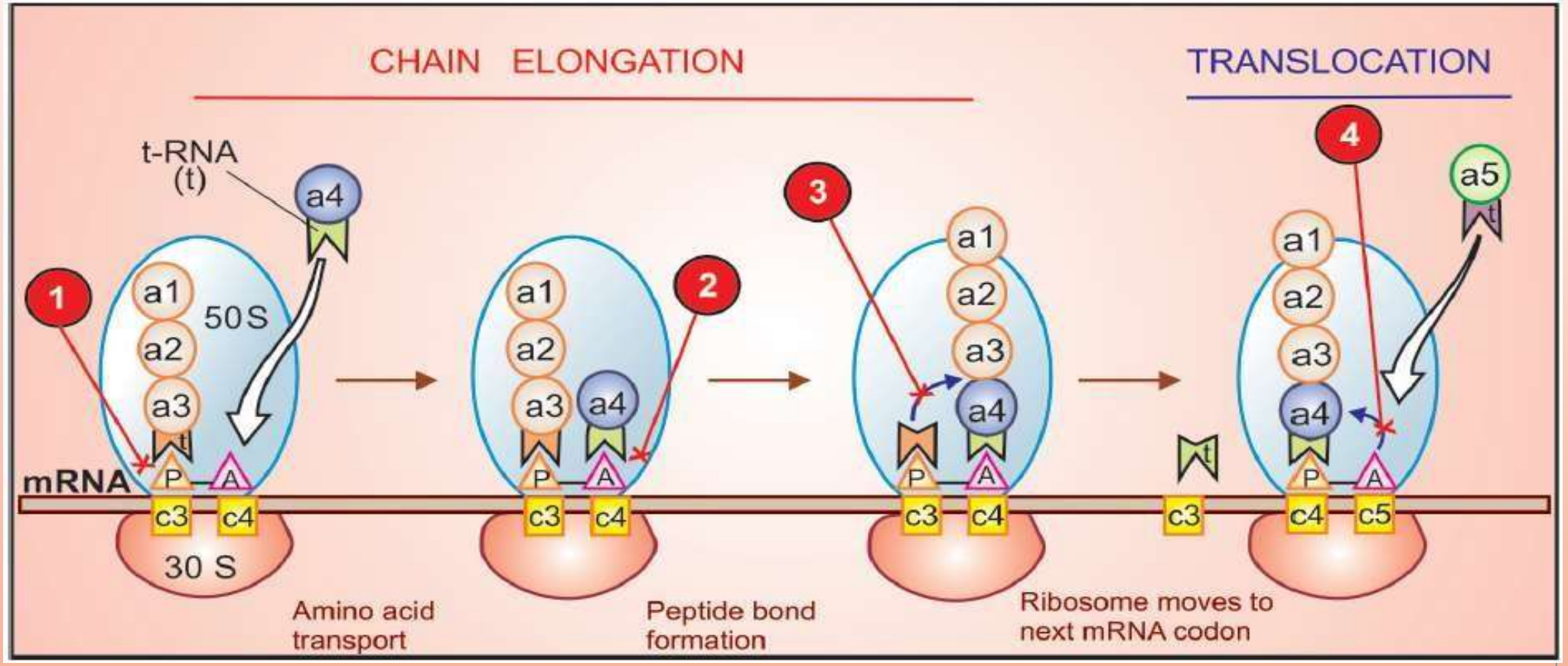
c. Thus very few modifications of the central ring are possible which do not violate the activity spectrum of aminoglycosides.



Common structure of aminoglycoside

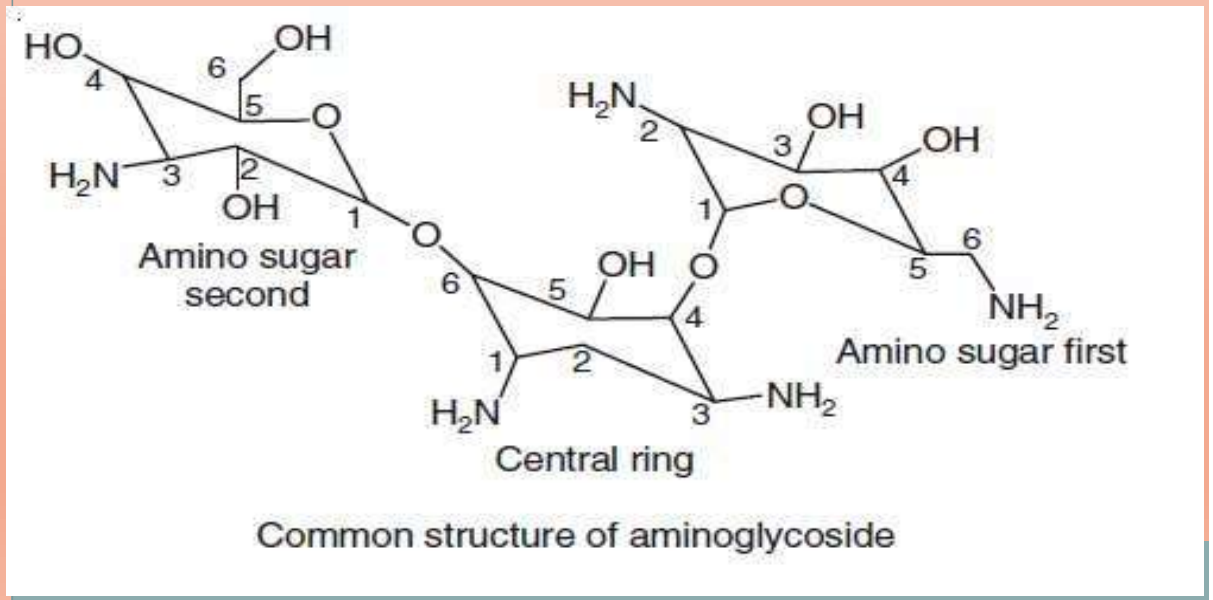
Mechanism of action

- The messenger RNA (mRNA) attaches to the 30S ribosome. The initiation complex of mRNA starts protein synthesis and polysome formation.
- The nacent peptide chain is attached to the peptidyl (P) site of the 50S ribosome. The next amino acid (a) is transported to the acceptor (A) site of the ribosome by its specific tRNA which is complementary to the base sequence of the next mRNA codon (C).
- The nascent peptide chain is transferred to the newly attached amino acid by peptide bond formation. The elongated peptide chain is shifted back from the 'A' to the 'P' site and the ribosome moves along the mRNA to expose the next codon for amino acid attachment.
- Finally the process is terminated by the termination complex and the protein is released.
- Aminoglycosides bind to several sites at 30S and 50S subunits as well as to their interface—freeze initiation, interfere with polysome formation and cause misreading of mRNA code.

