Organic Pharm. Chemistry III

Lecture 5

19th March 2023

10:30- 12:30

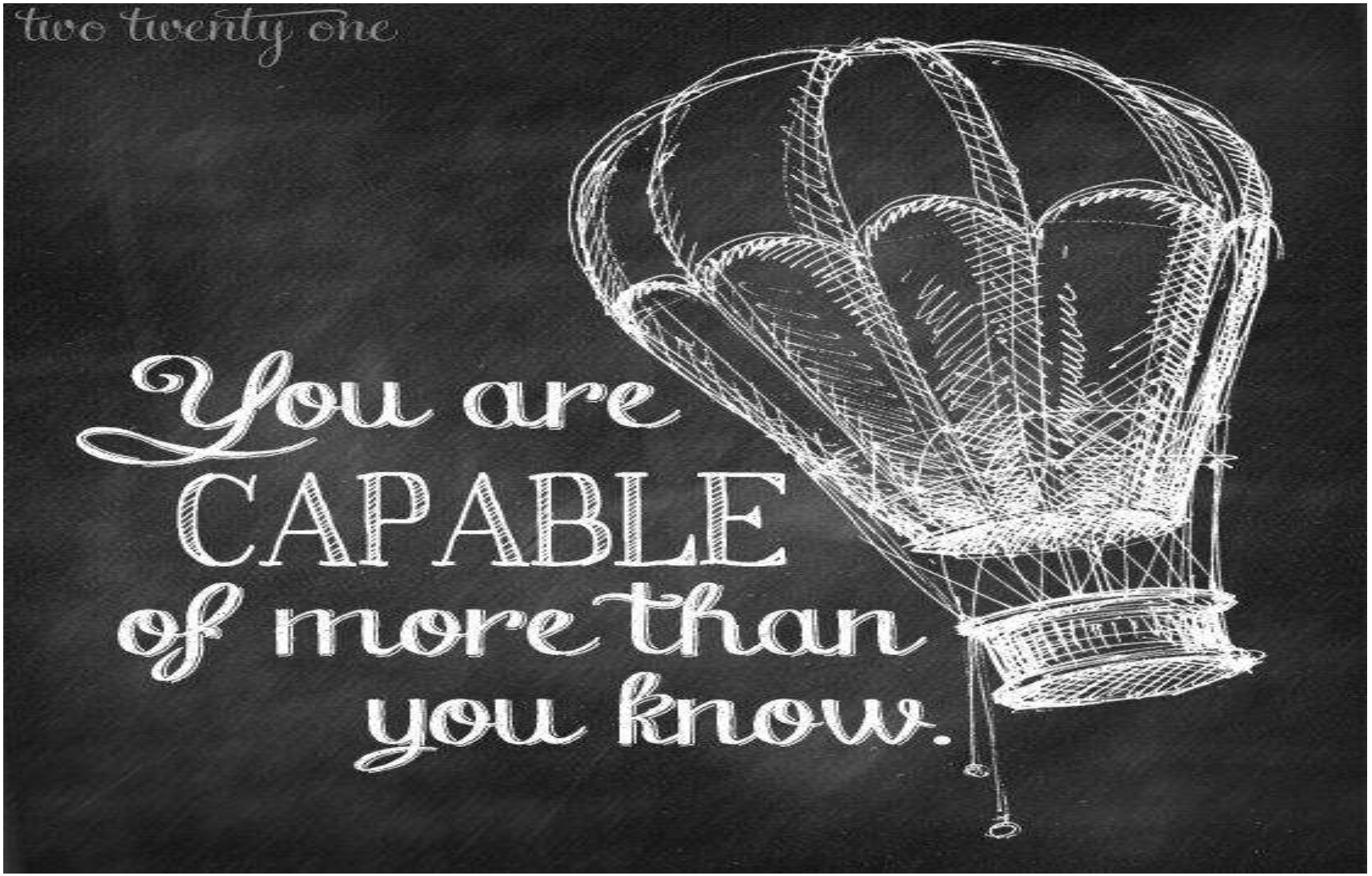
AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023

Inspired Quote of today !



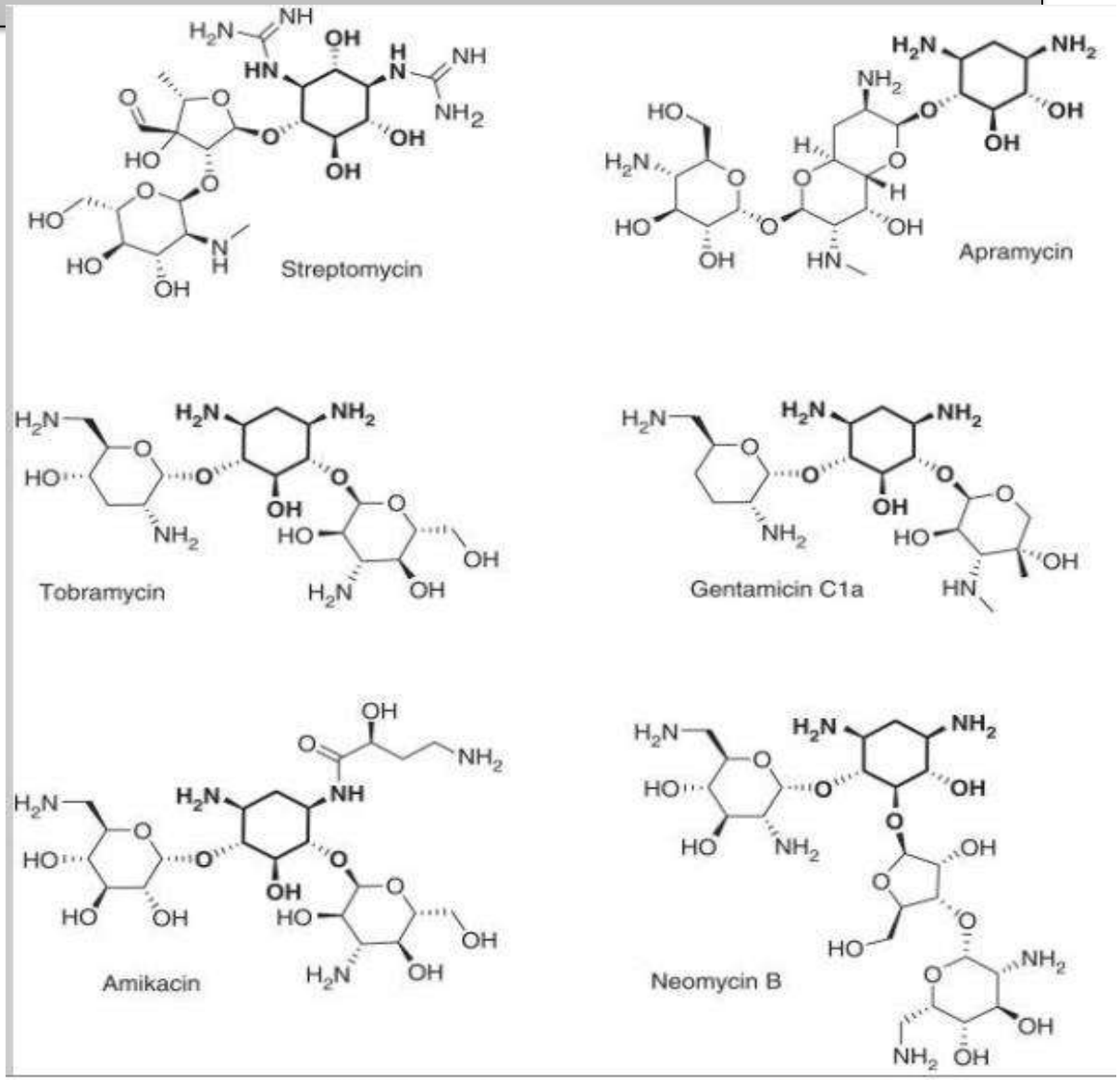
AMINOGLYCOSIDES

Objectives:

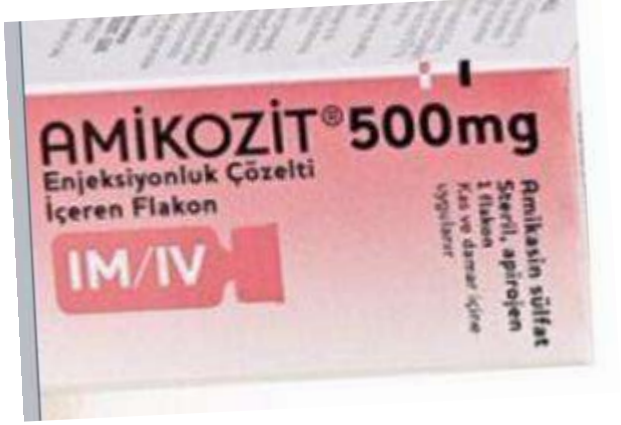
1. Understand the chemistry of drug with respect to their biological activity.
2. Know the Importance of SAR of Drugs
3. Know the metabolism, Adverse Effect and therapeutic value of drugs.

Learning Outcomes:

1. Students will Learn about the structures and Medicinal uses of Drugs.
2. Students will learn about relation of structure with its activity.



AMINOGLYCOSIDES



INTRODUCTION

• Include s**treptomycin, neomycin, kanamycin, amikacin, gentamicin, tobramycin, sisomicin, netilmicin**, and others

• Widely in combination with a β-lactam antibiotic

– serious infections with **gram-negative bacteria**

– combination with **vancomycin gram-positive endocarditis**

• Treatment of Tuberculosis.

Mechanism of Action

• Irreversible inhibitors of protein synthesis on either of the ways

– interference with the **initiation complex of peptide formation**

– misreading of mRNA - incorporation of incorrect amino acids into the peptide and results in a **nonfunctional or toxic protein**

– breakup of polysomes into nonfunctional monosomes

Mechanism of resistance

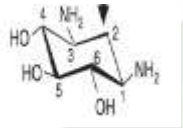
Production of a **transferase - inactivates the aminoglycoside**

• Impaired entry of aminoglycoside into the cell.

• Receptor protein on the 30S ribosomal subunit may be deleted or altered as a result of a mutation.

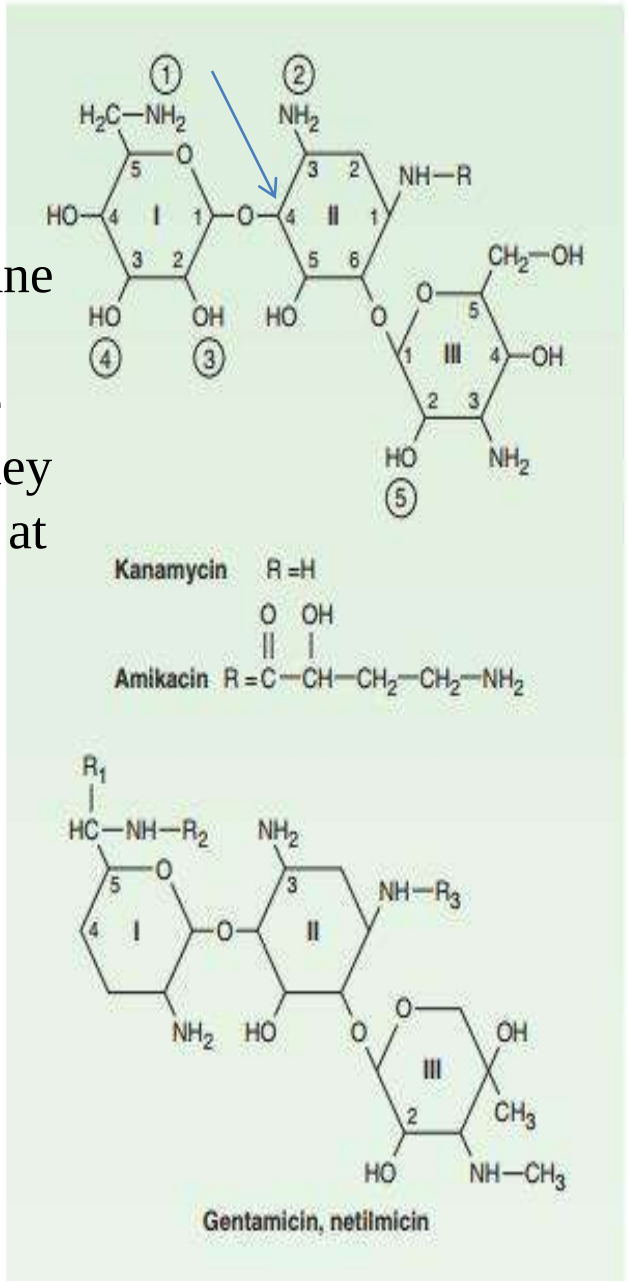
Physical and Chemical Properties

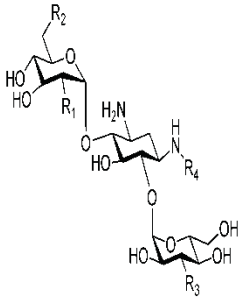
Aminoglycosides have a **hexose ring**, either streptidine (in streptomycin) or 2-deoxystreptamine (in other aminoglycosides), to which various amino sugars are attached by glycosidic linkages (Figures 1 and 2). They are water-soluble, stable in solution, and more active at alkaline than at acid pH.

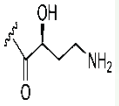


deoxystreptamine

FIGURE 1 Structure of streptomycin.

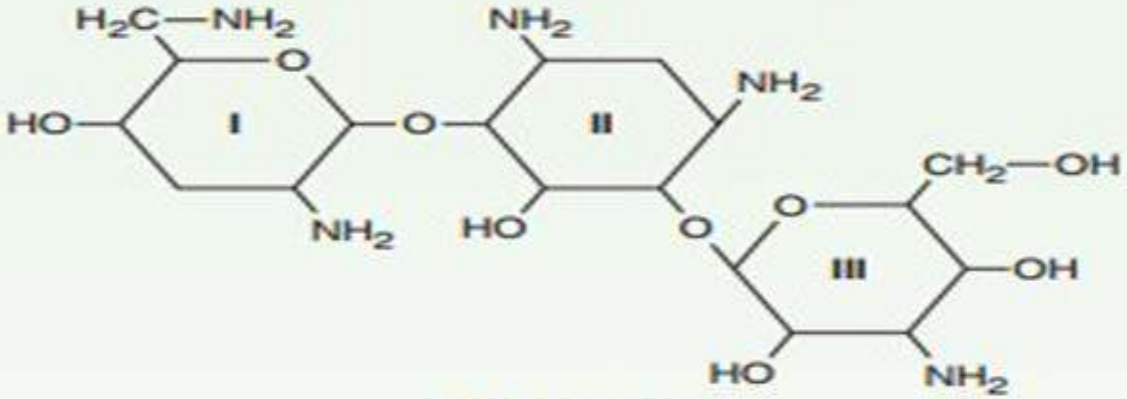




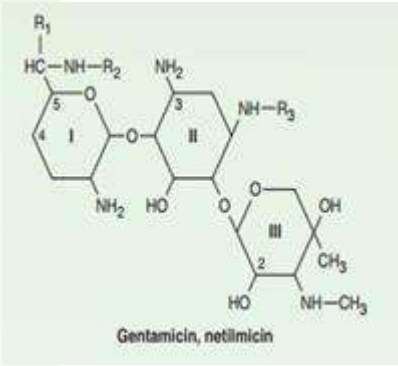
	R1	R2	R3	R4
kanamycin A	OH	NH_2	NH_2	H
kanamycin B	NH_2	NH_2	NH_2	H
kanamycin C	NH_2	OH	NH_2	H
kanamycin D	OH	NH_2	OH	H
amikacin	OH	NH_2	NH_2	

	Ring I			Ring II
	R_1	R_2	C4–C5 bond	R_3
Gentamicin C_1	CH_3	CH_3	Single	H
Gentamicin C_2	CH_3	H	Single	H
Gentamicin C_{1a}	H	H	Single	H
Netilmicin	H	H	Double	C_2H_5

Netilmicin



Tobramycin



Gentamicin, netilmicin

FIGURE 2 Structures of several important aminoglycoside antibiotics. Ring II is 2-deoxystreptamine. The resemblance between kanamycin and amikacin and between gentamicin, netilmicin, and tobramycin can be seen. The circled numerals on the kanamycin molecule indicate points of attack of plasmid-mediated bacterial transferase enzymes that can inactivate this drug. ①, ②, and ③, acetyltransferase; ④, phosphotransferase; ⑤, adenylyltransferase. Amikacin is resistant to modification at ②, ③, ④, and ⑤.

CLINICAL USES

Mycobacterial Infections

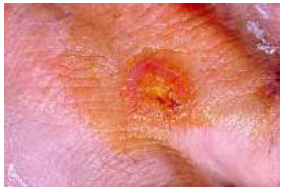
– second-line agent for treatment of **tuberculosis**

– 0.5–1 g/d - intramuscularly or intravenously

– combination with other agents to prevent emergence of resistance

• **Nontuberculous Infections**

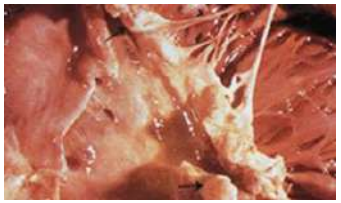
– plague, tularemia, and sometimes brucellosis : 1 g/d i/m in combination with an oral tetracycline.



– Combination with Penicillin

• enterococcal endocarditis

• 2-week therapy of viridans streptococcal endocarditis.



• enterococcal infections

ADVERSE REACTIONS

• Fever, skin rashes, and other allergic manifestations – hypersensitivity (**prolonged course of treatment)**

• Pain at the injection site

• Disturbance of vestibular function (irreversible)

— vertigo and loss of balance - proportion to the Age of the patient,

– Blood levels of the drug

– Duration of administration

• Pregnancy : deafness in the newborn – relatively Contraindicated

CLINICAL USE

• **Topical Administration**

– Solutions containing 1–5 mg/mL – infected surfaces or injected into joints, the pleural cavity, tissue spaces, or abscess cavities.

– limited to 15 mg/kg/d – **higher doses may be absorbed to produce systemic toxicity.**

– **neomycin-polymyxin-bacitracin combination**: applied to infected skin lesions or in the nares for suppression of staphylococci.

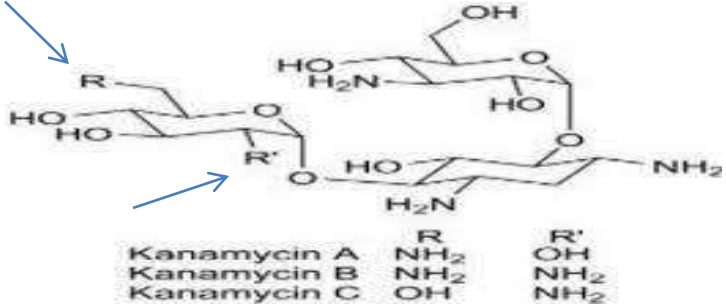
• **Oral Administration**

– Preparation for **elective bowel surgery** : 1 g of neomycin with 1 g of erythromycin base is given orally every 6–8 hours for 1–2 days combined.

– Reduces the aerobic bowel flora with little effect on anaerobes.

– Hepatic encephalopathy: coliform flora can be suppressed by giving 1 g every 6–8 hours.

Structure–Activity Relationship

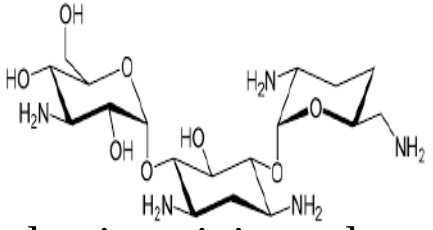


It is convenient to discuss sequentially aminoglycoside SARs in terms of **substituents in rings I, II, and III**.

Ring I is crucially **important for characteristic broad spectrum antibacterial activity, and it is the primary target for bacterial inactivating enzymes.**

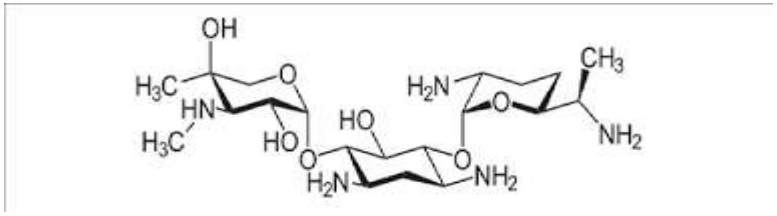
Amino functions at 6` and 2` are particularly important as **kanamycin B** (6`amino, 2`-amino) is more active than **kanamycin A** (6`-amino, 2`- hydroxyl), which in turn is more active than **kanamycin C** (6`-hydroxyl, 2`amino).

Methylation at either the 6`-carbon or the 6`-amino positions does not lower appreciably antibacterial activity and confers resistance to enzymatic acetylation of the 6`-amino group.



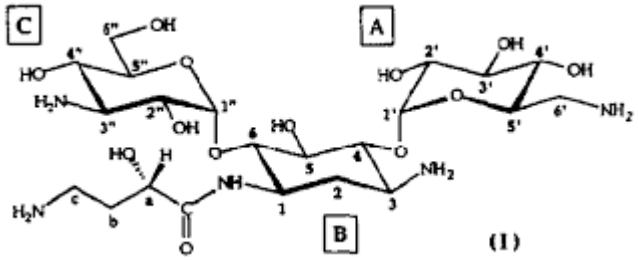
Removal of the 3`-hydroxyl or the 4`-hydroxyl group or both in the kanamycins (e.g., 3`,4`- dideoxykanamycin B or dibekacin) does not reduce antibacterial potency.

The **gentamicins** also lack oxygen functions at these positions, as do sisomicin and netilmicin, which also have a 4`5`-`dideoxykanamycin B or dibekacin) does not reduce antibacterial potency.



None of these derivatives is inactivated by phosphotransferase enzymes that phosphorylate the 3 hydroxyl group.

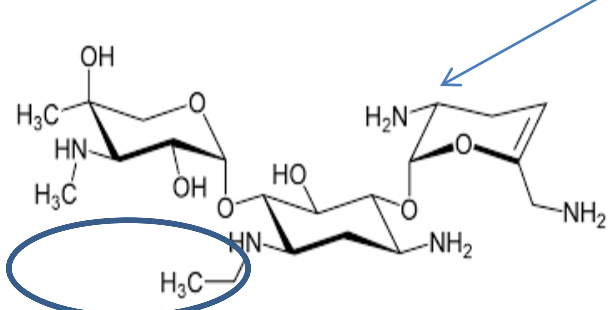
Evidently, the 3`-phosphorylated derivatives have very low affinity for aminoglycoside-binding sites in bacterial ribosomes.

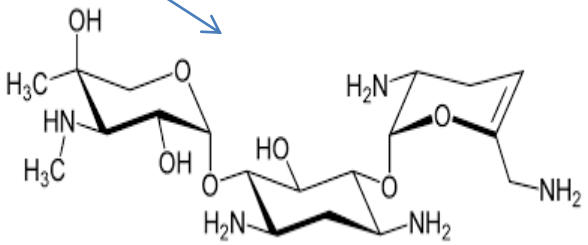


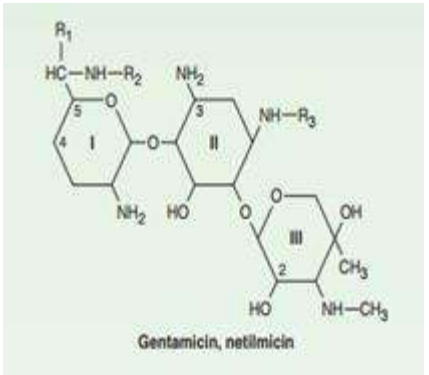
Ring II

Few modifications of **ring II** (deoxystreptamine) functional groups are possible without appreciable loss of activity in most of the aminoglycosides.

The **1-amino group of kanamycin A can be acylated** (e.g., **Amikacin**), however, with activity largely retained. Netilmicin (1-N-ethylsisomicin) retains the antibacterial potency of sisomicin and is resistant to several additional bacteria-inactivating enzymes.

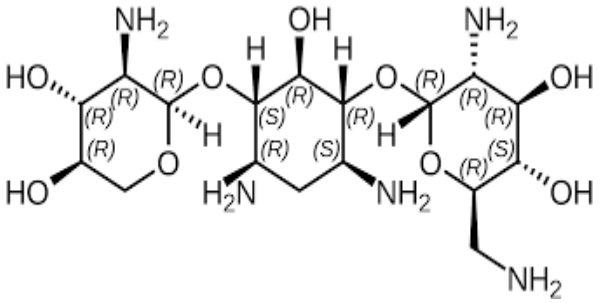






Ring III functional groups appear to be somewhat less sensitive to structural changes than those of either ring I or ring II.

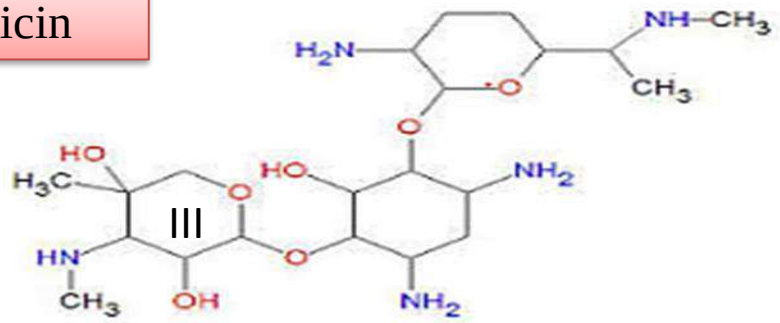
Seldomycin

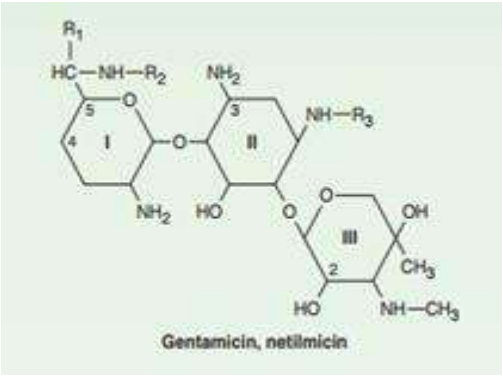


Although the 2``-deoxygentamicins are significantly less active than their 2``-hydroxyl counterparts, the 2``-amino derivatives (seldomycins) are highly active.

The 3``-amino group of gentamicins may be primary or secondary with high antibacterial potency. Furthermore, the 4``- hydroxyl group may be axial or equatorial with little change in potency.

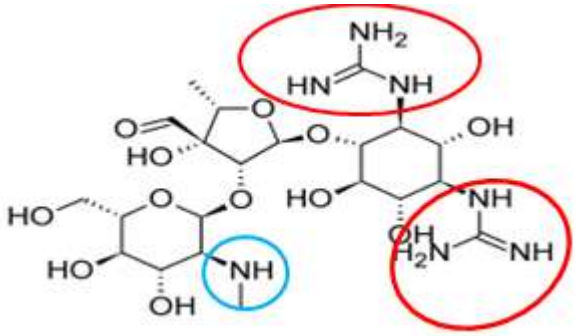
Gentamicin





Streptomycin Sulfate

Streptomycin Sulfate, Streptomycin acts as a **triacidic base** through the effect of its **two strongly basic guanidino groups** and the more weakly basic methylamino group



Acid hydrolysis yields streptidine and streptobiosamine

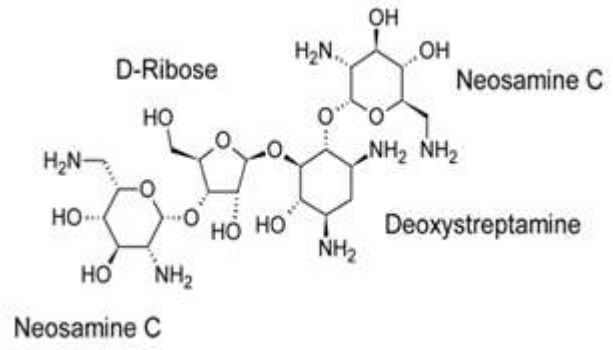
Because streptomycin is not absorbed when given orally or destroyed significantly in the GI tract, at one time it was used **rather widely in the treatment of infections of the intestinal tract.**

For **systemic action, streptomycin usually is given by intramuscular injection.**

Hydroxystreptomycin differs from streptomycin in having a hydroxyl group in place of one of the hydrogen atoms of the streptose methyl group. Mannisidostreptomycin has a mannose residue attached in glycosidic linkage through the hydroxyl group at C-4 of the N-methyl-L-glucosamine moiety.

Neomycin Sulfate

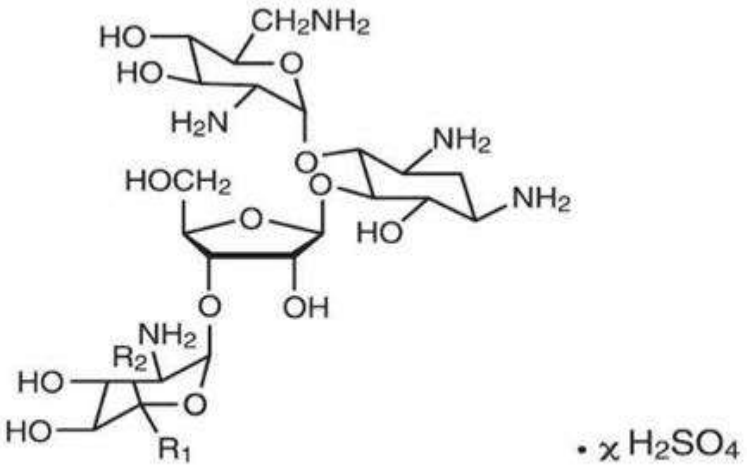
Neomycin Sulfate



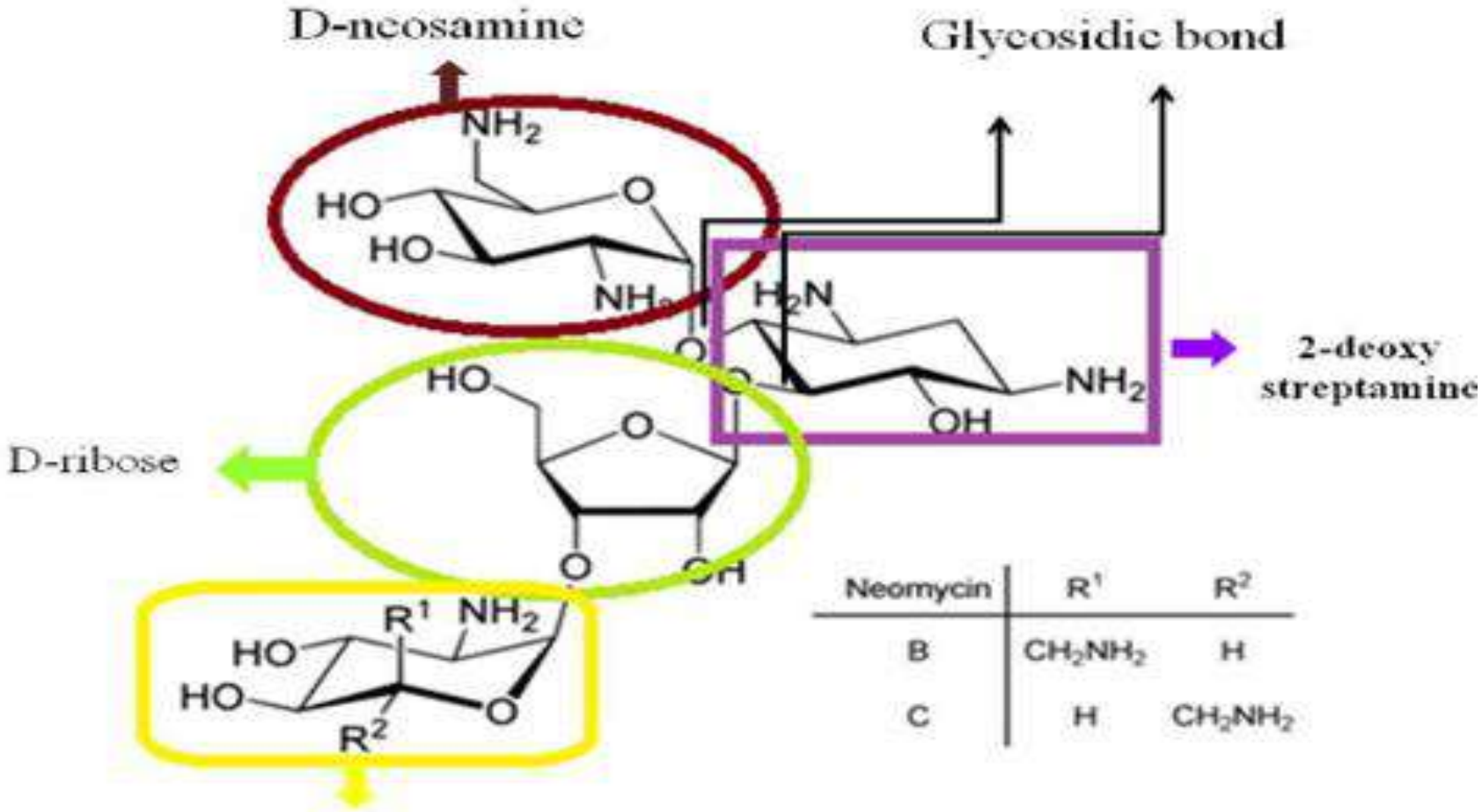
Neosamine C

It is considered **one of the most useful antibiotics for the treatment of GI infections,** It has broad-spectrum activity against various organisms and shows a low incidence of toxic and hypersensitivity reactions. It is absorbed very slightly from the digestive tract, so its oral use ordinarily does not produce any systemic effect.

neomycin B differs from neomycin C by **the nature of the sugar attached terminally to D-ribose**. That sugar, called **neosamine B**, differs from **neosamine C in its stereochemistry**.

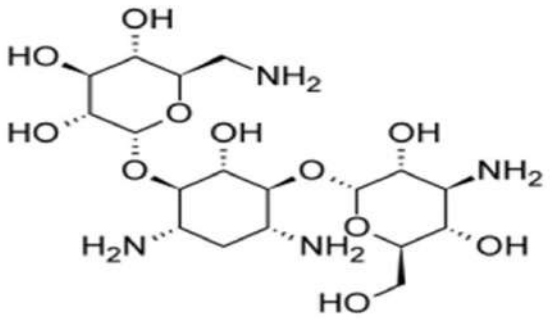


Neomycin B (R_1=H, R_2=CH_2NH_2)
Neomycin C (R_1=CH_2NH_2, R_2=H)



Neomycin	R^1	R^2
B	CH_2NH_2	H
C	H	CH_2NH_2

Kanamycin Sulfate



Chromatography showed that S. kanamyceticus **elaborates three closely related structures: kanamycins A, B, and C.** Commercially available kanamycin is almost pure kanamycin A, the least toxic of the three forms.

The kanamycins differ only in the **sugar moieties attached to the glycosidic oxygen on the 4-position of the central deoxystreptamine**

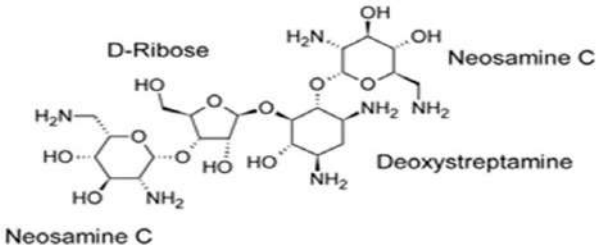
The kanamycins **do not have the D-ribose molecule that is present in neomycins and paromomycins. Perhaps this structural difference is related to the lower toxicity observed with kanamycins.**

Kanamycin **A** contains 6-amino-6-deoxy-D-glucose

Kanamycin **B** contains 2,6-diamino- 2,6-dideoxy-D-glucose

Kanamycin **C** contains 2-amino-2-deoxy-D-glucose

The use of kanamycin in the United States usually is restricted to infections of the **intestinal tract (e.g., bacillary dysentery) and to systemic infections arising from Gram negative bacilli.**

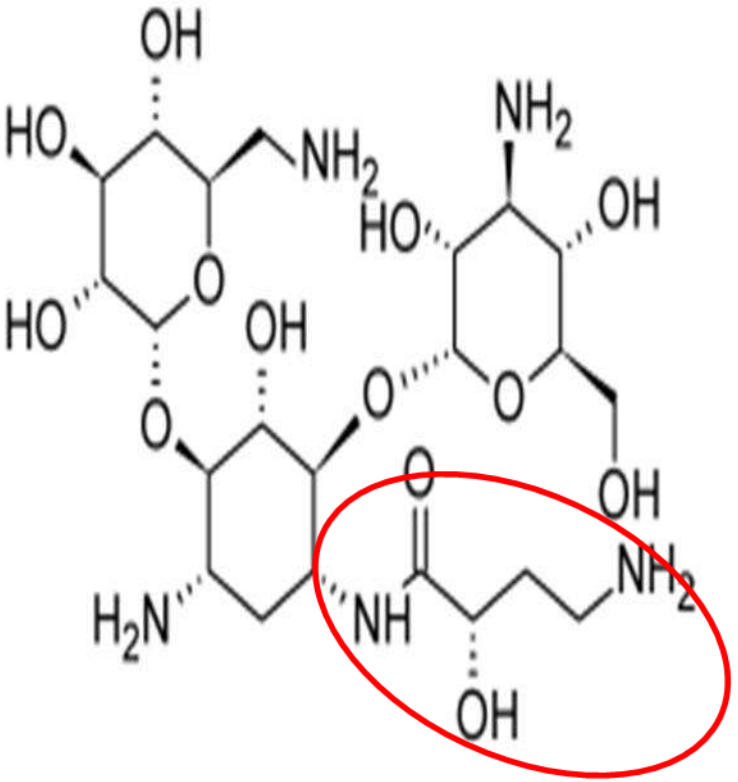


Amikacin

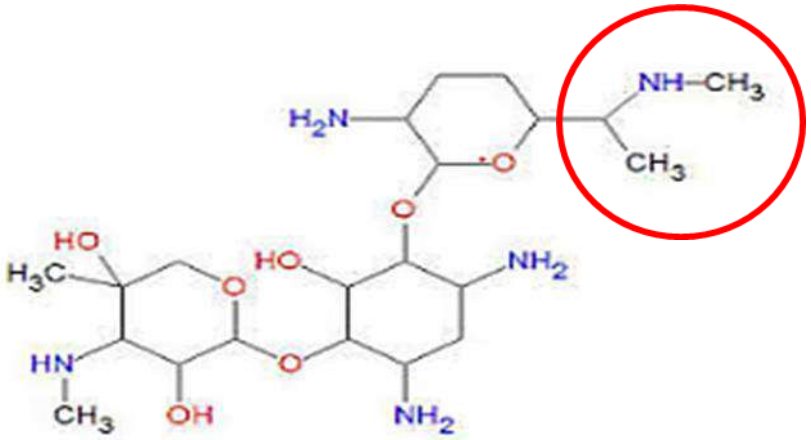
1-N-amino-α-hydroxy butyryl kanamycin A(Amikin),
is a semisynthetic aminoglycoside first **prepared in Japan**.

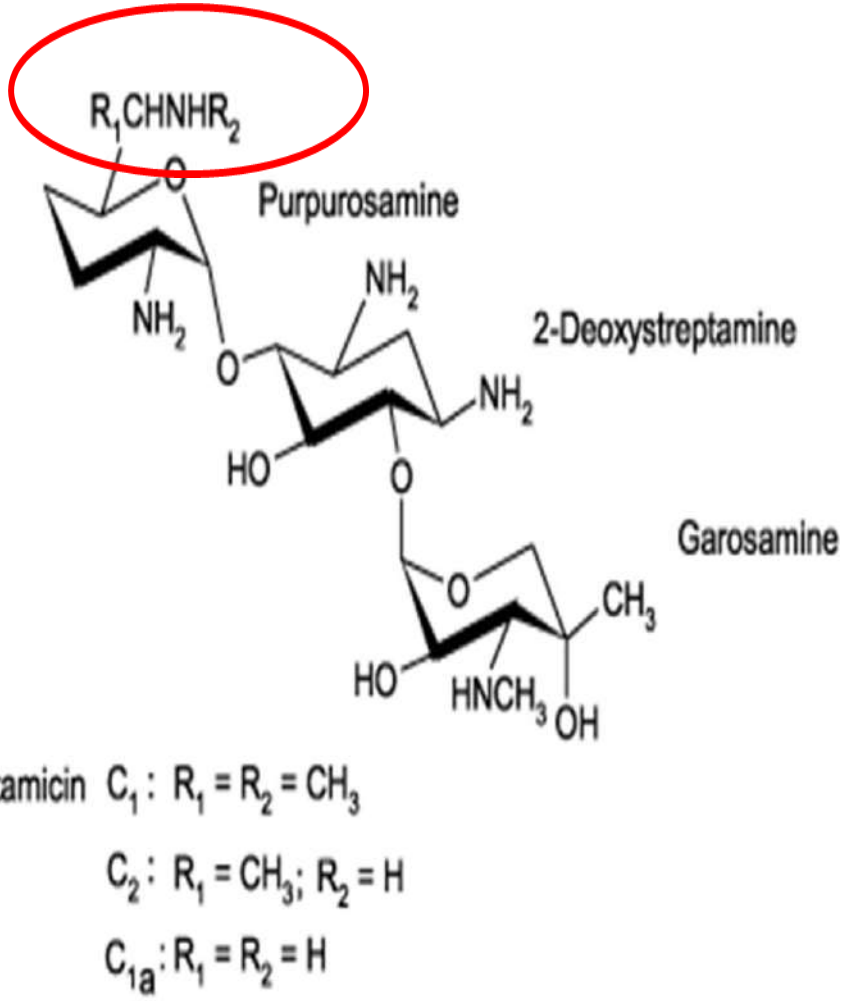
The synthesis formally involves simple acylation of the 1 amino group of the deoxystreptamine ring of kanamycin A.

The remarkable feature of Amikacin is that **it resists attack by most bacteria-inactivating enzymes and, therefore, is effective against strains of bacteria that are resistant to other aminoglycosides, including gentamicin and tobramycin.**



Gentamicin Sulfate





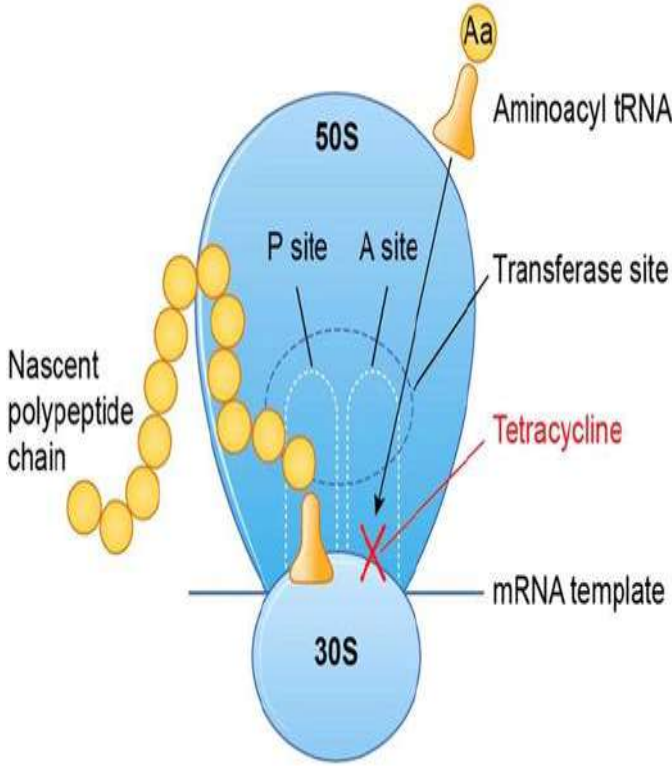
Gentamicin C_1: $R_1 = R_2 = CH_3$

C_2: $R_1 = CH_3$; $R_2 = H$

C_{1a}: $R_1 = R_2 = H$

Gentamicin is composed of a number of related gentamicin components and fractions which have varying degrees of antimicrobial potency. The main components of gentamicin include members of **the gentamicin C complex: gentamicin C1, gentamicin C1a, and gentamicin C2** which compose approximately 80% of gentamicin and have been found to have the highest antibacterial activity. Gentamicin A, B, X, and a few others make up the remaining 20% of gentamicin and have lower antibiotic activity than the gentamicin C complex

TETRACYCLINE



INTRODUCTION

Most important broad spectrum antibiotics obtained by:

-Fermentation procedures from Streptomyces spp.

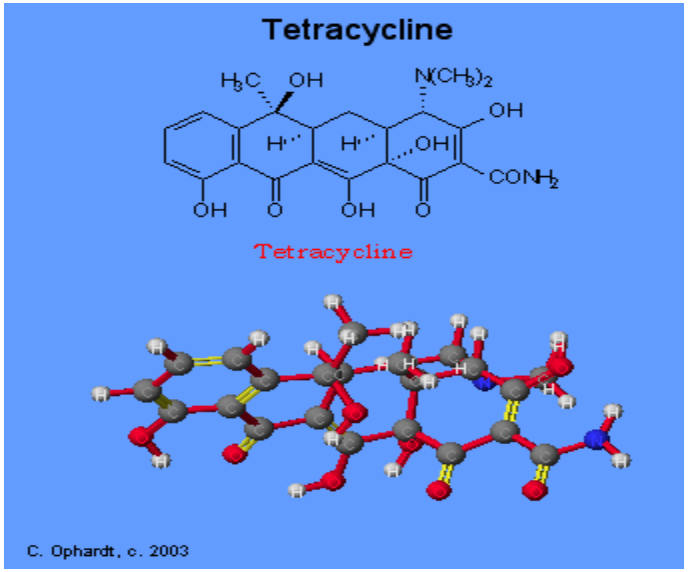
-Chemical transformation of natural products

Derivatives of octahydronaphthacene- A hydrocarbon system that comprises four-annulated six-membered rings.

Amphoteric compounds

MECHANISM OF ACTION

•Specific inhibitors of **bacterial protein synthesis**



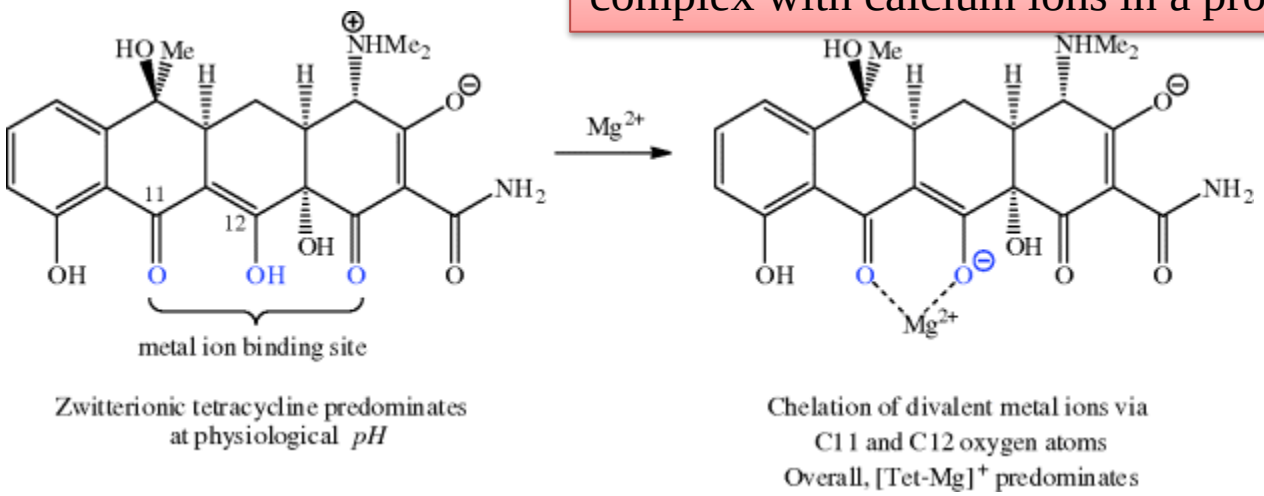
Stable chelate complexes are formed by the tetracyclines with many metals, including calcium, magnesium, and iron.

Such chelates are usually very insoluble in water, accounting for the impaired absorption of most (if not all) tetracyclines in the presence of milk; calcium-, magnesium-, and aluminum-containing antacids; and iron salts.

Soluble alkalinizers, such as sodium bicarbonate, also decrease the GI absorption of the tetracyclines.

The affinity of tetracyclines for calcium causes them to be incorporated into newly forming bones and teeth **as tetracycline–calcium orthophosphate** complexes

The mechanism of staining is tetracycline's ability to form a complex with calcium ions in a process called chelation



Zwitterionic tetracycline predominates at physiological *pH*

Chelation of divalent metal ions via C11 and C12 oxygen atoms
Overall, $[Tet\text{-}Mg]^+$ predominates

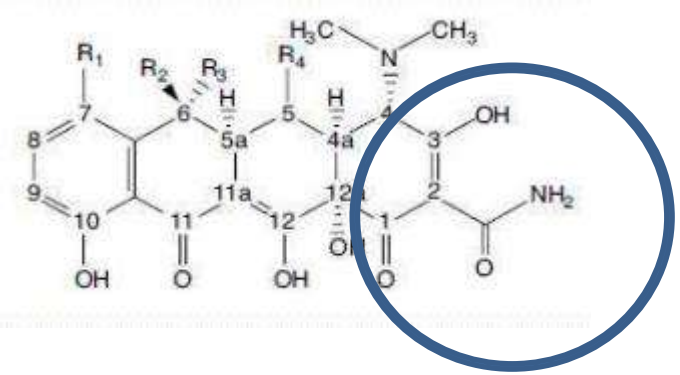
MECHANISMS OF RESISTANCE

A.) Efflux mediated by trans-membrane spanning

B.) Ribosomal protection

C.) Enzymatic oxidation

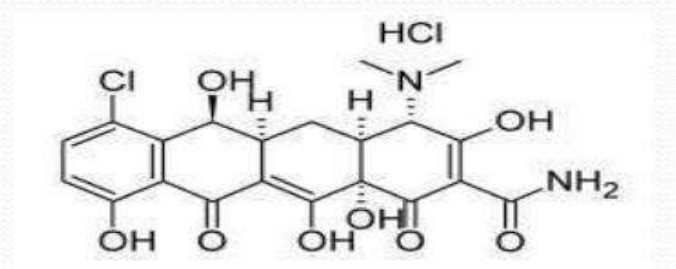
GENERAL STRUCTURE



STRUCTURE-ACTIVITY RELATIONSHIP

Positive effects:

- **<u>The enolized tricarbonylmethane system at C-1 to C-3 must be intact for good activity.</u>**

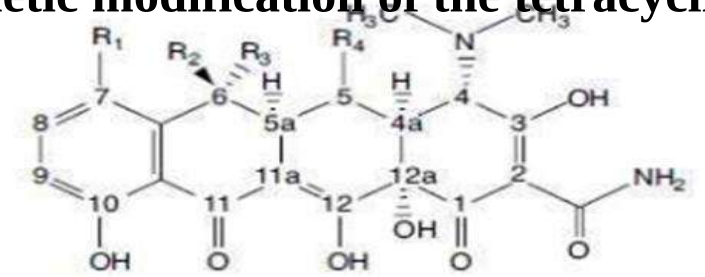


Aminoalkylation of the amide nitrogen yield derivatives that is more water soluble. (Rolitetracycline)

•Substituents at position 5,5a,6,7,8,9 (hydrophobic “northern and western” faces of the molecule).

•**More useful results have been achieved with the introduction of substituents at C-7.**

•**6-position: most fruitful site for semisynthetic modification of the tetracyclines.**



Negative effects:

•All derivatives containing fewer than four rings are inactive or nearly inactive.

•Replacement of the amide at C-2 with other functions (aldehyde or nitrile) reduces or abolishes activity.

•Monoalkylation of the **amide nitrogen** reduces activity proportionately to the size of the alkyl group.

•Removal of the 4-dimethylamino group reduces activity.

•Esters of the C-12a hydroxyl group are inactive, with the exception **of the formyl ester**

Alkylation at C-11a also leads to inactive compounds.

Dehydrogenation to form a double bond between C-5a and C-11a markedly decreases activity.

According to source:

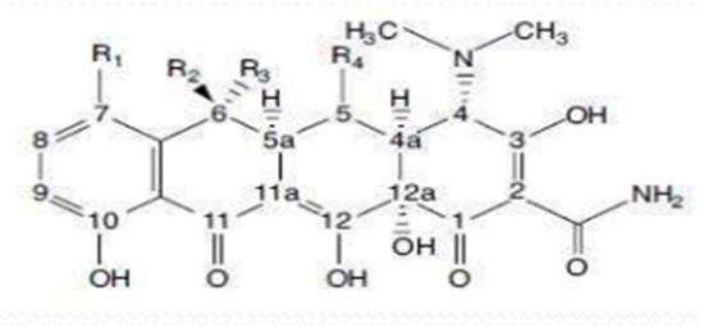
- **Naturally occurring**

-Tetracycline

-Chlortetracycline

-Oxytetracycline

-Demeclocycline



- **Semi-synthetic**

-Meclocycline

-Methacycline

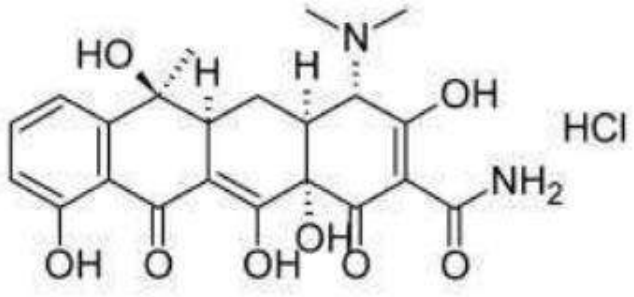
-Minocycline

-Rolitetracycline

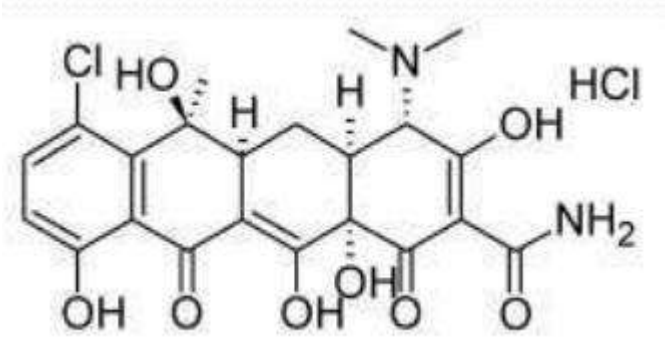
According to duration of Action

Short-acting: (half-life is 6-8hrs)

TETRACYCLINE



[4-dimethyl amino-1,4,4a,5,5a,6,11,12aoctahydro-3,6,10,12,12a-pentahydroxy-6- methyl-1,11 dioxo-2-naphthacenecarboxamide]

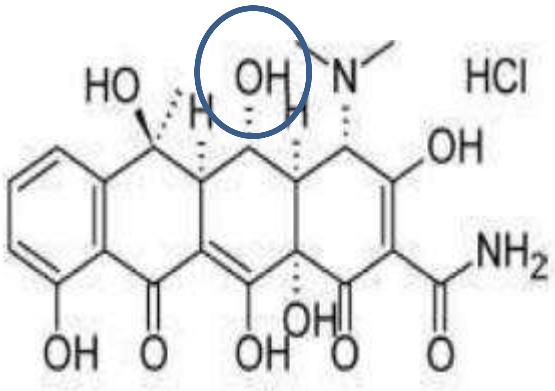


Chlortetracycline HCl

7-chloro-4-(dimethylamino)-1,4,4a,5,5a,6,11,12aoctahydro3,6,10,12,12a-pentahydroxy-6-methyl1,11-dioxo-2-naphthacenecarboxamide

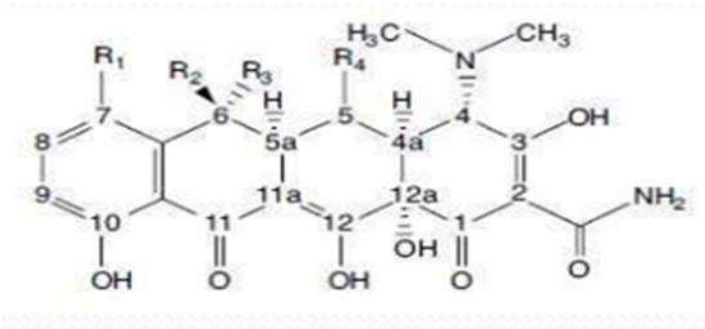
Oxytetracycline HCl

Chemical analog of chlortetracycline that showed similar antibiotic properties



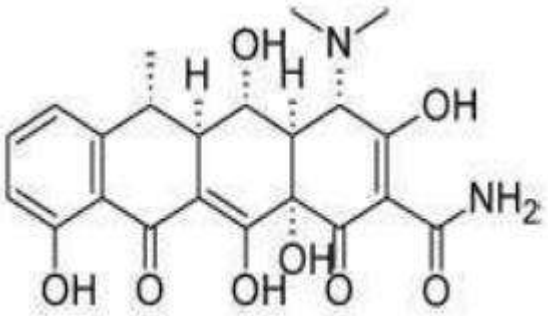
Long acting: (half-life is 16hrs)

Doxycycline



α-6-deoxy-5-oxytetracycline

Minocycline HCl



7-dimethylamino-6-demethyl-6-deoxytetracycline

CLINICAL USES

Primary Uses:

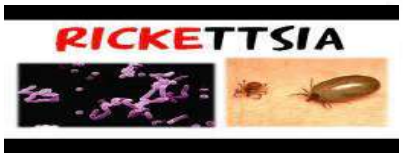
Treatment for infections caused by:

Mycoplasma pneumoniae

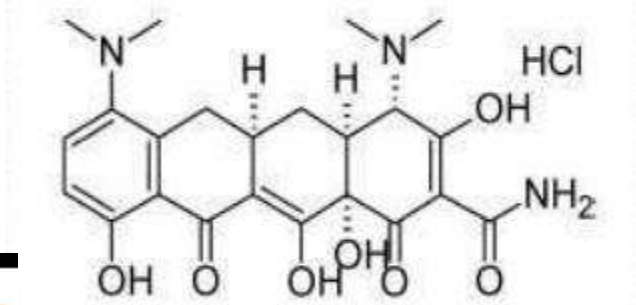
Chlamydiae

Ricketssiae

Vibrios



Spirochetes

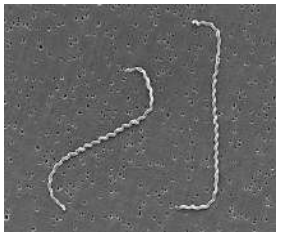


Secondary Uses:

Alternative drugs in the treatment of **syphilis**

Treatment of respiratory infections caused by susceptible organisms

Treatment of **leptospirosis**



Treatment of acne

Prophylaxis against infection in chronic Bronchitis

Selective Uses:

Tetracycline

Treatment of **gastrointestinal ulcers caused by Helicobacter pylori**

Doxycycline

Lyme disease

Prevention of malaria

Treatment of amebiasis

Currently an alternative to macrolides in the initial of community acquired pneumonias

Minocycline

Meningococcal carrier state

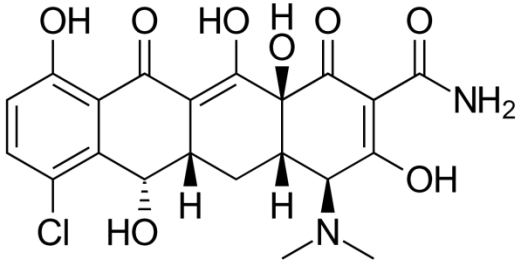
Demeclocycline

Inhibits the renal actions of antidiuretic hormone(ADH)

Management of patients with ADH-secreting Tumors

TOXICITY

Gastrointestinal disturbances



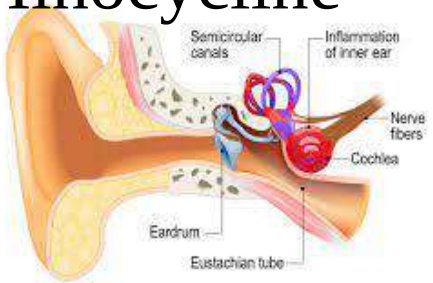
Bony structures and teeth

Hepatic toxicity

Renal toxicity

Photosensitivity- Especially **demeclocycline**

Vestibular toxicity- Doxycycline and Minocycline



Chloramphenicol

 Chloramphenicol is an antibiotic useful for the treatment of a number of bacterial infections.

 This includes use as an **eye ointment to treat conjunctivitis.**

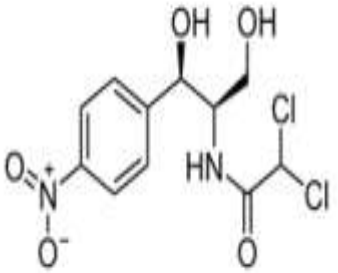
 By mouth or by injection into a vein, it is used to **treat meningitis, plague, cholera, and typhoid fever.**

History

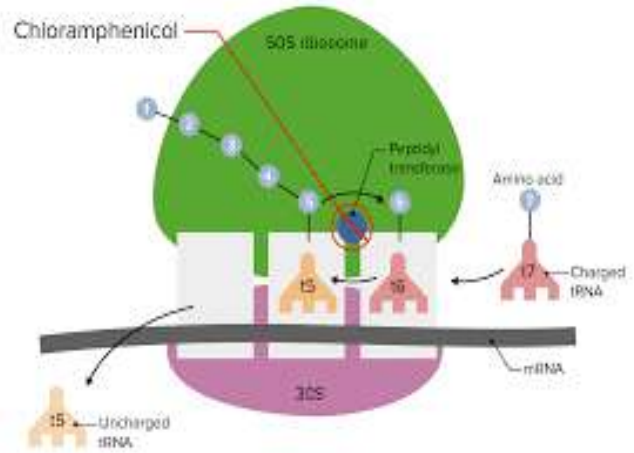
 Chloramphenicol was first isolated from Streptomyces venezuelae in 1947 and in 1949 a team of scientists at **Parke-Davis** including Mildred Rebstock published their identification of the chemical structure and their synthesis, making it the first antibiotic to be made instead of extracted from a microorganism.

 In 2007, the accumulation of reports associating **aplastic anemia and blood dyscrasia with chloramphenicol** eye drops lead to the classification of "probable human carcinogen" according to World Health Organization criteria, based on the known published case reports and the spontaneous reports submitted to the National Registry of Drug-Induced Ocular Side Effects.

Mechanism of Action

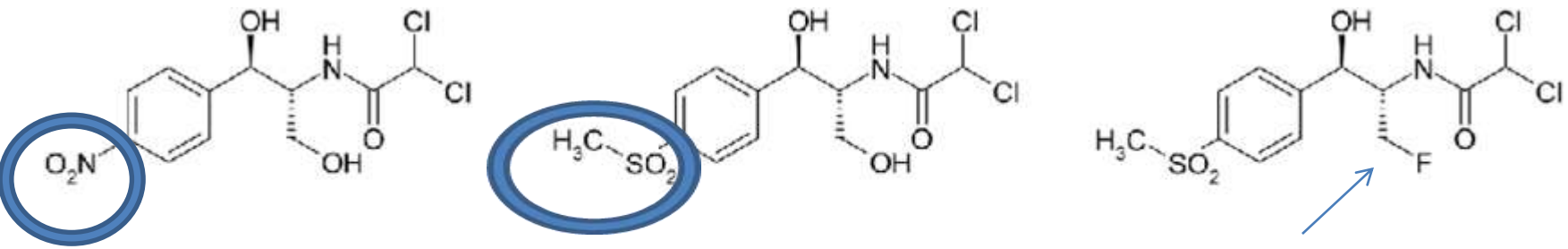


- Chloramphenicol is a bacteriostatic by **inhibiting protein synthesis.**
- It prevents protein chain elongation by inhibiting the peptidyl transferase activity of the bacterial ribosome.
- It specifically binds to A2451 and A2452 residues in the **23s rRNA of the 50s ribosomal** subunit, preventing peptide bond formation.
- Chloramphenicol directly interferes with substrate binding in the ribosome, as compared to macrolides, which sterically block the progression of the growing peptide.



Uses

- Chloramphenicol is an antibiotic.
- It's mainly used to treat eye infections (such as conjunctivitis) and sometimes ear infections.
- Chloramphenicol comes as eye drops or eye ointment



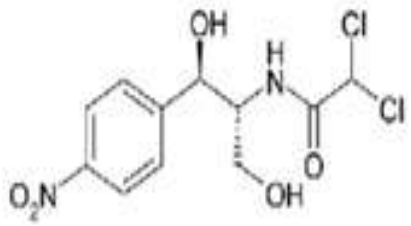
Chloramphenicol (CAP)

Thiamphenicol (TAP)

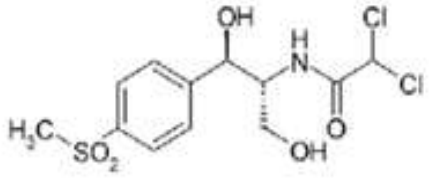
Florfenicol (FF)

General information

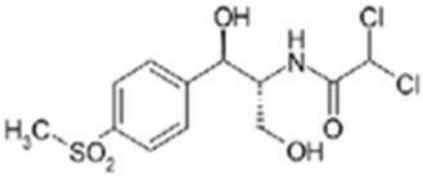
Thiamphenicol is a semisynthetic derivative of chloramphenicol, It differs in that the **NO2 group in the para position is replaced by a methylsulfonyl group**. The substitution in the para position of the molecule does not influence its effects on either protein or DNA synthesis in any recognizable way. The antibacterial spectrum is almost identical to that of chloramphenicol.



The two drugs are used in similar dosages, although there are large differences in their elimination. Glucuronidation is unimportant for thiamphenicol: over 90% of a therapeutic dose is excreted by the kidneys in unchanged form. The corresponding figure for chloramphenicol is only about 10%. **Thus, in contrast to chloramphenicol, the half-life of thiamphenicol is prolonged in patients with reduced renal function in whom accumulation can occur.**

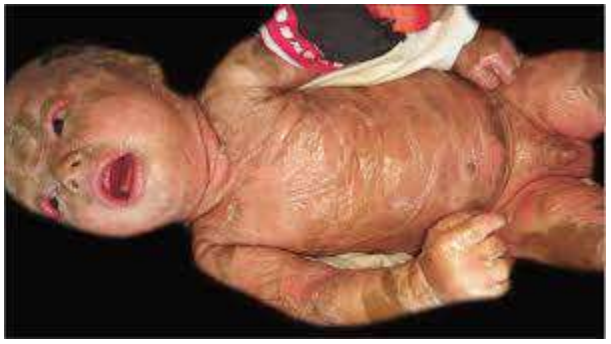


Thiamphenicol

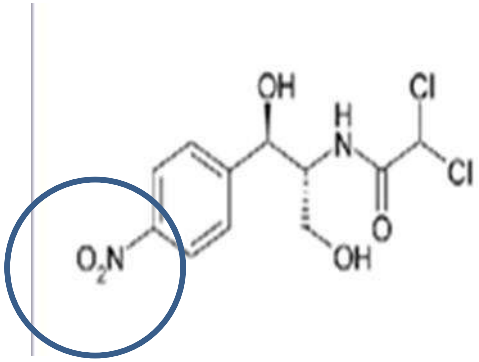


Thiamphenicol

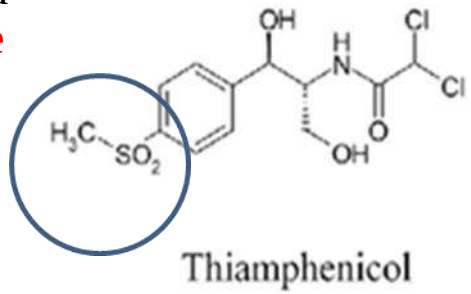
- The most important adverse reactions are **an immediate dose-related and reversible disturbance of erythropoiesis and peripheral neuropathy.** In contrast to chloramphenicol, aplastic anemia and **the gray syndrome** do not seem to occur with thiamphenicol. Thiamphenicol is a chloramphenicol analog with a range of activity similar to chloramphenicol, although it is generally 1–2 times less active. It has equal activity against Haemophilus, Bacteroides fragilis and Streptococcus. It differs pharmacokinetically in that it is not eliminated by **hepatic glucuronidation and is excreted unchanged in urine, so elimination is unaffected by liver disease.** Unlike chloramphenicol, thiamphenicol does not cause aplastic anemia in humans.



Florfenicol is a structural analog of **thiamphenicol** which has greater in vitro activity against pathogenic bacteria than chloramphenicol and thiamphenicol. It is also active against some bacteria that are resistant to chloramphenicol, especially **enteric bacteria**. Florfenicol is not susceptible to inactivation by chloramphenicol transacetylases; thus some organisms that **are resistant to chloramphenicol** through this mechanism are susceptible to florfenicol.

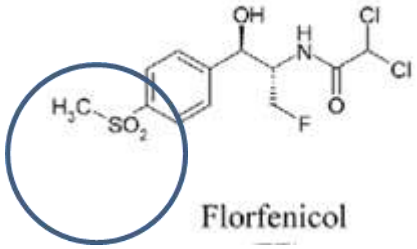


In dogs florfenicol is poorly absorbed after SC administration. It has a half-life of less than 5 h. The drug is well absorbed in cats after PO and IM administrations with a similar elimination half-life. It should not be given IV. Florfenicol can cause dose-related bone marrow suppression but has not been reported to cause fatal aplastic anemia in humans.



Thiamphenicol

Florfenicol shows promise as a replacement for other broad-spectrum antibacterials such as **sulfonamides and tetracyclines** that have been associated with toxicity and residue concerns in food animals. Currently it is approved for use only in cattle, aquaculture and pigs. In cattle it is used to treat infectious conjunctivitis and respiratory disease caused by bacteria **like Pasteurella and Haemophilus**.



Florfenicol

Organic Pharm. Chemistry III

Lecture 8

10th April 2023(15th April)

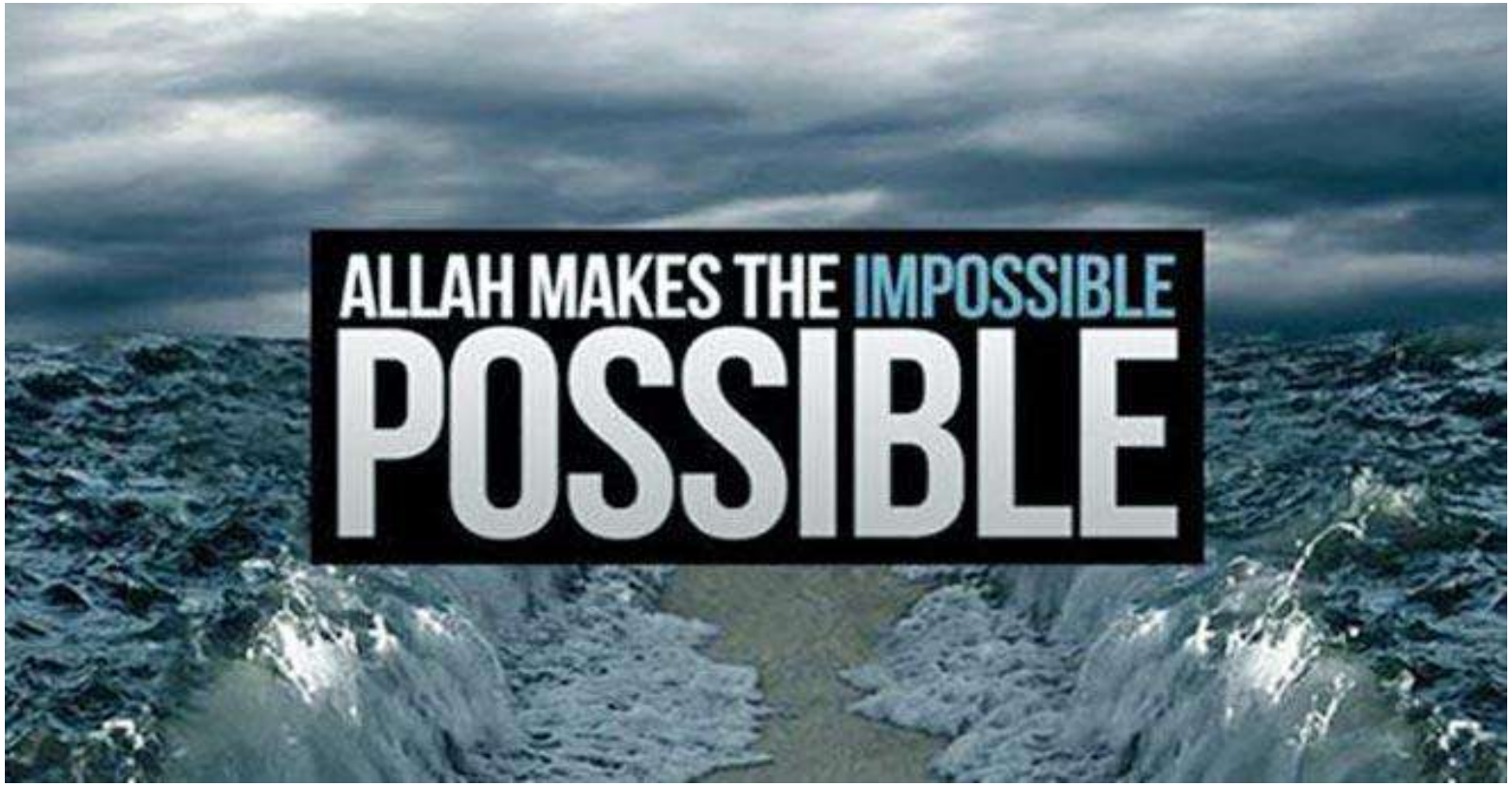
11:30- 2:30

AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023


ALLAH MAKES THE IMPOSSIBLE
POSSIBLE

INTRODUCTION:

Cancer: group of diseases characterized by uncontrolled growth and spread of abnormal cells that left untreated may lead to death.

Neoplesia: uncontrolled growth of new tissue the product of which is known as tumor & these tumors may be either malignant or benign.

Malignant tumors have the capability of invading surrounding tissues and moving to distant location in the body in process called metastasis that characteristic benign tumors does not posses.

Cancer occurs after normal cells have been transformed into neoplastic cells through alteration of their genetic material and the abnormal expression of certain genes. Neoplastic cells usually exhibit chromosomal abnormalities and the loss of their differentiated properties. These changes lead to uncontrolled cell division and many result in the invasion of previously unaffected organs, a process called metastasis.

Antineoplastic agents, cytotoxics or cytostatics are chemotherapeutic agents used for the treatment of cancer.

The goal of cancer chemotherapy is the selective destruction of malignant tumor cells while sparing normal host cells.

The modern treatment of cancer is being more directed towards **combination chemotherapy, in general, after surgery and radiation therapy, which are usually the initial treatments for most solid tumors**.

Cancer or tumor is neoplasm (from the Greek, new and formation), which means “a relatively autonomous growth of tissue”.

Cytostatics; Anticancer Agents

Cancer is believed to be a cause of several agents;

- certain chemical compounds,
- radiant energy (including even sun rays), certain viruses,
- pollution agents (water, air and food),
- alimentary deficiency, hereditary factors,
- and cellular mutation of unknown origin.
- Recently, oncogenes are also involved.

According to their localization and shape, malignant tumors receive different names:

- carcinoma (glandular tissue)
- sarcoma (connective tissue)
- Lymphoma (lymphatic ganglia)
- Leukemia (blood cells).

Advances in Cancer Chemotherapy

Treatment options of cancer:

Surgery: before 1955

Radiotherapy: 1955~1965

Chemotherapy: after 1965

Immunotherapy and Gene therapy

The Basic Concept of Cell Generation Cycle

The cycle of cell replication includes:

M（Mitosis）phase

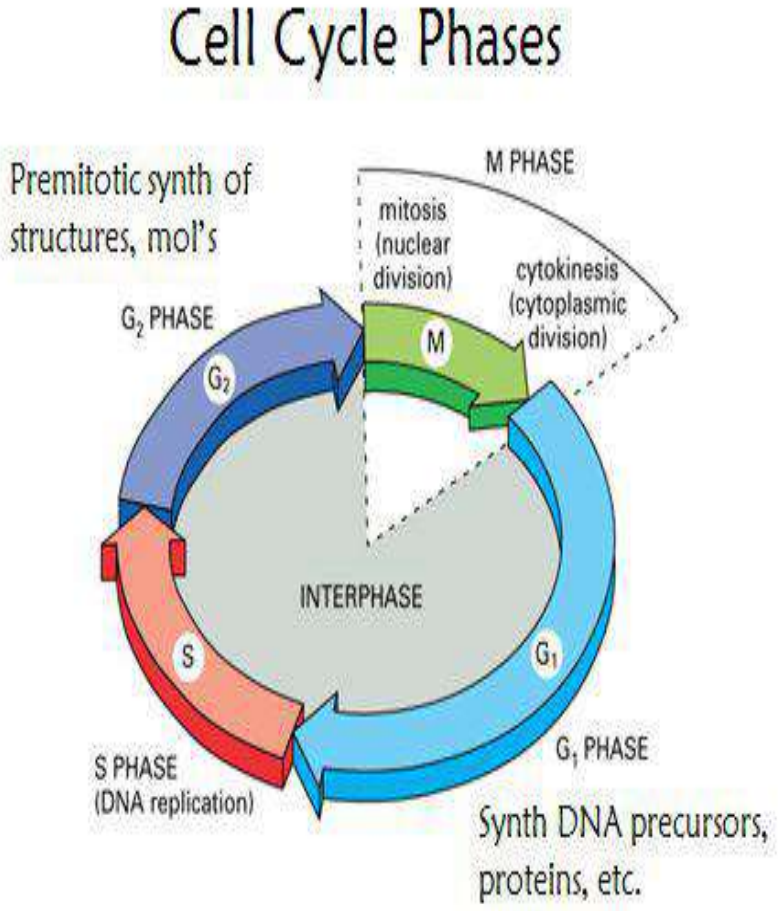
G1（Gap1, period before S）phase

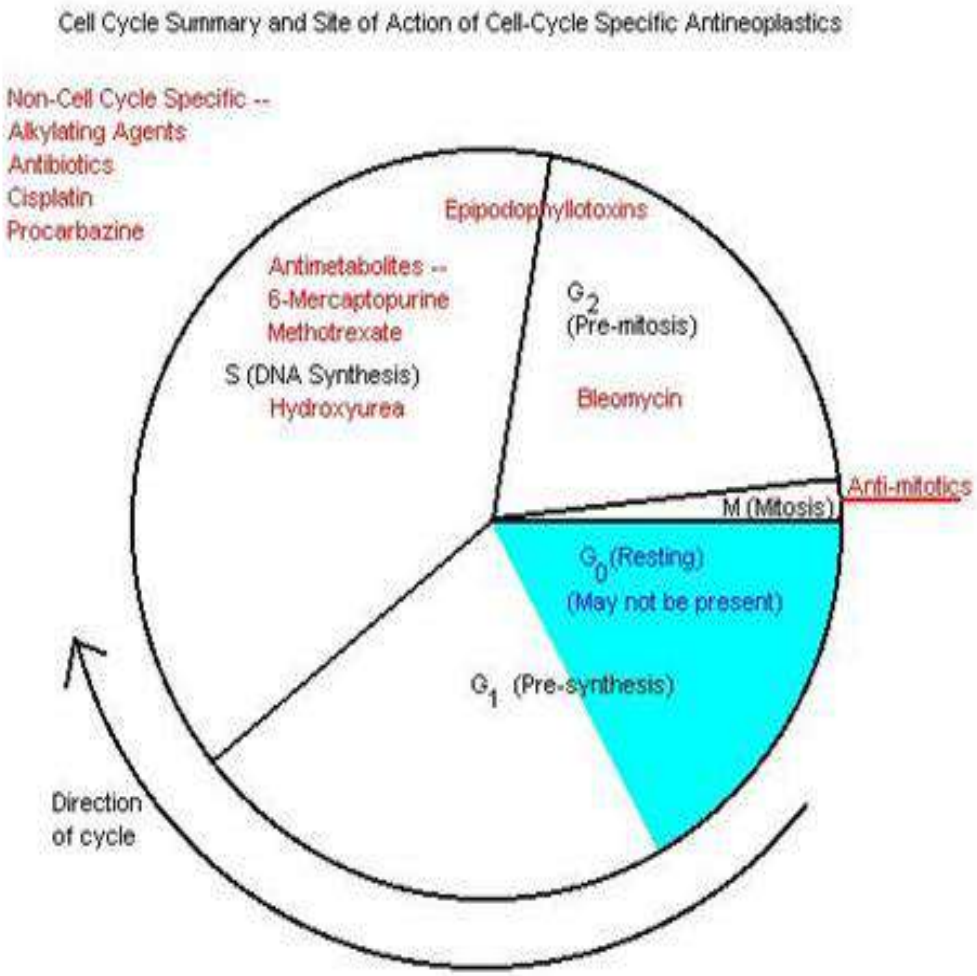
S（DNA synthesis）phase

G2（Gap2,period after S）phase

Growth Fraction (GF）

Cell cycle Phases
Site of Action of cell- cycle specific Antineoplastics





Anticancer Drugs

Affect cell division

Active on rapidly dividing cells

Most effective during S phase of cell cycle

Many cause DNA damage

Damage DNA → **initiate apoptosis**

Side effects greatest in other rapidly-dividing cells

Bone marrow toxicity

Impaired wound healing

Hair follicle damage

GIT epithelial damage

May themselves be carcinogenic

Classification of Anticancer:

Cell Cycle Nonspecific Agents (CCNSA)

drugs that are active throughout the cell cycle

- Alkylating Agents
- Platinum Compounds
- Antibiotics

Cell Cycle Specific Agents (CCSA)

drugs that act during a specific phase of the cell cycle

S Phase Specific Drug:

A.Antimetabolites, Topoisomerase Inhabitors

M Phase Specific Drug:

Vinca Alkaloids, Taxanes

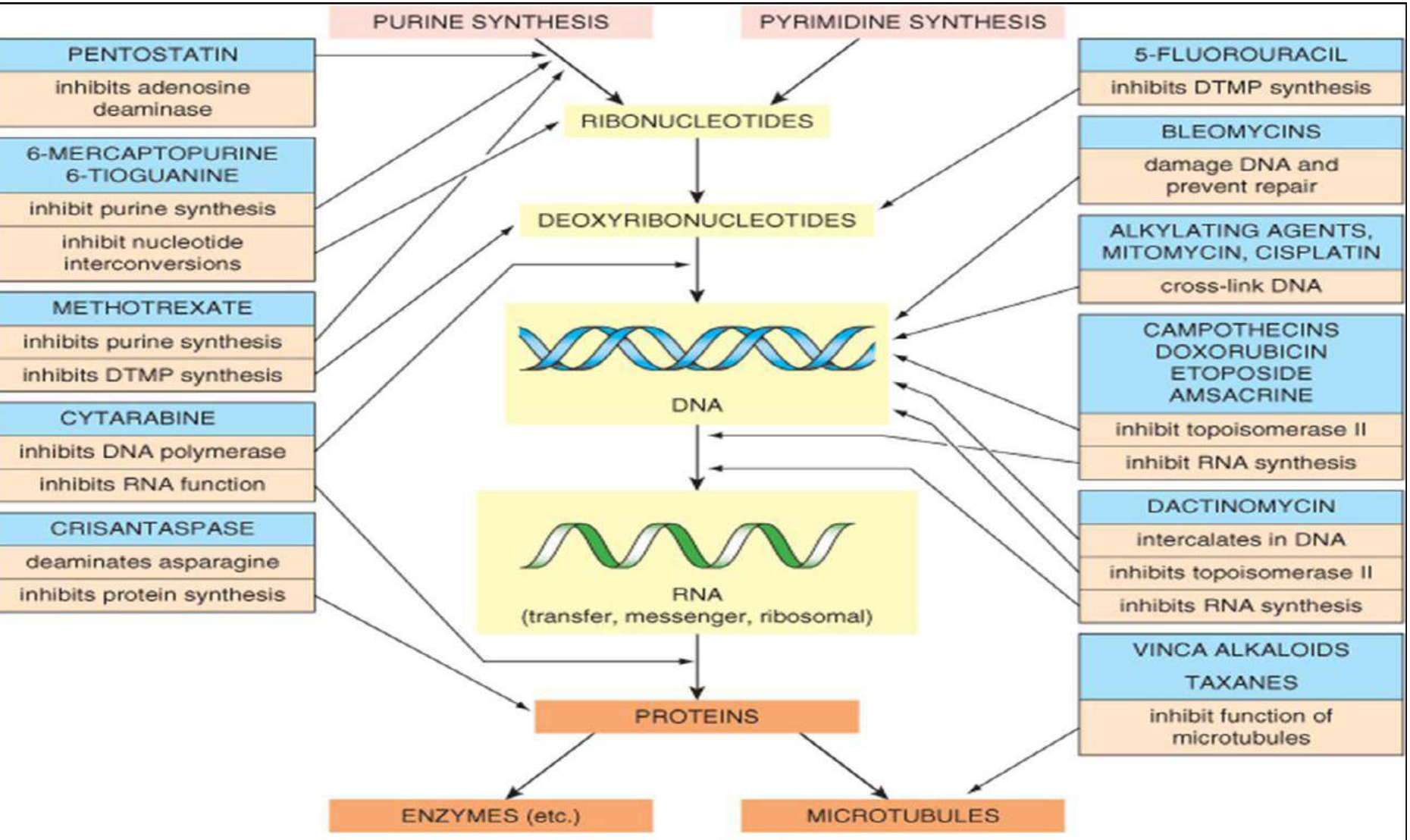
G2 Phase Specific Drug:

B.bleomycin

Classification of anticancer agents:

- Alkylating agents.
- Antimetabolites.
- Natural products.
- Sex hormones and analogues.
- Miscellaneous cytostatics.
- Radioisotopes.
- Immunosuppressive and immunostimulant drugs.

Site of action

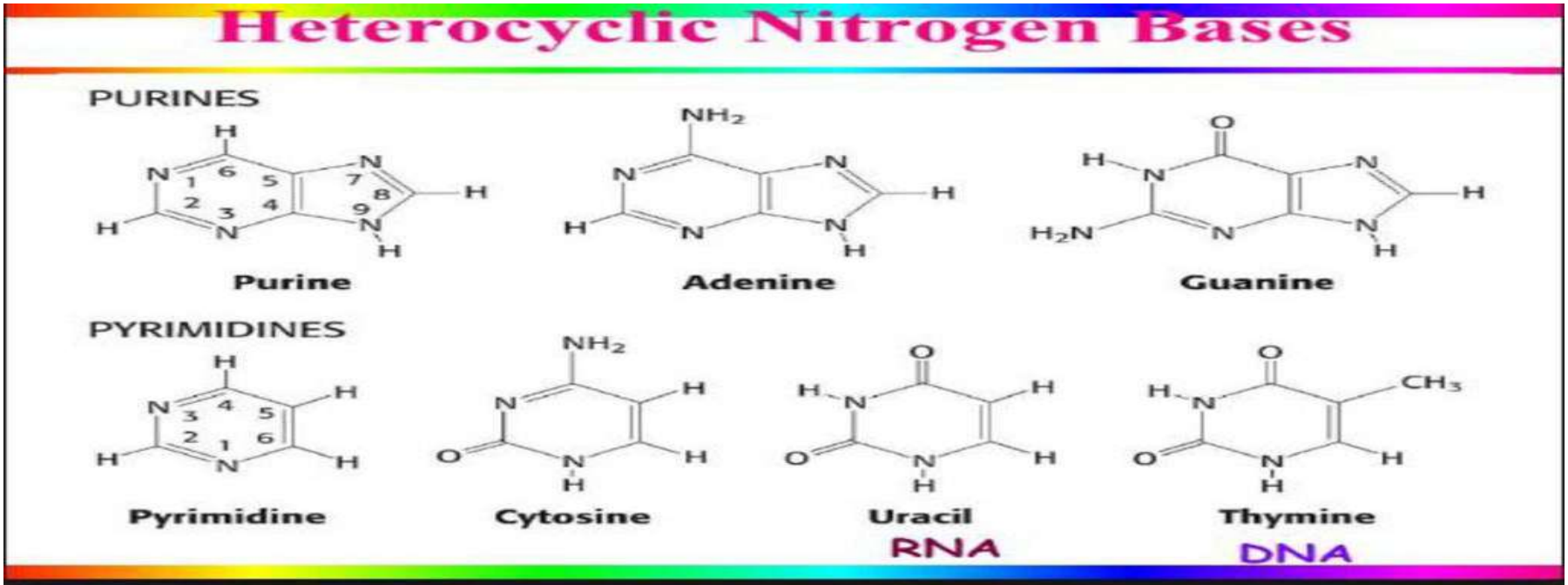


The alkylating agents

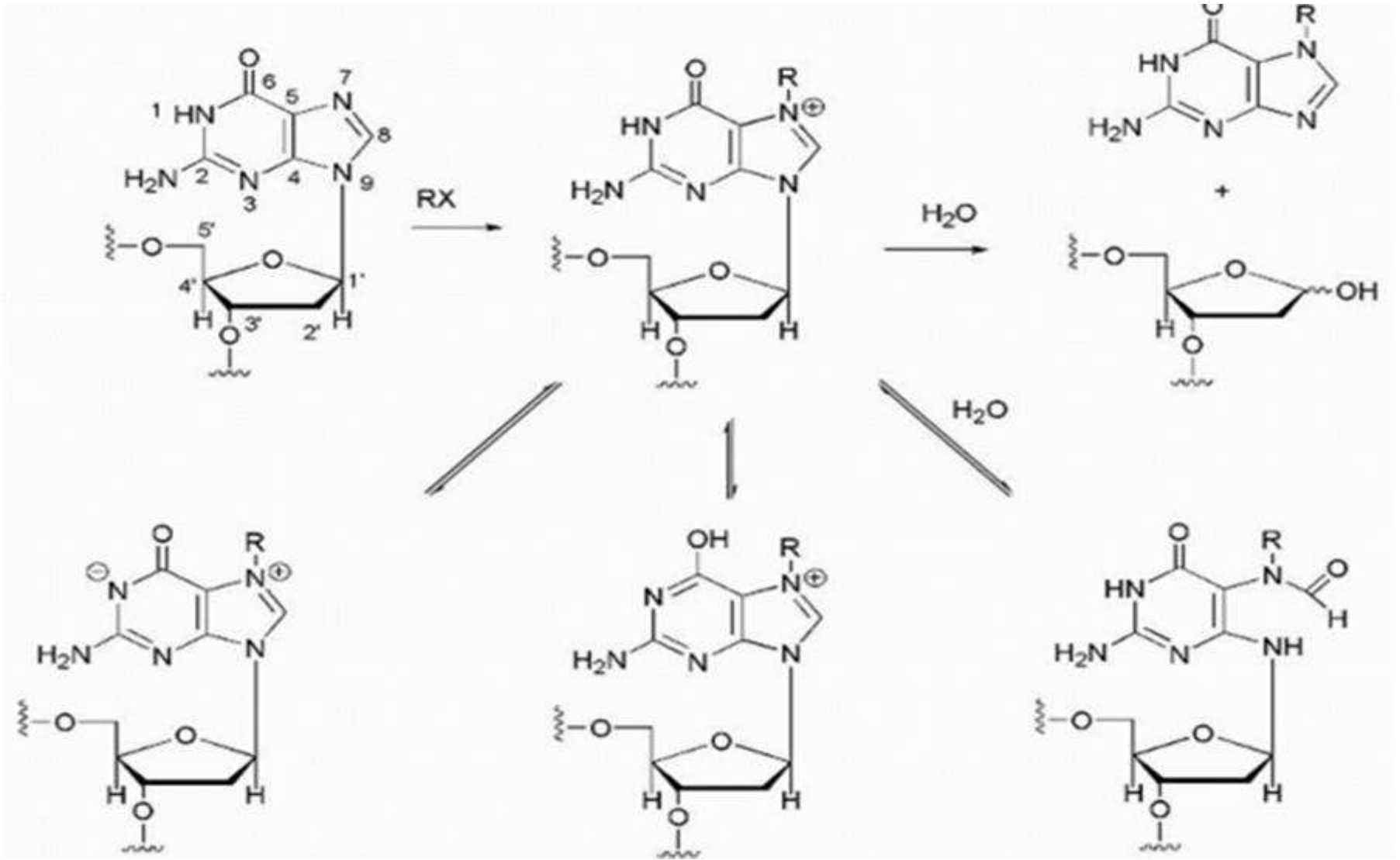
The alkylating agents are a class of drugs that are capable of forming covalent bonds with important biomolecules.

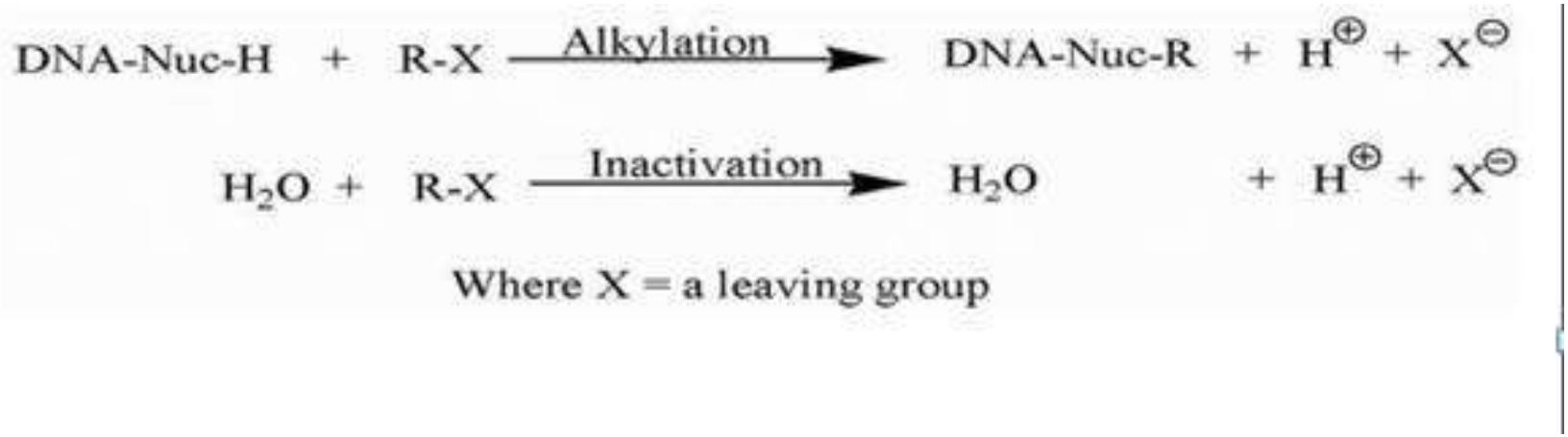
There are several potential nucleophilic sites on DNA, which are susceptible to electrophilic attack by an alkylating agent (N-2, N-3, and N-7 of guanine, N-1, N-3, and N-7 of adenine, 0-6 of thymine, N-3 of cytosine).

The most important of these for many alkylating **agents is the N-7 position of guanine** whose nucleophilicity may be enhanced by adjacent guanine residues. Alkylation converts the base to an effective leaving group so that attack by water leads to depurination and the loss of genetic information if the resulting depurination is not repaired by the cell (Scheme



Alkylation of 7- Guanine and subsequent depurination of DNA

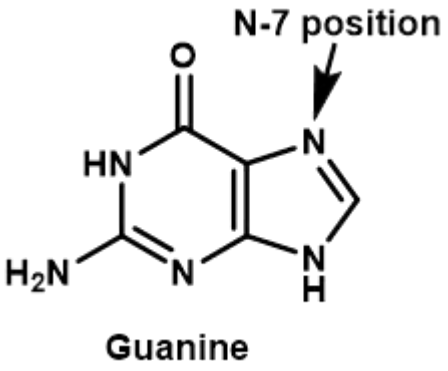




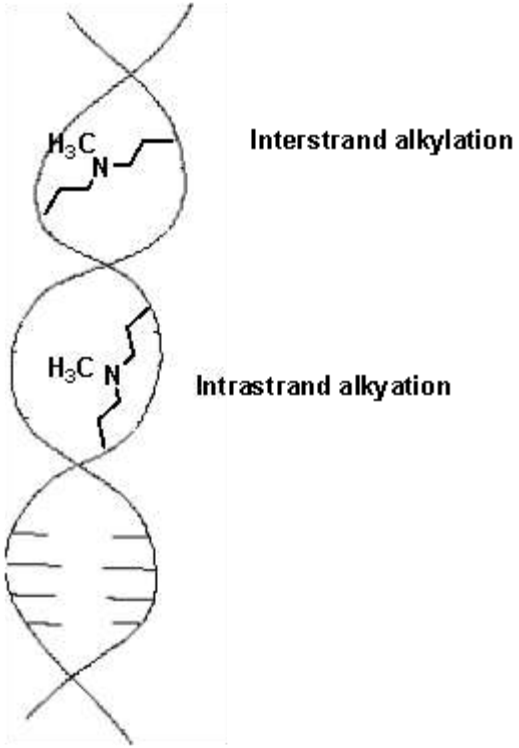
The general mechanism for alkylation involves nucleophilic attack by —N=, —NH2, —OH, —O—PO3H of DNA and RNA, while additional nucleophiles (—SH, COOH, etc.) present on proteins may also react (Scheme).

Anion formation increases the reactivity of the nucleophile compared with the un-ionized form (—O- is more nucleophilic than OH). Reaction with water is also possible, because it represents the nucleophile in greatest abundance in the body and this becomes more likely as the electrophile becomes more reactive.

Reaction involves displacement of a leaving group on the electrophile by the nucleophile. The reactivity of the electrophile is dependent in part on the ability of the leaving group to stabilize a negative charge.



Guanine



The N-7 position of guanine in DNA is strongly nucleophilic. Reaction orders depend on the structure of the alkylating agent. Other base positions of DNA attacked by alkylating agents are

- **N-2 & N-3 of guanine**
- **N-3 & N-1 & N-7 of adenine**
- **O-6 of thymine and N-3 of cytosine.**
- **Phosphate Oxygen of DNA is also alkylated to an appreciable extent**.

Alkylating agents are also; interact with enzymes and other proteins.

Alkylating agents could form inter- and intra-strand linkage in DNA.

Nitrogen mustards

Mustard gas

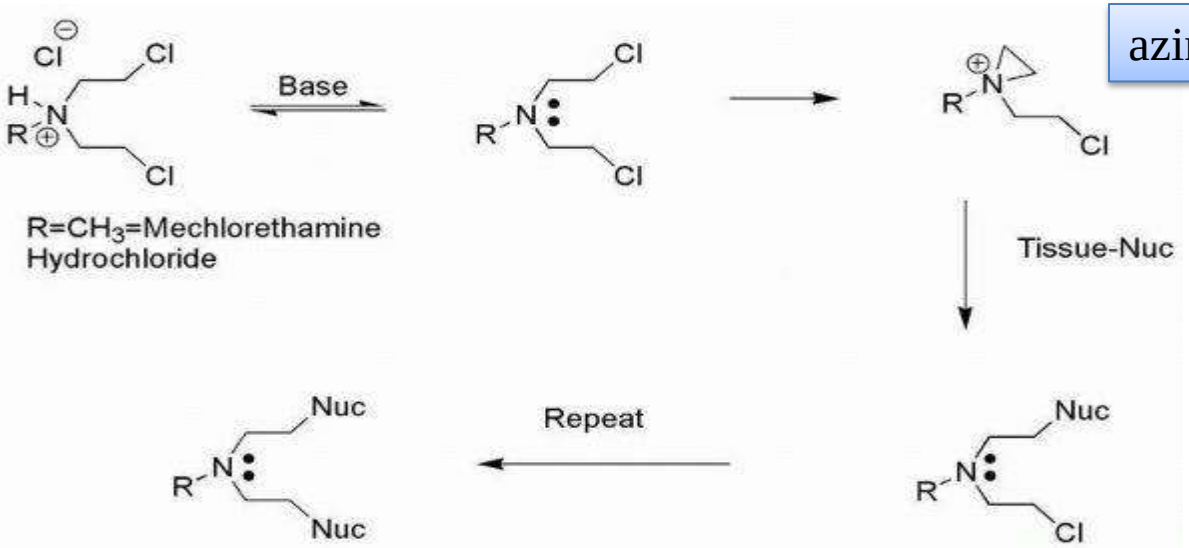
The nitrogen mustards are compounds that are chemically similar to sulfur mustard or mustard gas developed and used in World War I.

Investigation of sulfur mustard revealed that it possessed antineoplastic properties but because the compound existed as a gas at room temperature.Conversion of the sulfide to a tertiary amine allowed for the formation of salts, which exist as solids at room temperature allowing for easier handling and dosing.

Chlorambucil, melphalan, cyclophosphamide and ifosfamide.

The lack of selectivity of mechlorethamine led to attempts to improve on the agent. One rationale was to reduce the reactivity by reducing the nucleophilicity of nitrogen, thereby slowing aziridinium cation formation. This could be accomplished by replacement of the weakly electron-donating methyl group with groups that were electron withdrawing (-I). This is seen in the case of **chlorambucil and melphalan by attachment of nitrogen to a phenyl ring (Fig.)**

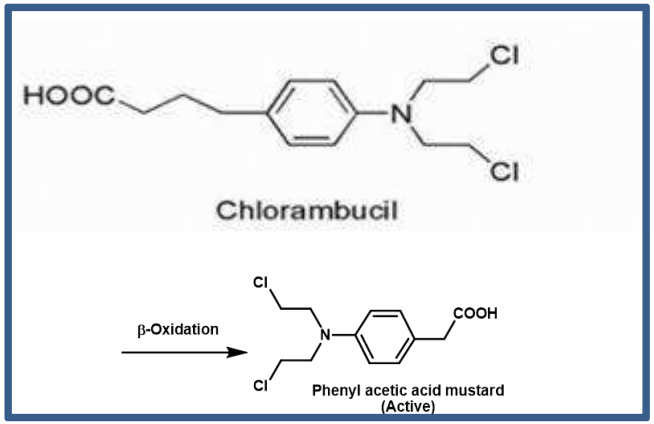
aziridinium cation

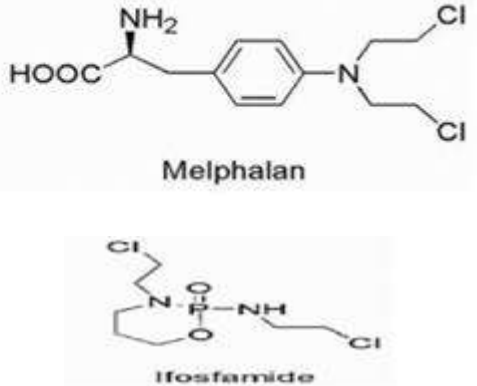


Attachment of more highly electron-withdrawing

functionalities was utilized in the case of **cyclophosphamide and ifosfamide**

In these cases, aziridinium cation formation is not possible until the electron-withdrawing function has been altered.

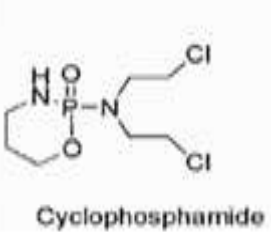




Chlorambucil is the slowest acting and least toxic of any nitrogen mustard.

Used for chronic lymphocytic leukemia.

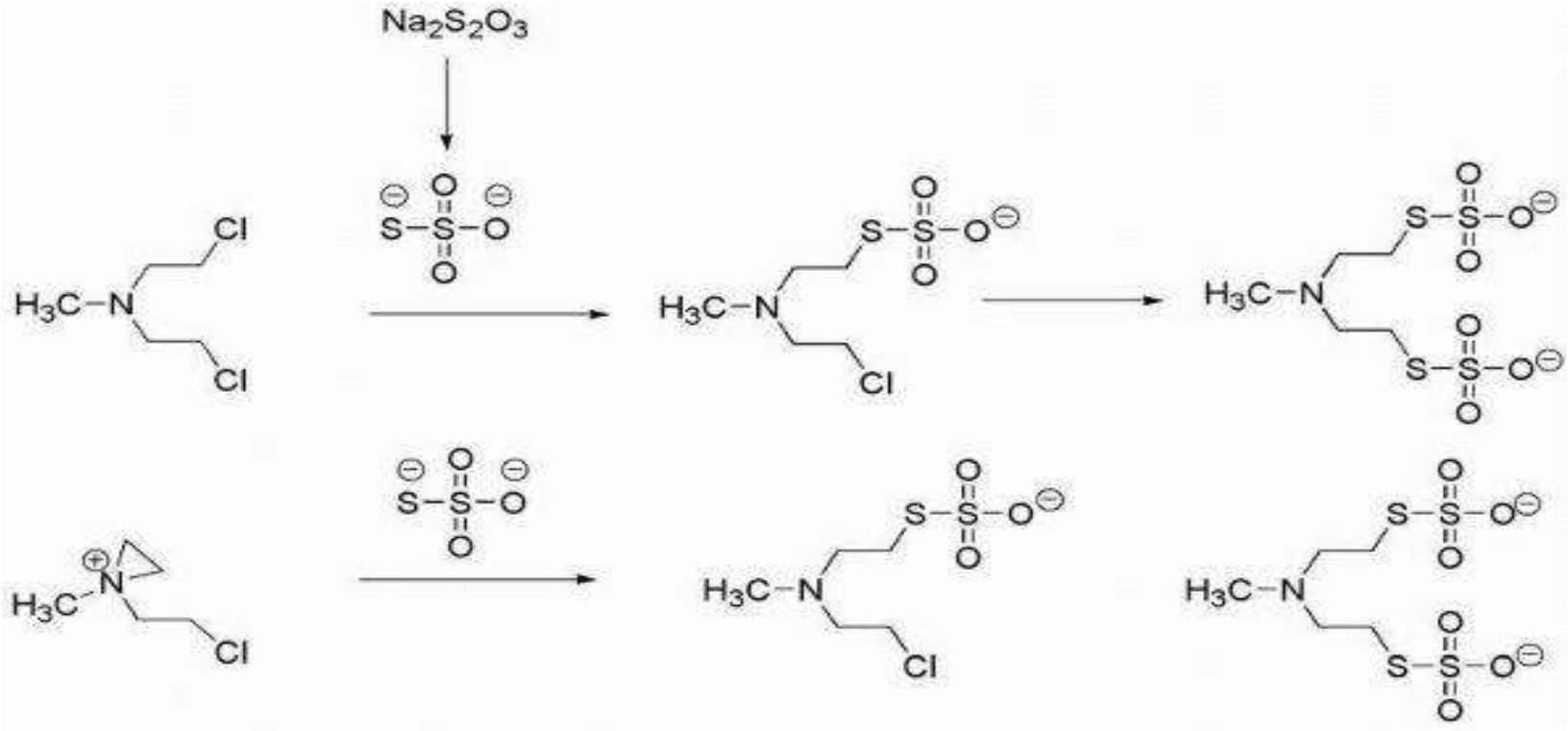
The butonic acid moiety is subjected to β-oxidation to the active phenylacetic acid mustard



Cyclophosphamide is not active until it is transformed by metabolic process to the active form by hepatic liver Cytochrome P-450 and thus, it could be classified as a bioprecursor prodrug . The metabolic 4-hydroxy derivative is first formed and undergoes the following modifications:

Mechlorethamine is highly reactive, in fact, too reactive and therefore nonselective, making it unsuitable for oral administration and necessitating direct injection into the tumor.

