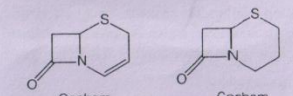
Nomenclatures

Cephalosporins are named in the following ways:

1. *Chemical abstracts*: 5-Thia-1-azobicyclo (4.2.0) octanes. Accordingly, cephalothin is 3-(Acetoxy methyl)-8-oxo-7-(2-thienyl) acetamido-5thia-1-aza-bicyclo[4.2.0]-oct-2ene-2-carboxylic acid.
2. *Cepham derivatives*: Cepham is the name given to the unsubstituted bicyclic lactam.

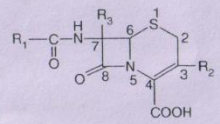


Classification

Cephalosporins are classified on the basis of their chemical structure, clinical pharmacology, antibacterial spectrum, or penicillinase resistance.

a. Orally administered: cephalexin, cephradine, and cefaclor
b. Parentrally administered: cephalothin, cephapirin, cephacetrile, and cefazedone. These agents aresensitivity to β-lactamase
c. Resistant to β-lactamase and parentrally administered: cefuroxime, cefamandole, cefoxitin
d. Metabolically unstable: cephalothin and cephairin

Clinically used cephalosporins



I. First-generation cephalosporins

These drugs have the highest activity against gram-positive bacteria and the lowest activity against gram-negative bacteria (Table 15.1)

II. Second-generation cephalosporins

These drugs are more active against gram-negative bacteria and less active against gram-positive bacteria than first-generation members (Table 15.2).

III. Third-generation cephalosporins

These drugs are less active than first-generation drugs against gram-positive organisms, but have a much-expanded spectrum of activity against gram-negative organisms (Table 15.3).

Table 15.1 First-generation cephalosporins.

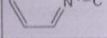
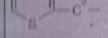
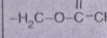
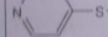
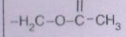
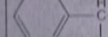
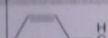
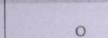
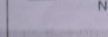
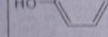
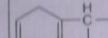
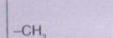
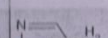
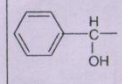
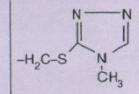
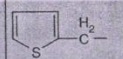
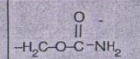
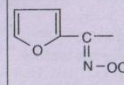
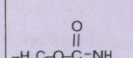
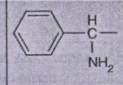
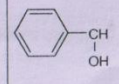
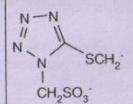
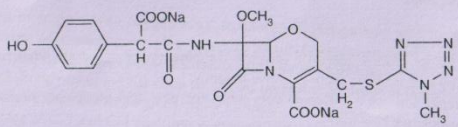
Name	R_1	R_2	R_3
Cephaloridine			–H
Cephalothin			–H
Cephapirin			–H
Cephalexin		$-CH_3$	–H
Cephaloglycine			–H
Cefadroxil		$-CH_3$	–H
Cephradine		$-CH_3$	–H
Cefazolin			–H
Cephradine		$-CH_3$	–H

Table 15.2 Second-generation cephalosporins.

Name	R_1	R_2	R_3
Cefamandole			–H
Cefoxitin			$-OCH_3$
Cefuroxime			–H
Cefaclor		–Cl	–H
Cefonicid			–H

Moxalactam



IV. Fourth-generation cephalosporins

Cefepime and cefpirome are new fourth-generation parenteral cephalosporins with a spectrum of activity which makes them suitable for the treatment of infections caused by a wide variety of bacteria (Table 15.4).

Table 15.3 Third-generation cephalosporins.

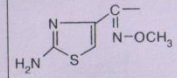
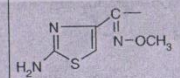
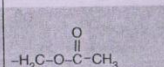
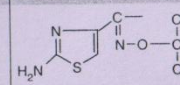
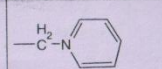
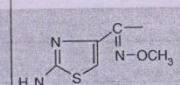
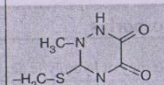
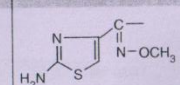
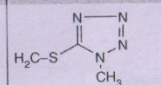
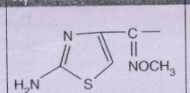
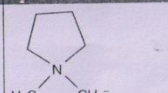
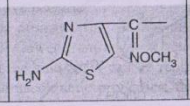
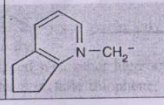
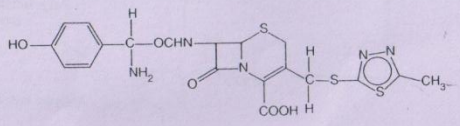
Name	R_1	R_2	R_3
Ceftizoxime		–H	–H
Cefotaxime			–H
Ceftazidime			–H
Ceftriaxone			–H
Cefmenoxime			–H

Table 15.4 Fourth-generation cephalosporins.

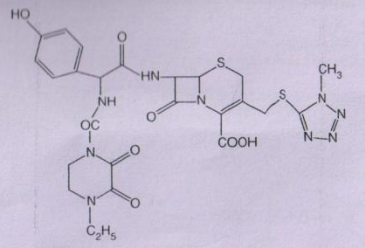
Name	R_1	R_2	R_3
Cefepime			–H
Cefpirome			–H

V. Miscellaneous

i. Cefaparole



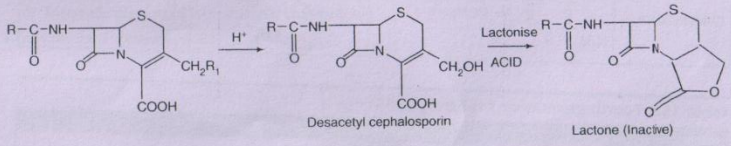
ii. Cefoperazone



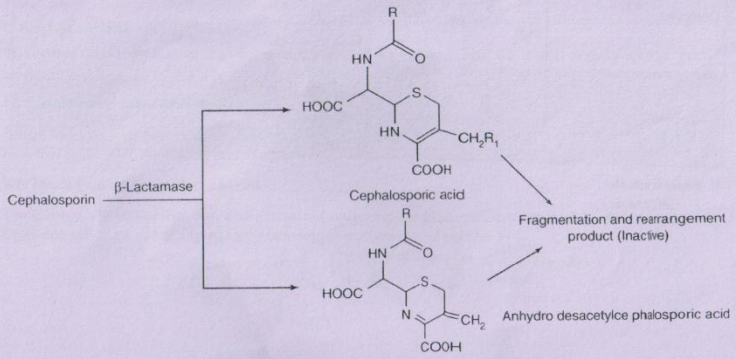
Degradation of Cephalosporins

Cephalosporins experience a variety of hydrolytic degradation reactions.