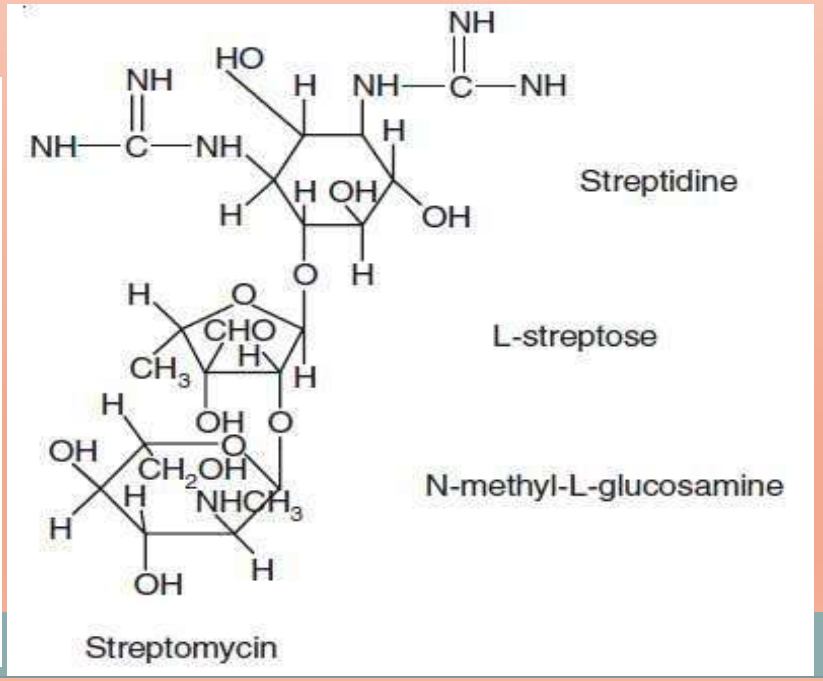

CHAIN ELONGATION
TRANSLOCATION
t-RNA
(t)
a4
a5
1
2
3
4
a1
a2
a3
50S
mRNA
P
A
c3
c4
c5
30 S
Amino acid
transport
Peptide bond
formation
Ribosome moves to
next mRNA codon

Streptomycin:

- The first aminoglycoside antibiotic to be used in chemotherapy
- Aminoglycosides are so named because their structures consist of amino sugars linked glycosidically.
- All have at least one aminohexose and some have pentose lacking an amino group (e.g. streptomycin, neomycin, and paromomycin).
- Each of clinically useful aminoglycosides contains a highly substituted 1,3-diaminocyclohexane central ring.



Common structure of aminoglycoside



Streptomycin

Structure of streptomycin

The aminoglycoside antibiotics contain two important structural features:

- **Amino sugar portion.**
- **Centrally placed hexose ring either 2-deoxystreptamine or streptidine.**

- This series includes streptomycin, gentamiycin, neomycin, kanamycin, tobramycin, amikacin, netilmicin, spectinomycin and framycetin.

- These consist of amino sugars linked glycosidically. They are all mixture of water soluble, basic carbohydrates that are closely related chemically.

- They inhibit the growth of gram-positive, gram-negative and mycobacteria. Except for the gentamicins, all are the products of species of streptomyces.

Unclassified Antibiotics Chloramphenicol)

These antibiotics retain their reputation in the chemotherapy, though do not retain common structural features with any other antibiotic. Due to their high clinical utility, they deserve special attention.

History

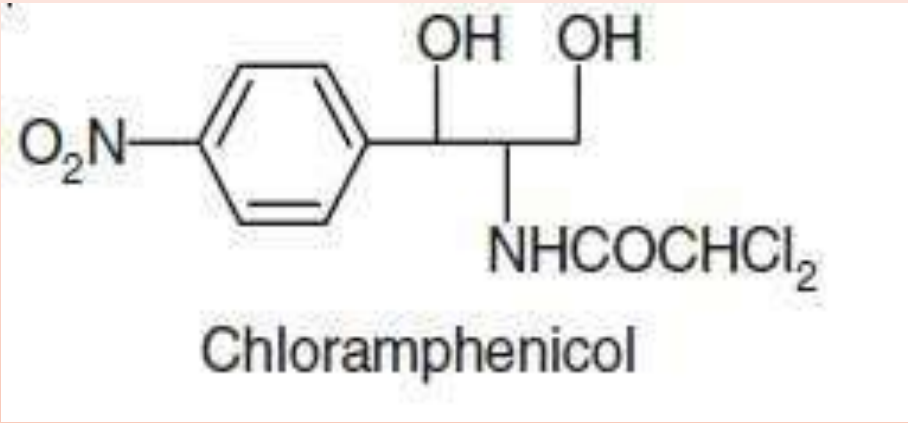
Originally isolated from *Streptomyces venezuelae by Ehlrich et al in* 1947, *chloramphenicol* (chloromycetin) is now produced totally by a synthetic route. It contains chlorine and is obtained from an actinomycete so named as chloromycetin.

Spectrum of Action

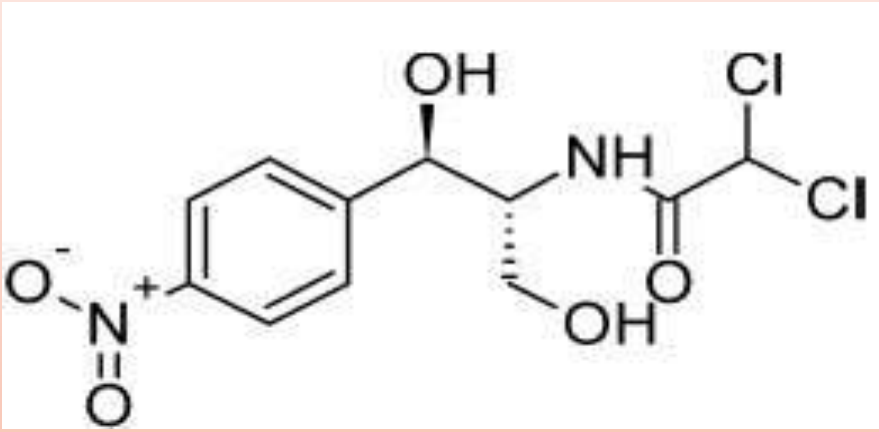
Chloramphenicol has a spectrum of activity resembling with that of the tetracyclines except that it exhibits a bit less activity against some gram-positive bacteria. It is specifically recommended for the treatment of serious infections caused by ***Hemophilus influenzae, Salmonella typhi (typhoid), Streptococcus pneumoniae and Neisseria meningitidis***.