In cases of extravasation (drug escapes from the tumor into the underlying tissue), the antidote sodium thiosulfate (Na2S2O3), a strong nucleophile, may be administered.

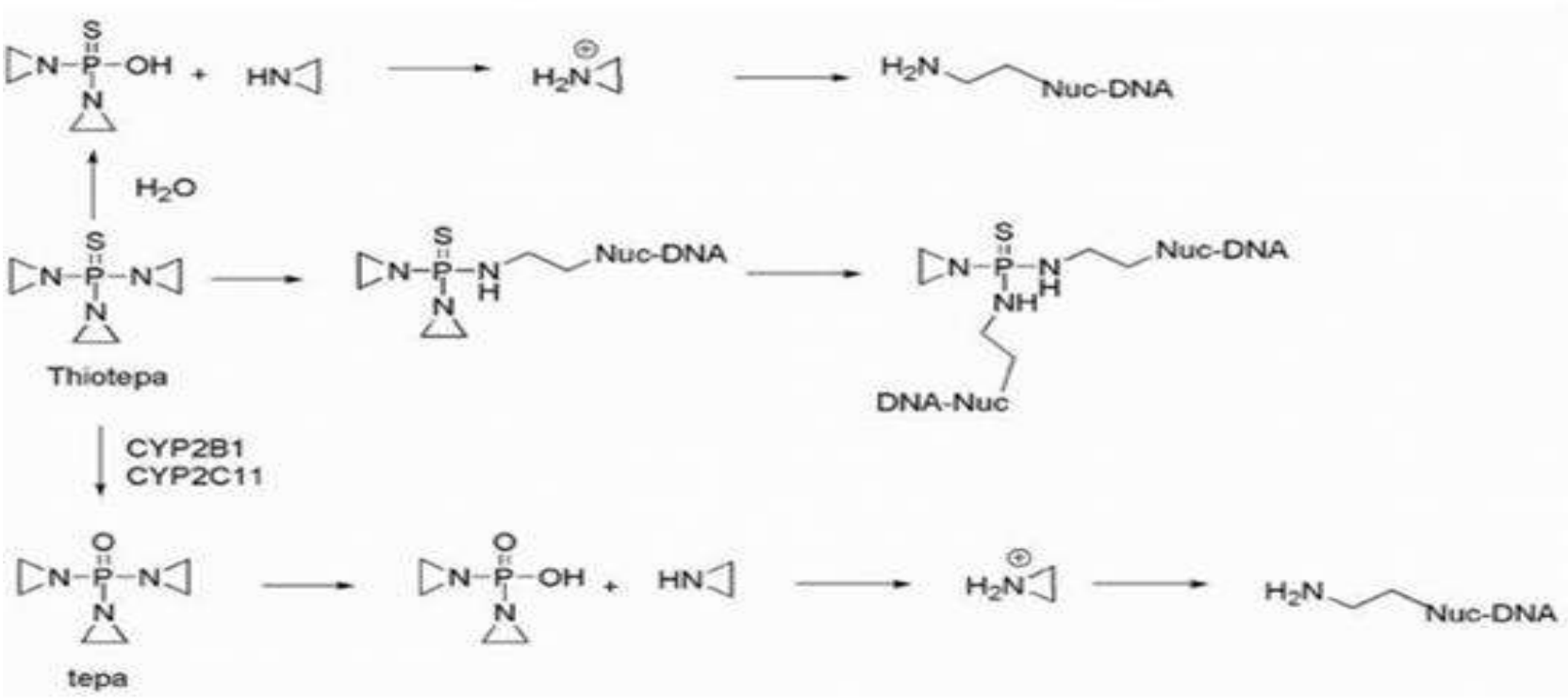


THIOTEPA

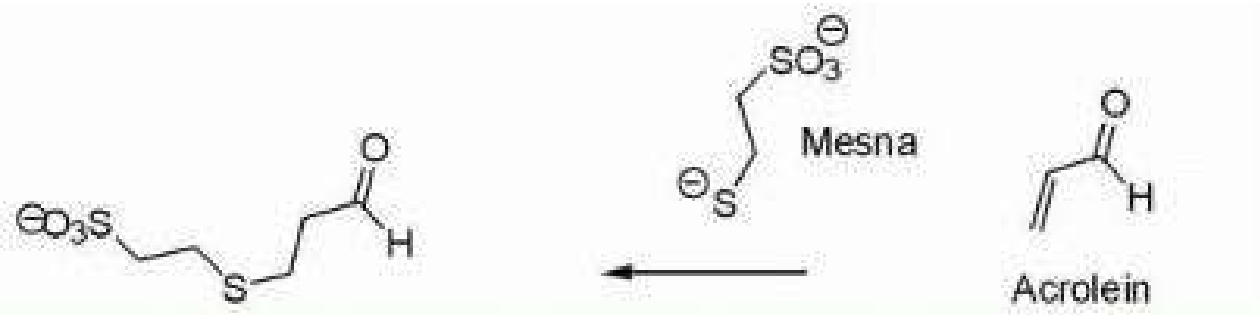
Thiotepa containing the thiophosphoramide functionality was found to be more stable than the oxa-analog (TEPA) but is metabolically converted to TEPA by desulfuration in vivo.

Thiotepa incorporates a less reactive aziridine ring compared with that formed in mechlorethamine. The adjacent thiophosphoryl is electron withdrawing and, therefore, reduces the reactivity of the aziridine ring system.

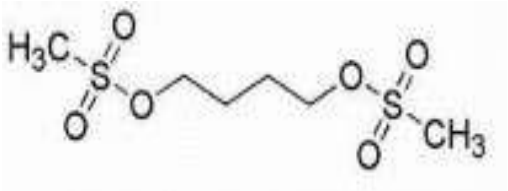
The conclusion that aziridine is the active alkylating agent once thiotepa has been converted to TEPA is based on the fact that when TEPA is incubated with DNA, no cross links are formed and only mono adducts are generated. The reactivity of aziridine generated by either route may be somewhat enhanced within cancer cells, **where the pH is normally reduced 0.2 to 0.4 pH** units resulting in an increase in reactivity toward nucleophilic attack.



Acrolein is also formed as a result of this process, which may itself act as an electrophile that has been associated with bladder toxicity. To decrease the incidence of kidney and bladder toxicity, the sulfhydryl (—SH) containing agent mesna may be administered and functions to react with the electrophilic species that may be present in the kidney

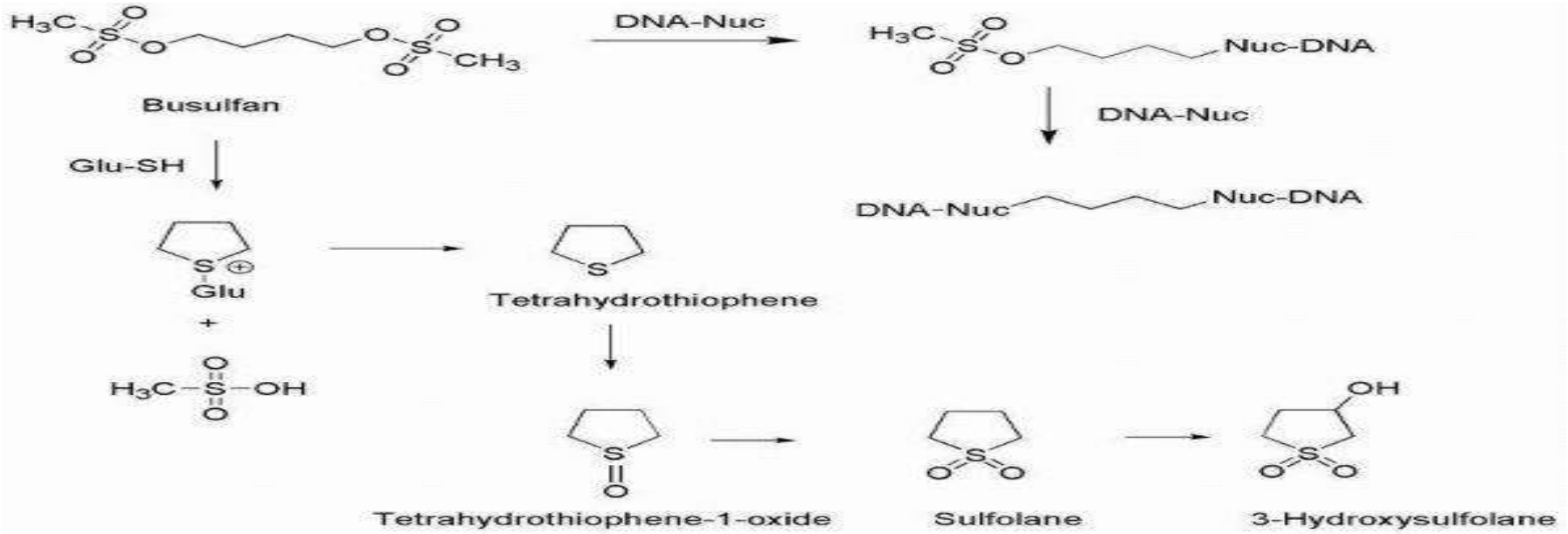


BUSULFAN



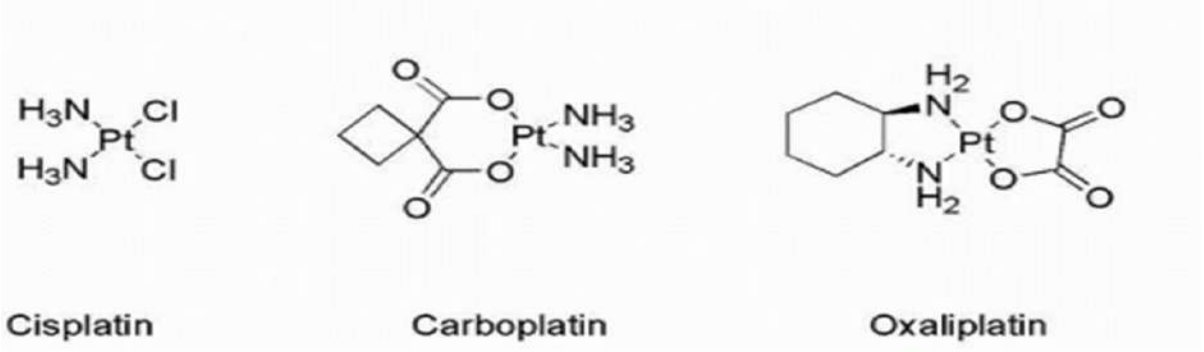
Busulfan utilizes two sulfonate functionalities as leaving groups separated by a four-carbon chain that reacts with DNA to primarily form intrastrand cross-link at 5′-GA-3′ sequences. The sulfonates are also subject to displacement by the sulfhydryl functions found in cysteine and glutathione, and metabolic products are formed as a result of nucleophilic attack by these groups to generate sulfonium species along with methane sulfonic acid.

This is followed by conversion to tetrahydrothiophene, and further oxidation products are subsequently produced to give the sulfoxide and sulfone. The cyclic sulfone known as sulfolane may be further oxidized to give 3-hydroxysulfolane



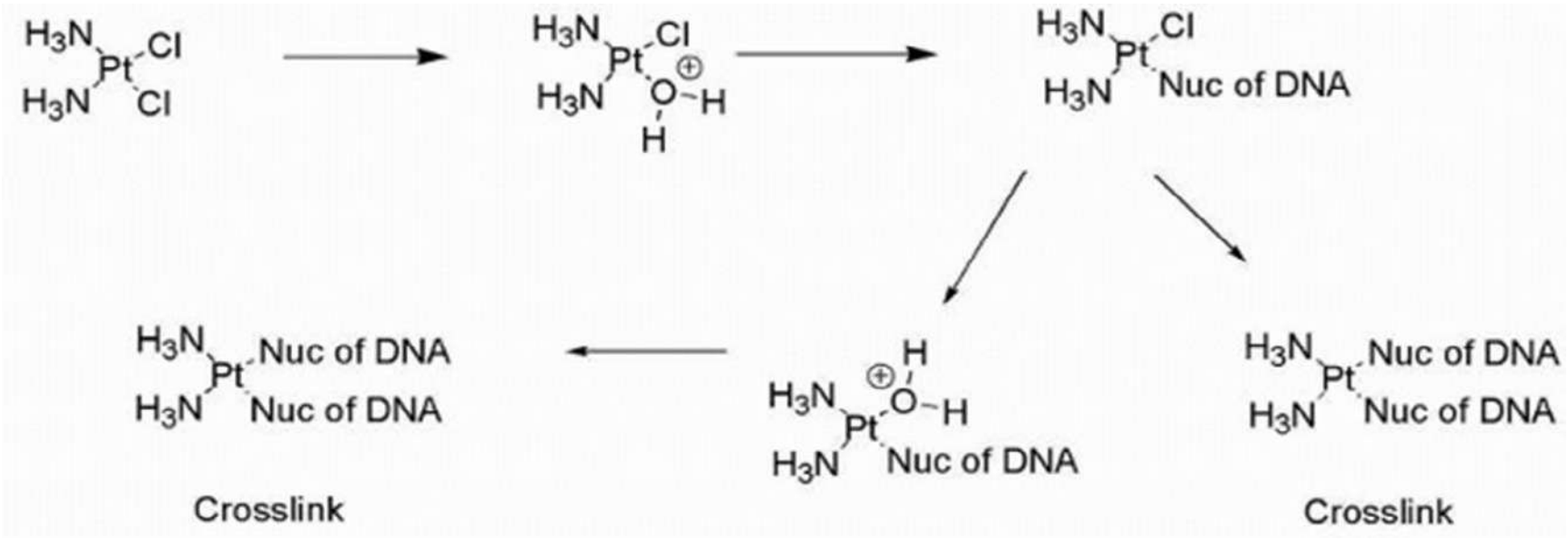
ORGANOPLATINUM COMPOUNDS

Compounds based on platinum that play a central role in many cancer treatment protocols.



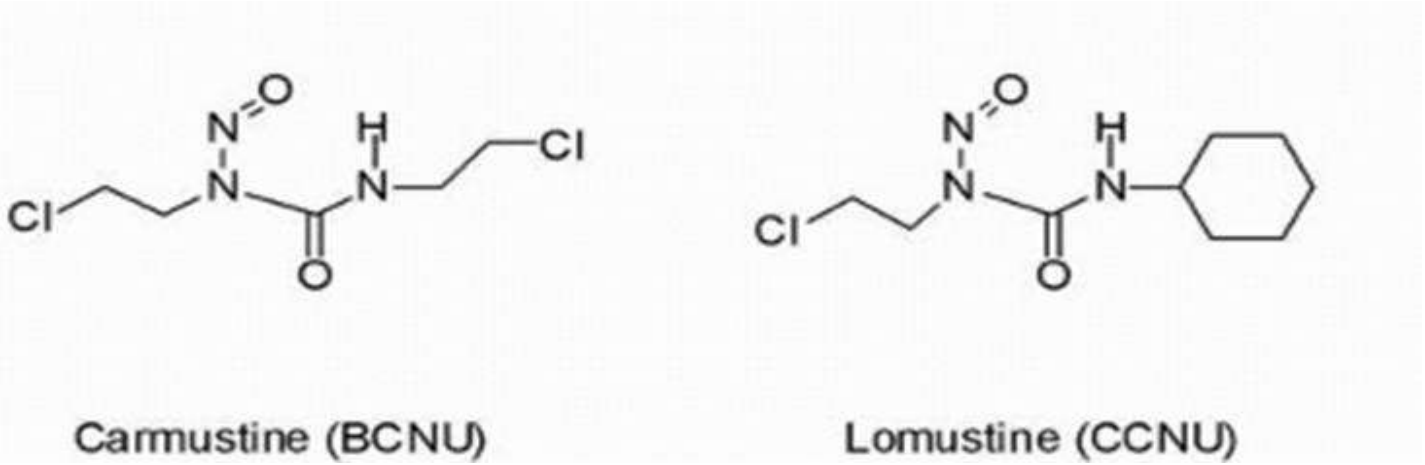
The dichloro species is maintained in the blood stream as a result of the relatively high chloride concentration . Movement into the tumor cells is accomplished by passive diffusion or carrier-mediated transport. Once inside the tumor cell, the drug encounters a lower chloride concentration and one chloro group is substituted by a water molecule in a process known as aquation. This serves to “trap” the molecule in the cell as a result of ionization. Reaction with DNA occurs preferentially at the N-7 of guanine of two adjacent guanine residues resulting in primarily (95%) intrastrand cross-links

Platinum (II) is considered to be a “soft” electrophile and as a result, its complexes are subject to attack by “soft” nucleophiles such as thiol groups found on proteins. This can result in significant protein binding (88%-95%) and inactivation caused by the presence of thiols in albumin, glutathione, and other proteins.



Cisplatin administration is also associated with significant nephrotoxicity and neurotoxicity that is dose limiting. These factors lead to the development of less reactive platinum compounds such as carboplatin and oxaliplatin in which the leaving group was incorporated into a chelate.

NITROSOUREAS



General structural requirement

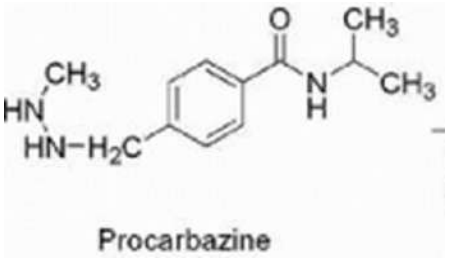
- Urea moiety.
- Nitroso group linked to the urea.
- Electronegative atom with good leaving properties

These compounds are reasonably stable at pH = 4.5 but undergo both acid and base catalyzed decomposition at lower and higher pH, respectively

alkylation of DNA involves abstraction of the NH proton, which is relatively acidic (pKa = 8-9), followed by rearrangement to give an isocyanate and a diazohydroxide.

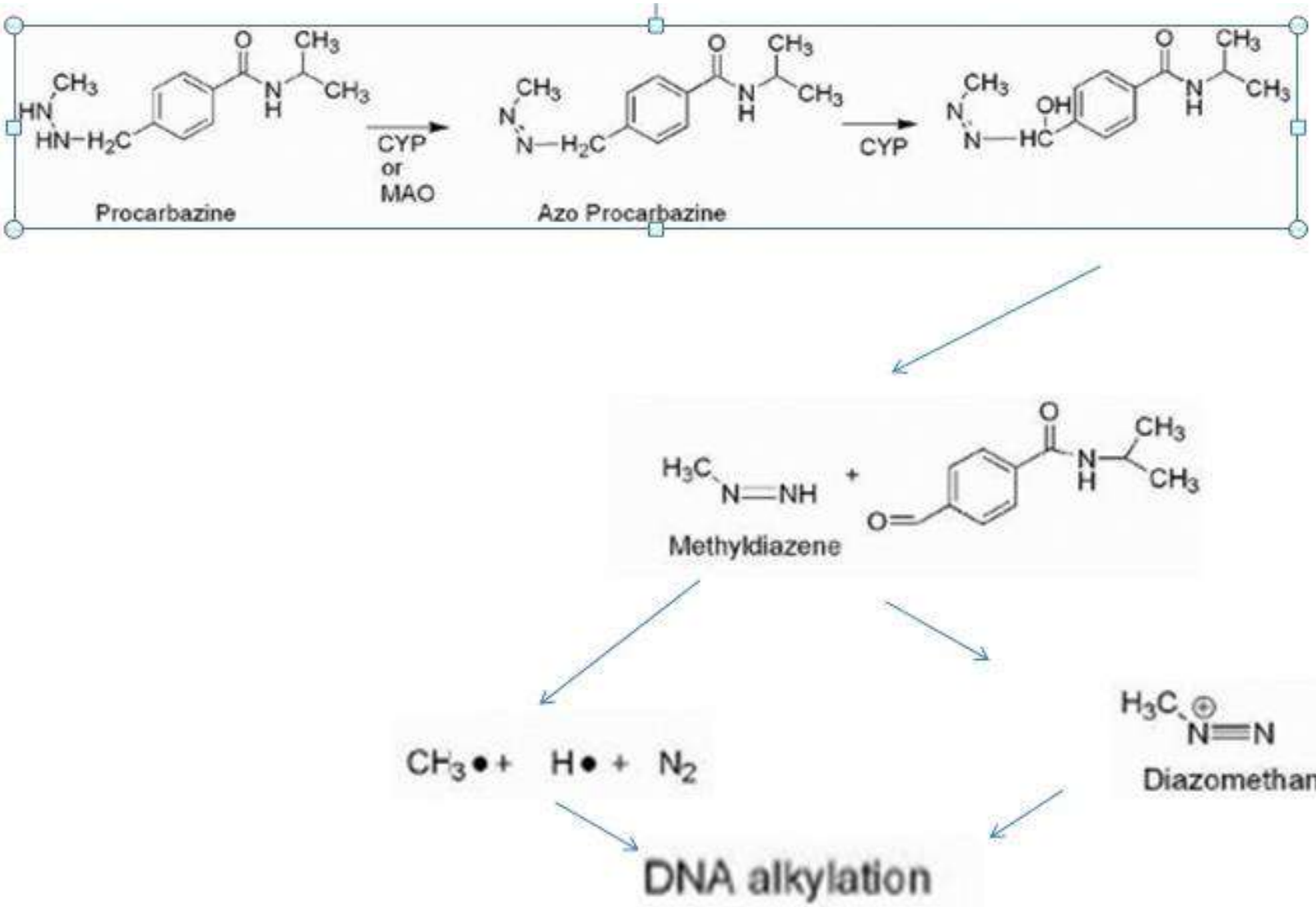
The diazohydroxide, upon protonation followed by loss of water, yields a diazo species that decomposes to a reactive carbocation . The isocyanate functions to carbamylate proteins and RNA, whereas the carbocation is believed to be the agent responsible for DNA alkylation.

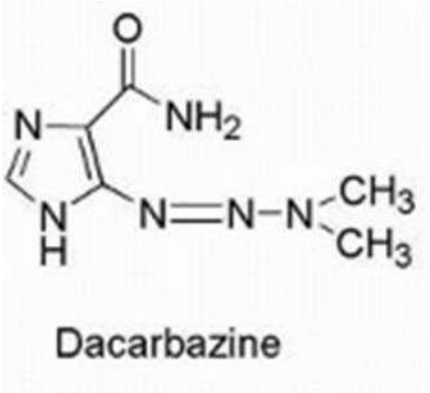
PROCARBAZINE, DACARBAZINE, AND TEMOZOLOMIDE



Procarbazine

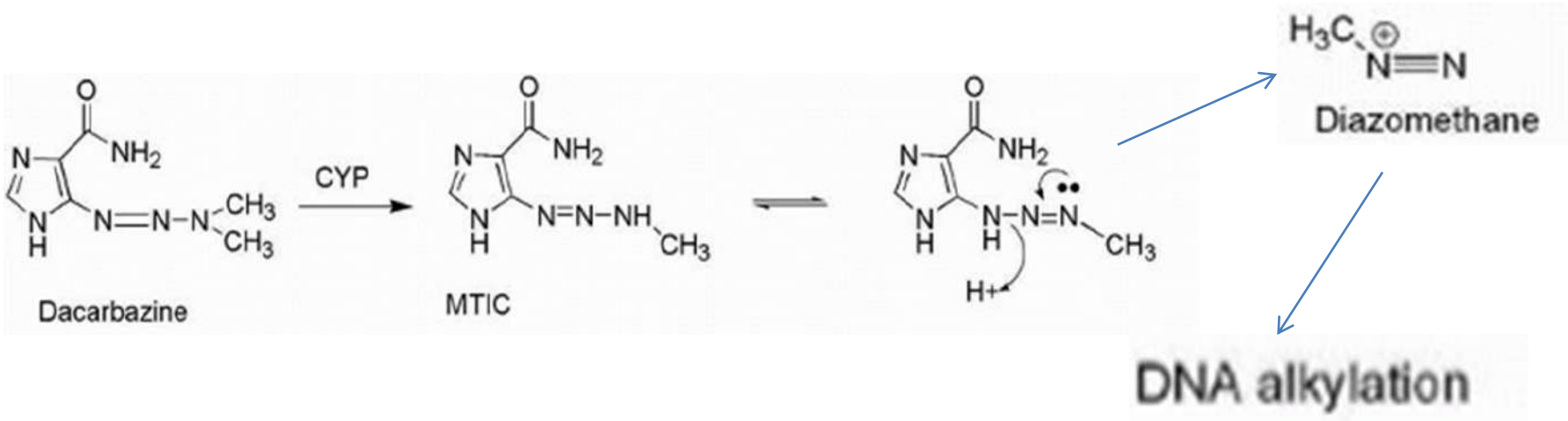
- **Procarbazine**
- oxidation of procarbazine does occur in the **liver and is mediated by CYP** and monoamine oxidase to give azo-procarbazine. This compound may also be generated nonenzymatically in an aerobic environment
- One such route involves CYP-mediated oxidation of the benzylic methylene carbon with subsequent decomposition to give methyldiazine and the aldehyde. The methyldiazine may then decompose by hemolytic bond cleavage to give methyl and hydrogen radicals along with nitrogen gas or be further oxidized to givethe diazo compound, which can decompose to give the methyl carbocation.
- Methylhydrazine covalently bound to the N-7 position of guanine especially on tRNA disrupting its function and preventing protein, RNA, and DNA synthesis.
- There was also a small amount of methylation at the O-6 position of guanine

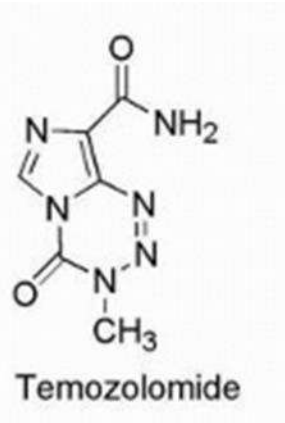

Procarbazine
CYP
or
MAO
Azo Procarbazine
CYP
Methyldiazene
+
$CH_3\bullet$ + $H\bullet$ + N_2
Diazomethane
DNA alkylation



Dacarbazine

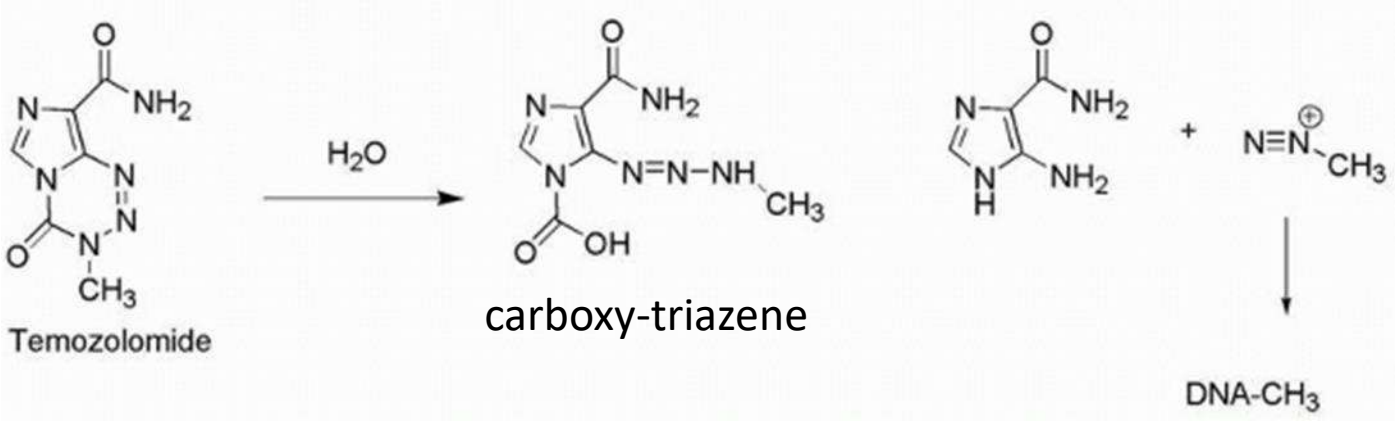
Activation of the agent occurs through the action of CYP to give the demethylated product monomethyl triazeno imidazole carboxamide (MTIC) Tautomerization allows for decomposition to give the aminocarboxamido-imidazole and diazomethane, which is capable of alkylating DNA .Methylation of DNA occurs at N-7, N-3 and O-6 of guanine among other sites





Temozolomide

Hydrolysis of temozolomide gives the carboxy-triazene, which spontaneously loses CO2 to give MTIC. temozolomide may be administered orally.

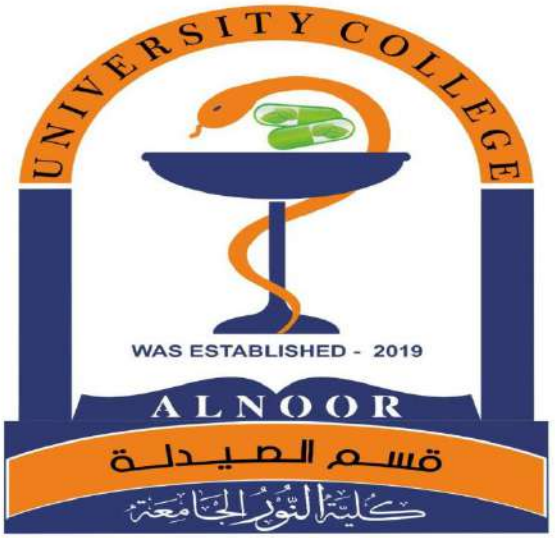


Organic Pharm. Chemistry III

Lecture 9

11th April 2023 (16th April)

11:30- 2:30



AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023

Lectures titles & Credit hours

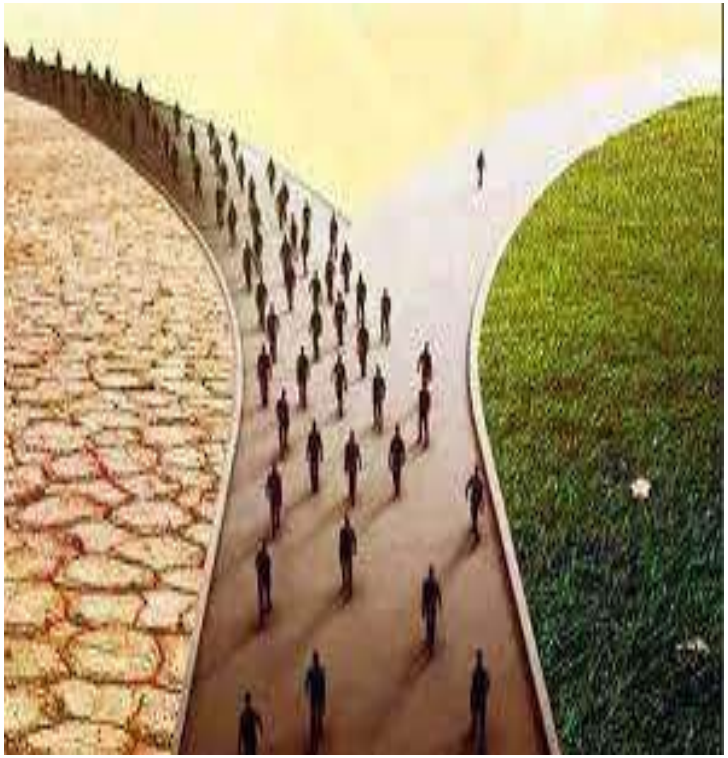
AlNoor University College
Pharmacy Department
Department of Pharmaceutical Chemistry
Title of the course: ***Organic Pharmaceutical Chemistry*** **III** Course number:
Level: 4th Class, 2nd Semester
Credit hours/week : **Theory 3** **Laboratory 1**
Reference text: ***Wilson and Gisvold Textbook of Organic Medicinal and Pharmaceutical Chemistry; Delgado JN, Remers WA, (Eds.); 10th ed., 2004.***

مدرس المادة :- أ.م.د.نهاد عبد الوهاب

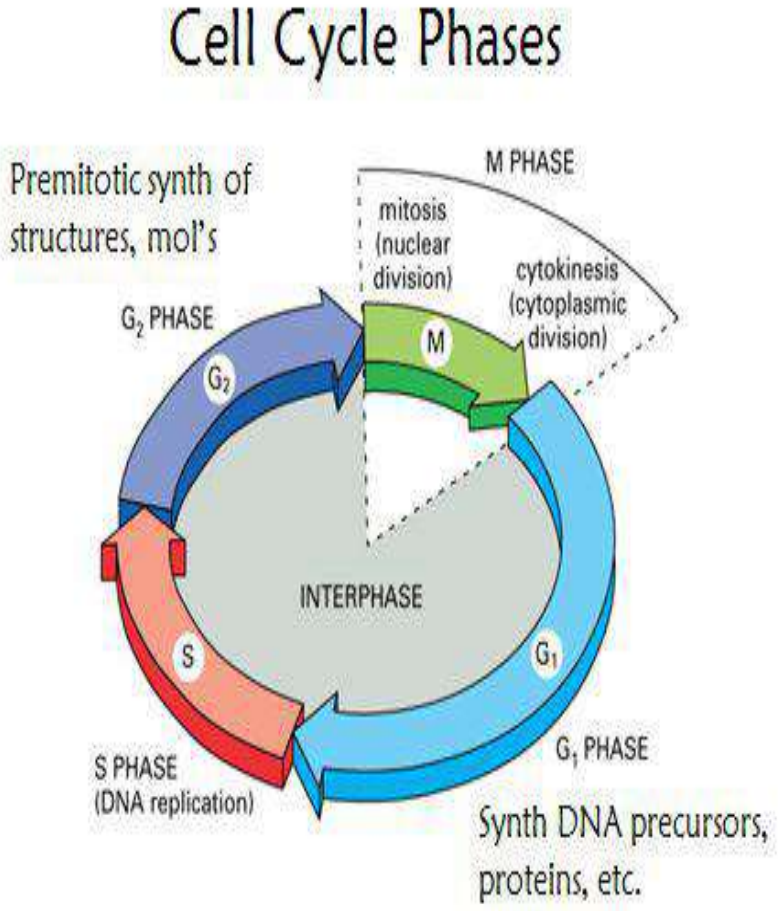
Objectives: To enable understanding mechanisms of drug action, including antibacterial, antifungal and antiviral agents, at molecular level, and the role of medicinal chemistry in the discovery and development of synthetic therapeutic agents. It also enables students to understand the concept of structure-activity relationship and its application in design and synthesis of new chemotherapeutic agents and hormone derivatives with potential biological activity.

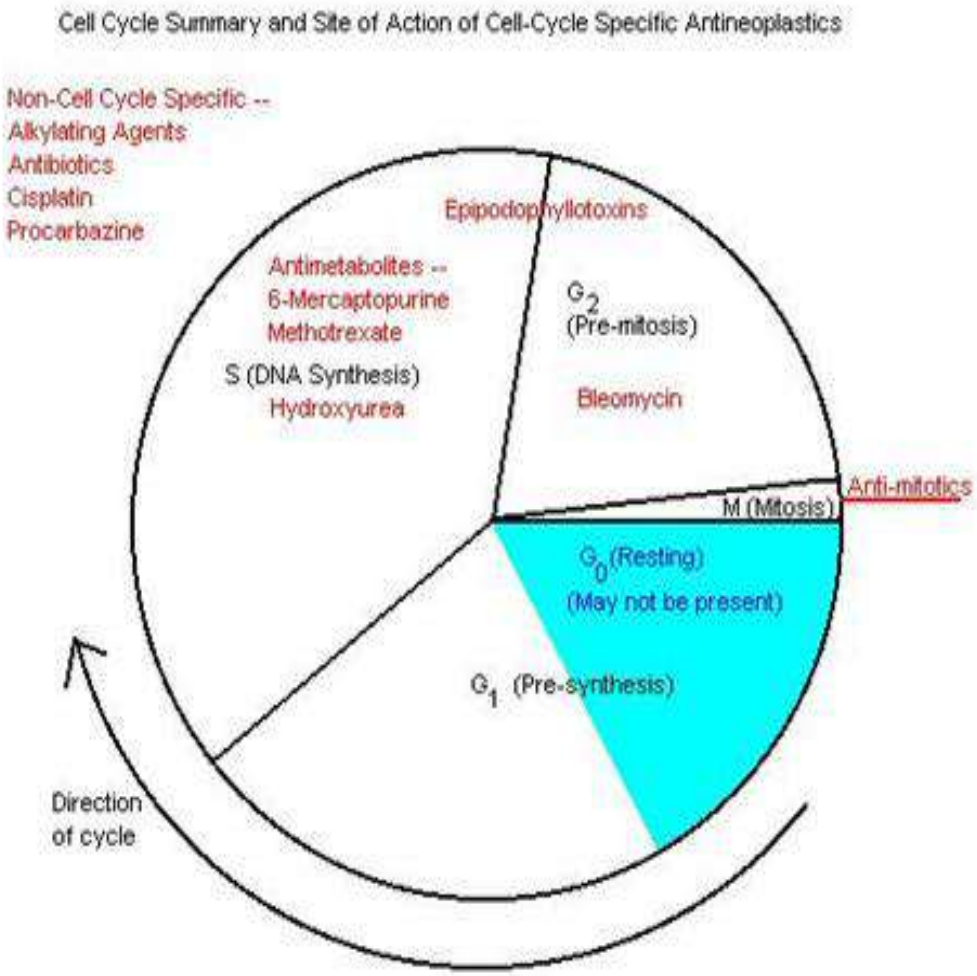
Lecture title	hours
Lecture title	**hours**
β-Lactam antibiotics (Penicillins); β-Lactamase inhibitors; Cephalosporins and Monobactams.	9
Aminoglycosides and Chloramphenicol; Tetracylines; Macrolides; Lincomycins and Polypeptides; Antiviral agents (properties of viruses, viral classification, products).	9
Sulfonamides (chemistry, nomenclature, mechanism of action, resistance, toxicity, side effects, metabolism, protein binding, distribution and SAR); products; Sulfones.	4
Anti-neoplastic agents: Alkylating agents; Antimetabolites; Antibiotics; Plant products; Miscellaneous compounds.	17
Hormones and related compounds; Future anti-neoplastic agents; Monoclonal antibodies; Gene therapy of cancer	6

Today’s quote



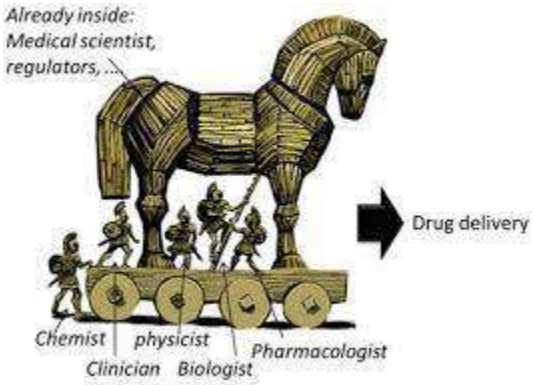
Cell cycle Phases
Site of Action of cell- cycle specific Antineoplastics

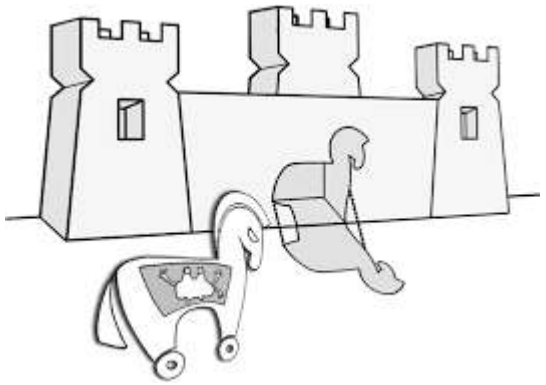




Classification of anticancer agents:

- Alkylating agents.(Lec. 1)
- Antimetabolites. (Lec. 2)
- Natural products.
- Sex hormones and analogues.
- Miscellaneous cytostatics.
- Radioisotopes.
- Immunosuppressive and immunostimulant drugs.





Anti Metabolites

- Most antimetabolites are effective cancer chemotherapeutic agents via interaction with the biosynthesis of nucleic acids. Therefore, several of the useful drugs used in antimetabolite therapy are purines, pyrimidines, folates, and related compounds.

- The antimetabolite drugs may exert their effects by several individual mechanisms **involving enzyme inhibition at active, allosteric, or related sites**. Most of these targeted enzymes and processes are involved in the regulatory steps of cell division and cell/tissue growth.
- **Often the administered drug is actually a prodrug form of an antimetabolite and requires activation in vivo to yield the active inhibitor.**

- The purine and pyrimidine antimetabolites are often compounds incorporated into nucleic acids and the nucleic acid polymers (DNA, RNA, etc.).

- The antifolates are compounds designed to interact at cofactor sites for enzymes involved in the biosynthesis of nucleic acid bases. The biosynthesis of these nucleic acid bases depend heavily on the availability of folate cofactors, hence antimetabolites of the folates find utility as antineoplastic agents.

2.A.Pyrimidine Drugs

The anticancer drugs based on pyrimidine structure are shown in Figure

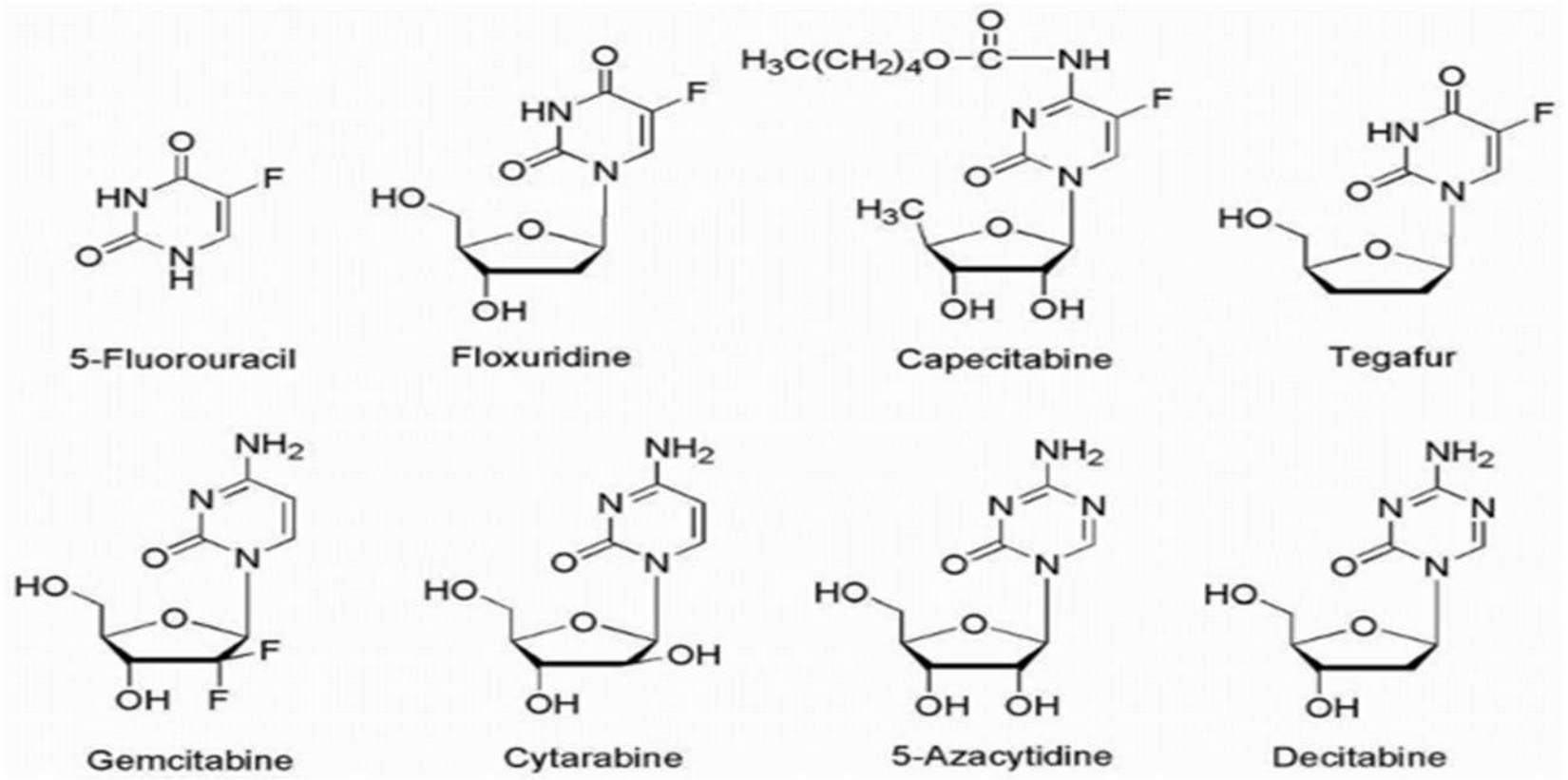
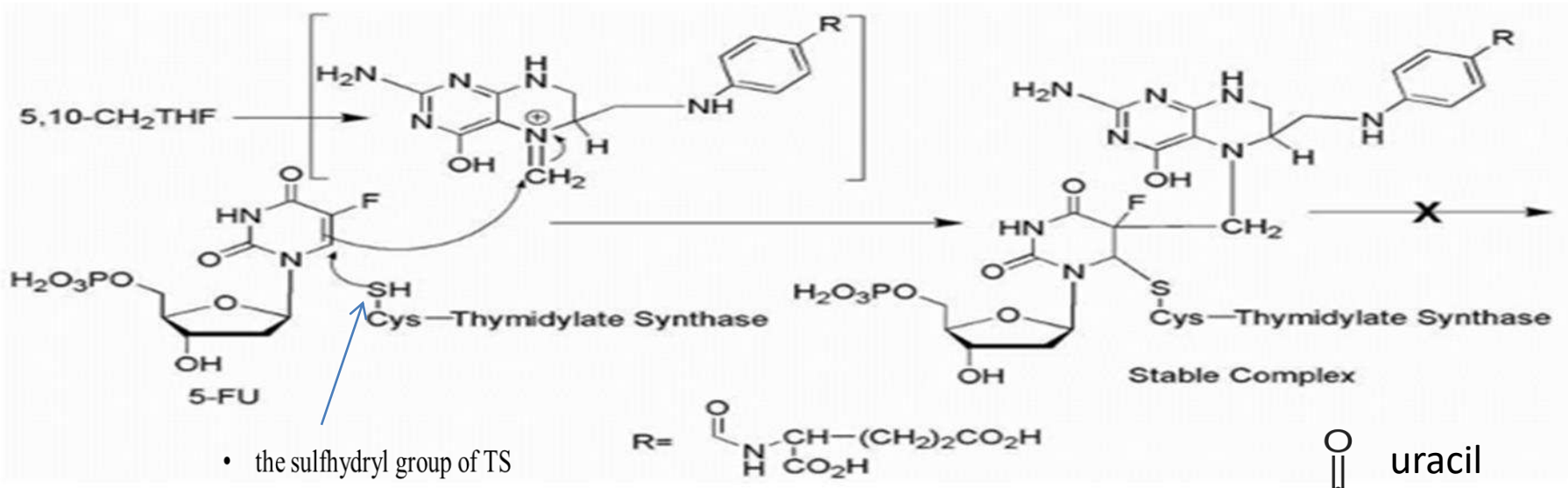


Figure 10.8 • Anticancer drugs based on pyrimidine and related compounds.



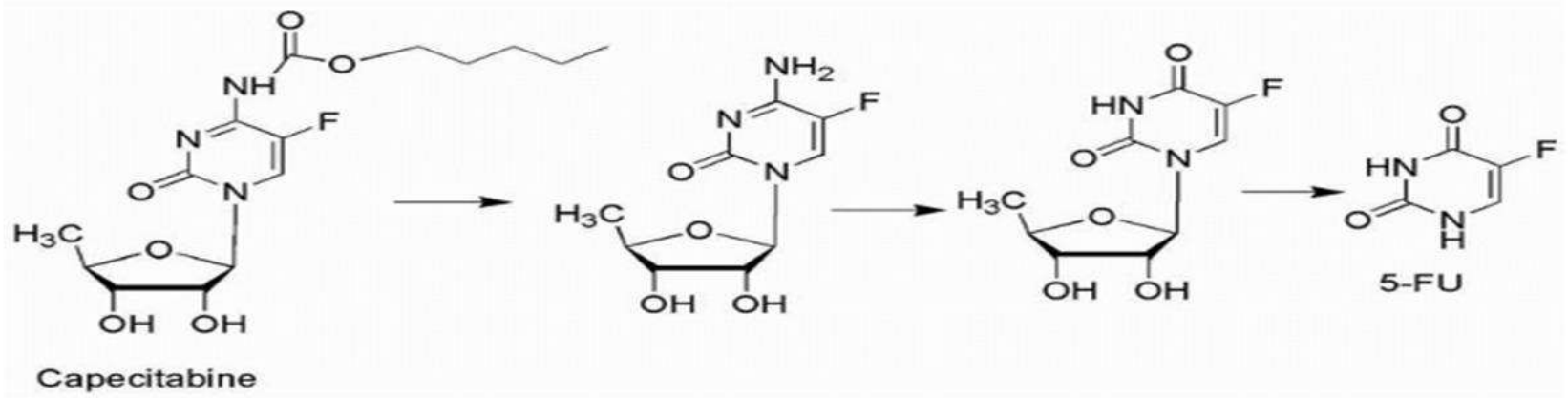
Scheme 10.19 • Mechanism of inhibition of TS by 5-fluorouracil.

Modification of the pyrimidine ring has also been explored for the development of potential anticancer drugs based on antimetabolite theory.

5-Fluorouracil is activated by conversion to the corresponding nucleotide species, 5-fluoro-2-deoxyuridylic acid .The resulting **5-fluoro-2′-deoxyuridylic acid is a powerful inhibitor of thymidylate synthetase(TS**), the enzyme that converts 2′-deoxyuridylic acid to thymidylic acid. In the inhibiting reaction, **the sulfhydryl group of TS adds via conjugate addition to the 6-position of the fluorouracil moiety** (Scheme 10.19), So the chemical mechanism of inhibition of TS by 5-fluorouracil is shown in Scheme 10.19

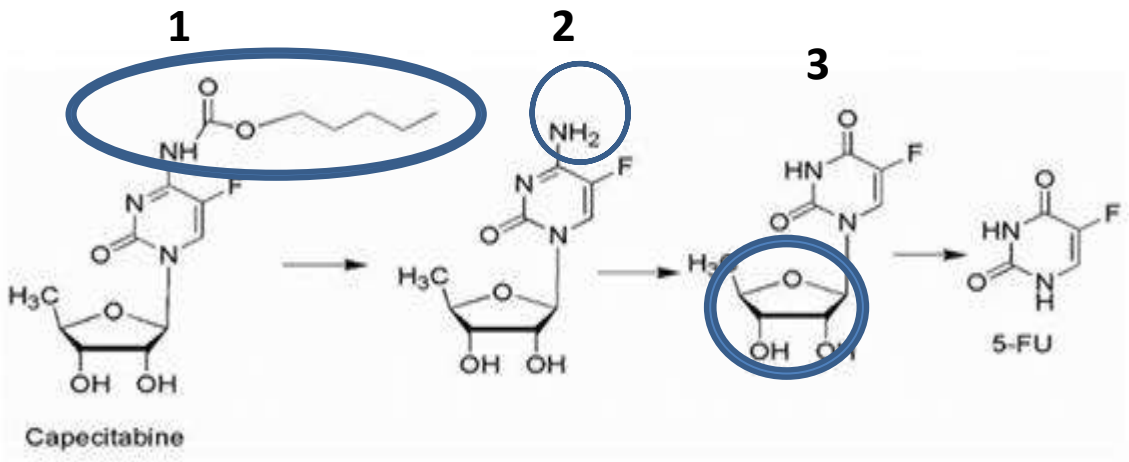
Attempts at chemical modification of 5-FU to protect from catabolic events have produced **several prodrug forms**, which are converted via in vivo metabolic and/or chemical transformation to the parent drug 5-FU.

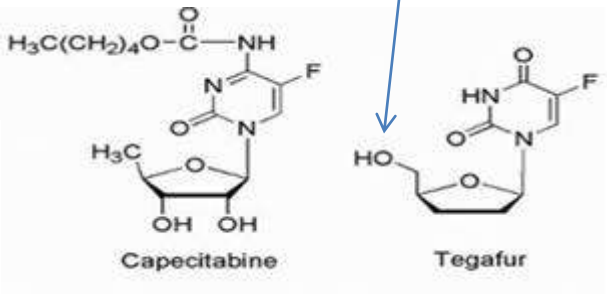
The carbamate derivative of 5′-deoxy-5-fluorocytidine is known as **capecitabine,** and it is converted to 5-FU through a series of activation steps. The activation sequence is shown in Scheme 10.21.



Scheme 10.21 • Metabolic activation of capecitabine to 5-FU.

The initial step is carbamate hydrolysis(1) followed by deamination(2), then hydrolysis of the sugar moiety(3) to yield 5-FU. **Some of these activation steps take place at a higher rate in tumor tissue leading to selective accumulation in those cells**. The last step in the sequence shown in Scheme 10.21 is catalyzed by **phosphorolases, and these enzymes occur in higher levels in colorectal tumors.** Despite this complex activation process**, capecitabine** still exhibits some of the significant toxicities of 5-fluorouracil. The tetrahydrofuran derivative **tegafur** is slowly converted to 5-FU but requires quite high doses to reach therapeutic plasma concentrations. Esters of the N-hydroxymethyl derivative of tegafur show greater anticancer activity than tegafur.

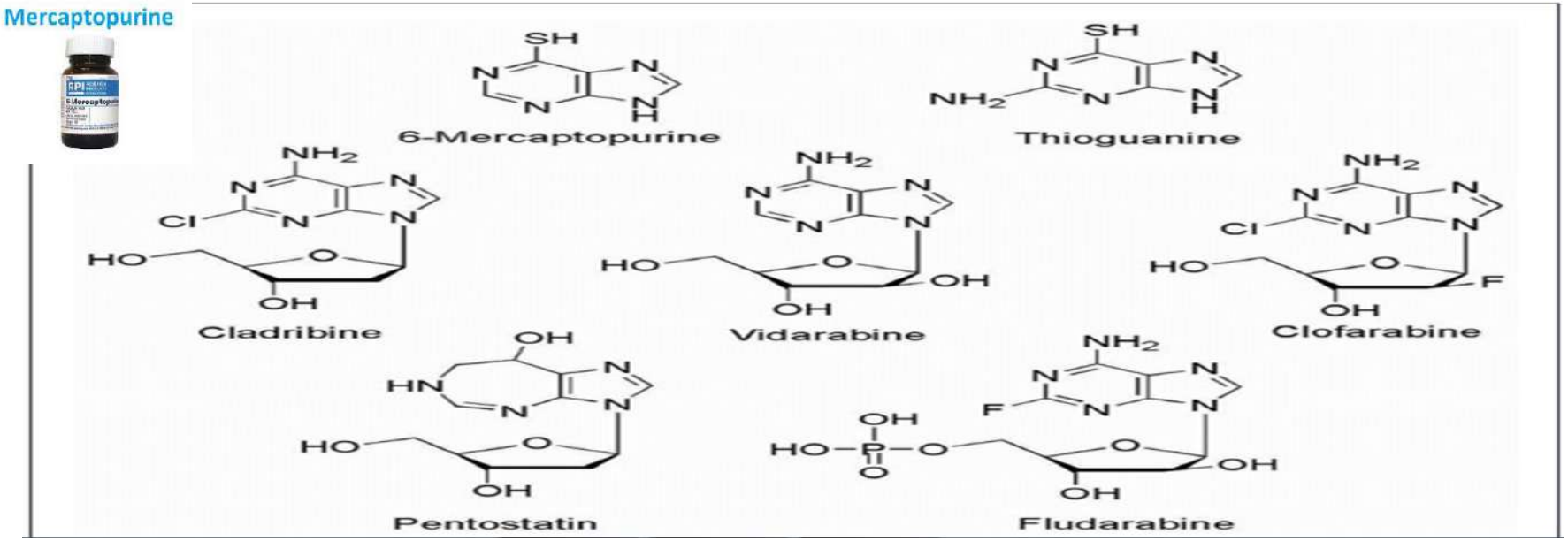




Gemcitabine is the result of fluorination of the **2′-position of the sugar moiety**. The mechanism of action for gemcitabine **includes alteration of the rate of incorporation into DNA as well as the rate of DNA processing and repair.**

2.B.Purine Drugs

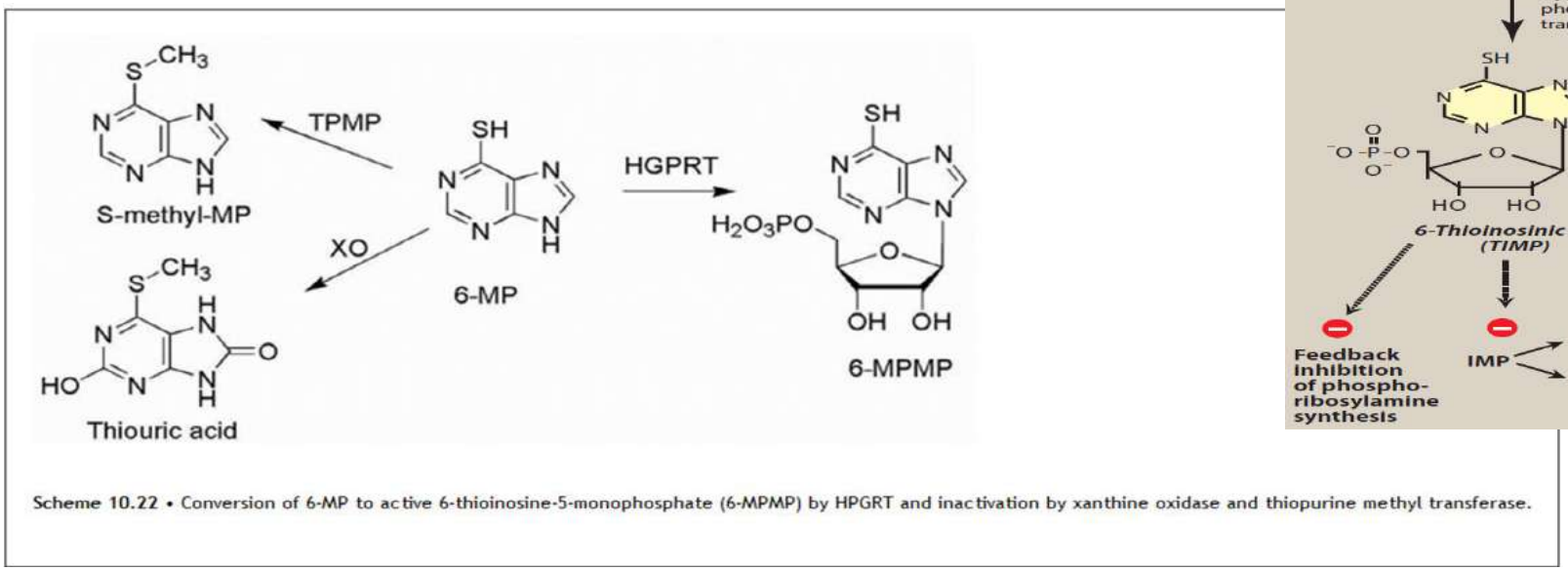
The anticancer drugs based on purine structure are shown in Figure 10.9.



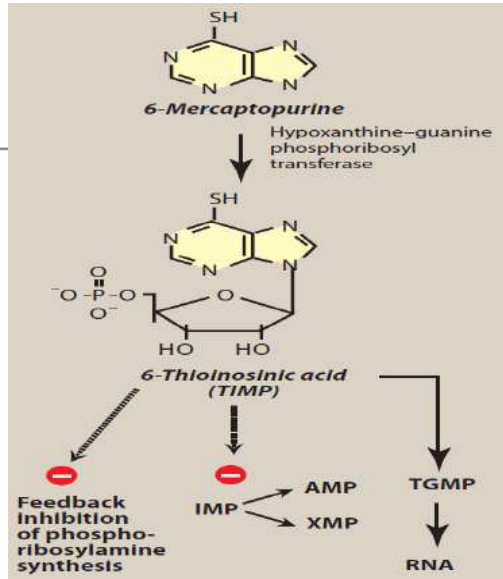
The design of antimetabolites based on purine structure began with isosteric thiol/sulfhydryl group to replace the 6-hydroxyl group of hypoxanthine and guanine.

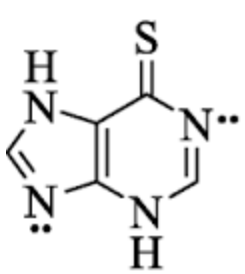
6-mercaptopurine (6-MP). This purine requires bioactivation to its ribonucleotide , 6-thioinosinate (6-MPMP), by the enzyme HGPRT. The resulting nucleotide . is a potent inhibitor of an early step in basic purine biosynthesis

Mercaptopurine competes with hypoxanthine and guanine for the enzyme hypoxanthine-guanine phosphoribosyltransferase (HGPRTase) and is itself converted to thioinosinic acid (TIMP).

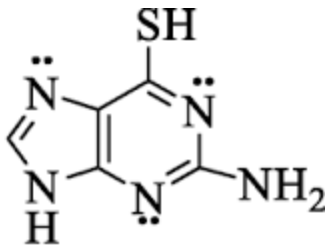


Scheme 10.22 • Conversion of 6-MP to active 6-thioinosine-5-monophosphate (6-MPMP) by HPGRT and inactivation by xanthine oxidase and thiopurine methyl transferase.

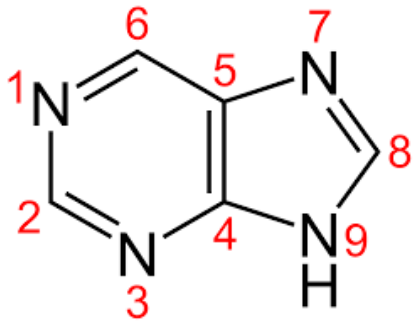




Mercaptopurine



6-Thioguanine



Purine

The major antineoplastic action of 6-MP appears to be related to the inhibition of purine biosynthesis.

Thioguanine (6-TG) is the 6-mercapto analog of guanine, analogous to 6-MP. Thioguanine is converted into its ribonucleotide by the same enzyme that acts on 6-mercaptopurine. It is converted further into the diphosphates and triphosphates. These species inhibit most of the same enzymes that are inhibited by 6-mercaptopurine.Thioguanine is also incorporated into RNA, and its 2′-deoxy metabolite is incorporated into DNA. The incorporation into RNA and DNA and the subsequent disruption of these polymers may account for a greater portion of the antineoplastic activity of thioguanine compared with 6-MP.

1. C. Folates

Folic acid and the structures of the major antifolate anticancer drugs are shown in Figure 10.10.

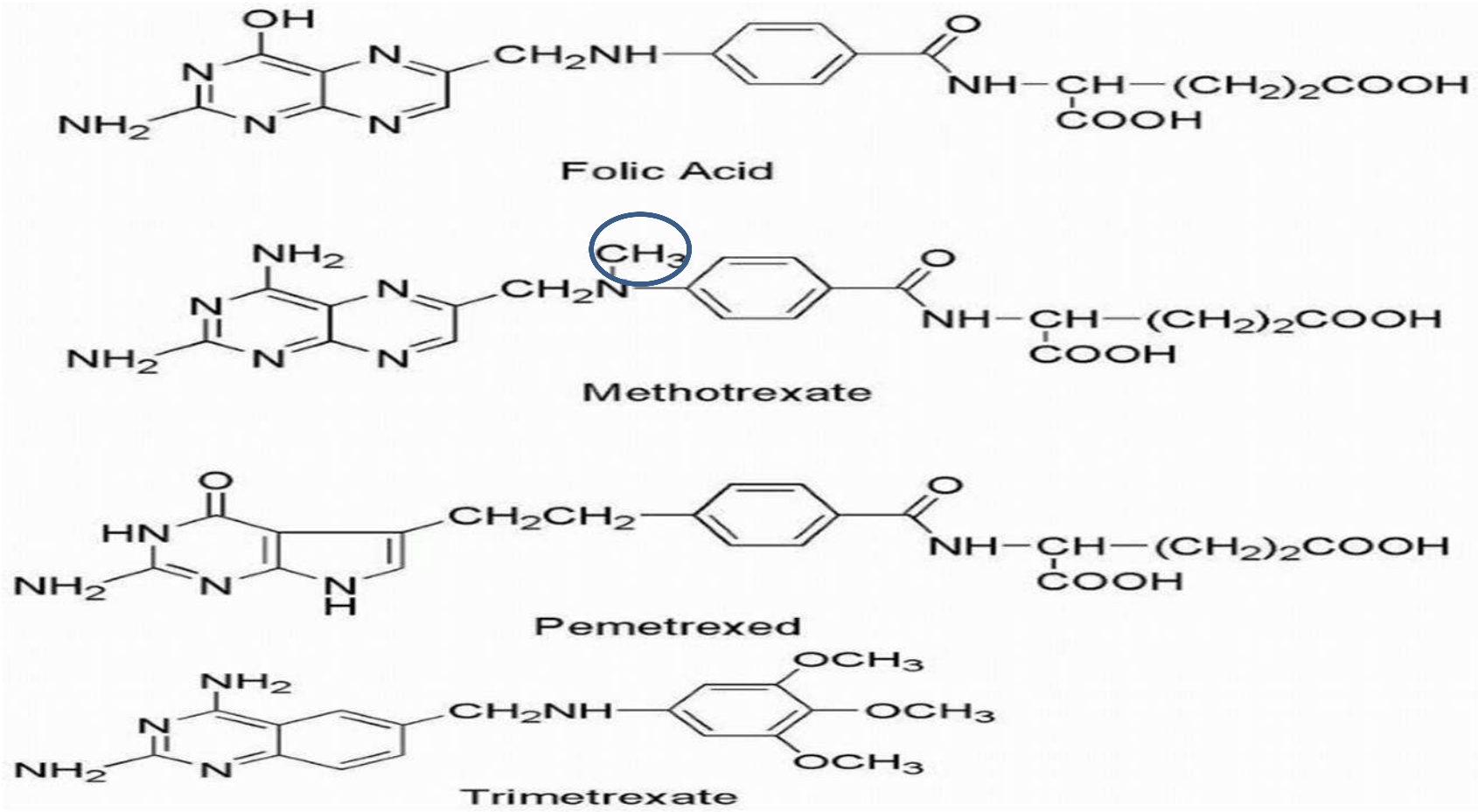
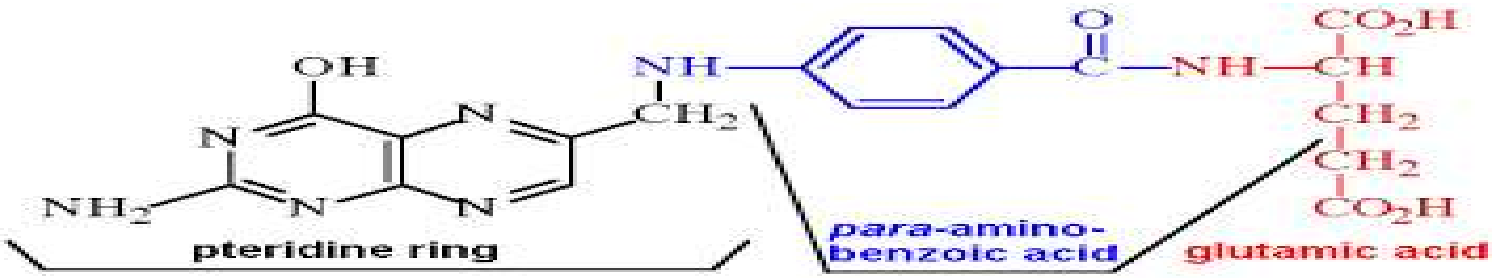
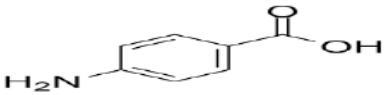
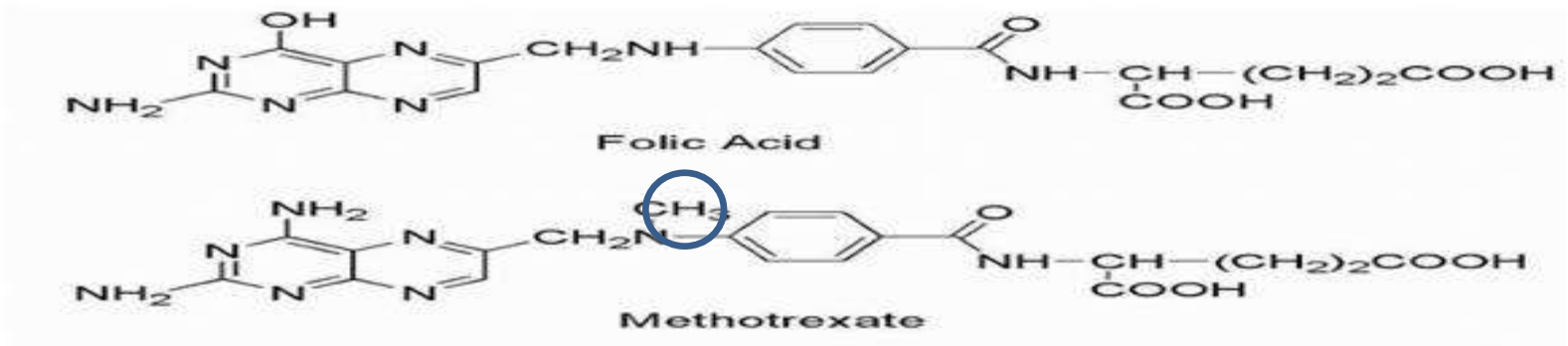


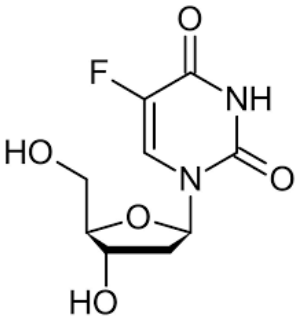
Figure 10.10 • Structures of folic acid and antifolate anticancer drugs.



Methotrexate is the classic antimetabolite of folic acid structurally derived by N-methylation of the para-aminobenzoic acid residue (PABA) and replacement of a pteridine hydroxyl by the bioisosteric amino group. The conversion of —OH to —NH2 increases the basicity of N-3 and yields greater enzyme affinity. This drug competitively inhibits the binding of the substrate folic acid to the enzyme DHFR, resulting in reductions in the synthesis of nucleic acid bases, perhaps most importantly, the conversion of uridylate to thymidylate as catalyzed by thymidylate synthetase. In addition, purine synthesis is inhibited because the N-10-formyl tetrahydrofolic acid is a formyl donor involved in purine synthesis.

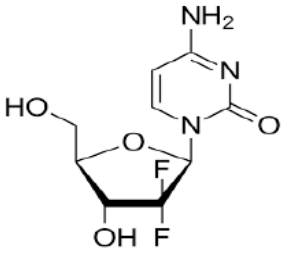






FLOXURIDINE (FLUORODEOXYURIDINE, FUDR)

The drug is available as a 500-mg vial of lyophilized powder. The drug is used to treat metastatic GI adenocarcinoma. The mechanism of action of this fluoropyrimidine deoxynucleoside analog involves metabolic conversion to 5-fluorouracil (5-FU) metabolites resulting in inhibition of TS thus disrupting DNA synthesis, function, and repair.



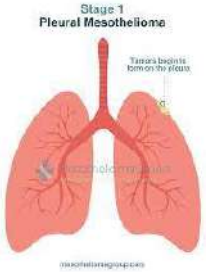
GEMCITABINE (DFDC, GEMZAR)

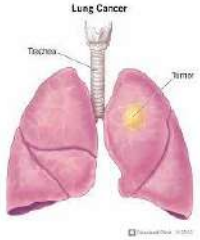
The drug is available as the hydrochloride salt in 200- and 1,000-mg lyophilized single-dose vials for IV use. Gemcitabine is used to treat bladder cancer, breast cancer pancreatic cancer. The mechanism of action of this fluorine-substituted deoxycytidine analog involves inhibition of DNA synthesis and function via DNA chain termination. The triphosphate metabolite is incorporated into DNA inhibiting several DNA polymerases and incorporated into RNA inhibiting proper function of mRNA. **Resistance can occur because of decreased expression of the activation enzyme deoxycytidine kinase or decreased drug transport as well as increased expression of catabolic enzymes.**

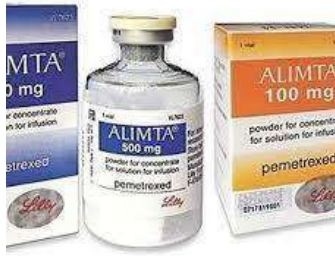
Pentostatin

PENTOSTATIN (2′-DEOXYCOFORMYCIN, DCF, NIPENT)

The drug is available in 10-mg vials for IV use. The drug is used to treat leukemias such as hairy cell leukemia, chronic lymphocytic leukemia, and lymphoblastic leukemia. The mechanism of action involves inhibition of the **enzyme adenosine deaminase yielding increased cellular levels of deoxyadenosine and deoxyadenosine triphosphate (dATP).** The increased levels of dATP are cytotoxic to lymphocytes. Pentostatin is a fermentation product of Streptomyces antibioticus. Resistance appears to involve decreased cellular transport or increased expression of catabolic enzymes. Acid instability prevents oral administration, and the drug is only administered by IV.







PEMETREXED (MTA, ALIMTA)

The drug is available in a 100-mg sterile vial for IV use. The drug appears to be effective against a range of tumors including mesothelioma, NSCLC, colorectal cancer, bladder cancer, and lung cancer. The mechanism of action involves inhibition of TS resulting in inhibition of thymidylate and DNA synthesis. This drug is a pyrrolopyrimidine analog of folate with antifolate activity. Resistance can occur by increased expression of TS, decreased binding affinity for TS, or decreased drug transport into cells.

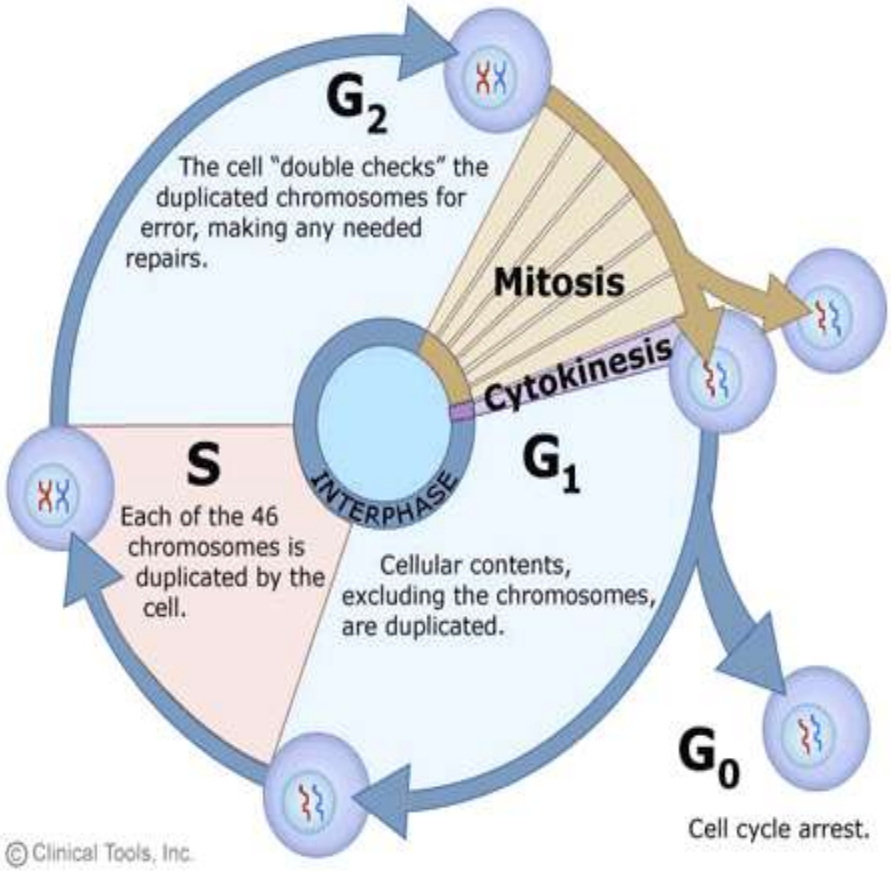
3. ANTIBIOTICS AND NATURAL PRODUCTS

A variety of the anticancer agents available today are derived from natural sources with several of these being obtained from microbial sources (antibiotics). Many of the antineoplastic antibiotics are produced by the soil fungus Streptomyces. Both the antibiotic and natural product classes have multiple inhibitory effects on cell growth; however, they primarily act to disrupt DNA function and cell division.

There are several mechanisms by which these agents target DNA, including intercalation, alkylation, and strand breakage either directly or as a result of enzyme inhibition. Intercalation is a process by which a planar molecule of the appropriate size inserts itself between adjacent base pairs of DNA and in so doing, it causes a local unwinding that may disrupt the normal template function of DNA. Intercalation requires that the drug induce a cavity between base pairs so that insertion may occur.

- There are several natural products that are capable of disrupting the formation and function of the mitotic spindle.

- These include the epipodophyllotoxins, the taxanes, and the vinca alkaloids.
- The mitotic spindle forms during the M phase of the cell cycle and is responsible for moving the replicated DNA to opposite ends of the cell in preparation for cell division.



1. Epipodophyllotoxins

The epipodophyllotoxins (Fig.)are semisynthetic derivatives of podophyllotoxin, which is isolated from the mayapple (mandrake) root and functions as an inhibitor of microtubule function.

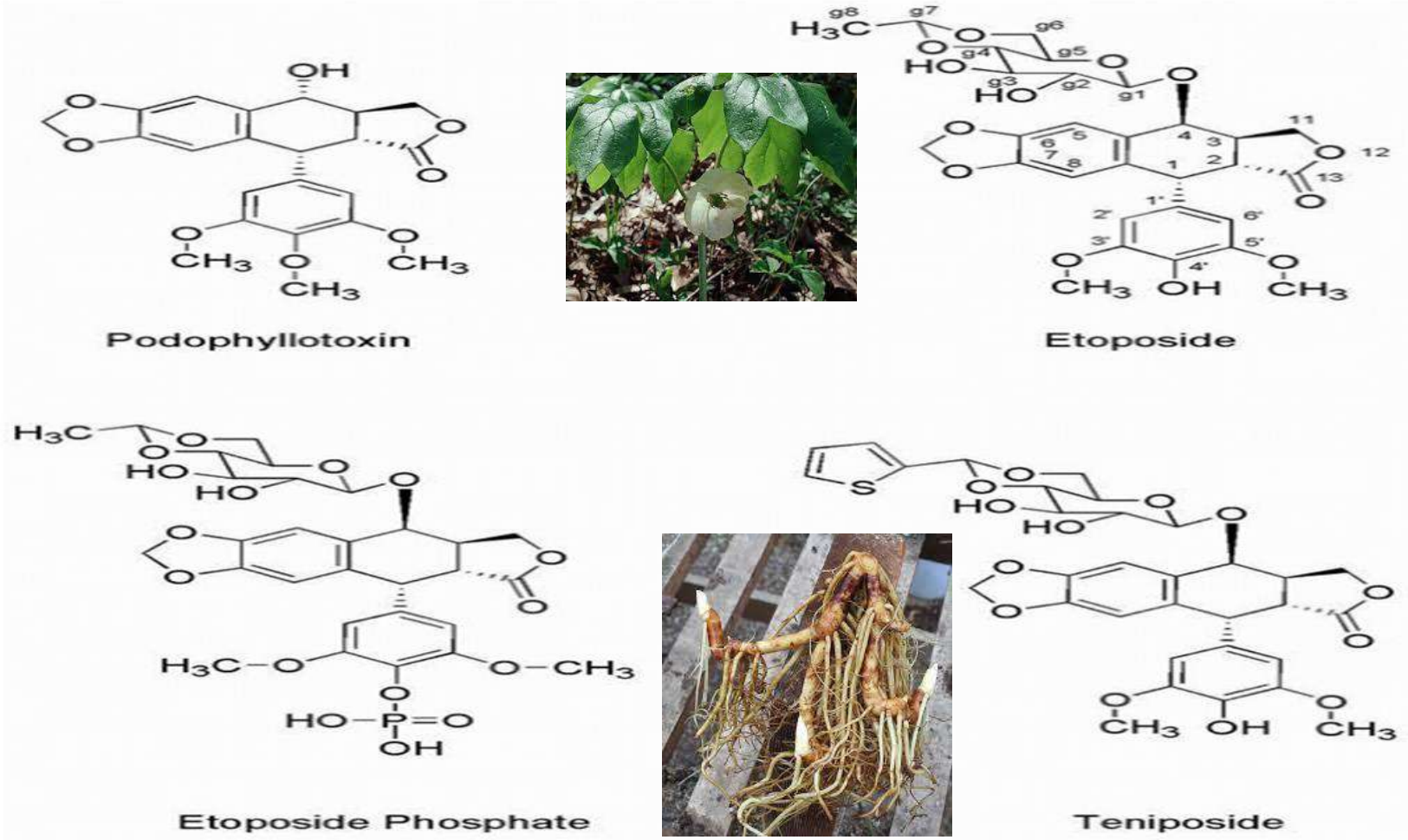


Figure 10.15 • Structures of the epipodophyllotoxins.

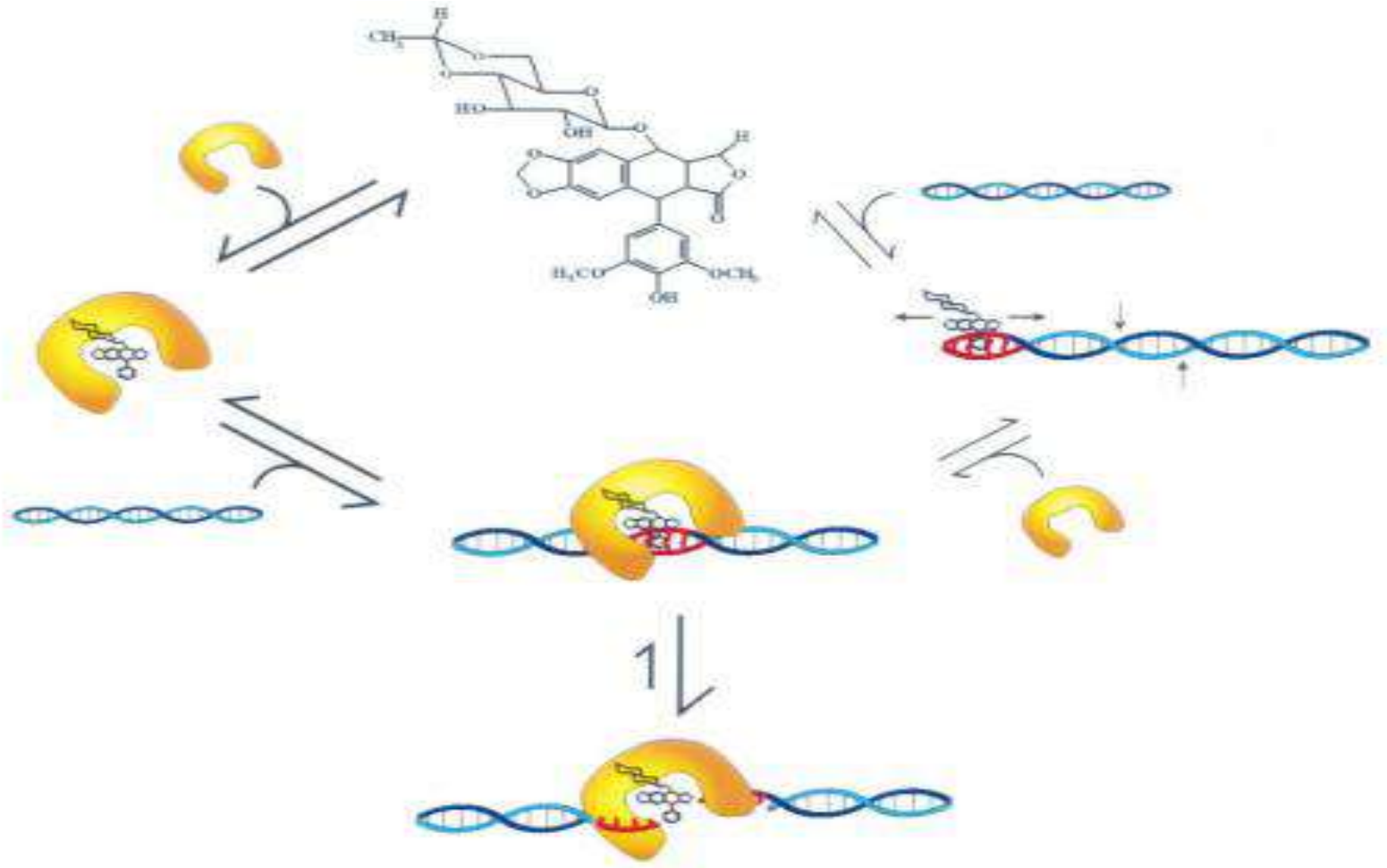
Chemical modification has led to compounds with a different mechanism of action, which involves inhibition of topoisomerase enzymes.

The change in mechanism was associated with removal of the 4′-methyl group of podophyllotoxin. Further alteration in podophyllotoxin involved the addition of the glycosidic portion of the molecules.

Etoposide acts on topoisomerase II stabilizing the cleavable complex leading to single- and double-strand breaks. If enough breaks are initiated, apoptosis is activated.

Etoposide is believed to bind to topoisomerase II in the absence of DNA, because it shows little tendency to interact with DNA alone. The etoposide-topoisomerase II complex then binds DNA, and strand cleavage occurs; however, the ligation step is inhibited. Binding of the drug occurs near the site at which the cleaved phosphodiester bond is held by the enzyme.

Pathway of etoposide-induced DNA cleavage complex formation



Camptothecins

The camptothecins (Fig. 10.16) are inhibitors of topoisomerase I and are used clinically for the treatment of various cancers.

The lead drug for this class of agents was **camptothecin,** which was isolated from Camptotheca acuminata, an ornamental tree found in China.

The mechanism of antitumor action was inhibition of topoisomerase I. Subsequently, **the incorporation of side chains containing basic amines led to the more water-soluble derivatives, topotecan and irinotecan** (Fig. 10.16).

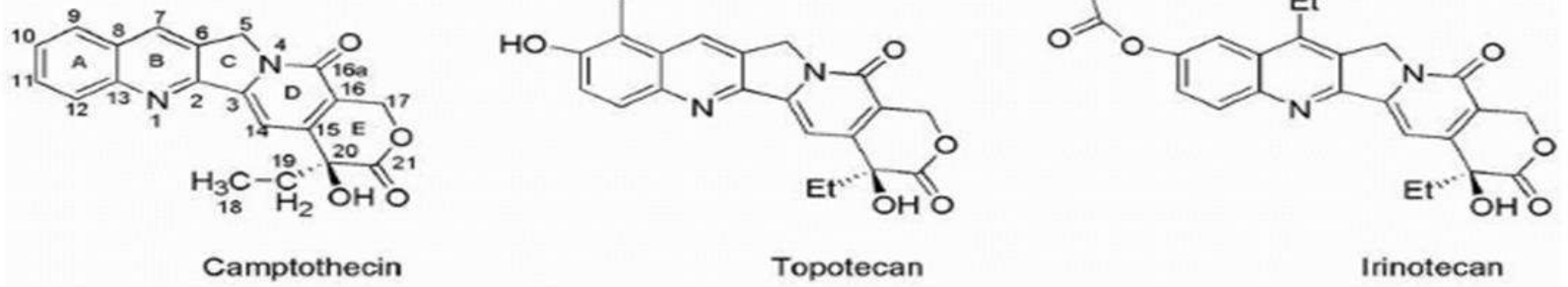
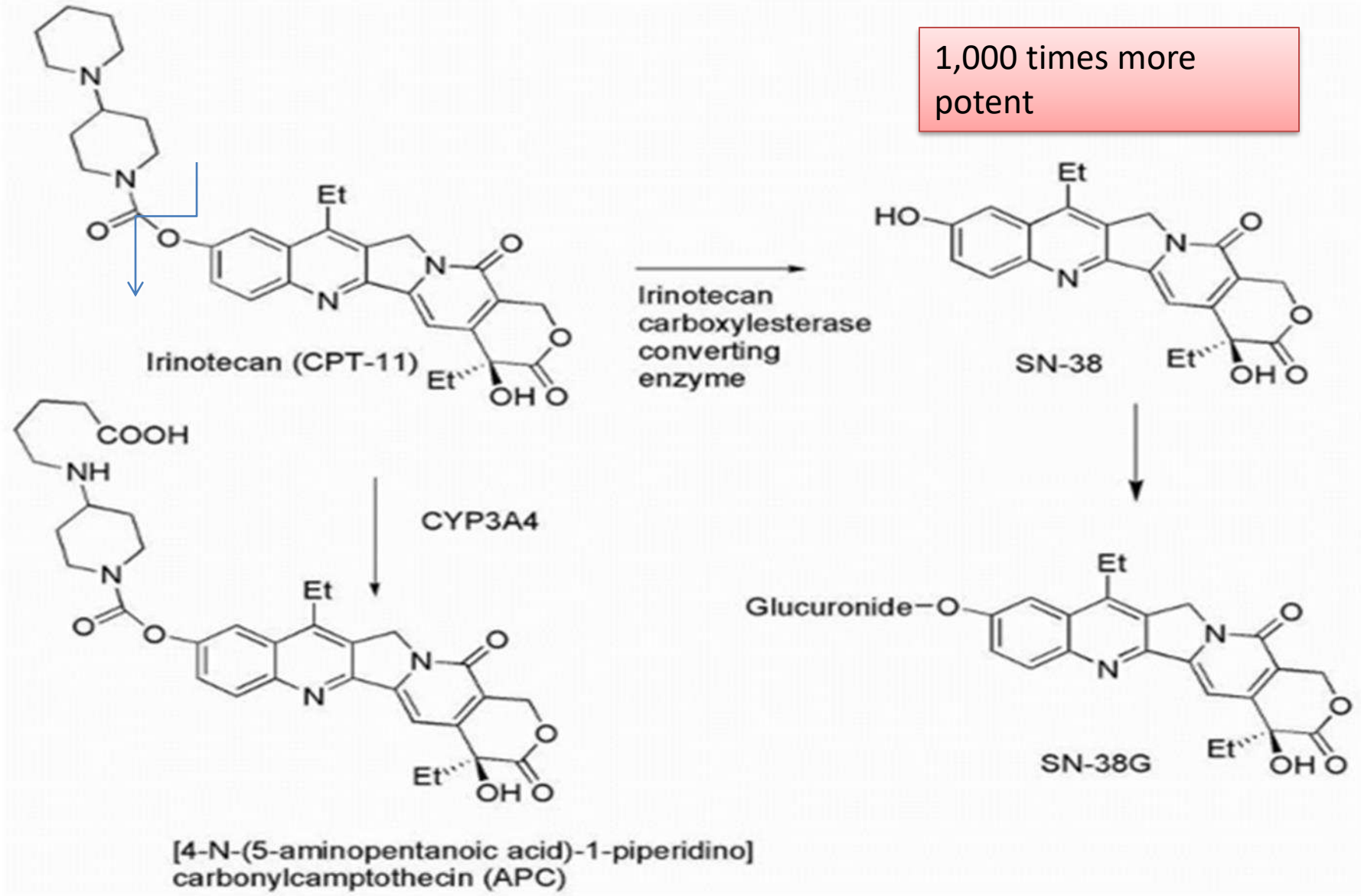


Figure 10.16 • Structures of topoisomerase I inhibitors.

Irinotecan undergoes hydrolysis of its carbamate moiety by irinotecan-converting enzyme to give SN-38, which is 1,000 times more potent than the parent compound (Scheme 10.29). There is wide interpatient variability in the extent of this transformation, which may explain differential responses to the agent. Further metabolism involves the glucuronidation (isozyme UGT1A1) of the resulting phenolic function of SN-38 to give SN-38G, which is inactive.

An additional metabolite forms as a result of CYP3A4-mediated conversion to [4-N-(5-aminopentanoic acid)-1-piperidino]carbonylcamptothecin (APC), which is 100 times less active than SN-38. There is also the additional complication that the parent and metabolites may exist as the lactones or as the inactive hydroxy acid forms.

1,000 times more potent



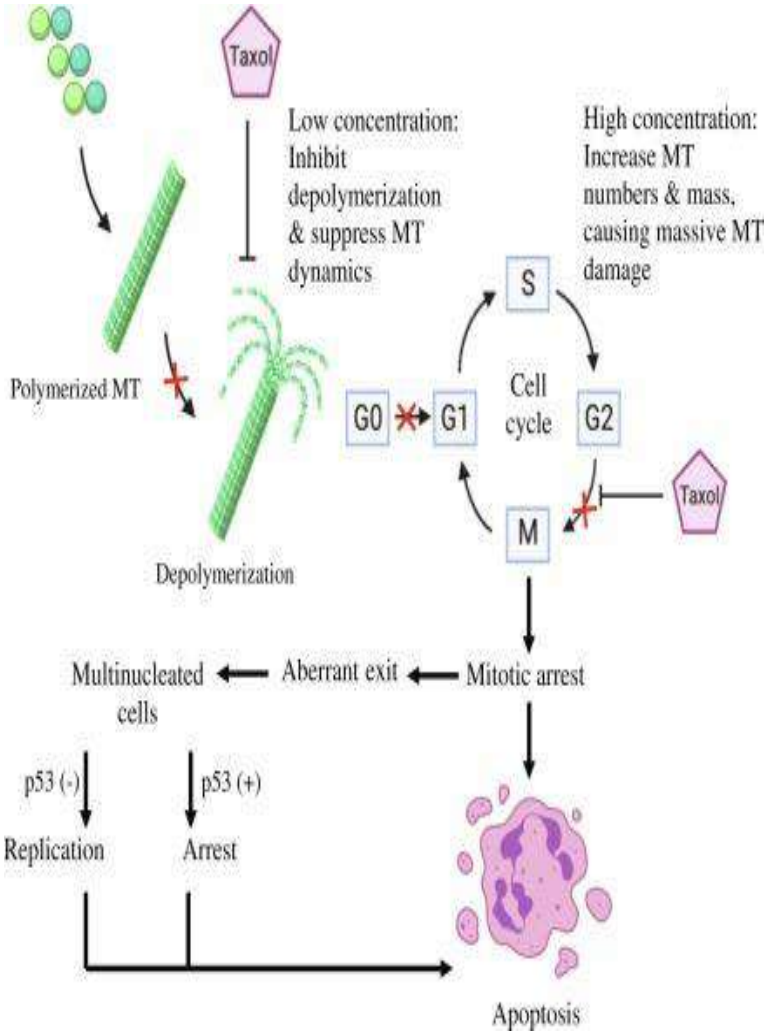
Scheme 10.29 • Metabolism of irinotecan.

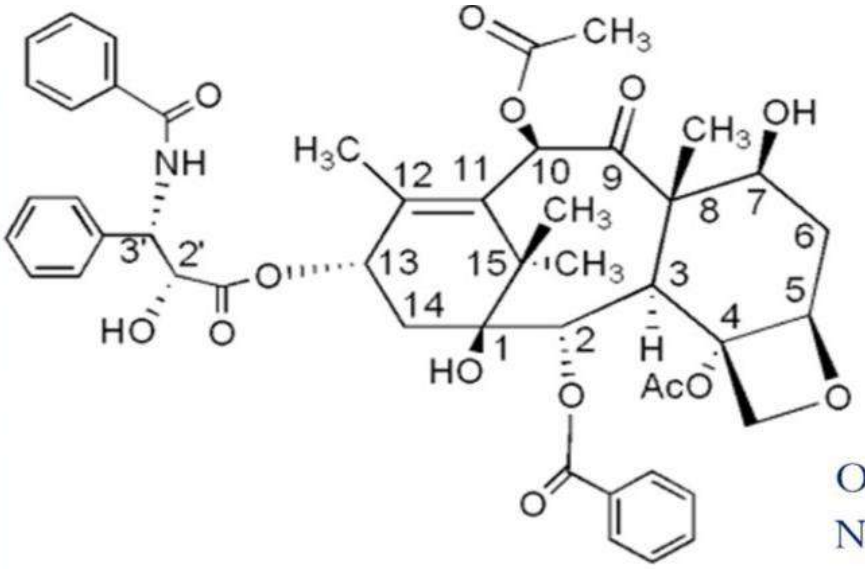
2.Taxanes

The taxanes or taxoids are a closely related group of antineoplastic agents that have a unique mechanism of action as inhibitors of mitosis and which are widely used in the therapy of ovarian, breast, lung, esophageal, prostate, bladder and head and neck cancers.

Taxanes exert their anticancer effect by inhibiting **the mitotic spindle—they bind to the microtubules and prevent their depolymerization, thus inhibiting mitosis and inducing apoptosis in cells undergoing the division process.** The drugs are cell-cycle specific in the M-phase.

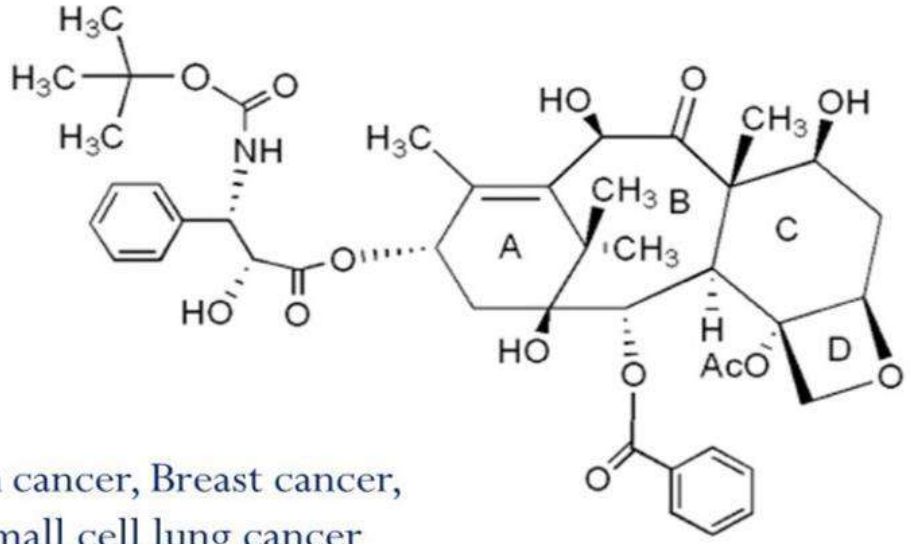
Briefly, the principal mechanism of Taxol is its ability to stabilize and prevent microtubules depolymerization, leading to cell cycle arrest at the G2/M phase and cell death. However, this effect could be cell lines and dose dependent.





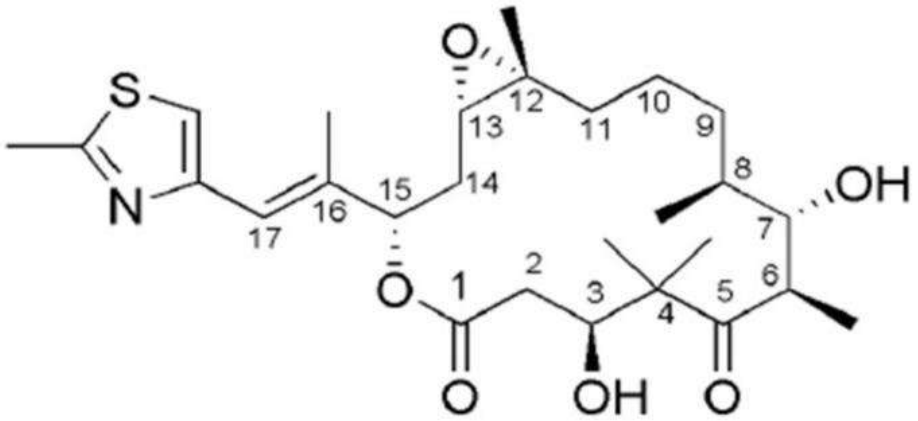
Paclitaxel

Ovarian cancer, Breast cancer,
Non –small cell lung cancer

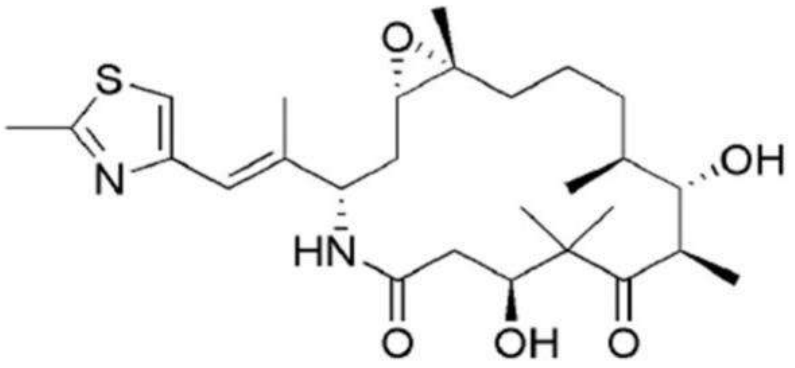


Docetaxel

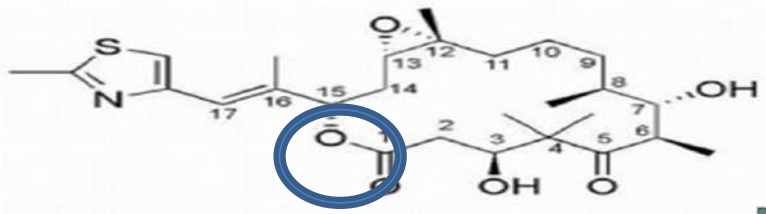
Semi-synthetic derivatives



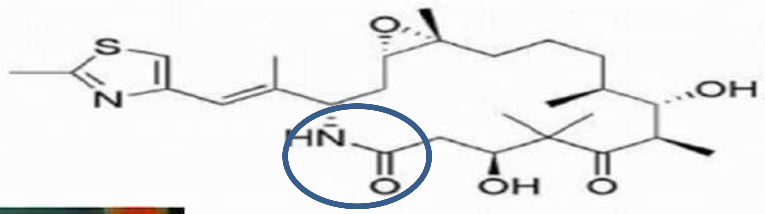
Epothilone B



Ixabepilone



Epothilone B



Ixabepilone

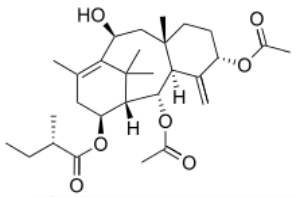
Figure 10.23 • Structures of epithilones.

The epothilones are macrocyclic lactones that have a mechanism of action similar to that of the taxanes but offer several advantages (Fig. 10.23).

Ixabepilone is the **semisynthetic amide analogue** of epothilone B that is isolated from the myxobacterium Sorangium cellulosum. The epothilones showed **potent in vitro activity but greatly decreased activity in vivo caused by metabolic instability via hydrolysis of the macrocyclic lactone**. Conversion to the lactam increased stability and maintained in vivo activity. **Ixabepilone has been recently approved for the treatment of metastatic breast cancer that is resistant to the taxanes**. Molecular modeling studies have been utilized to identify a common pharmacophore between the taxanes and epothilones.

Key structural components that assume comparable relative position are indicated in Table 10.2. Like the taxanes, ixabepilone binds to β-tubulin and stabilizes microtubules resulting in cell death. The current indications for the agent are in metastatic breast cancer in combination with capecitabine after the failure of an anthracycline and a taxane and as monotherapy in metastatic breast cancer after failure of an anthracycline, a taxane, and capecitabine.

