MScCH-09

Vardhman Mahaveer Open University, Kota

Drugs and Pharmaceuticals

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Vardhman Mahaveer Open University, Kota

Drugs and Pharmaceuticals

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Preface

The present book entitled "Drugs and Pharmaceuticals **"** has been designed so as to cover the unit-wise syllabus of MScCH-09 course for M.Sc. Chemistry (Final) students of Vardhman Mahaveer Open University, Kota. The basic principles and theory have been explained in simple, concise and lucid manner. Adequate examples, diagrammes , photographs and self-learning exercises have also been included to enable the students to grasp the subject easily. The unit writers have consulted various standard books and internet as their reference on the subject and they are thankful to the authors of these reference books.

Unit-1

Drug Discovery, Design and Developme

Structure of Unit:

1.1 Objectives

In this unit the students will be able to understand

- Drug Discovery with and without lead
- Identification of the active Part: The Pharmacophore
- Functional Group Modification
- Structural Activity Relationships
- Quantitative Structure activity Relationships
- About Pro-drugs and soft drugs

1.2 Introduction: Drug Discovery, Design, and Development

Medicinal chemistry is the scientific discipline that makes such drugs available through either discovery or design processes. Therapeutic agents are chemicals that prevent disease, assist in restoring health to the diseased, or alleviate symptoms associated with disease conditions.

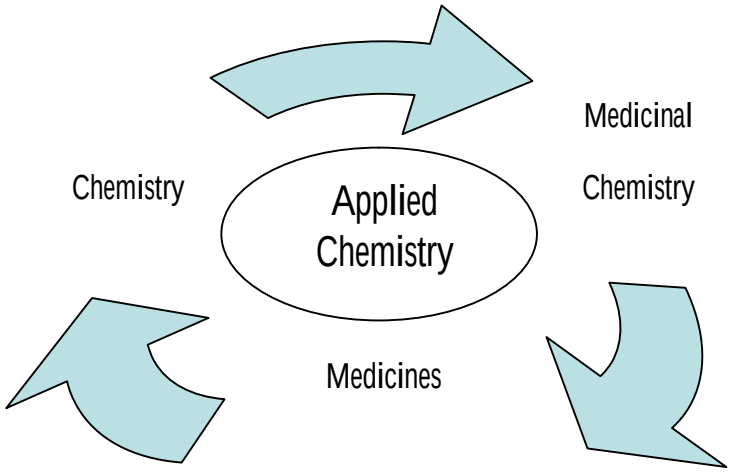


Fig.1.1: Relation Cycle of Applied Chemistry

Drug discovery is part luck and part structured investigation. In the twentieth century, a very large number of biologically active natural products were structurally modified in order to optimize their pharmacology, and novel drugs were prepared by use of increasingly advanced synthetic methods. Moreover, the rapidly growing understanding of the nature of disease mechanisms, how cells function, and how drugs interact with cellular processes has led to the rational design, synthesis, and pharmacological evaluation of new drug candidates. Most recently, new dimensions and opportunities have emerged from a deeper understanding of cell biology and genetics.

Drug design aims at developing a drug with high degree of chemotherapeutic index and specific action.

Drug design seeks to explain:

- Effects of biological compounds on the basis of molecular interaction in terms of molecular structures or precisely the physico-chemical properties of the molecules involved.
- Various processes by which the drugs usually produce their pharmacological effects.
- How the drugs specifically react with the protoplasm to elicit a particular pharmacological response.
- How the drugs usually get modified or detoxicated, metabolized or eliminated by the organism.
- Probable relationship between biological activities with chemical structure.

1.3 Development of new drugs

Drug development normally refers to the process of taking a compound that has been identified from the drug discovery process described above through the subsequent steps necessary to bring it to market. Typically, these additional major steps include preclinical development, clinical development, and regulatory approval.

The 'drug design' in a broader sense implies random evaluation of synthetic as well as natural products in bioassay systems, creation of newer drug molecules based on biologically-active-prototypes derived from either plant or animal kingdom, synthesis of congeners displaying interesting biological actions, the basic concept of isosterism and bioisosterism, and finally precise design of a drug to enable it to interact with a receptor site efficaciously.

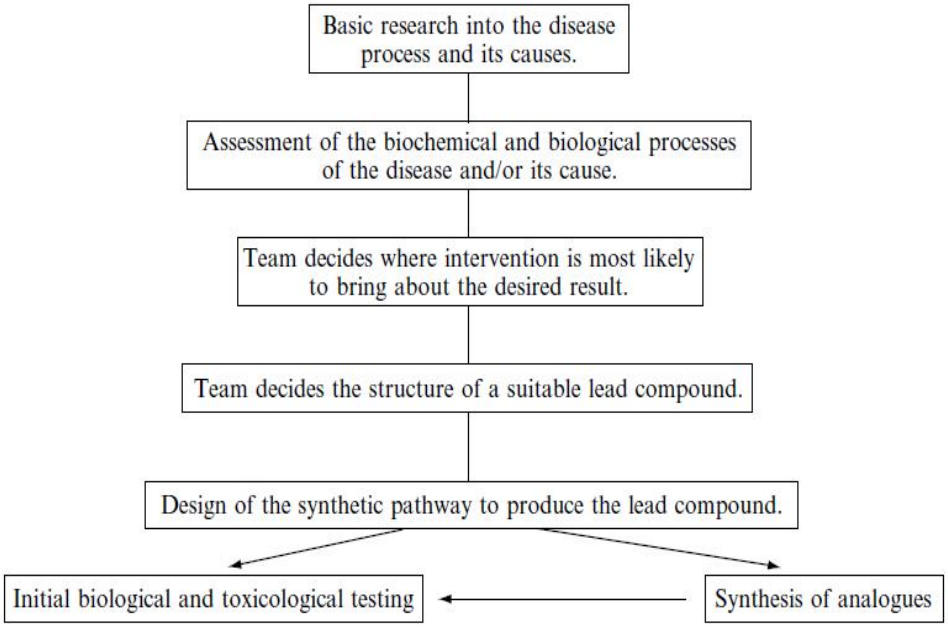


Fig.1.2: The general steps in the design of a new drug

Factors governing drug-design

- The smaller the expenditure of human and material resources involved evolving a new drug of a particular value, the more viable is the design of the programme.
- Experimental animal and clinical screening operations of the new drugs.
- Relationships between chemical features and biolgoical properties need to be established retrospectively.
- Quantitative structure-activity relationships (QSARs) vary to an appreciable extent in depth and sophistication based on the nature of

evaluation of structure or activity. A purposeful relation of structural variables must include steric factors, electronic features of component functional groups and, in general, the molecule as a whole.

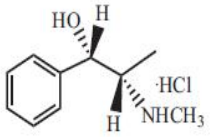
- The trend to synthesize a huge number of newer medicinal compounds indiscriminately for exploratory evaluation still prevails which exclusively reflects the creative genuineness and conceptual functions of a highly individualized expression of novelty by a medicinal chemist.
- Introduction of functional groups in a molecule that need not essentially resemble metabolites, but are capable of undergoing bonding interactions with important functional groups of biochemical components of living organisms affords an important basis for exploration.
- Disease etiologies and various biochemical processes involved prove useful.

1.4 Procedures followed in drug design

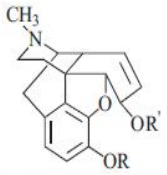
1.4.1 Drugs discovered without rational design

(a) Medicinal chemistry folklore

Medicinal chemistry, in its crudest sense, has been practiced for several thousand years. Man has searched for cures of illnesses by chewing herbs, berries, roots, and barks. Some of these early clinical trials were quite successful; however, not until the last 100–150 years has knowledge of the active constituents of these natural sources been known. The earliest written records of the Chinese, Indian, South American, and Mediterranean cultures described the therapeutic effects of various plant concoctions. Eg. *Ephedra sinica, Papaver somniferum, Rauwolfia serpentine, Colchicum autumnal, Digitalis purpurea* etc.

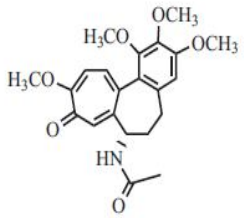


Pseudoephedrine Hydrochloride

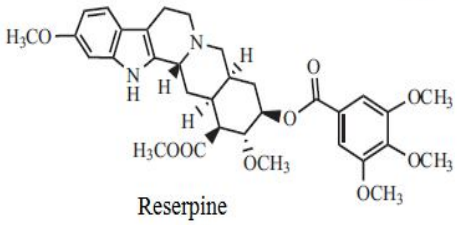


Morphine (R = R' = H)

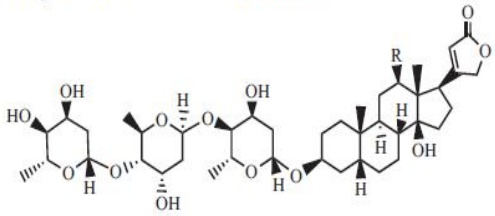
Codeine (R = CH_3, R' = H)



Colchicine



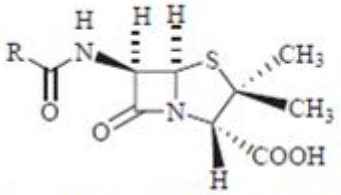
Reserpine



Digitoxine (R = H), Digoxine (R = OH)

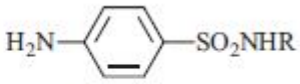
(b) Discovery of Penicillins

In 1928, Alexander Fleming noticed a green mold growing in a culture of *Staphylococcus aureus*, and where the two had converged, the bacteria were lysed. This led to the discovery of penicillin, which was produced by the mold. The original mold was *Penicillium notatum*, a strain that gave a relatively low yield of penicillin. It was replaced by *Penicillium chrysogenum.* Two reasons for the delay in the universal utilization of penicillin were the emergence of the sulfonamide antibacterials (sulfa drugs)



Penicillin V (R = $PhOCH_2$)

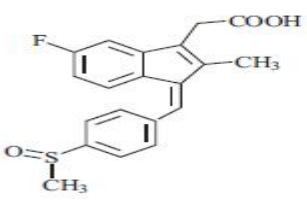
Penicillin G (R = CH_2Ph)



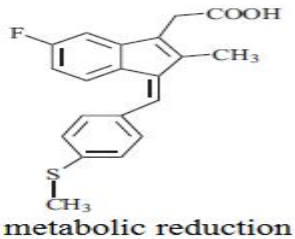
Sulfa Drugs

(C) Discovery of Drugs through Metabolism Studies

During drug metabolism studies, *metabolites* (drug degradation products generated in vivo) that are isolated are screened to determine if the activity observed is derived from the drug candidate or from a metabolite. For example, the anti-inflammatory drug sulindac is not the active agent; the metabolic reduction product is responsible for the activity.



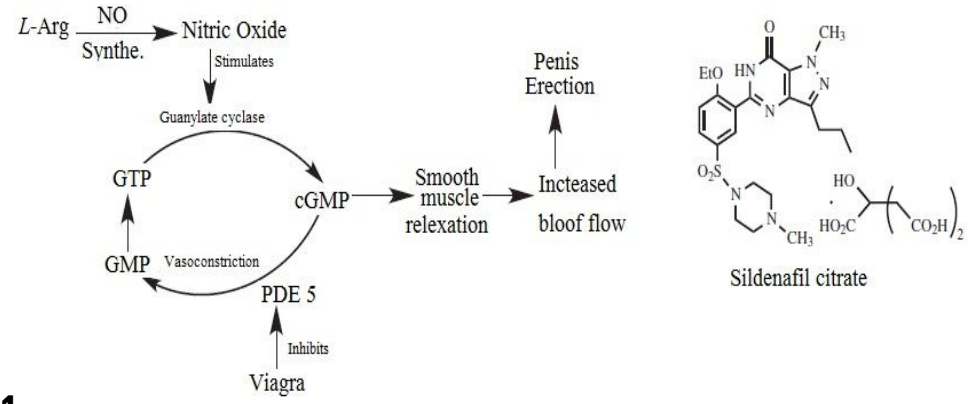
Sulindac



metabolic reduction product of Sulindac

(d) Discovery of Drugs through Clinical Observations

Sometimes a drug candidate during clinical trials will exhibit more than one pharmacological activity, that is, it may produce a side effect. This compound, then, can be used as a lead (or, with luck, as a drug) for the secondary activity. The impotence drug sildenafil citrate (Viagra) was designed for the treatment of angina and hypertension by blocking the enzyme phosphodiesterase-5, which hydrolyzes cyclic guanosine monophosphate (cGMP), a vasodilator that allows increased blood flow. In Phase II clinical trials, it was not as effective against angina as *Pfizer* had hoped, so it went back to Phase I clinical trials to see how high of a dose could be tolerated. It was during that clinical trial that the volunteers reported increased erectile function. Given the weak activity against angina, it was an easy decision to try to determine its effectiveness as the first treatment for erectile dysfunction.



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A rational approach to drug design may be viewed from different angles, namely:

(a) Quantum Mechanical Approach

Quantum mechanics (or wave mechanics) is composed of certain vital principles derived from fundamental assumptions describing the natural phenomena effectively. The properties of protons, neutrons and electrons are adequately explained under quantum mechanics. The electronic features of the molecules responsible for chemical alterations form the basis of drug molecule phenomena.

(b) Molecular Orbital Approach

The molecular orbital approach shows dependence on electronic charge as evidenced by the study of three volatile inhalation anaesthetics, and also on molecular conformation as studied with respect to acetylcholine by such

parameters as bond lengths and angles including torsional angles. Molecular orbital calculations are achievable by sophisticated computers, and after meticulous interpretations of results the molecular structure in respect of structure-activity analysis is established.

(c) Molecular Connectivity Approach

This approach establishes the presence of structural features like cyclization, unsaturation, skeletal branching, and the position and presence of heteroatom in molecules with the aid of a series of numerical indices. For example: an index was determined to possess a correlative factor in the SAR study of amphetamine-type hallucinogenic drugs. Molecular connectivity approach has some definite limitations, such as: electronegativity variance between atoms, non-distinguishable entity of *cis-trans* isomerism.

(d) Linear Free-Energy Approaches

This method establishes the vital link between the proper selections of physicochemical parameters with a specific biological phenomenon. However, such a correlation may not guarantee and allow a direct interpretation with regard to molecular structure, but may positively offer a possible clue towards the selection of candidate molecules for synthesis.

(e) Drug Design through Disjunction

Disjunction comes in where there is the systematic formulation of analogues of a prototype agent, in general, toward structurally simpler products, which may be viewed as partial or quasi-replicas of the prototype agent.

The method of disjunction is usually employed in three different manners, namely:

- Unjoining of certain bonds
- Substitution of aromatic cyclic system for saturated bonds
- Diminution of the size of the hydrocarbon portion of the
- parent molecule.

(f) Drug Design through Conjunction

This is known as the systematic formulation of analogues of a prototype agent, in general, toward structurally more complex products, which may be viewed as structures embodying, in a general or specific way, certain or all of the features of the prototype. In this type of drug-design, the main principle involved is the ‘principle of mixed moieties’. A drug molecule is essentially made up with two or more pharmacophoric moieties embedded into a single molecule.

Example. Acetylcholine is an effective postganglionic parasympathetic stimulant in doses that afford no appreciable changes in the ganglionic function; whereas hexamethonium possesses only a slight action at postganglionic parasympathetic endings in doses that produce a high degree of ganglionic blockade.

1.5 Concept of lead compounds and lead modification

Drugs are generally not discovered, lead compounds are discovered. In the modern drug discovery paradigm that we are discussing, a lead compound typically has most or all of the following characteristics:

- It interacts with the target in a manner consistent with that needed to achieve the desired effect.
- It is amenable to synthetic modifications needed to improve properties.
- It possesses, or can be modified to possess, physical properties consistent with its ability to reach the target after administration by a suitable route.

Interestingly, the 'drug discovery process' may be categorized into four distinct heads, namely:

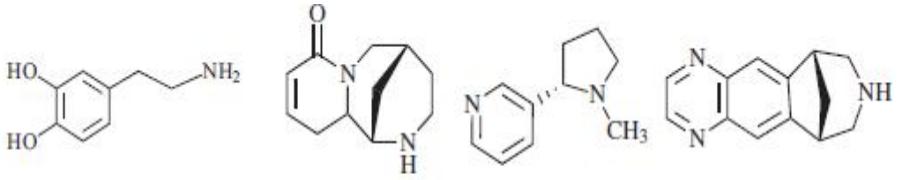
- Target identification and selection,
- Target optimization,
- Lead identification, and
- Lead optimization.

The concerted efforts encompassing various intangible and critical methodologies that ultimately relate to the activities, expertise, wisdom and integration of the individual scientist directly or indirectly involved in 'drug discovery process' virtually leads to advance drug discovery profiles.

Common sources of lead compounds are the following:

- The natural ligand or substrate for the target of interest. For example, dopamine is the natural ligand for the family of dopamine receptors.
- Another substance already known to interact with the target of interest. For example, the plant alkaloid cytisine was known to interact with nicotinic acetylcholine receptors.
- Comparing the three structures, one can also imagine that the structure of nicotine inspired some of the ideas for the modifications of cytisine on the way to the discovery of varenicline.

- *Random or targeted screening*- Screening refers to the exercise of conducting a biological assay on a large collection of compounds to identify those compounds that have the desired activity. Assays that rapidly measure binding affinities to targets of interest, called *high-throughput screens*, have been commonly used for this purpose.
- *Fragment-based screening*. Several screening methods using, for example, X-ray crystallography or NMR spectrometry have been developed to identify simple molecules (fragments) possessing typically modest affinity for a target, with the intent of connecting two or more of these fragments to create a useful lead compound.
- *Computational approaches.* Given knowledge of the binding site on the target (for example through X-ray crystallography) or of the structure of several known ligands, computational approaches may be used to design potential lead compounds.



Dopamine Cystine Nicotine Vareniciline

Lead Modification (Lead Optimization)

Once one or more lead compounds have been identified, the most notable parameters that may need to be optimized include: potency; selectivity; absorption, distribution, metabolism, and excretion (ADME); and intellectual property position. This process normally involves synthesizing modified versions (analogs) of the lead compound and assessing the new substances against a battery of relevant tests. It is not uncommon to synthesize and test hundreds of analogs in the lead optimization process before a drug candidate (a compound worthy of extensive animal testing) is identified.

(a) *Potency:* Potency refers to the strength of the biological effect, or put another way, how much (what concentration) of the compound is required to achieve a defined level of effectiveness.

(b) *Selectivity:* Unintended sites of action, noted above, refer to interactions with unidentified or unexpected targets. In addition, there may be off-targets that are related to the intended target, with which it would be disadvantageous for the drug to interact.

(c) *Absorption, Distribution, Metabolism, and Excretion (ADME): Absorption* refers to the process by which a drug reaches the bloodstream from its site of administration. *Distribution* refers to what "compartments" in the body the drug goes. *Metabolism* refers to the action of specific enzymes on a drug to convert it to one or more new molecules (called *metabolites*). *Excretion* refers to means by which the body eliminates an unchanged drug or its metabolites through urine, feces, exhalation and other secretions.

(d) *Intellectual Property Position:* Discovering a new drug and bringing it to market is an exceptionally expensive endeavor, to recover the costs and also be able to appropriately compensate investors who are financing the research (and incentivize potential new investors), it is imperative to obtain a patent on a drug that is progressing toward drug development. The patent gives the patent holder the legal means to prevent others from making, selling, or importing the drug, effectively granting the holder a monopoly, for a limited period of time, on selling the drug.

1.6 Concept of prodrugs and soft drugs

The term "prodrug" or "proagent" was introduced by *A. Albert* in 1958 to describe compounds that undergo biotransformation prior to eliciting a pharmacological effect. The formation of a prodrug provides a transient change of physicochemical and biological properties thereby altering or eliminating undesirable properties of the parent drug molecule. This prodrug to drug biotransformation may take place before absorption, during absorption, immediately after absorption, or at a specific site of action. The conversion of prodrugs to the active parent drug molecule can take place through various reactions such as hydrolytic cleavage either as a spontaneous or enzyme-mediated reaction.

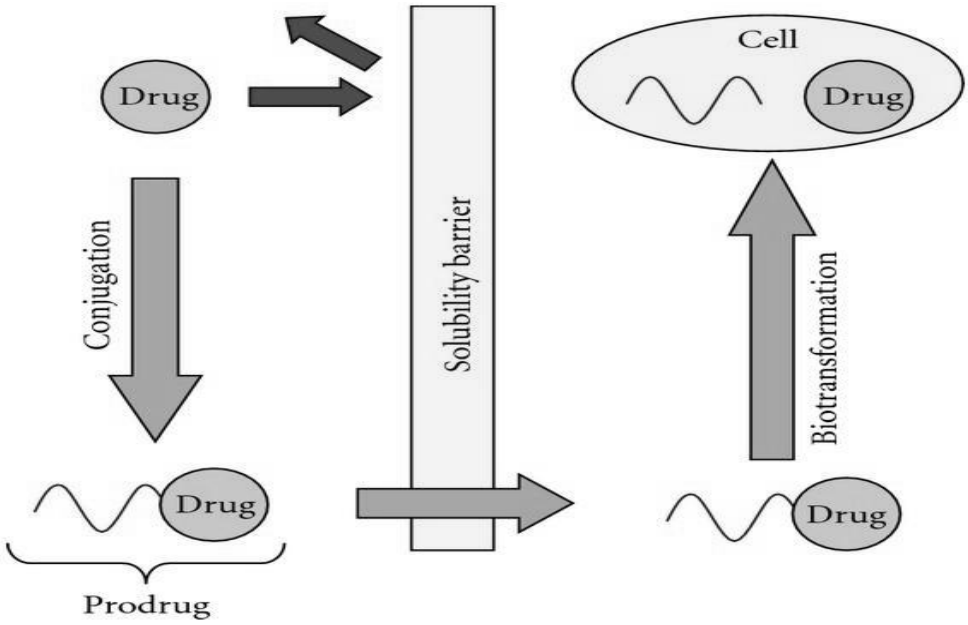


Fig.1.4: Prodrug principle

The prodrug approach has been successfully utilized to overcome significant barriers for many drugs aiming

1. To improve the oral bioavailability by increasing aqueous solubility, by increasing biomembrane permeability, and/or by improving (metabolic) stability
2. To increase the duration of pharmacological action
3. To decrease toxicity or adverse reactions
4. To obtain drug targeting using site-specific biotransformation or site specific transporters.

Types of Prodrugs-

(a) A *carrier-linked prodrug* is a compound that contains an active drug linked to a carrier group that can be removed enzymatically, such as an ester, which is hydrolyzed to an active carboxylic acid-containing drug.

(b) A *bioprecursor prodrug* is a compound that is metabolized by molecular modification into a new compound, which is the active principle or which can be metabolized further to the active drug. For example, if the drug contains a carboxylic acid group, the bioprecursor may be a primary amine, which is metabolized by oxidation to the aldehyde and then further metabolized to the carboxylic acid drug.

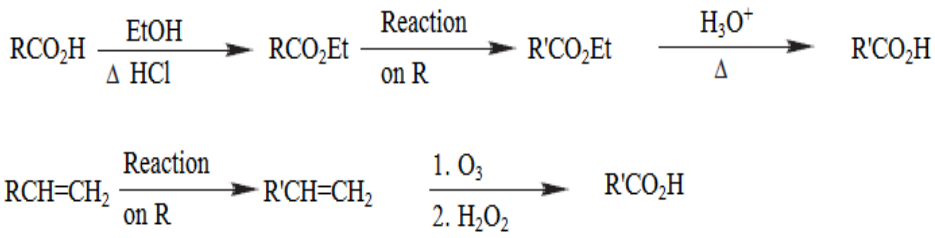


Fig.1.5: Protecting group analogy for a prodrug

Design of prodrugs: chemical considerations-

Prodrugs are most often taken into consideration after identification of a pharmacological active lead compound or structure.

- The first step in prodrug design is identification of functional groups such as hydroxyl, carboxyl, carbonyl, amide, NH-acidic, and/or amino groups in the active compound that are available for chemical derivatization.
- Potentially bioreversible derivatives such as esters, N-acyl, N-hydroxymethyl, or N-acyloxyalkyl derivatives, N-Mannich bases, enaminones, and lactones may be synthesized and subjected to further testing.
- The most important requirement for a prodrug is its ability to adequately regenerate the active drug *in-vivo*.
- It must be chemically stable in the bulk form and together with common excipients used in drug formulation leading to an acceptable shelf-life and, finally, the toxicity of the promoiety and the prodrug itself must be acceptable.

Prodrugs transformed by enzymatic reactions

The most common prodrugs are those requiring hydrolytic cleavage mediated by enzymatic catalysis. Drugs containing hydroxyl, carboxyl, or amino functional groups can be converted into prodrug esters or amides from which the active forms are readily regenerated by hydrolytic enzymes such as esterases, amidases, peptidases, or phosphatases.

Less often prodrugs are designed to undergo reductive or oxidative processes mediated by enzymes such as cytochrome P450, monoamine oxidases, azoreductases, or nitroreductases.

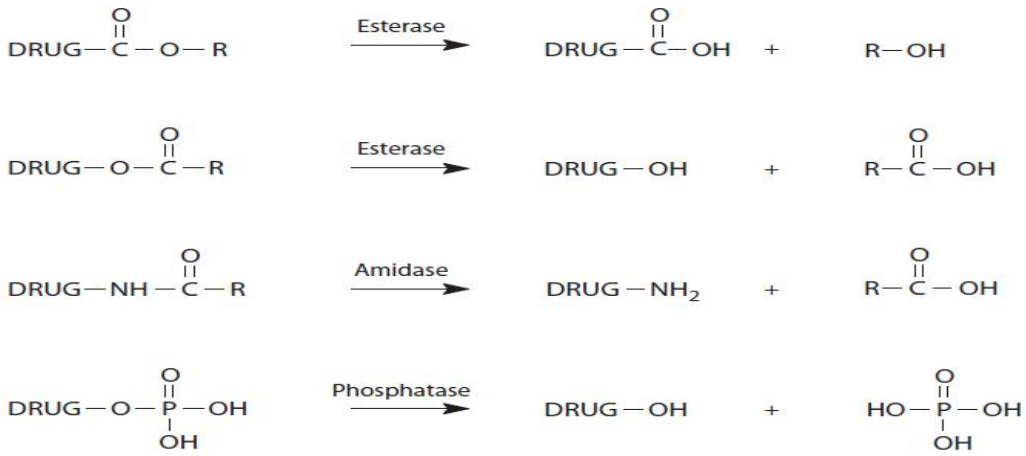
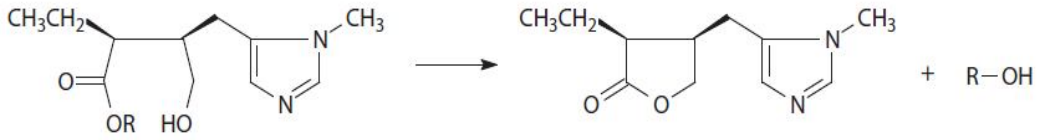


Fig.1.6: Schematic examples of prodrugs designed to undergo enzymatic-mediated hydrolysis

Prodrugs transformed by spontaneous reactions

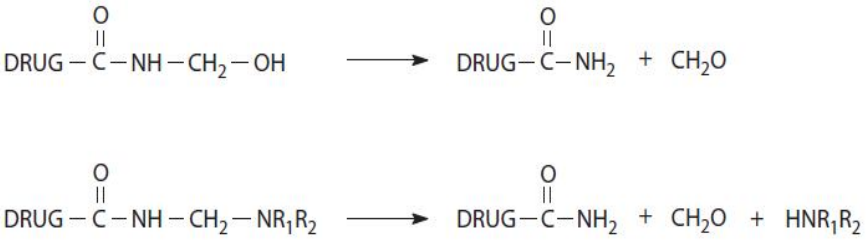
As an alternative to the enzymatic-mediated bioconversion, prodrugs may be designed to undergo spontaneous (or chemical) transformation dictated by the physicochemical environment such as the pH in various parts of the human body.

Some important examples of prodrugs biotransformed by nonenzymatic-mediated reactions are hydroxymethyl derivatives of NH-acidic drug molecules such as 5-fl uorouracil, phenytoin, N-Mannich bases derived from drugs containing amino or amide functions (e.g., tetracycline, carbamazepine) and ring-opened derivatives of cyclic drugs e.g. pilocarpine.



Pilocarpic acid ester Pilocarpine

Figure: Pilocarpic acid ester prodrug cyclization into pilocarpine



Schematic example of prodrugs to undergo spontaneous nonenzymatic transformation

Figure 1.7: Prodrugs transformed by spontaneous reactions

Design of Prodrugs: application of the prodrug principle-

The prodrug principle offers an opportunity for optimization of drug therapy for a variety of reasons. The intention of this section is to illustrate important features of the principle by providing selected examples of achievements by prodrug design and development.

(a) Design of prodrugs with improved bioavailability

The oral bioavailability (F) of a compound is determined by a number of parameters such as aqueous solubility (determines the amount of drug available at the site of absorption), permeability (the ability of the molecule to permeate biological membranes), and stability in the gastrointestinal tract.

Fick's first law of diffusion that describes the rate of diffusion across a (bio) membrane:

$$dQ/dt = P_{app} \times A \times (C - C_o)$$

where *dQ/dt* is the mass flux across the membrane, *A* is the membrane surface area, $C - C_o$ determines the initial concentration gradient between the apical & basolateral side of the epithelium and P_{app} is the permeability coefficient.

Thus, optimization of the flux and thereby the absorption potential is a balance between high aqueous solubility and adequate permeability characteristics. This has been successfully obtained by using the prodrug approach.

a. **Improved Aqueous Solubility**: For compounds suffering from dissolution rate or solubility limited absorption due to low aqueous solubility and/or high therapeutic dose introduction of a hydrophilic moiety may prove beneficial for the oral bioavailability. Example- Amprenavir phosphate (protease inhibitor) to fosamprenavir (ester prodrug).
b. **Improved Biomembrane Permeability (Passive Diffusion):** Due to the large surface area of the gastrointestinal epithelium, transcellular (passive) diffusion is a very important absorption principle for drug molecules. Thus, the ability of a compound to permeate this epithelium is of vital importance for the absorption of the drug. Example- Pivampicillin, talampicillin, and bacampicillin are the pivaloyloxymethyl, the phthalidyl, and the ethoxycarbonyloxyethyl esters, respectively, of the parent penicillin ampicillin.
c. **Improved Transporter-Mediated Permeability:** Various active transport mechanisms for amino acids, small peptides, monocarboxylic acids, monosaccharides, and nucleosides exist in the human body and it is generally recognized that such mechanisms play a major role in the absorption of many drug molecules. This has been used in prodrug

design attempting to provide chemical bioreversible modifi cations mimicking natural substrates for various active transporters with the aim of improving intestinal absorption of various drugs. Example- Levodopa (or l-dopa) is a prodrug of the neurotransmitter dopamine

d. **Increased Stability in the Gastrointestinal Tract:** The poor absorption of carbenicillin after oral administration can to a large extent be attributed to fast acid-catalyzed degradation in the gastrointestinal tract. More acid resistant Prodrugs such as carindacillin and carfecillin have been shown to signifi cantly increase the bioavailability of cabenicillin.

e. **Improved Metabolic Stability:** the prodrug approach can be applied to protect a drug against presystemic metabolism by directly blocking the susceptible part of the molecule or by blocking at an alternative position of the molecule. In both cases, a prodrug that is not a substrate for the metabolizing enzyme may be obtained. Example- Bioactivation of N-(N-acetyl- l-methionyl)-O,O-biscarbonyl dopamine.

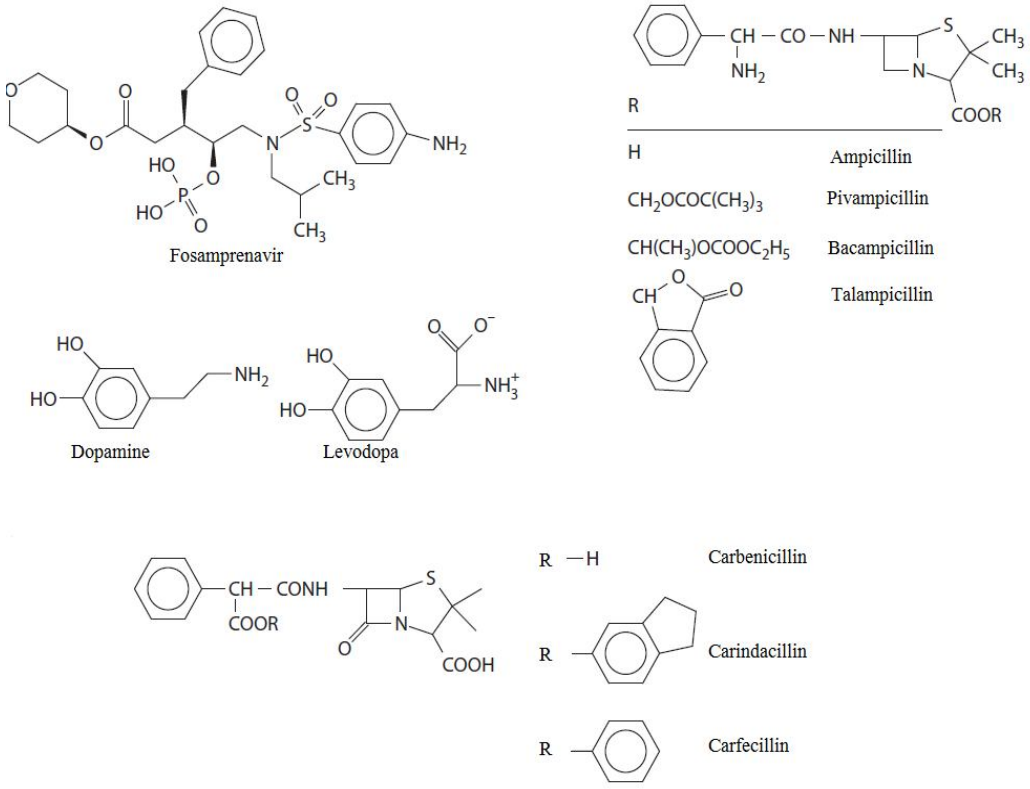
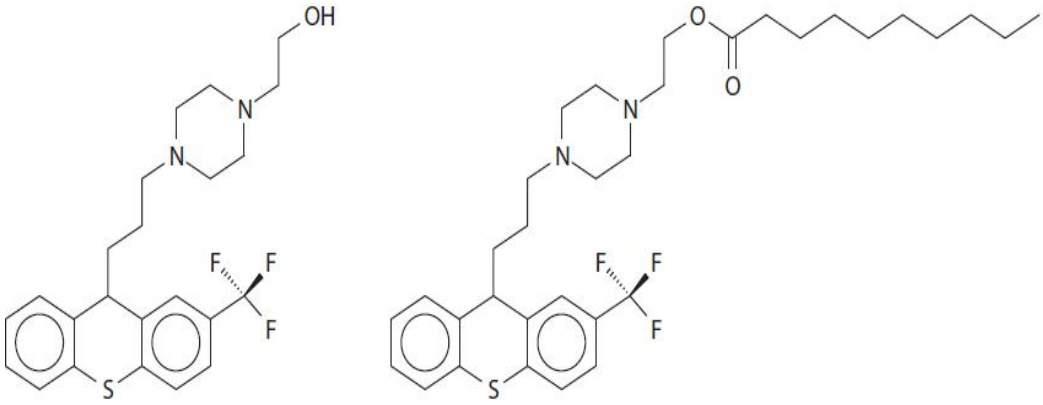


Fig.1.8: Design of prodrugs with improved bioavailability

(b) Design of prodrugs for prolonged drug action

The utility of prodrugs as a means to achieve prolonged pharmacological action of a drug has been effective mainly in terms of intramuscular (IM) injection preparations and other local administrations.

(i) **Intramuscular Depot Injections:** The principle has provided long-term (1–4 weeks) delivery of neuroleptic drugs such as haloperidole, flupentixole, and fluphenazine by IM preparations of highly lipophilic prodrug derivatives such as decanoate and other long-chain fatty acid esters formulated in oily vehicles. This has offered a significant increase in patient compliance and, thus, treatment of patients suffering from psychiatric disorders.

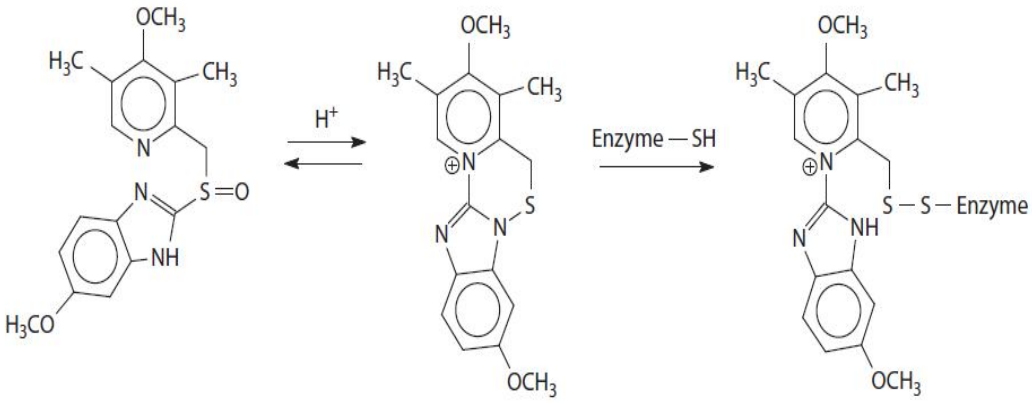


Flipentixole and its decanoate ester

(ii) **Macromolecular Prodrugs:** Conjugation of small drug molecules to high molecular weight promoieties such as polyethylene glycols (PEGs), polysaccharides such as dextrans or other polymers may be used to obtain prolonged drug action. Example- anticancer drugs such as paclitaxel.

(c) Design of produgs for drug targeting

Drug targeting by site-specific bioactivation was achieved using intelligent prodrug design of the blockbuster drug omeprazole, which is widely used in the treatment of gastric ulcers. Omeprazole specifically inhibits the enzyme gastric H+-K+-ATPase that is responsible for the gastric acid production and is located in the secretory membranes of parietal cells. Omeprazole itself does not inhibit this enzyme but is biotransformed within the acid compartments of the parietal cells into the active inhibitor cyclic sulfonamide, which reacts with cysteine thiol groups of the enzyme thereby inactivating it.



Omeprazole- Bioconversion and mechanism of action

Fig.1.9: Bioconversion and mechanism of action of Omeprazole

1.7 Summary

Brief overview and synopsis of points in drug discovery and development from screening and target identification through to drug approval and the potential for generics Armed with their understanding of the disease, scientists are ready to begin looking for a drug. They search for a molecule, or "lead compound," that may act on their target to alter the disease course. If successful over long odds and years of testing, the lead compound can ultimately become a new medicine. Lead compounds that survive the initial screening are then "optimized," or altered to make them more effective and safer. By changing the structure of a compound, scientists can give it different properties. For example, they can make it less likely to interact with other chemical pathways in the body, thus reducing the potential for side effects. For the purpose of this summary, drug development is defined as those activities required for advancing a biomedical concept to a medical product in commerce.

1.10 Glossary

- Drug design is the approach of finding drugs by design, based on their biological targets. Typically a drug target is a key molecule involved in a particular metabolic or signalling pathway that is specific to a disease condition or pathology, or to the infectivity or survival of a microbial pathogen.
- Drug development or preclinical development is defined in many pharmaceutical companies as the process of taking a new chemical lead through the stages necessary to allow it to be tested in human clinical trials,

although a broader definition would encompass the entire process of drug discovery and clinical testing of novel drug candidates.

- Computer Assisted Drug Design (CADD) is a three-dimensional puzzle where small drug molecules, ligands, are adjusted to the binding site of a protein.
- The process of drug discovery involves the identification of candidates, synthesis, characterization, screening, and assays for therapeutic efficacy.
- Ideal drug: Given by mouth and has a beneficial effect (safe & efficacious) in only-50%
- Promising drug candidate: Most site specific with best combination of target affinity, highest bioavailability and lowest toxicity.
- Membrane transport is mediated by specific integral membrane proteins – ion channels, porins, transporters (passive), pumps (active)
- Drug delivery is the method or process of administering a pharmaceutical compound to achieve a therapeutic effect in humans or animals.

1.11 Review questions / Comprehensive Questions

1. Define drug design. Jutify the following statements :Drug design aims at developing a drug with high degree of chemotherapeutic index and specific action.
2. Enumerate various steps of drug design. And development.
3. Discuss the various 'factors governing drug-design'.
4. Eloborate the 'rational approach to drug design'.
5. Describe Drug discovery without rational design.
6. 'Tailoring of Drugs' is the outcome of anunique blend of skill involving various configurational and stereochemical changes attributing its flexibility and overall dimension. Explain.
7. Discuss in detail Concept of lead compounds and lead modification.
8. Discuss the various possible approaches in designing newer drugs.
9. Discuss the basic concepts of 'prodrugs' with the help of suitable examples of parent drug molecule(s).
10. Write various applications of Prodrug.

1.12 References and Suggested readings

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Unit-2

Structure Activity Relationship

Structure of Unit:

2.1 Objectives

In this unit the students will be able to understand

- Structure activity relationship
- Ferguson principle
- Factors affecting biological activity
- Resonance and inductive effect
- Stereochemistry and its type
- Concept of isosterism and bioisoterism

2.2 Introduction: Structure Activity Relationship

A Structure-Activity Relationship (SAR) is the relationship between the chemical or 3D structure of a molecule and its biological activity. The aim of SAR is to identify the physicochemical properties of a drug and to see whether any of these properties has an effect on the drug's biological activity. If such a relationship holds true, an equation can be drawn up which identifies the relationship and allows the medical chemist to say with confidence that the property (or properties) has an important role in pharmacokinetics or

mechanism of action of drug. It also allows the medical chemist to some level of prediction. By identifying physicochemical properties, it should be possible to calculate in advance what the biological activity of novel analogue might be. There are two advantages of this. First, it allows the medical chemist to target efforts on analogues which should have improved activity and thus cut out the number of analogue that has to be made. Second, it allows modification of the effect or the potency of a bioactive compound (typically a drug) by changing its chemical structure. Medicinal chemists use the techniques of chemical synthesis to insert new chemical groups into the biomedical compound and test the modifications for their biological effects.

In an effort to interpret the SAR of a drug, two main approaches have emerged via (1) The group and moiety approach and (2) Integral approach.

The former places emphasis on significance of certain chemical groups in the drug molecule as a whole and particularly concerned with overall physico-chemical properties. Modulating the structure of drug implies introduction, elimination or substitution of certain groups in the drug. This may lead to the development of parallel drug with characteristics similar to the lead compound like vitamin analogues and hormone analogues. Hence the activity is maintained, although structure is changed. This can be expressed by an idea of bio-isosteric groups which generally have similar biological activity. The spectrum of action of the existing compound may be changed or side effects can be changed to main effects. E j. Ariens mentioned the following physico-chemicals parameters affecting drug-activity. The chemical properties of a drug are determinant for its biological action and activity. The various physico-chemical properties of bioactive compound in general, are parameters related to the interaction of the drug with its environment.

2.3 Ferguson Principle

Pharmacologically active compounds can be divided into two major groups:

(a) Structurally specific and

(b) Structurally non specific.

The structurally specific drugs bring about their effects by combining with a specific receptor. The SAR of such groups can only be varied with relatively narrow limits.

The structurally non specific drugs do not act on specific receptor. Instead, they penetrate into the cell or accumulate in cellular membranes, where they interfere by chemical or physical means, with some of the fundamental cellular

processes e.g. general anaesthetics, hypnotics, volatile insecticide and certain bacterial agents. The biological effect of such drugs is more closely correlated with the physical properties of the molecule than with the chemical structure e.g. cyclopropane, diethyl ether and chloroform, though having different structures, good general anaesthetics.

Ferguson suggest in 1939, that the potency of structurally none specific drugs was determined by their thermodynamic activity. His quantity is a measure of the proportion of the molecules which are free to react with enzymes system, nerve membranes and similar biologically important sites. The molecule which are not free to act in this way, are reacting with one another, with molecule of other solutes. It follows, therefore, that the thermodynamic activity of a drug in solution is not determine entirely by its concentration. In the case of volatile anaesthetics administer with air or oxygen, the thermodynamic activity is proportional to the relative saturation of a drug.

Ferguson theory predicts that the anaesthetic agents will show the same degree of biological activity if there concentrations are adjusted so that there thermodynamic activities are equal. This theory is also applicable to substances other than anaesthetics and it was originally applied to insecticides and antibacterial substances.

The physico-chemical parameters can be divided into three categories.

1) **Parameters which are an expression of the hydrophobic aggregation forces at site of action :**

   These include partition coefficient, surface activity, R_f value and the partial vapour pressure of the solution. Hydrophobic groups and hydrophobic aggregation forces represented by these parameters give relatively large contribution to binding energy. They allow, by variation in size of groups, a gradual change in lipid water balance which rules distribution by passive transport.

2) **Parameters which are an expression of the charge distribution in the molecule and thus of electrostatic force at site of action:**

   These include the redox potential, the base or acid dissociation constants, the electronic polarization, dipole moments, the inductive field effect and the resonance effect, especially in conjugated systems, the capacity of chelate formation, and H-bond formation and finally the characteristics in IR and NMR spectra. Electrostatic forces represented by theses parameters give a relatively low contribution to binding energy. They contribute more to the selectivity in drug-receptor

interaction and are essentially involved in substrate activation in enzymes and conformational skeletal important for induction of a stimulus in the macromolecular receptor molecules.

3) **Parameters which are an expression of spatial arrangement of the molecule:**

   These parameters represent spatial arrangement of various groups in the molecule and play a role in the possible steric hindrance on the intramolecular level. The location, size, volume and charge of particular group play a role here.

The intensity of pharmacological response elicited by many drugs is probably directly related to the concentration or activity of a drug in the immediate vicinity of the receptor site in the body. Since it is not possible to measure this concentration directly, the study of physico-chemical parameters presents a picture of indirect measurement of concentration of a drug at receptor site. It follows, therefore, that drug molecules exert their effect by influencing receptor sites in living systems through their physico-chemical properties.

2.4 Factors affecting bioactivity: Resonance Effect

Factor affecting bioactivity:

Various aspect of bioactivity applies to any chemical or synthetic origin. The aspect will be dealt as follows:

- Physicochemical properties: These are related to the transport of bioactive compound to its site of action, usually a receptor or other biomolecule at the cellular level. Under experimental conditions, either *in vivo* or clinical, or real life conditions, the extent to which a drug passes through semi-permeable membranes before reaching its site of action depends on its solubility. After the bioactive molecular entity has been identified, detailed data on solubility, partition coeficients and the electrolytic behaviour can be determined.
- Chemical parameters: The structural features of a compound can be related to its pharmacological properties, either qualitatively or quantitatively. The principles, concepts and numerical rules governing qualitative and quantitative relationships between structure and activity help explain the pharmacological activity of a new compound. It is important to evaluate the structure of a newly isolated plant compound. The basic aspect of molecular structures involved in bioactivity include: Resonance and Inductive effect.

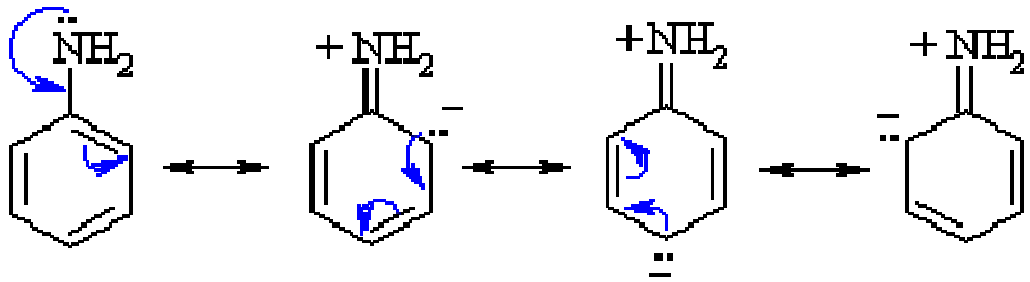
- Resonance is a phenomena that a molecule can be represented by two or more structures that differ only in their electron but not atomic arrangement. The electron density and electron distribution patterns explain the molecules activity.
- Inductive effect is a electro static phenomena caused by actual electron shifter displacements along chemical bonds. Both negative and positive inductive effects can lead to change bioactivity.
- Types of bonding: The phenomenon of biological activity is concerned with covalent and non-covalent interactions. Covalent bonds are formed enzymatically and are common to all biomolecules. Hydrogen bonds, ionic forces, hydrophobic bonding, charge-transfer interactions, all representing non-covalent interactions are also common to functional life processes.
- Spatial arrangement of the molecule : In terms of activity, it is important to have a good steric and electronic complementary between ligand and target bio molecule. Bioactive compounds interact with enzymes by fitting sterically into a binding pocket the space sterically provided by these targets.
- Thus the molecular dimensions, inter atomic distances, arrangements of electrons and the stereochemical properties of both ligand and target are decisive in determining biological activity

Resonance Effect

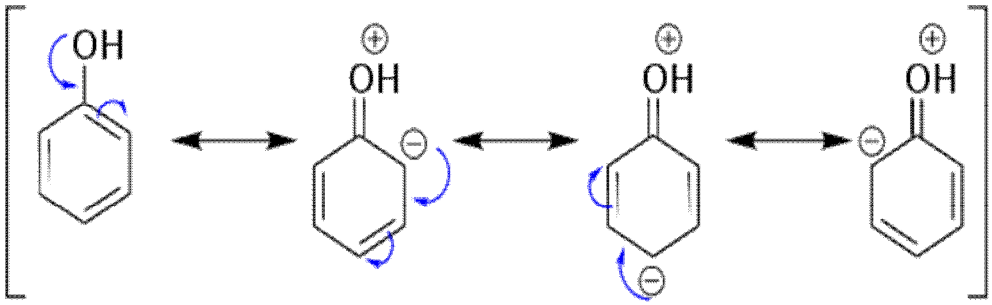
It has been observed that 'resonance' give rise to an altogether different distribution of electron density than would be the situation if there existed absolutely no resonance.

Example: The resonance effects, as observed in *two* electron donating functional moieties, such as: - (a) NH_2 (amino); and (b) –OH (hydroxyl), attached to an aromatic nucleus.

(a) Resonance structure of aniline



(b) Resonance structure of phenol



Explanation

For Resonance Structures of Aniline (a): In case, the first structure is the actual structure of aniline, the two unshared electrons of the N- atom would certainly reside exclusively on that particular atom. However, in true sense and real perspective the first structure is not the ideal and only structure for aniline but a hybrid one which essentially includes contribution from several canonical forms as shown, wherein the density of electrons of the unshared pair does not reside necessarily on the N-atom but get spread out around the phenyl ring. In nut shell, this observed density of electrons at one particular position (with a corresponding enhancement elsewhere) is invariably known as the 'resonance' or 'mesomeric effect'.

For Resonance Structure of Phenol (b): Here, the influence of R at the *para* position, and the electron-donating effect due to resonance is more marked, pronounced and significant as compared to the electron withdrawing influence due to induction.

Inductive Effect

The electronic effect of various substituents will clearly have an effect on a drug's ionization or polarity. This in turn may have an effect on how a drug can pass through cell membranes or how strongly it can interact with a binding site. It is therefore necessary to measure inductive effect of a substituent. **Hammett substitution constant** (σ) measures the electron withdrawing or electron donating ability of a substituent, and with reference to a specific substituent is usually defined by following equation:

$$\sigma_X = \log K_X = \log K_X/K_H - \log K_H$$

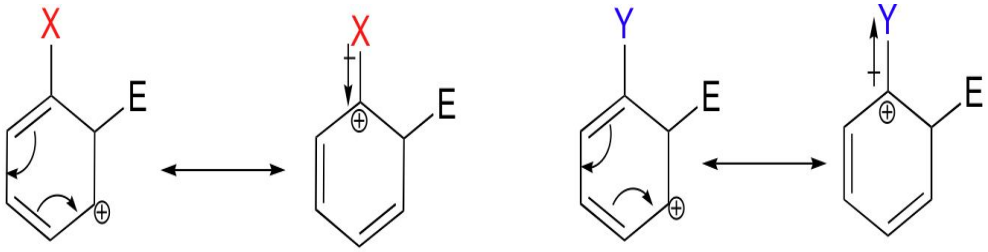
Hammett substitution constant may be determined conveniently by actual measurement of the dissociation of a series of substituted benzoic acids compared to the dissociation of benzoic acid itself. Benzoic acid is a weak acid and only partially ionizes in water. An equilibrium is setup between the ionized

and non-ionized forms, where the relative proportions of these species are known as equilibrium or dissociation constant K_H (the subscript H signifies that there are no substituents on the aromatic ring).

$$K_H = \frac{[PhCO^-_2]}{[PhCO_2H]}$$

When a substituent is present on the aromatic ring, this equilibrium is affected. Electron-withdrawing group, such as NO_2, CN, COOH, $CONH_2$, CONHR, $CONR_2$, CHO, COR SO_2R, SO_2OR, NO; results in aromatic ring having a strong electron withdrawing and stabilizing influence on the carboxylate anion. The equilibrium will therefore shift more to the ionized form such that the substituted benzoic acid is a stronger acid and has a larger K_X value.

If the substitution X is an electron-donating group, such as R, Ar, F, Cl, I, Br, SH, O^-, S^-, NR_2, NHR, NHCOR, OR, OH, OCOR; then the aromatic ring is less able to stabilize the carboxylate ion. The equilibrium shifts to the left and a weaker acid is obtained with a smaller K_X value.



X = electron donating group:
stabilizes the intermediate,
activates the ring

Y = electron withdrawing group:
destabilizes the intermediate,
deactivates the ring

Benzoic acids containing electron-withdrawing substituents will have larger K_X values than benzoic acid itself (K_H) and therefore the value of σ_X for an electron-withdrawing substituents will be positive.

Benzoic acids containing electron-donating substituents will have smaller K_X values than benzoic acid itself (K_H) and therefore the value of σ_X for an electron-donating substituents will be negative.

2.6 Stereochemistry

Stereoisomers are compounds containing the same number and kind of atoms, the same arrangement of bonds, but different three-dimensional structures; in other words, they only differ in the three-dimensional arrangements of atoms in

space. Stereo isomers are subdivided into two types, enantiomers and diastereoisomers.

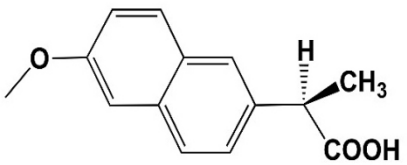
Enantiomers

Enantiomers are the compounds whose 3-D arrangement of atoms is such that they are nonsuperimposible mirror images. These are also termed as chiral compounds, enantiomers or antipode. These compounds posses identical physical as well as chemical properties except their ability to rotate plane polarized light in just opposite directions with almost equal magnitude, quantum and extent. Predominantly, when enantiomeric features are introduced strategically right into either a chiral environment or an asymmetric one, for instance: the human body, enantiomers shell evidently show marked and pronounced variant physical chemical properties thereby exhibiting appreciable and significant differences in their respective pharmacokinetic and pharmacodynamic behaviour.

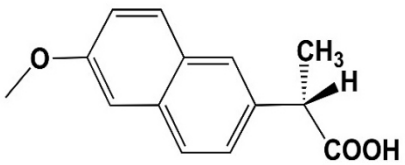
Thus, the presence of variant biological activities based on their diverse enantiomeric features in a drug substance may lead to:

- adverse side effects,
- toxicity caused due to one of the isomer,
- exhibit appreciable difference in absorbtion,
- show significant variation in serum protein binding,
- extent /degree of metabolism,
- conversion into a toxic substance (impaired metabolism),
- influence the metabolism of an altogether another drug.

For example naproxen sodium shows that the priority sequence in (S)-(+) naproxen sodium is to the left; and it exhibit activity as an antipyretic, analgesic and anti-inflammatory drug. In contrast, the (R)-(-) naproxen sodium is an inactive.



(*R*)-Naproxen

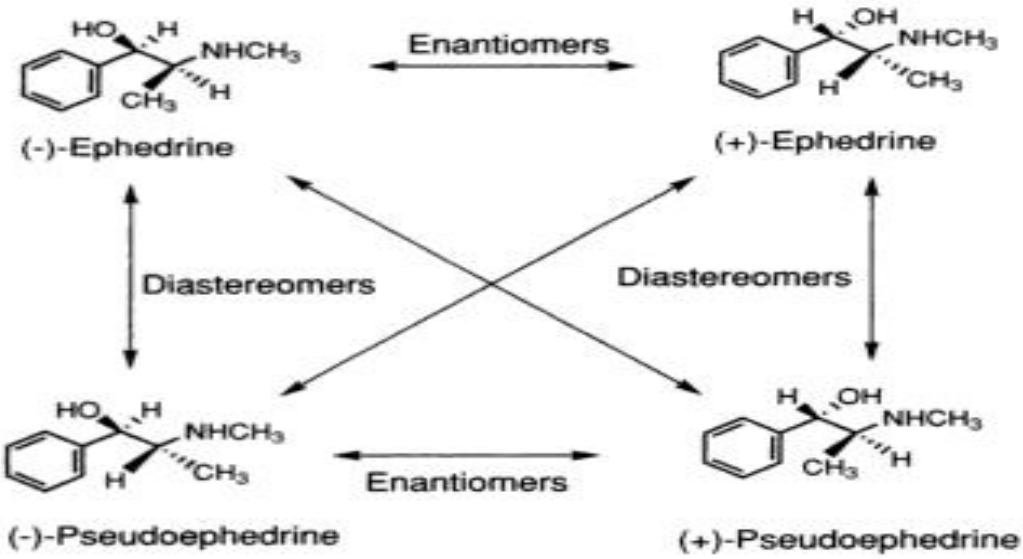


(*S*)-Naproxen

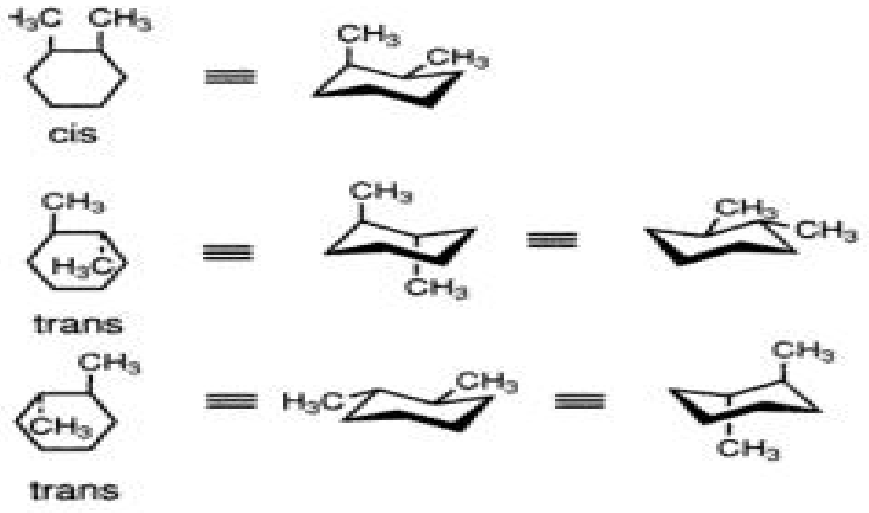
Diastereoisomers

Distereoisomers are all stereoisomeric compound which are not enantiomers. Thus, the term "diastereoisomer" includes compound containing both ring systems and double bonds simultaneously. Unlike enantiomers, diastereoisomers exhibit different physico-chemical properties, including melting point, boiling point, solubility and chromatographic behavior. These differences in physico-chemical properties allow the separation of diastereoisomers from mixtures utilizing standard chemical separation techniques such as column chromatography or crystallization. These isomers have different physico-chemical properties; thus, differences in biological activity.

Compound containing more than one chiral centre probably are the most common type of diastereoisomers used as drugs e.g. ephedrine and pesudoephrdrine. In (-) pseudoephrdine the two H-atoms are on the opposite side of the plane of the ring. However, in (-) ephedrine the said to two H-atoms are located on the same side i.e., beneath the ring plane; and hence, it shows biological activities (CNS-stimulatory actions).



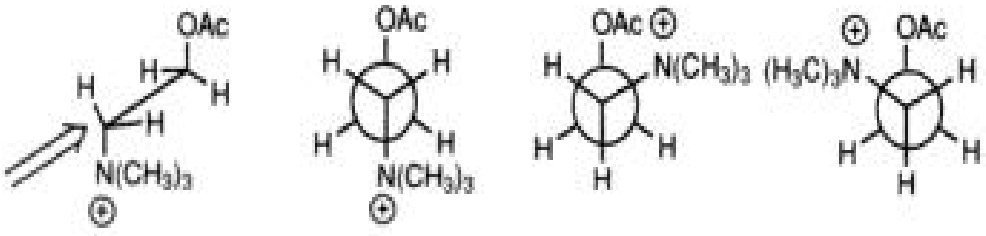
Diastereoisomers also can be found in cyclic compounds. For example, the cyclic alkane 1,2-dimethylcyclohexane can exist as *cis/trans*- diastereoisomer, and trans isomer also can exist as an enantiomeric pair. Each of trans-enantiomorphs are depicited in the two possible chair conformation for the cyclohexane ring. Cyclohexane ring can exhibit significant conformational freedom that allows for the possibility of conformational isomers.



Conformational Isomers

It is a dynamic process i.e, isomerization take place by rotation about one or more single bonds. Such bond rotation results in nonidentical spatial arrangement of atoms in molecule. This results in different conformation, whereas conversion of one enantiomer into another requires breaking of bonds, which has a higher energy requirement than rotation about single bond. For example, acetylcholine, each single bond within acetylcholine molecule is capable of undergoing rotation, and at room temperature, such rotation readily occur.

Acetylcholine



anti or staggered conformer

gauche or skew conformers

2.7 Isosterism

When a lead compound is first discovered for a particular disease state, it often lacks the required potency and pharmacokinetic suitable for making it a viable clinical candidate. The medical chemist therefore must modify the compound reduce the undesirable features without losing the desired biological activity. Replacement or modification of functional group with other groups having similar properties is known as 'isosteric' or 'biosteric replacement'.

Langmuir suggested that any two ions or molecules possessing essentially an identical number and arrangement of electrons must exhibit similar characteristics; e.g. CO and N; CO_2 and NO_2; and N^-_3 and NCO^-. Such isosteres which are isoelectric in nature must show good similarity in properties. To account for similarities between groups with the same number of valance electron but different numbers of atoms, Grimm developed his hydride displacement law which shows similar physical properties among closely related functional groups. Instead of considering only partial structures, Hinsberg applied the concept of isosterism to entire molecule and developed the concept of "ring equivalents" –that is, groups that can be exchanged for one another in a aromatic ring systems without drastic changes in physiochemical properties relative to the parent structure. Benzene and thiopene and pyridene illustrate this concept. A –CH=CH-group in benzene is replaced by divalent sulphur, -S, in thiophene, and a –CH= is replaced by trivalent –N= to give pyridine. The physical properties of benzene and thiopene are very similar than that of pyridine. For e.g., boiling point of benzene is 81.1^oC, thiophene is 84.4^oC while that of pyridine is 116^oC. Hinsberg therefore concluded that divalent ether -S must resemble –C=C in shape, these groups were considerd to be isosteric.

2.8 Bioisosterism

Bioisosteres are compounds or groups that essentially posses equal or near equal molecular shapes and volumes, approximately the same distribution of electrons and that exhibit similar physical characteristics such as hydrophobicity. Bioisostereic compounds affect the same biochemically associated systems as agonists and antagonists and thereby produce biological properties that are related to each other. It may be classified under two categories –classical and non-classical bioiosteres.

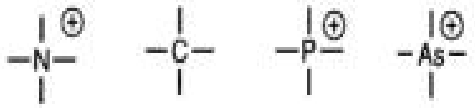
I. Classical bioisosteres

a) Monovalent atoms and groups
F, H, OH, NH, F, OH, NH OR CH_3 for SH, OH, Cl, Br, CF_3

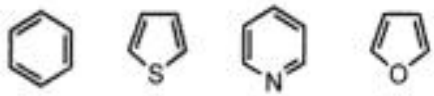
b) Divalent atoms and groups
-C=S, -C=O, -C=NH, -C=C-

c) Trivalent atoms and groups
-C=, -N=, -P=, -As=

d) Tetrasubstituted atoms



e) Ring equivalents



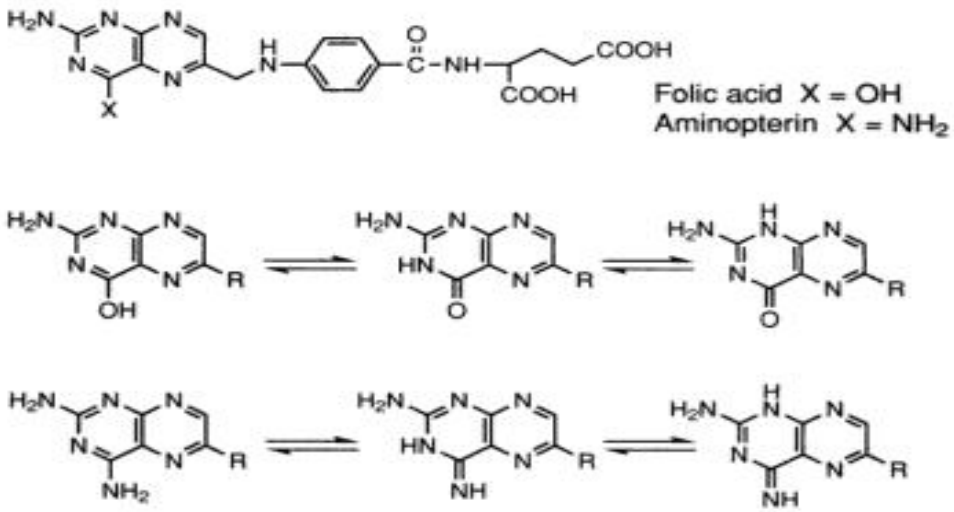
II. Non-classical bioisosteres

a) Exchangeable groups
b) Ring versus noncyclic structure

I. Classical bioisosteres

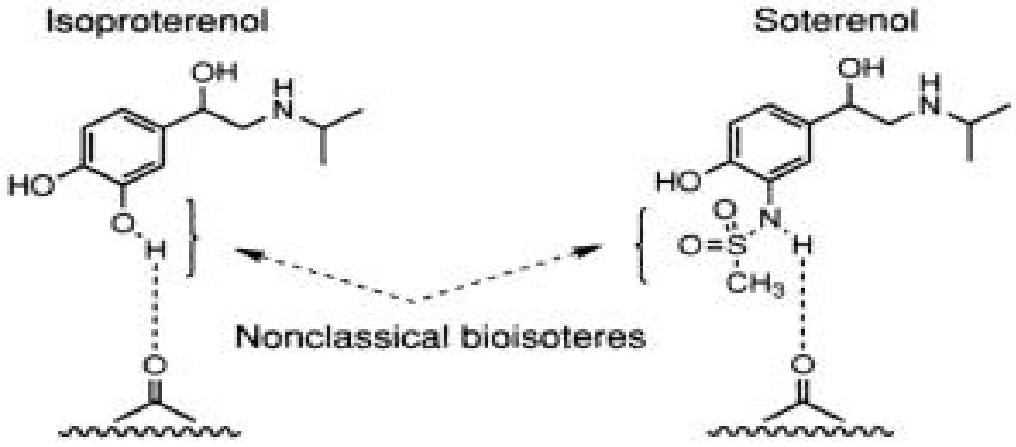
Functional groups that satisfy the original conditions of Langmuir and Grimm are referred to as classical bioisosteres. Substitution of hydrogen by fluorine is one of the most common

Monovalent isosteric replacement. A classical example of hydrogen replacement nby fluorine is development of antineoplastic agent 5-fluorouracil from uracil.



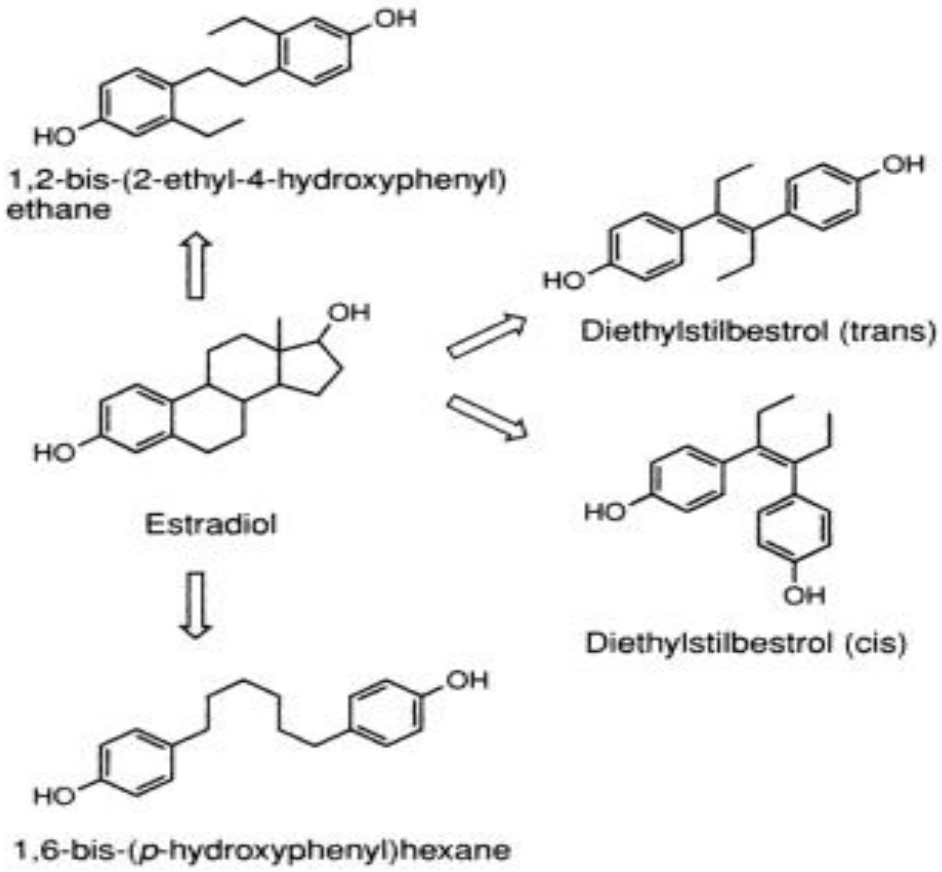
II. Non-classical bioisosteres

Non-classical bioisosteres do not obey electronic and steric definitions of classical bioisosteres and donot necessarily have the same number of atoms as the substituent they have the same of atoms as the substituent they replace. For example, non classical replacement of a sulphonamide group for a phenol in catecholamines. In this, steric factor appears to have less influence on receptor binding than acidity and hydrogen bonding potential of functional group on aromatic ring. Both groups are weakly acidic and capable of losing a proton and interacting with the receptor as anions or hydrogen bond donars at receptor.



Another example is the use of double bond to position essential functional groups in spatial configuration critical for activity this is shown with the naturally occurring hormone estradiol and the synthetic analogue diethylstilbestrol. The *trans* isomer of diethylstilbestrol has the same potency as estrsdiol, where *cis* isomer is only one-fourteenth as active. In *trans*

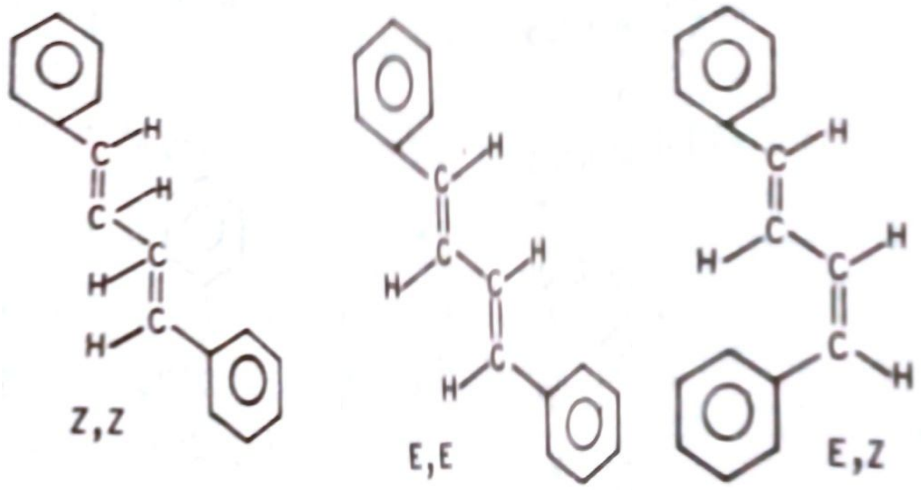
configuration, the phenolic hydroxyl group group mimic the correct orientation of the phenol and alcohol in estradiol. This is not possible with the *cis* isomer, and more flexible analogues have little or no activity.



2.9 Spatial arrangement

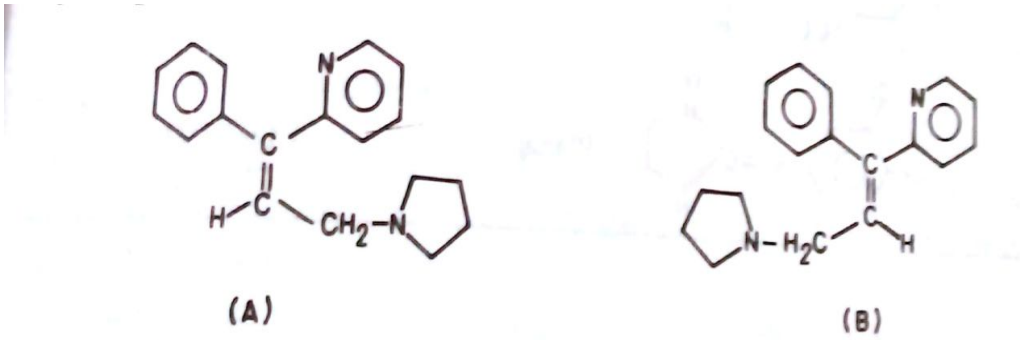
1. Planarity

It has been known that a series of closely related compounds differing in relative planarity are adsorbed on non biological medial to different extents. For example, in the case of three geometrical isomers of 1,4-diphenylbutadiene, the order of elution from an aluminium oxide column was the Z, Z isomer followed by the E, Z isomer and finally E, E isomer. In other words, the greatest absorbtion occurred with the most planar compound. Thus it was reasoned that differences in planarity may account for differences in bioactivities in closely related compounds.



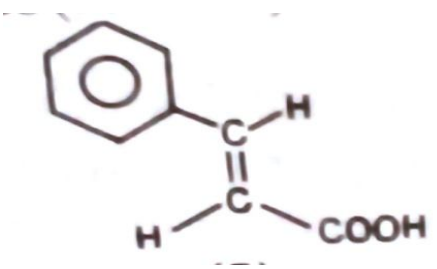
In some cases coplanarity of important functional groups is associated with marked bioactivities, e.g., certain antihistaminic compounds, and in the case of Bipyridine herbicides. On the other hand, examples may be found where a lack of coplanarity of important groups is necessary for significant biological activity, e.g., cinnamic acids are useful as plant growth hormones and clonidine and related compounds.

For example in certain antihismatic drugs in isomer (A) the 2-pyridyl function is coplanar with the olefinic double bond, whereas in isomer (B) there is no planarity of these two groups. The isomer (A) is far more active as an antihistaminic and this principle of coplanarity of the olefinic double bond and the heterocycle is found through this series of compounds.

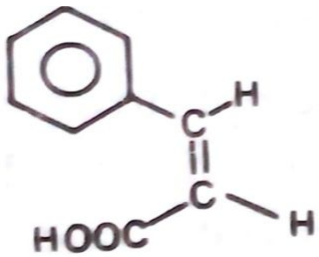


(2) Cinnamic acids:

They are useful as plant growth hormones and in the series of compounds, the aryl ring and the COOH group must not be coplanar. For example, E-cinnamic acid which is a planar moleculeis inactive.



E- Planner molecule
(inactive)



Z- Non planner molecule
(active)

Delocalized chemical bonding involves resonance for which conjugation is must. The resonance depends upon the substituent's at aromatic system. Hyperconjugation increases the stability of intermediates i.e. Carbocation and free radicals. *Tautomers* are isomers of a compound which differ only in the position of the protons. Keto form is more stable than enol form. Tautomerism mechanism observed in acid as well as in basic medium. Due to difference in electronegativities between two atoms forming an σ- bond gives Inductive effect.

2.10 Summary

Medicinal chemistry involves the discovery of new chemical entites for the treatment of disease and the systematic study of SARs of these compounds. Such studies provide the basis for development of better medicinal agents from lead compounds found via random screening, and rational design. The role of medicinal chemist is to increase the potency and duration of action of newly discovered compounds and to decrease adverse side effects. Without a thorough understanding of physico-chemical properties of organic functional groups that compromises any given structure. For a pharmacist, it is also important to understanding physico-chemical properties of medicinal agents that is to be dispensed. Such knowledge will help the practicing pharmacist not only to better understand the clinical properties of these compounds and also of that which are appearing in market.

2.11 Glossary

- A Structure-Activity Relationship (SAR) is the relationship between the chemical or 3D structure of a molecule and its biological activity.
- The SAR of a drug has two main approaches via (1) The group and moiety approach and (2) Integral approach.

- Pharmacologically active compounds can be divided into two major groups:
  (a) Structurally specific and, (b) Structurally non specific.
- The structurally specific drugs bring about their effects by combining with a specific receptor while structurally non specific drugs do not act on specific receptor. Instead, they penetrate into the cell or accumulate in cellular membranes, where they interfere by chemical or physical means, with some of the fundamental cellular processes
- Resonance gives rise to an altogether different distribution of electron density.
- The electronic effect of various substituents has an effect on a drug's ionization or polarity.
- Hammett substitution constant (σ) measures the electron withdrawing or electron donating ability of a substituent, and with reference to a specific substituent alkyl groups give +I effect while other electronegative groups give –I effect. The Hammett constant take into account both resonance and inductive effects.
- Stereoisomers are compounds containing the same number and kind of atoms, the same arrangement of bonds, but different three-dimensional structures.
- Enantiomers are the compounds whose 3-D arrangement of atoms is such that they are nonsuperimposible mirror images. These are also termed as chiral compounds, enantiomers or antipode.
- Distereoisomers are all stereoisomeric compound which are not enantiomers. Thus, the term "diastereoisomer" includes compound containing both ring systems and double bonds simultaneously. Unlike enantiomers, diastereoisomers exhibit different physico-chemical properties, including melting point, boiling point, solubility and chromatographic behavior.
- Replacement or modification of functional group with other groups having similar properties is known as isosteric or biosteric replacement.
- Bioisosteres are compounds or groups that essentially posses equal or near equal molecular shapes and volumes, approximately the same distribution of electrons and that exhibit similar physical characteristics

- It may be classified under two categories –classical and non-classical bioiosteres.
- Functional groups that satisfy the original conditions of Langmuir and Grimm are referred to as classical bioisosteres.
- Non-classical bioisosteres do not obey electronic and steric definitions of classical bioisosteres and donot necessarily have the same number of atoms as the substituent they have the same of atoms as the substituent they replace.
- a series of closely related compounds differing in relative planarity are adsorbed on non biological medial to different extents.

2.12 Review questions / Comprehensive Questions

1. Define structure activity relationship.
2. Explain the Ferguson principle.
3. What is cross conjugation? Explain with example.
4. What is resonance? How it effect the biological activity?
5. Explain the inductive effect.
6. Explain the types of stereoisomers with examples.
7. How spatial arrangement of compounds effects biological activity of compounds?
8. Give a comprehensive account of importance of 'Isosterism and Bioisosterism' in drug design.

2.13 References and Suggested readings

- Foyes Principle of Medicinal Chemistry (6th ed.)- Thomas L Lemke and DA Williams (Lipincott Williams & Wilkins).
- An Introduction to Medicinal Chemistry- Graham L. Patrick (Oxford University Press) 2006.
- Medicinal Chemistry (5th ed.)- Ashutosh Kar (New Age International Publisher) 2007.
- Principle of Medicinal Chemistry (18th ed.)- SS Kadam, KR Mahadik and KG Bothra (Nirali Prakashan) 2007.
- An Introduction to Drug Design- S.N. Pnadey and J.R. Dimmock (New Age International Publisher) 2011.

Unit-3

Theories of Drug Activity

Structure of Unit:

3.1 Objectives

In this unit the students will be able to understand

- Drug-Receptor Theory
- The Operational Model of Receptor Function
- Important Interactions (Forces) Involved in the Drug–Receptor Complex
- Determination of Drug–Receptor Interactions
- The Use of Mathematical Models in Pharmacology
- Pharmacodynamics of general receptors

3.2 Introduction: Theories of Drug Activity

Principles of drug action

Drugs (except those gene based) do not impart new functions to any system, organ or cell; they only alter the pace of ongoing activity. The basic types of drug action can be broadly classed as:

1. Stimulation refers to selective enhancement of the level of activity of specialized cells, e.g. adrenaline stimulates heart, pilocarpine stimulates salivary glands.
2. Depression means selective diminution of activity of specialized cells, e.g. barbiturates depress CNS, quinidine depresses heart.
3. Irritation connotes a nonselective, often noxious effect and is particularly applied to less specialized cells (epithelium, connective tissue).
4. Replacement refers to the use of natural metabolites, hormones or their congeners in deficiency states, e.g. levodopa in parkinsonism, insulin in diabetes mellitus, iron in anaemia.
5. Cytotoxic action for invading parasites or cancer cells, attenuating them without significantly affecting the host cells is utilized for cure/palliation of infections and neoplasms, e.g. penicillin, chloroquine, zidovudine, cyclophosphamide, etc.

Majority of drugs produce their action by interacting with a discrete target biomolecule, which usually is a protein. Such mechanism confers selectivity of action to the drug. Functional proteins that are targets of drug action can be grouped into four major categories, viz. enzymes, ion channels, transporters and receptors.

The largest number of drugs does not bind directly to the effectors, viz. enzymes, channels, transporters, structural proteins, template biomolecules, etc. but act through specific regulatory macromolecules which control the above listed effectors. These regulatory macromolecules or the sites on them which bind and interact with the drug are called 'receptors'.

Receptor is defined as a macromolecule or binding site located on the surface or inside the effector cell that serves to recognize the signal molecule/ drug and initiate the response to it, but itself has no other function. Though, in a broad sense all types of target biomolecules, including the effectors (enzymes, channels, transporters, etc.) with which a drug can bind to produce its action have been denoted as 'receptors' by some authors, such designation tends to steal the specific meaning of this important term.

Receptors have been divided into four major superfamilies: GPCRs, ligand-gated ion channels, tyrosine kinase receptors, and nuclear receptors. The first three receptor superfamilies are located in the cell membrane and the latter

family is located intracellularly. GPCRs are the largest class of receptors known; about 800 different human genes (~4% of the human genome) are predicted to be members of the GPCR family.

Pharmacological stimulation or inhibition of the earlier mentioned synaptic mechanisms are, however, likely to affect the function of the entire neurotransmitter system. Activation of neurotransmitter receptors may, in principle, represent the most direct and selective approach to the stimulation of a particular neurotransmitter system.