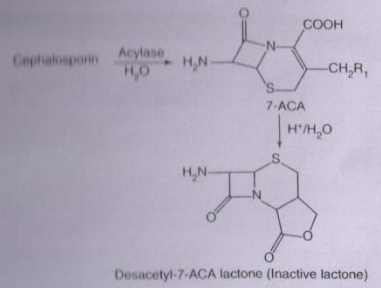
In strong acid solutions



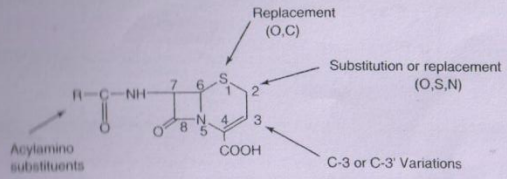
In the presence of β-lactamase



In the presence of acylase



SAR of Cephalosporins



1. 7-Acylamino substitution

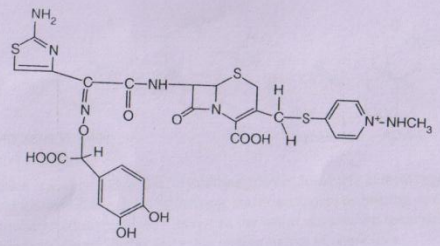
a. Acylation of amino groups generally increase the potency against gram-positive bacteria, but it is accompanied by a decrease in gram-negative potency.

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

c. Substitutions on the aromatic ring phenyl that increase 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; these include thiophene, tetrazole, furan, pyridine, and aminothiazoles.

e. The presence of catechol grouping can also enhance activity, particularly against *P. aeruginosa*, which not only exhibits antipseudomonas activity, but also retain some gram-positive activity, which is unused for a catechol cephalosporin.

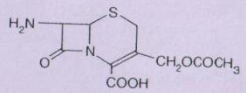


These compounds penetrate into the cell by utilizing the bacterial ion β-dependent ion transport system. There is a reduction of G^{-ve} activity when the lipophilicity of this side chain is increased and effects of polar α-substituents is enhanced (OH, NH_2, SO_3H, COOH).

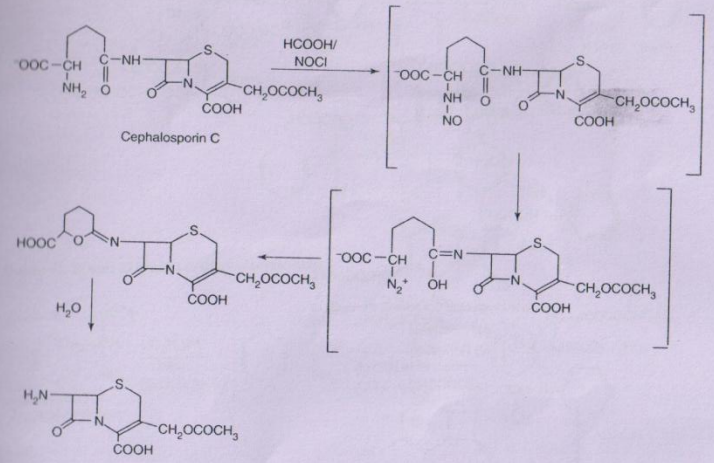
2. **Modification involving the C-3 substitution:** The nature of C-3 substituents influences pharmacokinetic and pharmacological properties as well as antibacterial activity. Modification at C-3 positionhas been made to reduce the degradation (lactone of desacetyl cephalosporin) of cephalosporins.
   a. The benzoyl ester displayers improved gram-positive activity, but lowered gram-negative activity.
   b. Pyridine, imidaozle replaced acetoxy group by azide ion yields derivative with relatively low gram-negative activity.
   c. Displacement with aromatic thiols of 3-acetoxy group results in an enhancement of activityagainst gram-negative bacteria with improved pharmacokinetic properties.
   d. Replacement of acetoxy group at C-3 position with-CH_3, Cl has resulted in orally activecompounds.
3. **Other modifications**
   a Introduction of C-7, a methoxy group, shows higher resistance to hydrolysis by β-lactamases.
   b. Oxidation of ring spectrum to sulphoxide or sulphone greatly diminishes or destroys the antibacterial activity.
   c. Replacement of sulphur with oxygen leads to oxacepam (latamoxet) with increased antibacterial activity, because of its enhanced acylating power. Similarly, replacement of sulphur with methylene group (loracavet) has greater chemical stability and a longer half-life.
   d. The carboxyl group position-4 has been converted into ester prodrugs to increase bioavailability of cephalosporins, and these can be given orally as well.
   e. Olefinic linkage at C-3 and C-4 positions is essential for antibacterial activity. Isomerization of the double bond to 2nd and 3rd positions leads to great losses in antibacterial activity.