Major toxicities associated with the use of ixabepilone have included peripheral neuropathy and myelosuppression occurring as neutropenia

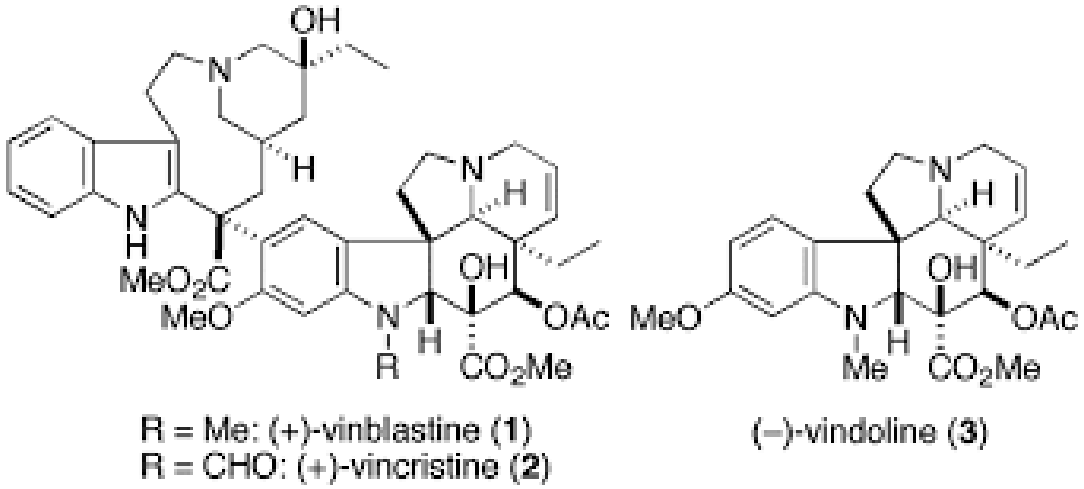


Paclitaxel	Epothilone B
C-15 gem-dimethyl	C-4 gem-dimethyl
C-1 OH	C-3 OH
C-9 carbonyl	C-7 OH
C-5 oxetane-oxygen	C-12,13 epoxide oxygen
C-3' phenyl ring	C-17 thiazole ring

3. Vinca alkaloids

The mechanism of action of vinca alkaloids is **to arrest dividing cells in metaphase by binding tubulin and preventing its polymerization into microtubules.**

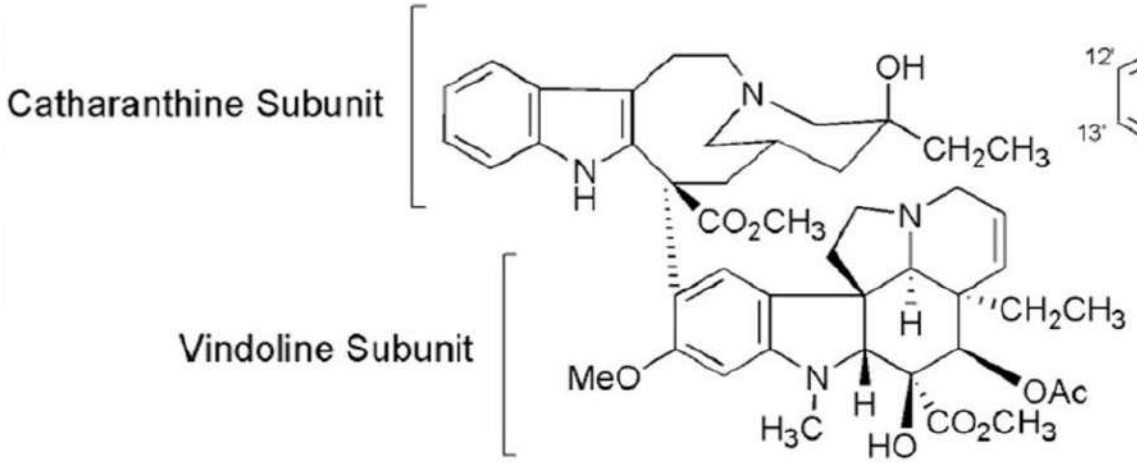
Vinca alkaloids, including vinblastine, vincristine, vindesine and vinorelbine, are widely used antineoplastic drugs, either as single agents or in combination with other drugs. The mechanism of action of these cell cycle-dependent agents is the inhibition of tubulin polymerisation into microtubules



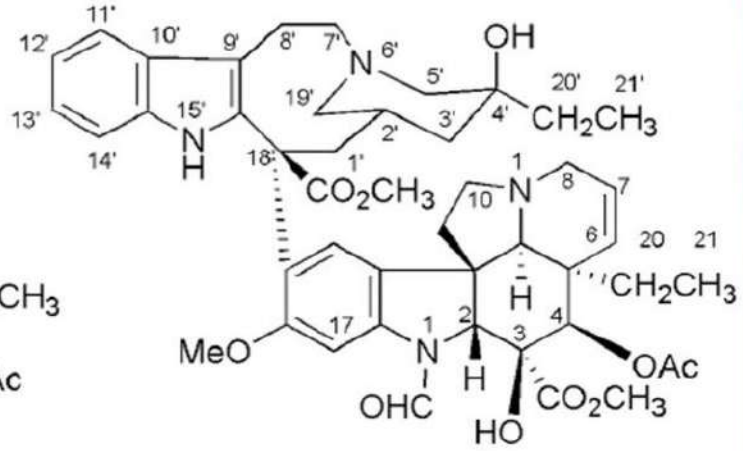
R = Me: (+)-vinblastine (**1**)
R = CHO: (+)-vincristine (**2**)

(–)-vindoline (**3**)

3. Vinca alkaloids.

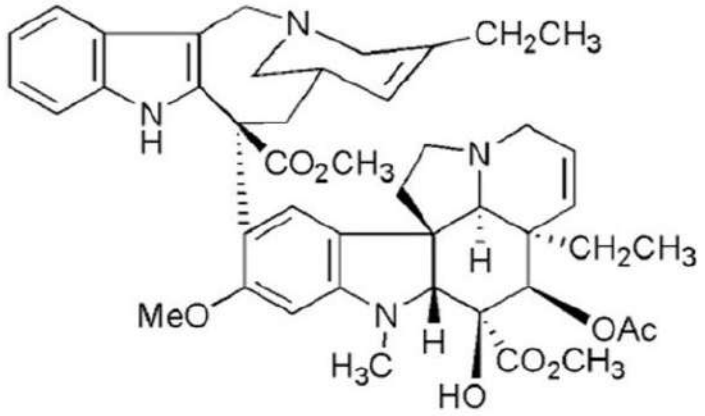


Vinblastine



Vincristine

Hodgkin's disease,
Acute lymphocytic leukemia,
Lung cancer



Vinorelbine

4.PROTEIN KINASE INHIBITORS

Imatinib (Fig. 10.26) was developed to specifically **inhibit tyrosine kinase(TK) and does so rather selectively by binding to the ATP-binding pocket and stabilizing an inactive form of the enzyme.**

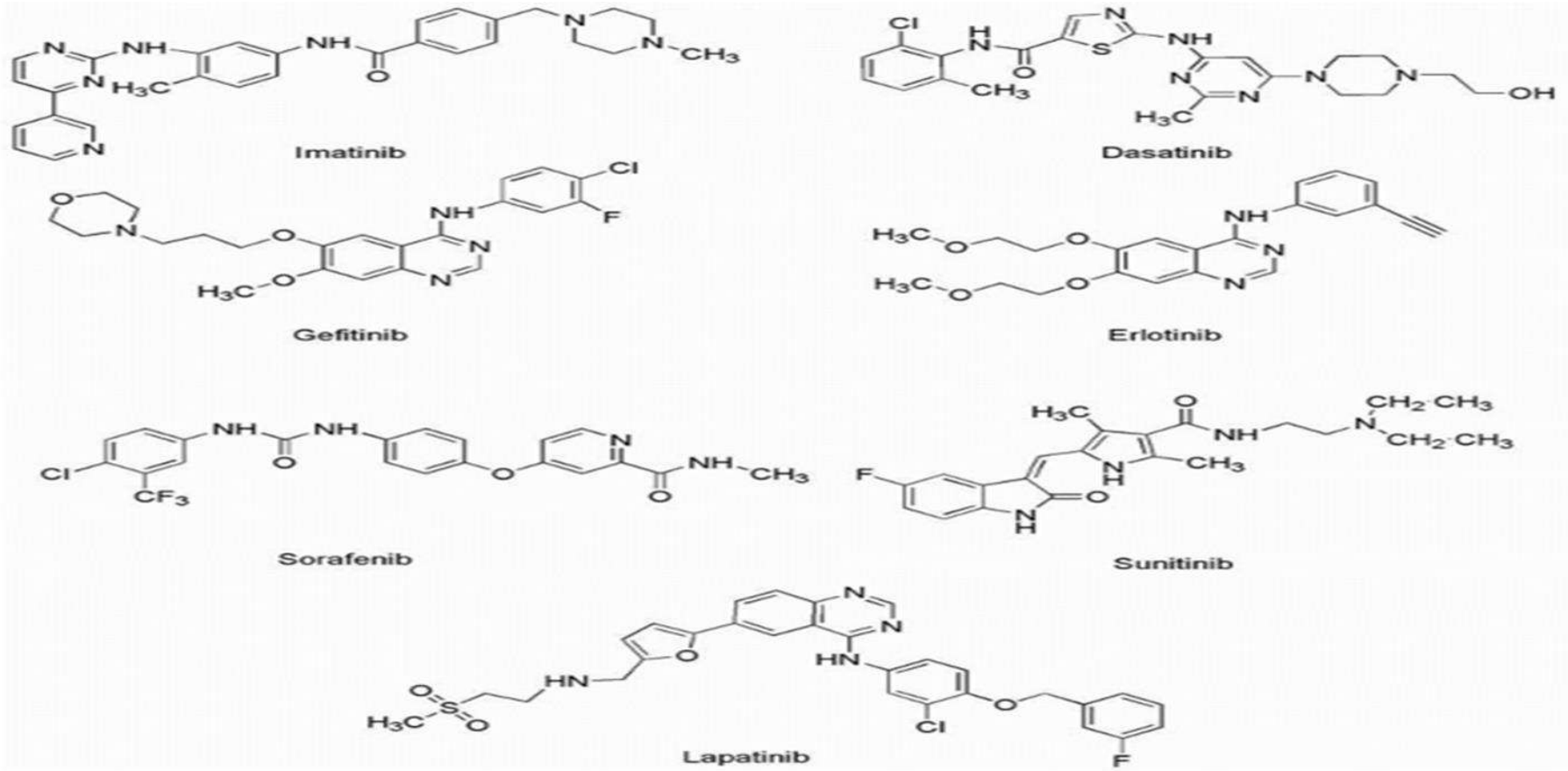
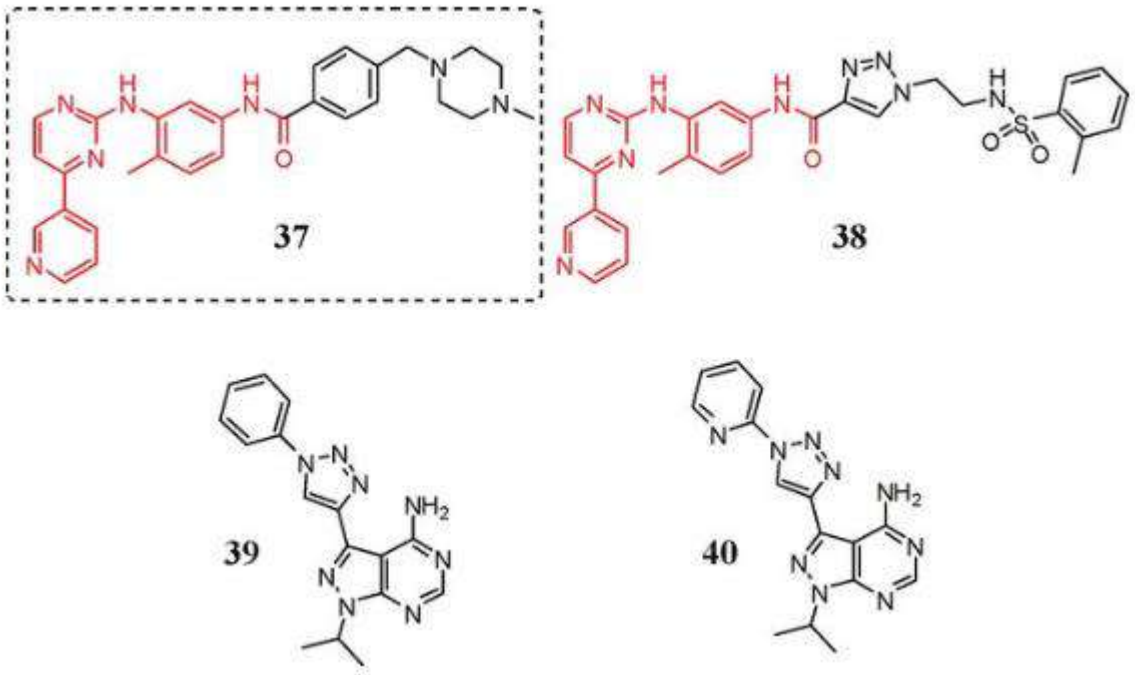


Figure 10.26 • Protein kinase inhibitors.

Protein kinases transfer a phosphoryl group from ATP onto target proteins and play a **critical role in signal transduction and other cellular processes.**

Protein kinases are enzymes that phosphorylate (add a phosphate, or PO4, group) to a protein and can modulate its function.



Chemical structures of protein kinase inhibitors. 37: Imatinib (a selective and potent inhibitor of Abl
38: A potent and selective click-inhibitor of Abl kinase identified from the click-inhibitor library. The Imatinib core structure is highlighted in red. 39 and 40: Click-inhibitors of PfPK7.

Organic Pharm. Chemistry III

Lecture 10 (last)

13th May 2023

11:30- 2:30

AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023

- Courage doesn’t always roar. Sometimes courage is the quiet voice at the end of the day saying, ‘I will try again tomorrow.’

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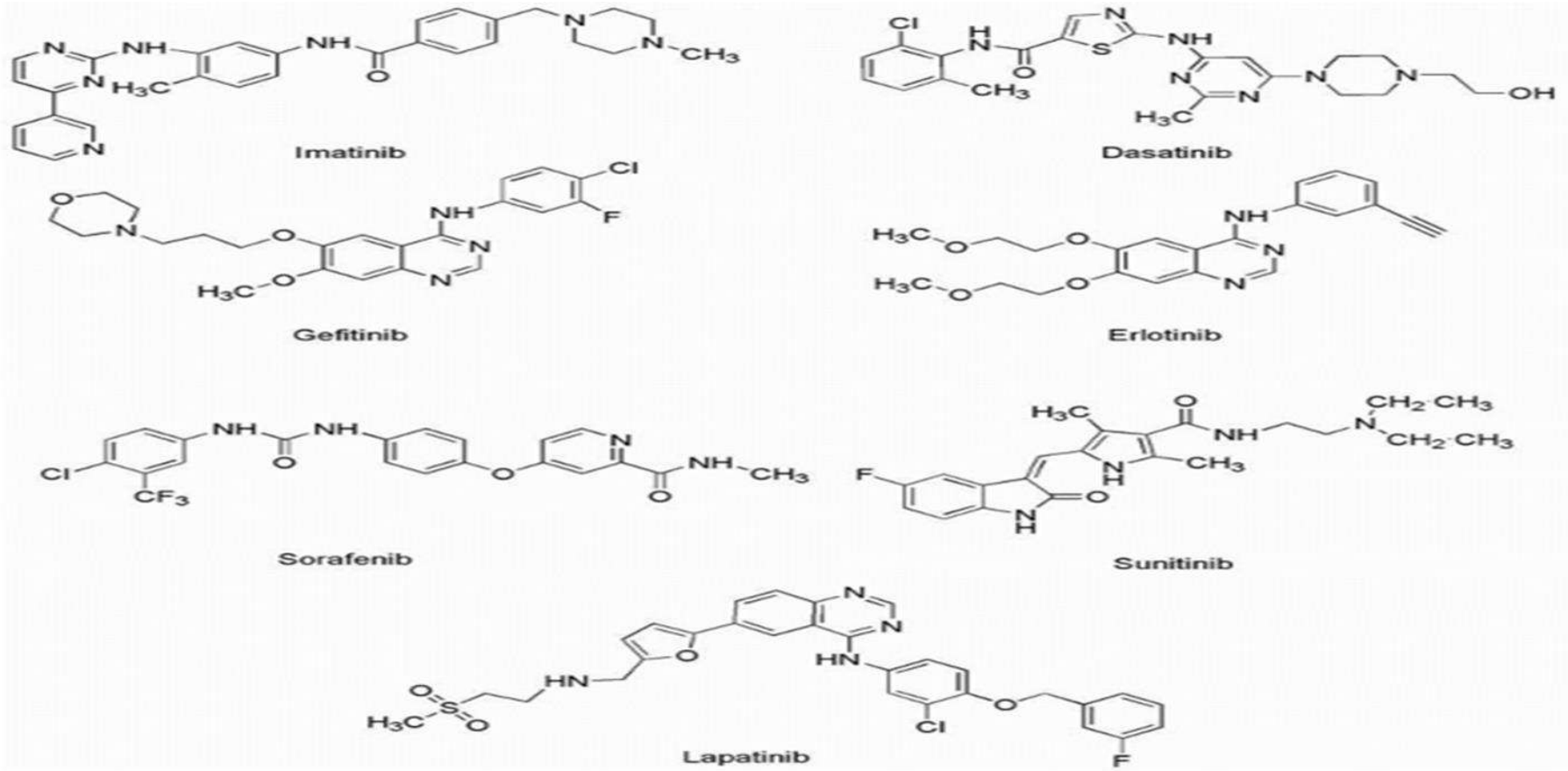

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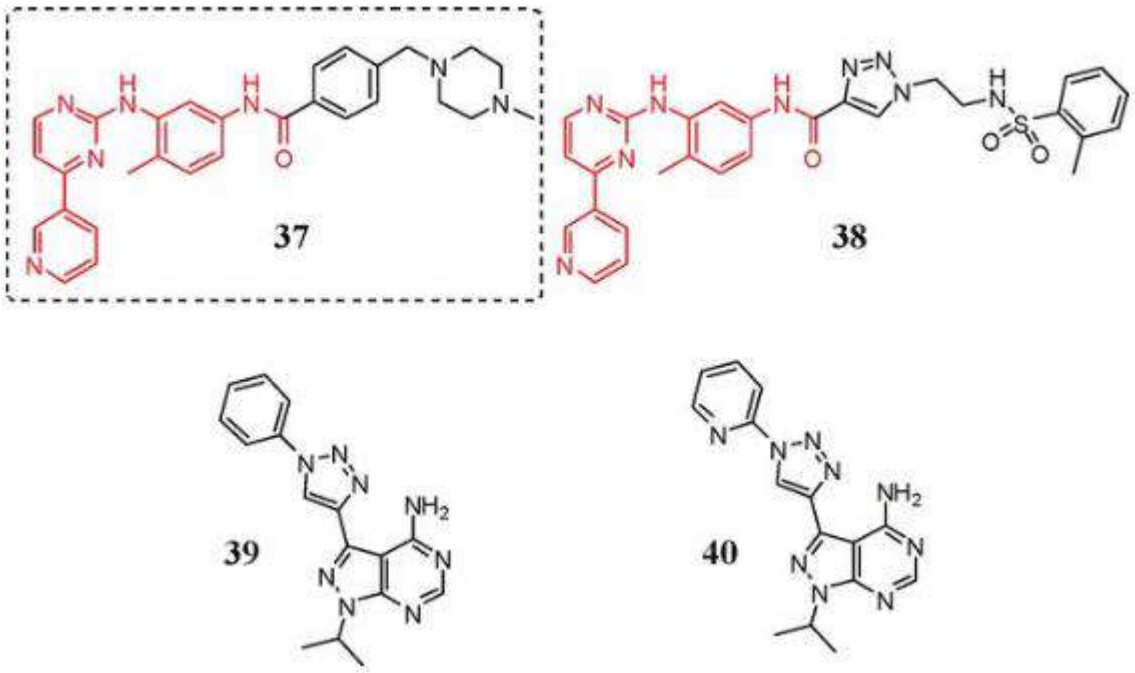

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Classification of anticancer agents:

- Alkylating agents.
- Antimetabolites.
- Natural products.
- Protein Kinase
- **Miscellaneous cytostatics.**
- Sex hormones and analogues
- Monoclonal antibody conjugates (Antibody-drug conjugates ADC)

Miscellaneous cytostatics.

1. TEMSIROLIMUS (CCI-779, TORISEL)
2. BORTEZOMIB (VELCADE) :Proteasomes]
3. Vorinostat
4. ASPARAGINASE (L-ASPARAGINASE, ELSPAR, L-ASNASE, CRISTANASPASE)
5. ESTRAMUSTINE PHOSPHATE SODIUM (EM, EMCYT)

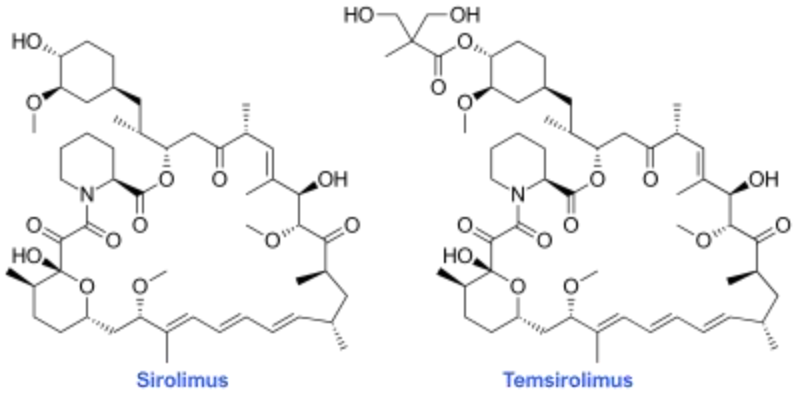
MISCELLANEOUS COMPOUNDS

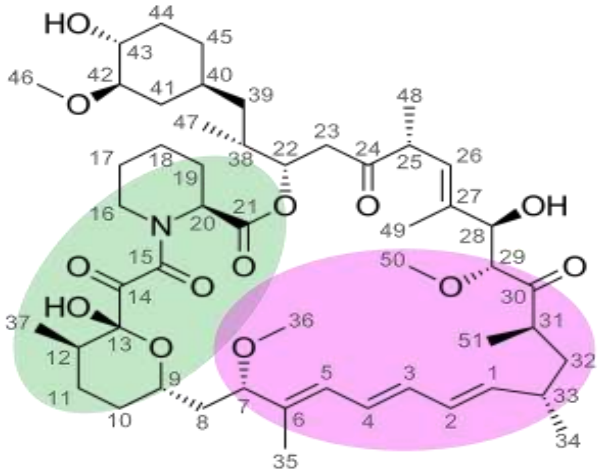
1. TEMSIROLIMUS (CCI-779, TORISEL)

Temsirolimus is an esterified derivative of rapamycin and in a similar manner binds initially to the protein .

Temsirolimus is a prodrug of rapamycin

It acts to inhibit the mammalian target of rapamycin (mTOR), a serine-threonine kinase that plays a crucial role in cell division. **It is somewhat unique in its method of kinase inhibition, because it actually binds to an allosteric modulator of the kinase rather than just binding to the ATP-binding site like most other kinase inhibitors**. Binding of temsirolimus inhibits the phosphorylating activity of mTOR





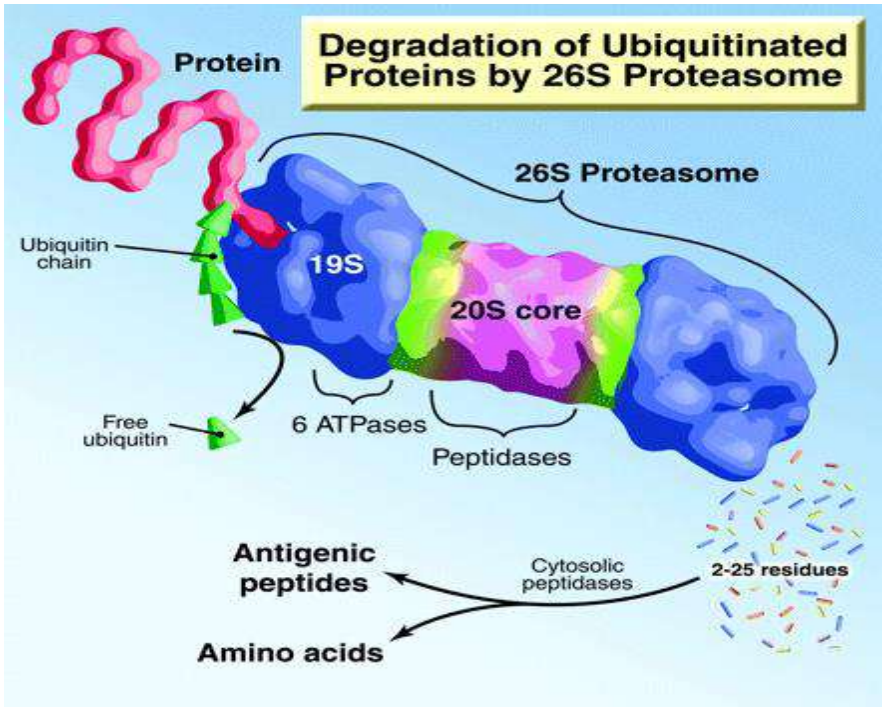
Binding sites of rapamycin: Green ring (pipecolate region) and purple ring represents binding region to mTOR

2. BORTEZOMIB (VELCADE)]

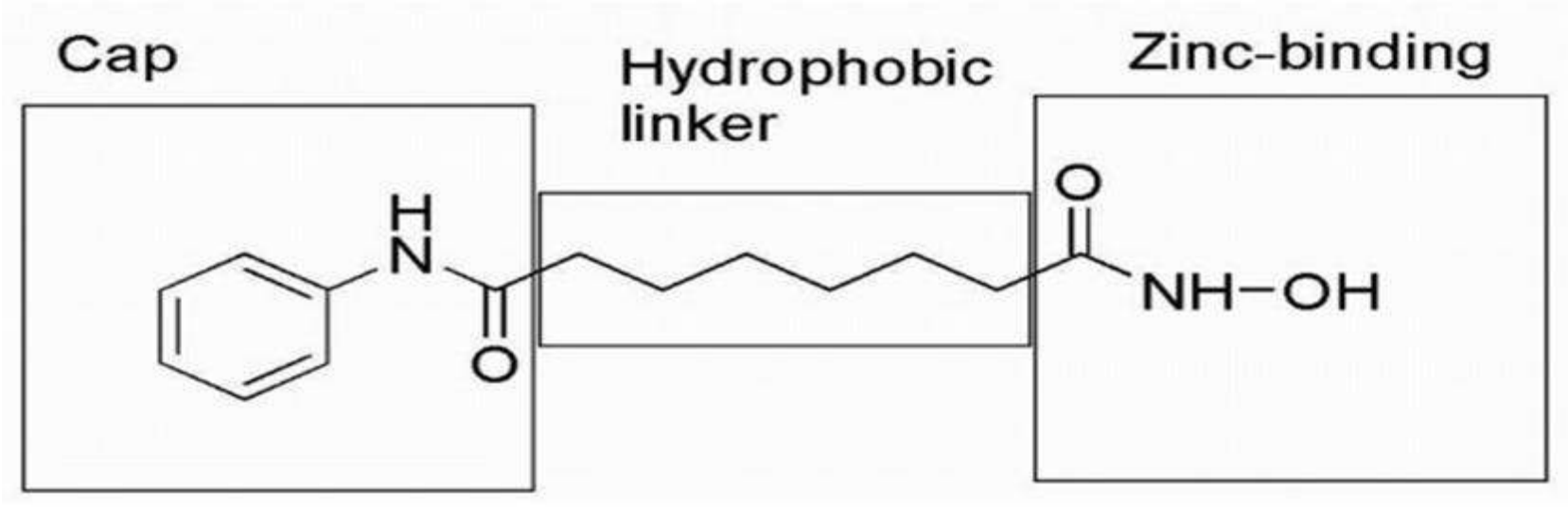
Proteasomes normally function to degrade proteins that are no longer needed by the cell. Such proteins are normally marked by the addition of ubiquitin, a 76 amino acid protein that is added to the ε-amino group of lysine residues on the target proteins.

The marked proteins are then hydrolyzed by the large barrel-shaped proteasomes to give peptides of 7 to 8 residues that may be further hydrolyzed and reutilized by the cell.

This process serves to regulate protein levels within the cell, remove defective proteins, and becomes important in maintaining normal signal transduction. Inhibition of the proteasomes results in the build up of ubiquitylated proteins, which disrupts cell-signaling processes and cell growth (Fig.)



3. Vorinostat fits the basic pharmacophore for the HDAC is (Fig.), which consists of a hydrophobic cap region connected to a zinc coordinating functionality by a hydrophobic linker.

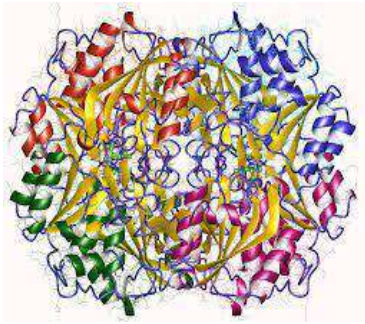


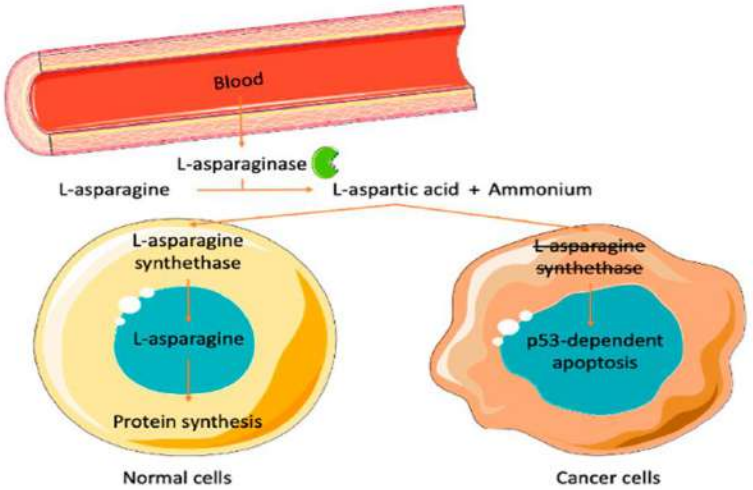
hydroxamate histone deacetylase inhibitors (Vorinostat). The structural design consists of a capping group, a carbon linker and a metal binding moiety

4. ASPARAGINASE (L-ASPARAGINASE, ELSPAR, L-ASNASE, CRISTANASPASE)

Asparaginase is available in 10-mL vials for intramuscular and IV use in the treatment of **acute lymphocytic leukemia**. Tumor cells are unable to synthesize asparagine, and therefore must utilize what is available in the extracellular environment.

The agent acts by hydrolyzing extracellular asparagine to aspartate and ammonia. The tumor cells are then **deprived of a necessary nutrient, and protein synthesis is inhibited leading to cell death.** The agent is specific for the G1 phase of the cell cycle. Resistance occurs because of the development of the tumor cells ability to **produce asparagine synthetase that allows them to synthesize the required amino acid.**

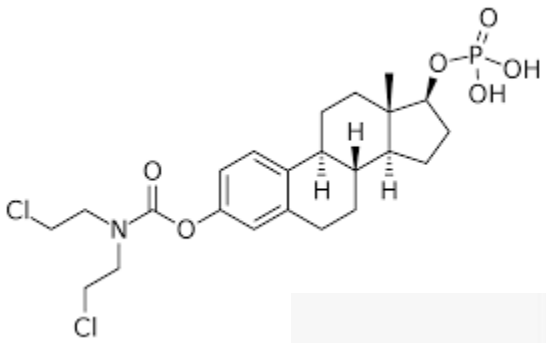


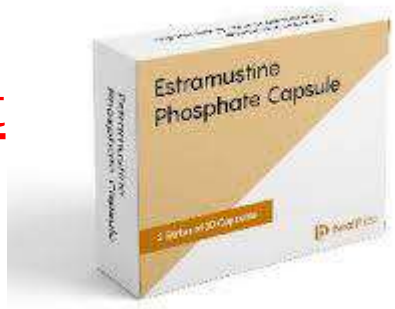


5. ESTRAMUSTINE PHOSPHATE SODIUM (EM, EMCYT)

Estramustine as the phosphate is available in **140-mg capsules** for the treatment of prostate cancer.

Although originally designed as an alkylating agent, it has been shown to be devoid of alkylating activity and functions as an inhibitor of **microtubule function by binding to microtubule associate proteins (MAPs) and also binds to tubulin at a site that is distinct from that of the vinca alkaloids but thought to partially overlap with that of pacilataxel**.





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5. ESTRAMUSTINE PHOSPHATE SODIUM (EM, EMCYT)

- **Sex hormones and analogues**

1. Glucocorticoids: Prednisolons and others
2. Estrogens
3. Selective estrogen receptor modulators
4. Selective estrogen receptor down modulators; Fulvestrant
5. Aromatase inhibitors : Aminoglutethamide
6. Antiandrogen : Flutamide , Bicalutamide
7. GnRH analogue ; Nafarellin, Leuprorelin, triptorelin
8. Progestins : Medroxyprogesterone acetate, hydroxyprogesterone caproate and megestrol

- **Monoclonal antibody conjugates (Antibody-drug conjugates ADC)**

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2nd line hormonal therapy for metastatic hormone dependent breast ca and endometrial cancer.

Hydroxyprogesterone — used in metastatic endometrial Cancer.

A/E: bleeding

9. GnRH antagonists — Cetorelix, Ganirelix, Abarelix

Cetrorelix, ganirelix and abarelix are antagonist of GnRH decrease the release of gonadotropins without causing initial stimulation can be used in prostatic cancer without the risk of flare up reaction.

Sex Hormones and Analogues

It involves the manipulation of the **endocrine system through exogenous administration of specific hormones**, particularly steroid hormones, or drugs which inhibit the production or activity of such hormones.

Because steroid hormones are powerful drivers of gene expression in certain cancer cells, changing the levels or activity of certain hormones can cause certain cancers to cease growing, or even undergo cell death.

1. Glucocorticoids such as prednisolone and dexamethasone have marked inhibitory effects on lymphocyte proliferation.
Used in the treatment of leukaemias and lymphomas.
Their ability to lower raised intracranial pressure, and to mitigate some of the side effects of anticancer drugs, makes them useful as supportive therapy .
Examples

Generic Name	Brand Name
dexamethasone	Dexamethasone Intensol
hydrocortisone	Cortef
methylprednisolone	Medrol
prednisolone	Orapred, Pediapred, Prelone
prednisone	Prednisone Intensol

2. Estrogens

Physiological antagonists of androgens.

Thus used to antagonize the effects of androgens in androgen dependent prostatic cancer.

The agonist is occasionally used to treat prostate **cancer through suppression of testosterone production.**

Diethylstilbestrol and ethinyloestradiol are two oestrogens used clinically in the palliative treatment of androgen-dependent prostatic tumours.

The latter compound has fewer side effects. These tumours are also treated with **gonadotrophin-releasing hormone analogues.**

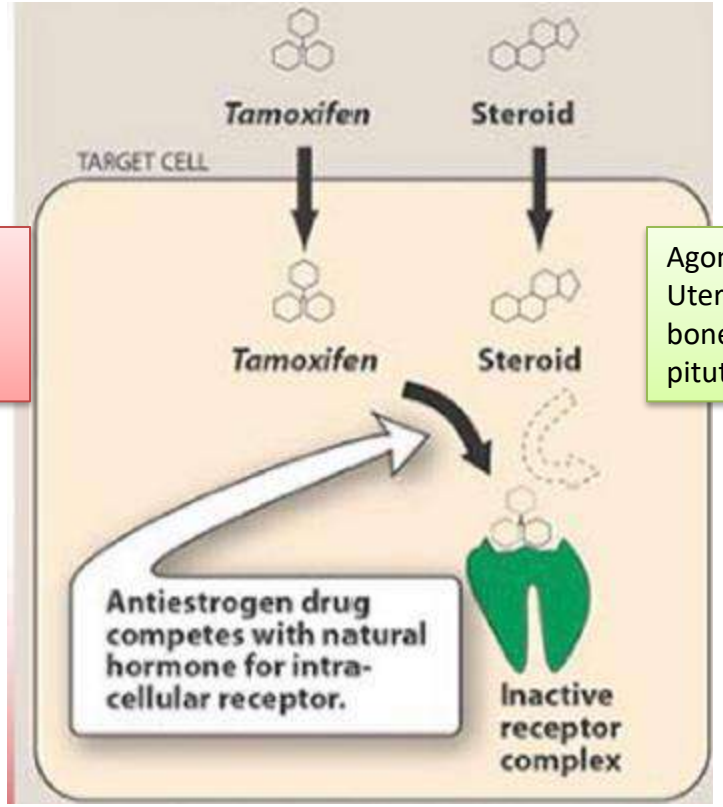
Oestrogens can be used to recruit resting mammary cancer cells into the proliferating pool of cells, thus facilitating killing by other cytotoxic drugs

3. Selective estrogen receptor modulators-

Tamoxifen, Toremifene

- ❑ Tamoxifen : Non steroidal antiestrogen

Antagonistic: Breast and blood vessels



Agonistic: Uterus, bone, liver, pitutary

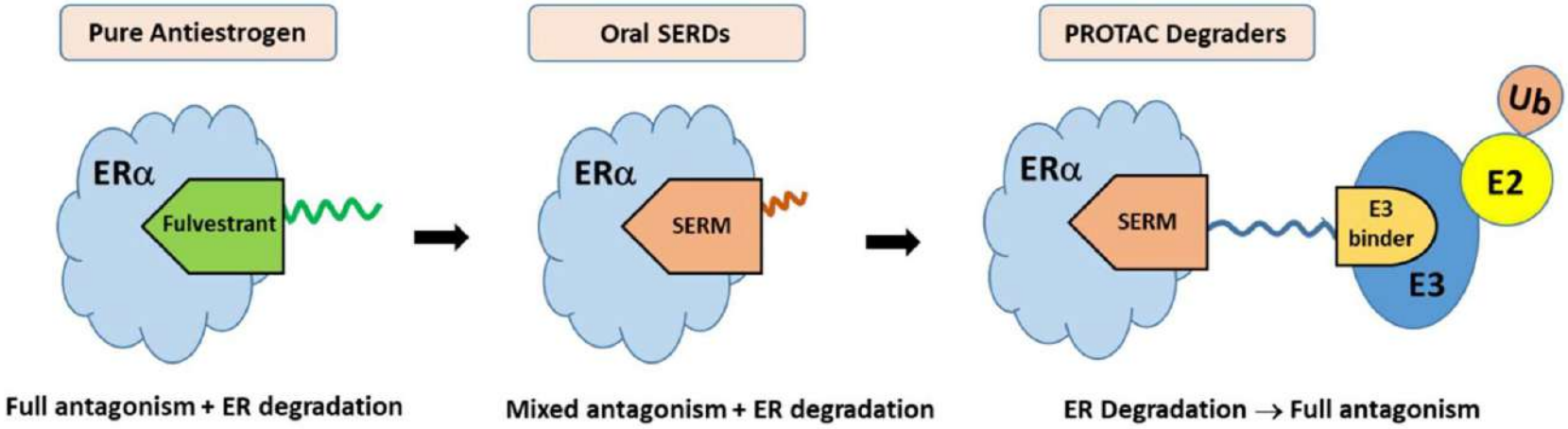
4. Selective estrogen receptor down regulators- Fulvestrant

- Pure estrogen antagonist

Uses: Metastatic ER+ Breast Cancer in postmenopausal women(**oestrogen receptor (ER)-positive breast tumours)**

MOA:

- Inhibits ER dimerization & prevents interaction of ER with DNA
- ER is down regulated resulting in more complete supression of ER responsive gene function.



5. Aromatase Inhibitors

* Aromatase is the enzyme responsible for conversion of **androstenedione** (androgen precursor) to estrone (estrogenic hormone).

- 1° gen.- Aminoglutethimide
- 2° gen.- Formestane, Fadrozole, Rogletimide
- 3° gen.- Exemestane, Letrozole,Anastrozole

© Aromatase inhibitors (Als) are a class of drugs used in the treatment of breast cancer and ovarian cancer in postmenopausal women.

© As breast and ovarian cancers require estrogen to grow, Aromatase inhibitors are **taken to either block the production of estrogen or block the action of estrogen on receptors.**

Aminogluthethimide-

Mechanism of Action:

- Inhibitor of adrenal steroid synthesis at the first step, conversion of cholesterol of pregnenolone.
- Inhibits the extra-adrenal synthesis of estrone and estradiol.
- Inhibits the enzyme aromatase that converts androstenedione to estrone.

ADR: Dizziness, Lethargy, Visual blurring, Rash

Therapeutic Uses: ER- and PR-positive metastatic breast cancer

6. Antiandrogens — Flutamide, Bicalutamide

- Antiandrogens, or **androgen antagonists**, first discovered in the 1960s, prevent androgens from expressing their biological effects on responsive tissues.
- Antiandrogens alter the androgen pathway **by blocking the appropriate receptors**, **competing for binding sites on the cell's surface, or affecting androgen production.**
- Antiandrogens are most frequently used to treat prostate cancer.

7. GnRH analogues — Nafarelin, Leuprorelin, triptorelin

NAFERELIN : nasal spray / SC inj

JFSH & LH release from pituitary- | the release of estrogen & testosterone.

USE : Breast Ca, Prostatic Cancer

8. Progestins: Medroxyprogesterone acetate, hydroxyprogesterone

caproate and megestrol . 2nd line hormonal therapy for metastatic hormone dependent breast ca

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Hydroxyprogesterone — used in metastatic endometrial Cancer.

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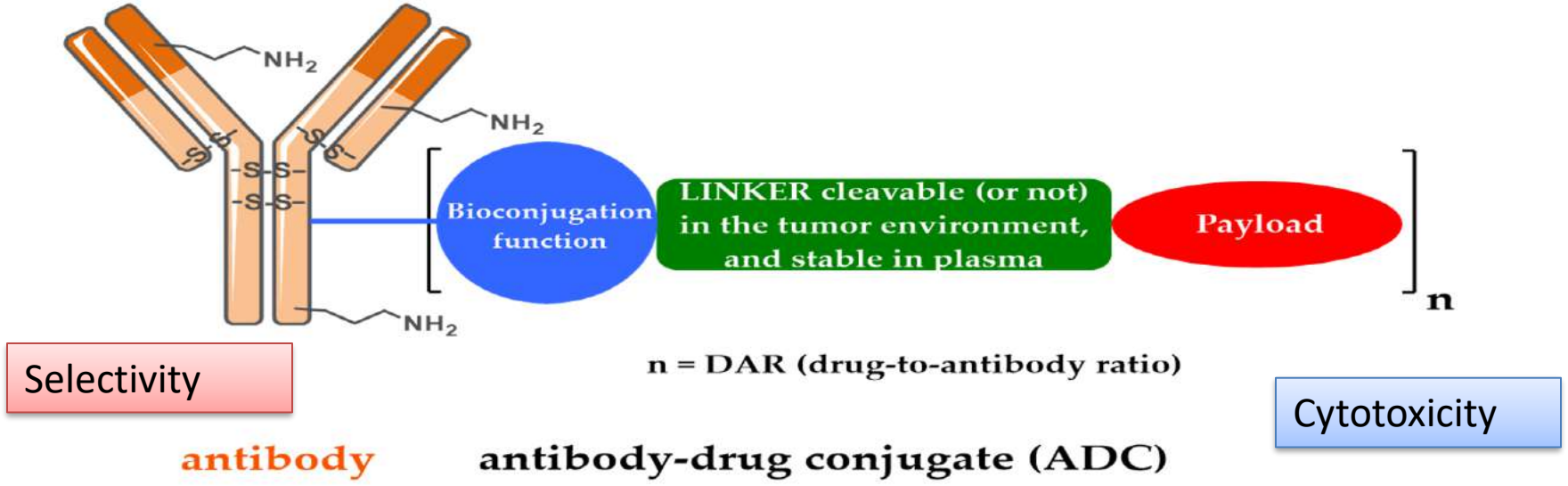
release of gonadotropins without causing initial stimulation can be used in prostatic ca without the risk of flare up reaction.

Antibody-drug conjugates

Antibody-drug conjugates or ADCs are a new class of drugs designed as a targeted therapy for the treatment of cancer.

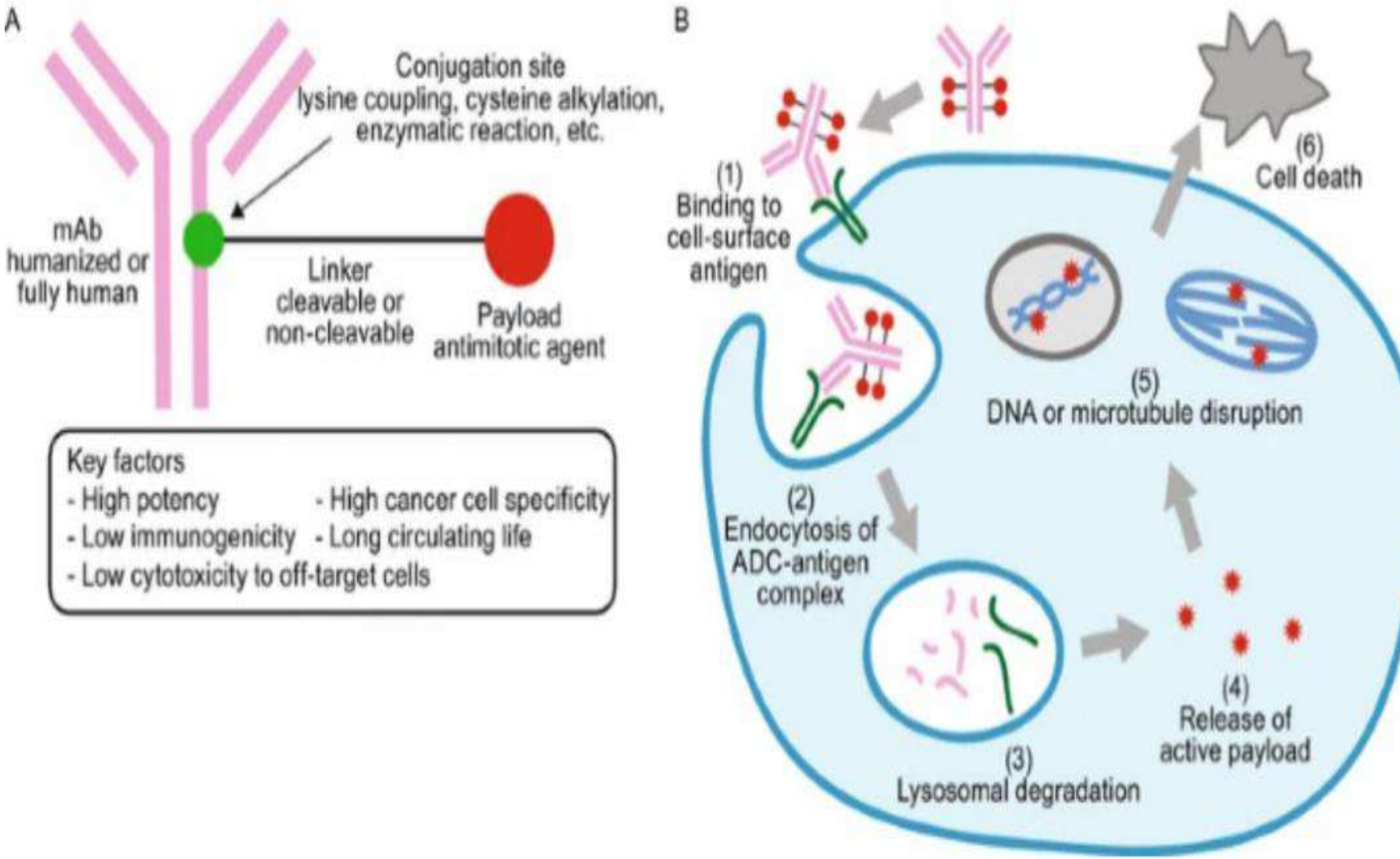
ADCs are complex molecules composed of an antibody linked, via a stable linker with labile bonds, to a cytotoxic (anticancer) drug.



Selectivity

Cytotoxicity

Structure and mechanism of action of ADCs



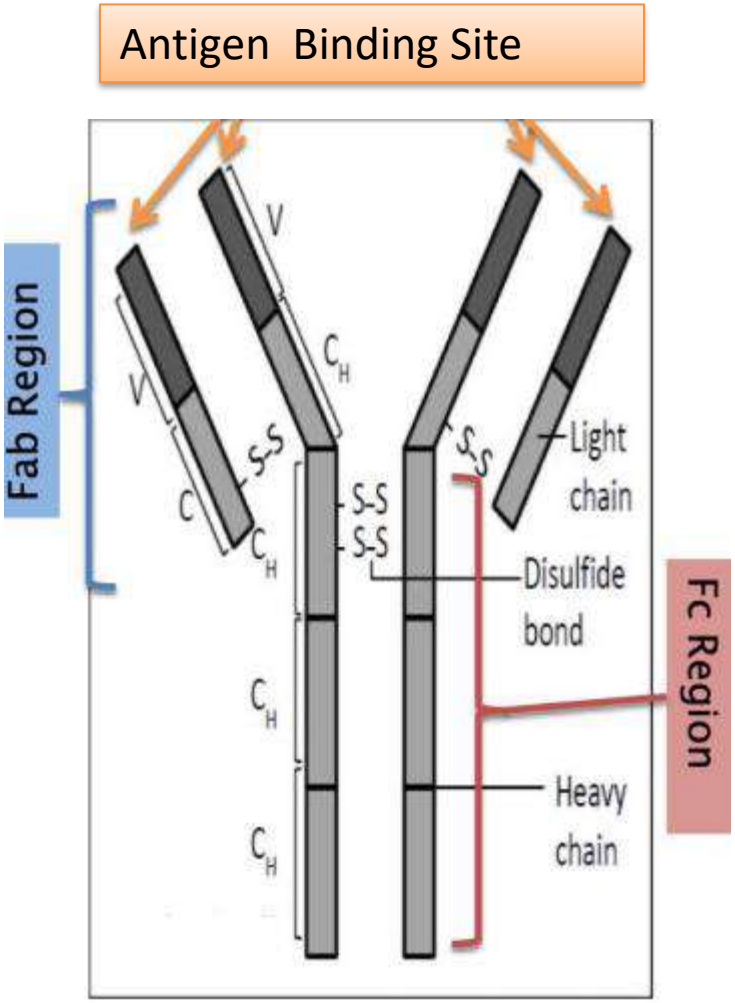
Antibodies are immunoglobulins made up of:

2 Light Chains (identical)

~25 KDa

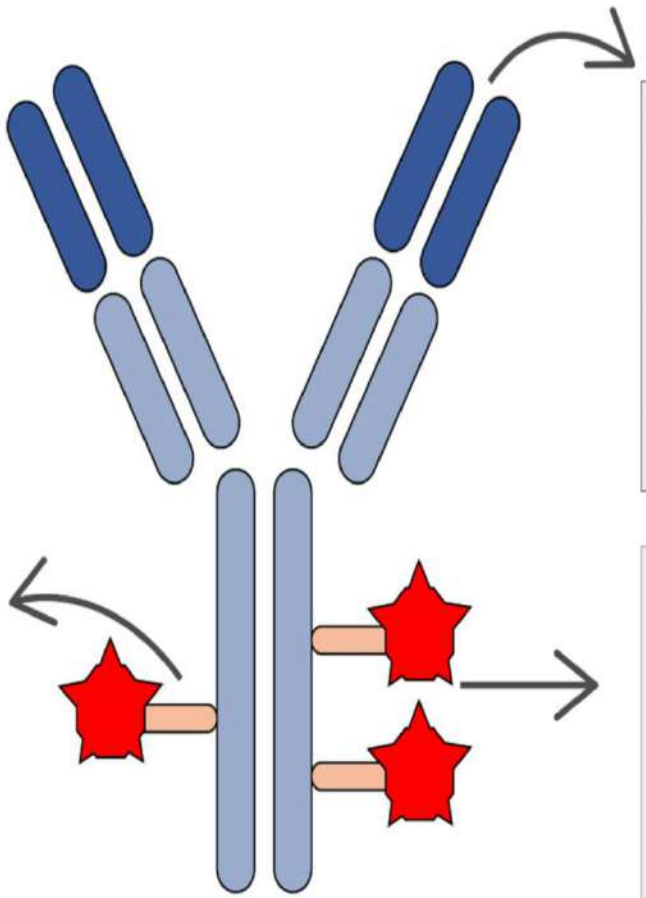
2 Heavy Chains (identical)

~50 KDa



From a biology perspective, the design of an effective ADC relies on :

Target antigen
-Tumor-specific and homogeneous expression pattern
-High levels of expression,
-Rapid internalization
-Minimal ectodomain shedding
-Non-internalizing ADCs challenge internalization as a strict property of target antigens



Antibody
-High specificity and affinity for the target antigen
-Favorable PK properties
-Minimal immunogenicity
-Can elicit effector functions
-The majority of ADCs employ human/humanized IgG1 mAbs
-Bispecific antibodies can be used for a more target-specific drug delivery

Linker
-Designed to covalently tether the cytotoxic molecule to the antibody scaffold
-Affects several physicochemical parameters ranging from stability in systemic circulation, to solubility and aggregation propensity
-Generally categorized into cleavable and non-cleavable

Payload
-The ultimate effector compound
-Super-toxic
-The main categories are: DNA-damaging agents and microtubule-disrupting agents
-Payload classes of FDA-approved ADCs include: Calicheamicins, Auristatins, Maytansinoids, Pyrrolobenzodiazepines and Camptothecin analogues
-Dual-drug ADCs achieve codelivery of warheads with different mechanism of action

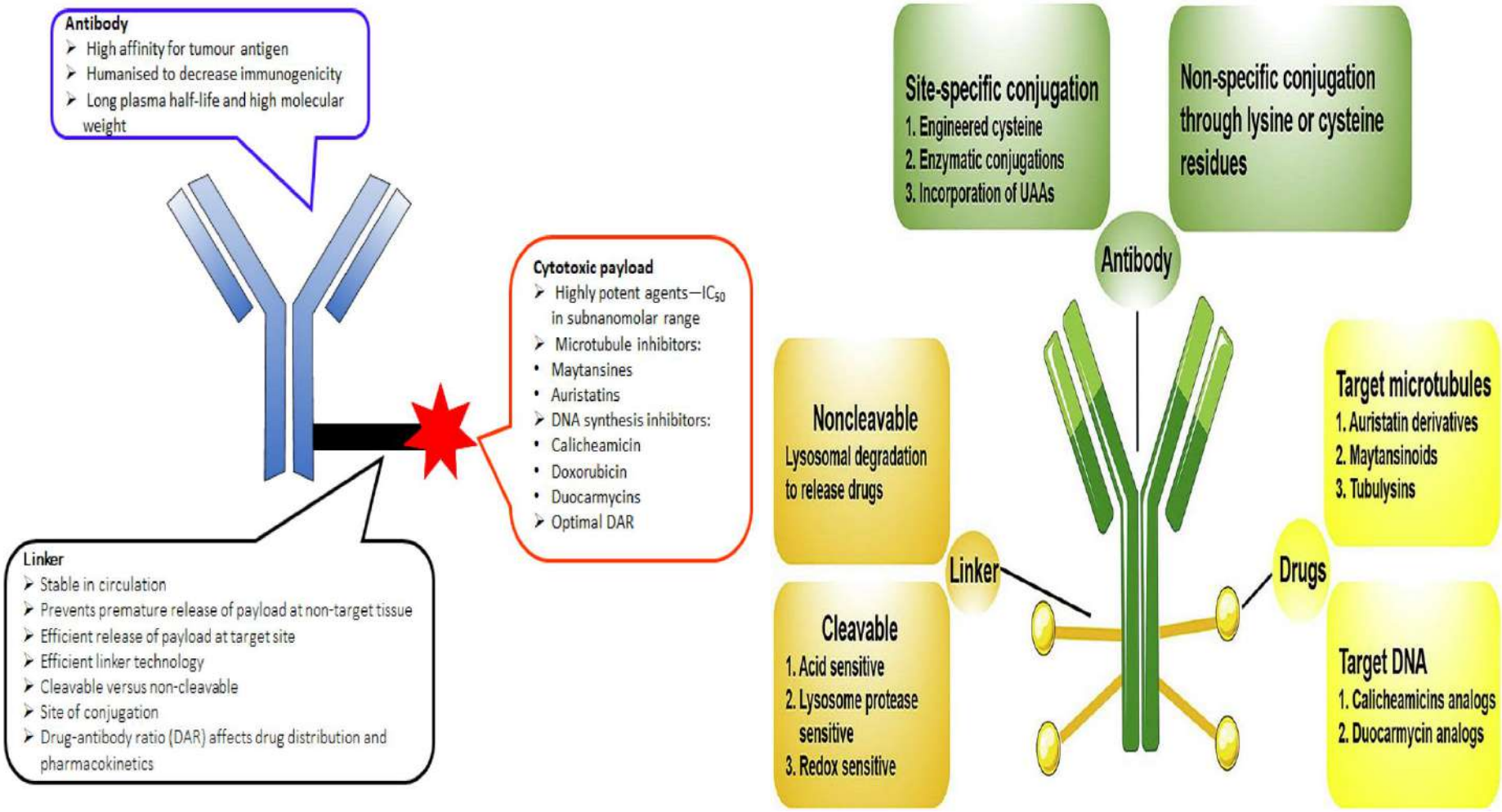
Target antigens

A successful ADC should target a well-internalized antigen with **low normal tissue expression and high expression on tumors.**

Antigen expression on normal tissues can be tolerated if expression on vital organs is minimal or absent.

Target antigens for ADCs in preclinical & clinical development	
Cancer	Target Antigens
Breast	CD174, GPNMB, CRIPTO & nectin-4 (ASG-22ME)
Ovarian	MUC16 (CA125), TIM-1 (CDX-014) & mesothelin
Lung	CD56, CD326, CRIPTO, FAP, mesothelin & GD2
Pancreatic	CD74, CD227 (MUC-1) & nectin-4 (ASG-22ME)
Prostate	PSMA, STEAP-1 & TENB2

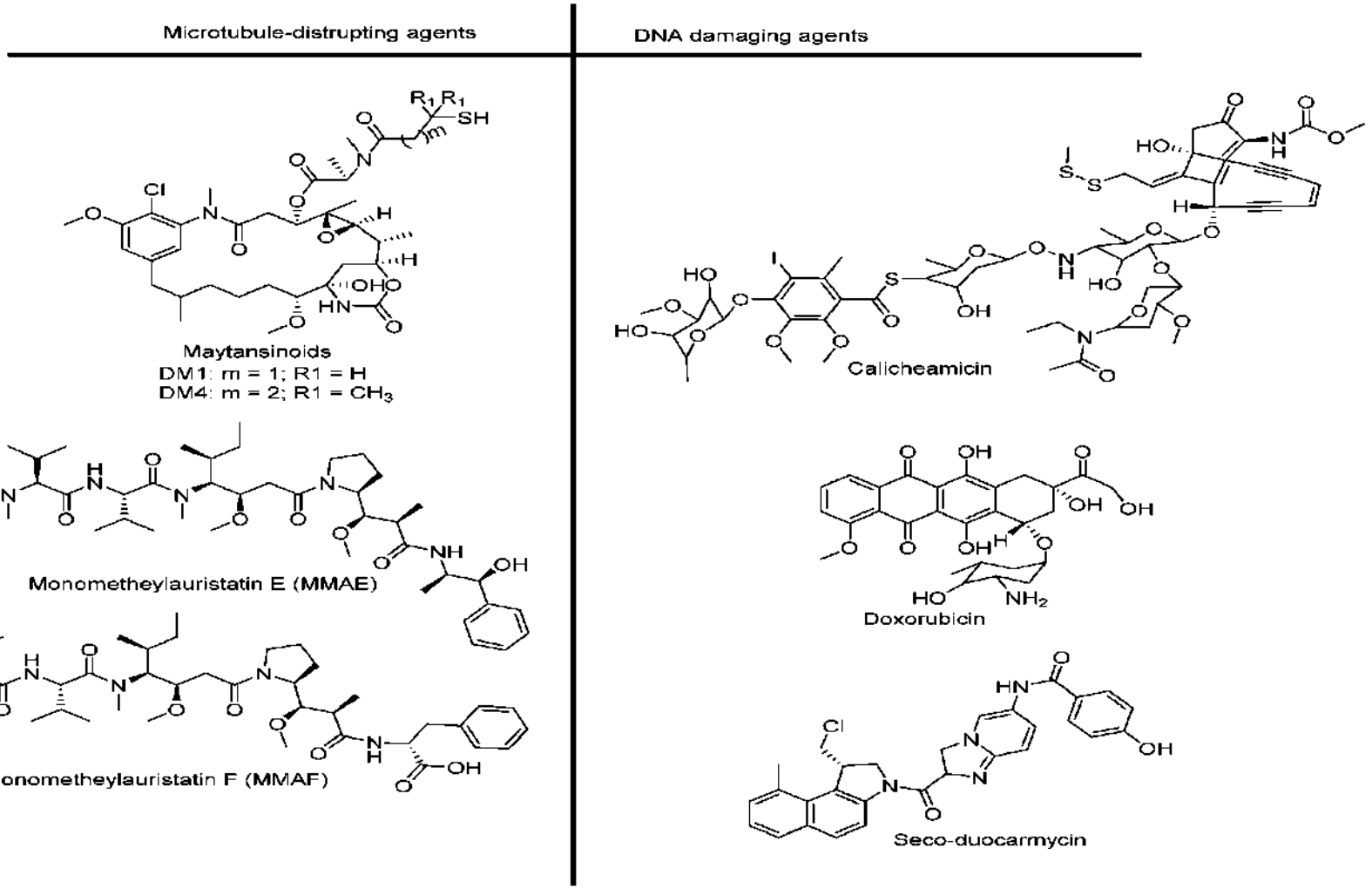
Schematic diagram of an antibody-drug conjugate (ADC).



Cytotoxic drugs used in ADC design

The drugs being used to construct ADCs generally fall into two categories:

1) Microtubule inhibitors
2) DNA-damaging agents



ADC advantages over Traditional Chemotherap

Traditional Chemotherapy

ADC

Damages the healthy cells

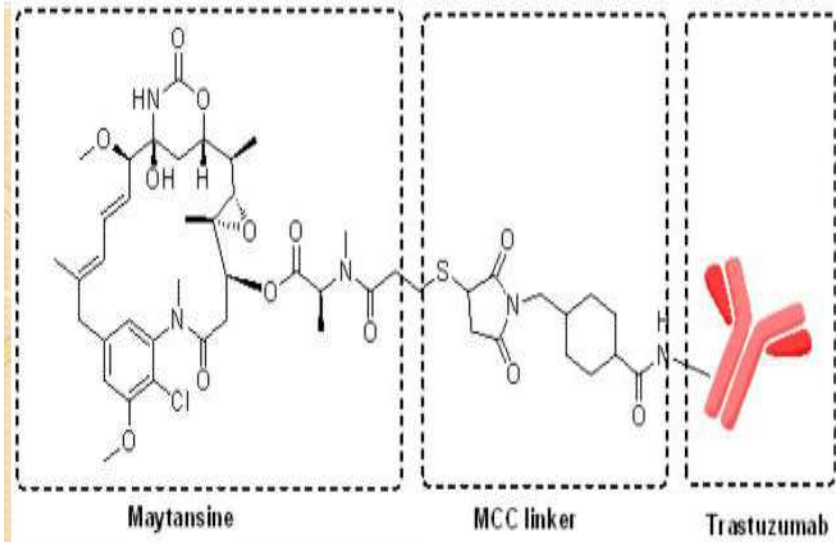
Rapid clearance

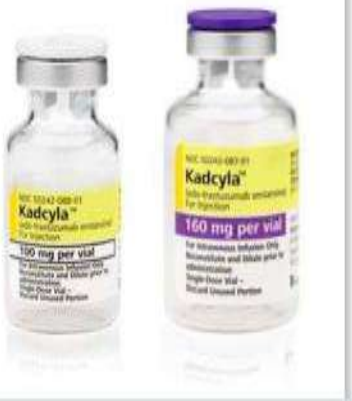
High binding affinity

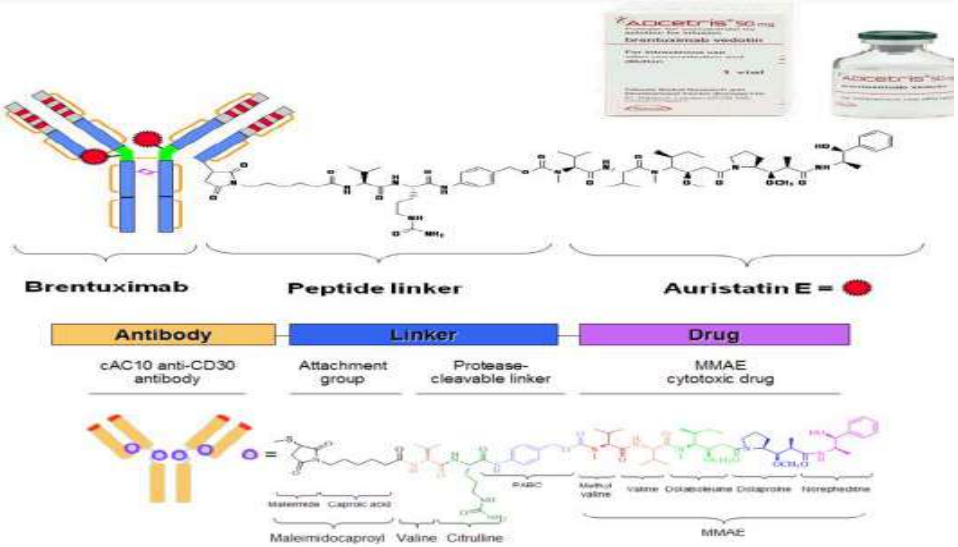
Specificity for tumor cells

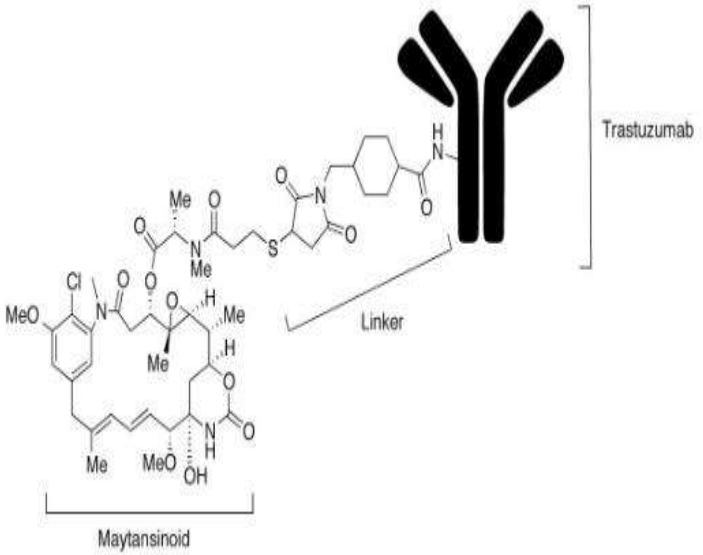
Long half-life

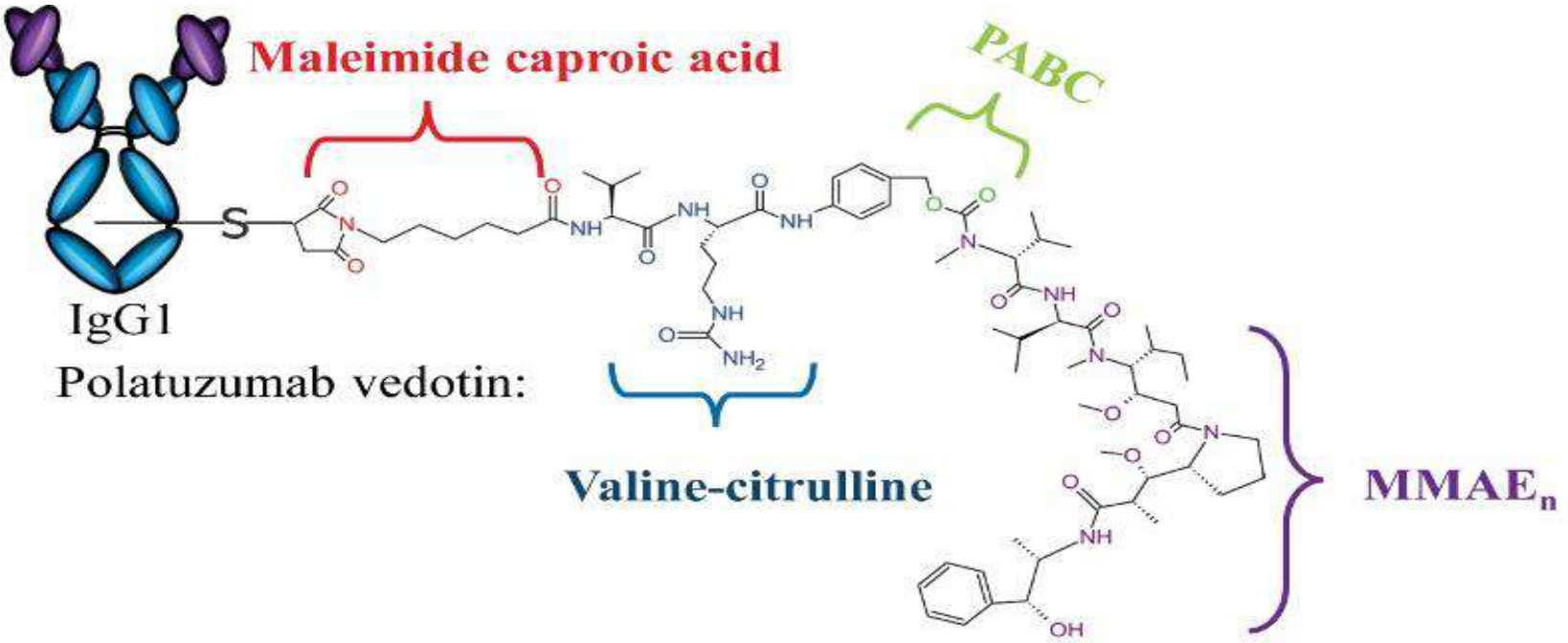
Agent	Linker	Warhead	Target
IMMU-110	Hydrazone	Doxorubicin	CD74
Mylotarg®	Hydrazone	Calicheamicin	CD33
CMC-544	Hydrazone	Calicheamicin	CD22
SAR3419	Disulfide	DM4	CD19
BT-062	Disulfide	DM4	CD138
BAY-94-9343	Disulfide	DM4	Mesothelin
SAR-566658	Disulfide	DM4	DS6
IMGN901	Disulfide	DM1	CD56
Kadcyla®	Thioether	DM1	HER2
IMGN529	Thioether	DM1	CD37
SGN-75	MC	MMAF	CD70
Adcetris®	Peptide (Val-Cit)	MMAE	CD30
RG-7596	Peptide (Val-Cit)	MMAE	CD79b
CDX-011	Peptide (Val-Cit)	MMAE	GPNMB
PSMA-ADC	Peptide (Val-Cit)	MMAE	PSMA
ASG-5ME	Peptide (Val-Cit)	MMAE	AGS-5
IMUU-130	Peptide (Phe-Lys)	SN-38	CEACAM5






Brentuximab
Peptide linker
Auristatin E =
Antibody
Linker
Drug
cAC10 anti-CD30 antibody
Attachment group
Protease-cleavable linker
MMAE cytotoxic drug
Maleimidocaproyl
Valine
Citrulline
MMAE


Trastuzumab
Linker
Maytansinoid

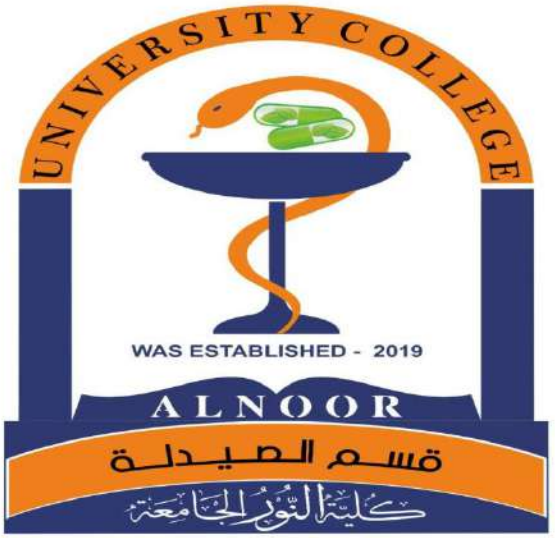

Maleimide caproic acid
PABC
IgG1
Polatuzumab vedotin:
Valine-citrulline
MMAE$_n$

Organic Pharm. Chemistry III

Lecture 3

12th March 2023

10:30- 12:30



AlNoor University College

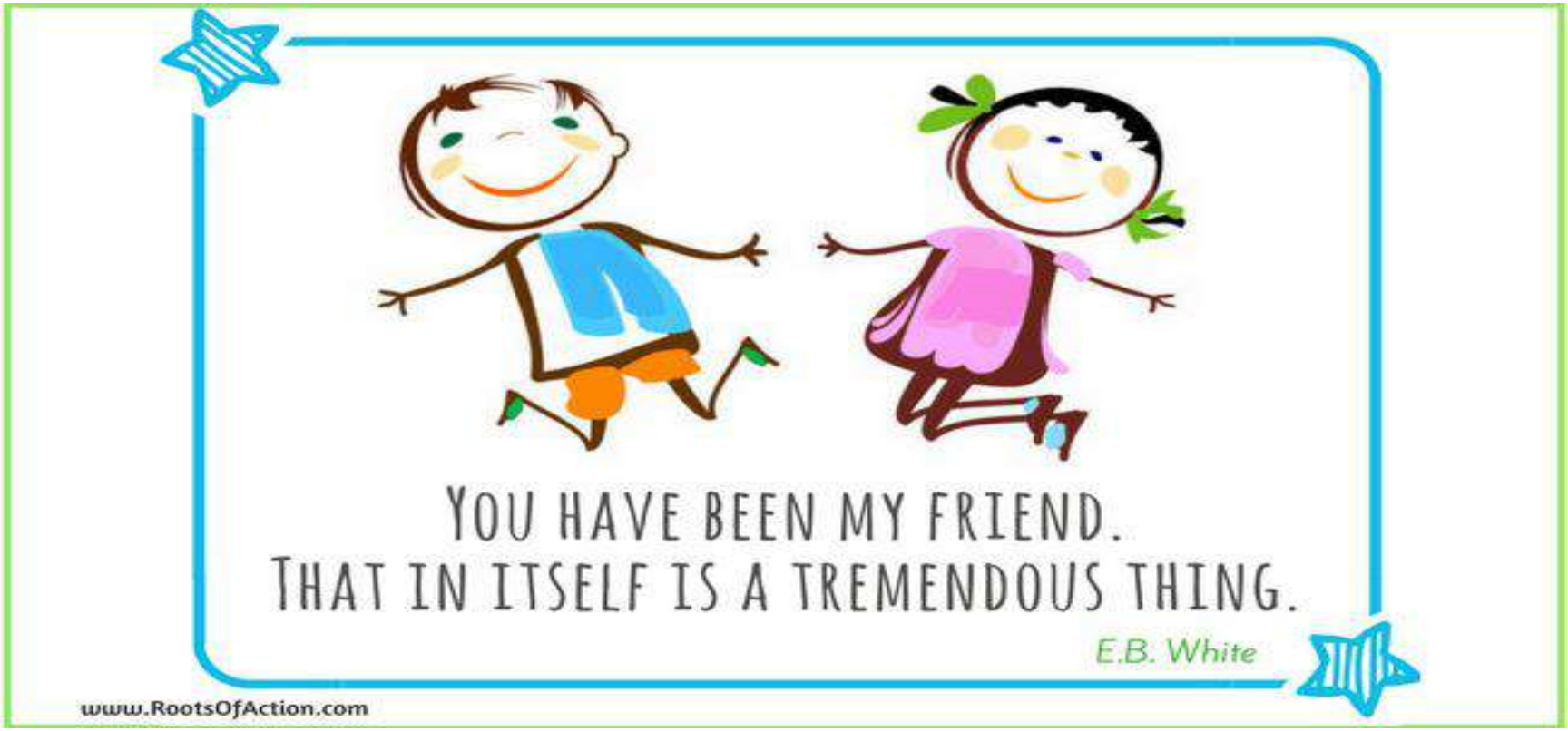
Pharmacy Department

2nd Course/ 4th Class

2022/2023

However difficult life may seem, there is always something you can do and succeed at."

- Stephen Hawking. ...



History

The cephalosporins are β-Lactam antibiotics that are closely related both structurally and functionally to the penicillins.

Mechanism of action, mechanism of resistance and some other properties of cephalosporins are identical to penicillins)

Cephalosporins are one of the most widely used antibiotics and are equal in importance to penicillin.

The cephalosporins are isolated from:

- Cephalosprium species
- Prepared semisynthetically.

In 1945

Giuseppe Brotzu`s discovered that cultures of Cephalosporium acremonium inhibited the growth of a wide variety of Gram-positive and Gram-negative bacteria.

Discovered from a fungal colony in Sardinian sewer water (1948)

Cephalosporin C identified in 1961

In 1948

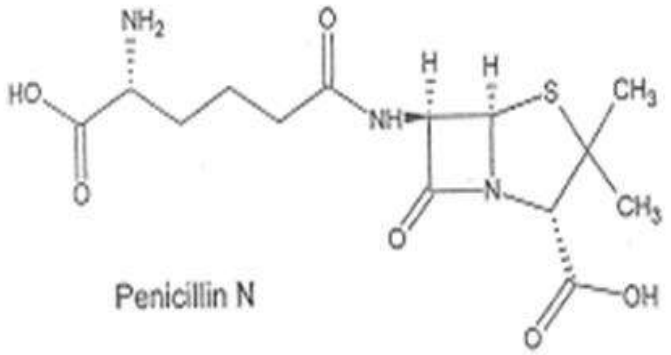
Abraham and his colleagues have been supplied cultures of the fungus and was isolated three principal antibiotic components:

- Cephalosporin P, (a steroid antibiotic that resembles fusidic acid) with minimal antibacterial activity.

- Cephalosporin N, later discovered to be identical with synnematin N (a penicillin derivative now called penicillin N)

- Cephalosporin C.

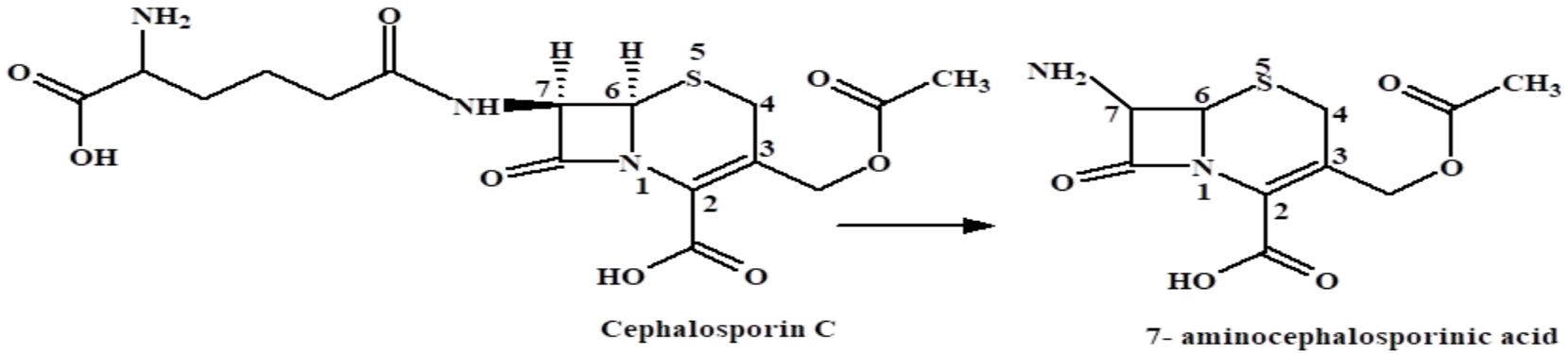
Penicillin N (Cephalosporin N)



Penicillin N

*Most of the antibiotics introduced since 1965 have been semisynthetic cephalosporins.

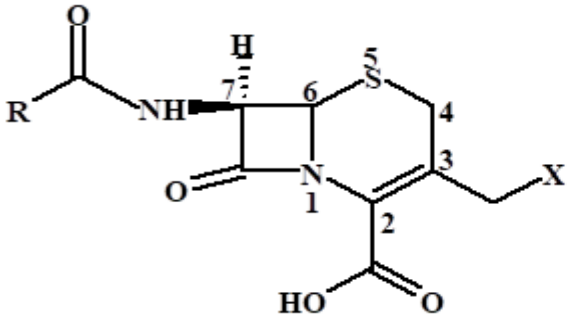
Cephalosporin C can be hydrolyzed by acid to 7-aminocephalosporanic acid.



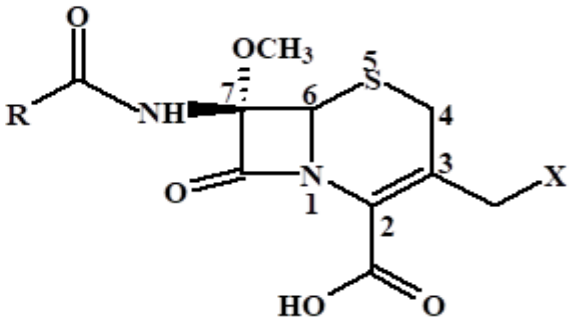
*Compounds containing 7-aminocephalosporanic acid are:

- Relatively stable in dilute acid.
- Highly resistant to penicillinase, regardless of the nature of their side chains and their affinity for the enzyme.

- Most cephalosporins are produced semisynthetically by the chemical attachment of side chains to 7-aminocephalosporanic acid.



Cephalosporins



Cephamycin

- Cephalosporins (7α-H) and cephamycins (7α-OCH3):
- Most natural cephalosporin and cephamycin are not used

clinically for side effects, but semi-synthetic products are used.

Objectives of pharmacomodulation:

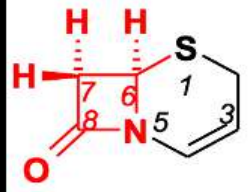
✓ Broaden the antibacterial spectrum,

✓ Increase resistance to -lactamases,

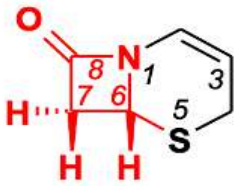
✓ Improve pharmacokinetics.

Structure of cephalosporins

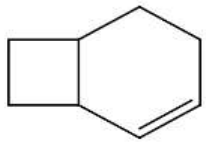
1. Nomenclature The different families of cephalosporins: according to the substitutions of the cepheme.



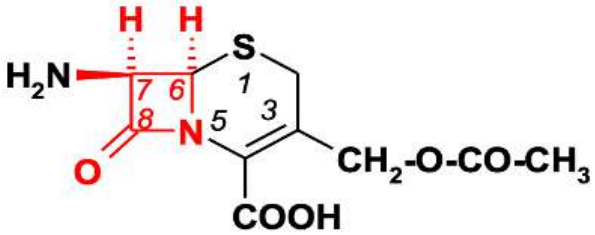
Δ3-cepheme



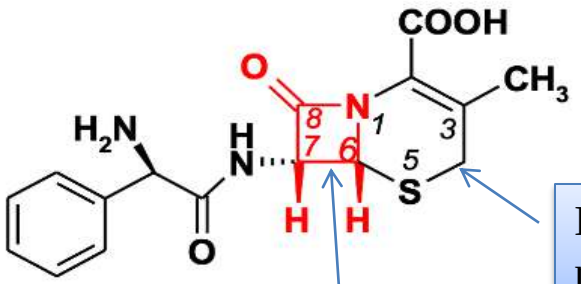
(6*R*)-5-thia-1-azabicyclo(4.2.0)oct-2-ene



bicyclo [4.2.0]oct-2-ene



7-aminocephalosporanic acid
7-ACA

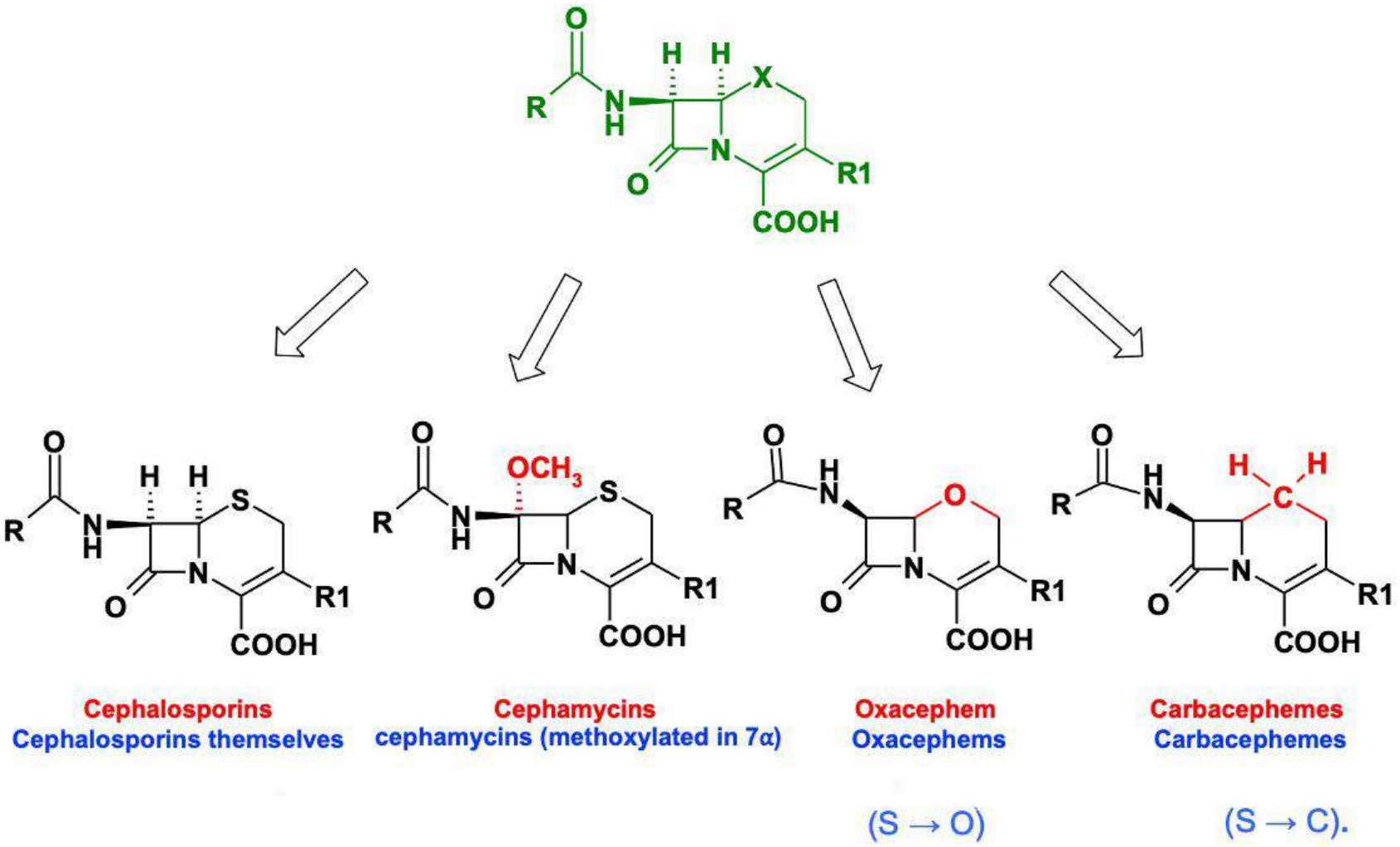


(6*R*,7*R*)-7-((*R*)-2-amino-2-(4-hydroxyphenyl)acetamido)
-3-methyl)-5-thia-1-azabicyclo(4.2.0)oct-2-ene

Dihydrothiazine ring

B- lactam ring

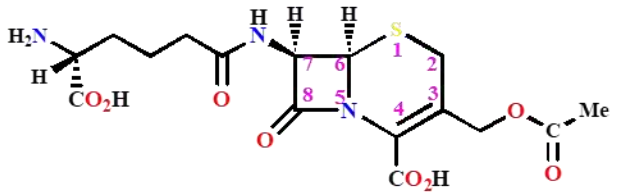
The different families of cephalosporins: according to the substitutions of cepheme.



Properties of Cephalosporin C

Disadvantages

- Polar due to the side chain - difficult to isolate and purify
- Low potency - limited to the treatment of urinary tract infections where it is concentrated in the urine
- Not absorbed orally



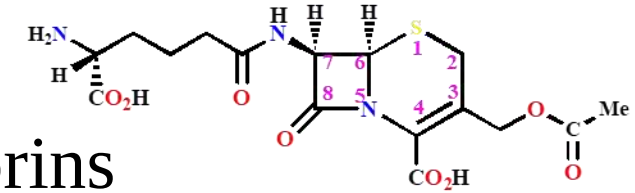
Advantages

- Non toxic
- Lower risk of allergic reactions compared to penicillins
- More stable to acid conditions
- More stable to b-lactamases
- Ratio of activity vs Gram -ve and Gram +ve bacteria is better

Conclusion

- Useful as a lead compound

SAR of Cephalosporins



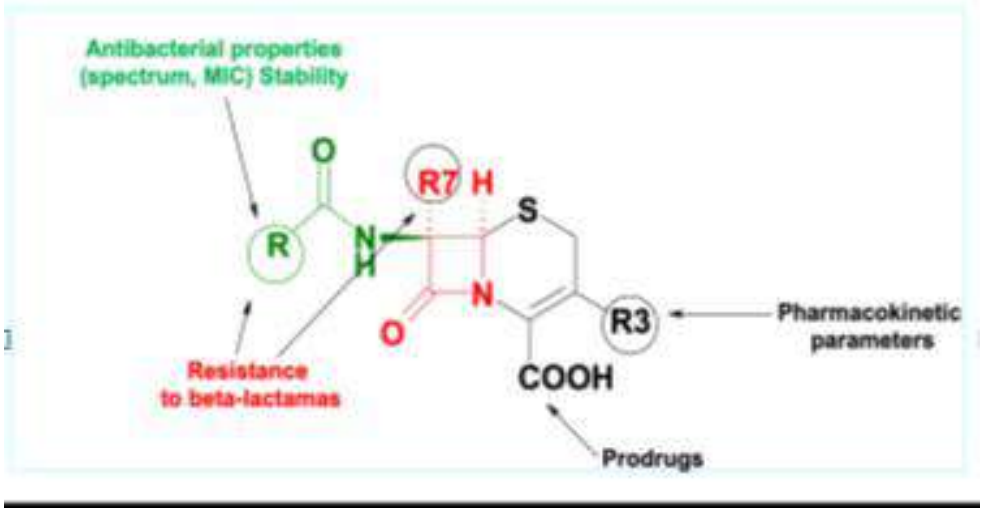
Cephalosporins

Similar to penicillins

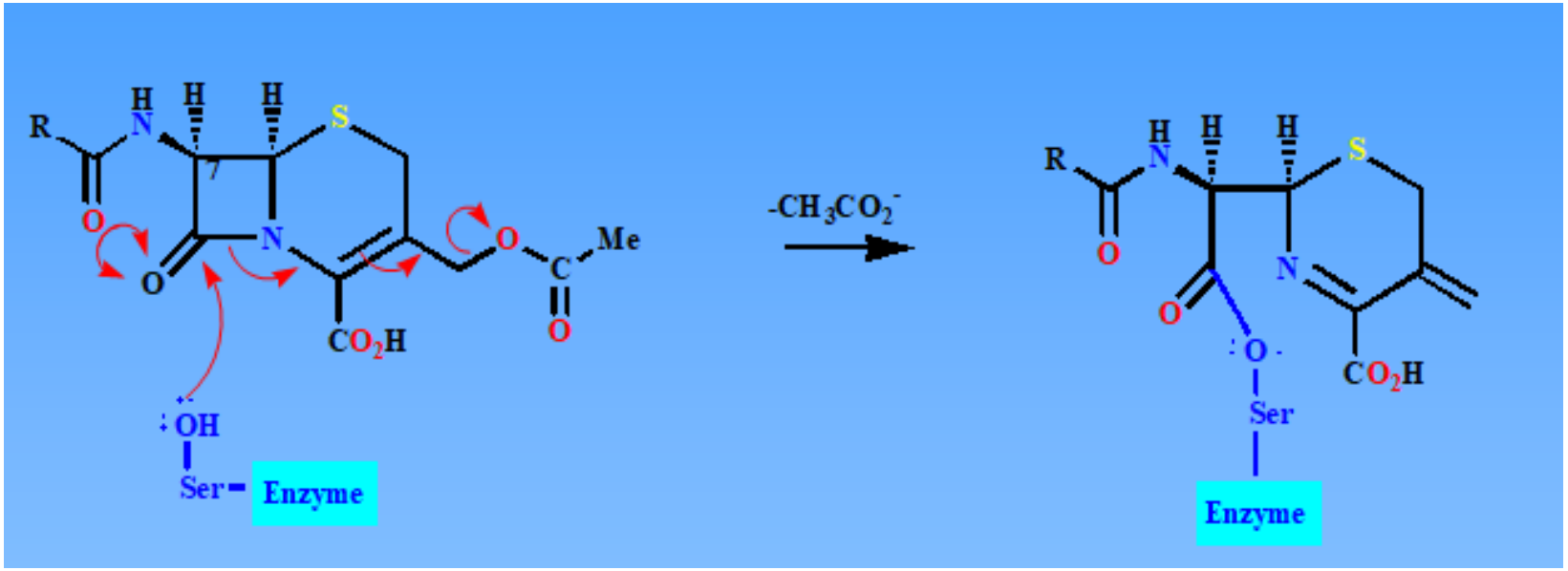
- The b-lactam ring is crucial to the mechanism
- The carboxylic acid at position 4 is important to binding
- The bicyclic system is important in increasing ring strain
- Stereochemistry is important
- The acetoxy substituent is important to the mechanism

Possible modifications

- 7-Acylamino side chain
- 3-Acetoxymethyl side chain
- Substitution at C-7



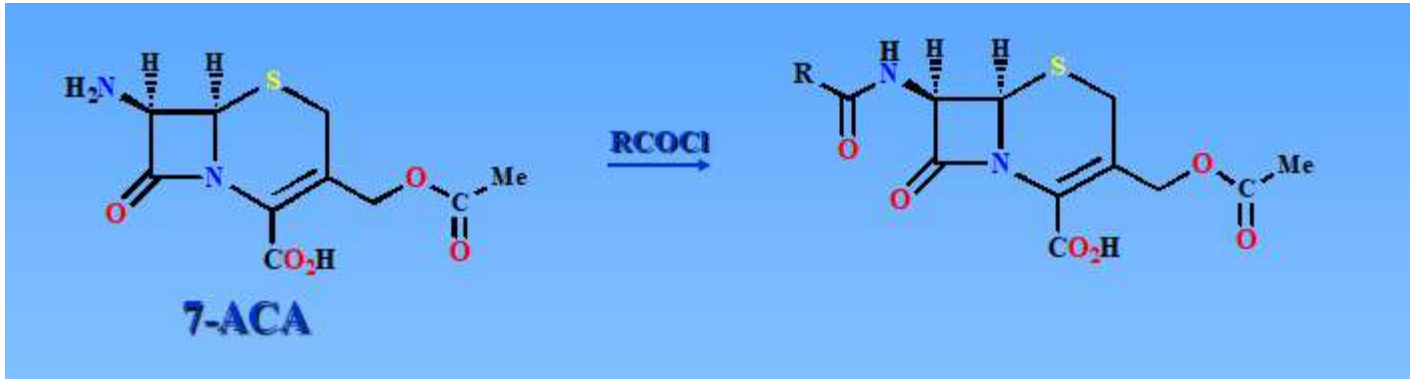
Mechanism of Action



The acetoxy group acts as a good leaving group and aids the mechanism

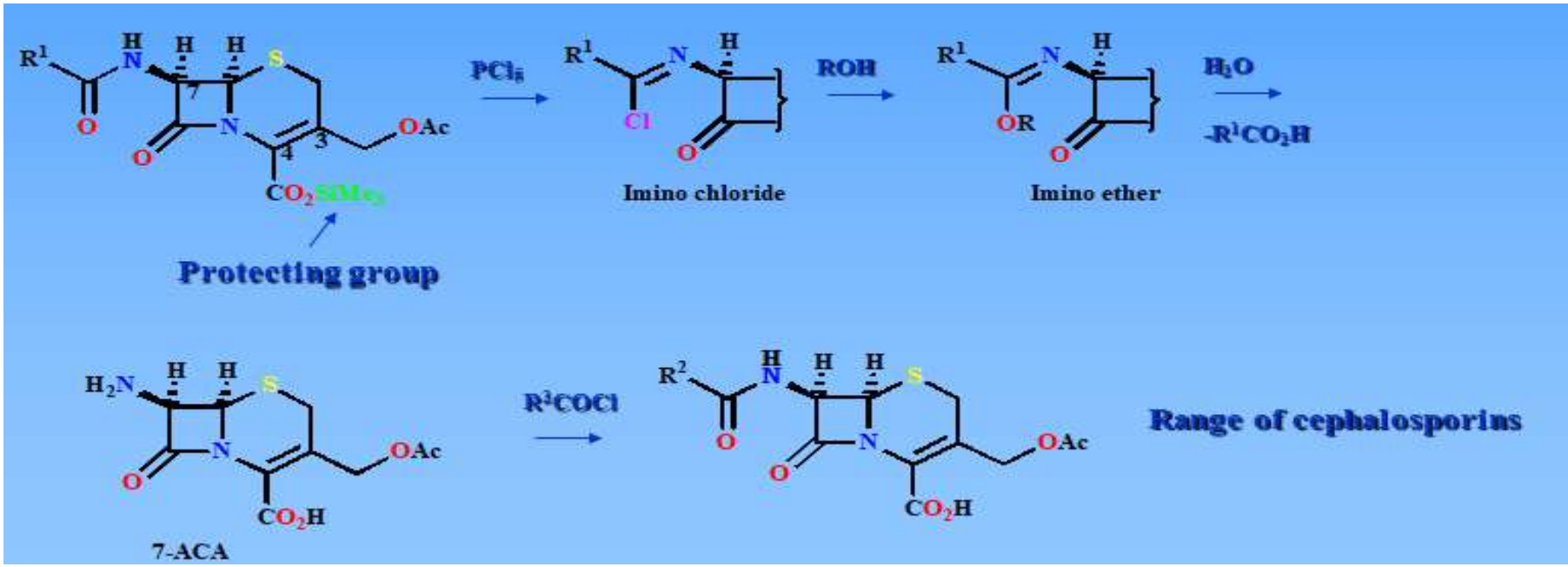
Variation of the 7-Acylamino Side Chain

- Not possible to generate analogues by fermentation
- Not possible to generate analogues by a full synthesis
- Restricted to semi-synthetic procedure



- 7-ACA not available by fermentation
- 7-ACA not available by enzymatic hydrolysis of cephalosporin C
- Generated by a chemical hydrolysis

- Generation of 7-ACA
- Need to hydrolyse a relatively unreactive secondary amide in the presence of a labile b-lactam ring



THE "GENERATIONS" OF CEPHALOSPORINS

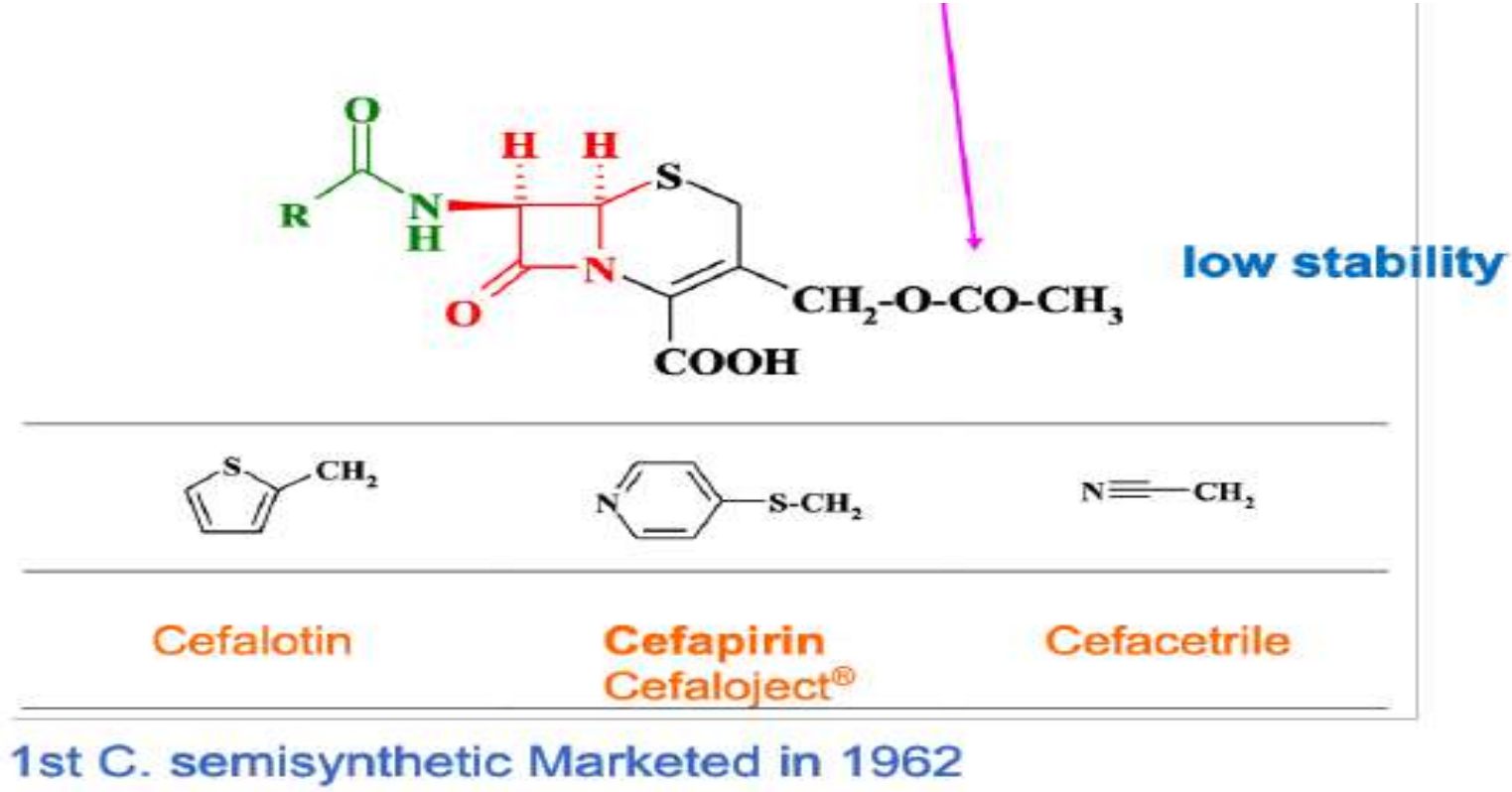
First generation cephalosporins

- Spectrum limited to Gram + cocci, mainly Staphylococci and S Streptococci, as well as some enterobacteria.
- Sensitivity to beta-lactamases, poor diffusion in the CSF.

The 3 groups of C1G

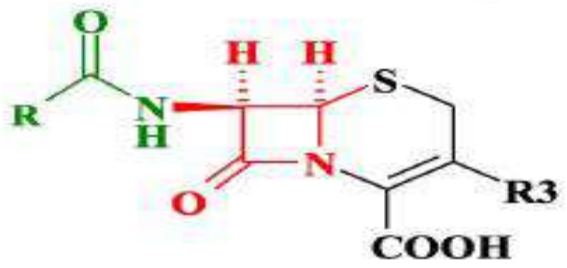
1st group: comprising an acetoxymethyl group in 3, hydrolyzed by esterases

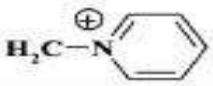
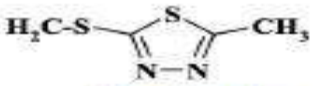
→ low stability.



2nd group:

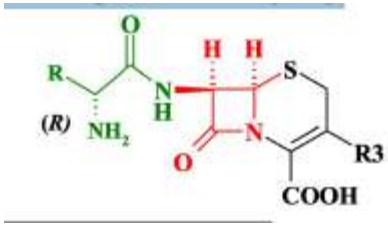
- comprising a heterocycle in 3 better stability therefore better pharmacokinetics.

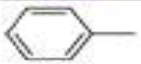
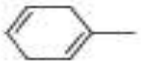
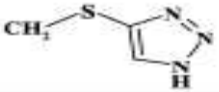


R	R3	DCI	ND
		Cefaloridin	
	 thiadiazole	Cefazolin	Cefacidal®

3rd group:

- with a a-amino side chain, contributing to activity by oral route



R	R3	DCI	ND
	CH_3	**Cefalexin** (anh ou 1 H_2O)	Cefacet® Ceporexine® Keforal®
	CH_3	**Cefadroxil** (1 H_2O)	Oracefal®
	CH_3	**Céfradin**	Dexef® Kelsef® Zeefra®
	Cl	**Cefaclor** (1 H_2O)	Alfatil® Alfatil ®
	 Thio-triazole	**Cefatrizine**	Cefaperos®

Useful antibacterial activity of C1G:

• Meti-S staphylococci

• Streptococci (except enterococci)

• Certain Enterobacteriaceae (not producing cephalosporinases):

Escherichia coli, Klebsiella

• Proteus mirabilis: MS or S

Therapeutic indications of C1G:

• ENT, respiratory, urinary, skin, acute or recurrent infections with sensitive germs

• In IM-IV (1st and 2nd group) / 4 - 8h: dosage = 2 - 12 g / day

• Oral (3rd group): dosage = 2 - 4 g / day

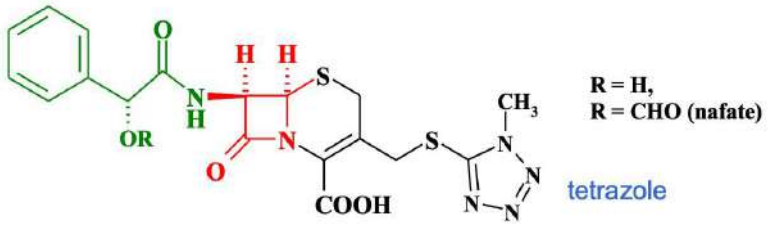
2nd GENERATION CEPHALOSPORINS

✓ Spectrum of activity of C1G but a little more extended for Enterobacteriaceae,

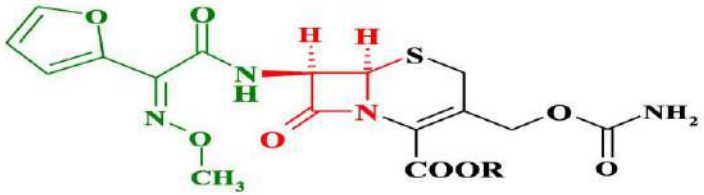
✓ Variable profile depending on the molecule.