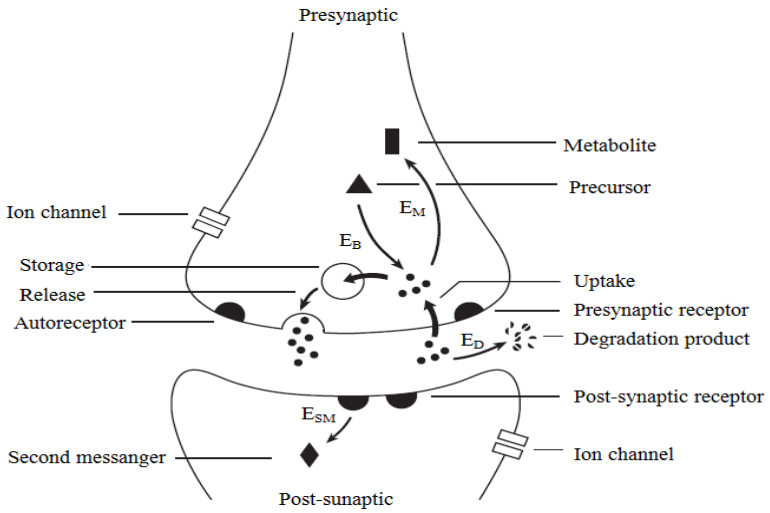


Fig. 3.1: Generalized schematic illustration of processes and mechanisms associated with an axosomatic synapse in the CNS. E, enzymes; E_M, metabolic; E_B, biosynthetic; E_D, degradation; E_{SM}, second messenger; (.) neurotransmitter.

3.3 Drug–Receptor Complex

To appreciate the mechanisms of drug action, it is important to understand the forces of interaction that bind drugs to their receptors. Because of the low concentration of drugs and receptors in the bloodstream and other biological fluids, the law of mass action alone cannot account for the ability of small doses of structurally specific drugs to elicit a total response by combination with all, or practically all, of the appropriate receptors. The enlightening calculation shown below supports the notion that something more than mass action is required to get the desired drug–receptor interaction.

One mole of a drug contains 6.02×10^{23} molecules (Avogadro's number). If the molecular weight of an average drug is 300 g/mol, then 15 mg (an effective dose for many drugs) will contain $6.02 \times 10^{23}(15 \times 10^{-3})/300 = 3 \times 10^{19}$

molecules of drug. The human organism is composed of about 3×10^{13} cells. Therefore, each cell will be acted upon by $3 \times 10^{19}/3 \times 10^{13} = 10^6$ drug molecules. One erythrocyte cell contains about 10^{10} molecules. On the assumption that the same number of molecules is found uniformly in all cells, then for each drug molecule, there are 1010/106 = 104 molecules of the human body! With this ratio of human molecules to drug molecules, Le Chatelier would have a difficult time explaining how the drug could interact and form a stable complex with the desired receptor.

The driving force for the drug–receptor interaction can be considered as a low energy state of the drug–receptor complex (Scheme given below), where k_{on} is the rate constant for formation of the drug–receptor complex, which depends on the concentrations of the drug and the receptor, and koff is the rate constant for breakdown of the complex, which depends on the concentration of the drug–receptor complex as well as other forces. The biological activity of a drug is related to its affinity for the receptor, i.e., the stability of the drug–receptor complex. This stability is commonly measured by how difficult it is for the complex to dissociate, which is represented by its K_d, the dissociation constant for the drug–receptor complex at equilibrium.

$$K_d = \frac{[\text{drug}][\text{receptor}]}{[\text{drug}-\text{receptor complex}]}$$

Because K_d is a dissociation constant, the smaller the K_d, the larger the concentration of the drug–receptor complex, the more stable is that complex, and the greater is the affinity of the drug for the receptor. K_d roughly represents the concentration of the drug required to reach an equilibrium of 50% in the drug–receptor complex.

$$\text{drug} + \text{receptor} \rightleftharpoons \text{drug}-\text{receptor complex}$$

Formation of the drug–receptor complex involves an elaborate equilibrium. Solvated ligands (such as drugs) and solvated proteins (such as receptors) generally exist as an equilibrium mixture of several conformers each. To form a complex, solvent molecules that occupy the binding site of the receptor must be displaced by the drug to produce a solvated complex; interactions between the drug and the receptor are stronger than the interactions between the drug and receptor with the solvent molecules. Drug–receptor complex formation is also entropically unfavorable; it causes a loss in conformational degrees of freedom for both the protein and the ligand, as well as the loss of three rotational and three translational degrees of freedom. Therefore, highly favorable enthalpic

contacts (interactions) between the receptor and the drug must compensate for the entropic loss.

Important Interactions (Forces) Involved in the Drug–Receptor Complex

Interactions involved in the drug–receptor complex are the same forces experienced by all interacting organic molecules and include covalent bonding, ionic (electrostatic) interactions, ion–dipole and dipole–dipole interactions, hydrogen bonding, charge-transfer interactions, hydrophobic interactions, cation–π interactions, halogen bonding, and van-der-waals interactions.

The spontaneous formation of a bond between atoms occurs with a decrease in free energy, that is, a noncovalent bond will occur only when there is a negative ΔG, which is the sum of an enthalpic term (ΔH) and an entropic term ($-T\Delta S$). The change in free energy (binding energy) is related to the binding equilibrium constant (K_{eq}) according to Equation. Therefore, at physiological temperature (37°C), changes in free energy of a few kilocalories per mole can have a major effect on the establishment of good secondary interactions. In fact, if the K_{eq} were only 0.01 (i.e., 1% of the equilibrium mixture in the form of the drug–receptor complex), then a ΔG^0 of interaction of -5.45 kcal/mol would shift the binding equilibrium constant to 100 (i.e., 99% in the form of the drug–receptor complex).

$$\Delta G^0 = -RT \int K_{eq}$$

The bonds formed between a drug and receptors are following:

1. The *covalent bond* is the strongest bond, generally worth anywhere from -40 to -110 kcal/mol in stability. It is seldom formed by a drug–receptor interaction, except with enzymes and DNA.
2. Drug and receptor groups will be mutually attracted provided they have opposite charges. This *ionic interaction* can be effective at distances farther than those required for other types of interactions, and they can persist longer. A simple ionic interaction can provide a $\Delta G0 = -5$ kcal/mol, which declines by the square of the distance between the charges.
3. As a result of the greater electronegativity of atoms, asymmetric distribution of electrons produces electronic dipoles. These dipoles in a drug molecule can be attracted by ions (*ion–dipole interaction*) or by

other dipoles (*dipole–dipole interaction*) in the receptor, provided charges of opposite sign are properly aligned.

4. *Hydrogen bonds* are a type of dipole–dipole interaction formed between the proton of a group X–H, where X is an electronegative atom, and one or more other electronegative atoms (Y) containing a pair of nonbonded electrons.
5. When a molecule (or group) that is a good electron donor comes into contact with a molecule (or group) that is a good electron acceptor, the donor may transfer some of its charge to the acceptor. This forms a *charge-transfer complex*, which, in effect, is a molecular dipole–dipole interaction.
6. When two nonpolar groups, such as a lipophilic group on a drug and a nonpolar receptor group, each surrounded by ordered water molecules, approach each other, these water molecules become disordered in an attempt to associate with each other. This increase in entropy, therefore, results in a decrease in the free energy ($\Delta G = \Delta H - T\Delta S$), which stabilizes the drug– receptor complex. This stabilization is known as a *hydrophobic interaction.*
7. In proteins, the most common aromatic group involved in a *cation–π interaction* is tryptophan (although phenylalanine, tyrosine, and histidine also participate), and the most common cation is arginine (although lysine is also important).
8. A covalently bonded halogen atom can act as an electron acceptor (Lewis acid) to undergo *halogen bonding* with an electron-rich donor atom, such as O, N, or S. The strength of these interactions is in the order H ≈ I > Br > Cl >> F.
9. As atoms from different molecules (such as a drug and a receptor) approach each other, the temporary dipoles of one molecule induce opposite dipoles in the approaching molecule, intermolecular attractions, known as *van der Waals forces*.

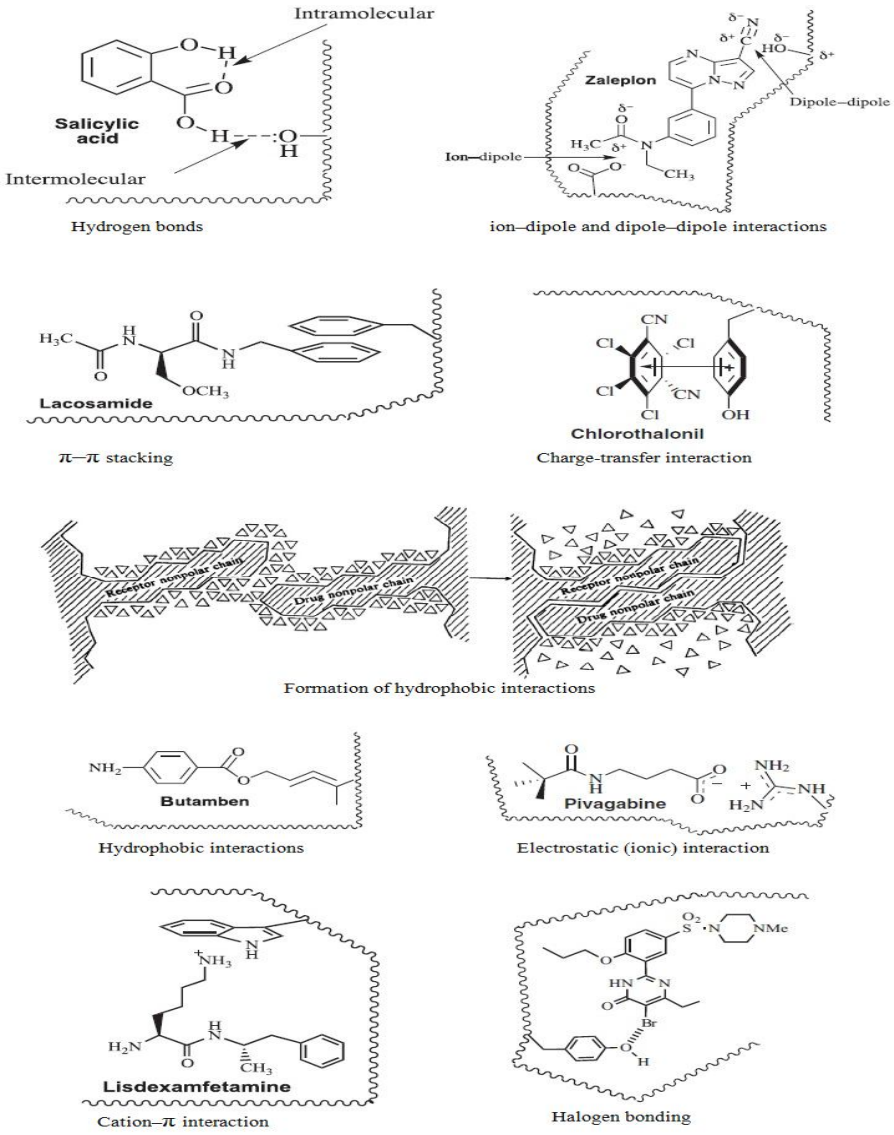


Fig. 3.2: Examples of forces involved in the Drug–Receptor Complex (Wavy line represents the receptor cavity)

3.4 Theories for Drug–Receptor Interactions

Before learning drug-receptor interactions, we should aware about following terms are used in describing drug-receptor interaction:

- *Agonist*: An agent which activates a receptor to produce an effect similar to that of the physiological signal molecule.
- *Inverse agonist*: An agent which activates a receptor to produce an effect in the opposite direction to that of the agonist.

- *Antagonist*: An agent which prevents the action of an agonist on a receptor or the subsequent response, but does not have any effect of its own.
- *Partial agonist*: An agent which activates a receptor to produce submaximal effect but antagonizes the action of a full agonist.
- *Ligand*: (Latin: ligare-to bind) Any molecule which attaches selectively to particular receptors or sites. The term only indicates affinity or binding without regard to functional change: agonists and competitive antagonists are both ligands of the same receptor.
- There are two general categories of compounds that interact with receptors: compounds that occur naturally within the body, such as hormones, neurotransmitters, and other agents that modify cellular activity *autocoids* and *xenobiotics*, compounds that are foreign to the body.

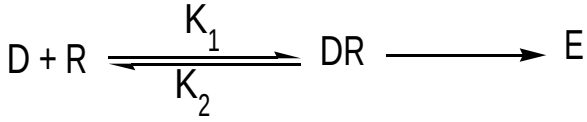
Over the years a number of hypotheses have been proposed to account for the ability of a drug to interact with a receptor and elicit a biological response. Several of the more important proposals are listed below.

- Occupancy Theory
- Rate Theory
- Induced-Fit Theory
- Macromolecular Perturbation Theory
- Activation - Aggregation Theory
- The Two-State (Multistate) Model of Receptor Activation
- The operational model of receptor function
- Classical model of receptor function
- The extended ternary complex model
- The ternary complex model
- Constitutive receptor activity
- Multistate receptor models and probabilistic theory
- Inactivation Theory

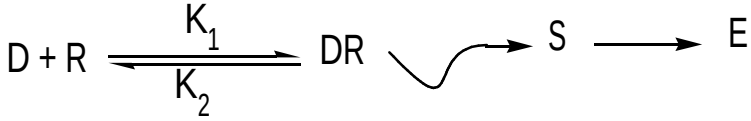
3.5 Occupancy Theory

After studying quantitative aspects of drug action, Clark (1937) propounded a theory of drug action based on occupation of receptors by specific drugs and that the pace of a cellular function can be altered by interaction of these

receptors with drugs which, in fact, are small molecular ligands. He perceived the interaction between the two molecular species, viz. drug (D) and receptor (R) to be governed by the law of mass action, and the effect (E) to be a direct function of the drug-receptor complex (DR) formed:



Subsequently, it has been realized that occupation of the receptor is essential but not itself sufficient to elicit a response; the agonist must also be able to activate (induce a conformational change in) the receptor. The ability to bind with the receptor designated as *affinity*, and the capacity to induce a functional change in the receptor designated as *intrinsic activity* (IA) or *efficacy* are independent properties. Competitive antagonists occupy the receptor but do not activate it. Moreover, certain drugs are partial agonists which occupy and sub-maximally activate the receptor. An all or none action is not a must at the receptor. A theoretical quantity(S) denoting strength of stimulus imparted to the cell was interposed in the Clark's equation:



Depending on the agonist, DR could generate a stronger or weaker S, probably as a function of the conformational change brought about by the agonist in the receptor. Accordingly:

Agonists have both affinity and maximal intrinsic activity (IA = 1), e.g. adrenaline, histamine, morphine. *Competitive antagonists* have affinity but no intrinsic activity (IA = 0), e.g. propranolol, atropine, chlorpheniramine, naloxone. *Partial agonists* have affinity and submaximal intrinsic activity (IA between 0 and 1), e.g. dichloroisoproterenol (on β adrenergic receptor). *Inverse agonists* have affinity but intrinsic activity with a minus sign (IA between 0 and -1), e.g. DMCM (on benzodiazepine receptor). It has also been demonstrated that many full agonists can produce maximal response even while occupying <1% of the available receptors. A large receptor reserve exists in their case, or a number of *spare receptors* are present.

The modified occupancy theory accounts for the existence of partial agonists and antagonists, but it does not account for why two drugs that can occupy the

same receptor can act differently, i.e., one as an agonist, the other as an antagonist.

Example of affinity and efficacy are given in following figure. Figure-A shows the theoretical dose–response curves for five drugs with the same affinity for the receptor (pK_D = 8), but having efficacies varying from 100% of the maximum to 20% of the maximum. The drug with 100% efficacy is a full agonist; the others are partial agonists. Figure-B shows dose–response curves for four drugs with the same efficacy (all full agonists), but having different affinities varying from a pK_D of 9 to 6.

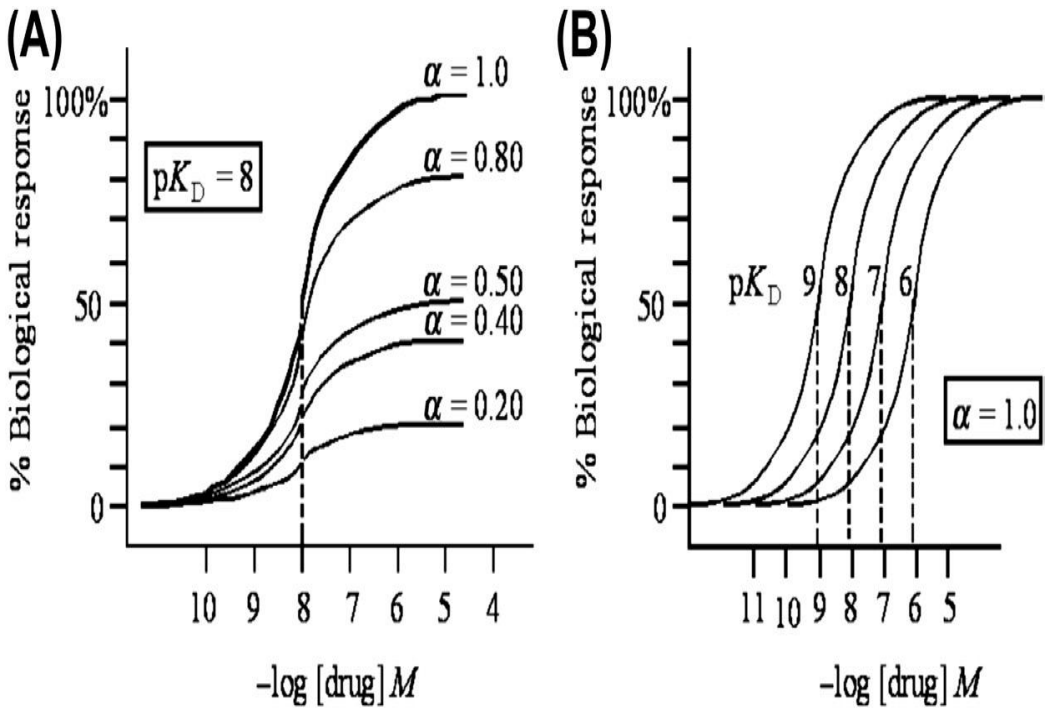


Fig. 3.3: Theoretical dose–response curves illustrate (A) drugs with equal affinities and different efficacies (the top compound is a full agonist, and the others are partial agonists) and (B) drugs with equal efficacies (all full agonists) but different affinities

3.6 Rate Theory

As an alternative to the occupancy theory, Paton proposed that the activation of receptors is proportional to the total number of encounters of the drug with its receptor per unit time. Therefore, the rate theory suggests that the pharmacological activity is a function of the rate of association and dissociation of the drug with the receptor and not the number of occupied receptors. Each

association would produce a quantum of stimulus. In the case of agonists, the rates of both association and dissociation would be fast (the latter faster than the former). The rate of association of an antagonist with a receptor would be fast, but the dissociation would be slow. Partial agonists would have intermediate drug–receptor complex dissociation rates. At equilibrium, the occupancy and rate theories are mathematically equivalent. As in the case of the occupancy theory, the rate theory does not rationalize why the different types of compounds exhibit the characteristics that they do.

3.7 Induced-Fit Theory

The induced-fit theory of Koshland was originally proposed for the action of substrates with enzymes, but it could apply to drug–receptor interactions as well. According to this theory, the receptor need not necessarily exist in the appropriate conformation required to bind the drug. As the drug approaches the receptor, a conformational change is induced, which orients the essential binding sites (Figure below). The conformational change in the receptor could be responsible for the initiation of the biological response (movement of residues to interact with the substrate). The receptor (enzyme) was suggested to be elastic, and could return to its original conformation after the drug (product) was released. The conformational change need not occur only in the receptor (enzyme); the drug (substrate) also could undergo deformation, even if this resulted in strain in the drug (substrate). According to this theory, an agonist would induce a conformational change and elicit a response, an antagonist would bind without a conformational change, and a partial agonist would cause a partial conformational change. The induced fit theory can be adapted to the rate theory. An agonist would induce a conformational change in the receptor, resulting in a conformation to which the agonist binds less tightly and from which it can dissociate more easily. If drug–receptor complexation does not cause a conformational change in the receptor, then the drug–receptor complex will be stable, and an antagonist will result. Other theories evolved from the induced-fit theory, such as the macromolecular perturbation theory, the activation– aggregation theory, and multistate models.

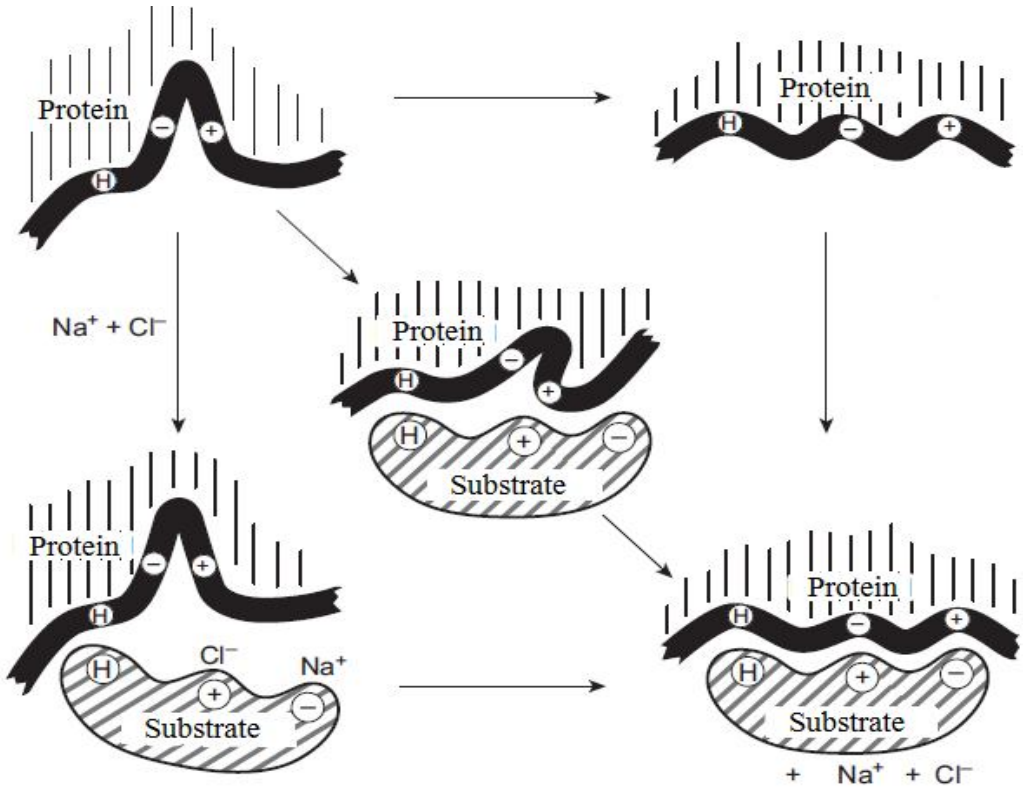


Fig. 3.4: Schematic of the induced-fit theory
(Koshland et. al.)

3. 8 Other theories & models of drug–receptor interactions

Macromolecular Perturbation Theory

Having considered the conformational flexibility of receptors, Belleau suggested that in the interaction of a drug with a receptor two general types of *macromolecular perturbations* could result: *a specific conformational perturbation* makes possible the binding of certain molecules that produce a biological response (an agonist) and *a nonspecific conformational perturbation* accommodates other types of molecules that do not elicit a response (e.g., an antagonist). If the drug contributes to both macromolecular perturbations, a mixture of two complexes will result (a partial agonist). This theory offers a physicochemical basis for the rationalization of molecular phenomena that involve receptors, but does not address the concept of inverse agonism.

Activation - Aggregation Theory

An extension of the macromolecular perturbation theory (which also is based on the induced-fit theory) is the activation–aggregation theory of Monad, Wyman, and Changeux and Karlin. According to this theory, even in the

absence of drugs, a receptor is in a state of dynamic equilibrium between an activated form (R_o), which is responsible for the biological response, and an inactive form (T_o). Using this theory, agonists bind to the R_o form and shift the equilibrium to the activated form, antagonists bind to the inactive form (T_o), and partial agonists bind to both conformations.

The Two-State (Multistate) Model of Receptor Activation

The revised *two-state model of receptor activation* proposes that, in the absence of the natural ligand or agonist, receptors exist in equilibrium (defined by equilibrium constant *L*) between an active state (R*), which is able to initiate a biological response, and a resting state (R), which cannot. In the absence of a natural ligand or agonist, the equilibrium between R* and R defines the basal activity of the receptor. A drug can bind to one or both of these conformational states, according to equilibrium constants K_d and K_d* for formation of the drug–receptor complex with the resting (D-R) and active (D-R*) states, respectively.

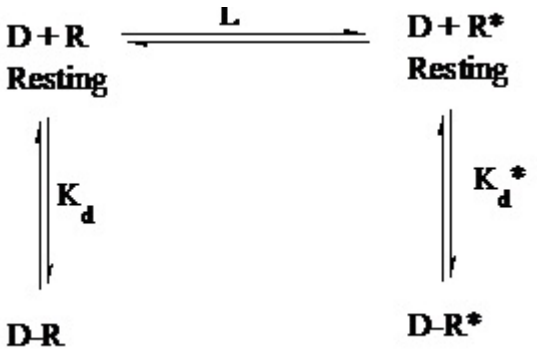


Fig. 3.5: Two-state model of receptor activation. D is the drug, R is the receptor, and L is the equilibrium between the resting (R) and the active (R*) state of the receptor

3.9 Summary

It is emphasized that drug activity is observed through a translation process controlled by cells. Different drugs have different inherent capacities to induce response (intrinsic efficacy). Thus, equal cellular responses can be achieved by different fractional receptor occupancies of these drugs. The ability to reduce stimulus-response mechanisms to single monotonic functions allows relative cellular response to yield receptor-specific drug parameters. It has been shown that most receptors can activate several different signaling pathways, which may also be selectively activated/ inhibited by drugs.

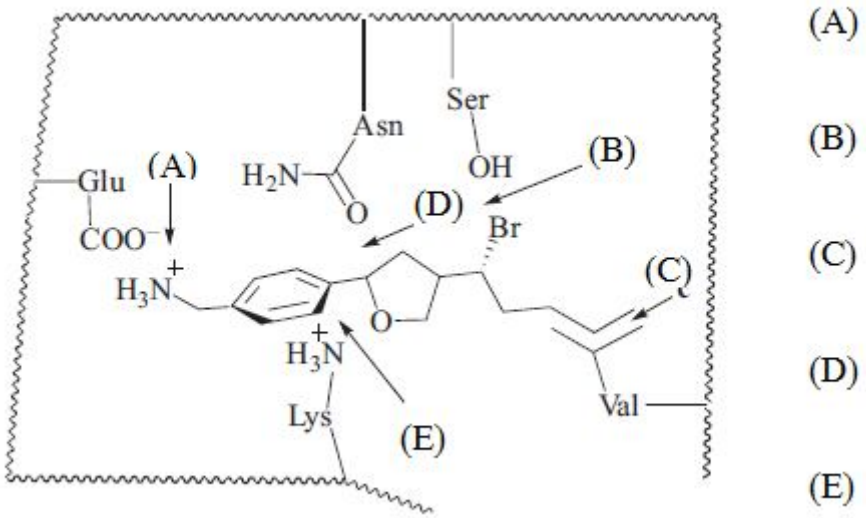
3. 10 Glossary

- Receptor is defined as a macromolecule or binding site located on the surface or inside the effector cell that serves to recognize the signal molecule/ drug and initiate the response to it, but itself has no other function.
- Agonist: An agent which activates a receptor to produce an effect similar to that of the physiological signal molecule.
- Inverse agonist: An agent which activates a receptor to produce an effect in the opposite direction to that of the agonist.
- Antagonist: An agent which prevents the action of an agonist on a receptor or the subsequent response, but does not have any effect of its own.
- Partial agonist: An agent which activates a receptor to produce submaximal effect but antagonizes the action of a full agonist.
- Ligand: (Latin: ligare-to bind) Any molecule which attaches selectively to particular receptors or sites.
- Affinity: The ability to bind with the receptor designated as affinity.
- Intrinsic activity or Efficacy: The capacity to induce a functional change in the receptor designated as intrinsic activity (IA) or efficacy.
- Agonists have both affinity and maximal intrinsic activity.
- Competitive antagonists have affinity but no intrinsic activity.
- Partial agonists have affinity and submaximal intrinsic activity.
- Inverse agonists have affinity but intrinsic activity with a minus sign.

3.11 Review questions / Comprehensive Questions

1. What is receptor? Give brief description about receptor.
2. What are different principles of drug action?
3. Discuss about Drug-Receptor complex.
4. Write different interactions/ forces involved in the drug–receptor complex.
5. What do you know about receptor theory? Enumerate different theories of drug–receptor interactions.
6. Discuss about Occupancy Theory.
7. Discuss about Rate Theory.
8. Discuss about Induced-Fit Theory
9. What is the importance of receptor theory in drug design?

10. Indicate what drug–receptor interactions are involved at every arrow shown. More than one kind of interaction is possible for each letter (A, B, C, D and E).



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Unit - 4

Quantitative Structure Activity Relationship

Structure of Unit:

4.1 Objective

- Aim of drug design is developing a drug with high degree of chemotherapeutic index and specific action.
- To design a drug on the basis as possible as reducing the minimum trial and error approach.

- To target the dedicated screening approach, is more or less random in nature and involves greater efficacy of therapeutic targets.

4.2 History

Drugs are not designed. Brown and Fraser in 1869 showed biological activity could be related to its structure convulsant action of naturally occurring alkaloids was lost after reaction with CH_3I. Ehrlich's big achievement in 1909 is the discovery of the specific cure for the treatment of syphilis. The random screening for a new chemical led to discovery of many important new drugs. The most convincing demonstration that is random screening plus systematic modification of the active compounds. The independent observation of Meyor and Overtone that in a rough way the narcotic potency of simple natural organic compounds parallels the lipophilicity was the enormous importance of the hydrophobic interaction in biology.

4.3 Introduction

The drug design aspect often involves molecular modeling and the use of quantitative structure activity relationship (QSAR) to better define the physicochemical properties that the crucial for biological activity. For identifying the effect, testing is usually done with specific receptor system or enzymes. The various approaches used in drug design include:

- Random screening of synthetic compounds or chemicals and natural products by bioassay procedures.
- Novel compounds preparation based on the known structures of biologically active, natural substance of the plant and animal origin.
- Preparation of structure analogues of lead with increasing biological activity.

There are number of procedures involved in drug design, the first step is the detection of some biological action in a group of compounds so as to serve as a lead. This is followed by molecular manipulations to increase or modify the activity. Identification of a lead nucleus depends upon the consideration of the following points:

1. Molecular structure of the drug
2. Behavior of the drug in the biophase
3. Geometric of the receptor
4. Drug-receptor interaction

5. Change in the structure on binding and
6. The observed biological response.

After following such a tedious process only fewer drug can reach to the level of clinical applicability. Such compounds have to be given extensive trials before they are tried on humans. This adds to the cost of research for new drugs. Broadly, this means that if the development of new drug is to remain economically feasible. The ratio of output to input must be increased.

The lead is a prototype compound that has the desired biological or pharmacological activity but may have many undesirable characteristics such as high toxicity, other biological activities, insolubility or metabolic problems.

QSAR is the study of how the physicochemical properties of a series of compounds affect their biological activity. Drug receptor interactions are a subset of structure- property correlation in which a variety of chemical and physical molecular property is employed to define the association between structure and properties. Quantitative values are measured or calculated for the physical features and these are related to biological activities using mathematical equations.

QSAR techniques employ powerful computers, molecular graphics and sophisticated software: they may be of enormous assistance to those trying to generate the large data bases resulting from the massive efforts in the drug research. QSAR is essentially a computerized statistically method which tries to explain the observed variance in the biological effect of certain classes of compounds as a function of molecular changes caused by the substituents. It assumed that potency of a certain biological activity exerted by a series of congeneric compounds is a function of various physicochemical parameters of the compounds.

The ability to examine multiple relationships between physical property and biological activity is given in Figure 4.1

4.4 QSAR parameters

Physical organic chemistry deals with characterization of the structure and prediction of the structure and prediction of the properties are usually found experimentally. Some property depends on the set of the selected descriptors. The structural information is coded in these properties. Therefore, good correlation of physicochemical properties with a particular set of indices may help in understanding the contribution of these invariants in determining the property.

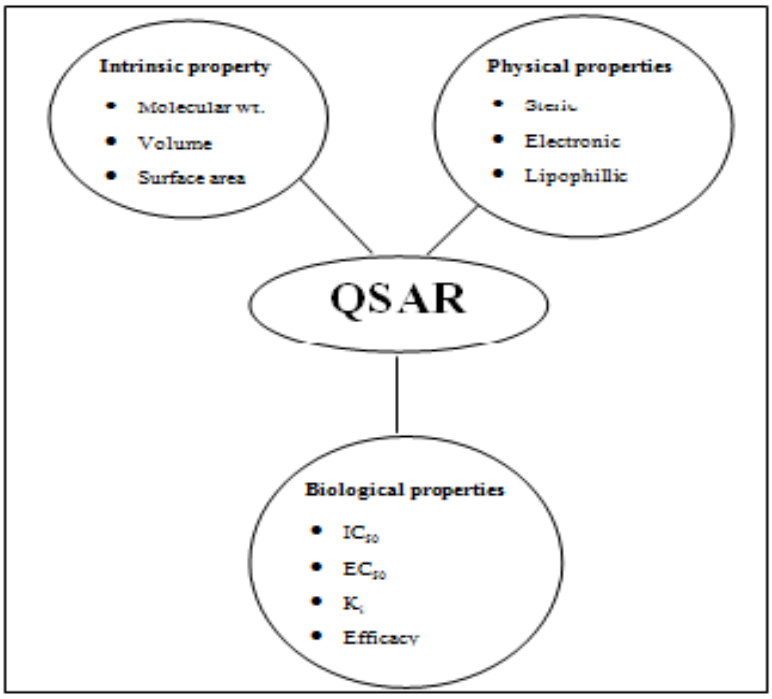


Fig. 4.1: The relationship between physical property and biological activity

4.5 Physicochemical parameters

Biological activity reflects the fundamental physicochemical properties of the bioactive compounds. For example enantiomers are related physico chemically very closely but they differ only by sterically. Various physicochemical parameters are used in QSAR studies are shown in Table 4.1

Table 4.1 Various physicochemical parameters and symbol used in QSAR study

S.No.	Physicochemical Parameters	Symbol
1.	Hydrophobic parameters	

	• Partition coefficients • Pie substituent constant • Solubility	Log p π S
2.	**Electronic parameters** • Ionization constant • Resonance effect • Inductive effect (field effect) • Ionization potential	 pKa R F I
3.	**Steric parameters** • Taft's steric substituent constant • Vander Waal radii • Molar refractivity • Molar volume	 E_S R MR MV

The physicochemical properties of a compound impact on the biological activity. The overall hydrophobic character of a compound influences how efficiency it can cross the cell membrane. The hydrophobic character and size of molecule may influence how well the compound interact fits into its binding site. While the electronic character of the substituent can influence the basicity of the compound, affecting both absorption and receptor binding.

There are many software programs that help in deriving equations but it is upto the medicinal chemist to decide what data to put in. the biological activity of each compound has to be included, but the chemist has to decide which physical feature might be more important to biological activity.

It is usually best to derive an initial equation based on only one or two physical features. The initial equation will give calculated activities close to the experimentally measured activities. The medicinal chemist can study these molecules and try to identify a physical feature with these molecules which have the others do not then search includes that property.

4.6 Hydrophobicity of the molecule

Hydrophobicity of a molecule is measured by log P value where P is known as the partition coefficient. Hydrophobicity of a drug is measured by distribution of compound between an aqueous and non aqueous solvent. Aqueous solution is water and non aqueous solution is n-octanol. Now P is the ratio of concentration of compound in n-octanol to concentration of compound in water.

$$P = \frac{\text{Concentration of compound in n-octanol}}{\text{concentration of compound in water}}$$

If greater proportions of drug dissolve in organic layer than higher the value of log P. higher value of log P and it indicates the higher *in vivo* activity. This is the indication that increasing hydrophobicity allow easier passage of drug through cell membrane in order to reach target site.

Most of the QSAR experiments are carried out on compounds that have a limited range of log P values.

Log activity = $- K_1 (\log P)^2 + K_2 (\log P) + K_3$

K_1, K_2 and K_3 are constant

In above equation $-(\log P)^2$ give negative impact on activity where as log P give positive effect when P is low, log P is more important than $-(\log P)^2$.

Substituent Hydrophobicity of the molecule

The partition coefficient describe the overall hydrophobicity of a molecule, but it is also possible to quantify the hydrophobic character of individual substituent that give hydrophobicity constant (π) for each substituent.

The substituent hydrophobicity constant is a measure of how hydrophobic substituent is related to hydrogen. The value is measured experimentally by comparing the log P values of a compound with and without the substituents.

The hydrophobicity constant (π) for the substituents (x) are obtained using the following equation:

$$\pi_x = \log P_X - \log P_H$$

P_H is the partition coefficient for standard compound and P_X is the partition coefficient for analogue containing the substituent X. If π is the positive then the substituents are more hydrophobic than hydrogen. If π is negative then the substituent is less hydrophobic than hydrogen.

Hydrophobicity constant can be used to calculate log P values for different compound avoiding the need to measure each log P value experimentally. For example log P value for parabromoanisol can be calculated as 2.97, given the log P value for benzene (2.13) and π constant for bromine and methoxy is 0.86 and -0.02 respectively. The relationship between biological activity and partition coefficient is given in Figure 4.2

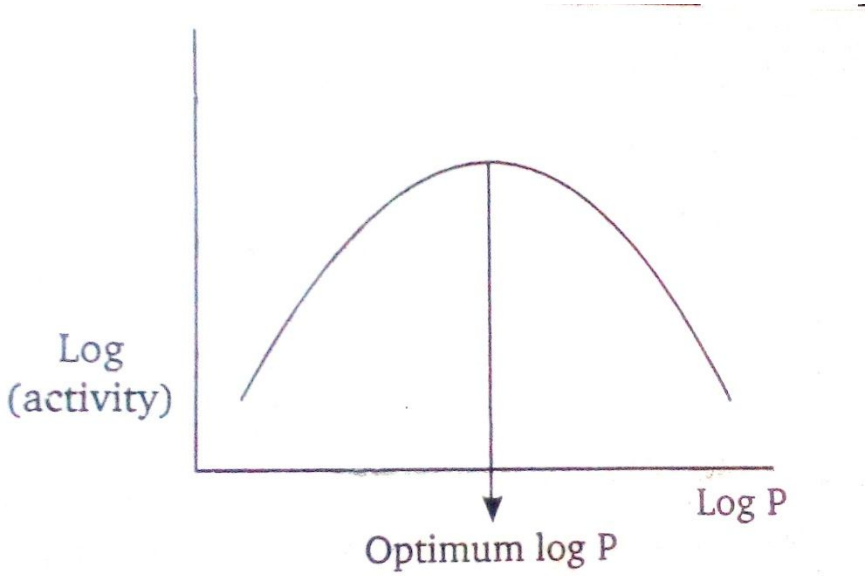


Fig. 4.2: Parabolic relationship between biologic activity (log 1/C) and partition coefficient (log P)

4.7 Electronic parameters

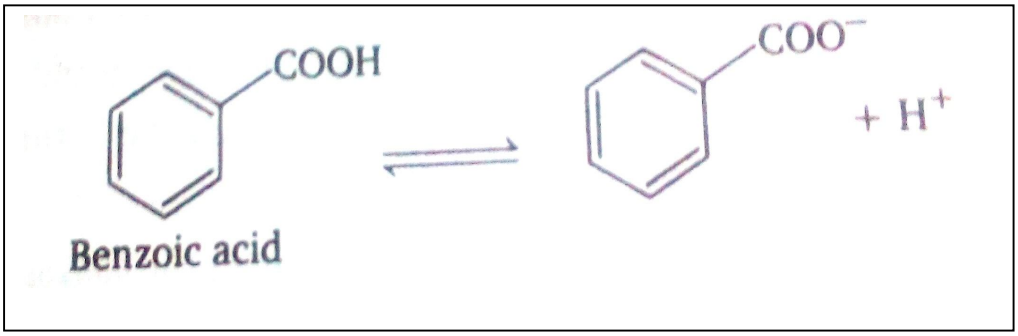
Electronic parameters mainly indicate the influence of polar characters of the drug on its biological activity. They affect:

- Metabolism and elimination pattern of the drug
- The drug receptor interaction

Hammet introduced constants as a quantitative measure of the electronic effects of substituents of aromatic rings on on reaction rates and equillibria. Hammet postulated that the electronic effect of a set of substituents on different organic reactions should be similar. The most commonly used electronic parameter is Hammet substituent constant σ which can be obtained from the dissociation constants K_X and K_H of the benzoic acids X-Ph-COOH and Ph-COOH respectively.

Electronic parameters mainly indicate the influence of polar characters of the drug on its biological activity. The electronic properties of aromatic substituents are describe by the Hammet substitution constant (σ). Hammet postulated that

the electronic effect of particular substituents in different organic reactions should be similar. He selected banzoic acid as a standard system to develop the numerical σ constant value.



Banzoic acid is a weak acid and partially ionizes in water. Equilibrium is attain between ionize and unionize form. The equilibrium dissociation constant K_H is showing the relative proportion of ionize or unionize proportion.

$$K_H = \frac{[PhCOO^-]}{[PhCOOH]}$$

The subscript H signifies that there are no substituents on the aromatic ring. Substituent on the aromatic ring affects this equilibrium. If electronic withdrawing group attached to benzene ring that will stabilize the carboxylate anion (stability $\propto$ 1/ amount of charge) and equilibrium will shift to ionize form and result in larger equilibrium constant. If an electron donating group is attached to ring than it destabilized the carboxylate anion and equilibrium will shift to left side results in smaller equilibrium constant.

The Hammet substituent constant (σ_X) for a particular substituent (X) is defined by the following equation:

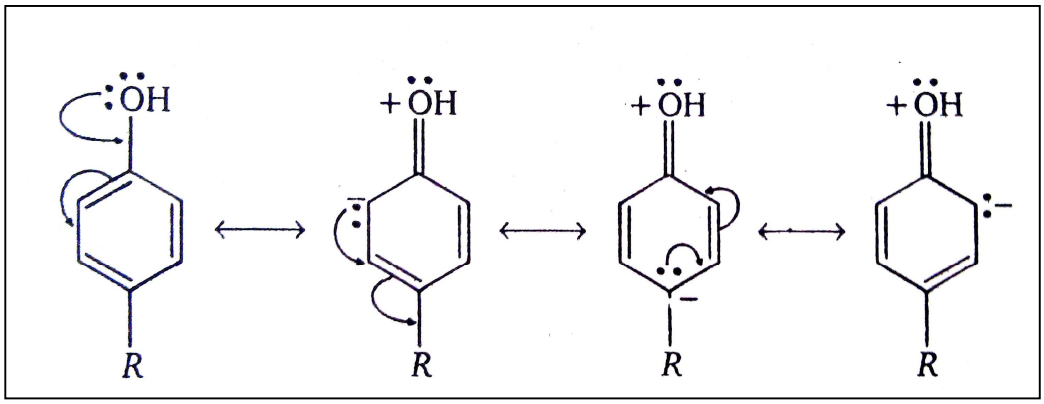
$$\sigma_X = \log \frac{Kx}{KH} = \log K_X - \log K_H$$

These constant are accurate for the molecular structures from which they are derived. Electron withdrawing group such as –Cl, -CN, $-CF_3$ have positive values while electronic donating group such as $-CH_3$, $-CH_2CH_3$ have negative value.

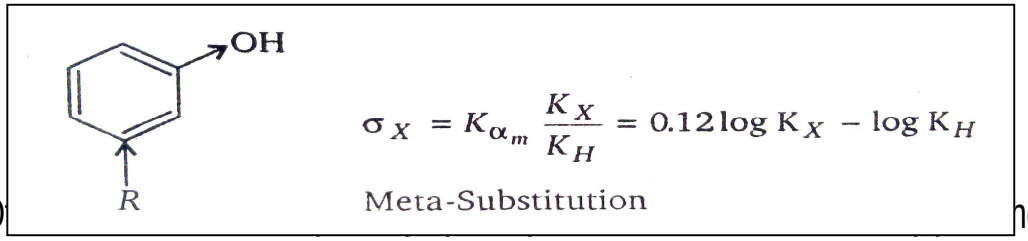
The value of the Hammet substituents depends upon the substituent's inductive effects and resonance effects and value also depends on the substituent position that is meta or para or rest of the molecule.

For example σ_m for phenol group is 0.12 it means electon withdrawing influence felt at the meta position due to induction. When phenol group is at the

para position (σ_p is –0.37) it means the group is electron donating at that position due to resonance. It should be noted that substitution at ortho position can have steric as well as electronic effect.



Inductive effect of phenol is predominates at meta position.



O[illegible]he Resonance effect (R) of aromatic substituent. Aliphatic electronic substituents constants have been obtained by measuring the rate of hydrolysis which is a measure of substituent's electronic effect which arises purely from inductive effect.

Electron donating groups reduce the rate of hydrolysis and have negative value. Electron donating groups increase the rate of hydrolysis and have positive values. Large substituents may also have a steric effect of hydrolysis by shielding the ester from attack.

Steric and electronic factors are separate out by measuring rate of hydrolysis is acidic and basic condition. Under basic condition steric and electronic factors are important whereas under acidic condition only steric factor are important.

4.8 Steric parameters

Steric substitution constant:

Steric features of drug markedly affect the drug receptor interaction reflecting the change in the onset and duration of biological action. Various parameters are used to describe the steric features of the substituents.

1. **Taft's constant (E_S):** which is derived from acid hydrolysis of aliphatic esters.

$$\text{Log } (K/K_0) = E_S$$

K= rate of acid hydrolysis of substituted ester

K_0 = rate of hydrolysis of parent ester

This parameter is useful in studying intra molecular steric effect.

Generally, E_S is standardized to the methyl group so that E_S for the CH_3 group is equal to zero. It is possible to standardize this parameter to hydrogen i.e. E_S (H) = 0.00 and adding 1.23 to every additional methyl group. Greater the positive value of E_S, the greater the steric effect. Greater steric effect affecting intra molecular and inter molecular hinderance to drug receptor interaction.

2. **Molar refractivity:** It is expressed by Lorentz equation

$$MR = \frac{(n^2-1)\,MW}{(n^2+2)\,d}$$

n = index of refraction at the sodium line

MW = molecular weight of the compound, and

d = density of compound

Greater the value of MR means the steric contribution of substituents.

3. **Molecular connectivity index** ($\aleph$):

It indicates the degree of branching in a given structure. Molecular connectivity describes the molecular structures in topological terms for calculation of connectivity index. The structural formula of the compound is written as skeleton formula without the hydrogen atom. It is known as hydrogen suppressed graph. Then valence number (δi) is indicated the atoms attached to each atom.

Such valence numbers of adjacent atoms are multiplied and the bond contribution is calculated by taking the reciprocal square root of the product $\delta i \delta j$.

$$\aleph = \sum(\delta i \delta j)^{-1/2}$$

Correlation of the physical properties with the variation in the structure depends not only on number of atoms in the structure but also upon arrangements of these atoms. Since size and shape of the molecule determines mainly many of the physical parameters govern the biological activity of drug, molecular connectivity index helps to quantify the effect of size and shape on the biological response.

Molecular connectivity index represents sub-structure environment, degree of branching, unsaturation, hetro-atoms and their position and presence of cyclic structures. The close correlation of molecular connectivity with partition coefficients and molar refractivity shows that connectivity index can be taken as measure of the lipophilic feature as well as polar interaction between the molecules. Hence, molecular connectivity index is such a parameter that expresses both, lipophilicity as well as steric feature of the drug molecule.

Effect of electronic and steric parameters on lipophilicity:

Inductive effect influences the overall lipophilicity of the molecule. In general, electron withdrawing groups increase π value when a hydrogen bonding group is involved. Thus in an aromatic skeleton having either nitro group or a hydroxyl group, the electron withdrawing inductive effects of the phenyl ring and the nitro group make the non bonded electrons on the hydroxyl group less available for H- bonding. It leads to a decrease in the affinity of this functional group for the aqueous phase. This then increases the log P or π value. Similarly delocalization of unbounded electrons (i.e. resonance effect) into aromatic systems decreases their availability for H-bonding with the aqueous phase. It leads to an increase in the log P or π value. Similarly, if a group stericaly shields non bonded electrons, then aqueous interactions will decreases and the π value will increase. However, crowding of functional groups involved in hydrophobic interactions will have the opposite effect. Conformational effects also can affect the πvalue.

4.9 Methods used in QSAR studies

Various methods used in QSAR analysis are as follows:

1. **Free energy models**
    - Hansch method
    - Free Wilson mathematical model
2. **Other statistical methods**

- Discriminant analysis
- Factor analysis
- Cluster analysis

3. **Pattern recognition**
4. **Topological methods**
5. **Quantum mechanical methods**
6. **Molecular modeling**

4.10 Hansch analysis (Theoretical method)

Hansch equation is widely used theoretically method in drug design. This method relates biological activity and physicochemical properties for example, degree of ionizations, molecular size or lipid solubility. Most common parameters are included in Hansch equation such as log P, π, σ, F, MR and E_S. Hansch proposed that the action of drug depending on two processes.

- First the journey from the point of entry in the body to the site of action.
- Secondly the interaction with the receptor site.

A typical Hansch equation would be represented as:

$$\text{Log } 1/C = K_1 (\log P)^2 + K_2 \log P + K_3 \sigma + K_4 E_S + K_5$$

Log 1/C = potency

K_1, K_2, K_3, K_4 and K_5 are constants.

These constant would be determined by computer in order to get the best fitting line. Activity is often measured by 1/C where C is the concentration of drug required to produce a specific effect. The more active the drug, the smaller the concentration required and larger value of 1/C.

It should be appreciated that a QSAR equation is only as good as the data that has been entered. Usually a range of compounds are synthesized to quantify the effect that two or three physical parameters have on biological activity. Hansch analysis may serve both to guide the medicinal chemist in future synthesis and testing of other compound in series and to the role of hydrophobic, electronic and steric factors in drug receptor interactions.

4.11 Free and Wilson analysis

Free and Wilson developed an approach to SAR. This method is based upon an additive mathematical model in which a particular substituent in a specific position is assumed to make an additive and contribution to biological activity of a molecule in a series of chemically related molecules. This method is based on the assumption that the introduction of a particular substituent at particular position that gives a quantitatively similar effect on biological potency of the whole molecule. Biological activity is expressed as:

Log (biological activity) = contribution of unsubstituted parent compound + contribution of corresponding substituent

This method is preferred when nothing is known about the mode of action or when the physicochemical properties of the substituents used are unknown. Best results with the free Wilson method are obtained in series with several positions are available for substitution and only if each substituent any location is present in at least two compound in a series.

4.12 Advantages of QSAR

QSAR helps to understand the forces that govern the activity in a series of compounds. It thus helps to reduce the work in drug design and ensure that every drug synthesized and pharmacologically tested is as meaningful as possible. The main area where QSAR provides:

1. **Forcasting of biological activity:** innumerable applications of QSAR has been reported where successful prediction of biological activity played an important role. Through the regression analysis, parameters or nature and position of substituent which may increase the activity can be guessed. The advanced techniques using computerized programs even give the structural features of the most possible active compound of the series.
2. **Selection of proper substituents:** Proper selection of substituents to develop a series leads to a decrease in the average number of analogs required to investigate the relationship between substituent parameters and the biological activity. Planning gives a good chance of finding out what combinations of parameters will optimized the potency.
3. **Bioisosterism:** With the introduction of QSAR, the qualitative concept of bioisosterism has turned to be more quantitative and constitutive. QSAR also helps to decide an isoester which will give better pharmacokinetic and pharmacodyanamic properties to lead molecule.

4. **Drug-receptor interactions:** In a series of compounds, QSAR studies help to predict in quantitative terms, the force involved in the drug receptor interaction if the substitution are involved in non-essential part of the drug molecule. If selection of parameter is proper, QSAR may also suggest at which position of the receptor, increased lipophilicity of the drug increase binding.
5. **Pharmacokinetic information:** The correlation between various types of parameter and the pharmacokinetic features of the drug can be done using QSAR.

4.13 Limitation of QSAR

The application of QSAR analysis may result in statistically valid equations. It is often difficult to interpret the relationship in biochemical terms. Failure of regression analysis in the prediction of biological activity of analogs results mainly due to

- A poorly designed series
- Improper condition of the biological testing
- Multiple mode of action

The most serious problem in QSAR is the lack of fundamental understanding of how to quantitatively describe substituent effect on drug-receptor interactions. A successful QSAR can provide only indirect (E_S, MV, MR) information about the three dimensional aspect of drug-receptor interactions.

Other effects (electronic or steric) have their own influences on the overall lipophilicity of the molecule. This may result in wrong correlation and interpretation of activity in a series that mainly depends upon lipophilicity for biological action. Electronic effect of a substituent may change both, the degree of ionization and the charge distribution.

Since the biological activity determination process is susceptible to considerable experimental variations, a non linear scatter may be observed during correlation of biological activity with physicochemical parameters. QSAR fails to explain this built in scatter mathematically.

Similarly, physiological active compounds on their way from the site of administration to the target sites, are known to undergo diverse chemical and biochemical and biochemical transformations. It is likely that they act differently on different bio-targets to exert same kind of activity.

4.14 Summary

Medicinal chemistry involves the discovery of new chemical entities for the treatment of disease and systematic study of the structure activity relationships of these compounds. Such studies provide the basis for development of better medicinal agents from lead compounds found via random screening, systemic screening and rational design. The role of the medicinal chemist is that of increasing the potency and duration of action of newly discovered compounds as well as decreasing adverse effects.

QSAR for drug receptor interactions are subset of structure property correlation in which a variety of chemical and physical molecular properties is employed to define the association between structure and property. QSAR are wide spread in medicinal chemistry since the advent of cheap and high speed computing technology in the past 20 year.

The QSAR approach can be extended with the recognition that the ligand occupies three dimensional space. The ability to determine or predict a pharmacophore map by the use of molecular modeling technique and the synthesis of rigid analogs then generates an hypothesis of bioactive confirmation from which comparative molecular field analysis can used to calculate the intermolecular interaction field that surround each molecule and subsequently the relationship between the biologic activity and the calculated fields is determined.

4.15 Glossary

- Lipophilicity: having an affinity for lipid
- Biophase: the period during which a effective concentration of a drug is maintained the vicinity of this site of action
- Enantiomer: molecules that are mirror image of one another
- Inductive effect: it is an experimentally observed effect of the transmission of charge through chain of atom in a molecule resulting in a permanent dipole in a bond.
- Resonance: it is a way of describing delocalized of electrons within certain molecule and poly atomic ions where the bonding cannot be expressed by a single Lewis formula.

- Pharmacophore: a part of a molecular structure that responsible for a particular biological or pharmacological interaction that it undergoes.
- Ligand: it is an ion or molecule that binds to a central metal atom to form a coordination complex.
- Vander Waal radius: it is an atom of is the radius of a imaginary hard sphere which can be used to model the atom for many purposes.
- H-bonding: it is bond that occurs between hydrogen and high electronegative atom (N, O and F).

4.16 Review questions / Comprehensive Questions

1. Define QSAR. Write about Hansch analysis.
2. Discuss in brief about physicochemical parameters used in QSAR.
3. Write a note on electronic parameters and steric parameters used in QSAR.
4. Discuss how resonance effect and inductive effect affect drug design.
5. Write methods used in QSAR technique.

4.17 References and Suggested readings

- Foye's Principles of Medicinal Chemistry, David A. Williams, Thomas L. Lemke (Fifth edition) 2005, B.I. Publication and Pvt. Ltd.
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Unit-5

Concepts of drug receptors

Structure of Unit:

5.1 Objectives

In this unit, we move from the general principles of drug action to the molecules that are involved in recognizing chemical signals and translating them into cellular responses. First, we consider the types of forces which are involved in drug receptor interaction. Next, we describe the main families of receptors and discuss the various forms of receptor-effectors linkage (signal transduction mechanisms) through which receptors are coupled to the regulation of cell function. The relationship between the molecular structure of a receptor and its functional linkage to a particular type of effectors system is a principal theme.

5.2 Introduction: Receptor

Pharmacodynamics is the study of drug effects. This includes physiological and biochemical effects of drugs and their mechanism of action at organ system.

Majority of drugs produce their effects by interacting with a discrete target biomolecule, which usually is a protein. Such mechanism confers selectivity of action to the drug. Functional proteins that are targets of drug action can be grouped into four major categories, as follows-

1. Ion channels

2. Enzymes
3. Carrier molecules (transporters)
4. Receptors

1. Ion channels

Proteins which act as ion selective channels participate in transmembrane signaling and regulate intracellular ionic composition. This makes them a common target of drug action. Drugs can affect ion channels either through specific receptors (ligand gated ion channels, G-protein operated ion channels) or by directly binding to the channel and affecting ion movement through it. (Fig. 5.2(a);B)

2. Enzymes

Almost all biological reactions are carried out under catalytic influence of enzymes. Enzymes are a very important target of drug action. Drugs can either increase or decrease the rate of enzymatically mediated reactions. (Fig. 5.2(a);A)

3. Transporters

Several substrates are translocated across membranes by binding to specific transporters (carriers) which either facilitate diffusion in the direction of the concentration gradient or pump the metabolite/ion against the concentration gradient using metabolic energy.

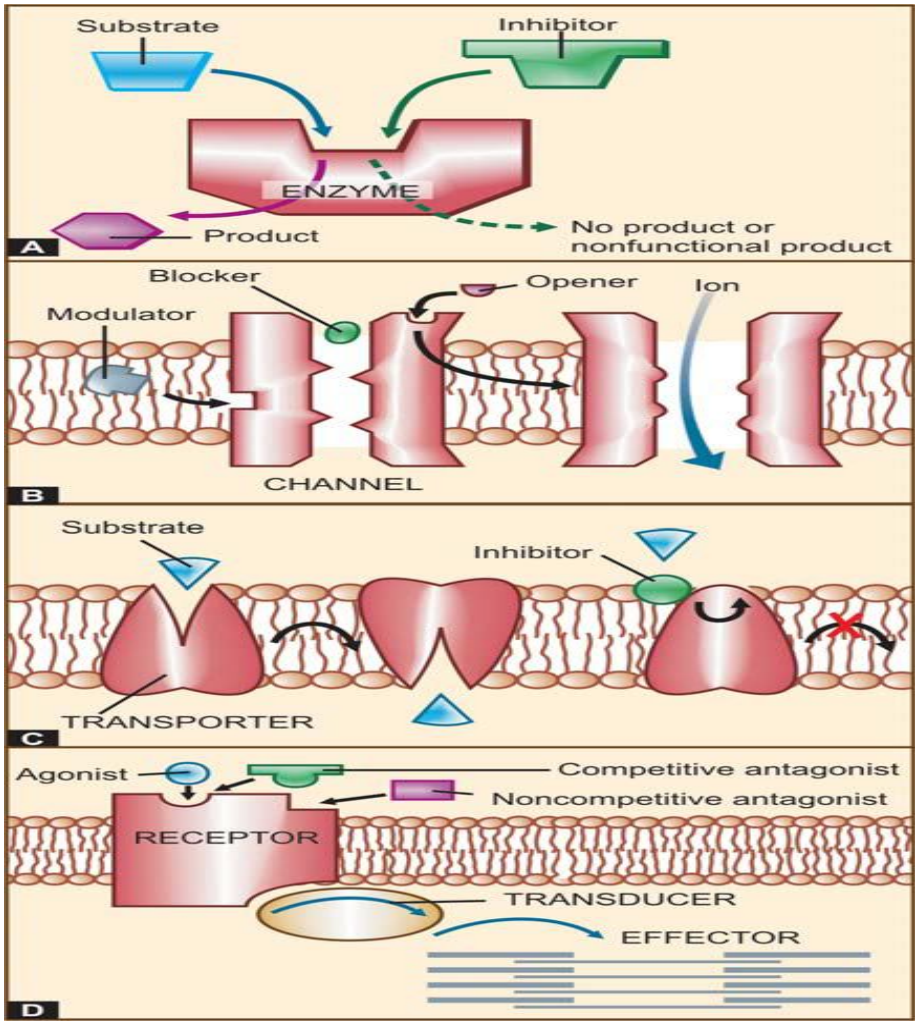

Fig. 5.2(a): Targets of drug action:(A) Enzyme; (B) Transmembrane ion channel; (C) Membrane bound transporter; (D) Receptor

Many drugs produce their action by directly interacting with the solute carrier (SLC) class of transporter proteins to inhibit the ongoing physiological transport of the metabolite/ion. (Fig. 5.2(a);C)

4. Receptors

Receptors have become the central focus of investigation of drug effects and their mechanisms of action (Pharmacodynamics). A fundamental concept of pharmacology is that to initiate an effect in a cell, most drugs combine with some molecular structure on the surface of or within the cell (Fig.5.2 (a);D). This molecular structure is called a receptor. A drug receptor is a specialized target macromolecule, present on the cell surface or intracellular, that binds a drug and mediates its pharmacologic actions.

Drug + Receptor $\Longrightarrow$ Drug-receptor complex $\Longrightarrow$ Effect

The following terms are used in describing drug-receptor interaction:

- **Agonist-** An agent which activates a receptor to produce an effect similar to that of the physiological signal molecule. Figure 5.2 (b)
- **Inverse agonist-** An agent which activates a receptor to produce an effect in the opposite direction to that of the agonist.

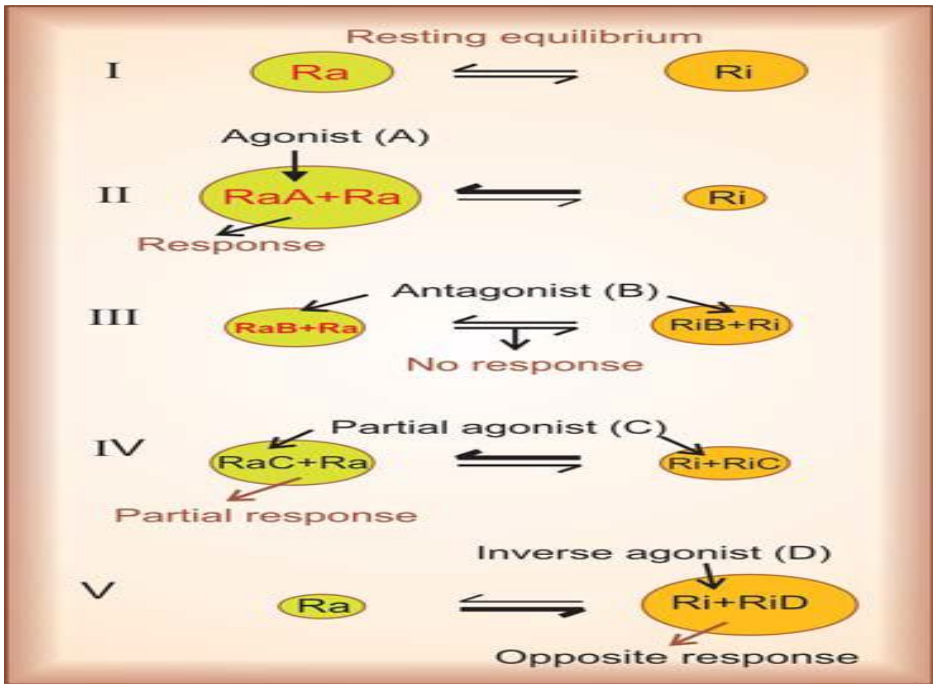

Fig. 5.2 (b): Illustrations of terms: Agonist; Antagonist; Inverse agonist; Partial agonist

- **Antagonist-** An agent which prevents the action of an agonist on a receptor or the subsequent response, but does not have any effect of its own. Figure 5.2 (b)
- **Partial agonist-** An agent which activates a receptor to produce sub maximal effect but antagonizes the action of a full agonist. Figure 5.2 (b)
- **Ligand-** Any molecule which attaches selectively to particular receptors or sites.

Although the term receptor is convenient, one should never lose sight of the fact that receptors are in actuality molecular substances or macromolecules in tissues that combine chemically with the drug. Since

most drugs have a considerable degree of selectivity in their actions, it follows that the receptors with which they interact must be equally unique. Thus, receptors will interact with only a limited number of structurally related or complementary compounds.

5.3 Forces involved in drug receptor interaction

Biological receptors are capable of combining with drugs in a number of ways, and the forces that attract the drug to its receptor must be sufficiently strong and long-lasting to permit the initiation of the sequence of events that ends with the biological response. Those forces are chemical bonds, and a number of types of bonds participate in the formation of the initial drug–receptor complex.

Covalent bonds

The covalent bond is strongest bond, generally worth anywhere from 40 to 110 kcal/mol in stability. It is seldom formed by a drug receptor interaction, except with enzymes and DNA.

Ionic interactions

For protein receptors at physiological pH basic groups such as the amino side chains of arginine, lysine, and , to much lesser extent histidine are protonated and, therefore provide a cationic environment acidic groups, such as caroboxylic acid side chains of aspartic acid and glutamic acid, are deprotonated to give anionic groups.

Drug and receptors group will be mutually attracted provides they have opposite charges. This ionic interaction can be affective at distance further than those required for other types of interactions and they can persist longer.

1. Ion-Dipole and Dipole-Dipole interactions

As a result of greater electro negativity of atoms such as oxygen, nitrogen, sulfur and halogens relative to that carbon, C-X bonds in drug and receptors, where X is an electronegative atom, will have an asymmetric distribution of electrons; these produces an electronic dipoles.

2. Hydrogen Bonds

Hydrogen bonds are a type of dipole-dipole interaction formed between the proton of a group X-H where X is an electronegative atom, and other electronegative atoms Y containing a pair nonbonded electrons.

3. Charge-Transfer Complexes

When molecule that is a good electron donor comes into contact with a molecule that is good electron acceptor, the donor may transfer some of its charges to the acceptor. This forms a charge-transfer complex, which, in effect, is a molecular dipole-dipole interaction.

4. Hydrophobic Interactions

In the presence of a non polar molecules or region of a molecule, the surrounding water molecules orient themselves and, therefore are in a higher energy state than when only other water molecules are around. When to non polar groups, each surrounded by a ordered water molecules, approach each other, these water molecules becomes disordered in an attempt to associate with each other, this increase in entropy, therefore, results in a decrease in the free energy that stabilizes the drug-receptor complex.

5. Vander Walls or London Dispersion Forces

Atoms in nonpolar molecules may have a temporary nonsymmetrical distribution of electron density, which results in the generation of a temporary dipole. As atoms from different molecules approach each other, the temporary dipoles of one molecule induce opposite dipoles in the approaching molecule consequently, intermolecular attractions, and known as Vander walls forces.

5.4 Types of receptor

Based on molecular structure and the nature of this linkage (the transduction mechanism), we can distinguish four receptor types, or super families are follows:

Type1: Ligand-gated ion channels

Type2: G-protein-coupled receptors

Type3: Kinase-linked receptors

Type4: Nuclear receptors

Table 5.4: Types of Receptor

	Type 1 Ligand-gated ion channels	**Type 2 G-protein-coupled receptors**	**Type 3 Kinase-linked receptors**	**Type 4 Nuclear receptors**
Location	Membrane	Membrane	Membrane	Intracellular

Effector	Ion channel	Channel or enzyme	Enzyme	Gene transcription
Coupling	Direct	G-protein	Direct	Via DNA
Examples	Nicotinic acetylcholine receptor, gamma-aminobutyric acid type A	Muscarinic acetylcholine receptor (mAChR), adrenoceptors	Insulin, growth factor, cytokine receptors	Steroid, thyroid hormone receptors
Structure	Oligomeric assembly of subunits surrounding central pore	Monomeric (occasionally dimeric) structure comprising seven transmembrane helices	Single transmembrane helix linking extracellular receptor domain to intracellular kinase domain	Monomeric structure with separate receptor and DNA-binding domains

Type 1: Ligand-gated ion channels

The ligand-gated ion channels are also known as ionotropic receptors. These are membrane proteins with a similar structure to other ion channels but incorporating a ligand-binding (receptor) site, usually in the extracellular domain.

Type 2: G-protein-coupled receptors

The G-protein-coupled receptors (GPCRs) are also known as metabotropic receptors or seven-transmembrane-spanning (heptahelical) receptors. They are membrane receptors that are coupled to intracellular effector systems via a G-protein.

Type 3: Kinase-linked and related receptors

There is a large and heterogeneous group of membrane receptors responding to protein mediators. They comprise an extracellular ligand-binding domain linked to an intracellular domain by a single transmembrane helix. In many cases, the intracellular domain is enzymic in nature (with protein kinase or guanylate cyclase activity).

Type 4: Nuclear receptors

The nuclear receptors regulate gene transcription. The term nuclear receptor is something of a misnomer since some are actually located in the cytosol and migrate to the nuclear compartment when a ligand is present.

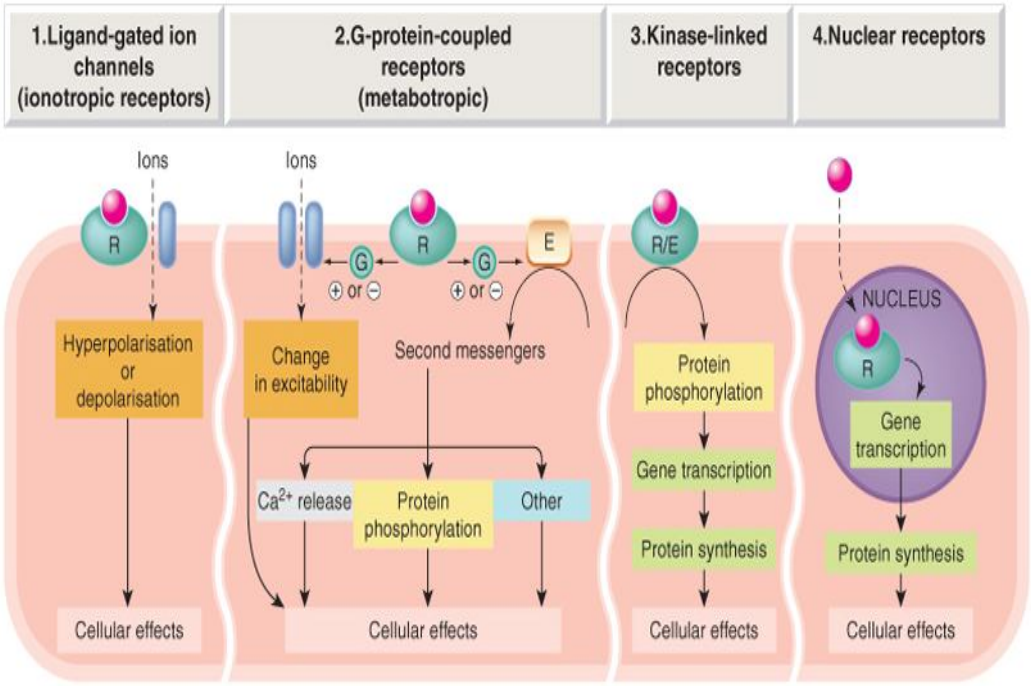

Fig.5. 4: Types of receptor-effector linkage

5.5 G-protein coupled receptors (GPCR)

These are a large family of cell membrane receptors which are linked to the effector (enzyme/ channel/carrier protein) through one or more GTP-activated proteins (G-proteins) for response effectuation. The molecule has 7 α-helical membrane spanning hydrophobic amino acid (AA) segments which consists 3 extracellular and 3 intracellular loops. Fig. 5.5(a)

The agonist binding site is located somewhere between the helices on the extracellular face, while another recognition site formed by cytosolic segments binds the coupling G-protein. The G proteins float in the membrane with their exposed domain lying in the cytosol, and are heterotrimeric in composition (α, β and γ subunits). In the inactive state GDP is bound to their exposed domain; activation through the receptor leads to displacement of GDP by GTP.

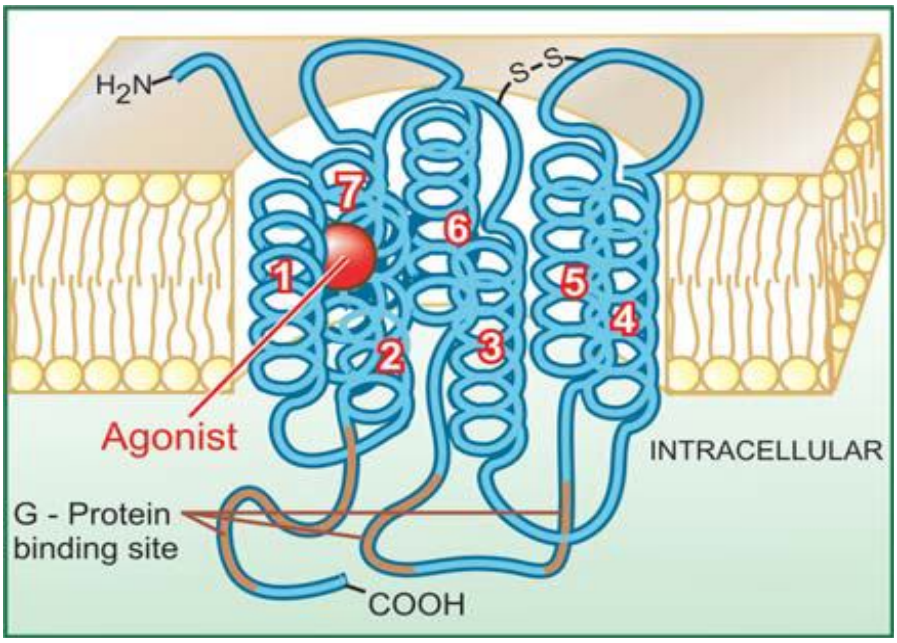

Fig. 5.5(a): Diagrammatic representation of G-protein coupled receptor molecule

The active α- subunit carrying GTP dissociates from the other two subunits and either activates or inhibits the effector. The βγ subunits have also been shown to modulate certain effectors like receptor operated K+ channels, adenylylcyclase (AC) and phospholipase C.

Number of G proteins distinguished by their α subunits have been described. The important ones with their action on the effector are:

G_s : Adenylyl cyclase ↑, Ca2+ channel ↑

G_i : Adenylyl cyclase ↓, K+ channel ↑

G_o : Ca2+ channel ↓

G_q : Phospholipase C ↑

G_{13} : Na+/H+ exchange ↑

There are three major effector pathways through which GPCRs function.

• Adenylyl cyclase (AC): cAMP pathway-

Activation of AC results in intracellular accumulation of second messenger cAMP which functions mainly through cAMP-dependent protein kinase (PKA). The PKA phosphorylates and alters the function of many enzymes, ion channels, transporters and structural proteins to manifest as increased contractility/impulse generation (heart), relaxation (smooth muscle), glycogenolysis, lipolysis, inhibition of secretion/mediator release, modulation of junctional transmission, hormone synthesis, etc. Fig. 5.5(b)

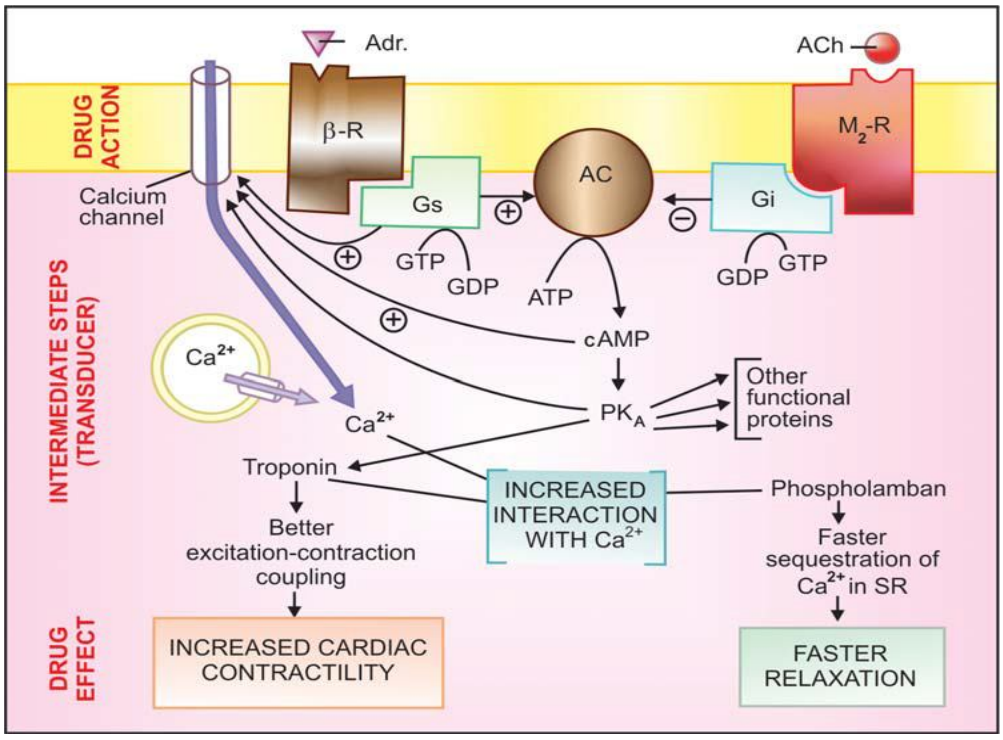

Fig. 5.5(b): The action-effect sequence of two G-protein coupled (β adrenergic and muscarinic M2) receptor activation in myocardial cell

• Phospholipase C pathway: IP3-DAG pathway-

Activation of phospholipase C (PLC) hydrolyses the membrane phospholipid phosphatidyl inositol 4, 5-bisphosphate (PIP_2) to generate the second messengers inositol 1,4,5-trisphosphate (IP_3) and diacylglycerol (DAG). The IP_3 mobilises Ca^{2+} from intracellular organellar depots and DAG enhances protein kinase C (PKc) activation by Ca^{2+}. Cytosolic Ca^{2+}. PKc and other effectors—mediates/modulates contraction, secretion/ transmitter release, eicosanoid synthesis, neuronal excitability, intracellular movements, membrane function, metabolism, cell proliferation, etc. Figure 5.5(c)

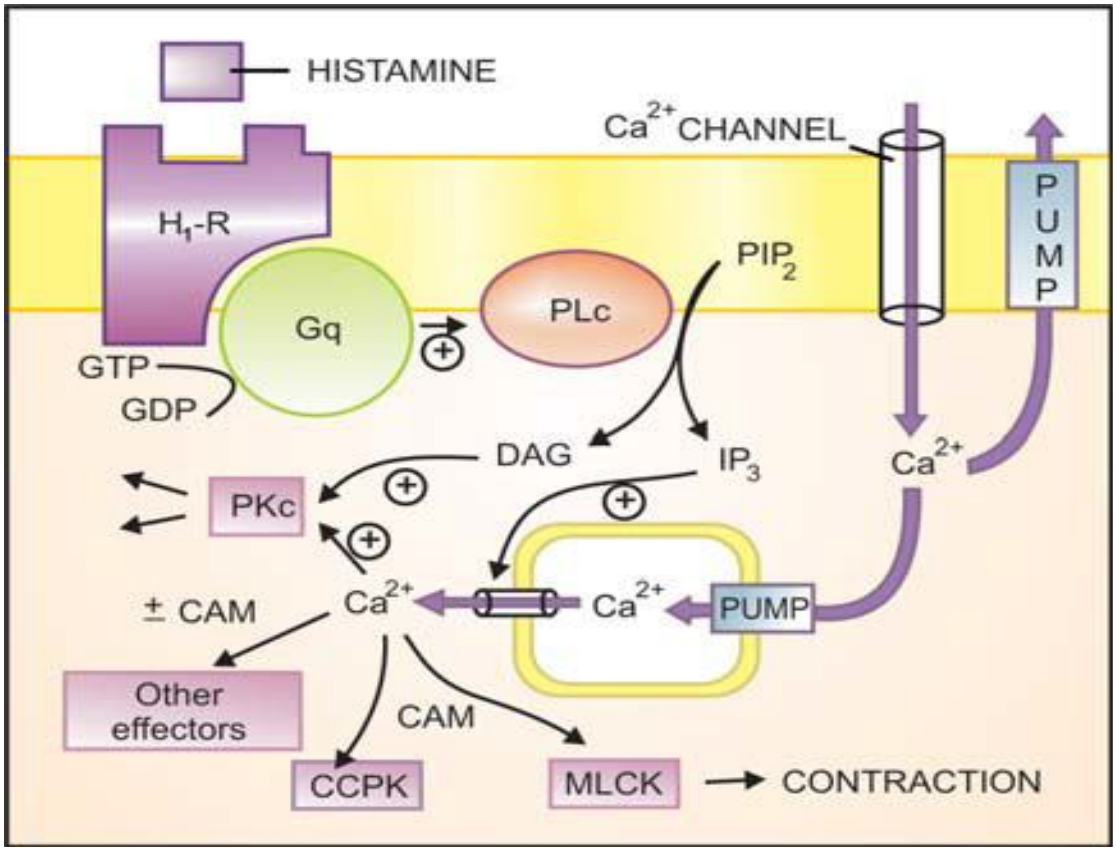

Figure 5.5(c): The important steps of phospholipase C(PLc) pathway of response effectuation (in smooth muscle): histamine receptor (H_1R); phosphatidyl inositol 4, 5-bisphosphate (PIP_2); inositol 1, 4, 5-trisphosphate (IP_3); diacylglycerol (DAG); calmodulin (CAM); myosin light chain kinase (MLCK); calcium-calmodulin protein kinase (CCPK).

- **Ion Channel pathway-**

The activated G proteins can also open or close ionic channels specific for Ca^{2+}, K^+ or Na^+, without the intervention of any second messenger like cAMP or IP3, and bring about hyperpolarization/depolarization/ changes in intracellular Ca^{2+}.

5.6 Ligand gated ion channel receptor

These cell surface receptors, also called ligand gated ion channels, enclose ion selective channels (for Na+, K+, Ca2+ or Cl^-) within their molecules. Agonist binding opens the channel (Fig. 4.4) and causes depolarization/hyperpolarization/ changes in cytosolic ionic composition, depending on the ion that flows through.

