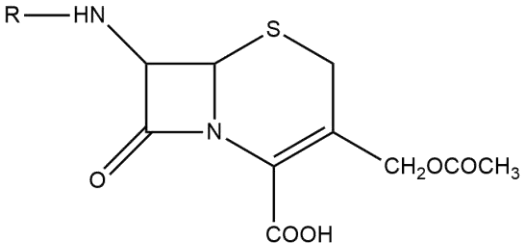
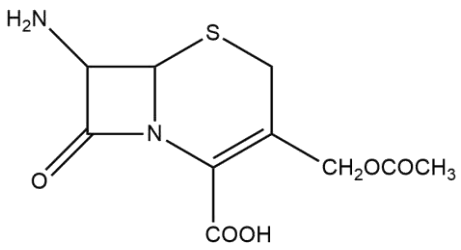
2.3. CEPHELOSPORIN

These are β-lactam antibiotics closely related (structurally and functionally) to the penicillins. Cephalosporins have 7-amino cephalosporanic acid nueleus and are obtained from the fungus *Cephalosporium*.



Cephalosporins



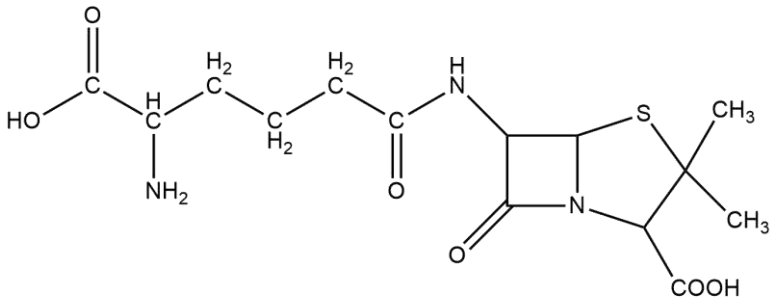
7-amino cephalorosporanic acid

2.3.1. Introduction, History & Development

- In 1945, Giuseppe Brotzu's discovery found that *Cephalosporium acremonium* cultures inhibited the growth of wide variety of Gram-positive and Gram-negative bacteria.
- The cephalosporins were isolated from the fungus *Cephalosporium acremonium* in 1948 by Pro Tzu, Newton, and Abraham (1953).
- The molecular modification of cephalosporin-C (Main Product) gave origin to semisynthetic substances.
- The development was started after modification of 7-ACA side chains with cephalothin as the first drug introduce by Eli Lilly and Company in 1964.
- Cephalosporins have similar fundamental structural requirements (β-lactam) as penicillin with little differences, it has cephems (dihydro 1,3 thiazene ring fused with β-lactam) ring while penicillin has penam ring.
- It has also similar mechanism of action as penicillin they mainly inhibit the cross-linking of the peptidoglycan units in bacterial cell walls by inhibiting transpeptidase enzyme. However, they bind in the target proteins other than penicillins binding proteins.
- The cephalosporins are much more acid stable than the corresponding penicillins

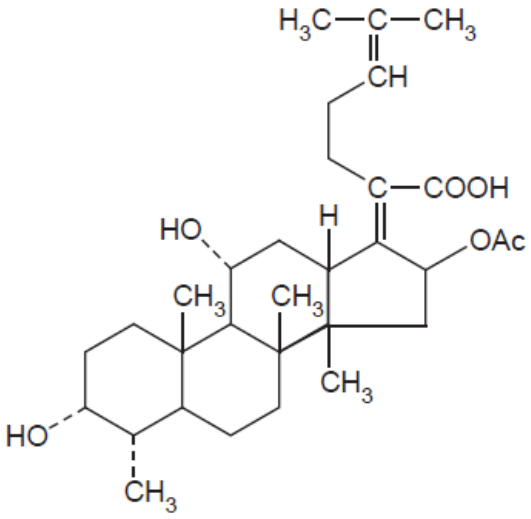
Cephalosporins can be divided into three classes:

1. *Cephalosporin N*: It has a penicillin-like structure being a derivative of 6-aminopenicillanic acid.



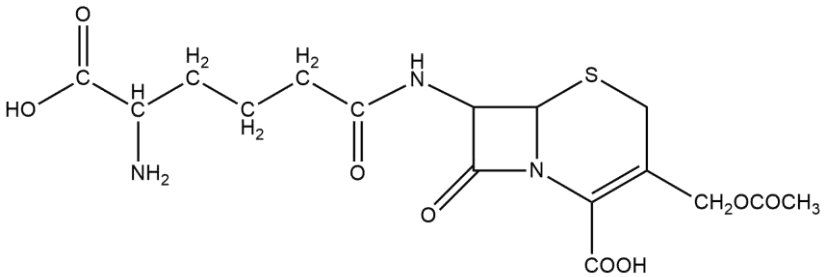
Cephalosporin N

2. *Cephalosporin P*: An acidic antibiotic, which is steroidal in nature.