STRUCTURES:

✓ **a-hydroxylated cephalosporin: cefamandole**



✓ Alkoxyiminated cephalosporin: cefuroxime and its esters

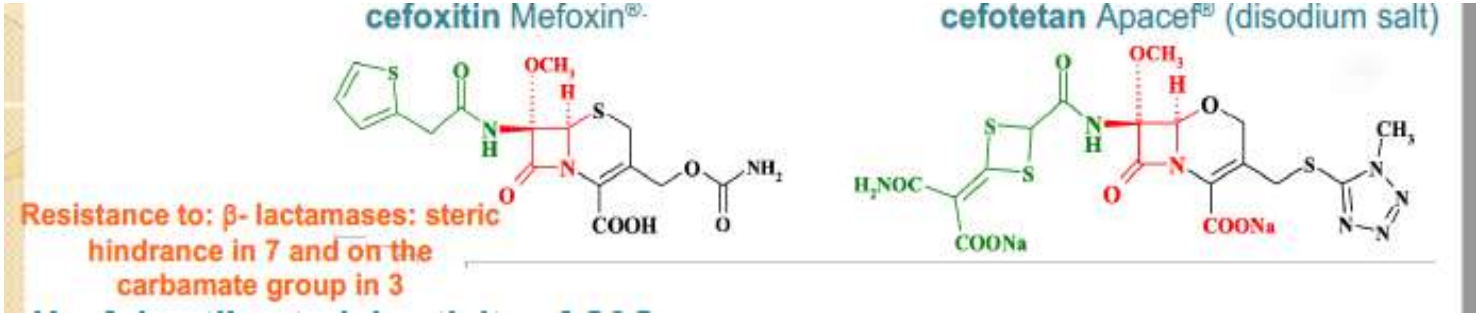


Alkoxyimine: stabilizes/b lactamases and shifts spectrum to Gram-

R = H cefuroxime (Parentral sod. Salt) Zinnat®

R= CH(CH3)-O-C(O)-CH3 cefuroxime-axetil (Oral)

✓ Cephamycines :



Useful antibacterial activity of C2G:

- Meti-S staphylococci, streptococci
- Enterobacteriaceae: E. coli, Proteus mirabilis
- H. influenzae: MS or S
- Cefoxitin: anaerobic Gram - bacteria and enterobacteriaceae

Therapeutic indications of C2G:

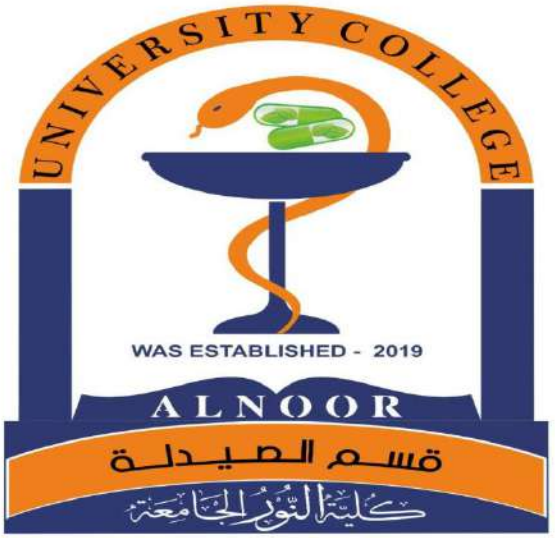
- Infections caused by germs resistant to C1G and ampicillin, in monotherapy or in combination (aminoglycosides)
- Antibiotic prophylaxis in surgery (CV and orthopedics)
- In IM-IV / 8h: dosage = 2 - 6 g / day
- In Oral route (cefuroxime-axetil): ENT and broncho-pulmonary infections
- dosage = 0.5 - 2 g / day

Organic Pharm. Chemistry III

Lecture 4

12th March 2023

10:30- 12:30



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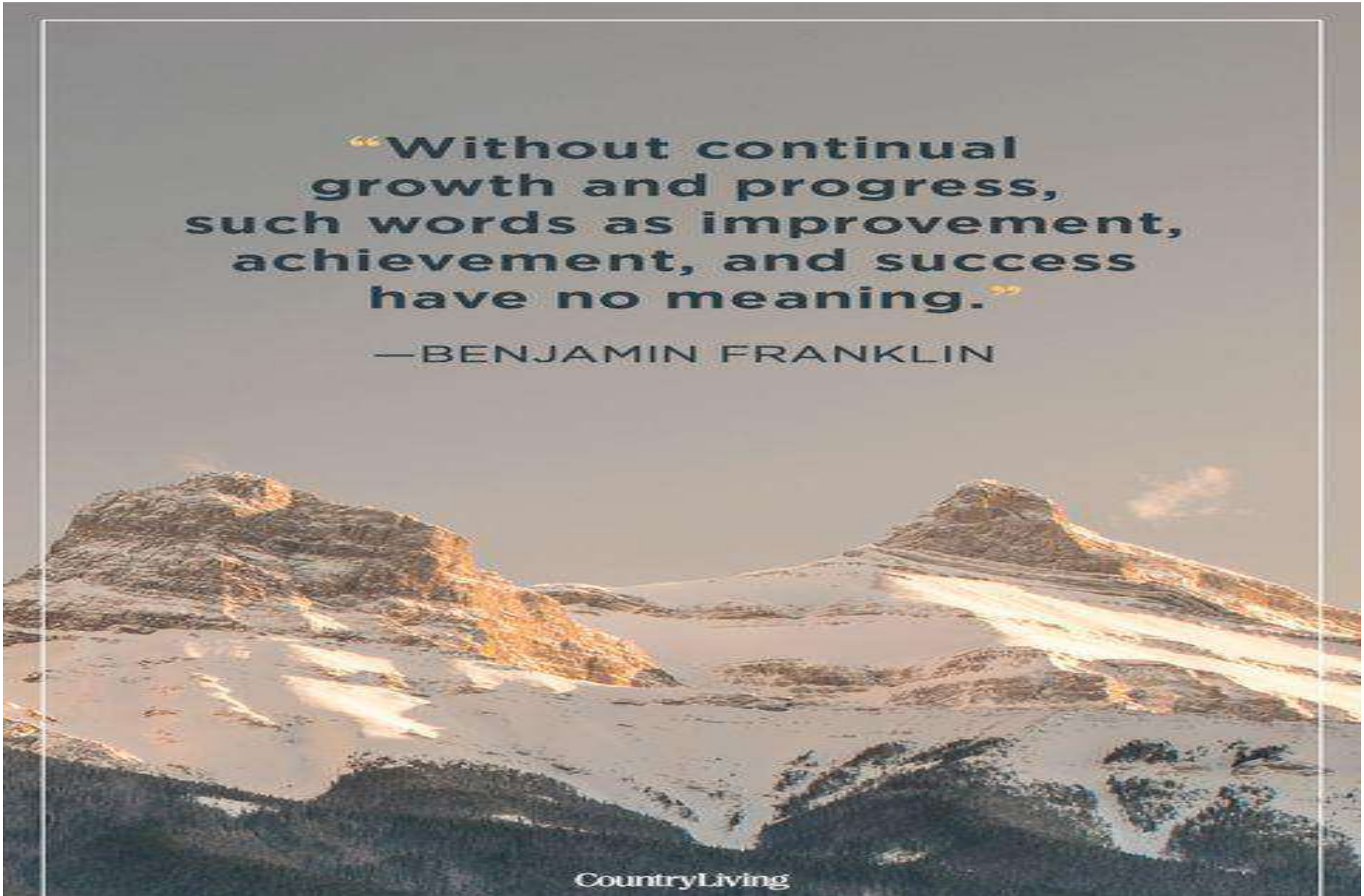
2nd Course/ 4th Class

2022/2023


"Without continual growth and progress, such words as improvement, achievement, and success have no meaning."
—BENJAMIN FRANKLIN
CountryLiving

Third generation cephalosporins

- Considerable increase in activity

•High stability to beta-lactamases: penicillinases, certain cephalosporinases (enterobacteria)

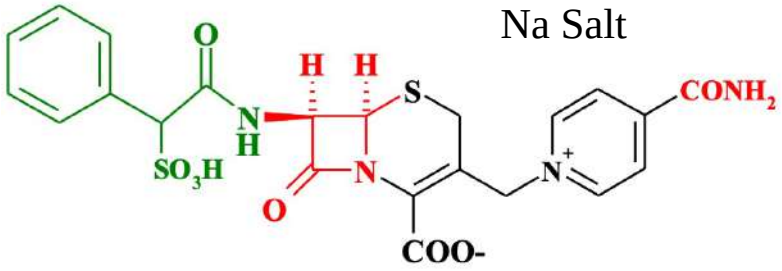
• Dissemination in sites inaccessible to C1G and C2G (meninges)

• Severe hospital infections

Third generation cephalosporins

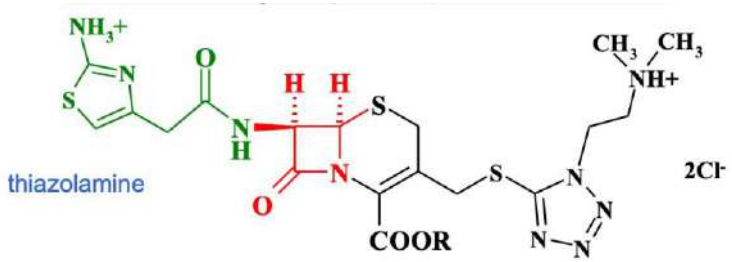
1. STRUCTURES

• Cephalosporin a-sulfonic: cefsulodine P



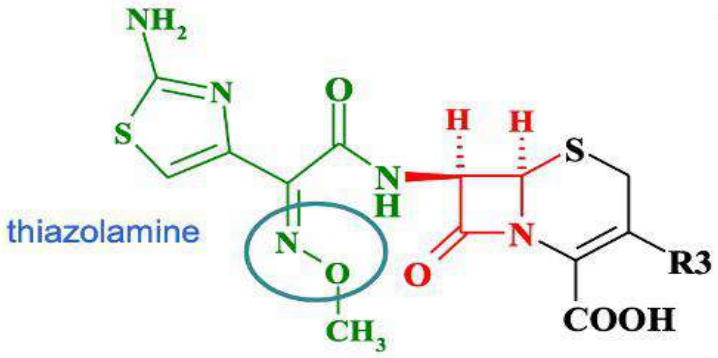
H-bond between NH at 7 and the sulfonyl residue: resistance to b lactamases

- Aminothiazolyl cephalosporin: cefotiam

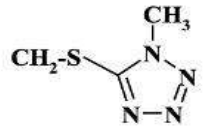
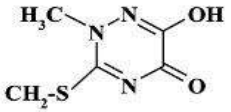


R = H cefotiam,
R = CH(CH$_3$)-O-CO-O-C$_6$H$_{11}$
cefotiam-hexétil
Taketiam® Texodil®

- Aminothiazolyl cephalosporins (oximes)



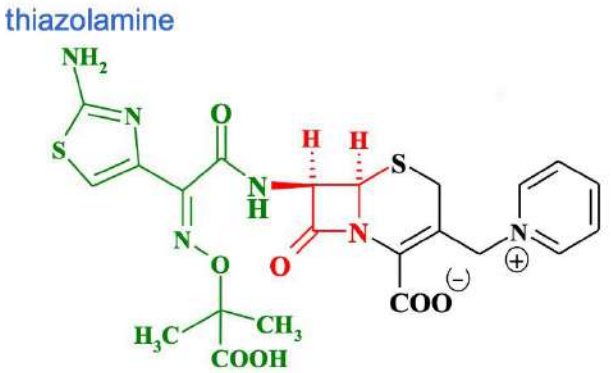
Cefpodoxime-proxetil
COO-CH(CH$_3$)-O-C(O)-O-CH(CH$_3$)$_2$

R3	DCI	ND
H	ceftizoxime	
CH$_2$-O-CH$_3$	cefpodoxime (proxetil)	Orélox®
CH$_2$-O-CO-CH$_3$	cefotaxime	Claforan®
	cefmemoxime	
	ceftriaxone	Rocéphine®

- Aminothiazolyl-cephalosporins (oximes) continued

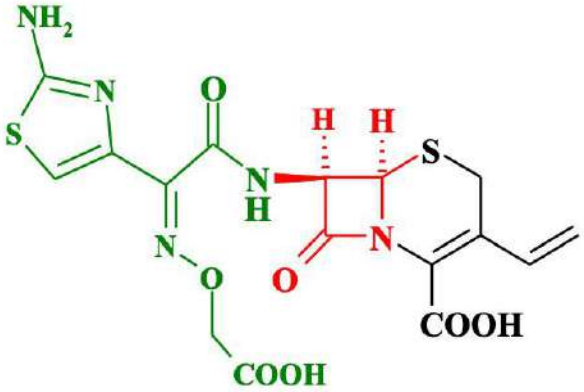
Ceftazidime Fortum® Fortumset® (pentahydrate)

Good activity on Pseudomonas aeruginosa



Cefixime Oroken® (trihydrate)

Vinyl: better stability in an acidic environment, administration by oral route.



.2. Useful antibacterial activity of C3Gs:

• Gram + cocci: staphylococci, penis-S pneumococci

• Gram-cocci : N. gonorrhoeae, meningitidis

• Gram-bacilli: enterobacteria (E. coli, Proteus, Shigella, Salmonella), H. influenzae, P. aeruginosa

• Spirochetes: Borrelia (Lyme disease).

Pharmacokinetic data of C3G:

• Good diffusion (CSF, bile) • t1 / 2 very variable

Therapeutic indications of C3G:

• Severe, localized or generalized infections resistant to other blactams (in particular hospital infections) in monotherapy or in combination

• Antibiotic prophylaxis in surgery

• In IM-IV / 8 - 12 h: dosage = 2 - 6 g / day for oral forms (cefixime and cefpodoxime-proxetil):

OTHER 3rd GENERATION CEPHALOSPORINS

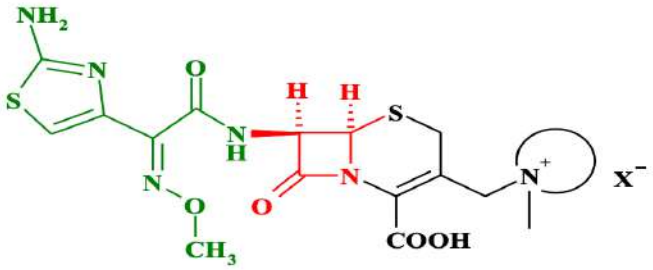
✓ Better stability vis beta-lactamases,

✓ Increased affinity for PLPs,

✓ Good activity on P. aeruginosa,

✓ Better activity on gram-cocci (staphylococci and streptococci).

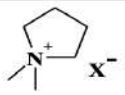
1. STRUCTURES



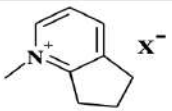
.2. **Useful antibacterial activity:**

• Cocci at Gram + and at Gram –

• Gram bacilli -: enterobacteria



Cefepime (2 HCl)
Axepim®



Cefpirome (sulfate)
Cefrom®

INDICATIONS: Treatment of severe infections, localized or generalized, with bacteria resistant to other b-lactams (in particular hospital infections) as mono therapy or in combination.

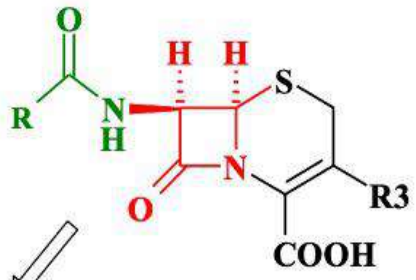
Dosage: 2 - 4 g / d

SIDE EFFECTS

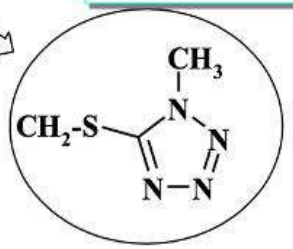
They are rather well tolerated.

Local intolerance reactions:
IM: pain,
IV: phlebitis.

Allergic manifestations: fever, vasculitis, urticaria, angioedema, anaphylactic shock,



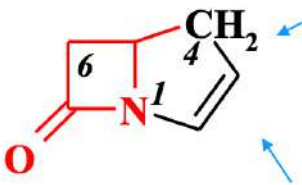
- bleeding, haemorrhages
- anti-abuse effect



Gastrointestinal (diarrhea) and hepatic disorders.

HEMISYNTHETIC BETA-LACTAMES OF CARBAPENEMS

STRUCTURE-ORIGIN

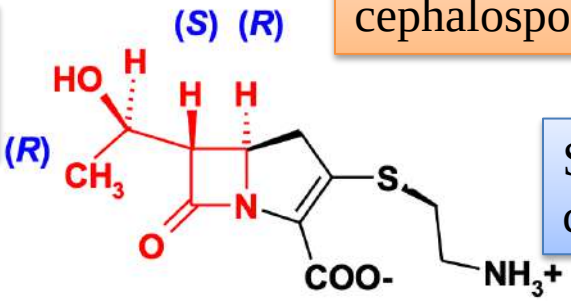


Replacement of S by CH2

Double bond in position equivalent to that of cephalosporins

Isolation of thienamycin: *Streptomyces cattleya*

Reverse configuration in 6 compared to P and C



Side chain: cysteamine

INTERESTS:

✓ Strong activity on Gram - bacteria;

✓ Bactericidal effect (binding to PLP2; *cysteamine*),

✓ Resistance to b -lactamases in Gram germs -.

But chemical instability due to the cysteamine residue, hence IMIPENEM

Protection of the amine in the form of amidine:

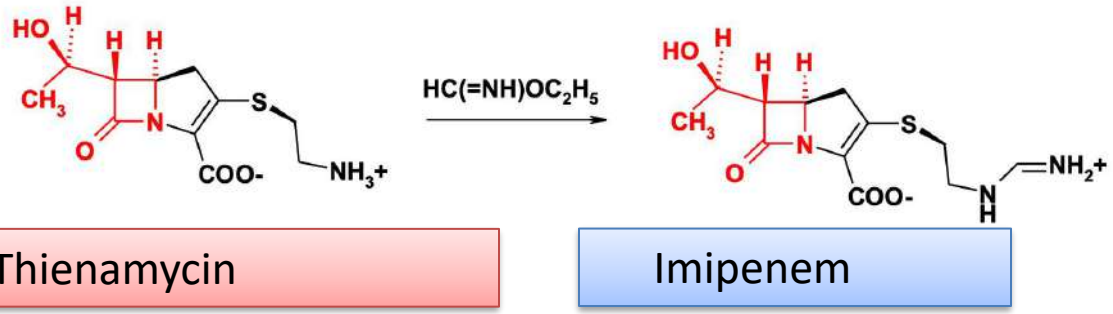
✓ ***Stability***,

✓ ***Better bacteriological activity.***

ACCESS ROAD

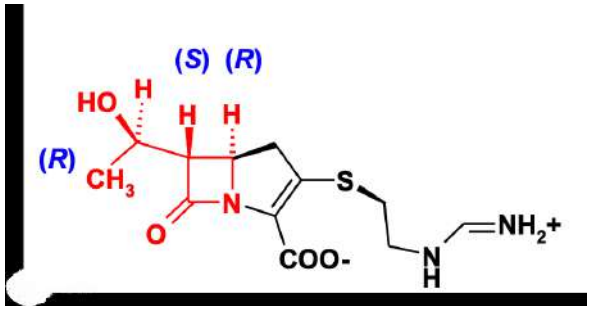
✓ Fermentation, **but total synthesis is preferred,**

✓ **Passage amine** → ***amidine*:**



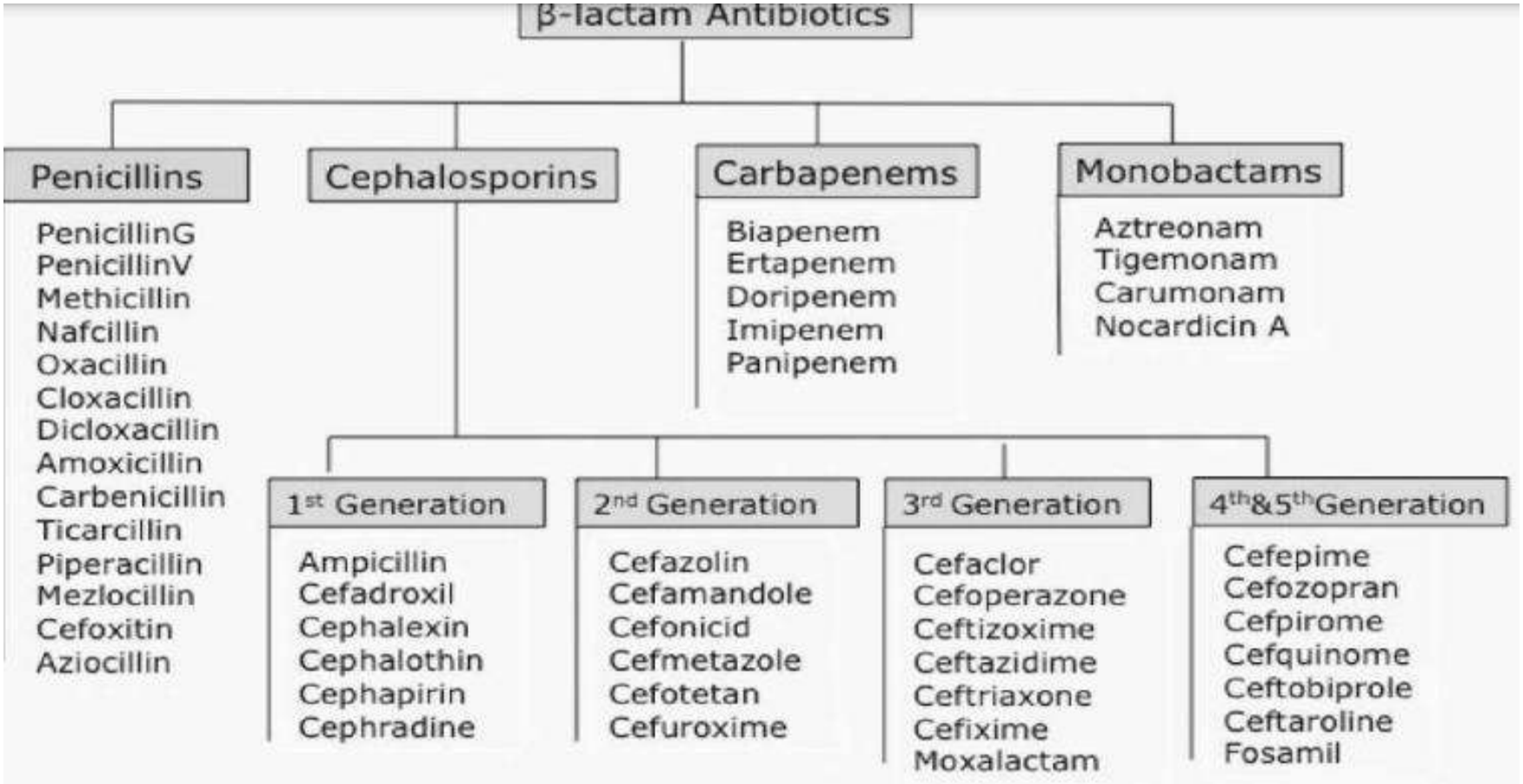
Thienamycin

Imipenem



IMIPENEM

BIOLOGICAL ACTIVITY
Good penetration through the porins of the outer membrane of Gram -
germs,
✓ Excellent stability vis-à-vis b-lactamases,
✓ High bactericidal activity
✓ Marked post-antibiotic effect (growth latency after elimination of the antibiotic),


β-lactam Antibiotics
Penicillins
PenicillinG
PenicillinV
Methicillin
Nafcillin
Oxacillin
Cloxacillin
Dicloxacillin
Amoxicillin
Carbenicillin
Ticarcillin
Piperacillin
Mezlocillin
Cefoxitin
Aziocillin
Cephalosporins
1st Generation
Ampicillin
Cefadroxil
Cephalexin
Cephalothin
Cephapirin
Cephradine
2nd Generation
Cefazolin
Cefamandole
Cefonicid
Cefmetazole
Cefotetan
Cefuroxime
3rd Generation
Cefaclor
Cefoperazone
Ceftizoxime
Ceftazidime
Ceftriaxone
Cefixime
Moxalactam
4th&5th Generation
Cefepime
Cefozopran
Cefpirome
Cefquinome
Ceftobiprole
Ceftaroline
Fosamil
Carbapenems
Biapenem
Ertapenem
Doripenem
Imipenem
Panipenem
Monobactams
Aztreonam
Tigemonam
Carumonam
Nocardicin A

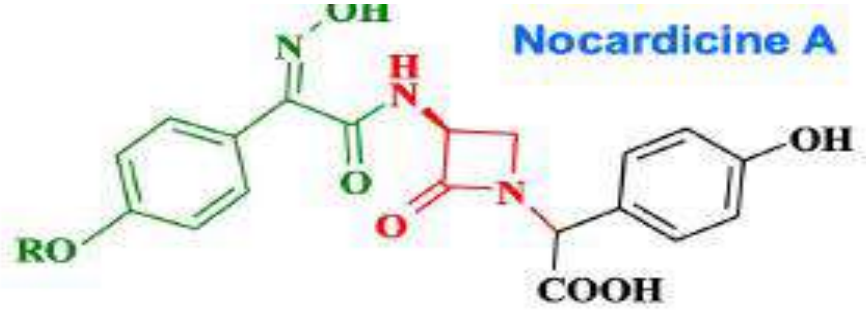
HEMISYNTHESIS BETA-LACTAMES Of MONOBACTAMES

MONOBACTAMES

I. ORIGIN

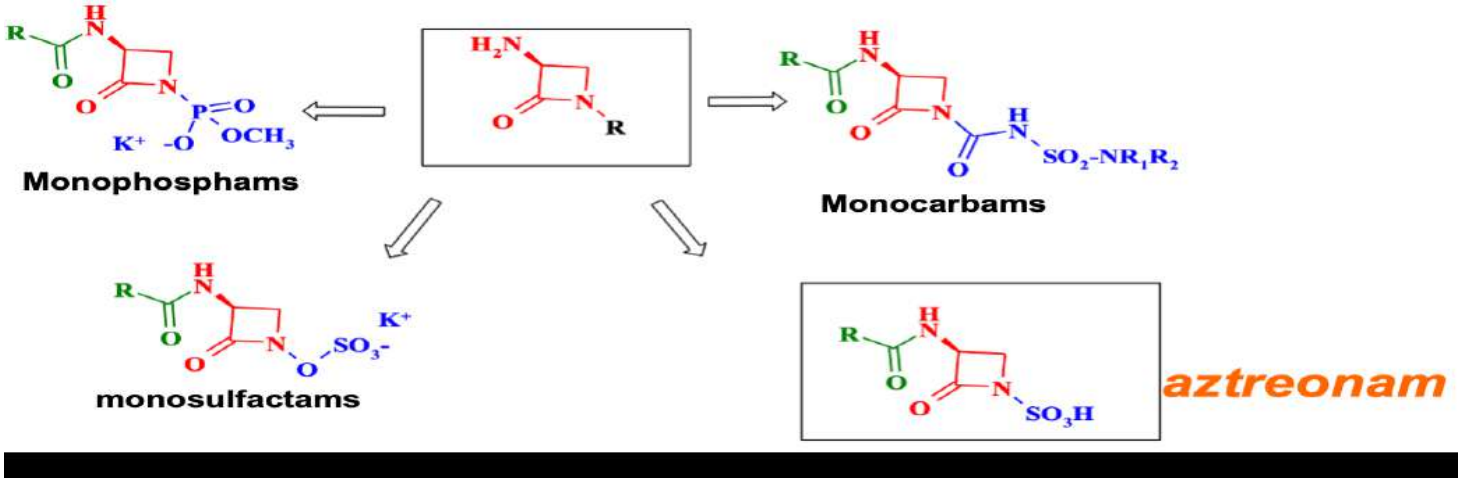
nocardicins (1976);

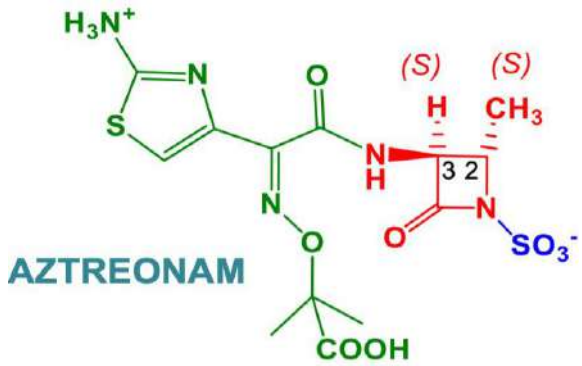
sulfazecin and SQ 26180 (1981).



(2-Oxo-3-aminoazetidin-1-yl) -2-phenylacetic acid

2. CLASSIFICATION - STRUCTURES



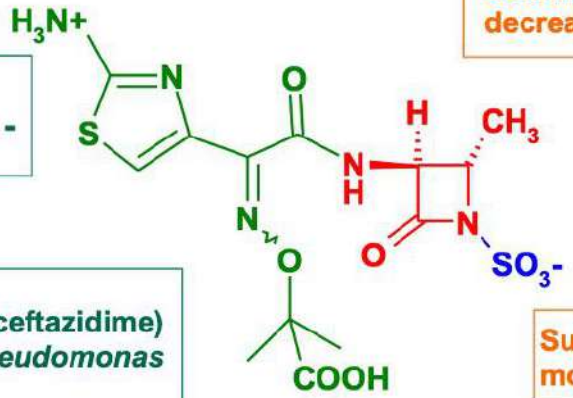


- Internal salts: zwitterion

3-Amino-2-oxo azetidine-1-sulfonic acid

Stability /b-lactamases, particularly against Gram-, is obtained by substituting the 2-position.

SAR



2-Aminothiazole spectrum → Gram -

Stability against beta-lactamases, but decreased Gram + activity.

Isobutyric chain (ceftazidime) binding on PLP 3 *Pseudomonas* activity

Sulfonic acid "Activation of the molecule"

Lack of inducing power of β- -lactamases

Selective spectrum exclusively on gram bacteria -:

✓ Gram bacilli - : enterobacteria Proteus, Salmonella, Shigella, E.coli, Pseudomonas, Haemophilus;

✓ Gram-cocci: N. gonorrhoeae and meningitidis.

INDICATIONS of AZTREEONAM

✓ Severe gram-negative infections (nosocomial infections),

✓ As monotherapy based on the antibiogram data,

✓ **First-line** and in combination (aminoglycoside) in very severe conditions before the antibiogram.

✓ dosage: 3-8 g / d IM or IV (/ 6-8 h).

REACTIONS OF AZTREONAM

= **well tolerated molecules 7%** of prescriptions (in 2% of cases, discontinuation of treatment)

✓ Gastrointestinal effects: diarrhea, nausea and / or vomiting;

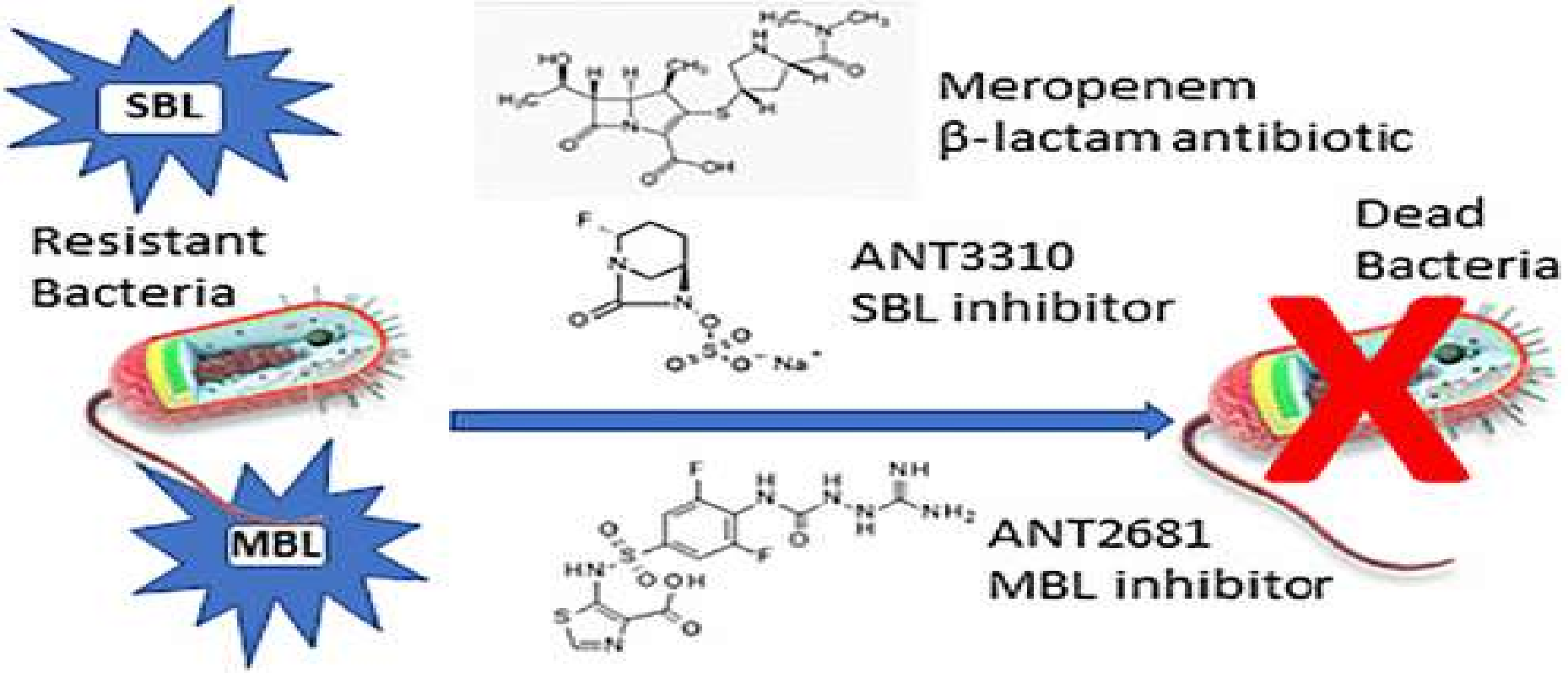
✓ Superinfection by gram + germs;

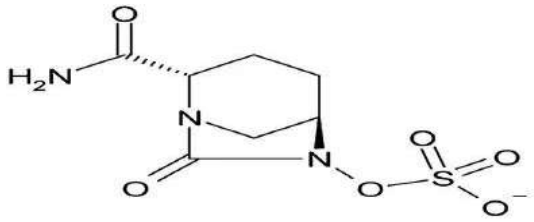
✓ Immuno-allergic reactions (2%)

- low immunogenicity,

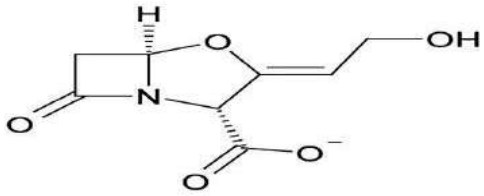
- virtual absence of cross reactions with other beta-lactams

HEMISYNTHETIC BETALACTAMES INHIBITORS OF BETALACTAMASES

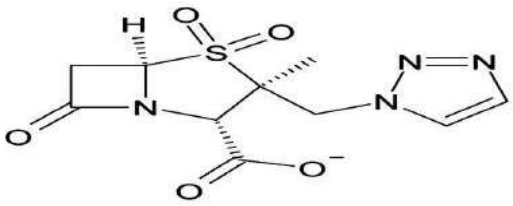




Avibactam



Clavulanic acid



Tazobactam

BETA-LACTAMASES INHIBITORS

INTRODUCTION

1.1. Inactivation of beta-lactams

(penicillins, cephalosporins, monobactams)

1.2. Structure of beta-lactamases

✓ **Serine enzymes** exhibiting a great structural analogy with the PLPs.

✓ **Classified according to**:

- Isoelectric pH,
- Enzymatic constants,
- The amino acid sequence,
- Sensitivity or insensitivity to different inhibitors.

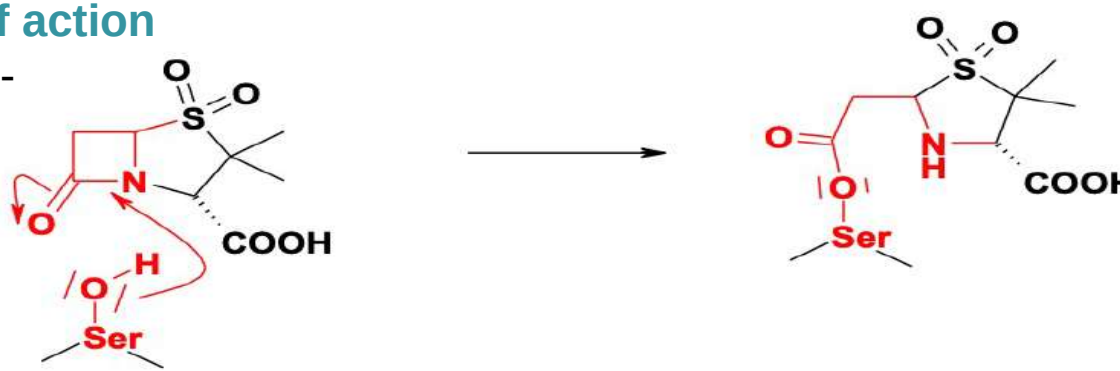
1.3. Beta-lactamase biosynthesis

✓ ***Constitutive*** (case of Gram - bacteria),

✓ ***Inducible*** (induction by methicillin S. aureus betalactamase

1.4. Biomolecular mechanism of action

Attach Nu of CO of lactame of OH-
Of serine



BETA-LACTAMASES INHIBITORS

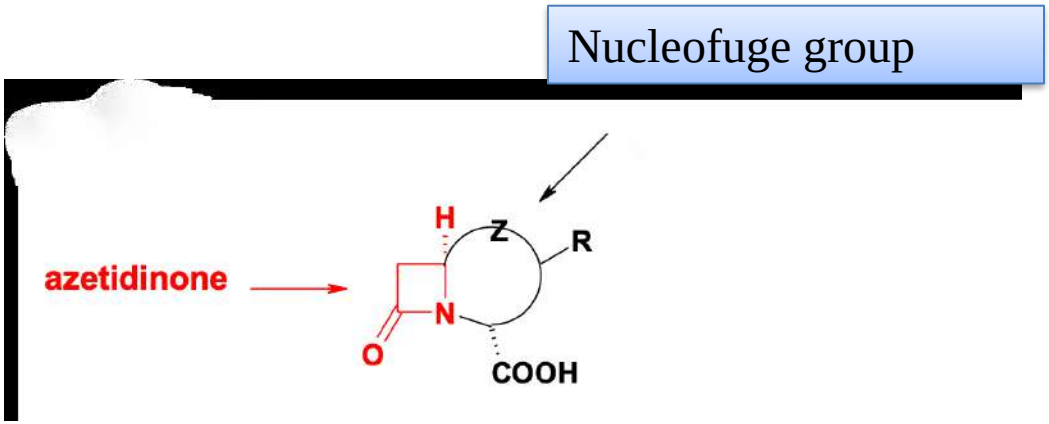
- STRATEGIES TO LIMIT THE ACTION OF BETA-LACTAMASES

Design ABTs capable of resisting their action: - penicillins - 2nd and 3rd G cephalosporins.

Decrease their biosynthesis: beta-lactam + chloramphenicol combination

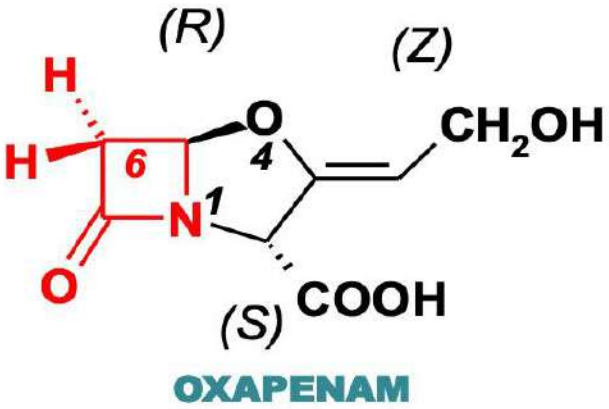
Inhibit them by:
- competitive inhibitors (reversible)
- non-competitive inhibitors = suicide substrates (irreversible)

SAR of **b-lactamase inhibitors**



In chemistry, a nucleofuge (from nucleo- 'atomic nucleus', and fuge 'to run away/escape') is a leaving group which retains the lone pair of electrons from its previous bond with another species.

BETA-LACTAMASES INHIBITORS



3.1. OXAPENAME: clavulanic acid

Obtained by fermentation of Streptomyces clavuligerus **Augmentin ® , Claventin ®**

3.1.1. PHYSICO-CHEMICAL CHARACTERS

✓ **Potassium clavulanate**, very soluble in water,

✓ **Identification**: IR and NMR

✓ **Dosage**: HPLC

3.1.2. BIOLOGICAL ACTIVITY

✓ Irreversible inhibitor **(= suicide substrate**) of beta-lactamases,

✓ Active at low concentration especially on penicillinases,

✓ Weak antibiotic effect by itself.

THERAPEUTIC INTEREST

In combination with penicillin

Amoxicillin - clavulanic acid
Augmentin®

✓Oral absorption of the 2 active ingredient and **similar bioavailability**.

✓**Improvement in the performance** of amoxicillin against beta-lactamase-producing strains.

✓ **Oral** route (1.5 - 2 g) and **IV** (1 - 12 g).

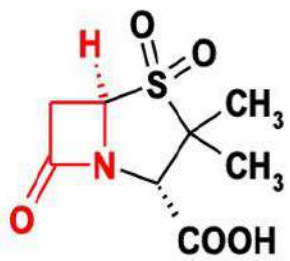
Ticarcillin - clavulanic acid
Claventin®

✓**Improved activity** of carboxypenicillin.

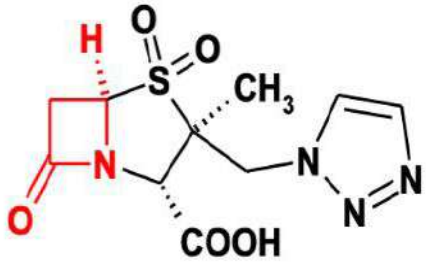
✓ Lack of selection in the hospital environment of bacteria resistant to beta-lactamases.

✓ **Slow IV**; dosage: 9 - 18 g.

SULFONYLPENICILLINIC ACIDS



SULBACTAM and TAZOBACTAM

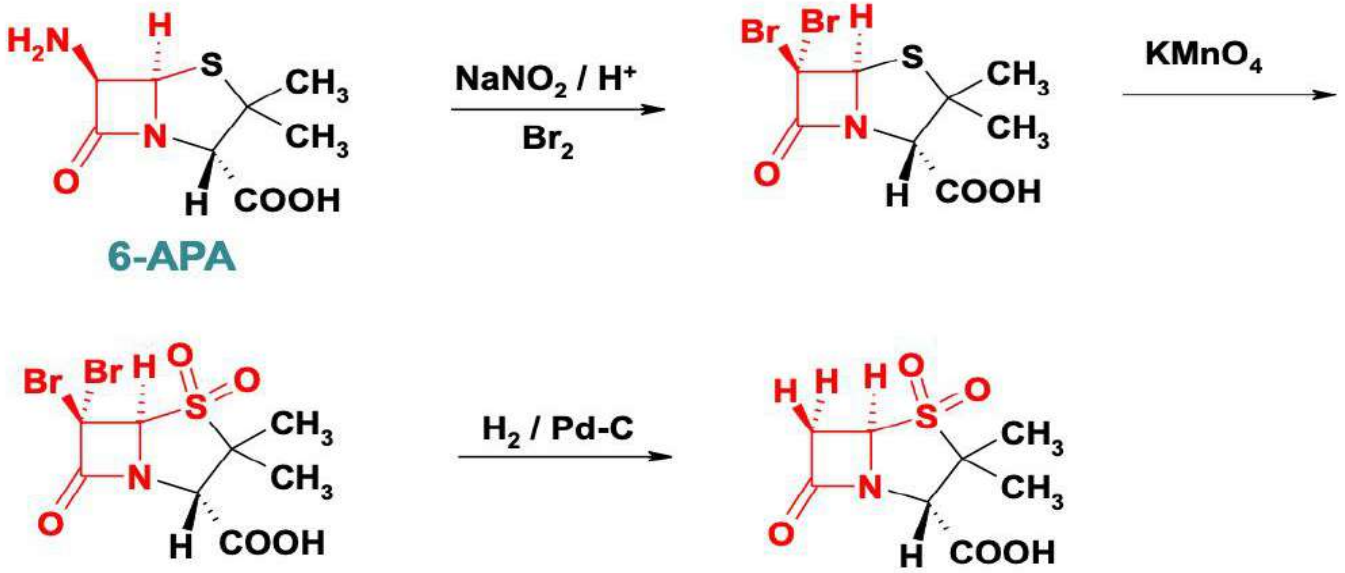


✓ Oxidation derivatives of pename (= sulfone),

✓ Used in the form of sodium salts in combination.

. Route of access to sulbactam

Halogenating diazotization - oxidation - hydrogenolysis



Biological activity of sulfonylpenicillins

✓ Irreversible inhibitor of most of the penicillinases produced by Gram + and - bacteria,

✓ Intrinsic antibiotic effect (sulbactam).

. Therapeutic benefit of the combination

In association : ampicillin - sulbactam
Unacim® (1 g) (0.5 g)

✓Poor digestive absorption of sulbactam,

✓ **Improvement of the performance** of ampicillin, especially against *S. aureus*,

✓ Oral and slow IV; dosage: 0.75 - 1.5 g / day.

Piperacillin - tazobactam Tazocilline®

✓ Slow IV; dosage = **12 g**

USEFUL ANTIBACTERIAL ACTIVITY OF THE ASSOCIATION

✓ Spectrum of associated beta-lactam + irreversible inhibition of betalactamases, produced by germs responsible for severe hospital infections: *Staphylococcus* meti-S, Enterobacteriaceae, anaerobic bacteria including *B. fragilis*

PRECAUTIONS FOR USE

✓ Dividing the doses to avoid high doses of clavulanic acid responsible for diarrhea.

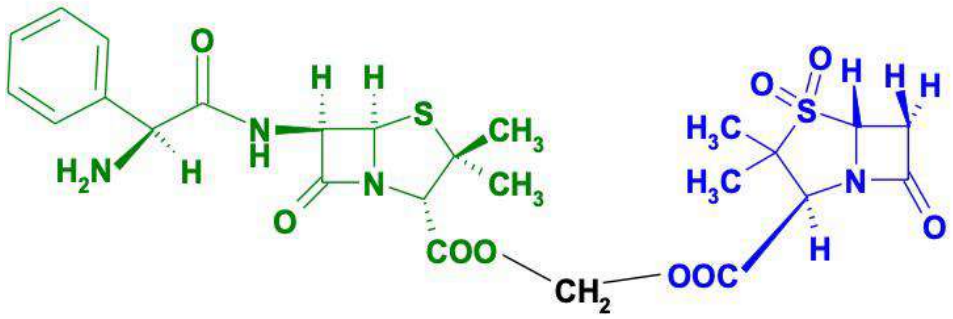
✓ Separate administration of aminosides.

✓ Presence of aspartame (excipient in powders):

✓ **Contraindication**: subjects with phenylketonuria.

CONCLUSIONS

Example of a prodrug: ampicillin + sulbactam (sultamicillin) linked through a methylene bridge.



Organic Pharm. Chemistry III

Lecture 6

26th March 2023

10:30- 12:30

AlNoor University College

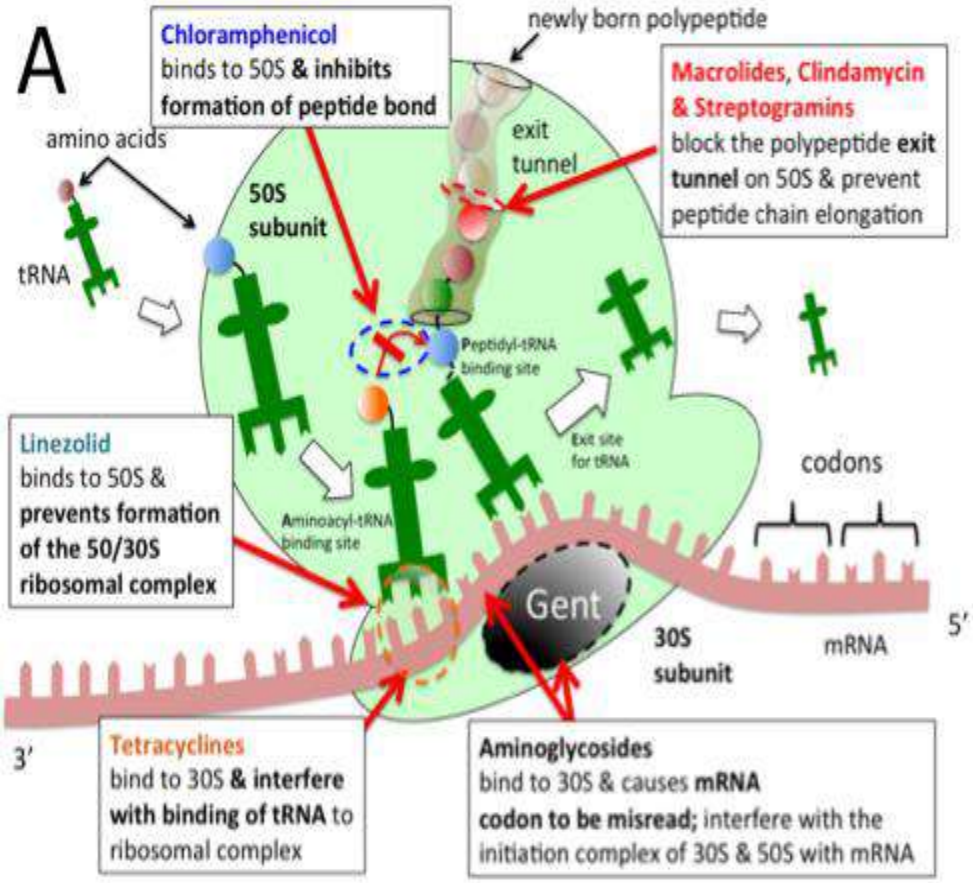
Pharmacy Department

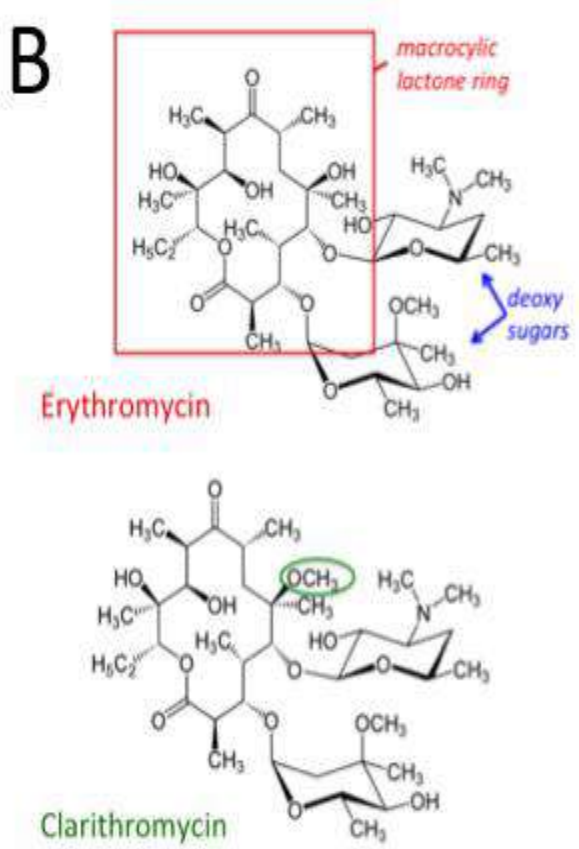
2nd Course/ 4th Class

2022/2023

أسأل الله لكم في شهر رمضان.. حسنات تتكاثر وذنوب تتناثر وهموم تتطاير وأن يجعل بسمتكم سعادة وصمتكم عبادة وخاتمتكم شهادة ورزقكم في زيادة وبكل زخة مطر وبعدد من حج واعتمر.. أدعو الله أن يتقبل صالح العمل

Macrolides


A
Chloramphenicol
binds to 50S & inhibits formation of peptide bond
newly born polypeptide
Macrolides, Clindamycin & Streptogramins
block the polypeptide exit tunnel on 50S & prevent peptide chain elongation
amino acids
exit tunnel
50S subunit
tRNA
Peptidyl-tRNA binding site
Exit site for tRNA
Aminoacyl-tRNA binding site
Linezolid
binds to 50S & prevents formation of the 50/30S ribosomal complex
codons
Gent
30S subunit
mRNA
5′
3′
Tetracyclines
bind to 30S & interfere with binding of tRNA to ribosomal complex
Aminoglycosides
bind to 30S & causes mRNA codon to be misread; interfere with the initiation complex of 30S & 50S with mRNA


B
macrocylic lactone ring
deoxy sugars
Erythromycin
Clarithromycin

Introduction

 The macrolides are a class of natural products that consist of a large macrocyclic lactone ring to which one or more deoxy sugars, usually **cladinose and desosamine, may be attached.**

 The lactone rings are usually 14-, 15-, or 16-membered.

 Macrolides belong to the polyketide class of natural **products(Polyketides, a diverse group of heteropolymers with antibiotic and antitumor properties,)**

History

 The first macrolide discovered was erythromycin, which was first used in 1952.

 Erythromycin was widely used as a substitute to penicillin in cases where patients were allergic to penicillin or had penicillin-resistant illnesses.

 Later macrolides developed, including **azithromycin and clarithromycin, stemmed from chemically modifying erythromycin; these compounds were designed to be more easily absorbed and have fewer side-effects (erythromycin caused gastrointestinal side-effects in a significant proportion of users).**

Mechanism of action

 Macrolides are protein synthesis inhibitors.

 The mechanism of action of macrolides is inhibition of bacterial protein biosynthesis, and they are thought to do this by preventing peptidyltransferase from adding the growing peptide attached to tRNA to the next amino acid (similarly to chloramphenicol as well as inhibiting bacterial ribosomal translation. Another potential mechanism is premature dissociation of the peptidyl-tRNA from the ribosome.

 Macrolide antibiotics do so by binding reversibly to the P site on the 50S subunit of the bacterial ribosome.

This action is considered to be bacteriostatic.

 Macrolides are actively concentrated within leukocytes, and thus are transported into the site of infection

- The macrolides are a group of antibiotics produced by various strains of Streptomyces and having a macrolide ring structure linked to one or more sugars . They act by inhibiting protein synthesis, specifically by blocking the 50S ribosomal subunit. They are broad spectrum antibiotics.

- Azithromycin,
- Clarithromycin,
- Examples:Erythromycin, Roxithromycin etc.....

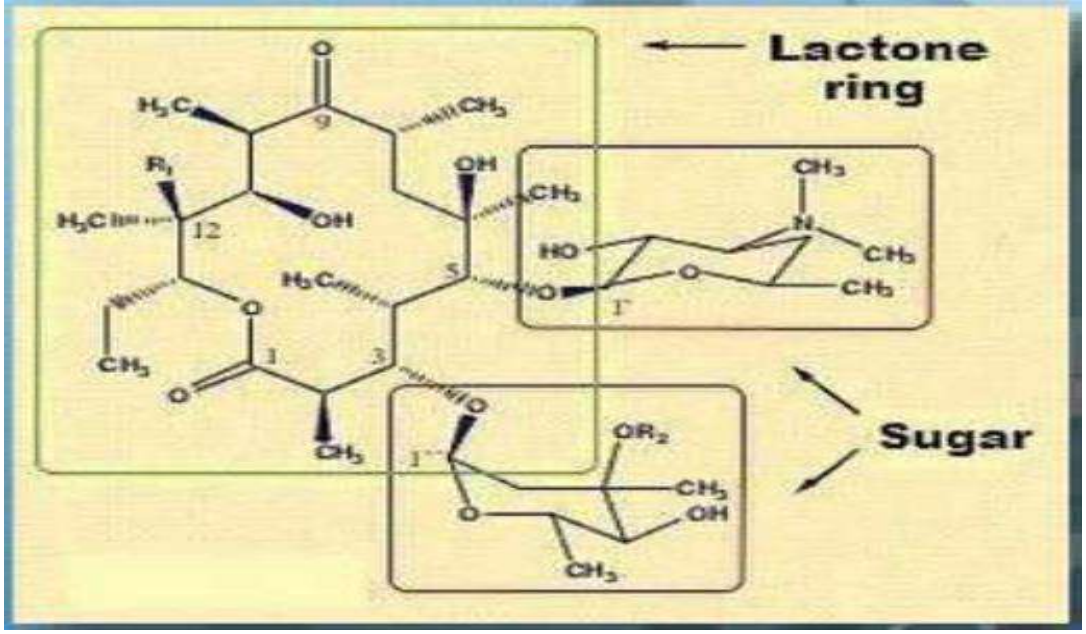
Objectives:

1. Understand the chemistry of drug with respect to their biological activity.
2. Know the Importance of SAR of Drugs
3. Know the metabolism, Adverse Effect and therapeutic value of drugs.
4. Understand the importance of Drug design.

Learning Outcomes:

1. Students will Learn about the structures and Medicinal uses of Drugs.
2. Students will learn about relation of structure with its activity.
3. Students will Learn about the designing of drugs.

SOURCE:



These Are Produced By Streptomyces species.

CHEMISTRY:

- A macrocyclic lactone ,usually having 12 to17atoms.
- A ketone group.
- One or two amino-sugars linked to the nucleus.
- A neutral sugar linked either to amino sugar or to lactone ring.
- The presence of the dimethylamino moiety on the sugar residue, which explains the basicity of these compounds and consequently formation salts.

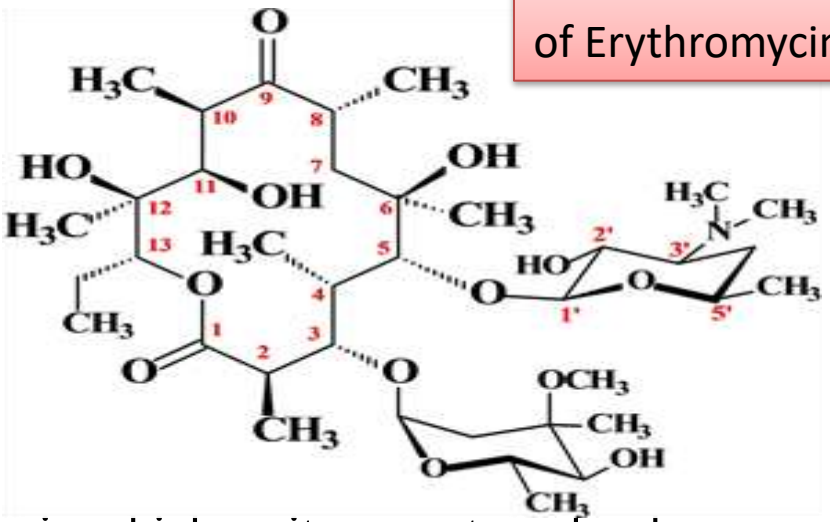
SAR

- As macrolide are unstable in acidic pH, a no. of strategies have been utilized to improve the acidic stability of erythromycin.
- The addition of hydroxylamine

to the ketone to form oxime

e.g. roxithromycin

- Alteration of c-6hydroxyl group: nucleophilic functionality which initiates erythromycin degradation.
- The azalides (azithromycin)are

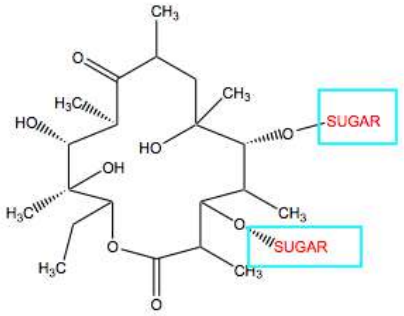


Chemical structure of Erythromycin A.

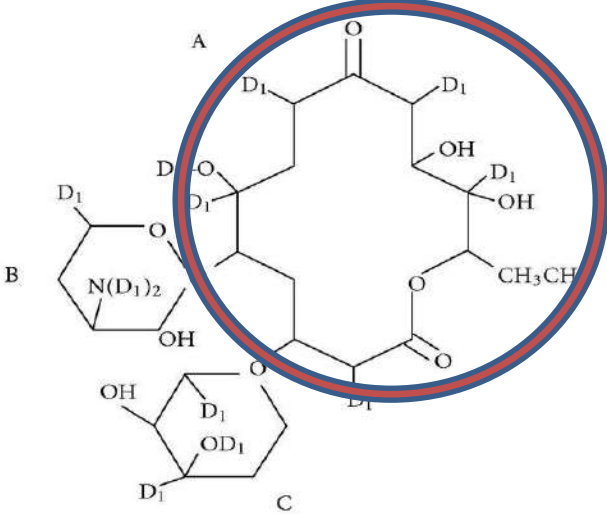
Semi-synthetic 15 -membered congeners in which a nitrogen atom has been introduced to expand a 14-membered precursor-leads to an extended spectrum of action.

Physical & Chemical properties

- Water insoluble molecules.
- Occurs as crystalline powders.
- Stable in aqueous solutions at or bellow room temperature.
- Unstable in acidic conditions and forms internal cyclic ketal



Macrolide Backbone



A: Macrocyclic lactone ring
B: Desosamine
C: Cladinose
D_1: CH_3
D_2: H

Macrolide

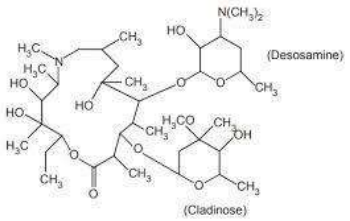
Is having 3 parts

Lactone ring

It is a large non-planar strainless ring with 12-16 atoms

Aglycone part

Ketone group



Glycosidically linked amino sugars

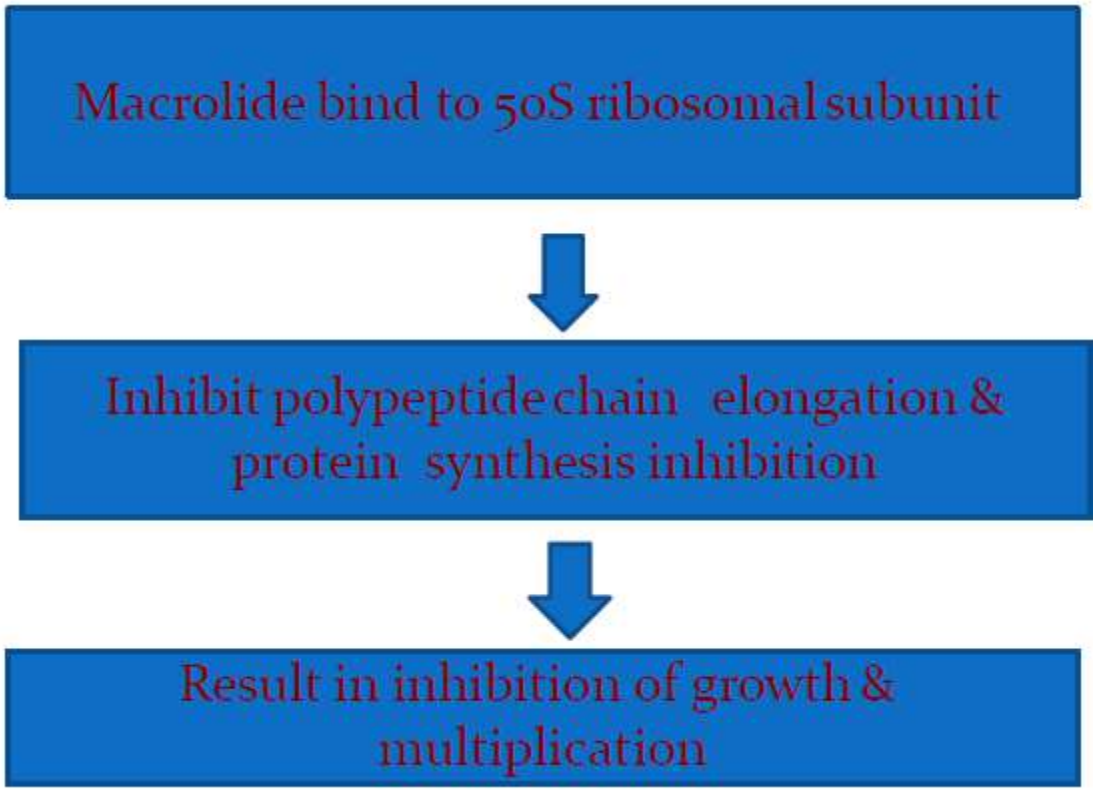
It is having 2 deoxy sugars
1) L-Cladinose (neutral sugar)
2) d- Desosamine(amino sugar)

Glycone part

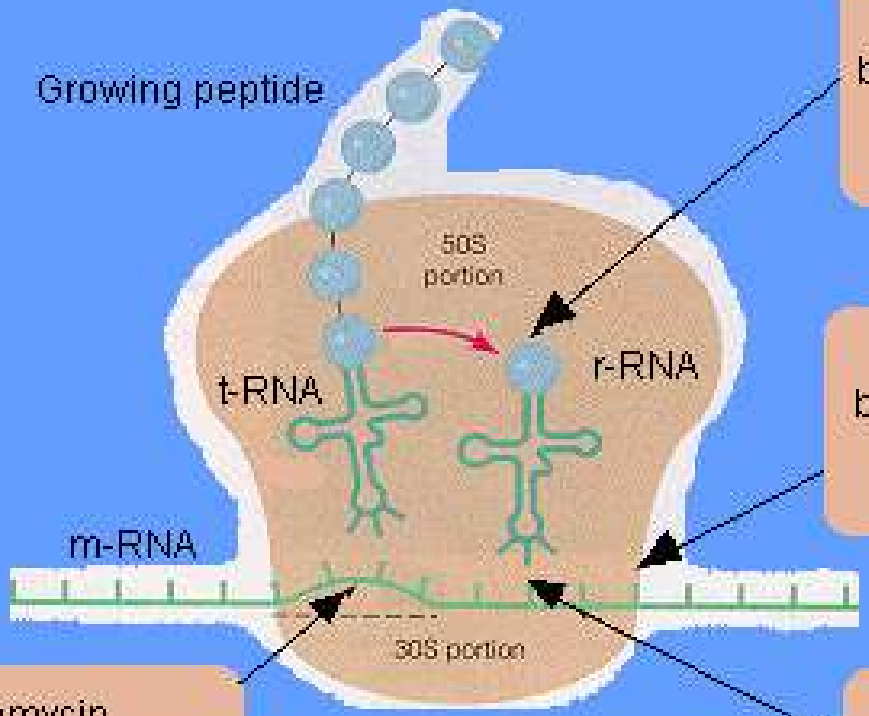
The amino group present is dimethyl amino group .
It forms salts with acids.
Eg: 1) Glucoheptanoic acid
2) Lactobionic acid

Mechanism of action

- Macrolide is protein biosynthesis inhibitors



Inhibition of Protein Synthesis by Antibiotics



Chloramphenicol binds to 50S r-RNA and inhibits formation of peptide bond

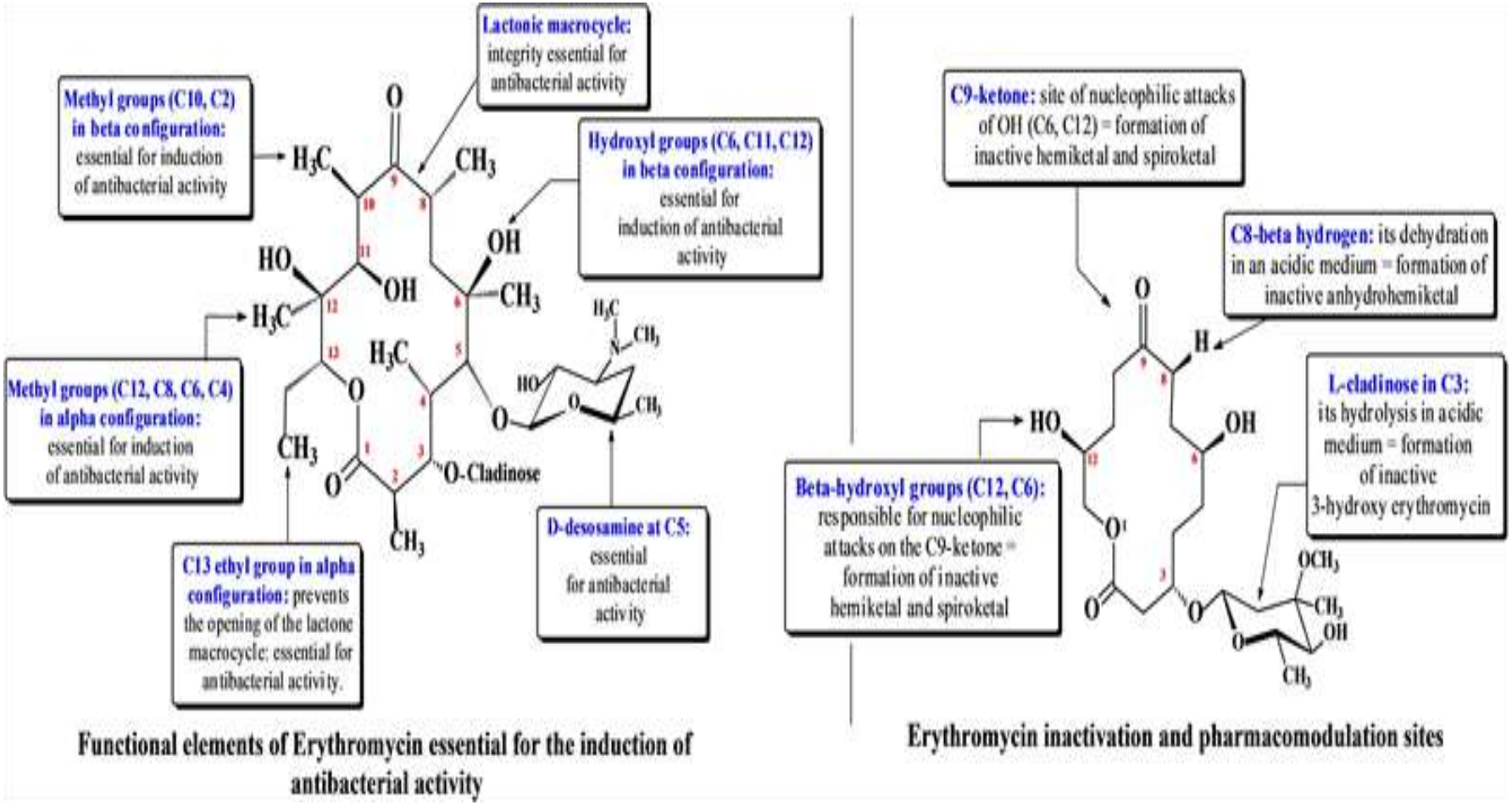
Erythromycin binds to 50S r-RNA and prevents movement along m-RNA

Streptomycin changes shape of 30S r-RNA and causes m-RNA to be read incorrectly

Tetracycline interfers with the t-RNA anticodon reading of m-RNA codon

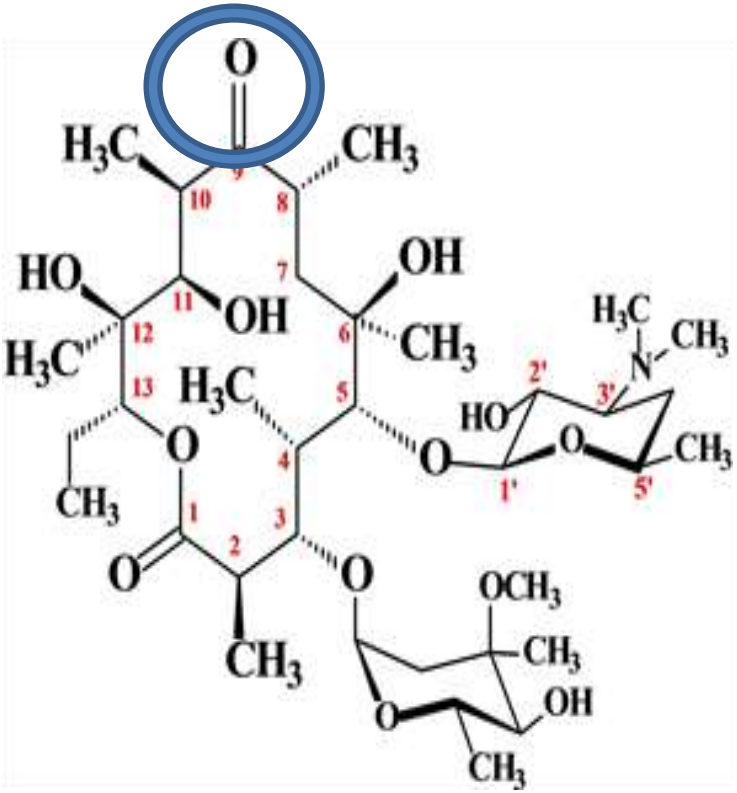
C. Ophardt, c. 2003

Study of Structure-Activity Relationships



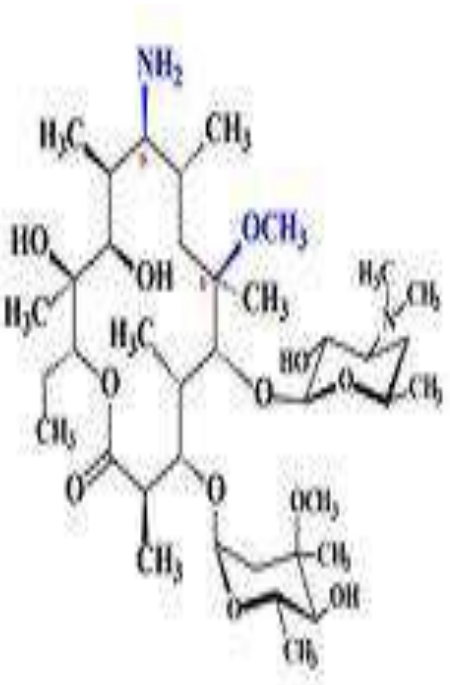
Weak point of Erythromycin A

- It is the ketone function in position C9, hydroxyl groups in C6 and C12 and hydrogen in position C8 in its beta conformation. These entities are responsible **for the acetalization reaction which leads to the hemiketal and spiroketal derivatives responsible for the digestive intolerance of Erythromycin A .**
- Another weak point of Erythromycin A is the presence of L-cladinose in position 3. Indeed, in gastric acid medium, the hydrolysis of the latter occurs to form an inactive 3-hydroxy erythromycin A derivative; which would inevitably favour the appearance of drug-resistant germs



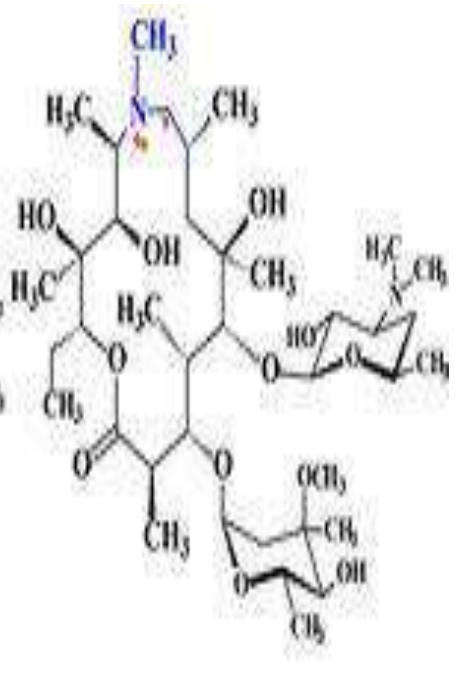
Pharmacochemical Evolution of Erythromycin A to Neomacrolides

Roxithromycin



Erythromycylamine

Dirithromycin



Azithromycin

Pharmacochemical Evolution of Erythromycin A to Neomacrolides

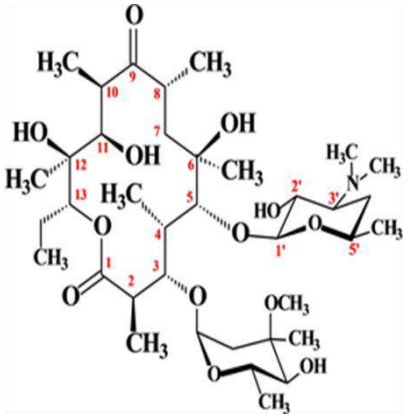
In order to obtain more effective hemisynthesis derivatives or neomacrolides of Erythromycin A, various chemical variations were made at its pharmacomodulable sites. **These include chemical variations at the sites carried by carbons C3, C6, C8, C9, C11 and C12.**

These different pharmacomodulations have led to neomacrolides **having higher pharmacokinetic performance, better digestive tolerance and/or a broader antibacterial spectrum compared to Erythromycin A**.

The neomacrolides are classified according to **the size of the 14- or 15-membered rings macrocycle**. Thus the 14-membered ring derivatives are composed of **Clari**thromycin**,** **Rox**ithromycin, Erythromy**cylamine**, **Dir**ithromycin and **Flur**ithromycin.

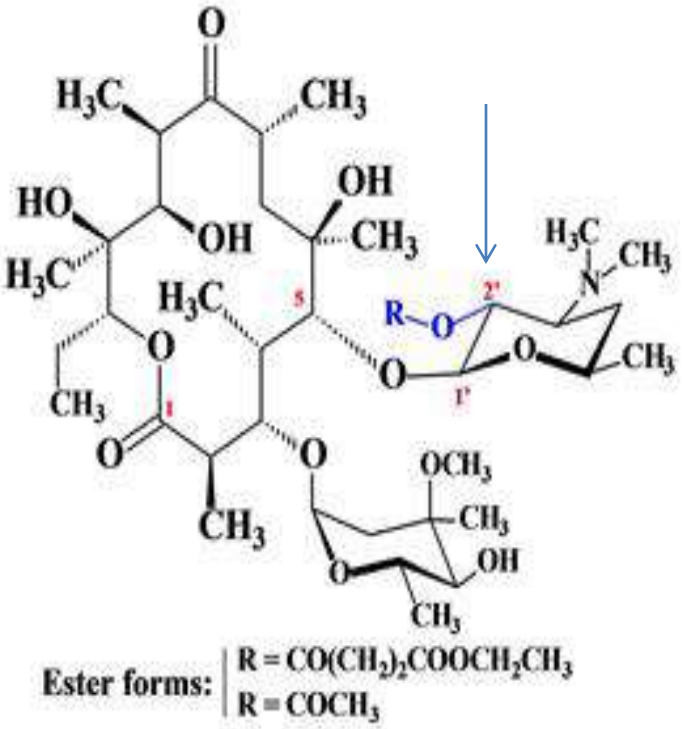
Only one 15-membered neomacrolide is currently used in therapy, it is **Azithromycin**. The different pharmacomodulations undertaken around the chemical structure of Erythromycin A can be summarized in

- **one esterification and salification,**
- **chemical modulation of the C6 hydroxyl function,**
- **halogenation of C8 and**
- **chemical modulation of the ketone function in C9.**



I.Esterification and Salification of Erythromycin A

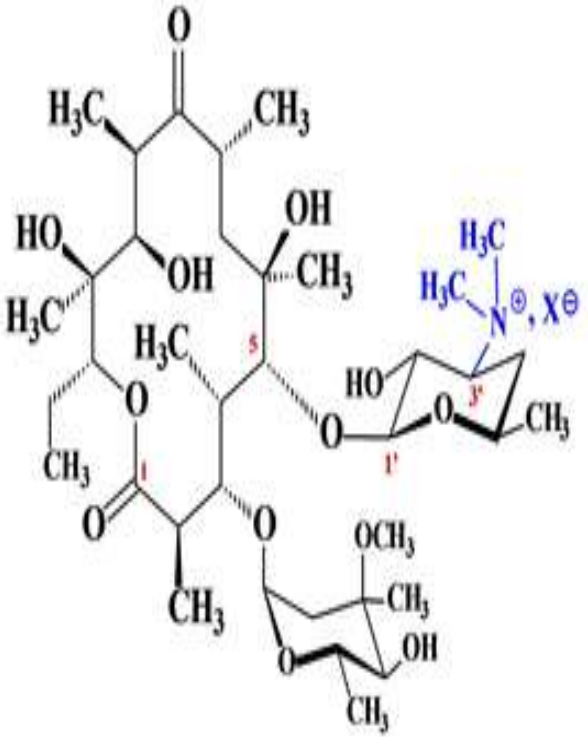
In order to **mask the bitter taste** of Erythromycin A, the hydroxyl in position C2'of D-desosamine (Figure) has been **esterified** .The Erythromycin esters obtained have shown better stability in an acid medium, but still require repeated administration .Indeed, pharmacokinetic evaluations of **Erythromycin stearate and Erythromycin ethyl succinate administered orally have shown that their absorption is <u>influenced by meals</u>** **.**In addition, stearate, ethyl succinate, estolate, propionate and acistrate of Erythromycin A have **enabled preparing intramuscular injectable forms**



In order to obtain formulations for intravenous use,

various salts of Erythromycin A have been prepared. The salification concerned the **dimethylamine group at C3'of D-desosamine**. The salts for therapeutic use obtained were in particular **salts of glucoheptonate** .

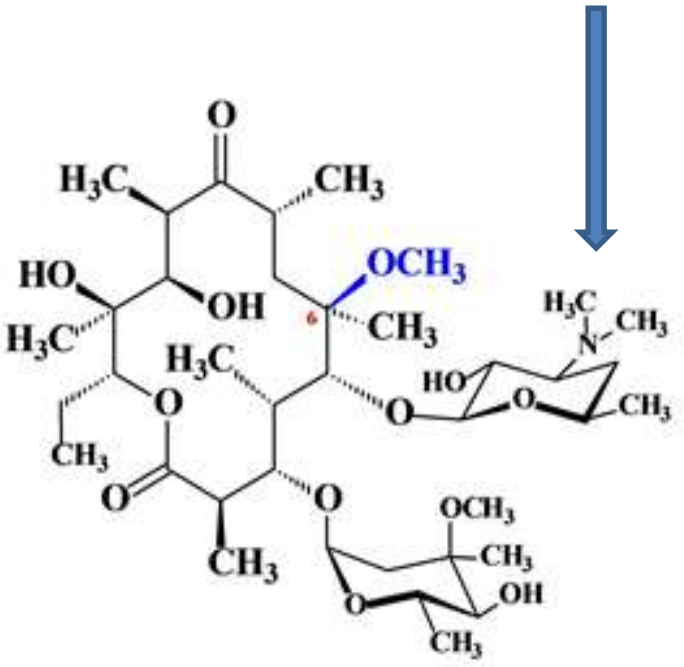
A stoichiometric mixture of Erythromycin base with one of these salts is able **to obtain an intravenous formulation.** However, these salts have the drawback of having inconsistent pharmacokinetics and of causing **venous irritations like the phlebitis**



II. Chemicals Modulations at C6 and C8: Clarithromycin and Flurithromycin

The hydroxyl at C6 involved in gastric acid medium in the hemi acetalization reaction with the ketone function at C9, was blocked by O-methylation thus leading to **O6-methyl erythromycin A or Clarithromycin** (Figure).This new molecule has shown an activity greater than or equal to that of Erythromycin A against respiratory pathogens with an increased antibacterial activity against Haemophilus influenzae, due to its active metabolite 14-hydroxyclarithromycin which acts in synergy with the parent molecule .In addition, Clarithromycin is more stable in an acid environment and therefore has fewer gastrointestinal side effects .

However, the presence of hydroxyl at position C12 and the ketone function at position C9 lead to a degradation of Clarithromycin in an acid medium, to pseudo-Clarithromycin which is inactive.

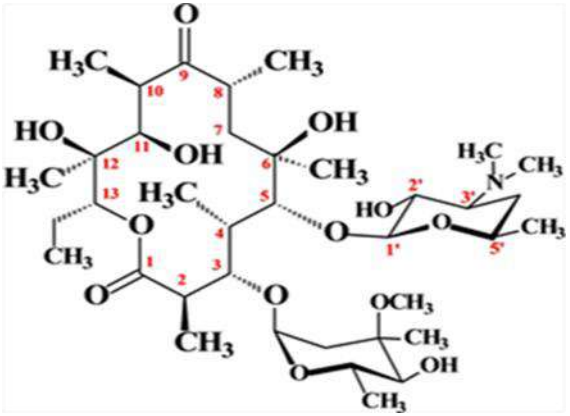


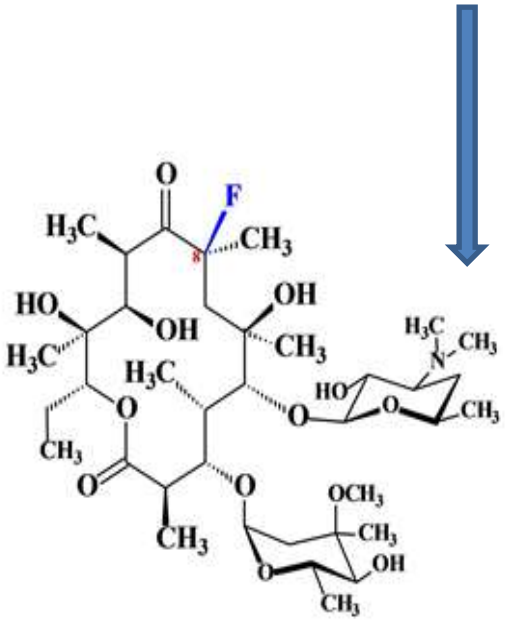
Clarithromycin

Chemicals Modulations at C6 and C8: Clarithromycin and Flurithromycin (cont.)

The hydrogen at β at the C8 position involved in the degradation of Erythromycin A in gastric acid medium has been replaced by a halogen atom, in this case fluorine. This fluorine atom minimizes the metabolism of the molecule.This strategy made it possible to obtain Flurithromycin .**Flurithromycin has the advantage of being more stable in an acid environment**. Indeed, the dehydration in C8 of Erythromycin A in anhydrohemiketal is prevented by the addition of the fluorine atom. Thus, the enolization of the ketone in C9 is annihilated. This improved stability is characterized by **a prolonged half-life time at the origin of its once-daily administration.**

Flurithromycin exhibited two to four times the antibacterial activity of Erythromycin A due to its inhibitory effects on the formation of the bacterial ribosome 30S subunit

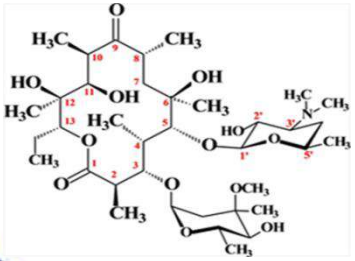


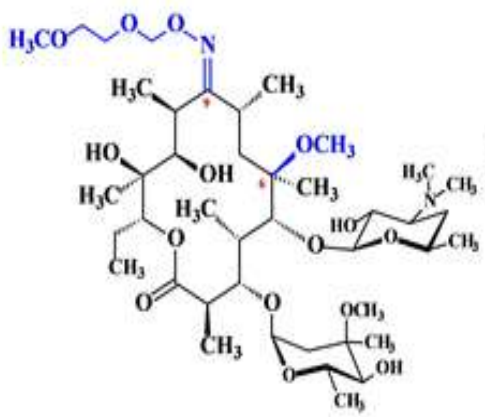


Flurithromycin

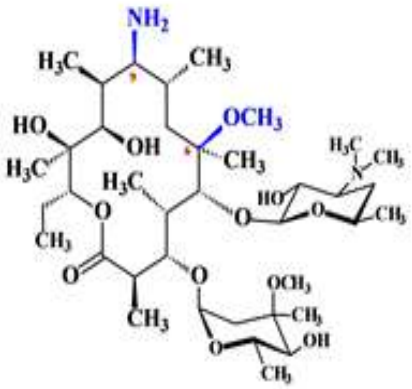
Chemical Modulation of the Ketone Function in C9

In order to prevent the formation of the inactive 6,9-hemiketal, and therefore to compensate for the degradation in gastric acid medium of Erythromycin A, its **ketone function at C9 has previously been transformed into an N-9-oxime derivative.** The latter, serving as a synthesis intermediary, subsequently underwent several chemical variations. This chemical variation at C9 ketone has led to the development of **Roxithromycin, Clarithromycin, Erythromycylamine, Dirithromycin and Azithromycin.**

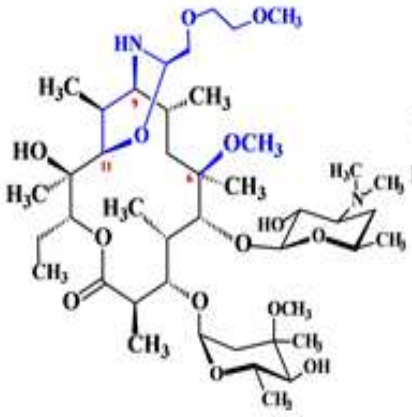




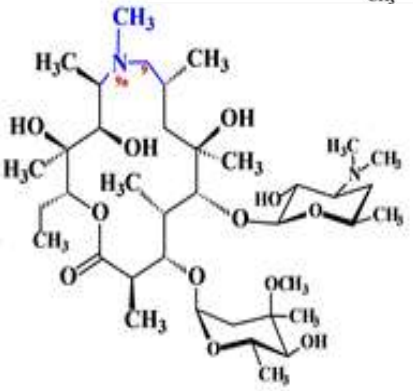
Roxithromycin



Erythromycylamine



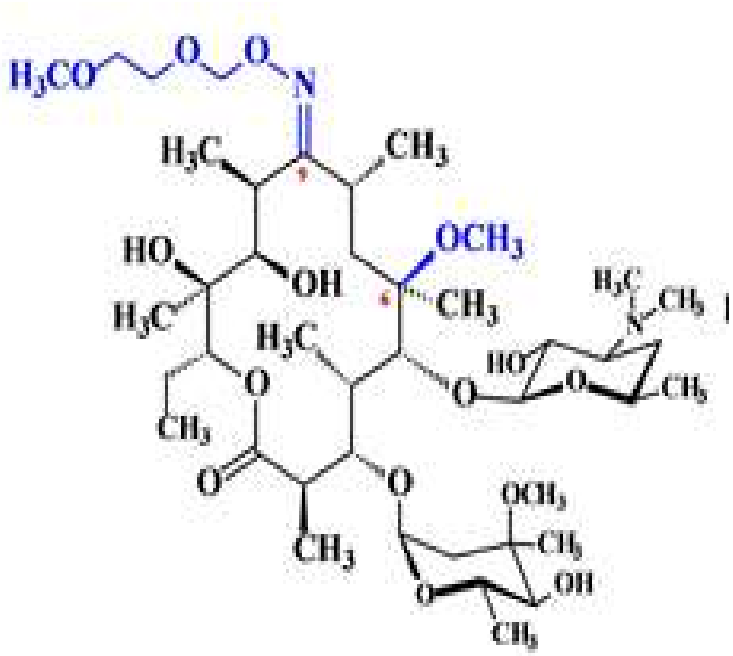
Dirithromycin



Azithromycin

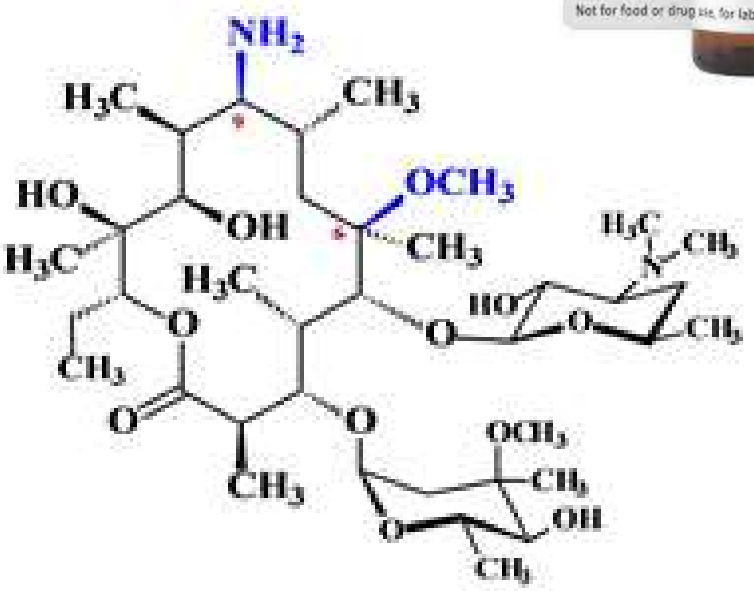
The first was an O-alkylation reaction of the hydroxyl of the oxime function, thus leading to the production of ether oximes including **Roxithromycin, the first hemisynthetic macrolide**

Roxithromycin has better stability in an acid environment and good digestive absorption, hence an increase in its bioavailability by oral route ranging **from 72% to 85% .**In addition, Roxithromycin has a weaker binding to cytochrome P450 associated with an exaltation of the elimination half-life time of the order of 12 hours, resulting in **twice-daily administration**. On the other hand, its spectrum of antibacterial activity remains **equivalent to that of Erythromycin A**



Roxithromycin

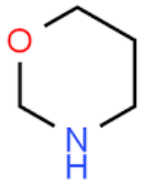
The second reaction undertaken on the N-9-oxime derivative of Erythromycin A was its reduction to an amine function. This modulation made it possible to obtain Erythromycylamine, which is very active on bacterial germs .Unfortunately, this N-9-amine from Erythromycin A has the **major drawback of being poorly absorbed orally due to the presence of the protonable amine function**, which is therefore difficult to cross the digestive membranes



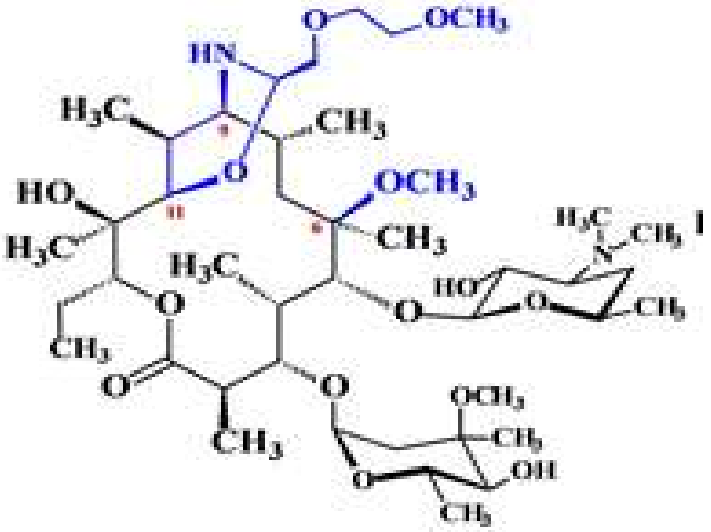
Erythromycylamine

To improve membrane crossings, Erythromycylamine underwent **heterocyclization through its amine functions at C9, hydroxyl at C11 and under the action of an acetaldehyde**. Such heterocyclization led to the formation of a **bicycle of Erythromycin A or 1,3-oxazinane of Erythromycin A also called Dirithromycin** .

1,3-oxazinane



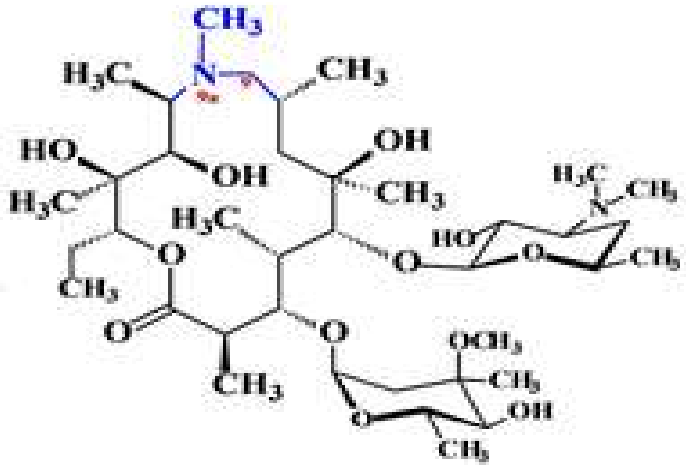
Dirithromycin is **actually a prodrug,** because in an acidic environment, it rapidly transforms into Erythromycylamine .It appears to have a good tissue concentration after oral administration, a low potential for interaction with cytochrome P450 a large volume of distribution (11 - 100 L/kg) and a half-life of high elimination (20 - 50 hours), **resulting in once-daily administrations.** However, the antibacterial activity spectrum of Dirithromycin in vitro is similar to that of Erythromycin A with low oral bioavailability (10%)



Dirithromycin

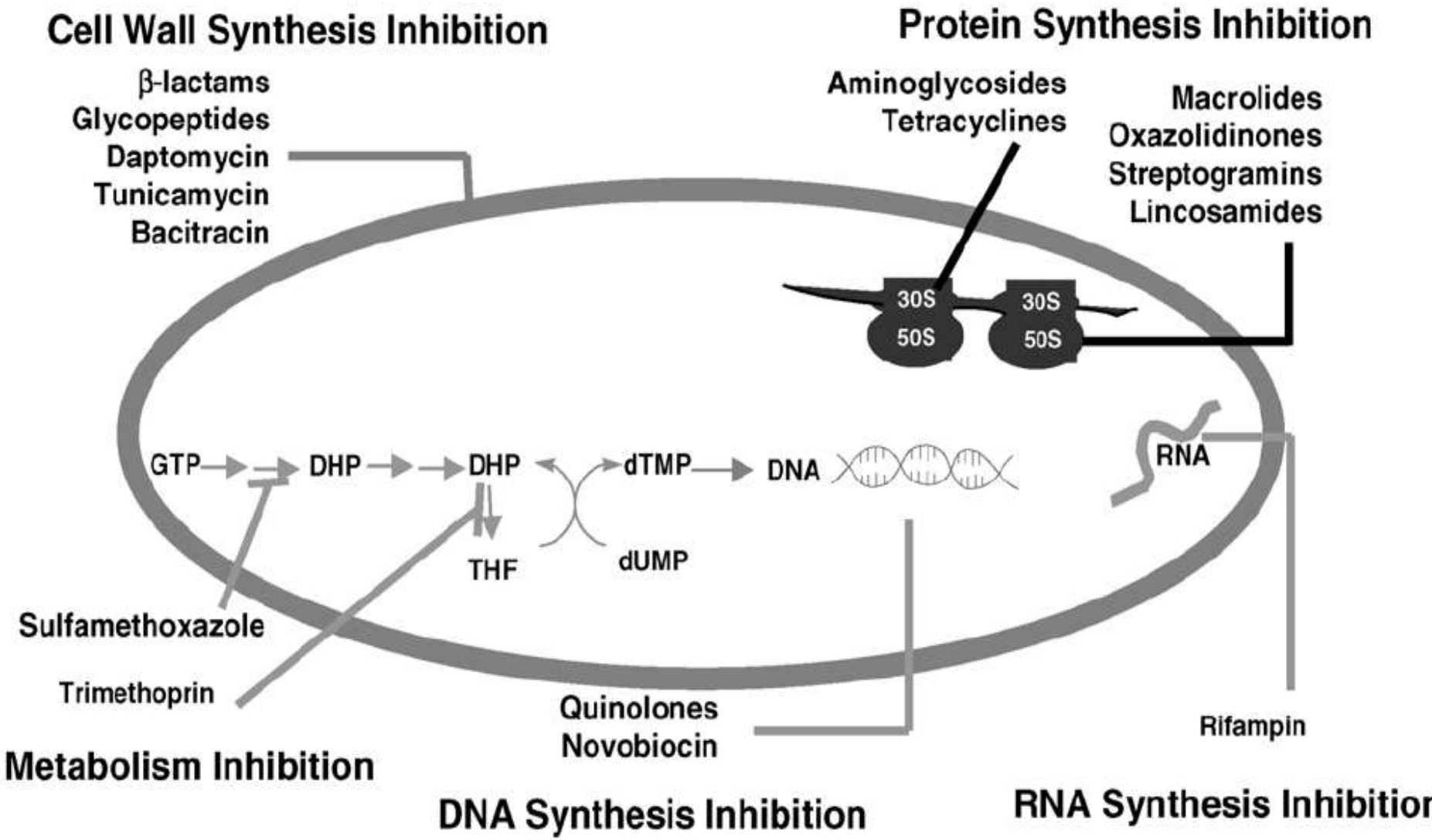
The latest reaction to the N-9-oxime derivative of Erythromycin A was that of the Beckmann rearrangement. It led to the enlargement of the lactonic macrocycle from 14-membered ring to a new macrocycle with 15-membered ring following the introduction of a nitrogen atom which undergoes methylation in the end. These are the **Azalides, the leader of which is Azithromycin.**

In the end, Azithromycin or 9-deoxo-9a-aza-9a-methyl-homoerythromycin A has **become one of the most effective antibiotics in the world and currently contributes, due to its exceptional therapeutic properties, to the improvement of quality of life globally**.



Azithromycin

Antibacterial Targets:



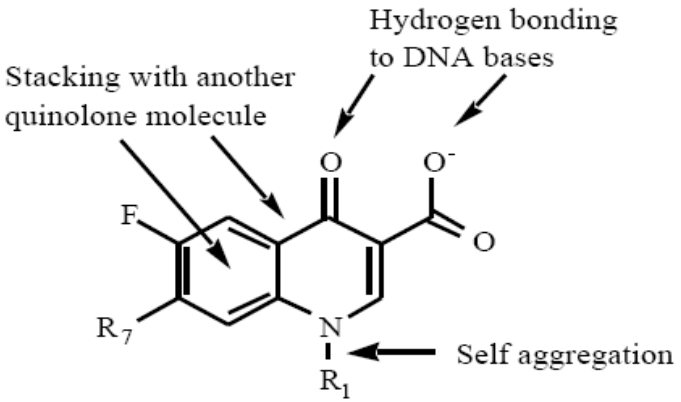
Bacterial DNA Synthesis: Gyrase and Topo IV: Quinolone Inhibitors

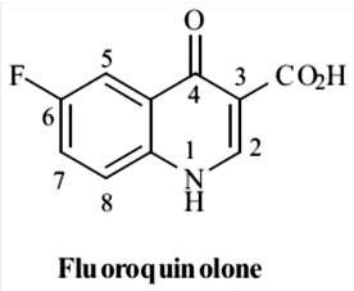
Cooperative Binding Model:

- drug binds to cleaved single-stranded DNA and thereby traps the enzyme
- four quinolone molecules bind cooperatively to DNA via H-bonds to DNA bases
- quinolones stack onto each other to form pairs
- quinolones aggregate via substituents at 1 and 8 position

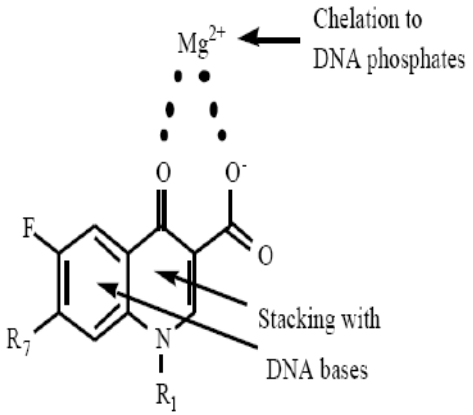
Mg2+ Bridge Model:

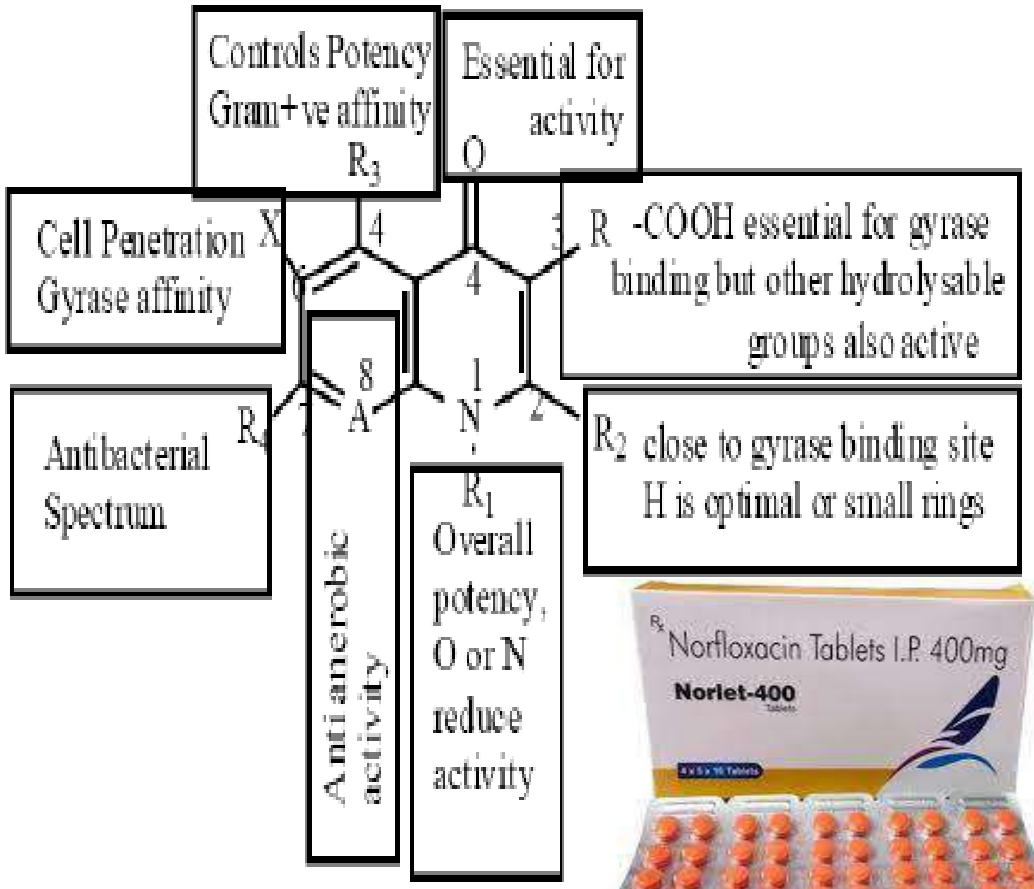
- drug binds to DNA phosphates via chelation of Mg2+
- quinolone ring stacks onto DNA bases
- quinolone binding induces a conformational change in the gyrase-DNA complex
- cleavage of DNA is not necessary for drug binding





Fluoroquinolone





R; -COOH, -COOR'; Coumarin, penems, carbapenems or monobactam etc.