It possesses marked effectiveness against several gram-negative bacteria and also exhibits antirickettsial activity. Its ability to penetrate into CNS presents an alternative therapy for meningitis.

SAR of Chloramphenicol



Chloramphenicol



SAR of Paranitrophenyl Group

1. Replacement of the nitro group by other substituents leads to reduction in activity.
2. Shifting of the nitro group from the para positions also reduces the antibacterial activity.
3. Replacement of phenyl group by the alicylic moieties results in less potent compounds.
4. The p-nitrophenyl group may be replaced by other aryl structures without appreciable loss of activity.

SAR of Dichloroacetatamido side chain

1. Other dihalo derivatives of the side chain are less potent though major activities are retained.

2.While in case of trihalo derivatives, Hanch et al in the light of QSAR calculations claimed that the 2 -NHCOCF3 derivative would be about 1.7 times as active as the chloramphenicol.

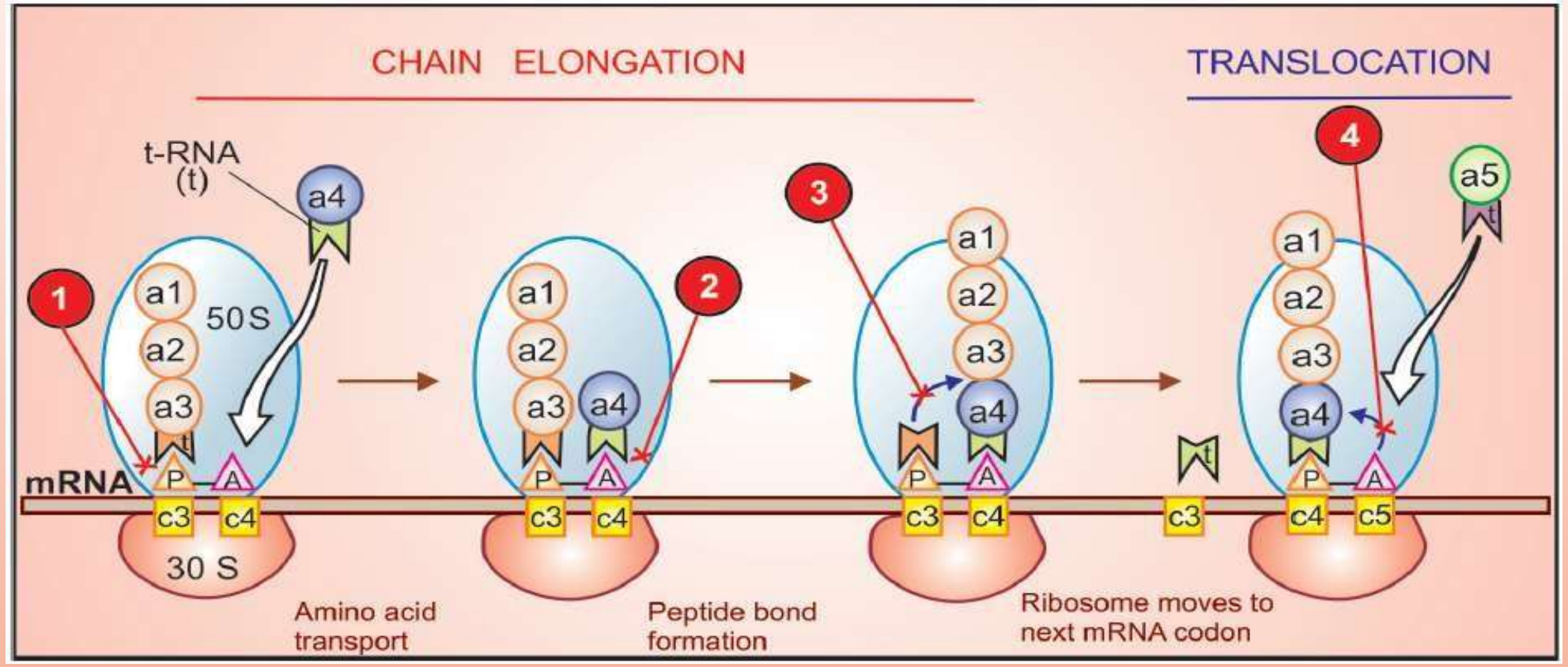
SAR of 1, 3-propanediol

1.The primary alcoholic group on C-1 atom if modified, results in a decrease in activity hence the alcoholic group seems to be essential for activity.

2.Of the four stereoisomers of chloramphenicol the antibacterial activity resides in only D-threo compound. Other isomers are inactive compounds.