Cephalosporin P

3. *Cephalosporin-C*: It is a true cephalosporin and it is a derivative of 7 amino-cephalosporanic acid, alfa-amino adipic acid.

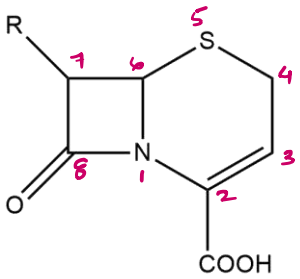


Cephalosporin-C

2.3.2. Structure and Nomenclature

System 1:

Cephem: 5-Thia-1-azobicyclo (4.2.0) oct-2-ene-8-one system



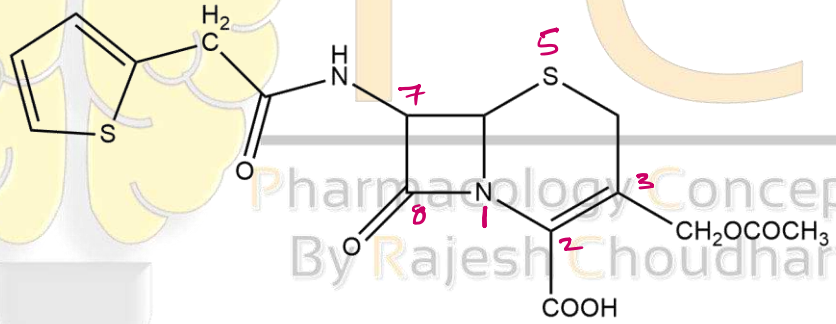
Cephem

8-oxa-5-Thia-1-azobicyclo (4.2.0) oct-2-ene-2- carboxylic acid

(Cephem-2- carboxylic acid)

Example:

Cephalothin

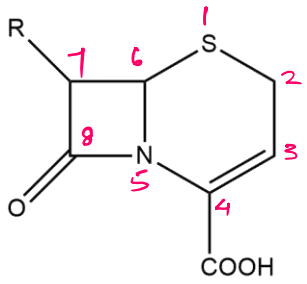


Cephalothin

3-(Acetoxy methyl)-8-oxo-7-(2-thienyl) acetamido-5thia-1-aza-bicyclo[4.2.0]-oct-2ene-2-carboxylic acid

System 2

Cephem: 1-Thia-5-azobicyclo (4.2.0) oct-3-ene-8-one system

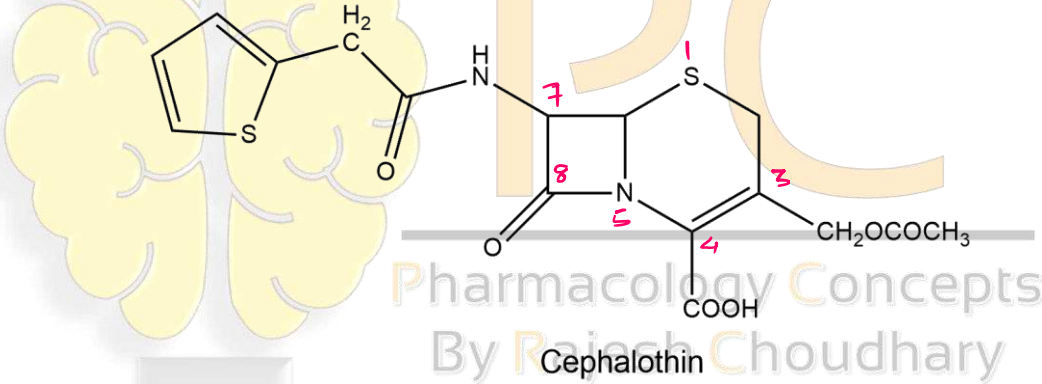


Cephem

8-oxa-1-Thia-5-azobicyclo (4.2.0) oct-3-ene-4- carboxylic acid

(Cephem-4- carboxylic acid)