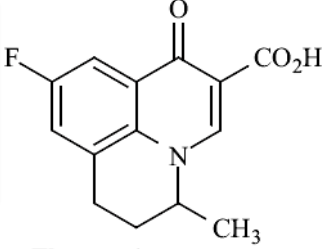
R_1: halo atom, halo substituted aromatic ring, hetro atom, (substituted) C_{1-6} alkyl, (Substituted) C_{3-6} cycloalkyl, (Substituted) aryl etc.

R_2: H, -SCH_3, C_{1-6} alkylthio, or R_2 & R_1 may join to form a ring.

Synthetic compounds discovered 1962 as a by-product of an antimalarial program.

- Not used as antibiotics until discovery **of improved 6-fluoroquinolones in 1980s (norfloxacin).**
- Orally bioavailable; excellent distribution; active against G+/-
- Resistance development can be rapid by target modification (mutation in the quinolone resistance determining region QRDR of gyrase/topo IV) and by active efflux.

Bacterial DNA Synthesis: Gyrase and Topo IV: Quinolone Inhibitors



Flumequine

1)First generation: nalidixic acid, oxolinic acid, pipedinic acid, **flumequine,** ...

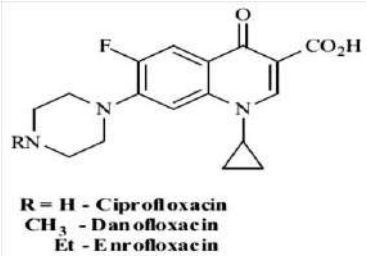
2)Second generation (**most potent against *Pseudomonas***): norfloxacin, ciprofloxacin, enoxacin, fleroxacin, **ofloxacin,** levofloxacin, lomefloxacin, ...

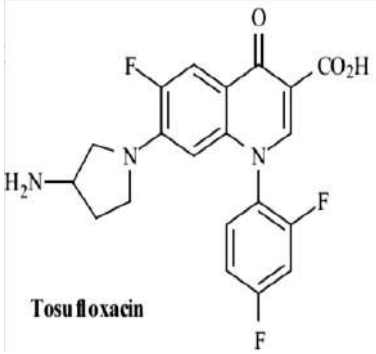
3)Third generation (**more potent** against *Pneumococcus* and anaerobes): sparfloxacin, tosufloxacin, gatifloxacin, pazufloxacin, grepafloxacin, ...

4)Fourth generation (**most potent** against *Pneumococcus* and anaerobes): trovafloxacin, clinafloxacin, sitafloxacin, moxifloxacin, **gemifloxacin**

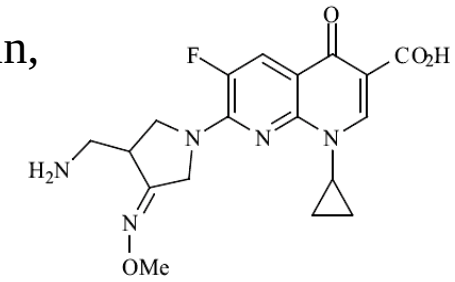
Respiratory Q (active against Streptococcus, Staphylococcus, Haemophilus): levofloxacin

Antipseudomonas Q (active against *Pseudomonas*, *Haemophilus*): ciprofloxacin, ofloxacin



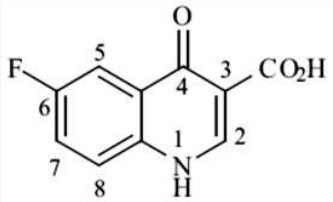


Tosufloxacin



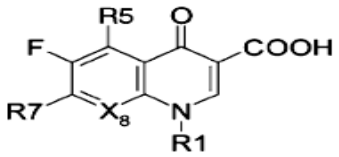
Gemifloxacin (GMFX)

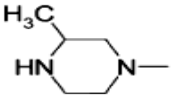
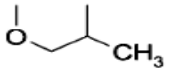
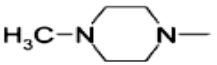
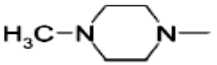
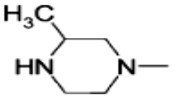
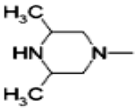
Phototoxicity of fluoroquinolones with halogen substituents at 8-position)



Fluoroquinolone

Structure–phototoxicity relationships in female Balb/c mice receiving a single intravenous administration of quinolone irradiation for 4 h

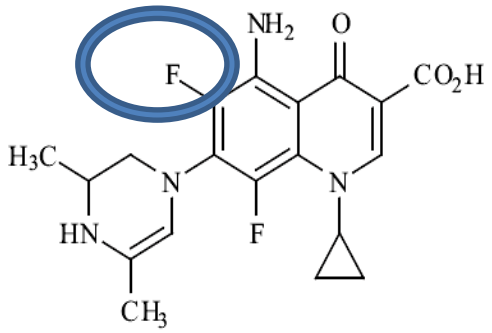


Drug	X8	R1	R5	R7	Phototoxicity
Gatifloxacin	$COCH_3$		H		–
Ofloxacin					–
Ciprofloxacin	CH		H		+
Norfloxacin	CH	C_2H_5	H		+
Enoxacin	N	C_2H_5	H		++
Fleroxacin	CF	CH_2CH_2F	H		++
Lomefloxacin	CF	C_2H_5	H		+++
Sparfloxacin	CF		NH_2		+++

Phototoxic potential was assessed by the results of the thickness and histopathological findings of the auricle at 96 h post-dose. The auricular thickness of lomefloxacin and sparfloxacin was estimated at 48 h post-dose because the auricles showed focal loss and could not be measured after this timepoint. (–) none, (+) mild, (++) moderate, (+++) severe.

Phototoxicity of fluoroquinolones with halogen substituents at 8-position

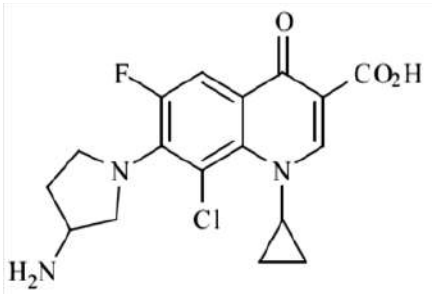
- Sparfloxacin (Zagam, Mylan/Aventis); approved 1996; withdrawn 1998 due to phototoxicity
- Clinafloxacin (Warner Lambert/Pfizer); stopped clinical development due to phototoxicity



Sparfloxacin

• Animal **studies show joint & cartilage damage in weight-bearing joint**s of young animals (dogs; effect animal- & dose-dependent)

- All fluoroquinolones have shown this toxicity
- Mechanism unclear
- Fluoroquinolones not approved for use in children (except in CF)
- Compassionate use cases suggest that this toxicity is very rare in children

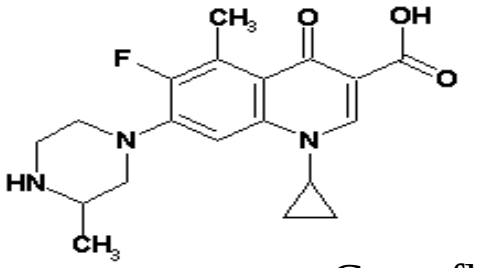


CI-960 (Clinafloxacin)

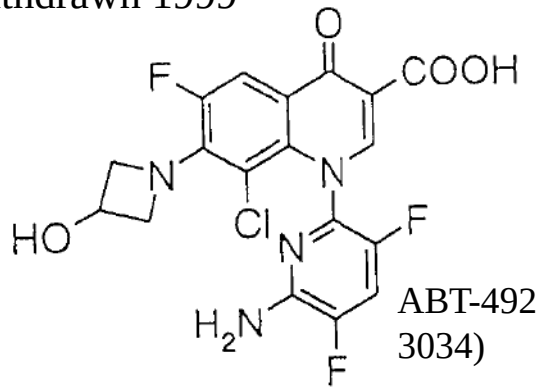
ABT-492 (WQ-3034)

• QT interval prolongation (hERG) observed with some fluoroquinolones

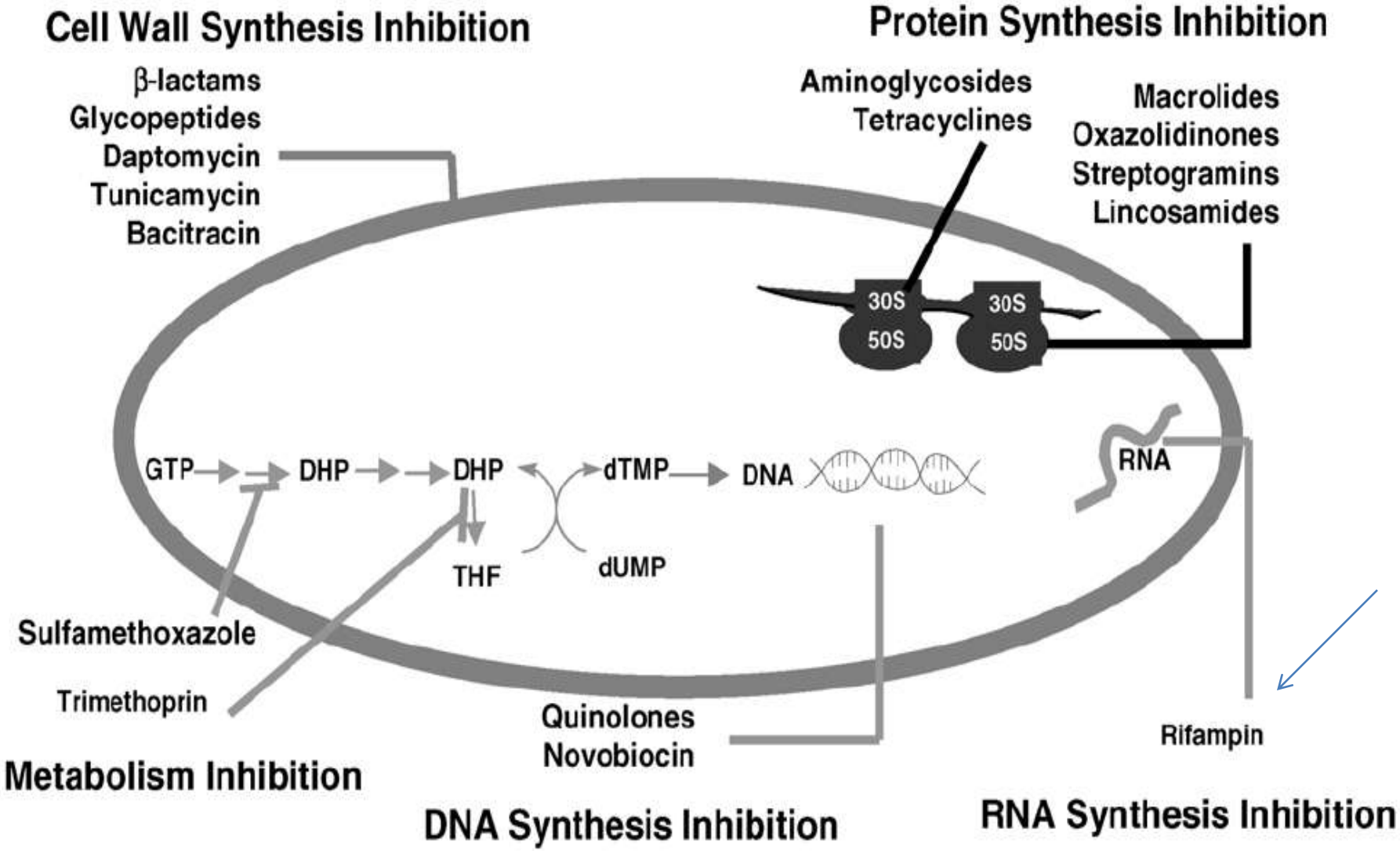
- Grepafloxacin (Raxar, Glaxo); approved 1997; voluntarily withdrawn 1999

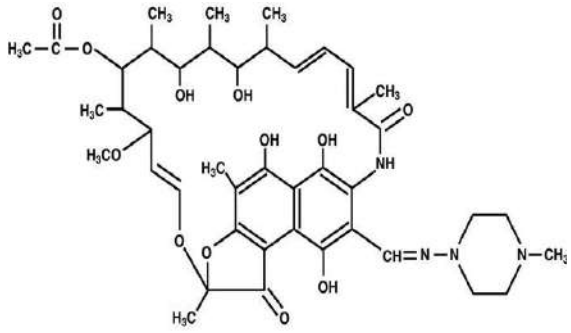


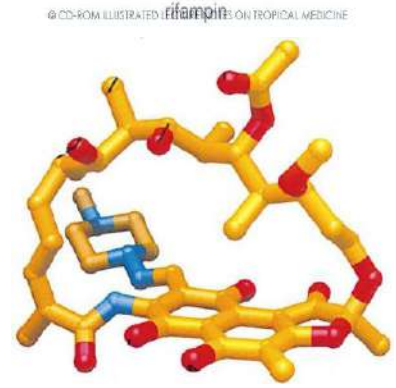
Grepafloxacin

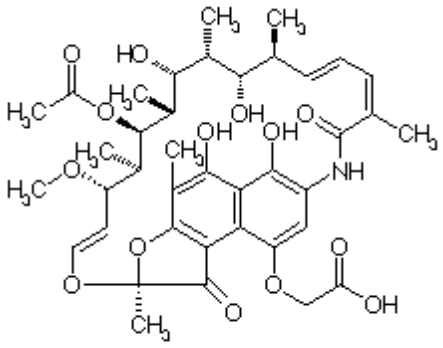


RNA SYNTHESIS INHIBITION: Rifamycins









Rifampin

- **Ansamycin class antibiotic**
- semisynthetic rifamycin derivative
- isolated 1957 from *Nocardia mediterranei*
- inhibits selectively **bacterial RNA polymerase** by binding to the ß-subunit, >12Å away from the active site
- binding site is highly conserved among bacteria but not in eukaryotic RNAPs
- blocks the exit path of elongating RNA when transcript is 2-3 nucleotides in length
- **bacteriostatic**
- active against G+/-, however MICs for G- are higher because of reduced outer membrane penetration
- primarily used against Mycobacteria (tuberculosis, leprosy) and Meningococci
- resistance develops easily by RNAP mutations that reduce target affinity
- often used in combination with other bactericidal antibiotics

The Organic Chemistry of Drug Design and Drug Action

BY:

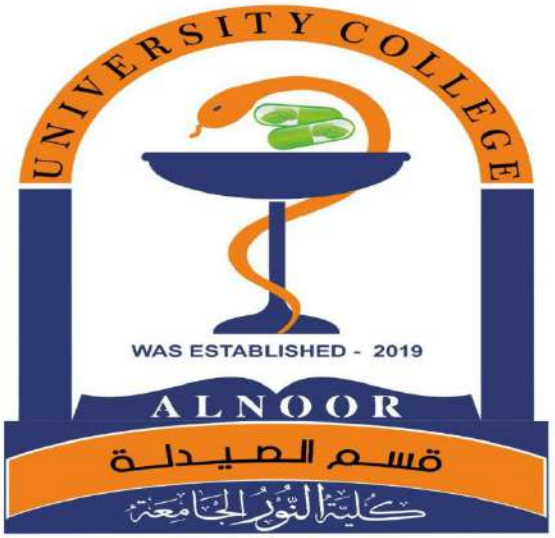
Dr. Nohad A AlOmari (B.Pharm/MSc & PhD in Medicinal Chemistry)

Organic Pharm. Chemistry III

Lecture 1

20th February (5th March)

10:30- 12:30



AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023

Today ‘s Inspiring Quote

Lectures titles & Credit hours

AlNoor University College
Pharmacy Department
Department of Pharmaceutical Chemistry
Title of the course: ***Organic Pharmaceutical Chemistry*** **III** Course number:
Level: 4th Class, 2nd Semester
Credit hours/week : **Theory 3** **Laboratory 1**
Reference text: ***Wilson and Gisvold Textbook of Organic Medicinal and Pharmaceutical Chemistry; Delgado JN, Remers WA, (Eds.); 10th ed., 2004.***

مدرس المادة :- أ.م.د.نهاد عبد الوهاب

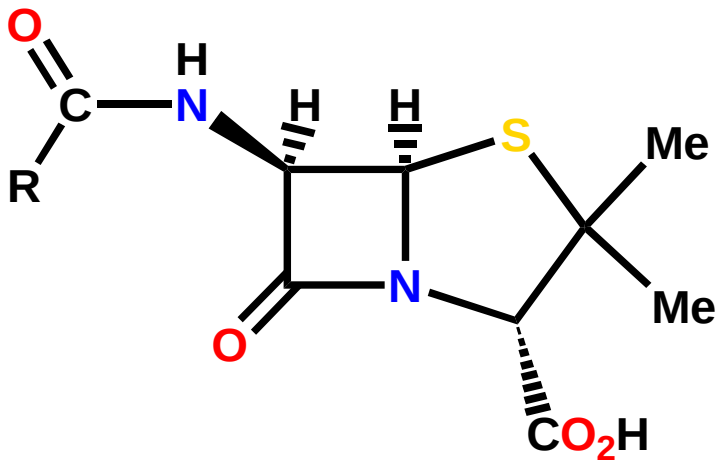
Objectives: To enable understanding mechanisms of drug action, including antibacterial, antifungal and antiviral agents, at molecular level, and the role of medicinal chemistry in the discovery and development of synthetic therapeutic agents. It also enables students to understand the concept of structure-activity relationship and its application in design and synthesis of new chemotherapeutic agents and hormone derivatives with potential biological activity.

Lecture title	hours
Lecture title	**hours**
β-Lactam antibiotics (Penicillins); β-Lactamase inhibitors; Cephalosporins and Monobactams.	9
Aminoglycosides and Chloramphenicol; Tetracylines; Macrolides; Lincomycins and Polypeptides; Antiviral agents (properties of viruses, viral classification, products).	9
Sulfonamides (chemistry, nomenclature, mechanism of action, resistance, toxicity, side effects, metabolism, protein binding, distribution and SAR); products; Sulfones.	4
Anti-neoplastic agents: Alkylating agents; Antimetabolites; Antibiotics; Plant products; Miscellaneous compounds.	17
Hormones and related compounds; Future anti-neoplastic agents; Monoclonal antibodies; Gene therapy of cancer	6

PENICILLINS

Lec. 1

20/02/2023



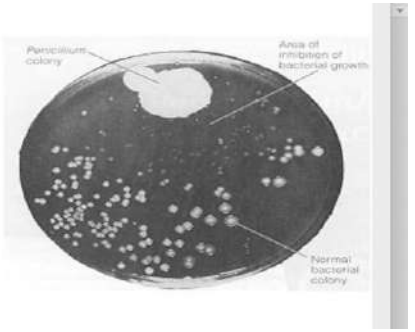
INTRODUCTION TO PENICILLINS

•Antibacterial agents which inhibit bacterial cell wall synthesis
•**Discovered by Fleming from a fungal colony (1928)**
•Shown to be non toxic and antibacterial
•Isolated and purified by Florey and Chain (1938)
•First successful clinical trial (1941)
•Produced by large scale fermentation (1944)
•Structure established by X-ray crystallography (1945)
•1945: Nobel Prize A. Fleming / H. Florey;
•E. B. Chain
•1947: Nobel Prize in Chemistry Robert
•Robinson performs the first synthesis of
•penicillin.
•Full synthesis developed by Sheehan (1957)
•Isolation of 6-APA by Beechams (1958-60)
- development of semi-synthetic penicillins
•Discovery of clavulanic acid and b-lactamase inhibitors


Fleming


Florey

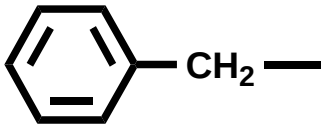

Chain




Robison

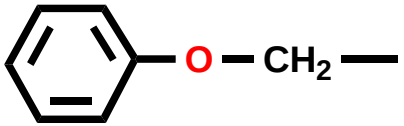
STRUCTURE

R =

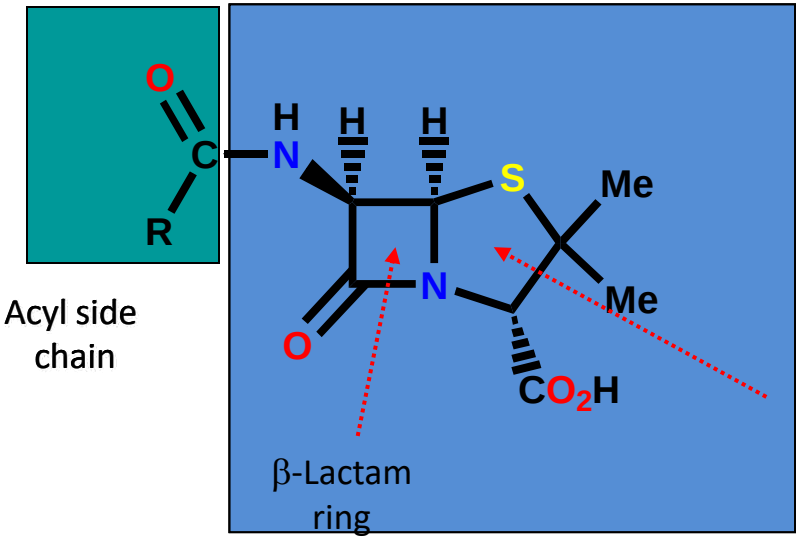


Benzyl penicillin (Pen G)

R =



Phenoxymethyl penicillin (Pen V)



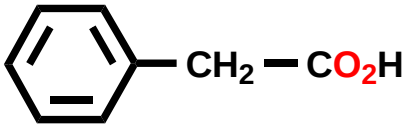
Acyl side chain

6-Aminopenicillanic acid (6-APA)

Thiazolidine ring

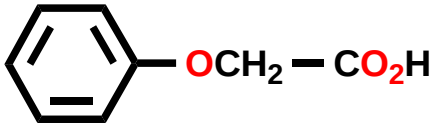
β-Lactam ring

Side chain varies depending on carboxylic acid present in fermentation medium



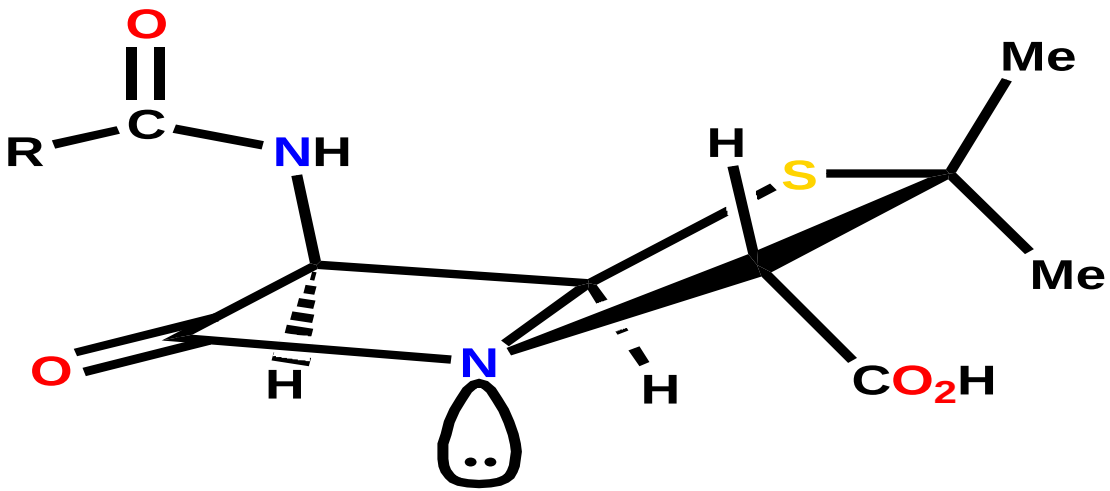
present in corn steep liquor

→ Penicillin G



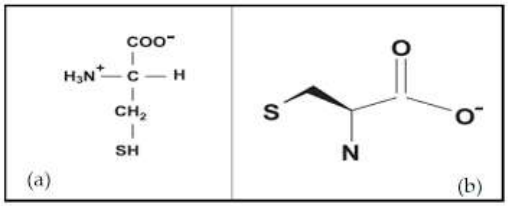
→ Penicillin V
(first orally active penicillin)

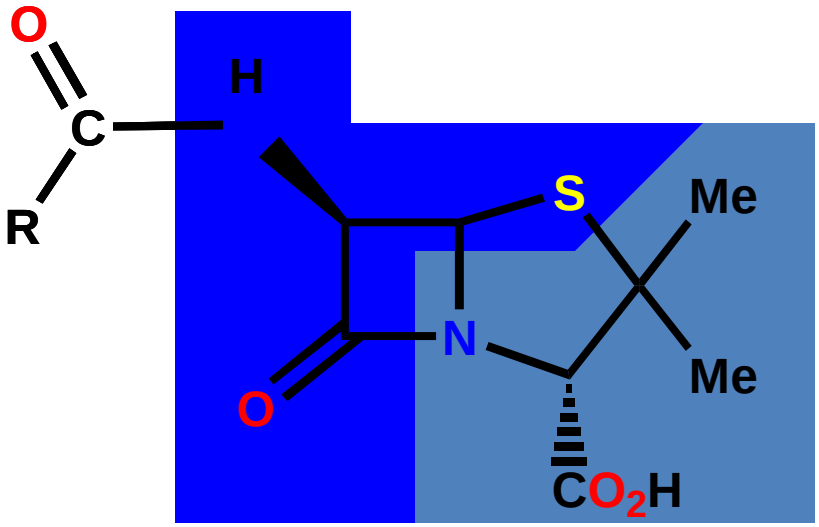
Shape of Penicillin G



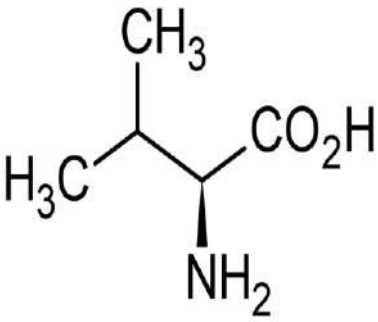
Folded ‘envelope’ shape

Biosynthesis of Penicillins





CYS



VAL

Properties of Penicillin G

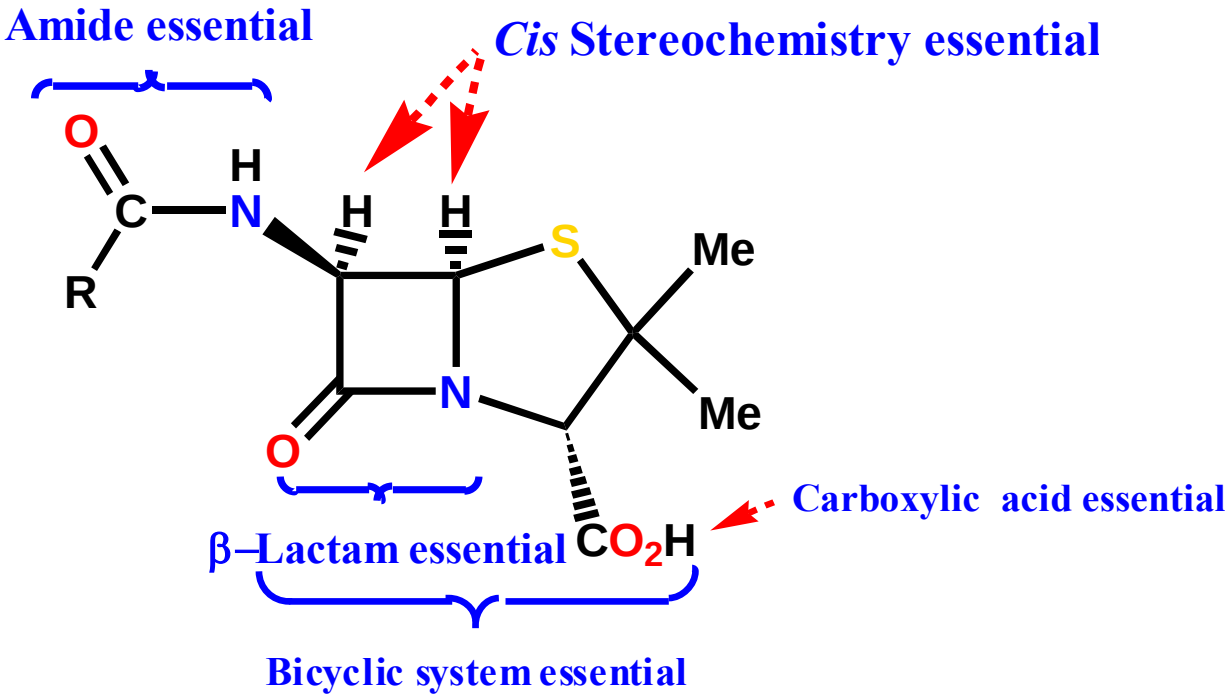
- Active vs. Gram +ve bacilli and some Gram -ve cocci
- Non toxic
- Limited range of activity
- Not orally active - must be injected
- Sensitive to ß-lactamases
  (enzymes which hydrolyse the ß-lactam ring)
- Some patients are allergic
- Inactive vs. *Staphylococci*

Drug Development

Aims

- To increase chemical stability for oral administration
- To increase resistance to ß-lactamases
- To increase the range of activity

SAR

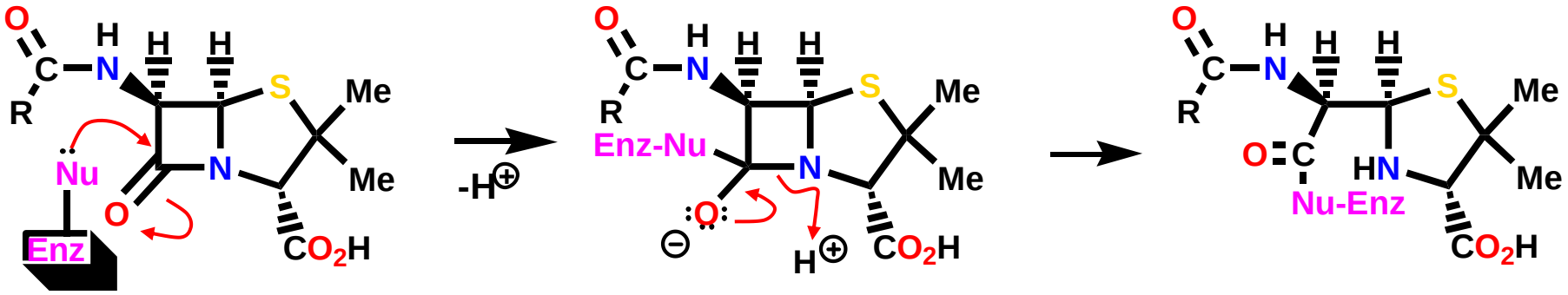


Conclusions

- Amide and carboxylic acid are involved in binding
- Carboxylic acid binds as the carboxylate ion
- Mechanism of action involves the ß-lactam ring
- Activity related to ß-lactam ring strain (subject to stability factors)
- Bicyclic system increases b-lactam ring strain
- Not much variation in structure is possible
- Variations are limited to the side chain (R)

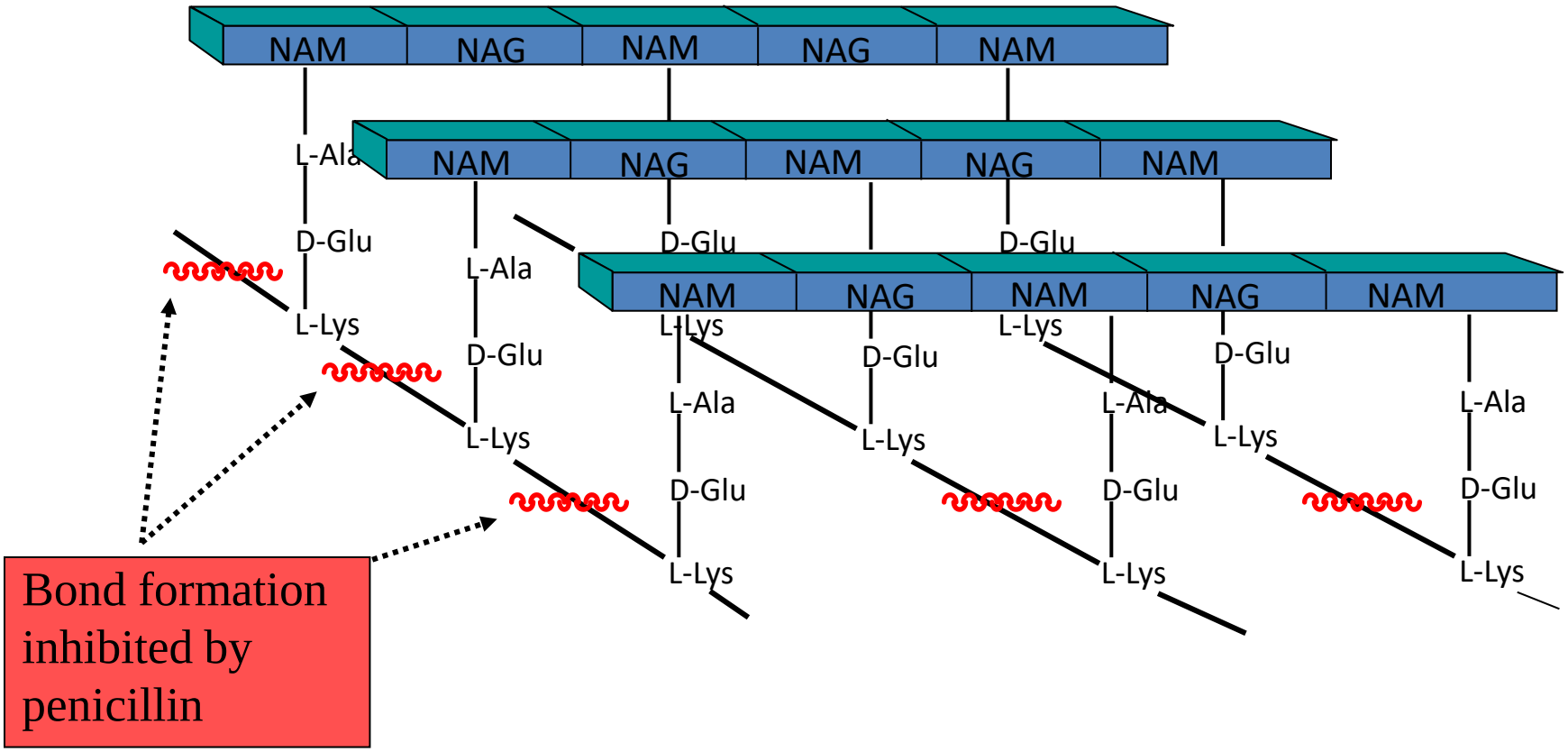
Mechanism of action

•Penicillins inhibit a bacterial enzyme called the transpeptidase enzyme which is involved in the synthesis of the bacterial cell wall
•The ß-lactam ring is involved in the mechanism of inhibition
•Penicillin becomes covalently linked to the enzyme's active site leading to irreversible inhibition

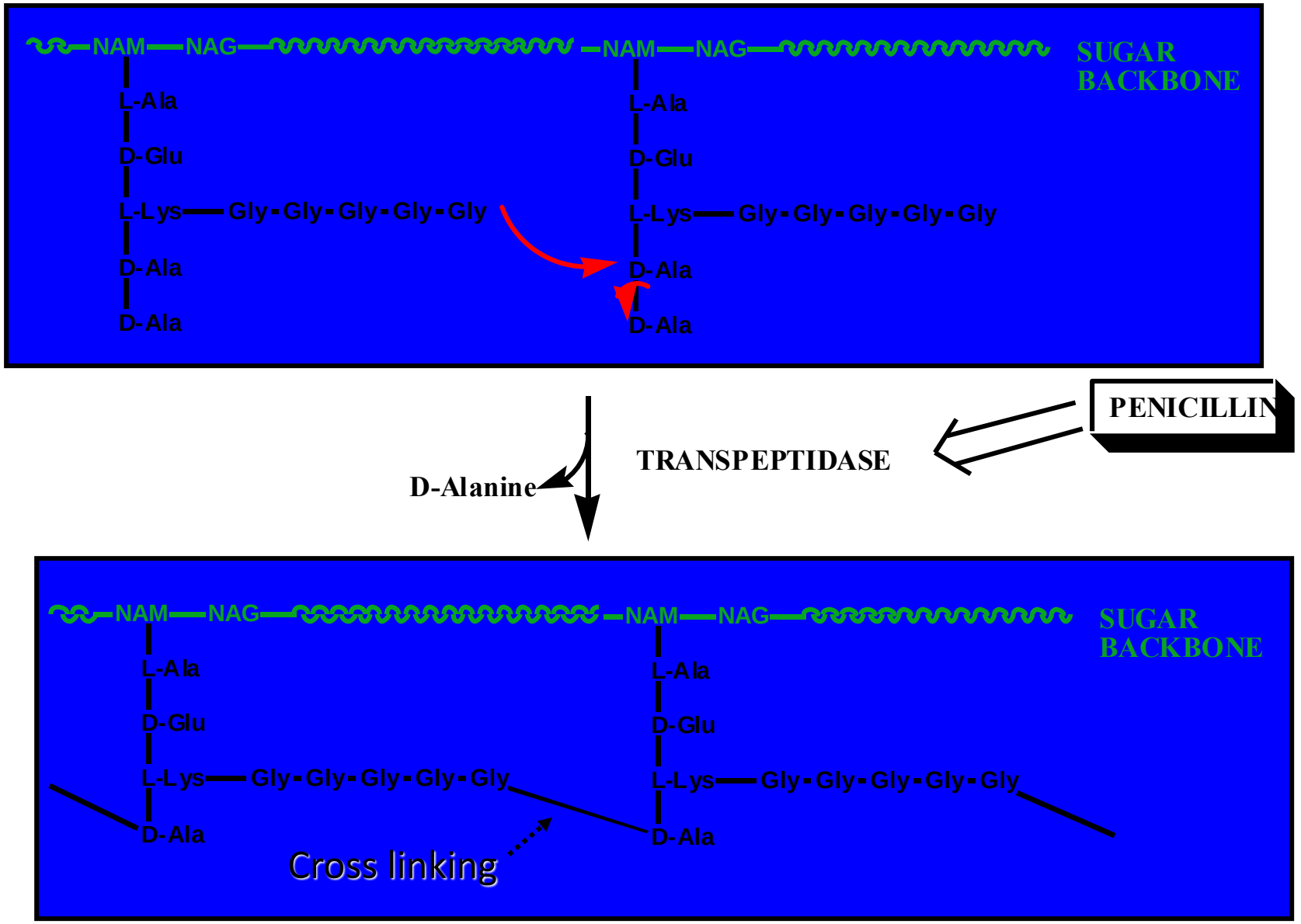


•Covalent bond formed to transpeptidase enzyme
•Irreversible inhibition

Mechanism of action - bacterial cell wall synthesis



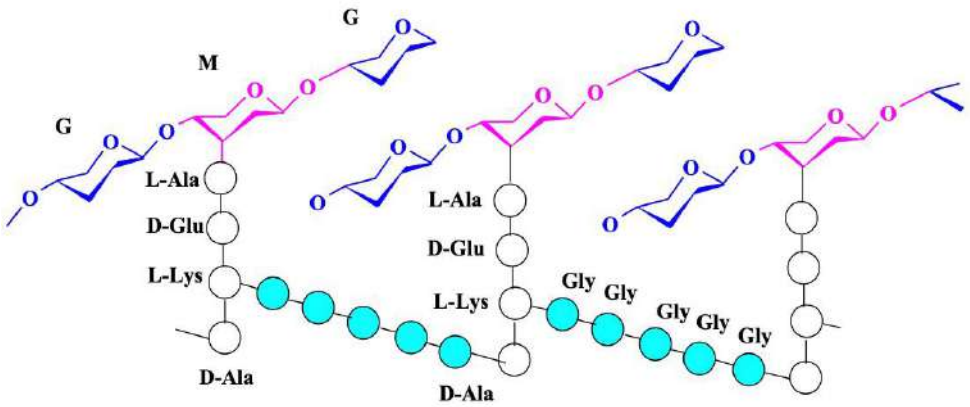
Mechanism of action - bacterial cell wall synthesis

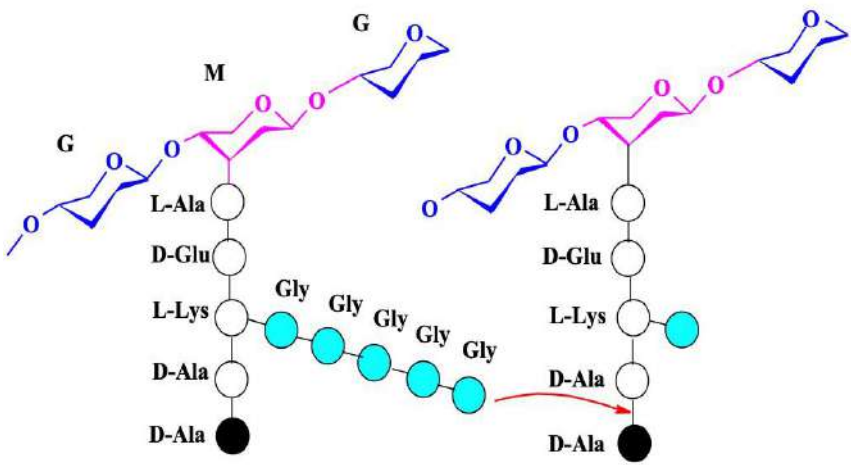


Mechanism of action - bacterial cell wall synthesis

•Penicillin inhibits final cross linking stage of cell wall synthesis
•It reacts with the transpeptidase enzyme to form an irreversible covalent bond
•Inhibition of transpeptidase leads to a weakened cell wall
•Cells swell due to water entering the cell, then burst (lysis)
•Penicillin possibly acts as an analogue of the L-Ala-g-D-Glu portion of the pentapeptide chain. However, the carboxylate group that is essential to penicillin activity is not present in this portion.

•https://www.google.com/search?q=why+penicillin+inhibit+transpeptidase+enzyme&rlz=1C1GCEA_enIQ982IQ982&sxsrf=AJOqlzVi_UmRLUEmoIMWzlOfZXb0Vfhddg:1677689572191&source=lnms&tbm=vid&sa=X&ved=2ahUKEwiznfnXmLv9AhU_R_EDHXmXA18Q_AUoAXoECAEQCg&biw=1292&bih=735&dpr=0.8#fpstate=ive&vld=cid:640ee296,vid:a81nHSqQuvI

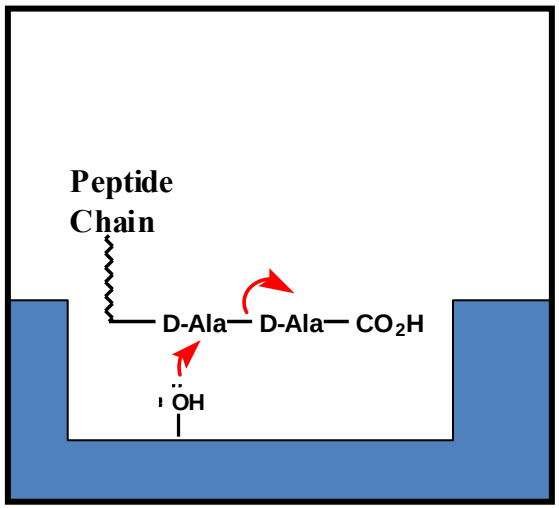


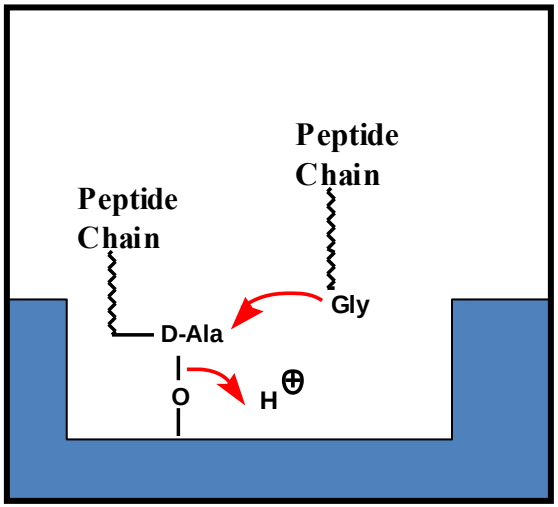


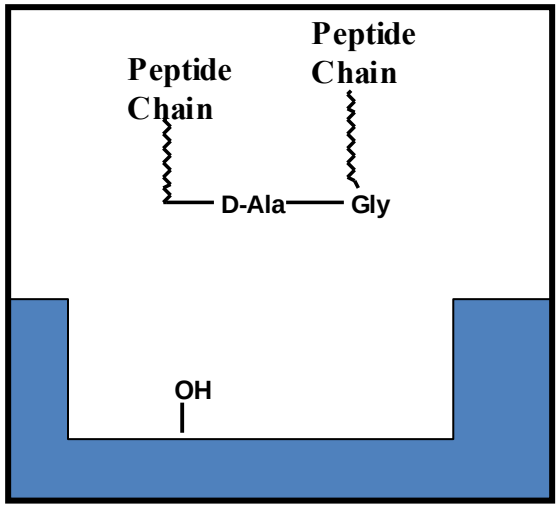
Mechanism of action - bacterial cell wall synthesis

Alternative theory- Pencillin mimics D-Ala-D-Ala.

Normal Mechanism



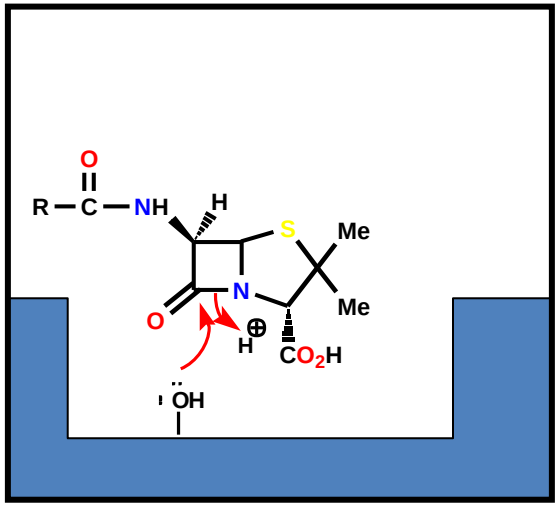


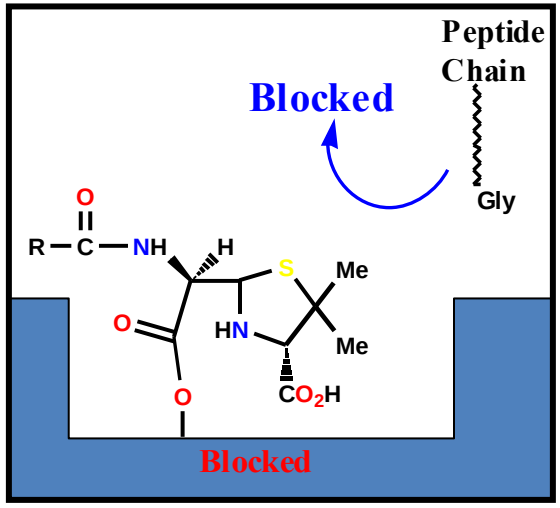


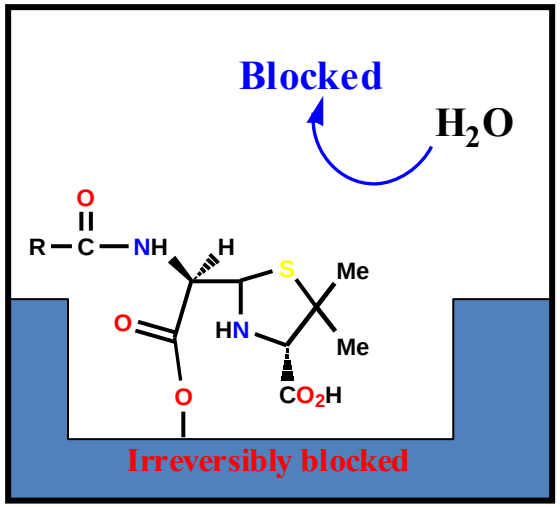
Mechanism of action - bacterial cell wall synthesis

Alternative theory- Pencillin mimics D-Ala-D-Ala.

Mechanism inhibited by penicillin

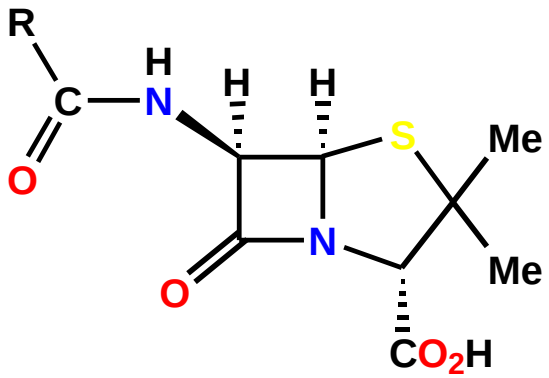




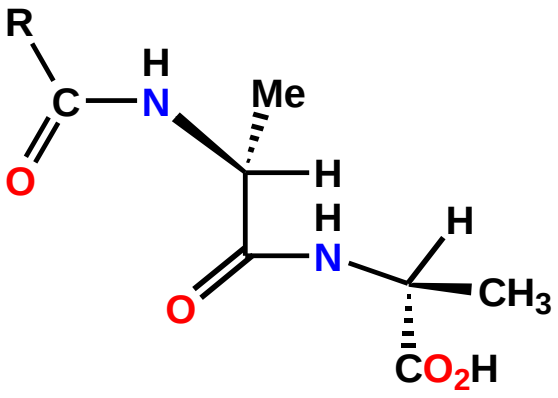


Mechanism of action - bacterial cell wall synthesis

Penicillin can be seen to mimic acyl-D-Ala-D-Ala

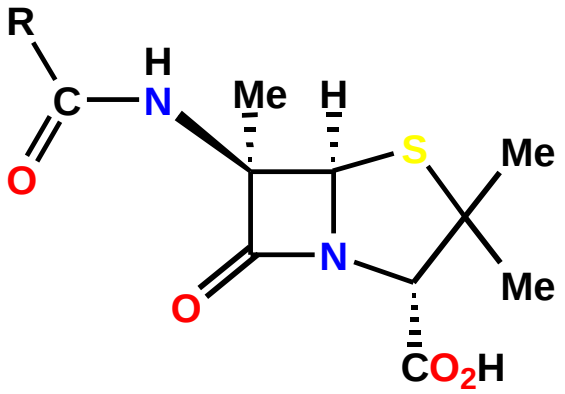


Penicillin



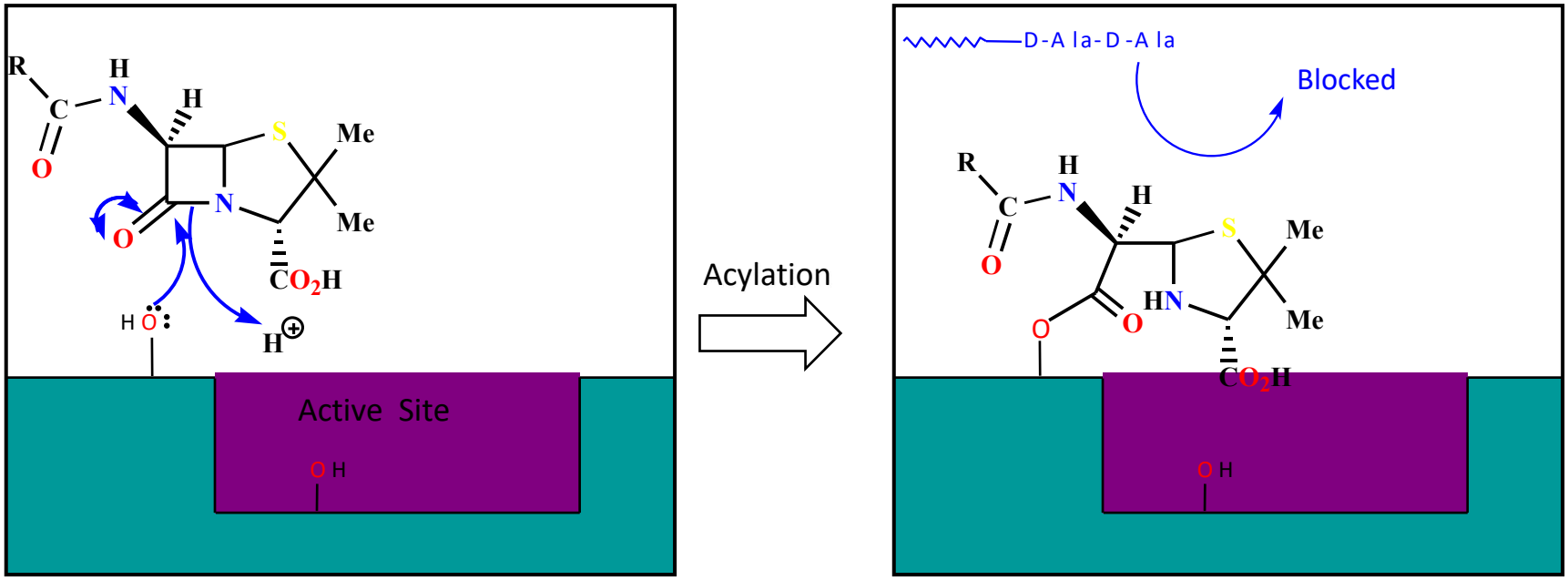
Acyl-D-Ala-D-Ala

But 6-methylpenicillin is inactive despite being a closer analogue



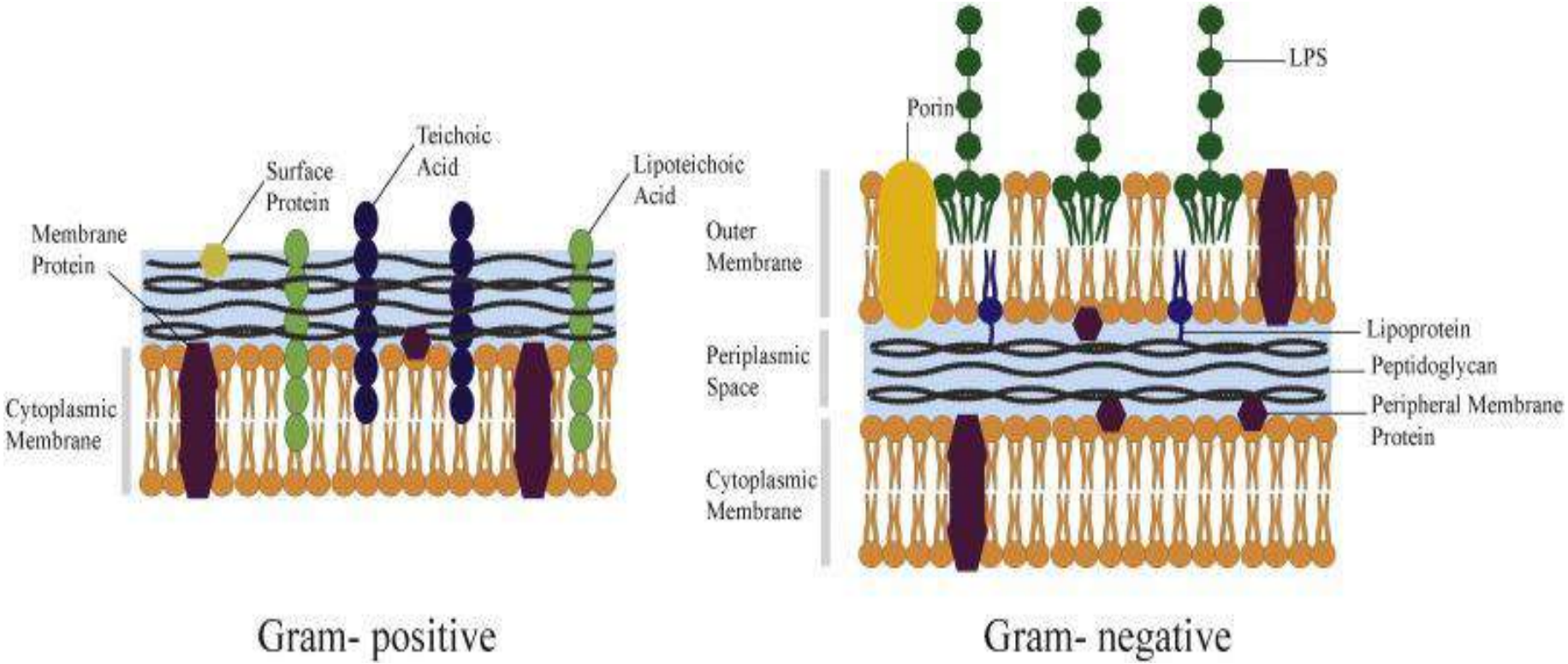
Mechanism of action - bacterial cell wall synthesis

Penicillin may act as an ‘umbrella’ inhibitor



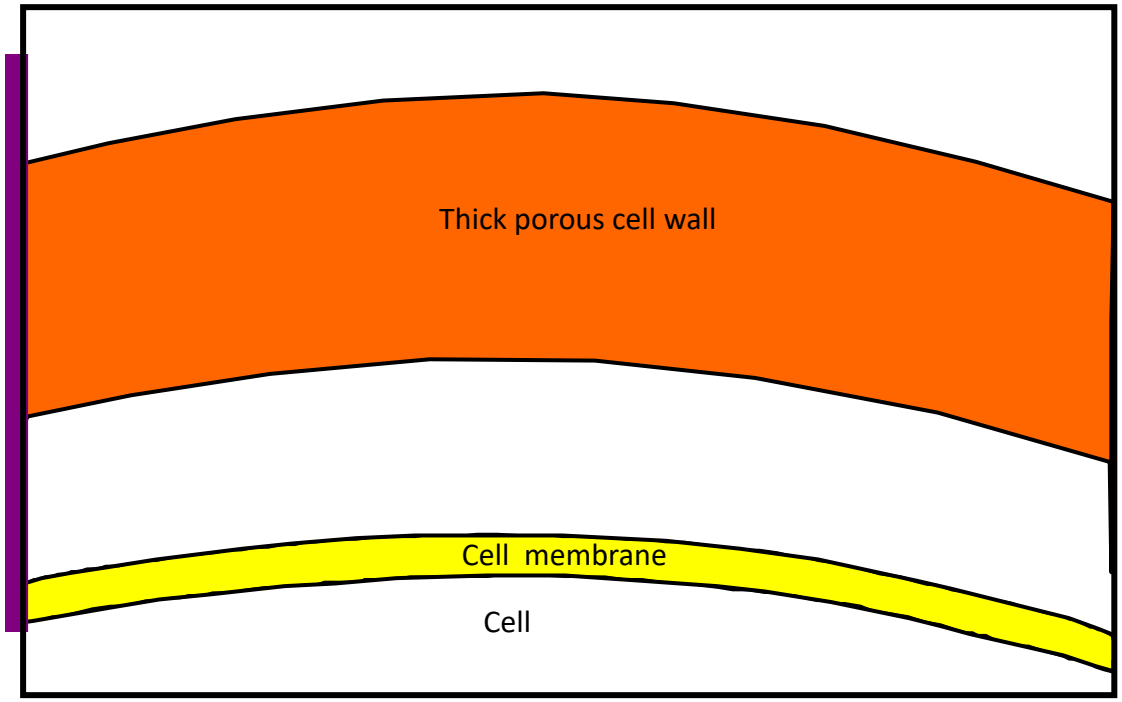
Gram +ve and Gram -ve Cell Walls

•Penicillins have to cross the bacterial cell wall in order to reach their target enzyme.
•Cell walls are porous and are not a barrier.
•The cell walls of Gram +ve bacteria are thicker than Gram -ve cell walls, but the former are more susceptible to penicillins



Gram +ve and Gram -ve Cell Walls

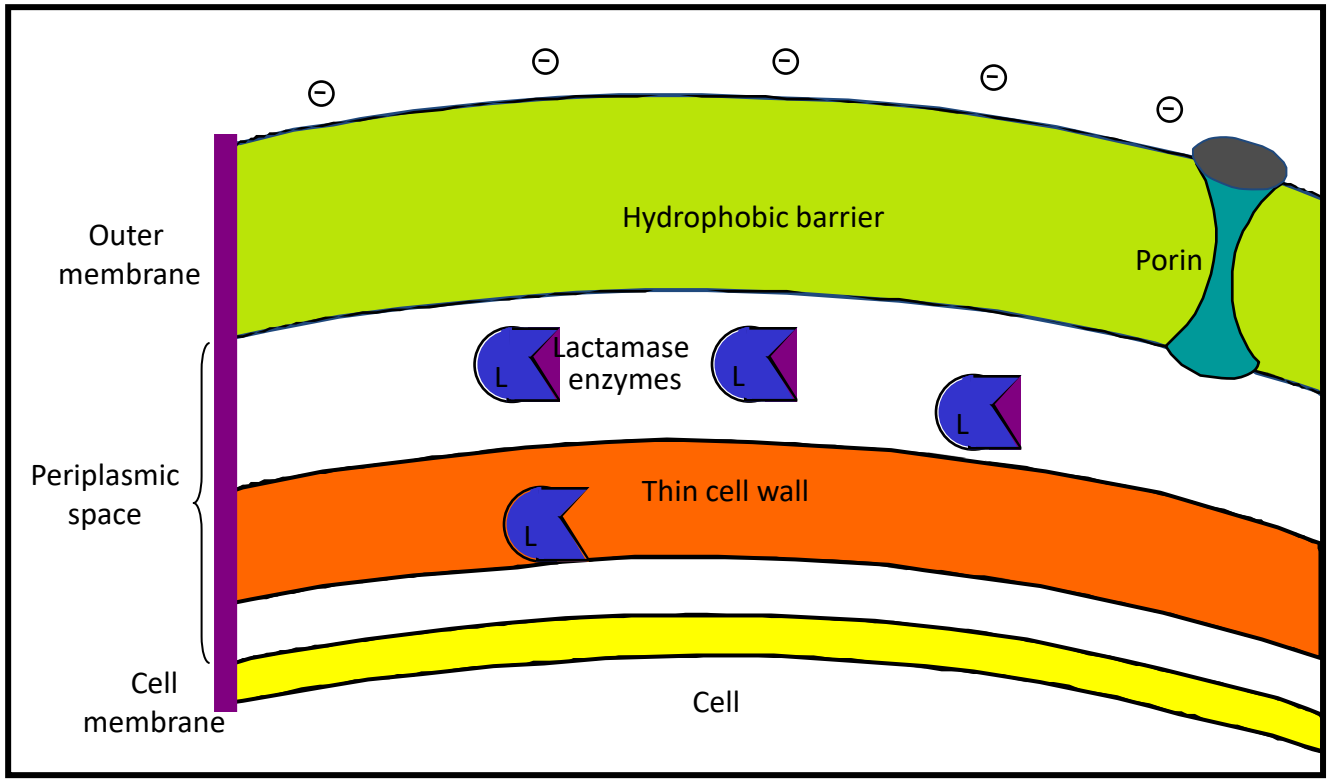
Gram +ve bacteria



- Thick cell wall
- No outer membrane
- More susceptible to penicillins

Gram +ve and Gram -ve Cell Walls

Gram -ve bacteria



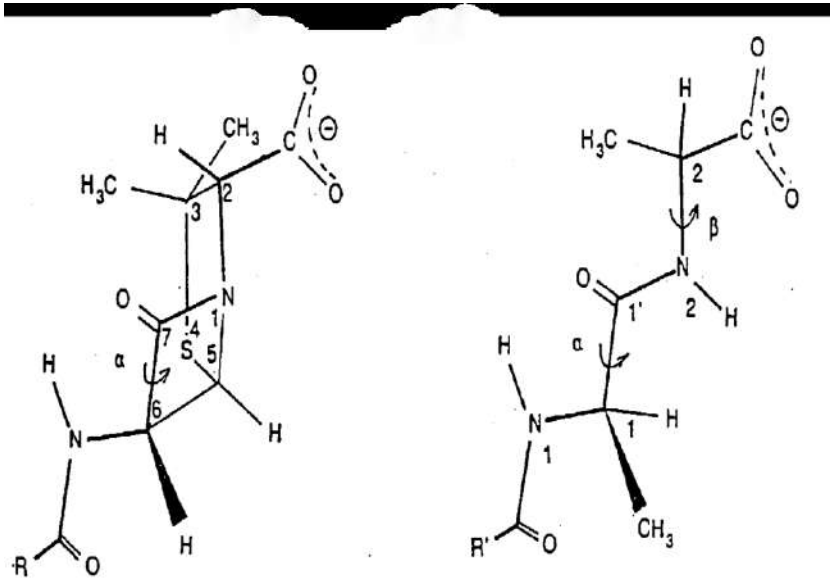
- Thin cell wall
- Hydrophobic outer membrane
- More resistant to penicillins

Mechanisms of action

Bacteriostatic action

b-lactams are **reversible transpeptidase** inhibitors (PLP) which bind pentaGly to the tetrapeptide.

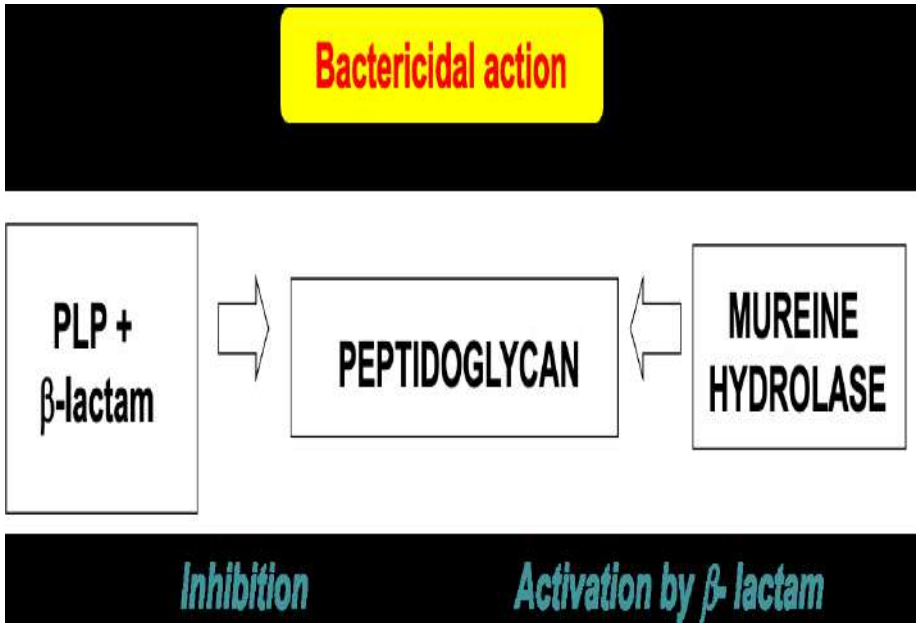
The PLP substrate is therefore the D-**Ala-D-Ala** residue of the precursor



Bactericidal action

Other mechanisms of action

The b-lactams induce the **release of inhibition of various autolytic systems**, in particular **murein hydrolase** which splits the bond between *N*-acetylmuramic acid and alanine → disorganization of the peptidoglycan → lethal effect → **Bactericidal** action



Resistance to Penicillins

Factors

•Gram -ve bacteria have a lipopolysaccharide outer membrane preventing access to the cell wall
•Penicillins can only cross via porins in the outer membrane
•Porins allow small hydrophilic molecules such as zwitterions to cross

•High levels of transpeptidase enzyme may be present
•The transpeptidase enzyme may have a low affinity for penicillins
(e.g. PBP 2a for *S. aureus*)

•Presence of b-lactamases
•Concentration of b-lactamases in periplasmic space

•Mutations
•Transfer of b-lactamases between strains
•Efflux mechanisms pumping penicillin out of periplasmic space

Penicillin Analogues - Preparation

1) By fermentation

- Vary the carboxylic acid in the fermentation medium
- Limited to unbranched acids at the a-position i.e. RCH_2CO_2H
- Tedious and slow

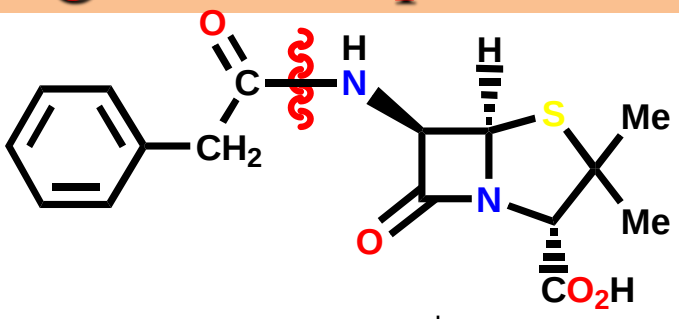
2) By total synthesis

- Only 1% overall yield
- Impractical

3) By semi-synthetic procedures

- Use a naturally occurring structure as the starting material for analogue synthesis

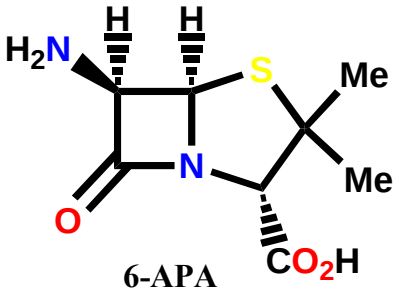
Penicillin Analogues - Preparation



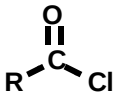
Penicillin G

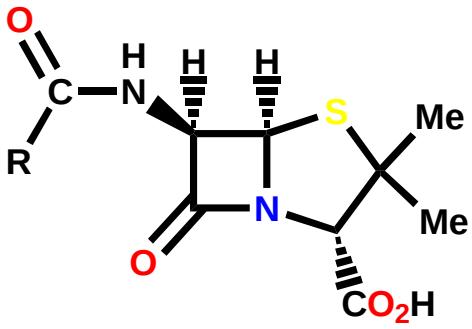
Penicillin acylase
or chemical hydrolysis

Fermentation ⟶



6-APA



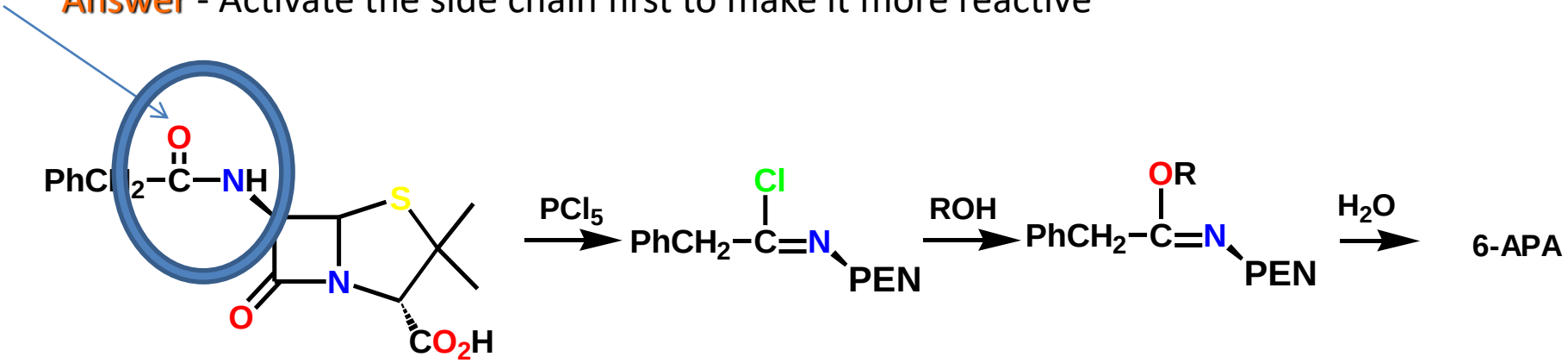


Semi-synthetic penicillins

Penicillin Analogues - Preparation

Problem - How does one hydrolyse the side chain by chemical means in presence of a labile b-lactam ring?

Answer - Activate the side chain first to make it more reactive



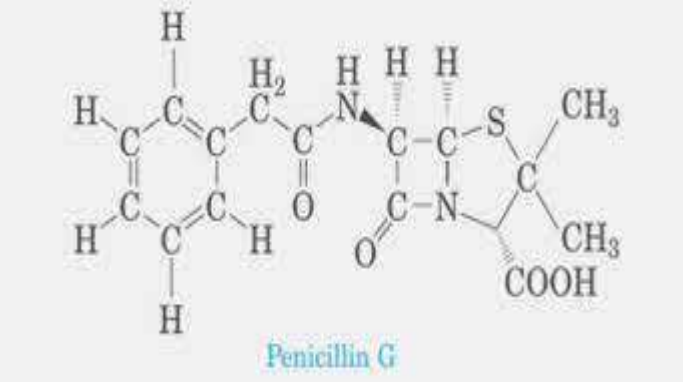
Note - Reaction with PCl_5 requires the involvement of a lone pair of electrons from nitrogen. Not possible for the β-lactam nitrogen.

Problems with Penicillin G

•It is sensitive to stomach acids

•It is sensitive to b-lactamases - enzymes which hydrolyse the b-lactam ring

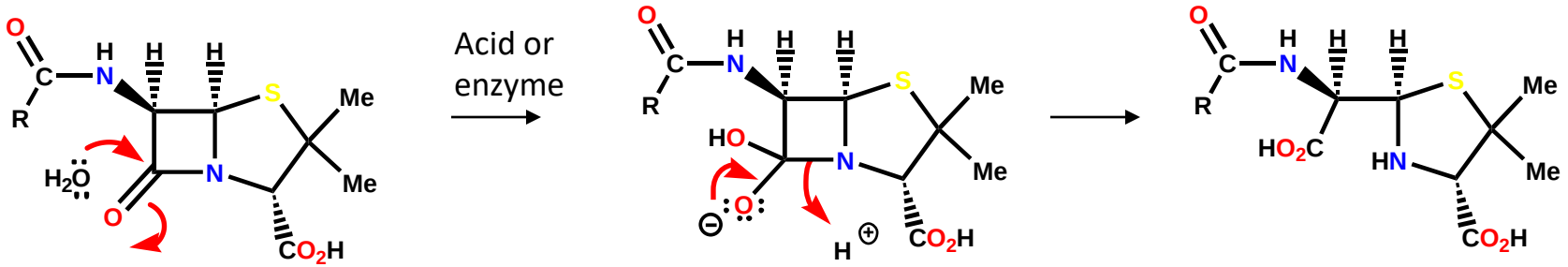
• It has a limited range of activity



Problem 1 - Acid Sensitivity

Reasons for sensitivity

1) Ring strain



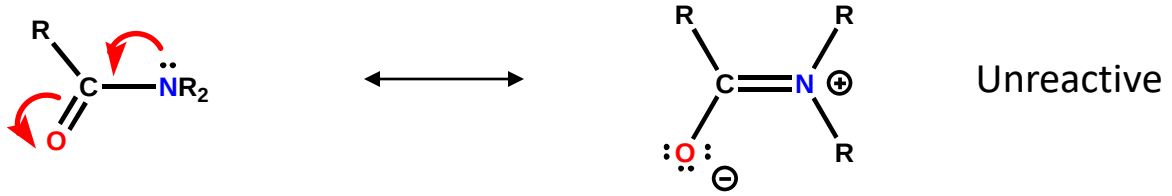
Problem 1 - Sensitivity

Reasons for sensitivity

2) Reactive b-lactam carbonyl group

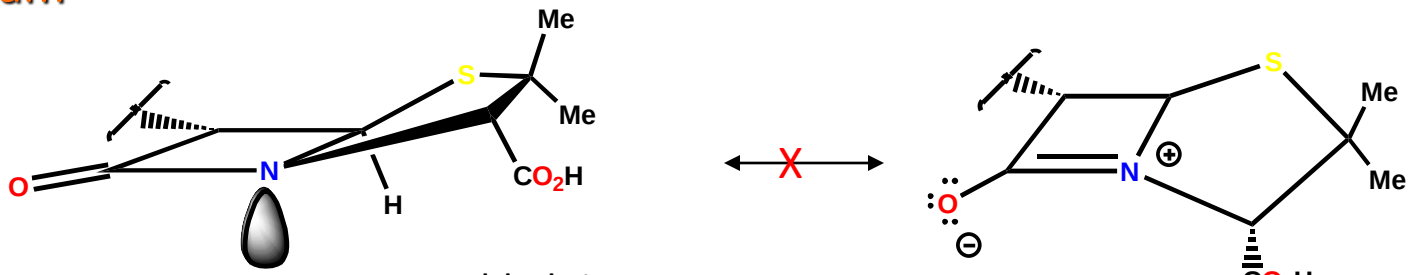
Does not behave like a tertiary amide

Tertiary amide



Unreactive

β-Lactam



Folded ring system

Impossibly strained

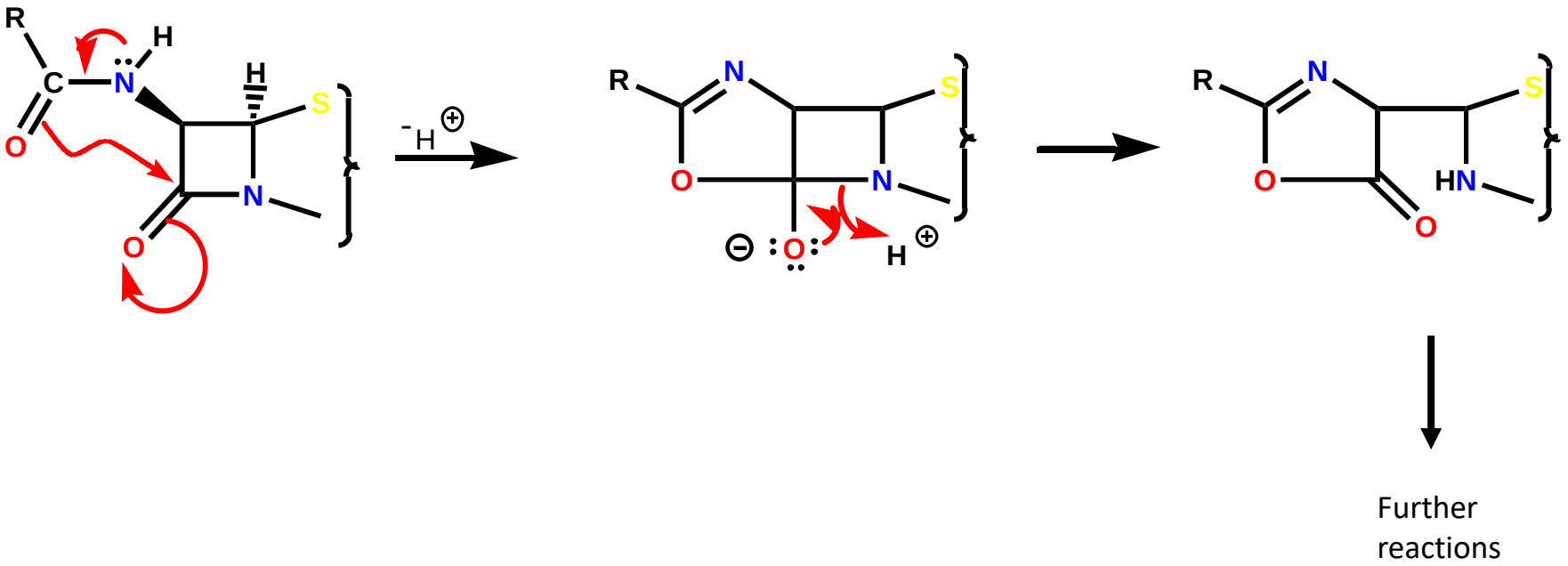
- Interaction of nitrogen's lone pair with the carbonyl group is not possible
- Results in a reactive carbonyl group

Problem 1 - Sensitivity

Reasons for sensitivity

Acyl side chain

Neighboring group participation in the hydrolysis mechanism



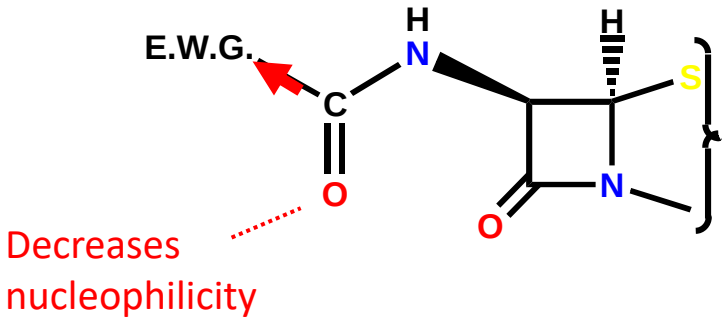
Problem 1 - Sensitivity

Conclusions

- The b-lactam ring is essential for activity and must be retained
- Cannot tackle factors 1 and 2
- Can only tackle factor 3

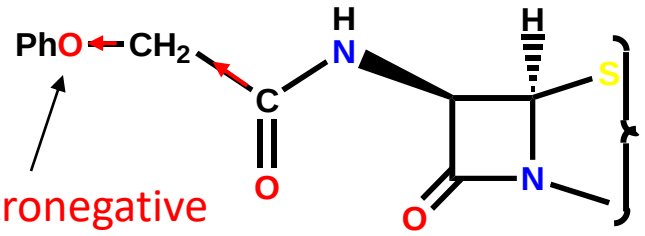
Strategy

Vary the acyl side group (R) to make it electron-withdrawing to decrease the nucleophilicity of the carbonyl oxygen



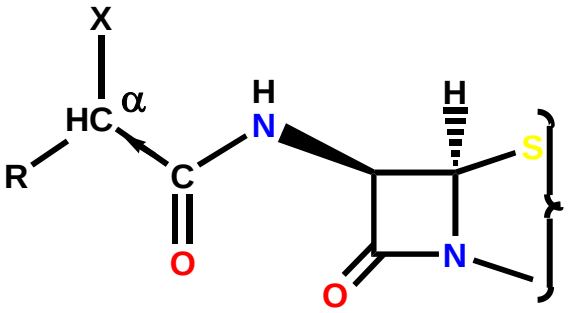
Problem 1 - Sensitivity

Examples



electronegative oxygen

Penicillin V
(orally active)



X = NH_2, Cl, PhOCONH, Heterocycles, CO_2H

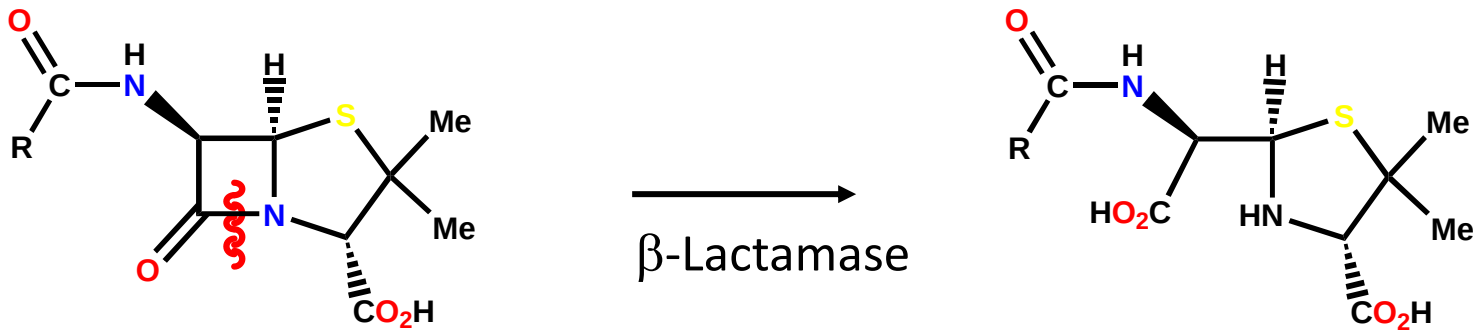
- Better acid stability and orally active
- But sensitive to b-lactamases
- Slightly less active than penicillin G
- Allergy problems with some patients

Very successful semi-synthetic penicillins
e.g. ampicillin, oxacillin

Problem 2 - Sensitivity to β-Lactamases

b-Lactamases

- Enzymes that inactivate penicillins by opening b-lactam rings
- Allow bacteria to be resistant to penicillin
- Transferable between bacterial strains (i.e. bacteria can acquire resistance)
- Important with respect to *Staphylococcus aureus* infections in hospitals
- 80% *Staph*. infections in hospitals were resistant to penicillin and other antibacterial agents by 1960
- Mechanism of action for lactamases is identical to the mechanism of inhibition for the target enzyme
- But product is removed efficiently from the lactamase active site



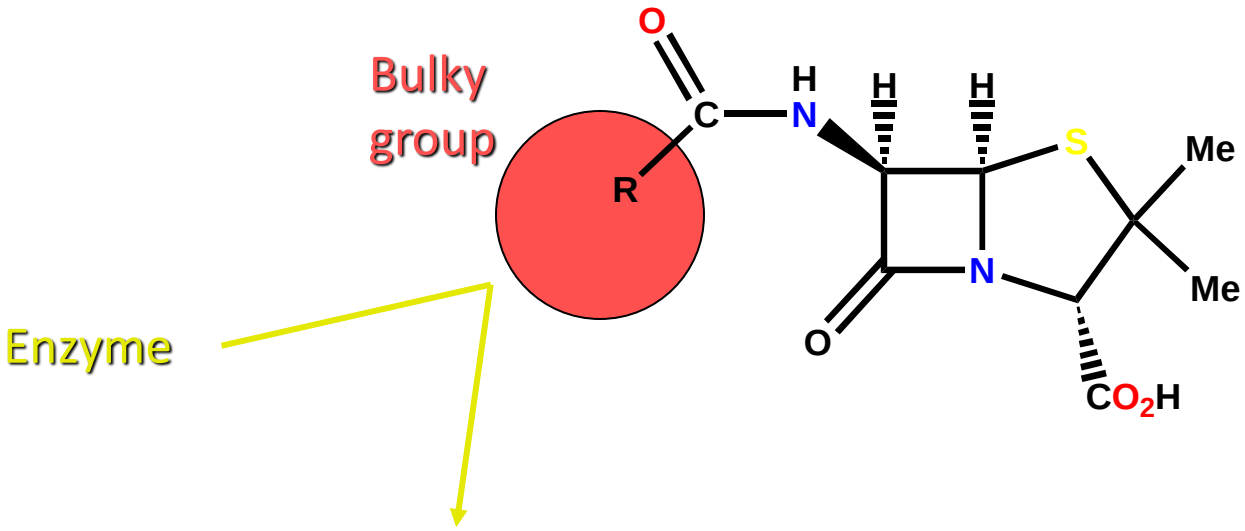
Problem 2 - Sensitivity to β-Lactamases

Strategy

•Use of steric shields

•Block access of penicillin to the active site of the enzyme by introducing bulky groups to the side chain

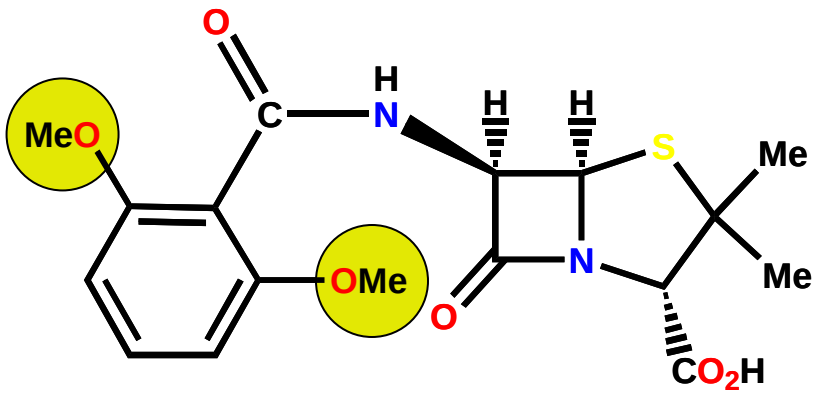
•Size of shield is crucial to inhibit reaction of penicillins with b-lactamases, but not with the target transpeptidase enzyme



Problem 2 - Sensitivity to b-Lactamases

Examples - Methicillin (Beechams - 1960)

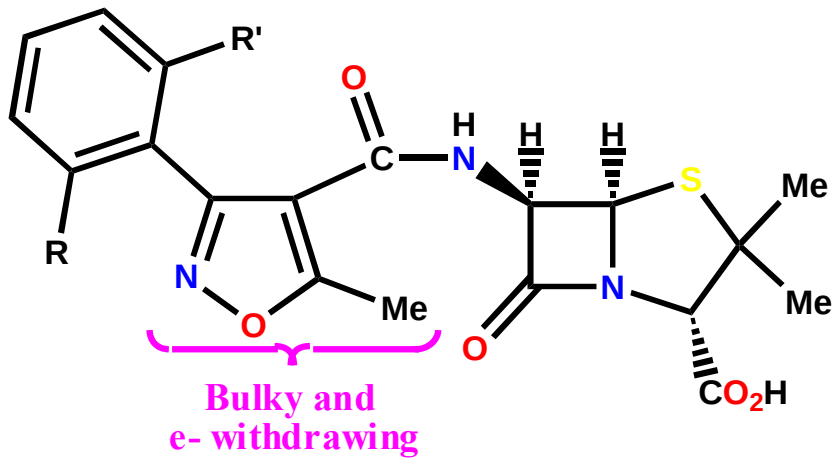
ortho groups important



- Methoxy groups block access to b-lactamases but not to transpeptidases
- Binds less readily to transpeptidases compared to penicillin G
- Lower activity compared to Pen G against Pen G sensitive bacteria
- Poor activity vs. some *streptococci*
- Inactive vs. Gram -ve bacteria
- Poor range of activity
- Active against some penicillin G resistant strains (e.g. *Staphylococcus*)
- Acid sensitive since there is no electron-withdrawing group
- Orally inactive and must be injected

Problem 2 - Sensitivity to β-Lactamases

Examples - Oxacillin



Oxacillin R = R' = H
Cloxacillin R = Cl, R' = H
Flucloxacillin R = Cl, R' = F

- Orally active and acid resistant
- Resistant to b-lactamases
- Active vs. *Staphylococcus aureus*
- Less active than other penicillins
- Inactive vs. Gram -ve bacteria
- Nature of R & R' influences absorption and plasma protein binding
- Cloxacillin better absorbed than oxacillin
- Flucloxacillin less bound to plasma protein, leading to higher levels of free drug

Organic Pharm. Chemistry III

Lecture 2

27th February (12th March)

10:30- 12:30

AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023

Today‘s Inspiring Quote

Problem 3 - Range of Activity

Factors

1) Cell wall may have a coat preventing access to the cell
2) Excess transpeptidase enzyme may be present
3) Resistant transpeptidase enzyme (modified structure)
4) Presence of b-lactamases
5) Transfer of b-lactamases between strains
6) Efflux mechanisms

Strategy

- The number of factors involved make a single strategy impossible
- Use trial and error by varying R groups on the side chain
- Successful in producing broad spectrum antibiotics
- Results demonstrate general rules for broad spectrum activity.

Problem 3 - Range of Activity

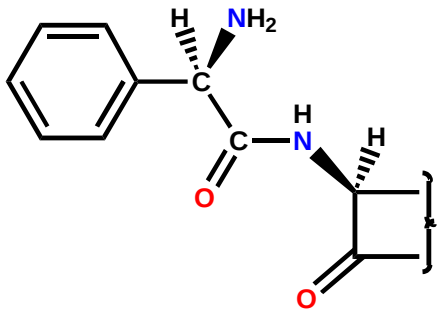
Results of varying R in Pen G

1) Hydrophobic side chains result in high activity vs. Gram +ve bacteria and poor activity vs. Gram -ve bacteria
2) Increasing hydrophobicity has little effect on Gram +ve activity
   but lowers Gram -ve activity
3) Increasing hydrophilic character has little effect on Gram +ve activity
   but increases Gram -ve activity
4) Hydrophilic groups at the a-position (e.g. NH_2, OH, CO_2H) increase activity vs Gram -ve bacteria

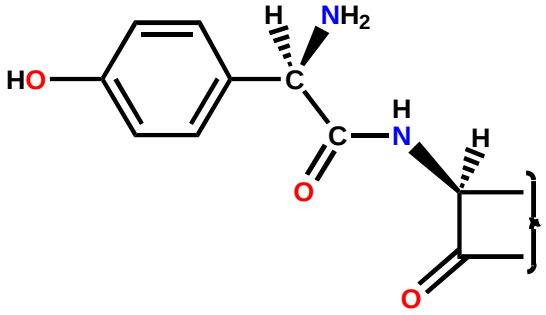
Problem 3 - Range of Activity

Examples of Broad Spectrum Penicillins

Class 1 - NH_2 at the α-position
Ampicillin and amoxicillin (Beechams, 1964)



Ampicillin (Penbritin)
2nd most used penicillin



Amoxicillin (Amoxil)

Problem 3 - Range of Activity

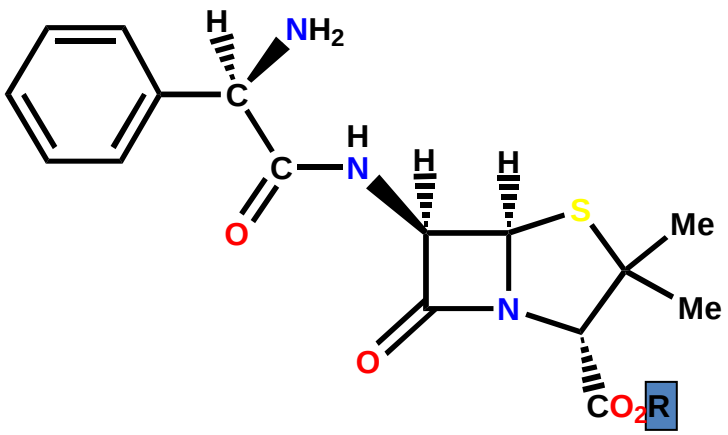
Examples of Broad Spectrum Penicillins

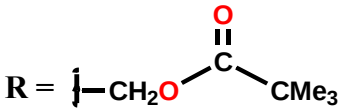
Properties

- Active vs Gram +ve bacteria and Gram -ve bacteria which do not produce b-lactamases
- Acid resistant and orally active
- Non toxic
- Sensitive to b-lactamases
- Increased polarity due to extra amino group
- Poor absorption through the gut wall
- Disruption of gut flora leading to diarrhea
- Inactive vs. *Pseudomonas aeruginosa*

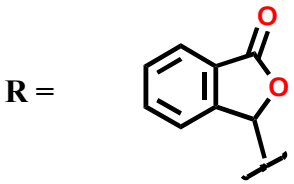
Problem 3 - Range of Activity

Prodrugs of Ampicillin (Leo Pharmaceuticals - 1969)

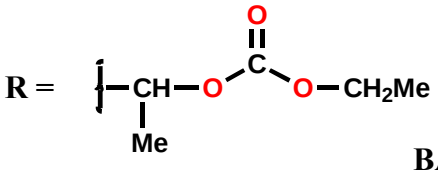




PIVAMPICILLIN



TALAMPICILLIN



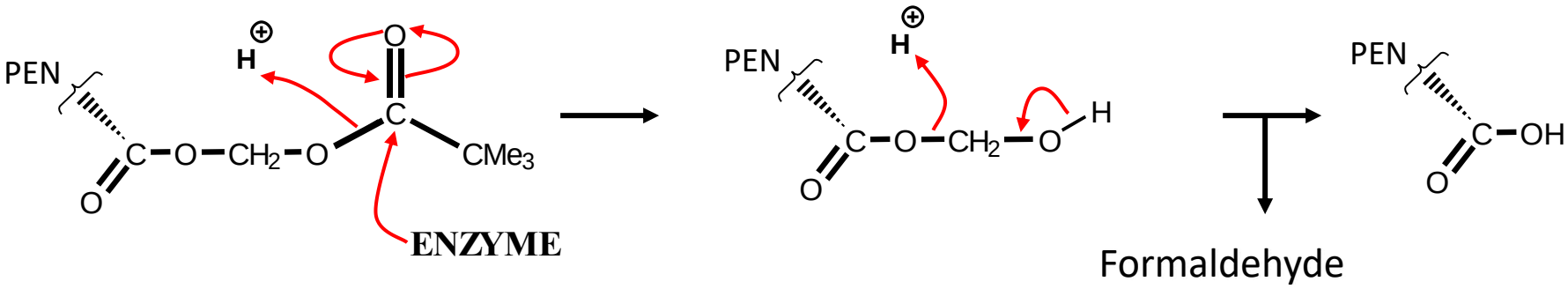
BACAMPICILLIN

Properties

- Increased cell membrane permeability
- Polar carboxylic acid group is masked by the ester
- Ester is metabolised in the body by esterases to give the free drug

Problem 3 - Range of Activity

Mechanism of prodrug activation



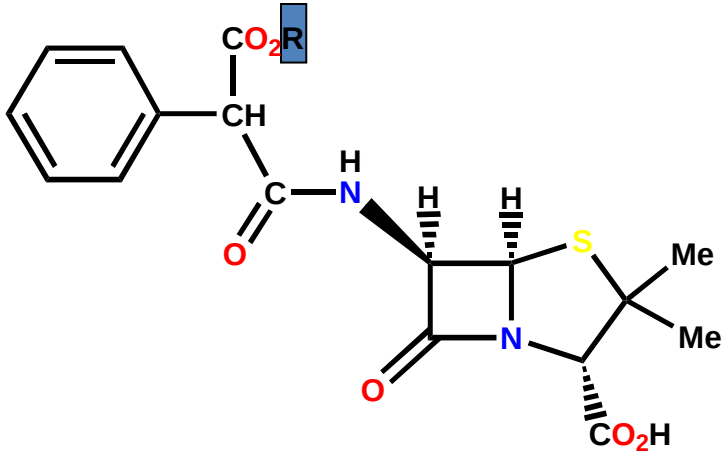
- Extended ester is less shielded by the penicillin nucleus
- Hydrolysed product is chemically unstable and degrades
- Methyl ester of ampicillin is not hydrolysed in the body
- Bulky penicillin nucleus acts as a steric shield for methyl ester

Problem 3 - Range of Activity

Examples of broad spectrum penicillins

Class 2 - CO_2H at the a-position (carboxypenicillins)

Examples



R = H Carbenicillin

R = Ph Carfecillin

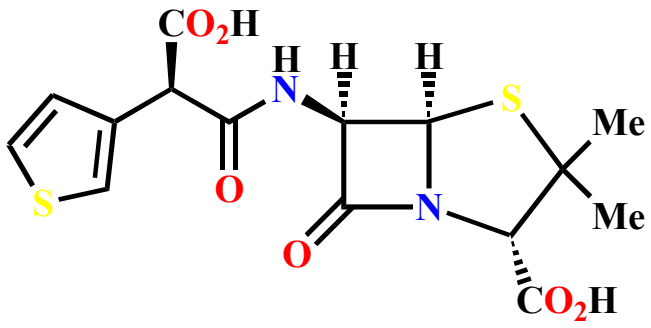
- Carfecillin = prodrug for carbenicillin
- Active over a wider range of Gram -ve bacteria than ampicillin
- Active vs. *Pseudomonas aeruginosa*
- Resistant to most b-lactamases
- Less active vs Gram +ve bacteria (note the hydrophilic group)
- Acid sensitive and must be injected
- Stereochemistry at the a-position is important
- CO_2H at the a-position is ionised at blood pH

Problem 3 - Range of Activity

Examples of broad spectrum penicillins

Class 2 - CO_2H at the a-position (carboxypenicillins)

Examples



TICARCILLIN

- Administered by injection
- Identical antibacterial spectrum to carbenicillin
- Smaller doses required compared to carbenicillin
- More effective against *P. aeruginosa*
- Fewer side effects
- Can be administered with clavulanic acid

Problem 3 - Range of Activity

Examples of broad spectrum penicillins

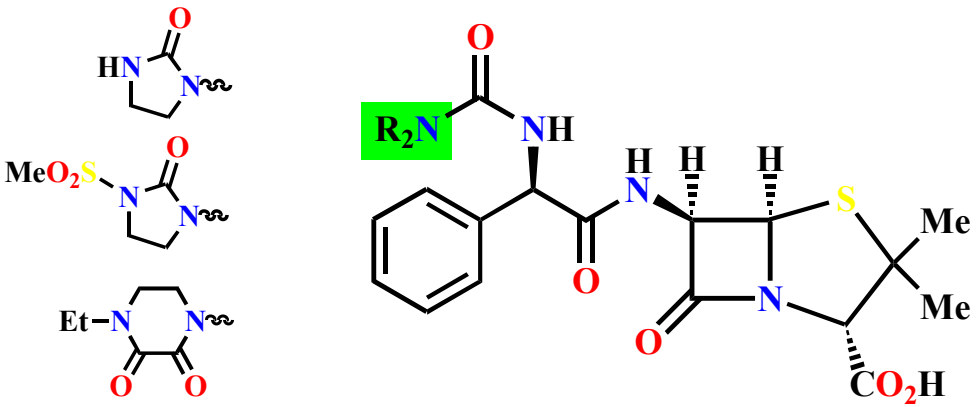
Class 3 - Urea group at the a-position (ureidopenicillins)

Examples

Azlocillin

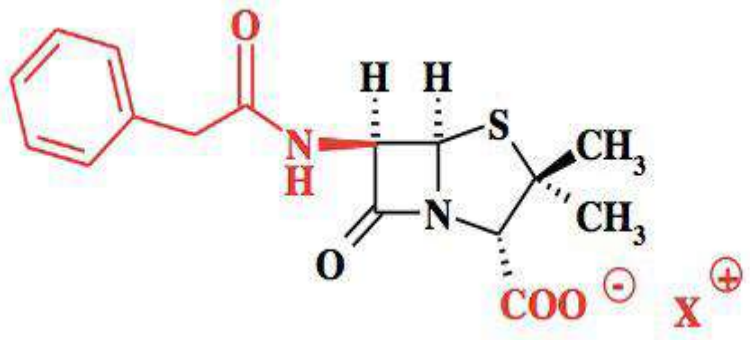
Mezlocillin

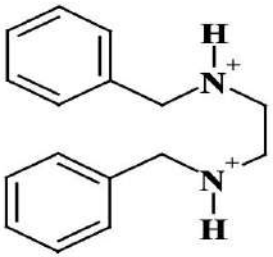
Piperacillin



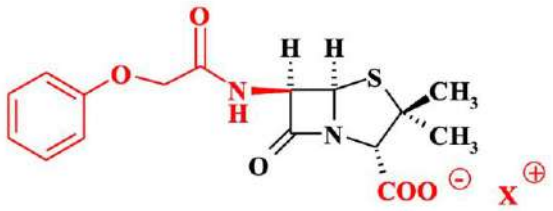
- Administered by injection
- Generally more active than carboxypenicillins vs. streptococci and *Haemophilus* species
- Generally have similar activity vs Gram -ve aerobic rods
- Generally more active vs other Gram -ve bacteria
- Azlocillin is effective vs *P. aeruginosa*
- Piperacillin can be administered alongside tazobactam

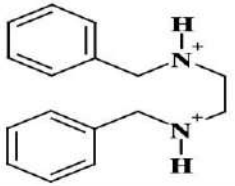
Penicillin G (benzylpeenicillin)



X ⊕	DCI	ND
	benzylpenicillin	Pénicillin G®...
Na^+		
$C_6H_5CH_2$ $C_6H_5CH_2CH_2$ NH_2^+	benethamin - penicillin	Biclinocilin ®
1/2 	benzathin - penicillin	Extencillin ®

Penicillin V (phenoxymethylpenicillin



X ⊕	D.C.I.	ND
K^+	phenoxymethyl penicillin potassi	Ospen®
1/2 	benzathin - penicillin	Oracillin ®

The hydrophobic character of Penicillin V is directly related to the binding to plasma proteins, renal elimination: (competition with probenecid, or diuretic), therefore their half-life increased.
Administrable orally, narrow antibacterial spectrum, limited to Gram + germs.

Indications :

- Penicillin G : infections caused by susceptible germs ENT / pulmonary / soft tissue
- Penicillin V : streptococcal infections

Adverse effects: especially allergic.

B. Semisynthetic penicillins

We want molecules that can be used orally, slowly eliminated, resistant to Beta- lactamases, with a broad spectrum and shifted to Gram-negative bacteria.

Classification of synthetic penicillins

1 Penicillins G (benzylpenicillin) and **V** (phenoxymethylpenicillin): sensitive to penicillinases, penicillin V is resistant to acid hydrolysis → oral route.

2. Group M penicillins: narrow spectrum, resistant to penicillinases (hindered penicillins), isoxazolpenicillins (oxacillin, cloxacillin).