5.7 Enzyme-linked receptors

This class of receptors has a subunit with enzymatic property or binds a JAK (Janus-Kinase) enzyme on activation. The agonist binding site and the catalytic site lie respectively on the outer and inner face of the plasma membrane. These two domains are interconnected through a single transmembrane stretch of peptide chain. There are two major subgroups of such receptors:

a) Those that have intrinsic enzymatic activity.

b) Those that lack intrinsic enzymatic activity, but bind a JAK-STAT kinase on activation.

a) Intrinsic enzyme receptors:

The intracellular domain is either a protein kinase or guanylyl cyclase. On binding the peptide hormone to the extracellular domains, the monomeric receptors move laterally in the membrane and form diamers. Dimerization activates tyrosine-protein kinase (t-Pr-K) activity of the intracellular domains so that they phosphorylate tyrosine (t) residues on each other, as well as on several SH2 domain substrate proteins (SH2-Pr). The phosphorylated substrate proteins then perform downstream signaling function. Figure 5.7(a)

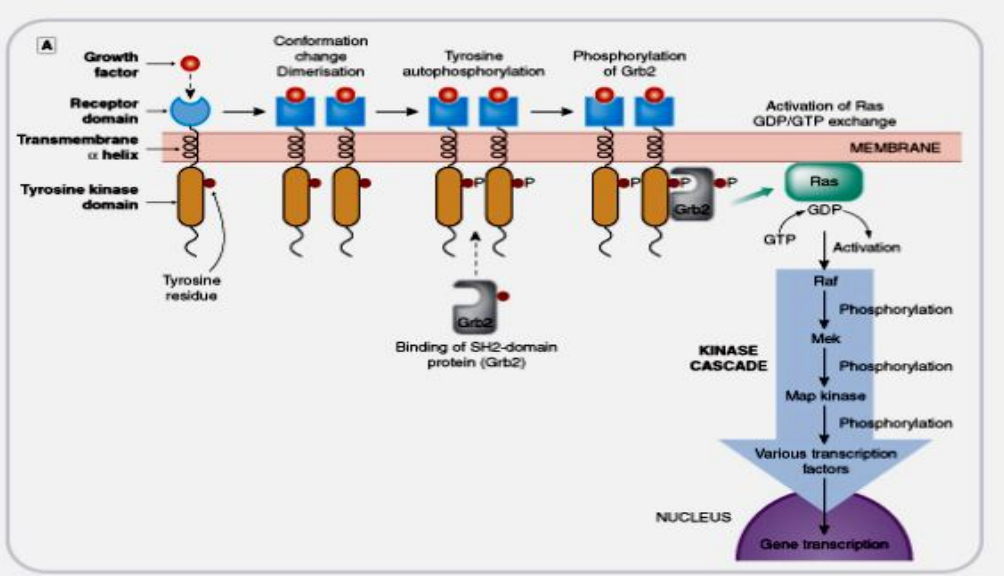

Fig. 5.7(a): Transduction mechanisms of kinase-linked receptors :(a) Intrinsic enzyme receptors

b) **JAK-STAT kinase binding receptor:**

The intracellular domain of these receptors lacks intrinsic protein kinase activity. Signal molecule binding to the extracellular domain induces receptor dimerization which activates the intracellular domain to bind free moving JAK (Janus Kinase) molecules. The activated JAK phosphorylate tyrosine residues on the receptor which then binds another protein STAT (signal transducer and activator of transcription). Tyrosine residues of STAT also get phosphorylated by JAK. The phosphorylated STAT dimerize, dissociate from the receptor and move to the nucleus to regulate transcription of target genes. Figure 5.7(b)

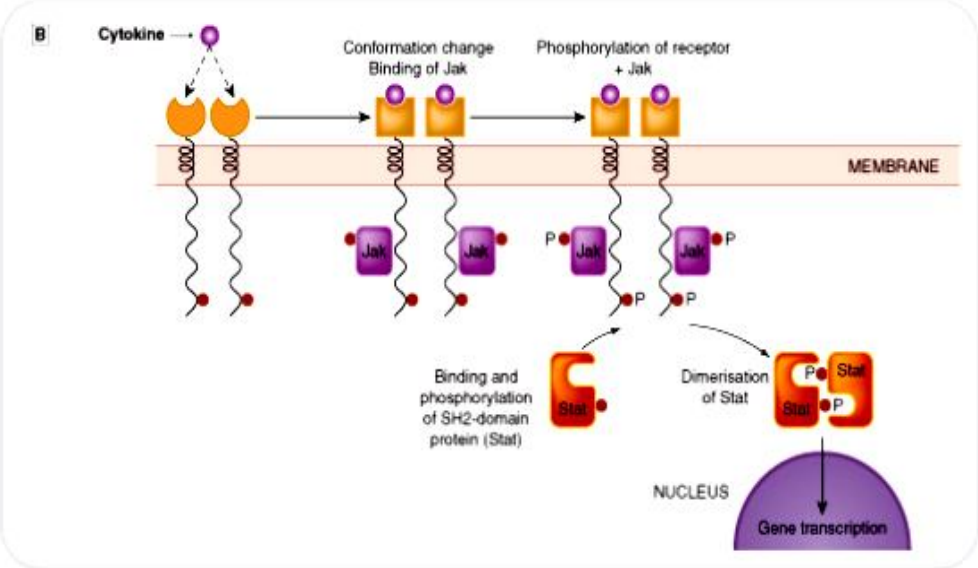

Fig. 5.7(b): Transduction mechanisms of kinase-linked receptors: (b) JAK-STAT kinase binding receptor

5.8 Nuclear receptor

These are intracellular (cytoplasmic or nuclear) soluble proteins which respond to lipid soluble chemical messengers that penetrate the cell. The receptor protein (specific for each hormone/ regulator) is inherently capable of binding to specific genes, and exposes the DNA binding regulatory segment located in the middle of the molecule. Attachment of the receptor protein to the genes facilitates their expression so that specific mRNA is synthesized on the template of the gene. This mRNA moves to the ribosomes and directs synthesis of specific proteins which regulate the activity of target cells.

The glucocorticoid (G) penetrates the cell membrane and binds to the glucocorticoid receptor (GR) protein that normally resides in the cytoplasm. The GR has a steroid binding domain; binding of the steroid to GR dissociates

the complexed proteins (HSP90, etc. Fig. 5.8. The steroid binding domain is exposed, promoting dimerization of the occupied receptor. The steroid bound receptor diamer translocates to the nucleus and interacts with specific DNA sequences called 'glucocorticoid responsive elements' (GREs). The expression of these genes is consequently altered resulting in promotion (or suppression) of their transcription. The specific mRNA thus produced is directed to the ribosome where the message is translated into a specific pattern of protein synthesis, which in turn modifies cell function.

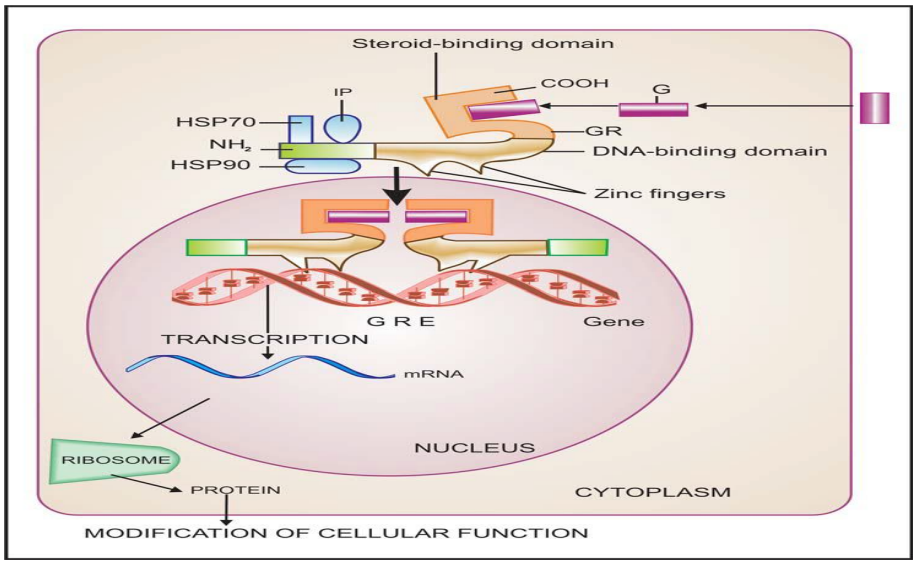

Fig. 5.8: Transduction mechanism of nuclear receptor : Glucocorticoid receptor

5.9 Summary

Receptor is the component of a cell or organism that interacts with a drug and initiates the chain of events leading to the drug's observed effects. Receptors mediate the actions of both pharmacologic agonists and antagonists. Some drugs and many natural ligands, such as hormones and neurotransmitters, regulate the function of receptor macromolecules as agonists; i.e., they activate the receptor to signal as a direct result of binding to it. Some agonists activate a single kind of receptor to produce all of their biologic functions, whereas others selectively promote one receptor function more than another. Other drugs act as pharmacologic antagonists; ie, they bind to receptors but do not activate generation of a signal; consequently, they interfere with the ability of an agonist to activate the receptor. Biological receptors are capable of combining with drugs in a number of ways, and the forces that attract the drug to its receptor, those forces are chemical bonds, and a number of types of bonds such as

hydrophobic interactions, charge-transfer complexes, hydrogen bonds, ion-dipole and dipole-dipole interactions, ionic interactions, covalent bonds, vander walls or london dispersion forces participate in the formation of the initial drug–receptor complex.

5.10 Glossary

- **Depolarisation-** Rapid inflow of sodium ions and cell becomes positive.
- **Dimer-** A combination of two identical molecules to form a single compound.
- **Hyperpolarisation-** Rapid outflow of potassium ions and cell becomes negative.
- **Transcription-** Synthesis of m-RNA molecule that is a complimentary copy of a DNA gene.

5.11 Review questions / Comprehensive Questions

1. What is Pharmacodynamics?
2. Explain mechanism of drug target action.
3. Define receptor. Discuss about GPCR.
4. Explain the transduction mechanism of nuclear receptor.
5. Discuss the molecular structure of GPCR.
6. Explain the transduction mechanism of ligand gated ion channel receptor.
7. Discuss the chemistry of elementary treatment of drug receptor interaction.
8. Enlist the forces that are involved in drug receptor interaction.
9. Explain the transduction mechanism of kinase linked receptor.
10. Discuss the drug-receptor transduction mechanism.

5.12 References and Suggested readings

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Unit - 6

Pharmacokinetic I

Structure of Unit:

6.1 Objectives

In this unit, we set out some general principles underlying the interaction of drugs with living systems. We discuss drug translocation, absorption, distribution, and chemical transformation by drug metabolism, and other processes involved in drug elimination. The aim of drug therapy is to prevent, cure, or control various disease states. To achieve this goal, adequate drug doses must be delivered to the target tissues so that therapeutic, yet nontoxic levels are obtained. The clinician must recognize that the speed of onset of drug action, the intensity of the drug's effect, and the duration of the drug action are controlled by four fundamental pathways of drug movement and modification in the body. First, drug absorption from the site of administration permits entry of the therapeutic agent (either directly or indirectly) into plasma (input). Second, the drug may then reversibly leave the blood stream and distribute into the interstitial and intracellular fluids (distribution). Third, the drug may be metabolized by the liver, kidney, or other tissues. Finally, the drug and its metabolites are eliminated from the body (output) in urine, bile, or feces.

6.2 Introduction: Pharmacokinetics

Pharmacokinetics (Greek: *Kinesis*—movement) — What the body does to the drug. Pharmacokinetics is the quantitative study of drug movement in, through and out of the body. This refers to movement of the drug in and alteration of the drug by the body; includes absorption, distribution, binding/localization/storage, biotransformation (metabolism) and elimination of the drug. (Fig. 6.1).

After entry of the drug into the systemic circulation either by intravascular injection or by absorption from any extravascular site, drug is followed different processes by which changes in its plasma concentration called disposition processes.

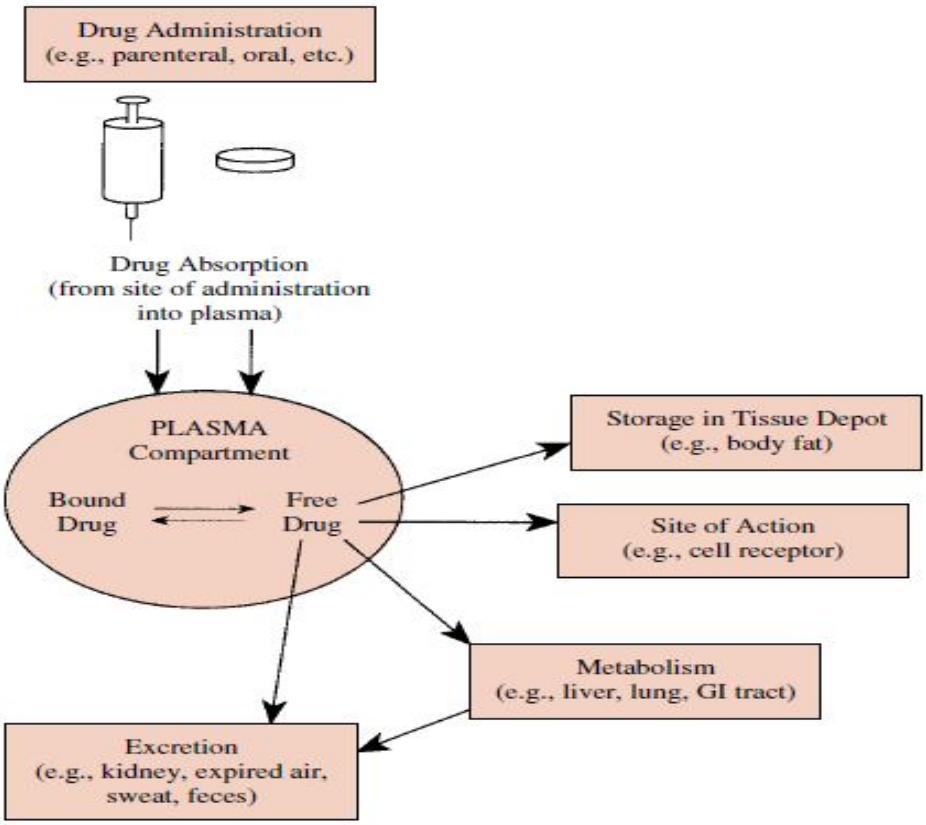

Fig. 6.1: Representation of drug absorption, distribution, metabolism and elimination: Drug disposition

Drug disposition is divided into four stages:

- Absorption from the site of administration
- Distribution within the body
- Metabolism

- Excretion

6.3 Absorption

Absorption is the movement of the drug from its site of administration to the blood stream. Rate and efficiency of absorption depend on the route of administration. For intravenous delivery, absorption is complete, that is, the total dose of drug reaches the systemic circulation. Drug delivery by other routes may result in only partial absorption and thus lower bioavailability.

Routes of drug administration-

The route of administration is determined primarily by the properties of the drug (such as water or lipid solubility, ionization, etc.) and by the therapeutic objectives. There are two major routes of drug administration, enteral and parenteral. (Figure 6.3)

A. Enteral

1. **Oral:**

   Giving a drug by mouth is the most common route of administration, but it is also the most variable, and requires the most complicated pathway to the tissues. Some drugs are absorbed from the stomach; however, the duodenum is often the major site of entry to the systemic circulation because of its larger absorptive surface.

2. **Sublingual:**

   Placement under the tongue allows the drug to diffuse into the capillary network and therefore to enter the systemic circulation directly. Administration of an agent by this route has the advantage that the drug bypasses the intestine and liver and is not inactivated by metabolism.

3. **Rectal:**

   Fifty percent of the drainage of the rectal region bypasses the portal circulation; thus the biotransformation of drugs by the liver is minimized. Both the sublingual and the rectal routes of administration have the additional advantage that they prevent the destruction of the drug by intestinal enzymes or by low pH in the stomach. The rectal route is also useful if the drug induces vomiting when given orally or if the patient is already vomiting.

4. Parenteral:

Parenteral administration is used for drugs that are poorly absorbed from the gastrointestinal (GI) tract, and for agents such as insulin that are unstable in the GI tract. Parenteral administration is used for treatment of unconscious patients and under circumstances that require a rapid onset of action. Parenteral administration provides the most control over the actual dose of drug delivered body.

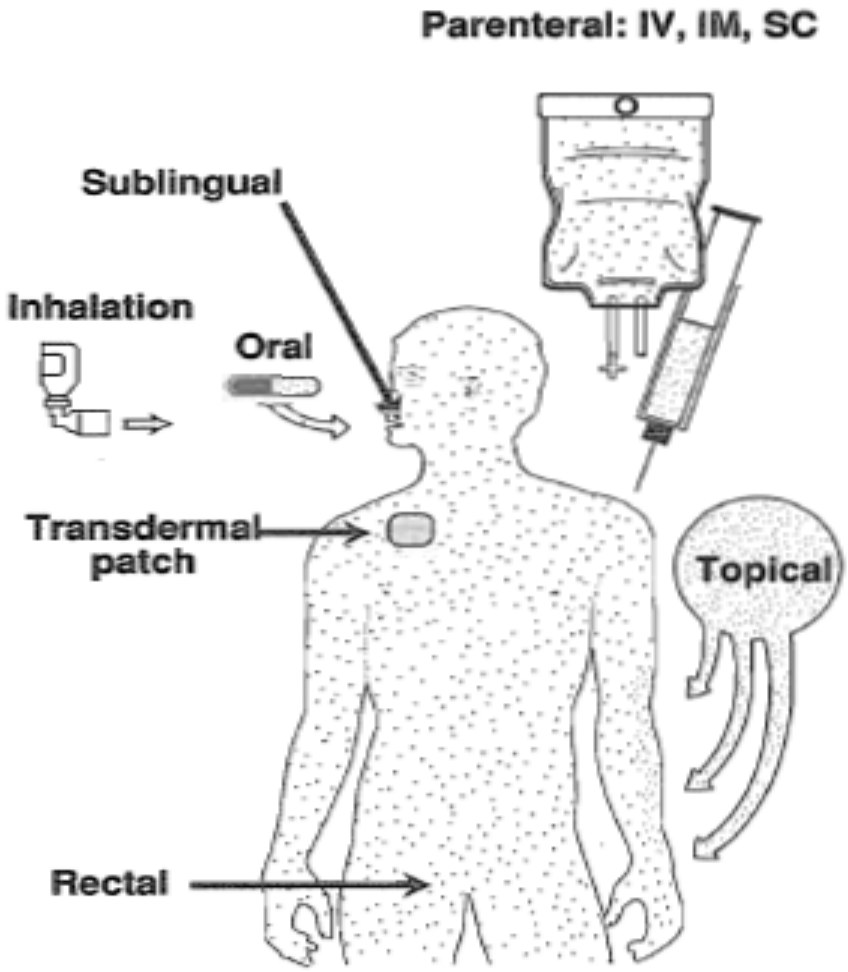

Fig. 6.3: Commonly used routes of drug administration. (IV=intravenous; IM=intramuscular; SC= subcutaneous)

The three major parenteral routes are intravascular (intravenous or intra-arterial), intramuscular, and subcutaneous.

a) **Intravascular:** Intravenous (IV) injection is the most common parenteral route. For drugs that are not absorbed orally, there is often no other choice. With IV administration, the drug avoids the GI tract and, therefore, first-pass metabolism by the liver. This route permits a rapid effect and a maximal degree of control over the circulating levels of the drug.

b) **Intramuscular (IM):** Drugs administered intramuscularly can be aqueous solutions or specialized depot preparations-often a suspension of drug in a nonaqueous vehicle, such as ethylene glycol or peanut oil. Absorption of drugs in aqueous solution is fast, whereas that from depot preparations is slow. As the vehicle diffuses out of the muscle, the drug precipitates at the site of injection. The drug then dissolves slowly, providing a sustained dose over an extended period of time.

c) **Subcutaneous (SC):** This route of administration, like that of IM injection, requires absorption and is somewhat slower than the IV route. SC injection minimizes the risks associated with intravascular injection.

B. Other

1. **Inhalation**:

Inhalation provides the rapid delivery of a drug across the large surface area of the mucous membranes of the respiratory tract and pulmonary epithelium, producing an effect almost as rapidly as by intravenous injection. This route of administration is used for drugs that are gases (for example, some anesthetics), or those that can be dispersed in an aerosol. The route is particularly effective and convenient for patients with respiratory complaints (for example, asthma or chronic obstructive pulmonary disease) as drug are delivered directly to the site of action and systemic side effects are minimized.

2. **Intranasal:**

Desmopressin is administered intranasally in the treatment of diabetes insipidus; salmon calcitonin, a peptide hormone used in the treatment of osteoporosis, is available as a nasal spray.

3. **Intrathecal/Intraventricular:**

It is sometimes necessary to introduce drugs directly into the cerebrospinal fluid (CSF), such as methotrexate in acute lymphocytic leukemia.

4. **Topical:**

Topical application is used when a local effect of the drug is desired. For example, clotrimazole is applied as a cream directly to the skin in the treatment of dermatophytosis, and atropine is instilled directly into the eye to dilate the pupil and permit measurement of refractive errors.

5. **Transdermal:**

This route of administration achieves systemic effects by application of drugs to the skin, usually via a transdermal patch. The rate of absorption can vary markedly depending upon the physical characteristics of the skin at the site of application. This route is most often used for the sustained delivery of drugs, such as the antianginal drug, nitroglycerin.

Factor affecting drug absorption-

- **Blood flow to the absorption site**: Blood flow to the intestine is much greater than the flow to the stomach; thus absorption from the intestine is favored over that from the stomach.
- **Total surface area available for absorption:** Because the intestine has a surface rich in microvilli, it has a surface area about 1,000 times that of the stomach; thus absorption of the drug across the intestine is more efficient.
- **Contact time at the absorption surface:** If a drug moves through the GI tract very quickly, as in severe diarrhoea, it is not well absorbed. Conversely, anything that delays the transport of the drug from the stomach to the intestine delays the rate of absorption of the drug.
- **Aqueous solubility**: A drug given as watery solution is absorbed faster than when the same is given in solid form or as oily solution.
- **Concentration:** Passive diffusion depends on concentration gradient; drug given as concentrated solution is absorbed faster than from dilute solution.
- **Route of administration:** Drug absorption is slower through enternal route than parenteral route.

6.4 Transport across the membrane

All pharmacokinetic processes involve transport of the drug across biological membranes. Biological membrane this is a bilayer (about 100 Å thick) of phospholipid and cholesterol molecules, the polar groups (glyceryl phosphate attached to ethanolamine/choline or hydroxyl group of cholesterol) of these are oriented at the two surfaces and the nonpolar hydrocarbon chains are embedded in the matrix to form a continuous sheet. Extrinsic and intrinsic protein molecules are adsorbed on the lipid bilayer Glycoproteins or glycolipids are formed on the surface by attachment to polymeric sugars, aminosugars or sialic acids. The specific lipid and protein composition of different membranes differs according to the cell or the organelle type. The proteins are able to freely float through the membrane: associate and organize or vice versa. Some of the intrinsic ones, which extend through the full thickness of the membrane, surround fine aqueous pores. Paracellular spaces or channels also exist between certain epithelial/endothelial cells. Other adsorbed proteins have enzymatic, carrier, receptor or signal transduction properties.

Drugs are transported across the membrane by:

1. Passive diffusion and filtration
2. Specialized transport

1. Passive diffusion-

The drug diffuses across the membrane in the direction of its concentration gradient. The drug molecules moves from higher concentration to lower concentration. This process does not require energy. More soluble drugs attains higher concentration in the membrance and diffuse quickly. ex diazepam. **(**Fig. 6.4)

- **Filtration-**

Filtration is passage of drug through aquous pores in the membrane is through paracellular spaces. This can be accelerated by hydrodynamic flow of the solvents. Pore size is about 4A. Filtration is depends upon the molecular size and weight of the drug. If the drug molecules are smaller than the pores, they are filtered easily through the membrane. The intestinal mucosal cells and RBCS have very small pores about 40A however capillaries have large paracelular spaces. **(**Fig. 6.4)

2. Specialized transport-

This can be carrier mediated or by pinocytosis. All the cell membrance express a transmembrane protein serve as a carriers or transporters. Transports combines with their substracts, undergo a conformational changes carrying the substrate to the other side of the membrane where the substrate dissociate and the transports returns back to its original state. Depends upon the requirement of energy, carrier transport in two types-

- Facilitated diffusion
- Active transport

- **Facilitated diffusion-**

This is a type of carrier mediated transport. It does not require energy. The drug attaches to a carriers in the membrane, which facilitates its diffusion across the membrane. The molecules transport from lower concentration to higher concentration. **(**Fig. 6.4) Eg: absorption of vit.B12 from gut, transport of amino acids into the brain.

- **Active diffusion-**

The drug molecules move from a region of low concentration to high concentration against the concentration gradient. It requires energy. **(**Fig. 6.4) Eg. Transport of choline into cholinergic neurons.

- **Pinocytosis-**

It is the process of transport across the cell in particulate from by formation of vesicles. **(**Fig. 6.4)

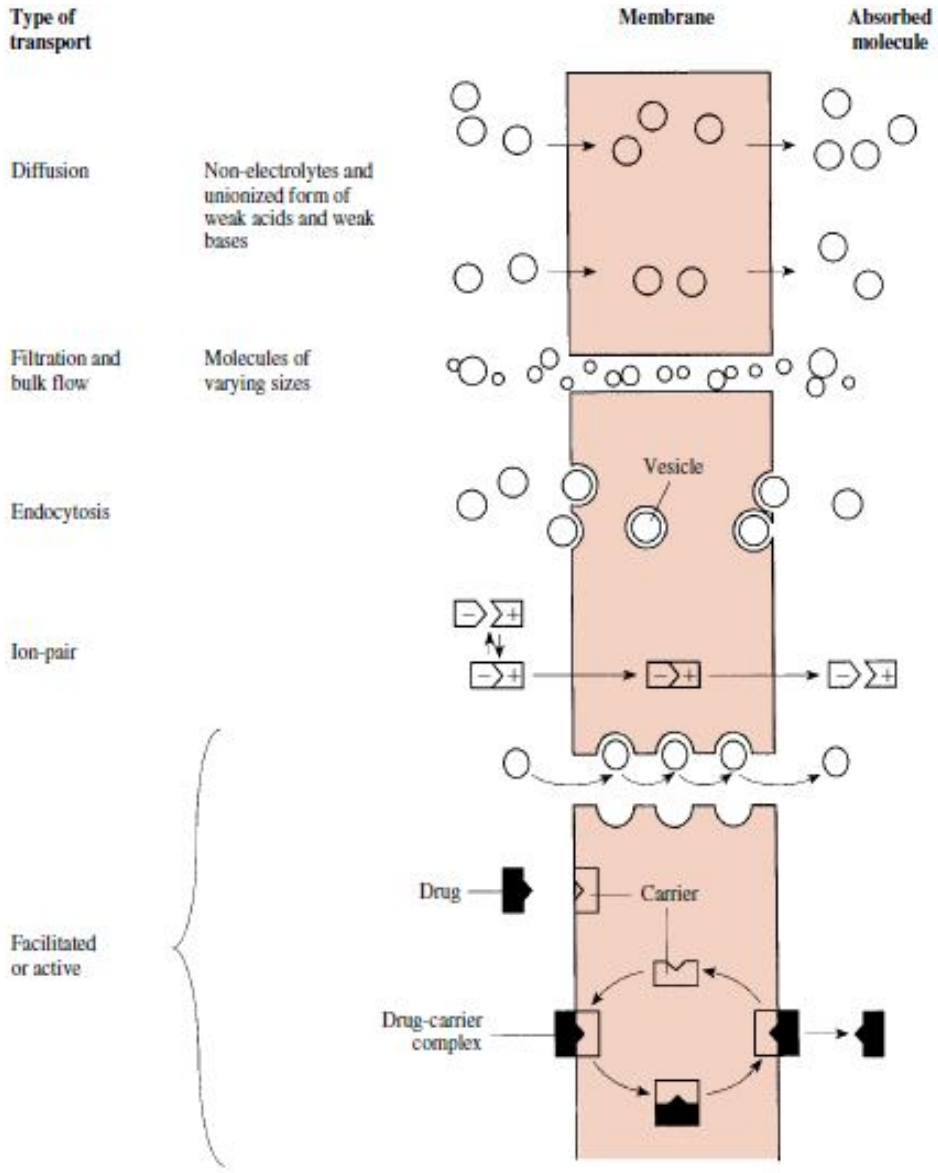

Fig. 6.4: Transport of the drug across the membrane

6.5 Distribution

Distribution is the delivery of drug from the systemic circulation to tissues. Once a drug has gained access to the blood stream, it gets distributed to other tissues that initially had no drug, concentration gradient being in the direction of plasma to tissues. Distribution is defined as the reversible transfer of drugs between body fluid compartments. After drug absorption, a drug enters the systemic circulation and is distributed in the body fluids. Distribution of a drug depends on its-

- Lipid solubility

- Ionization at physiological pH
- Extent of binding to plasma and tissue proteins
- Presence of tissue specific transporters
- Differences in regional blood flow

Apparent volume of distribution-

Is defined as the hypothetical volume of blood fluid into which a drug is uniformly distributed at a concentration equal to that in plasma, assuming the body to be a single compartment.

$$V = \frac{\text{dose administered i.v.}}{\text{plasma concentration}}$$

Redistribution-

Highly lipid soluble drug such as thiopentone , on i.v administration immediately gets distributed to areas of high blood flow such as brain and causes general anaesthesia. Immediately within a few minutes, it recrosses the BBB and gets distributed into the blood and then to the less perfused tissues such as muscle and adipose tissue.

Binding of drugs to plasma proteins

Most drugs found in the vascular compartment are bound reversibly with one or more of the macromolecules in plasma. Although some drugs simply dissolve in plasma water, most are associated with plasma components such as albumin, globulins, transferrin, ceruloplasmin, glycoproteins, and α and β lipoproteins. While many acidic drugs bind principally to albumin, basic drugs frequently bind to other plasma proteins, such as lipoproteins and α1-acid glycoprotein (α1-AGP), in addition to albumin. The extent of this binding will influence the drug's distribution and rate of elimination because only the unbound drug can diffuse through the capillary wall, produce its systemic effects, be metabolized, and be excreted.

Blood-brain barrier (BBB)-

In order to enter the brain, drugs must pass through the endothelial cells of the capillaries of the central nervous system (CNS) or be actively transported. Lipid-soluble drugs readily penetrate into the CNS, since they can dissolve in the membrane of the endothelial cells. Ionized or polar drugs generally fail to enter the CNS, since they are unable to pass through the endothelial cells of the

CNS, which have no slit junctions. These tightly juxtaposed cells form tight junctions that constitute the so called blood-brain barrier.

Blood-Testis Barrier-

The existence of a barrier between the blood and testes is indicated by the absence of staining in testicular tissue after the intravascular injection of dyes. Morphological studies indicate that the barrier lies beyond the capillary endothelial cells and is most likely to be found at the specialized Sertoli–Sertoli cell junction. It appears that Pgp, the efflux transporter protein, also plays a role in forming this blood-testis barrier. This protein probably plays a role in preventing certain chemotherapeutic agents from reaching specific areas of the testis and thus hinders treatment of the neoplasm.

Placental Barrier-

The blood vessels of the fetus and mother are separated by a number of tissue layers that collectively constitute the placental barrier. Drugs that traverse this barrier will reach the fetal circulation. The placental barrier, like the blood-brain barrier, does not prevent transport of all drugs but is selective, and factors that regulate passage of drugs through any membrane (e.g., pKa, lipid solubility, protein binding) are applicable here.

In general, substances those are lipid soluble cross the placenta with relative ease in accordance with their lipid–water partition coefficient and degree of ionization. Highly polar or ionized drugs do not cross the placenta readily. However, most drugs used in labor and delivery are not highly ionized and will cross. They are generally weak bases with pKa values of about 8 and tend to be more ionized in the fetal bloodstream, since the pH of fetal blood is around 7.3 as compared with the maternal blood pH of 7.44. Differences in maternal and fetal blood pH can give rise to unequal concentrations of ionizable drugs in the mother and the fetus.

Factors influencing drug distribution-

These include:

1 Capillary permeability,
2 Blood flow–tissue mass ratio (i.e., perfusion rate),
3 Extent of plasma protein and specific organ binding,
4 Regional differences in ph,
5 Transport mechanisms available, and
6 The permeability characteristics of specific tissue membranes.

6.6 Metabolism

Metabolism (Biotransformation) means chemical alteration of the drug in the body. It is needed to render nonpolar (lipid-soluble) compounds polar (lipid insoluble) so that they are not reabsorbed in the renal tubules and are excreted. The primary site for drug metabolism is liver; others are—kidney, intestine, lungs and plasma. Some agents are initially administered as inactive compounds (pro-drugs) and must be metabolized to their active forms.

Reactions of drug metabolism

The kidney cannot efficiently eliminate lipophilic drugs that readily cross cell membranes and are reabsorbed in the distal tubules. Therefore, lipid-soluble agents must first be metabolized in the liver using two general sets of reactions, called Phase I and Phase II reactions.

- **Phase I/Nonsynthetic/ Functionalization reactions:** a functional group is generated or exposed—metabolite may be active or inactive.
- **Phase II /Synthetic/Conjugation reactions:** metabolite is mostly inactive; except few drugs, e.g. glucuronide conjugate of morphine and sulfate conjugate of minoxidil are active.

Phase I: Phase I reactions function to convert lipophilic molecules into more polar molecules by introducing or unmasking a polar functional group, such as -OH, or -NH2. Phase I metabolism may increase, decrease, or leave unaltered the drug's pharmacologic activity.

Phase II: This phase consists of conjugation reactions. If the metabolite from Phase I metabolism is sufficiently polar, it can be excreted by the kidneys. However, many metabolites are too lipophilic to be retained in the kidney tubules. A subsequent conjugation reaction with an endogenous substrate, such as glucuronic acid, sulfuric acid, acetic acid or an amino acid results in polar, usually more water-soluble compounds that are most often therapeutically inactive. Glucuronidation is the most common and the most important conjugation reaction. (Fig. 6.6)

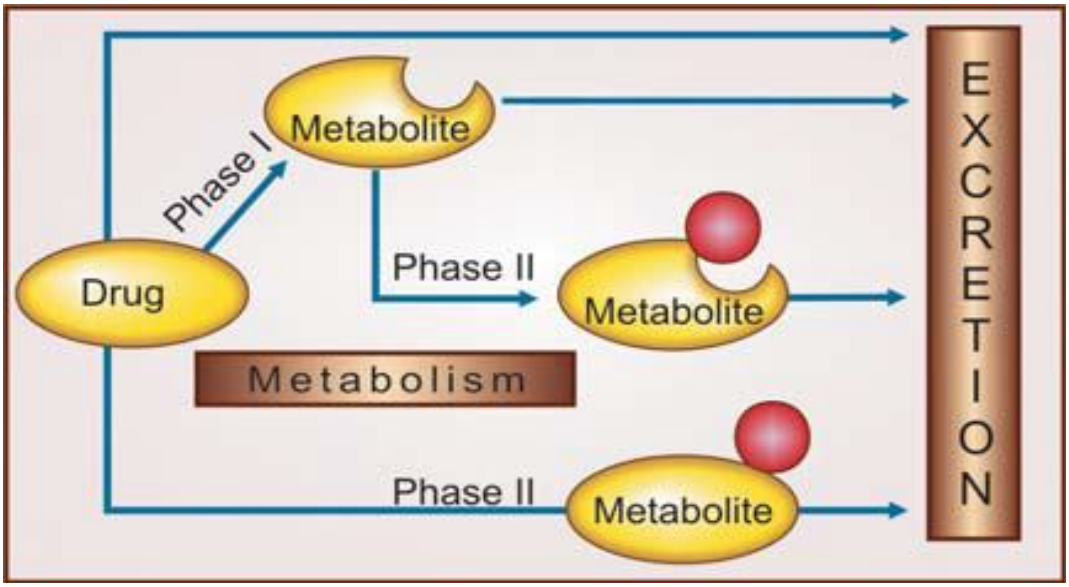

Fig. 6.6: Metabolism of a drug by phase I and phase II reactions

6.7 Elimination

Removal of the drug and its metabolite from the body is known as drug excretion. The main channel of excretion of drugs are the kidneys, others include lungs, bile, feces, sweat, saliva, tears etc.

Drugs and their metabolites are excreted in:

- **Urine-** It is the most important channel of excretion for majority of drugs.
- **Faeces-** Most of the drug present in faeces is derived from bile. Macromolecules (MW > 300) are preferentially eliminated in the bile. Liver transports into bile organic acids (especially drug glucuronides), organic bases and steroids by nonspecific active transport mechanisms. Some drugs are excreted directly in colon, e.g. anthracene purgatives, heavy metals.
- **Exhaled air-** Gases and volatile liquids such as general anaesthetics, paraldehyde, alcohol are eliminated by lungs. Lungs also serve to trap and extrude any particulate matter injected i.v.
- **Saliva and sweat-**These are of minor importance for drug excretion. However, Lithium, pot. Iodide, rifampin and heavy metals are excreted through this way.
- **Milk-** Most drugs enter breast milk by passive diffusion, such as more lipid soluble and less protein bound drugs. However, the excretion of drug in milk is received by the suckling infant. Milk has a lower pH

(7.0) than plasma, basic drugs are more concentrated in it. Although, the total amount of drug reaching the infant through breast feeding is generally small and majority of drugs can be given to lactating mothers without ill effects on the infant.

Renal excretion-

The amount of drug or its metabolites ultimately present in urine is the sum total of glomerular filteration, tubular reabsorption and tubular secretion. (Fig. 6.7)

Glomerular filtration- In the capillaries of glomerulus, larger pores are found which are able to filter all non protein bound drugs. In renal failure or after the age of 50 glomerular rate decreases progressively.

Tubular reabsorption- Lipid-soluble drugs filtered at the glomerulus back diffuse in the tubules because 99% of glomerular filtrate is reabsorbed, but nonlipidsoluble and highly ionized drugs are unable to do so.

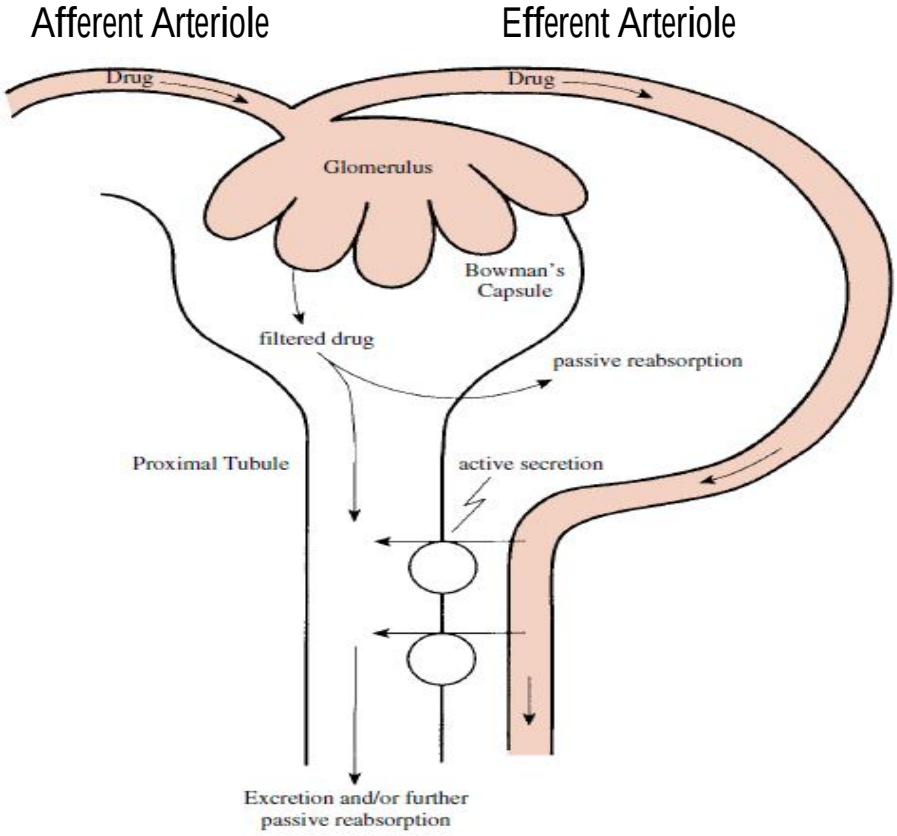

Fig. 6.7*:* Renal excretion of drugs

Tubular reabsorption depends on lipid solubility and ionization of the drug at the existing urinary pH. If changes in urinary pH occur, it affects tubular reabsorption of drugs in the following way:

- Weak acids ionize more and are less reabsorbed in alkaline urine.
- Weak bases ionize more and are less reabsorbed in acidic urine.

Tubular secretion- This is the active transfer of organic acids and bases. Tubular transport mechanisms are not well developed at birth. Duration of action in many drugs, example penicillin, aspirin cephalosporins etc. is longer in neonates. These systems mature during infancy.

Clinical Implications of Renal Excretion

The rate of urinary drug excretion will depend on the drug's volume of distribution, its degree of protein binding, and the following renal factors:

1. Glomerular filtration rate
2. Tubular fluid pH
3. Extent of back-diffusion of the unionized form
4. Extent of active tubular secretion of the compound
5. Possibly, extent of active tubular reabsorption

6.8 Pharmacokinetics of Elimination

Drug elimination is the sum total of metabolic inactivation and excretion. The pharmacokinetics of elimination of a drug gives an idea to devise rational dosage regimens and to modify them according to individual needs.

Pharmacokinetic of elimination, such as :

1. Clearance (CL)
2. Plasma half life
3. Bioavailability (F)

1 Clearance [CL]-

The clearance of a drug is defined as the fraction of the apparent volume of the distribution from which the drug is removed in unit time.

CL = Rate of elimination / plasma concentration of the drug

- **First order kinetics-**

The rate of elimination is directly proportional to the drug concentration, CL remains constant; or a constant fraction of the drug present in the body is eliminated in unit time.

- **Zero order kinetics-**

The rate of elimination remains constant irrespective of drug concentration, CL decreases with increase in concentration; or a constant amount of the drug is eliminated in unit time.

2. Plasma half life (t1/2)-

It is the time required for the plasma concentration of the drug to decrease by 50% of its original value.

Clinical importance of plasma half-life –

- Determine the duration of drug action
- Determine the frequency of drug administration
- Estimate the time required to reach the steady

3. Bioavailability (F)-

Bioavailability refers to the rate and extent of absorption of a drug from a dosage form. It is a measure of the fraction (F) of administered dose of a drug that reaches the systemic circulation in the unchanged form. Bioavailability of drug injected i.v. is 100%, but is frequently lower after oral ingestion because—

- The drug may be incompletely absorbed.
- The absorbed drug may undergo first pass metabolism in the intestinal wall/liver or be excreted in bile.

6.9 Summary

When a drug enters the body, the body begins immediately to work on the drug: absorption, distribution, metabolism (biotransformation), and elimination. These are the processes of pharmacokinetics. These processes modify specific pharmacokinetic parameters. Absorption is the passage of the drug from its site of administration into the blood; distribution is the delivery of the drug to the tissues. Drug metabolism changes the chemical structure of a drug to produce a drug metabolite, which is frequently but not universally less pharmacologically active. Metabolism also renders the drug compound more water soluble and therefore more easily excreted. The two basic parameters are clearance, the measure of the ability of the body to eliminate the drug; and volume of distribution, the measure of the apparent space in the body available to contain the drug.

6.10 Glossary

- **Antianginal-** Preventing or relieving angina pectoris.
- **Dermatophytes-** They are fungi that can cause infections of the skin, hair and nails.
- **Depot preparations-** A substance in a form that tends to keep in at the site of injection so that absorption occurs over a long period.
- **Extra vascular route-** Outside a vessel.
- **Portal circulation-** The circulation of blood through liver.

6.11 Review questions / Comprehensive Questions

1. What is pharmacokinetics?
2. Discuss the factors of absorption, distribution, metabolism and excretion.
3. Discuss the pharmacokinetic parameters of elimination.
4. Define drug disposition.
5. Enumerate the methods of route of drug administration.
6. Discuss the merits and demerits of route of drug administration.
7. Write a short note on parenteral route.
8. Discuss the processes which are involved in translocation of the drug.
9. Explain the metabolic reactions.
10. What are the phase II reactions? Give example.

6.12 References and Suggested readings

- Basic & Clinical Pharmacology (9th ed.)- Bertram G.Katzung (Mc Graw Hill Publisher) 2004.
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Unit - 7

Pharmacokinetic-II

Structure of Unit:

7.1 Objectives

In this unit the students will be able to understand

- We study different parameters of pharmacokinetics by which drug has maximum efficacy and minimum toxicity.
- Using these parameters calculate the dose of drug in individual patient that is safe for patients.
- Pharmacokinetic is very useful in development of formulation by which drug show maximum effectiveness.
- We describe the main pathways of drug elimination by the kidney and biliary excretion.

- We presents a simple approach to quantitative pharmacokinetics, explaining how drug clearance determines the steady-state plasma concentration
- How the characteristics of absorption and distribution and excretion, determine the time course of drug concentration in the blood before and after steady state and how these vary with different dosing regimens.

7.2 Introduction

The goal of therapeutics is to achieve a desired beneficial effect with minimum adverse effects. For this purpose clinician must determine the dose that most closely to achieve this goal. A rational approach of this objective combines the principles of pharmacokinetic with pharmacodyanamic to clarify the dose effect relationship. Pharmacodyanamics is related to concentration-effect part of the interaction whereas the pharmacokinetic deals with dose-concentration part. The pharmacokinetic process of absorption, distribution and elimination determine how rapidly and for how long the drug will appear at target organ. The pharmacodyanamic concept of maximum response and sensitivity determine the magnitude of the effect at a particular concentration. The relationship between dose, drug concentration and effects allows the clinician to consider the various pathologic and physiologic condition of a particular patient that makes him or her different from the average individual in responding to a drug.

7.3 Pharmacokinetics

The standard dose of the drug is based on trials in healthy volunteers and patient with average ability to absorb, distribute and eliminate the drug. This dose will not be suitable for every patient. Dose of drug in individual patient depends on physiologic (e.g. Maturation of organ functions in infants) and pathologic conditions (e.g. Heart failure, renal failure). The two basic parameters are clearance and volume of distribution. Pharmacokinetic is defined as the kinetics of drug absorption, distribution, metabolism and excretion and their relationship with the pharmacological, therapeutic and toxicological response in human being.

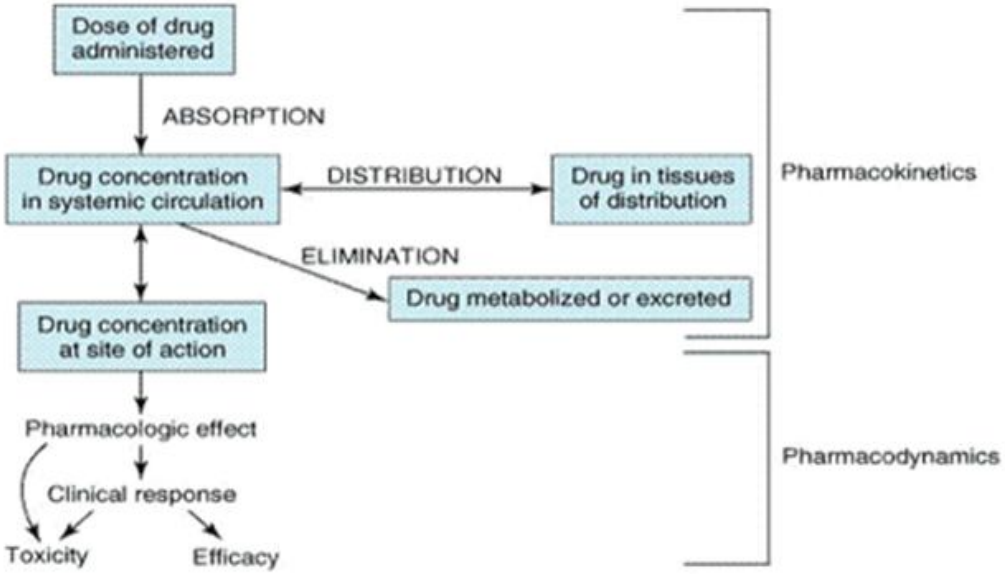


Fig.7.1: The relationship between dose and effect can be separated into pharmacokinetic (dose-concentration) and pharmacodynamic (concentration-effect) components.

Pharmacokinetic means movement of drug. It means pharmacokinetic is the study and characterization of the time course of drug absorption, distribution, metabolism and excretion. It is a quantitative study of drug movement inside and outside of the body. Pharmacokinetic consideration determines the route of drug administration, dose, time of peak action, duration of action and frequency of administration of drug. Pharmacokinetic is used in clinical setting to enhance the safe and effective therapeutic management of individual patient.

There are some important pharmacokinetic parameters like absorption, distribution, metabolism and excretion.

7.4 Absorption

Absorption is movement of drug from its site of administration into the circulation, not only part of administered dose that gets absorbed, but also the rate of absorption is important except when given intravenously, the drug has to cross the biological membranes.

7.5 Disposition

Disposition means what happen with drug after absorption. In other terms it is a process that tends to lower the plasma concentration of drug. In this parameter distribution, metabolism and excretion processes are includes that reduce the plasma concentration of drug. After absorption drug reached into blood stream, drugs are simultaneously distributed throughout the body and eliminated.

7.6 Distribution

After administration of drug in the blood stream, it is ready to distribute to other tissues. The transfer of drug from blood to extracellular fluids and tissue is called distribution. Distribution of drugs depends upon its

- Solubility in lipids
- Differences in regional blood flow
- Binding to plasma and tissue proteins
- Ionization at physiological pH

The distribution of drug continuous till equilibrium occurs between unbound drug in plasma and tissue fluids. A drug is circulated to various organ and tissue from blood. When the process of distribution is complete, different organ and tissues contain varying concentration of drug which can be varied according to blood perfusion rate and volume of tissue. There is direct relationship between the concentration of drug in plasma (C) and amount of drug in the body (X).

$$X \alpha C$$
$$X = V_d \times C$$

V_d is proportionality constant having the unit volume and popularly called as apparent volume of distribution.

V_d is defined as the hypothetical volume of body fluid into which drug is dissolved or distributed. Volume of distribution is calculated as follows:

$$\frac{\text{Amount of drug in body}}{\text{Plasma drug concntration}}$$

The unit of measurement of volume of distribution is ml.

The transfer of drug from blood to extracellular fluids and tissue is called distribution.

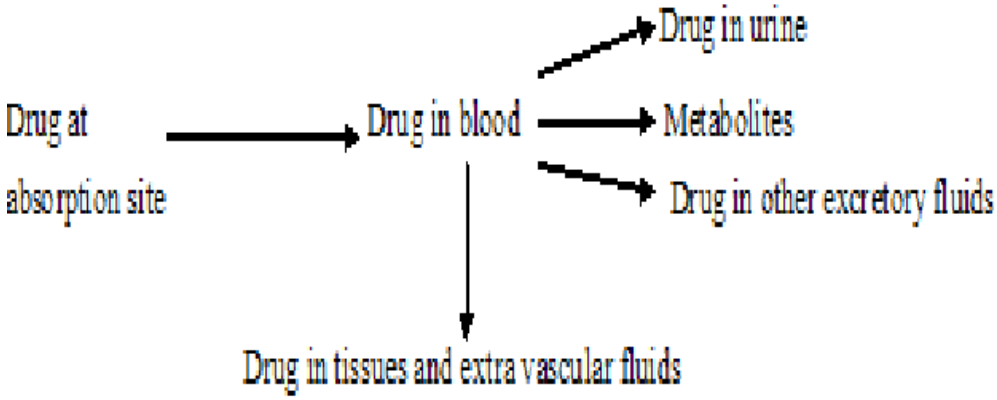


Fig. 7.2: Schematic representation of drug absorption, distribution and elimination

7.7 Drug elimination and clearance

Drug elimination:

It is a process in which transfer of pure drug or metabolite product of drug from blood to urine or other excretory compartment. Drugs are removed from the body by various elimination processes. Drug elimination refers to irreversible removal of drug from the body by all route of elimination. Drug elimination is usually divided into two major components: biotransformation and excretion.

Biotransformation or drug metabolism is the process by which the drug is the drug is chemically converted in the body to a metabolite. Biotransformation is usually an enzymatic process. The enzymes involved in the biotransformation of drugs are located mainly in the liver.

Drug excretion is the removal of intact drug. Nonvolatile drugs are excreted mainly by renal excretion. Volatile drug such as gaseous anaesthetics or drugs with high volatility, are excreted via lungs into aspired air. Drug elimination is described in terms of clearance from well stirred compartments containing uniform drug distribution.

Clearance:

Clearance describes the process of drug elimination from the body or from a single organ. Clearance may be defined as volume of drug in fluid cleared from the body per unit time. The units of clearance are milliliters per minute (ml/min) or liter per hour (L/hr). Drug elimination rate in terms of amount of drug removed per unit time (mg/min).

Clearance is calculated as:

$$Cl = \text{Rate of elimination} / C$$

Where C is plasma concentration

Clearance may also be defined as the rate of drug elimination divided by plasma drug concentration.

$$Cl_T = \text{Elimination rate / Plasma concentration} = \frac{dD_E/dt}{C_p}$$

Where D_E is amount of drug eliminated and dD_E/dt is the rate of elimination. Drug clearance is a pharmacokinetic term for describing drug elimination from the body. Drug clearance (body clearance, total body clearance or Cl_T) considered the entire body as a single drug eliminating system from which many unidentified elimination process may occur.

It is important to note the additive character of clearance. Elimination of drug from the body may involve processes occurring in the kidney, the lung, the liver, and other organs. Dividing the rate of elimination at each organ by the concentration of drug presented to it yields the respective clearance at that organ. Added together, these separate clearances equal total systemic clearance:

$$CL_{renal} = \frac{\text{Rate of elimination}_{kidney}}{C}$$

$$CL_{liver} = \frac{\text{Rate of elimination}_{liver}}{C}$$

$$CL_{other} = \frac{\text{Rate of elimination}_{other}}{C}$$

$$CL_{systemic} = CL_{renal} + CL_{liver} + CL_{other}$$

Renal Clearance: Renal clearance is defined as the volume of blood or plasma that is cleared of drug per unit time through the kidney.

$$Cl_R = \text{Excretion rate/ Plasma concentration} = \frac{dD_U/dt}{C_P}$$

Renal drug excretion: Renal excretion is major route of elimination for many drugs. The process by which the drug is excreted via the kidney may include

- Glomerular filteration

- Active tubular secreation
- Tubular reabsorption

Renal clearance is the ratio of "sum of sum rate of glomerular filteration and active screation minus rate of reabsorption to plasma drug concentration"

$$\textbf{Renal clearance} = \frac{\text{Rate of filtration} + \text{Rate of secreation} - \text{Rate of reabsorption}}{\text{plasma drug concentration}}$$

The two major sites of drug elimination are the kidneys and the liver. Clearance of unchanged drug in the urine represents renal clearance. Within the liver, drug elimination occurs via biotransformation of parent drug to one or more metabolites, or excretion of unchanged drug into the bile, or both. For most drugs, clearance is constant over the concentration range encountered in clinical settings, i.e., elimination is not saturable, and the rate of drug elimination is directly proportional to concentration.

$$\text{Rate of elimination} = CL \times C$$

This is usually referred to as first-order elimination. When clearance is first-order, it can be estimated by calculating the area under the curve (AUC) of the time-concentration profile after a dose. Clearance is calculated from the dose divided by the AUC.

The different kinetic models are describes by clearance:

1. **Capacity-Limited Elimination:** For drugs that exhibit capacity-limited elimination (eg, phenytoin, ethanol), clearance will vary depending on the concentration of drug that is achieved. Capacity-limited elimination is also known as saturable, dose- or concentration-dependent, nonlinear, and Michaelis-Menten elimination.

   Most drug elimination pathways will become saturated if the dose is high enough. When blood flow to an organ does not limit elimination, the relation between elimination rate and concentration (C) is expressed mathematically as follow:

   $$\text{Rate of elimination} = \frac{V_{max} \times C}{K_m + C}$$

   The maximum elimination capacity is V_{max}, and K_m is the drug concentration at which the rate of elimination is 50% of V_{max}. At concentrations that are high relative to the K_m, the elimination rate is

almost independent of concentration—a state of "pseudo-zero order" elimination. If dosing rate exceeds elimination capacity, steady state cannot be achieved: The concentration will keep on rising as long as dosing continues. This pattern of capacity-limited elimination is important for three drugs in common use: ethanol, phenytoin, and aspirin. Clearance has no real meaning for drugs with capacity-limited elimination, and AUC cannot be used to describe the elimination of such drugs.

2. **Flow-Dependent Elimination:** In contrast to capacity-limited drug elimination, some drugs are cleared very readily by the organ of elimination, so that at any clinically realistic concentration of the drug, most of the drug in the blood perfusing the organ is eliminated on the first pass of the drug through it. The elimination of these drugs will thus depend primarily on the rate of drug delivery to the organ of elimination. Such drugs can be called "high-extraction" drugs since they are almost completely extracted from the blood by the organ. Blood flow to the organ is the main determinant of drug delivery, but plasma protein binding and blood cell partitioning may also be important for extensively bound drugs that are highly extracted.

There are several other route of drug excretion called non renal route of drug excretion. The various such excretion processes are:

1. **Biliary excretion:** the ability of liver to excrete the drug in bile is expressed by biliary clearance.

$$\text{Biliary clearance} = \frac{\text{Biliary clearance rate}}{\text{Plasma drug concentration}}$$

2. **Pulmonary excretion:** Gaseous and volatile substances such as the general anaesthetics are absorbed through the lungs by simple diffusion.
3. **Salivary excretion:** Excretion of drug through saliva is a passive diffusion process. The bitter after taste in the mouth of patient on medication is an indication of drug excretion in saliva. The process is responsible for side effects such as black hairy tongue in patient receiving antibiotics.
4. **Mammary excretion:** Excretion of drug in milk is important since it can entered into the breast feeding infant.
5. **Skin/ dermal excretion:** Drugs excreted through the skin via sweat. Passive excretion of drugs and their metabolites through skin is responsible

to some extent for the urticaria and dermatitis and other hypersensitivity reactions.

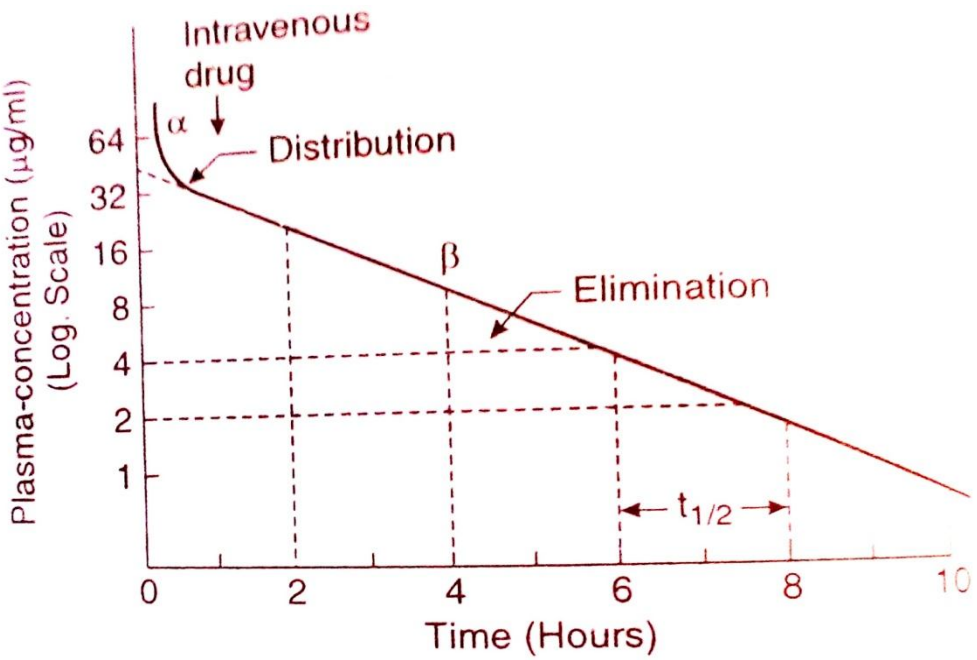
6. **Gastrointestinal excretion:** Excretion of drug through GIT usually occur after parenteral administration when the concentration gradient favorable for passive diffusion.
7. **Genital excretion:** reproductive tract and genital secretions may contain the excreted drugs. Some drugs have been detected in semen.

7.8 Plasma half life

It is the time taken by the drug for its plasma concentration to be reduced to half of its original value. At least distribution half life and elimination half life can be calculated.

When the drug is given intravenously, which have one compartment distribution and first order of elimination. A plot is drawn between plasma concentration and time, which show two slopes:

- Due to distribution, initially declining an α- phase.
- Due to elimination, later less declining ß-phase



The elimination half life is called 'half life' of drugs.

Elimination $t_{1/2}$ is:

$$T_{1/2} = \frac{\ln 2}{K}$$

Where ln 2 is natural logarithm of 2 or 0.693

K= elimination rate constant of the drug i.e., the fraction of total amount of drug in the body which are removed in per unit time.

$$K = Cl/V_d$$

$$\text{Now } t_{1/2} = 0.693 \times V_d / Cl$$

Repeated drug administration: if drug is repeated in ashort duration of time, it accumulate in the body unless elimination balances input and a steady state plasma concentration (C_{ss}) is attained. C_{ss} is expressed as:

$$C_{SS} = \frac{\text{Dose rate}}{\text{Clearance}}$$

If the therapeutic plasma concentration of drug and clearance is known than the dose rate can be obtained as:

$$\text{Dose rate} = \text{Target } C_{SS} \times \text{Clearance}$$

If a drug is taken orally only a fraction of dose reaches in systemic circulation in the active form, in this case:

$$\text{Dose rate} = \frac{\text{Target Css} * \text{Clearance}}{\text{Fraction}}$$

Target level strategy: For those drugs whose effects are not easily quantifiable and safety margin is small than target level strategy is best to achieve a certain plasma concentration.

Drugs with small $t_{1/2}$, means upto 2-3 hours, are administered at conventional intervals. Generally 6-12 hours are required to achieve target levels.

Drugs whose $t_{1/2}$, is longer a dose which is sufficient to attain the target concentration after single administration. If repeated administration of dose will be given than drug will accumulate and produce toxicity. If the dose is such as to attain target level at steady state, the therapeutic effect will be delayed by about 4 half lives. These drugs are generally taken by initial loading dose and maintenance dose.

Loading Dose: Loading dose is a single or few rapidly repeated doses which are given in the beginning to attain target concentration, it may be expressed.

$$\text{Loading dose:} = \frac{\text{Target plasma concentration X Volume}}{\text{Fraction of drug (F)}}$$

Maintenance dose:

In most clinical situations, drugs are administered in such a way as to maintain a steady state of drug in the body, ie, just enough drug is given in each dose to replace the drug eliminated since the preceding dose. Thus, calculation of the appropriate maintenance dose is a primary goal. Clearance is the most important pharmacokinetic term to be considered in defining a rational steady state drug dosage regimen. At steady state, the dosing rate ("rate in") must equal the rate of elimination ("rate out").

Maintenance dose = Clearance x C_{SS}

= Dosing rate x dosing interval

Thus, if the desired target concentration is known, the clearance in that patient will determine the dosing rate. If the drug is given by a route that has a bioavailability less than 100%, then the dosing rate will be predicted.

Note that the steady-state concentration achieved by continuous infusion or the average concentration following intermittent dosing depends only on clearance. The volume of distribution and the half-life need not be known in order to determine the average plasma concentration expected from a given dosing rate or to predict the dosing rate for a desired target concentration.

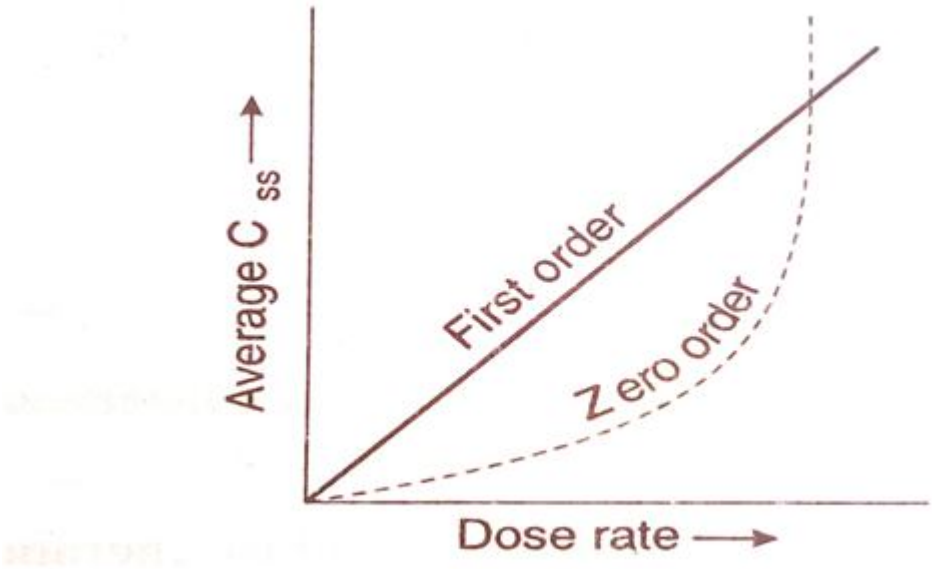


Fig.7. 3: Relationship between dose rate and average steady state plasma concentration of drugs eliminated by first order and zero order kinetics

7.9 Bioavailability

Bioavailability is defined as the fraction of unchanged drug reaching the systemic circulation after administration by any route. The area under the blood concentration- time curve (area under the curve, AUC) is a common measure of the extent of bioavailability for a drug given by a particular route. For an intravenous dose of the drug, bioavailability is assumed to be equal to unity. For a drug administered orally, bioavailability may be less than 100% because

- Incomplete extent of absorption
- First-pass elimination

Extent of Absorption: After oral administration, a drug may be incompletely absorbed. This is mainly due to lack of absorption from the gut. Other drugs are either too hydrophilic (eg, atenolol) or too lipophilic (eg, acyclovir) to be absorbed easily, and their low bioavailability is also due to incomplete absorption. If too hydrophilic, the drug cannot cross the lipid cell membrane, if too lipophilic; the drug is not soluble enough to cross the water layer adjacent to the cell. Drugs may not be absorbed because of a reverse transporter associated with P-glycoprotein. This process actively pumps drug out of gut wall cells back into the gut lumen. Inhibition of P-glycoprotein and gut wall metabolism, eg., by grapefruit juice, may be associated with substantially increased drug absorption.

First-Pass Elimination: After absorption of drug across the gut wall, the portal blood delivers the drug to the liver prior to entry into the systemic circulation. A drug can be metabolized in the gut wall (eg, by the CYP3A4 enzyme system) or even in the portal blood, but most commonly it is the liver that is responsible for metabolism before the drug reaches the systemic circulation. In addition, the liver can excrete the drug into the bile. Any of these sites can contribute to this reduction in bioavailability, and the overall process is known as first-pass elimination.

Rate of absorption: The rate of absorption is determined by the site of administration and the drug formulation. Both the rate of absorption and the extent of input can influence the clinical effectiveness of a drug. The mechanism of drug absorption is said to be zero-order when the rate is independent of the amount of drug remaining in the gut. In contrast, when the full dose is dissolved in gastrointestinal fluids, the rate of absorption is usually proportional to the gastrointestinal concentration and is said to be first-order.

Oral formulation of a drug from different manufacturers or different batches from the same manufacturer may have the same quantity (chemically equivalent) of drug but may not yield same blood level (biologically equivalent).

If the rate and extent of bioavailability of drug is not different under suitable test conditions, two preparation of drug are considered bioequivalent.

As the figure given below, B is more slowly absorbed than A, B may not produce therapeutic effect and C is absorbed to lesser extent that means has lower bioavailability.

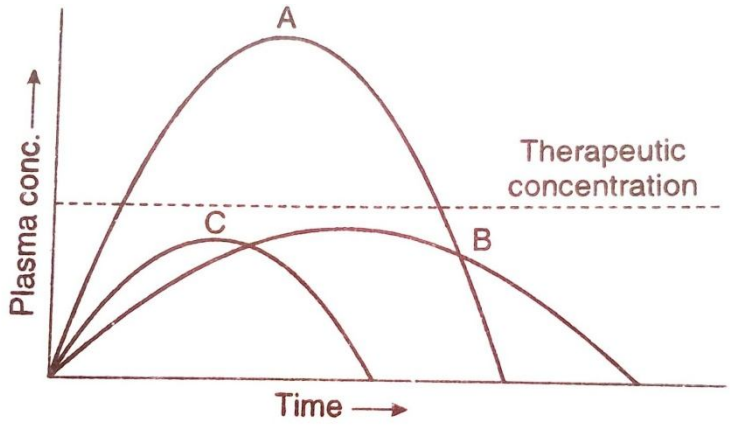


Fig.7. 4: Plasma concentration time curve showing bioavailability differences between three cases of drug having same amount

7.10 Pharmacokinetic in drug development process

To use a drug for longer time, it is generally advantageous to modify a drug by following manner:

- If frequency of administration of a drug is reduced, it may be used for a longer period.
- It is better to use a contraceptive monthly or quarterly rather than to be taken daily.
- A single monitoring dose is less likely to be forgotten than a 6 or 8 hourly regimen.
- Drug effect could be maintained overnight without disturbing sleep.
- Large fluctuations in plasma concentration are avoided.

The processes which are utilized for developing a drug and its action are given as follows:

1. **By prolonging absorption from site of administration:**

a. **Oral:** Drug particles are coated with resins, plastic materials etc. which disperse release of the active ingredients in the GIT. A semi permeable membrane is used to control the release of drug from the tablet and capsule. This technique may prolong the action by 4 to 6 hours and not beyond this time because in that time drug particles reach in the colon.

b. **Parenteral:** The subcutaneous and intramuscular injection of drug in insoluble form or as oily solution pallet implantation and biodegradable implant may develop a drug action.

c. **Transdermal drug delivery:** The drug which is used as ointment, in adhesive patchs, or strips applied on skin is becoming popular.

2. By increasing plasma protein binding: Development of drugs have been made by increasing plasma protein binding which may be slowly released in the free active form e.g. Sulphadoxine.
3. **By retarding rate of metabolism:** Some small chemical modifications may affect the rate of metabolism without affecting the biological action, such as addition of ethinyl group in estradiol makes it longer acting and suitable for use as oral contraceptive.
4. **By retarding renal excretion:** The tubular secretion of drugb being an active process which can be reduced by a competing substance, for instance, probenecid prolongs time of action of penicillin and ampicillin.

7.11 Summary

The basic tenet of pharmacokinetic is that the magnitude of both the desired response and toxicity are functions of drug concentration in blood. It is not only the efficacy of a drug at the site of action that determine the intensity and duration of its pharmacologic or therapeutic effects but also the amount of drug and the rate at which the drug gets to the site of action. The vital process of the body may delay the transport of drug molecules across membranes, convert drug molecule into metabolites, and remove them from body as metabolites and/or unchanged form. In this results, therapeutic failure as a result of drug concentration being too low or unacceptable toxicity as a consequence of too high drug concentration. Between these limits of concentration lies a region associated with therapeutic success. The region may be regarded as a therapeutic range or “therapeutic window”

7.12 Glossary

Area under the curve: It represents the total integrated area under the plasma level time profile and expresses the total amount of drug that comes into the systemic circulation after its administration.

Pharmacodyanamic: It is the study of drug effects

Heart failure: In ability of the heart to circulate blood effectively enough to meet the body metabolic needs.

Renal failure: Raise in the serum creatinin level of 25% or more

Physiological pH: pH of the blood circulation

Blood perfusion: The blood goes into the particular organ by the artery.

Steady state concentration: The concentration of drug in plasma approaches a constant value.

Urticaria: Multiple swollen raised areas on the skin that is intensely itchy

Dermatitis: An inflammatory rash marked by the itching and redness.

7.13 Review questions / Comprehensive Questions

1. Define- (a) disposition and (b) distribution.
2. It is better to express V_d in liters/Kg body weight. Why?
3. What are various non renal route of drug excretion?
4. What is the reason bitter after taste of medicament in patient on drug therapy?
5. What are the importances of pharmacokinetic in drug development process?
6. Explain the extent of absorption and first pass elimination.
7. Write and explain the renal excretion.
8. Express the relationship between pharmacokinetic and pharmacodyanamic.
9. Why drugs show lower bioavailability?

7.14 References and Suggested readings

- Foye's Principles of Medicinal Chemistry, David A. Williams, Thomas L. Lemke (Fifth edition) 2005, B.I. Publication and Pvt. Ltd.
- Biopharmaceutics and Pharmacokinetics A Treatise, D. M. Bramankar and Sunil B. Jaiswal (First edition) 1995 Vallabh Prakashan

- Essential of Medical Pharmacology (Seventh edition)- KD Tripathi (JAYPEE Publisher) 2013.
- Basic & Clinical Pharmacology (ninth edition)- Bertram G.Katzung (Mc Graw Hill Publisher) 2004.

Unit - 8

Pharmacodynamics-I

Structure of Unit:

8.1 Objectives

The aim of this chapter is to present current approaches to

- Pharmacodynamics
- Mechanisms of drug action
- Enzyme
- Enzyme stimulation
- Enzyme inhibition
- Sulfonamides

8.2 Introduction: Pharmacodynamics

Pharmacodynamics is the study of drug effects. It attempts to elucidate the complete action-effect sequence and the dose-effect relationship. Modification of the action of one drug by another drug is also an aspect of pharmacodynamics.

Pharmacodynamics is the study that establishes and elucidates relationships between concentrations of a drug at the receptor or target organ (effect site) and the intensity of its pharmacological effect. Pharmacodynamic studies can provide a means for identifying important pharmacological and toxicological

properties of a drug in animals and humans, including, e.g., efficacious target concentrations, drug safety margin, potential risk factors, and the presence of active metabolites. The effect site (site of action) of a drug can be a target receptor/enzyme(s) or an organ(s) where the initial pharmacological responses to the drug are produced.

Enzymes are special types of receptors. The receptors discussed in Chapter 3 are mostly membrane-bound proteins that interact with natural ligands and agonists to form complexes that then elicit a biological response. Subsequent to the response, the ligand is released intact. Enzymes, most of which are soluble and found in the cytosol of cells, interact with substrates to form complexes, but, unlike receptors, it is from these enzyme–substrate complexes that enzymes catalyze reactions, thereby transforming the substrates into products that are released. Therefore, the two characteristics of enzymes are their ability to recognize a substrate and to catalyze a reaction with it.

8.3 Enzymes

Enzymes are natural proteins that catalyze chemical reactions; ribonucleic acids (RNA) also can catalyze chemical reactions. The first enzyme to be recognized as a protein was jack bean urease, which was crystallized in 1926 by Sumner and was shown to catalyze the hydrolysis of urea to CO_2 and NH_3.

Almost all biological reactions are carried out under catalytic influence of enzymes; hence, enzymes are a very important target of drug action. Drugs can either increase or decrease the rate of enzymatically mediated reactions. However, in physiological systems enzyme activities are often optimally set. Thus, stimulation of enzymes by drugs, that are truly foreign substances, is unusual. Enzyme stimulation is relevant to some natural metabolites only, e.g. pyridoxine acts as a cofactor and increases decarboxylase activity. Several enzymes are stimulated through receptors and second messengers, e.g. adrenaline stimulates hepatic glycogen phosphorylase through β receptors and cyclic AMP. Stimulation of an enzyme increases its affinity for the substrate so that rate constant (kM) of the reaction is lowered. Apparent increase in enzyme activity can also occur by enzyme induction, i.e. synthesis of more enzyme protein. This cannot be called stimulation because the kM does not change.

How do Enzymes Work?

Following scheme shows a generalized scheme for an enzyme-catalyzed reaction, where S is the starting reactant (substrate), E is the enzyme, TS is the transition state, and P is the product. In general, enzymes function by lowering

transition state energies and energetic intermediates and by raising the ground state energy (ground state destabilization). The transition state for an enzyme-catalyzed reaction, just as in the case of a chemical reaction, is a high energy state having a lifetime of about 10^{-14} s, the time for one bond vibration. There is no spectroscopic method that can detect the transition state structure in an enzyme.

$$E + S \overset{K_s}{\rightleftharpoons} E^*S \overset{k_{cat}}{\rightleftharpoons} E^*TS \rightleftharpoons E^*P \rightleftharpoons E + P$$

An enzyme-catalyzed reaction always is initiated by the formation of an enzyme–substrate (or E·S) complex, from which the catalysis takes place. The concept of an E·S complex was originally proposed independently in 1902 by Brown and Henri; this idea is an extension of the 1894 *lock and key hypothesis* of Fischer in which it was proposed that an enzyme is the lock into which the substrate (the key) fits. These curiosities led Koshland in 1958 to propose the induced-fit hypothesis discussed in relationship to receptors in Chapter-3, when a substrate begins to bind to an enzyme, interactions of various groups on the substrate with particular enzyme functional groups are initiated, and these mutual interactions induce a conformational change in the enzyme.

Enzyme catalysis is characterized by two features: *specificity* and *rate acceleration*. The active site contains moieties that are responsible for both of these properties of an enzyme, namely, amino acid residues and, in the case of some enzymes, cofactors. A *cofactor*, also called a *coenzyme*, is an organic molecule or a metal ion that binds to the active site, in some cases covalently and in others non-covalently, and is essential for the catalytic action of those enzymes that require cofactors.

8.4 Mechanisms of enzyme catalysis

Once the substrate binds to the active site of the enzyme via the interactions noted in Chapter 3, there are a variety of mechanisms that the enzyme can utilize to catalyze the conversion of the substrate to product. The most common mechanisms are approximation, covalent catalysis, general acid–base catalysis, electrostatic catalysis, desolvation, and strain or distortion. All of these act by stabilizing the transition state energy or destabilizing the ground state (which is generally not as important as transition state stabilization).

- *Approximation* is the rate enhancement by proximity, that is, the enzyme serves as a template to bind a substrate so that it is close to reactive groups

of the enzyme (or to bind multiple substrates so that they are close to each other) in the reaction center.

- *Covalent catalysis* involves covalent bond formation as a result of attack by an enzyme nucleophile at an electrophilic site on the substrate.
- In any reaction where proton transfer occurs, *general acid catalysis* and/or *general base catalysis* can be an important mechanism for specificity and rate enhancement. There are two kinds of acid–base catalysis: general catalysis and specific catalysis. If catalysis occurs by a hydronium (H_3O^+) or hydroxide (HO^-) ion and is determined only by the pH, not the buffer concentration, it is referred to as *specific acid or specific base catalysis*, respectively.
- An enzyme *electrostatic catalyzes* a reaction by stabilization of the transition state and by destabilization of the ground state.
- The *desolvation* hypothesis posits that an enzyme active site, which is largely or completely devoid of water, can mimic the reaction environment found in the gas phase.
- *Strain or distortion* of the bound substrate is essential for catalysis, play an important role in the reactivity of molecules. The much higher reactivity of epoxides relative to other ethers demonstrates this phenomenon and is another type of ground state destabilization.

A very important bacterial enzyme in medicinal chemistry is the peptidoglycan transpeptidase, the enzyme that catalyzes the cross-linking of peptidoglycan strands to make the bacterial cell wall.

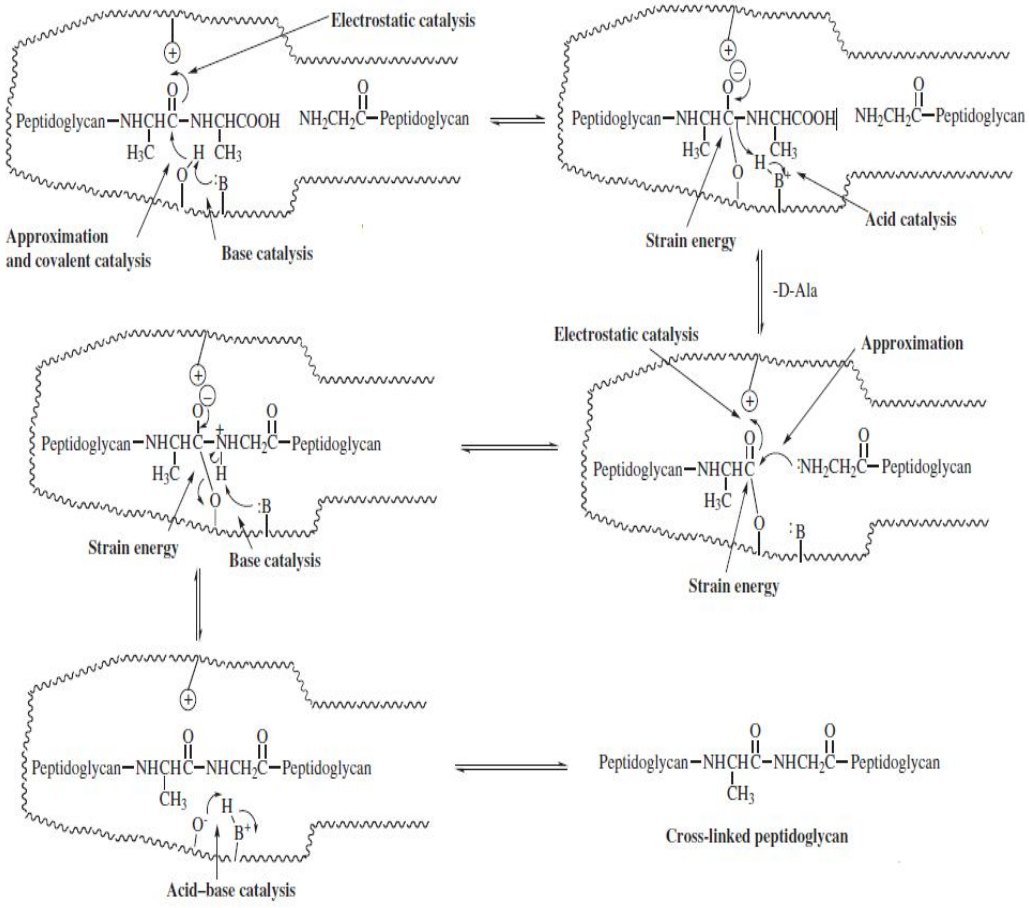


Fig. 8.1: Hypothetical mechanism for peptidoglycan transpeptidase

8.5 Elementary treatment of enzyme stimulation

Many of the top 100 drugs sold worldwide for elementary treatment of enzyme stimulation. In recent years, enzyme inhibitors not only have provided an increasing number of potent therapeutic agents for the treatment of diseases, but also have significantly advanced the understanding of enzymatic transformations. The Elementary treatment of enzyme stimulation is current approaches to socalled rational inhibitor design, which uses knowledge of enzymic mechanisms and structures in the design process.

These enzymes constitute the various metabolic pathways that, in concert, provide the requirements for the viability of the cell. A selective treatment of enzyme stimulation may block either a single enzyme or a group of enzymes, leading to the disruption of a metabolic pathway(s). This will result in either a decrease in the concentration of enzymatic products or an increase in the concentration of enzymatic substrates. The effectiveness of treatment of enzyme stimulation as a therapeutic agent will depend on

1. The potency of the inhibitor,
2. Its specificity toward its target enzyme,
3. The choice of metabolic pathway targeted for disruption, and
4. The inhibitor or a derivative possessing appropriate pharmacokinetic characteristics.

Clearly, the choice of target enzyme is also of prime importance for chemotherapy, Good bioavailability of the drug is also crucial for the drug to reach its site of action in the body in effective therapeutic concentrations. For example, highly polar or charged compounds, such as phosphorylated compounds, frequently cannot readily cross cell membranes and are therefore generally less useful as drugs. Physical approaches to facilitate the transport of this class of compounds into the cell include the use of liposomes or nanoparticles. Chemical approaches may also be employed. These include the use of prodrugs, in which functional groups on the inhibitor are modified in such a manner that they are able to be taken up by the cell and, later, metabolically converted to the active drug.