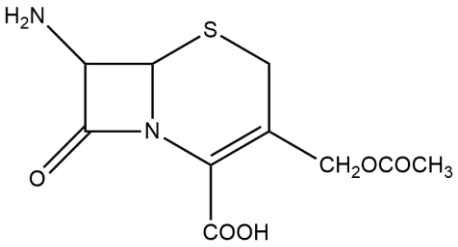
Cephalothin



Cephalothin

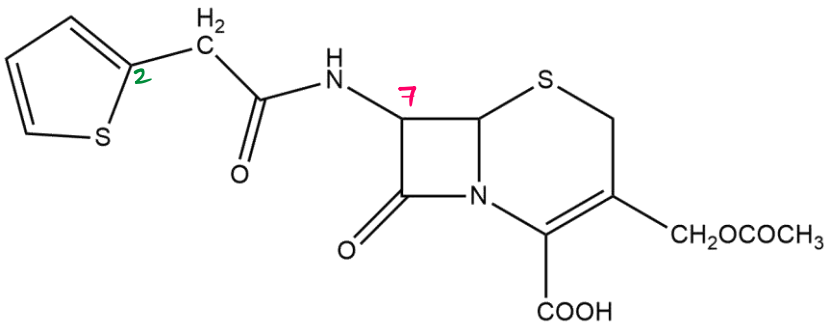
3-(Acetoxy methyl)-8-oxo-7-(2-thienyl) acetamido-1-thia-5-aza-bicyclo[4.2.0]-oct-3-ene-4-carboxylic acid