3. Group A penicillins (aminopenicillins): **spectrum broadened** and shifted to **Gram -** bacteria, **orally active**.

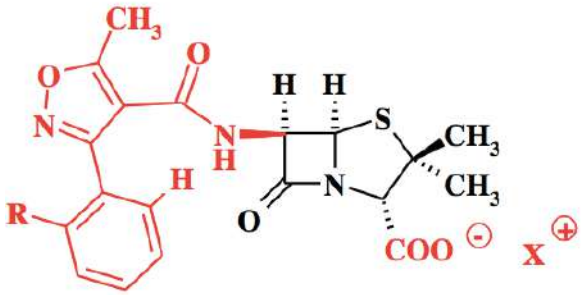
- **Ampicillin** (and combination Sulbactam)
- **Pro--ampicillins** characterized by better pharmacokinetics: pivampicillin, bacampicillin
- **Ampicillin analog**: **amoxicillin** (and a combination of clavulanic acid)

4. Ureidopenicillins: mezlocillin, piperacillin (and combination with tazobactam).

HEMISYNTHETIC BETA LACTAMS

5. Carboxypenicillins: antipyocyanics (anti *Pseudomonas aeruginosa*), Ticarcillin (and Associate with clavulanic acid).

6. Amidinopenicillins: mecillinam and its ester: pivmecillinam.



Access routes to penicillins

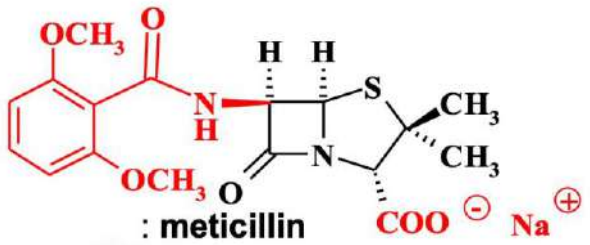
1. Natural penicillins: by fermentation of selected strains of *Penicillium notatum*, *P. chrysogenum* with addition of precursors:

- Phenylacetic acid ⟶ penicillin G
- Phenoxyacetic acid ⟶ penicillin V

2.Hemisynthetic penicillins: acylation of 6-aminopenicillanic acid (6-APA) obtained enzymatically (bacterial or fungal amidases).

Penicillins M

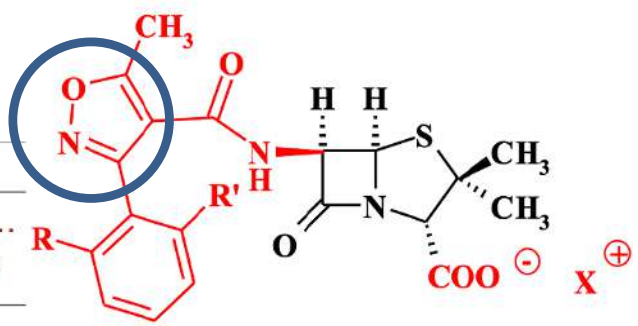
The more crowded the molecule,
the more it is protected from beta lactamases,
this is the steric shield.



Narrow spectrum, sterically hindered, resistant to penicillinases and therefore effective on *Staphylococcus aureus*, which produces penicillinase. Less effective than natural penicillins on strains that do not produce penicillinases. Oral or parenteral route at a dose of 2 to 3 g / d and more.

Isoxazolyl Penicillins

R	R'	D.C.I.	ND
H	H	oxacillin	Bristopen®…
Cl	H	cloxacillin	Orbenin ®



Therapeutic benefit: **infections with Staphylococcus aureus** producing resistant and sensitive penicillinases, methicillin.

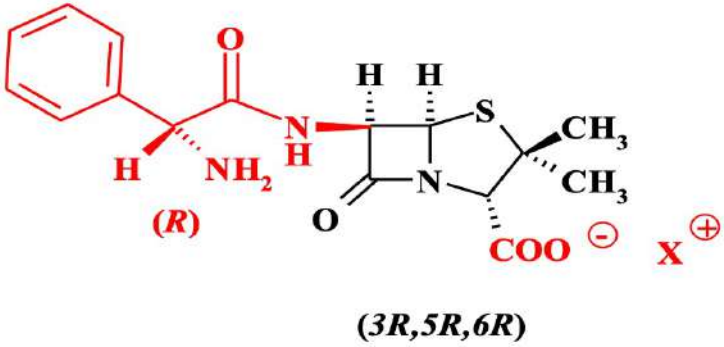
5. Group A penicillins (aminopenicillins)

Hydrophilic molecules capable of crossing porins, broad spectrum, active on Gram - bacteria but inactive on staphylococci producing penicillinases.

Orally active (stable in acidic medium: amine).

5.1 Aminopenicillins: Ampicillin

Used as trihydrate and sodium salt trihydrate.



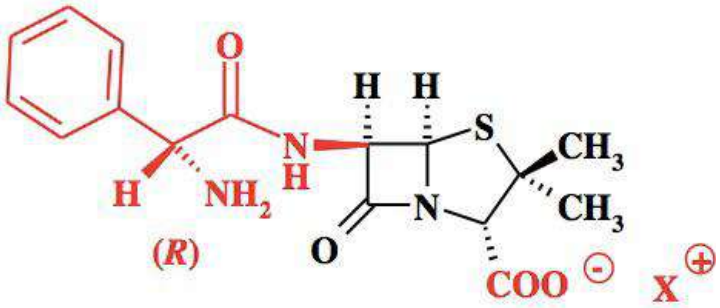
(3R,5R,6R)

Antibacterial spectrum:

active on many Gram germs - , but

frequent resistance (Klebsiella, Enterobacter, Serratia, Pseudomonas aeruginosa, Bacteroides fragilis ...)

Pharmacokinetic data:



• Better tissue diffusion (CSF)

• Urinary and biliary elimination (10 - 20% dose)

• low bioavailability (40%)

• Better duration of action than penicillin G (t1 / 2 = 1h)

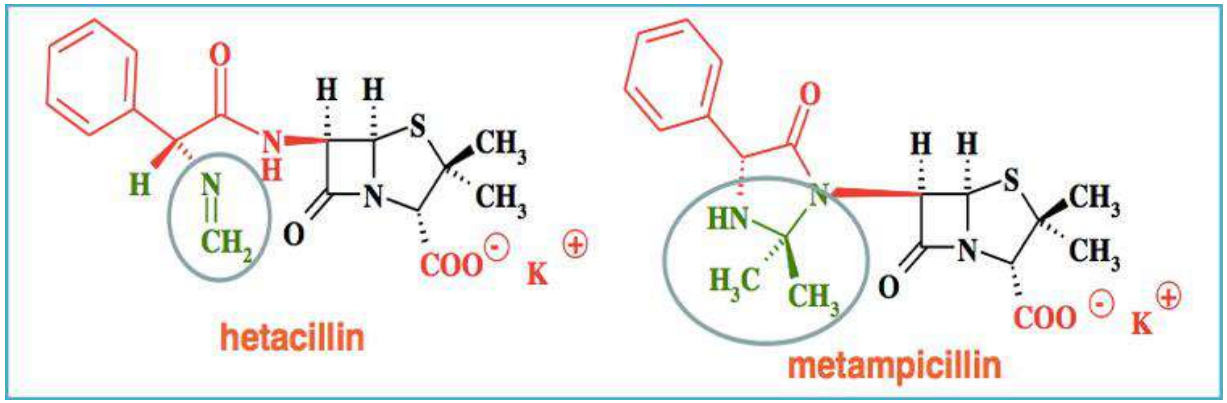
• Binding to plasma proteins (20%)

Therapeutic indication: especially urinary and pulmonary. Dosage: 2 - 4 g / d orally, up to 12 g / d intravenously in severe infections.

5.2. Prodrugs / Pro-ampicillins

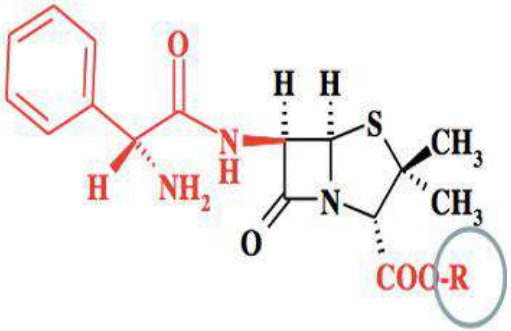
✓ Characterized by better pharmacokinetics.

✓ obtained by protection of the amine functions:



✓ Protection of acid functions (esters):

R	D.C.I.	ND
$-CH_2-O-CO-C(CH_3)_3$	pivampicillin	Pro-ampi®
$-CH(CH_3)-O-CO-OC_2H_5$	bacampicillin	

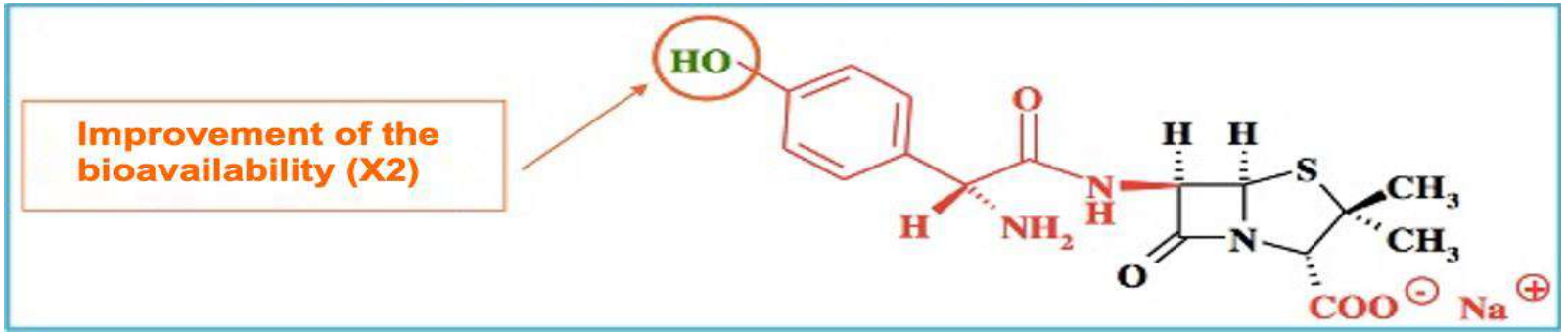


Very good bioavailability (90 - 100%), higher plasma levels.

Dosage: 1 - 3 g / day

Structural analogues of ampicillin:

amoxicillin We added a phenol group which is polar.



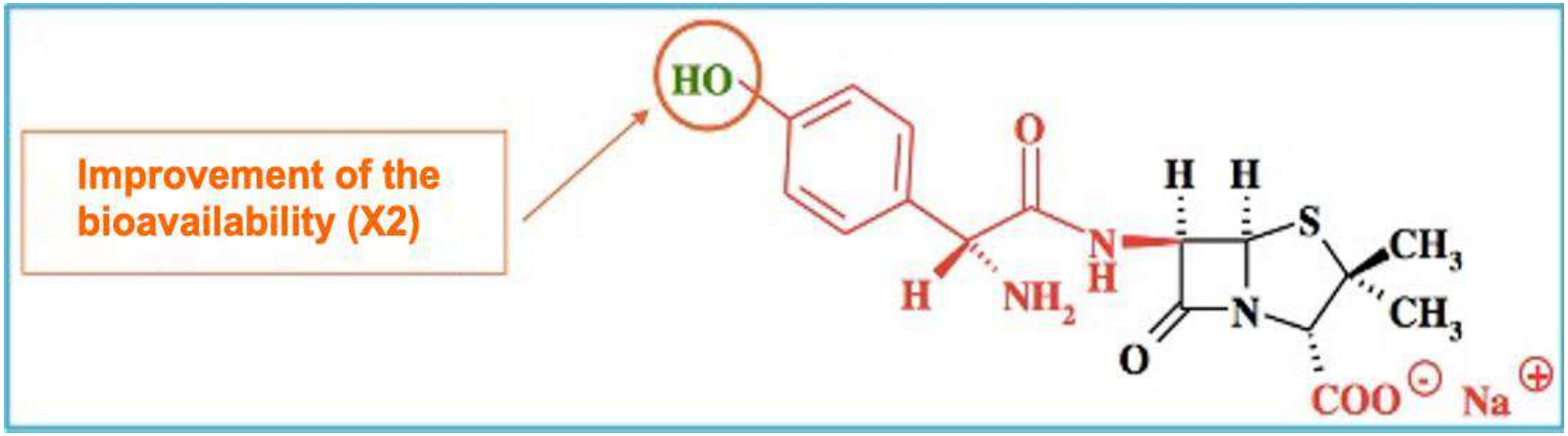
Therapeutic use of group A

• Gram-germ infections: ENT, bronchopulmonary, urogenital, surgical ...

• In association with aminoglycosides and macrolides: septicemia, meningitis, endocarditis.

• Rashes of maculopapular erythema type, quite frequent.

5.3. Structural analogues of ampicillin: amoxicillin We added a phenol group which is polar.

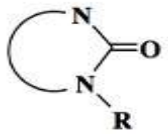
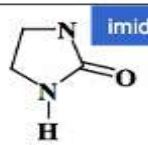
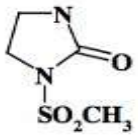
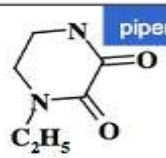


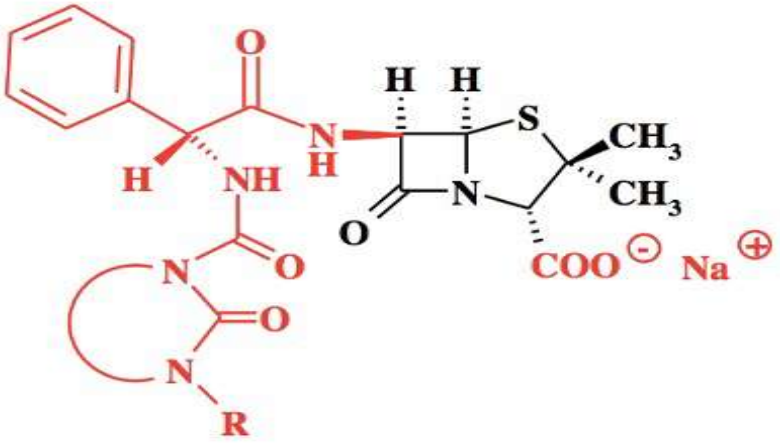
5.4; Therapeutic use of group A

- **Gram-germ infections**: ENT, bronchopulmonary, urogenital, surgical ...
- In association with aminoglycosides and macrolides: septicemia, meningitis, endocarditis.
- Rashes of maculopapular erythema type, quite frequent.

Ureidopenicillins

- Benzyl C carrying a **ureide** group linked to **a heterocycle**.
- **Spectrum extended to Gram germs** - resistant to group A.
- Diffusion in the **cerebro spinal fluid**.
- **Intravenous administration**.

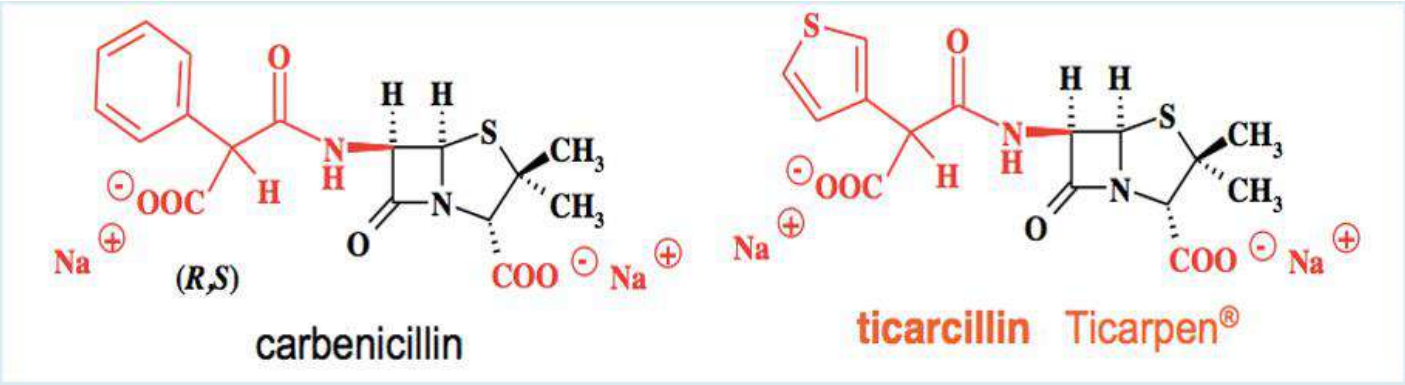
	D.C.I.	ND
 imidazolidinone	Azlocillin	
	Mezlocillin	
 piperazinedione	**Piperacillin**	Piperillin



Useful antibacterial activity: spectrum of ampicillin and ***Serratia***, ***Enterobacter***, ***Citrobacter***, ***Pseudomonas aeruginosa*** but ***inactivated by penicillinases*** (and not by cephalosporinases).

Main indications: **first-line in severe infections outside hospital**, localized or generalized, mono or plurimicrobial, in monotherapy or in combination (septicemia, pneumopathies, meningitis, abdominal, urogenital, pelvic infections, etc.) with sensitive germs (in particular **Gram bacilli - and anaerobes**).
Dosage: 6 to 15 g / day.
In combination with tazobactam Tazocilline®**.**

Carboxypenicillins **P-C incompatible** acid molecules with aminoside solutions. Water soluble, hygroscopic powders, **unstable** solutions.



PHYSICO-CHEMICAL CHARACTERISTICS

✓ powders soluble in water (disodium salt), hygroscopic,

✓ unstable solutions (a few hours in in an acidic environment),

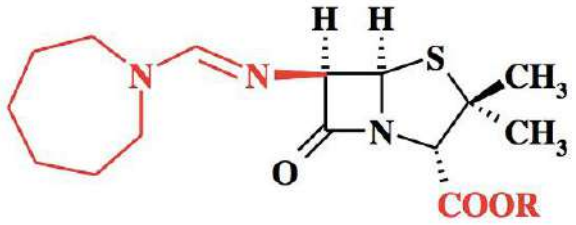
✓ P-C incompatibilities with amino side solutions.

Indications: reserved for serious **infections with Gram-resistant** bacteria including *Pseudomonas aeruginosa* and *Proteus-R* and **anaerobic Gram + bacteria**.

Dosage: 5 - 20 g / d every 8 hours.

Combination with clavulanic acid Claventin® , Augmentin®

Amidinopenicillins Narrow spectrum, limited to certain Enterobacteriaceae, loss of activity against Gram + bacteria, treatment of lower urinary tract infections with sensitive Enterobacteriaceae.



R	D.C.I.	ND
Na^+	mecillinam	
$-CH_2-O-CO-C(CH_3)_3$ (HCl)	pivmecillinam	Selexid®

✓ Narrow spectrum, limited to certain Enterobacteriaceae,

✓ loss of activity on Gram + germs

✓ treatment of lower urinary tract infections caused by sensitive enterobacteriaceae.

Organic Pharm. Chemistry III

Lecture 8

3rd April 2023

10:30- 1:30

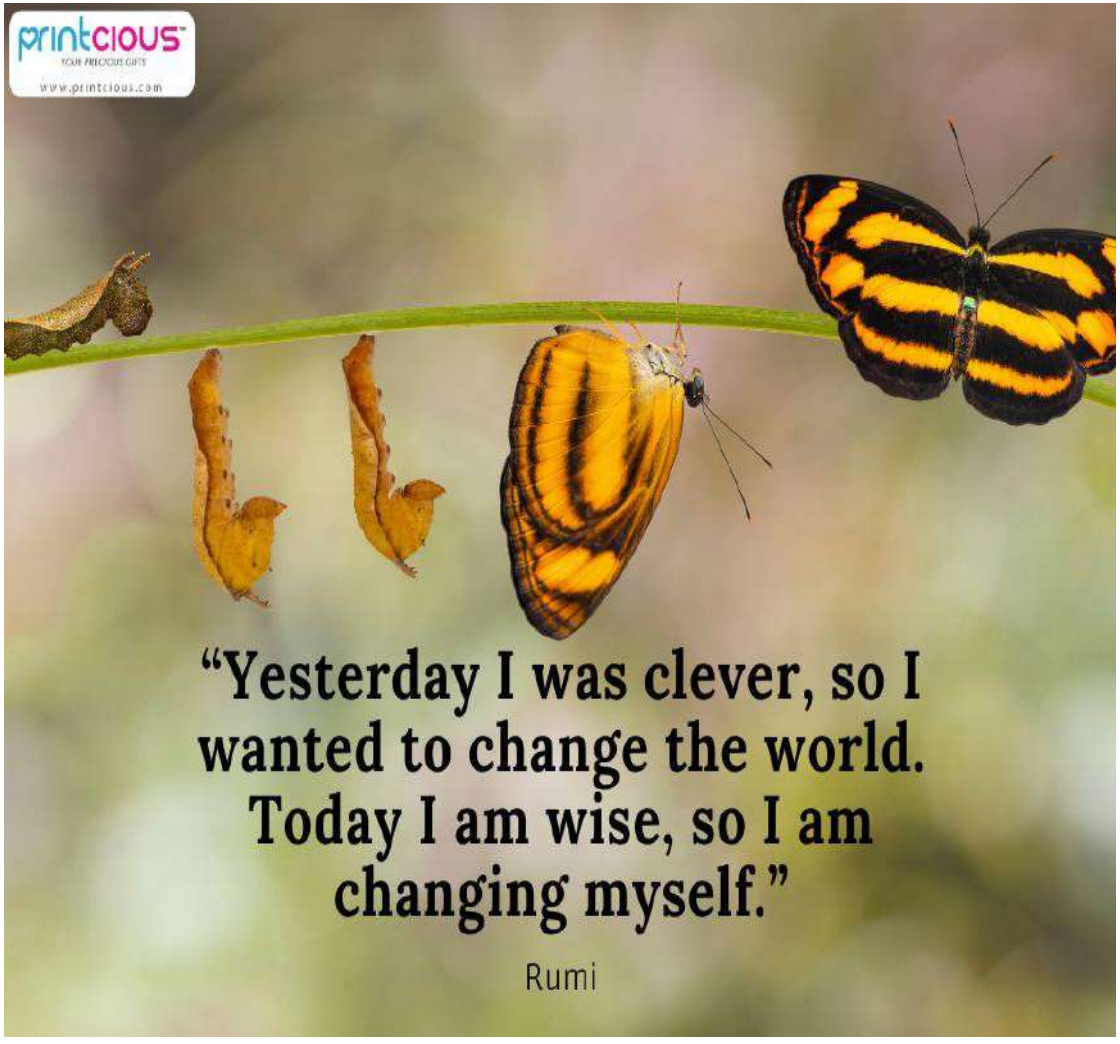
AlNoor University College

Pharmacy Department

2nd Course/ 4th Class

2022/2023

Today’s inspired Quote !

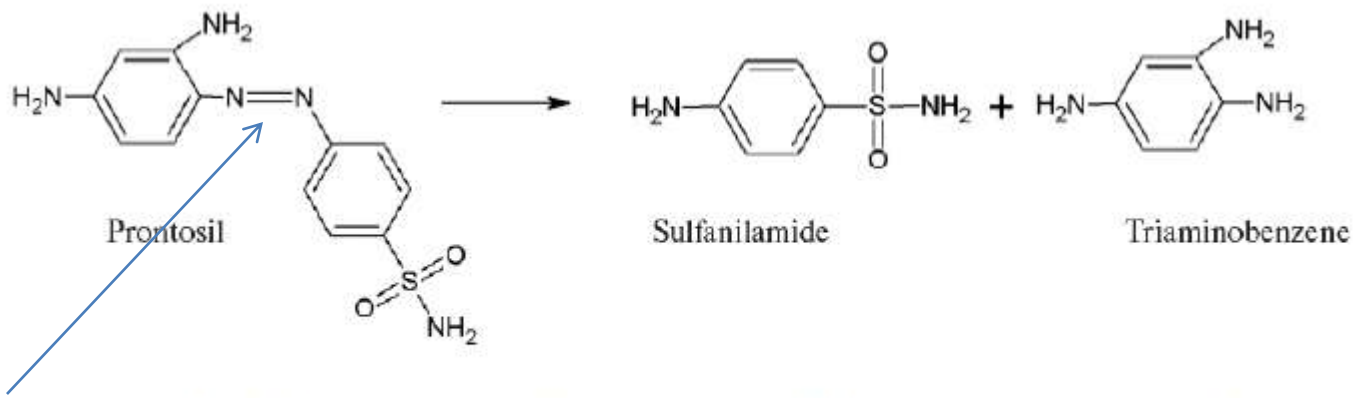


Introduction

Sulfonamides (Sulphonamides) are a group of man-made (synthetic) medicines that contain the **sulfonamide** chemical group. They may also be called sulfa drugs.

- Some sulfonamides are also devoid of antibacterial activity, e.g., the anticonvulsant (Sultiame). The sulfonylureas and thiazide diuretics are newer drug groups based upon the antibacterial sulfonamides.

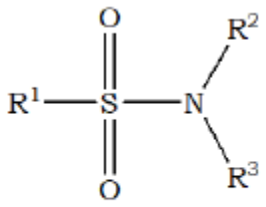
- The first sulfonamide was trade named Prontosil, which is a prodrug Prontosil, the first commercially available antibacterial with a relatively broad effect (against Gram-positive cocci but not against enterobacteria).



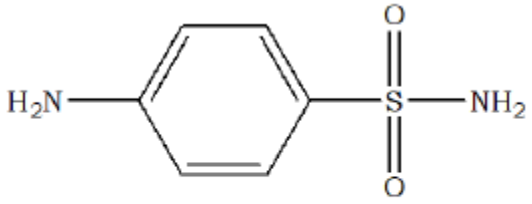
Azo reduction of prontosil is one example, where intestinal bacteria convert inactive therapeutics into their pharmacologically active form. **Bacterial *azoreductases*** present in the distal gut cleave the N-N double bond and produce active metabolite sulfanilamide.

Allergies to sulfonamides are common. The overall incidence of adverse drug reactions to sulfa antibiotics is approximately 3%, close to penicillin.

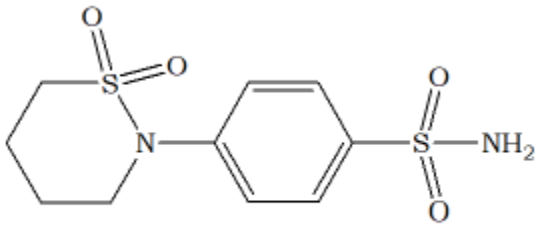
- The general structure of Sulfonamides



Sulfonamide functional group



Sulfanilamide



Sultiame

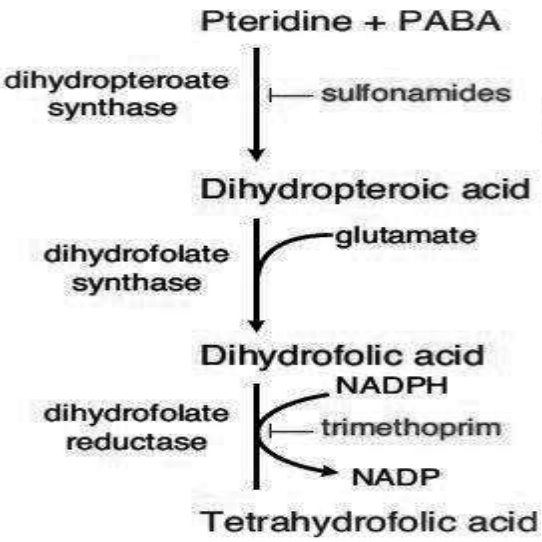
- Sir Gerhard Domagk, (born October 30, 1895, Lagow, Brandenburg, Germany-died April 24, 1964, Burgberg, near Königsfeld, Germany), German bacteriologist and pathologist who was awarded the 1939 Nobel Prize for Physiology or Medicine for his discovery (announced in 1932) of the antibacterial effects of Prontosil, the first of the sulfonamide drugs.

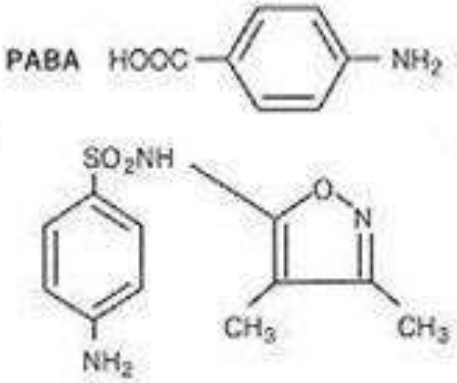
- **Trimethoprim** exerts a **synergistic effect** with **sulfonamides**.
- **- Trimethoprim** is often given in combination with **sulfamethoxazole**, which inhibits the preceding step in bacterial protein synthesis-given together, sulfamethoxazole and trimethoprim inhibit two consecutive steps in the biosynthesis of bacterial nucleic acids and proteins. As a monotherapy **trimethoprim is considered bacteriostatic**, but **in combination with sulfamethoxazole is thought to exert bactericidal activity.**

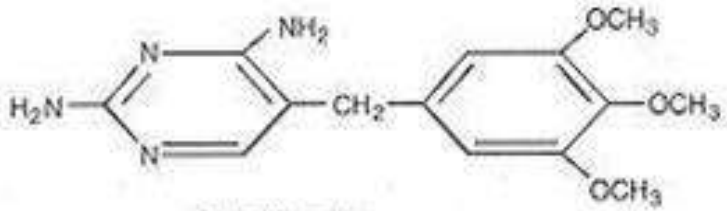
Mechanism of action: Sulfonamides & Trimethoprim

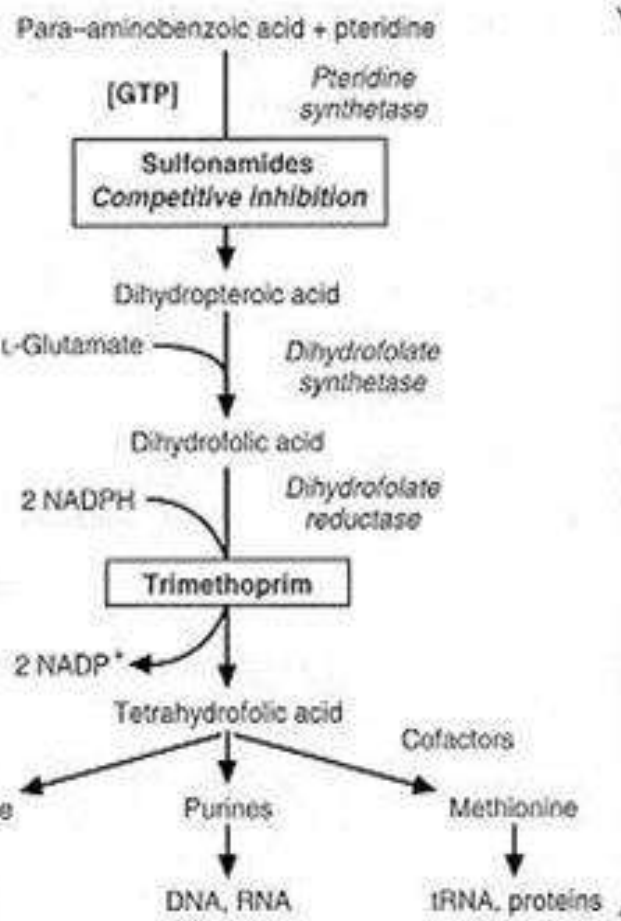
- Sulfanilamide is a competitive inhibitor of bacterial enzyme dihydropteroate synthetase. This enzyme normally uses para-aminobenzoic acid (PABA) for synthesizing the necessary folic acid. The inhibited reaction is normally necessary in these organisms for the synthesis of folic acid. Without it, bacteria cannot replicate.

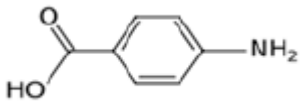
- Trimethoprim is a reversible inhibitor of dihydrofolate reductase, one of the principal enzymes catalyzing the formation of tetrahydrofolic acid (THF) from dihydrofolic acid (DHF).
- Tetrahydrofolic acid is necessary for the biosynthesis of bacterial nucleic acids and proteins and ultimately for continued bacterial survival-inhibiting its synthesis, then, results in **bactericidal activity**. Trimethoprim binds with a much stronger affinity to bacterial dihydrofolate reductase as compared to its mammalian counterpart, **allowing trimethoprim to selectively interfere with bacterial biosynthetic processes**.



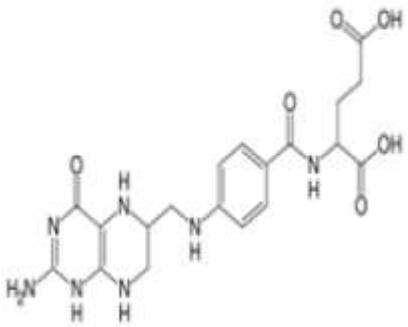




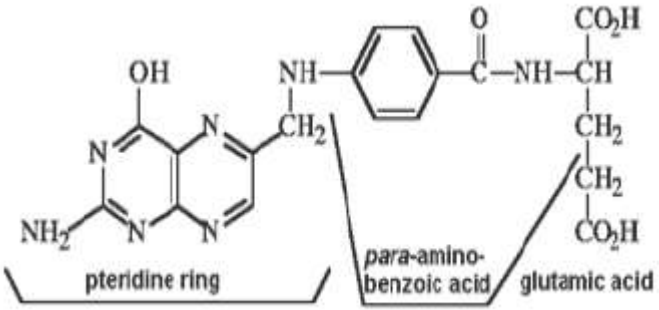




4-Aminobenzoic acid (PABA)



Tetrahydrofolic acid

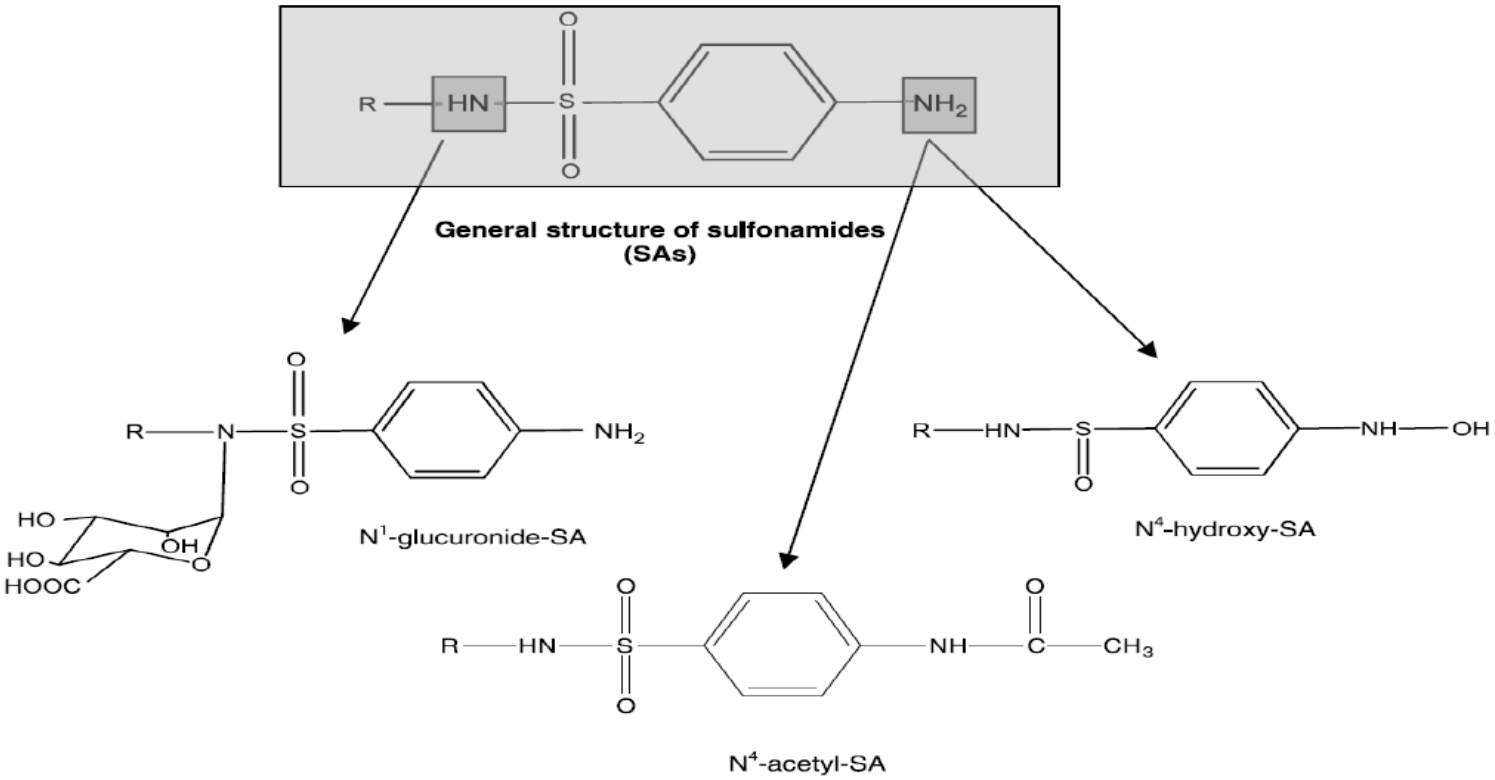


Folic acid

Absorption, Distribution, Metabolism & Excretion

- Sulfonamides class of drugs is absorbed rapidly from the GI tract.
- Approximately 70-100% of an oral dose is absorbed, and sulfonamide can be found in the urine within 30 min of ingestion.
- Peak plasma levels are achieved in 2-6 h, depending on the drug.
- The small intestine is the major site of absorption, but some of the drug is absorbed from the stomach. Absorption from other sites, such as the vagina, respiratory tract, or abraded skin, is variable and unreliable.
- All sulfonamides are bound in varying degree to plasma proteins, particularly to albumin.
- Sulfonamides are metabolized in the liver. The major metabolite is the **N_4-acetylated sulfonamide**. Acetylation results in products that have no antibacterial activity but retain the toxic potential of the parent substance.

- Sulfonamides are eliminated from the body partly as the unchanged drug and partly as metabolic products. The largest fraction is excreted in the urine, and the $t_{1/2}$ depends on renal function.
- - In acid urine, the older sulfonamides are insoluble and crystalline deposits may form. Small amounts are eliminated in the feces, bile, milk, and other secretions.

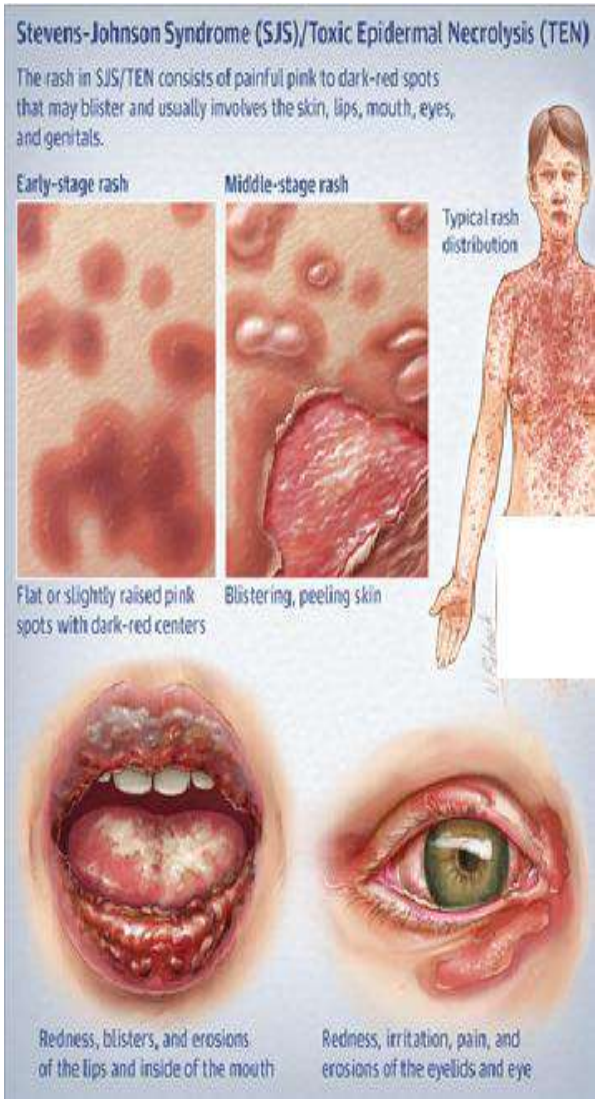


Side effects

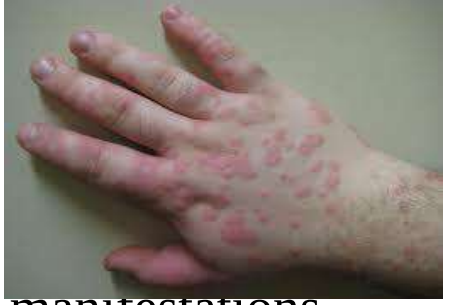
- - Sulfonamides have the potential to cause a variety of untoward reactions, including urinary tract disorders, haemopoietic disorders, and hypersensitivity reactions.

- When used in large dose, it may develop a strong allergic reaction. One of the most serious is *Stevens Johnson syndrome* (or toxic epidermal necrolysis).
- - Some of the original sulfonamide drugs were derived from azo dyes and had the interesting effect of temporarily turning the patient red.

- **N.B- Stevens-Johnson syndrome** (SJS) is a life-threatening condition affecting the skin, in which due to cell death the epidermis separates from the dermis. The syndrome is thought to be a hypersensitivity complex affecting the skin and the mucous membranes.



Adverse reactions



i) The most common manifestation

of a hypersensitivity reaction to sulfa drugs

are rash and hives. However, there are several life-threatening manifestations of hypersensitivity to sulfa drugs, including Stevens-Johnson syndrome, toxic epidermal necrolysis, agranulocytosis, hemolytic anemia, thrombocytopenia, and fulminant hepatic necrosis, among others

ii) The sulfonamide antibiotic chemical structures are implicated in the hypersensitivity reactions associated with the class.

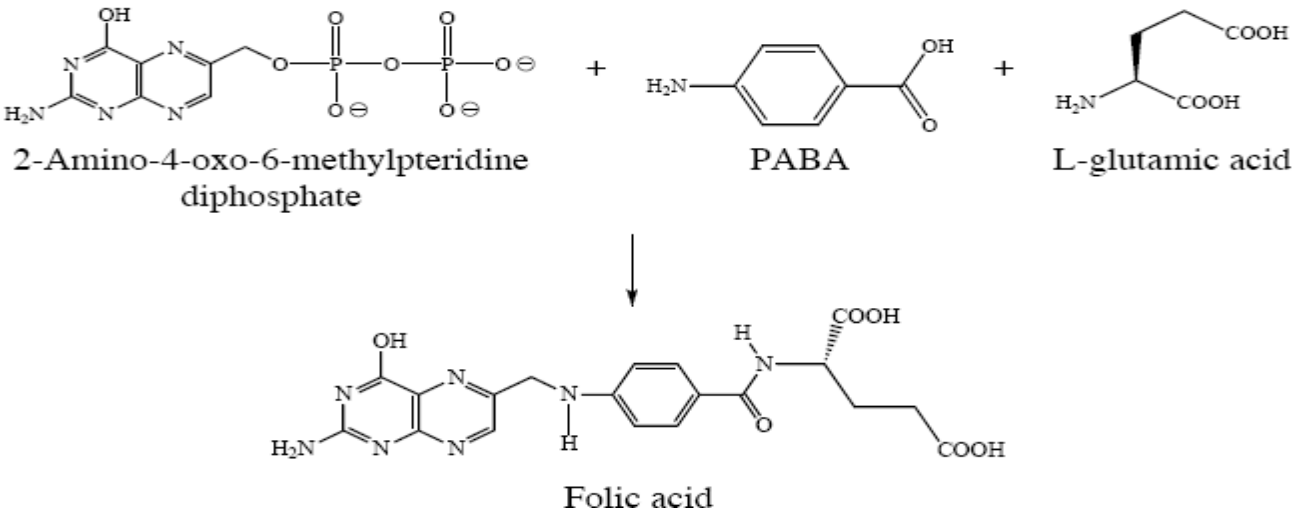
- The first is the N_1 **heterocyclic ring**, which causes a type I hypersensitivity reaction.
- The second is the N_4 **amino nitrogen** that, in a stereospecific process, forms reactive metabolites that cause either direct cytotoxicity or immunologic response .

➢ **Note By:**

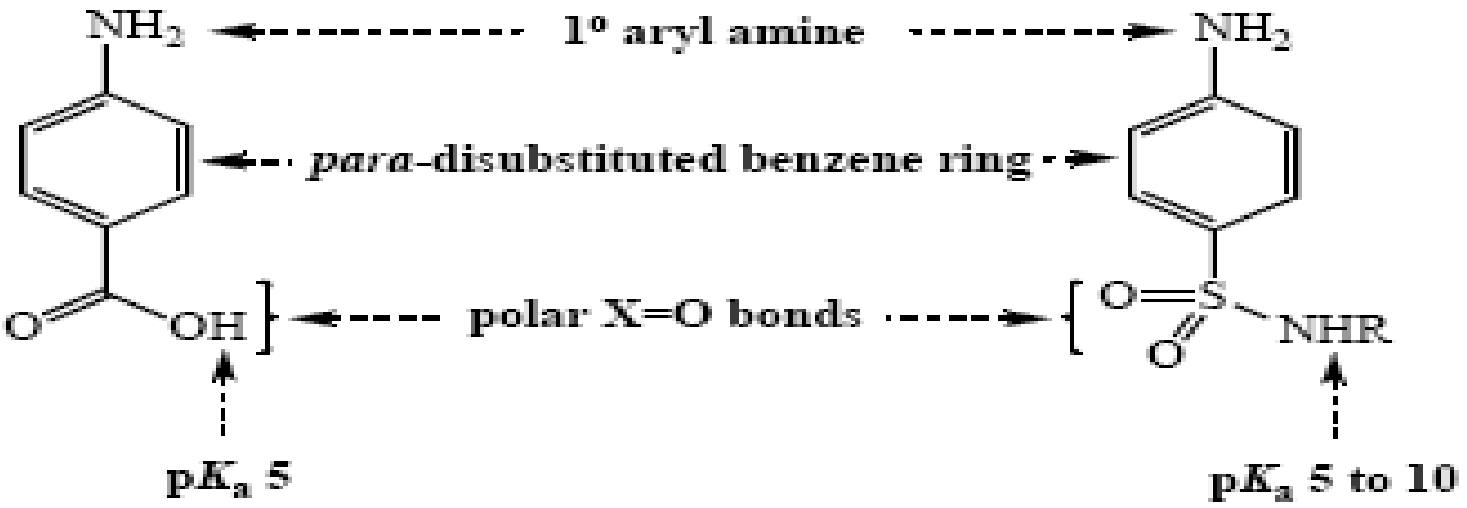
- **Folic acid:** Folic acid and folate (the anion form) are forms of the water-soluble Vitamin B9. These occur naturally in food and can also be taken as supplements. Folate gets its name from the Latin word folium.

- # Biological roles of folate
- i) Folate is necessary for the production and maintenance of new cells. This is especially important during periods of rapid cell division and growth such as infancy and pregnancy. Folate is needed to synthesize DNA bases (most notably thymine, but also purine bases) needed for DNA replication. Thus folate deficiency hinders DNA synthesis and cell division, affecting most notably bone marrow and cancer, both of which participate in rapid cell division.
- ii) In the form of a series of **tetrahydrofolate** (THF) compounds, folate derivatives are substrates in a number of single- carbon-transfer reactions, and also are involved in the synthesis of **dTMP** (**2′-deoxythymidine-5′-phosphate**) from **dUMP** (**2′-deoxyuridine-5′-phosphate**). It is a substrate for an important reaction that involves **vitamin B_{12}** and it is necessary for the synthesis of **DNA**, required for all dividing cells.

- ***Why sulfonamide or sulfonamide inhibit folic acid synthesis mechanism?***
- - Cells use folic acid as a single-carbon atom building block for the construction of nucleic acids and other biological molecules. Inhibition of this process **prevents growth and reproduction** but does not directly lead to cell death. Bacteria synthesize folic acid from 2-amino-4-oxo-6-methylpteridine diphosphate, *p*-aminobenzoic acid (PABA), and L-glutamic acid. Because sulfa drugs are structural mimics of PABA they may bind to dihydropteroate synthetase, one of the enzymes necessary for folic acid synthesis (reversible and competitive inhibition). With this enzyme inhibited, folic acid synthesis is prevented, and cell growth and reproduction are halted.



- In addition, the two molecules are of nearly identical length (6.7 Å for PABA versus 6.9 Å for sulfanilide), both are roughly flat, and both have an equal distribution of charge (δ_+ on the NH_2 group and δ_- on the COOH or SO_2NHR groups). This effect can be seen more clearly by examining the electrostatic potential surfaces of these molecules.

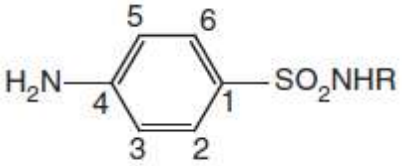


Why bacterial dihydrofolate reductase is many times more sensitive to Trimethoprim than is equivalent enzyme in humans?

- - In microorganism human one of the key enzymes *dihydrofolate reductase* which reduces dihydrofolate to tetrahydrofolate is many more sensitive to folate antagonist trimethoprim in bacteria than in human because of **IC_{50}** values (the concentration causing 50% inhibition).

Inhibitor	**IC_{50} (micro mol/lit) for *dihydrofolate reductase* enzyme**		
	Human	***Protozoal***	***Bacteria***
Trimethoprim	260	0.07	0.005
Pyrimethamine	0.7	0.0005	2.5
Methotrexate	0.001	0.1	Inactive

SAR of Sulphonamides

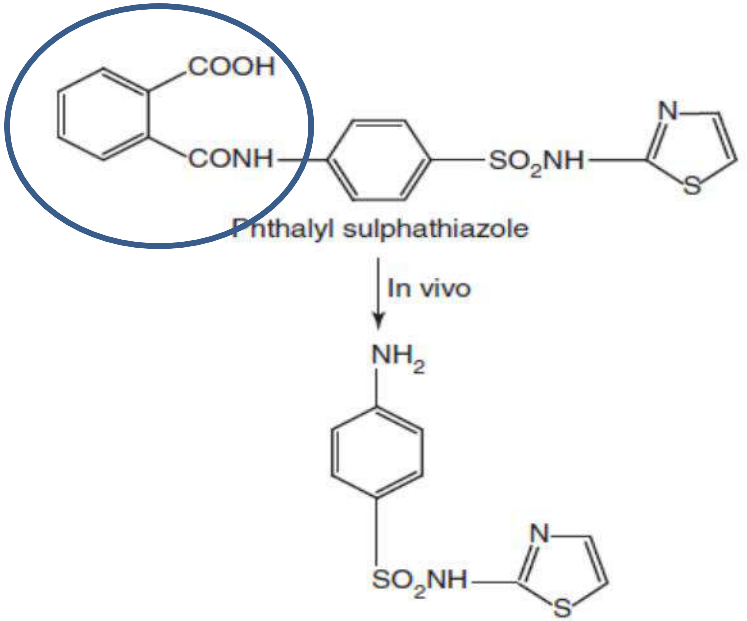


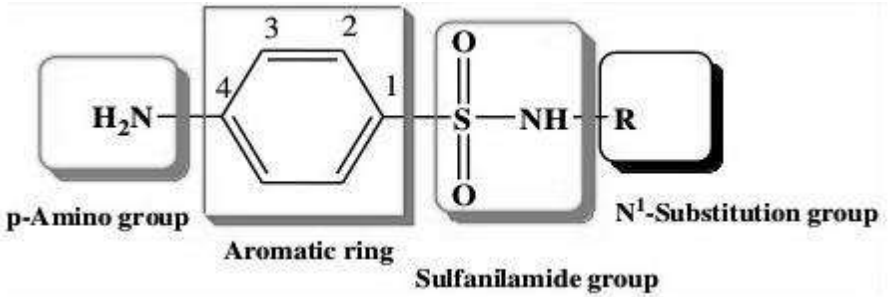
✦ The major features of SAR of sulphonamides include the following:

- Sulphanilamide skeleton is the minimum structural requirement for antibacterial activity.

- The amino- and sulphonyl-groups on the benzene ring are essential and should be in **1** and **4 position**.

The **N-4** amino group could be modified to be **prodrugs,** which are converted to free amino function *in-vivo*.





Sulphur atom should be directly linked to the benzene ring.

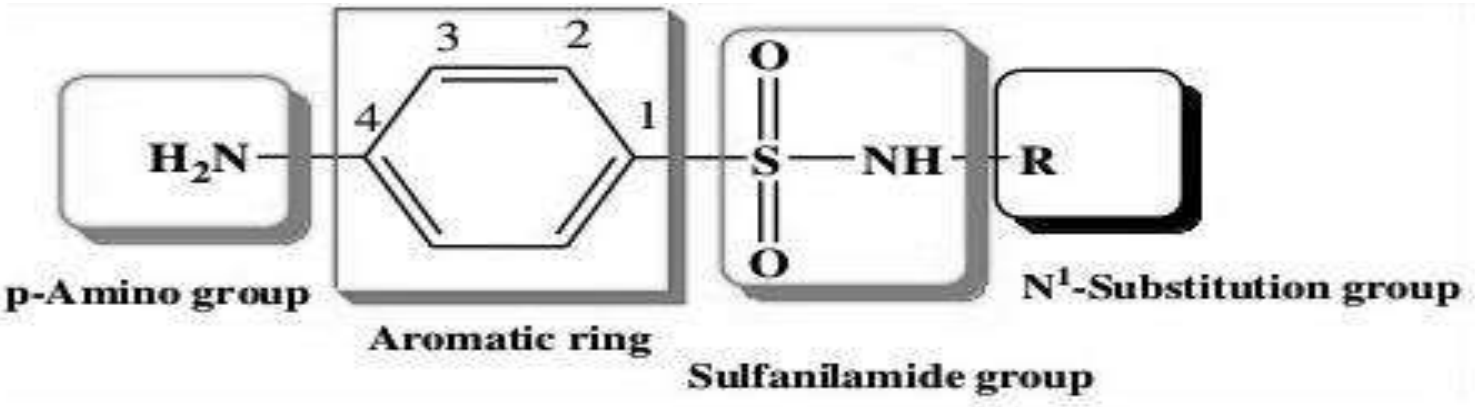
- Replacement of benzene ring by other ring systems or the introduction of additional substituents on it **decreases or abolishes its activity.**
- Exchange of the **–SO_2NH** group by **–CONH** reduces the activity.
- On **N-1**-substituted sulphonamides, activity varies with the nature of the substituent at the amino group. With substituents imparting **electron-rich characters to SO_2 group, bacteriostatic activity increases.**

Heterocyclic substituents lead to highly potent derivatives, while sulphonamides, which contain a single benzene ring at **N-1 position**, are considerably more toxic than heterocyclic ring analogues.

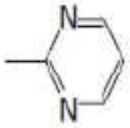
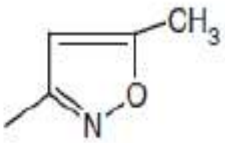
- ✓ - The free aromatic amino groups should reside para to the sulphonamide group. Its replacement at ortho or meta position results in compounds devoid of antibacterial activity.
- ✓ - The active form of sulphonamide is the ionized, maximum activity that is observed between the pKa values **6.6–7.4.**
- ✓ - Substitutions in the benzene ring of sulphonamides produced inactive compounds.
- ✓ - Substitution of free sulphonic acid **(–SO_3H)** group for sulphonamido function destroys the activity, but replacement by a sulphinic acid group **(–SO_2H)** and acetylation of **N-4** position retains back the activity.
- ✓ - *Meta-Sulphonamides* bind to the basic centres of arginine, histidine, and lysine sites of proteins. The binding groups are alkyl, alkoxy, and halides. The binding affects the activity of sulphonamides; protein binding appears to modulate the availability of the drug and its half-life.
- ✓ - The lipid solubility influences the pharmacokinetic and antibacterial activity, and so increases the half-life and antibacterial activity in vitro.

Sulphonamides can be classified on the basis of the chemical structure

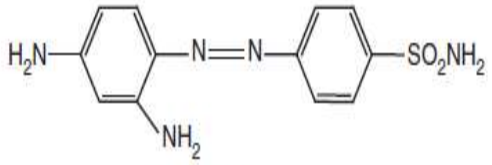
- (i) ***N-1 substituted sulphonamide:*** Sulphadiazine, Sulphacetamide, Sulphadimidine.
- (ii) ***N-4 substituted sulphonamides (prodrugs):*** Prontosil.
- (iii) ***Both N-1 and N-4 substituted sulphonamides***: Succinyl sulphathiazole, Phthalylsulphathiazole.
- (iv) ***Miscellaneous:*** Mefenide sodium.



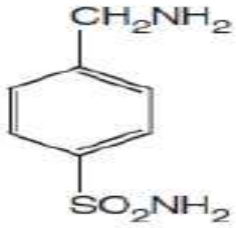
N-1 Substituted sulphonamides

Name	R	R^1
Sulphanilamide	–H	–H
Sulphacetamide	–H	$-COCH_3$
Sulphadiazine	–H	
Sulphamethoxazole	–H	

N-4 substituted suphonamides

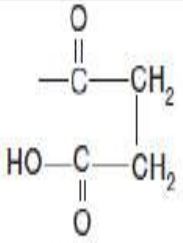
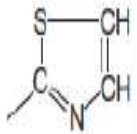
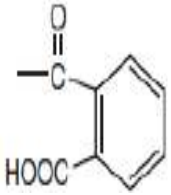
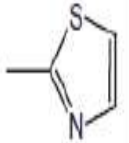


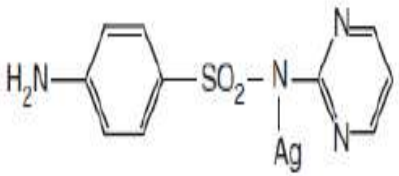
Prontosil



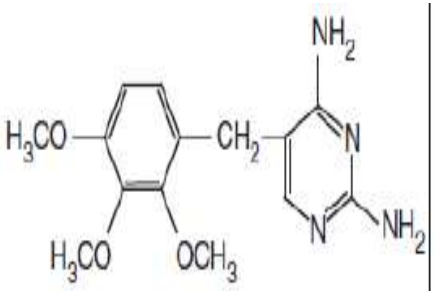
Mafenide

N-1 and N-4 substituted suphonamides

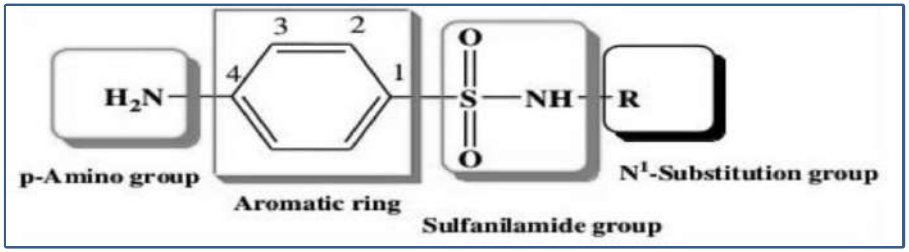
Name	R	R_1
Succinyl sulphathiazole		
Phthalylsulphathiazole		



Silver sulphadiazine



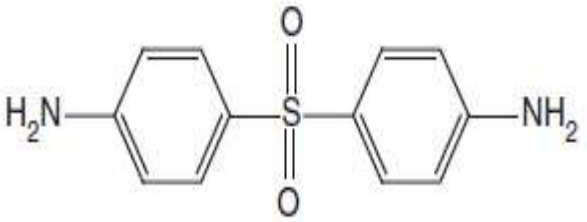
Trimethoprim



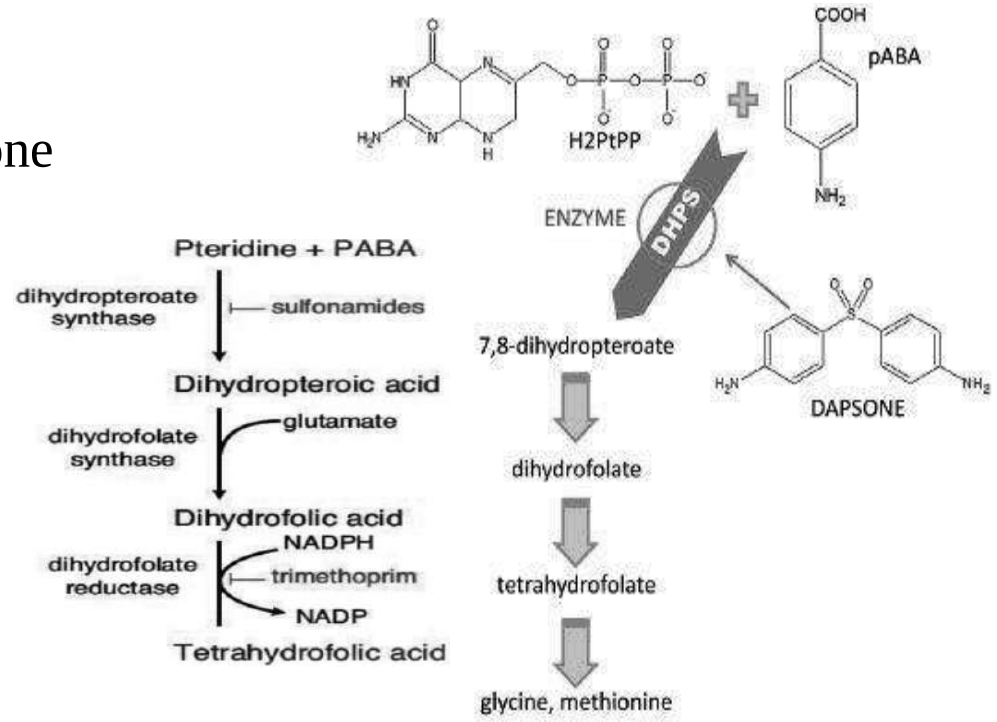
- **Dapsone (DDS, Diaminodiphenyl sulphone)**

Mechanism of action

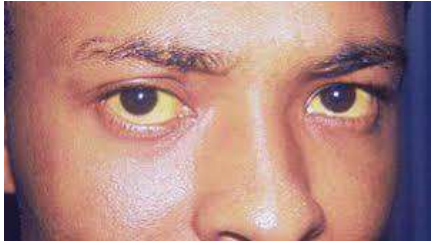
- As an antibacterial, Dapsone inhibits bacterial synthesis of **dihydrofolic acid**, via competition with **para-aminobenzoate** for the active site of ***dihydropteroate synthase***.
- As an anti-inflammatory, Dapsone inhibits the enzyme ***myeloperoxidase***. As part of the respiratory burst that neutrophils use to kill bacteria



4-(4-Aminophenylsulfonyl)benzenamine



Adverse effects



The most prominent side-effects of this drug are dose-related **hemolysis** (which may lead to hemolytic anemia) and **methemoglobinemia**.

- Toxic hepatitis and cholestatic jaundice.
- Other adverse effects include nausea, headache, and rash (which are common), and insomnia, psychosis, and peripheral neuropathy

• Dosage

- The dose as tablets is 25 or 100 mg. For adults the dose consumed is 50 mg per day orally. For lepromatous leprosy, 100 mg Dapsone + 600 mg Rifampin and/or clofazimine 100 mg daily for at least 2 years followed by Dapsone monotherapy.

For borderline tuberculoid disease,

Dapsone 100 mg daily + Rifampin 600 mg

once monthly for 6 months.

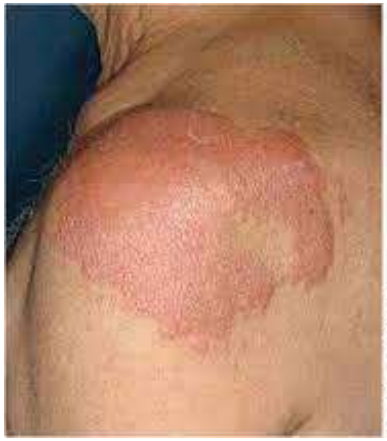
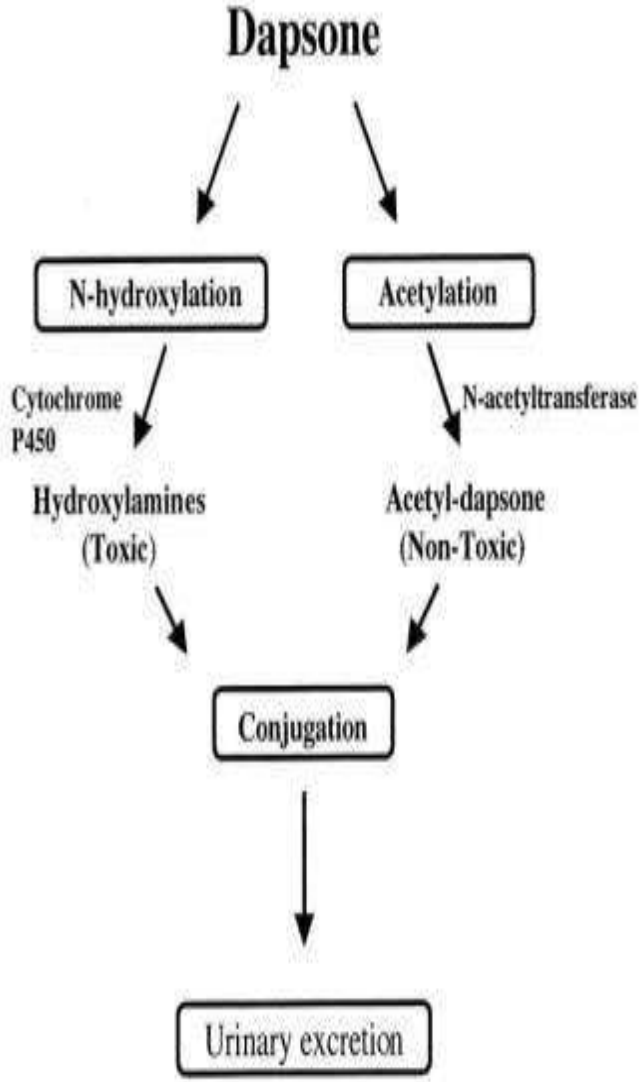
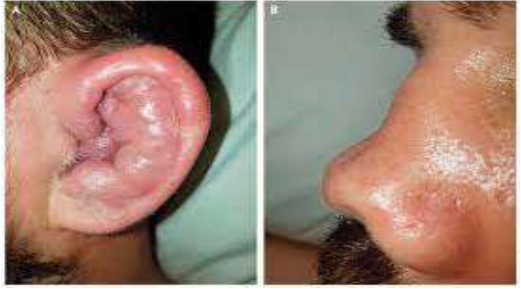


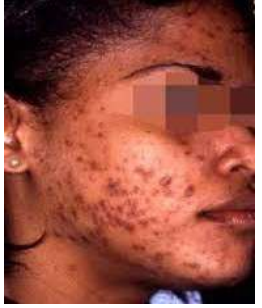
FIGURE 2: Le
shoulder les
Erythematou
plaques with
sharply
demarcated
edges.

Pharmacokinetics

- **-** The major metabolic product of Dapsone results from **N-acetylation** in the liver by ***N-acetyltransferase***.
- **-** It also undergoes **N-hydroxylation** to **hydroxylamine derivative**. These metabolic reactions are catalyzed by **CYP3A4** isoforms.
- **-** The urine consists of small amounts of Dapsone and the metabolites, that is, **N-acetyldiamino-diphenyl sulphone** and **N-hydroxy-diamino-diphenyl sulfone**, as well as glucuronide and sulphate of each of these substances.







- Uses

- Dapsone is commonly used in combination with rifampicin and clofazimine for the treatment of **leprosy**.

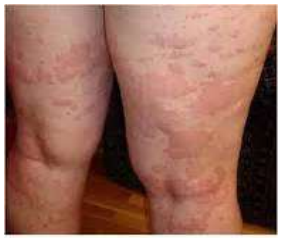
- It is also used to both treat and prevent **pneumocystis pneumonia** and **toxoplasmosis**.

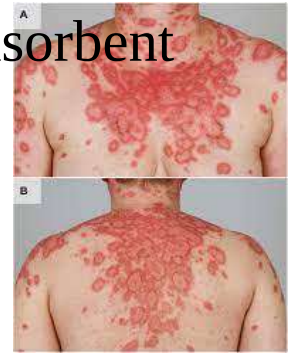
- Dapsone by mouth was one of the first medications used to treat moderate to severe **acne vulgaris** and useful in the prevention of malaria.

- Dapsone also used to treat Autoimmune disease (like Cutaneous lupus erythematosus, Idiopathic thrombocytopenic purpura, Chronic spontaneous urticaria, Relapsing polychondritis).

- Dapsone also used in treatment of Dermatitis herpetiformis and generalized granuloma annulare.

- Dapsone has been used as a monomer in the design of dye adsorbent polymers.





Trimethoprim/Sulphamethoxazole

- **Trimethoprim/Sulphamethoxazole** (**TMP/SMX**), also known as **Co-Trimoxazole**.

- Co-trimoxazole is available in oral and intravenous preparations with the standard single-strength tablet containing **80 mg of trimethoprim** combined with **400 mg of sulfamethoxazole**.

- It is an antibiotic used to treat a variety of bacterial infections.

- It consists of one part trimethoprim to five parts sulfamethoxazole.

- It is used for urinary tract infections, skin infections, travellers' diarrhoea, respiratory tract infections, and cholera, among others.

- It may be used both to treat and prevent pneumocystis pneumonia in people with HIV/AIDS.

Co-Trimoxazole can be used to treat or prevent:

▪ Lung infections (pneumonia or PJP) caused by a bacterium called *Pneumocystis jirovecii* (previously P. carinii).

▪ It is used for skin infections, travellers' diarrhoea, and cholera.

▪ Infections caused by a bacterium called Toxoplasma (toxoplasmosis).

▪ An infection called nocardiosis which can affect the lungs, skin and brain.

▪ It may be used both to treat and prevent pneumocystis pneumonia in people with HIV/AIDS.

- It can be given by orally or intravenously.

- **Common side effects** include nausea, vomiting, rash, and diarrhoea.

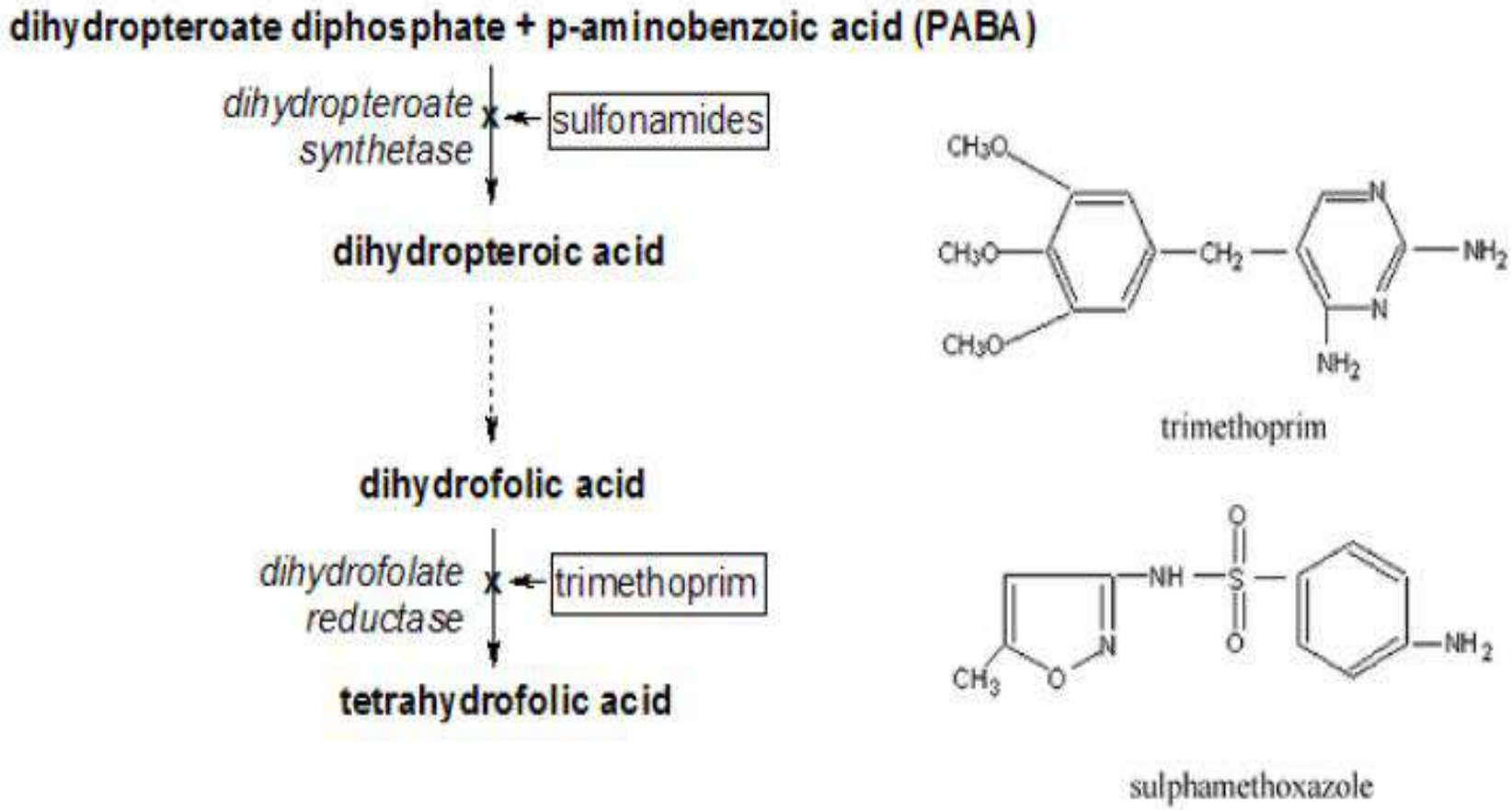
- Severe allergic reactions and *Clostridium difficile diarrhoea* may occasionally occur.

- Its use near the end of pregnancy is not recommended.

- It appears to be safe for use during breastfeeding as long as the baby is healthy.

- TMP/SMX generally results in bacterial death. It works by blocking the making of folate by the bacteria.

- **Mechanism of Action**



Drugs	Uses
Sulphamethizole	Cystitis, Genital tract inflammation, Gonorrhea, Nephritis, Prostatitis, Urinary Tract Infection and Vaginal Inflammation
Sulphaisoxazole	Urinary Tract Infections, Meningococcal Meningitis, Acute Otitis Media, Trachoma, Inclusion Conjunctivitis, Nocardiosis, Chancroid, Toxoplasmosis, Malaria and Other Bacterial Infections.
Sulfamethazine	For the treatment bacterial infections causing bronchitis, prostatitis, Bacterial Conjunctivitis, Endometritis, Furuncle, Streptococcal Sore Throat, Ulcers and Urinary Tract Infections.
Sulfacetamide	For the treatment of Bacterial Vaginitis, Keratitis, Acute Conjunctivitis, Acne Vulgaris, Conjunctivitis, Trachoma, Superficial Ocular Infections and Blepharitis.
Sulphapyridine	For the treatment of Dermatitis Herpetiformis, Benign Mucous Membrane Pemphigoid and Pyoderma Gangrenosum.

Sulphamethoxazole	Sulfamethoxazole is indicated in combination with trimethoprim, in various formulations, for the following infections caused by bacteria with documented susceptibility: urinary tract infections, acute otitis media in pediatric patients (when clinically indicated), acute exacerbations of chronic bronchitis in adults, enteritis caused by susceptible Shigella, prophylaxis and treatment of *Pneumocystis jiroveci* pneumonia, and travelers' diarrhea caused by enterotoxigenic *E. coli*. Additional indications include the adjunctive treatment of cholera, treatment of bacillary dysentery, nocardiosis, and second-line treatment of brucellosis in combination with gentamicin or rifampicin.
Sulphadiazine	For the treatment of rheumatic fever, Nocardiosis, Plague, Plasmodium Infections, Toxoplasmosis, Trachoma, Urinary Tract Infection, Wound Infections, Bacterial otitis media caused by Haemophilus influenzae, Prophylaxis of Rheumatic fever Recurrent Rheumatic fever and meningococcal meningitis.
Mafenide	Indicated for use as an adjunctive topical antimicrobial agent to control bacterial infection when used under moist dressings over meshed autografts on excised burn wounds (Second Degree Burns and Third-Degree Burns).
Sulfasalazine	For the treatment of Crohn's disease, Crohn's Disease (CD), Polyarticular juvenile rheumatoid arthritis, chronic or unspecified, Proctitis, Rheumatoid Arthritis, Severe Ulcerative Colitis, Mild Ulcerative Colitis, Moderate Ulcerative colitis

Organic Pharmaceutical Chemistry II

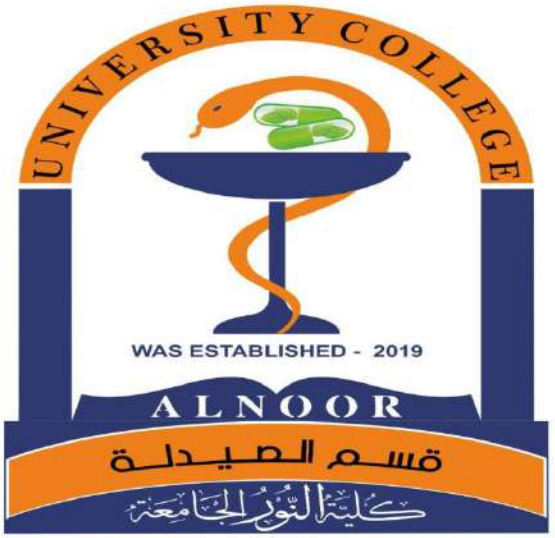
BY:

Dr. Nohad A AlOmari (B.Pharm/MSc & PhD in Medicinal Chemistry)

Organic Pharm. Chemistry II

Lecture 1

16th September 2023(8:30- 11:30)(11:30-2:30)



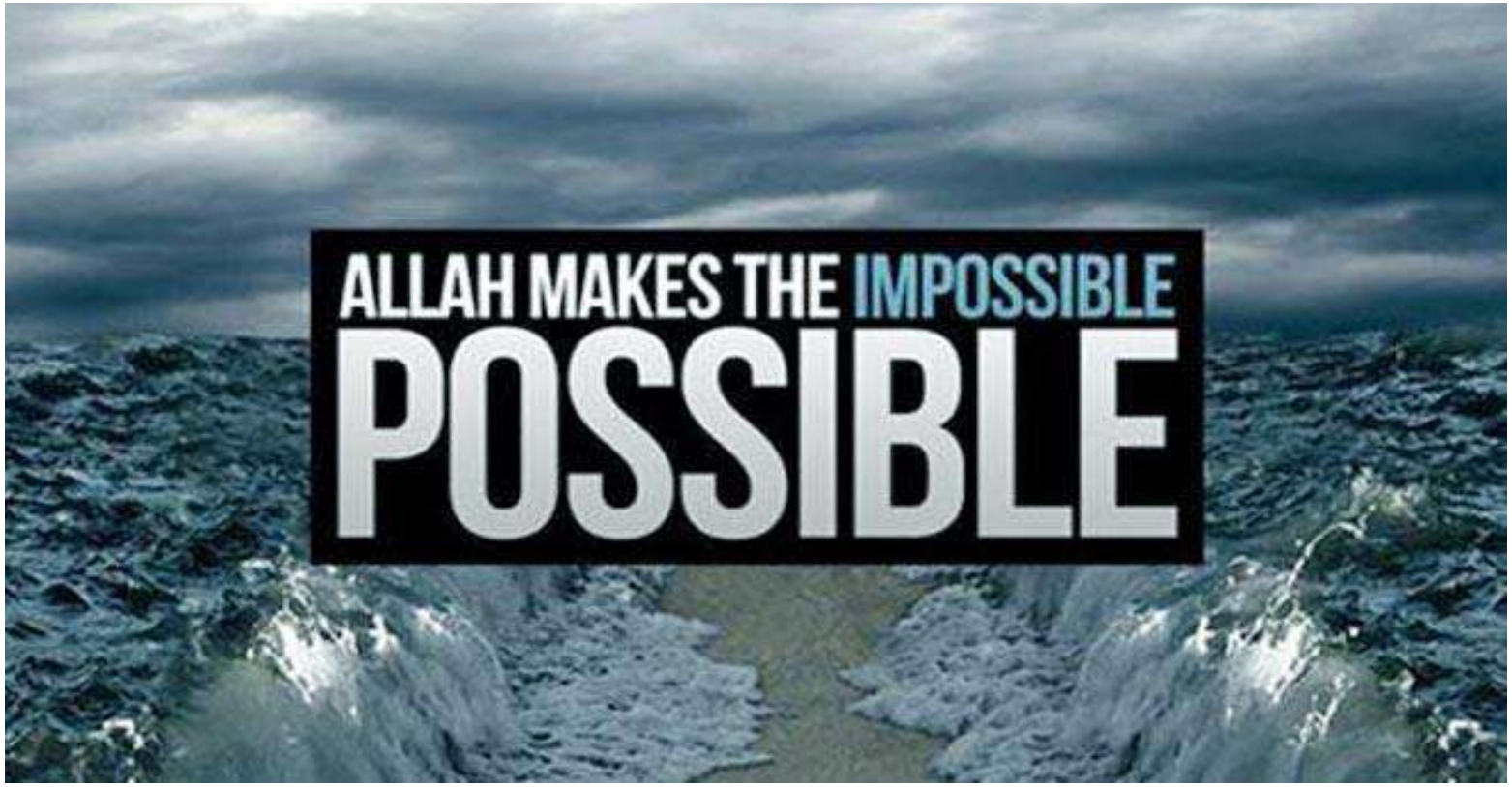
AlNoor University College

Pharmacy Department

1st Course/ 4th Class

2023/2024

Quote of Today



University of Baghdad
College of Pharmacy
Department of Pharmaceutical Chemistry
Title of the course: ***Organic Pharmaceutical Chemistry* II** Course number: **412**
Level: 4th Class, 1st Semester
Credit hours: **Theory 3 hours** **Laboratory 1 hour**
Tutors:
Reference text: ***Wilson and Gisvold Textbook of Organic Medicinal and Pharmaceutical Chemistry; Delgado JN, Remers WA, (Eds.); 10th ed., 2004.***

Objectives: **The course is devoted to the discovery and development of new agents for treating diseases, and enables translating the drug structural formula into therapeutic effect. Additionally, it focuses on the methods of preparation for some pharmaceutical agents.**

No	Lecture title	hours
1.	Cholinergic agents, cholinergic receptors and their subtypes.	2
2.	Cholinergic agonists; stereochemistry and structure-activity relationships (SAR); products; cholinesterase inhibitors.	5
3.	Cholinergic blocking agent; structure-activity relationships (SAR); Solanaceous alkaloid and analogues; synthetic cholinergic blocking agents and products; ganglionic blocking agents (neuromuscular blocking agents).	6
4.	Analgesic agents (SAR of morphine, SAR of meperidine type molecules; SAR of methadone type compounds; N-methylbezomorphans, antagonist type analgesics in benzomorphans).	3
5.	Analgesic receptors, endogenous opioids; Products; Antitusive agents; Anti-inflammatory analgesics.	7
6.	Adrenergic agents (Adrenergic neurotransmitters); Adrenergic receptors; Drugs affecting Adrenergic neurotransmission; Sympathomimetic agents; Adrenergic receptor antagonists.	11
7.	CNS depressant; Benzodiazepines and related compounds; Barbiturates; CNS depressant with skeletal muscle relaxant properties; Antipsycotics; Anticonvulsants.	9

Reference Books

- Wilson and Gisvold's Textbook of Organic Medicinal and Pharmaceutical Chemistry - 12th Edition Beale, John M. , Block, John H.

Extra Reading Assignments:

1.Foye, W.O. (1995) Principles of Medicinal Chemistry, Chapter 17. Williams & Wilkins , Philadelphia, pp. 321-344.

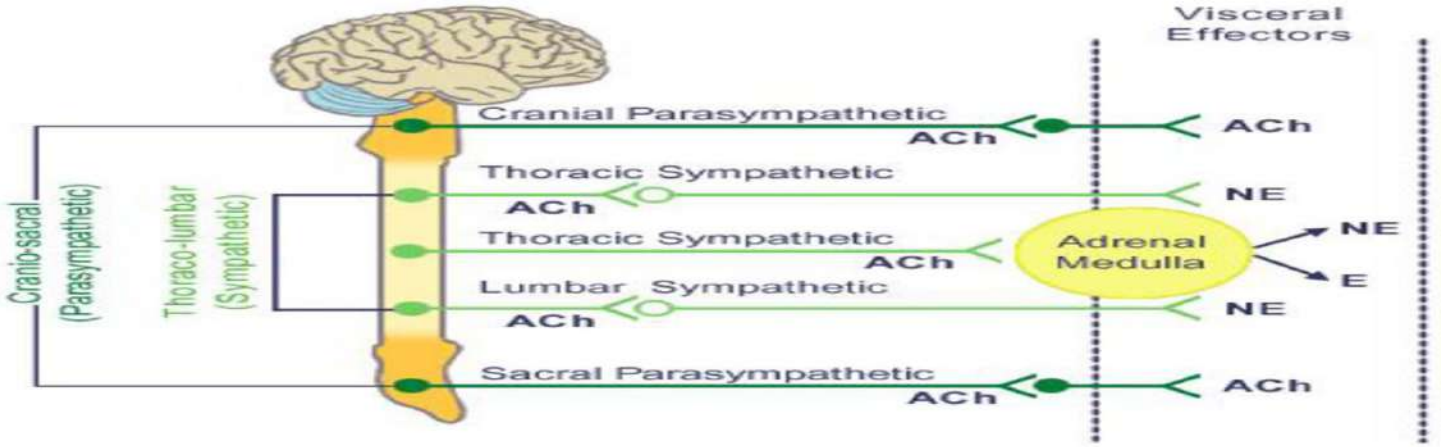
2. Goodman & Gilman’s The Pharmacological Basis of Therapeutics, 9th ed., McGraw Hill, 1996, New York, pp. 115-118.

Introduction:

-Definition of Cholinergic agent.

-Cholinergic receptors (Muscarinic and Nicotinic receptors); their subtypes, locations and functions.

-Synthesis, Structure activity relationship (**SAR**) of Acetyl choline (Ach).



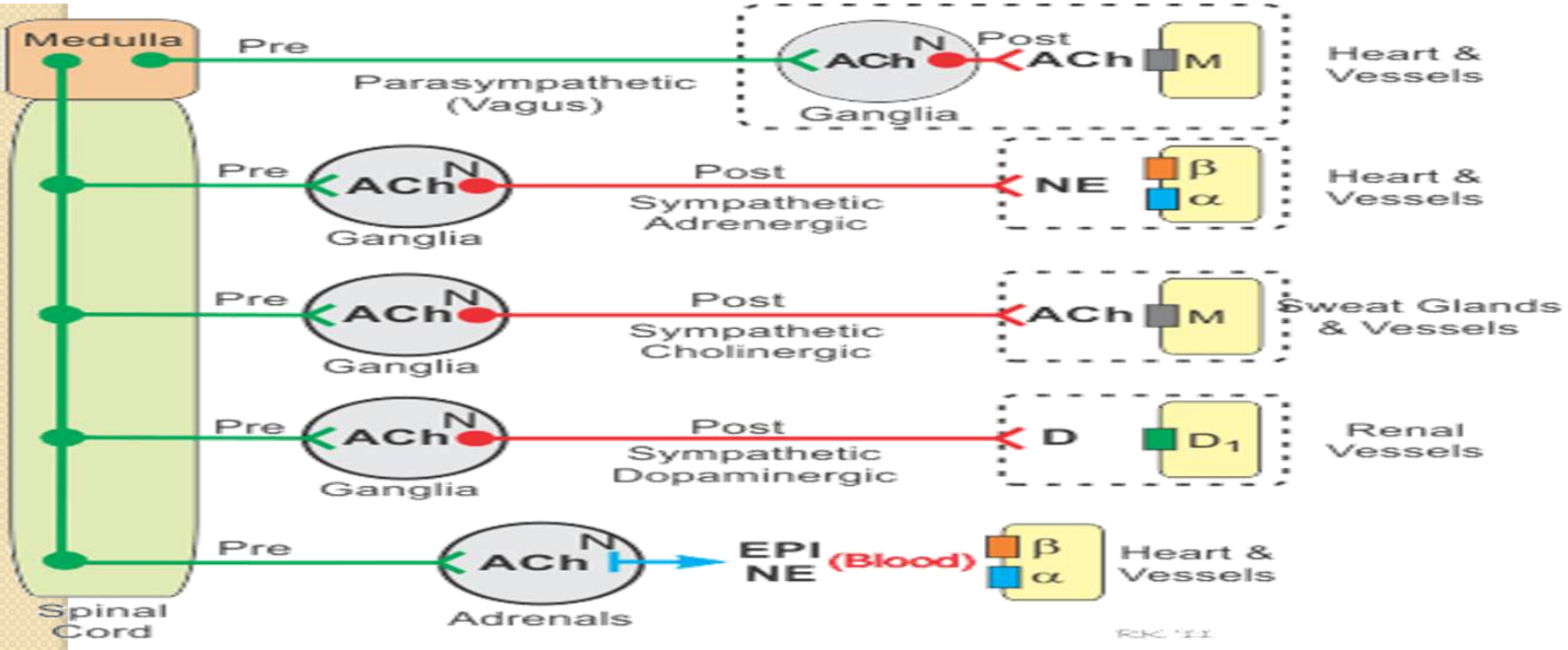
that **Ach** is the transmitter secreted by **all autonomic preganglionic** nerve terminals. Hence, **all** synapses between pre- and post-ganglionic fibres are **cholinergic**.

Similarly, **parasympathetic postganglionic** fibres secrete **Ach**, i.e. synaptic junctions between parasympathetic postganglionic fibres and effector organs are also **cholinergic**.

By contrast, virtually all **sympathetic postganglionic** neurons secrete NE: Thus, synapses between sympathetic postganglionic fibres and effectors are **adrenergic**. Note that there are some **exceptions to this latter rule:** Sympathetic postganglionic fibres innervating certain **sweat glands** (eccrine glands) secrete Ach, as do some of the sympathetic postganglionic fibres controlling blood vessel dilatation in skeletal muscle.

To Summarize:

Type of axon	Transmitter	Action
Preganglionic-symp + parasymp.	Ach	Exc. only
Postganglionic-symp. (most)	NE	Exc./Inhib.
Postganglionic-symp. (some)	Ach	Exc./Inhib.
Postganglionic-parasymp.	Ach	Exc./Inhib.



CNS = central nervous system; Pre = preganglionic; Post = postganglionic; ACh = acetylcholine; N = nicotinic receptor; NE = norepinephrine; EPI = epinephrine; D = dopamine; M = muscarinic receptor; β = β-adrenoceptor; α = α-adrenoceptor; D_1 = dopaminergic receptor

Pharmacological effects of Ach

A) Through the muscarinic receptor

Cardiac effects

- Bradycardia,
- decrease of atrioventricular conduction.
- decrease of the strength of atrium contractions.

Blood vessels

- Acetylcholine injection causes release of nitric oxide (NO) which dilates blood veins

- **Effects on smooth muscles**
- intestine: an increase in tone with sometimes an increase in the peristaltic contractions. This can lead to Nausea and vomiting.
- ureters: increase in tone.
- bronchi: bronchoconstriction. (An aerosol of acetylcholine can cause an attack of asthma)
- **Effects on secretions**
- Acetylcholine increases digestive (abundant saliva), bronchial, cutaneous (sweat) and lacrimal (tears) secretions.
- **Effects on the eye**
- Acetylcholine induces a decrease of iris diameter or miosis which can lower the intra-ocular pressure

- Through the nicotinic receptor
- **In Autonomic NS**, Ach allows nerve transmission
- **In neuromuscular junctions**
  - At low dose Ach allow skeletal muscle movement (important for breathing)
  - At high does, it causes muscle paralysis!
- **In Brain**, cholinergic deficiency causes Alzheimer disease

Autonomic nervous system

The component of the CNS (Central Nervous System) that functions below the conscious level, controlling several key physiological processes:

1. Distribution of blood flow & tissue perfusion
2. Regulation of blood pressure
3. Control of visceral smooth muscle (eyes, bladder, bowels)
4. Control of endocrine and exocrine glands
5. Control of metabolic energy (glycolysis, neoglucogenesis etc)

Biologic responses to parasympathetic stimulation:

- Constriction of pupil (miosis), ciliary body ("accommodation" of lens)
- Contraction of smooth muscle in the GI ("peristalsis") and urinary tract
- Constriction of the bronchioles ("broncho constriction")
- Slowing of heart rate ("bradycardia")
- Increased secretion by the gland

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Autonomic nervous system

Muscarinic Receptors

Acetylcholine is the transmitter at **three different sites**, autonomic ganglia, parasympathetic postganglionic nerve terminals and skeletal muscle motor nerve terminals.

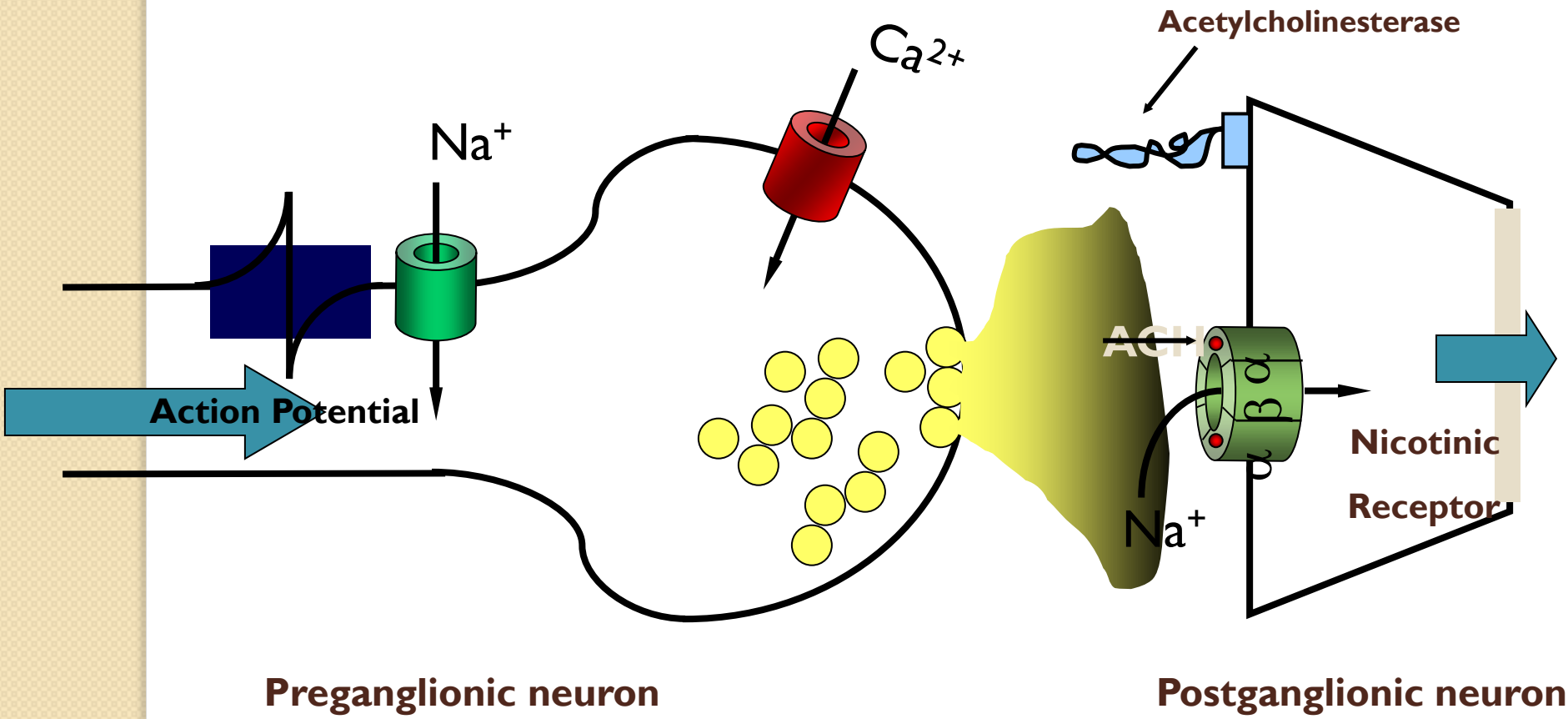
The action of acetylcholine on visceral effectors (smooth muscle of GIT, cardiac muscle and exocrine gland) resembles the action of the naturally occurring plant alkaloid muscarine. Therefore these receptors of acetylcholine on visceral effectors are called **muscarinic receptors** .

Atropine blocks this receptor of acetylcholine. Acetylcholine in large dose exhibits its effects through ganglionic stimulation.

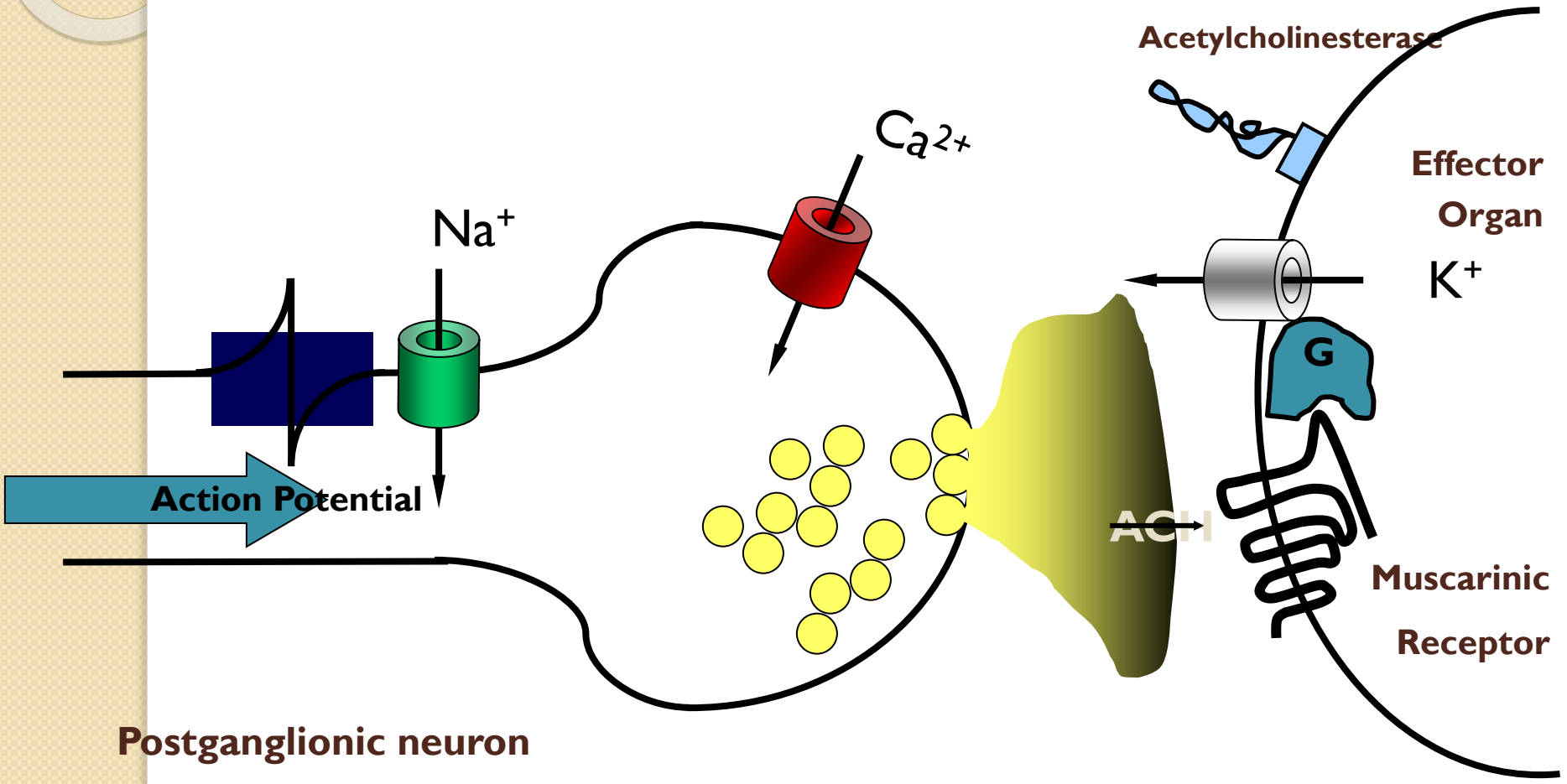
This response resembles the effect of naturally occurring alkaloid nicotine. Thus, the response of acetylcholine on parasympathetic ganglia, sympathetic ganglia and adrenal medulla and neuromuscular junction are called **nicotinic receptors** .

At the skeletal muscle motor plate the cholinergic receptors are nicotinic in nature; d-tubocurarine blocks these receptors in the skeletal muscle. The ganglionic nicotinic receptors are blocked by hexamethonium.