Mechanism of action

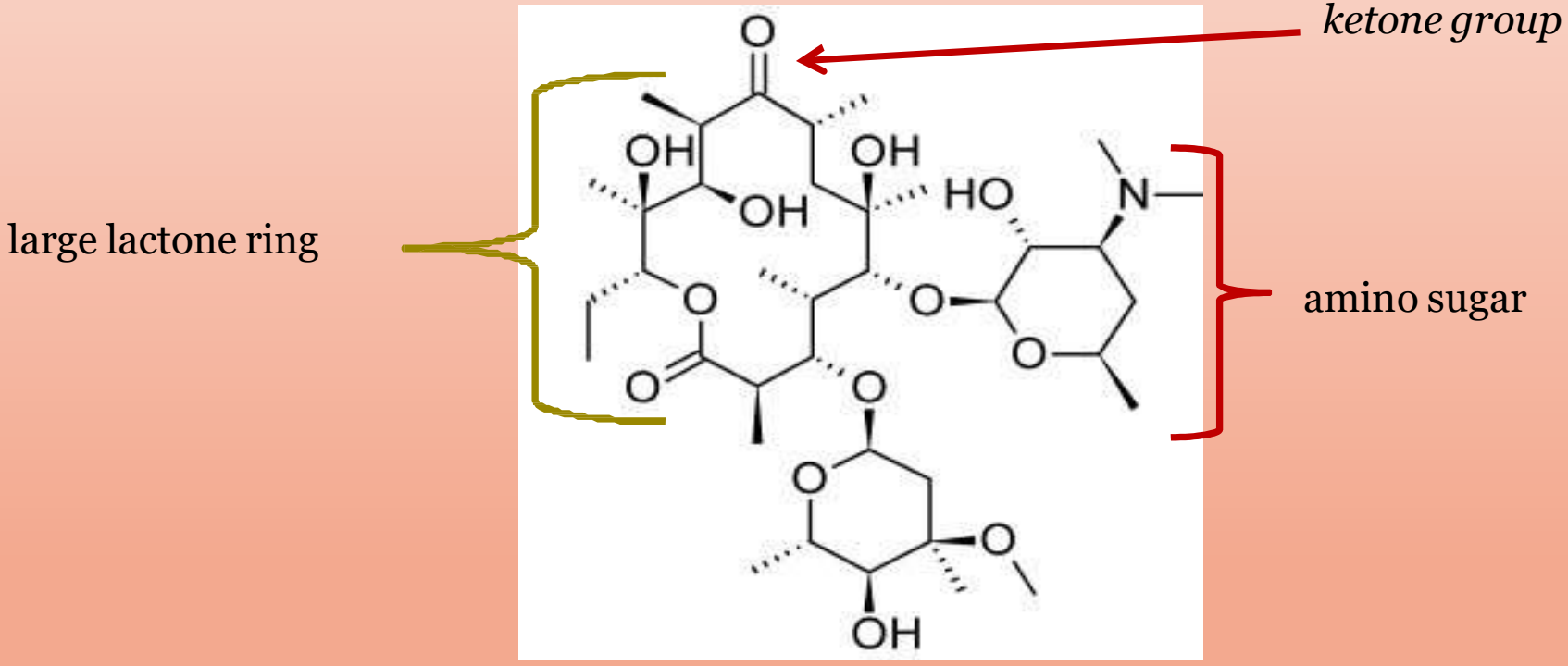
- The messenger RNA (mRNA) attaches to the 30S ribosome. The initiation complex of mRNA starts protein synthesis and polysome formation.
- The nacent peptide chain is attached to the peptidyl (P) site of the 50S ribosome. The next amino acid (a) is transported to the acceptor (A) site of the ribosome by its specific tRNA which is complementary to the base sequence of the next mRNA codon (C).
- The nascent peptide chain is transferred to the newly attached amino acid by peptide bond formation. The elongated peptide chain is shifted back from the 'A' to the 'P' site and the ribosome moves along the mRNA to expose the next codon for amino acid attachment.
- Finally the process is terminated by the termination complex and the protein is released.
- **Chloramphenicol binds to 50S subunit—interferes with peptide bond formation and transfer of peptide chain from 'P' site.**


CHAIN ELONGATION
TRANSLOCATION
t-RNA
(t)
a4
a5
1
2
3
4
a1
a2
a3
a4
50S
mRNA
P
A
t
c3
c4
c5
30 S
Amino acid
transport
Peptide bond
formation
Ribosome moves to
next mRNA codon

MACROLIDES

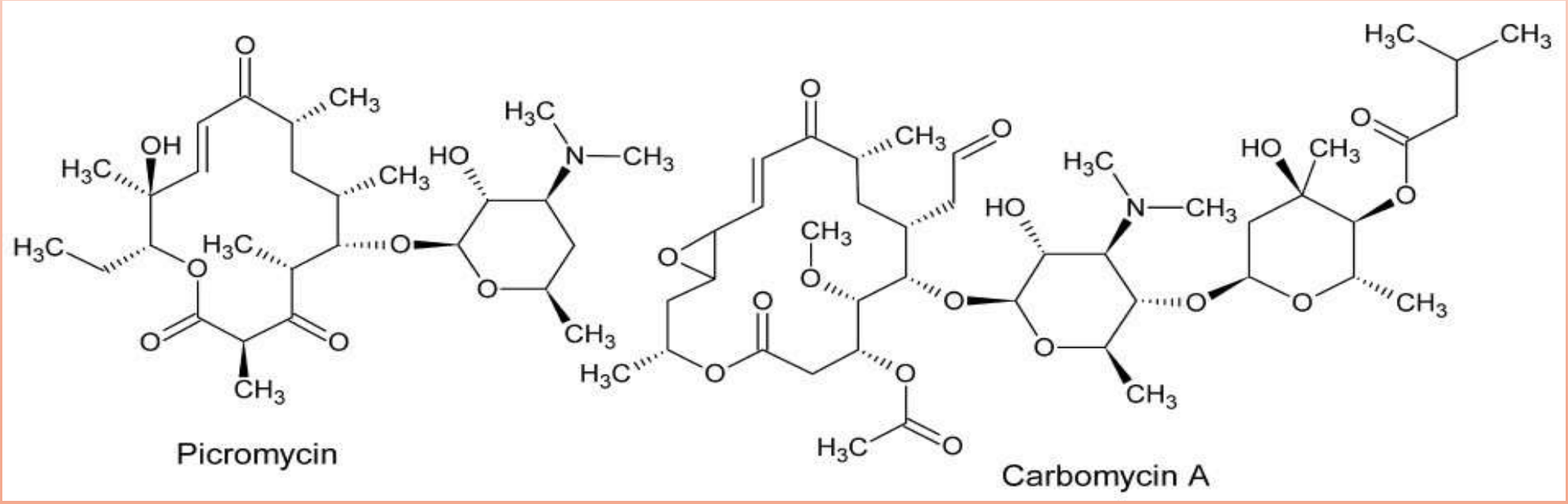
- Among the many antibiotics isolated from the actinomycetes is the group of chemically related compounds called the *macrolides*.
- In1952, erythromycin and carbomycin were reported as new antibiotics
- Currently, more than 40 such compounds are known
- Semisynthetic derivatives of erythromycin are clarithromycin and azithromycin etc.

- **Chemistry Macrolide antibiotics**
- The macrolide antibiotics have three common chemical characteristics
  (a) a large lactone ring (which prompted the name *macrolide)*
  (b) a ketone group, and
  (c) a glycosidically linked amino sugar.