The human body, even though its defenses are constantly on guard, is still susceptible to invasion by foreign pathogens. Since the development of the sulfa drugs (sulfonamides), enzyme inhibitors have played a vital role in controlling these infectious agents. Following tables provide a list of enzyme inhibitors that have been used in the treatment of the various diseases caused by these agents. All these compounds needed to satisfy the usual requirements for specificity and low toxicity.

Table-8.1: Examples of Enzyme Inhibitors Used in the Treatment (elementary treatment of enzyme) of Bacterial, Fungal, Viral, and Parasitic Diseases

S. No.	Clinical Use	Enzyme Inhibited/ treated	Inhibitor/Treatment
1.	Antibacterial	Dihydropteroate synthetase	Sulphonamides
2.	Antibacterial	Dihydrofolate reductase	Trimethoprim, methotrexate
3.	Antibacterial	Alanine racemase	D-Cycloserine
4.	Antibacterial	Transpeptidase	Penicillins, cephalosporins
5.	Antifungal	Fungal sterol l4a-	Clotrimazole,ketoconazol

		demethylase	e
6.	Antifungal	Fungal squalene epoxidase	Terbinafine, naftifine
7.	Antiviral	Thymidine kinase and thymidylate kinase	Idoxuridine
8.	Antiviral	DNA, RNA polymerases	Cytosine arabinoside
9.	Antiviral	Viral DNA polymerase	Acyclovir, vidarabine
10	Antiviral	HIV reverse transcriptase	Dideoxyinosine, zidovudine
11	Antiviral	HIV protease	Saquinavir
12	Antiviral	Influenza virus neuraminidase	Zanamavir, oseltamivir
13	Antiprotozoal	Pyruvate dehydrogenase	Organoarsenical agents
14	Antiprotozoal	Ornithine decarboxylase	a-Difluoromethylornithine

Table- 8.2: Examples of Enzyme Inhibitors/Trearment Used in the Cancer

S. No.	Type of Cancer	Enzyme Inhibited/ treated	Inhibitor/Treatment
1.	Benign prostatic hyperplasia	Steroid 5a-reductase	Finasteride
2.	Estrogen-mediated breast cancer	Aromatase	Arninoglutethimide, fadrozole
3.	Leukemia, osteosarcoma, head, neck, and breast cancer	Dihydrofolate reductase	Methotrexate
4.	Colorectal cancer	Thymidylate synthase	5-Fluorouracil
5.	Leukemia	Glutamine-PRPP amidotransferase	6-Mercaptopurine, azathioprine
6.	Small-cell lung cancer, non- Hodgkin's lymphoma	Topoisomerase-II	Etoposide
7.	Hairy-cell leukemia	Adenosine deaminase	Pentostatin

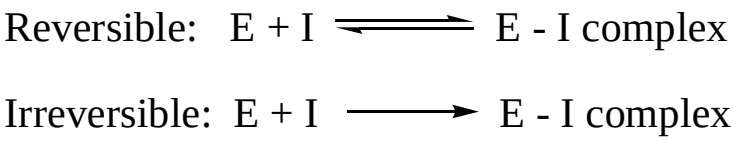
Table-8.3: Examples of Enzyme Inhibitors/Treatment Used in Various Human Disease States

S. No.	Clinical Use	Enzyme Inhibited/ treated	Inhibitor/Treatment
1.	Epilepsy	GABA transaminase	Vinyl GABA
2.	Epilepsy	Carbonic anhydrase	Sulthiame
3.	Epilepsy	Succinic semialdehyde dehydrogenase	Sodium valproate
4.	Antidepressant	Monoamine oxidase (MAO)	Tranylcypromine, phenelzine
5.	Antihypertensive	Angiotensin converting enzyme	Captopril, enalaprilat
6.	Cardiac disorders	Na^+, K^+-ATPase	Cardiac glycosides
7.	Gout	Xanthine oxidase	Allopurinol
8.	Ulcer	H^+, K^+- ATPase	Omeprazole
9.	Hyperlipidemia	HMG-CoA reductase	Atorvastatin, simvastatin
10	Anti-inflammatory	Prostaglandin synthase, Cyclooxygenase (COX) I and II	Aspirin, naproxen, ibuprofen
11	Arthritis	Cyclooxygenase (COX) II	Celecoxib
12	Glaucoma	Acetylcholinesterase	Neostigmine
13	Glaucoma	Carbonic anhydrase II	Acetazolamide, dichlorphenamide

8.6 Enzyme inhibition

Any compound that slows down or blocks enzyme catalysis is an *enzyme inhibitor*. If the interaction with the *target enzyme* (specific enzyme for inhibition) is irreversible (usually covalent), then the compound is a special type of enzyme inhibitor referred to as an *enzyme inactivator*.

Enzyme inhibitors can be grouped into two general categories: reversible and irreversible inhibitors. As the name implies, inhibition of enzyme activity by a *reversible inhibitor* is reversible, suggesting that noncovalent interactions are involved. An *irreversible enzyme inhibitor*, also called an *enzyme inactivator*, is one that prevents the return of enzyme activity for an extended period, suggesting the involvement of a covalent bond.

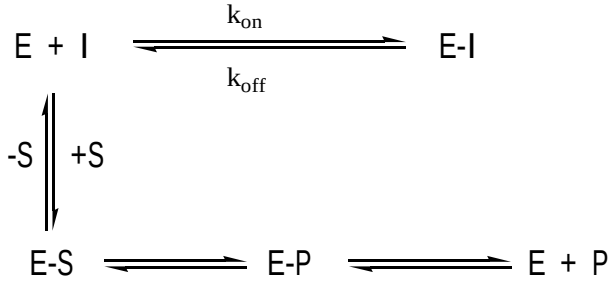


Reversible Enzyme Inhibition

The inhibitors form a dynamic equilibrium system with the enzyme. The inhibitory effects of reversible inhibitors are normally time dependent because the removal of unbound inhibitor from the vicinity of its site of action by natural processes will disturb this equilibrium to the left.

The most common enzyme inhibitor drugs are the reversible type, particularly ones that compete with the substrate for active site binding. These are known as *competitive reversible inhibitors*, typically compounds that have structures similar to those of the substrates or products of the target enzymes and which bind at the substrate binding sites, thereby blocking substrate binding.

As in the case of the interaction of a substrate with an enzyme, an inhibitor (I) also can form a complex with an enzyme (E). The equilibrium constant K_i (k_{off}/k_{on}) is a *dissociation* constant for breakdown of the E-I complex; therefore, as discussed for the K_d of drug–receptor complexes, the *smaller* the K_i value for an inhibitor I, the more potent the inhibitor.

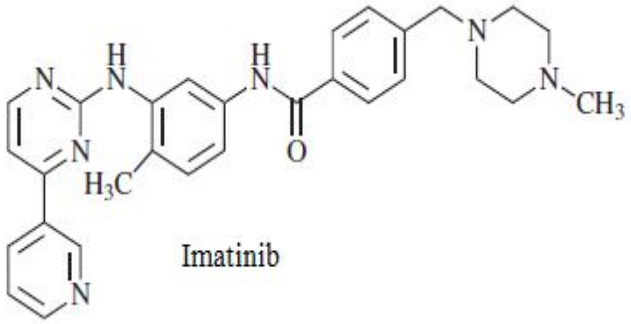


Interaction of the inhibitor with the enzyme can occur at a site other than the substrate-binding site (i.e., at an *allosteric binding site*) and still result in

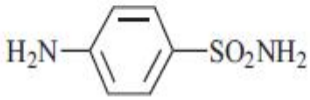
inhibition of substrate turnover. When this occurs, often as a result of an inhibitor-induced conformational change in the enzyme to give a form of the enzyme that does not bind the substrate properly, then the inhibitor is a noncompetitive reversible inhibitor.

Examples of Competitive Reversible Inhibitor Drugs

- Simple Competitive Inhibition: Epidermal growth factor receptor tyrosine kinase as a target for cancer
- Stabilization of an Inactive Conformation: Imatinib, an Antileukemia Drug
- Alternative Substrate Inhibition: Sulfonamide Antibacterial Agents (Sulfa Drugs)



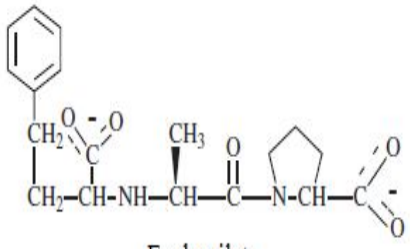
Imatinib



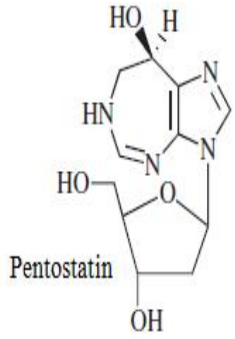
Sulfanilamide

Transition State Analogs and Multisubstrate Analogs: The enzyme achieves this rate enhancement by changing its conformation so that the strongest interactions occur between the substrate and enzyme active site *at the transition state* of the reaction.

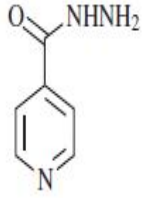
- Enalaprilat is a very potent slow, tightbinding inhibitor of angiotensin-converting enzyme (ACE)
- Pentostatin: an antineoplastic agent is a potent inhibitor of the enzyme adenosine deaminase (adenosine aminohydrolase).
- Isoniazid: The presently accepted mechanism of action of isoniazid proposes the oxidation by the heme-containing peroxidase enzyme KatG to form acyl radical.



Enalaprilat

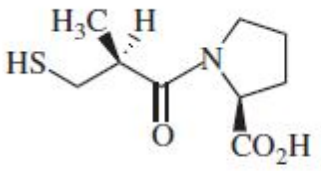


Pentostatin

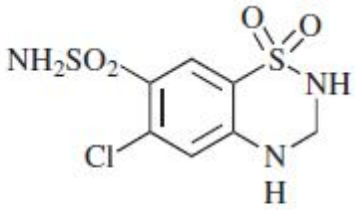


Isoniazid

Slow, Tight-Binding Inhibitors: The equilibrium between enzyme and inhibitor is reached slowly, and inhibition is time-dependent, reminiscent of the kinetics for irreversible inhibition. *Tight-binding inhibitors* are those inhibitors for which substantial inhibition occurs when the concentrations of inhibitor and enzyme are comparable. *Slow, tight-binding inhibitors* have both properties, can bind noncovalently or covalently. Example- Captopril, Hydrochlorothiazide

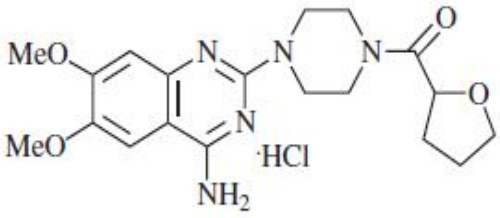


Captopril

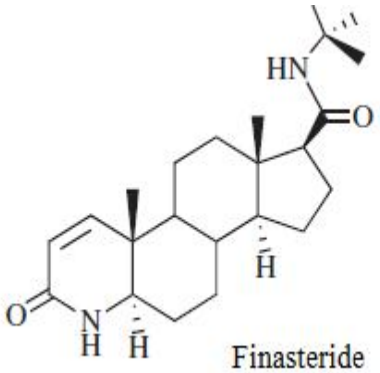


Hydrochlorothiazide

Dual-Acting Drugs: Dual-Acting Enzyme Inhibitors: When there are two related enzymes whose inhibition would give an enhanced effect compared to the sum of the effects of inhibiting either enzyme alone, it may be beneficial to design a single inhibitor of both enzymes, a compound known as a *dual-acting enzyme inhibitor*.



Terazosin HCl



Finasteride

Irreversible Inhibition

A competitive irreversible enzyme inhibitor, also known as an *active-site directed irreversible inhibitor* or an *enzyme inactivator*, is a compound whose structure is similar to that of the substrate or product of the target enzyme and which generally forms a covalent bond to an active site residue (a slow, tight-binding inhibitor, however, often is a noncovalent inhibitor that can be *functionally* irreversibly bound). Vigabatrin is a good example of this phenomenon.

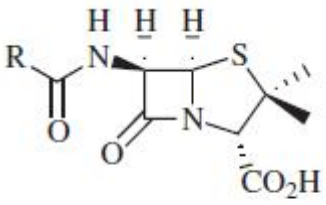
An *affinity labeling agent* is a reactive compound that has a structure similar to that of the substrate for a target enzyme. Subsequent to reversible E·I complex formation, it reacts with active site nucleophiles (usually amino acid side

chains), generally by acylation or alkylation (SN_2) mechanisms, thereby forming a stable covalent bond to the enzyme.

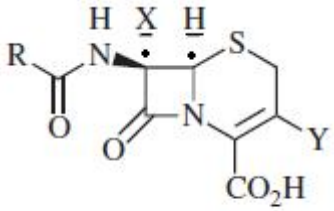
$$E + I \underset{}{\overset{K_I}{\rightleftharpoons}} E{\bullet}I \xrightarrow{k_{inact}} E - I$$

Selected Affinity Labeling Agents:

Penicillins and Cephalosporins/Cephamycins: The penicillins, cephalosporins, and cephamycins all have in common the β-lactam ring and are known collectively as β-*lactam antibiotics*. They inactivate an enzyme that is essential for bacterial growth, but which does not exist in animals, namely, the peptidoglycan transpeptidase.

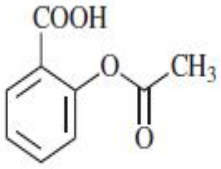


Penicillins

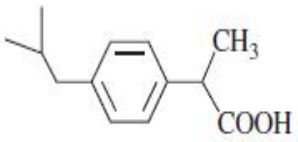


Cephalosporins

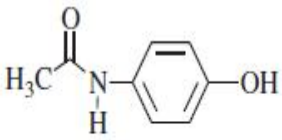
Aspirin and other NSAIDs inhibits prostaglandin-synthatase or cyclooxygenase (COX), which in inhibition of prostaglandin (PG) biosynthesis.



Aspirin



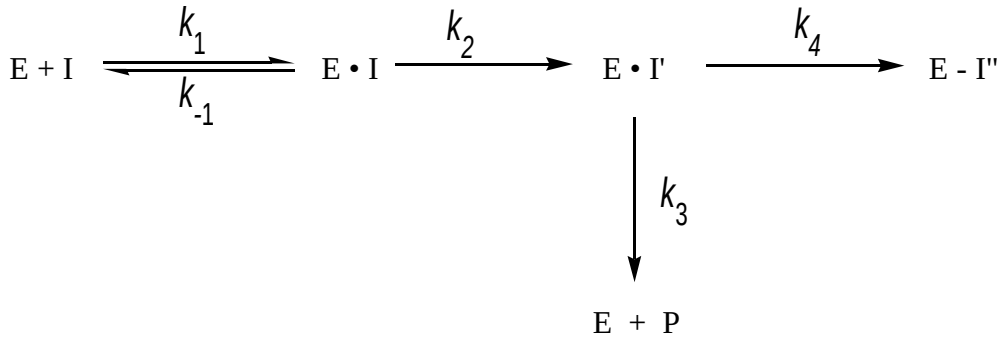
Ibuprofen



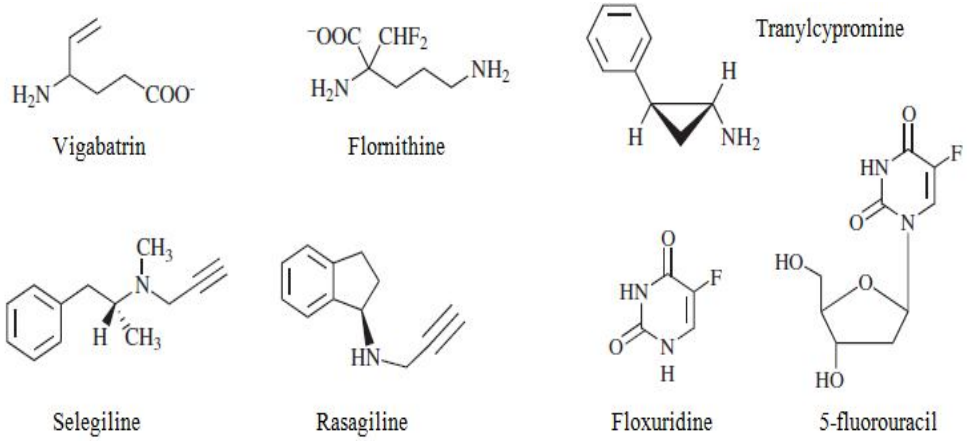
Acetaminophen

***Mechanism-Based Enzyme Inactivators*:**

A *mechanism-based enzyme inactivator* is an unreactive compound that bears a structural similarity to the substrate for a specific enzyme. Once the inactivator binds to the active site, the target enzyme, via its normal catalytic mechanism, converts the compound into a product that inactivates the enzyme prior to escape from the active site.



Examples of mechanism-based enzyme inactivators are vigabatrin (anticonvulsant), eflornithine (antiprotozoal), tranylcypromine (antidepressant), selegiline & rasagiline (antiparkinson), floxuridine & 5-fluorouracil (antitumor), etc.



8.7 Sulfonamides

In the early 1930s, Gerhard Domagk, head of bacteriological and pathological research at the Bayer Company in Germany, who was trying to find agents against streptococcal infections, tested a variety of azo dyes. One of the dyes, Prontosil, showed dramatically positive results, and successfully protected mice against streptococcal infections. However, Bayer was unwilling to move rapidly on getting Prontosil onto the drug market. As Albert tells it, when, in late 1935, Domagk's daughter cut her hand and was about to die of a streptococcal infection, her father gave her Prontosil! Although she turned bright red from the dye, her recovery was rapid, and the effectiveness of the drug became quite credible. In 1939, Domagk was awarded the Nobel Prize in Medicine for this achievement.

Prontosil is a prodrug, a compound that requires metabolic activation to be effective. Furthermore, they demonstrated that sulfanilamide was as effective as Prontosil.

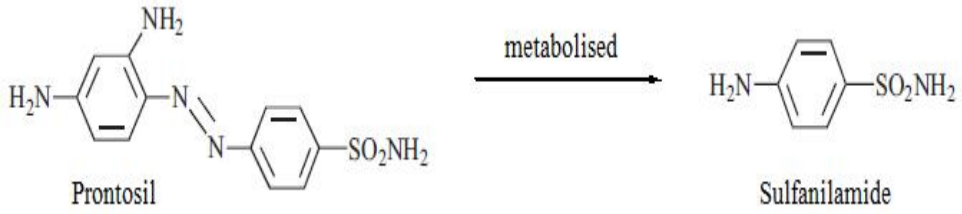


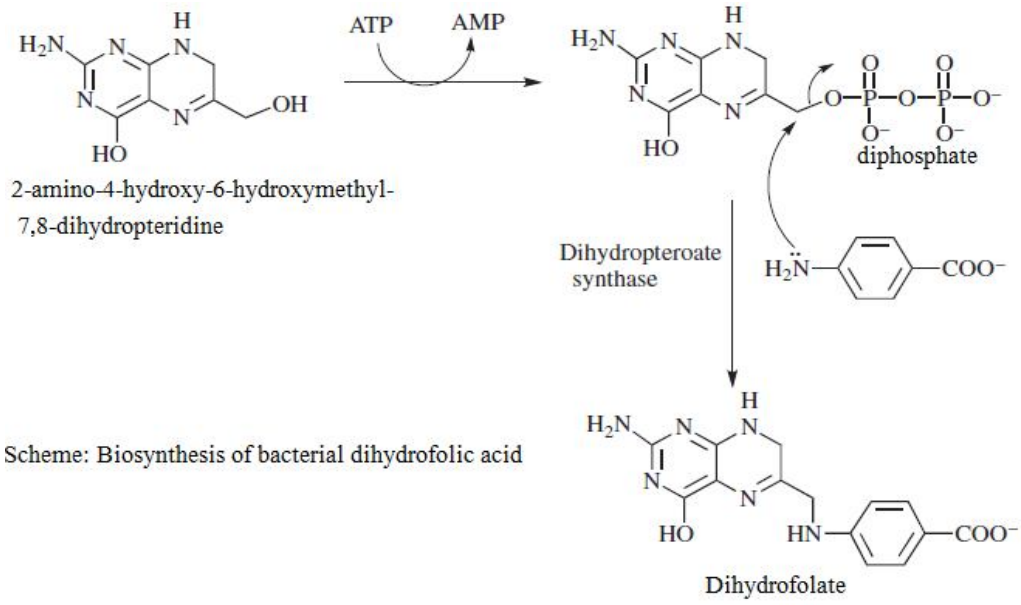
Fig. 8.2: Prodrug concept for sulfonamides

Mechanism of Action

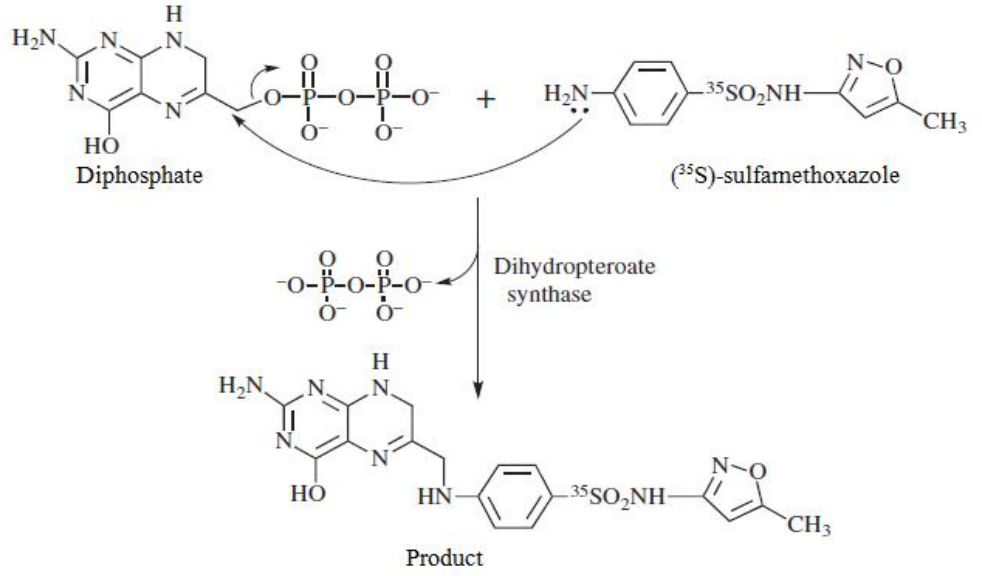
Many bacteria synthesize their own folic acid (FA) of which para aminobenzoic acid (PABA) is a constituent, and is taken up from the medium. Woods and Fildes (1940) proposed the hypothesis regarding sulfonamide action. Sulfonarnides, being structural analogues of PABA, inhibit bacterial folate synthase -FA is not formed and a number of essential metabolic reactions suffer. Sulfonamides competitively inhibit the union of PABA with pteridine residue to form dihydropteroic acid which conjugates with glutamic acid to produce dihydrofolic acid. Also, being chemically similar to PABA, the sulfonamide may itself get incorporated to form an altered folate which is metabolically injurious. Human cells also require FA, but they utilize preformed FA supplied in diet and are unaffected by sulfonamides. Evidences in favour of this mechanism of action of sulfonamides are:

(a) PABA, in small quantities, antagonizes the antibacterial action of sulfonamides.

(b) Only those microbes which synthesize their own FA and cannot take it from the medium are susceptible to sulfonamides. Pus and tissue extracts contain purines and thymidine which decrease bacterial requirement for FA and antagonize sulfonamide action. Pus is also rich in PABA.



Scheme: Biosynthesis of bacterial dihydrofolic acid



Scheme: Dihydropteroate synthase use of sulfamethoxazole in place of *para*-aminobenzoic acid

Fig. 8.3: Mechanism of action of sulfonamides

Resistance to sulfonamides: Most bacteria are capable of developing resistance to sulfonamides. Prominent among these are gonococci, pneumococci, Staph. aureus, meningococci, E. coli, Shigella and some Strep. pyogenes, Strep. viridans and anaerobes. The resistant mutants either:

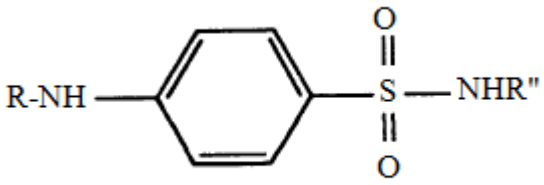
- Produce increased amounts of PABA, or

- Their folate synthase enzyme has low affinity for sulfonarnides, or
- Adopt an alternative pathway in folate metabolism.

Resistance developed in vivo is quite persistent. Sensitivity patterns have changed depending on the extent of use. When an organism is resistant to one sulfonamide, it is resistant to them all.

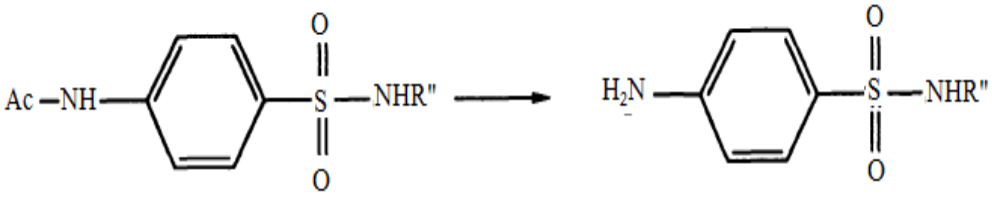
Structure-activity relationships (SAR):

The synthesis of a large number of sulfonamide analogues led to the following conclusions.



Sulfonamide Analogues

- The p-amino group is essential for activity and must be unsubstituted (i.e. R = H). The only exception is when R = acyl (i.e. amides). The amides themselves are inactive but can be metabolized in the body to regenerate the active compound. Thus amides can be used as sulfonamide prodrugs.
- The aromatic ring and the sulfonamide functional group are both required.
- The aromatic ring must be para-substituted only.
- The sulfonamide nitrogen must be secondary.
- R" is the only possible site that can be varied in sulfonamides.



Scheme: Metabolism of acyl group to regenerate active compound

- Changing the nature of the group R" has also helped to reduce the toxicity of some sulfonamides. The primary amino group of sulfonamides is acetylated in the body and the resulting amides have reduced solubility which can lead to toxic effects. For example, the metabolite formed from sulfathiazole (an early sulfonamide) is poorly soluble and can prove fatal if it blocks the kidney tubuls.

- Sulfadiazine was also found to be more active than sulfathiazole and soon replaced it in therapy.

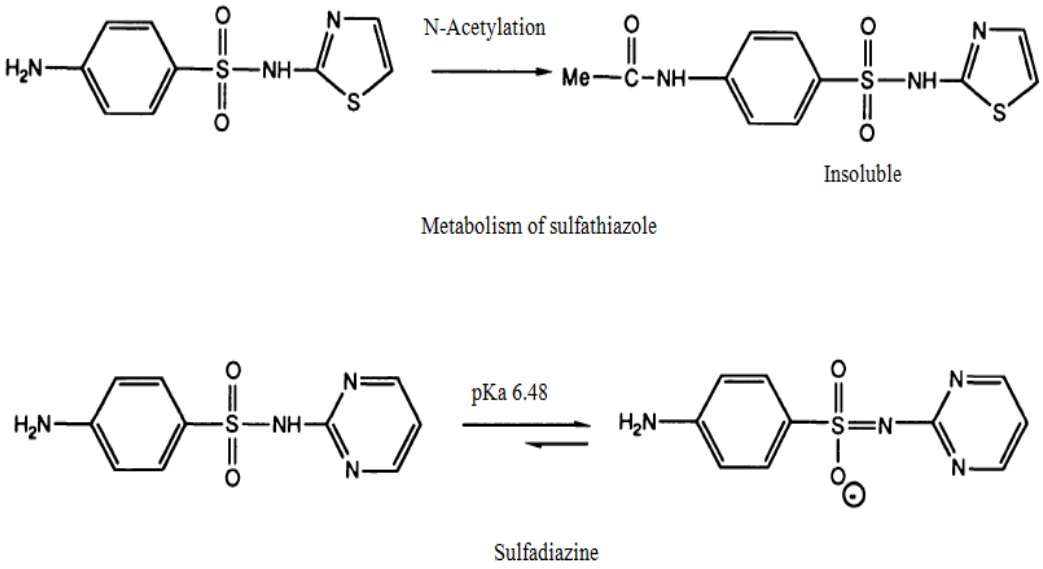


Fig. 8.4: SAR of Sulphonamides

Classification

Sulphonamides i.e., the systemic antibacterial drugs may be classified broadly on the basis of their site of action as described in the sections that follows:

1. ***Sulphonamides for General Infections*:** These sulphonamides are invariably employed against the streptococcal, meningococcal, gonococcal, staphylococcal and pneumococcal infections. Examples: sulfanilamide, sulfapyridine, sulfathiazole, sulfadiazine, sulfamerazine, sulfadimidine, sufalene, sulfamethizole etc.
2. ***Sulphonamides for Urinary Infections:*** A number of sulphonamides have been used extensively for the prevention and cure of urinarytract infections over the past few decades. They are used sometiems as a prophylactic before and after manipulations on the urinary tract. A few such sulphanilamide analogues belonging to this category shall be dealt with here. Examples: sulfacetamide, sulfafurazole, sulfisoxazole acetyl, sulfacitine, etc.
3. ***Sulphonamides for Intestinal Infections:*** A plethora of insoluble sulphonamide analogues, for instance phthalylsulfathiazole and succinylsulfathiazole, are not readily absorbed from the gastrointestinal tract. However, the release of active sulphonamide in high concentration, obtained due to hydrolysis in large intestine, enables their application for intestinal infections and also for pre-operative preparation of the bowel for surgery.

Examples: sulfaguanidine, phthalylsulfathiazole, succinylsulfathiazole, phthalylsulfacetamide, salazosulfapyridine, etc.

4. ***Sulphonamides for Local Infection:*** There are some sulphonamides which are used exclusively for certain local applications. Examples: Sulfacetamide sodium, Mafenide, etc.
5. ***Sulphonamide Related Compounds:*** There are some sulphonamides which essentially differ from the basic sulphonamide nucleus, but do possess anti-bacterial properties. Examples: Nitrosulfathiazole, dapsone, silver sulfadiazine, etc.

Uses of Sulfonamides:

Systemic use of sulfonamides alone (not combined with trimethoprim or pyrimethamine) is rare now though they can be employed for suppressive therapy of chronic urinary tract infection, for streptococcal pharyngitis and gum infection; such uses are outmoded. Combined with trimethoprim (as cotrimoxazole) sulfamethoxazole is used for many bacterial infections, P. jiroveci and nocardiasis. Along with pyrimethamine, certain sulfonamides are used for malaria and toxoplasmosis. Ocular sulfacetamide sod. (10-30%) is a cheap alternative in trachoma/ inclusion conjunctivitis, though additional systemic azithromycin or tetracycline therapy is required for eradication of the disease. Topical silver sulfadiazine or mafenide are used for preventing infection on burn surfaces.

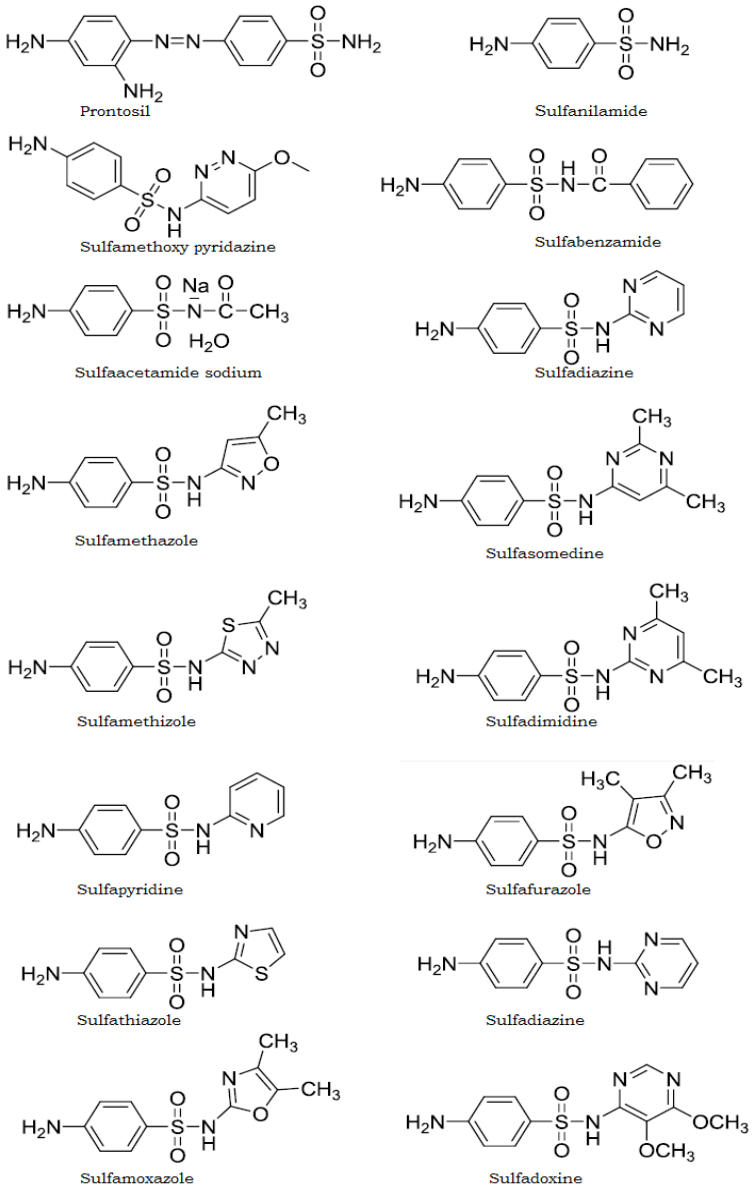
ADME of Sulfonamides:

***Absorption*:** Normally given orally or applied topically; some soluble salts given parenterally, readily absorbed from GIT, achieving peak blood levels in 30 min, except for those remain in intestine.

***Distribution*:** Highly plasma protein bound; will displace other bound drugs and bilirubin. Sulfonamides given in late term can induce neonatal jaundice distributed in total body water, readily enter CNS, synovial and ocular fluid, fetal circulation and milk.

***Metabolism*:** Primarily by acetylation at free amino group; some oxygenation of aromatic ring and/or side chain. Acetylated metabolites inactive

***Elimination*:** Majority eliminated unchanged concentrated in urine; useful in urinary tract infections; older sulfonamides actually formed crystals in tubules and ureter.



8.8 Summary

Pharmacodynamics deals with the relationship between drug concentrations at the effect site or concentrations (usually unbound concentrations) in plasma in equilibrium with effect site concentrations, and the magnitude of the observed pharmacological effect of the drug. Almost all biological reactions are carried out under catalytic influence of enzymes; hence, enzymes are a very important target of drug action. Since the development of the sulfa drugs (sulfonamides), enzyme inhibitors have played a vital role in controlling these infectious agents. Sulfonamides, also known as sulfa drugs, have a history that dates back to almost 70-80 years. A sulfonyl group plays a very important role as a key constituent of number of biologically active molecules.

8.9 Glossary

- *Pharmacodynamics* is the study of drug effects.
- *Enzymes* are natural proteins that catalyze biochemical reactions, works as receptor.
- *Coenzyme* is an organic molecule or a metal ion that binds to the active site, in some cases covalently or non-covalently.
- Any compound that slows down or blocks enzyme catalysis is an *enzyme inhibitor.*
- *Sulfonamide* (Sulfa drugs) grouping is derived from a sulfonic acid group by replacing its hydroxyl group with an amino group.

8.10 Review questions / Comprehensive Questions

1. Define pharmacodynamics. What is the role of pharmacodynamics in drug design?
2. What are enzymes? Discuss the mode of enzyme action.
3. What do you understand about elementary treatment of enzyme stimulation?
4. Describe different types of enzyme inhibition with suitable examples.
5. What is the role of enzyme inhibition in drug discovery & design?
6. Define sulfonamides. Classify them with examples.
7. Describe mechanism of action of sulfonamides with suitable scheme/reaction.
8. Discuss the SAR and uses of sulfonamides.

8.11 References and Suggested readings

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Unit-9

Pharmacodynamics – II

Structure of Unit:

9.1 Objectives

In this unit the students will be able to understand

- Meaning of xenobiotics
- Membrane disrupters
- Concept of biotransformation
- Metabolizing enzymes
- Types of biotransformation reactions
- Role of biotransformation in new drug development

9.2 Introduction: Pharmacodynamics

Pharmacodynamics-

It is the study of drug effect on the body. This includes physiological and biochemical effects of drugs and their mechanism of action at organ system. Drugs (except those gene based) do not impart new functions to any system,

organ or cell, they only alter the pace of ongoing activity. The basic types of drug action can be categorized as:-

Stimulation- It refers to the drugs which stimulate activity of specialized organs.

Depression- It means drugs which suppress activity of specialized organs.

Irritation- This is a noxious effect and particularly applied to less specialized cells.

Replacement- It means use of natural metabolites or hormones in deficiency states.

Cytotoxic Action- It refers to selective cytotoxic action for parasites or cancer cell, without affecting the host cells.

Mechanism of drug action

Majority of drugs act by interacting with a discrete target. The target sites at which drugs act may vary with different types of drugs.

Common sites for drug action are-

- Cell Membranes and Walls
- Enzymes
- Receptors
- Nucleic Acids

9.3 Membrane active drugs

Both types of cells either eukaryotic or prokaryotic consists a membrane called cytoplasmic membrane or plasma membrane. It acts as a barrier and separates the internal contents (Intracellular fluid) of cell from external medium (extracellular fluid).

The cell wall, a rigid external covering protect the fragile membranes of microorganism. The cell wall and plasma member are together known as the cell envelope.

Transport of drug ions and biomolecules across the membranes occur by –

- Passive diffusion and filtration
- Specialized transport

Many drugs exert their pharmacological effect by affecting the membrane transport processes.

Example – (A) Local anesthetics

(B) Cardiotonic drugs (Digitals Glycosides)

Local anesthetics are the drugs which reversibly block impulse conduction upon topically or local application and cause loss of sensory perception, especially of pain in a restricted area of the body. Local anesthetics are sodium channel blockers. They act by blocking the voltage sensitive sodium channel and reducing the influx of ions, thereby prevent conduction of action potential. Examples- Procaine Lidocaine

Digitalis selectively binds to extracellular face of the membrane associated Na^+ K^+ ATPase of myocardial fibers and inhibits this enzyme. Inhibition of this cation pump results in progressive accumulation of Na^+ intracellularly. This indirectly results in intracellular Ca^{+2} accumulations. Thus digitalis increases force of cardiac contraction.

Many drugs act either disrupting the structure of membranes and walls or inhibit the synthesis of cell membrane or blocking ion channels. These drugs produce their action by one of the following way-

- Inhibit enzymes which are assisting in production of compounds necessary for maintaining the integrity of membrane.
- Inhibit processes of cell wall formation; so that incomplete cell wall formed through which loss of vital cellular material and finally death of cell occur.
- Make cell membrane porous by forming channels through it which leads loss of vital cellular material and subsequent death of cell.

Membrane disrupters are –

(I) Antifungal agents

- Azoles
- Allylamines
- Phenols

(II) Antimicrobials-

- Ionophoric Antibiotics
- Cell wall Synthesis Inhibitors

Antifungal agents

Fungal infections usually involve the skin and mucous membranes of the body. The fungal microorganisms are believed to damage the cell membrane, leading

to a loss of essential cellular components. Antifungal agents counter this attack by both fungi static and fungicidal action.

Azoles

Imidazoles- Clotrimazole Econazole Miconazole

Triazoles- Fluconazole Itraconazole Voriconazole

These agents inhibit the fungal cytochrome P450 enzyme 'lanosterol l4-demethylase' and thus impair the biosynthesis of ergosterol for the cytoplasmic membrane leading to a cascade of membrane abnormalities in the fungus. Azoles exhibit fungi static activity at low concentration and fungicidal activity at higher concentrations.

Allylamines

They act by inhibiting squalene epoxidase non-competitively, the enzyme for the squalene epoxidation stage in the biosynthesis of ergosterol in the fungi. This leads to an increase in squalene concentration in the membrane with subsequent loss of membrane integrity, which allows loss of cell contents to occur. Examples- Terbinafine

Phenols

There are numerous phenolic antifungal agents. The mechanism by which the membrane destruction occurs is not well known. They are believed to acts by disrupting bacterial membranes and denaturing bacterial proteins and the death of the cell.

Antimicrobials-

Antibacterial antibiotics normally act by either making the plasma membrane of bacteria more permeable to essential ions and other small molecules by Ionophoric action or by inhibiting cell wall synthesis. Those compounds that act on the plasma membrane also have the ability to penetrate the cell wall structure. In both cases, the net result is a loss in the integrity of the bacterial cell envelope, which leads to irreversible cell damage and death. Antimicrobials agents' are-

- Ionophoric Antibiotics
- Cell wall Synthesis Inhibitors

Ionophoric Antibiotics

Ionophores are substances that can penetrate a membrane and increase its permeability to ions. They transport ions in both directions across a membrane. Consequently, they will only reduce the concentration of a specific ion until its

concentration is the same on both sides of a membrane. This reduction in the concentration of essential cell components of a microorganism is often sufficient to lead to the destruction of the organism.

Ionophores are classified as-

1. Channel Ionophores
2. Carrier Ionophores

Channel Ionophores form channels across the membrane through which ions can diffuse down a concentration gradient. Example- gramicidin

Carrier Ionophores pick up an ion on one side of the membrane, transport it across, and release it into the fluid on the other side of the membrane. Example- Valinomycin

Cell wall synthesis inhibitors

All β-lactam antibiotics interfere with the synthesis of bacterial cell wall. The bacteria synthesize UDP-N-acetylmuramic acid pentapeptide and UDP-N-acetyl glucosamine. The peptidoglycan residues are linked together forming long strands and UDP is split off. The final step is cleavage of the terminal D-alanine of the peptide chains by transpeptidases; the energy so released is utilized for establishment of cross linkages between pepide chains of the neighbouring strands. This cross linking provides stability and rigidity to the cell wall. The β-lactam antibiotics inhibit the transpeptidases so that cross linking does not take place.

Example-Penicillins Cephalosporins Cycloserine, Vancomycin Bacitracin.

Many bacteria are resistant to antibiotics especially to b-lactam antibiotics. Bacteria that have developed a resistance to β-lactam antibiotics are treated using either incorporating a β-lactamase inhibitor, such as clavulanic acid or sulbactam, or a lactamase resistant drug, such as vancomycin.

9.4 Xenobiotics

Xenobiotics (substances foreign to the body)

Humans are exposed daily to a wide verity of foreign compounds through exposure to environmental contaminants as well as in diets. All chemical substances that are not nutrients for body called xenobiotics or exogenous compounds. Drugs are also considered xenobiotics and are extensively metabolized in humans.

Xenobiotics are Categories as followings -

- Drugs
- Food constituents
- Food additives
- Chemicals of abuse (ethanol, coffee, tobacco etc)
- Agrochemicals (fertilizers, insecticides, herbicide etc)
- Industrial chemicals (solvents, dyes, monomers, polymers)
- Pollutants

In general these are lipophilic chemicals that in the absence of metabolism would not be efficiently eliminated, and thus would accumulate in the body, resulting in toxicity. The ability of humans to metabolize and clear drug is a natural process that involves the same enzymatic pathways and transport systems that are utilized for normal metabolism of dietary constituents. With few exceptions, all xenobiotics are subjected to one or multiple pathways that constitute the phase I and Phase II metabolic systems, which modify xenobiotics enzymatically in body. The chemical modification of xenobiotics is called biotransformation or metabolism.

9.5 Biotransformation (Metabolism)

The terms biotransformation and metabolism are often used synonymously; particularly when applied to drugs. Biotransformation or Metabolism of a drug is complex and important process whereby living organism's effect chemical change to a molecule. The product of such a chemical change is called a metabolite.

The general principle of drug metabolism is to change the non polar (lipid soluble) compounds in to polar (hydrophilic), water soluble derivatives via several chemical reactions, and promoting their excretion in to urine by kidneys. The term detoxification means inactivation of drug molecule and excretion of its less toxic metabolites. The term is discarded as the chemical reactions in the body can same times yield metabolites of greater toxicity or activity than original molecule. Metabolism is a wider term which refers for the chemical alteration that occur within the body and its processes alters drugs in two ways-

1-Conversion of lipophilic compounds in to hydrophilic-

Metabolic reactions convert a drug molecule progressively more water soluble and so favors its elimination in the urine.

2-Alteration in Biological Activity-

Inactivation –

Inactivation means conversion of pharmacologically active substance to an inactive substance or less active.

Examples – Paracetamol Lidocaine

Active metabolites–

Metabolic reactions convert pharmacologically active drug to one or more active metabolites.

Examples – Codeine Morphine Diazeparn

Activation of Inactive drug –

Metabolic reactions convert inactive to an active substance i.e. Prodrug.

Examples – Dopamine Aspirin

9.6 Sites and enzymes of metabolism

Sites of biotransformation –

Liver is the most important site, for metabolism of almost all drugs due to presence of large variety of enzymes in it. Other site involved in metabolism is kidney, plasma, intestine, skin, lungs, and spleen.

Enzymes of metabolism –

The drug metabolizing enzymes are following-

- Microsomal Enzymes
- Non-microsomal Enzymes

Microsomal Enzymes – These are present in endoplasmic reticulum, liver, intestinal mucosa, kidney and lungs. These enzymes catalyze a majority metabolic reaction such as oxidation, reduction, hydrolysis, and glucuronidation.

Example – Monooxygenases Cytochorome P 450

Non-microsomal Enzymes –

These are present in the cytoplasm and mitochondria of hepatic cells and also in other tissues. These enzymes catalyse few oxidations, reduction a number of hydrolytic reactions and conjugation reactions other than glucuronidation.

Examples – Esterases Amidases

9.7 Biotransformation Reactions

The metabolism of xenobiotics involves two sequential steps-

1. Phase I reactions

2. Phase II reactions

Phase I reactions

These reactions usually convert the parent drug to a more polar metabolite by introducing or unmasking a functional group (-OH, -NH2, -SH). The polar group created serves then as an anchor point for the second metabolic step.

Phase-II reactions

These reactions consists of conjugation of reactive groups, present either in the parent molecule or after Phase-I transformation. If phase I metabolites are sufficiently polar, they may be readily excreted. However, many phase I products are not eliminated rapidly and undergo a subsequent reaction in which an endogenous substrate such as glucuronic acid, sulfuric acid, acetic acid, or an amino acid combines with the newly incorporated functional group to form a highly polar conjugate.

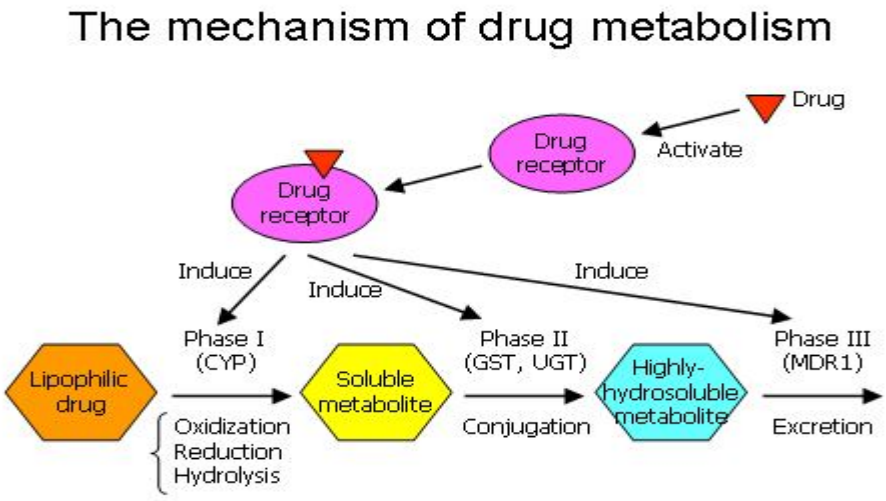


Fig.9.1: Mechanism of drug action

PHASE-I REACTIONS (NONSYNTHETIC REACTIONS)

(I) OXIDATION REACTIONS

Oxidation is the most common metabolic reaction which involves addition of O_2 or removal of hydrogen from the drug.

Microsomal oxidation:

The smooth endoplasmic reticulum of cells in many organs, specially the liver, contains membrane-associated enzymes which are responsible for dug oxidation. The primary components of this enzyme system are cytochrome P-450 reductase and cytochrome 450. This enzyme system has been termed a mixed function oxygenase.

Examples- Phenytoin Pentobarbital

Non-microsomal oxidation:

Soluble enzymes in mitochondria of cells can metabolize few compounds.

Examples

- Xanthine oxidase converts hypxanthine to xanthine and xanthine to uric acid.
- Tyrosine hydroxylase hydroxylates tyrosine to dopa.

Various oxidation reactions are:

1-Hydroxylation

$RCH_2CH_3 \rightarrow RCH_2CH_2OH$

Aromatic hydroxylation

Examples- Phenobarbital Phenylbutazone Amphetamine

Aliphatic hydroxylation

Examples- Chlorpropamide Ibuprofen Digitoxin

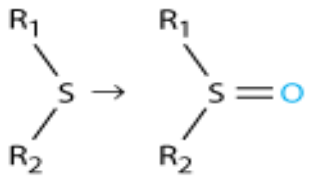
2-Oxygenation at N or S atoms

- **Oxygenation at N atom**

  $RNH_2 \longrightarrow RNHOH$

  Examples-Aniline

- **Oxygenation at S atom**



Examples- chlorpromazine

3-Oxidative dealkylation

- **N- Dealkylation**

$RNHCH_3 \rightarrow RNH_2 + CH_2O$ Examples- Morphine

- **O-Dealkylation**

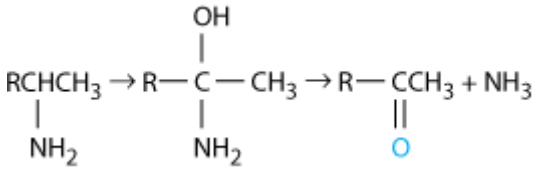
$ROCH_3 \rightarrow ROH + CH_2O$ Examples- Codeine

- **S-Dealkylation**

$RSCH_3 \rightarrow RSH + CH_2O$

Examples- 6-Methylthiopurine

4-Oxidative deamination



Examples- Amphetamine

(II) REDUCTION REACTIONS

This reaction is the converse of oxidation and occurs in both the microsomal and nonmicrosomal metabolizing system. It is less common than oxidation.

Examples:

Microsomal reduction- Chloramphenicol

Nonmicrosomal reduction- Chloral hydrate

1-Aldehydes Reduction

$RCHO \longrightarrow RCH_2OH$

Examples- Chloral hydrate

2-Azo Reduction

$R_1N = NR_2 \longrightarrow R_1NH_2 + R_2NH_2$

Examples- Azo gantrisin

3-Nitro Reduction

$O_2NR \longrightarrow H_2NR$

Examples- Chloramphenicol

(III) HYDROLYSIS REACTIONS

This is cleavage of drug molecule by taking up a molecule of water.

Hydrolysis

Nonmicrosomal hydrolases:

- Procainamide (Amidases)
- Proinsulin (Peptidases)

Microsomal hydrolases:

- Cyclization
- Decyclization

1-**Ester Hydrolysis**

$R_1COOR_2 \longrightarrow R_1COOH + HOR_2$

Examples- Procaine aspirin

2-Amide Hydrolysis

$R_1CONHR_2 \rightarrow R_1COOH + H_2NR_2$

Examples- lidocaine Indomethacin

3-Cyclization

This is formation of ring structure from a straight chain compound.

Example- Proguanil

4-Decyclization

This is opening up of ring structure of the cyclic drug molecule,. This is generally a minor pathway.

Examples- Phenytoin

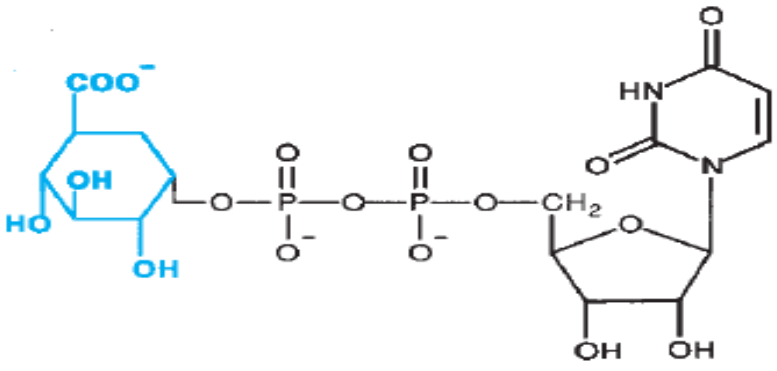
PHASE II REACTIONS (SYNTHETIC REACTIONS / CONJUGATE REACTIONS)

In these reactions a drug or its metabolite is couple (conjugated) enzymatically with an endogenous substance usually a carbohydrate or an amino acid or a derivative of these, resulting almost invariably in inactivation of the parent drug. Nearly all conjugates are inert and water soluble and pass in urine or bile.

These involve conjugation of the drug or its phase I metabolite with an endogenous substrate to form a polar highly ionized organic acid, which is easily excreted in urine or bile

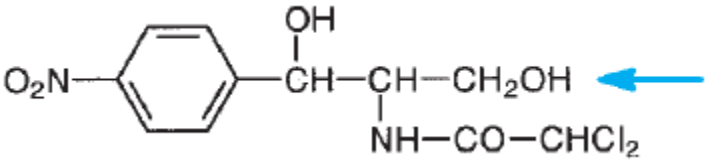
1-GLUCURONIDE CONJUGATION

Glucuronidation is a major pathway of xenobiotic biotransformation. Glucuronidation requires the cofactor uridine diphosphate-glucuronic acid (UDP glucuronic acid), and the reaction is catalyzed by UDP glucuronosyltransferases (UGTs). The site of glucuronidation is generally an electron-rich nucleophilic heteroatom (O, N, or S). Therefore, substrates for glucuronidation contain such functional groups as aliphatic alcohols and phenols (which form O-glucuronide ethers), carboxylic acids (which form O-glucuronide esters), primary and secondary aromatic and aliphatic amines (which form N-glucuronides), and free β -glucuronidase.



Cofactor- Uridine-5'- diphospho-D-glucronic acid (UDP-GA)

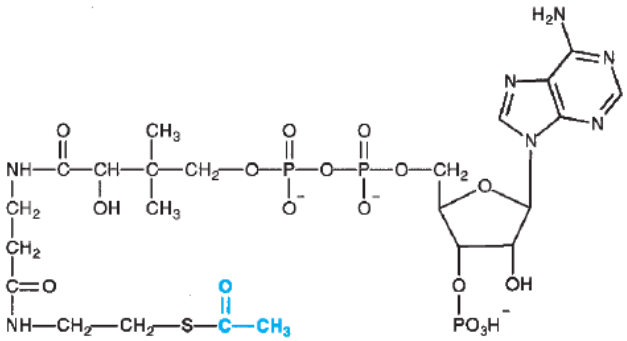
Example- Chloramphenicol



The arrow indicates the site of glucuronidation.

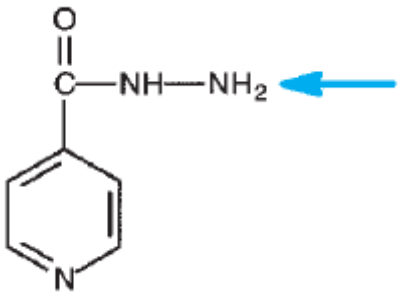
2-ACETYLATION

Compounds having amino or hydrazine residues are conjugated with the help of acetyl coenzyme-A.



Cofactors- Acetyl coenzyme-A

Example-Isoniazid

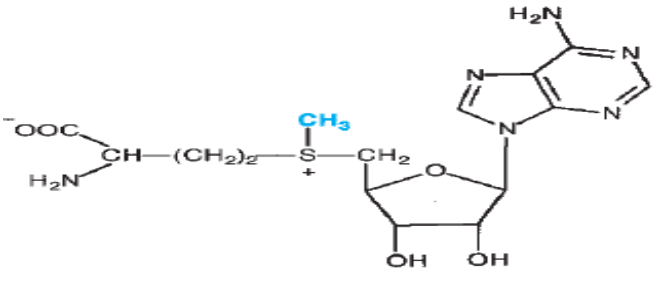


The arrow indicates the site of Acetylation.

3-METHYLATION

Methylation differs from most other phase II reactions because it generally decreases the water solubility of xenobiotics and masks functional groups that might otherwise be conjugated by other phase II enzymes.

The amines and phenols can be methylated; methionine and cysteine acting as methyl donors. Example – Adrenaline Histamine Nicotinic Acid Methyldopa

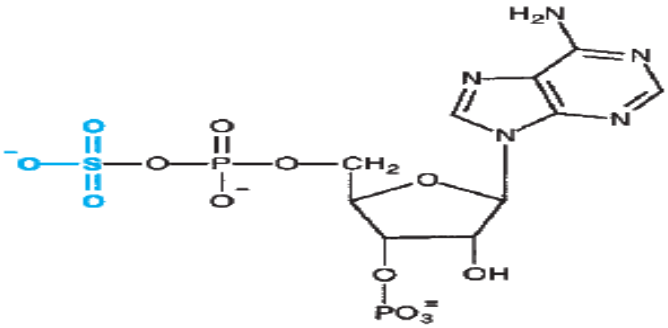


Cofactors- S-Adenosylmethionine (SAM)

Histamine *N*-Methylhistamine

4-SULFATE CONJUGATION

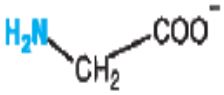
Sulfate conjugation generally produces a highly water soluble sulfuric acid ester. The reaction is catalyzed by sulfotransferases (SULTs) enzymes. The phenolic compounds and steroids are sulfated by sulfate conjugation. Sulfate conjugation involves the transfer of sulfonate not sulfate (i.e. SO3- not SO4-) from PAPS to the xenobiotic.



Cofactors- 3-phosphoadenosine- 5-phosphosulfate (PAPS)

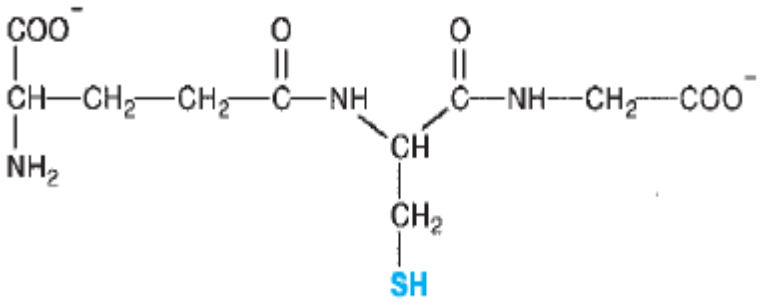
5- GLYCINE CONJUGATION

In glycine conjugation salicylates and other drugs having carboxylic acid group are conjugated with glycine.



6- GLUTATHIONE CONJUGATION

Glutathione conjugation is carried out by glutathione s-transferase forming a mercapturate is normally a minor pathway. However, it serves to inactivate highly reactive quinone or epoxide intermediates formed during metabolism of certain drugs, e.g. paracetamol. When large amount of such intermediates are formed (in poisoning or after enzyme induction), glutathione supply falls short- toxic adducts are formed with tissue constituents - tissue damage.



Glutathione

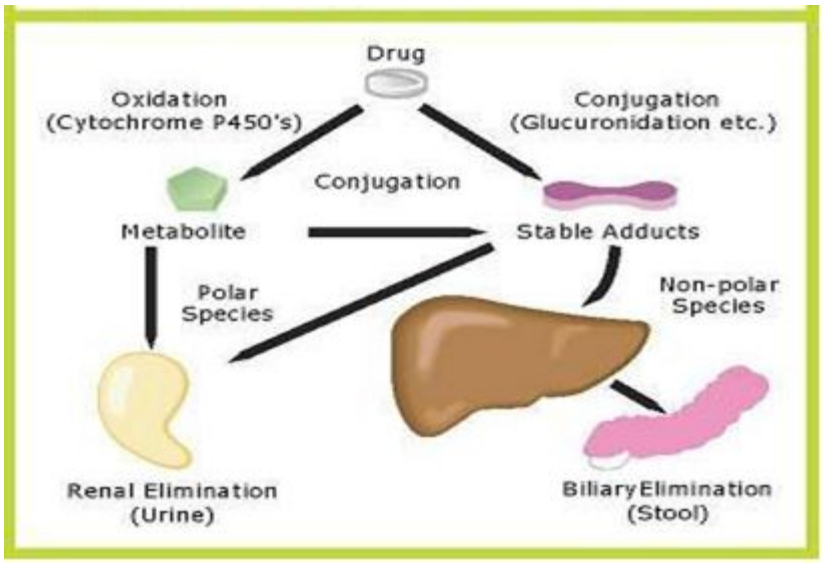


Fig. 9.2: Drug metabolism pathways

9.8 Significance of Drug Metabolism

The metabolic changed-drugs have been of considerable interest and of great practical value in the search for new and improved medicines.

Significance of drug development related to metabolism are-

- The Azo dye, prontosil which is inactive in vitro, is converted in the body to the active sulphanilamide by metabolic reduction process.
- The metabolic acetylation of the sulphonamides served in the development of compunds which are acetylated to a lesser extent and acetylated derivatives are more soluble, hence reduce kidney damage to crystallization in the renal tubules.

- Analgesic properties of phenacetin depends on its conversion by O-dealkylation to produce an active metabolite, acetaminophen i.e., p-hydroxyacetanilide.
- The antidepressant properties of imipramine and amitriptyline, both tertiary amines are to be mediated by their secondary amine metabolites, called desipramine and notriptyline.
- Cholroguanide shows its antimalarial activity when it is converted into 1-(chlotpheny)-2,4-diamino-6-dimethy1-dihydro-1,3,5 triazine by the human body.
- Arsine-oxide is a therapeutically useful compound resulted from arseno compound-As =As-, when it undergoes oxidation reaction, Arsine-oxide is although more toxic but a superior therapeutic compound developed by drug metabolism process.
- The introduction of mandelic acid as a genito-urinary antiseptic drug showed the observation that it gets excreted unchanged and in the acidic pH of urine, it has significant bactericidal properties. Thus the metabolism of a drug may have a profound effect on its pharmacological activity. It plays an important and rapidly exploring role in the development of new drugs.

9.9 Summary

Majority of drugs exert their pharmacological effect either by affecting the membrane transport processes or disrupting the structure of membranes and walls or inhibit the synthesis of cell membrane or blocking ion channels. During the process of biotransformation, the molecular structure of a drug is changed from one that is absorbed to one that can be readily eliminated from the body. The liver has enzymes that facilitate chemical reactions such as oxidation, reduction, and hydrolysis of drugs (phase I). It has other enzymes that attach substances to the drug, producing reactions called conjugations (phase II).

The knowledge of the metabolic fate of a drug is highly important, since the metabolism can generate toxic species, activate the drug or lead to the loss of pharmacological activity. The liver is the principal, but not the only, site of drug biotransformation. The biotransformation reactions are very important in the development for new medicines.

9.10 Glossary

- **Antibiotics-** These are substances produced by microorganisms, which selectively suppress the growth of or kill other microorganisms at very low concentrations.
- **Antifungal-** Agents counter fungal infection attack by both fungi static and fungicidal action.
- **Biotransformation-** It refers to chemical alteration of drug molecule.
- **Conjugate Reactions**- Drug or its metabolite is couple enzymatically with an endogenous substance
- **Cytochrome P 450**- Heme-containing protein**,** P-it is pink**,** 450- absorption spectrum has a maximum at 450 nm.
- **Cytoplasmic membrane-** It is a barrier between internal and external medium of cell.
- **Detoxification-** It means inactivation of drug molecule.
- **Drugs-** Chemicals alter the pace of ongoing activity of body in diseases state.
- **Local Anesthetics-** Local anesthetics are the drugs which reversibly block impulse conduction in a restricted area of the body.
- **Xenobiotics-** All chemical substances those are foreign to the body.

9.11 Review questions / Comprehensive Questions

1. What is biotransformation?
2. Explain biotransformation reactions.
3. Define xenobiotics and discuss about metabolizing enzymes.
4. Discuss role of metabolism in new drug development process.
5. Discuss the mechanism of action of membrane active agents.
6. Write note on-
    A. Antifungal agents
    B. Cell wall Synthesis Inhibitors
7. Explain the effect of biotransformation on the biological activity.

9.12 References And Suggested Readings

- Textbook of Organic Medicinal and Pharmaceutical Chemistry (11th ed.)- Wilson and Gisvold's.
- Basic & Clinical Pharmacology (9th ed.)- Bertram G. Katzung (Mc Graw Hill Publisher) 2004.
- Essential of Medical Pharmacology (7thed.)- KD Tripathi (JAYPEE Publisher) 2013.

Unit-10

Antineoplastic agents

Structure of Unit:

10.1 Objectives

In this chapter we deal with cancer and anticancer therapy, emphasising first the pathogenesis of cancer before proceeding to describe the drugs used therapeutically. Finally we consider the extent to which new knowledge of cancer biology is leading to new treatments.

10.2 Introduction: Cancer

The medical term for 'tumor' or 'cancer' is neoplasm, which is defined as growth or mass of abnormal tissue formed due to excess and uncoordinated cell proliferation. Cancer is a disease in which there is uncontrolled multiplication and spread within the body of abnormal forms of the body's own cells. It is one of the major causes of death in the developed nations-at least one in five of the population of Europe and North America can expect to die of cancer.

The anticancer drugs either kill cancer cells or modify their growth. Most anticancer drugs are antiproliferative-most damage DNA and thereby initiate apoptosis. They also affect rapidly dividing normal cells and are thus likely to depress bone marrow, impair healing, depress growth, cause sterility and hair loss, and be teratogenic. Most cause nausea and vomiting.

10.3 Characteristic of tumors

Cancer cells manifest, to varying degrees, four characteristics that distinguish them from normal cells:

- Abnormal uncontrolled cell divison
- Dedifferentiated and loss of function
- Ability to spread to other parts of the body
- Metastasis

10.4 Types of tumors

A. **Malignant tumor-** these are cancerous tumor which are fast grouing and invade other body parts. They cannot be removed by surgery. These tumors may lead to death.

B. **Non malignant tumor or benign tumor-** These are non-cancerous tumor which are slow growing and do not invade other parts of the body. They are removed by normal surgery and does not lead to death. According to the embryologic origin the name for cancers is divided into two general categories-

- Sarcoma- Sarcoma is a cancer which aries from the abnormal growth of mesodermal tissue.
- Carcinoma- Carcinoma is a cancer which arises from ecto or endodermal cells is called a carcinoma.

10.5 Carcinogenic agents

These agents causes cancer or tumor, as follows-

1. Chemical agents-

- **Initiators**- These agents are initiate the growth of tumors. eg:polygenic aromatic hydrocarbons, azodyes, aromatic amines, alkylating agents.
- **Promoters-** These agents are initiate the growth of already present tumor, but itself does not initiate tumor growth. eg:phenolic compounds,artificial swetners(saccharin and cyclamate) and phenobarbitol.

2. Physical agents-

- Radiation
- Mechanical injury for tissues
- Ionizing and U.V. radiation

3. Hormonal agents-

- Oestrogen hormone
- Anabolic ateroids

4. Biological agents-

- Stones in gall bladder and urinary tract
- Viral infections

10.6 Pathogenesis

The basis for tumor formation is change in gentic factors which leads to non lethal genetic damage of cells. There are two types of regulatory genes in the body which protects during development of cancer,these two genes are the primary targets-

- Growth promoting proto onchogene
- Growth suppressor genes or entionchogens.

10.7 Cancer Chemotherapy

Cancer is a disease characterized by a shift in the control mechanisms that govern cell survival, proliferation, and differentiation. Cells that have undergone neoplastic transformation usually express cell surface antigens that

may be of normal fetal type, may display other signs of apparent immaturity, and may exhibit qualitative or quantitative chromosomal abnormalities, including various translocations and the appearance of amplified gene sequences. Such cells proliferate excessively and form local tumors that can compress or invade adjacent normal structures. A small subpopulation of cells within the tumor can be described as tumor stem cells. They retain the ability to undergo repeated cycles of proliferation as well as to migrate to distant sites in the body to colonize various organs in the process called metastasis.

Cancer chemotherapy means anticancer agents or antineoplastic agents. Cancer chemotherapy is aimed either to kill cancer cells or modify their growth. Anticancer drugs generally attack the metabolic sites that are essential in cell replication. The drugs used in chemotherapy of cancer destroys an constant fraction of malignant cells (Fig. 10.7).

Cancer chemotherapeutic agents-

(1)Drugs acting directly on cells (Cytotoxic drugs)

(A)ALKYLATING AGENTS

- Mechlorethamine

(a) Nitrogen mustards (Mustine HCl)

- Cyclophosphamide,
- Chlorambucil,
- Melphalan

(b) **Ethylenimine**

- Thio-TEPA

(c) **Alkyl sulfonate**

- Busulfan

(d) Nitrosoureas

- Carmustine (BCNU),
- Lomustine (CCNU)

(e) **Triazine**

- Dacarbazine (DTIC)

(B)ANTIMETABOLITES

(a) **Folate antagonist**

- Methotrexate (Mtx)

(b) Purine antagonist

- 6-Mercaptopurine (6-MP)
- 6-Thioguanine (6-TG)
- Azathioprine
- Fludarabine

(c) Pyrimidine antagonist

- 5-Fluorouracil (5-FU)
- Cytarabine (cytosine arabinoside)

(C) VINCA ALKALOIDS

- Vincristine (Oncovin)
- Vinblastine

(D) TAXANES

- Paclitaxel
- Docetaxel

(E) EPIPODOPHYLLO TOXIN

- Etoposide

(F) CAMPTOTHECIN ANALOGUES

- Topotecan, Irinotecan

(G) ANTIBIOTICS

- Actinomycin
- Doxorubicin
- Daunorubicin
- Mitoxantrone
- Bleomycins,
- Mitomycin C

(H) MISCELLANEOUS

- Hydroxyurea
- Procarbazine
- L-Asparaginase
- Cisplatin

(II) Drugs altering hormonal milieu

(A) GLUCOCORTICOIDS

- Prednisolone and others

(B) ESTROGENS

- Fosfestrol
- Ethinylestradiol

(C) SELECTIVE ESTROGEN RECEPTOR MODULATORS

- Tamoxifen

(D) SELECTIVE ESTROGEN RECEPTOR DOWN REGULATORS

- Fulvestrant

(E) AROMATASE INHIBITORS

- Anastrozole
- Exemestane

(F) ANTIANDROGEN

- Flutamide
- Bicalutamide

(G) 5-ALPHA REDUCTASE INHIBITOR

- Finasteride
- Dutasteride

(H) GNRH ANALOGUES

- Nafarelin
- Triptorelin

(I) PROGESTINS

- Hydroxyprogesterone acetate

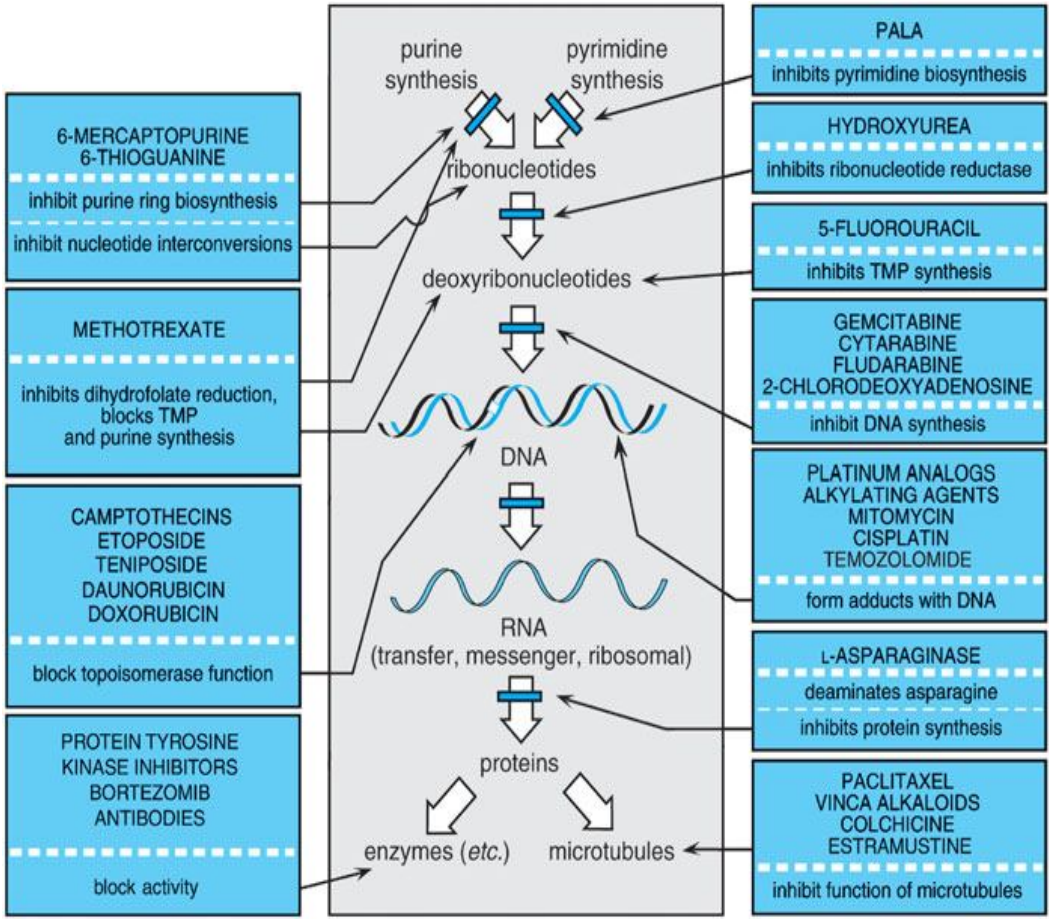


Fig.10.1: Summary of the mechanisms and sites of action of some chemotherapeutic agents useful in neoplastic disease. PALA = N-phosphonoacetyl-L-aspartate; TMP = thymidine monophosphate.

10.8 Special problems

Majority of the cytotoxic drugs have more profound effect on rapidly multiplying cells,because the most important target of action are the nucleic acids and their precursors; rapid nucleic acid synthesis occurs during cell division. Many cancers (especially large solid tumours) have a lower growth fraction (lower percentage of cells are in division) than normal bone marrow, epithelial linings, reticuloendothelial (RE) system and gonads. These tissues are particularly affected in a dose-dependent manner by majorityof drugs; though, there are differences in susceptibility to individual members.

1. **Bone marrow:** Depression of bone marrow results in granulocytopenia, agranulocytosis, thrombocytopenia, aplastic anaemia. This is the most serious toxicity; often limits the dose that can be employed. Infections and bleeding are the usual complications.

2. **Lymphoreticular tissue:** Lymphocytopenia and inhibition of lymphocyte function results in suppression of cell mediated as well as humoral immunity.
3. **Oral cavity:** The oral mucosa is particularly susceptible to cytotoxic drugs because of high epithelial cell turnover. Many chemotherapeutic drugs produce stomatitis as an early manifestation of toxicity. The gums and oral mucosa are regularly subjected to minor trauma, and breaches are common during chewing. Oral microflora is large and can be the source of infection. Neutropenia and depression of immunity caused by the drug indirectly
4. Increase the chances of oral infections. Thrombocytopenia may cause bleeding gums. Xerostomia due to the drug may cause rapid progression of dental caries.
5. **GIT:** Diarrhoea, shedding of mucosa, haemorrhages occur due to decrease in the rate of renewal of the mucous lining. Drugs that frequently cause mucositis are—bleomycin,actinomycin D, daunorubicin, doxorubicin, fluorouracil and methotrexate. Nausea and vomiting are prominent with many cytotoxic drugs. This is due to direct stimulation of CTZ by the drug as well as generation of emetic impulses/mediators from the upper g.i.t.
6. Skin Alopecia occurs due to damage to the cells in hair follicles. Dermatitis is another complication.
7. **Gonads:** Inhibition of gonadal cells causes oligozoospermia and impotence in males; inhibition of ovulation and amenorrhoea are common in females. Damage to the germinal cells may result in mutagenesis.
8. **Foetus:** Practically all cytotoxic drugs given to pregnant women profoundly damage the developing foetus → abortion, foetal death, teratogenesis.
9. **Carcinogenicity:** Secondary cancers, especially leukaemias, lymphomas and histocytic tumours appear with greater frequency many years after the use of cytotoxic drugs. This may be due to depression of cell mediated and humoral blocking factors against neoplasia.
10. **Hyperuricaemia:** This is secondary to massive cell destruction (uric acid is a product of purine metabolism). Gout and urate stones in the urinary tract may develop. Allopurinol is protective by decreasing uric acid synthesis. In addition to these general toxicities, individual drugs may produce specific adverse effects, e.g. neuropathy by vincristine, cardiomyopathy by doxorubicin, cystitis and alopecia by cyclophosphamide.

10.9 Action of Antineoplastic agents

Anticancer drugs act in many ways-

a) They can react with the nuclei iof cells as well as with the cell membrances and other cell organelles.

b) Antitumor drugs can act at all phases of the cell cycle (fig-10.9) by inhibiting cellular process : such as by inhibiting the growth of the cell components such as DNA,RNA and protein.

c) They act by distrupting DNA dependent enzymes such as DNA or RNA polymerase, which are essential for replication and transcription of the cellular DNA.

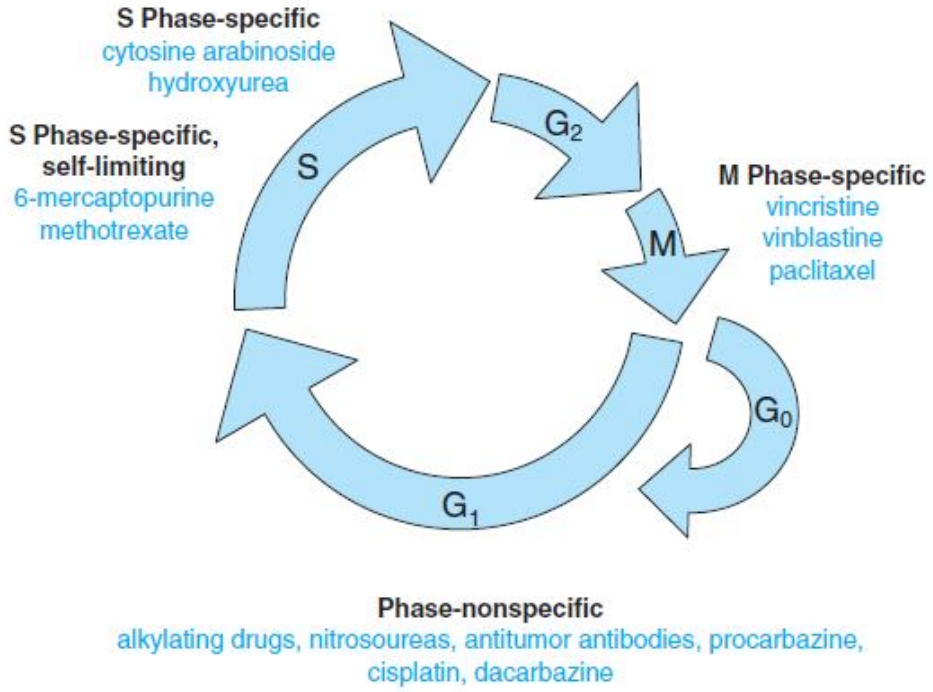


Fig. 10.9: The Cell Cycle and the relationship of antitumor drug action to the cycle G1 is the gap period between mitosis and the beginning of DNA synthesis. Resting cells (cells that are not preparing for cell division) are said to be in a subphase of G1, G0. S is the period of DNA synthesis, G2 the premitotic interval, and M the period of mitosis.

10.10 Role of Alkylating agents in treatment of cancer

These are called as polyfunctional alkyleting agents and characterized by containing at least one and usually two or more reactive allyl groups in the molecule which perform omportant cellular functions. Alkylating agents act upo DNA,RNA and certain enzymes. These compounds produce highly

reactive carbonium ion intermediates which transfer alkyl groups to cellular macromolecules by forming covalent bonds (Fig. 10.10). Alkylating agents can damage bone marrow and cautiously used in the treatment of cancers of lymphoid tissues. The alkylating agents are thought to react with the 7 position of guanine in each of the double strands of DNA causing cross-linking. This interferes in the separation of the strands and prevent mitosis. Alkylating agents have cytotoxic and radiomimetic action.

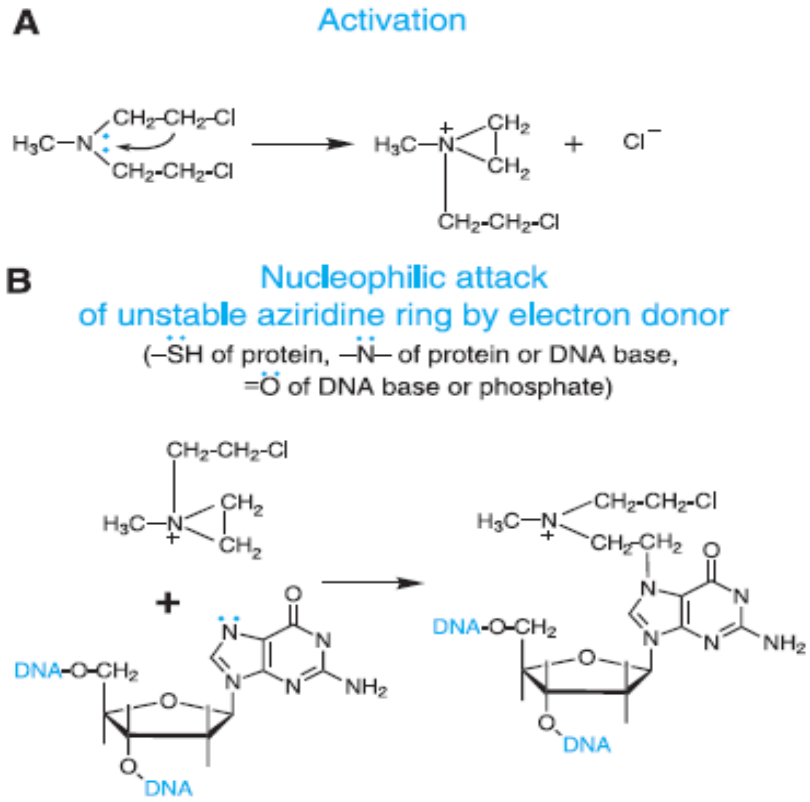
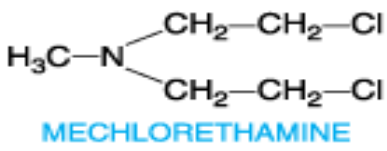


Fig. 10.10: Mechanism of action of Alkylating agents

Mechlorethamine-

It is the nitrogel mustard drug and also known as mustine hydrochloride.The principal use of mechlorethamine is in combination chemotherapy of hodgkin's disease and the non-hodgkin's lymphomas. It has veterinary use,including treatment of lymphosarcoma and mast cell sarcoma in dogs and leukesis in chickens.

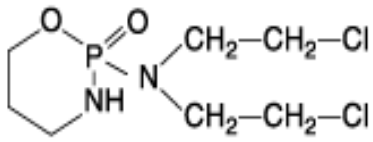
Structure-



Cyclophosphamide-

Cyclophosphamide consist of a nitrogen and phospharamide moiety in its structure. It is used to treat lymphosarcomas and hodgkin's disease as well as breast,ovarian and lung cancer.