System 3: By using Cephem or cephalosporanic acid



7-amino cephalorosporanic acid

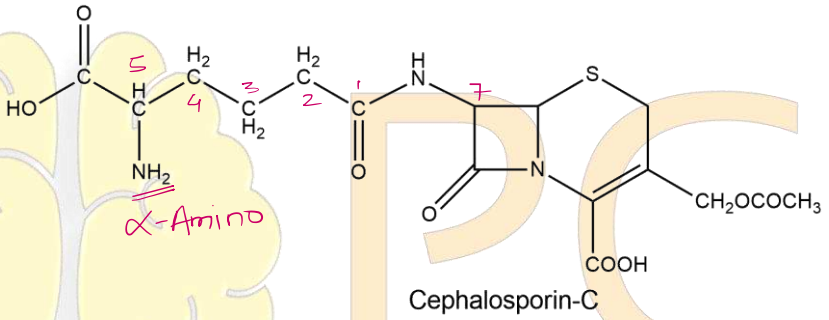
Cephalothin



Cephalothin

7-(2-thienyl) acetamido- cephalosporanic acid

Cephalosporin C

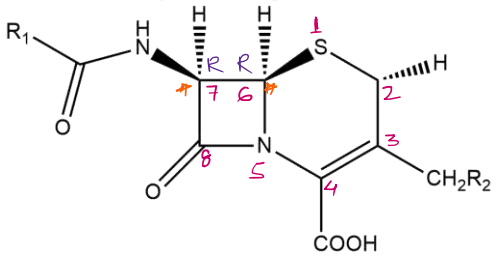


Cephalosporin-C

7-alfa-amino adipic amino or

7-(5-amino-5-cabboxy valiramido)- cephalosporanic acid

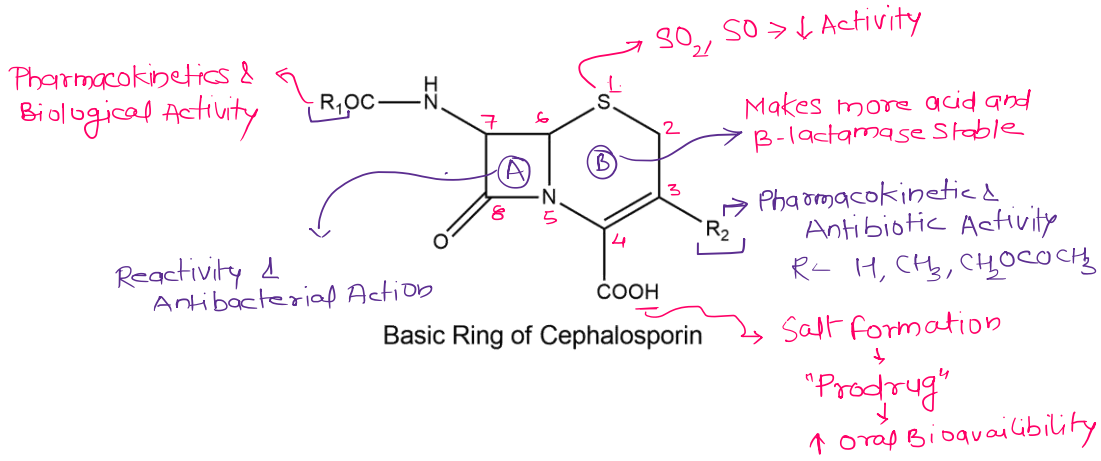
2.3.3. Stereochemistry



Cephalosporin

- Chiral Centre: at C6 and C7
- Absolute Configuration: 6R:7R

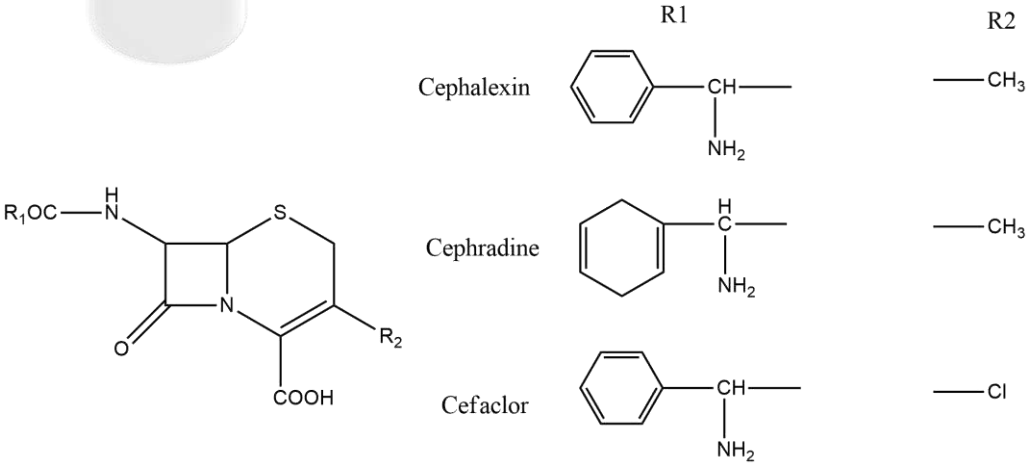
2.3.4. Structural Activity Relationship of Cephalosporin



1. β-lactam ring essential required for PBP reactivity, antibacterial activity, mechanism of action, and drug resistance
2. There are several sites for modification: A) Acylamino side chain, B) C-3 substitution, C) Sulfur atom, D) Carboxy group at C-4

A. Acylamino Substitution (R1)

1. Phenyl Glycyl Group [C6H5-CH(NH2)-COOH]: The addition of amino group and a hydrogen to α and α1 position produces basic compound, which is protonated under acidic conditions of stomach. The ammonium ion improves the stability of β-lactum of cephalosporins and make active orally. Activity against positive bacteria is increased and gram negative is decreased by acylation of amino group. E.g. **Orally Active**: cephalexin, cephradine, and cefaclor



2. When the new acyl groups are derived from carboxylic acids, it shows good spectrum of antibacterial action for gram-positive bacteria.

3. Substitutions on the aromatic ring phenyl that increase lipophilicity provide higher gram-positive activity and generally lower gram-negative activity.

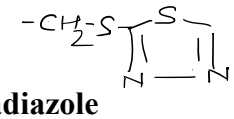
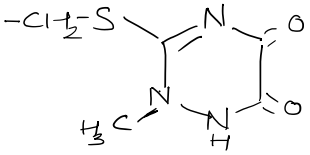
4. The phenyl ring in the side chain can be replaced with other **heterocycles** with improved spectrum of activity and pharmacokinetic properties; these include thiophene (Cephalothin, Cephaloridine) tetrazole (Cefazolin), furan (Cefuroxime), and pyridine (Cephapirin).

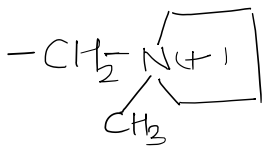
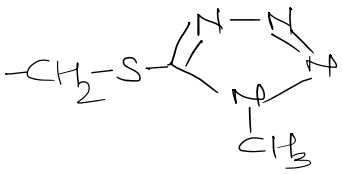
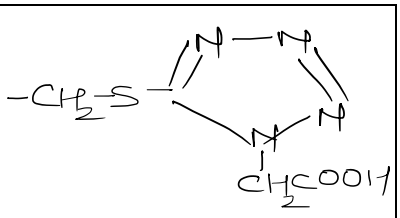
5. Introducing of methoxy imine or N-alkoxy acid imine increase the gram -ve activity and decrease the grame +ve activity. E.g., **Methoxy imines:** Cefuroxime, Cefotaxime, Ceftizoxime, Ceftriaxone. **N-alkoxy imine**: Cefixime and Ceftazidime