Synthesis and Drug Profile

7-Aminocephalosporinic acid (7ACA)

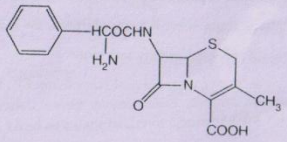


Synthesis

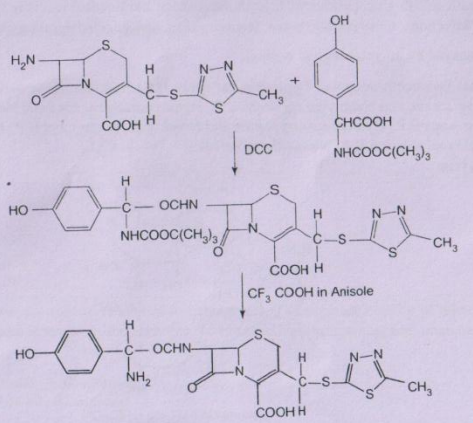


I. First-generation cephalosporins

i. Cephalexin (Keflex, Keforal)



Synthesis



Adverse reactions of the cephalosporins

The cephalosporins produce a number of adverse effects. Examples are the following:

1. *Allergic manifestation*: The cephalosporins should be avoided or used with caution in individuals allergic to penicillins. When cefamandole or cefoperazone is ingested with alcohol, a disulphiram-like effect is seen, because these cephalosporins block the second step in alcohol oxidation, which results in the accumulation of acetaldehyde.
2. *Bleeding*: Bleeding can occur with cefemandole or ceforperazone because of antivitamin K effects. But the administration of the vitamin overcomes this problem.

iii. Aminoglycoside antibiotics

The aminoglycoside antibiotics contain one or more amino sugars linked to an aminocytitol ring by glycosidic bonds. These are broad-spectrum antibiotics; in general, they have greater activity against gram-negative than gram-positive bacteria. The development of streptomycin, the first antibiotic of this group, was a well-planned work of Waksman (1944) and his associates, who isolated it from a strain of *Streptomyces griseus*.

The aminoglycoside can produces severe adverse effects, which include nephrotoxity, ototoxicity, and neuro effects. These properties have limited the use of aminoglycoside chemotherapy to serious systemic indications. Some aminoglycosides can be administered for ophthalmic and topical purposes.

Production of streptomycin

Initially, streptomycin was produced by surface cultures but nowadays streptomycin is produced by submerged cultures. The yield of streptomycin depends on the medium used. Again as in penicillin the culture medium must contain protein materials such as soya bean meal and cottonseed meal, in addition to other constituents. The medium used is composed of 1% glucose, 1% peptone, 0.3% meat extract or 1–2% corn steep liquor and 0–5% sodium chloride. The culture solution is kept in large vats, growth of the microorganism begins at 24–28°C and the maximum yield is achieved after 3–5 days.

Isolation of streptomycin

After separating the mycelium and other waste materials, the antibiotic from the filtrate is removed either by adsorption on charcoal or on base-exchange resins. Using dilute aqueous or alcoholic mineral acids, it is eluted from the adsorbent and the acidic eluate is purified by passing it through anion exchange resin. The pure streptomycin is isolated either as sulphate or as the crystalline trihydrochloride with calcium chloride. Aseptic handling must be done during the production and isolation of the drug.

In order to get completely sterile drug, the crystalline compounds (obtained above) are redissolved to give 25% solution, which is free from undesired impurities (heavy metals, colour and other impurities), by passing through a Seitz filter, and then freeze dried. The freeze dried powder is transferred aseptically to small vials as in case of penicillin.

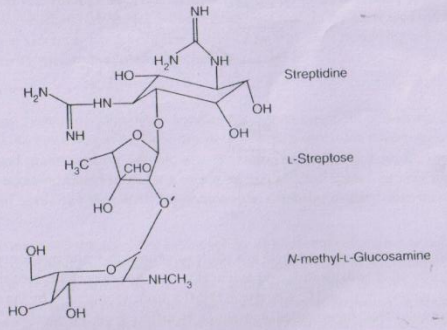
Streptomycin is a colourless, water-soluble, levorotatory base and has not yet been obtained as a crystalline compound. Its aqueous solutions are stable in the pH range 1–10 and streptomycin in fact is more stable than penicillin.

Mode of action: The aminoglycosides exhibit bactericidal effects as a result of several phenomena. Ribosomal binding on 30s and 50s subunits as well as the interface produces misreading; this disturbs the normal protein synthesis. Cell membrane damage also plays an integral part in ensuring bacterial cell death. Some examples of aminoglycoside antibiotics are listed in Table 15.5.

Table 15.5 Examples of aminoglycoside antibiotics.

Name	Source
Streptomycin	Streptomyces griseus
Neomycin	S. fradiae
Kanamycin	S. kanamyeleticus
Gentamysin	Micromonospora purpura
Netilmicin	Micromonospora species
Tobramycin (Nebramycin)	S. tenebrarius
Framycetin (Soframycin)	S. decaris
Paromomycin	S. rimosus and S. paramomycinus
Amikacin	It is 1-L-(-) 4-amino-2-hydroxy butyryl kanamycin

iv. Streptomycin and dihydrostreptomycin

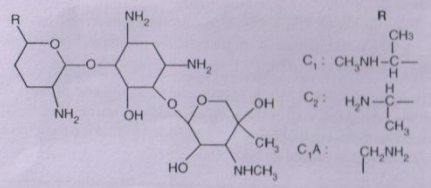


Properties and uses: Streptomycin sulphate is a white hygroscopic powder, very soluble in water, and practically insoluble in ethanol. The organism, *S. griseus*, releases the other substances, such as hydroxystreptomycin, mannisidostreptomycin, and cycloheximide, but do not reach up to the required activity/potency level. The development of resistant strains of bacteria and chronic toxicity constitutes major drawbacks of this category. It is an aminoglycoside antibacterial also used as antitubercular drug.

Assay: It is assayed by microbiological method.

Dosage forms: Streptomycin injection B.P.

v. Gentamycins

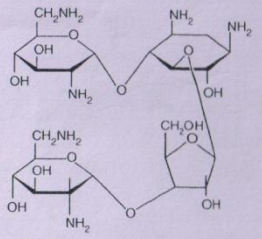


Properties and uses: Gentamycin is a mixture of C_1, C_2, and C_1A compounds, obtained commercially from *Micromonospora purpurea*. Gentamycin sulphate exists as white hygroscopic powder, soluble in water, and practically insoluble in alcohol, although it is a broad-spectrum antibiotic. It is used in the treatment of infections caused by gram-negative bacteria of particular interest and has a high degree of activity against *P. aeruginosa* , where the important causative factor is burned skin. It is used topically in the treatment of infected bed-sores, pyodermata, burns, and in the infection of the external eye.