Structure-

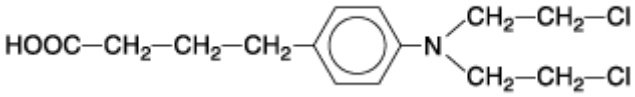


CYCLOPHOSPHAMIDE

Chlorambucil-

It has properties similar to that of mechlorethamine . chlorambucil is the drug of choice in chronic lymphatic lukaemia. It is also used in hodgkin's disease and some solid tumors. It has some immunosuppressive property.

Structure-

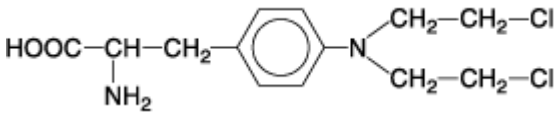


CHLORAMBUCIL

Melphalan-

It is an phenylalanmine nitrogen mustard. It is very effective in multiple mycloma,breast cancer and advanced ovarian cancer.

Structure-



MELPHALAN

Thiotepa-

It is an ethyleneimine compound. It is used for superficial bladder tumours.

Busulphan-

It is an alkyl sulphonate.

Structure-

$CH_3SO_2OCH_2CH_2CH_2CH_2OSO_2CH_3$

Busulphan

It is highly effective on granulocyte and is used in chronic myclocytic bukemia.

Nitrosoureas-

Nitrosoureas (carmustine and lomustine) are highly lipid soluable alkylating agents. Nitrosoureas have cumulative depressive effect on bone narrow. They have clinical activity against. The lymphomas, malignant melanoma and Hodgkin's disease.

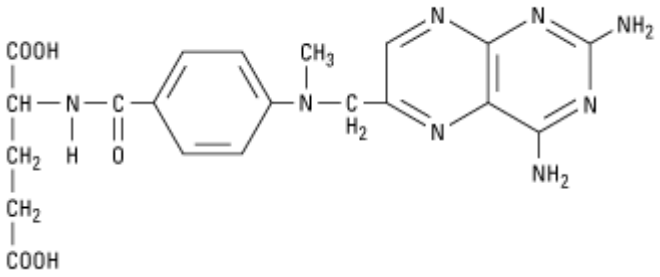
10.11 Role of antimetabolites in treatment of cancer

Antimetabolites are structurally related to normal components of DNA or of coengymes involved in the nuclic acid aynthesis. Antimetabolites inhibits a metabolic pathway which is essential for survival or reproduction of cancer cells through inhibition of folate, purine, pyrimidine and pyrimidine nucleoside which are also required for DNA synthesis. Antimetabolites can kill the cancer cells without damaging the host cells.

Methotrexate-

It is an analogue of folic acid. It is an antimetabolite of tolic acid.

Structure-



Methotrexate

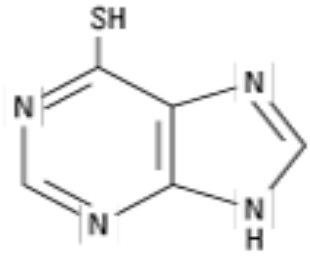
In the cell, folic acid is first of all reduced to dihydrofolic acid and then to tetrahydrofolic acid. Methotrexate is able to inhibit the enzyme dihydrofolate reductase and does not allow the formation of tetrahydrofolate which has been essential for synthesis of purine and pyrimidine and then inhibits the DNA ,RNA and protein synthesis.

Methotrexate used in choriocarcinoma, the acute leukemias,,osteosarcoma and head, neck and breast cancer.

6-mercaptopurine(6-mp) –

It is an analogue of purine , which is essential component of DNA called adenine. The drug is converted in the cells to ribonucleotide of 6-mp, which then suppresses the denovo biosynthesis of purines and DNA.

Structure-



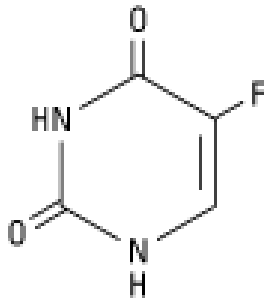
6-mercaptopurine

It is effective in choriocarcinoma leukemias and chronic myelocytic leukemia.

5-Flurouracil-

It is a pyrimidine analogue.

Structure-



5-Flurouracil

5-Fluorouracil converted into

↓

5-dump(deoxyuridine monophosphate)

↓

and interfere with DNA synthesis and functions by inhibiting thymidylate synthetase enzyme

The drug is mainly used in solid tumors of breast, colon, urinary bladder, liver etc. it is also effective in treating superficial basal cell carcinomas by topical application.

10.12 Carcinolytic antibiotics

These are products obtained from microorganisms and have antitumour activity. They intercalate between DNA strands and interfere with its template function.

Dactinomycin-

It is very potent antineoplastic antibiotic obtained from the species of streptomyces. The drug interfere with the movement of RNA polymerase along the gene and thus preventing transcription. It may also cause strand breaks and stabilize DNA-topoisomerase-2 complex.

It is effective in combination with methotrexate against gestational choriocarcinomr.

Bleomycin-

Bleomycin are a group of metal-chelating geycopeptide antibiotics obtained from streptomyces verticillus. They have antitumour activity. These antibiotics degrade performed DNA, causing chain fragmentation and release of free bases.

Bleomycin is mainly used in the treatment of testicular tumors. it is also highly effective in squamous cell carcinoma of skin, oral cavity, head ,neck, genitourinarytract, esophagus and hodgkin's lymphoma.

Mithramycin-

It is an antibiotic, produced by streptomyces argillaceous. It is binds to DNA and is more inhibitory to RNA.

It is useful in hypercalcemia, and paget's disease of bone.

Anthramycin-

it is derived from streptomycetes. The drug binds in the narrow groove of DNA with the guanine by a covalent binding and the drug inhibits nucleic acid synthesis.

It is used as a antitumor agent.

Mitomycins-

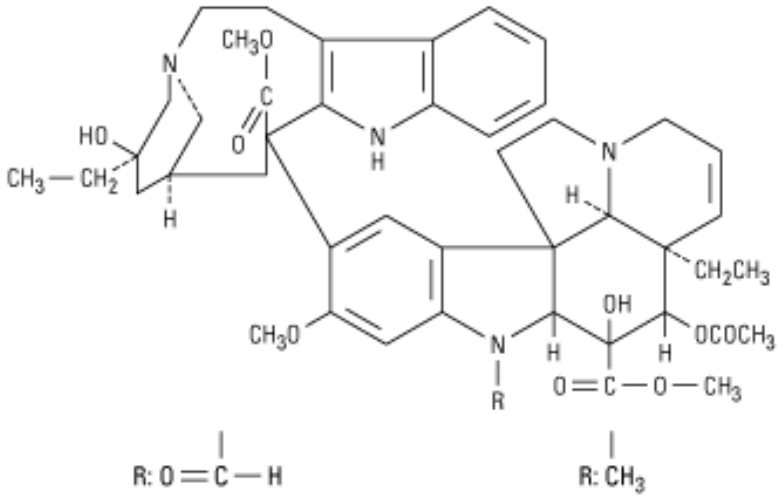
It is obtained from streptomyces caespitosus. It cross links DNA and may also degrade DNA by generating free radicals. It is highly toxic drug used in resistant cancers of stomach, cervix, colan, rectum, and bladder etc.

10.13 Mitotic inhibitor

Mitotic inhibitors such as 'vincristine' and vinblatine, are the main vinca alkaloids used in cancer chemotherapy. They bind to microtubular protein-'tubulin' which prevents its polymerization, causes disruption of mitotic spindle and interfese with cytoskeletal function interfese with cytoskeletal function in mitosis and metaphase arrest occurs, vincristine is mainly used in treatment of childhood acute leukaemia. It is also used in lymposarcoma, Hodgkin,s disease

and carcinoma lung. Vinblastine is used in combination Hodgkin,s disease and testicular carcinoma.

Structure-



Vincristine **Vinblastine**

Vinblastine-

Vinblastine is an alkaloid obtained from vinca rosea. Its mode of action involves depolymerization of microtubules, which are important part of mitotic spindle. The drug useful in Hodgkin,s disease and other lymphomas.

Vincristine-

Vincristine is an alkaloid derived from vinca rosea, causes arrest of mitotic cycle(spindle poison) .

Vinblastine and vincristine

↓

Binds to β tubulin (drug tubulin complex)

↓

Disruption of mitotic spindle

↓

Chromosome fail to move apart during mitosis

↓

Metaphase arrest

↓

Cell division is inhibited

Use: This drug useful in acute lukaemia in children.

10.14 Summary

Cancer is a disease characterized by a shift in the control mechanisms that govern cell survival, proliferation, and differentiation. The terms cancer, malignant neoplasm and malignant tumour are synonymous; they are distinguished from benign tumours by the properties of dedifferentiation, invasiveness and the ability to metastasis. There are three main approaches to treating established cancer- surgical excision, irradiation and chemotherapy-and the role of each of these depends on the type of tumour and the stage of its development. Chemotherapy can be used on its own or as an adjunct to other forms of therapy. Cancer Chemotherapeutic agents are: alkylating agents and related compounds, which act by forming covalent bonds with DNA and thus impeding DNA replication; antimetabolites, which block or subvert one or more of the metabolic pathways involved in DNA synthesis; cytotoxic antibiotics, i.e. substances of microbial origin that prevent mammalian cell division; plant derivatives (vinca alkaloids, taxanes, camptothecins): most of these specifically affect microtubule function and hence the formation of the mitotic spindle; Hormones-of which the most important are steroids, namely glucocorticoids, estrogens and androgens-and drugs that suppress hormone secretion or antagonise hormone action.

10.15 Glossary

- The term cancer refers to a malignant neoplasm (new growth).
- A small subpopulation of cells within the tumor can be described as tumor stem cells.
- Metastases are secondary tumours formed by cells that have been released from the initial or primary tumour and have reached other sites through blood vessels or lymphatics, or as a result of being shed into body cavities.
- Alkylating agents have alkyl groups that can form covalent bonds with cell substituents; a carbonium ion is the reactive intermediate.
- Antimetabolites block or subvert pathways in DNA synthesis.
- Carcinolyting agents inhibits DNA and RNA synthesis.

10.16 Review questions / Comprehensive Questions

1. What is antineoplastic agents?
2. Explain and classify the antineoplastic agents.
3. Define cancer. And discuss about carcinogenic agent
4. Discuss the role of alkylating agents.
5. Discuss the role of antimetabolites.
6. Discuss the mitotic inhibitors and carcinolytic agents.
7. Discuss the mechanism of action of cancer chemotherapeutic agents.
8. What is neoplasm?

10.17 References and Suggested readings

- Essential of Medical Pharmacology (7th ed.)- KD Tripathi (JAYPEE Publisher) 2013.
- Basic & Clinical Pharmacology (9th ed.)- Bertram G.Katzung (Mc Graw Hill Publisher) 2004.
- The Pharmacological basis of Therapeutics (10th ed.)- Goodman & Gilman's (Mc Graw Hill Publisher) 2001.
- Pharmacology(4th ed.)- Lippincott's (Wolters Kluwer Publisher) 2002.
- Textbook of Organic Medicinal and Pharmaceutical Chemistry (11th ed.)- Wilson and Gisvold's.

Unit - 11

Synthesis Antineoplastic agents

Structure of Unit:

11.1 Objectives

In this unit the students will be able to understand

- Cancer
- Types of cancer treatment
- Cancer Chemotherapy (Background)
- Cancer Chemotherapy
- Antineoplastic Agents
- Mechanism of action
- Synthesis of anticancer drugs
- Recent development in cancer treatment

11.2 Introduction: Antineoplastic agents

Cancer remains one of the major causes of death in the world and it has been estimated that there will be 15 million new cases and 10 million deaths in 2020. *Cancer* is a disease present in people and animals in which the structure and normal function of body tissues are disrupted. *Cancer* is a term used for diseases in which abnormal cells divide without control and are able to invade

other tissues. Cancer cells can spread to other parts of the body through the blood and lymph systems, this process is called metastasis.

Categorized based on the functions/locations of the cells from which they originate:

- Carcinoma - skin or in tissues that line or cover internal organs. E.g., Epithelial cells. 80-90% reported cancer cases are carcinomas.
- Sarcoma - bone, cartilage, fat, muscle, blood vessels, or other connective or supportive tissue.
- Leukemia - White blood cells and their precursor cells such as the bone marrow cells, causes large numbers of abnormal blood cells to be produced and enter the blood.
- Lymphoma - cells of the immune system that affects lymphatic system.
- Myeloma - B-cells that produce antibodies- spreads through lymphatic system.
- Central nervous system cancers - cancers that begin in the tissues of the brain and spinal cord.
- A neoplasm, or tumour is an abnormal mass of tissue, the growth of which exceeds and is uncoordinated with that of the normal tissue and continues in the same manner after cessation of the stimuli which have initiated it.
- A malignant tumour grows rapidly and continuously, and even when it has impoverished its host and source of nutrition, it still retains the potentiality for further proliferation. Besides, malignant tumours invade and destroy neighbouring tissues and possess no effective capsule, a malignant tumour readilty ulcerate and tend sooner or later to disseminate and form metastases.

The causation of neoplasms are many, for instance : the genetic factors e.g., retinoblastoma is determined by a Mendelian dominant factor and so are the multiple benign tumours ; the chemical carcinogens e.g., arsenic, soot, coal tar, petroleum lubricating oil ; the polycyclic hydrocarbon carcinogens e.g., 1, 2, 5, 6-dibenzanthracene, 3, 4-benzpyrene.

The important characteristics differentiating cancer cells from normal cells are:

1. Limitless replicative potential: Cells begin to divide uncontrollably because the mechanisms that control growth are disrupted.
2. Self-sufficiency in growth signals;

3. Nonresponsiveness to normal growth-inhibitory signals;
4. Escape from senescence and apoptotic death;
5. Ability to invade normal tissues and metastasize to distant sites; and
6. Support angiogenesis within the growing tumor.

There has been a tremendous growth in different aspects of cancer research, cancer chemotherapy vis-a-vis a better understanding of the intricacies of the 'tumour biology' that has ultimately led to not only the legitimate evolution but also the explicite elucidation of the probable mechanisms of action for the antineoplastic agents. In fact, the various strategies involved to augment the speedy as well as meaningful progress in the develoment of antineoplastic agents may be accomplished as follows:

- Fundamental basis for the more rational approach in the design of newer drugs,
- Large collaborative investigations, concerted integrated research based on recent developments and advances in the clinical techniques, and
- Combination of such privileged advantages with improved preliminary screening methodologies.

As on data nearly ten different types of 'neoplasms' may be 'cured'* with the aid of chemotherapy in patients quite satisfactorily, namely: leukemia in children, Hodgakin's disease, Burkitt's lymphoma, Ewing's sarcoma, choriocarcinoma in women, lymphosarcoma, mycosis fungoides, rhabdomyosarcoma, testicular carcinoma, and retinoblastoma in children.

Treatment of cancer includes surgical intervention, radiation, immunotherapy, and chemotherapy using neoplastic drugs. Chemotherapy is currently used in addition to surgical intervention in order to remove possible metastatic cells that still remain. Moreover, some types of tumors are currently treated first with chemotherapeutic agents. All characteristics targeted by recent therapies. In addition, the modulation of resistance with new drugs to potentate existing chemotherapeutic agents and/or the chemoprevention of primary tumors provide exciting new targets for cancer therapeutics. Salient-Features of Antineoplastic Drugs are as follows:

- Block the biosynthesis or transcription of nucleic acids to check cell-division through direct interference with mitotic spindles,
- Both mitosis phases and cells that are engaged in DNA-synthesis are found to be highly susceptible to these antineoplastic agents, and

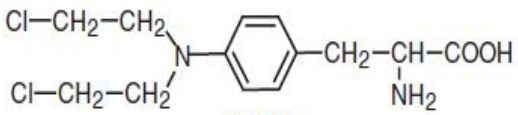
- In the resting state the not-so-fast growing tumours invariably possess good number of cells.

11.3 Classification of Antineoplastic agents

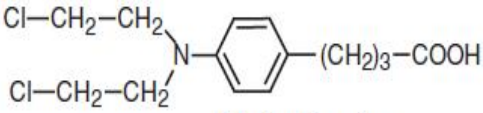
1. Alkylating Agents:

Alkylating agents are chemically reactive compounds that combine most readily with nucleophilic centres a fully saturated carbon atom of the alkylating group becoming attached to the nucleophile.

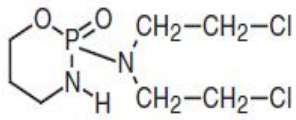
(i) Mustards: Nitrogen mustards were selected for the clinical application for the treatment of neoplasms because they presented fewer problems in handling, besides their respective hydrochlorides and other salts are generally stable solids having low vapour pressure and high solubility in water. A few important nitrogen mustards used as antineoplastic agents are discussed below, for instance: Mechlorethamine hydrochloride, Mephalan, Cyclophosphamide, Ifosamide and Chlorambucil.



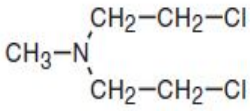
Melfalan



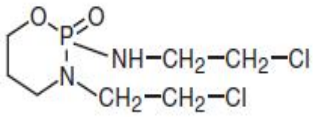
Mechorethamine



Cyclophosphamide

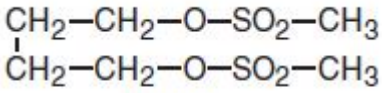


Chlorambucil



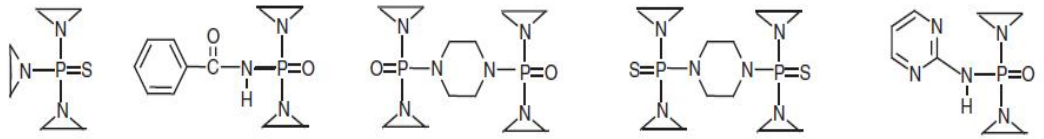
Ifosfamide

(ii) Methanesulphonates: The most important alkylating agent in this group is Busulfan.



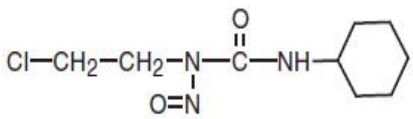
Busulfan

(iii) Ethylenimines: Ethylenimines are highly reactive alkylating reagents. They alkylate DNA at position N_7 of guanine, analogous to mechlorethamine. Ethyenimines exhibit cytostatic action and suppress development of proliferating, as well as malignant tissues. They disrupt the metabolism of nucleic acids and block mitotic cell division. They are used for breast and ovarian cancer, nonoperable tumors, and other reoccurrences and metastases. Example- triethylenphosphortriamide or thiotepa, benzotef, dipin, thiodipin, fosphemide, etc.

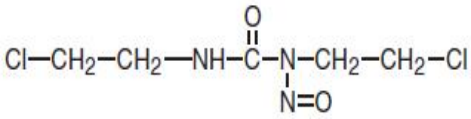


Thiotepa Benzotef Dipin Thiodipin Fosphemide

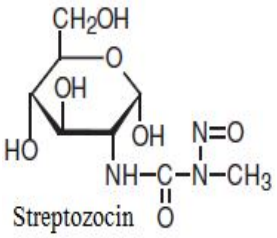
***(vi) Nitrosoureas*:** It is believed that in the body, nitrosoureas break down to β-chloroethanol and alkylisocyanate. The resulting β-chloroethanol is a highly reactive alkylating agent, and the alkylisocyanates are carbamoylating agents for proteins, which also exhibit certain cytotoxic activity. Example- Carmustine, Lomustine, Streptozocin, Cisplatin, Carboplatin, etc.



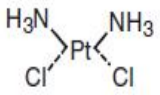
Lomustine



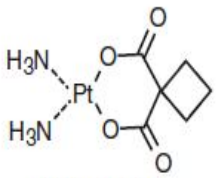
Carmustine



Streptozocin



Cisplatin

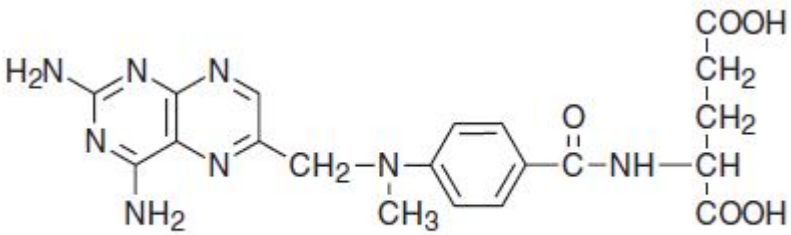


Carboplatin

2. Antimetabolites:

Antimetabolites are structural analogs of ordinary cellular metabolites such as folic acid, pyrimidines and pyrines, which after being introduced in the body, begin to imitate the structure of ordinary metabolites. They compete with metabolites to block important reactions leading to formation of DNA/RNA. In general, following are the various classes of antimetabolites usually employed in the treatment, namely:

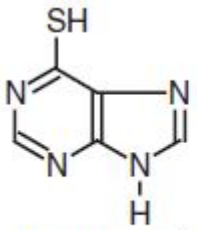
(i) ***Antifolic acid compounds*:** Folic acid antagonists, in particular methotrexate, act by competitively binding with the enzyme dehydrofolate reductase in place of folic acid.



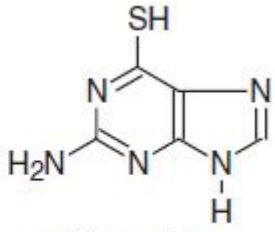
Methotrexate

(ii) ***Analogues of Purines*:** These compounds inhibit synthesis of purine nucleotides, which are made up of purine bases and phosphorylated ribose.

Purine, mercaptopurine and thioguanine turned out to be the most effective in chemotherapy for cancer, both compounds must be transformed into nucleotides by adding a phosphoribosyl fragment.

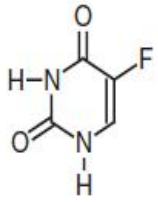


Mercaptopurine

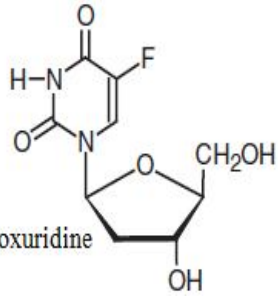


Thioguanine

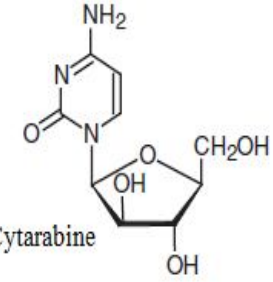
(iii) ***Analogues of Pyrimidines*****:** It is possible that the most important mechanism of action of fluorinated pyrimidines is the inhibition of thymidylate synthetase synthesis, thus affecting the process of DNA production. Example- fluorouracil, fluoxuridine, Cytarabine, etc.



Fluorouracil



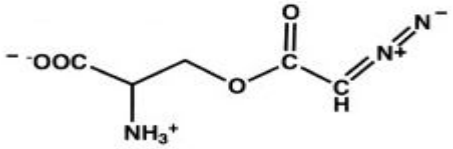
Fluoxuridine



Cytarabine

(iv) ***Amino acid antagonists*****:** The amino acid antagonists broadly act as glutamine antagonists in the synthesis of formylglycinamidine ribotide from glutamine and formylglycinamide ribotide.

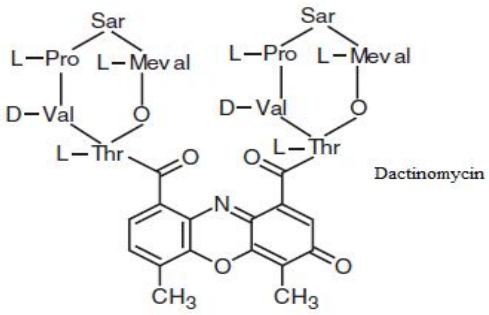
Example- Azaserine.



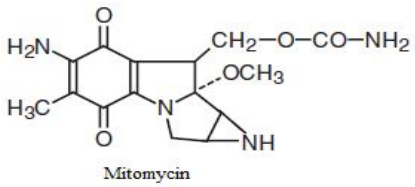
Azaserine

3. Antibiotics:

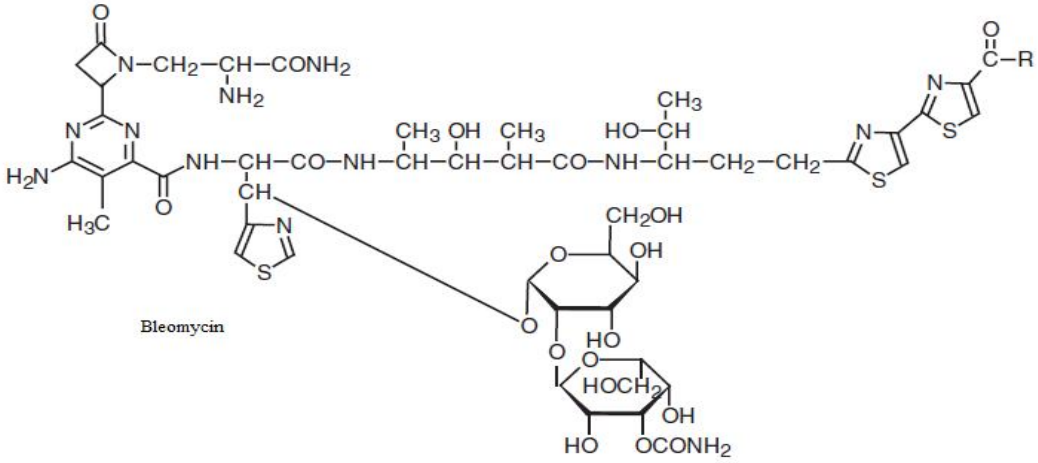
A number of antibiotics possess pronounced cytostatic properties, and they are extremely effective in treating certain tumors. Included in this group are dactinomycin, anthracyclins (daunorubicin and doxorubicin), bleomycin, etc.



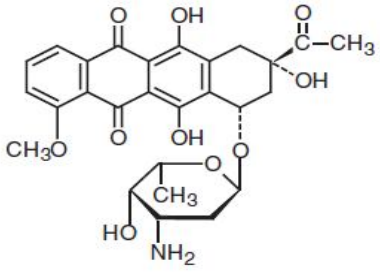
Dactinomycin



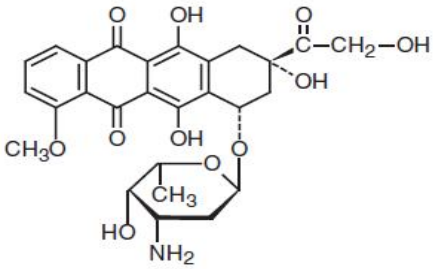
Mitomycin



Bleomycin



Daunorubicin



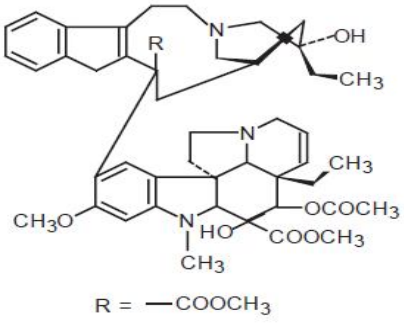
Doxorubicin

4. Plant products:

Vinblastine and vincristine are alkaloids isolated from plants of the periwinkle family (*Vinca rosea*). These compounds cause cells to stop at metaphase and inhibit assembly of microtubules, and likewise, failure of mitotic spindle formations. They inhibit synthesis of nucleic acids and proteins. Vinblastine and vincristine differ only in the substituent on the nitrogen atom of the indol fragment of the molecule, and are used in combination with other chemotherapeutic agents. They are mainly used for leukoses, myelomas, sarcomas, cancer of various organs, and for lymphomas.

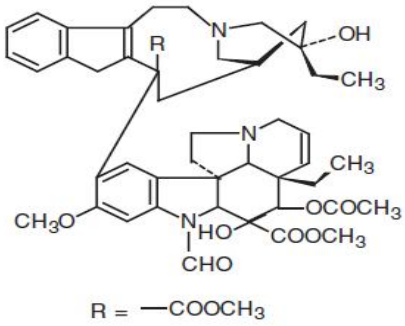
Etoposide and teniposide are synthetic derivatives of the extract of the American mandragora plant (May Apple). The mechanism of their action has not been completely explained; however, they act on the enzyme topoisomerase II, which disturbs the twisting of DNA. In addition, they inhibit DNA and RNA

synthesis, as well as transport of nucleotides to cells. Cytotoxic action on normal cells is observed only in very high doses. These drugs exhibit significant activity in lymphomas, leukemia, Kaposi's sarcomas, and in testicular cancer.



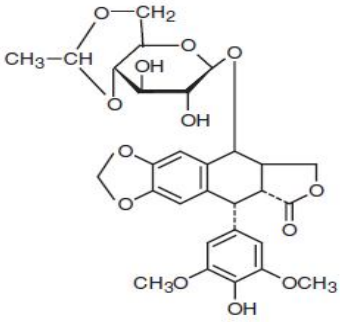
R = —COOCH3

Vinblastine

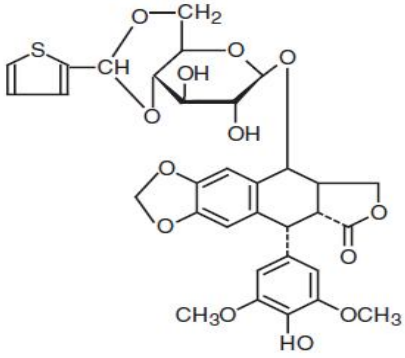


R = —COOCH3

Vincristine



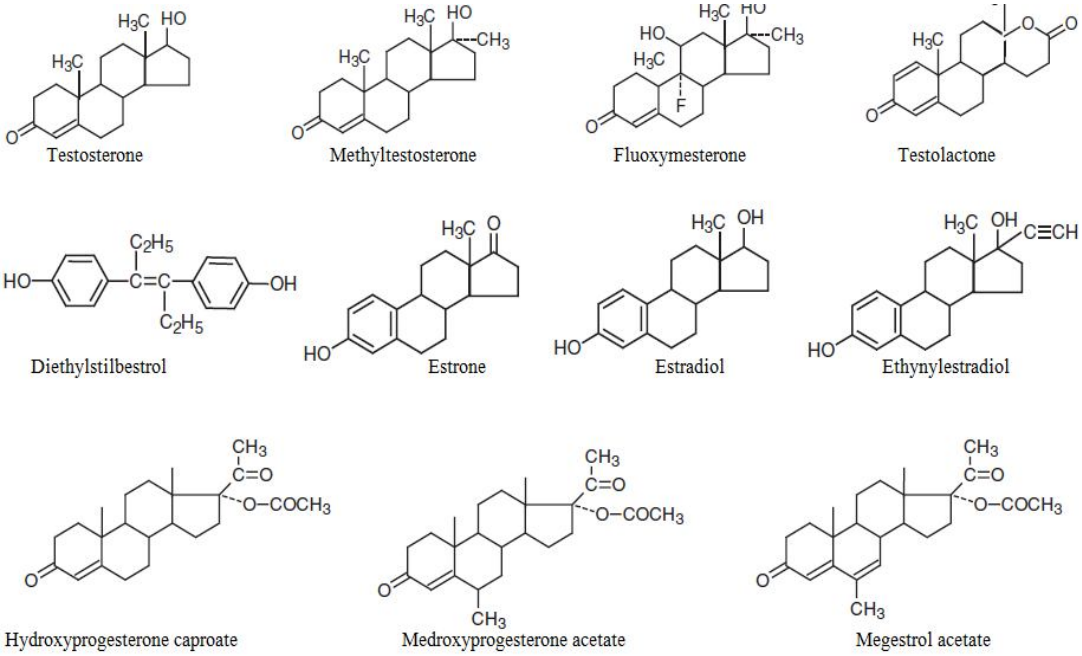
Etoposide



Teniposide

5. Hormones:

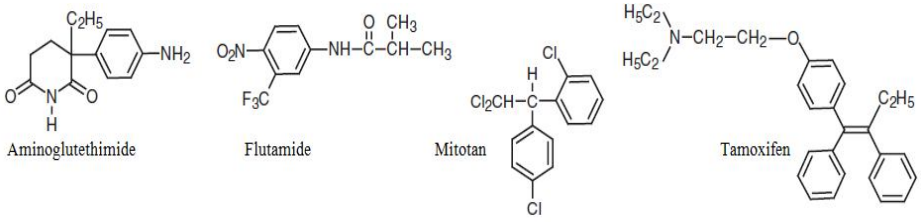
Hormonal drugs are successfully used for complex treatment of malignant tumors. Tumors can be both hormone-dependent, as well as hormone-sensitive. Hormone-dependent tumors regress in the absence of hormonal activity. In particular, the antiestrogen drug tamoxifen prevents stimulation of cancerous breast tumor cells by estrogens. This also applies to aminoglutethimide, an inhibitor of corticosteroid synthesis by the adrenal glands. Hormonal drugs that inhibit growth of certain human tumors are steroids, including androgens, estrogens, progestins, and corticosteroids, although only glucocorticoids are used. Moreover, neither cortisol nor cortisone is used to treat malignant tumors, but instead prednisone, prednisolone, methylprednisolone, and dexametasone are used.



11.4 Synthesis approach of antineoplastic agents

5. **Non-hormonal drugs:**

In addition to hormonal drugs, five other nonsteroids that have a direct relationship to this section are also used in cancer chemotherapy. They are aminoglutethimide, flutamide, mitotan, tamoxifen, and leuprolide.



Synthesis of mechlorethamine:

Mechlorethamine, bis-(2-chloroethyl)methylamine, is made by reacting methylamine with ethylene oxide, forming bis-(2-hydroxyethyl)methylamine, which upon reaction with thionyl chloride turns into the desired mechlorethamine.

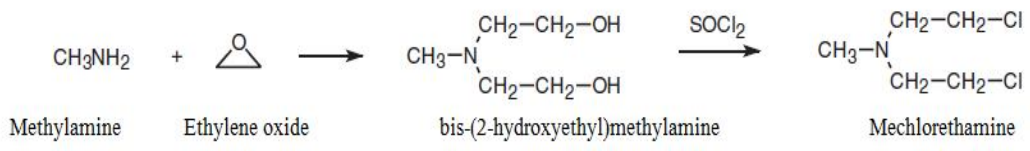


Fig.11.1: Synthesis of mechlorethamine

Mechlorethamine is widely used intravenously in combination with other drugs to treat Hodgkin's disease, lymphosarcoma, leukemia, and bronchogenic carcinoma.

Synthesis of cyclophosphamide: Cyclophosphamide, 2-[bis-(2-chloroethyl) amino]tetrahydro-2H-1,3,2- oxazaphosphorin-2-oxide, is made by reacting bis(2-chloroethyl)amine with phosphorous oxychloride, giving N,N-bis-(2-chloroethyl)dichlorophosphoramide, which upon subsequent reaction with 3-aminopropanol is transformed into Cyclophosphamide.

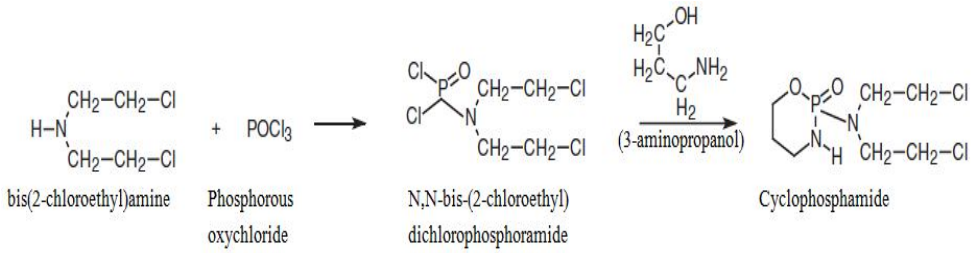


Fig. 11.2: Synthesis of cyclophosphamide

The distinctive chemical structure of this drug gives it selective antineoplastic activity. Present in the blood, it is practically inactive, although upon penetrating cancerous cells and reacting with a relatively large number of phosphamidases, it cleaves, essentially releasing a cytostatic substance, bis-(2-chloroethyl)amine. This means that the alkylating action of this drug is specifically directed toward cancerous cells.

It is used for chronic lymphatic leukemia, Hodgkin's disease, Burkitt's lymphoma, multiple myeloma, and cancer of the breast, neck, ovaries, etc.

Synthesis of melphalan:

Melphalan, L-3-[p-[bis-(2-chloroethyl)amino]phenyl]alanine, is a structural analog of chlorambucil in which the butyric acid fragment is replaced with an aminoacid fragment, alanine. This drug is synthesized from L-phenylalanine, the nitration of which with nitric acid gives 4-nitro-L-phenylalanine. Reacting this with an ethanol in the presence of hydrogen chloride gives the hydrochloride of 4-nitro-L-phenylalanine ethyl ester, the amino group of which is protected by changing it to phthalamide by a reaction with succinic anhydride to give intermediate. The nitro group in this molecule is reduced to an amino group using palladium on calcium carbonate as a catalyst. The resulting aromatic amine is then reacted with ethylene oxide, which forms a bis-(2-hydroxyethyl)-amino derivative. The hydroxy groups in this molecule are replaced with chlorine atoms upon reaction with thionyl chloride, after which treatment with hydrochloric acid removes the phthalamide protection, giving melphalan.

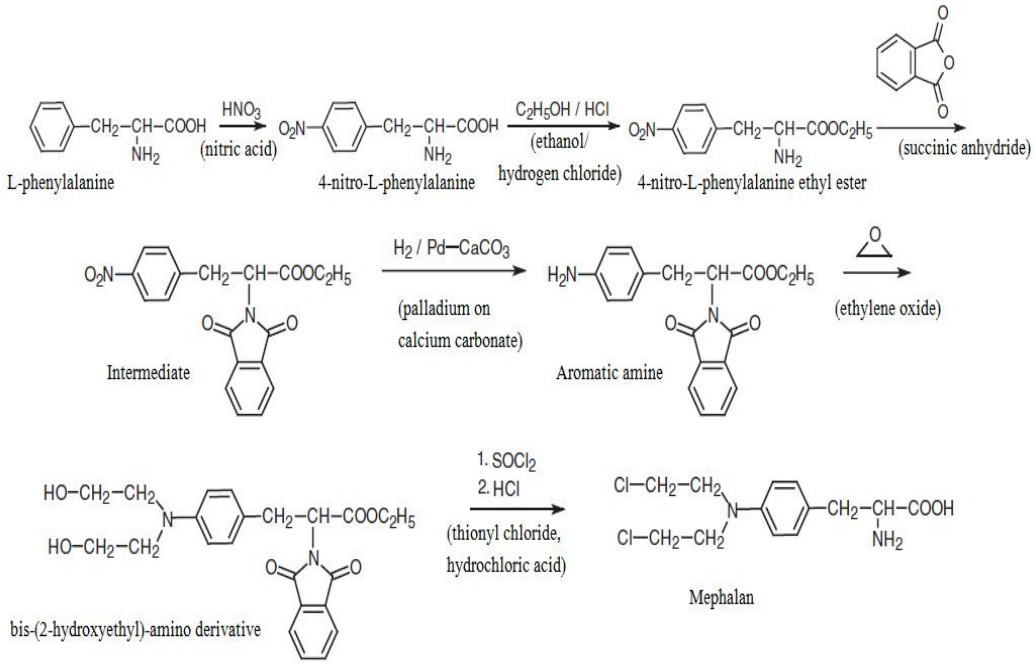


Fig. 11.3: Synthesis of melphalan

Melaphalan is used intravenously and orally to treat multiple myeloma and cancers of the breast, neck, and ovaries. A synonym of this drug is alkeran. The racemic form of this drug, D,L-3-[p-[bis-(2-chloroethyl)amino]phenyl]alanine, is also widely used under the name sarcolysine or racemelfalan.

Synthesis of Fluorouracil:

Fluorouracil, 4-fluorouracil, is made by condensing the ethyl ester of fluoroacetic acid with ethylformate in the presence of potassium ethoxide, forming hydroxy-methylenfluoroacetic ester, which cyclizes by reacting it with S-methylisothiourea to 2-methylthio-4-hydroxy-5-fluoropyrimidine, which is subsequently hydrolyzed by hydrochloric acid to fluorouracil. An alternative method of synthesizing 5-fluorouracid is direct fluorination of uracil with fluorine or trifluoromethyl hypofluoride.

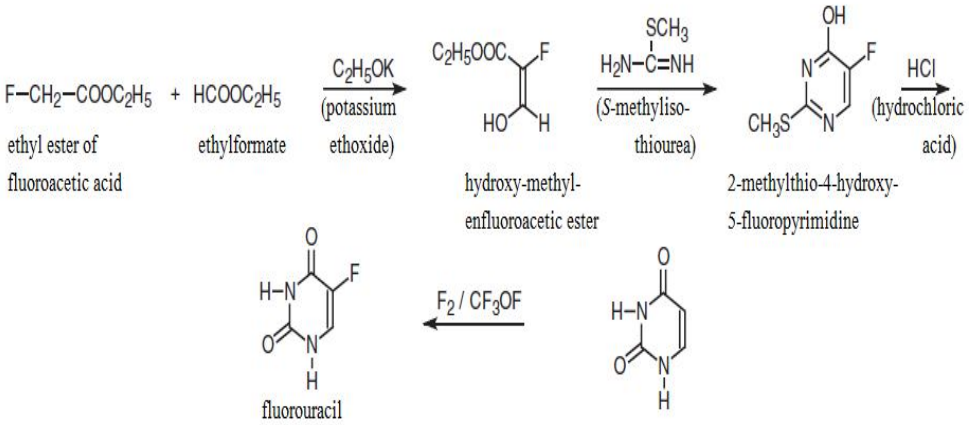


Fig. 11.4: Synthesis of Fluorouracil

Fluorouracil acts by inhibiting synthesis of pyrimidine, and thus the formation of DNA. Fluorouracil is used to treat carcinomas of the head, neck, colon, rectum, breast, stomach, bladder, pancreas, and for actinic and solar creatitis.

Synthesis of mercaptopurine:

Mercaptopurine, 6-purinthiol, is made from uric acid, which is synthesized from barbituric acid. Barbituric acid is easily made by condensing urea with malonic ester and then nitrosylating it with nitrous acid. The nitroso derivative is reduced by hydrogen (obtained in situ by reacting tin with hydrochloric acid) to an amine (uramil), and then reacted with isocyanic acid, which forms pseudouric acid. This undergoes cyclization to uric acid when heated in the presence of hydrochloric acid. Upon reacting phosphorous pentachloride with uric acid, 2,6,8-trichloropurine is formed. The three chlorine atoms in trichloropurine differ significantly in terms of reactivity for nucleophilic substitution. The chlorine atom at C6 is much more active than the chlorine atom at C2, and this is more active than the chlorine atom at C8, which allows subsequent manipulation by them. Interaction of 2,6,8-trichloropurine with sodium hydroxide allows to replace the chlorine atom at C6, forming the dichloro-derivative, which is then reduced by hydriodic acid to hypoxanthine. Upon reaction with phosphorous pentasulfide, hypoxanthine is transformed into mercaptopurine.

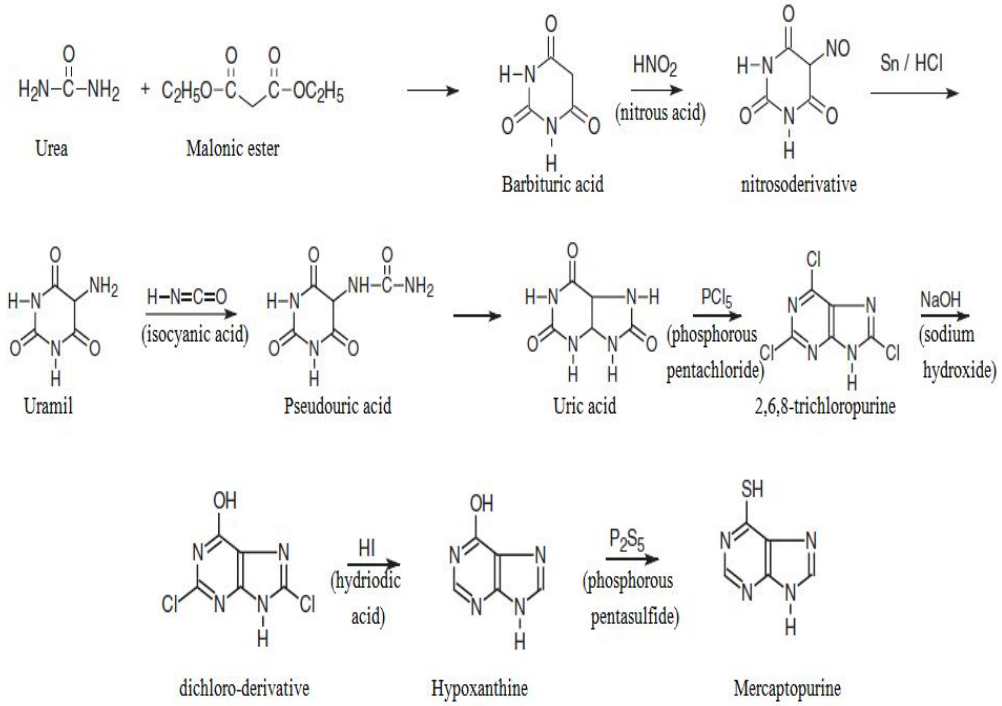


Fig. 11.5: Synthesis of mercaptopurine

In the body, mercaptopurine is converted into an active form of the drug, nucleotide 6-thioinosin-5-phosphate. Nucleotide 6-thioinosin-5-phosphate inhibits the first step in the synthesis of inosin-5-phosphate by negative feedback, preventing its transformation to adenosine or guanine nucleotides, which are necessary for synthesizing DNA. Thus, mercaptopurine inhibits synthesis and interconversion of purine nucleotides, which leads to a halt in DNA synthesis in proliferating cells during the cell cycle.

Mercaptopurine is used for treatment of lymphobastomas, myeloblastoma leucosis, and to treat neuroleukemia.

11.5 General toxicity of cytotoxic drugs

An understanding of toxicities, adverse effects, and special dosing considerations of existing anticancer compounds is important to the design of effective drug combinations and to the interpretation of the toxicological profile of new chemical entities. Most cytotoxic anticancer agents are dosed to maximum tolerated levels to achieve maximum cell kill.

Majority of the cytotoxic drugs have more profound effect on rapidly multiplying cells, because the most important target of action are the nucleic acids and their precursors; rapid nucleic acid synthesis occurs during cell division. Many cancers (especially large solid tumours) have a lower growth fraction (lower percentage of cells are in division) than normal bone marrow, epithelial linings, reticuloendothelial (RE) system and gonads. These tissues are particularly affected in a dose-dependent manner by majority of drugs; though, there are differences in susceptibility to individual members.

1. **Bone marrow:** Depression of bone marrow results in granulocytopenia, agranulocytosis, thrombocytopenia, aplastic anaemia. This is the most serious toxicity; often limits the dose that can be employed. Infections and bleeding are the usual complications.
2. **Lymphoreticular tissue:** Lymphocytopenia and inhibition of lymphocyte function results in suppression of cell mediated as well as humoral immunity.
3. Because of action (1) and (2) + damage to epithelial surfaces, the host defence mechanisms (specific as well as nonspecific) are broken down susceptibility to all infections is increased. Of particular importance are the opportunistic infections due to low pathogenicity organisms Infections by fungi (*Candida* and others causing deep mycosis), viruses (*Herpes zoster,*

cytomegalo virus), *Pneumocystis jiroveci* (a fungus) and *Toxoplasma* are seen primarily in patients treated with anticancer drugs.

4. **Oral cavity:** The oral mucosa is particularly susceptible to cytotoxic drugs because of high epithelial cell turnover. Many chemotherapeutic drugs produce stomatitis as an early manifestation of toxicity. The gums and oral mucosa are regularly subjected to minor trauma, and breaches are common during chewing. Oral microflora is large and can be the source of infection. Neutropenia and depression of immunity caused by the drug indirectly increase the chances of oral infections. Thrombocytopenia may cause bleeding gums. Xerostomia due to the drug may cause rapid progression of dental caries.
5. **GIT**: Diarrhoea, shedding of mucosa, haemorrhages occur due to decrease in the rate of renewal of the mucous lining. Drugs that frequently cause mucositis are-bleomycin, actinomycin D, daunorubicin, doxorubicin, fluorouracil and methotrexate. Nausea and vomiting are prominent with many cytotoxic drugs. This is due to direct stimulation of CTZ by the drug as well as generation of emetic impulses/mediators from the upper g.i.t. and other areas.
6. **Skin:** Alopecia occurs due to damage to the cells in hair follicles. Dermatitis is another complication.
7. **Gonads:** Inhibition of gonadal cells causes oligozoospermia and impotence in males; inhibition of ovulation and amenorrhoea are common in females. Damage to the germinal cells may result in mutagenesis.
8. **Foetus:** Practically all cytotoxic drugs given to pregnant women profoundly damage the developing foetus may results abortion, foetal death, teratogenesis.
9. **Carcinogenicity**: Secondary cancers, especially leukaemias, lymphomas and histocytic tumours appear with greater frequency many years after the use of cytotoxic drugs. This may be due to depression of cell mediated and humoral blocking factors against neoplasia.
10. **Hyperuricaemia**: This is secondary to massive cell destruction (uric acid is a product of purine metabolism). Gout and urate stones in the urinary tract may develop. Allopurinol is protective by decreasing uric acid synthesis. In addition to these general toxicities, individual drugs may produce specific adverse effects, e.g. neuropathy by vincristine, cardiomyopathy by doxorubicin, cystitis and alopecia by cyclophosphamide.

11.6 Recent development in cancer chemotherapy

In 2013, WHO launched the Global Action Plan for the Prevention and Control of Noncommunicable Diseases 2013-2030 that aims to reduce by 25% premature mortality from cancer, cardiovascular diseases, diabetes and chronic respiratory diseases.

There are many new approaches to cancer therapy, some at the very early research stage and others at later phases of development. These approaches range from novel drugs, biological agents and drug delivery systems, to gene targeting, gene therapy and, more recently, the possibility of targeting cancer stem cells.

New types of cancer treatment

- Hormonal Treatments: These drugs are designed to prevent cancer cell growth by preventing the cells from receiving signals necessary for their continued growth and division. E.g., Breast cancer – tamoxifen after surgery and radiation
- Specific Inhibitors: Drugs targeting specific proteins and processes that are limited primarily to cancer cells or that are much more prevalent in cancer cells.
- Antibodies: The antibodies used in the treatment of cancer have been manufactured for use as drugs. E.g., Herceptin, avastin
- Biological Response Modifiers: The use of naturally occuring, normal proteins to stimulate the body's own defenses against cancer. E.g., Abciximab, rituxmab
- Vaccines: Stimulate the body's defenses against cancer. Vaccines usually contain proteins found on or produced by cancer cells. By administering these proteins, the treatment aims to increase the response of the body against the cancer cells.

11.7 Summary

The main goal of anti-neoplastic drug is to eliminate the cancer cells without affecting normal tissues. Early diagnosis is the key. Combination therapy and adjuvant chemotherapy are effective for small tumor burden. Because chemotherapeutic agents target not only tumor cells, but also affect normal dividing cells including bone marrow, hematopoietic, and GI epithelium.

Treatment of cancer includes surgical intervention, radiation, immunotherapy, and chemotherapy using neoplastic drugs.

11.8 Glossary

- *Chemotherapy* means the use of any drug (such as aspirin or penicillin) to treat any disease, but to most people chemotherapy refers to drugs used for cancer treatment. It's often shortened to "chemo."
- Two other medical terms used to describe cancer chemotherapy are *Antineoplastic* (meaning anti-cancer) therapy and *cytotoxic* (cell-killing) therapy.
- *Alkylating agent*: a reactive chemical that forms a covalent bond with chemical moieties on the biological target (usually a protein). For instance, b-haloalkylamines generate an aziridinium ion in aqueous base that inserts into –SH, –CHOH, or other chemical structures in peptides. Once inserted, the effects of the alkylating agent are irreversible.
- *Gene* the sequence of DNA that codes for a complete protein.
- *Antimetabolites*: Interfere with the formation of key biomolecules including nucleotides, the building blocks of DNA.

11.9 Review questions / Comprehensive Questions

1. What is a neoplasm? What are the causations of neoplasm ?
2. How would you classify the 'antineoplastic agents' ? Give the structure, chemical name and uses of one important member from each category.
3. How would you classify 'Antimetabolities' ?
4. Give the structure, chemical name and uses of the following: Methotrexate, Meracaptopurine, Fluorouracil, Azaserine.
5. Discuss the synthesis of Meracaptopurine, Fluorouracil.
6. Classify the 'plant products' employed in the treatment of malignant disease. Give structure, name and uses of one potent drug from each category.
7. Give a comprehensive account of 'hormones' that are potent as antineoplastic agents. Support your answer with suitable examples.
8. Give a brief account of Pharmacokinetics, pharmacodynamic and mode of action of antineoplastic agents.

11.10 References and Suggested readings

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Unit - 12

Cardiovascular Drugs

Structure of Unit:

12.1 Objectives

In this unit the students will be able to understand

- Cardiovascular system (Heart)
- Cardiac output
- Blood Pressure
- Various types of cardiovascular diseases
- Class of drugs inhibit peripheral sympathetic function
- Centrally acting sympatholytics
- Directly acting vasodilators
- Prevention of blood coagulation
- Lysis of blood clot

12.2 Introduction: Cardiovascular system

The Cardiovascular system consists of heart, blood and blood vessels. It is a transport system which through blood transports respiratory gases, nutrients and excretory products to various parts of the body.

HEART –

Heart is surrounded by outer covering called pericardium. It contains two layers known as visceral pericardium and parietal pericardium.

Middle layer- Myocardium

Inner layer– Endocardium

Chambers of Heart-

- Atrium – 2 (Right and left)
- Ventricle – 2 (Right and left)

Functions-

It acts as a pump and maintains a constant circulation of blood throughout the body.

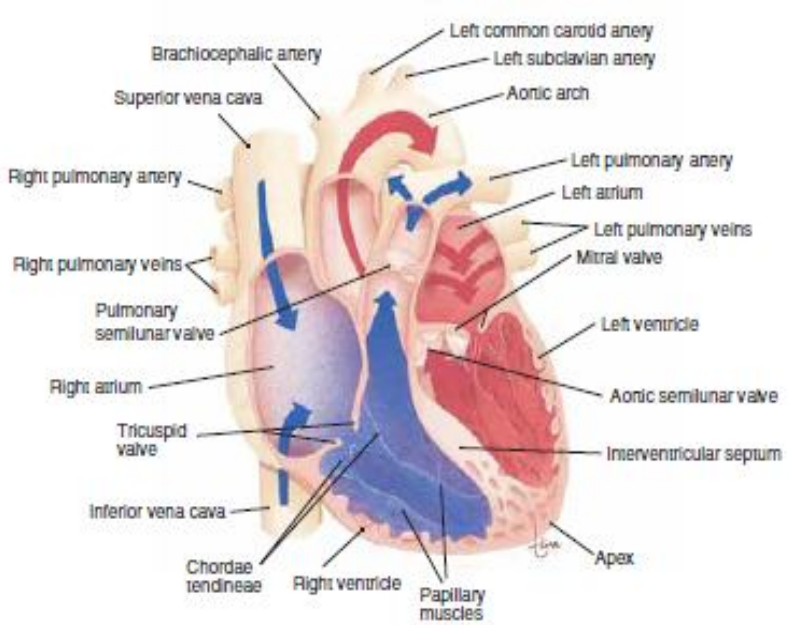


Fig. 12.1: Frontal section of the heart in anterior view, showing internal structures

CARDIAC OUTPUT –

It is defined as the quantity of blood pumped by heart in one minute. Cardiac output depends on –

1- Venous return – Amount of blood returned to the heart through veins.

2- Force of heart contraction
3- Peripheral resistance offered by blood vessels.

BLOOD PRESSURE –

The hydrostatic pressure exerted by blood on the walls of a blood vessel. Blood pressure generates due to contraction of ventricles.

- **Systolic blood pressure–** It is maximum pressure recorded during ventricular systole.
- **Diastolic blood pressure–** It maximum pressure recorded during ventricular diastole.

Factors affecting blood pressure -

1. Blood volume
2. Cardiac output
3. Peripheral resistance
4. Elasticity of blood vessels
5. Diameter of lumen of blood vessels

12.3 Cardiovascular Diseases

Cardiovascular diseases can be classified as followings-

- Disorders of Heart
- Disorders of blood vessels
- Disorders of blood pressure

12.3.1 Disorders of Heart

1-Heart Failure

2-Ischemic heart disease

- Myocardial Infarction
- Angina Pectoris

3-Cardiac arrhythmias

4-Congenital heart disease

5-Infective heart disease (Endocarditis)

1-HEART FAILURE

It is defined as impairment of cardiac output (function) so that heart is unable to maintain adequate circulation of blood to meet body's metabolic need.

Cardiac output depends upon –

- Preload
- Afterload
- Strength of myocardial muscle.

Types of heart failure –

1-Systolic and diastolic heart failure
2-Acute and chronic heart failure
3-Left sided and right sided heart failure
4-Forward and backward heart failure
5-High output and low output heart failure

Etiology-

1-Intrinsic pump Failure
2-Increased work load
3-Impaired Filling of heart

2-ISCHEMIC HEART DISEASE

Ischemic heart disease (Coronary arterial disease) is defined as the cardiac disability due to imbalance between demand and supply of oxygenated blood.
It is a syndrome consisting of-

- Myocardial Infarction
- Angina pectoris
- Chronic heart disease with heart failure
- Sudden cardiac death

MYOCARDIAL INFARCTION-

It is defined as ischemic necrosis of the myocardium due to sudden occlusion of branch of coronary artery. An acute thrombus at the site of atherosclerotic obstruction is common cause. The term heart attack used alternatively.

ANGINA PECTORIS –

It is a pain syndrome due to an imbalance between o_2 supply and oxygen demand in a portion of myocardium.

Types of angina pectoris –

Stable Angina (Classical)

It occurs during exercise due to increased demand of oxygen.

Prinzenetals angina (Variant) –

It occurs at rest due to spasm of coronary arteries.

Unstable Angina –

Repeated progressive episodes of angina occur, at rest or increased frequency and duration of anginal attack. In most cases, it is common due to rupture of atheromatous plaque and platelet aggregation in coronary artery, leading to progressive thrombosis.

O_2 Supply decreases

- Coronary atherosclerosis
- Coronary vasospasm
- Coronary thrombosis

O_2 Supply increases

- Increases heart rate
- Ventricular contractility
- Ventricular hypertrophy
- Ventricular wall tension

ARRHYTHMIAS

Arrhythmias are irregularities in cardiac rhythm.

Arrhythmias arises due to-

a. **Delayed after depolarization**- It is caused by an inward current associated with abnormally raised intracellular Ca^{+2}, which trigger ectopic beats.
b. **Re-entry –** It is occur due partial conduction block. Re–entry facilitated when parts of the myocardium are depolarized, conduction then depending on slow Ca^{+2}current.
c. **Ectopic pacemaker activity -** In arrhythmias, pacemaker activity can arise in other part of heart than SA node & conducting tissue. It is encouraged by sympathetic activity.
d. **Heart block –** It is result from damage to the atrioventricular node or ventricular conducting system.

Clinically arrhythmias divided as –

On the basis of site of origin

- Supra ventricular
- Ventricular

On the basis of heart rate

Tachycardia– It means increased heart rate, due to either increased automaticity, after depolarization or re-entry of an impulse.

Bradycardia– It means decreased heart rate, due to reduced automaticity or abnormal slowing/blockade of impulse conduction.

CONGENITAL HEART DISEASE-

These are present since birth and accounts for 0.5% of newborn children. It is attributed to multifactorial inheritance involving genetic and environmental factors like rubella drugs alcohol etc.

INFECTIVE ENDOCARDITIS

It is defined as invasion of heart valve or endocardium by bacteria.

Types-

- Acute endocarditis
- Chronic endocarditis

12.3.2.1 Disorders of blood vessels-

ATHEROSCLEROSIS-

It means hardening of arteries. It is slowly progressive vascular disease, characterized by deposition of fibro fatty plaque in the inner layer of medium or large sized muscular arteries. It is composed of blood, calcium deposits, cholesterol esters, smooth muscle cells and covering of Fibrous plaques. Its growth causes a reduction in blood flow and weakening of affected areas of vessel wall.

EMBOLUS –

It is a clot moving in circulation and become impacted in small vessels.

THROMBUS –

It is a clot obstructing a blood vessel at the point where it is actually formed.

12.3.4 Disorders of blood pressure

A-HYPERTENSION –

It is a very common disorder, affecting worldwide population. It increases the risk for various other cardiovascular diseases. Hypertension is defined as abnormally higher blood pressure in arteries.

Category	Blood pressure (mmHg)

	Systolic	Diastolic
Normal	<120	<80
Pre hypertension	120-139	80-89
Hypertension (Mild)	140-159	90-99
Moderate	160-179	100-109
Severe	180-209	110-119
Emergency	210<	120<

Types –

Primary/Essential hypertension- 90% to 95% of all cases of hypertension is primary hypertension, which is a persistently elevated blood pressure that cannot be attributed identifiable cause.

Secondary hypertension– The remaining 5-10% of cases is secondary hypertension, which has an identifiable underlying cause. Several disorders cause secondary hypertension.

1-Obstruction of renal blood Flow – Damage of renel tissue may cause the kidneys to release excessive amounts of renin in to the blood. The resulting high level of angiotensin II causes vasoconstriction, thus increasing systemic vascular resistance.

2-Hypersecretion of aldosterone– It stimulates excessive re absorption of salts and water by kidneys, which increases the volume of body fluids.

3-Hypersecretion of epinephrine and nor epinephrine – These increase heart rate, contractility and systemic vascular resistance.

B-HYPOTENSION –

It is a defined as the lowering of blood pressure. It is occur as a complication of other diseases- shock, Myocardial infarction etc. Hypotension leads to inadequate blood supply to brain. It may produce brief unconsciousness (Fainting). If this is prolonged death may occur.

12.4 Drugs inhibitors of peripheral sympathetic Function

Drugs inhibitors of peripheral sympathetic Function

1 Ganglionic blocking drugs
2 Adrenergic neuronal blockers
3 Adrenergic receptor blockers
   - α- Adrenoceptor blockers

- β- Adrenoceptor blockers
- α + β- Adrenoceptor blockers

1-GANGLIONIC BLOCKING DRUGS

These agents act at nicotinic receptors (N_N type) of autonomic ganglia and block both parasympathetic and sympathetic ganglia and produce complex effects.

Classification –

A. Competitive blockers

1-Quaternary ammonium compounds

- Hexamethonium
- Pentolinium

2-Amines (secondary/tertiary)

- Mecamylamine
- Pempidine

3-Monosulfonium compound

- Trimethaphan

B. Persistent depolarizing blockers

- Nicotine (large dose)
- Anticholinesterases (large dose)

Uses – They were used for hypertension and peptic ulcer but now totally replaced due to intolerable side effects. There is at present no clinical relevance of Ganglionic blockers. Trimethaphan occasionally used to produce controlled hypotension and in hypertensive emergency due to aortic dissection.

2-ADRENERGIC NEURONAL BLOCKERS

Reserpine –

It is an alkaloid extracted from root of Rauwolfia serpentina. Reserpine enters adrenergic neuron, binds to the intra neuronal vesicles which store monoamines (NA, 5-HT, DA) and inhibits the vesicular catecholamine uptake and storage. The monoamines are leak in to cytoplasm and destroyed MAO enzyme.

Uses –

Antihypertensive effect is due to depletion of biogenic amines.

Mild antipsychotic effect is due to DA depletion.

Guanethidine-

It is a polar guanidine compound which is taken up into the adrenergic nerve endings by active amine transport, and has three important facets of action:

a. Displaces NA from storage granules.
b. Inhibits nerve impulse coupled release of NA.
c. Engages and blocks NA uptake mechanism at the axonal membrane.

Uses –

It has gone out of use now due to marked side effects.

3-ADRENERGIC RECEPTOR BLOCKERS

- **α- ADRENOCEPTOR BLOCKERS**

These agents block receptors, thus inhibiting receptors mediated responses of sympathetic stimulation and adrenergic agonists.

Classification –

(I) Nonselective ($\alpha_1$and α_2 blockers)-

(A)Reversible-

- Phentolamine
- Tolazoline

(B)Irreresible-

- Phenoxybenzamine

(II) Selective α -receptors blockers –

(A) α_1-Blockers –

- Prazocin
- Terazcin
- Doxazocin

(B) α_2-Blockers -

- Yohimbine

Phentolamine –

It is an imidazoline derivative. It competitively blocks the effect of noradrenaline at adrenergic receptors.

Tolazaline –

It is similar to phentolamine and is rarely used.

Phenoxybenzamine –

It cyclizes spontaneously in the body produce a highly reactive intermediate ethyleniminium which covalently binds to α-receptors and cause irreversible blockade. It also inhibits the reuptake of NA into adrenergic nerve endings.

Prazocin –

It is potent and highly selective α_1-adrenergic blocker. Prazocin block sympathetically mediated vasoconstriction and produce fall in BP. It also inhibits phophodiesterase which degrades cyclic AMP. Rise in smooth muscle cyclic AMP could contribute to its vasodilator action.

Terazocin –

It is chemically and pharmacologically similar to prazocin but due to longer duration of action, only daily single dose required.

Doxazocin –

It is longer acting congener of prazocin with similar pharmacological activity.

Yohimbine –

Yohimbine is an alkaloid. It is competitively blocks α_2 receptors. It is rarely used clinically.

Uses of α -blockers –

- Pheochromocytoma
- Hypertension
- Benign prostatic hypertrophy
- Secondary shock
- Peripheral vascular disease
- Congestive heart failure
- Male sexual dysfunction

β- ADRENOCEPTOR BLOCKERS

These drugs blocks adrenergic responses or sympathetic stimulation mediated by β- receptors.

Classification –

Nonselective –

- Propranolol
- Sotalol

- Timolol
- Pindolol

β_1- Selective (Cardio selective)-

- Atenolol
- Metoprolol
- Bisoprolol
- Esmolol
- Celiprolol
- Nebivolol

Actions of β – blockers –

Propranolol is the prototype drug.

Cardiovascular system-

HEART-

- Decrease heart rate
- Decrease myocardial contractility Force
- Decrease cardiac output
- Decrease automaticity of ectopic foci
- Decrease cardiac work so that reduce O_2 demand of myocardium.

BLOOD VESSELS – Continuous administration leads to a decrease peripheral vascular resistance in hypertensive patients, reduce blood pressure.

Pindolol –

It has intrinsic sympathomimetic activity (Partial agonistic activity), Stimulate β - receptors partially in the absence of catecholamine's. It also has membrane stabilizing activity (local anaesthetic activity).

Esmolol –

Selective β - blocker and has short duration of action. It has no membrane stabilizing effect. It is poorly lipid soluble.

Atenolol –

- No membrane stabilizing effect
- Longer duration of action
- More potent

- Poorly Lipid soluble

Therapeutic uses of **β**- blockers –

- Hypertension
- Angina pectoris and Myocardial Infarction
- Cardiac arrhythmias
- Congestive heart Failure
- Pheochromocytoma
- Hypertrophic obstructive cardiomyopathy

α + β -ADRENOCEPTOR BLOCKERS

These agents are capable of blocking both **α** and **β** receptors.

Lobetalol –

- Competitive blocker at β_1 β_2 and α_1 adrenergic receptors.
- Partial agonistic activity at β_2 receptors
- times more potent in blocking **β** than **α**
- Moderately potent hypotensive and is especially useful in pheochromocytoma and essential hypertension

Carvidilol–

- It also block β_1 β_2 and α_1 -adrenergic receptors
- No intrinsic sympathomimetic activity
- Antioxidant, membrane stabilizing and vasodilator activity
- Used as cardio protective in CHF.

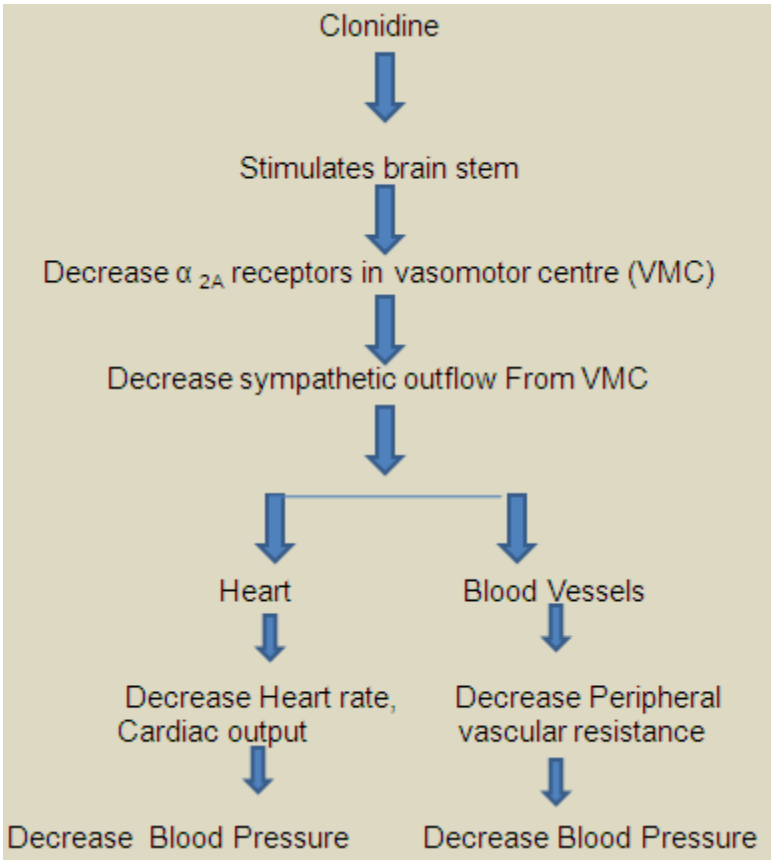
12.5 Central intervention of Cardiovascular Output

These are centrally acting sympatholytics which blocks adrenergic responses.

CLONIDINE –

It is an imidazoline derivative.

Mechanism of action –

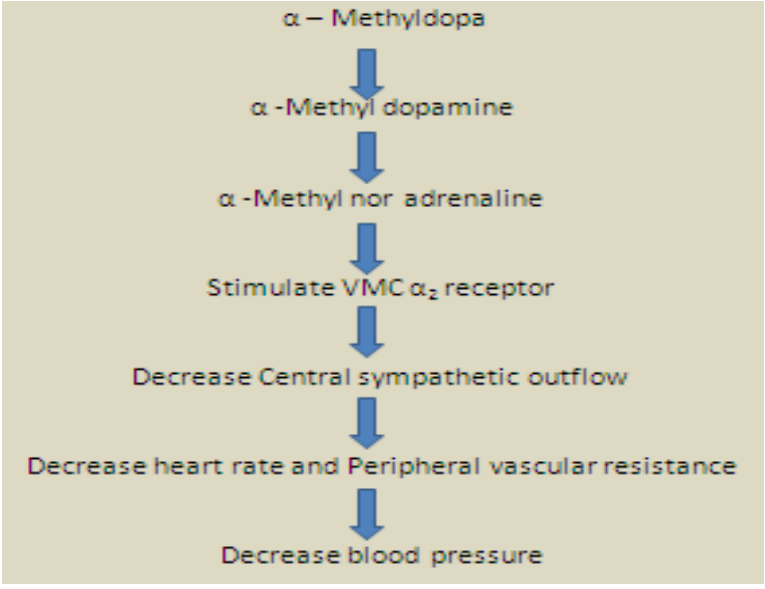


Uses - Clonidine occasionally used with a diuretic in combination for hypertension due to frequent side effect.

METHYLDOPA –

It is the **α** -methyl analogue of dopa, the precursor of DA and NA. The **α** -methyl - NA formed in the brain from methyldopa acts on central α_2 receptors to decrease efferent sympathetic activity.

Mechanism of action –



Uses – Methyldopa used to treat hypertension during pregnancy.

12.6 Directly Acting Arteriolar Dilators

These drugs directly produce vasodilatory effect.

Minoxidil

Diazoxide

Hydralazine

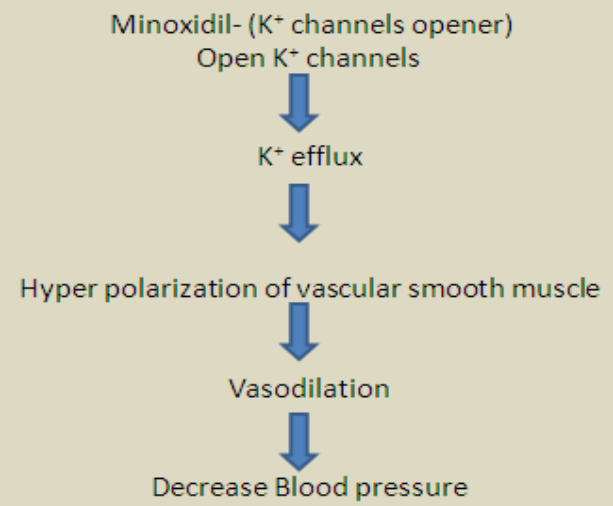
Sodium nitroprusside

MINOXIDIL-

It is a powerful arteriolar dilator

Minoxidil is a prodrug converted to an active metabolite by sulfate conjugation.

Mechanism of action-



Uses -

1-It is used in combination with B-blockers or diuretics.

2-Topically used to male pattern boldness.

DIAZOXIDE

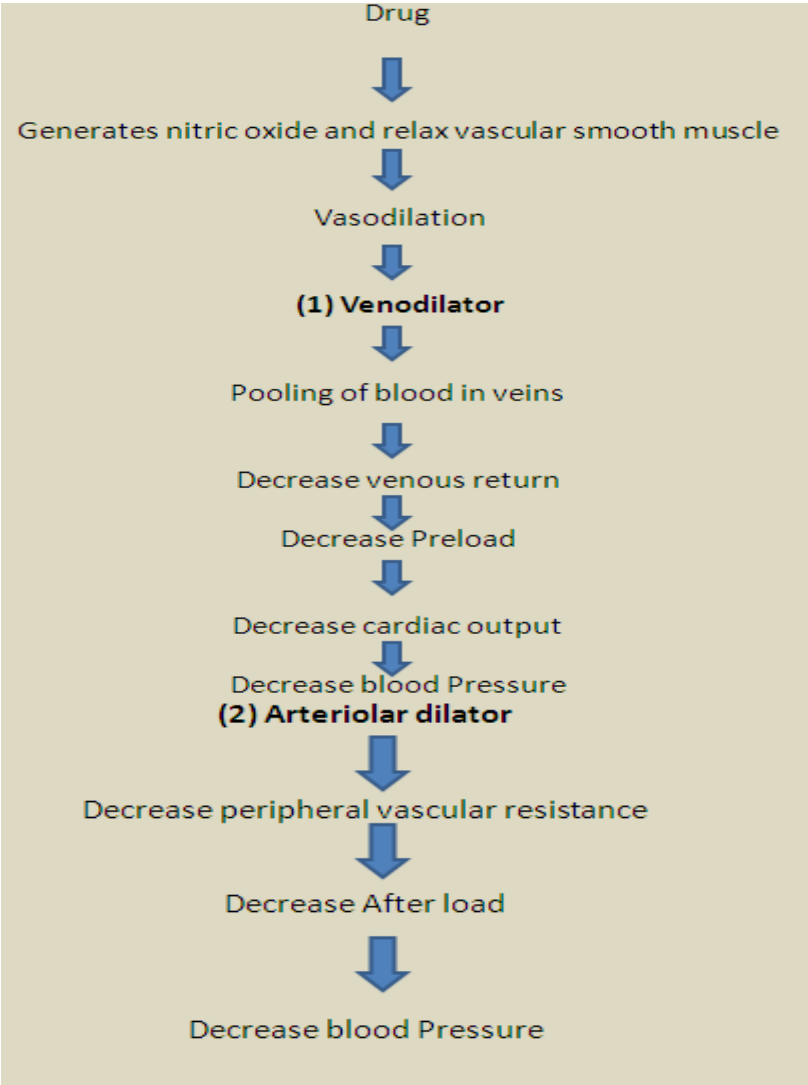
- It is K^+ Channel opener arteriolar dilator
- Mechanism of action similar to minoxidil

Uses - Hypertensive emergencies.

SODIUM NITROPRUSSIDE-

It is a powerful arteriolar and venodilator.

Mechanism of action-



HYDRALAZINE-

It is a directly acting arteriolar vasodilator with little action on venous capacitance vessels, reduces total peripheral resistance. It causes greater reduction of diastolic than systolic BP.

Uses-

It is not used alone. Large doses are not recommended for long periods.

It is used in moderate-to-severe hypertension not controlled by the first line drugs.

Usually, low doses are added to the diuretic and □-blocker already being administered.

12.7 Clotting Prevention and Lysis

Haemostasis and blood coagulation involve complex interactions between the injured vessel wall, platelets and coagulation factors.

ANTICOAGULANTS

These are drugs used to reduce the coagulability of blood. They may be:

- Heparin
- Heparinoids
- Danaparoid
- Lepirudin
- Bishydroxycoumarin (dicumarol)
- Warfarin sodium
- Acenocoumarol (Nicoumalone)
- Ethylbiscoumacetate
- Phenindione

Uses-

- Deep vein thrombosis and pulmonary embolism
- Myocardial infarction (MI)
- Atrial fibrillation (AF)

FIBRINOLYTICS (Thrombolytics)

These are drugs used to lyse thrombi/clot to recanalize occluded blood vessels (mainly coronary artery). They are work by activating the natural fibrinolytic system.

The clinically important fibrinolytics are:

- Streptokinase
- Urokinase
- Alteplase (rt-PA)
- Reteplase
- Tenecteplase

Uses-

- Acute myocardial infarction

- Deep vein thrombosis
- Pulmonary embolism
- Peripheral arterial occlusion

ANTIPLATELET DRUGS (Antithrombotic drugs)

These are drugs which interfere with platelet function and are useful in the prophylaxis of thromboembolic disorders.

The clinically important antiplatelet drugs are:

- Aspirin
- Clopidogrel
- Dipyridamole
- Abciximab
- Ticlopidine

12.8 Summary

The Cardiovascular system is a transport system which consists of heart, blood and blood vessels. Various cardiovascular diseases disturb hearts normal function. These disorders may be related to cardiac output, blood vessels and blood pressure. These consist of different type of factors and causes. Many types of drugs are used to correct conditions in which heart not work properly such as- peripheral and central sympathetic function inhibitors, directly acting arteriolar dilator and agents that prevent clotting and produce lysis of blood clots.

12.11 Glossary

- Afterload**: -** Systemic peripheral resistance or pulmonary circulation resistance.
- Aldosterone: - The most biologically active mineralocorticoid hormone secreted by adrenal cortex.
- Atrial Fibrillation (AF): - AF is an irregular and often rapid heart rate that commonly causes poor blood flow to the body.
- Benign Prostatic Hypertrophy: - BPH also called benign enlargement of the prostate. It involves hyperplasia of prosthetic cells resulting in the formation of large nodules in the transition zone of the prostate.

- Cardiac Rhythm: - The predominant electrical activity of the heart.
- Haemostasis: - Arrest of blood loss
- Pheochromocytoma: - It is a tumour or adrenal medulla which release excessive adrenaline and noradrenaline. This increase concentration of catecholamines can cause intermittent or persistant hypertension.
- Preload - Volume and pressure of blood in ventricles before systolic contraction of heart.
- Thrombosis: - Thrombosis is the formation of a blood clot inside a blood vessel obstructing the flow of blood.
- Vasospasm: - It is refers to a condition in which a blood vessels spasm leads to vasoconstriction.

12.12 Review questions / Comprehensive Questions

1. What is hypertension? Describe its types.
2. Explain the mechanism of action of followings-
   A. Minoxidil
   B. Diazoxide
   C. Hydralazine
   D. Sodium nitroprusside
3. Write short note on-
   A. □- adrenoceptor blockers
   B. □- adrenoceptor blockers
   C. □ + □- adrenoceptor blockers
4. Enlist cardiac disorders Explain cardiac arrhythmias.
5. What is ischemic heart disease? Explain myocardial infarction and angina pectoris.

12.13 References and Suggested readings

- Essential of Medical Pharmacology (7th ed.)- KD Tripathi (JAYPEE Publisher) 2013.
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Unit - 13

Synthesis of Cardiovascular Drugs

Structure of Unit:

13.1 Objectives

- In this chapter we discuss the drugs which are used for different type of cardiac disorder.
- Here we study about the method of synthesis of different cardio vascular drugs.

13.2 Introduction: Cardiovascular Drugs

Cardiovascular drugs are the group, which have major action on the heart or blood vessels, or those used primarily for cardiovascular disorders, so that they check the total output of the heart as well as the distribution of blood to certain parts of the circulatory system. Amyl nitrate, sorbitrate, diltiazem, quinidine, verapamil, methyldopa, atenolol and oxyprenolol are some important cardiovascular drugs, which can be synthesized in the following manner:

13.3 Synthesis of Amyl nitrate:-

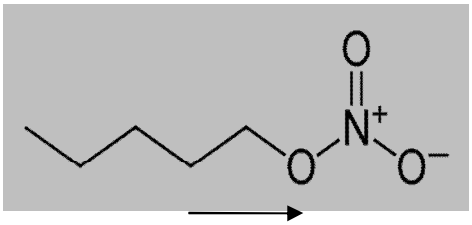
Amyl nitrate have been found effective in both relieving and preventing the painful angina attacks. Although it is short acting antianginal agent, when oxygen is insufficient to meet the myocardial work load, which can occur due to vasospasm of the coronary vessels. The amyl nitrate induces the vasodilation of the coronary vessels.

The most prominent action of this drug is exerted on vascular smooth muscle. Nitrate dilates veins more than arteries which cause peripheral pooling of blood, this reduces the preload on heart and diastolic size and pressure therefore decrease cardiac work load.

Alkyl nitrates are employed as reagents in organic synthesis. Amyl nitrate is used as an additive in diesel fuel, where it acts as an 'ignition improver' by accelerating the ignition of fuel

Structure: - Amyl nitrate is the chemical compound with the formula $CH_3(CH_2)_4ONO_2$. This molecule consists of the 5-carbon amyl group attached to a nitrate functional group. It is the ester of amyl alcohol and nitric acid.

Synthesis:-



$C_5H_{11}OH$ + $NaNO_3$ + H_2SO_4

Amyl alcohol Sod. nitrate Amyl Nitrate

The amyl nitrate can be prepared by esterification of simple amyl alcohol with nitric acid or sodium nitrate in conc. Sulphuric acid. In the synthesis of amyl nitrate, the amyl alcohol is react with sodium nitrate in presence of sulphuric acid and the reaction give amyl nitrate, sodium thiosulphate and water molecule as final product.

This drug can be hydrolysed easily; therefore moisture should be avoided on storage.

13.4 Synthesis of Diltiazem:-

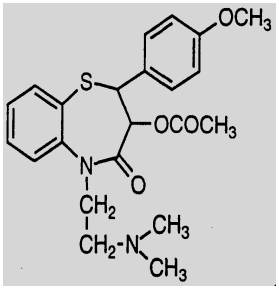
Diltiazem is in a group of drugs called calcium channel blockers. It works by relaxing the muscles of your heart and blood vessels. Diltiazem is used to treat hypertension (high blood pressure), angina (chest pain), and certain heart rhythm disorders.