5. The L-isomer of an α-amino α1-hydrogen derivative of cephalosphorins was 30–40 fold stable than D-isomer.

B. R2 Substitution at C-3

1. Alteration on R2 interfere the chemical/acid stability/instability/pharmacokokinetic properties.

-H, -CH3, -Cl, -CH2OCH23 -CH=CHR Acid stable, metabolic stable, oral active derivatives	-CH2OCOCH3 Acid and Metabolic unstable Orally inactive E.g., Cephalothin, Cephapirin, Cefotaxime	-CH2OCONH2 Acid unstable Little metabolic stable Orally inactive e.g., Cefuroxime, Cefoxitin
 **thio-thiadiazole** Acid unstable, metabolic stable, oral inactive e.g. Cefazolin	 **Thio-triazine** Acid Stable, somewhat metabolic stable, orally inactive e.g. Ceftriaxone	 Acid Stable, somewhat metabolic stable, poorly oral active e.g. Cefaloridine, Ceftazidime

 Oral inactive Anti-pseudomonal activity E.g., Cefepime	 **N-methyl thio-tetrazole** E.g., Cefamandole, Cefotetan, Cefoperazone Metabolic stable Acid Unstable Not orally Active	 **thio-tetrazole-l-lactic acid** E.g., Ceforanide Metabolic stable Acid Unstable Not orally Active

2. Heterocyclic substitution enhance protein binding and half life

3. The benzoyl ester displayers improved gram-positive activity, but lowered gram-negative activity.

4. Pyridine, imidaozle replaced acetoxy group by azide ion yields derivative with relatively low gramnegative activity

C. Others

1. Double bond between C3-C4 is essential for activity, changing the position may lead to loss of activity.

2. Oxidation of S atom may lead to decrease the activity

3. replace the S atom with O enhanced the antibacterial activity

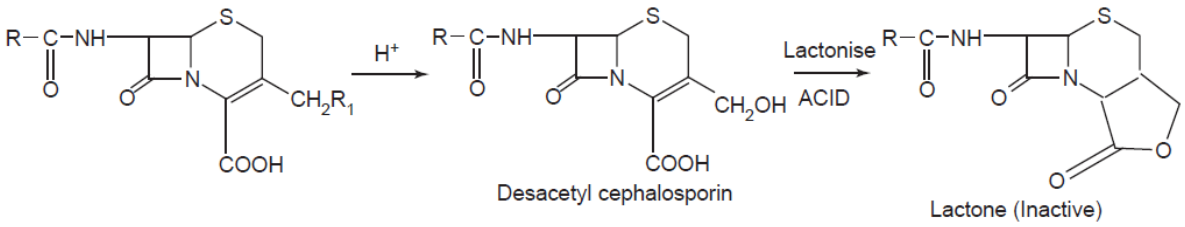
4. Carboxy group at C-4 helps to preparation of Salt and Prodrug to enhance the oral bioavailability.

5. Methoxy group at C-7, shows higher resistance to hydrolysis by β-lactamase

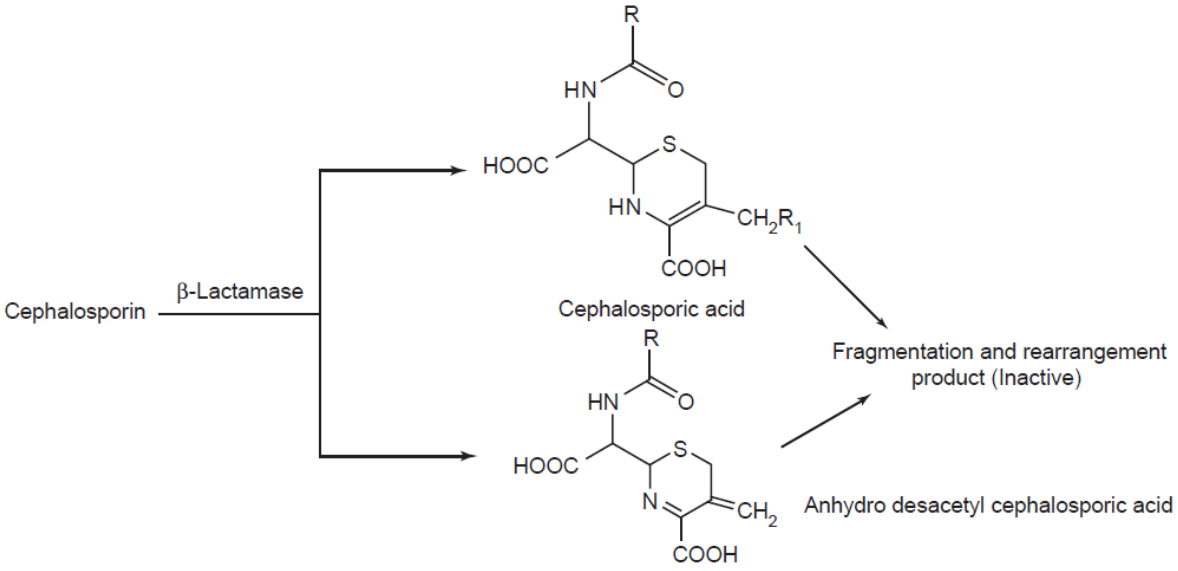
2.3.5. Chemical Degradation of Cephelosporin