Assay: It is assayed by microbiological method.

Dosage forms: Gentamicin cream B.P., Gentamicin ear drops B.P., Gentamicin and Hydrocortisone acetate ear drops B.P., Gentamicin eye drops B.P., Gentamicin injection B.P., Gentamicin ointment B.P.

vi. Neomycin



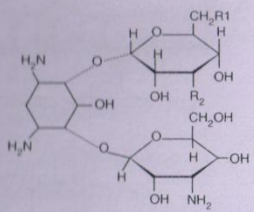
Properties and uses: Neomycin sulphate is a white or yellowish-white hygroscopic powder, very soluble in water, very slightly soluble in alcohol, and practically insoluble in acetone. Neomycin is a mixture of closely related epimers, neomycin B, and C. Neomycin B differ from neomycin C by the nature of the sugar attached terminally to ribose. This sugar called neosamine B differs from neosamine C in its stereochemistry. In

neomycin B, the neobiosamine moiety contains β-L-iodopyranosyl, whereas in neomycin C the configuration is inverted and it is 2-D-glucopyranosyl. It is photosensitive and its main use is in the treatment of the ear, eye, and skin infections; these include burns, wounds, ulcer, and infected dermatoses.

Assay: It is assayed by microbiological method.

Dosage forms: Dexamethasone and Neomycin ear spray B.P., Hydrocortisone and neomycin cream B.P., Hydrocortisone acetate and Neomycin ear drops B.P., Hydrocortisone acetate and Neomycin eye drops B.P., Neomycin eye drops B.P., Hydrocortisone acetate and Neomycin eye ointment B.P., Neomycin eye ointment B.P., Neomycin oral solution B.P., Neomycin tablets B.P.

vii. Kanamycin

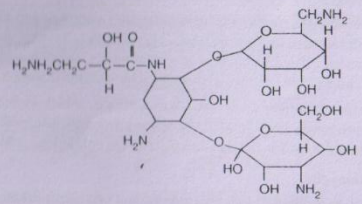


Kanamycin A=R_1=NH_2 ; R_2=OH
Kanamycin B=R_1=R_2=NH_2
Kanamycin C=R_1=OH; R_2=NH_2

Properties and uses: Kanamycin sulphate is a white crystalline powder, soluble in water, practically insoluble in acetone and in alcohol. The mixture consists of three related structures, that is, Kanamycin A, B, and C. The kanamycins do not possess D-ribose molecule that is present in neomycins and paramomycins. The use of kanamycins is restricted to infections of the intestinal tract and to systemic infections.

Assay: It is assayed by microbiological method.

viii. Amikacin

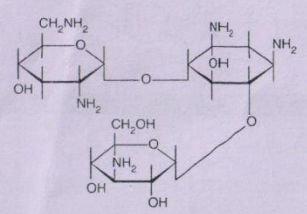


Properties and uses: Amikacin is a semisynthetic drug derived form kanamycin A. It retains 50% of the original activity of kanamycin A. L-Isomer is more active than D-isomer. It resists attack by most bacterial-

inactivating enzyme. Therefore, it is very effective and less ototoxic than other aminoglycosides.

Dosage forms: Amikacin sulphate injection I.P.

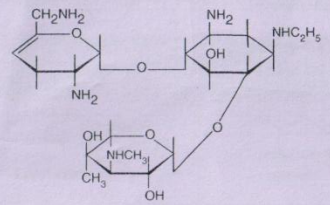
ix. Tobramycin



Properties and uses: Its activity is similar to gentamycin. The superior activity of tobramycin against *P. aeruginosa* may make it useful in the treatment of bacterial oesteromyelitis, and pneumonia caused by *P. species*.

Dosage forms: Tobramycin injection I.P.

x. Netilmicin (1-*N*-ethylsisomicin)



Properties and uses: Netilimycin sulphate is a white or yellowish-white hygroscopic powder, very soluble in water, practically insoluble in acetone and alcohol. It is similar to gentamycin and tobramycin. The majority of the aminoglycoside inactivating enzymes do not metabolize it. It is useful for the treatment of serious infections due to susceptible enterobacteria and other aerobic gram-negative bacilli. **Assay:** It is assayed by microbiological method.

SAR of Aminoglycoside Antibiotics

The aminoglycosides consist of two or more amino sugars joined in glycoside linkage to a highly substituted 1,3-diaminocyclo hexane (aminocyclitol), which is a centrally placed ring. The ring is a 2-deoxy streptamine in all aminoglycosides except streptomycin and dihydrostreptomycin, where it is streptidine.

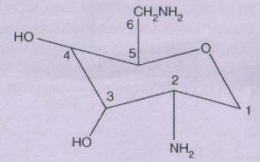
Thus,

- In kanamycin and gentamycin families, two amino sugars are attached to 2-deoxy streptamine.
- In streptomycin, two amino sugars are attached to strepidine.

- In neomycin family, there are amino sugars attached to 2-deoxy streptamine.

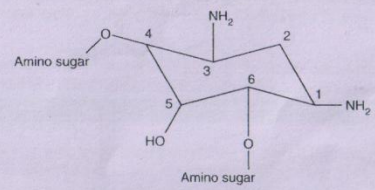
The aminoglycoside antibiotics contain two important structural features. They are amino sugar portion and centrally placed hexose ring, which is either 2-deoxystreptamine or streptidine.

1. Amino sugar portion



i. The amino function at C-6 and C-2 positions serve as major target sites for bacterial inactivating enzymes.
ii. Methylation at C-6 position does not decrease the activity; instead, increases enzyme resistance.
iii. Cleavage of 3-hydroxyl or the 4-hydroxyl or both groups does not affect the activity.