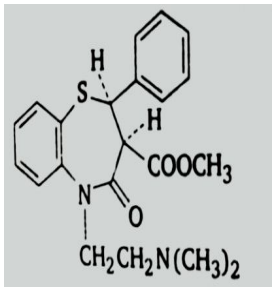
Diltiazem is a calcium channel blocker for the treatment of myocardial insufficiency and angina pain, Ca^{++} (calcium ion) is inhibited to influx into

myocardial cells. Since 1960, calcium ions are known to play a critical role in many physiological functions. Physiologic calcium is found in a variety of location such as intracellular and extracellular, both.

Diltiazem interfere the movement of Ca^{++} ions into the cell.

Structure:-





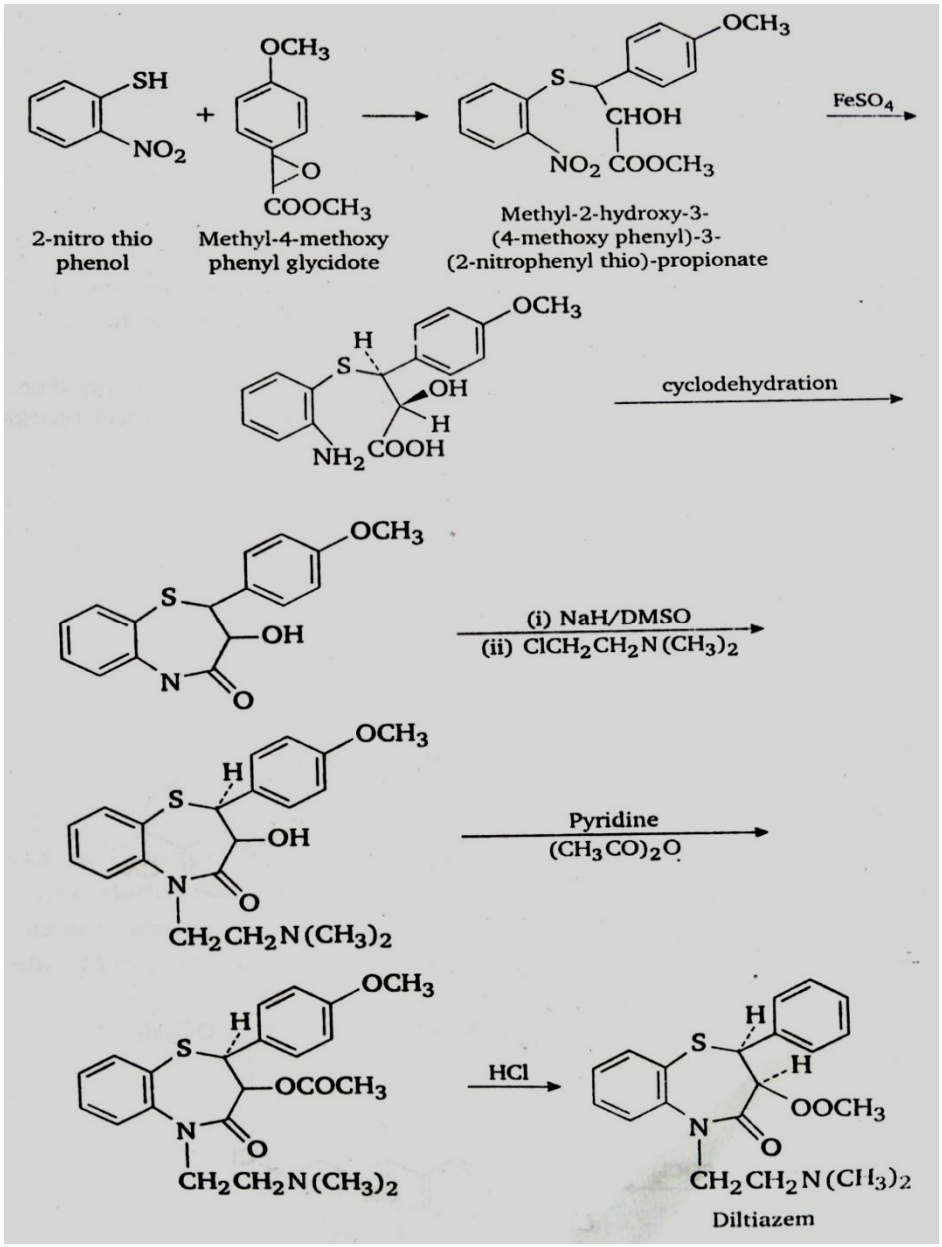
Do not use diltiazem if you have certain heart conditions such as "sick sinus syndrome" or "AV block" (unless you have a pacemaker), low blood pressure, or if you have recently had a heart attack.

Diltiazem may impair your thinking or reactions. Be careful if you drive or do anything that requires you to be alert. Do not stop taking this medication without first talking to your doctor. If you stop taking diltiazem suddenly, your condition may become worse.

Diltiazem may be only part of a complete program of treatment that also includes diet, exercise, and other medications. Follow your diet, medication, and exercise routines very closely.

If you are being treated for high blood pressure, keep using diltiazem even if you feel well. High blood pressure often has no symptoms.

Synthesis: - Diltiazem can be prepared in the following way:



Before taking this medicine:-

Should not use this medication if allergic to diltiazem or if have:

- certain heart conditions, especially "sick sinus syndrome" or "AV block" (unless you have a pacemaker);
- low blood pressure; or
- If recently had a heart attack.

Uses- Diltiazem is used to prevent chest pain (angina). It may help to increase your ability to exercise and decrease how often you may get angina attacks. Diltiazem is called a calcium channel blocker. It works by relaxing blood vessels in the body and heart and lowers the heart rate. Blood can flow more easily and your heart works less hard to pump blood.

Diltiazem may also be used to control your heart rate if you have a fast/irregular heartbeat (such as atrial fibrillation).

13.5 Synthesis of Verapamil:-

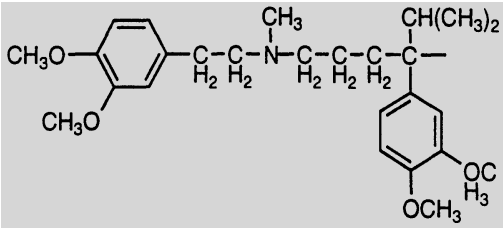
Verapamil is a calcium channel blocker. It works by relaxing the muscles of your heart and blood vessels.

It is a calcium channel blocker which is used in the treatment of angina. Verapamil is also effective and widely used for the treatment of superaventricular arrhythmias chemically; it is a derivative of papaverine and was originally conceived as vasodilator for angina.

After oral administration, verapamil is well absorbed and is metabolized to a great extent in liver (first –pass effect) so that only about 20% of an oral dose is bioavailable. This first pass effect disappears in prolonged use.

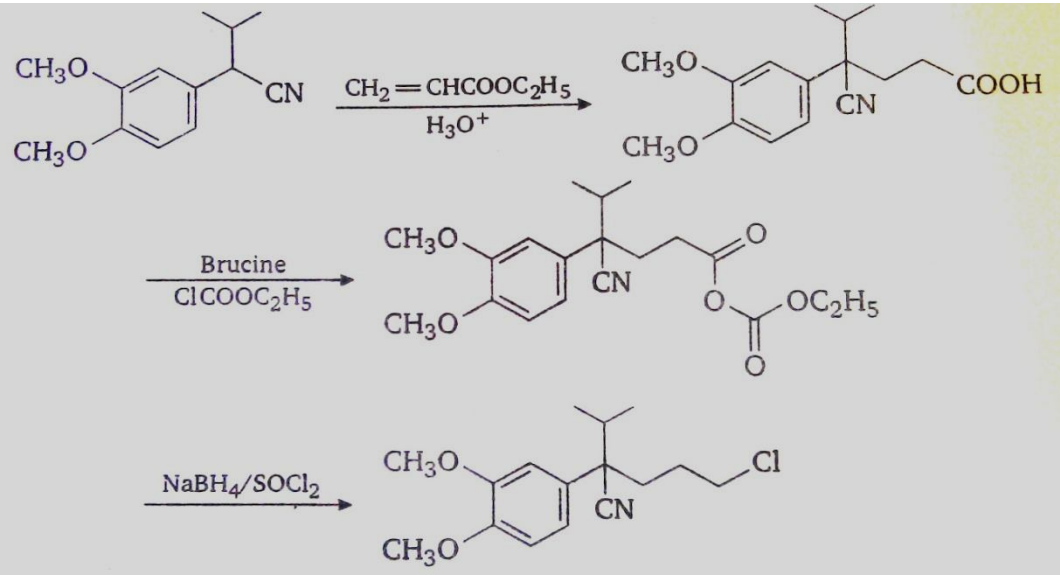
One of the major metabolite of verapamil is norverapamil. The therapeutic effects of verapamil after oral administration are observed with 2 hours.

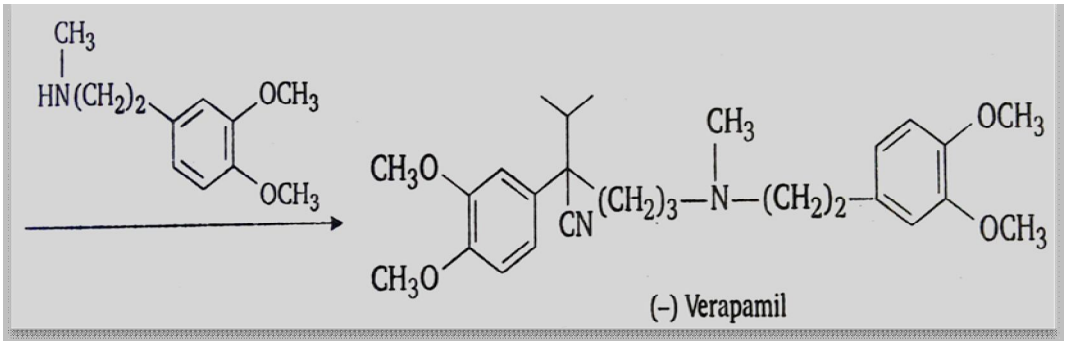
Structure:-



Verapamil is a phenylalkylamine derivative, consisting a chiral *i.e.*, one asymmetric centre. Its L-isomer is more potent as a calcium channel blocker than the other enantiomer.

Synthesis: - verapamil can be prepared as follows:





Verapamil is used to treat hypertension (high blood pressure), angina (chest pain), and certain heart rhythm disorders

Before taking this medicine:-

Should not use verapamil if allergic to it, or if have a serious heart condition such as:

- "Sick sinus syndrome" or "AV block" (unless you have a pacemaker);
- Severe heart failure;
- Slow heartbeats that have caused you to faint; or
- Certain heart rhythm disorders of the atrium (the upper chambers of the heart that allow blood to flow into the heart).

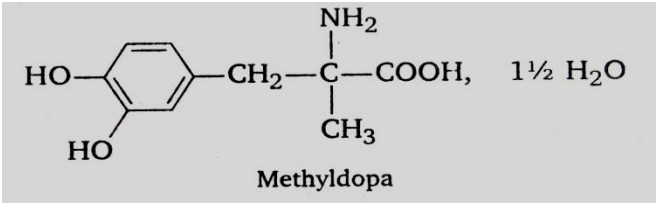
13.6 Synthesis of Methyldopa: -

It is the alpha-methyl analogue of dopa, which is a precursor of dopamine. It has been one of the oldest and most widely used antihypertensive agents. It is alpha methyl derivative of dopa (3, 4-dihydroxy phenyl alanine). The chemical name of methyl dopa is (-)-3,-(3, 4-dihydroxyphenyl)-2-methyl-L-alanine sesqui hydrate.

It is supposed that methyldopa shows its hypotensive effect within the central nervous system by its conversion to a metabolite called alpha-methyl nor-adrenaline which acts as a potent O_2 –adrenergic agonist, this may result in a decrease in sympathetic out flow from the CNS. Methyldopa is used in the treatment of moderate to severe hypertension. It may be administered as tablets.

Methyldopa is an alpha-2 receptor agonist. It reduces elevated blood pressure by relaxing and dilating (widening) blood vessels. Blood flows more freely and at a lower pressure through dilated blood vessels.

Structure:-



When the blood transfers from arteries to the tissue capillaries and veins, it results in excessive stimulation of sympathetic nervous system (CNS). This may cause arteriole contraction and increased peripheral resistance to the flow of blood. As a result, blood pressure increases. Methyldopa is used for contracting this condition i.e., reducing blood pressure and its accompanying symptoms.

Treating high blood pressure, it may be used with other high blood pressure medicines.

Do not use methyldopa if:

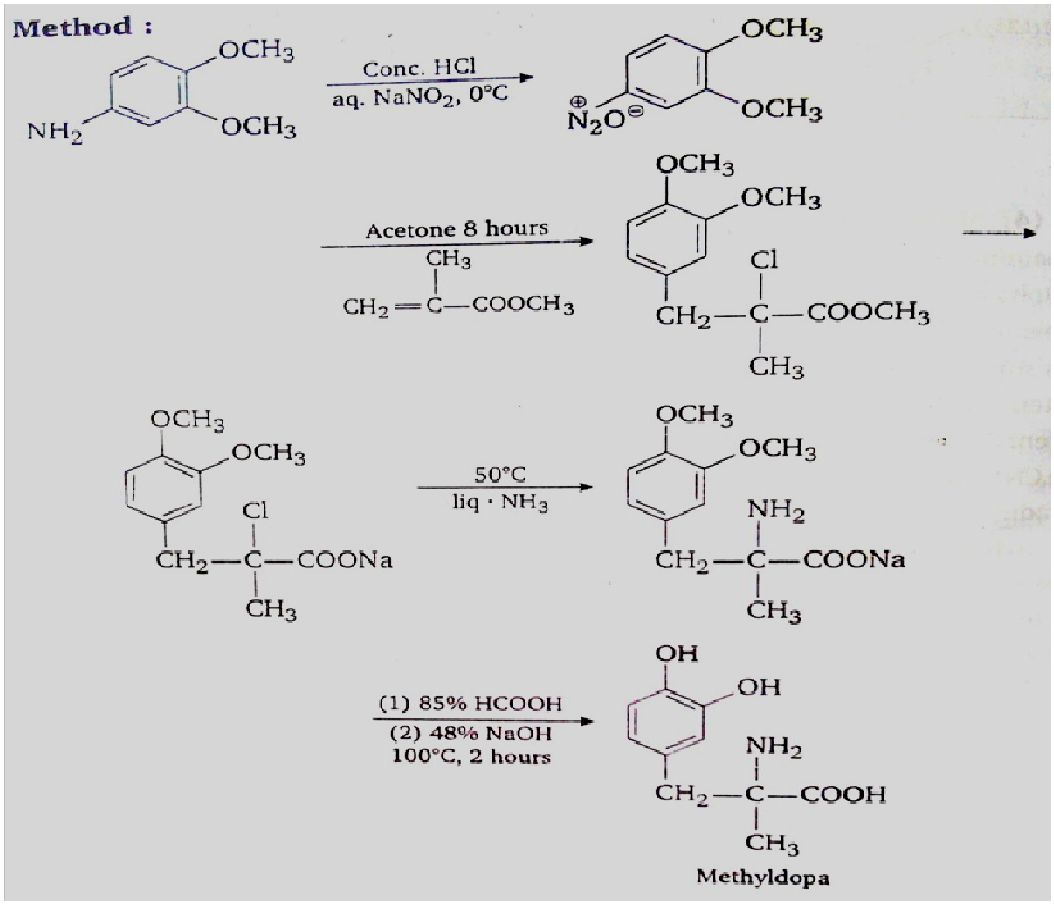
- Allergic to any ingredient in methyldopa
- Have liver disease, severe kidney problems, a liver reaction caused by past use of methyldopa, or a history of anemia caused by immune system
- Receiving enteral feedings
- Taking a monoamine oxidase inhibitor (MAOI) (eg. phenelzine)

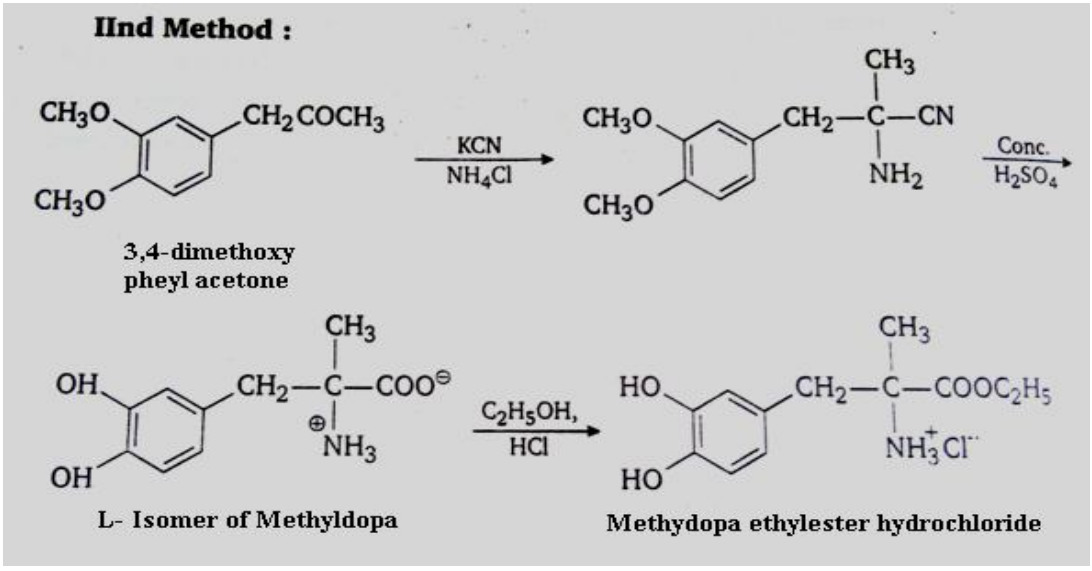
Before using methyldopa:

Some medical conditions may interact with methyldopa. Tell your doctor or pharmacist if you have any medical conditions, especially if any of the following apply to you:

- If pregnant, planning to become pregnant, or are breast-feeding
- If taking any prescription or nonprescription medicine, herbal preparation, or dietary supplement
- If have allergies to medicines, foods, or other substances
- If have hemolytic anemia or other blood problems, liver problems, kidney problems, or tumors on your adrenal glands

Synthesis: - Methyldopa can be prepared as given below:

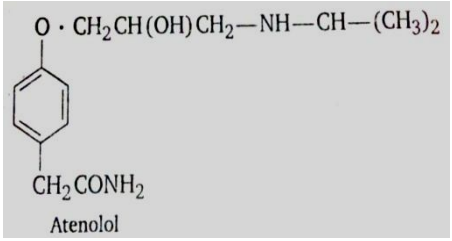




13.7 Synthesis of Atenolol:-

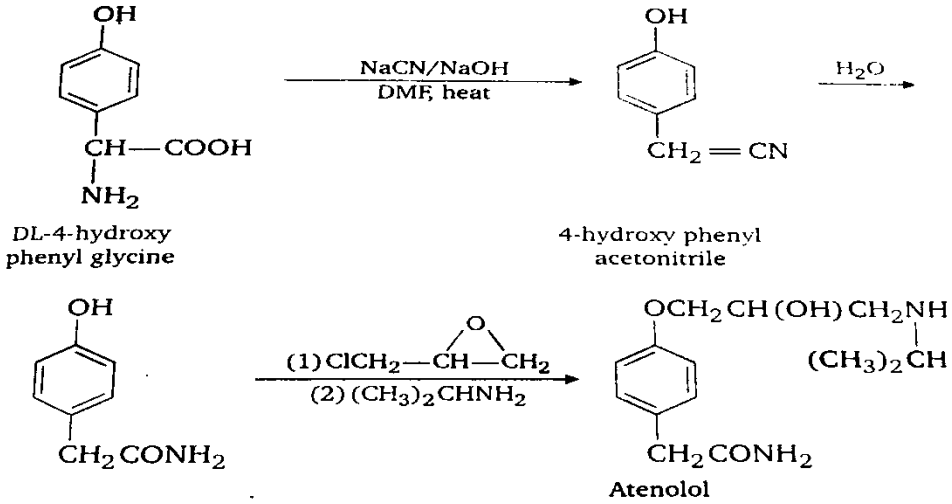
Atenolol is anti arrhythmic agent and known as beta-adrenoreceptor blocking agent. Its half life is 6 to 9 hours. Since atenolol is hydrophilic in nature, therefore, very little quantity of this drug penetrates into the brain.

Structure:-



Atenolol

Synthesis:-



Atenolol

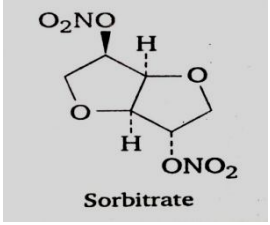
Uses- its principle effect has been to reduce cardiac activity by decreasing or preventing beta-adrenoreceptor stimulation. This drug also finds use in the treatment of angina pectoris for reducing the oxygen consumption and for increasing the exercise tolerance of heart. Atenolol is also useful in the treatment of hypertension.

13.8 Synthesis of Sorbitrate

This drug is also known as isosorbide dinitrate. Sorbitrate is used for both treatment and prevention of painful angina attacks. It is a long acting antianginal agent. This drug can be administered sub lingually i.e., transdermal and buccal administration routes. The action of sorbitratre may last for 4 to 6 hours. Sorbitrate is metabolized primarily in the liver by glutathione-nitrate reductase. This enzyme catalizes the denitration of the parent drug to yield two metabolites, 2- and 5- isosorbide mononitrate.

Sorbitrate is chemically 1,4:3,6-dianhydro-D-glucitol-2,5-dinitrate. It may get exploded when it gets subjected to percussion or excessive heat. Diluted sorbitrate has been a dry mixture of isosorbide dinitrate with lactose or some other suitable inert excipient. This allows for safe handling of this drug. This drug is also useful as adjunctive therapy in congestive heart failure.

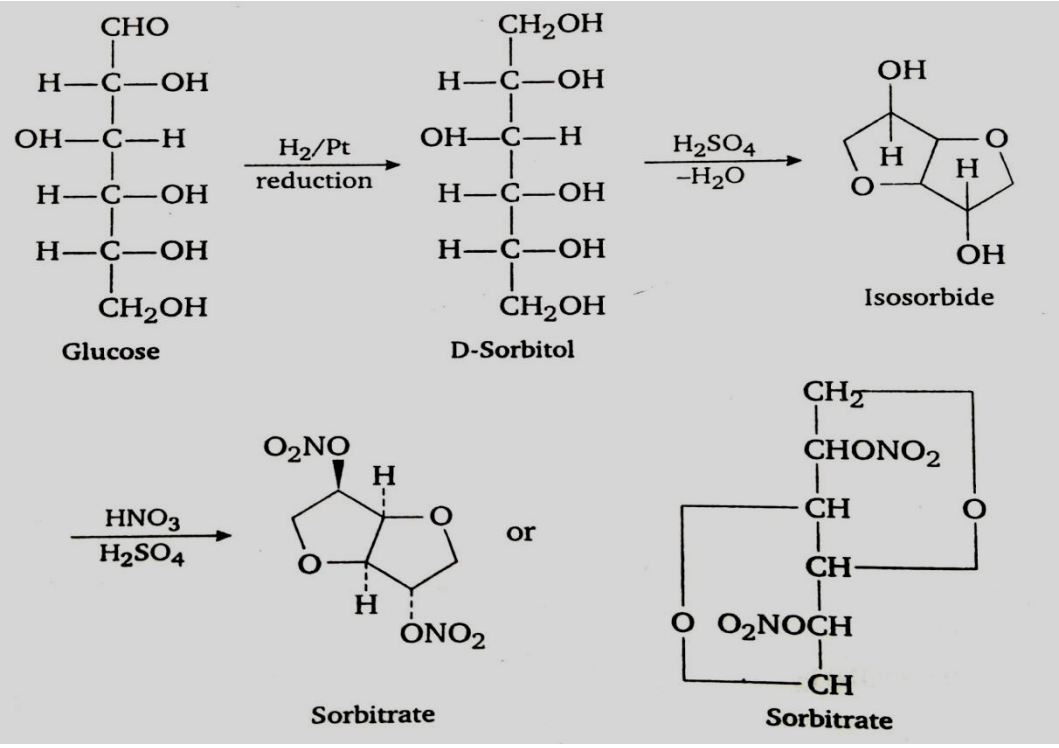
Structure:-



Sorbitrate

- Isosorbide dinitrate is in a group of drugs called nitrates.
- Isosorbide dinitrate dilates (widens) blood vessels, making it easier for blood to flow through them and easier for the heart to pump.
- Isosorbide dinitrate is used to treat or prevent attacks of chest pain (angina).
- Only the sublingual tablet should be used to treat an angina attack that has already begun.
- Isosorbide dinitrate regular and extended-release tablets are used to prevent angina attacks but will not treat an angina attack.
- Do not use isosorbide dinitrate if taking sildenafil (Viagra). Serious, life-threatening side effects can occur if take isosorbide dinitrate while you are using sildenafil.

Synthesis:- Sorbitrate can be prepared as follows-



13.9 Synthesis of Quinidine:-

Quinidine is an alkaloid, found in cinchona bark (cinchona officinalis L) and is a close relative of quinine. In fact, quinidine and quinine are diastereomers of one another.

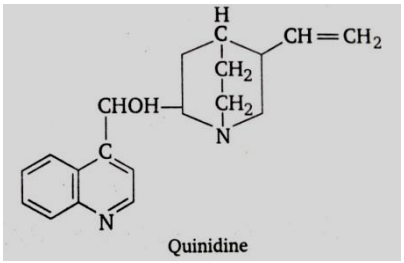
Structurally quinidine and quinine are similar, and quinidine is dextro-isomer of quinine. Despite this similarity, quinidine and quinine differ markedly in their effects on cardiac muscle. Frey (1918) found quinidine superior to quinine for this purpose.

Quinidine consists of quinoline ring and the bicyclic quinuclidine ring system with a hydroxyl methylene bridge connecting these two components. Examination of quinidine shows 2 basis nitrogen atoms. The character of quinidine is basic; hence it is always used in more water soluble salt forms. These salts are gluconate, polygalacturonate, and quinidine sulphate.

The use of intravenous quinidine is rare, however, in special situations it may be administered intravenously as the gluconate salt.

Quinidine is metabolized mainly in liver. Its renal excretion is also significant. Quinidine has some alpha-adrenergic blocking property. At higher dose it directly dilates blood vessels which cause fall in BP.

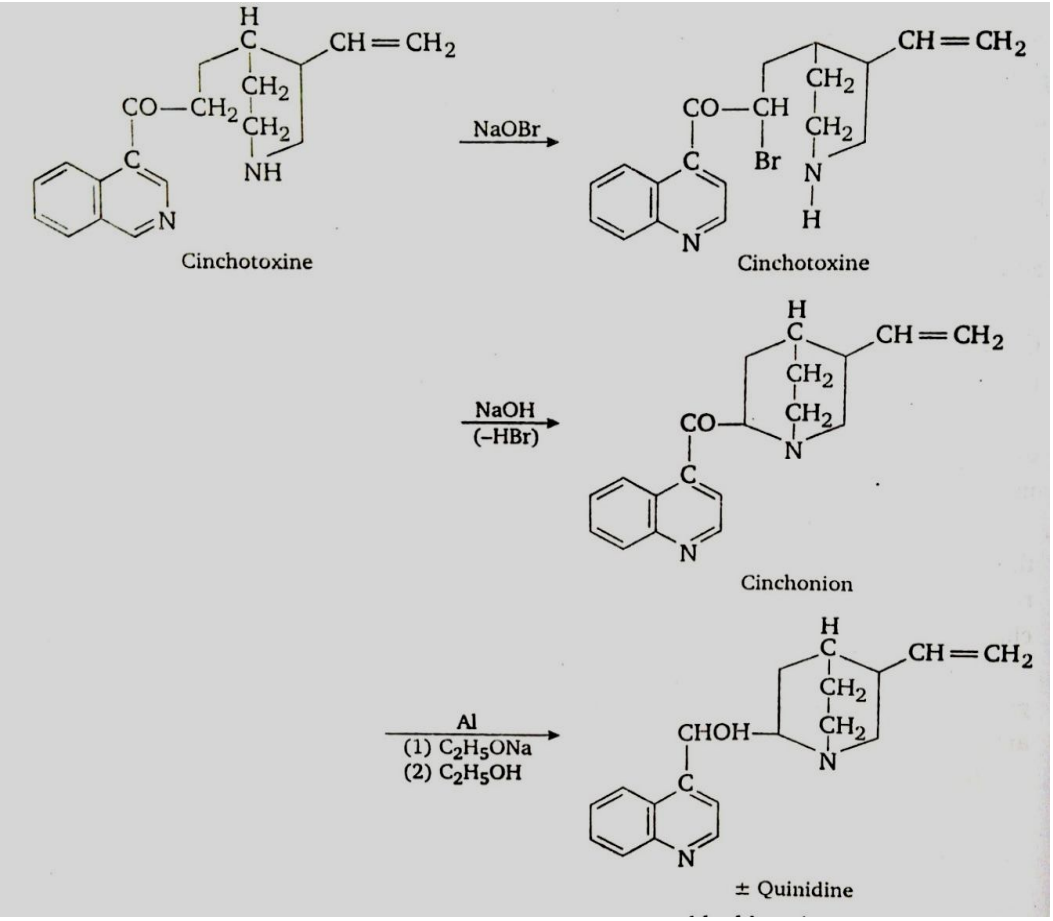
Structure:-



Quinidine

Uses- quinidine is effective in a large number of atrial and ventricular arrhythmias, but is not a preferred drug for treatment of acute arrhythmias. It is mainly used to maintain sinus rhythm.

Synthesis:-



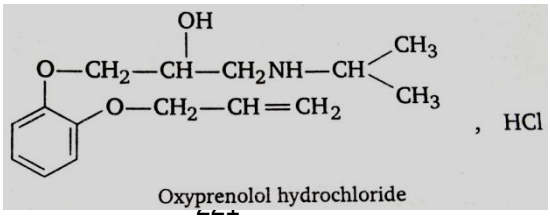
13.10 Synthesis of Oxyprenolol:-

Oxyprenolol is a beta-adrenergic blocking drug. The branch of the autonomic nervous system in which norepinephrine is the neurotransmitter between the nerve ending and the effector muscle is known as the adrenergic nervous system.

The adrenergic nervous system plays an important role in regulating many physiological functions, including blood pressure, heart rate and force, bronchial tone and gastrointestinal motility.

Oxyprenolol acts as competitive inhibitors of the effects of catecholamine at beta-receptor sites. The principal effect of this drug has been to reduce cardiac activity by decreasing or preventing beta-adrenoceptor stimulation and find use in the treatment of cardiac arrhythmias. Oxyprenolol is also useful in angina pectoris for reducing the oxygen consumption and for increasing the exercise tolerance of the heart.

Structure: -



Oxyprenolol consist of O-allyl oxyphenoxy moiety. Officially it is found as hydrochloride salt.

13.11 Summary

In this unit we have discussed the synthesis of cardiovascular drugs. During the study of this unit we synthesise the different cardiovascular agent which are used for the treatment of cardiac disorders like cardiac arrhythmia, angina pectoris, hypertension etc. Methyldopa exhibits variable absorption from the gastrointestinal tract. It is metabolized in the liver and intestines and is excreted in urine. Isosorbide dinitrate regular and extended-release tablets are used to prevent angina attacks but will not treat an angina attack. Avoid drinking alcohol, It can increase some of the side effects of isosorbide dinitrate. Atenolol is used to treat angina (chest pain) and hypertension (high blood pressure). It is also used to treat or prevent heart attack.

13.12 Glossary

- **Antianginal Agent:-** these are the drugs used in the treatment of angina pectoris (angina is a type of chest pain caused by reduced blood flow to the heart muscle).
- **Asymmetric Centre:-** An atom having a spatial arrangement of ligands which is not superimposable on its mirror image.
- **Cardiac Arrhythmias:-** An arrhythmia is an irregular heartbeat- the hear may beat too fast (tachycardia), too slowly (bradycardia), too early (premature contraction) or too regularly (fibrillation). Arrhythmias are heat-rhythm problems, they occur when the electrical impulses to the heart that coordinate heartbeats are not working properly, making the heartbeat too fast/slow or inconsistently
- **Diastereomers:-** these are stereoisomers that are not mirror images of one another and are non-superimposable on one another
- **Enantiomer:-** these are chiral molecules that are mirror images of one another.
- **Esterification: -** the process of combining an organic acid (RCOOH) with an alcohol (ROH) to form an ester (RCOOR) and water.
- **Haemolytic Anemia :-** this is a form of anemia due to hemolysis, the abnormal breakdown of red blood cells (RBCs), either in the blood

vessels (intravascular hemolysis) or elsewhere in the human body (extravascular)

- **Sick Sinus Syndrome: -** this is also known as sinus node disease or sinus node dysfunction- is the name for a group of heart rhythm problems (arrhythmias) in which the sinus node- the heart's natural pacemaker- doesn't work properly.
- **Sinus Rhythm: -** it is normal beating of the heart, as measured by an electrocardiogram (ECG).
- **Supraventricular Arrhythmias: -** this is supraventricular tachycardia which means that from time to time your heartbeats very fast for a reason other than exercise, high fever, or stress. For most people who have SVT, the heart still works normally to pump blood through the body.

13.13 Review questions /comprehensive questions:-

1. Write the preparation and use of amyl nitrate.
2. How methyldopa synthesises?
3. Give the synthesis of isosorbide dinitrate with its structure and uses.
4. How sorbitrate is synthesised from glucose?
5. Give the synthesis of quinidine from cinchotoxine with its uses.
6. Give any one synthesis of antiarryhthmic agent.
7. Write short note on oxyprenolol hydrochloride.

13.14 References and suggested readings

- Textbook of Organic Medicinal and Pharmaceutical Chemistry (11th ed.)- Wilson and Gisvold's.
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- A Book of medicinal chemistry volume I; (first edition 2008-2009) Dr. Anees Ahmad Siddiqui, Seemi Siddiqui, Dr. R. Rajesh, published by birla publication pvt. Ltd.
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Unit-14

Local Anti-infective Drugs

Structure of Unit:

14.1 Objectives

This chapter deals with drugs used to treat infections caused by bacteria, fungus, protozoan, virus and helminthes etc We give first some necessary information about bacteria, fungus, protozoan, virus and helminthes etc. after then we describe the various types of drugs and their general mode of action.

14.2 Introduction: Local Anti-infective Drugs

The anti-infective drugs act against the infections by bacteria, fungus, protozoan, virus and helminthes etc. drugs of this class differ from all others in that they are designed to inhibit/kill the infecting organism and to have no or minimal effect on the recipient or host. Local anti-infective drugs can be classified in many ways:

- Antifungal Drugs- Antifungal drugs are used for deep systemic and superficial fungal infections.
- Antiviral Drugs- Antiviral Drugs act against the infection causes due to viruses.

- Antimalarial drug- These are drugs used for prophylaxis, treatment and prevention of relapses of malaria.
- Antiamoebic drugs- These are drugs useful in infection caused by the protozoa Entamoeba histolytica.
- Antigiardiasis drugs- These are drugs act against giardiasis, caused by Giardia lamblia
- Antitrichomoniasis drugs- These are drugs effective against Trichomonas vaginalis is another flagellate protozoon which causes vulvovaginitis.
- Antileishmaniasis drugs- These are drugs act against Visceral leishmaniasis (kala-azar) caused by Leishmania donovani.
- Antihelminthics drugs- Anthelmintics are drugs that either kill (vermicide)
  or expel (vermifuge) infesting helminths.
- Antileprotic Drugs- These are drugs act against leprosy, caused by Mycobacterium leprae.

14.3 Antifungal Drugs

Antifungal drugs are used for deep systemic and superficial fungal infections. Infections caused by fungi are called mycoses and they can be divided into superficial infections (affecting skin, nails, scalp or mucous membranes) and systemic infections (affecting deeper tissues and organs). Systemic mycoses fungal disease is systemic candidiasis caused by a yeast like organism. Other more infections are cryptococcal meningitis or endocarditis, pulmonary aspergillosis and rhinocerebral mucormycosis.

Classification of Antifungal Drugs-

1. Antibiotics

A. Polyenes

- Amphotericin B (AMB)
- Nystatin

B. Heterocyclic benzofuran

- Griseofulvin

2. Antimetabolite

- Flucytosine (5-FC)

3. Azoles

A. Imidazoles (topical)

- Clotrimazole
- Miconazole
- Ketoconazole **(systemic)**

B. Triazoles (systemic)

- Fluconazole

4. Allylamine

- Terbinafine

5. Other topical agents

- Tolnaftate
- Quiniodochlor
- Sod. Thiosulfate

General mode of action of Antifungal agents-

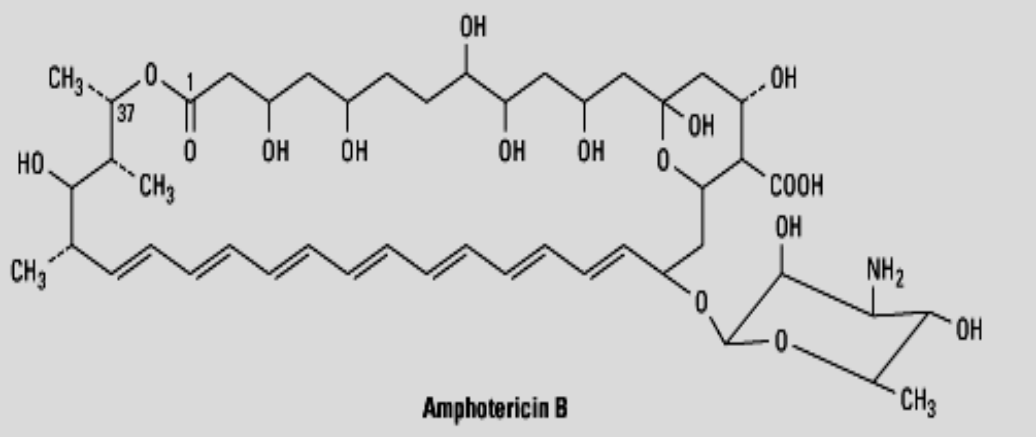
Polyene antibiotics:

Amphotericin B:

Amphotericin B is naturally obtained polyene macrolide antibiotic from streptomyces nodous. Though, it is potentially toxic, it is the drug of choice for the treatment of the systemic mycoses.

Mechanism of Action:

Amphotericin bind to the ergosterol present in the cell membranes and interferes with permeability and with transport function. This from of the cell, resulting in cell death. Amphotericin has a selective action, binding to the membranes of fungi and some protozoan, less ability mammalian cells and not at all to bacteria. Figure14.3 (a)



Amphotericin B

It is used topically for oral, vaginal and cutaneous candidiasis and ostomycosis. It is most effective for various types' systemic mycoses. Amphotericin B is most effective drug for resistant cases of kala azar and mucocutaneous leishmaniasis.

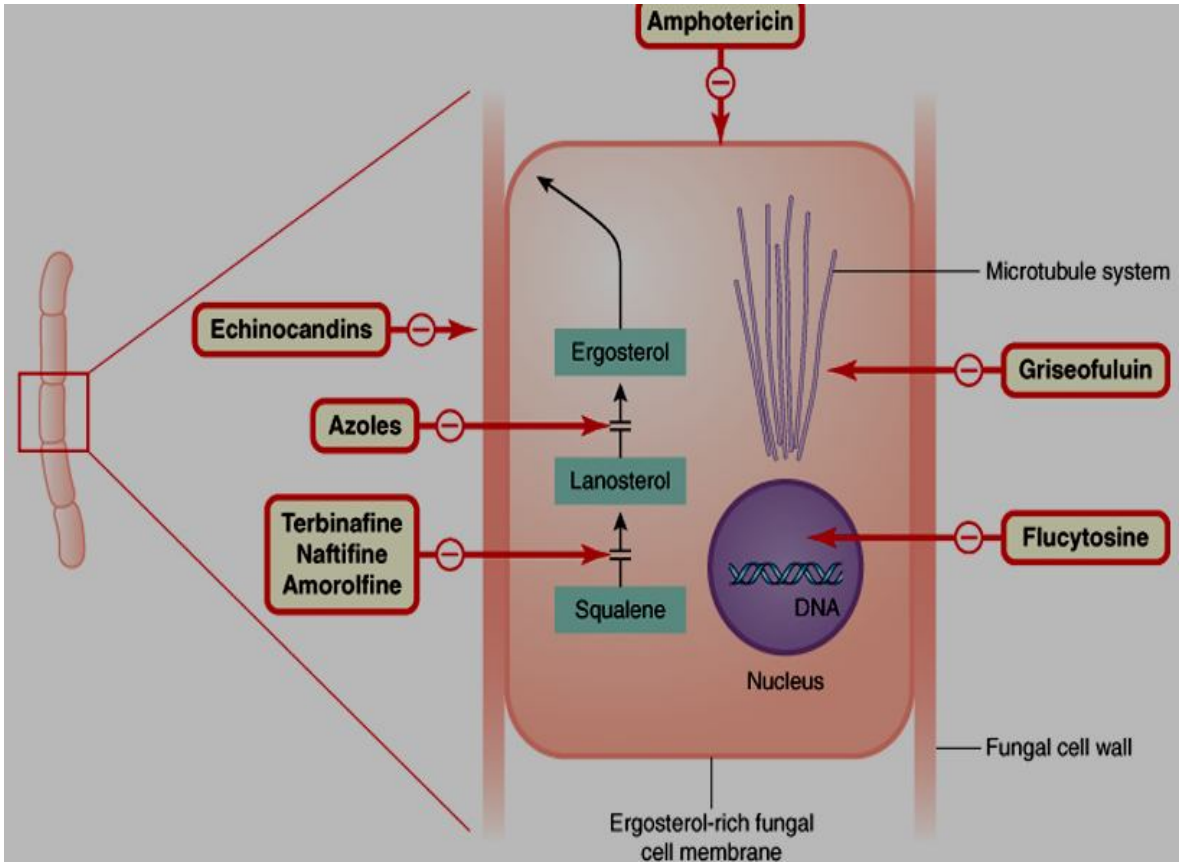


Fig.14.3: General mode of action of Antifungal agents

Griseofulvin :

Griseofulvin is a narrow-spectrum antifungal agent isolated from cultures of penicillium griseofulvum. It is the first chemical compound to cure efficiently infections due to the superficial dermatophytes (ringworm), when administered orally.

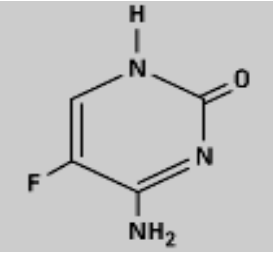
Mechanism of Action:

Griseofulvin interferes with the mitosis by interacting with microtubules. It also has a weak anti-inflammatory effect. It acts mainly on the growing fungal cells. It accumulates in the infected, newely synthesized, keratin-containing tissues, making them unsuitable for the growth of the fungi. Therapy must be continued until normal tissue replaces infected tissues. Figure14.3 (a)

Griseofulvin is used orally for ringworm infections. It is ineffective topically. Majority of the tinea infections are treated with Griseofulvin.

Flucytosine:

Flucytosine is a synthetic fluorinated pyrimidine that is given orally and is effective against systemic infections due to yeast.



Flucytosine

Mechanism of Action:

Flucytosine is taken up by fungal cells and is converted into antimetabolite 5-fluorouracil in them but not in human cells. 5-fluorouracil inhibits thymidylate synthetase and thus DNA synthesis. Figure14.3 (b)

Flucytosine is useful in cryptococcal meningitis and systemic candidiasis including urinary tract infections.

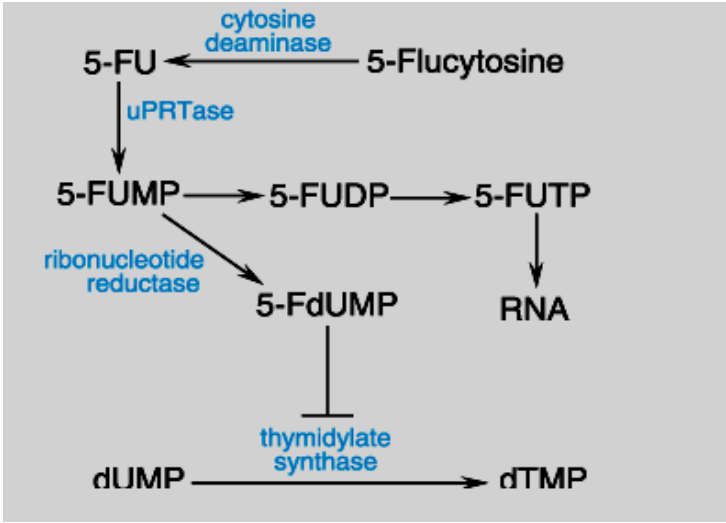


Figure14.3 (b): Mechanism of action of 5-Flucytosine

Azoles :

Azoles are group of synthetic fungistatic agents with a board spectrum of activity and the most extensively used antifungal drugs. The important drugs of this group are fluconazole, itaconazole, ketoconazole, miconazole, clotrimazole and econazole.

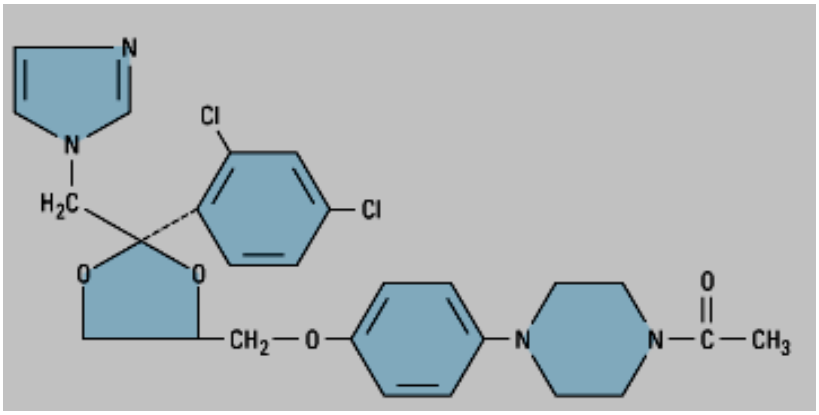
Mechanism of Action :

The mechanism of all azoles (imidazoles and triazoles) is the same. They inhibit the fungal cytochrome P450 3A (CYP3A) enzyme, lanosine 14α-demethylase which is responsible for the conversion of lanosterol to ergosterol which is main sterol in the fungal cell membrane. This inhibition disrupts

membrane function and increases permeability. The net effect s an inhibition of replication azoles also inhibit the transformation of candidal yeast cells into hyphae the invasive and pathogenic form of the parasite.

Ketoconazole :

Ketoconazole was the first azole, which is orally effective broad spectrum antifungal drug, useful is both dermatophytosis and deep mycosis.

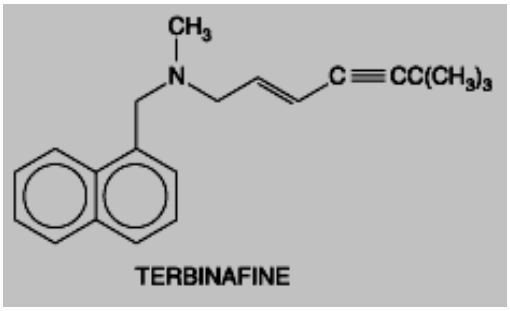


Ketoconazole

In addition to its antifungal activity, it also inhibit gonadal and adrenal steroid synthesis in humans. Thus, it suppresses testosterone and cortisol synthesis.

Terbinafine :

Terbinafine is administered both orally and topically and is effective against dematophytes and candida.



It acts by selectively inhibiting the enzyme squalene epoxidase, which is involved in the synthesis of ergosteral from squalene in the fungal cell wall. The accumulation of squalene within the cell is toxic to the organism. It acts as a fungicidal agent.

It is used topically for tinea and pityriasis infections. Onchomycosis is also treated. It is also used to treat the fungal infections of the nails.

14.4 Antiviral Drugs

Antiviral drugs act against the infection causes due to virus. Viruses are obligate intracellular parasites consisting essentially of nucleic acid (either RNA or DNA) enclosed in a protein coat or capsid. The nuclein acid and coat together is termed as nucleocapsid. Some viruses my also contain antigenic viral glycoproteins as well as host phospholipids acquired when the virus nucleocapsid buds through the nuclear or plasma membrane of the host cell.

Viruses do not have metabolic machinery of their own. They not only take nutrition from the host cell but also direct its metabolic machinery to synthesize new viral particles. Most viruses survive outside the host cell only for a short time. After penetration into the target cell, viral replication occurs in following steps:

- Uncoating of viral nucleic acid strands
- Early, regulatory protein synthesis
- Replication of viral RNA or DNA)
- Late structural protein synthesis
- Coating and maturation of viral particles and
- Release of viral particles from the cell

Antiviral agents can target any one of the above steps. To be effective therapy is to be started in the incubation period(prophylactic).

Classification of Antiviral Drugs-

1. ANTI-HERPES VIRUS-

- Idoxuridine
- Acyclovir
- Valacyclovir

2. ANTI-RETROVIRUS-

(a) Nucleoside reverse transcriptase inhibitors (NRTIs):

- Zidovudine (AZT)
- Didanosine

(b) Nonnucleoside reverse transcriptase inhibitors(NNRTIs):

- Nevirapine
- Efavirenz

(c) Protease inhibitors:

- Ritonavir
- Indinavir
- Nelfinavir

3. ANTI-INFLUENZA VIRUS-

- Amantadine
- Rimantadine*

4. NONSELECTIVE ANTIVIRAL DRUGS-

- Ribavirin
- Adefovir dipivoxil
- Interferon α

General mode of action of Antiviral agents-

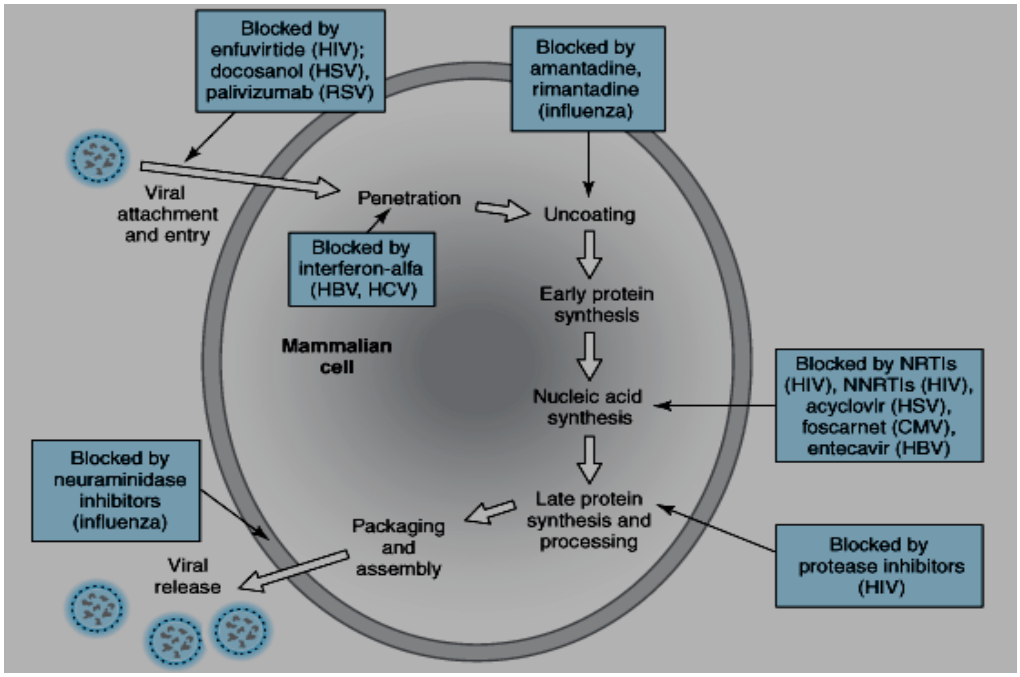


Fig.14.4 (a): General mode of action of Antiviral agents

1. Anti-Herpes Virus Agents:

Herpes viruses are associated with diseases such as cold sores, viral encephalitis and genital infections. Genital infections are a hazard to the newborn during parturition. The drugs that are effective against these viruses act mainly during the acute phase of viral infections. These drugs act mainly by inhibiting viral DNA polymerase or inhibition of attachment to the host cell. Figure14.4(b)

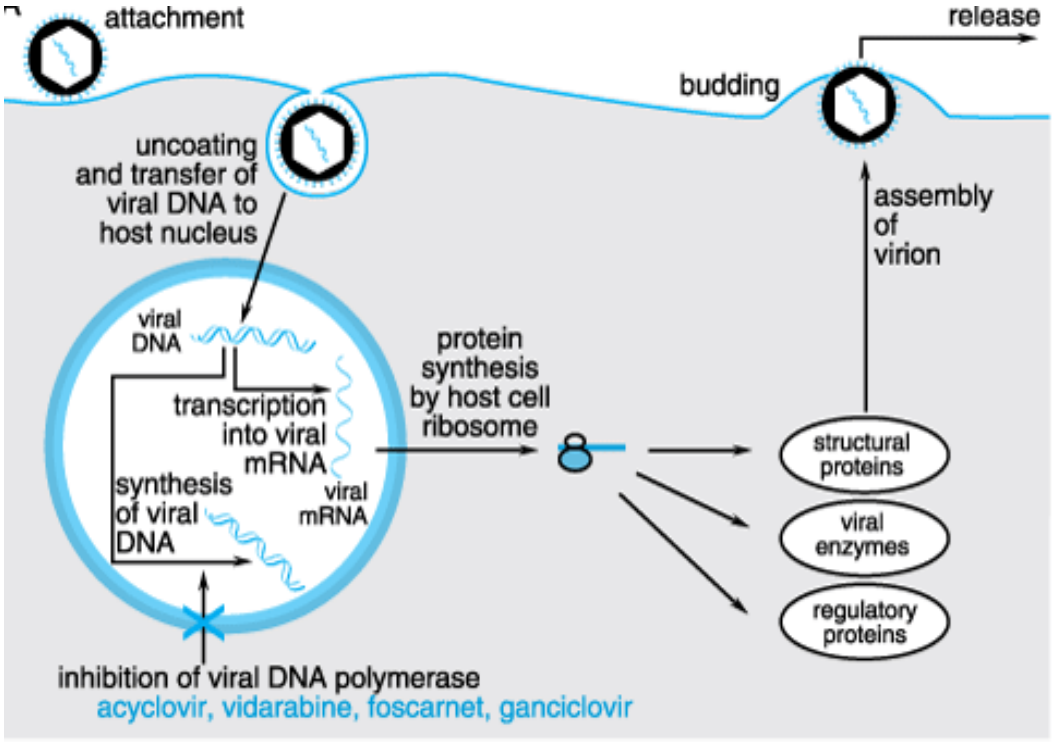
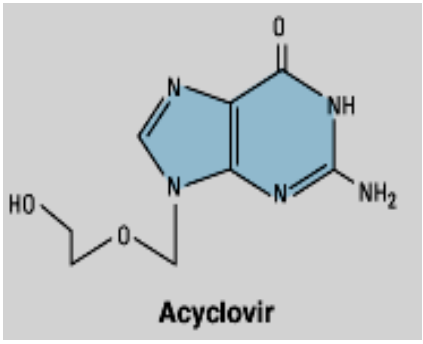


Fig.14.4(b): General mode of action of Anti-Herpes Virus agents

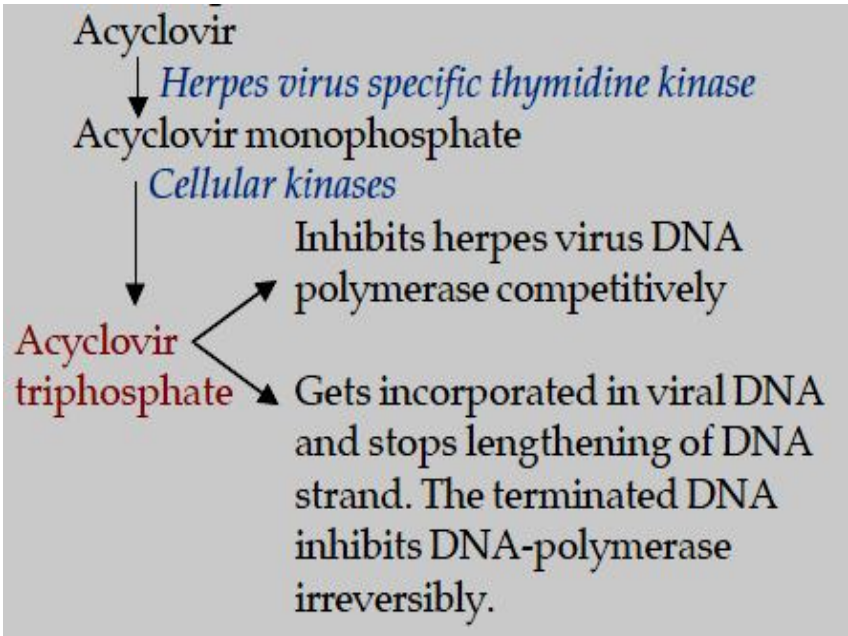
Acyclovir-

Acyclovir (9-[(2-hydroxy-ethoxy) methyl]-9H-guanine) is an acyclic guanine nucleoside analog with a high specificity for Herpes simplex and Varicella zoster viruses.



Mechanism of action-

Acyclovir is converted to its active metabolite via three phosphorylation steps. First, viral thymidine kinase converts acyclovir to acyclovir monophosphate. Next, host cell enzymes convert the monophosphate to the diphosphate and then to the active compound, acyclovir triphosphate, the active metabolite that inhibits DNA synthesis and viral replication. Figure14.4(b)



Oral acyclovir is useful in the treatment of HSV-1 and HSV-2 infections, such as genital herpes, herpes encephalitis, herpes keratitis, herpes labialis, and neonatal herpes.

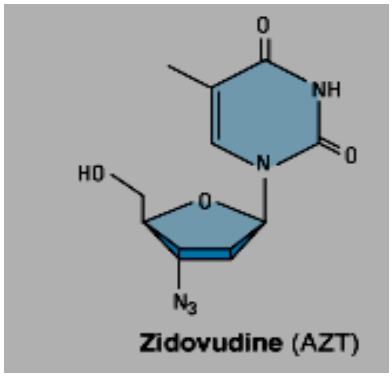
2. Anti-retrovirus drugs-

These are drugs active against human immunodeficiency virus (HIV) which is a retrovirus.

Nucleoside reverse transcriptase inhibitors (NRTIs)-

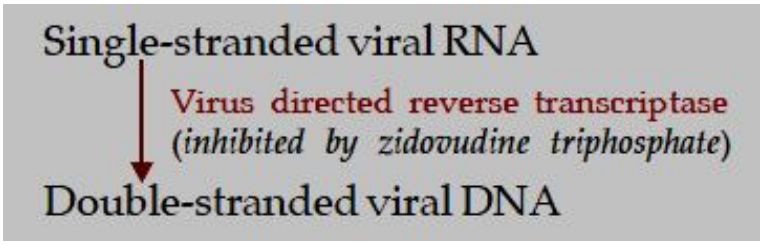
Zidovudine-

It is a thymidine analogue (azidothymidine, AZT), the prototype NRTI.



Mechanism of action-

After phosphorylation in the host cell—zidovudine triphosphate selectively inhibits viral reverse transcriptase (RNA-dependent DNA polymerase) in preference to cellular DNA polymerase. Figure14.4(c)



Zidovudine is used in HIV infected patients only in combination with at least 2 other anti retrovirus drugs.

Didanosine (ddI)-

It is a purine nucleoside analogue which after intracellular conversion to didanosine triphosphate competes with ATP for incorporation in viral DNA, inhibits HIV reverse transcriptase and terminates proviral DNA. Figure14.4(c)

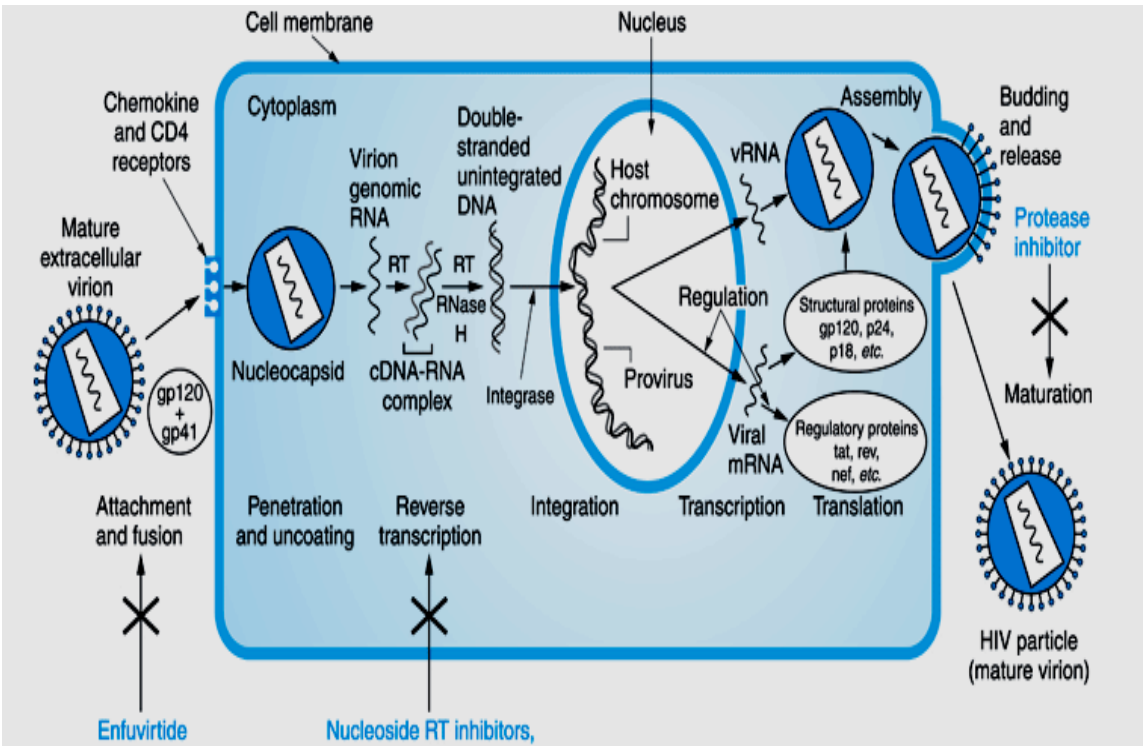


Fig.14.4(c): General mode of action of Anti-Retro Virus agents

Non-nucleoside reverse transcriptase inhibitors (NNRTIs)-

Nevirapine (NVP) and Efavirenz (EFV) –

These are nucleoside unrelated compounds which directly inhibit HIV reverse transcriptase Figure14.4(c) without the need for intracellular phosphorylation.

Retroviral protease inhibitors (PIs)-

An aspartic protease enzyme encoded by HIV is involved in the production of structural proteins and enzymes (including reverse transcriptase) of the virus. The large viral polyprotein is broken into various functional components by this enzyme. This protease acts at a late step in HIV replication, i.e. maturation of the new virus particles when the RNA genome acquires the core proteins and enzymes. They bind to the protease molecule, interfere with its cleaving function. Figure14.4(c)

3. Anti influenza virus agents-

Amantadine and Rimantadine-

Amantadine (Symmetrel) is a synthetic tricyclic amine, and rimantadine (Flumadine) is its α-methyl derivative. Both drugs inhibit the replication of the three antigenic subtypes of influenza A (H1N1, H2N2 and H3N2) and have negligible activity against influenza B.

Mechanism of action-

Inhibition of the viral M2 protein, an integral membrane protein that acts as a H+ channel. Blockade of the M2 protein prevents the acid-mediated dissociation of the ribonucleoprotein complex that occurs early in replication. In certain strains, the pH changes that result from M2 inhibition alter the conformation of hemagglutinin, hence inhibit viral assembly. Figure14.4(d)

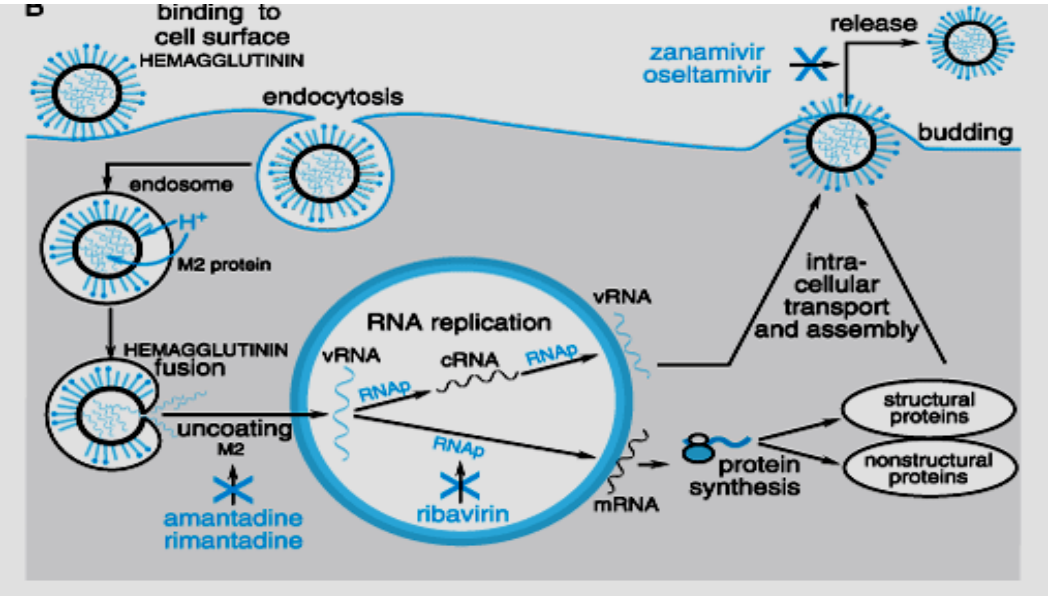


Fig.14.4 (d): General mode of action of Anti influenza Virus agents

14.5 Antimalarial drug

There are the drugs used for prophylaxis, treatment and preventing of relapses of malaria. Malaria caused by 4 species of the protozoal parasite plasmodium, is endemic in most parts of india and other tropical countries. Malaria is a mosquito-borne acute infectious disease caused by four species of the protozoal parasite plasmodium. The symptoms of malaria include fever, shivering, pain in the joints, headache, repeated vomiting, generalized convulsions and coma.

Classification of Antimalarial drugs-

1. 4-AMINOQUINOLINES-

- Chloroquine
- Amodiaquine

2. QUINOLINE-

- Mefloquine

3. CINCHONA ALKALOID-

- Quinine
- Quinidine

4. BIGUANIDES-

- Proguanil

5. DIAMINOPYRIMIDINES-

- Pyrimethamine

6. 8-AMINOQUINOLINE-

- Primaquine
- Bulaquine

7. SULFONAMIDES-

- Sulfadoxine and sulfone
- Dapsone

8. TETRACYCLINES-

- Tetracycline
- Doxycycline

9. SESQUITERPINE –

- Artesunate
- Artemether

10. AMINO ALCOHOLS-

- Halofantrine
- Lumefantrine

11. MANNICH BASE-

- Pyronaridine

12. NAPHTHOQUINONE-

- Atovaquone

General mode of action of Antimalarial agents-

Antimalarial drugs attack the parasites at its various stages of life cycle in the human host. Antimalarials which act on erythrocytic schizogony are calle erythrocytic schizontocides, those which act on preerythrocytic and also exoerythrocytic stages in liver are called tissue schizontocides, whereas those which kill gametocytes in blood are called gametocides. Figure14.5

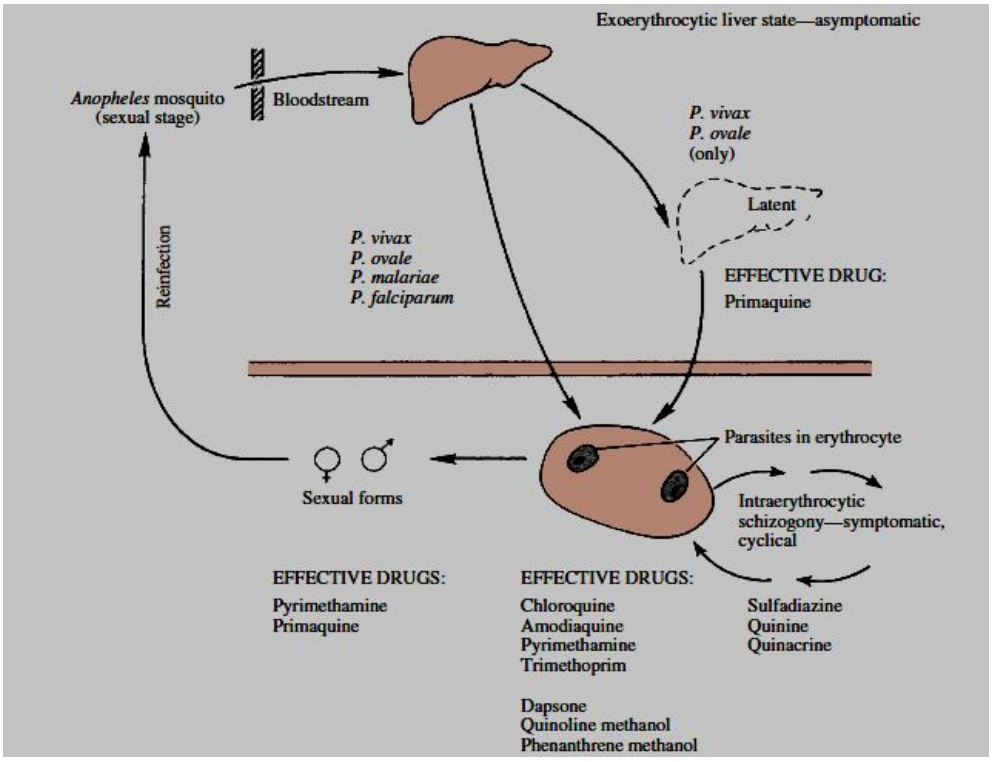
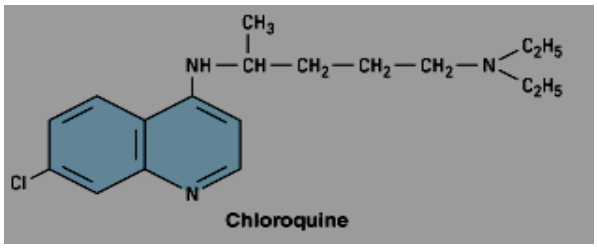


Fig.14.5: Life cycle of malaria parasites and locations where specific drugs are effective

Chloroquine :

Chloroquine is a potent blood schizonticidal agent effective against the erythrocytic forms of all four parasites but has no action against sporozoites, hypnozoites or gametocytes.



Chloroquine

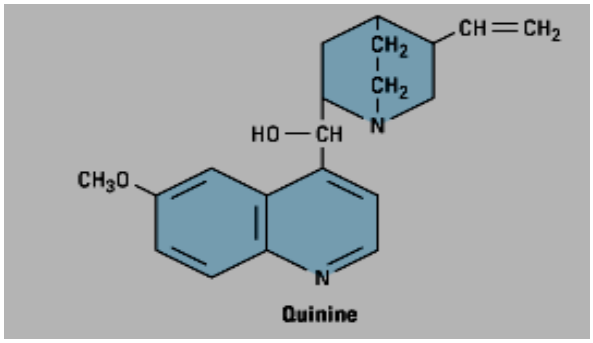
Mechanism of action :

The mechanism of action of chloroquine is complex. Chloroquine which is unionized at netural PH diffuses freely into the parasite lysosome. At the acidic PH of the lysosome, it is converted to a protonated, membrance impermeable form and is trapped inside the parasite. At high concentrations, it inhibits the synthesis of protein RNA and DNA synthesis. Chloroquine acts mainly on haem disposal by preventing digestion of haemoglobin by the parasite and thus reducing the supply of amino acid necessary for survival of parasite. It also inhibits the enzyme haem polymerase that polymerises toxic free haem to haemozoin, rendering it harmless. Figure14.5

Chloroquine is the drug of choice for clinical cure and suppression of all types of malaria. It is also used in giardiasis, rheumatoid arthritis and to control acute manifestations of lepra reaction.

Quinine :

Quinine is an alkaloid obtained from cinchona bark.



Quinine

It is the oldest of all agents used in the treatment of malaria and is still used for cerebral malaria and chloroquine-resistant P.falciparum malaria. Quinine is a blood schizonticidal agent and is effective against erythrocytic forms of all the four species of falciparum but has no effect on exoerythrocytic forms or gametocytes.

14.6 Antiamoebic drugs

Amoebiasis (amoebic dysentery) is an infectious disease caused by protozoa, entamoeba histolytica, which is produced by the ingestion of cysts of this organism. The ingested cysts develop into trophozoites and adhere to the colonial epithelial cells in the intestine. These trophozites then lyses the host cell and invades the submucosa. This produces the amoebic ulcers and cause acute dysentery or chronic intestinal amoebiasis. The parasite may also pass into blood stream and invades the liver causing liver abscesses. Other organs like lung, spleen, kidney and brain are rarely involved in extraintestinal amoebiasis. Some individuals act as carriers of parasite, without developing any disease, but the cysts that are present in their faces affect others. The cysts can survive outside the body for atleast a week in a moist and cool environment. Amoebiasis can be acute or chronic with patients showing various degrees of illness and symptoms such as mild diarrhoea to fulminating diarrhoea. The use of drugs in treating amoebiasis is aimed not only in ill patients but also on carriers. Different drugs are available for treating acute, chronic and extra-intestinal infections and also in the carrier state.

1. TISSUE AMOEBICIDES

(a) For both intestinal and extraintestinal amoebiasis

- Nitroimidazoles: Metronidazole
- Alkaloids: Emetine

(b) For extraintestinal amoebiasis

- Chloroquine

2. LUMINAL AMOEBICIDES

(a) Amide

- Diloxanide furoate
- Nitazoxanide

(b) 8-Hydroxyquinolines

- Quiniodochlor

(c) Antibiotics

- Tetracyclines

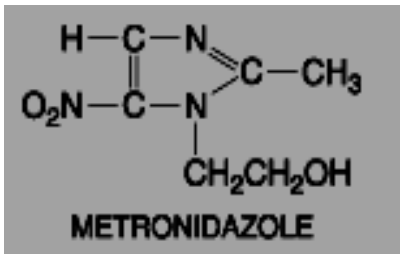
General mode of action of Antiamoebic drugs -

The mode of action of these drugs is that they are selectively toxic to anaerobic microorganisms. After entering into the cell by diffusion the drug gets reduced by some redox proteins which operate only in anaerobic microbes and exerts

cytotoxicity by damaging DNA and other critical biomolecules. DNA helix destabilization and strand breakage has been occurred in susceptible organisms.

Metronidazole-

Metronidazole is the drug of choice in the treatment of different forms of amoebiasis. It kills the trophozites of E.histolytica but has no effect on the cysts.



METRONIDAZOLE

It is often used in combined with diloxanide furoate for the treatment of amoebiasis.

Mechanism of action:

Metronidazole is selectively toxic to anaerobic organisms and amoebae. Some anaerobic protozoan parasites possess ferrodoxin like, low-redox potential, electron transport proteins that participate in metabolic electron removal reactions. The nito group of metronidazole accepts electrons, forming reduced cytotoxic compounds that bind to proteins and DNA causing cell death.

It is used in treatment of amoebiasis, giardiasis, trichomonas vaginitis and pseudomembranous enterocolitis. It is also used in treatment of many anaerobic bacterial infections and in peptic ulcers.

Diloxanide Furoate-

Diloxanide furoate (Furamide) is an amebicide that is effective against trophozoites in the intestinal tract.

14.7 Antigiardiasis drugs

Giardia lamblia is a flagellate protozoon which mostly lives as a commensal in the intestine. It is transmitted from man too man by faecal contamination of food and water. It sometimes invades the mucosa and causes diarrhoea. Many drugs which are useful in amoebiasis are also used in the treatment of giardiasis.

The drugs are-

1. Metronidazole
2. Mepacrine
3. Quinodocholor

4. Furazolidone

14.8 Antitrichomoniasis drugs :

Trichomonas vaginalis is another flagellate protozoon which causes vulvovaginits. The main trichomonas organism that causes disease in human is T.vaginalis. Virulent strains of the organism can cause inflammation of the vagina in females and sometimes of the urethra in males.

The drugs are-

1. **DRUGS USED ORALLY-**

- Metronidazole

2. **DRUGS USED INTRAVAGINALLY-**

- Diiodo hydroquin, Quiniodochlor, Clotrimazole

14.9 Antileishmaniasis drugs

Visceral leishmaniasis (kalazar) caused by leishmania donovani occurs in tropical and subtropical region of the world. Leishmaniasis is transmitted by the bite of the female sand fly phlebotomus. In this fly the parasite present in the flagellate extracellular (promastigote) form, while in human it is found extracellularly with in macrophages in the nonflagellate (amastigote) form. Mucocutaneous and dermal leishmaniasis are caused respectively by L. Brazilliensis and L. Tropica.

In visceral type (liver and speen), the Parasite is present in blood stream and causes very serious problems, such as hepatomegaly, anaemia and intermittent fever. In mucocutaneous form, large ulcers are produced in mucous membrances. In cutaneous form, simple skin infections that may heal spontaneously are observed.

Drugs used in the treatment of leishmaniasis are-

1. **ANTIMONIALS**-

- Sodium stibo gluconate

2. **DIAMIDINE-**

- Pentamidine

3. **OTHERS-**

- Amphotericine B
- Ketoconazole
- Miltefosine

General mode of action of Antileishmaniasis drugs-

Antimonials: Sodium stibogluconate-

Sodium stibogluconate is the drug of choice for kala-azar (visceral leishmaniasis). The drug inhibited SH-dependent enzymes and may cause oxidative damage of the parasite by producing free radicals. It has been shown to block glycolytic and fatty acid oxidation pathway.

Miltefosine:

Miltefosine is effective in treatment of both cutaneous and visceral leishmaniasis. It probably acts by interfering with cell signaling pathways.

Diamidine: Pentamidine-

It interacts with kinetoplast DNA and inhibits topoisomerase II or interferes with aerobic glycolysis.

Pentamidine is used in antimony-resistant leishmaniasis.

14.10 Antihelminthics drugs

Helminthic infections are the major health problem in topical countries and many people are suffering from worm infestations. Helminths (worms) can cause various gastrointestinal and general symptoms. They also cause blood loss, nutritional deficiencies, urticaria allergic manifestations and even intestinal obstruction. Humans are the primary hosts for many helminth infections. Most of the worms reproduce sexually in human host, producing eggs and larvae which pass out of the body and infect secondary or intermediate host. Helminthic infections are rarely fatal, but are major cause for ill health.

The drugs which are useful against helminth are-

1. Mebendazole
2. Albendazole
3. Thiabendazole
4. Ivermectin
5. Pyrantel
6. Metronidazole
7. Praziquantel

General mode of action of Anthelminthic drugs-

Anthelminthic drugs aim the metabolic targets of the parasite that are either absent or have different characteristics than those of the host. The drug must be

able to penetrate the cuticle of the worm or gain access to its alimentary tract. These drugs act by causing paralysis of the worm or by damaging its cuticle which leads to partial digestion or rejection by immune system. They also interfere with the metabolism of the worms, as the metabolic requirements vary greatly between different species. The anthelminthic drug that kills the worm is called vermicidal and that affects the worm causing its expulsion is known as vermifuge.

Benzimidazoles :

Benzimidazoles are broad-spectrum anthelminthic drugs and include mebendazole, thiabendazole and albendazole.

These drugs mainly bind to free β-tubulin and inhibits its polymerisation, thus interfering eith microtubule-dependent glucose uptake by the worm. These have a selective inhibition on microtubular function of helminths than in mammalian tissue.

Mebendazole :

Mebenazole is effective in the treatment of ascariasis, enterobiasis, trichuriasis and in hook worm infestation. It also has some action against S.stercoralis. it acts very slowly and the affected parasites are expelled with faces. It also affects the ova of trichuris and Hook worm and is also effective against larvae of trichinella spiralis and Echinococcus granulosus(Hydatid worm).

Praziquantel:

Praziquantel is a broad-spectrum anthelminthic and is the drug of choice for treatment of schistosomiasis and for cestode infections like cysticercosis. It acts mainly by increasing the permeability of the cell membrance of the paralysis and death of the worm. It also modifies the parasite rendering it susceptible to the immune response of the host. The drug is not only active against adult schistosomes but also their immature forms that cause infections in humans by penetrating the skin.

Pyrantel:

Pyrantel is a derivative of tetrahydropyrimidine and is effective against round worms, pin worms and hook worms. It is less active against strongyloides and has no action against Trichuris and other worms. Pyrantel acts by depolarizing the neuromuscular junction in the helminths, causing spasm and paralysis. It also possess some anticholinesterase activity. The paralyzed worms are expelled from the hosts intestinal tract.

14.11 Antileprotic Drugs

These drugs act against Mycobacterium leprae, causing leprosy disease.
Drugs are-

1. SULFONE-

- Dapsone (DDS)

2. PHENAZINE DERIVATIVE-

- Clofazimine

3. ANTITUBERCULAR DRUGS-

- Rifampin, Ethionamide

4. OTHER ANTIBIOTICS-

- Ofloxacin
- Minocycline
- Clarithromycin

Dapsone (DDS)-

It is diamino diphenyl sulfone (DDS).

DAPSONE

Mechanism of Action-

The sulfones are structural analogues of PABA and are competitive inhibitors of folic acid synthesis. Folic acid functions as a coenzyme in the transfer of one-carbon units required for the synthesis of thymidine, purines, and some amino acids and consist of three components: a pteridine moiety, PABA, and glutamate. The sulfonamides, as structural analogues, competitively block PABA incorporation; sulfonamides inhibit the enzyme dihydropteroate synthase, which is necessary for PABA to be incorporated into dihydropteroic acid, an intermediate compound in the formation of folinic acid. Since the sulfonamides reversibly block the synthesis of folic acid, they are bacteriostatic drugs.

Sulfones are bacteriostatic and are used only in the treatment of leprosy.

14.12 Summary

There are different drugs that are used in the treatment of infection. Some drugs are act on the site of infection locally. Drugs follow different path of mechanism by which infection may be cure. Pharmacotherapy of local infective disease such as antiviral, antifungal, antimalarial, antileprotic, antigiardiasis, antileishmaniasis, antiamoebic and antiprotozoans etc. may be cure by drugs used alone or in combination. Anti-infective drugs may act by penetration into the cell of infective agent and distrupt their cell and cell wall components and inhibit their genetic material and their metabolic function.

14.13 Glossary

- **Anti-infective-** Any agent that is used to combat infection.
- **Antiinflammatory-** An agent that counteracts inflammation.
- **Anaemia-** A reduction in the mass of circulating red blood cells.
- **Bacteriostatic-** Inhibiting bacterial growth.
- **Diarrhoea-** The passage of fluid or unformed stools.
- **Fungistatic-** Inhibiting fungi growth.
- **Urinary tract infection-** Infection of the kidneys, ureters, or bladder by microorganisms.
- **Vaginitis-** Inflammation of the vagina.