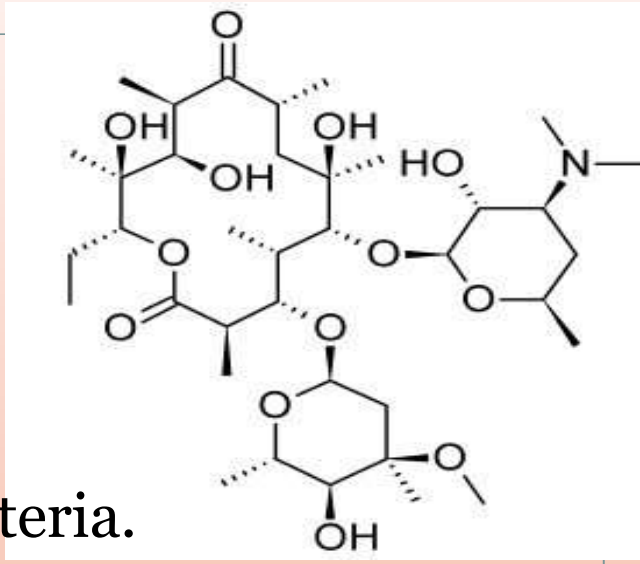


Erythromycin

- Usually, the lactone ring has 12, 14, or 16 atoms in it
- Olefinic group may be conjugated with the ketone function
- Because of the dimethylamino group on the sugar moiety, the macrolides are bases that form salts with pKa values between 6.0 and 9.0
- free bases are only slightly soluble in water

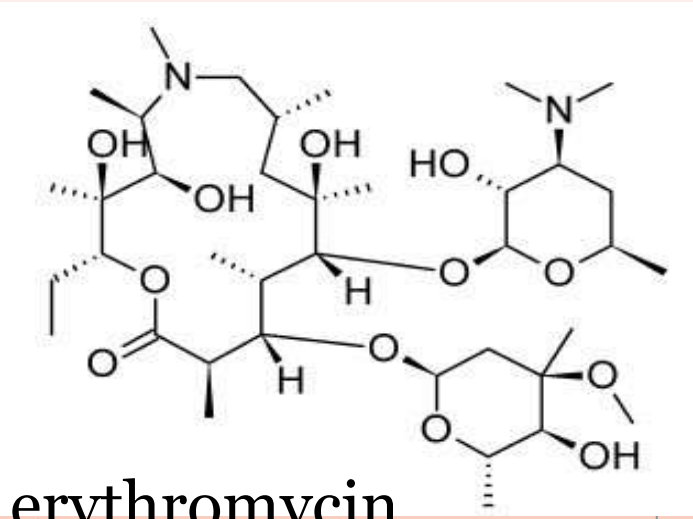


Erythromycin



- Isolated from *Streptomyces erythraeus*.
- treatment of various upper respiratory and soft-tissue infections caused by Gram-positive bacteria.
- It is also effective against many venereal diseases, including gonorrhea and syphilis
- **The amino sugar attached through a glycosidic link to C-5 is desosamine (3-dimethylamino-D-*xylo-hexose) gives a basic* character to erythromycin**
- **The other carbohydrate structure linked as a glycoside to C-3 is called *cladinose* (3-methoxy-3-C-methyl-L-*ribo-hexose) and is* unique to the erythromycin molecule.**
- Two such analogs have been found, erythromycins B and C

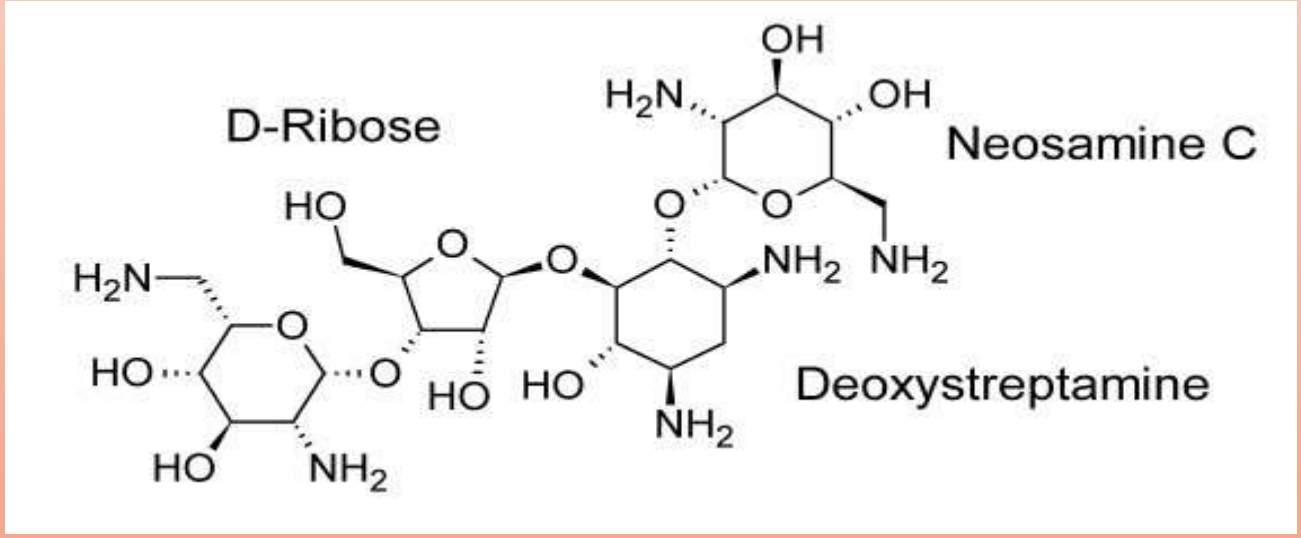
Azithromycin



- Azithromycin is a semisynthetic derivative of erythromycin.
- **Prepared by Beckman rearrangement , followed by *N-methylation and reduction* of the resulting ring-expanded lactam.**
- **It is a prototype of a series of nitrogen-containing, 15-membered ring macrolides known as *azalides***
- **Replacement of 9-keto group by weakly basic tertiary amine increases the stability of azithromycin to acid-catalyzed degradation.**
- These changes also increase the lipid solubility of the molecule.