2. Centrally placed hexose ring (aminocyclitol ring)



i. Various modifications at C-1 amino group have been tested. The acylation (e.g. amikacyn) and ethylation (e.g. 1-*N*-ethylsisomycin) though does not increase the activity helps to retain the antibacterial potency.
ii. In sisomicin series, 2-hydroxylation and 5-deoxygenation result in the increased inhibition of bacterial inactivating enzyme systems. Thus, very few modifications of the central ring are possible, which do not violate the activity spectrum of aminoglycosides.

3. Tetracycline antibiotics

Tetracyclines have a ring system of four linear annelated six-membered rings and are characterized by a common octahydronaphthacenes skeleton. They are potent, broad-spectrum antibacterial agents effective against gram-positive and gram-negative aerobic and anaerobic bacteria. As a result, the tetracyclines are drugs of choice or well-accepted alternatives for a variety of infectious diseases. Among these, they also play a role in the treatment of sexually transmitted and gonococcal diseases, urinary tract infections, bronchitis, and sinusitis remain prominent.

The majority of the marketed tetracyclines (tetracycline, chlorotetracycline, oxytetracycline, and demeclocycline) are naturally occurring compounds obtained by the fermentation of *Streptomyces spp.* broths. The semisynthetic tetracyclines (methacycline, deoxycycline, minocycline) have the advantage of longer duration of antibacterial action. However, all these tetracyclines exhibit a similar profile in terms of antibacterial potency. In general, their activity encompasses many strains of gram-negative *E. coli, Proteus, Klebsiella,*

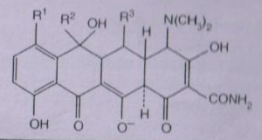
Enterobacter, Niesseria, and *Serratia* spp., as well as gram-negative *Streptococci* and *Staphylococci.* Of particular interest is the potency of tetracylines against *Haemophilus, Legionella, Chlamydia,* and *Mycoplasma.*

Production of tetracyclines: Chlortetracycline (aureomycin) is produced by a strain of *S. aureofaciens.* It is isolated by the following stages: (i) adsorption on activated charcoal or magnesium silicate; (ii) washing the chromatogram with an acidified organic solvent such as methanol or acetone; (iii) selection of the fraction which appears to be yellow under UV light; (iv) finally recovery of the antibiotic by extraction with butanol, precipitation with dry ether resolution in dilute hydrochloric acid and freezing to remove water. Tetracycline is produced by *S. aureofaciens* as well as other *Streptomyces* organisms; while oxytetracycline (terramycin) is produced by *S. rimosus.*

Classes of tetracyclines

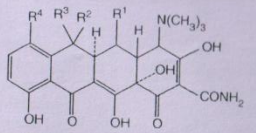
I. Natrual tetracyclines (biosynthetic)
II. Semisynthetic tetracyclines
III. Protetracyclines

I. Natural tetracyclines



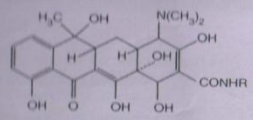
S. No.	Drug	R^1	R^2	R^3
1.	Tetracycline	–H	$-CH_3$	–H
2.	Chlortetracycline	–Cl	$-CH_3$	–H
3.	Oxytetracycline	–H	$-CH_3$	–OH
4.	Bromotetracycline	–Br	$-CH_3$	–H
5.	Dexamethyltetracycline	–H	–H	–H
6.	Dexamethylchlortetracycline	–Cl	–H	–H

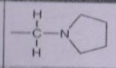
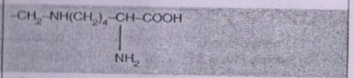
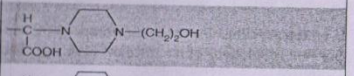
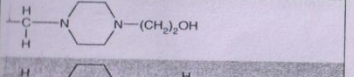
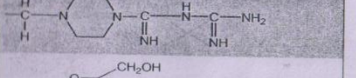
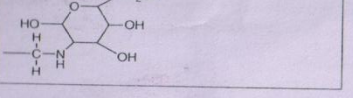
II. Semisynthetic tetracyclines



S. No.	Drug	R^1	R^2	R^3	R^4
1.	Doxycycline	–OH	–H	$-CH_3$	–H
2.	Minocycline	–H	–H	–H	$-N-(CH_3)_2$
3.	Methacycline	–OH	$=CH_2$	–	–H
4.	Meclocycline	–OH	$=CH_2$	–	–Cl
5.	Sancycline	–H	–H	–H	–H

III. Protetracyclines

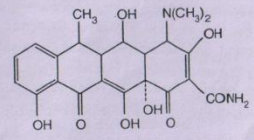


S. No.	Drug	R
1.	Rolitetracycline	
2.	Lymecycline	
3.	Clomocycline	–CH2–OH
4.	Apicycline	
5.	Pipacycline	
6.	Guamecycline	
7.	Meglucycline	

General mode of action of tetracyclines: In bacterial protein synthesis, the messenger RNA attaches itself to 30S ribosomes. The initiation complex of mRNA starts the protein synthesis and polysome formation of the nascent peptide that is attached to 50S ribosomes. Its specific tRNA transports the next amino acid to the acceptor site of the ribosome, which is complementary to the base sequence of the next mRNA codon. The nascent peptide is transferred to the newly attached amino acid by peptide bond formation. Tetracyclines bind to 30S ribosomes and the attachment of aminoacyl tRNA to mRNA ribosome complex is interfered.