14.14 Review questions / Comprehensive Questions

1. What are the local anti-infective agents?
2. Enlist the local anti-infective agents.
3. Discuss the general mode of action of local anti-infective agents.
4. Explain the mechanism of action and uses of NNRTIs.
5. Classify the antiviral drugs.
6. Discuss the mechanism of action and uses of azoles.
7. Discuss the general mode of action of anti protozoal drugs.
8. Write the mechanism of action and uses of quinine.
9. Enlist the drugs which are used in the treatment of giardiasis and trichnosomiasis.
10. What are the antihelminthics? Discuss their general mode of action.

14.15 References and Suggested readings

1. Basic & Clinical Pharmacology (9th Ed.)- Bertram G.Katzung (Mc Graw Hill Publisher) 2004.
2. The Pharmacological basis of Therapeutics (10th Ed.)- Goodman & Gilman's (Mc Graw Hill Publisher) 2001.
3. Pharmacology (5th Ed.)- H.P Rang and M.M Dale (Elsevier publisher) 2003.
4. An introduction to medicinal chemistry (3rd Ed.)- Graham L.Patrik 2006. Principles of medicinal chemistry (3rd ed.)- William O. Foye (Varghese publisher) 1989.
5. Essential of Medical Pharmacology (7th ed.)- KD Tripathi (JAYPEE Publisher) 2013.

Unit-15

Synthesis of Local Anti-Infective Drugs

Structure of Unit:

15.1 Objective

- In this unit we will discuss about the drugs which are used for local infections and also their method of synthesis and uses.
- Here we study that how sulphonamides are prepared.
- Here we also explain the use and adverse effect of these drugs.

15.2 Introduction

Anti-infective is an agent that is capable of acting against infection, either by inhibiting the spread of an infectious agent or by killing the infectious agent outright. Drugs of this class differ from all others in that they are designed to inhibit/kill the infecting organism and to have no or minimal effect on the recipient or host. These drugs are used for superficial fungal infections. Fungal infection is due to fungi, which are plant-like, non photosynthetic eukaryotes

growing either in colonies of single cell (yeasts) or in filamentous multicellular aggregates (molds). Drugs which are used for local infections are sulphonamides, chloroquin, primaquin, econazole, fluconazol etc.

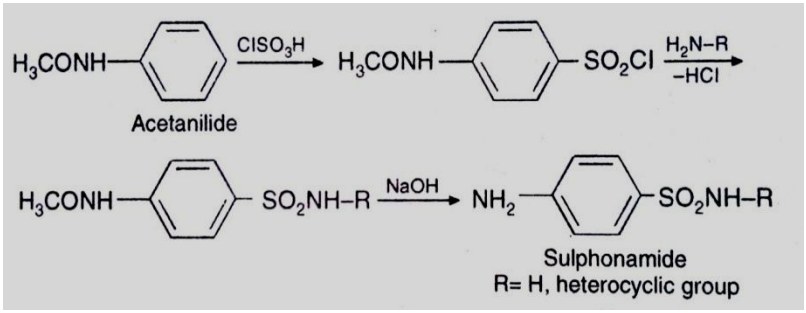
15.3 Sulphonamide

The sulphonamides are synthetic bacteriostatic antibiotics with a wide spectrum against most gram-positive and many gram-negative organisms. The sulphonamides and sulphone antibacterial as well as the 2,4-diaminopyrimidine antifolates continue to be successful chemotherapeutic agents.

Synthesis:-

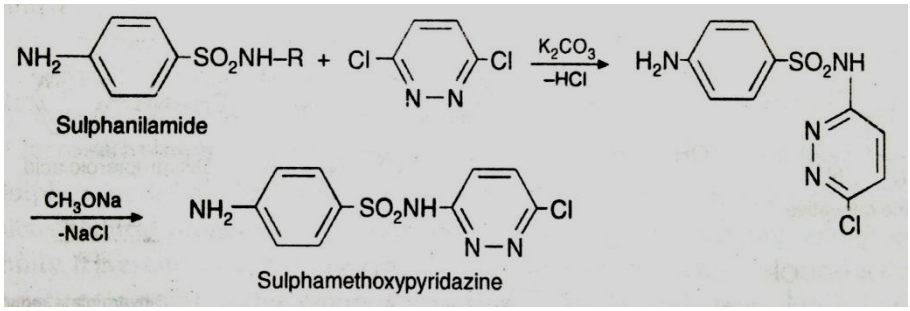
General methods of preparation:-

Method-I



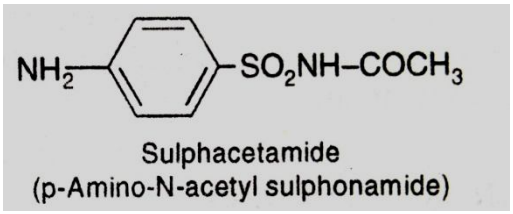
Acetanilide on treatment with chlorosulphonic acid gives sulphonyl chloride derivative, which on reaction with appropriate primary amine gives sulphonamide derivative. Saponification gives the final target compound.

Method-II



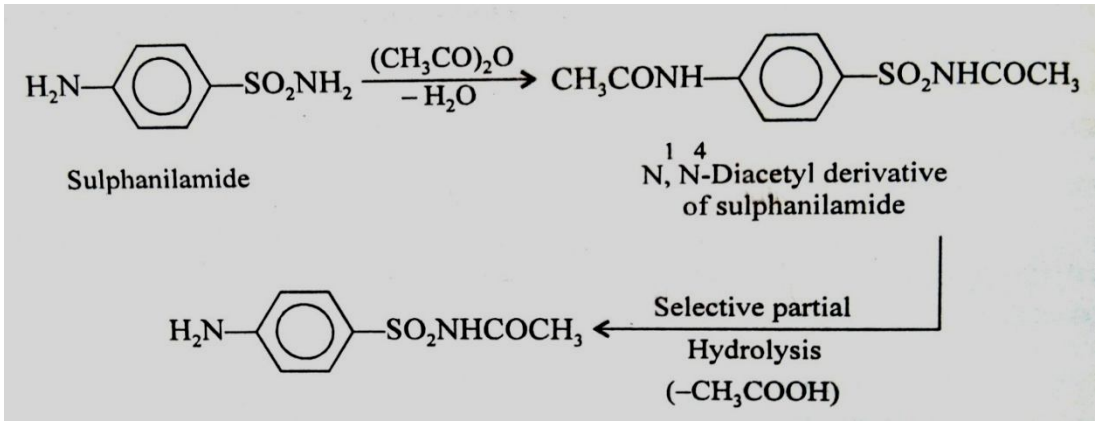
- **Synthesis of local anti-infective sulphonamides with their structure**

A. Structure of sulphacetamide:-



Sulphacetamide
(p-Amino-N-acetyl sulphonamide)

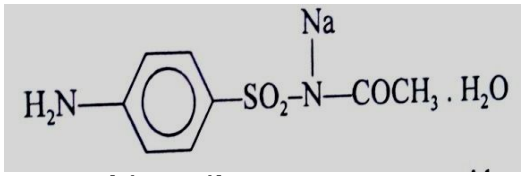
Synthesis of sulphacetamide:-



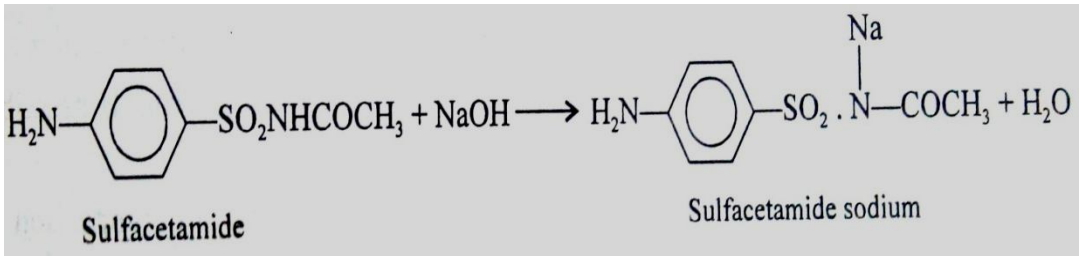
It is prepared by the selective and partial hydrolysis of the N1, N4-diacetyl derivatives of sulphanilamide which is obtained by acetylation of sulphanilamide.

It possesses general characteristic of a sulphonamide and was formerly used in the treatment of bacterial infections of the urinary tract.

B. Structure of sulphacetamide sodium:-



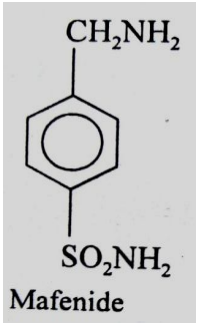
Synthesis of sulphacetamide sodium:-



It may be prepared by heating together sulphacetamide and sodium hydroxide in equimolar concentration.

It is chiefly employed by local application in injuries or infections of the eyes at various strengths ranging from 10% to 30%, also used in the treatment of acute conjunctivitis.

C. Structure of Mafenide:-

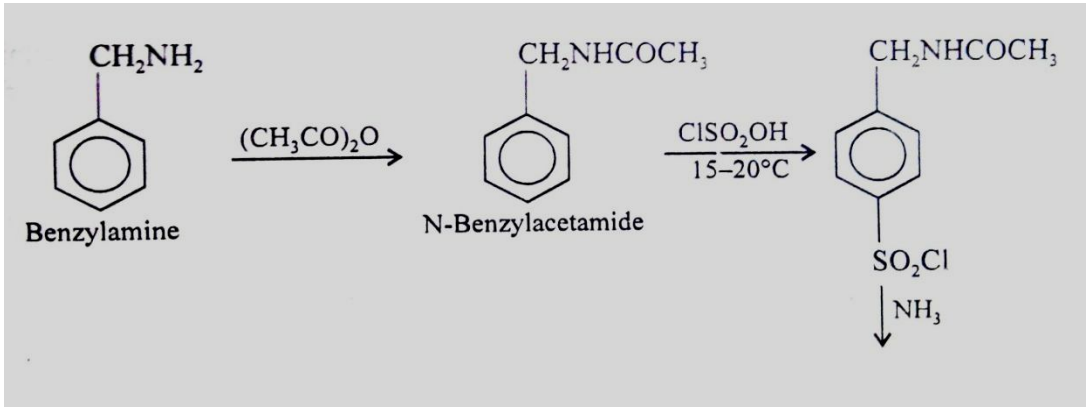


Mafenide

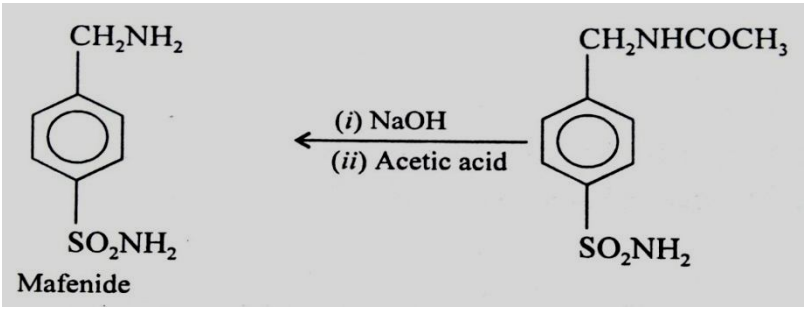
N-benzylacetamide is prepared by the acetylation of benzylamine, which on treatment with chlorosulfonic acid at 15-20°c yield p-benzylamide sulphonyl chloride. This on amination gives the corresponding sulphonamide derivative, which upon hydrolysis with sodium hydroxide and subsequent neutralization with acetic acid yields mafenide.

It is used in the treatment and cure of gas gangarene.

Synthesis of Mafenide:-

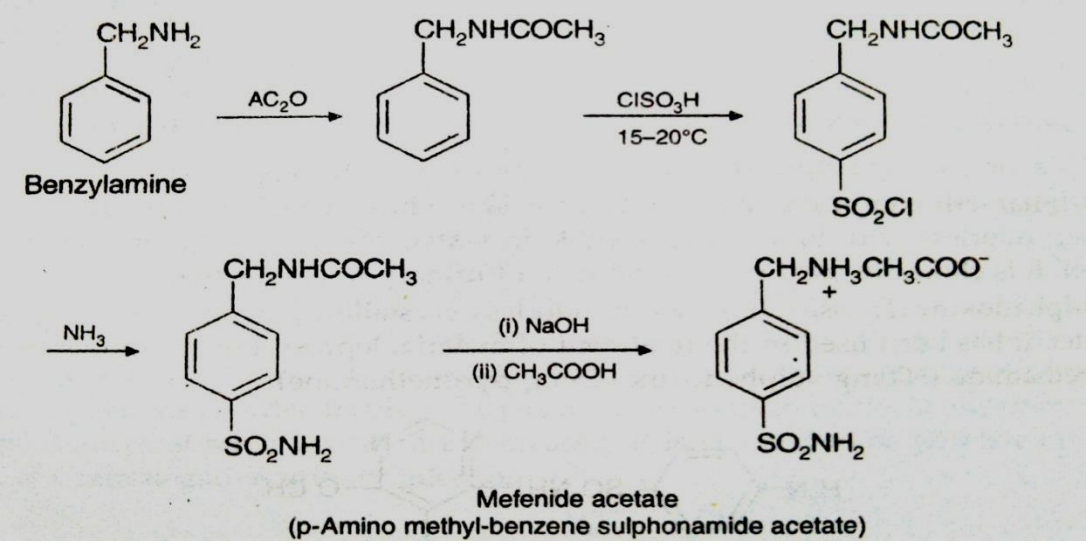


Benzylamine N-Benzylacetamide

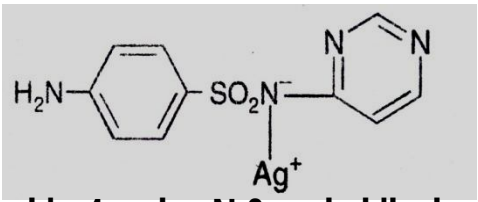


Mafenide

Synthesis of Mafenide acetate:-



D. Structure of silver sulfadiazine:-



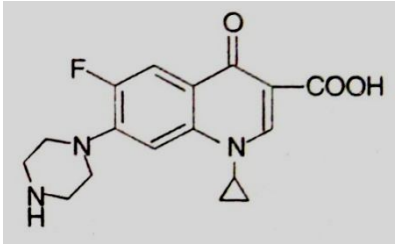
Benzenesulfonamide, 4-amino-N-2-pyrimidinyl, mono silver (1+) salt

It is an effective topical antimicrobial agent especially against "pseudomonas species". it find its extensive use in burn therapy because it attacks the pseudomonas radically which is perhaps considered to be the ultimate cause of failures in the treatment of burn case.

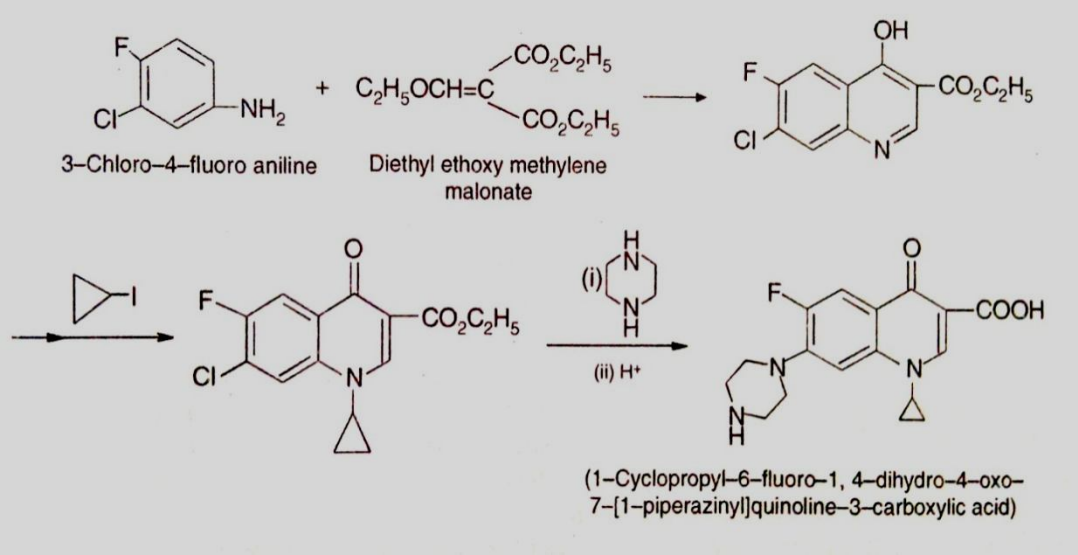
15.4 Ciprofloxacin:- (ciproxin)

It is a bactericidal drug. Its mode of action depends upon blocking bacterial DNA replication by binding itself to an enzyme called DNA gyrase.

Structure of ciprofloxacin:-



Synthesis:-



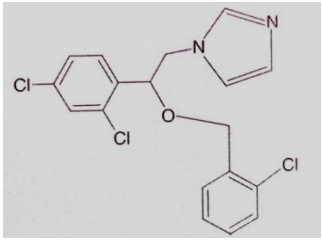
Ciprofloxacin is a broad spectrum antibiotic that is active against both gram-positive and gram-negative bacteria. The major adverse effect seen with the use of ciprofloxacin is gastro-intestinal irritation, common with many other such antibiotics.

It is effective for the treatment of urinary tract infection and prostatitis and for acute diarrheal disease caused by E.coli shigela, salmonella and campylobacter.

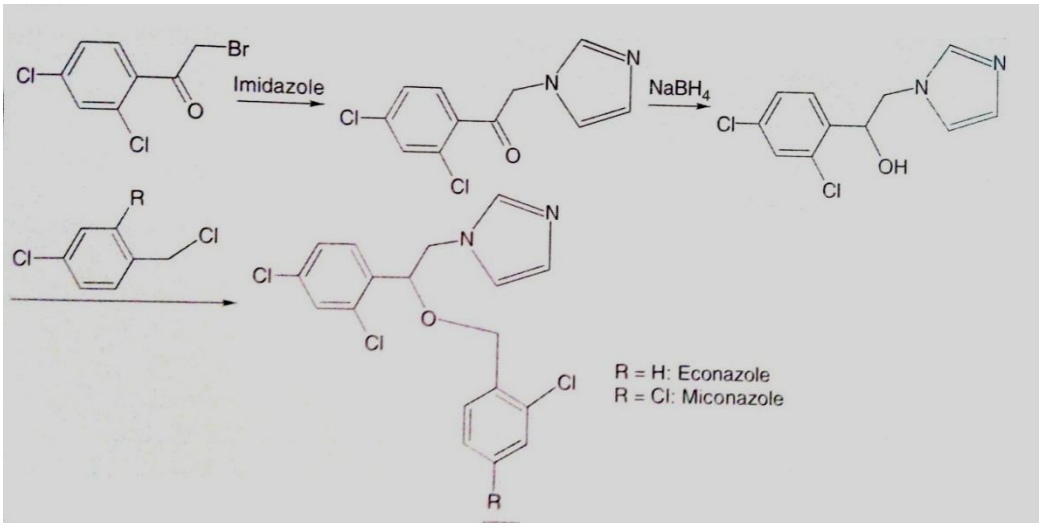
15.5 Econazole

It is deschloro derivative of miconazole. Its actions are like miconazole. It is available as a water insoluble cream (1%) to be applied twice a day.

Structure of Econazole:-



Synthesis of Econazole

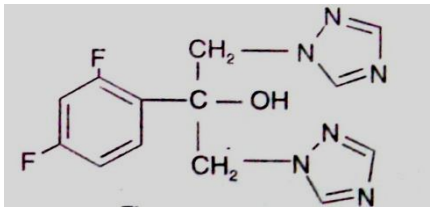


Econazole is synthesised from 2,4-dichlorophenacyl. It reacts with hydrogen bromide and give 2,4-dichlorophenacyl bromide, which is reacted with imidazole to make 1-(2,4-dichlorobenzoylmethyl)-imidazole. Reducing the carbonyl group in this molecule with sodium bromohydride gives 1-(2,4-dichlorophenyl)-3-(1-imidazolyl)-ethanol and the hydroxyl group is alkylated by 4-chlorobenzylchloride using base such as sodium hydride to make econazole.

15.6 Fluconazole

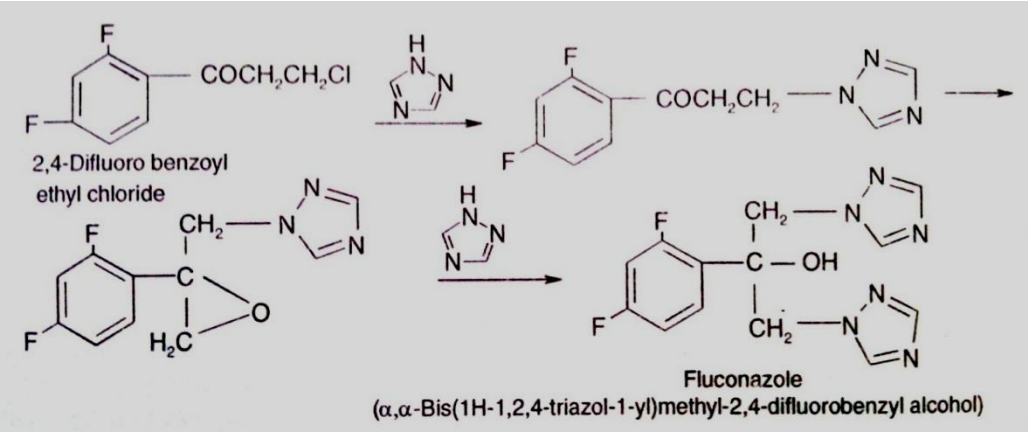
Fluconazole is a widely used bis-triazole antifungal agent. Fluconazole is generally considered to be a fungistatic agent. It is principally active against candida speices and crytococcus species. fluconazole has useful activity against coccidioides immitis and is often used to suppress the meningitis produced by that fungus.

Structure of Fluconazole:-

IUPAC- 2-(2,4-  diflurophenyl)-1,3-bis(1H-1,2,4-triazol-1-yl) propane-2-ol

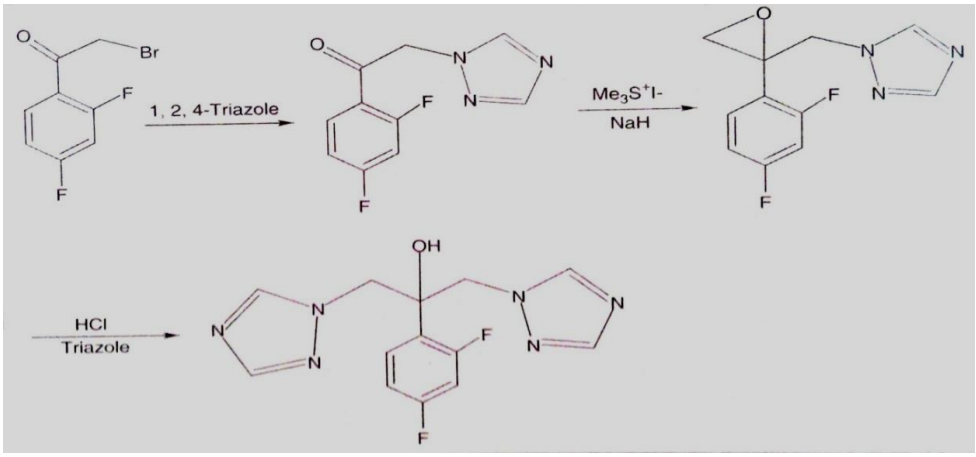
Synthesis:-

Method-I



It is prepared from 2,4-diflurophenacyl chloride. Displacement of chlorine by triazole affords intermediate condensation of carbonyl group with the yield from trimethylsulphonium iodide, leads to an addition product.

Method-II



The anion formed on the carbonyl oxygen then internally displaces dimethyl sulphide to give on oxiran or (epoxide). Reaction of this with triazole leads to epoxide ring opening with consequent incorporation of the second triazole, and affords fluconazole.

The drug is indicated for the treatment of systemic candida infection and the treatment of cryptococcal meningitis.

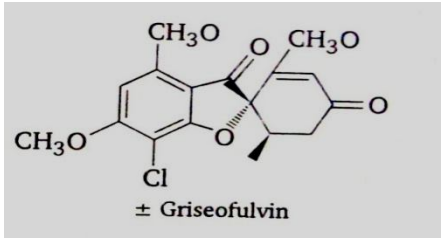
The adverse effect profile of fluconazole has remained very favourable. Nausea and vomiting occur. Allergic rash, eosinophillia and thrombocytopenia have been encountered in patients with AIDS.

15.7 Griseofulvin:- (fulvicin)

It is obtained from the mould "penicillium griseofulvum". Griseofulvin is a fungistatic drug that cause disruption of the mitotic spindle by interacting with polymerised microtubules.

It is a white, tasteless powder and slightly soluble in water. It is naturally occurring spirane. The drug is principally fungistatic, effective against the dermatophytes- Trichophyon, Microsporum and Epidermophyton.

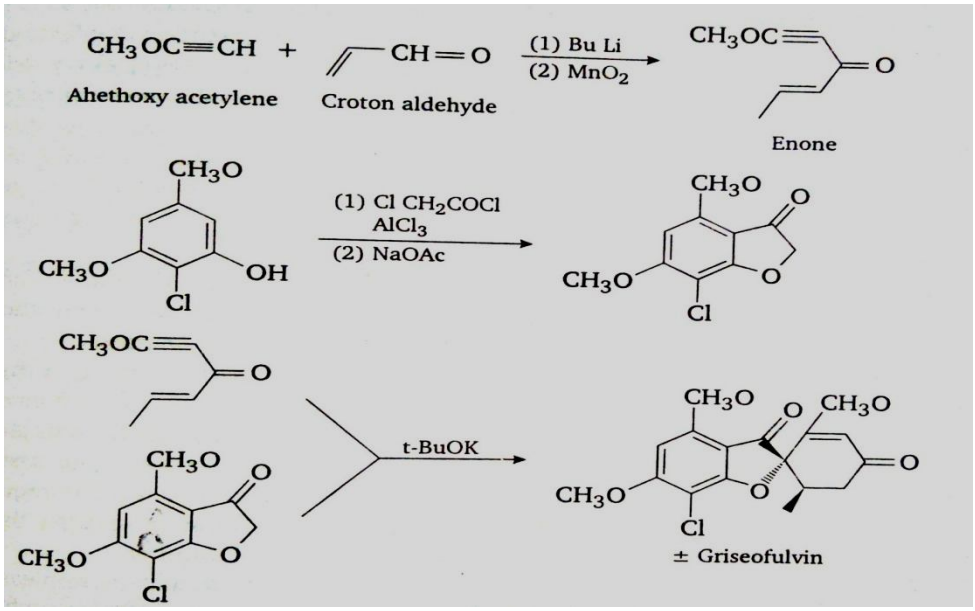
Structure:-

IUPAC 7-chloro- 

2,4,6-trimethoxy

6methylspiro[benzofuran-2(3H),1 (2)cyclohexen]-3,4-dione

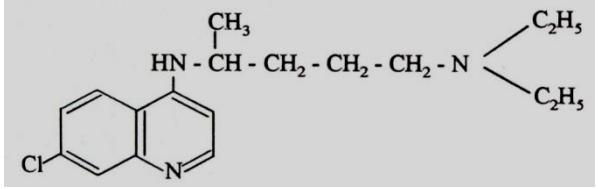
Synthesis:-



15.8 Chloroquin:- (Resochin)

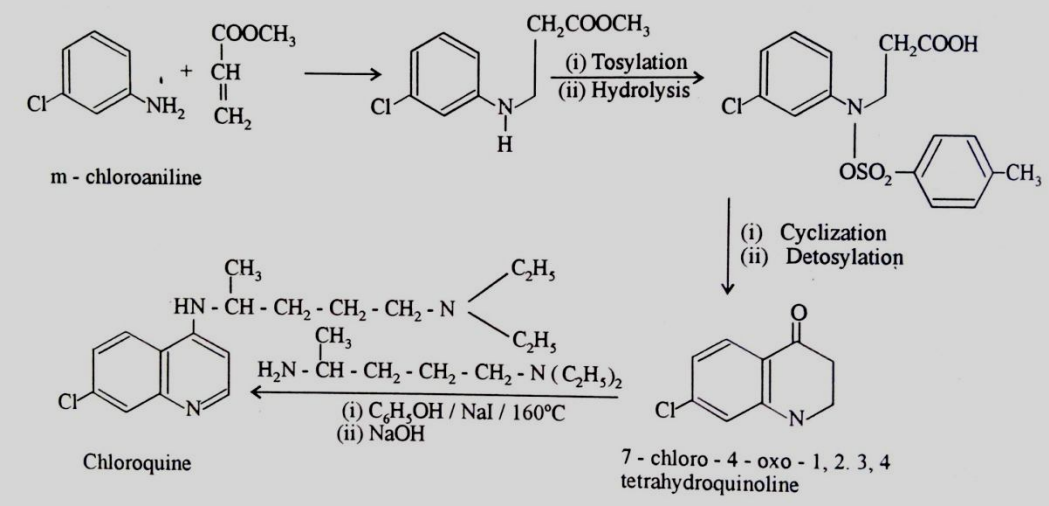
It is white crystalline powder with bitter taste, slowly decolourise on exposure to light.

Structure:-



IUPAC Name- 7-chloro-4-[4-(diethyl amino)-1-methylbutylamino] quinoline.

Synthesis:-



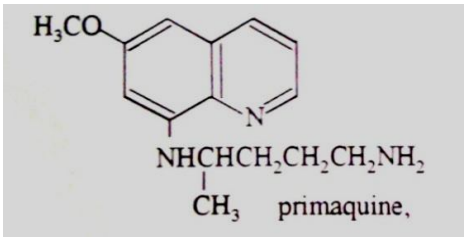
Chloroquin is made by reacting 4,7-dichloroquinoline with 4-dimethylamino-1-methylbutylamine at 180°c.

The drug completely cures faciparum malaria. the major metabolite, monodesethyl chloroquine has also antimalarial activity.

15.9 Primaquin

It is an antimalarial drug which is specifically kills the primary exoerythrocytic stages 0f p. vivax, p. falciparum, p. Malariae and p. Ovale, and the secondary exoerythrocytic form of all except p. falciparum, which has no secondary forms.

Structure of Primaquin:-



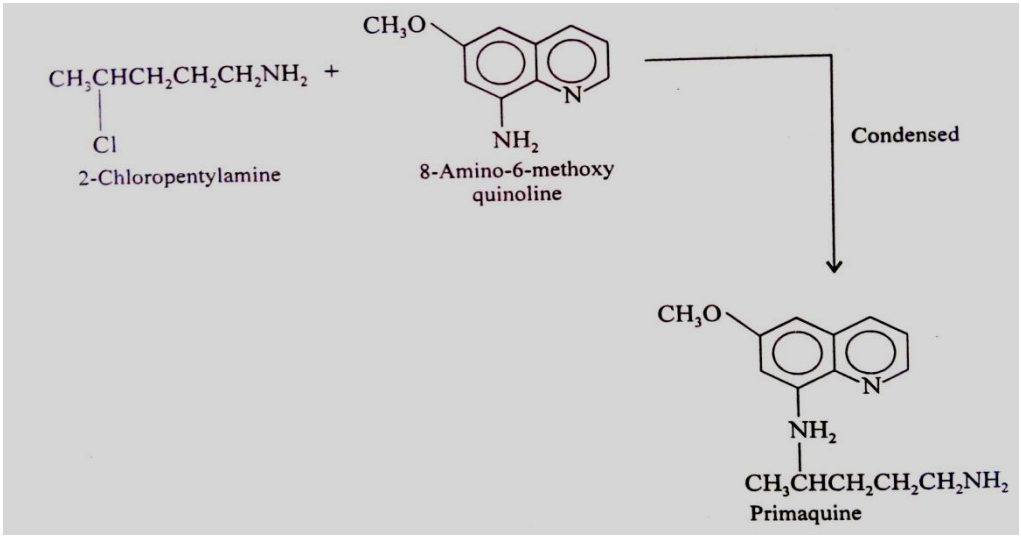
IUPAC Name-8-[(4-amino-1-methylbutyl)amino]-6-methoxy quinoline

Synthesis :-

The synthesis of primaquin may be accomplished by either of the two following methods:-

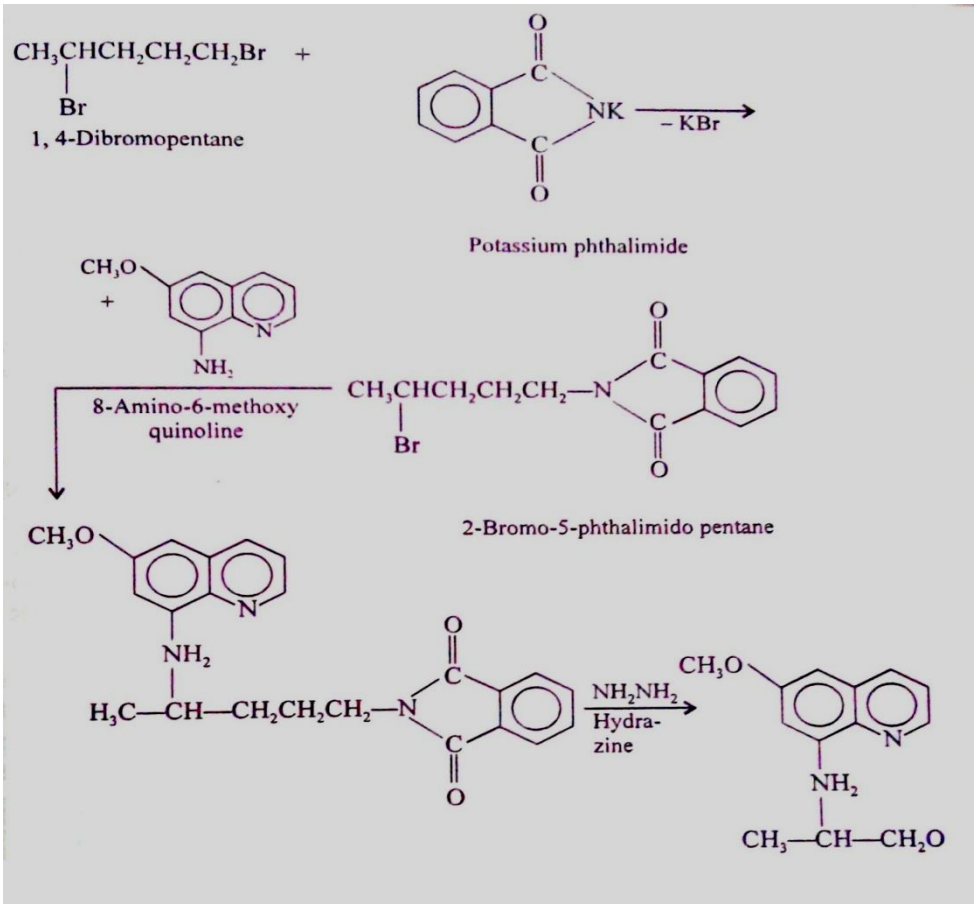
Method I- From 2-chloropentylamine:-

It may also be prepared by the condensation of 2-chloro-pentylamine with 8-amino-6-methoxy quinoline to obtain primaquin base which on treatment with bimolar quantity of phosphoric acid yield primaquin.

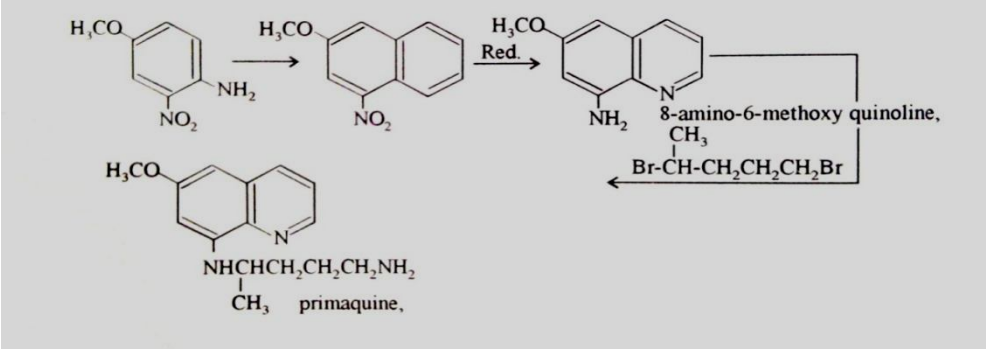


Method-II Elderfield's from 1,4-dibromopentane:-

2-bromo-5-phthalimido pentane is prepared by the interaction of 1,4-dibromopentane with potassium phthalimide, which on reaction with 8-amino-6-methoxy quinoline yields the condensed product. Further treatment with hydrazine eliminates the phthalimido residue and yields the primaquin base which on reaction with a double molar quantity of phosphoric acid forms the official compound.



Method- III



15.10 Summary

The antimicrobial activity of sulphonamides extends to many microbial species having a folic acid pathway which consist of many gram positive and gram negative cocci and bacilli, fungi, mycobacteria, some large viruses, and protozoa.

The use of sulphonamides includes the treatment of acute and chronic gram-positive and gram negative bacterial infections, consisting of leprosy, malaria, trachoma, toxoplasmosis and cocci diosis

Fluconazole can be administered orally as well as intravenously in severe infections. It is longer acting, safer and more efficacious drug. Econazole has broad spectrum antifungal activity.

Chloroquin is the drug of all types of malaria except that caused by P. falciparum, because P. falciparum has acquired significant resistance against chloroquin.

15.11 Glossary

- **Gangrene: -** is a potentially life-threatening condition that arises when a considerable mass of body tissue dies (necrosis). This may occur after an injury or infection, or in people suffering from any chronic health problem affecting blood circulation.
- **Eosinophillia: -** is a condition in which the eosinophil count in the peripheral blood exceeds 4.5×108/L.
- **Meningitis: -** is an acute inflammation of the protective membranes covering the brain and spinal cord, known collectively as the meninges.

The inflammation may be caused by infection with viruses, bacteria, or other microorganisms, and less commonly by certain drugs.

- **Mitotic Spindle: -** The collectively term for all the spindle fibers that form during mitosis. It is a spindle-shaped structure that develops outside the nucleus during mitosis.
- **Prostatitis: -** is inflammation of the prostate gland. Prostatitis is classified into acute, chronic, asymptomatic inflammatory prostatitis, and chronic pelvic pain syndrome.
- **Saponification: -** Saponification is a process that produces soap, usually from fats and lye.
- **Urticaria: -** commonly referred to as hives, is a kind of skin rash notable for pale red, raised, itchy bumps. Hives may cause a burning or stinging sensation. They are frequently caused by allergic reactions.

15.12 Review questions/comprehensive question-

1. What are local anti-infective agents with examples? Give synthesis of any one local anti infective agent.
2. Give the different methods for the synthesis of primaquin as local anti-infective agent.
3. Give the synthesis of chloroquin with its structure.
4. What are sulphonamides? Which type of sulphonamides are used as local anti infective agent.
5. Give the synthesis of Mafenide.
6. Give the synthesis of any one local anti infective agent belongs to sulphonamide category.
7. Give the structure and synthesis of chloroquin.
8. Give the synthesis of following-
    - Sulphacetamide sodium.
    - Econazole.
    - Flconazole
    - Chloroquin.

15.13 References and suggested readings

- Textbook of Organic Medicinal and Pharmaceutical Chemistry (11th ed.)- Wilson and Gisvold's.
- Introduction to medicinal chemistry, Alex Gringauz-1996
- A Book of Medicinal Chemistry; (second edition) D. Srirm and P. Yogeswari, published by pearson education.
- A Book of medicinal chemistry volume II; (first edition 2008-2009) Dr. Anees Ahmad Siddiqui, Seemi Siddiqui, Dr. R. Rajesh, published by birla publication pvt. Ltd.
- A Text book of medicinal chemistry; (third edition 2003) P. Parimoo, published by CBS publication.
- A Text book of medicinal chemistry (*synthesis and Biochemical Approach*) Vol. II; (third edition 2004, reprint 2006) Surendrs N. Pandeya, published by S. G. Publisher.
- Principle of medicinal chemistry-vol. I; (eighteenth edition, september 2007, reprint edition- March 2008, August 2008,) Dr. S. S. Kadam, Dr. K. R. Mahadik, Dr. K. G. Bothara, published by- Nirali prakashan.
- Medicinal Chemistry; (revised third edition-2005, reprint-2008) Ashutosh Kar, published by New age international (P) Ltd.
- Essential of medical Pharmacology; (seventh edition-2013) K. D. Tripathi, Published by- JAYPEE brothers medical publishers (P) Ltd.

Unit-16

Psychoactive Drugs

Structure of Unit:

16.1 Objectives

In this unit the students will be able to understand

- Meaning of Psychoactive Drugs
- Different types of Neurotransmitters
- General Mechanism of Neurochemical Transmission
- CNS depressants
- Different types of General Anesthetics
- Mechanism of action of Sedative – Hypnotics
- Mechanism of action of Anti-anxiety drugs
- Biochemical theories of affective disorders

16.2 Introduction: Psychoactive Drugs

Psychoactive or Psychotropic drugs are the agents which affectmental processes and are used to treat mental disorders.

Psychoactive drugs may be classified on the basis of primary use as following-

1-Antipsychotics (Neuroleptics/Ataractic/Major Tranquillizer)

2-Antidepressants (Thymoleptics)
3-Antianxiety drugs (Anxiolytics/Minor Tranquillizer)
4-Antimanic (Mood Stabilizer)

16.3 Neurotransmitters (Mediators)

These are chemical messengers through with nerve transmit their message across synapse and neuroeffector junction. Neurotransmitters are synthesized by nerve cells, actively transported along the axons and stored in the synaptic vesicles. They are released by exocytosis in response to the action potential and diffuse across the synaptic cleft. Classically, neurotransmitters fall into two major categories:

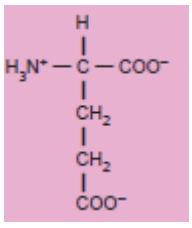
Excitatory- Ion channels are opened to permit net influx of positively charged ions, leading to depolarization with a reduction in the electrical resistance of the membrane; and

Inhibitory- Selective ion movements lead to hyper polarization, also with decreased membrane resistance.

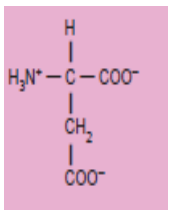
Classification-

1-Excitatory Neurotransmitter –

Glutamate

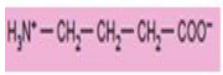


Aspartate

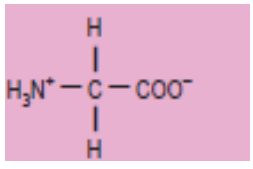


2-Inhibitory Neurotransmitter –

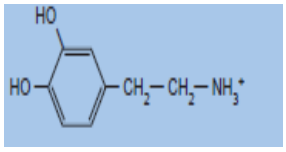
GABA



Glycine

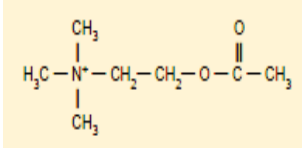


Dopamine

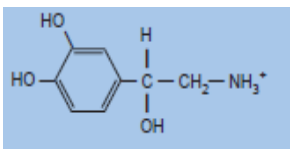


3-Excitatory and Inhibitory Neurotransmitter –

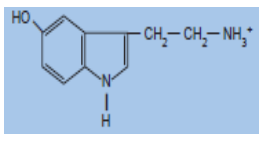
Acetylcholine



Serotonin



Noradrenaline



16.4 Neurochemical Transmission

Neurohumoral transmission implies that nerves transmit their message across synapses and neuroeffector junctions by the release of chemical messengers.

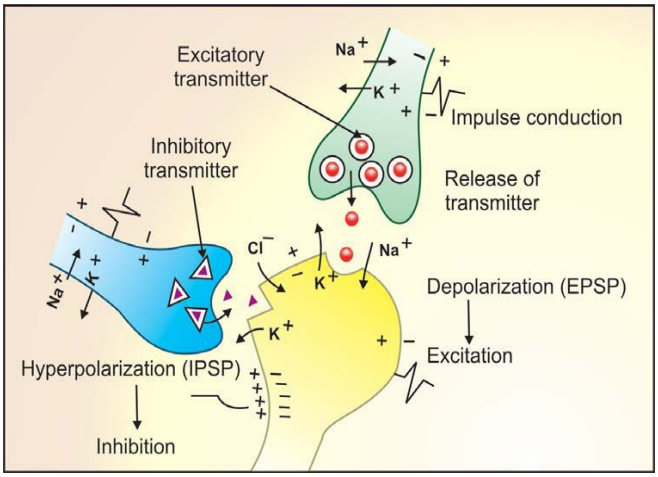


Fig.16.1: Diagrammatic representation of steps in excitatory and inhibitory Neurochemical transmission.

Excitatory postsynaptic potential (EPSP)

When an excitatory transmitter binds with specific receptor on post junctional membrane, the membrane permeability to Na^+ or Ca^{+2}increases, influx of cations causes depolarization followed by K+ efflux.

Inhibitory postsynaptic potential (IPSP)

When an inhibitory transmitter binds with specific receptor on post junctional membrane, the membrane permeability to K^+ or Cl^- increases, K^+ moves out and Cl^- in moves and hyper polarization occur.

Basic steps in Neurochemical transmission-

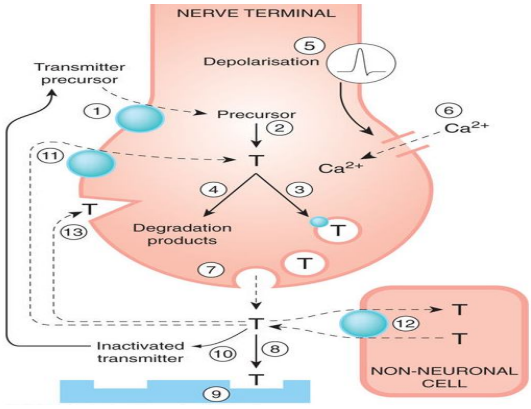


Figure16-2: Diagrammatic representation steps of Neurochemical transmission.

Neurochemical transmission steps are as followings-

- Uptake of precursors
- Synthesis of transmitter
- Uptake/Transport of transmitter into vesicles
- Degradation of surplus transmitter
- Depolarization by propagated action potential
- Influx of Ca2+ in response to depolarization
- Release of transmitter by exocytosis
- Diffusion to postsynaptic membrane
- Interaction with postsynaptic receptors
- Inactivation of transmitter
- Reuptake of transmitter or degradation products by nerve terminals
- Uptake of transmitter by non-neuronal cells; and interaction with presynaptic receptors

The transporters (11 and 12) can release transmitter under certain conditions by working in reverse.

Some neurotransmitters bind to their receptors and act quickly to open or close ion channels in the membrane. Others act more slowly via second-messenger systems to influence chemical reactions inside cells. The result of either process can be excitation or inhibition of postsynaptic neurons. Many neurotransmitters are also hormones released into the bloodstream by endocrine cells in organs throughout the body.

Acetylcholine-

It is released by many PNS neurons and by some CNS neurons. ACh is an excitatory neurotransmitter at some synapses, such as the neuromuscular junction, where the binding of ACh to inotropicreceptors opens cation channels. It is also an inhibitory neurotransmitter at other synapses, where it binds to metabotropic receptors coupled to G proteins that open K^+ channels. The enzyme acetyl cholinesterase inactivates ACh by splitting it into acetate and choline.

Glutamate (glutamic acid)

Excitatory synaptic transmission is mediated by glutamate, which is present in very high concentrations in excitatory synaptic vesicles. Glutamate is released into the synaptic cleft by Ca^{+2}-dependent exocytosis. The released glutamate acts on postsynaptic glutamate receptors and is cleared by glutamate transporters present on surrounding glia. In glia, glutamate is converted to glutamine by glutamine synthetase, released from the glia, taken up by the nerve terminal, and converted back to glutamate by the enzyme glutaminase.

Gamma amino butyric acid (GABA) and **glycine** are important inhibitory neurotransmitters. At many synapses, the binding of GABA to inotropic receptors opens Cl^-channels. GABA is found only in the CNS, where it is the most common inhibitory neurotransmitter. One-third of all brain synapses use GABA.

Like GABA, the binding of glycine to inotropic receptors opens Cl channels. About half of the inhibitory synapses in the spinal cord use the amino acid glycine; the rest use GABA.

Norepinephrine (NE)–

A smaller number of neurons in the brain use epinephrine as a neurotransmitter. Norepinephrine also serves as hormones. Cells of the adrenal medulla, the inner portion of the adrenal gland, release them into the blood.

Dopamine (DA)-

Brain neurons containing this neurotransmitter are active during emotional responses, addictive behaviors and pleasurable experiences. In addition, dopamine-releasing neurons help regulate skeletal muscle tone and some aspects of movement due to contraction of skeletal muscles. The muscular stiffness that occurs in Parkinson disease is due to degeneration of neurons that release dopamine.

Nor epinephrine and dopamine are classified chemically as catecholamines. Inactivation of catecholamine occurs via reuptake into synaptic end bulbs. Then they are either recycled back into the synaptic vesicles or destroyed by the enzymes. The two enzymes that break down catechol amines are catechol-O-methyltransferase (COMT) and monoamine oxidase (MAO).

Serotonin-

It is also known as 5-hydroxytryptamine (5-HT), concentrated in the neurons in a part of the brain called the raphe nucleus. It is thought to be involved in

sensory perception, temperature regulation, control of mood, appetite, and the induction of sleep.

16.5 CNS depressants

These are drugs which produce central nervous system depressant effect. Manifestations of CNS depression are-

- Drowsiness
- Sedation
- Hypnosis
- Disorientation
- Confusion
- Unconsciousness
- Coma
- Death

CNS depressants-

- General Anesthetics
- Sedative – Hypnotics
- Anti-anxiety drugs

16.6 General Anesthetics

Drugs produce reversible loss of all sensations and state of unconsciousness. The physiologic state induced by general anesthetics typically includes analgesia, amnesia, loss of consciousness, inhibition of sensory and autonomic reflexes, and skeletal muscle relaxation. An ideal anesthetic drug would induce loss of consciousness smoothly and rapidly, while allowing for prompt recovery of cognitive function after its administration is discontinued. The anesthetic technique will vary according to the proposed type of diagnostic, therapeutic, or surgical intervention.

Stages of Anesthesia-

- Stage of analgesia
- Stage of excitement
- Stage of surgical anesthesia
- Stage of medullary depression

CLASSIFICATION-

(I) INHALATIONAL

(A) GAS

- Nitrous oxide
- Ether

(B) VOLATILE LIQUIDS

- Halothane
- Enflurane
- Isoflurane
- Desflurane
- Sevoflurane

(II) INTRAVENOUS

(A) INDUCING AGENTS

- Thiopentone sod.
- Methohexitone sod.
- Propofol
- Etomidate

(B) SLOWER ACTING DRUGS

(1)Benzodiazepines

- Diazepam
- Lorazepam
- Midazolam

(2) Dissociative anaesthesia

- Ketamine

(3) Opioid analgesia

- Fentanyl

Mechanism of Action –

The main site of action of anesthetics is reticular formation, which normally maintains a state of consciousness. Most anesthetics depress reticular formation by enhancing the activity of inhibitory transmitters and blocking the activity of excitatory transmitters.

Ligand gated ion channels are the major targets of anesthetic action. The GABA-A receptor gated Cl^- channel is the most important of these. Many

inhalational anesthetics, barbiturates, benzodiazepines and propofol potentiate the action of inhibitory transmitter GABA to open Cl^- channels. These interact with its own specific binding site on the GABA-A receptor Cl^- channel complex, but none binds to the GABA binding site as such; though some inhaled anesthetics and barbiturates (but notbenzodiazepines) can directly activate channels.

Action of inhibitory transmitter which activates Cl^- channels in the spinal cord and medulla is augmented by barbiturates, propofol and many inhalational anesthetics. This action may block responsiveness to painful stimuli resulting in immobility of the anesthetic state. N_2O and ketamine do not affect GABA or glycine gated Cl^- channels. They selectively inhibit the excitatory NMDA type of glutamate receptor. This receptor gates mainly $Ca+^2$ selective cation channels in the neurons, inhibition of which appears to be the primary mechanism of anesthetic action of ketamine as well as N_2O. The volatile anesthetics have little action on this receptor.

INHALATIONAL ANAESTHETICS

Inhalational Gas

Nitrous oxide-

It is a colourless, odourless, noninflammable gas.

Nitrous oxide is a good analgesic

Onset of action is quick andsmooth

Recovery is rapid

Potency is low

Generally used as a carrier and adjuvant to other anesthetics

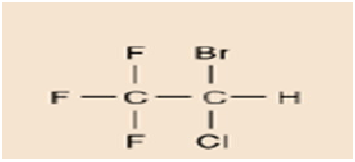
Volatile liquids

Ether-

(C_2H_5 - O - C_2H_5)

It is a highly volatile produces irritating vapours which are inflammable and explosive. Ether is a potent anesthetic, produces good analgesia. Now it is not used in developed countries due to unpleasant and inflammable properties.

Halothane

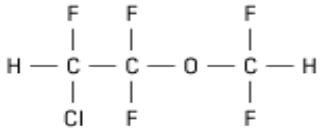


It is nonirritant, noninflammable and potent anesthetic.For induction 2-4% and for maintenance 0.5-1% is delivered by the use of a special vaporizer. It is currently one of the most popular anesthetic suitable for children as well as adults.

Enflurane-

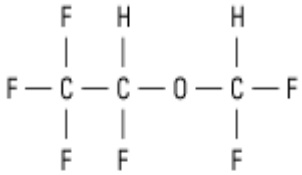
This is the substitute of halothane with rapid acting property. It is replaced by isoflurane due to its tendency to provoke seizures.

Isoflurane-



Isoflurane is an isomer of Enflurane. It has similar properties, but more potent, more volatile and less soluble in blood. It produces relatively rapid induction and recovery and is administered through a special vaporizer, used for maintenance.

Desflurane-



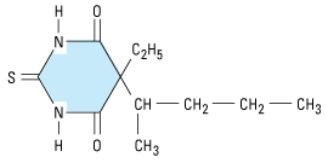
It is a newer congener of isoflurane. Thermostatically heated special vaporizer is used to deliver desflurane in the carrier gas (N2O + O2) mixture. It is a alternative to isoflurane for routine surgery as well, especially prolonged operations.

INTRAVENOUS ANAESTHETICS

INDUCING AGENTS

These are drugs which on i.v. injection produce loss of consciousness, are generally used for induction because of rapidity of onset of action. They also use to reduce the amount of maintenance anesthetic.

Thiopentone sodium-

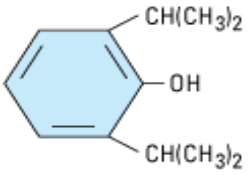
It must be prepared  freshly before injection. Thiopentone is a commonly used inducing

agent. It can be employed as the sole anesthetic for short operations that are not painful.

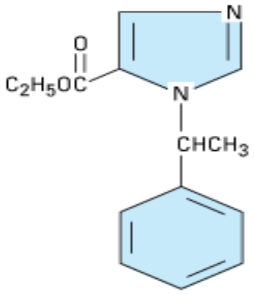
Methohexitone sodium- It is more potent and has a rapid action than thiopentone.

Propofol- It is an oily liquid employed as a 1% emulsion. Pain during injection can be minimized by combining with Lidocaine

Etomidate-



It is has a rapid onset and short duration of action. It is a poor analgesic and has not found much favour.



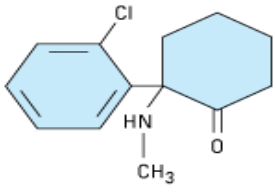
SLOWER ACTING DRUGS

Benzodiazepines (BZDs) –

BZDs are frequently used for inducing maintaining and anesthesia. BZDs are poor analgesics, an opioid or N_2O is usually added if the procedure `is painful. The action of BZDs can be reversed by flumazenil. They are useful for endoscopies, cardiac catheterization, fracture setting etc. they include diazepam, lorazepam etc.

Ketamine-

It is pharmacologically related to the hallucinogen phencyclidine. The primary site of action is in the cortex and sub cortical areas. Ketamine has been used for short operations. It is good for repeated use; particularly suitable for burn dressing.



Fentanyl-

This is related to pethidine, short acting potent opioid analgesic given at the beginning of painful surgical procedures. It is frequently used to supplement anesthetics in balanced anesthesia.

16.7 Sedative – Hypnotics

Sedative – Drugs that reduce excitement and relax the individual (person) without producing sleep.

Hypnotics – Drugs that produce or maintains sleep, similar to normal sleep.

A drug in small dose may induce sedation while same drug in large dose may act as hypnotic.

CLASSIFICATION

1. Barbiturates

A. Long acting

- Phenobarbitone

B. Short acting

- Butobarbitone
- Pentobarbitone

C. Ultra-shortacting

- Thiopentone
- Methohexitone

2. Benzodiazepines

A. Hypnotic

- Diazepam
- Flurazepam
- Nitrazepam
- Alprazolam
- Temazepam
- Triazolam

B. **Antianxiety**

- Diazepam
- Chlordiazepoxide

- Oxazepam
- Lorazepam
- Alprazolam

C. Anticonvulsant

- Diazepam
- Lorazepam
- Clonazepam
- Clobazam

3. Newer nonbenzodiazepine hypnotics

- Zopiclone
- Zolpidem
- Zaleplon

Mechanism of action-

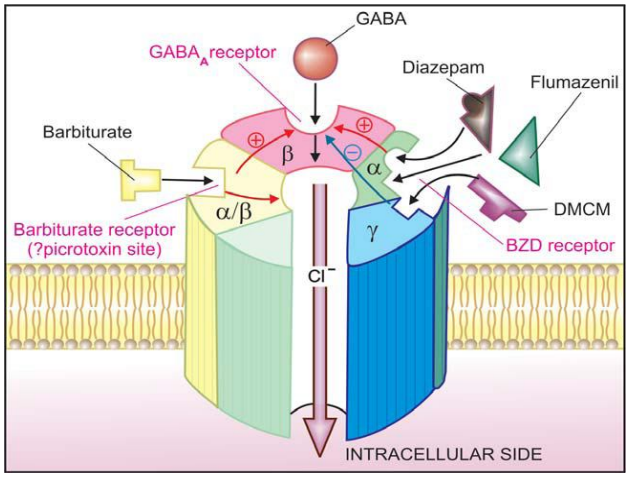
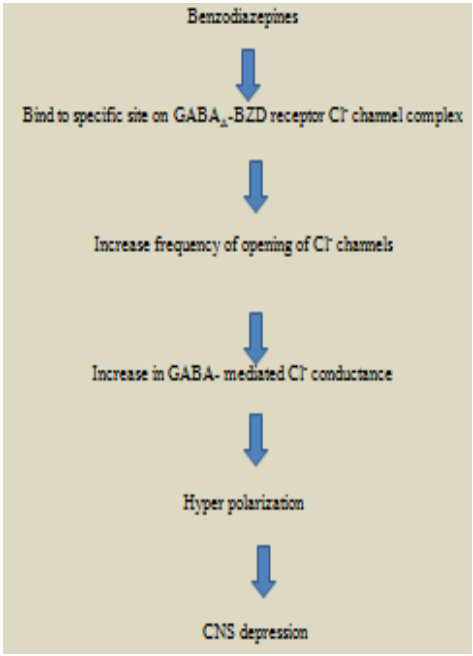


Fig. 16.3: Schematic depiction of $GABA_A$-benzodiazepine receptor-chloride channel complex

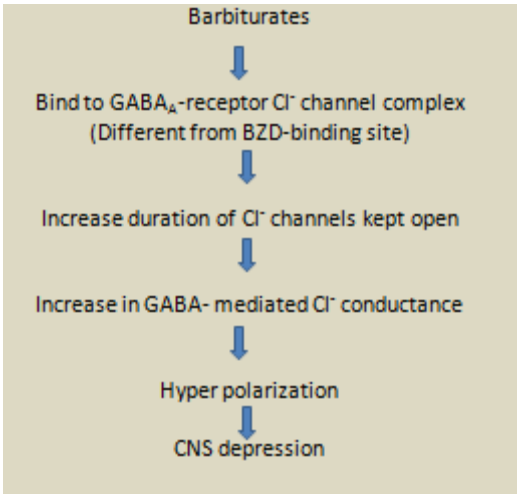
Benzodiazepines



Therapeutic uses-

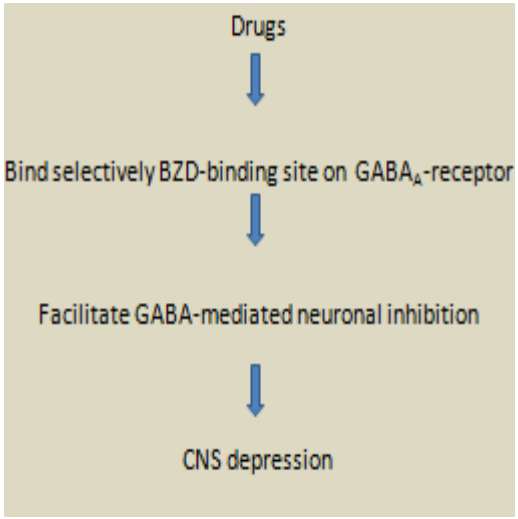
Benzodiazepines decrease time required to fall asleep. The total sleep time is increased. Now Benzodiazepines are preferred drugs for treatment of short-term insomnia. Long term use of Benzodiazepines not recommended but suitable for occasionally use by air travelers, shift workers.

Barbiturates-



Therapeutic uses- Barbiturates cause marked alteration in sleep latency and duration. At present Barbiturates not used for the treatment of insomnia.

Non- benzodiazepines-



16.8Antianxiety drugs

Anxiety – It is an emotional state associated with uneasiness, discomfort and fear about same defined or undefined threat.

Antianxiety drugs-

These are drugs, mostly mild CNS depressants, which produce a restful state of mind without interfering with normal mental or physical functions and control the symptoms of anxiety. The drugs differ markedly closely resemble to sedative-hypnotics.

CLASSIFICATION

1. **Benzodiazepines**
   - Diazepam
   - Chlordiazepoxide
   - Oxazepa
   - Lorazepam
   - Alprazolam
2. **Azapirones**
   - Buspirone
   - Gepirone

- Ispapirone

3. **Sedative antihistaminic**
   - Hydroxyzine
4. **β-blocker**
   - Propranolol

Benzodiazepines-

Benzodiazepines act on limbic system and facilitate inhibitory effect of GABA.These drugs are useful in short-term treatment of anxiety.

Azapirones-

It is a new class of antianxiety drugs. It does not interact with BZD receptor.The mechanism of anxiolytic action is not clearly known, but may be dependent on its selective partial agonistic action on 5-HT_{1A} receptors. By stimulating presynaptic 5-HT_{1A} autoreceptors, it reduces the activity of dorsal raphe serotonergic neurones. These mainly used in the treatment of generalized anxiety states. Its effect may take time to develop. So it is not effective in acute cases.

Sedative antihistaminic-

H_1 antihistaminic Hydroxyzine have selective anxiolytic action. It may be used in reactiveanxiety or that associated with marked autonomicsymptoms.

β- blocker-

Propranolol and other nonselective β-blockers are mainly used to reduce the symptoms of anxiety which are occur due to sympathetic over activity, potentiate anxiety symptoms. They may be used for performance/situational anxiety or as adjuvant to BZDs.

16.9 Neurochemistry of Mental Diseases

The term "psychosis" denotes a variety of mental disorders. The pathogenesis of these conditions is unknown. In 1880s some experiments had suggested a possibility of chemical factors affecting the pathology of psychoses. Research over the past decades has resulted in interesting theories involving amines and their receptors. It should be pointed out, that these concepts are still theories, not established explanations of the disease state. The basic concept is that neurotransmitter imbalances within the brain are the main causes of psychiatric disorders.

Biochemical theories of affective disorders-

The Amine Hypothesis

In the early 1950s after the introduction of reserpine, it became apparent that the drug could induce depression. Studies revealed that the principle mechanism of action of reserpine was to inhibit the neuronal storage of amine neurotransmitters such as serotonin and nor epinephrine. Reserpine induced depression and depleted stores of amine neurotransmitters; therefore, it was reasoned, depression must be associated with decreased functional amine-depended synaptic transmission. This idea provided the basis for what became known as the amine hypothesis of depression. Drugs that increased amine function in appropriate synaptic areas would relieve depression.

Both depression and mania are associated with changes in brain monoamines. The monoamine hypothesis proposes that, in depression, there is deficiency of the neurotransmitters nor adrenaline and serotonin in the brain which can be altered by antidepressants. Drugs that depression can modify affect amine storage, release or uptake. Thus the concentration of amines in nerve endings or at postsynaptic receptors is enhanced. In support of the monoamine hypothesis are the findings that amphetamines, which release presynaptic nor adrenaline and dopamine from stores and prevent their reuptake, have a weak antidepressant effect. The importance of serotonin is illustrated by the finding that depressed patients may exhibit down-regulation of some postsynaptic serotonin receptors.

Specific serotonin reuptake inhibitors, act predominantly by preventing serotonin reuptake and have more limited effects on noradrenaline reuptake. Tricyclic antidepressant in general inhibits nor adrenaline reuptake, but effects on serotonin reuptake vary widely; desipramine and protriptyline have minimal potential for raising serotonin concentrations. MAOIs increase the availability of nor adrenaline and serotonin by preventing their destruction by the monoamine oxidase type, a enzyme in the presynaptic terminal.

The amine hypothesis has provided the major experimental models for the discovery of new antidepressant drugs. As a result, all currently available antidepressants, excepts bupropion, are classified as having their primary actions on the metabolism reuptake, or selective receptor antagonism of serotonin, nor epinephrine, or both.

Schizophrenia

In schizophrenia, there appears to be over activity of dopamine systems in areas of the brain associated with complex mental and emotional functions. This is the dopamine hypothesis of schizophrenia.

Dopamine systems in the brain

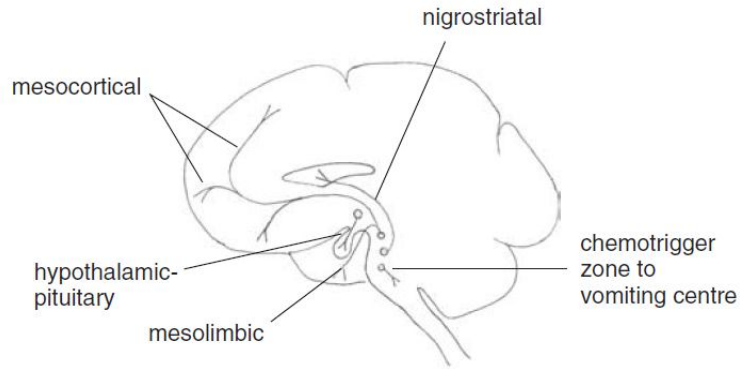


Fig.16.4: Dopamine pathway in brain

More recently, it has been suggested that schizophrenia may be a developmental disorder of the prefrontal cortex where there is actually a deficiency of dopamine, which leaves dopamine activity in the mesolimbic pathway unbalanced.

There is some evidence for the involvement of serotonin (5HT) and possibly other transmitters interacting with dopamine pathways. Most anti-psychotic drugs seem to work by blocking dopamine receptors in the brain, although some of the new atypical anti-psychotics also block serotonin receptors.

Dopamine receptors

There are two main types of dopamine receptor: the D1 type, which includes D1 and D5, and is excitatory; and the D2 type, which includes D2, D3 and D4, and is inhibitory. The significance of different types of dopamine receptors is still unclear, but drugs that are effective in schizophrenia appear to have an affinity for D2 type receptors.

The Dopamine Hypothesis of Schizophrenia:

Based on the efficacy of antipsychotic drugs, efforts continue to link the disorder with abnormalities of amine neurotransmitter function, especially that of dopamine. The dopamine hypothesis for schizophrenia is the most fully developed of several hypotheses and is the basis for much of the rationale for

drug therapy. Several evidences suggests that excessive dopaminergic activity plays a role in the disorder:

- Many antipsychotic drugs strongly block postsynaptic D2 receptors in the central nervous system, especially in the mesolimbic-frontal system.
- Drugs that increase dopaminergic activity, such as levodopa (a precursor), amphetamines (releasers of dopamine), and apomorphine (a direct dopamine receptor agonist), either aggravate schizophrenia or produce psychosis in some patients.
- Dopamine receptor density has been found postmortem to be increased in the brains of schizophrenics who have not been treated with antipsychotic drugs.
- Positron emission tomography (PET) has shown increased dopamine receptor density in both treated and untreated schizophrenics when compared with such scans of non schizophrenic persons.
- Successful treatment of schizophrenic patients has been reported to change the amount of homovanillic acid (HVA), a metabolite of dopamine, in the cerebrospinal fluid, plasma, and urine.

The dopamine hypothesis is far from complete, however. If an abnormality of dopamine physiology were completely responsible for the pathogenesis of schizophrenia, antipsychotic drugs would do a much better job of treating patients—but they are only partially effective for most and ineffective for some patients. The cloning and characterization of multiple dopamine receptor types may permit more direct testing of the dopamine hypothesis if drugs can be developed that act more selectively on each receptor type. The traditional antipsychotics bind D2 50 times more avidly than D1 or D3 receptors. Until recently, the main thrust in drug development was to find agents that were more potent and more selective in blocking D2 receptors. The fact that several of the atypical antipsychotic drugs have much less effect on D2 receptors and yet are effective in schizophrenia has redirected attention to the role of other dopamine receptors and to no dopamine receptors, especially serotonin receptor subtypes that may mediate synergistic effects or protect against the extra pyramidal consequences of D2 antagonism.

Current brain imaging and biochemical studies in patients do not support a single biologic abnormality as common to most depressions. Rather, prevailing hypotheses emphasize an underlying role for several brain circuits that have a

propensity to become dysfunctional, in individuals with a range of genetic predispositions. It is likely that several pathophysiologic processes will ultimately be identified to account for the presentation of mental disorders. As a result of these considerations, the direction of research has changed to a greater focus on compounds that may act on several transmitter-receptor systems. The great hope is to produce drugs with greater efficacy and fewer adverse effects, especially extra pyramidal toxicity.

16.10 Summary

Drugs acting in the central nervous system are the most widely used group of pharmacologic agents. The mechanisms by which various drugs act in the CNS have not always been clearly understood. In the last three decades, however, dramatic advances have been made in the methodology of CNS pharmacology. Neurotransmitters mediate signal from one neuron to another. Neurotransmitters are either excitatory or inhibitory. Pharmacological agents which are use in the treatment of mental disorders called Psychoactive Drugs. The physiologic state induced by general anesthetics typically includes analgesia, amnesia, loss of consciousness, inhibition of sensory and autonomic reflexes, and skeletal muscle relaxation. General anesthetics are usually administered by intravenous injection or by inhalation. Both the inhaled and the intravenous anesthetics can depress spontaneous and evoked activity of neurons in many regions of the brain. The sedative-hypnotic class cause sedation or to encourage sleep. Anxiety is an emotional state, unpleasant in nature, associated with uneasiness. The anxiolytic and sedative-hypnotics drugs are closely resemble. There are some biochemical hypotheses which explain the role of neurotransmitters in mental disorders pathogenesis.Neurotransmitter functions provide potential targets for pharmacologic therapy.

16.11 Glossary

- Ataractic: Drugs producing state of mental calm and tranquility.
- Coma: A state of unconsciousness from which one cannot be aroused.
- Confusion: Not being aware of or oriented to time, place or self
- Disorientation: Inability to estimate direction or location, or to be cognizant of time or of persons.
- Drowsiness: The of almost falling asleep

- Neuroleptics: Drug producing symptoms similar to central nervous system (CNS) depressant diseases.
- Thymoleptics: Synonym of antidepressant drugs.
- Tranquilizers: Drugs reducing mental tension without affecting normal function.

16.12 Review questions / Comprehensive Questions

1. Define and classify Psychoactive Drugs.
2. Classify neurotransmitters and write briefly about Neurochemical transmission.
3. Define and classify CNS depressant drugs.
4. Explain the mechanism of action of followings-
   A. General Anesthetics
   B. Sedative – Hypnotics
   C. Anti-anxiety drugs
5. Write in detail about biochemical theories of affective disorders.
6. Write short note on-
   A. Inhalational anesthetics
   B. Dopamine Hypothesis of Schizophrenia

16.13 References and Suggested readings

- Basic & Clinical Pharmacology (9th ed.)- Bertram G.Katzung (McGraw Hill Publisher) 2004.
- Principles of medicinal chemistry, volume II-Dr. S.S. Kadam, Dr. K.R. Mahadik, Dr. K.G. Bothara (Niraliprakashan) 2007.
- Pharmacology (5th ed.)- H.P Rang and M.M Dale (Elsevier publisher) 2003.
- Essential of Medical Pharmacology (7th ed.)- KD Tripathi (JAYPEE Publisher) 2013.

Unit-17

Antipsychotic Drugs

Structure of Unit:

17.1 Objectives

In this unit the students will be able to understand

- Meaning of Psychosis
- Symptoms of Schizophrenia
- Neruoleptic Drug Classes and Mechanism of Action
- Characteristics of different Neruoleptic Drugs
- Meaning and Types of Depression
- Treatment of Depression
- Serendipity and Drug Development
- Stereochemistry of Psychotropic Drugs

17.2 Introduction:

The term psychosis refers to a variety of mental disorders characterized by one or more of the following symptoms: diminished and distorted capacity to process information and draw logical conclusions, hallucinations, delusions, incoherence or marked loosening of associations, catatonic or disorganized behavior, and aggression or violence.

- Cognitive disorders (Acute and chronic organic brain syndromes)
- Functional disorders
    i. Schizophrenia (split mind)
    ii. Paranoid states

17.3 Schizophrenia – (split mind)

It is a chronic disorder, typically arising in early adulthood and progressing throughout the rest of the life.
The symptoms of schizophrenia are followings-
Positive symptoms– Not normally found in healthy individuals.

- Hallucinations
- Delusions
- Thought disorder

Negative symptoms– Normally present in healthy individuals.

- Impoverishment of thought
- Blunted Emotion
- Attention deficit
- Lack of motivation or initiative

17.4 Neruoleptic- Antipsychotic Drugs

The neruoleptic drugs (antipsychotic drugs or major tranquilizers) are used in the treatment of all type of psychosis, especially schizophrenia but are also effective in some other psychotic states, such as manic states with psychotic symptoms such as grandiosity or paranoia and hallucinations, and delirium.
CLASSIFICATION –
1-Phenothiazines

a. **Aliphatic side chain**
    - Chlorpromazine
    - Triflupromazine
b. **Piperidine side chain**
    - Thioridazine
c. **Piperazine side chain**
    - Trifluoperazine

- Fluphenazine

2-Butyrophenones –

- Haloperidol
- Trifluperidol

3-Thioxanthenes-

- Flupenthixol

4-Other heterocyclic's-

- Pimozide
- Loxapine

5-Atypical antipsychotics

- Clozapine
- Risperidone
- Olanzapine
- Quetiapine
- Aripiprazole
- Ziprasidone

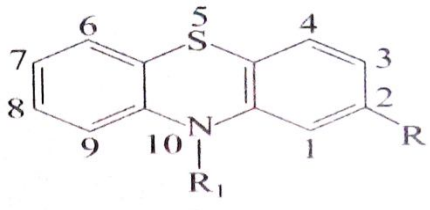
Mechanism of action

The antipsychotic actions of neruoleptic drugs appear to reflect a blockade at dopamine and/or serotonin receptors. All of the older and most of the newer neruoleptic drugs block dopamine receptors in the brain and the periphery. The neruoleptic drugs bind to these receptors to varying degrees. Most of the newer atypical agents appear to exert part of their unique action through inhibition of serotonin receptors (5-HT), particularly 5-HT_{2A} receptors.

17.5 Distinctive Features of Neuropleptics

Antipsychotic drugs differ in potency and in their propensity to produce different effects.

1-Phenothiazines



(A) Aliphatic side chain

Chlorpromazine (Prototype drug)

R= Cl

$R_1 = (CH_2)_3N(CH_3)_2$

In schizophrenia patients CPZ

- Reduces agitation and aggressiveness
- Reduces spontaneous movements
- Suppresses hallucinations and delusions
- Corrects disturb thought and behavior
- Relives anxiety

TriFlupromazine

R= CF_3

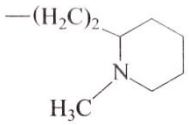
$R_1 = (CH_2)_3N(CH_3)_2$

- More potent than CPZ
- Used mainly as antiemetic

(B) Piperidine side chain

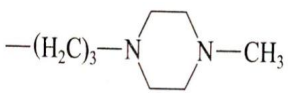
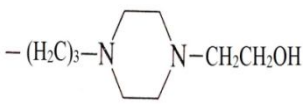
Thioridazine

R = SCH_3

R_1 = 

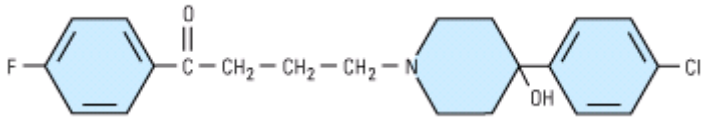
- Low potency
- Central anticholinergic action
- Risk of eye damage limits long-term use

(C) Piperazine side chain

	R	**R_1**
Trifluoperazine	**CF_3**	
Fluphenazine	**CF_3**	

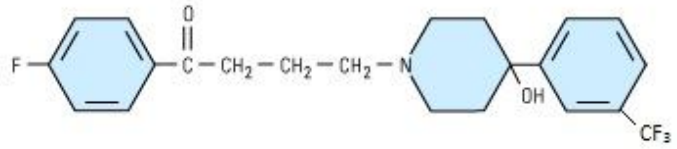
These are high potency piperazine side chain phenothiazines. They have minimum autonomic actions. Extra pyramidal side effects are marked.

2-Butyrophenones –
Haloperidol



- Widely used Potent antipsychotic drug
- Does not cause weight gain
- Used for acute schizophrenia, acute mania, senile psychoses.

TriFluperidol

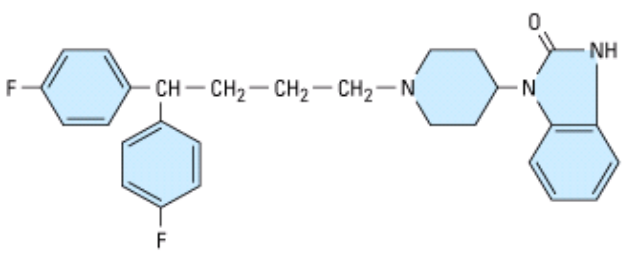


- Pharmacologically similar to haloperidol
- Slightly more potent

3-Thioxanthenes-
Flupenthixol

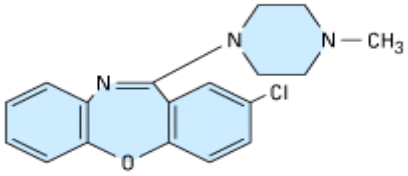
- It is less sedating than CPZ
- Indicated in schizophrenia and other psychoses
- Infrequently used now

4-Other heterocyclic's-
Pimozide



- Long duration of action
- Preferred for maintenance therapy

Loxapine

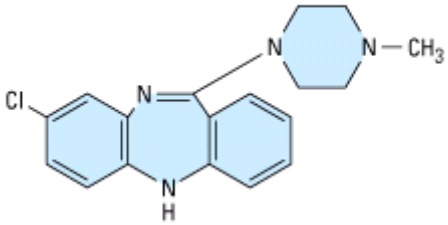


- Antipsychotic activity like CPZ
- Quick and short lasting actions

5-Atypical Antipsychotics

- These have weak D2 blocking activity
- Potent 5-HT_2 antagonistic activity
- Improve the impaired cognitive function in psychotics.
- Minimal extra pyramidal side effects

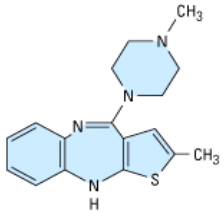
Clozapine



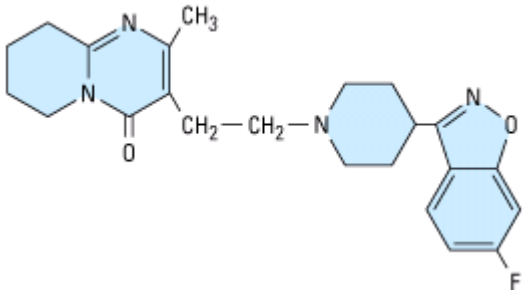
- Mainly block 5-HT_2 receptors
- Weak D_2 blocking activity
- Suppresses both positive and negative symptoms of schizophrenia
- Used as a reserve drug in resistant schizophrenia

Olanzapine

Resembles clozapine in blocking multiple monoaminergic, muscarinic and H_1receptors. Beneficial in both positive and negative symptoms of schizophrenia. Approved for use in mania

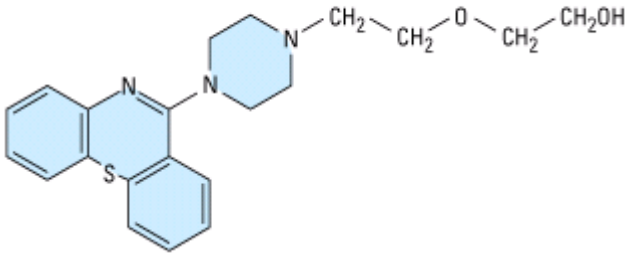


Risperidone



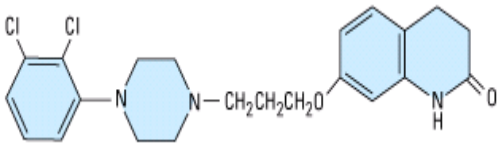
Block multiple monoaminergic, muscarinic and H_1receptors, This blockade may contribute to efficacy as well as side effects. Used for the treatment of schizophrenia and short term treatment of mania associated with bipolar disorder.

Quetiapine



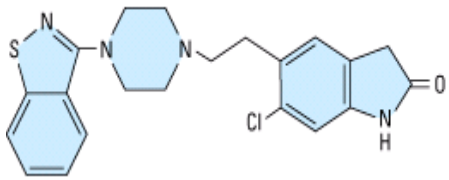
- Short-acting atypical antipsychotic
- Blocks 5-HT_1A, 5-HT_2, D_2, α_1, α_2 and H_1 receptors in the brain
- D_2 blocking activity is low
- Quetiapine has not been found to benefit negative symptoms of schizophrenia
- Used in mania/bipolar disorder

Aripiprazole



- Quite long-acting
- Partial agonist at $D_2$and 5-HT_{1A} receptor
- Antagonist at 5-HT_2 receptor
- Indicated in schizophrenia as well as mania and bipolar illness

Ziprasidone



- It is the newer atypical antipsychotic with combined D_2+ 5-HT $_{2A}$/ + H1 + α_1 blocking activity.
- In comparative trials, its efficacy in schizophrenia has been rated equivalent to haloperidol
- It is also indicated in mania

17.6 Depression

It is an affective disorder refers to a pathological change in mood state. Depression is characterized by sad mood, loss of interest and pleasure worthlessness, guilt, physical and mental slowing, self destructive thoughts etc. Depression is a heterogeneous disorder that has been characterized in a variety of ways. A simplified classification based on presumed origin is as follows:

1- Brief reactive or secondary depression (most common), occurring in response to real stimuli such as grief, illness, etc.