Similar to penicillin, cephalosporin undergoes variety of hydrolytic degradation

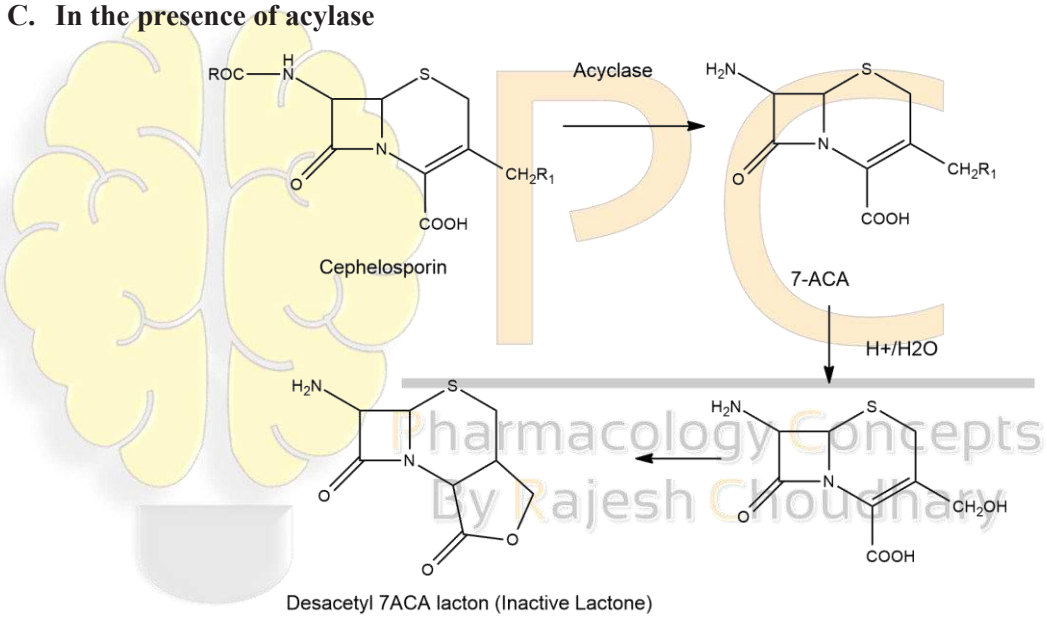
A. In the presence of strong acid



B. In the presence of beta-lactamase enzyme



C. In the presence of acylase



2.3.6. Classification and Important Products

(1) First Generation Cephalosporins:

- These have greater activily against gram positive and less activity against gram negative microorganisms.
- These are effective against *E.coli, Proteus, Klebsiella, Staphylococci, Streptococci* and *Pneumococci*.

- These are ineffective against *Salmonella, Shigella, Anaerobes* and *Pseudomonas*. These were developed in 1960s.
- e.g. Cefazolin, Cephalexin, Cephalothin, Cephaloridine, Cephradine, Cefadroxil and Cephapirin.

(2) Second Generation Cephalosporins :

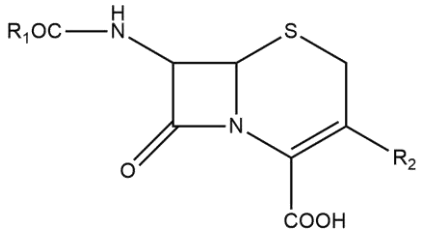
- These have greater activity against gram negative microorganism including *H. influenza, Enterobacter aerogenes* and some *Neisseria species*.
- Second generation cephalosporins were introduced subsequent to first generation cephalosporins. These are inactive against *anaerobes* and *Pseudomona aerugionosa*.
- e.g. Cefaclor, Cefoxitin, Cefamandole and Cefuroxime.