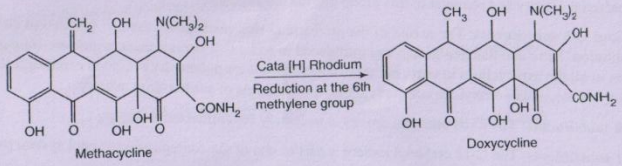
Physicochemical properties: The tertracyclines are yellow, bitter, odourless, and light sensitive crystalline compounds. These are amphoteric due to the acidic and the basic substituents, and have low solubility in water (0.5 mg/ml). They form water-soluble salts with strong acids and bases. In each tetracycline, there are

ii. Doxycycline (Vibramycin)



4-(Dimethylamino)-3,5,10,12,12a-pentahydroxy-6-methyl-1,11-dioxo-1,4,4a,5,5a,6,11,12a-octahydrotetracene-2-carboxamide

Synthesis



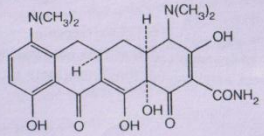
Methacycline → Doxycycline

Properties and uses: It was first obtained in small yields by a chemical transformation of oxytetracycline. The 6α-methyl epimer is more than three times as active as its β epimer.

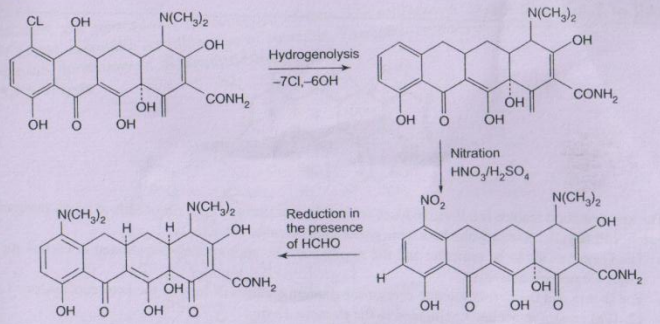
Dose: In adults, the oral dosage is 100 mg every 12 h.

Dosage forms: Doxycycline HCl capsules I.P., Doxycycline HCl tablets I.P.

iii. Minocycline (Cynomycin, Minolox)



Synthesis

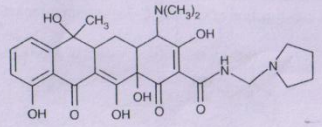


Minocycline

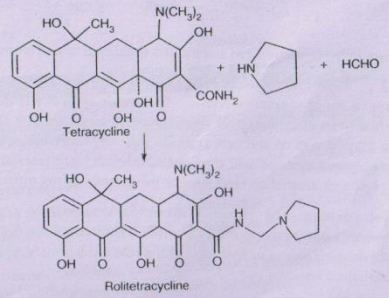
Properties and uses: It is a yellow crystalline powder with slightly bitter taste, soluble in water. It is very active against gram-positive bacteria. It is especially effective against *Mycobacterium marinum*. As a prophylactic against streptococcal infections, it is the drug of choice. It lacks the 6-hydroxyl group, therefore, it is stable to acids and does not dehydrate or rearrange to anhydro or lactone forms.

Dose: The dose orally for adults is 200 mg.

iv. Rolitetracycline

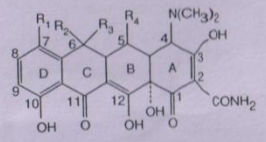


Synthesis



Tetracycline → Rolitetracycline

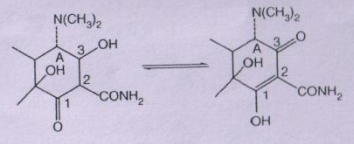
SAR of Tetracyclines



The key structural feature is a linearly fused tetracyclic nucleus and each ring needs to be six membered and purely carbocyclic. A tetracyclic backbone skeleton is essential for activity.

- The D-ring needs to be aromatic and the A-ring must be appropriately substituted at each of its carbon atoms for notable activity.
- The B-ring and the C-ring tolerate certain substitutent changes as long as the keto-enol systems (at C-11, 12, 12a) remain intact and conjugated to the phenolic D-ring.
- The D, C, B-ring phenol, keto-enol system is imperative and the A-ring must also contain a conjugated keto enol system.
- Specifically, the A-ring contains a tricarbonyl derived keto-enol array at positions C-1, 2, and -3. Other structural requirements for good antibacterial activity include a basic amine function at C-4 position of the A-ring.

Modification of C-1 and C-3 position: The keto-enol system between carbon atom 1 and 3 is highly delocalized and extremely slow to react. The keto-enol tautomerism of ring A is a feature common to all biologically active tetracyclines, blocking this system by forming derivatives at C-1 and C-3 results in loss of antibacterial activity. A–C = O function of C-1 and C-3 is essential for activity.



In addition, equilibrium between nonionized and Zwitter ionic structure of tetracycline is essential for activity.

Modification of C-2 position: The carboxamide moiety is present in all naturally occurring tetracyclines and this group is crucial for antibacterial activity. The amide is best left unsubstituted or monosubstitution is acceptable in the form of activated alkylaminomethyl amide (mannich bases). An example includes rolitetracycline large alkyl group on the carboxamide that may alter the normal keto-enol equilibrium of the C-1, 2, and 3 conjugated systems and diminishes inherent antibacterial activity. The replacement of carboxamide group or dehydration of carboxamide to the corresponding nitrile results in a loss of activity.

Modification of C-4 position: The naturally occurring tetracyclines contain α-C-4 dimethyl amino substituents that favourably contribute to the keto-enolic character of the A-ring. Replacement of dimethyl amino group with a hydrazone oxime or hydroxyl group leads to a pronounced loss of activity probably due to the increase in heteroatom basicity.

Modification of C-4a position: The α-hydrogen at C-4a position of tetracyclines is necessary for useful antibacterial activity.

Modification of the C-5 and C-5a positions: Many naturally occurring antibacterial tetracyclines have an unsubstituted methylene moiety at the C-5 position. However, oxytetracycline contains C-5 α–hydroxyl group, was found to be a potent compound, and has been modified chemically to some semisynthetic tetracyclines. Alkylation of the C-5 hydroxyl group results in a loss of activity. Ester formation is only acceptable if the free oxytetracycline can be liberated in vivo; only small alkyl esters are useful. The configuration of the naturally occurring tetracyclines places the C-5a hydrogen atom in a α-configuration. Epimerization is detrimental to antibacterial activity.