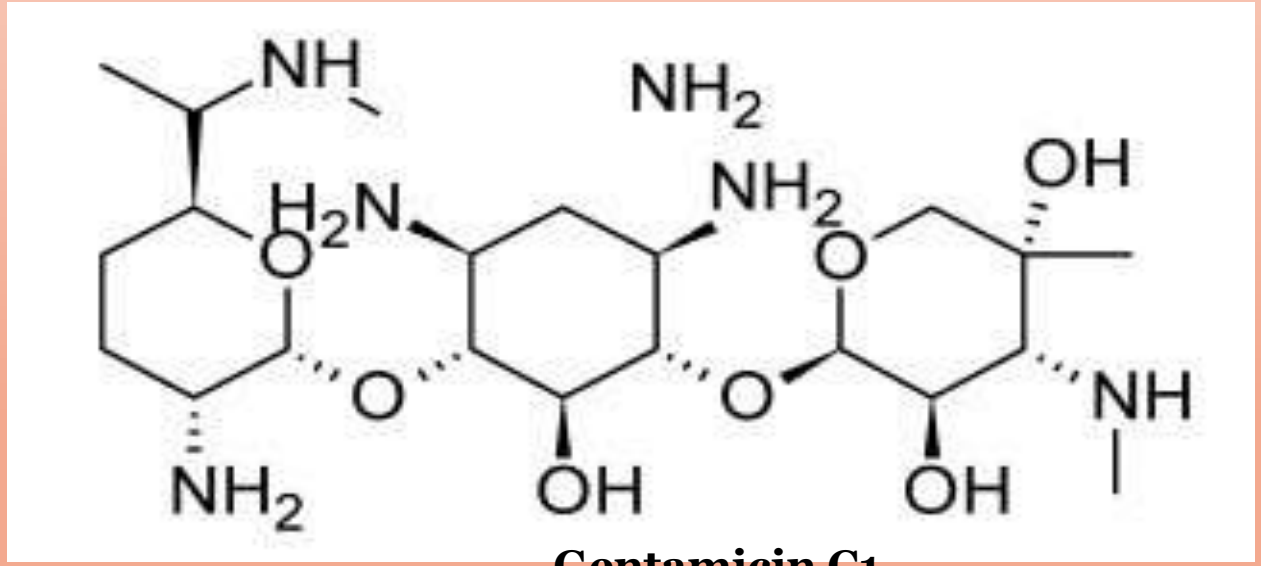
Neomycins

- Neomycin, as produced by *S. fradiae, is a mixture of* closely related substances.
- Included in the "neomycin complex" is neamine (originally designated *neomycin A) and* neomycins B and C.

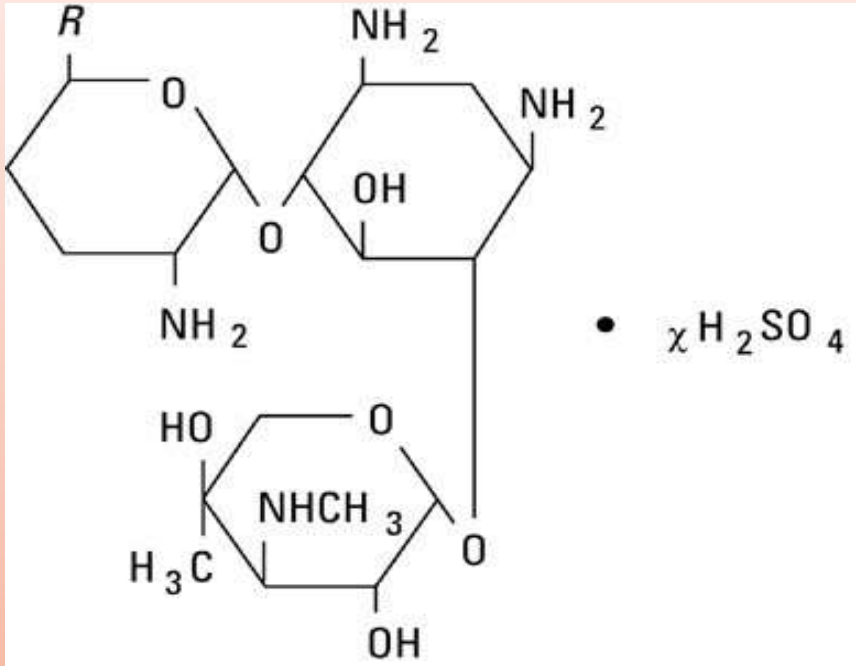


Gentamicin

- It is obtained commercially from *Micromonospora purpurea*.
- it has a broad spectrum of activity against many common pathogens, both Gram-positive and Gram negative.
- Gentamicin sulfate is a mixture of the salts of compounds identified as gentamicins C1, C2, and C_{1a}.



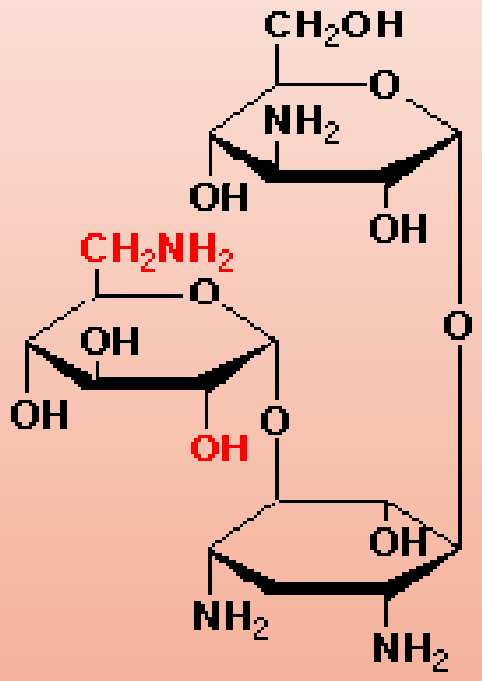
Gentamicin C1



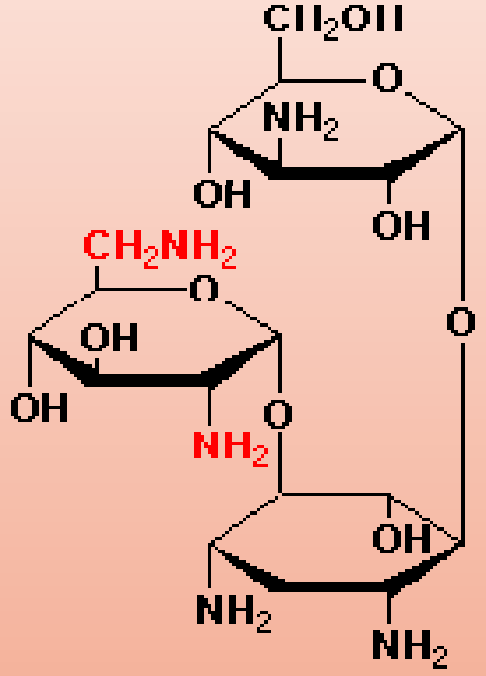
• χH_2SO_4

Gentamicin	R
C_1	$H_3C-HN-\underset{\displaystyle CH_3}{\overset{\displaystyle CH_3}{C}}-H$
C_2	$H_2N-\underset{\displaystyle CH_3}{\overset{\displaystyle CH_3}{C}}-H$
C_{1A}	CH_2NH_2