Parasympathetic Ganglionic Synapse

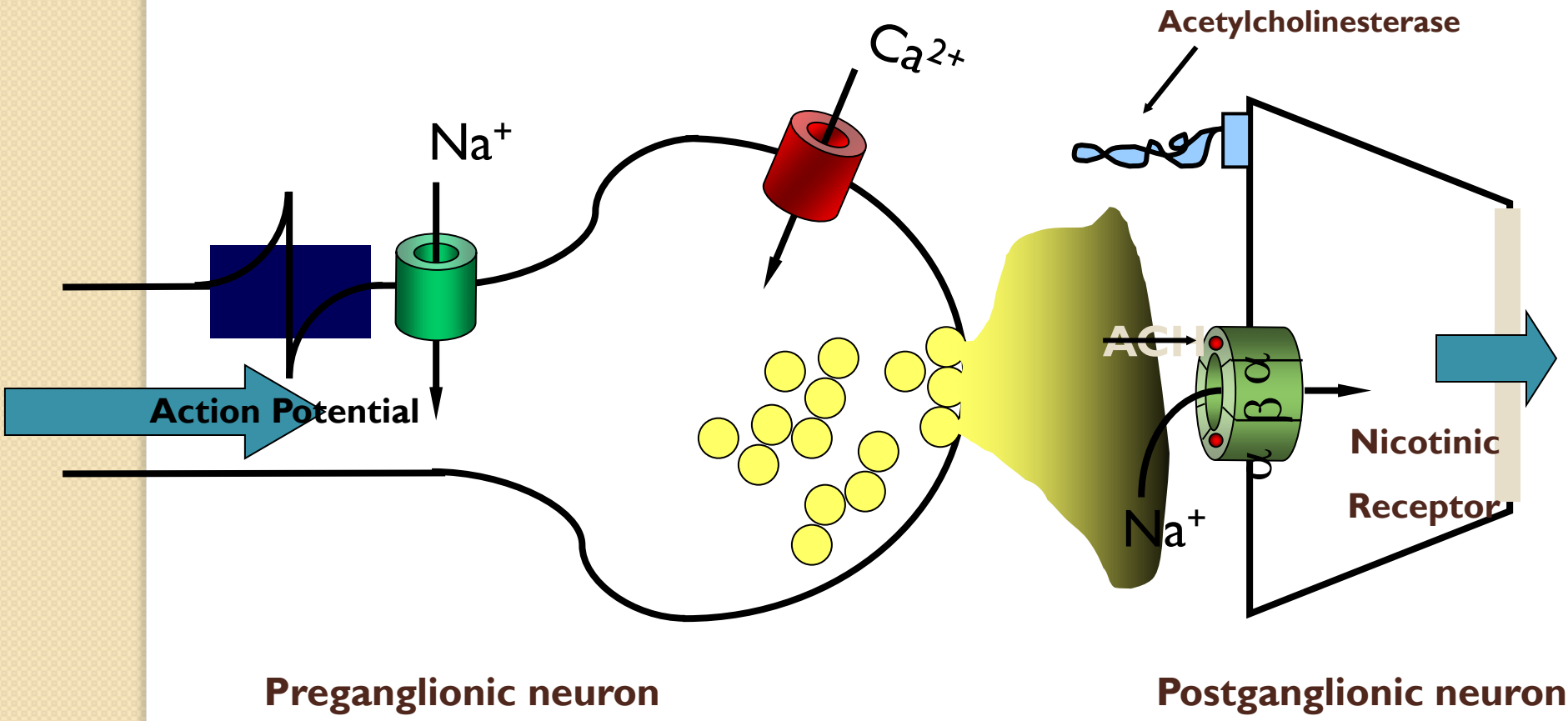


Parasympathetic Organ Synapse



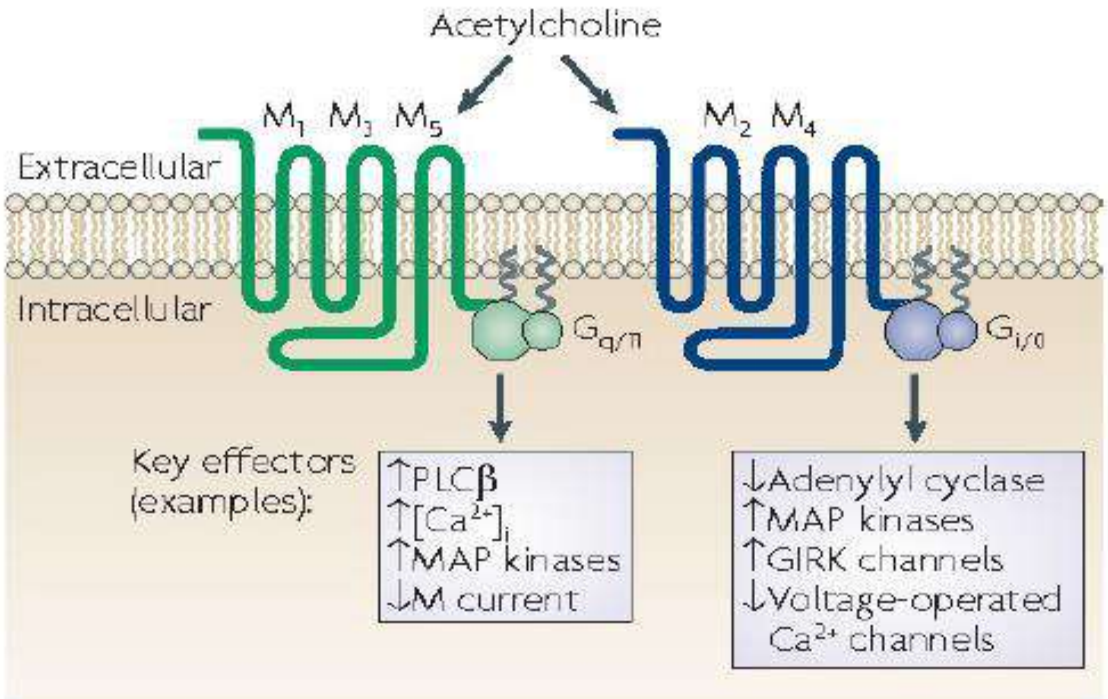
Postganglionic neuron

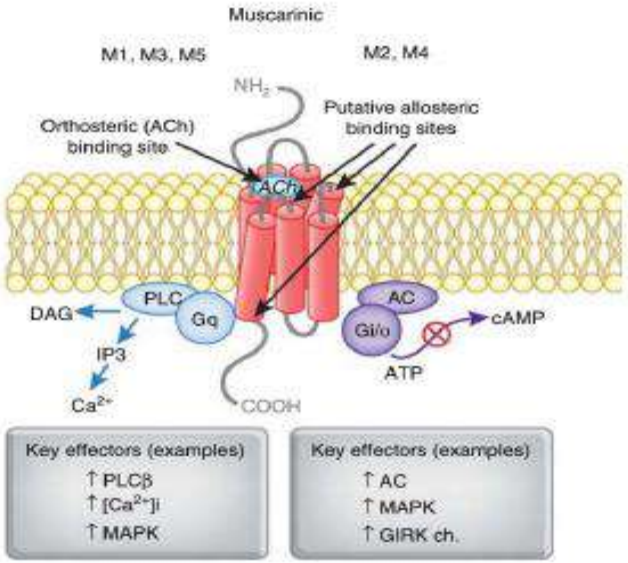
Sympathetic Ganglionic Synapse



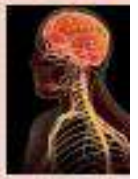
Muscarinic receptor (M); mitogen-activated protein kinase (MAPK or MAP kinase); Phospholipase C (PLC); G-Protein-Coupled Inwardly Rectifying Potassium (GIRK)

Subclassification of mAch receptors



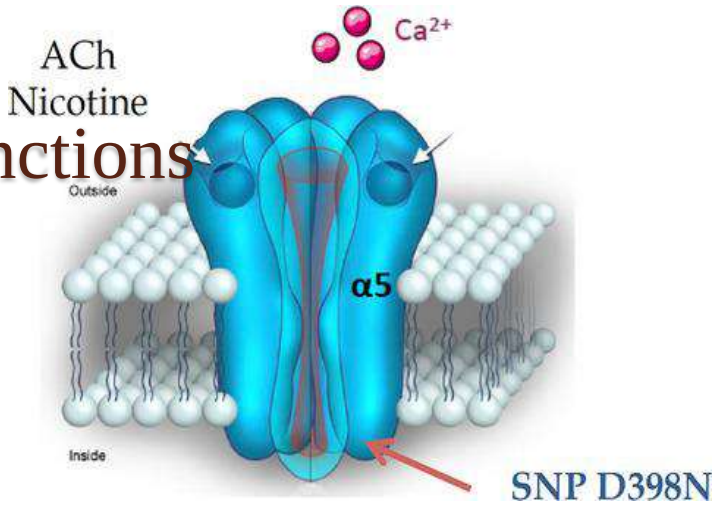


Muscarinic receptors subtypes and functions

Receptor	Tissue location	Function
M1	CNS, gastric and salivary glands, autonomic ganglia, enteric nerves	↑ Cognitive function ↑Seizure activity, ↑Secretions ↑ Autonomic ganglia depolarization ↓ DA release and locomotion 
M2	Autonomic nerve terminals; CNS; heart; smooth muscle	↑ Smooth muscle contraction Neural inhibition in periphery via autoreceptors and heteroreceptor ↓ Ganglionic transmission Neural inhibition in CNS, ↓ Heart rate ↑ Tremors hypothermia & analgesia
M3	CNS (< other mAChRs), smooth muscle, glands, heart	↑ Smooth muscle contraction (e.g., bladder) ↑ Salivary gland secretion ↑ Food intake, body fat deposits Inhibits dopamine release Synthesis of nitric oxide
M4	CNS	Inhibition of autoreceptor- and heteroreceptor-mediated transmitter release in CNS, Analgesia, Cataleptic activity; Facilitates dopamine release
M5	Low levels in CNS & periphery; predominate mAChRs in dopaminergic neurons of substantia nigra & ventral tegmentum area	Mediates dilation of cerebral arteries Facilitates dopamine release Augments drug seeking behavior and reward

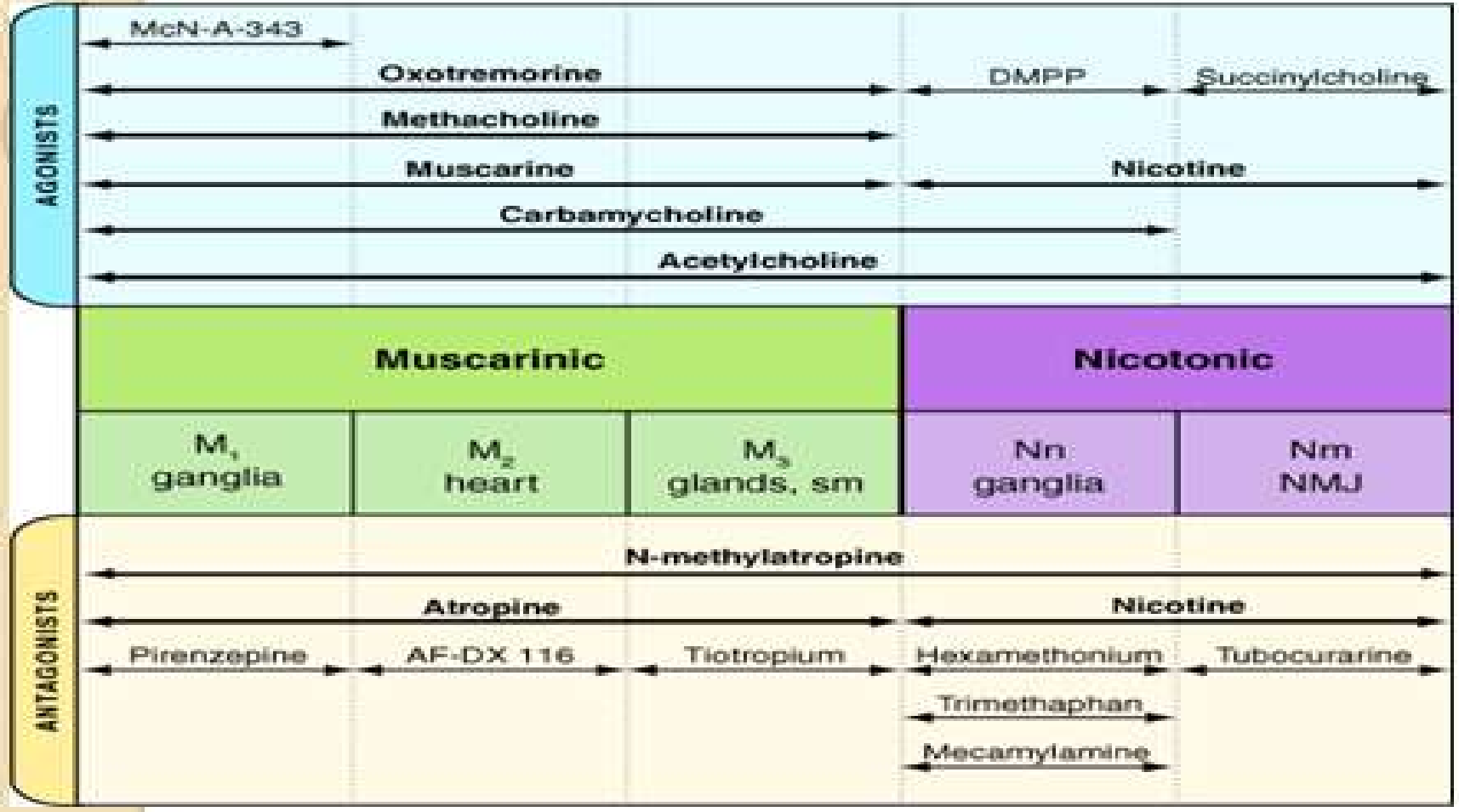
Nicotinic receptors subtypes and functions

Receptor	Location	Membrane Response
Skeletal muscle (N_M) $(\alpha_1)_2\beta_1\varepsilon\delta$ $(\alpha_1)_2\beta_1\gamma\delta$	Skeletal neuromuscular junction (post-junctional)	Excitatory; end plate depolarization; contraction (skeletal muscle)
Peripheral neuronal (N_N) $(\alpha_3)_2(\beta_4)_3$	Autonomic ganglia; adrenal medulla	Excitatory; depolarization firing of postganglionic neuron; depolarization & secretion of catecholamines
Central neuronal (CNS) $(\alpha_4)_2(\beta_4)_3$ (α-bungarotoxin insensitive)	CNS; pre- & postjunctional	Pre- & postsynaptic excitation; prejunctinal control of transmitter release
$(\alpha_7)_5$ (α-bungarotoxin sensitive)	CNS; pre- and postsynaptic	Same as central neuronal

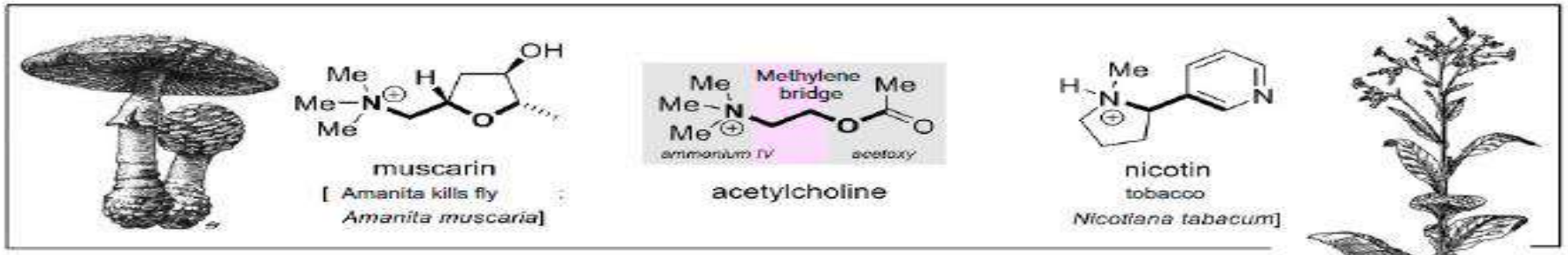


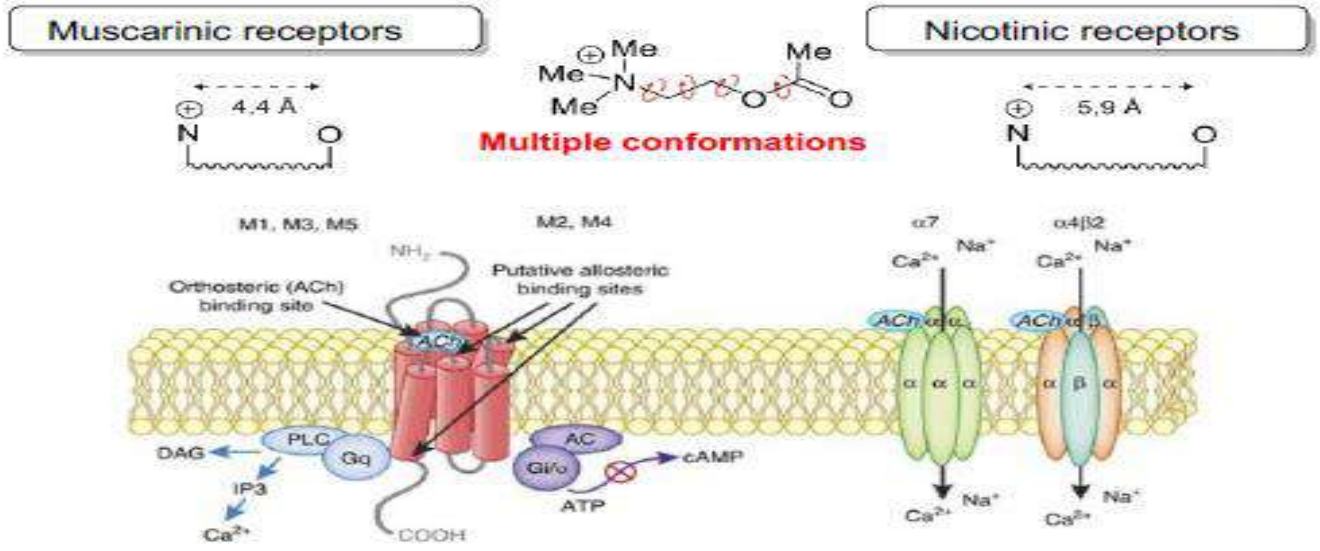
- They are agonized by nicotine
- They also occur in the CNS and Autonomic NS plus are exclusive in neuromuscular junction, and are part of a ligand gated ion channel receptors
- Physiological functions depend upon muscle-type or neuronal-type
- Muscle-type nicotinic AChRs are localized at neuromuscular junctions and allow muscle contraction and maintain muscle tone; **(thus these are targets for muscle relaxants)**
- Neuronal type are involved in cognitive function, learning and memory, arousal, reward, motor control and analgesia.

Muscarinic Receptors Vs Nicotinic Receptors



Muscarinic Receptors Vs Nicotinic Receptors

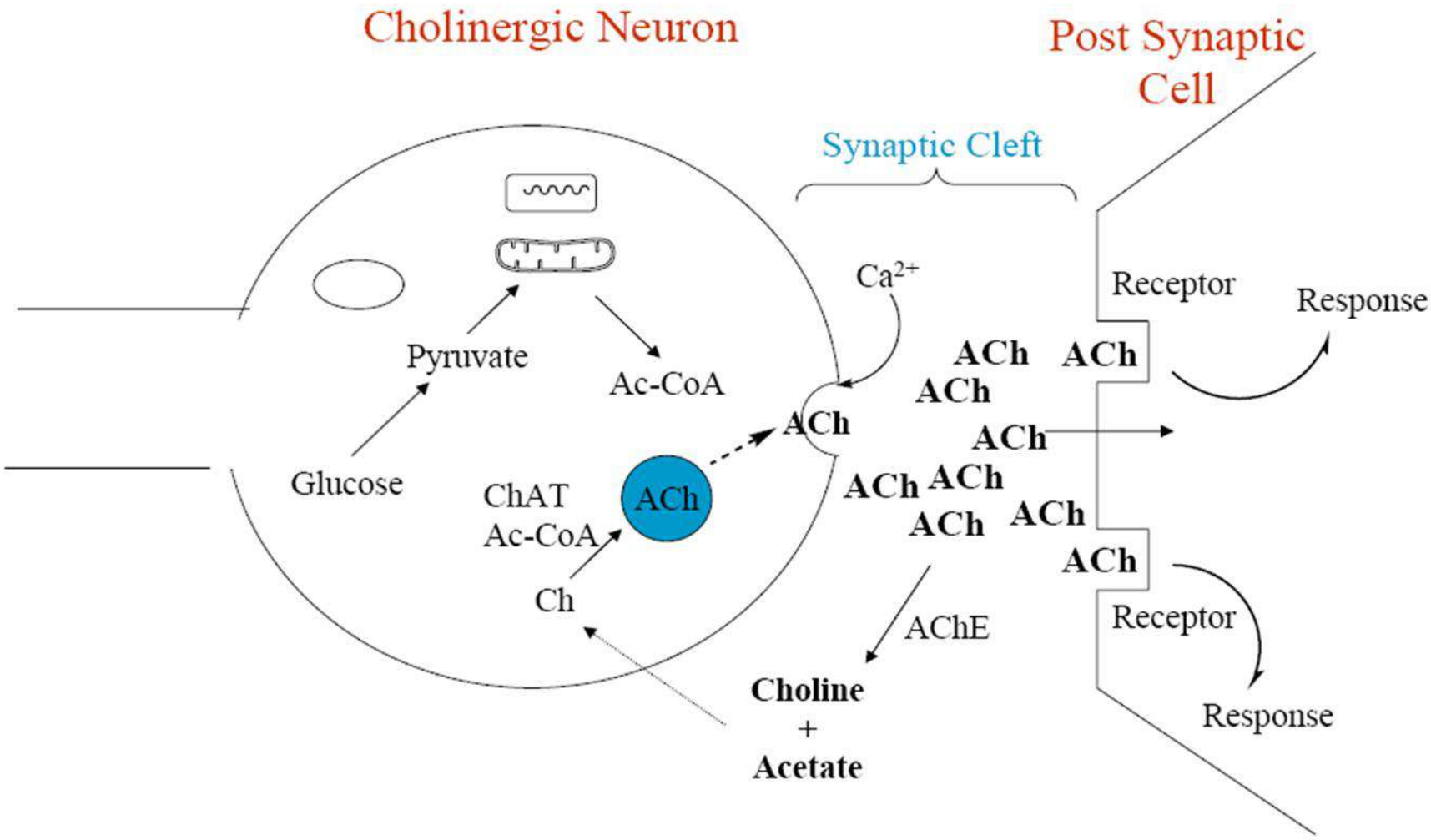




1. **Muscarinic receptors**
   Activates cholinergic receptors on smooth muscle and cardiac muscle

2. **Nicotinic receptors**
   Activates cholinergic receptors at nerve synapses and on skeletal muscle

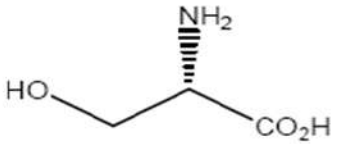
Schematic of Acetylcholine Biosynthesis, Action, Hydrolysis and Reuptake



ACh: Acetylcholine; Ch: Choline; ChAT: Choline acetyl transferase; Ac-CoA: Acetyl-*S*-Coenzyme A

Biosynthesis & Hydrolysis of Acetylcholine

Biosynthesis of Acetylcholine:

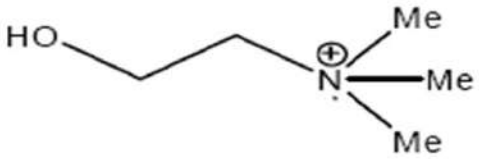


L-Serine

Serine Decarboxylase

Choline-*N*-Methyl-Transferase

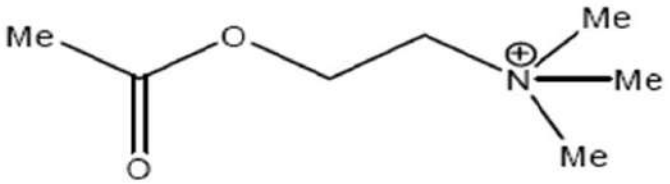
S-Adenosylmethionine (Me donor)



Choline

Choline Acetyl Transferase

Acetyl-*S*-Coenzyme A (Acetyl donor)

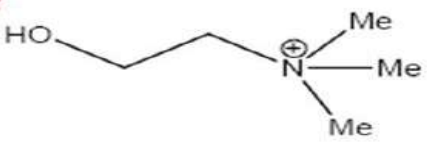


Acetylcholine

Hydrolysis of Acetylcholine:

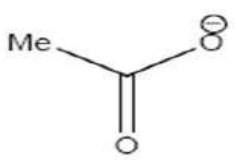
Acetylcholine

Acetylcholine Esterase (AChE)



Choline

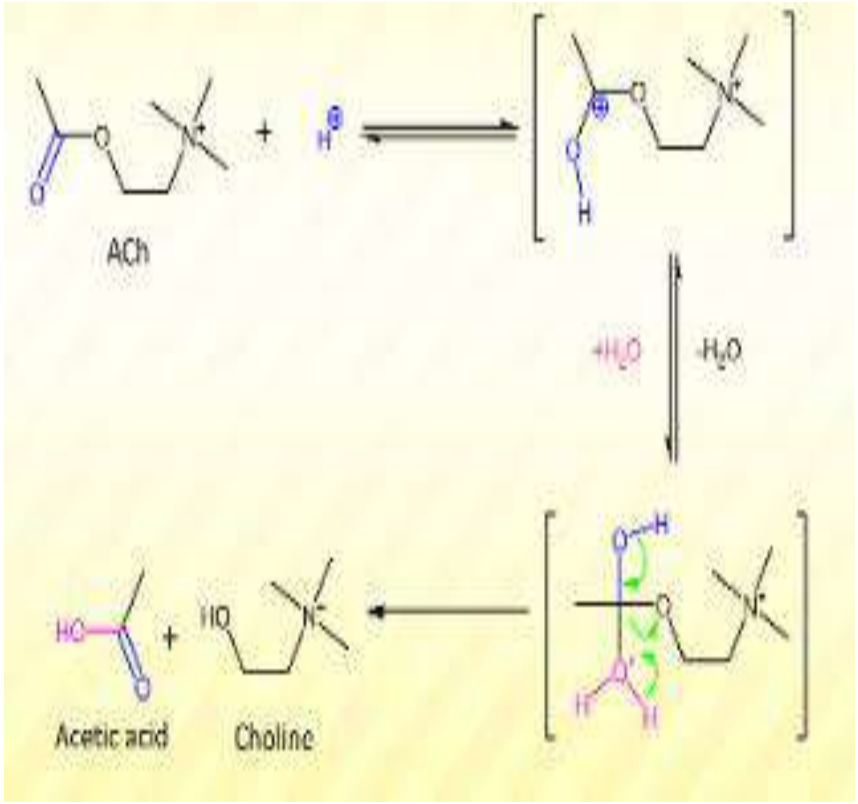
+



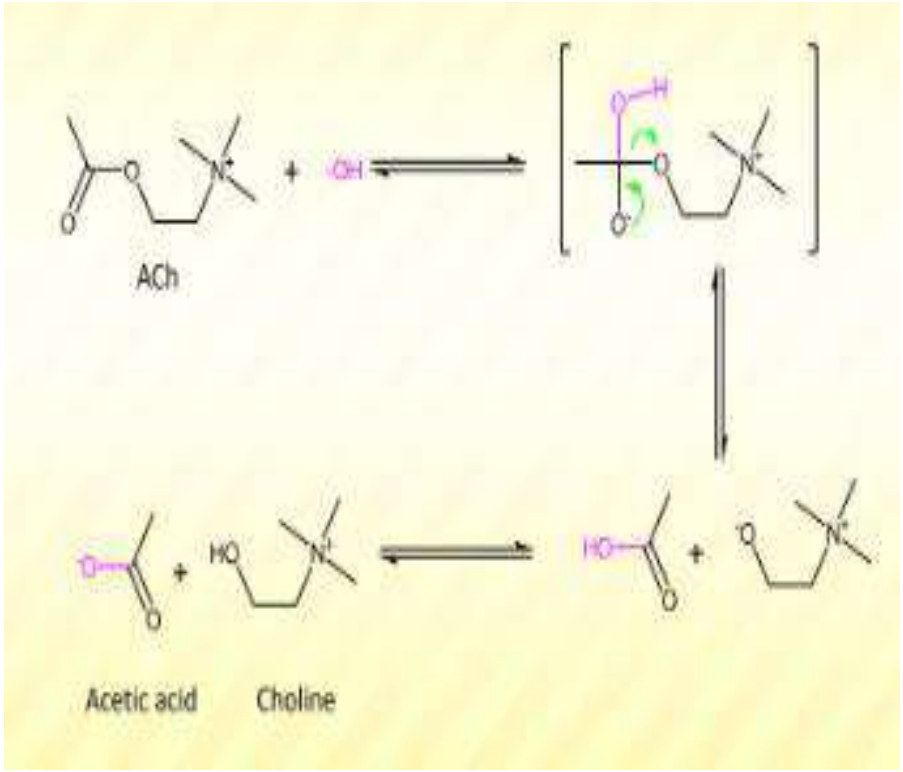
Acetate

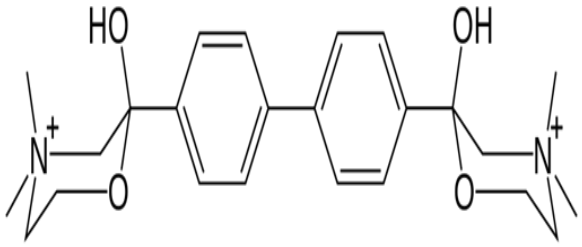
Hydrolysis of Ach

Acid catalyzed hydrolysis of Ach

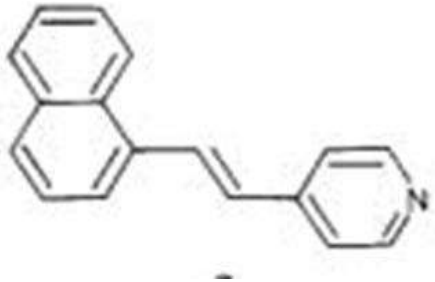


Base catalyzed hydrolysis of Ach



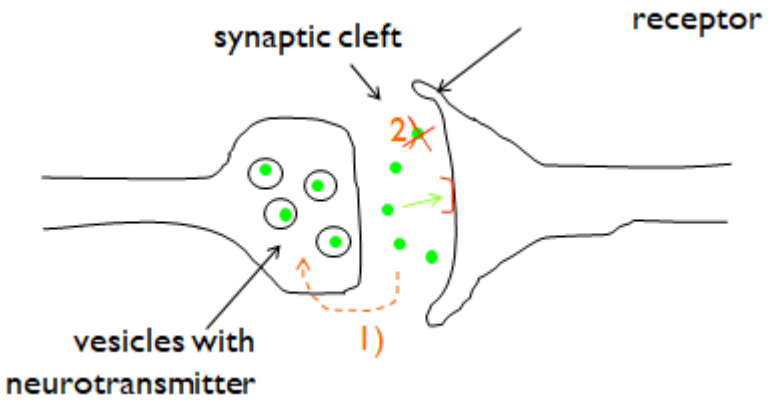


Hemicholinium HC-3

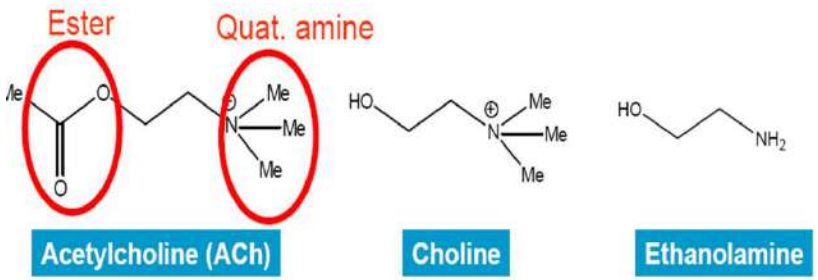


Trans-N-Methyl -4-naphthylvinyl)pyridinium iodide

Several quaternary ammonium bases act as competitive inhibitors of choline uptake. Hemicholinium(HC-3), bisquaternary cyclic hemiacetal, and triethyl analogue of choline, 2-hydroxyethyltriethylammonium, act at the presynaptic membrane to inhibit the high affinity uptake of choline into the neuron (1). Cause a delayed paralysis at repetitively activated cholinergic synapse and can produce respiratory paralysis in test animals. The delayed block is caused by the depletion of stored Ach, which may be reversed by choline.



- **Chemistry**
- Acetylcholine is a choline molecule that has been acetylated at the oxygen atom. Because of the charged ammonium group, acetylcholine does not penetrate lipid membranes. Because of this, when the molecule is introduced externally, it remains in the extracellular space and at present it is considered that the molecule does not pass through the blood–brain barrier.



Stereochemistry of Ach:-

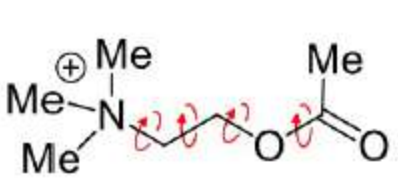
Early models for cholinergic receptor did not account for the observed stereo selectivity of the receptor for agonist and antagonist.

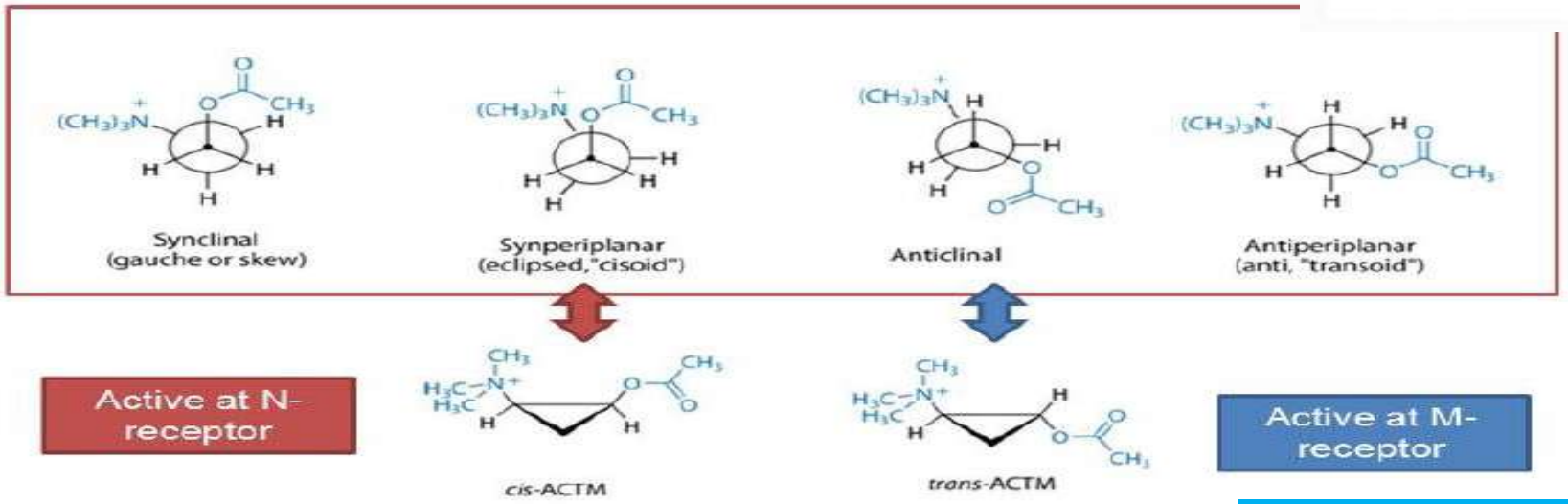
Even though Ach dose not exhibit optical isomerism but many synthetic & naturally occurring agonists and antagonists are optical isomers.

One of the enantiomer is many times more active than other. Thus the stereochemistry of cholinergic ligands is important for receptor binding.

The stereochemistry of cholinergic ligands provides rational basis for design of cholinergic drugs as well as to describe properties and function of cholinergic receptors.

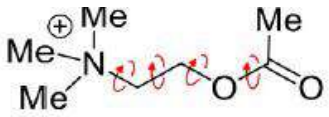
Conformations of Ach





ACTM:2-acetoxycycloproply-1-trimethylammonium

Defines separation of ester and N

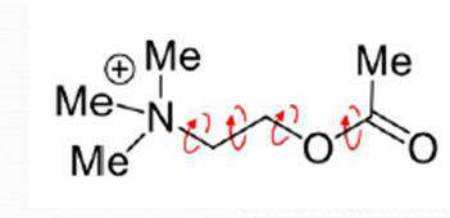


Muscarinic receptor

Nicotinic receptor

Separation between ester and quaternary nitrogen groups is important for binding . This distance is differ in Muscarinic and nicotinic receptors

Acetylcholine is not used in clinical practice why??

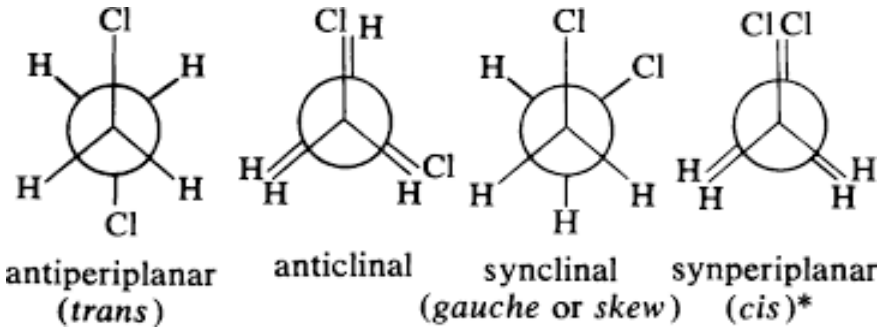


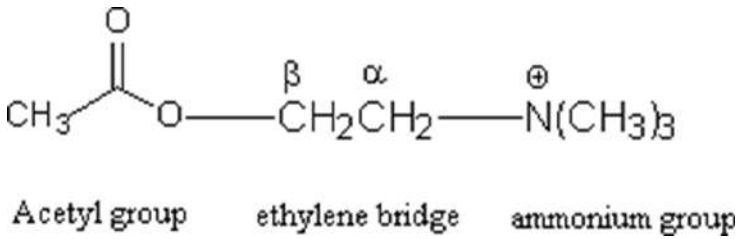
because:

1. It is very toxic
2. The doses required are very high
3. It is very rapidly hydrolyzed
4. It is very costly

STRUCTURE ACTIVITY RELATIONSHIP

- Acetylcholine can exist in a number of conformations. Four of these conformations are synplanar, synclinal, anticlinal, and antiplanar.
- The most active isomer is the (+) *trans* enantiomer and it is identical to synclinal conformation of acetylcholine.
- The muscarinic receptors and acetylcholinesterase display stereoselectivity, the (S) enantiomer of methacholine is equipotent with acetylcholine, while the R (–) enantiomer is about 20-fold less potent.



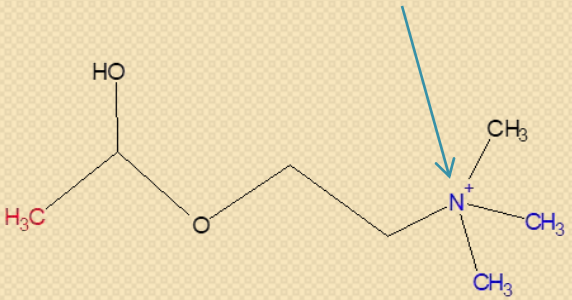


I. Modification of Quaternary Ammonium Group

The quaternary ammonium group is essential for intrinsic activity, and contributes to the affinity of the molecule for the receptors, partially through the binding energy and partially because of its action as a detecting group.

The trimethyl ammonium group is the optimal functional moiety for the activity, although some exceptions are known (e.g. pilocarpine, nicotine, and oxotremorine), and it shows maximal muscarinic activity.

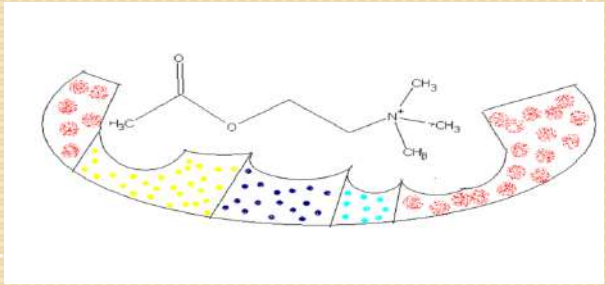
Placement of primary, secondary, or tertiary amines leads to decrease in activity.

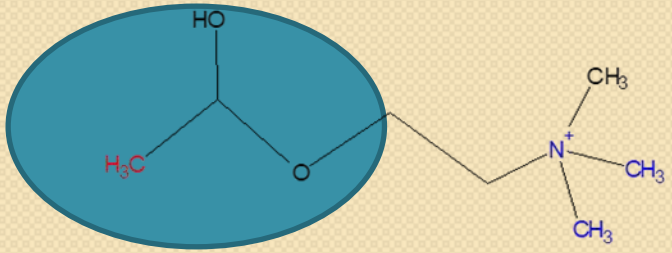


II. Modification of acyloxy group

The ester group of ACh contributes to the binding of the compound to the muscarinic receptor.

Replacement of methyl group by ethyl or large alkyl groups produces inactive compounds. Esters of aromatic or higher molecular weight acids possess cholinergic antagonist activity.





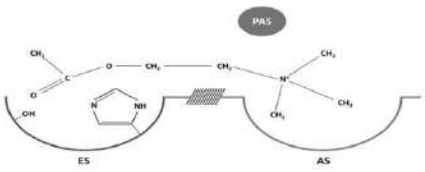
III. Modification of ethylene bridge

The methyl ester is rapidly hydrolyzed by cholinesterase to choline and acetic acid. To reduce susceptibility to hydrolysis, carbamate esters of choline (carbachol) were synthesized and were found to be more stable than carboxylate esters.

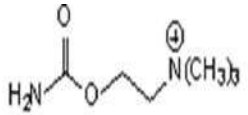
Placement of α-substitution in choline moiety results in a reduction of both nicotinic and muscarinic activity, but muscarinic activity to a greater extent.

Incorporation of β-substitution leads to reduction of nicotinic activity to greater extent.

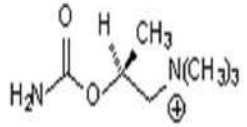
Replacement of ester group with ether or ketone produces chemically stable and potent compounds.



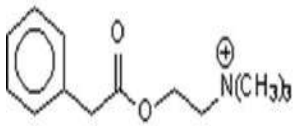
Replacement of acetyl function gives rise to agonist, partial agonist, or antagonist [please see the size and the nature of the functional group replacement]



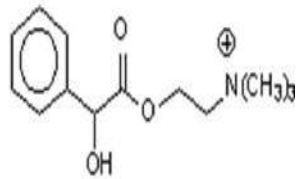
Carbachol
(nicotinic & muscarinic agonist)



Bethanechol
(muscarinic agonist)



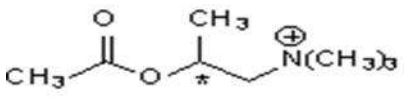
partial agonist / antagonist



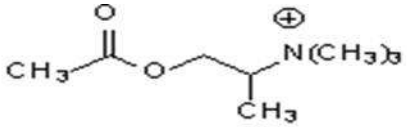
muscarinic antagonist

Schematic representation of AChE binding sites: ES – esteratic site, AS – anionic substrate binding site, PAS – peripheral anionic binding site.

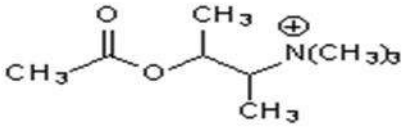
III. Modification of ethylene bridge



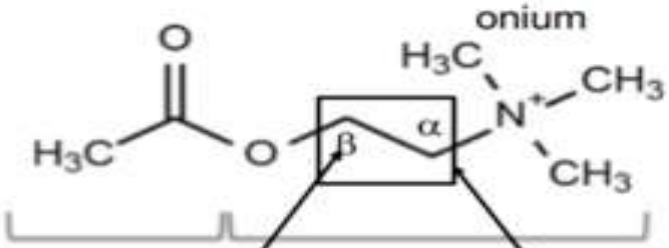
Muscarinic agonist
S(+) 240 X more active
than R(-) isomer



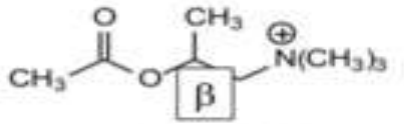
Nicotinic agonist



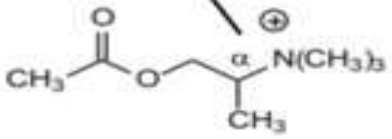
inactive
(both muscarinic or nicotinic)



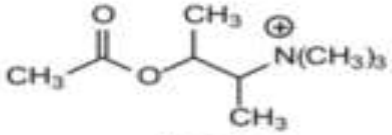
- β methyl substitution produces selective muscarinic agonist
- α methyl substitution produces selective nicotinic agonist



Methacholine
S(+) 240 X more active
than R(-) isomer



Nicotinic agonist



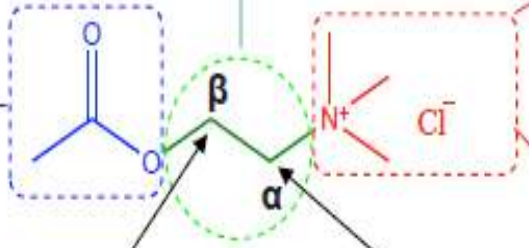
inactive
(both muscarinic or nicotini

Outlines of SAR of ACh

There should be a two-carbon units betweeen the oxygen atom and the nitrogen atom; **Ethylene group**

Quaternary amine group
A molecule must possess a nitrogen atom capable of bearing a positive charge, preferably a quaternary ammonium salt.

Acyloxy group
The molecule should have an oxygen atom, preferably an ester-like oxygen capable of participating in a hydrogen bond.



For maximum potency, the size of the alkyl groups substituted on the nitrogen should not exceed the size of a methyl group.

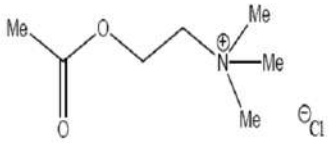
Inclusion of methyl group in beta carbon to N makes ***muscarinic*** selective

Inclusion of methyl group in beta carbon to N makes ***Nicotinic*** selective

The overall size of the molecule cannot be altered, much bigger molecules have poorer activity

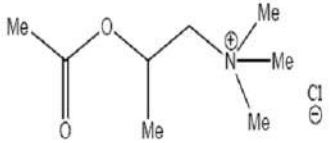
A larger third alkyl group is tolerated but more than one large alkyl groups leads to loss of activity

Cholinergic Agonists



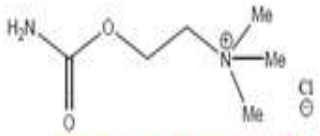
Acetylcholine chloride

- Prototypical muscarinic (and nicotinic) agonist, but a poor therapeutic agent.
- Chemical/enzymatic instability; Low bioavailability (poorly absorbed); Nonselective action.
- Quick onset and short duration of action.
- Use: In ocular surgery, causes complete miosis in seconds. Instilled directly in the anterior chamber.



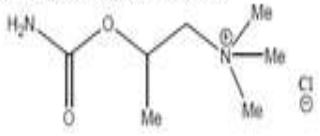
Methacholine chloride

- More stable than acetylcholine.
- More selective action (muscarinic > nicotinic).
- Racemic drug. *S*-enantiomer more active.

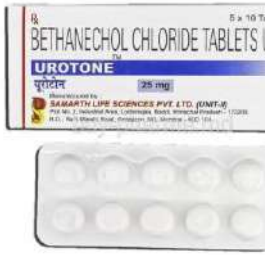


Carbachol chloride

- Potent agonist activity. Nonselective (Muscarinic / Nicotinic)
- Also acts indirectly by promoting ACh release and anticholinesterase (weak) activity.
- Increased hydrolytic stability (carbamate linkage more stable than ester)
- Uses: Topically for glaucoma; intraocular for miosis in surgery
- Side Effects: Corneal edema; decreased vision.

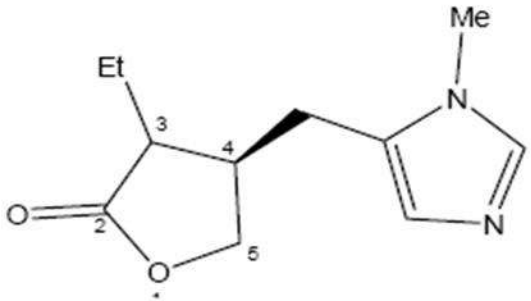


Bethanechol chloride



- Potent muscarinic agonist. Orally effective, also administered by subcutaneous injection.
- Increased hydrolytic stability (carbamate and steric bulk)
- Stimulant of GI tract smooth muscle and urinary bladder.
- Uses: For the relief of post-surgical urinary retention and abdominal distention.
- Low toxicity, no serious side effects. Should be used with caution in asthmatic patients.

Pilocarpine

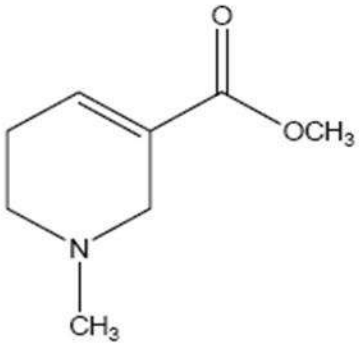


Pilocarpine

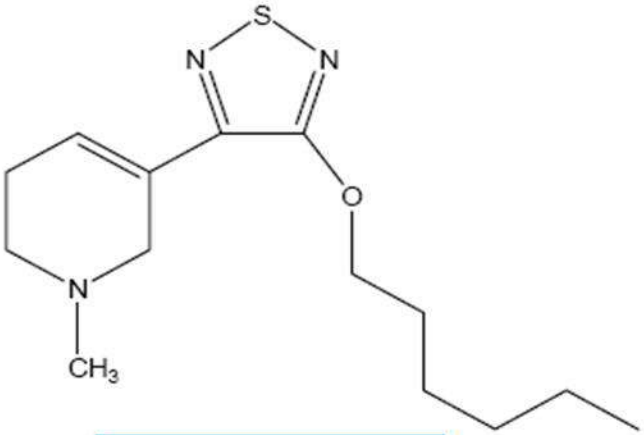
- Natural product. Isolated from the leaves of *Pilocarpus jaborandi*.
- Unstable to alkali (lactone hydrolysis) and bases (epimerization at C-3).
- Available as ophthalmic solution, gel, tablet and Ocusert delivery system.
- Systemic effects include copious sweating, salivation and gastric secretion.
- Used in the treatment of glaucoma and xerostomia (dry mouth).

Future Muscarinic Agonists:

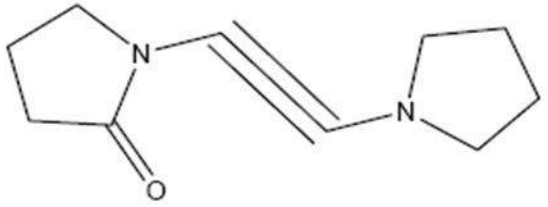
Current research interest is focused on developing agents with selective affinity for muscarinic receptors in the brain. Potentially useful in the treatment of Alzheimer's disease and other cognitive disorders.



Arecoline

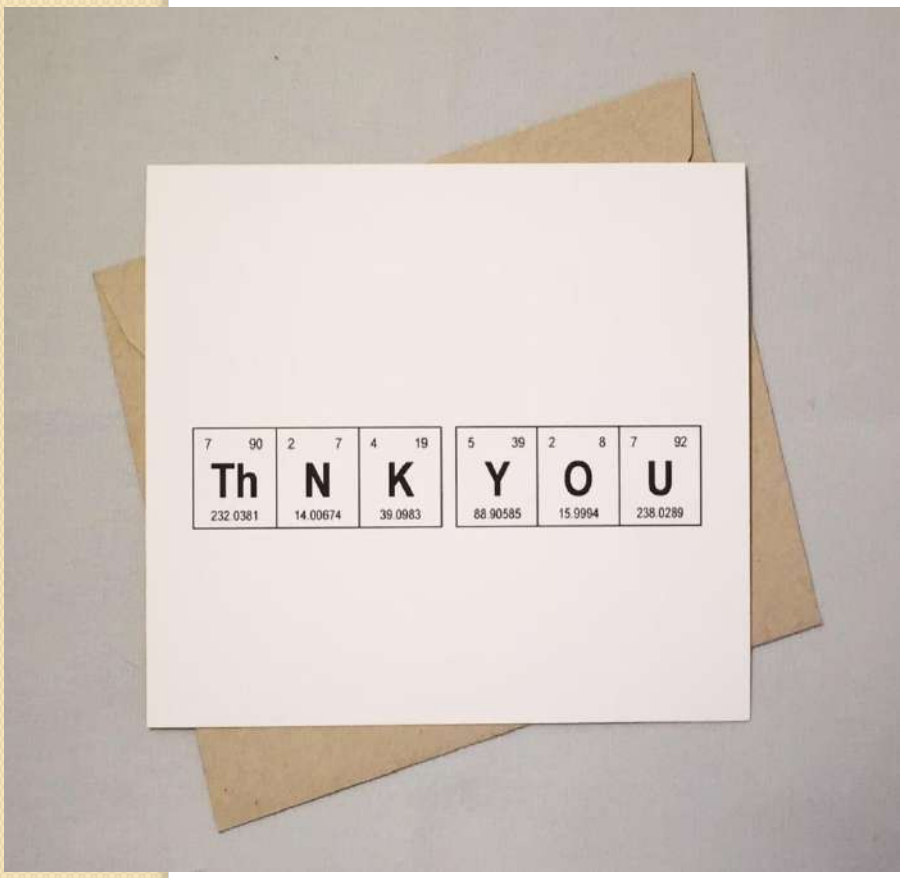


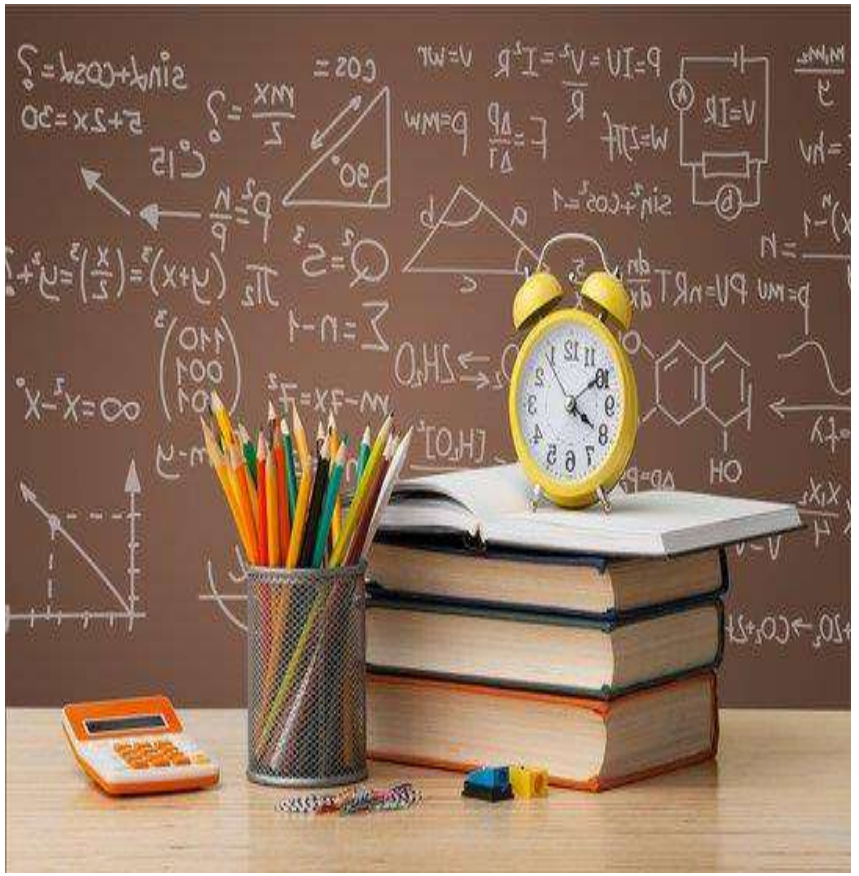
Xenomeline



Oxotremorine

Requisitions for Lec. 2
((classification of cholinergic drugs , introduction to anticholinergic Drugs))

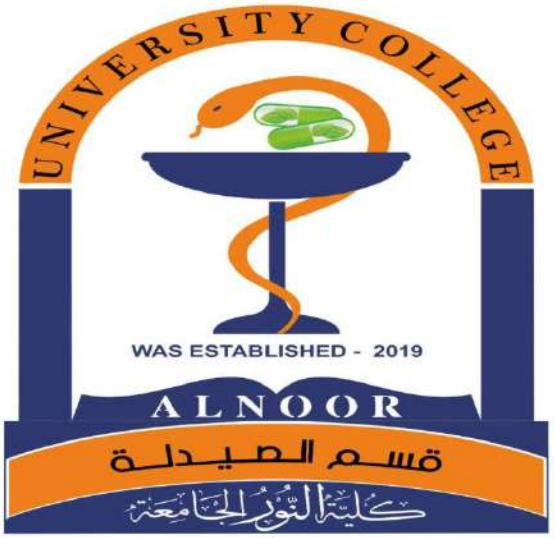




Organic Pharm. Chemistry II

Lecture 2

30th September 2023(8:30- 11:30)(11:30-2:30)



AlNoor University College

Pharmacy Department

1st Course/ 4th Class

2023/2024

Design of cholinergic agonists

Use of steric shields

Rationale

Shields protect ester from nucleophiles and enzymes

Shield size is important

Must be large enough to hinder hydrolysis

Must be small enough to fit binding site

Requirements

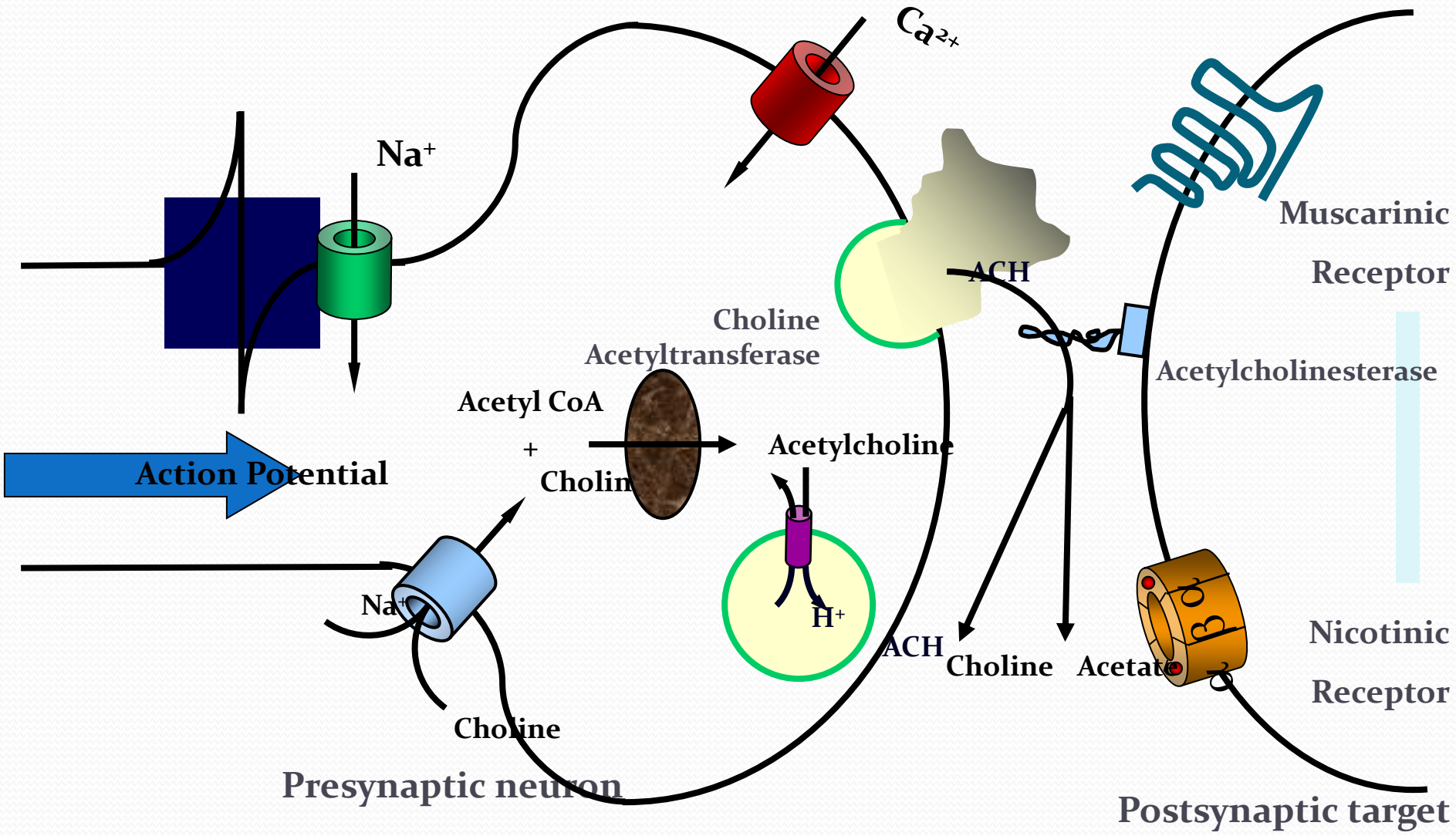
Correct size

Correct pharmacophore - ester and quaternary nitrogen

Increased stability to acid and esterases

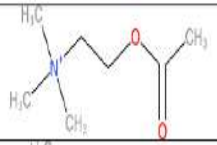
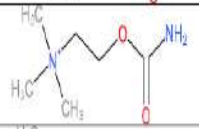
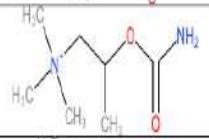
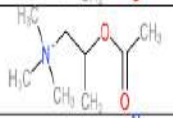
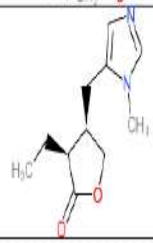
Increased selectivity

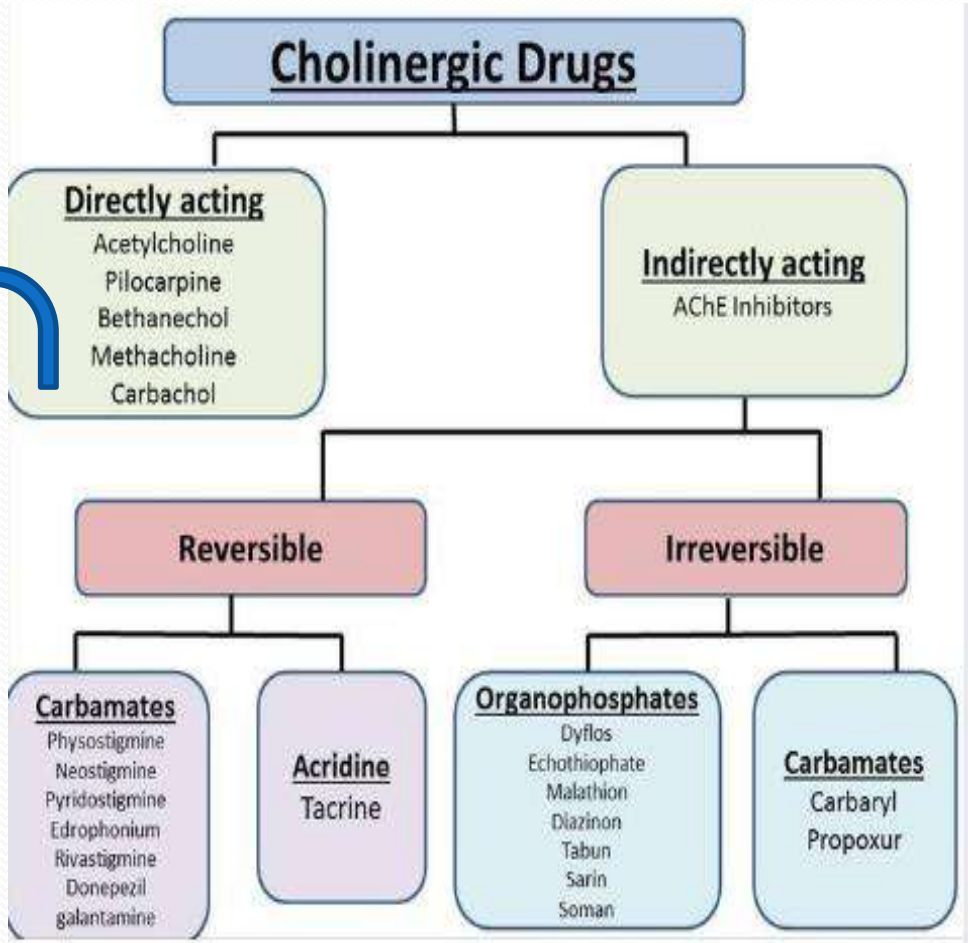
Pharmacologic manipulation of the cholinergic system



Cholinomimetics = parasympathomamitcs

- Direct Acting

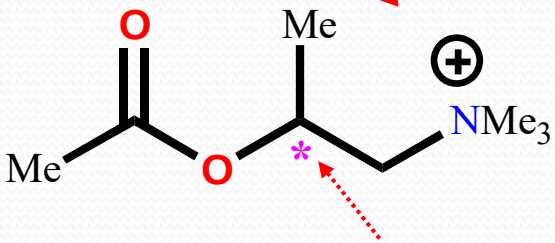
DRUG	STRUCTURE	CLASS
Acetylcholine		Choline esters
Carbachol		Choline esters
Bethanechol		Choline esters
Methacholine		Choline esters
Pilocarpine		Alkaloidal derivative



Design of cholinergic agonists (DIRECT)

hinders binding to esterases and provides a shield to nucleophilic attack

Methacholine



asymmetric centre

Properties

Three times more stable than acetylcholine

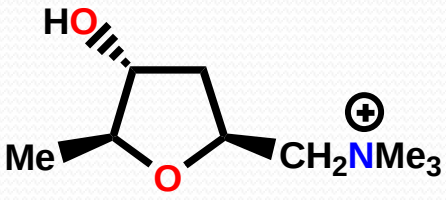
Increasing the shield size increases stability but decreases activity

Selective for muscarinic receptors over nicotinic receptors

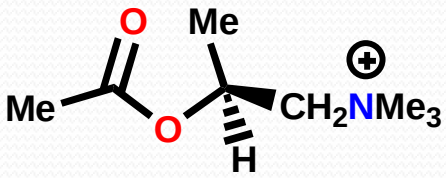
***S*-enantiomer is more active than the *R*-enantiomer**

Stereochemistry matches muscarine

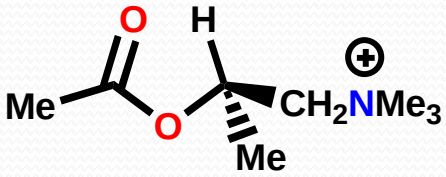
Not used clinically



MUSCARINE

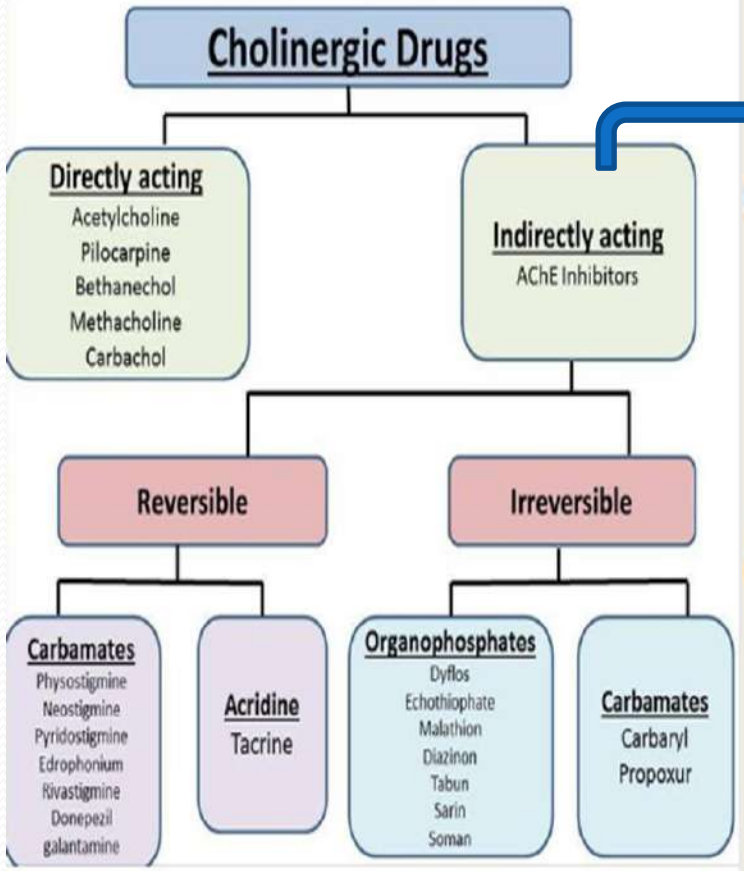


(*S*)



(*R*)

Indirect Cholinergic Drugs



Cholinergic Drugs – Indirect acting

- **Cholinesterase inhibitors** or **reversible anticholinesterases:**
  1. **Natural:** Physostigmine
  2. **Synthetic:** Neostigmine, Pyridostigmine, Distigmine, Rivastigmine, Donepezil, Gallantamine, Edrophonium, Ambenonium, Demecarium

1. Irreversible anticholinesterases:

1. **Organophosphorous Compounds (OPC)** – Diisopropyl fluorophosphate (DFP), Ecothiophate, Parathion, malathion, diazinon (insecticides and pesticides)
2. **Tabun, sarin, soman (nerve gases in war)**
3. **Carbamate Esters:** Carbaryl and Propoxur (Baygon)

ANTICHOLINESTERASES AGENTS

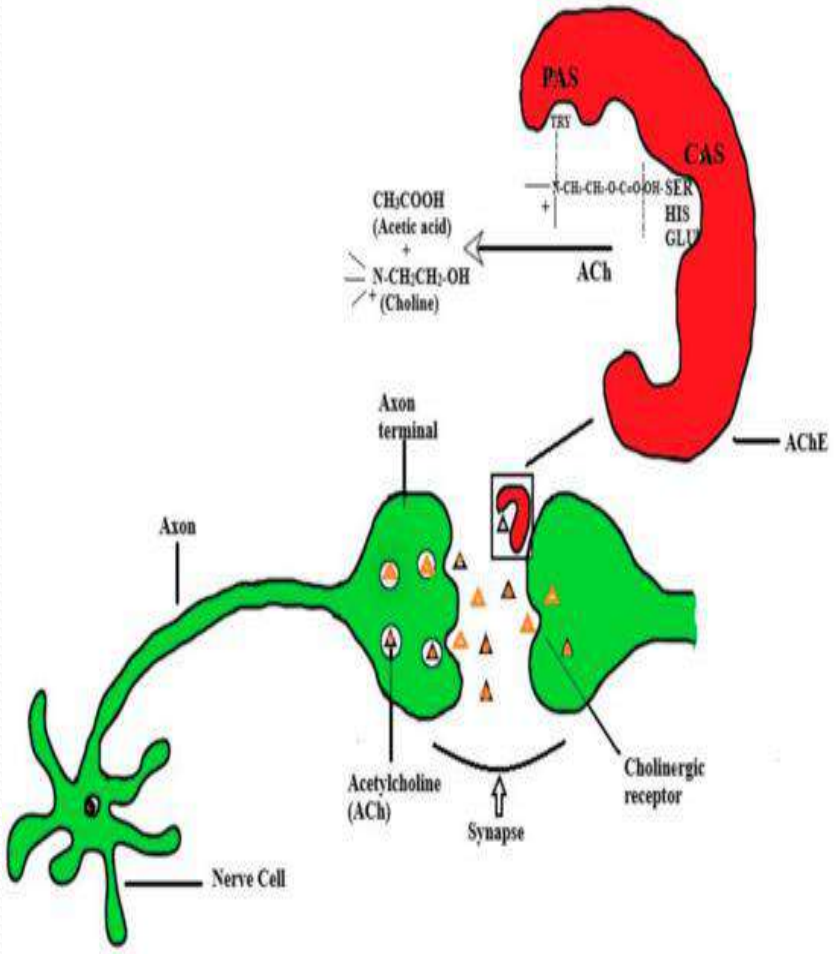
Reversible :

Short :
Edrophonium

Medium : Neostigmine, Physostigmine, Pyridostigmine, Tacrine

Cholinesterase inhibitors or reversible anticholinesterases:

1. **Natural:** Physostigmine
2. **Synthetic:** Neostigmine, Pyridostigmine, Distigmine, Rivastigmine, Donepezil, Gallantamine, Edrophonium, Ambenonium, Demecarium

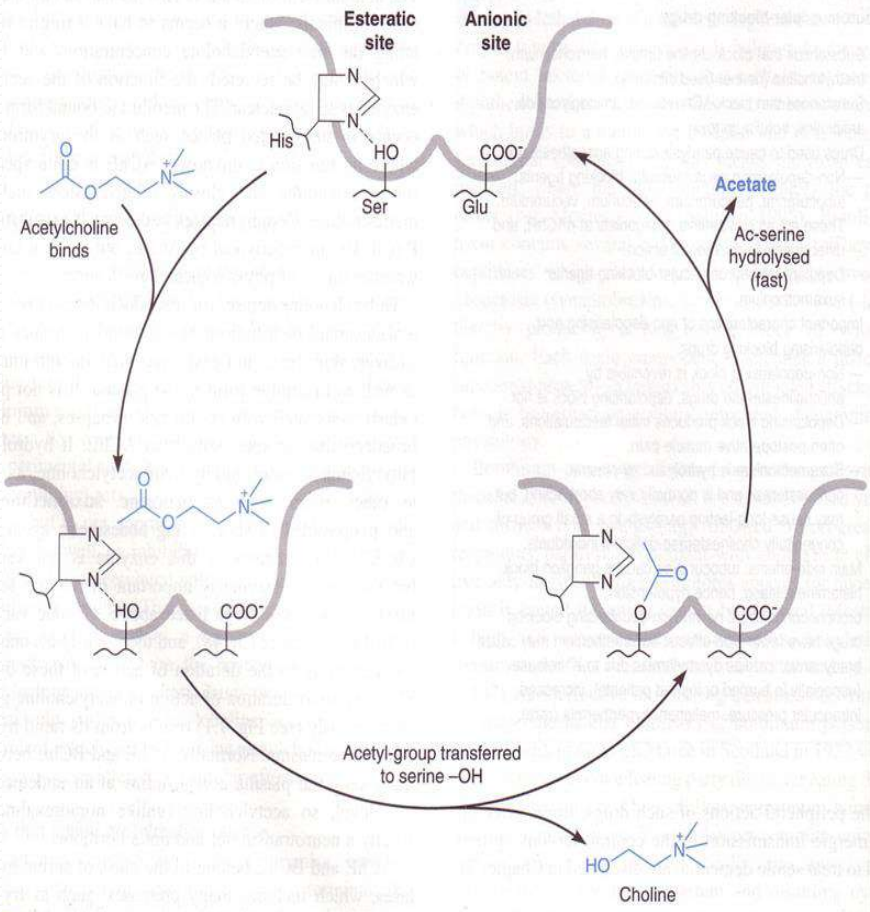


ANTI-CHOLINESTERASES AGENTS

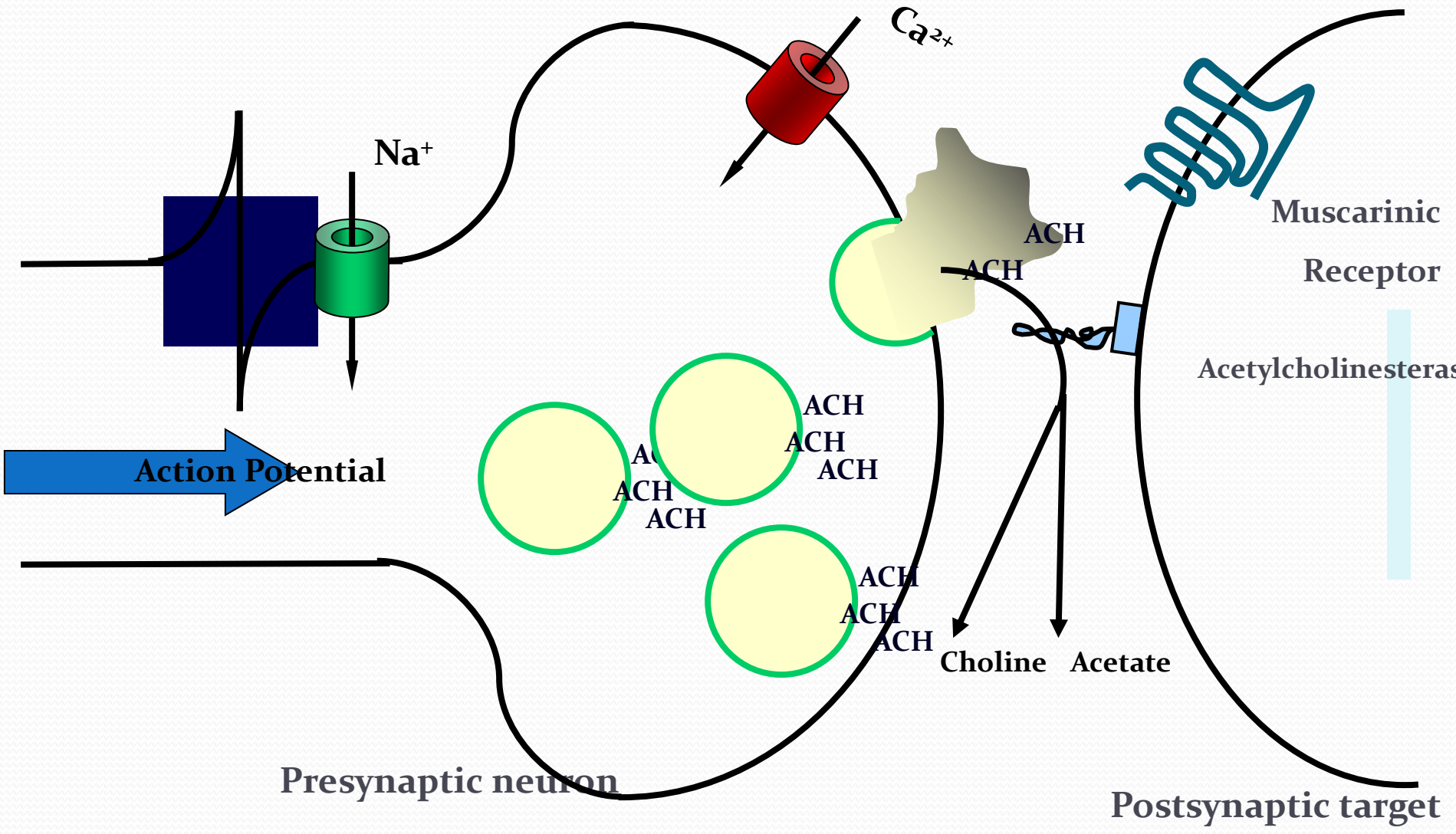
Mechanism of action :
Acetyl cholinesterase (AchE) is an enzyme with **anionic** and **esteratic site.**

Acetylcholine (Ach) involves attraction of the positive charge N+ of Ach and anionic site; acetylation of serine leading to the acetylated enzyme.

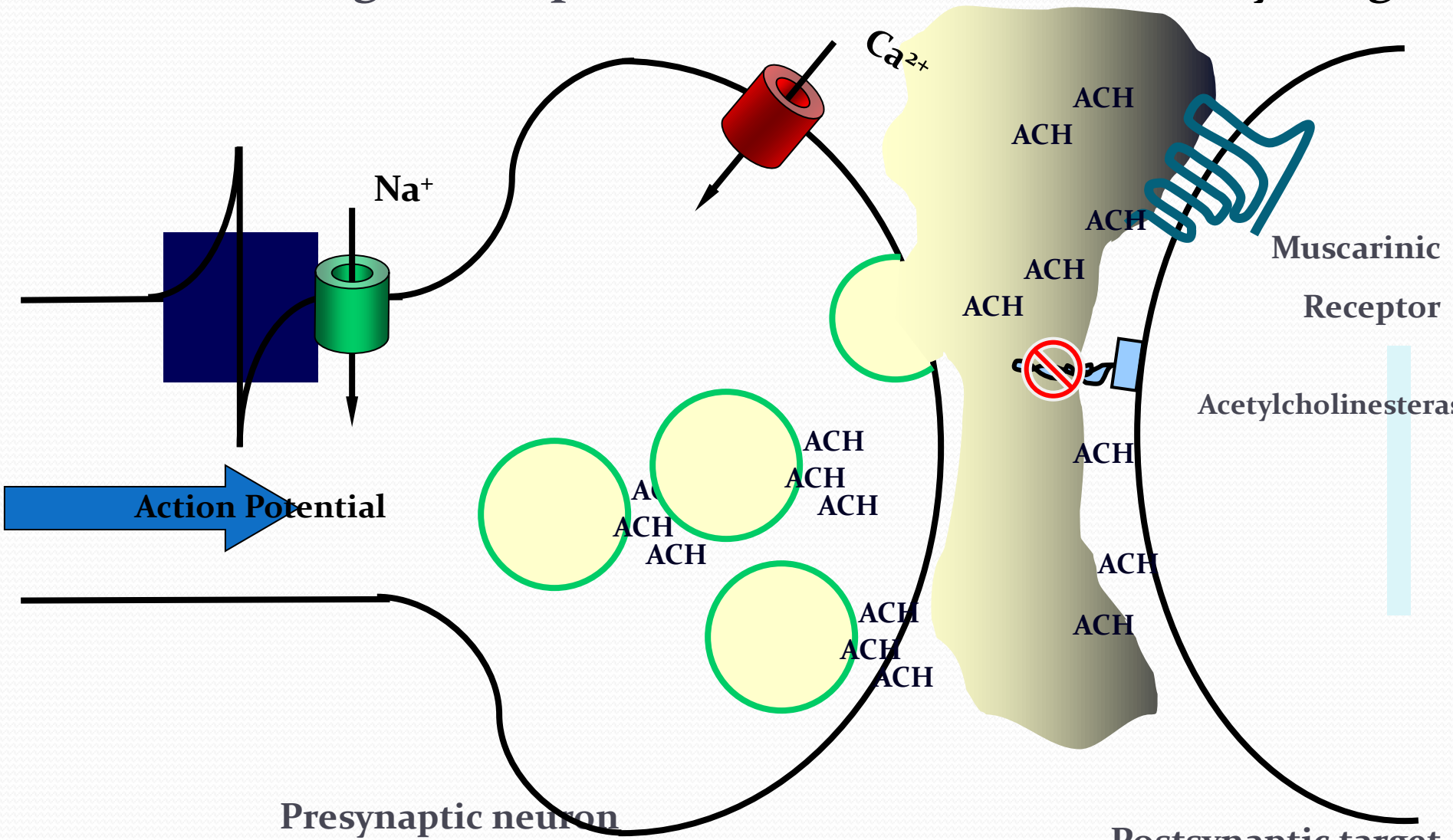
The acetylated enzyme reacts with the water to produce acetic acid and free enzyme within milliseconds.



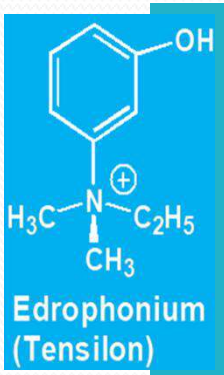
Pharmacologic manipulation of AChE: **No inhibition**



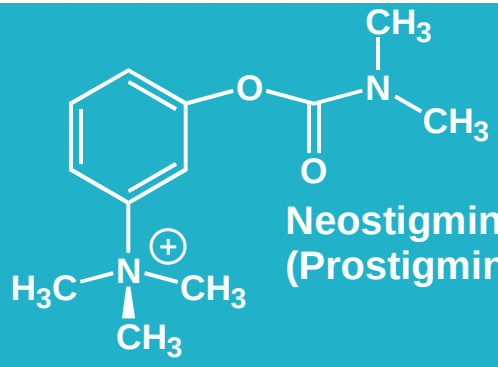
Pharmacologic manipulation of AChE: **Inhibition by drugs**



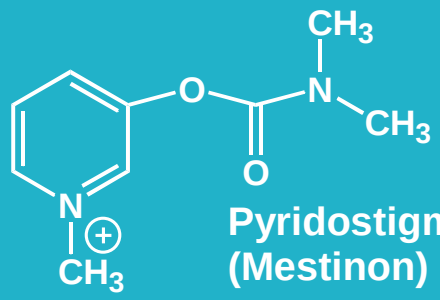
Acetylcholinesterase inhibitors



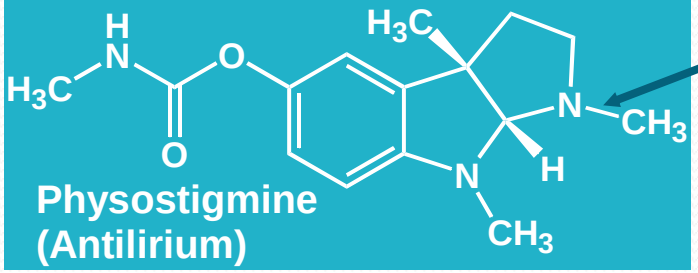
Edrophonium (Tensilon)



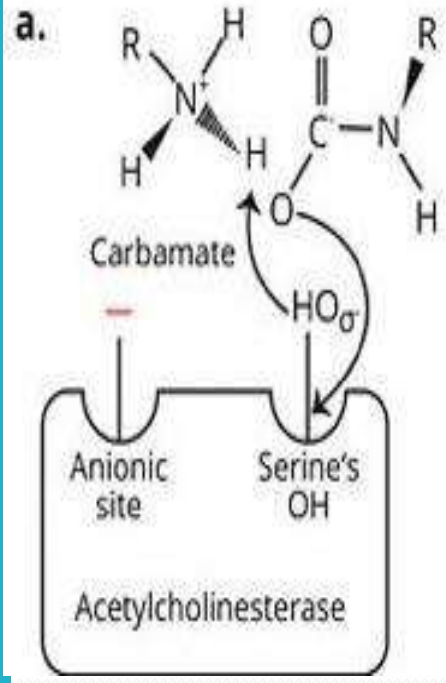
Neostigmine (Prostigmin)

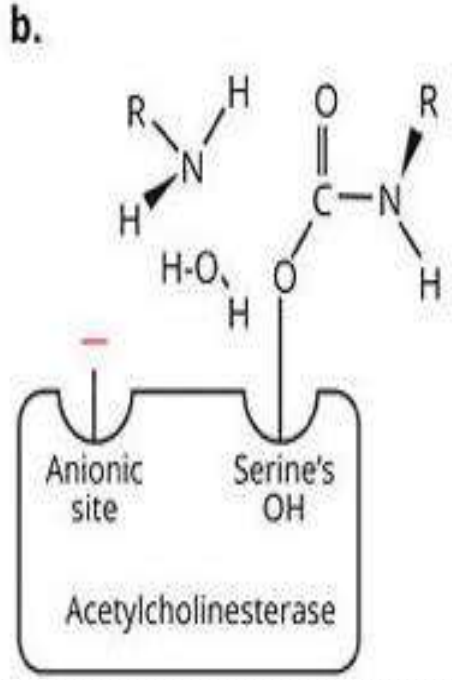


Pyridostigmine (Mestinon)



Physostigmine (Antilirium)



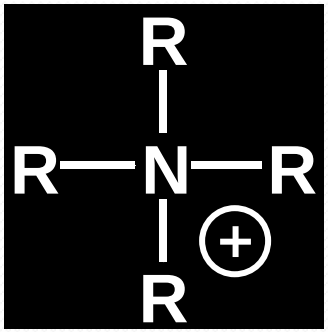


Most basic Nitrogen; protonated at physiological pH.

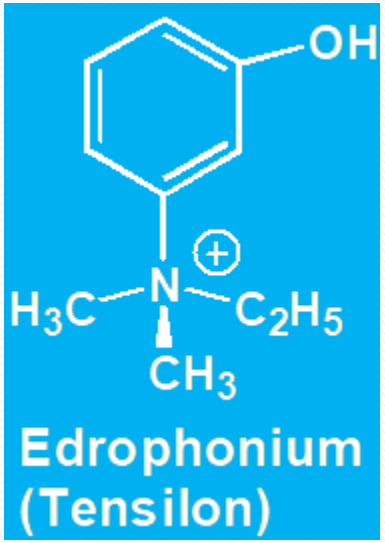
Edrophonium, Neostigmine and Physostigmine

They combine with the ChE and carbamylated enzyme is slow to hydrolyze and free the enzyme (~30 mins).

Acetylcholinesterase inhibitors

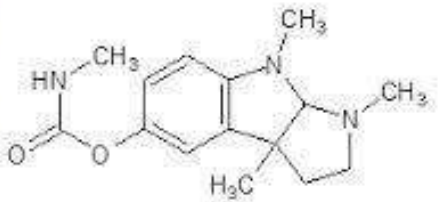


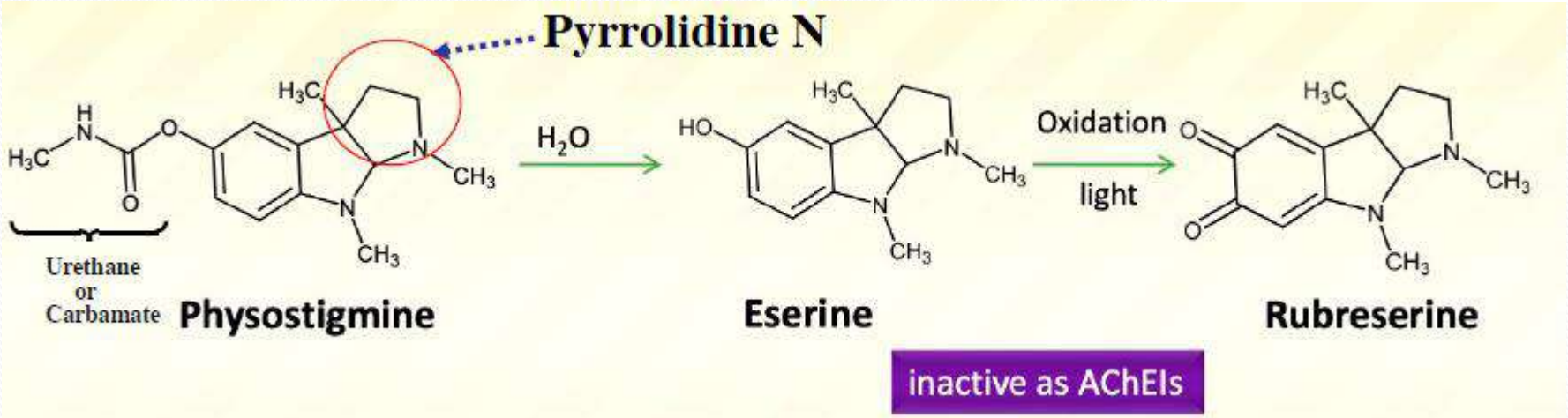
R	Relative Potency
CH_3	1.0
C_2H_5	5.0
C_3H_7	100
C_4H_9	50



- Quaternary ammonium alcohol
- Simplest structures
- Bind to anionic site and block ACh binding
- Reversible(Non-covalent)

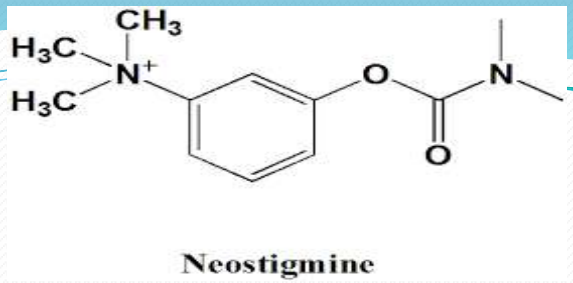
Physostigmine



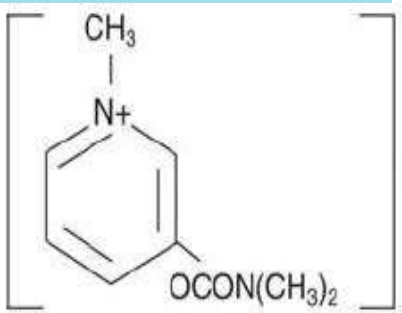


Acetylcholinesterase is carbamylated at a slow rate and the carbamylated AChE also is regenerated quite slowly
Because physostigmine is a tertiary amine rather than a quaternary ammonium salt, it is more lipophilic than other AChEIs and can diffuse across the blood-brain barrier.
It is investigated for use in the treatment of Alzheimer's disease.

- **Sensitive to heat , light, moisture etc.**
- **Fully ionised (can't cross BBB)**
- **More stable to hydrolysis (extra N- methyl group increase stability)**
- **Antidote for Atropine poisoning**
- **Topical application of glaucoma**



Neostigmine



Pyridostigmine

Neostigmine

- Chemically more stable than Neostigmine .
- Longer DoA
- Neostigmine administered orally or IV
- Prevent atony of intestinal , skeletal
- And bladder musculature , also as urinary stimulant .
- Mostly used for treatment of MG

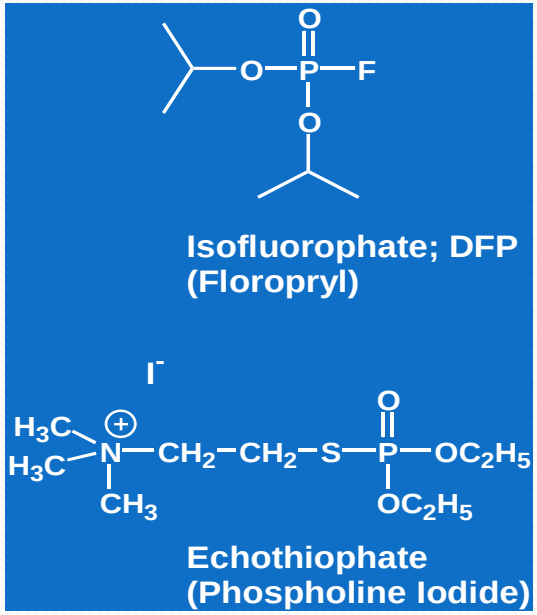
Carbaryl
Widely used as insecticidal used on house plants and vegetables as well as for fleas and ticks on pets .

Pyridostigmine

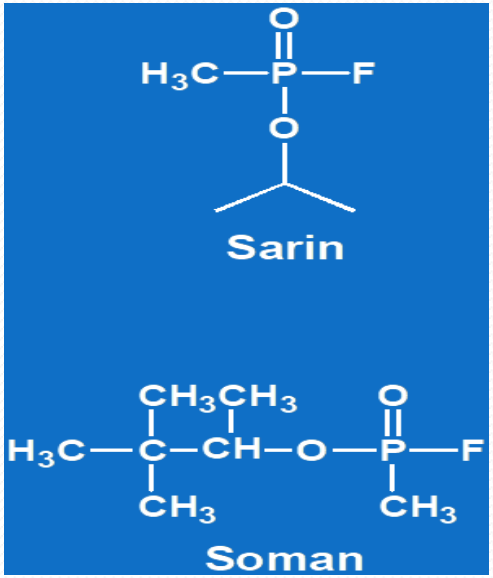
Myasthenia Gravis

- Pyridostigmine is a medication that treats myasthenia gravis (MG).
- Pyridostigmine (Mestinon) is slightly longer-acting (with a half-life of 4 hours) and has fewer cholinergic side effects than neostigmine bromide and other anticholinesterase preparations. Unlike physostigmine, pyridostigmine has no unwanted CNS effects because it does not cross the blood-brain barrier.
- HW??
- Compare among them ?

Acetylcholinesterase inhibitors

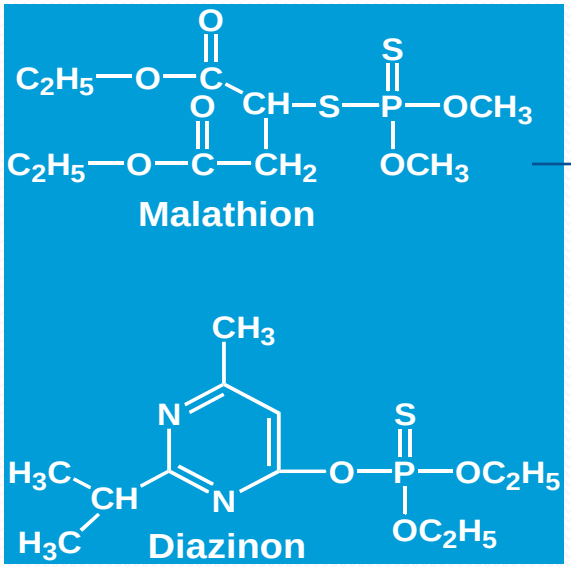


- Organophosphates
- Irreversible
- Covalent modification to AChE
- Longer acting
- Used in the treatment of glaucoma



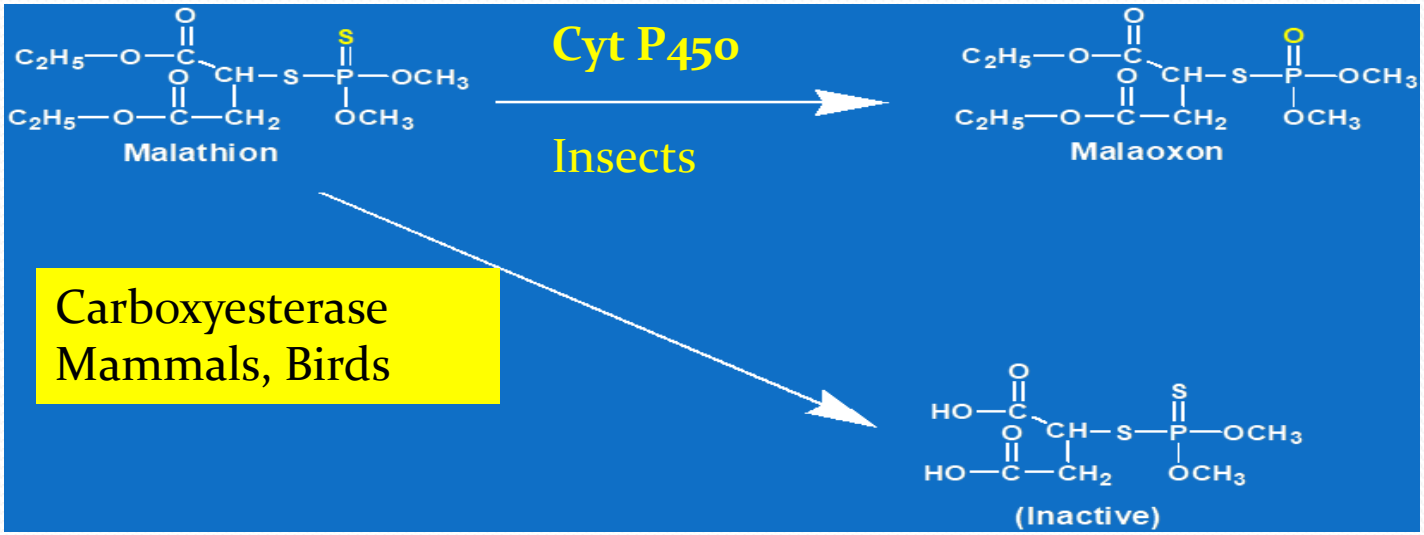
- Organophosphates
- Nerve gases
- Irreversible
- Covalent modification to AChE

Acetylcholinesterase inhibitors

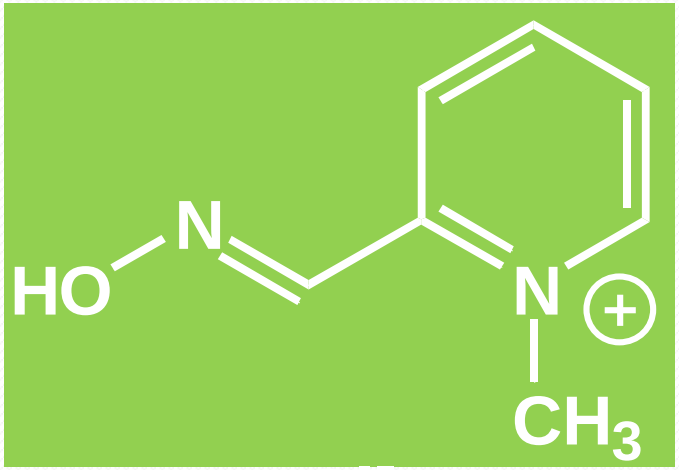


- Organophosphates
- Insecticides
- Irreversible
- Covalent modification to AChE
- Rapidly inactivated in mammals

Biotransformation of Insecticides



Antidote for AChE “poisoning”

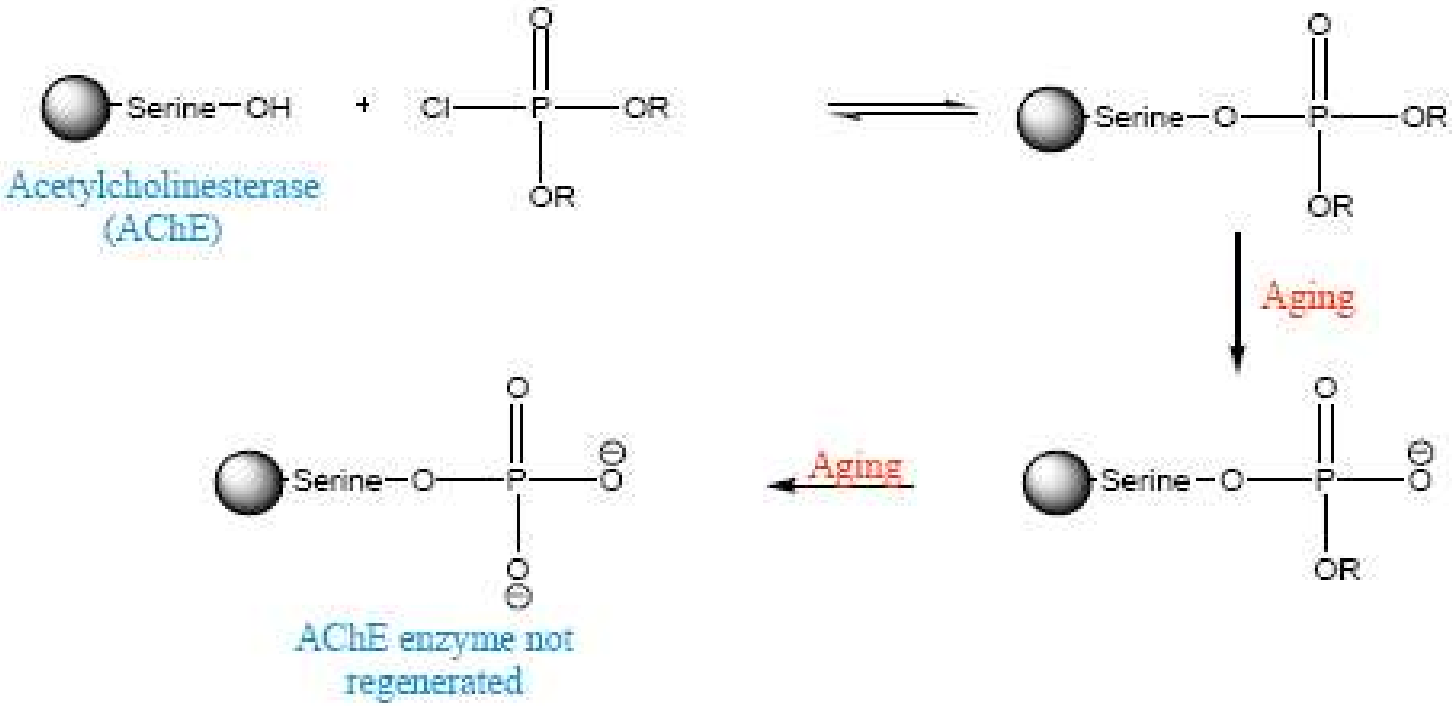


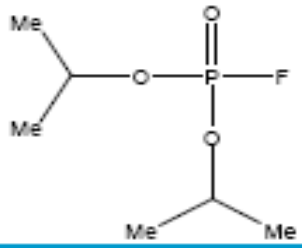
- Pralidoxime Chloride (Protopam; 2-pyridine aldoxime methyl chloride; 2-PAM)
- Antidote for pesticide or nerve gas poisoning
- Most effective if given within a few hours of exposure

Irreversible Inhibitors

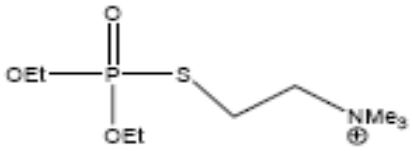
Compounds containing phosphoryl or phosphonic halides that can react with AChE to form AChE-phosphate complexes stable to hydrolytic cleavage.

Mainly used as agricultural insecticides and nerve gas agents.



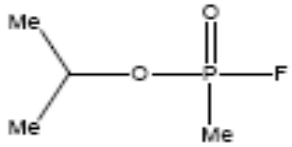


Diisopropylfluorophosphate

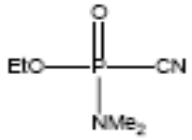


Ecothiophosphate

- Inhibits AChE irreversibly
- Long lasting inhibition (up to ~ 4 weeks)
- Applied topically to the eye in glaucoma treatment



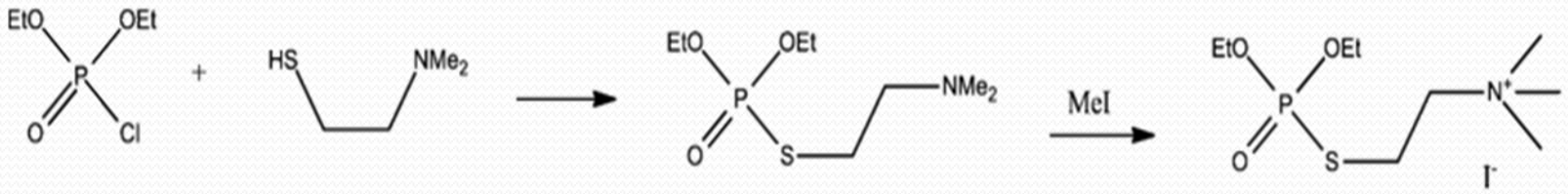
Isopropylmethylphosphono-fluoridate (Sarin®)

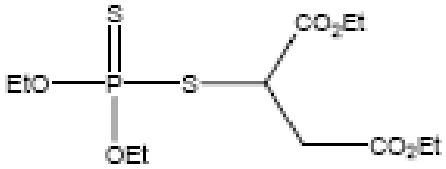


Ethyl-*N*-dimethylphosphor-amide cyanidate (Tabun®)

- Highly potent and irreversible AChE inhibitors
- Extremely toxic (nerve gas)

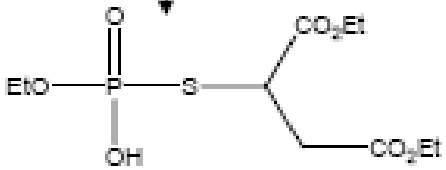
Synthesis of Ecothiophosphate



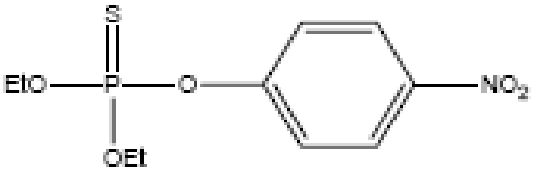


Malathion
(Weak AChE inhibitor)

Microsomal oxidation



Malaoxon
(10,000 times more active AChE inhibitor)



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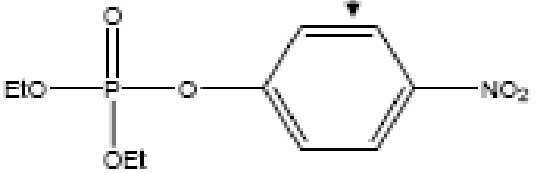
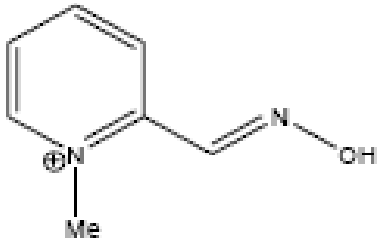


Paraoxon
(High AChE inhibitory activity)

- Bioactivated by microsomal oxidation
- Main use as agricultural insecticide. Malathion is also used in lice infestation.
- Highly poisonous. Pralidoxime (2-PAM) is a mechanism based antidote for poisoning

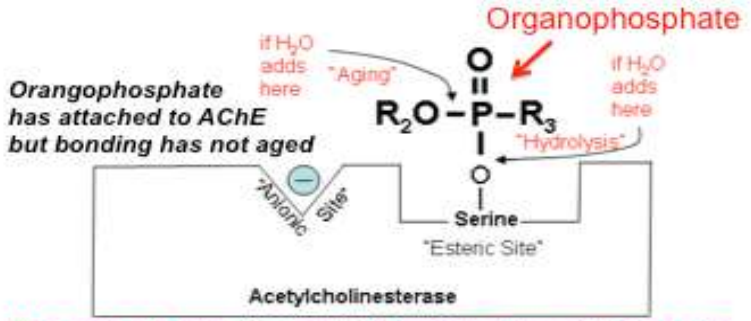


Pralidoxime (2-PAM)

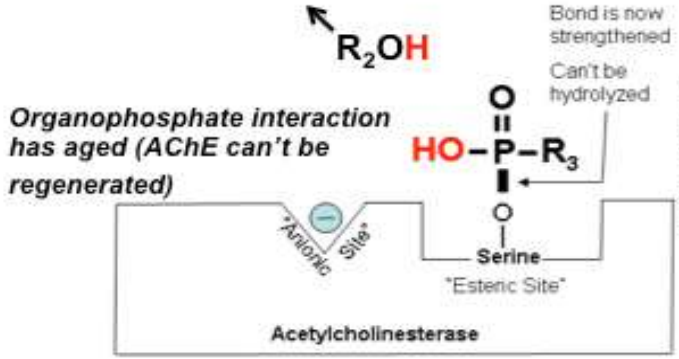
- Effective antidote for poisoning by parathion and related pesticides
- Most effective by intramuscular, intravenous, or subcutaneous administration
- Treatment is effective if initiated within few hours
  - Pralidoxime Chloride (Protopam; 2-pyridine aldoxime methyl chloride; 2-PAM)
  - Antidote for pesticide or nerve gas poisoning
  - Most effective if given within a few hours of exposure

- The phosphorylated ChE reacts very slowly or not at all with the water.
- If more OH groups in the form of Oximes are provided, reactivation occurs faster.
- Pralidoxime (PAM) attaches to the **anionic site** in presence of Organophosphorus compounds and set the enzyme free.
- PAM is ineffective in case of physostigmine poisoning as anionic site is not free.