Modification at the C-6 position: The C-6 position is tolerant of a variety of substituents. The majority of tetracyclines has α-methyl group and α β-hydroxyl group at this position. Demeclocyclin is a naturally occurring C-6 demethylated chlortetracycline with an excellent activity. The C-6 methyl group contributes little to the activity of tetracycline. Similarly, the C-6 hydroxyl group also appears to offer little in terms of antibacterial activity and removal of this group affords doxycycline.

C-7 and C-9 substituents: The nature of the aromatic D-ring predisposes the C-7 position to electrophilic substitution. Nitro and halogen groups are introduced in some C-7 tetracyclines, which are among the most potent of all the tetracyclines in vitro, but these compounds were potentially toxic and carcinogenic. The C-7 acetoxy, azido, and hydroxyl tetracyclines are inferior in terms of antibacterial activity.

C-10 substituents: The C-10 phenolic moiety is necessary for antibacterial activity.

C-11 substituents: The C-11 carbonyl moiety is part of one of the conjugated keto-enol system required for antibacterial activity.

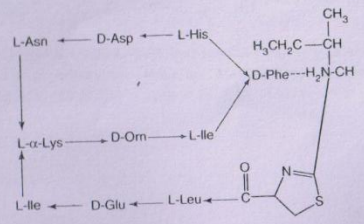
C-11a substituents: In general, modifications at the C-11a position of tetracycline are not tolerated.

C-12/12a substituents: As with the C-11 position, the C-12 is a part of the keto-enol system that is vital for the drug uptake binding and observed antibacterial activity. The C-12a hydroxyl group is needed for antibacterial activity although this moiety can be esterified to provide tetracyclines with increased lipophilicity. Antibacterial properties are retained if the alkyl ester is small in size and readily undergoes hydrolysis to liberate free tetracycline.

4. Polypeptide antibiotics

The compounds have complex polypeptide structure. These are resistant to animal and plant proteases. These contain lipid moieties besides amino acids that are not found in peptides of animal and plant origins. Examples: bacitracin, polymycin, amphomycin, tyrothricin, and vancomycin.

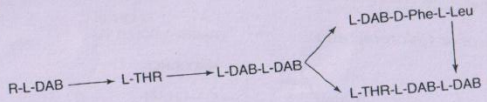
i. Bacitracin



Properties and uses: Bacitracin is a white hygroscopic powder, soluble in water and alcohol. Bacitracin antibiotic is isolated from the fermentation broth of a culture of tracyl-1 strain of *Bacillus subtilis*. It is found to be a complex mixture of at least 10 polypeptides (A, A_1, B, C, D, E, F_1, F_2, F_3, and G), of which bacitracin A fraction is believed to be the most abundant and the most potent. A divalent ion Zn^{++} enhances its activity. Although bacitracin is occasionally employed for topical application (often in combination with neomycin, polymycin, and tyrothicin) for the treatment of burns, ulcer, and wounds, it can cause serious necrosis of the kidney tubules; if it is given systematically (i.e. I .V route) an oral administration is not feasible due to its lack of absorption from the GI tract. A variety of gram-positive *cocci* and *bacilli* are sensitive to bacitracin. It should be stored in airtight containers due to its hygroscopic nature.

Assay: It is assayed by microbiological method.

ii. Polymyxin



Polymyxin B_1:R = (+)-6-methyloctanoyl

Polymyxin B_2:R = 6-methylpeptanoyl

DAB = α–γ–Diaminobutyric acid

Properties and uses: Polymycin sulphate is a white hygroscopic powder, soluble in water, and slightly soluble in ethanol. The polymyxins are cyclic peptides holding a fatty acid side chain. This is a group of relatively simple basic, cationic, detergent peptides that are produced by *Bacillus polymyxia*. At least, five polymyxins (A, B, C, D, and E) are known, but only polymyxin B and polymyxin E are of clinical utility. Both polymyxin B and polymyxin E (colistin) are mixtures of two components and is used in the treatment of bacterial meningitis, urinary tract infection, burns, wounds, and gastroenteritis. Polymyxin may affect renal tubules and central nervous system (CNS), and because of their nephrotoxicity associated with their systemic use, they are primarily employed to treat topical infections.

Assay: It is assayed by adopting liquid chromatography technique.

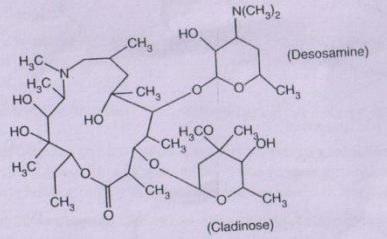
5. Macrolide antibiotics

The macrolide antibacterial agents are extremely useful chemotherapeutic agents for the treatment of a variety of infectious disorders and diseases caused by a host of gram-positive bacteria, both *cocci* and *bacilli*; they also exhibit useful effectiveness against gram-negative *cocci*, specially, *neisseria* spp. The mac-rolides are commonly administered for respiratory, skin, tissue, and genitourinary infections caused by these pathogens.

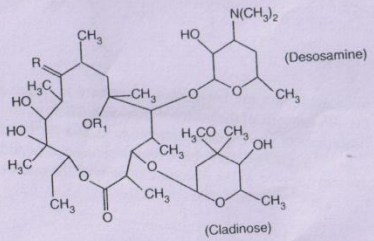
Chemistry: They are characterized by five common chemical features.