(3) Third Generation Cephalosporins:

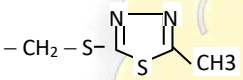
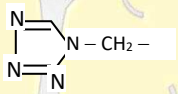
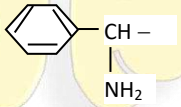
- These antibiotics offer wider coverage against gram negative bacilli and are less active on gram positive cocci. These were introduced in 1980s.
- e.g. Cefixime, Ceftriaxone, Cefotaxime, Ceftrazidime, Ceftizoxime and Cefoperazone.

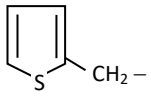
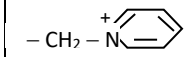
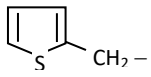
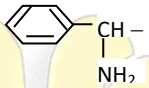
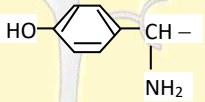
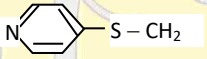
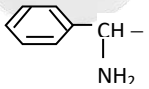
(4) Fourth Generation Cephalosporins:

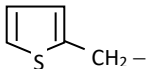
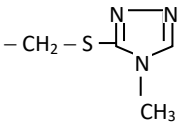
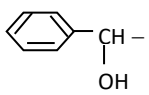
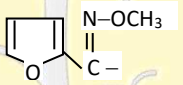
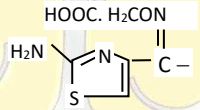
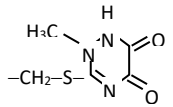
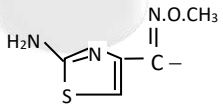
- These are newly developed cephalosporins, have same properties like those of third generation cephalosporins but more resistant to *β-lactamases*.
- Fourth generation cephalosporins are inactive against methicillin resistant *Staphylococci*.
- e.g. Cefepime and Cefprome.

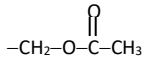
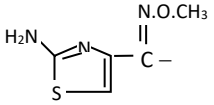
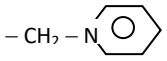
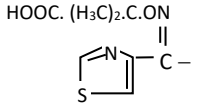
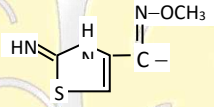
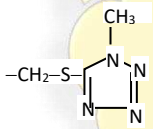
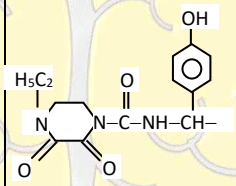


Basic Ring of Cephalosporin

COMPOUND	R2 at C3	R_1 at C7	IUPAC NAME	DOSE	USE
FIRST GENERATION					
1. Cefazolin[$]			3-[(5-methyl-1,3,4-thiadiazol-2-yl)sulfanylmethyl]-8-oxo-7-[[2-(tetrazol-1-yl)acetyl]amino]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	250-500 mg i.m./i.v. every 8 hr.	Used in infections caused by *Klebsiella*, *E.coli* and *P. mirabilis*.
2. Cephalexin*	–CH_3		7-[[2-amino-2-phenylacetyl]amino]-3-methyl-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	250 mg -1g. every 8 hr.	Used in the infections caused by *Streptococci*, *Staphylococci*, *Gonococci*, *Meningococci*, *C. diphtheriae, Clostridia, E. Coli, Klebsiella, Salmonella* etc. (less active against *penicillinase* producing *Staphylococci* and *H. influenzae*).

3. Cephalothin[$]	$-CH_2-OCO-CH_3$		3-(acetyloxymethyl)-8-oxo-7-[(2-thiophen-2-ylacetyl)amino]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	1-2 g i.v. 6 hr. (i.m. is very painful).	Used mainly in infections caused by *penicillinase* producing *Staphylococci*.
4. Cephaloridine[$]			8-oxo-3-(pyridin-1-ium-1-ylmethyl)-7-[(2-thiophen-2-ylacetyl)amino]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylate	0.5 – 2 g i.m. or i.v. 6-12 hr.	Similar to cephalexin (highly susceptible to β-*lactamases*).
5. Cephradine[#]	$-CH_3$		7-[[(2*R*)-2-amino-2-cyclohexa-1,4-dien-1-ylacetyl]amino]-3-methyl-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	250 mg–1g. oral/ im/i.v. two to four times daily.	Similar to cephalexin
6. Cefadroxil*	$-CH_3$		7-[[(2*R*)-2-amino-2-(4-hydroxyphenyl)acetyl]amino]-3-methyl-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	500 mg – 1 g two times daily.	Similar to cephalexin
7. Cephapirin[$]	$-CH_2-O-CO-CH_3$		3-(acetyloxymethyl)-8-oxo-7-[(2-pyridin-4-ylsulfanylacetyl)amino]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	1 – 2 g 6 hr. (i.m. is very painful).	Similar to cephalothin
SECOND GENERATION					
8. Cefaclor*	–Cl		7-[[(2*R*)-2-amino-2-phenylacetyl]amino]-3-chloro-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	250–500 mg every 8 hr.	Used in the infections caused by *H. influenzae E.coli and P. mirabilis.*