Organophosphate Aging – chemical stabilization of phosphate bond to AChE occurs over time

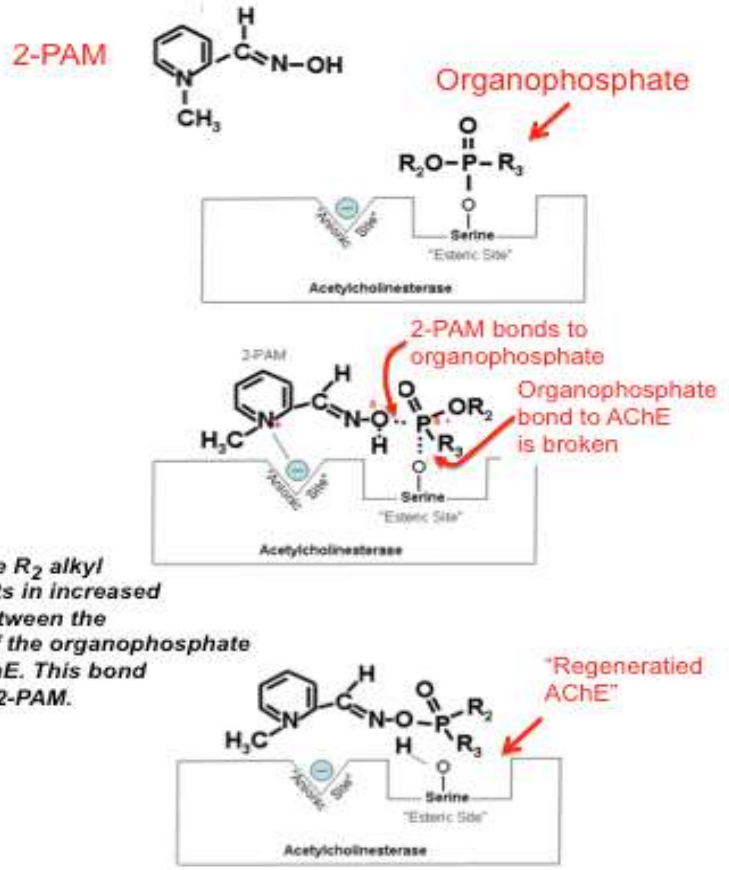


The rate of aging is unique for each organophosphate compound, and can occur over minutes to days depending on the agent



The departure of the R_2 alkyl group (aging) results in increased electron sharing between the phosphate group of the organophosphate & the serine on AChE. This bond can't be broken by 2-PAM.

Pralidoxime (2-PAM) prevents aging & regenerates AChE



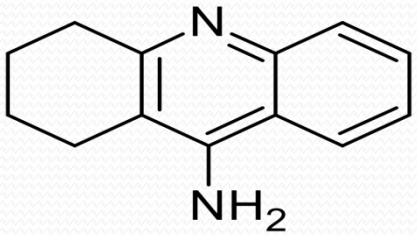
Treatment of Alzheimer's Disease

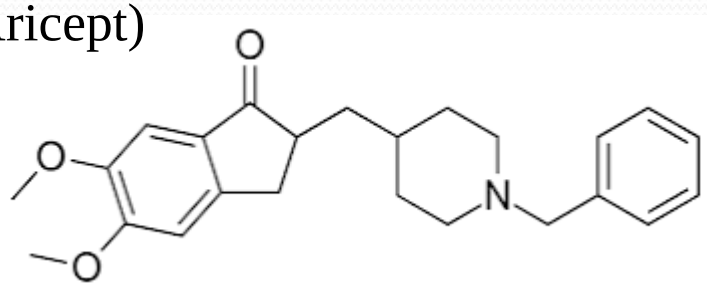
Donzepil (Aricept)

- Bind to anionic site and block ACh binding
- Reversible
- Non-covalent
- Enhances cognitive ability
- Does not slow progression of disease
- Newer agent: Donepezil (Aricept)

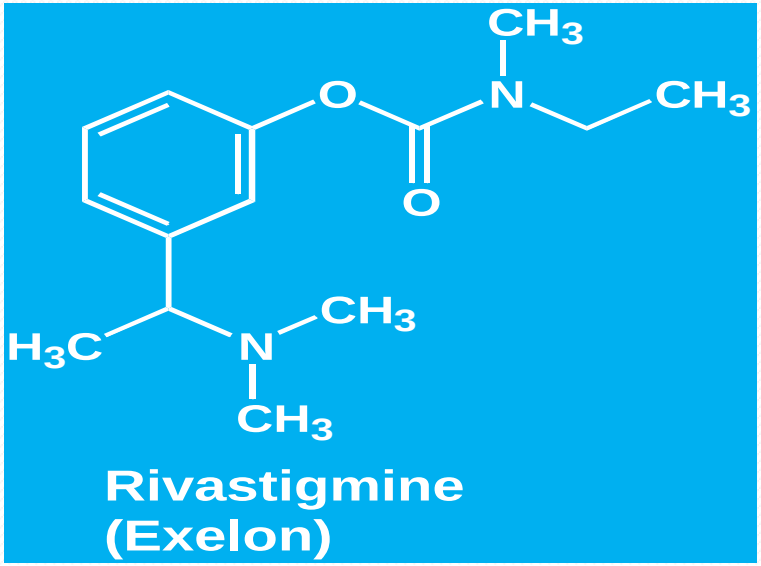
Tacrine (Cognex)

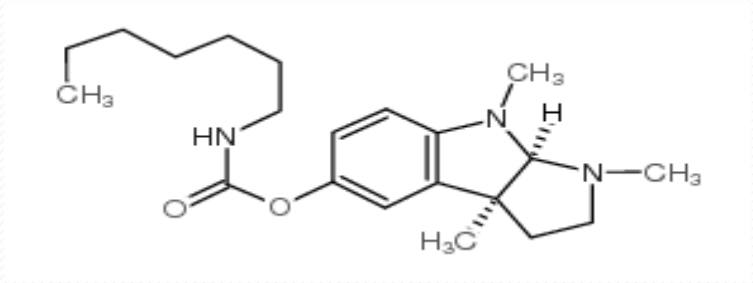
nowadays there are not specific studies justifying why the protonation of the cationic species takes place on the central ring of the N atom while the Cl atom is bonded to the N of NH_2 in the HCl species instead …





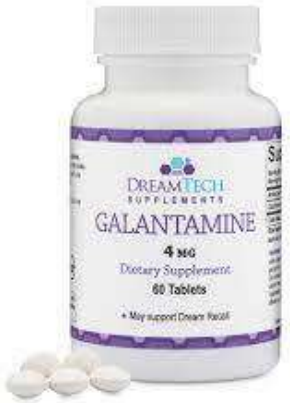
Treatment of Alzheimer's Disease

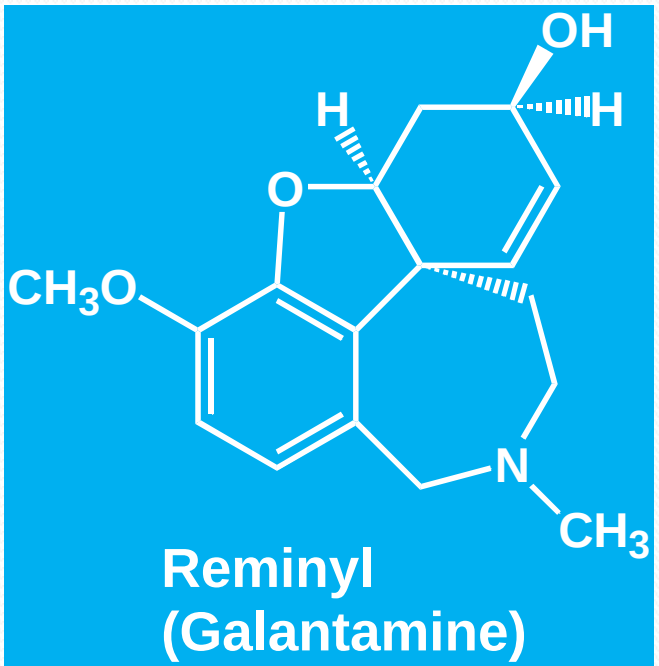




- Reversible carbamate AChE inhibitor
- Enhances cognitive ability by increasing cholinergic function
- Loses effectiveness as disease progresses
- Side Effects: Nausea, vomiting, anorexia, and weight loss
- Newer long-acting carbamate: Eptastigmine

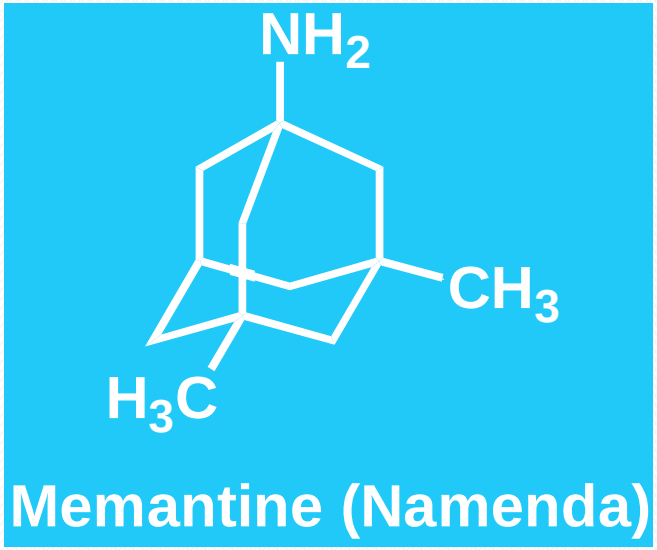
Treatment of Alzheimer's Disease





- **Reversible competitive AChE inhibitor**
- **Extract from daffodil (Narcissus pseudonarcissus) bulbs**
- **Loses effectiveness as disease progresses**
- **May be a nicotinic receptor agonist**
- **Inhibitors of P450 enzymes (3A4, 2D6) will increase galantamine bioavailability**

Treatment of Alzheimer's Disease



- ***N*-methyl-D-aspartate (NMDA) receptor antagonist**
- **NMDA receptors are activated by glutamate in the CNS in areas associated with cognition and memory**
- **Neuronal loss in Alzheimer's may be related to increased activity of glutamate**
- **May slow progression of the disease**
- **Favorable adverse effect profile**

Memantine addresses dysfunction in glutamatergic transmission, while the AChEIs serve to increase pathologically lowered levels of the neurotransmitter acetylcholine.

Cholinergic Antagonists

Lecture Outlines

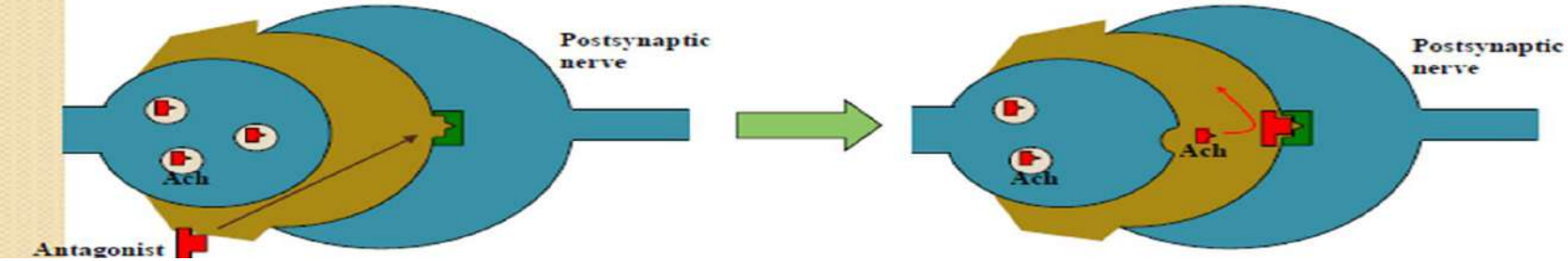
- Introduction
- Muscarinic antagonists
  - Therapeutic application
  - Structure Activity Relationship (SAR) studies
  - Specific muscarinic antagonists
- Nicotinic antagonists
  - Introduction
  - Discovery
  - Different classes of nicotinic antagonists

Learning Objectives

After completion of this topic students should be able to

1. Identify the structures and discuss therapeutic uses of various classes muscarinic and nicotinic antagonists
2. Identify acetylcholine binding residues at the muscarinic receptor
3. Discuss structure activity/pharmacokinetic relationships of antimuscarinics including
   1. Substitution at alpha carbon with respect to ester group
   2. Substitution on the nitrogen
4. Identify chemical class of any muscarinic antagonist
5. Identify the metabolites (active/inactive) of specific antimuscarinics
6. Identify the structural requirements of a compound to be a neuromuscular blocker
7. Discuss how nicotinic antagonists bind at the receptor site
8. Compare and contrast depolarizing and non-depolarizing neuromuscular blockers
9. Identify the metabolites of (active and inactive) of steroid and tetrahydro isoquinoline type neuromuscular blockers

- Drugs which bind to cholinergic receptor but do not activate it
- Prevent acetylcholine from binding
- Opposite clinical effect to agonists - lower activity of acetylcholine



Muscarinic antagonists are commonly referred to as

- Anticholinergics
- Antimuscarinics
- Cholinergic blockers
- Antispasmodics
- Parasypatholytics

Clinical Effects

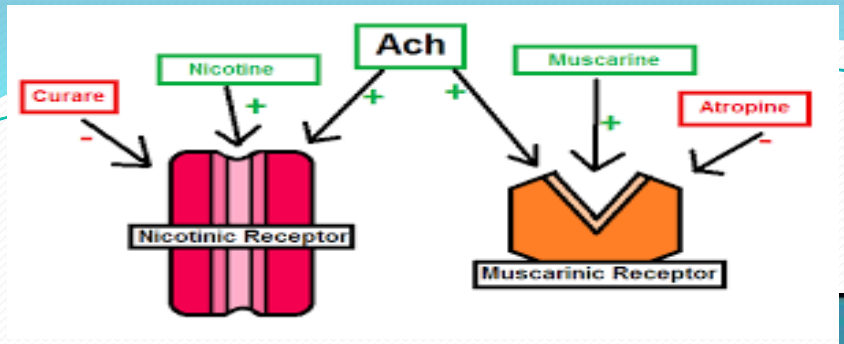
- Decrease of saliva and gastric secretions
- Relaxation of smooth muscle
- Decrease in motility of GIT and urinary tract
- Dilation of pupils

Uses

- Shutting down digestion for surgery
- Ophthalmic examinations
- Relief of peptic ulcers
- Treatment of Parkinson's Disease
- Anticholinesterase poisoning
- Motion sickness

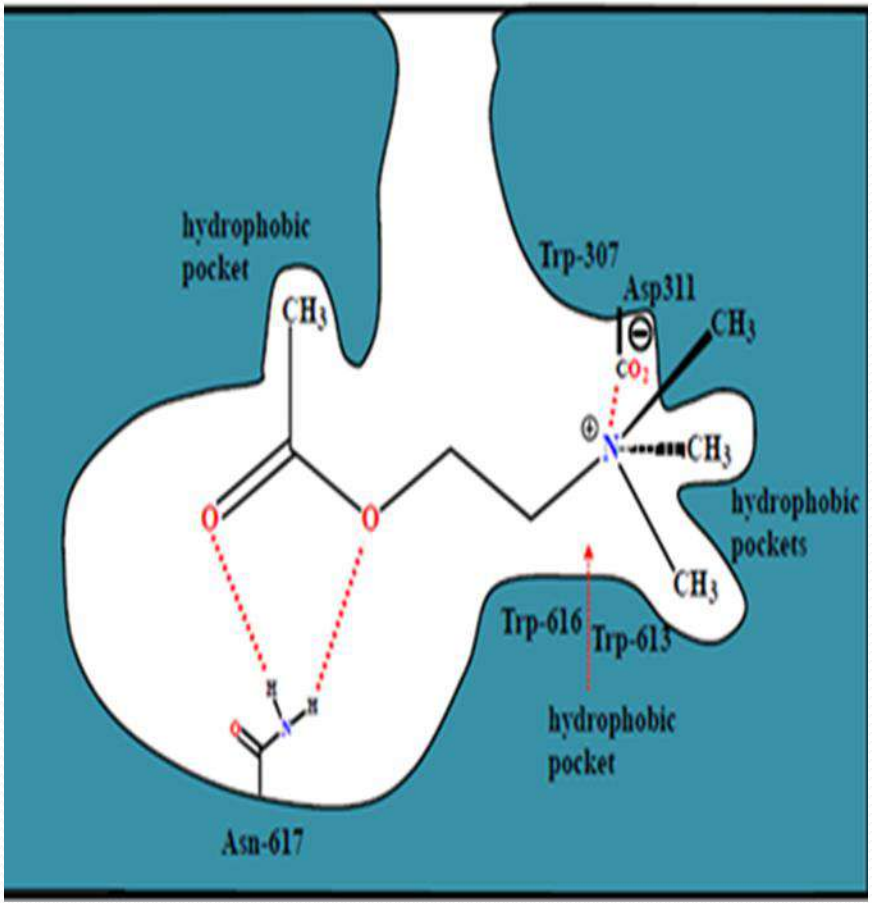
What are the therapeutic applications of antimuscarinics?

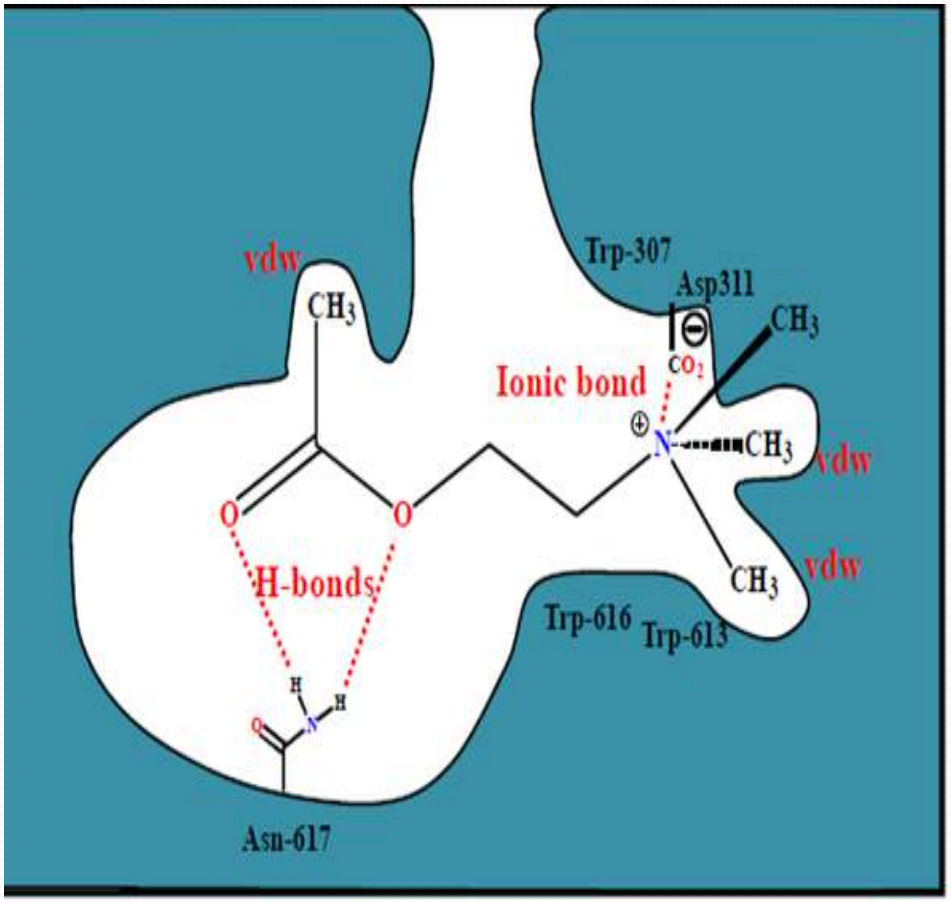
Organ	M-Receptor Effect	Antimuscarinic effect	Therapeutic indication
Eye			
Iris circular muscle	Contracts	Relaxes	
Ciliary muscle	Contracts	Relaxes	Mydriatic
Heart			
Sinoatrial node	Decelerates	Accelerates	Bradycardia
Atrial contractility	Decelerates	Accelerates	Bradycardia
Bronchiole smooth muscle	Contracts	Relaxes	Asthma Allergic rhinitis
Gastrointestinal tract			
Smooth muscle	Contracts	Relaxes	GI antispasmodic
Secretions	Increases	Decreases	
Sphincters	Relaxes	Contracts	
Bladder smooth muscles	Contracts	Relaxes	Overactive bladder



Muscurinic Antagonists

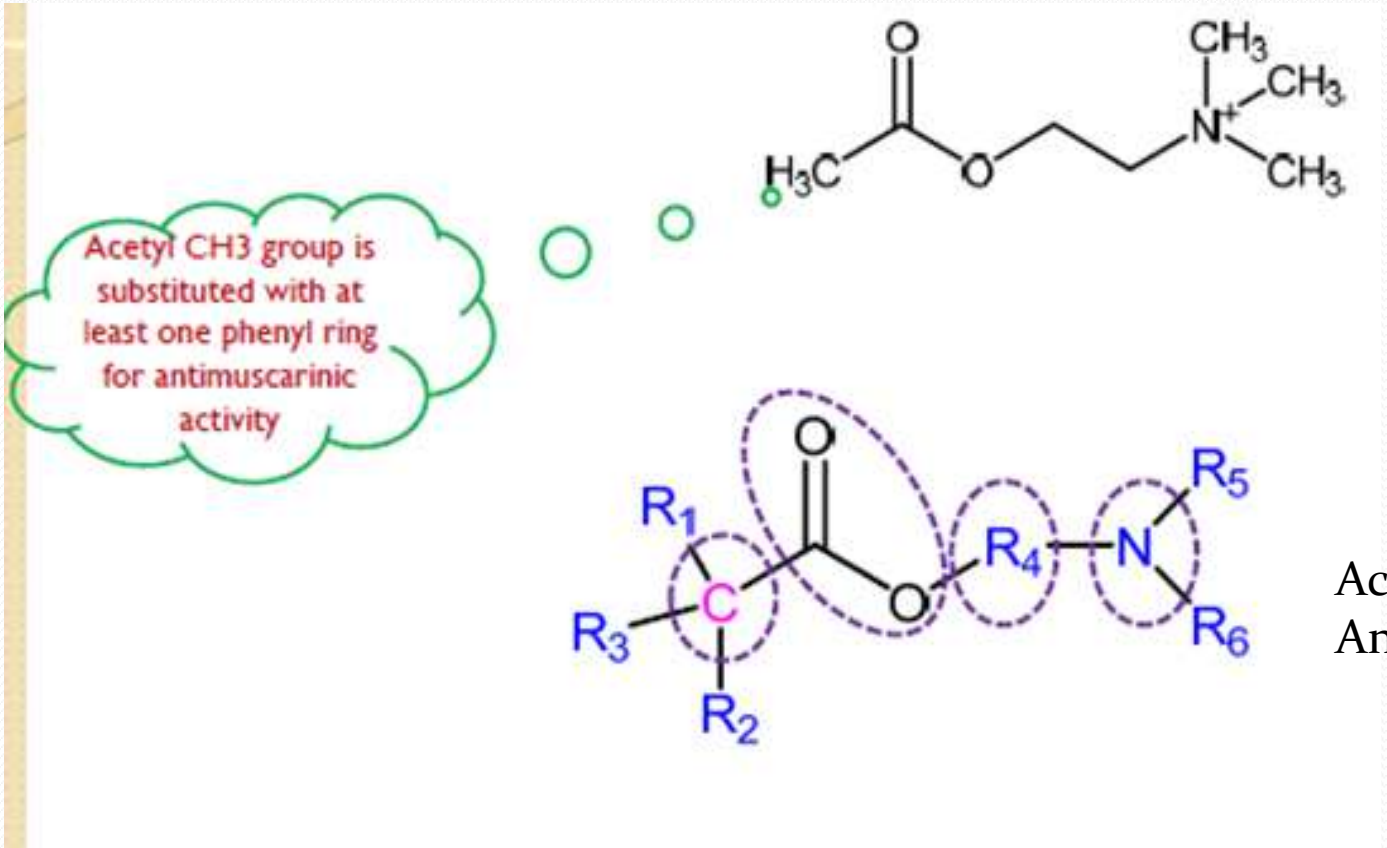
Binding Site (Muscurinic)





It is postulated that muscarinic antagonists bind to the Asp and contain hydrophobic substituents that bind to a hydrophobic pocket in the receptor, which does not allow the change in conformation needed to transfer the agonist signal to the coupled G protein

Structural Activity Relationship



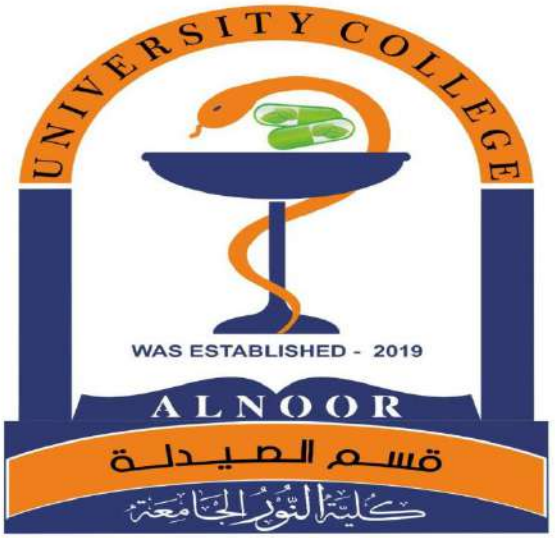
Ach

Ach Muscurinic Antagonists

Organic Pharm. Chemistry II

Lecture 3

7th October 2023(8:30- 11:30)(11:30-2:30)



AlNoor University College

Pharmacy Department

1st Course/ 4th Class

2023/2024

Anticholinergic

- Introduction
- Muscarinic antagonists
  - Therapeutic application
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What is muscarinic antagonist

- An agent that have high binding affinity for the muscarinic receptor but have no intrinsic activity.

What are they competing with to occupy muscarinic receptor?

- Acetylcholine
- They are competitive (reversible) antagonists of acetylcholine
- Their pharmacological actions are opposite to that of the muscarinic agonists

Muscarinic Antagonists

- Drugs which bind to cholinergic receptor but do not activate it
- Prevent acetylcholine from binding
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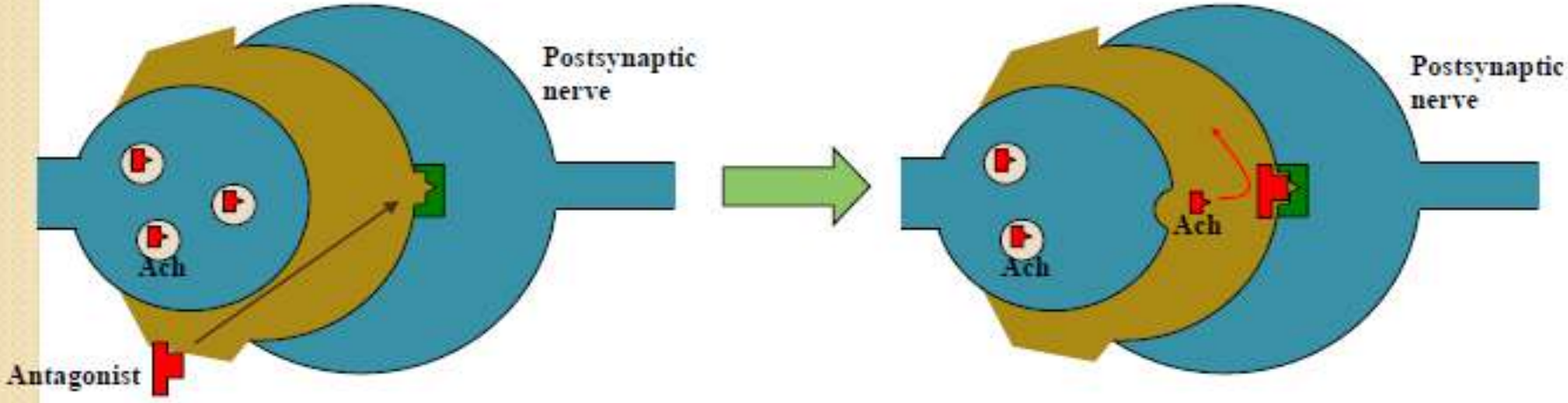

Muscarinic Antagonists

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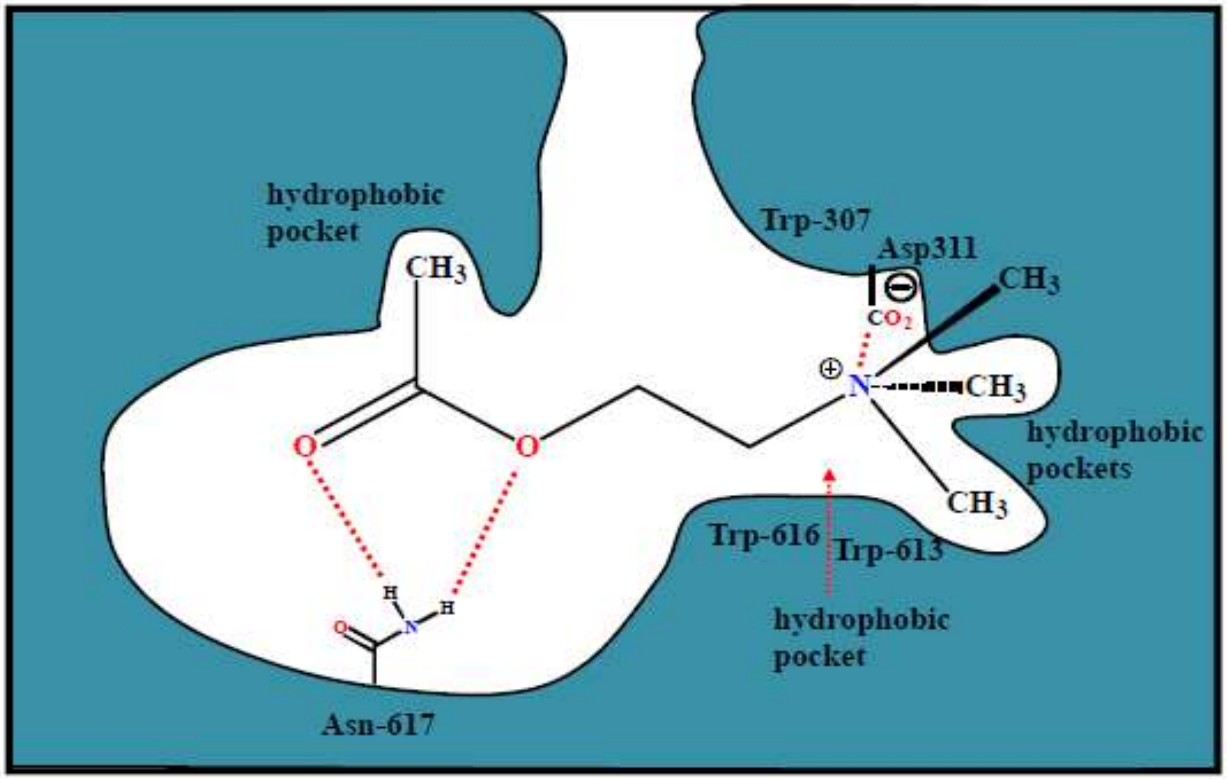
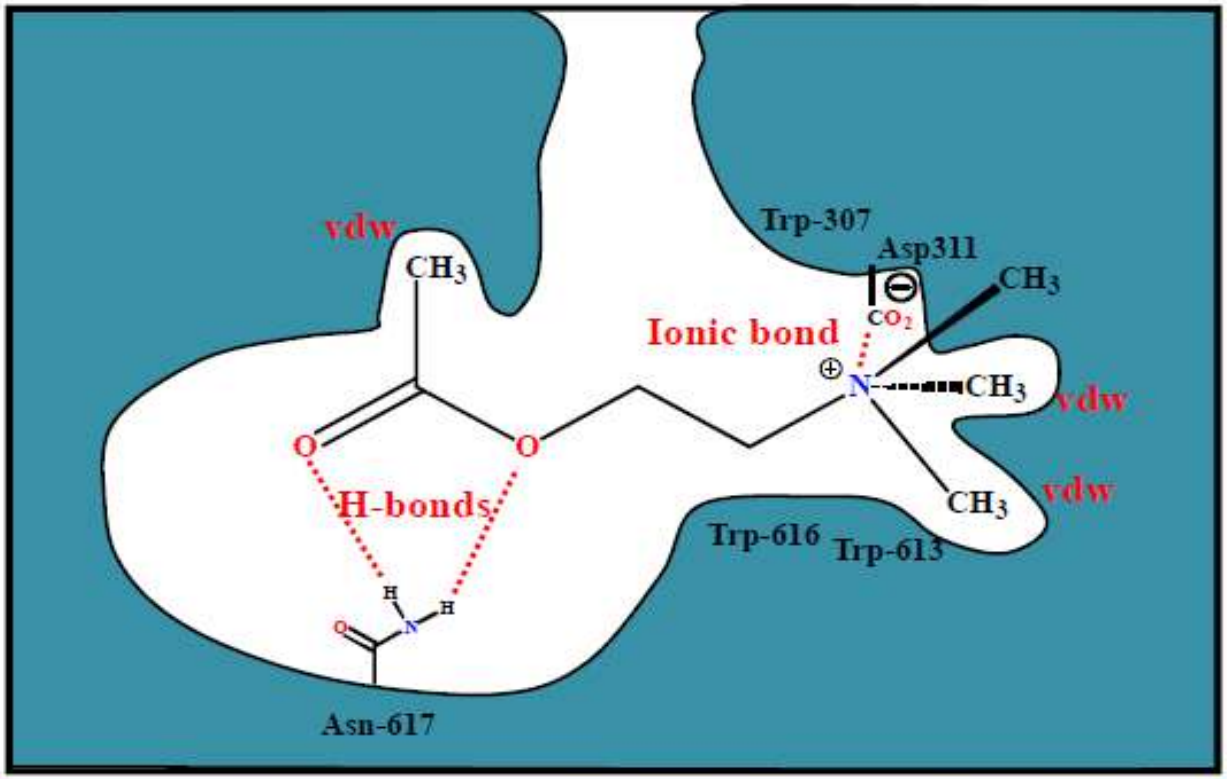
Binding site (muscarinic)



Binding site (muscarinic)



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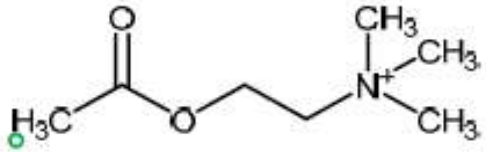
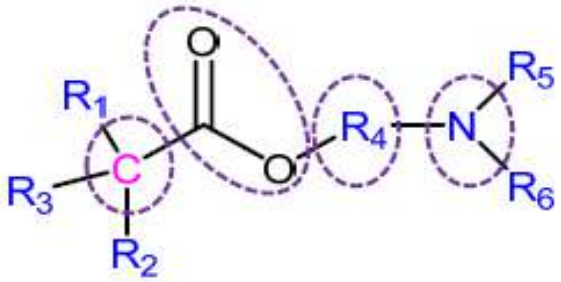
Structure Activity Relationship (SAR) Studies



Acetylcholine (ACH)

Acetyl CH3 group is substituted with at least one phenyl ring for antimuscarinic activity



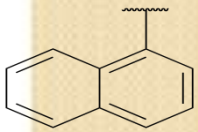
Acetylcholine analogue antimuscarinics

Substitutions at α-carbon with respect to ester group

1. May be a hydrogen atom, a hydroxyl group, a hydroxymethyl group, or a carboxamide
2. Hydroxyl group or a hydroxymethyl group, the antagonist usually is more potent

OH-, OCH3-

R_2 and R_3 should be carbocyclic or heterocyclic rings (phenyl, cyclohexyl, cyclopentyl) for maximal antagonist potency



Substitution of naphthalene rings at R_2 and R_3 affords inactive compounds, because of steric hindrance at the muscarinic receptor.

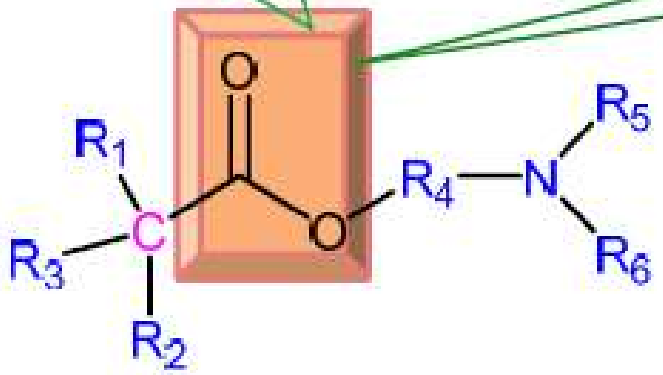
Bigger R_2 and R_3 groups bind to the hydrophobic region outside the Ach receptor site

The hydroxyl group at R_1 presumably increases binding strength by participating in a hydrogen bond interaction at the receptor.

Changes at ester group

This substituent may also be an ether oxygen, or it may be absent completely.

Ester group provides most potent anticholinergic activity

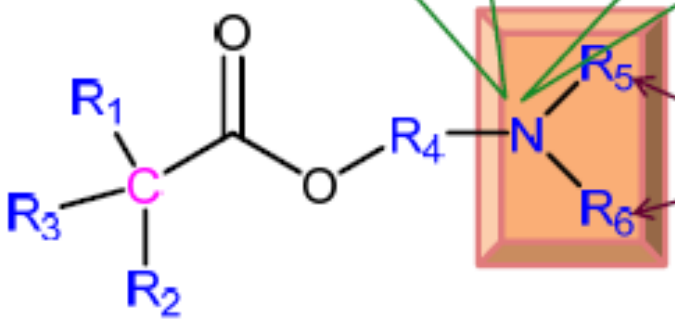


What about cholinomimetic Please compare??

Substitution at the amine group

Compounds possessing the quaternary ammonium group exhibit nicotinic antagonist activity at high doses.

Quaternary ammonium compounds possess most potent anticholinergic activity



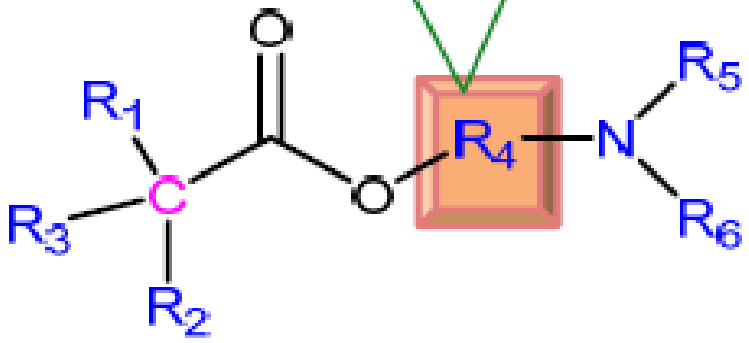
Methyl, ethyl, propyl, or isopropyl groups are tolerated

Tertiary amines also possess antagonist activity, presumably by binding to the receptor in the protonated form.

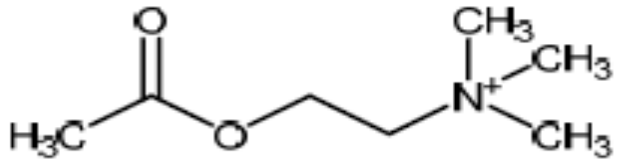
Quaternary ammonium drugs are primarily used in the treatment of ulcers or other conditions for which a reduction in gastric secretions and reduced motility of the gastrointestinal tract are desired

Changes at R_4 position

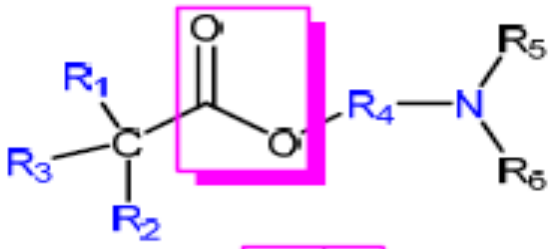
Optimum chain length is 2-4 carbons. Two carbon chain possess the most antagonistic activity.



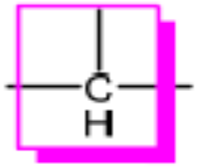
The Pharmacophore for all classes of antimuscarinics



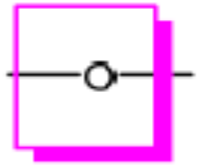
Acetylcholine (ACH)



R_1 = OH : Aminoalcohol ester
R_1 = $CONH_2$: Aminoamide ester



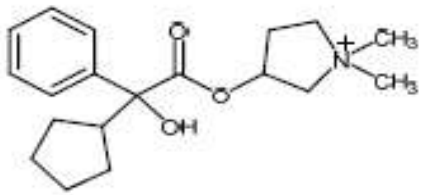
R_1 = OH : Aminoalcohol
R_1 = $CONH_2$: Aminoamides



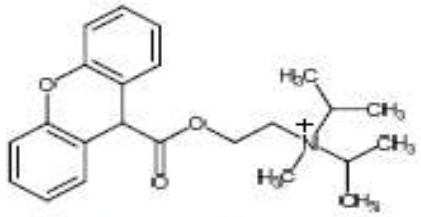
R_1 = OH : Aminoalcohol ether
R_1 = $CONH_2$: Aminoamide ether

Acetylcholine analogue antimuscarinics

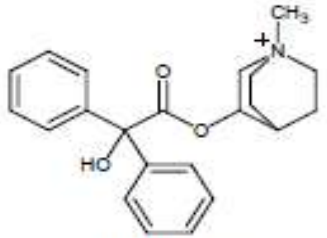
Identify which chemical class the following compounds belong to ..



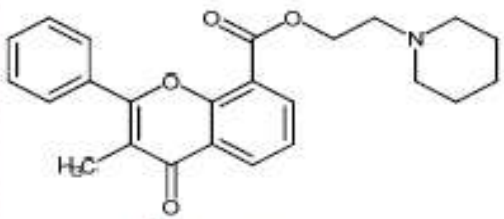
Glycopyrrolate
(Nodapton, Robinal, Tarodyl)



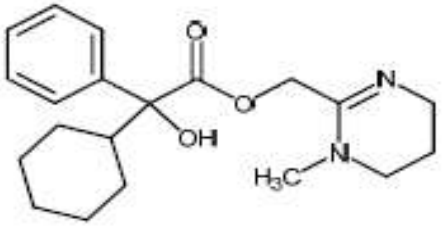
Propantheline
(Corrigast, Pro-Banthine, Pantheline)



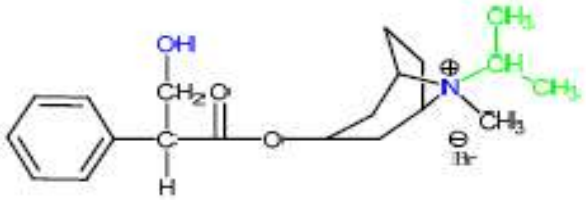
Clidinium
(Quarzan)



Flavoxate
(Bladderon, Spasuret, Urispas)

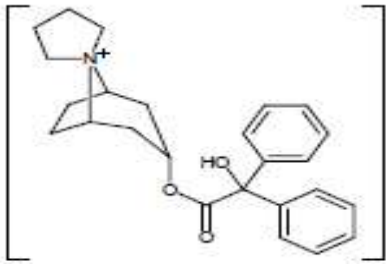


Oxyphencyclimine
(Antulcus, Setrol, Daricon)



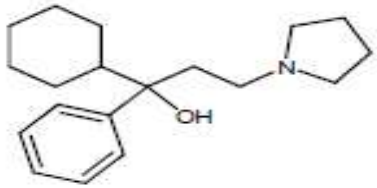
Ipratropium
(Atem, Atrovent, Narilet)

More Drugs …



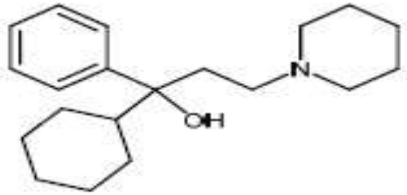
Trospium Chloride
(Sanctura)

Urinary and GI antispasmodic



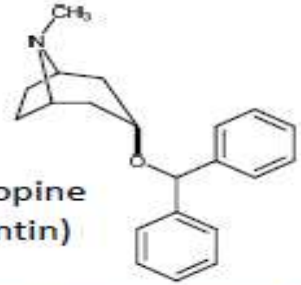
Procyclidine
(Kemadrin)

Antiparkinsonism



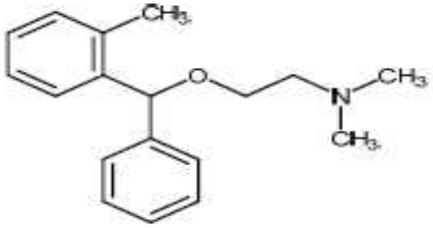
Trihexyphenidyl
(Artane)

Antiparkinsonism



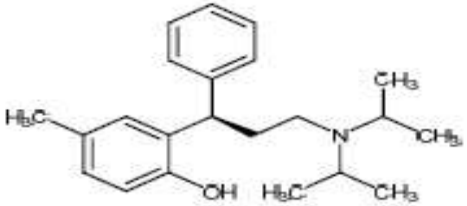
Benztropine
(Cogentin)

Antiparkinsonism



Orphenadrine
(Norflex)

Muscle relaxant (skeletal); antihistaminic



Tolterodine
(Detrol)

Urinary incontinence

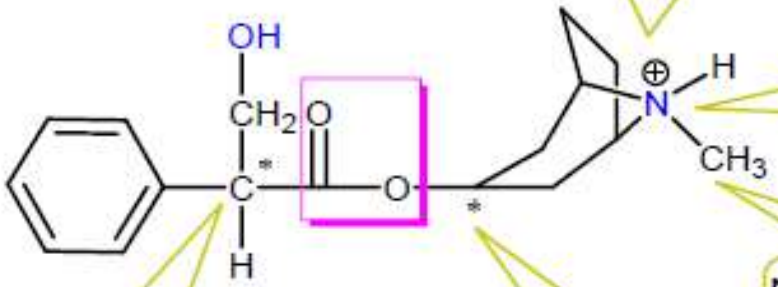
Muscarinic receptor subtype selectivity

- With the exception of solifenacin and darifenacin (selective M_3 antagonists), all other agents discussed in this chapter display no marked selectivity for muscarinic receptor subtypes.

SPECIFIC ANTIMUSCARINIC DRUGS

Atropine: Classic Prototype for Antimuscarinics

Amino alcohol ester pharmacophore



Quaternary Nitrogen binds to the anionic Asp residue in the muscarinic receptor

When the Nitrogen made is quaternary, molecule looses oral availability, but effective by inhalation

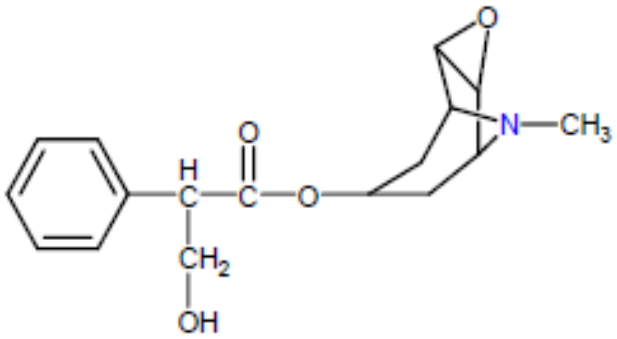
Methyl is optimal substitution on Quaternary Nitrogen

R-isomer is 100 fold more active than and *S*- isomer

There is little difference between *R*- and *S*- configurations

- The naturally occurring alkaloid, (–)-hyoscyamine, upon base-catalyzed racemization gives (±)-hyoscyamine or atropine.
- **Uses:** antidote for anticholine esterase poisoning, Decrease GIT motility, Dilation of pupils

Scopalamine (Hyoscine)



- Chemically and pharmacologically similar to atropine
- CNS depressant where as other antimuscarinics are CNS stimulants
- Used in the treatment of motion sickness, parkinsonism
- Used as a truth drug (CNS effects)

Antimuscarinic drugs used in the treatment of urinary incontinence

1.Tolterodine
2. Oxybutynin
3. Solifenacin
4. Darifenacin

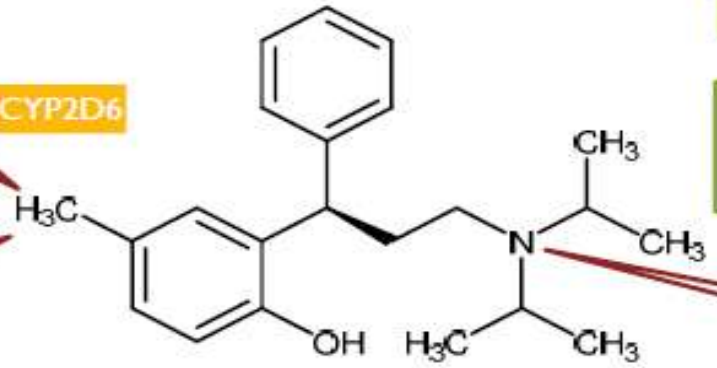
Tolterodine (Detrol®)

Oxidation to hydroxymethyl Tolterodine

Active

CYP2D6

Further oxidation to Carboxylic acid



N-dealkylation

Non-selective muscarinic antagonist

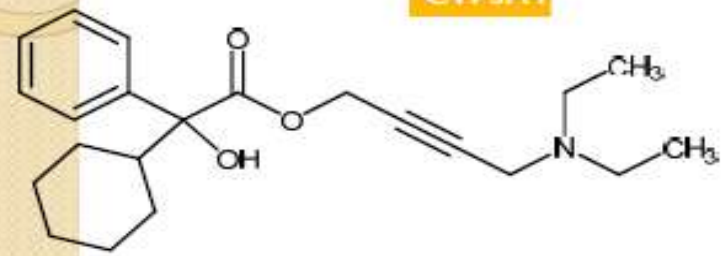
Metabolites:
1. 5-hydroxymethyl (Active)
2. N-dealkylated-5-carboxylic acid

- Acts on M2 and M3 subtypes of muscarinic receptors whereas most anti-muscarinic treatments for overactive bladder only act on M3 receptors making them more selective.

Known Side Effects:	Hyposalivation(Dry mouth) Decreased gastric motility (Upset stomach) Headache , Constipation , Dry eyes , Drowsiness
Half Life:	2-4 hrs

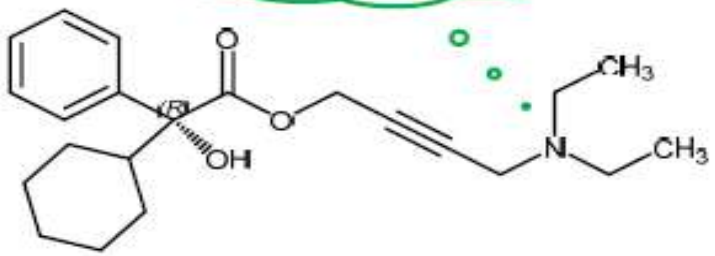
Oxybutynin (Ditropan®)

CYP3A4

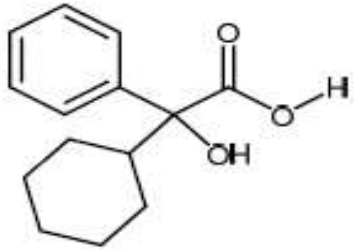


N-deethylation

Active



Non-selective muscarinic antagonist

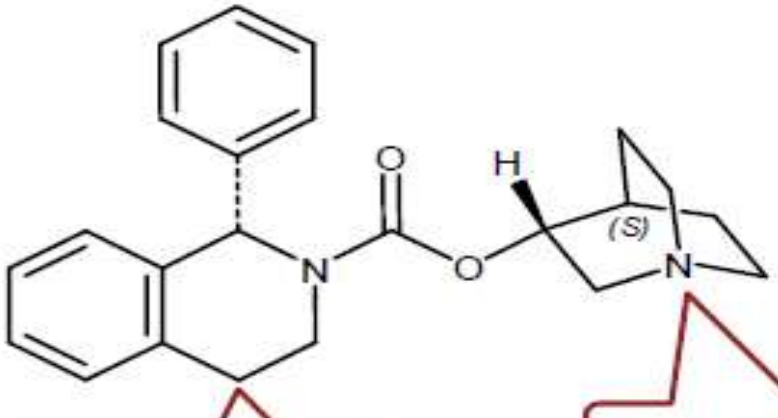


Inactive

The (*R*)-enantiomer is a more potent anticholinergic than the (*S*)-enantiomer, but marketed as racemic mixture

Half Life : 2-5 hrs

Solifenacin (Vesicare)



Selective M3 antagonist

CYP3A4

Active

4-hydroxylation

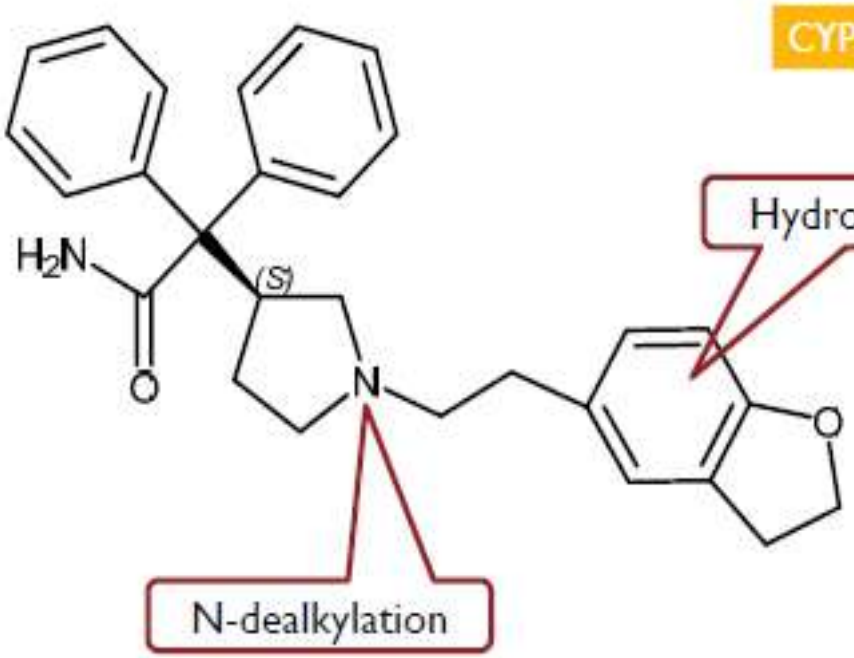
N-oxide, N-glucuronide

Half Life: 55 hrs

Metabolites:

1. 4-hydroxy derivative (active),
2. N-glucuronide
3. N-oxide
4. 4-hydroxy-N-oxide

Darifenacin(Enablex®)

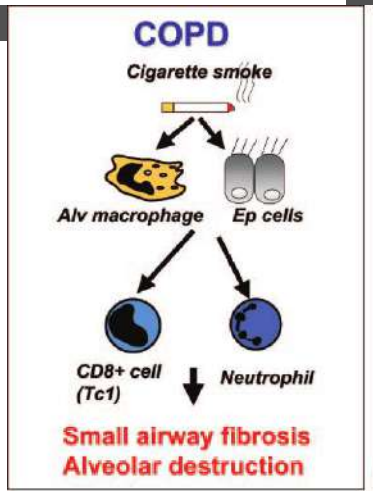


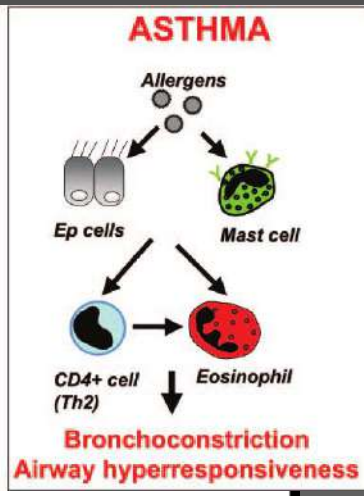
Selective M3 antagonist

Antimuscarinic drugs used in the treatment of bronchial asthma

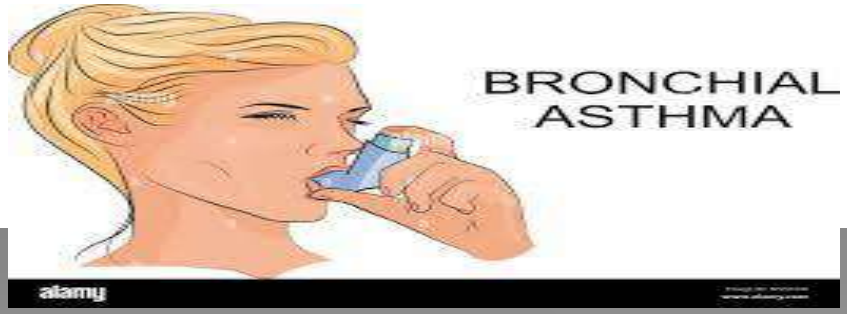
- Ipratropium hydrobromide
- Tiotropium bromide

M_3 receptors: Bronchiole constriction

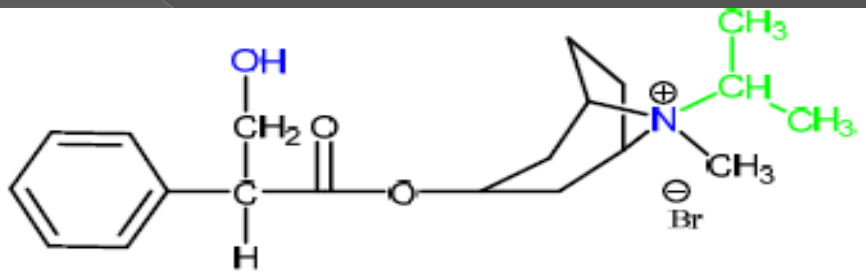




Therapeutic use: block cholinergic bronchiole constriction (anticholinergics) and allow adrenergic bronchiole dilation to help overcome the pulmonary constriction associated with asthmatic attack.



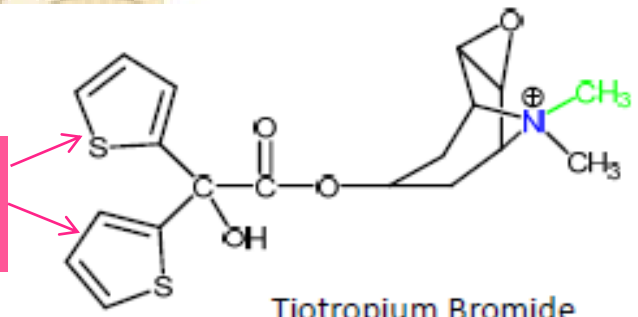
Ipratropium Hydrobromide (Atrovent®)



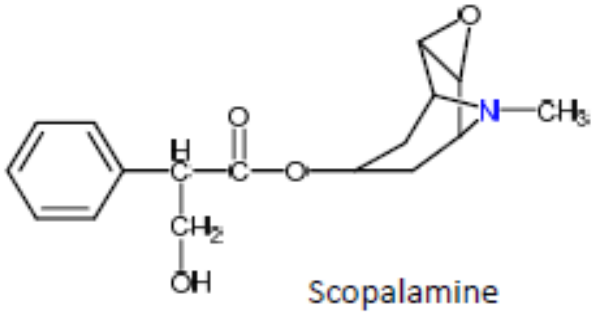
- N-isopropyl analogue of Atropine
- Quaternary cationic nature makes it highly hydrophilic and poorly absorbed from the lungs after inhalation via solution or aerosol, so bronchodilation effect can be considered to be a local, site-specific effect
- Ipratropium is indicated primarily for the relief of bronchospasms associated with COPD and has seen little application for the treatment of asthma
- Reaches the circulation minimally and is partially metabolized to inactive esterase products

Tiotropium Bromide (Spiriva®)

Di Thienyl



Tiotropium Bromide

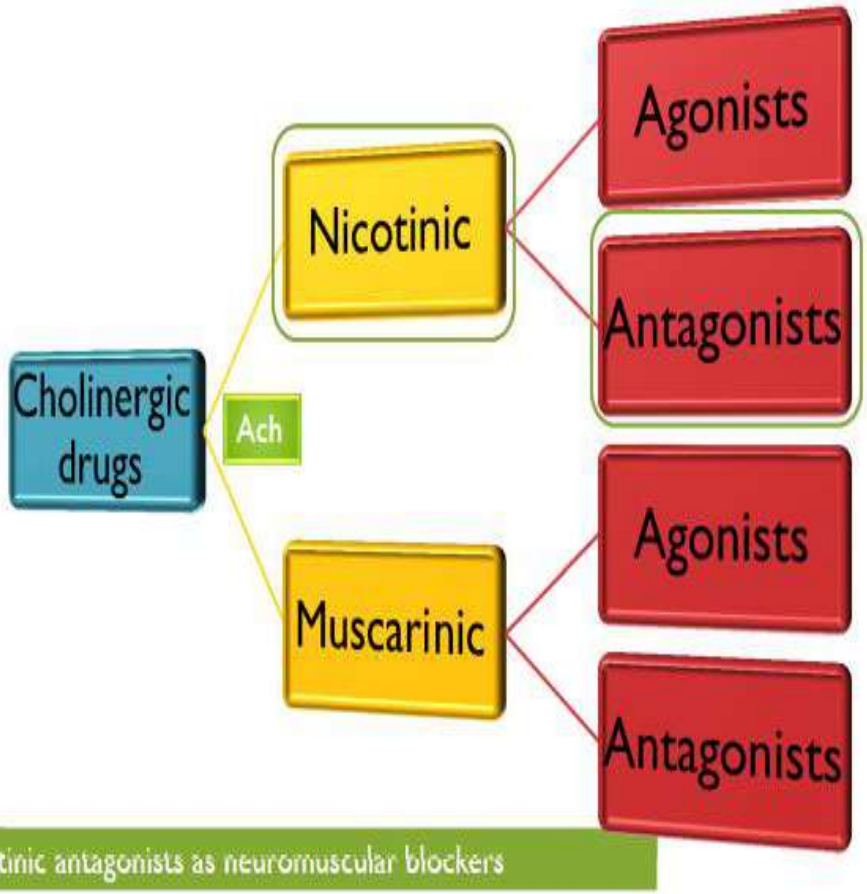


Scopalamine

- Dithienyl derivative of N-methyl scopolamine, a quaternary analogue of naturally occurring scopolamine in Atropa belladonna
- Indicated primarily for the relief of bronchospasms associated with COPD and can be considered to be a site-specific, local medication to the lung.
- Longer duration of action than ipratropium (24 versus <4 hours, respectively).
- Tiotropium is metabolized by both CYP3A4 and CYP2D6, followed by glutathione conjugation to a variety of metabolites

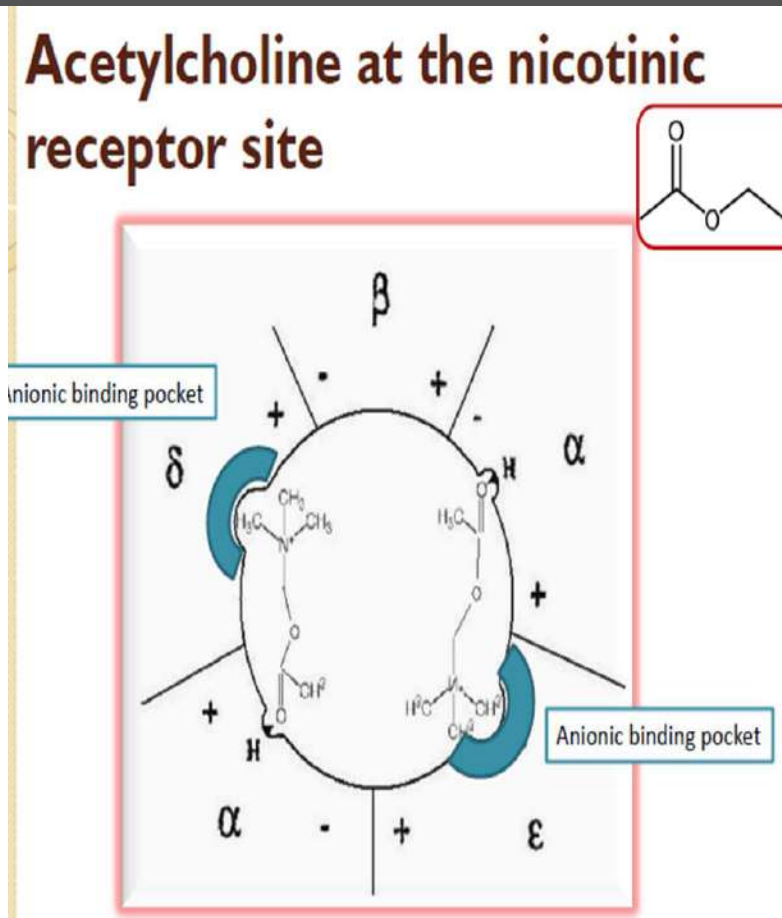
NICOTINIC ANTAGONISTS

Cholinergics



Nicotinic antagonists as neuromuscular blockers

Acetylcholine at the nicotinic receptor site



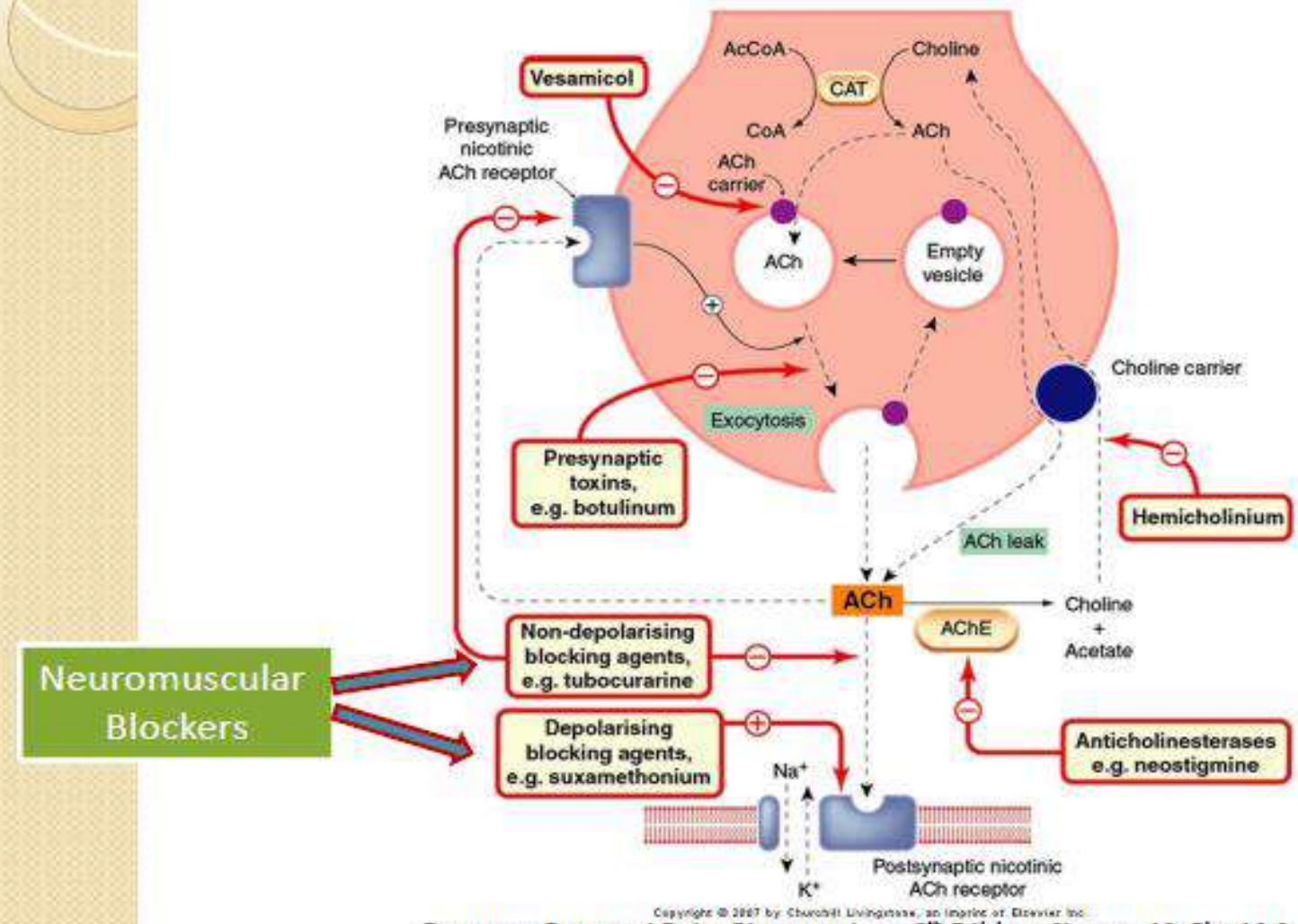
Nicotinic Antagonists

- Compounds that bind to cholinergic nicotinic receptors with no intrinsic efficacy (competitive antagonists with Ach)

- Two subclasses of nicotinic antagonists
  - Skeletal neuromuscular blocking agents and
  - Ganglionic blocking agents.

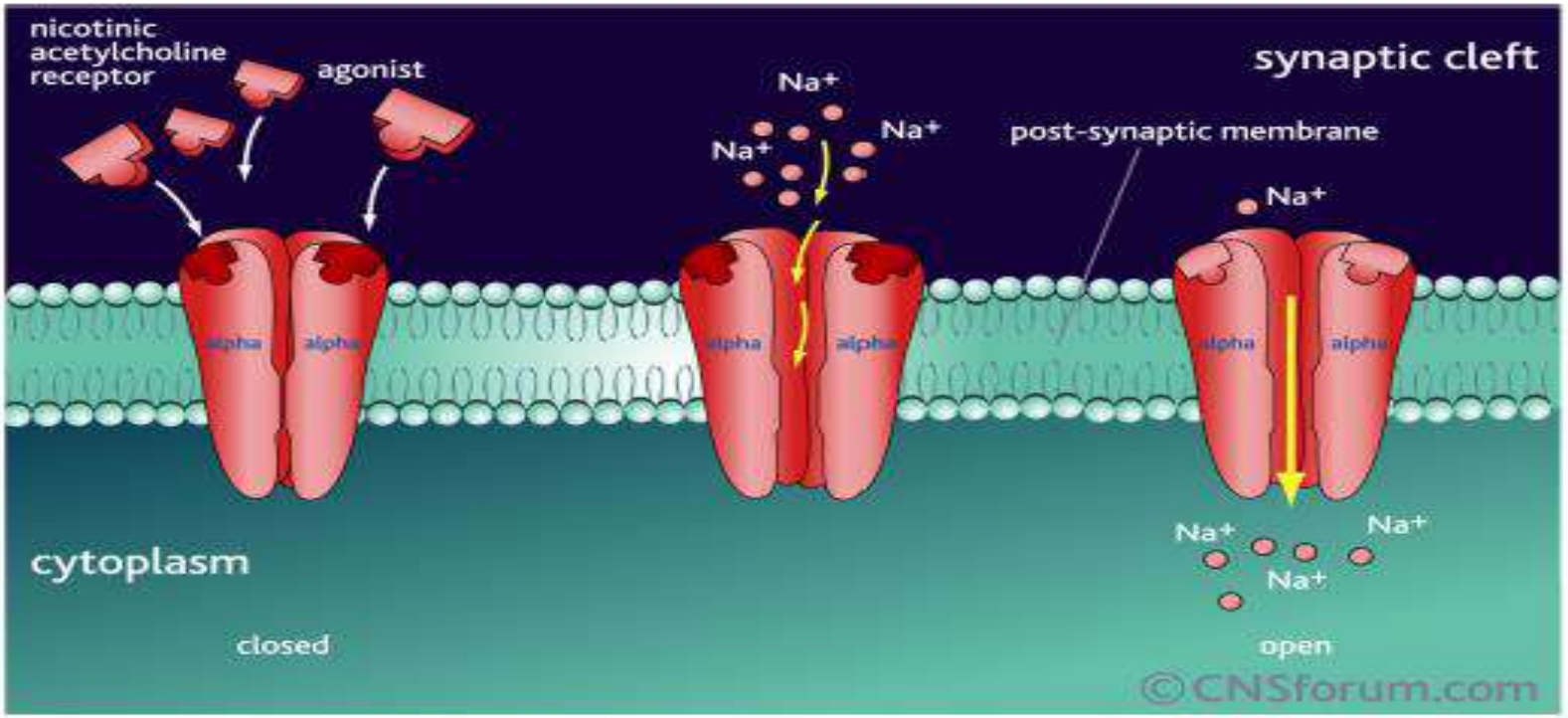
- DONOT confuse with skeletal muscle relaxants that produce their effects through the CNS

Events and sites of drug action at a nicotinic cholinergic synapse



Resource: Rang and Dales Pharmacology, 6th Edition, Chapter 10, Fig. 10.2

Neuromuscular Blockers (NMBs)



Depolarizing blockers: Depolarizes the post synaptic membrane with the inflow of sodium ions

Non-depolarizing blockers: Blocks the sodium ions conductance

Therapeutic application

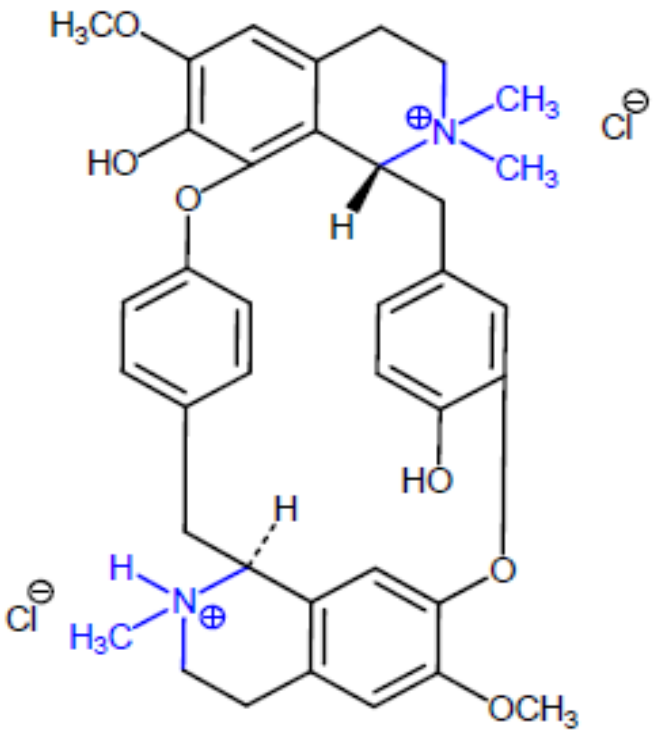
- Neuromuscular blocking agents are used primarily as an adjunct to general anesthesia.
  - Produces skeletal muscle relaxation which facilitates operative procedures

- Questions to be considered in choosing NMB
  1. Will the compound produce the desired neuromuscular blockade?
  2. What is its duration of action?
  3. What are its adverse effects?
  4. What is its relative cost?

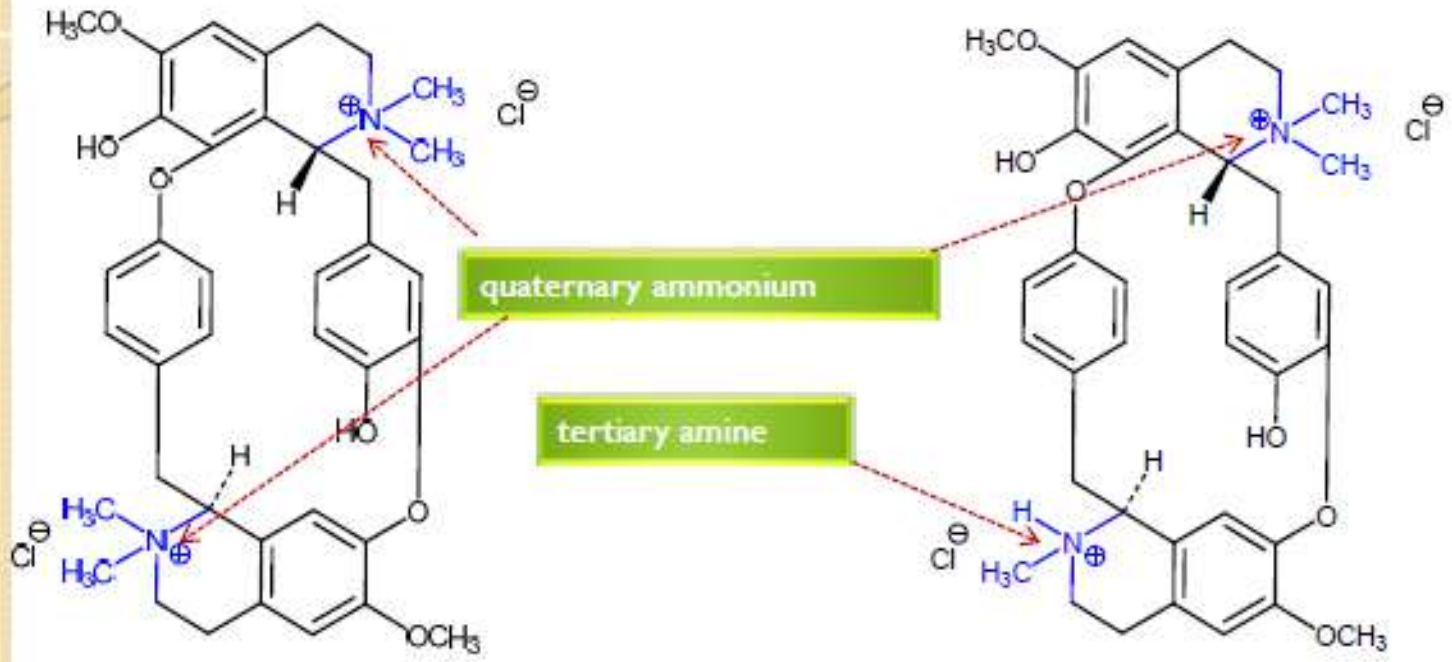
Discovery: Tubocurarine – the first neuromuscular blocker

- The neuromuscular blocking effects of extracts of curare were first reported as early as 1510

- It was demonstrated that curare extracts prevented skeletal muscle contractions by an effect at the neuromuscular junction



Tubocurarine structure



Incorrect structure: 1935

Correct Structure: 1970

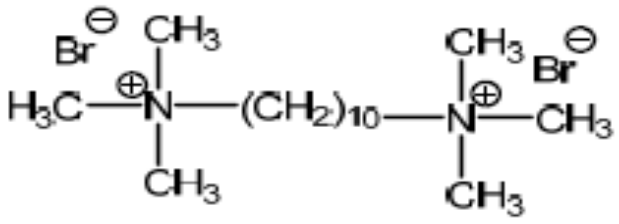
- Nevertheless, the incorrect structure of tubocurarine served as the model for the synthesis of all the neuromuscular blocking agents in use today

Rationale for designing new compounds

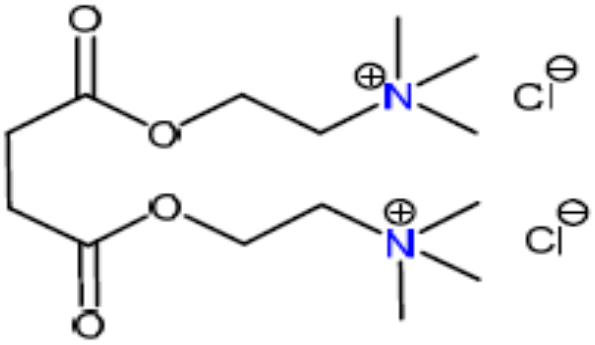
- Bis-quaternary ammonium compound having two quaternary ammonium salts separated by 10 to 12 carbon atoms (similar to the distance between the nitrogen atoms in tubocurarine) was a requirement for neuromuscular blocking activity
- The rationale for this structural requirement was that nicotinic receptors possessed two anionic-binding sites, both of which had to be occupied for a neuromuscular blocking effect.
- Nicotinic antagonist may be a
  - Depolarizing neuromuscular blocker
  - Non-depolarizing neuromuscular blocker

Depolarizing agents

- Decamethonium Bromide
- Succinylcholine Chloride (Anectine®)



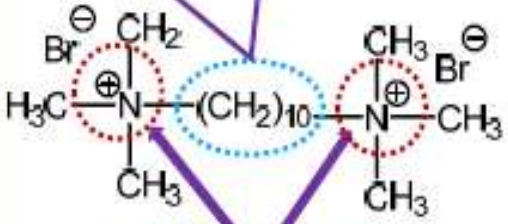
Decamethonium Bromide



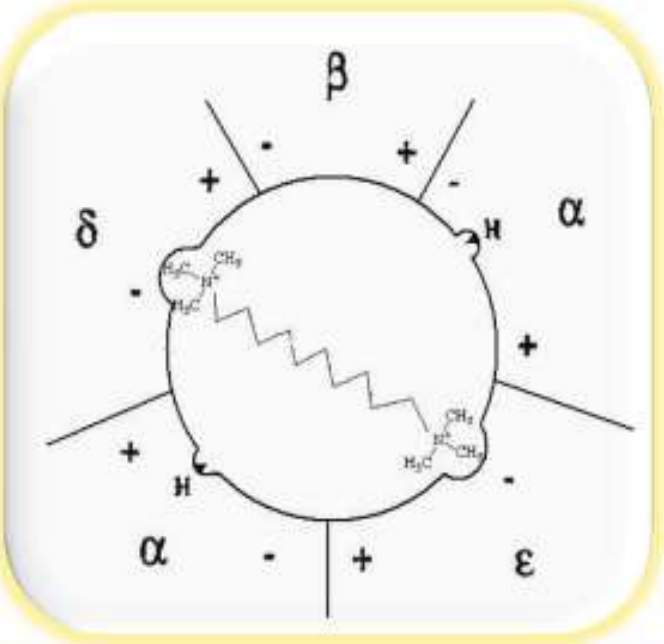
Succinylcholine Chloride

Decamethonium Bromide

10 carbon bridge between two nitrogens

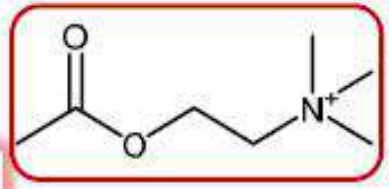


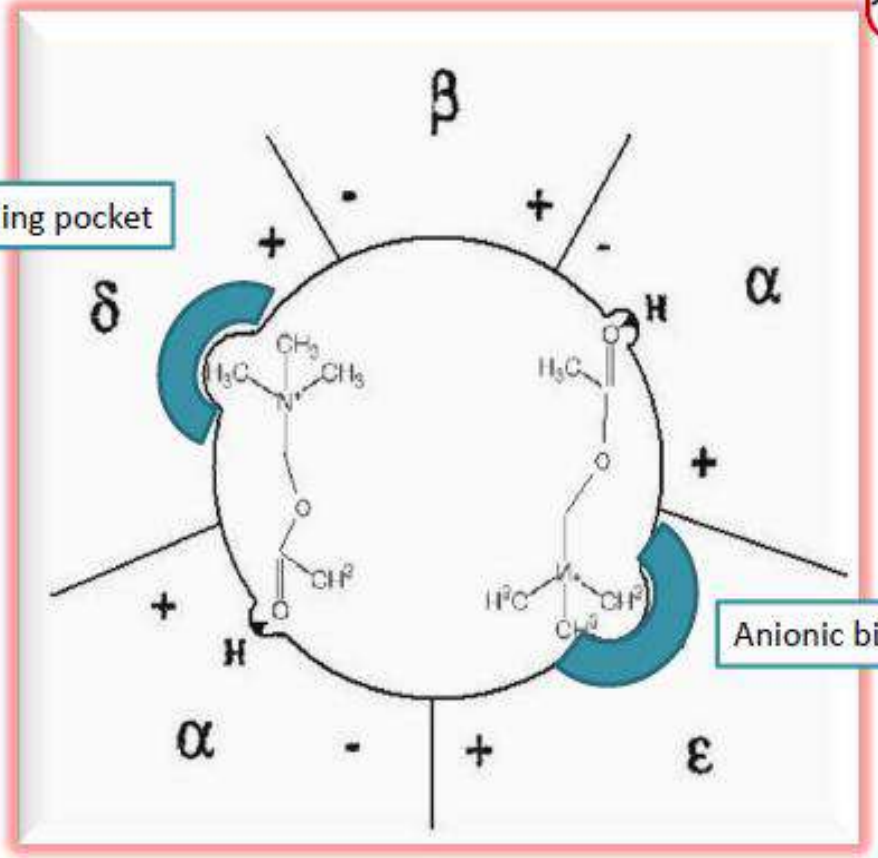
Bis-quaternary ammonium salt



Decamethonium at the nicotinic receptor

Acetylcholine at the nicotinic receptor site

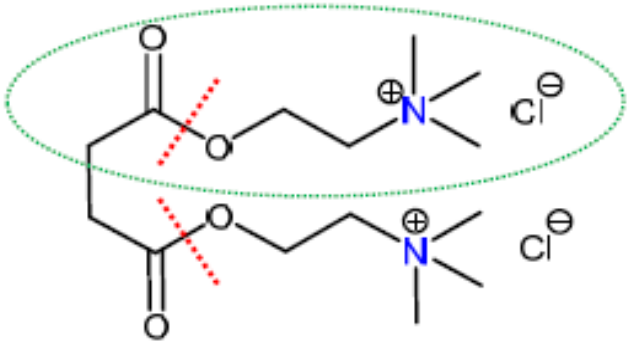




Anionic binding pocket

Anionic binding pocket

Succinyl choline



- Dimer of acetylcholine bonded through their α-carbon adjacent to ester
- Rapidly hydrolyzed to inactive metabolites, both in aqueous solution and plasma esterases (shorter duration of action, 6-8 min)
- Through succinylcholine has 10 atoms between two nitrogens, it still reported to catalyze with two molecules at the receptor site.

Non-depolarizing agents

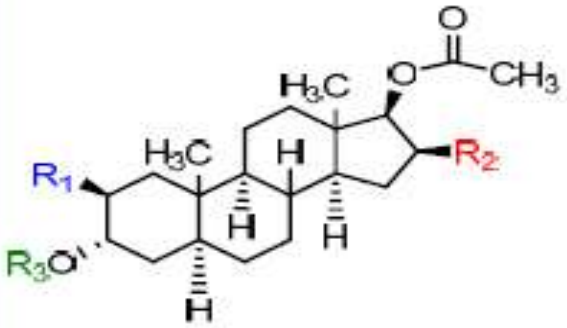
Aminosteroids

- Pancuronium bromide (Pavulon)
- Vecuronium bromide (Norcuron)
- Rocuronium bromide (Zemuron)
- Pipecuronium bromide (Arduan)

Tetrahydroisoquinolines

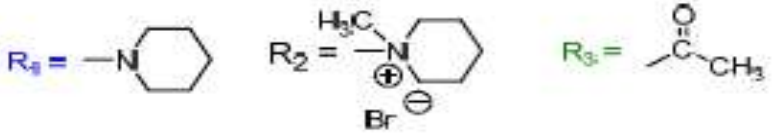
- d-Tubocurarine and metocurine (Metubine Iodide)
- Atracurium besylate (Tracrium)
- Cis-atracurium
- Mivacurium chloride (Mivacron)
- Doxacurium chloride (Nuromax)

Amino steroid NM blocking agents

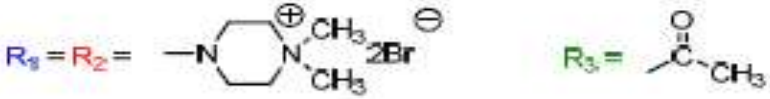


Pancuronium bromide

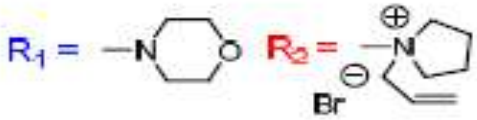
Vecuronium bromide

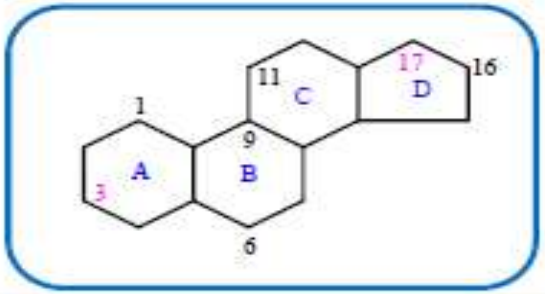


Pipecuronium bromide



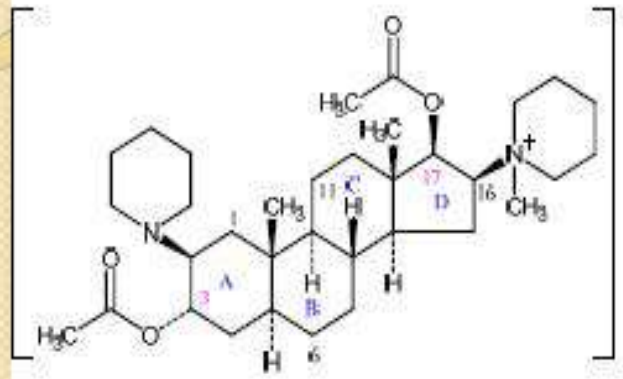
Rocuronium bromide

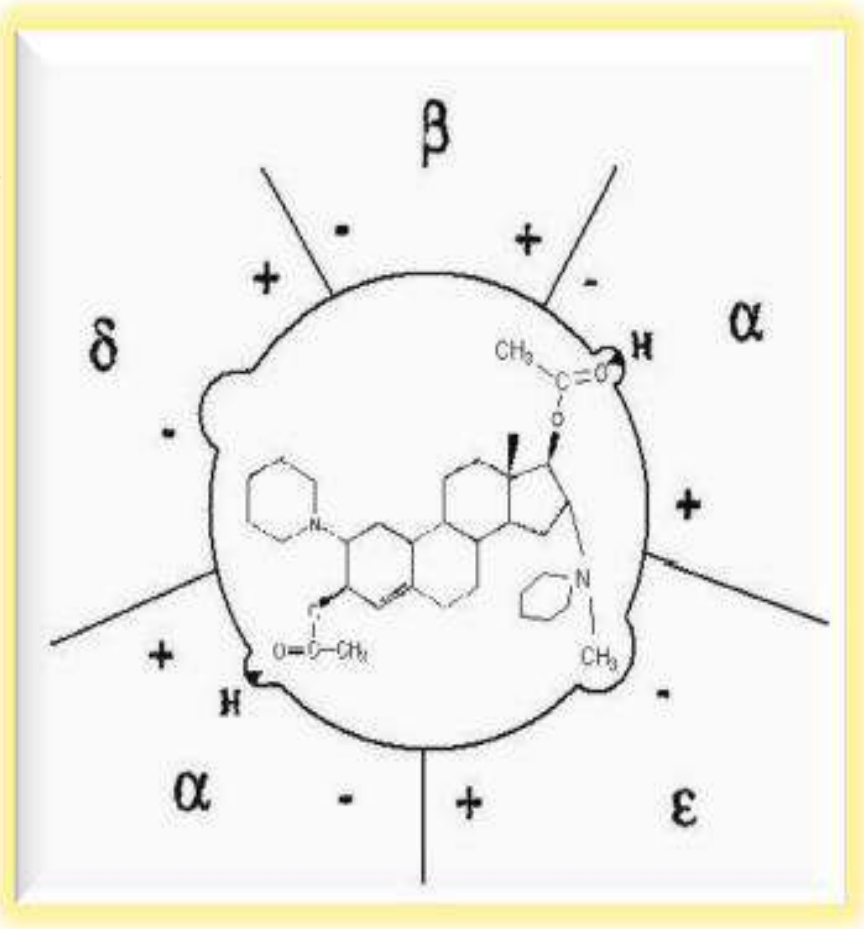




Monoquaternary aminosteroids are less potent but has faster onset of action

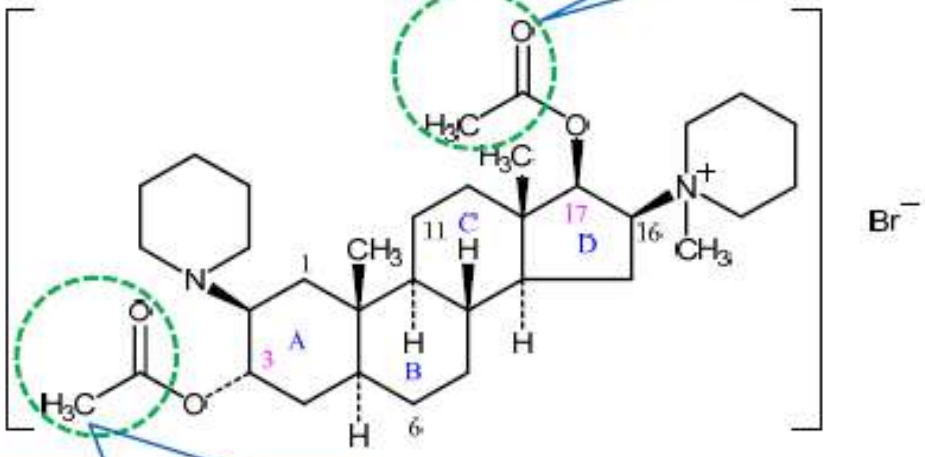
Vecuronium at the nicotinic receptor





Vecuronium (Norcuron)

17 deacetylated metabolite is active



3-deacetylated metabolite is active

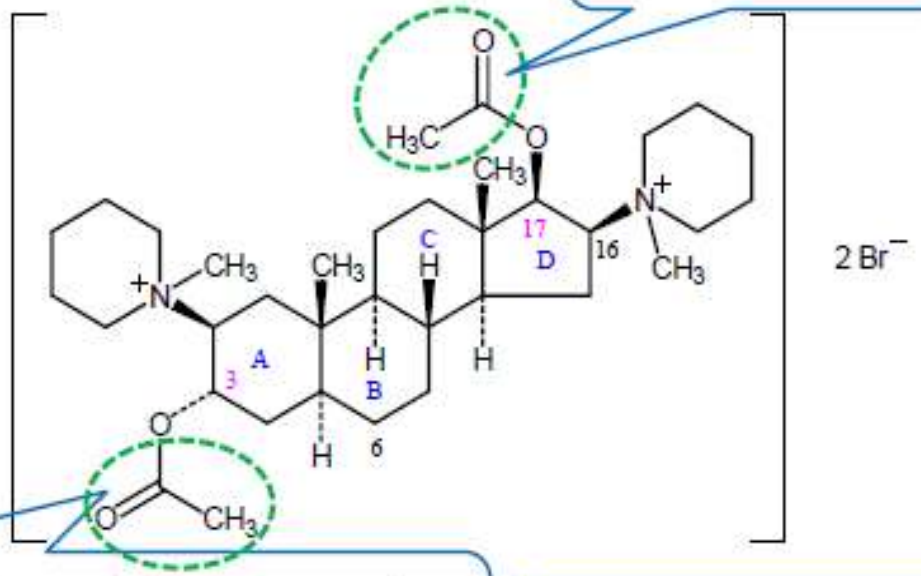
Metabolites:

- 3-hydroxy metabolite (Active)
- 17-hydroxy metabolite (Active)
- 3, 17-dihydroxy metabolite (Active)

Accumulation is responsible for prolonged neuromuscular blockade in patients receiving long-term therapy

Pancuronium Bromide (Pavulon)

17 deacetylated metabolite is inactive



Pancuronium

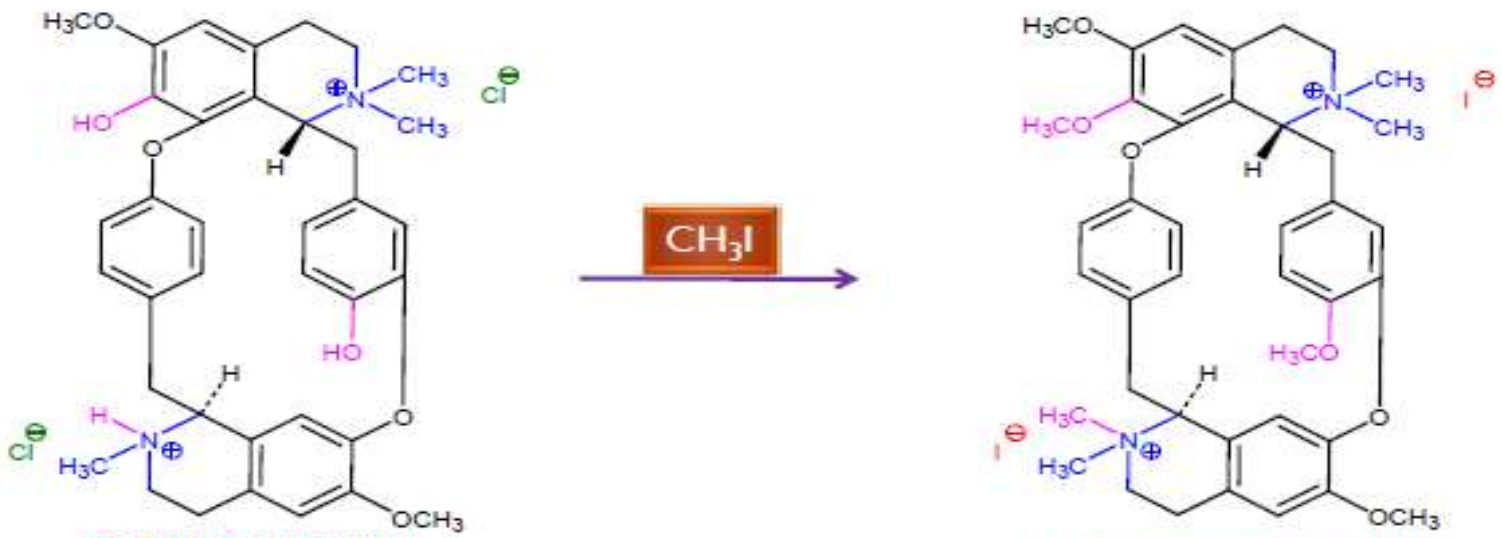
3-deacetylated metabolite is active as long as 17 deacetylation does not occur

Metabolites:

- 3-hydroxy metabolite (Active)
- 17-hydroxy metabolite (Inactive)
- 3, 17-dihydroxy metabolite (Inactive)

Tetrahydroisoquinolines

d-Tubocurarine and metocurine (Metubine Iodide®)



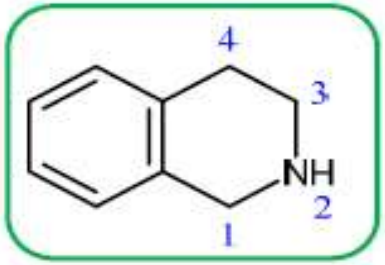
d-Tubocurarine

- Long duration of action
- Mostly eliminated unchanged via kidney and bile, <1% gets demethylated in the liver

Metocurine iodide

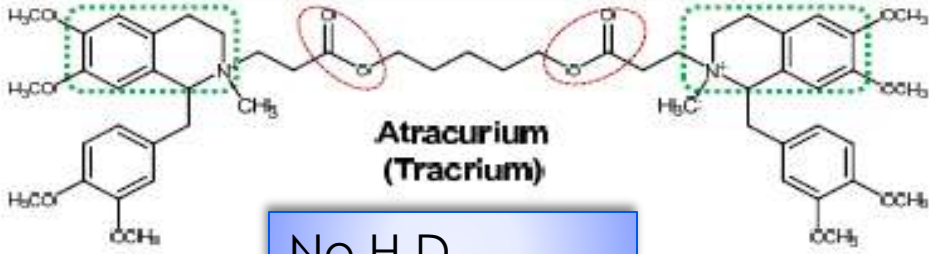
- Long duration of action
- 4 fold more potent than d-tubocurarine
- Eliminated unchanged via kidney

Tetrahydroisoquinolines

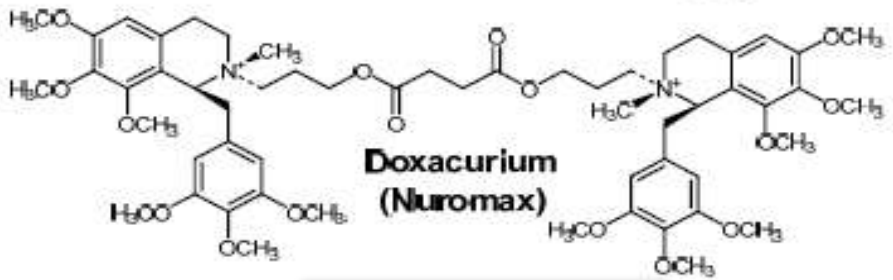


1,2,3,4 - tetrahydroisoquinoline

Hoffman Degredation (HD)

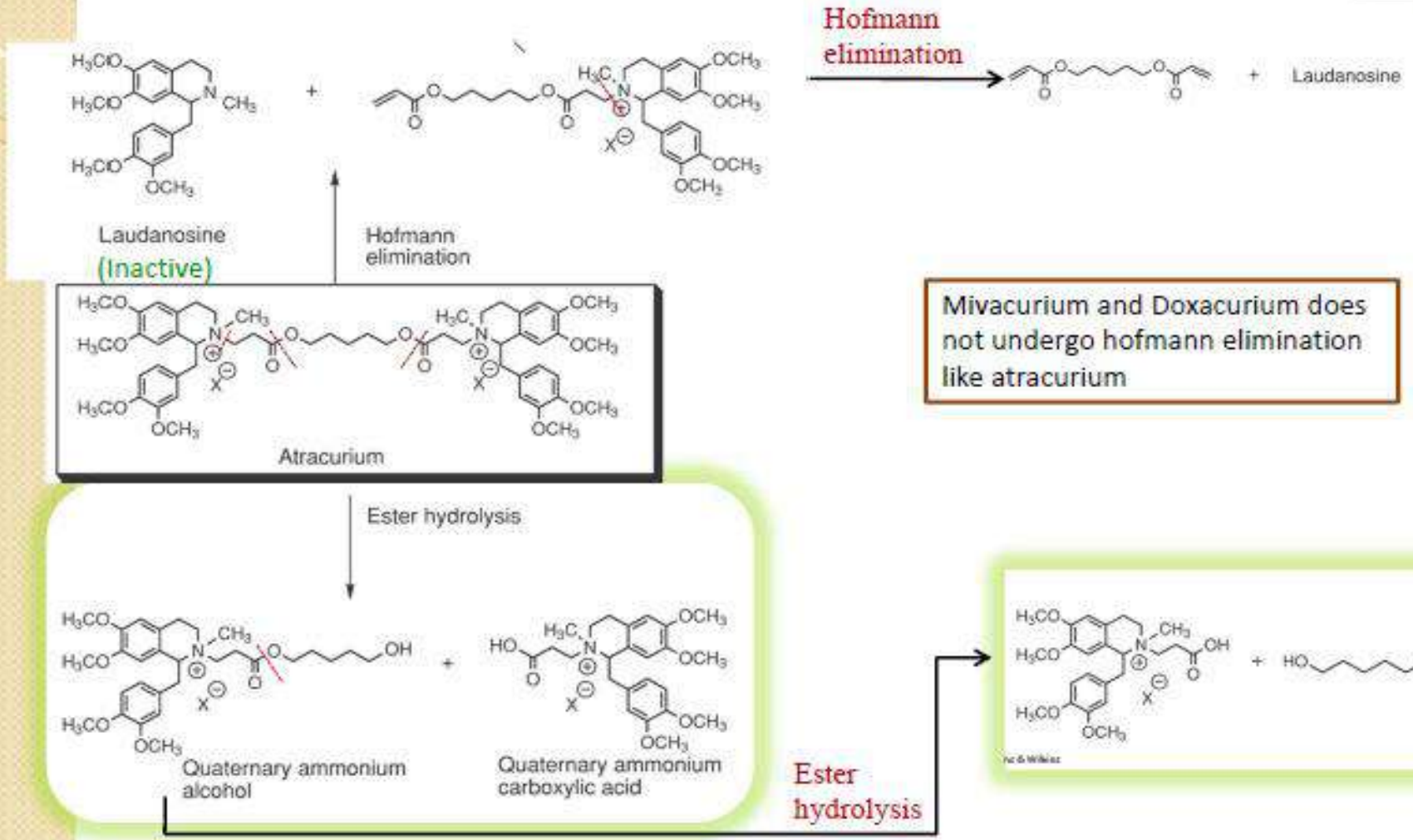


No H D



No HD

Atracurium metabolism



Structure Activity Relationships

- Contain at least one +ve charged quaternary amine (4 C atoms attached to nitrogen) like Ach
- N^+ attracted to Alpha sub unit of AChR (-ve charge)

Bis quaternary amines

- *Succinylcholine, Pancuronium, Atracurium*

Mono quaternary amines

- *D-Tubocurarine, Vecuronium, Rocuronium*
- In acidosis, Tertiary amine protonated → + charge → increased potency
- Bridging structure in between two amines is lipophilic and determines potency
- 10-12 carbon bridge between two nitrogens is optimal for maximal neuromuscular blockade.

Metabolism of neuromuscular blockers

Agent	Duration of action (min)	Metabolites	Mode of elimination
Succinylcholine	6–8	Ester hydrolysis (inactive)	Hydrolysis by plasma cholinesterases
d-Tubocurarine	80–120	Mostly unchanged	Renal elimination, liver clearance
Vecuronium	30–40	3-hydroxy , 17-hydroxy, 3,17-dihydroxy (all active)	Liver metabolism and clearance, renal elimination
Pancuronium	120–180	3-hydroxy (active) 17-hydroxy, 3,17-dihydroxy (inactive)	Renal elimination, liver metabolism and clearance
Pipecuronium	80–100	Primarily unchanged	Renal elimination, liver metabolism and clearance
Rocuronium	30–40	Primarily unchanged	Liver metabolism and clearance
Atracurium	30–40	Ester hydrolysis products, Laudanosine (all inactive)	Hofmann degradation, hydrolysis by plasma cholinesterases
Mivacurium	12–18	Ester hydrolysis products (inactive)	Hydrolysis by plasma cholinesterases
Doxacurium	90-120	Ester hydrolysis products (Inactive)	Renal elimination, liver metabolism

Duration of action: 0-29 min: Short acting; 30-40 min: Intermediate acting; over 40 min: long acting

QUESTIONS ???