1. A macrocyclic lactone usually has 12–17 atoms, hence the name macrolide.
2. A ketone group.
3. One or two amino sugars glycosidically linked to the nucleus.
4. A neutral sugar linked either to amine sugar or to nucleus.
5. The presence of dimethyl amino moiety on the sugar residue, which explains the basicity of these compounds, and consequently the formation of salts. The antibacterial spectrum of activity of the more potent macrolides resembles that of penicillin. Examples: erythromycin, oleandomycin, clarithromycin, flurithromycin, dirithomycin, azithromycin.

i. Azithromycin



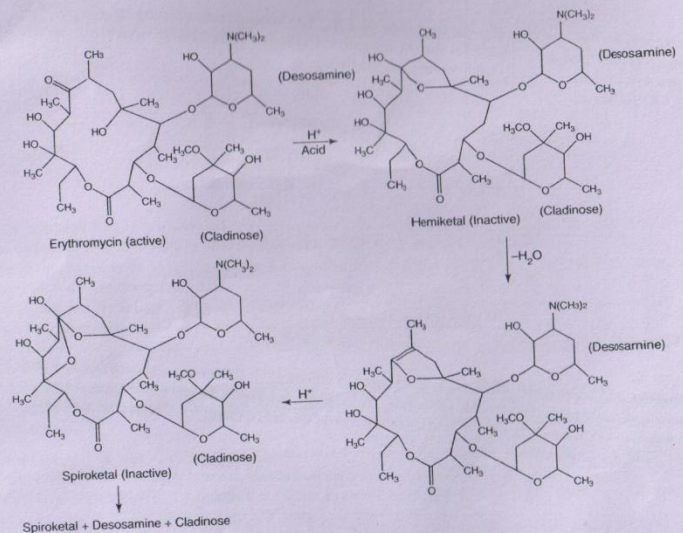
Properties and uses: Azithromycin is a white powder, practically insoluble in water, soluble in anhydrous ethanol and methylene chloride. It is very stable under acidic conditions, is less active against *Streptococci* and *Staphylococci* than erythromycin, and is far more active against respiratory infections due to *H. influenzae* and *Chlamydia trachomatis*.



Name	R	R_1
Erythromycin	=O	–H
Roxithromycin	$CH_3OCH_2CH_2OCH_2O$–=	–H
Clarithromycin	=O	$–CH_3$

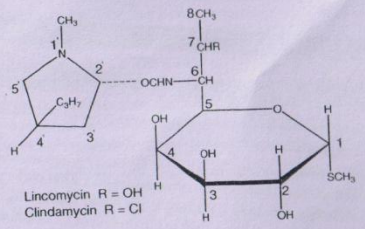
Acid degradation of erythromycin

Erythromycin is unstable in the acid media. The C-6 hydroxyl group reversibly attacks the C-9 ketone giving rise to a hemiketal intermediate. Dehydration prevents regeneration of the parent erythromycin and the C-12 hydroxyl group can subsequently add to produce a spiroketal species. The cladinose group is cleaved from the macrocycle and more harsh conditions lead to the release of desosamine. Useful antibacterial activity last till the dehydration of the hemiketal and the spiroketal is weakly active.



Mode of action: Macrolide antibiotics are bacteriostatic agents that inhibit protein synthesis by binding irreversibly to a site on the 50S subunits of the bacterial ribosome. Thus, inhibiting the translocation steps of protein synthesis at varying stages of peptide chain elongation (hinder the translocation of elongated peptide chain back from 'A' site to 'P' site). The macrolides inhibit ribosomal peptidyl transferase activity. Some macrolides also inhibit the translocation of the ribosome along with the mRNA template.

6. Lincomycins



Properties and uses: The antibiotic lincomycin is obtained from *Actinomycetes, Streptomyces*, and *Lincolnesis*. The ability of lincomycin to penetrate into bones, adds to its qualities and it gets promoted in the chemotherapy of bone and joint infections by penicillin resistant strains of *S. aureus*. Variation of the substituents on pyrrolidine portion and C-5 side chain affects the activity. Some of the examples are as follows:

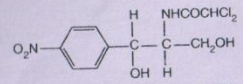
i. *N*-demethylation imparts activity against gram-negative bacteria.
ii. Increase in the chain length of the propyl substitutent at C-4 position in pyrrolidine moiety up to *n*-hexyl increase *in vivo* activity.
iii. The thiomethyl ether of α-thiolincosamide moiety is essential for activity.
iv. Structural modifications at C -7 position, such as introduction of 7S chloro or 7 R-OCH_3 change the physiochemical parameters of the drug (i.e. partition coefficient), and thus, alter the activity spectrum and pharmacokinetic properties. The usual side effects include skin rashes, nausea, vomiting, and diarrhoea.

Dosage forms: Lincomycin HCl capsules I.P.

7. Other antibiotics

Examples of other antibiotics are chloramphenicol, rifampicin and mupirocin.

Chloramphenicol or Chloromycetin



N-(1,3-Dihydroxy-1-(4-nitrophenyl)propan-2-yl)dichloroacetamide

Chloramphenicol has a spectrum of activity resembling that of the tetracyclines except that it exhibits a bit less activity against some gram-positive bacteria. It is isolated from *Salmonella venezuelae* by Ehrlich et al in 1947. It contains chlorine and is obtained from an actinomycete, and thus, named as chloromycetin. It is specifically recommended for the treatment of serious infections caused by *H. influnzae, S. typhi* (typhoid), *S. pneumoniae, and N. meningitides*. Its ability to penetrate into the CNS presents an alternative therapy for meningitis and exhibits antirickettsial activity.

Isolation of chloramphenicol: It is extracted by adopting counter-current principle using amyl acetate. The amyl acetate solution is concentrated, washed with sulphuric acid, alkali and finally with water and then evaporated. Chloramphenicol is sufficiently stable in hot water to be used at about 93°C. The crude crystals are recrystallized from water containing charcoal.

Properties and uses: Chlorampenicol is a white or greyish-white or yellowish-white crystalline powder or fine crystals, slightly soluble in water, soluble in alcohol and propylene glycol. It was the first, and still is the only therapeutically important antibiotic to be produced in competition with microbiological processes. It contains a nitrobenzene moiety and is a derivative of dichloroacetic acid. Since it has two chiral centres, four isomers are possible. The D-(-) threo is the biologically active form. It is used in the treatment of typhoid fever caused by *S. typhi*. The most serious adverse effect of chloramphenicol is bone marrow depression and fatal blood dyscrasias.

Assay: Dissolve the sample in water, dilute with the same solvent, and measure the absorbance at the maximum of 278 nm using ultraviolet spectrophotometer.

Dose: Usual adult dose is 500 mg every 6 h.