Kanamycin A



Kanamycin B



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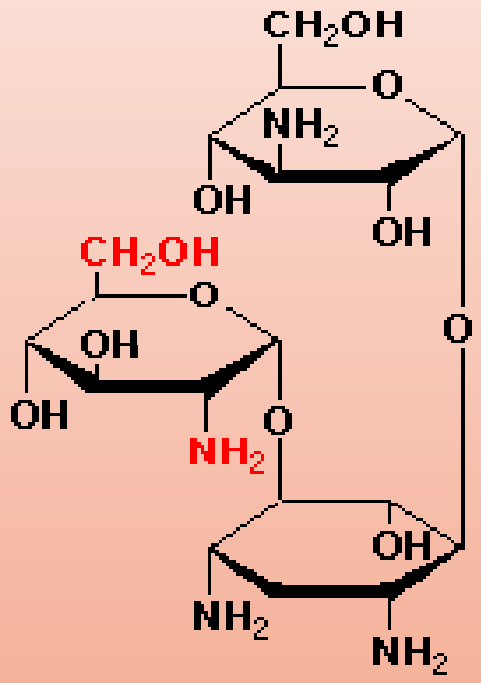
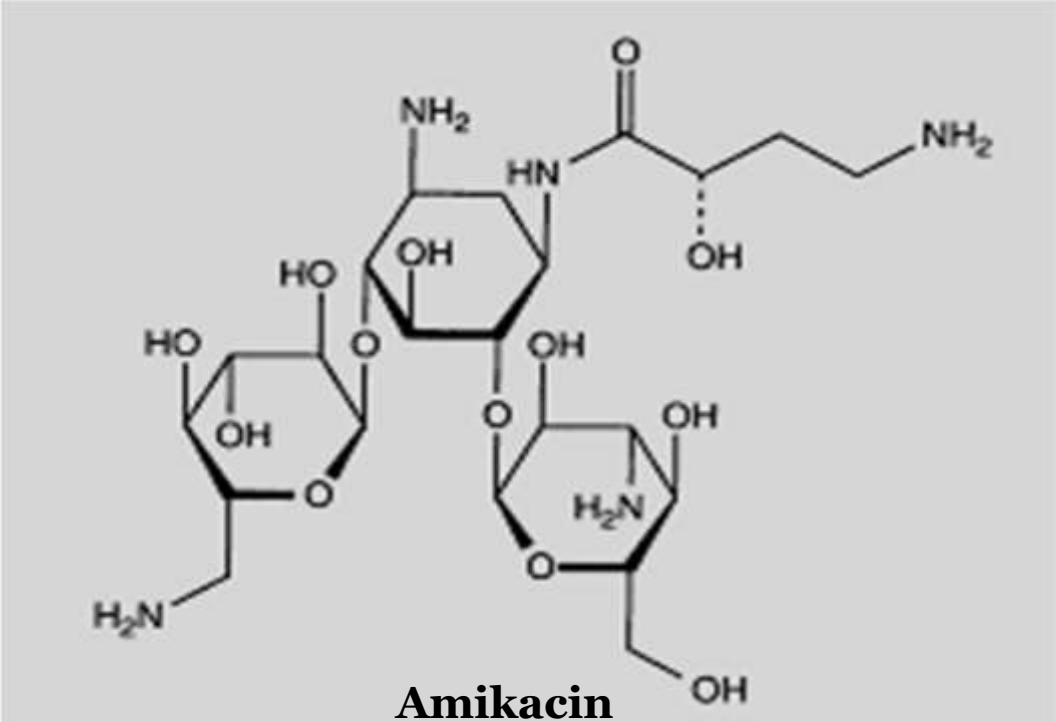
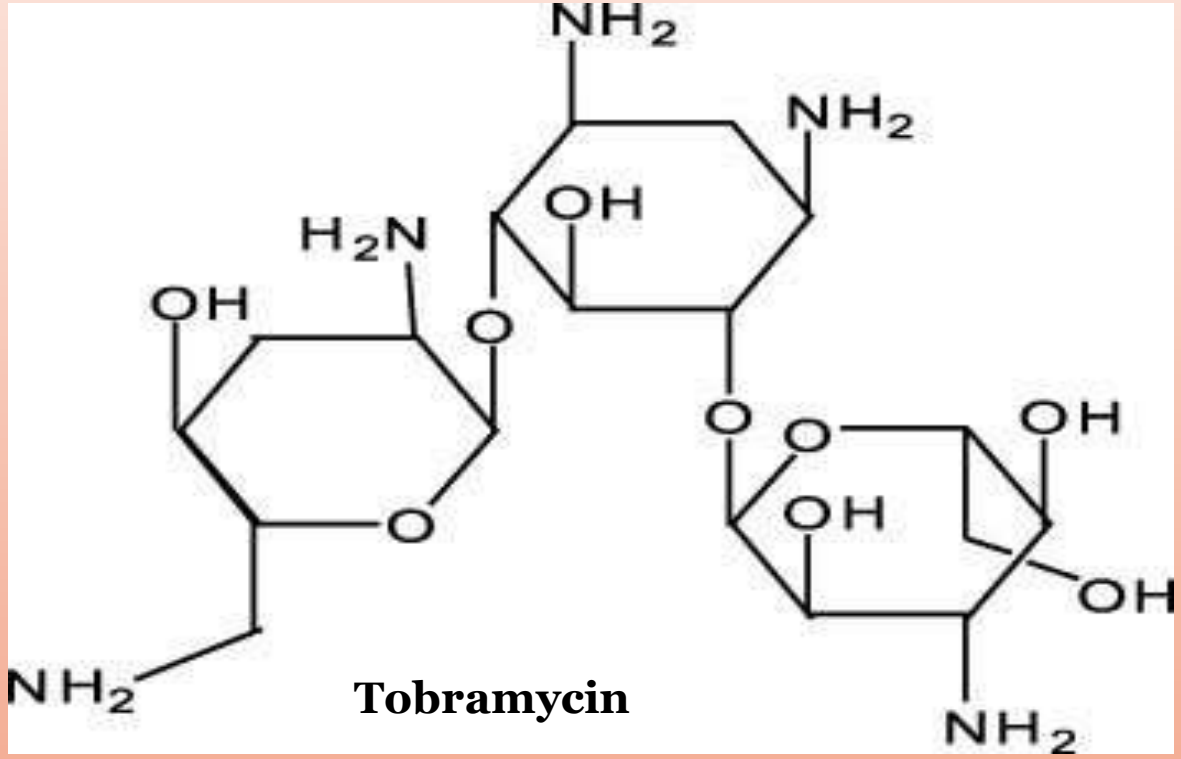
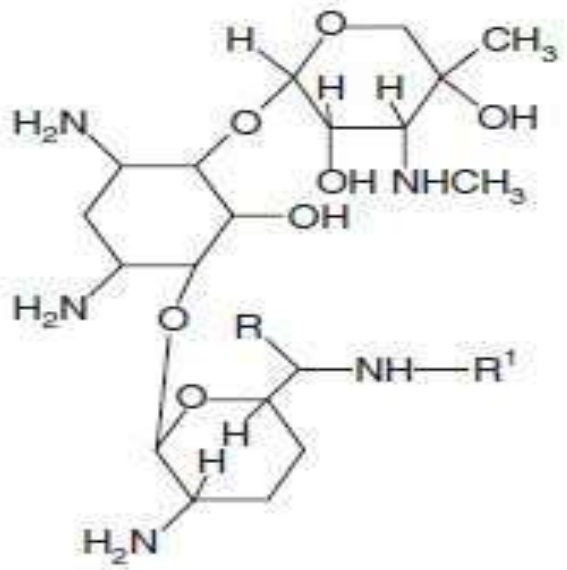