9. Cefoxitin[$]	–CH_2– $OCONH_2$		3-(carbamoyloxymethyl)-7-methoxy-8-oxo-7-[(2-thiophen-2-ylacetyl)amino]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	1-2 g im/i.v 6-8 hr.	Used in the infections caused by *B. fragilis*, *Serratia* and indole positive *Proteus* (Highly resistant to *β-lactamases*).
10. Cefamandole[$]			7-[[(2*R*)-2-hydroxy-2-phenylacetyl]amino]-3-[(1-methyltetrazol-5-yl)sulfanylmethyl]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	1-2g im/iv 6 hr.	Used in the infections caused by *H.influenzae*, *Klebsiella, E.coli,* indole positive *Proteus* and *Enterobacter*.
11. Cefuroxime[$]	–CH_2–O–CO– NH_2		3-(carbamoyloxymethyl)-7-[[(2*Z*)-2-(furan-2-yl)-2-methoxyiminoacetyl]amino]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	250 – 500 mg 12 hr.	It is active against *H. influenzae*, *Meningococci* & *Pneumococci*. It is also used in gonorrhoea.
THIRD GENERATION					
12. Cefixime*	– CH = CH_2		7-[[(2*Z*)-2-(2-amino-1,3-thiazol-4-yl)-2-(carboxymethoxyimino)acetyl]amino]-3-ethenyl-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	200 mg every 12 hr. or 400 mg once a day.	Used in the infections caused by *E.coli*, *Klebsiella, H. influenzae, M.catarrhalis, N. gonorrhoea & N.meningitidis.*
13. Ceftriaxone[$]			7-[[(2*Z*)-2-(2-amino-1,3-thiazol-4-yl)-2-methoxyiminoacetyl]amino]-3-[(2-methyl-5,6-dioxo-1*H*-1,2,4-triazin-3-yl)sulfanylmethyl]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	500 mg – 1 g every 12 hr.	Used in *gonorrhoea* and infections, caused by *H.influenzae*, *N.meningitidis* and *Strep. pneumoniae*.

14. Cefotaxime[$]			3-(acetyloxymethyl)-7-[[(2*Z*)-2-(2-amino-1,3-thiazol-4-yl)-2-methoxyiminoacetyl]amino]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	1-2g i.m./iv every 6-12 hr.	It is active against anaerobic gram negative and gram positive bacteria.
15. Ceftazidime[$]			7-[[(2*Z*)-2-(2-amino-1,3-thiazol-4-yl)-2-(2-carboxypropan-2-yloxyimino)acetyl]amino]-8-oxo-3-(pyridin-1-ium-1-ylmethyl)-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylate	500 mg – 2 g i.m./i.v. every 8 hr.	Highly active against *Pseudomonas*. It is also active against *Enterobactericeae.*
16. Ceftizoxime[$]	–H		7-[[(2*Z*)-2-(2-amino-1,3-thiazol-4-yl)-2-methoxyiminoacetyl]amino]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	500 mg–1g i.m./i.v. every 8-12 hr	Similar to cefotaxime.
17. Cefoperazone[$]			7-[[(2*R*)-2-[(4-ethyl-2,3-dioxopiperazine-1-carbonyl)amino]-2-(4-hydroxyphenyl)acetyl]amino]-3-[(1-methyltetrazol-5-yl)sulfanylmethyl]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	1 – 2g i.m./i.v. every 12 hr.	Most active against *Pseudomonas*. Used in the treatment of infections caused by *S.typhi* and *B. fragilis* (*suseptible* to β-*lactamases*).

***** Administered orally, **$** Administered parenterally, # ***Cephradine is the only cephalosporin available both for oral and parenteral administration***