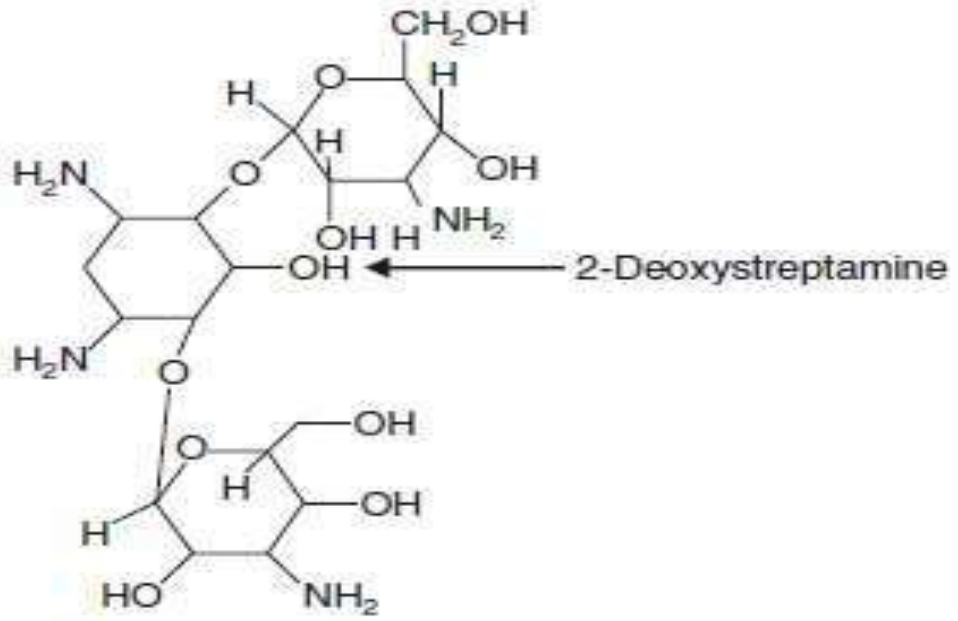
Amikacin



Tobramycin



Gentamicins



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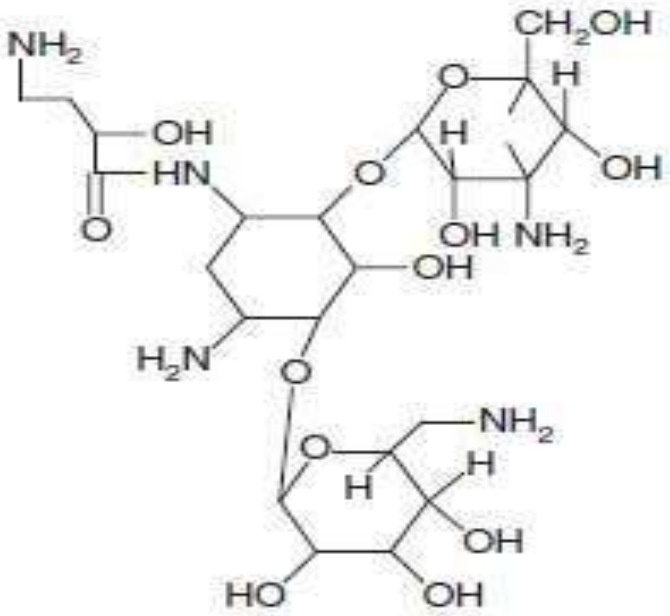

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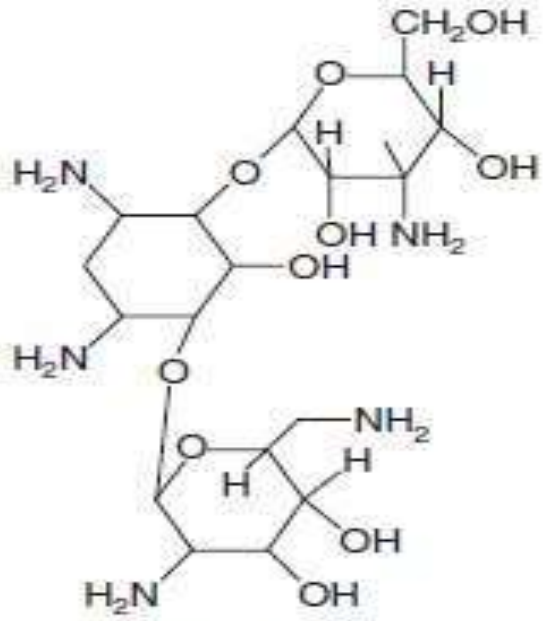

Tobramycins