2- Melancholic and recurrent depression, a genetically determined biochemical disorder manifested by an inability to experience ordinary pleasure or to cope with ordinary life events; and

3-Depression associated with bipolar affective (manic-depressive) disorder.

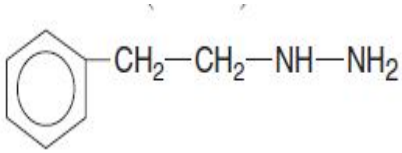
17.5 Antidepressant Drugs

These drugs enhance mood in depressive state and increased output of behavior.

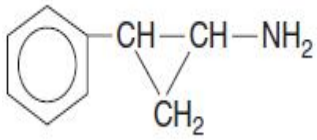
CLASSIFICATION

I. Reversible inhibitors of MAO-A (RIMAs)

Phenelzine

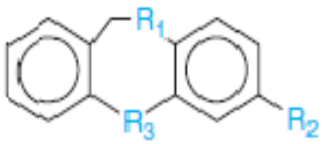


Tranylcypromine



II. Tricyclic antidepressants (TCAs)

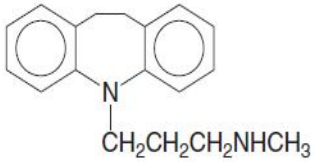
A. NA + 5-HT reuptake inhibitors



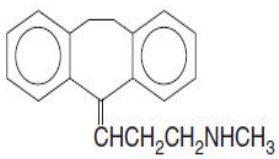
DRUG	R_1	R_2	R_3
Imipramine	C	H	$N(CH_2)_3N(CH_3)_2$
Amitriptyline	C	H	$C=CH(CH_2)_2N(CH_3)_2$
Clomipramine	C	Cl	$N—(CH_2)_3N(CH_3)_2$
Doxepin	O	H	$C=CH(CH_2)_2N(CH_3)_2$

B. Predominantly NA reuptake inhibitors

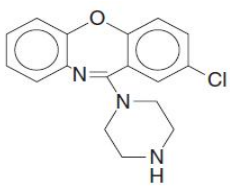
Desipramine



Nortriptyline

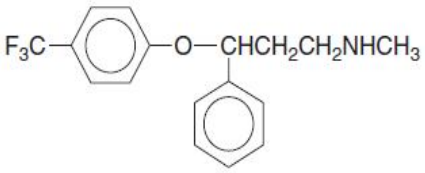


Amoxapine

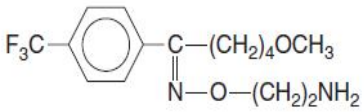


III. Selective serotonin reuptake inhibitors (SSRIs)

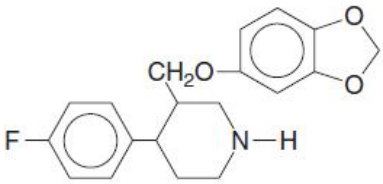
Fluoxetine



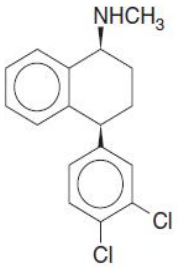
Fluvoxamine



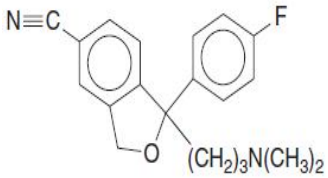
Paroxetine



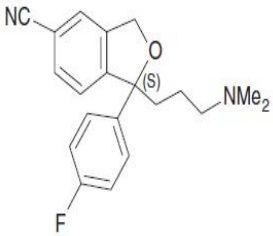
Sertraline



Citalopram

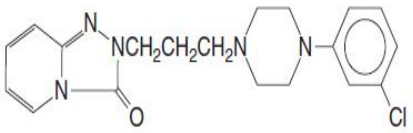


Escitalopram

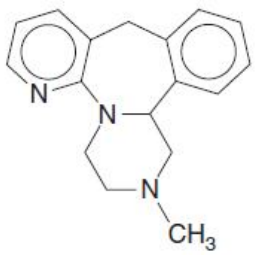


IV. Atypical antidepressants

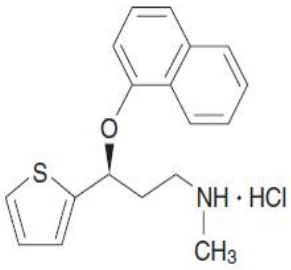
Trazodone



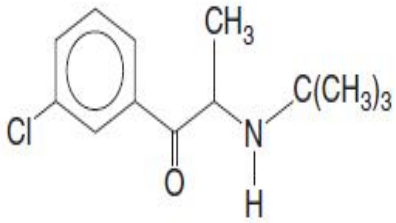
Mirtazapine



Duloxetine



Bupropion



I. Reversible inhibitors of MAO-A (RIMAs)

MAO is a mitochondrial enzyme regulates the metabolic degradation of catecholamine's, 5-HT, and other endogenous amines in the CNS and peripheral tissues. Hepatic MAO has a crucial defensive role in inactivating circulating monoamines and compounds. Two isoenzyme forms of MAO have been identified, MAO-A and MAO-B.

Dopamine is degraded equally by both isoenzymes. Inhibition of this enzyme system by MAO inhibitors causes a reduction in metabolism and a subsequent increase in the concentrations of biogenic amines. Selective MAO-A inhibitors are more effective in treating major depression than type MAO-B inhibitors.

II. Tricyclic Antidepressants (TCAs)

Mechanism of action

The TCAs and related drugs inhibit active reuptake of biogenic amines into their respective neurons and thus potentiate them. Reuptake inhibition results in increased concentration of the amines in the synaptic cleft in the CNS and periphery.

III. Selective Serotonin Reuptake Inhibitors (SSRIs)

These drugs inhibit serotonin transporter (SERT) and block reuptake of 5-HT into the neuron. This blockade increase the availability of 5-HT at receptors in the CNS and enhance serotoninergic activity.

IV. Atypical antidepressants

Trazodone –

- Block 5-HT reuptake and 5-HT_2 antagonist
- Block α_1-adrenergic receptors

Mirtazapine

- Blocks α_2-autoreceptors on noradrenergic neurons and heteroreceptors on 5-HT neuron increases NA and 5-HT release

Duloxetine

- SNRI - 'Serotonin and nor adrenaline Reuptake Inhibitor'

- It inhibits uptake of these amines into the neuron

Bupropion

- Inhibits reuptake of DA and NA into the neuron

17.8 Buturophenones– Serendipity And Drug Development

Serendipity (i.e. chance)

Frequently lead compounds are found as a result of serendipity. Serendipitous observation means that an agent developed for another purpose has a desirable but unexpected clinical effect.

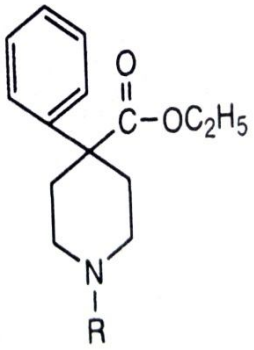
Psycho pharmacological drugs discover by serendipity

- Amphetamine
- Buspirone
- Carbamazejine
- Chlordiazepoxide
- Chlorpramine
- Clozapine
- Fluoxetine
- Haloperidol
- Imipramine
- Iproniazid
- Reserpine
- Valproic acid

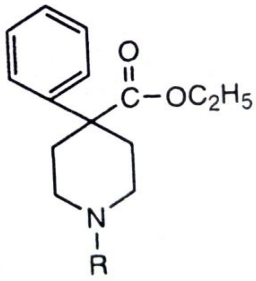
Buturophenones– Serendipity and Drug Development

The potent neuropleptics Buturophenones are classic examples of serendipitously discovered lead compound. Paul Jansen undertook a systemic study of the 4-phenylpiperidine molecule, particularly to evaluate the limit of N-substitution of normepridine.

R = CH_3

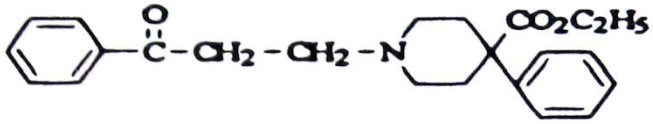


Replacement of methyl of meperidine with p-aminophenylethyl moiety (anileridine) and a phenylaminopropyl group (piminodine) afforded active analgesics property.

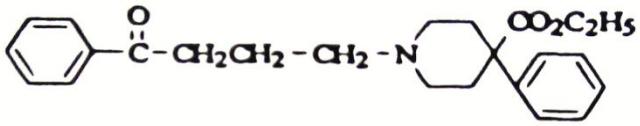


Piminodine R = $CH_2CH_2CH_2NHC_6H_5$
Anileridine R = $CH_2CH_2\,pC_6H_4NH_2$

First molecular modification produced a normepridine propiophenone analog which is 100 times more potent than morphine.

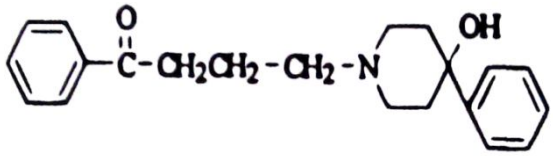


Lengthening the chain by one CH_2 produced a butyrophenone which show analgesic potency more than morphine but also surprisingly produce CPZ-like activity in animal models at low level.

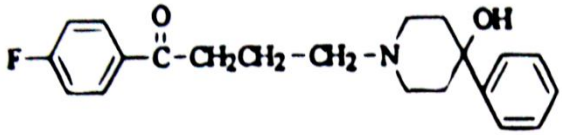


Replacement of

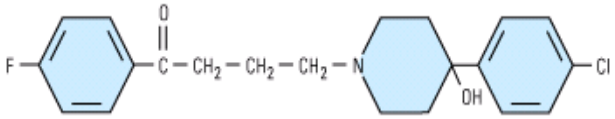
the 4-carboethoxy group with OH led to disappearance of analgesic activity while show neruoleptic activity comparable to CPZ.



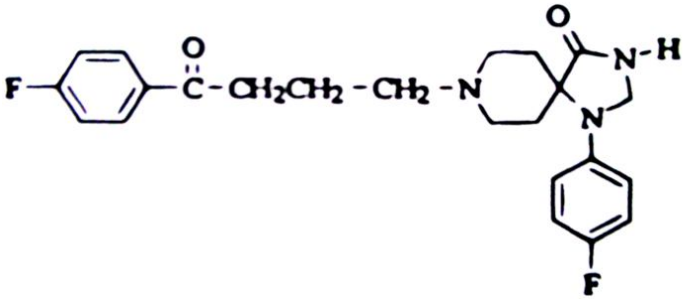
The next step was to determine the proper positions (Para) for halogen substituent in both aromatic rings to increase the effectiveness of above compound. First halogen substitution was on ketonic phenyl ring p-F atom.



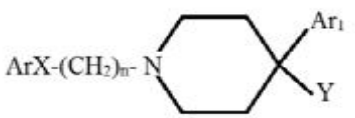
Halogen substitution on both rings- Haloperidol



Further modification of the 4-piperidine position resulted in spirotriazolodecanes carrying a keto function where the 4-OH was on earlier compounds. Here potencies exceed CPZs by several thousand times.



Structure Activity Relationship (SAR)

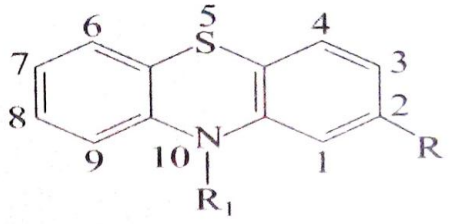


1. When Ar is an aromatic ring system, antipsychotic activity is seen, substitution of fluoro at Para position increases the activity.
2. When X = carbonyl (c=o), optimum antipsychotic activity is occur.
3. When n=3, optimum activity is seen, change in length of chain (longer or shorter) decreases the activity.
4. Aliphatic amino nitrogen when incorporated into cyclic form maximum activity is seen.
5. Ar_1 is an aromatic and required. It should be directly attached to forth position or occasionally separated from it by one interventing atom.
6. The group Y can vary and help in activity.

17.9 Stereo chemical aspects of psychotropic Drugs

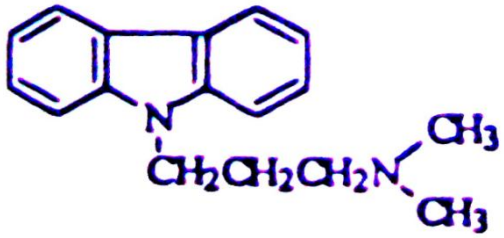
Antipsychotic drugs have various neuro pharmacological properties as a result of their structural diversity. The chemical structure of antipsychotic drugs plays an essential role in their receptor-binding affinities in the brain and their therapeutic profiles and metabolic side effects in schizophrenia treatment.
In the determination of type of psychotropic activity obtained stereo chemical and other geometric factors are significantly important. The tricyclic 6-6-6 system of the phenothiazines shows that in planar ring systems optimal neruoleptic activity occurs.

$R = R_1 = H$

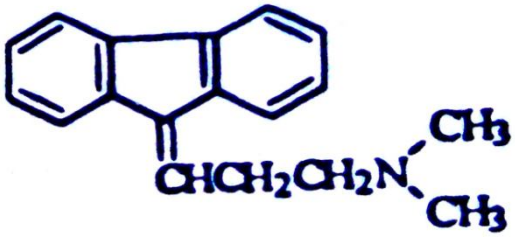


However, compounds with total rigid planarity, as in a 6-5-6 system are invariably inactive.

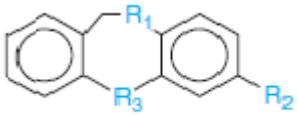
Examples-
Carbazole derivatives



Fluorene derivatives



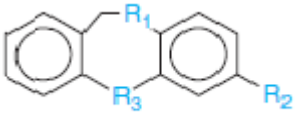
It might be postulated that neruoleptic receptors are planar but have rather strict size requirements for binding.
Ring systems with relatively few deviations from planarity, by angling of the atoms in the middle ring, begin to show some thymoleptic effects while still maintaining neruoleptic activity.
Examples-



Drug	R_1	R_2	R_3
Amitriptyline	C	H	C=CH $(CH_2)_2N(CH_3)_2$
Doxepin	O	H	C=CH $(CH_2)_2N(CH_3)_2$

More pronounced twisting of the ring system usually results in exclusive antidepressant action. A non planar angled thymoleptic receptor concept might be involved.

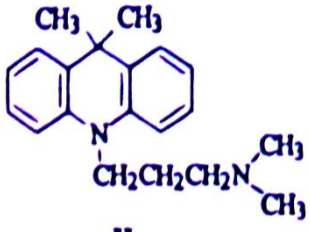
Examples-
Imipramine



R_1 = C
R_2 = H
R_3 = N—$(CH2)_3N(CH_3)_2$

Acridine derivatives

I



II

Compound I displays good antipsychotic activity.
Compound II, however, shows antidepressant activity similar to that of imipramine. The two additional methyl groups at position 9 result in a twisting of the three-ring system to a sufficient degree that the requirements of the postulated non planar angled antidepressant receptor site are accommodated.
Two situations arise in compound II that may explain the clinical observations.

First, it can be assumed that the planar acridine ring simply cannot accommodate two bulky methyl groups at the 9 position and maintain flatness. Thus a twisting must occur that results in loss of co planarity of the three rings.
Second, it can be argued that the bulk of the methyl groups prevent a good fit to the planar neruoleptic receptors.
The framework of the tricyclic system at a fundamental level, seen as angled or flexed, to varying degrees.

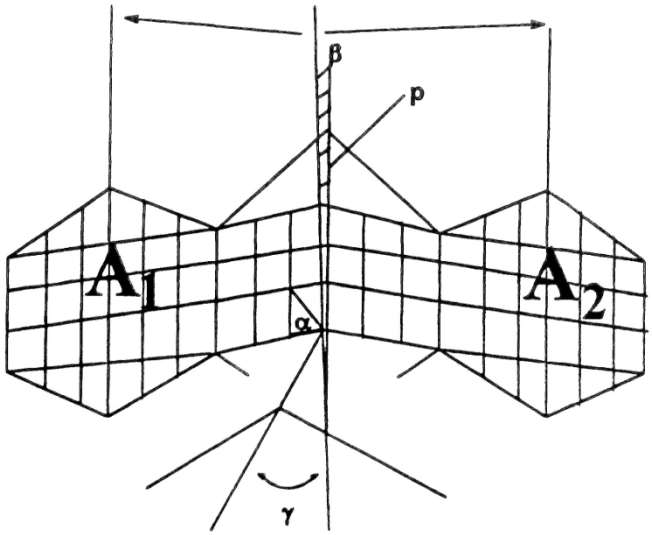


Fig.17.1: Steric parameters of tricyclic psychotropic drugs

Flexure represents the degree of non planarity between the A_1 and A_2 rings. Annulation is the angle at which these two rings attached to middle ring. Torsion is the degree to which the molecule is twisted out of symmetry.

When the angle of flexure is small the system is relatively flat and clinically neruoleptic. Further twisting out of the plane results in thymoleptic properties. Angles of annulation and torsion do not appear to have that much bearing on therapeutic effects.

The insufficient neuro-chemical knowledge is the main problem in development of safer and especially more effective compounds. Research is active in areas such as α-$_2$ adrenoceptors, several HT receptors, GABA receptors, lithium mimetic, certain anticonvulsant calcium antagonists, neuropeptides (e.g., endorphins, GH), and even corticosteroids.

17.10 Summary

The term "psychosis" denotes a variety of mental disorders. Schizophrenia is a particular kind of psychosis characterized mainly by a clear sensorium but a marked thinking disturbance. Major depression is one of the most common psychiatric disorders. The symptoms of depression are often subtle and unrecognized both by patients and by physicians.

The antipsychotic and antidepressant drugs affect cortical, limbic, hypothalamic, and brainstem mechanisms that are of fundamental importance in the regulation of arousal, consciousness, affect, and autonomic functions. Physiological and pharmacological modifications of these brain regions may have important behavioral consequences and useful clinical effects regardless of the underlying cause of any mental disorder.

17.11 Glossary

- **Bipolar disorder :** Bipolar disorder (manic depression) is a mental illness that brings severe high or low moods and changes in sleep, energy, thinking, behavior.
- **Catatonic :** It is characterized by marked lack of movement, activity, or expression.
- **Cognition :** It is the mental act or process by which knowledge is acquired, including perception, intuition, and reasoning.
- **Delusions :** It is the act of deluding or state of being deluded.

- **Extra pyramidal :** Extra pyramidal side effects are drug induced moment disorders such as tremor, akathisia, Parkinsonism etc.
- **Partial agonist :** Partial agonists are drugs that have only partial efficacy at the receptor.

17.12 Review questions / Comprehensive Questions

1. Classify antipsychotic drugs. Explain mechanism of action, effects and uses of chlorpromazine.
2. Write briefly on-
   a. Haloperidol
   b. Clozapine
   c. Risperidone
   d. Olanzapine
3. Classify antidepressants. Explain the mechanism of action of each class with example.
4. Write briefly on-

a. Reversible inhibitors of MAO-A (RIMAs)

b. Tricyclic antidepressants (TCAs)

5. Explain role of serendipity in finding of lead compounds with special emphasis on development of Buturophenones,
6. Explain that the chemical structure of antipsychotic drugs plays an essential role in their receptor-binding affinities and type of psychotropic activity obtained.

17.13 References and Suggested readings

- Essential of Medical Pharmacology (7th ed.)- KD Tripathi (JAYPEE Publisher) 2013.
- Basic & Clinical Pharmacology (9th ed.)- Bertram G.Katzung (Mc Graw Hill Publisher) 2004.
- The Pharmacological basis of Therapeutics (10th ed.)- Goodman & Gilman's (Mc Graw Hill Publisher) 2001.
- Foye's Principles of Medicinal Chemistry, David A. Williams, Thomas L. Lemke (Fifth edition) 2005, B.I. Publication and Pvt. Ltd.
- Introduction to medicinal chemistry: how drugs act and why by Alex Gringauz.

Unit-18

Synthesis Of Antipsychotic Drugs

Structure of Unit:

18.1 Objective

- In this chapter we discuss the drugs which are used for different type of Psychotic disorder.
- Here we study about the method of synthesis of different Antipsychotic drugs.
- Here we also explain the use and adverse effect of these drugs.

18.2 Introduction

These drugs are used for the treatment of major psychosis. They are also called as "major tranquilizers". Since they reduce agitation and disturbed behavior seen in schizophrenia. These drugs have the pharmacological property of antagonizing the action of dopamine and this is responsible for most of their effects on the nervous system.

Most of these agents have also antiemetic, sympatholytic and α-adrenergic blocking actions.

The most important type of psychosis are –

- Schizophrenic
- Affective disorders (eg. depression, mania)
- Organic psychoses.

Classification:-

[A]	Typical neuroleptics :–	Phenothiazines (eg. Chlorpromazine) Butyrophenones (eg. Haloperidol) Thioxathenes (eg. Fluperthioxol)
[B]	Atypical neuroleptics :-	Benzamides (eg. Sulpiride) Diphenylbutyl piperazines (eg. Pimozide) Dibenzyldiazepines (eg. Clozapine)

18.3 Diazepam

Diazepam is a benzodiazepine (ben-zoe-dye-AZE-eh-peens). It affects chemicals in the brain that may become unbalanced and cause anxiety.

Diazepam is used to treat anxiety disorders, alcohol withdrawal symptoms, or muscle spasms. Diazepam is sometimes used with other medications to treat seizures.

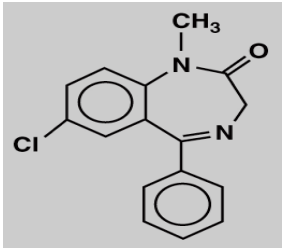
Do not drink alcohol while taking diazepam. This medication can increase the effects of alcohol.

Before taking this medicine :-

Should not use this medication if allergic to diazepam or similar drugs (Ativan, Klonopin, Restoril, Xanax, and others), or if have:

- Myasthenia Gravis (A Muscle Weakness Disorder);
- Severe Liver Disease;
- Narrow-Angle Glaucoma;
- A Severe Breathing Problem; Or
- Sleep Apnea (Breathing Stops during Sleep).

Structure of Diazepam

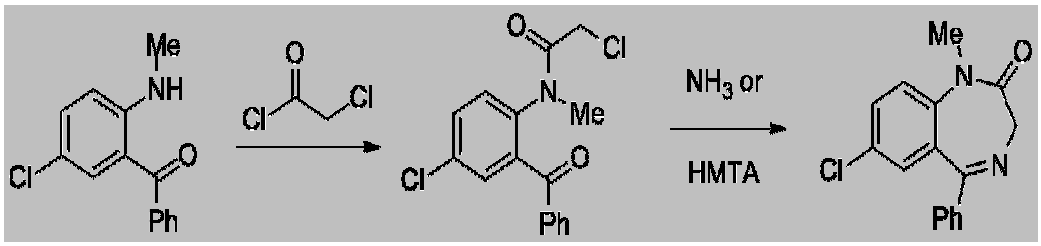


IUPAC Name :- (7-chlon-1, 3-dihydro-1-methyl-5-phenyl-2H-1,4-benzodiazepine-2-one)

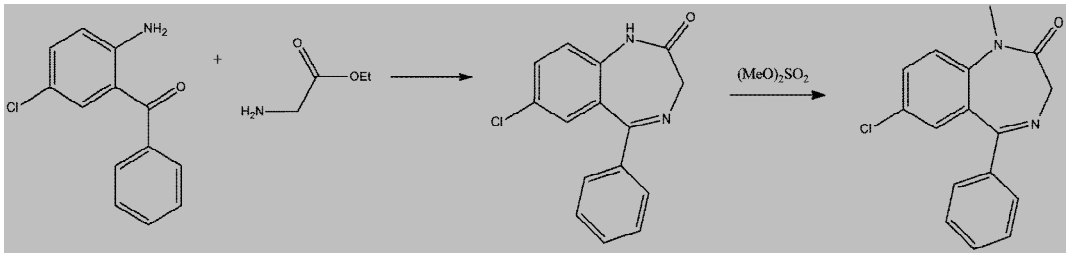
Do not give this medication to a child younger than 6 months old.

The sedative effects of diazepam may last longer in older adults. Accidental falls are common in elderly patients who take benzodiazepines. Use caution to avoid falling or accidental injury while you are taking this medicine.

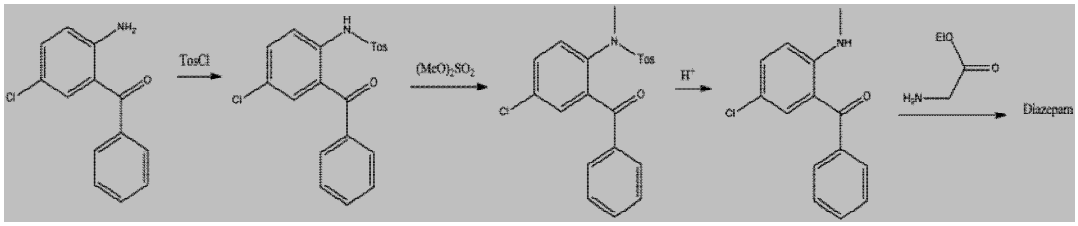
Synthesis of Diazepam:-



One of the more straightforward approaches to this compound involves first the acetylation of aminobenzophenone with chloroacetyl chloride to give the chloromethyl amide. Heating this compound with ammonia or its latent equivalent, hexamethylene tetramine (HMTA), can be envisaged to involve the initial displacement of chlorine to give a glycineamide. Cyclization by imine formation then affords diazepam. Support for this sequence comes from the observation that a modest yield of diazepam can be obtained on heating with glycine ethyl ester in pyridine.



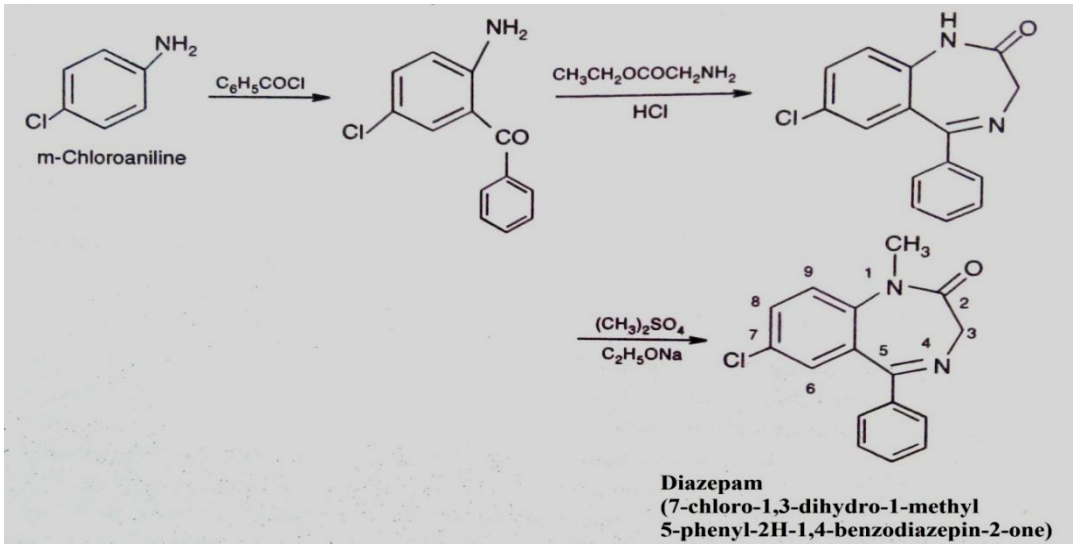
The second way consists of the direct cyclocondensation of 2-amino-5-chlorobenzophenone with glycine ethyl ester hydrochloride. The amide nitrogen atom of the obtained 7-chloro-1,3-dihydro-5-phenyl-2H-1,4-benzodiazepin-2-one, is methylated by dimethylsulfate, which leads to the formation of diazepam.



The first method differs from the second in that the methylation of nitrogen is accomplished before the cyclocondensation reaction.

To do this, the initial 2-amino-5-chlorobenzophenone is first tosylated by p-toluenesulfonylchloride and the obtained tosylate transformed into the N-sodium salt, which is then alkylated by dimethylsulfate. The resulting 2-N-tosyl-N-methyl-5-chlorobenzophenone is hydrolyzed in an acidic medium, giving 2-methylamino-5-chlorobenzophenone, which undergoes cyclocondensation by reaction with ethyl ester of glycine hydrochloride, forming the desired diazepam.

Another method of synthesis is- from m-chloraniline:-



2-amino-5-chlorobenzophenone is reacted with glycine ethyl ester in presence of pyridine to give the cyclized nordiazepam, which is methylated with dimethyl Sulffate.

Use: - It is used for the control of anxiety and tension states, the relief of muscle spasm.

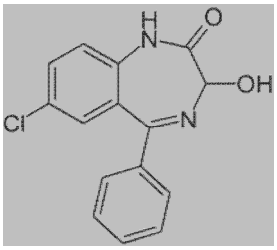
Properties: - off white to yellow crystalline powder melting point 131-135. Sparingly soluble in water, freely soluble in chloroform and alcohol.

18.4: Oxazepam

It is one of the metabolites of diazepam and used in the relief of psychoneuroses. It has reduced toxicity as compared to diazepam because the 3-OH group is easily eliminated as the glucuronide which is in active pharmacologically.

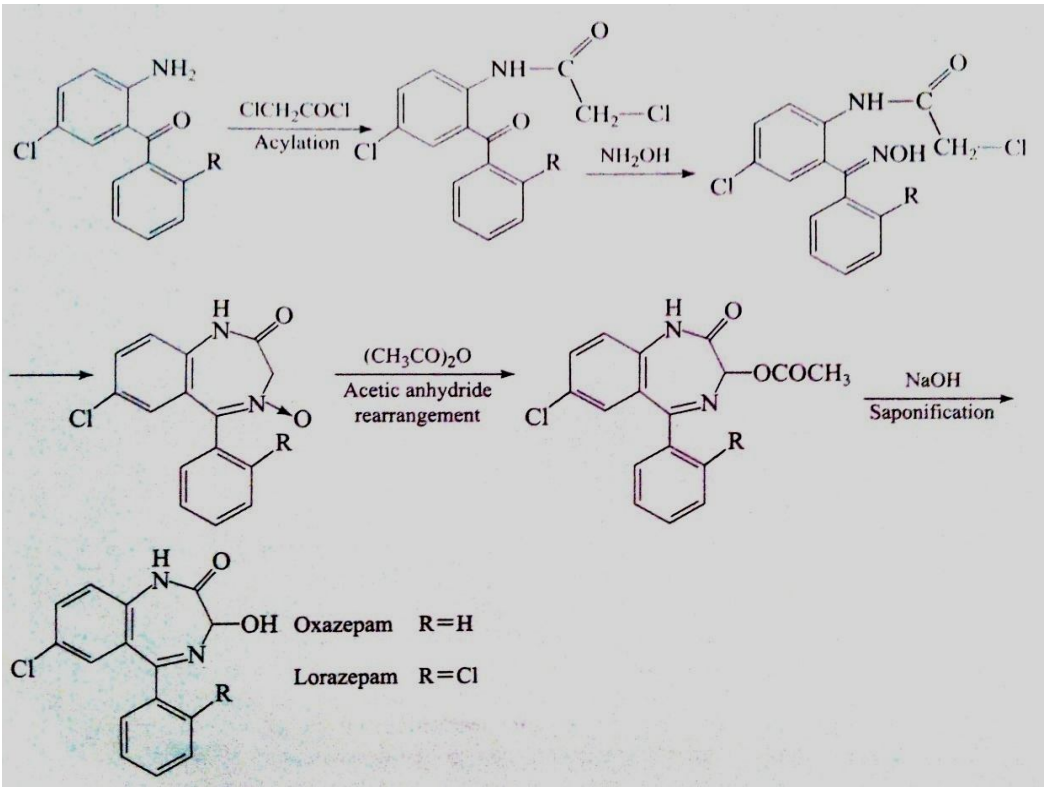
This benzodiazepine is slowly absorbed on oral administration. Also penetration in brain is slow. It has a short duration of action. So it is also used mainly in short lasting anxiety states.

Structure of Oxazepam



IUPAC Name-7-chloro-1, 3-dihydro-3- hydroxy-5- pheyl-1, 4-benzodiazepin-2-one.

Synthesis of Oxazepam



Side effects-

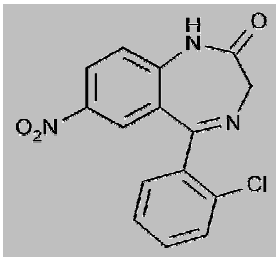
The side effects of oxazepam are similar to those of other benzodiazepines, and may include dizziness, drowsiness, headache, memory impairment, paradoxical excitement, and anterograde amnesia, but does not affect transient global amnesia. Side effects due to rapid decrease in dose or abrupt withdrawal from oxazepam may include abdominal and muscle cramps, convulsions, depression, inability to fall asleep or stay asleep, sweating, tremors, or vomiting.

18.5: Clonazepam (clonopin):-

Clonazepam is in a group of drugs called benzodiazepines (ben-zoe-dye-AZE-eh-peens). It affects chemicals in the brain that may become unbalanced and cause anxiety.

Clonazepam is used to treat seizure disorders or panic disorder.

Structure of Clonazepam:-



IUPAC Name:- 5-(2-chlorophenyl)-7-nitro-2,3-dihydro-1,4-benzodiazepin-2-one

Should not use this medication:-

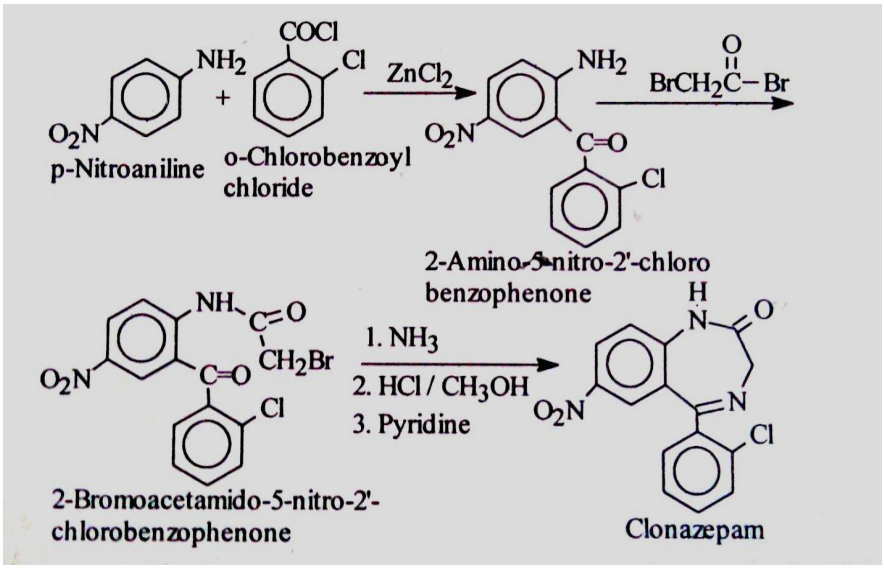
- If have severe liver disease or narrow-angle glaucoma, or
- If allergic to clonazepam or other benzodiazepines, such as alprazolam (Xanax), chlordiazepoxide (Librium), clorazepate (Tranxene), lorazepam (Ativan), or oxazepam (Serax).

The sedative effects of clonazepam may last longer in older adults. Accidental falls are common in elderly patients who take benzodiazepines. Use caution to avoid falling or accidental injury while taking clonazepam.

It is a yellow colored crystalline powder, insoluble in water acid soluble in alcohol. It is useful either alone or in combination with other anti-convulsant drugs for the treatment of petitmal epilepsy, not responsive to a valporate or ethosuximide.

Do not drink alcohol while taking clonazepam. This medication can increase the effects of alcohol.

Synthesis of Clonazepam:-



In the synthesis of clonazepam first p-nitroaniline is react with o-chlorobenzoyl chloride in the presence of zinc chloride which give 2-amino-5-nitro-2-chloro-benzophenone, this again react, with 1,2-dibromomethanone and give 2-bromoacetamido-5-nitro-2-chlorobenzophenone. At last in the presence of ammonia, methanol and pyridine it give the main compound i.e. Clonazepam

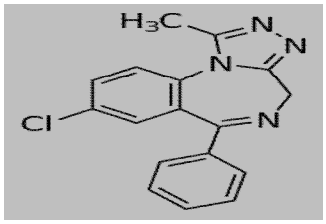
Adverse effects:- Include drowsiness, "glassy eyed" appearance, confusion, chest congestion, hair loss, anorexia, and anemia

18.6 Alprazolam (Xanax)

Alprazolam belongs to a group of drugs called benzodiazepines (ben-zoe-dye-AZE-eh-peens). It works by slowing down the movement of chemicals in the brain that may become unbalanced. This results in a reduction in nervous tension (anxiety).

Alprazolam is used to treat anxiety disorders, panic disorders, and anxiety caused by depression.

Structure of Alprazolam:-



IUPAC Name- 8-chloro-1-methyl-6-pheyl-4H-5-triazolo(4,3-a) benzodiazepine

Do not drink alcohol while taking alprazolam. This medication can increase the effects of alcohol. Alprazolam may be habit-forming and should be used only by the person for whom it is prescribed.

It is a recently introduced antianxiety drug. In addition to anxiolytic effect, it has a mood elevating action. Also it produces less drowsiness.

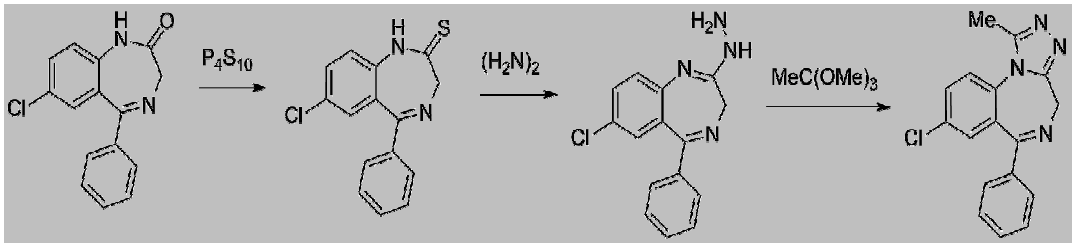
The drug is highly potent as anoxiolytic on a milligram basis. Oxidative metabolism of the methyl group to the methyl alcohol followed by conjugation is rapid and the duration of action is short.

Should not take alprazolam:

- If pregnant, because it could harm the unborn baby.
- If have Narrow-angle glaucoma;
- If also taking itraconazole (Sporanox) or ketoconazole (Nizoral); or
- If allergic to alprazolam or to other benzodiazepines, such as chlordiazepoxide (Librium), clorazepate (Tranxene), diazepam (Valium), lorazepam (Ativan), or oxazepam (Serax).

Synthesis

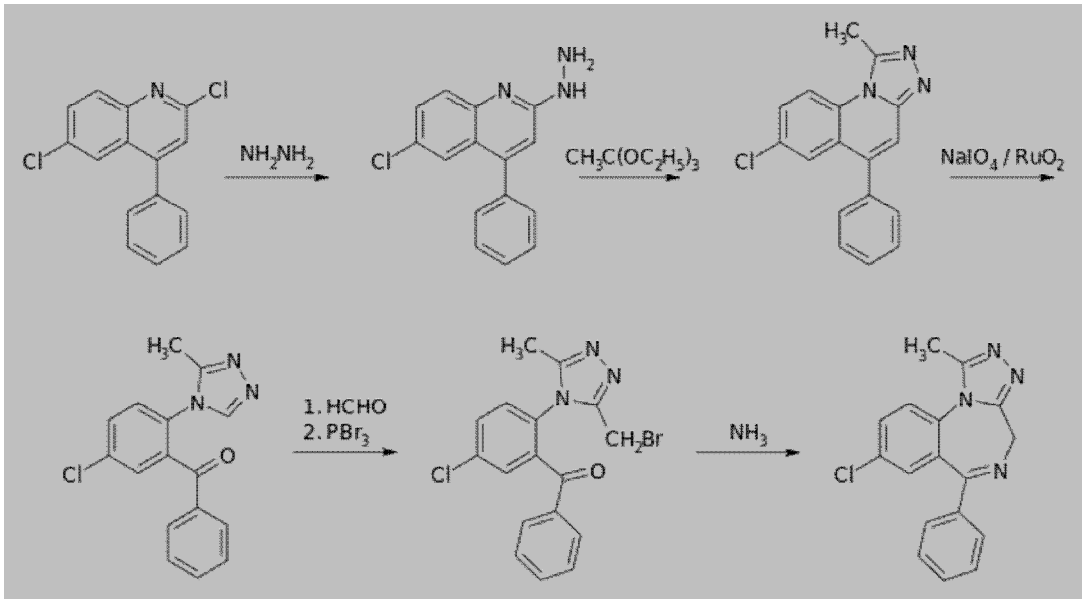
Alprazolam is a chemical analogue of triazolam that differs by the absence of a chlorine atom in the *o*-position of the 6-phenyl ring. The same scheme that was used to make triazolam can be used to make alprazolam, with the exception that it begins with 2-amino-5-chlorobenzophenone.



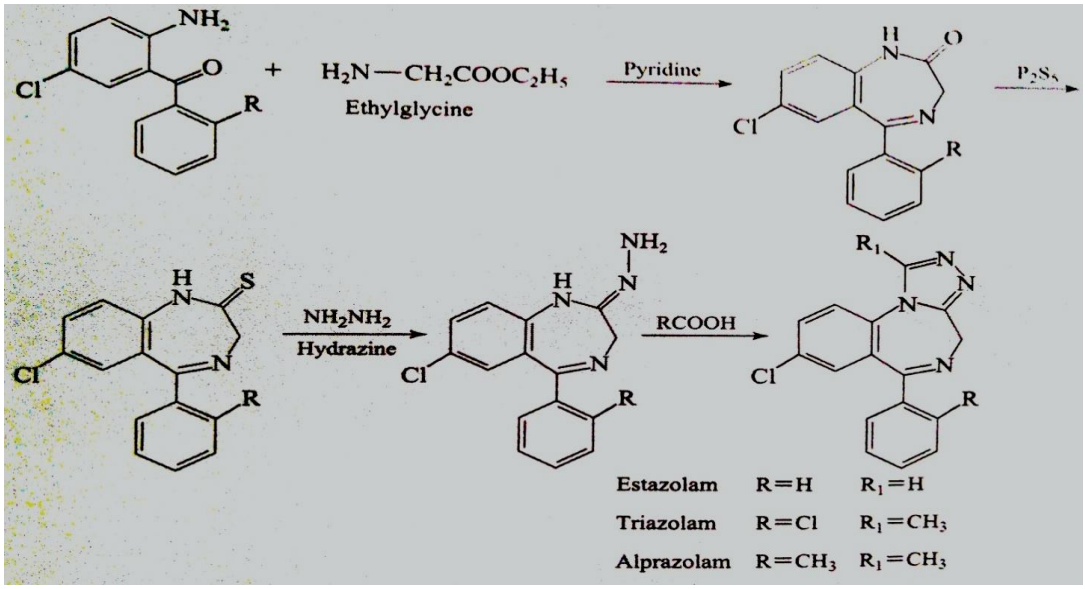
Another way of making alprazolam has been suggested, which comes from 2,6-dichloro-4-phenylquinoline, the reaction of which with hydrazine gives 6-chloro-2-hydrazino-4-phenylquinoline. Boiling this with triethyl orthoacetate in xylene leads to the heterocyclization into a triazole derivative. The resulting product undergoes oxidative cleavage using sodium periodate and ruthenium dioxide in an acetone–water system to give 2-[4-(3'-methyl-1,2,4-triazolo)]-5-chlorobenzophenone. Oxymethylation of the last using formaldehyde and subsequent substitution of the resulting hydroxyl group by phosphorus tribromide, gives 2-[4- (3'-methyl - 5' – bromomethyl - 1, 2, 4 – triazolo)] – 5 - chlorobenzophenone.

Substitution of the bromine atom with an amino group using ammonia and the spontaneous, intramolecular heterocyclization following that reaction gives alprazolam.

Method I

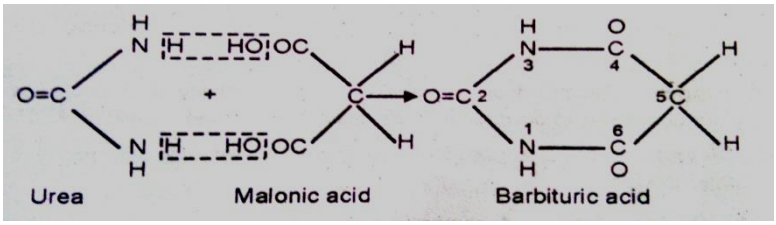


Method II



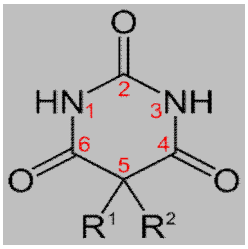
18.7 Barbiturates

Barbiturates are derivatives of barbituric acid (malonyl urea) which is obtained by the condensation of urea and malonic acid.



Barbituric acid itself does not possess hypnotic activity. But hypnotic activity is produced if the hydrogen atoms at position 5-are replaced by alkyl or aryl groups.

General structure of barbiturates-



Use of barbiturates

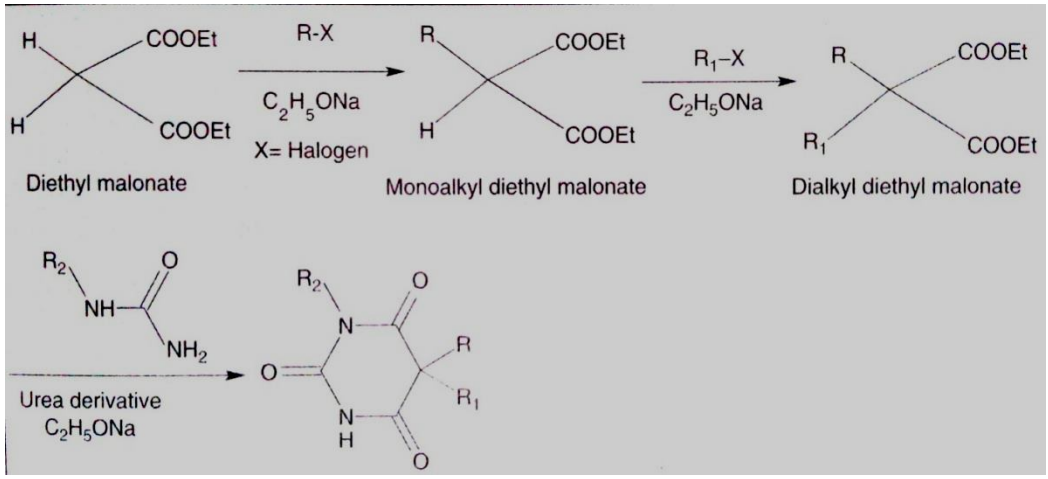
- At low doses barbiturates are indicated for the relief of anxiety and tension.
- At high doses they are used for the management of insomnia (sleeplessness)
- Long duration acting barbiturates are also used as anti-convulsant agents.

Classification

[A]	**Long Acting :-** (6 or more hrs)	Phenobarbitone Mephobarbitone Metharbital
[B]	**Intermediate duration:-** (3-6 hrs)	Butabarbital Amobarbital Aprobarbital Hexobarbital
[C]	**Short Acting :-** (les than 3 hrs)	Buto barbitone Secobarbitone Pento barbitone

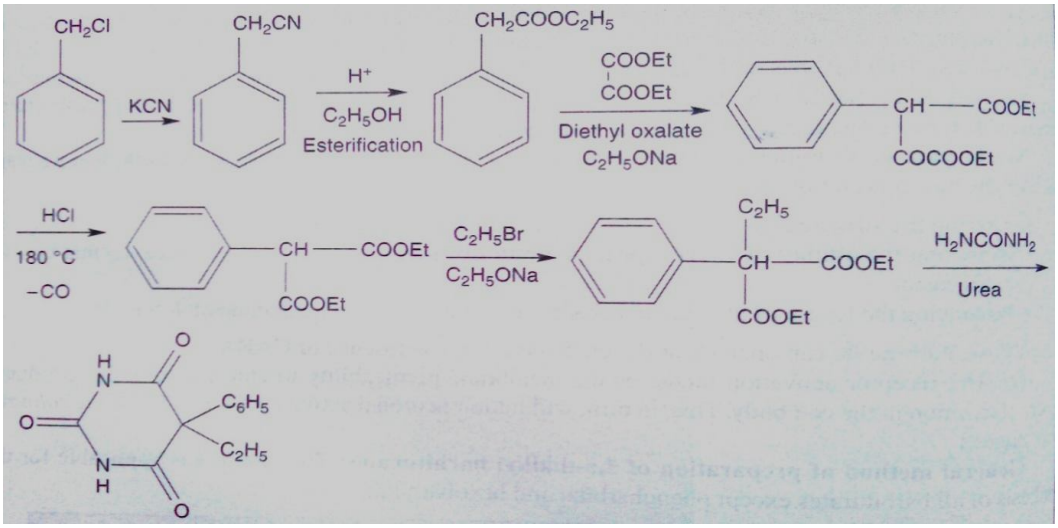
General method of preparation of 5,5-dialkyl barbiturates :-

This method is applicable for the synthesis of all barbiturates except Phenobarbital and hexobarbital.



If	R	R_1	R_2
Phenobarbital	C_2H_5	C_6H_5	H
Butabarbital	C_2H_5	$CH_3CH_2CH(CH_3)$-	H
Pentobarbital	C_2H_5	$CH_3CH_2CH_2CH(CH_3)$-	H

Synthesis of Phenobarbital (5-ethyl-5-phenyl barbituric acid):-



Benzyl chloride on reaction with potassium cyanide affords benzyl cyanide. Ethanolysis of benzyl cyanide in the presence of acid gives phenyl acetic acid ethyl ester. The methylen group of which undergoes acylation using the diethyl

oxalate, giving diethyl ester of phenyloxobutandionic acid, which upon heating easily loses carbon oxide and turns into phenylmalonic ester.

Alkylation of the obtained product using ethyl bromide in the presence of sodium ethoxide leads to the formation of α–phenyl–α–ethyl malonic ester, the condensation of which with urea gives Phenobarbital.

18.8 Summary

Atypical antipsychotic agents are used to treat the symptoms schizophrenia and bipolar disorder. Clonazepam may cause harm to an unborn baby, and may cause breathing or feeding problems in a newborn. But having seizures during pregnancy could harm both mother and baby. Do not start or stop taking clonazepam during pregnancy without medical advice. Oxazepam is contraindicated in myasthenia gravis, chronic obstructive pulmonary disease, and limited pulmonary reserve, as well as severe hepatic disease.

18.9: Glossary

- **Agitation:-** It is defined as the state of feeling irritated or restless (A state of anxiety or nervous excitement).
- **Anemia:-** It is usually defined as a decrease in the amount of red blood cells (RBCs) or the amount of hemoglobin in the blood. It can also be defined as a lowered ability of the blood to carry oxygen
- **Anterograde Amnesia:-** Anterograde Amnesia is a loss of the ability to create new memories after the event that caused the amnesia, leading to a partial or complete inability to recall the recent past, while long-term memories from before the event remain intact
- **Anti-Convulsants :-** It is also known as antiepileptic drugs or as antiseizure drugs. These are a diverse group of pharmacological agents used in the treatment of epileptic seizures.
- **Myasthenia Gravis:-** It is characterized by weakness and rapid fatigue of any of the muscles under voluntary control. Myasthenia gravis is caused by a breakdown in the normal communication between nerves and muscles.
- **Narrow-Angle Glaucoma:-** This is the second most common form of glaucoma. Patients often have acute attacks of eye pain due to sudden

increase in eye pressure. Between attacks the eye pressure is normal. An attack of this type of glaucoma is an emergency. Untreated, it may cause blindness in a day or two.

- **Paradoxical :-** A paradoxical reaction is an adverse reaction to a substance, almost always to a drug, that is exactly the opposite of the intended effect. A paradoxical reaction to any substance can be frightening and uncomfortable.
- **Psychoneurosis:-** Psychoneurosis is the emotional maladaptation due to unresolved unconscious conflicts. This leads to disturbances in thought, feelings, attitudes, and behavior.
- **Schizophrenia:-** It is a mental disorder that generally appears in late adolescence or early adulthood- however, it can emerge at any time in life. It is one of many brain diseases that may include delusions, loss of personality (flat affect), confusion, agitation, social withdrawal, psychosis, and bizarre behavior.
- **Transient Global Amnesia:-** It is a sudden, temporary episode of memory loss that cannot be attributed to a common neurological condition, such as epilepsy or stroke.

18.10 Review questions /comprehensive questions

1. Give the general method of synthesis of barbiturates.
2. What is the method for synthesis of Diazepam?
3. Give the synthesis of any two-
    a. Alprazolam
    b. Clonazepam
    c. Oxazepam
4. Give the synthesis of phenobarbitone.
5. Give the chemical synthesis of diazepam from chloroaniline.
6. Discuss about the chemistry of diazepam and its synthesis.

18.11 References and suggested readings

- Textbook of Organic Medicinal and Pharmaceutical Chemistry (11th ed.)- Wilson and Gisvold's.

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- A Book of medicinal chemistry volume I; (first edition 2008-2009) Dr. Anees Ahmad Siddiqui, Seemi Siddiqui, Dr. R. Rajesh, published by birla publication pvt. Ltd.
- A Text book of medicinal chemistry; (third edition 2003) P. Parimoo, published by CBS publication.
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- Principle of medicinal chemistry-vol. II; (eighteenth edition, september 2007, reprint edition- March 2008, August 2008,) Dr. S. S. Kadam, Dr. K. R. Mahadik, Dr. K. G. Bothara, published by- Nirali prakashan.
- Medicinal Chemistry; (revised third edition-2005, reprint-2008) Ashutosh Kar, published by New age international (P) Ltd.
- Essential of medical Pharmacology; (seventh edition-2013) K. D. Tripathi, Published by- JAYPEE brothers medical publishers (P) Ltd.

Unit-19

Antibiotics

Structure of Unit:

19.1 Objectives

- Antibiotics inhibit the growth and survival of microorganism without serious toxicity to the host.
- Antibiotics and antimicrobial agents to keep infectious disease under control.
- To study about different cell wall biosynthesis inhibitor that cures the infection.
- Antibiotics that inhibit protein synthesis by which housekeeping protein not synthesize and indirectly kill the bacteria such as tetracycline, aminoglycoside etc.

19.2 Introduction

Paul Ehrlich introduced the term chemotherapy in 1907 in referring to antiparasitic therapy. Now refers more broadly to the use of any chemical compound that selectively acts on microbes or cancer. Ehrlich had previously developed selective chemical stains for the microscopic examination of

Mycobacterium tuberculosis and other microorganisms, using the coal-tar derivative dyes. The search for safe, effective chemotherapeutic drugs is hindered by humans share with all living organisms. Success requires exploitation of metabolic or structural differences between normal human cells and disease-producing cells. Humans were not the first to exploit the selective toxicity of chemicals. Many fungi and bacteria make toxic substances that kill or suppress the growth of competing microorganisms or facilitate infection of a host. Plants make a vast array of toxins for their self-defence. Exploitation of the selective toxicity of chemicals is an ancient and widely employed technique.

History of chemotherapy may be divided into 3 phases:-

(a) The period of empirical use of 'mouldy curd' by Chinese on boils, chaulmoogra oil by Hindus in leprosy, chenopodium by Aztecs for intestinal worms, mercury by Paracelsus for syphilis, cinchona bark for fevers.

(b) Ehrlich's phase of dyes and organometallic compounds (1890-1935): with the discovery of microbes and they are the cause of many diseases. Ideas of Ehrlich that if certain dyes could selectively stain microbes, they could also be selectively toxic to these organisms and tried methylene blue, trypan red etc. Ehrlich eventually discovered the arsenical compound- atoxyl for sleeping sickness (Trypanosoma brucei, the tsetse fly–borne parasite that causes African trypanosomiasis), arsphenamine in 1906 and neoarsphenamine in 1909 for syphilis.

(c) The modern era of chemotherapy was demonstrating the therapeutic effect of Prontosil, a sulphonamide dye in pyogenic infection. It was soon realised that the active moiety was paraamino benzene sulphonamide, and the dye part was not essential. Fleming 1929 found that a diffusible substance was elaborated by Penicillium mould which could destroy Staphylococcus on the culture plate. The well known observation of a clear zone inhibition of (lysis) of bacterial colonies surrounding a colony of contaminating air borne.

19.3 What is Antibiotic ?

Antibiotics are microbial metabolites or synthetic analogs which suppress the growth and survival of microorganism at low concentration without serious toxicity to the host. This definition excludes other natural substances which also inhibit microorganisms but are produced by higher forms eg. Antibodies or even those produced by microbes but are needed in higher concentration eg. Lactic acid, Ethanol. Antibiotics are among the most frequently prescribed

medications today although microbial resistance due to misuse threatens their continued efficacy. In many cases the clinical utility of natural antibiotics has been enhanced through medicinal chemical manipulation of the original structure leading to broader antimicrobial spectrum, greater potency, lesser toxicity, more convenient administration etc. Examples of such semi synthetic antibiotics are amoxicillin and doxycycline.

19.4 Classification of antibiotics

Antimicrobial drugs can be classified according to their chemical structure and their mechanism of actions.

(A) On the basis of Chemical structure

1. Sulphonamide and related drugs: Sulfadiazine, sulfamethoxazole and others
2. Diaminopyrimidines: Trimethoprim, pyrimethamine
3. Quinolones: Nalidixic acid, Norfloxacin, Ciprofloxacin
4. β- lactam antibiotics: Penicillins, Cephalosporins, Monobactams, Carbapenems
5. Tetracyclines: Oxytetracycline, Doxycycline, etc.
6. Nitrobenzene derivative: Chloramphenicol
7. Aminoglycosides: Streptomycin, Gentamicin, Amikacin, Neomycin, etc.
8. Macrolide antibiotics: Erythromycin, Clarithromycin, Azithromycin, etc.

(B) On the basis of mechanism of action

1. Inhibit cell wall synthesis: Penicillins, Cephalosporins, Cycloserine, Vancomycin Bacitracin.
2. Cause leakage from cell membranes: Poly-peptides-Polymyxins, Colistin, Bacitracin Polyenes-Amphotericin B, Nystatin, Hamycin.
3. Inhibit protein synthesis: Tetracycline, Chloramphenicol, Aminoglycosides, Erythromycin, Clindamycin, Linezolid.

19.5 Bacterial cell wall biosynthesis

The bacterial cell wall differs in structure and functions compared to outer layer of mammalian cells. This makes easy to provide a number of potentially attractive target for selective chemotherapy of bacterial infection. The bacterial cell wall is chemically distinct from mammalian cell walls construction. The

initial units of cell wall constructed within the cell and final assembly takes place outside the inner membrane.

According Gram staining bacteria are differentiate in two types: Gram (+) bacteria and Gram (-) bacteria. The difference in two bacteria is composition of bacterial cell wall.

Gram (+) Bacteria:

The cell wall of Gram (+) bacteria is simpler than Gram (-) bacteria. On the outside of the cell is a characteristic carbohydrates and proteins that together make up antigenic determinants that differ from species to species. The next barrier that the wall presents is peptidoglycan layer. This is a spongy, gel-forming layer consisting of a series of alternating sugars (N-acetyl glucosamine [NAG] and N-acetylmuramic acid [NAM]) linked in a long chain. To the free lactic acid carboxyl moieties of N-acetylmuramic acid units is attached with a series of amino acids of which L-alanin-D-glutamine-L-lysine-D-alanin through peptide linkage. D-stereochemistry of the glutamate and the terminal alanin is important feature in protecting the peptidoglycan from hydrolysis by host peptidase.

The terminal D-alanyl unit is bonded to the lysyl unit of an adjacent tetrapetide strand through a pentaglycyl unit. The last step in the biosynthesis is a transamidation wherein the terminal amino moieties on the last glycine of the A strand displaces the terminal D-ala unit on the nearby B strand. This step is catalysed by a cell wall transamidase (one of the penicillin binding protein, PBP) which form a covalent bond during the synthesis phase with a particular serine hydroxyl on the enzyme. Completion of the catalytic cycle by cross linking through displacement of the enzyme and substitution by a glycine residue regenerates the enzyme and produces a thickened three dimensional cell wall.

Gram (-) Bacteria:

The cell wall of gram –ve bacteria is more complex and more lipoidal. These cells usually contain an additional, outer membrane. The outer layer contains complex lipopolysaccharides that encode antigenic responses. This exterior layer also contains a number of enzymes and proteins.

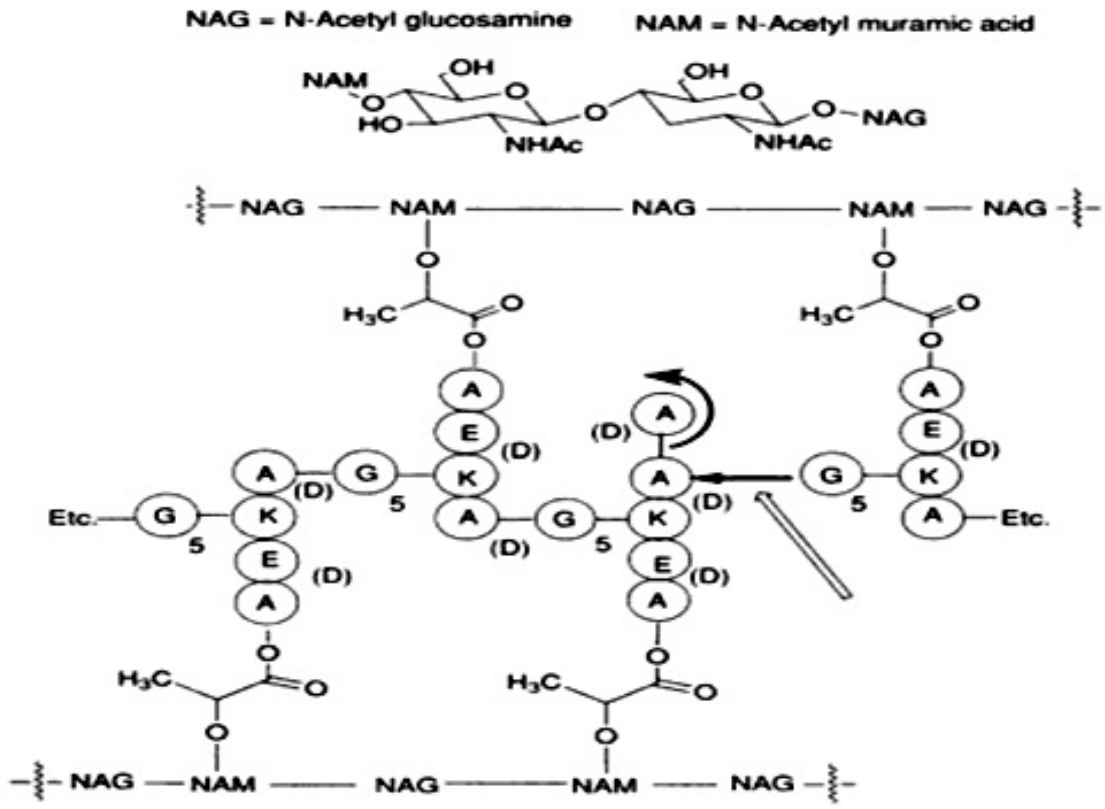
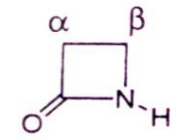


Fig. 19.1: Biosynthesis of bacterial cell wall

19.6 Cell wall biosynthesis inhibitors

β- Lactam antibiotics

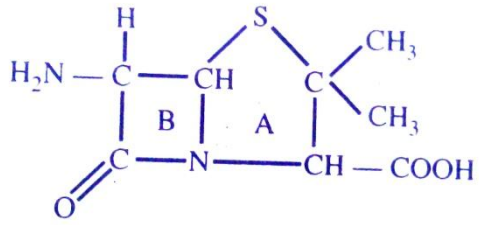
The name lactam means cyclic amides and the term lactone means cyclic esters. According to nomenclature, second carbon in an aliphatic carboxylic acid was designated as alpha, the third beta. β- Lactam is a cyclic amide with four atoms in its ring. The contemporary name for this system is azetidinone. It is note that beta lactam ring proved to be the main component of pharmacophore so the term possesses medicinal as well as chemical significnce. The beta lactam antibiotics are penicillin and cephalosporin. The penicillin subclass of β- Lactam antibiotics is characterised by presence of a substituted 5-membered thiazolidine ring fused to the β- Lactam ring. This fusion and the chirality of the β- Lactam ring results in the molecule representing possessing a "V" shape.



beta -lactam
azetidinone

19.7 Penicillin

Penicillin had been discovered in 1929 and is a member of β- Lactam antibiotics The original fermentation-derived penicillins were produced by uncontrolled fermentation of the fungus *Penicillium chrysogenum.* Penicillin can be considered as the amido derivatives of the 6-aminopenicillanic acid.



6-Aminopenicillanic acid (6-APA)

In the basic skeleton, a thiazolidine ring (A) is fused with β- Lactam ring (B) which is a four membered cyclic amide. Classification of penicillin is given in Table 19.1

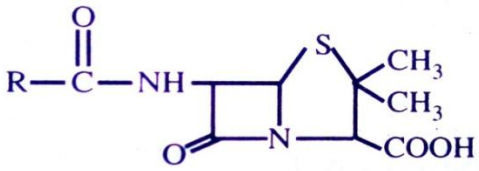
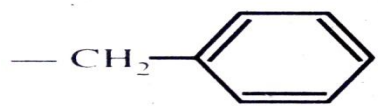
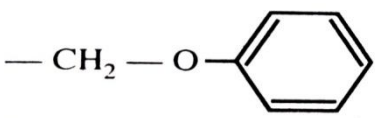
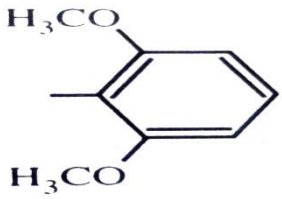
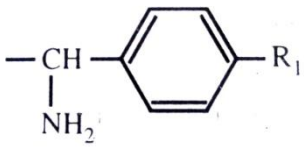
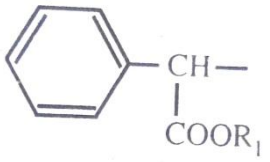
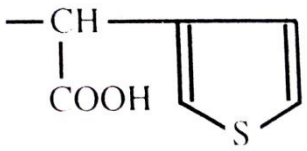
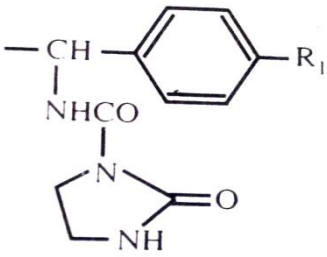


Table 19.1: Classification of penicillin antibiotic

S.No.	Name	Nature of substituent, R
1	Penicillinase susceptible penicillins: i. Penicillin G ii. Penicillin V	 

2.	Penicillinase resistant penicillins: i. Methicillin	
3.	Amino penicillins: i. Ampicillin (R_1= H) ii. Amoxicillin (R_1= OH)	
4.	Carboxypenicillins: i. Carbenicillin (R_1= H) ii. Ticracillin	 
5.	Uriedopenicillins: Azlocillin	

Mechanism of action:

Molecular mode of action of the β- Lactam antibiotics is a selective and irreversible inhibition of the enzyme processing the developing peptidoglycan layer. Just before the cross-linking occurs, the peptide unit from the lactate carboxyl of a muramic acid unit terminate in a D-ala-D-ala unit. This is cleaved between these two amino acids by hydrolysis catalysed by a cell wall transamidase. This is one of the penicillin binding proteins that normally present in bacterial inner membrane and perform construction, repair and

housekeeping functions maintaining cell integrity and playing a vital role in cell growth and division.

Uses:

1. Streptococcal infections like pharyngitis, otitis media, scarlet fever, rheumatic fever respond to ordinary doses of PnG given for 7-10 days. For subacute bacterial endocarditis (SABE) caused by *Streptococci viridans* or *faecalis* treated with high doses (10-20 MU i.v. daily).
2. Pneumococcal infections
3. Meningococcal infections are still mostly responsive. Meningitis and other infections may be treated with intravenous injection of high doses.
4. Gonorrhoea PnG has become unreliable for treatment of gonorrhoea due to spread of resistant strain.
5. Syphilis *Trypanosoma pallidum* has not shown any resistance and PnG is the drug of choice. Early and latent syphilis is treated either with daily injection of 1.2 MU of procaine penicillin.

19.8 Cephalosporins

Penicillin which has outstanding biological antibiotics properties that it entered clinical uses with comparatively little change. Cephalosporin was produced by *Cephalosporium arcemonium.* One of the constituents had the useful property of activity against penicillin resistant cultures due to its stability to β-lactamases. The nature of side chain produced 7-aminocephalosporanic acid (7-ACA) which analogous to 6-aminopenicilllanic acid. Many of the compounds produced in this way. They differ from one another in antimicrobial spectrum, β- lactamase stability, absorption from GI tract, metabolism, stability and side effect. The cephalosporins have their β- lactam ring attached to a 6-membered dihydrothiazine ring.

In the cephalosporin antibiotic two rings are fused. Ring A is dihydrothaizine ring and ring B is β-lactam ring. By addition of different side chain at position 7 of β-lactam ring and position 3 of dihydrothiazine ring, a large number of semisynthetic compound have been produced. Basic structure of cephalosporin is given below. Classification of cephalosporin is given in Table 19.1

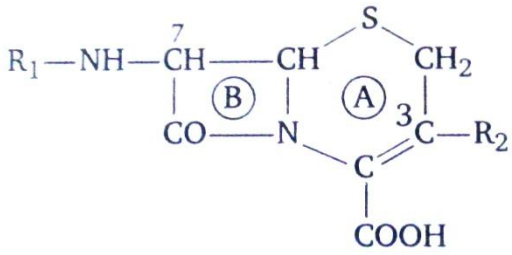
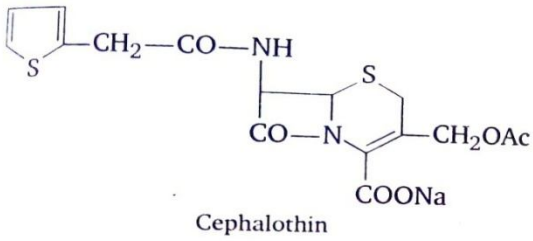


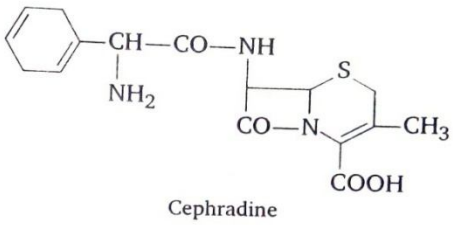
Table 19.1: Classification of cephalosporins

First Generation	Second Generation	Third Generation
• Cefazolin • Cephalexin • Cephaprin • Cephalothin	• Cefaclor • Cefanocid • Cefoxitin • Cefuroxine axetil	• Cefoperazone • Cefotaxime • Ceftriaxome • Cefixime

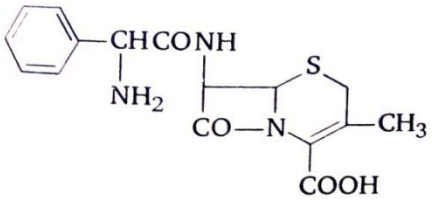
Structures of some cephalosporins:



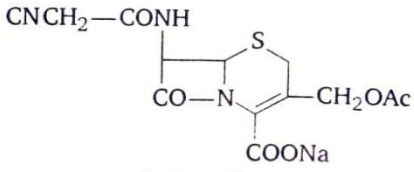
Cephalothin



Cephradine



Cephalexin



Cephacetrile

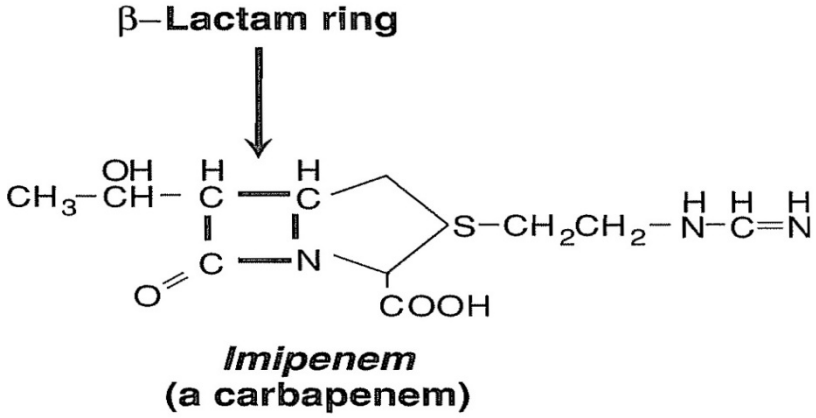
Mechanism of action: The cephalosporins are believed to act in manner similar to penicillins by binding to penicillin binding proteins followed by cell lysis. Cephalosporins are bactericidal.

Uses:

Cephalosporin is used in upper respiratory tract, skin and related soft tissue, urinary tract infection, bones and joints, as well as septicemias and intraabominal and bile tract infection caused by susceptible Gram (+) organism.

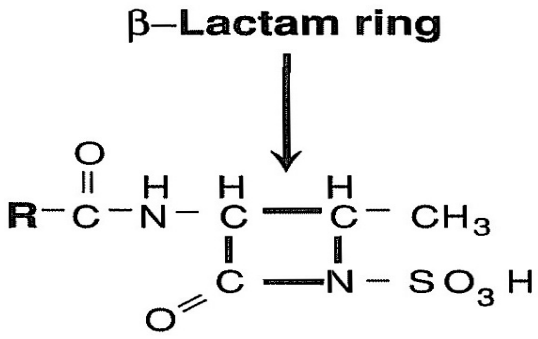
Carbepenem: Carbepenam are synthetic beta lactam antibiotics that differ from the penicillins in that the sulfur atom of the thiazolidine ring has been

externalized and replaced by a carbon atom. Imepenem is the only drug is available.



Imipenem
(a carbapenem)

Monobectam: The monobactams, of which aztreonam is the only commercially available example, are unique because the β-lactam ring is not fused to another ring. Monobactams also disrupt cell wall synthesis. The drug's narrow antimicrobial β-Lactam ring spectrum precludes its use alone in empiric therapy. Aztreonam is resistant to the action of β-lactamases.



Aztreonam
(a monobactam)

19.9 Antibiotics inhibiting protein synthesis

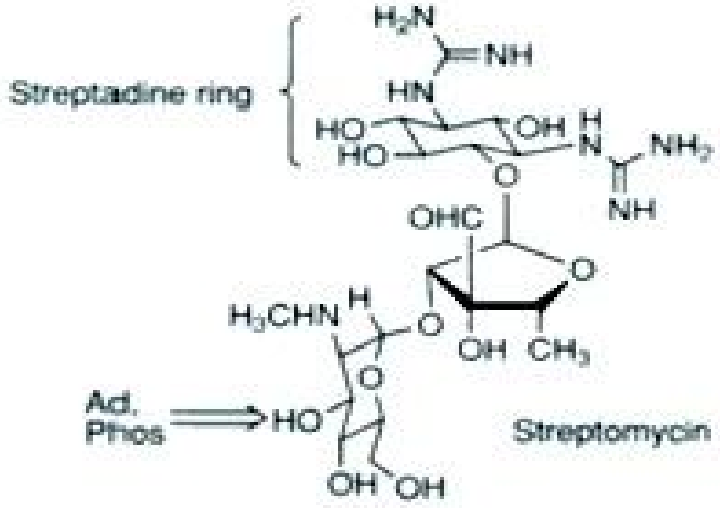
Some antibiotic show their lethal effect on bacteria by inhibiting ribosomally mediated protein synthesis. Eukaryotic organism made their essential proteins on ribosomal organelles and the sequence of biochemical steps in both biologic classes is closely analogous to prokaryotic organisms. Eukaryotic organisms have 80S ribosome while prokaryotic organisms have 70S ribosome. The binding sites for the important antibiotics lie on the proteins or the ribosomal DNA of the bacterial 70S ribosome. Interference with the bacterial protein biosynthesis prevents repair, cellular growth, and reproduction and in clinical achievable doses is bactericidal or bacteriostatic. Some of the protein synthesis

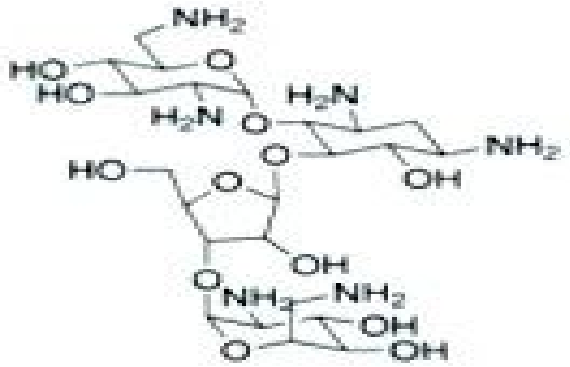
inhibitors are synthetic compounds and other were first isolated from the fermented culture filtrate of various organisms as antibiotic.

Aminoglycosides:

The aminoglycoside class of antibiotics contains a pharmacophoric 1,3-diaminoinositol moiety. The chemistry, spectrum, potency, toxicity and pharmacokinetics of these agents are a function of the specific identity of diaminoinositol unit.

Several aminoglycoside antibiotics are discovered such as Streptomycin, amikacin, gentamycin, neomycin and tobramycin etc. the structures of aminoglycoside antibiotic are given below.





Neomycin B

Mechanism of action: All members of this family are believed to inhibit bacterial protein synthesis by the mechanism determined for streptomycin. Susceptible organisms have an oxygen dependent systems that transport the antibiotic across the cell membrane. Antibiotics than binds to the separated 30S

ribosomal subunit and interfering with assembly of the functional ribosomal apparatus or causing the 30S subunit of the complete ribosome to misread the genetic code.

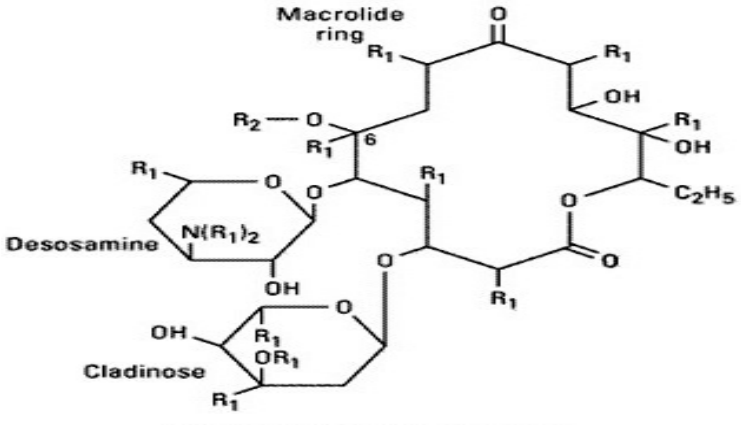
Uses:

In most other situations, e.g. urinary tract infection, peritonitis, septicaemias, etc. where streptomycin was used earlier, gentamicin or one of the newer aminoglycosides is now preferred due to low potency and widespread resistance to streptomycin.

Macrolides:

The macrolides are a group of antibiotics with a macrocyclic lactone structure. The clinical important members of this class have two or more characteristic sugars attached to the 14-membered ring. The 14 membered ring macrolides are biosynthesized from propionic acid units so that every second carbon of erythromycin bears a methyl group with one exception is oxygen bearing.

Erythromycin was the first of these to find clinical application both as the drug of first choice and as an alternative to penicillin in individuals who are allergic to β-lactam antibiotics. The new members of this family, clarithromycin (a methylated form of erythromycin) and azithromycin (having a larger lactone ring) have some features in common with and others that improve on, erythromycin. Recently, dirithromycin, a macrolide similar to erythromycin in antibacterial spectrum, but with the advantage of one-daily dosage, has been approved.



Erythromycin ($R_1 = CH_3$, $R_2 = H$)
Clarithromycin (R_1, $R_2 = CH_3$)

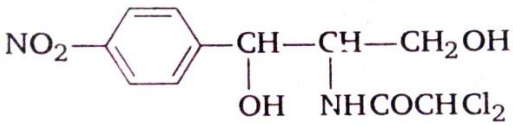
Mechanism of action: the macrolide inhibiting bacteria by interfering with programmed ribosomal protein biosynthesis by inhibiting translocation of aminoacyl t-RNA following binding to the 50S subtype.

Uses: macrolide are used for treatment of upper and lower respiratory tract and soft tissue infection, caused by gram (+) microorganism like S*treptococcus pyogens* and S*treptococcus pneumonia*.

Chloramphenicol

Chloramphenicol was obtained from *streptomyces venezuelae*. It has nitro benzene substitution which is responsible for its antibacterial activity. It is soluble in alcohol but poorly soluble in water. Chloramphenicol succinate, which is used for parenteral administration, is highly water-soluble. It is hydrolyzed in vivo with liberation of free chloramphenicol.

Chloramphenicol is broad spectrum antibiotic that is active against gram-positive and gram-negative bacteria, rickettsiae, micoplasma and Chlamydia. It is inactive against pseudomonas, many proteus viruses and fungi. It is highly active agaist salmonella including *S. typhi*. It is less active against gram positive cocci, spirochetes and certain anterobacteriaceae. It is inactive on antamoeba and plasmodia.



Chloramphenicol

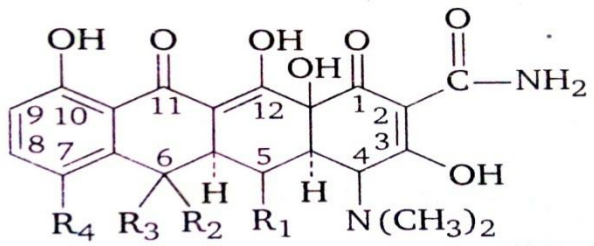
Mode of action:- Chloramphenicol inhibits bacterial protein synthesis by interfering with 'transfer' of the elongating peptide chain to the newly attached aminoacyl-tRNA at the ribosome mRNA complex. It attaches to the 50s ribosome and hence hinders the access of aminoacyl-tRNA to the acceptor side for amino acid incorporation. Most probably, by acting as a peptide analogue, it prevents formation of peptide bonds.

Use- it is a drug of choice for typhoid fever. It is also used in intraocular infections, H. influenza meningitis, anaerobic infections, external ear infections and urinary tract infection. It may be considered for treatment of serious rickettsial infections, such as typhus or Rocky Mountain spotted fever, in children for whom tetracyclines are contraindicated. Chloramphenicol is occasionally used topically in the treatment of eye infections because of its wide antibacterial spectrum and its penetration of ocular tissues and the aqueous humor.

Tetracycline:

All are obtained from soil actinomycetes. The first to be introduced was chlortetracycline in 1948 under the name aureomycin (because of the golden yellow colour of S. aureofaciens colonies producing it). It contrasted markedly

from penicillin and streptomycin (the other two antibiotics available at that time) in being active orally and in affecting a wide range of microorganisms-hence called 'broadspectrum antibiotic'. Tetracycline antibiotic is characterized by highly functionalized partially reduced naphthacene ring system from which both the family name and numbering system is derived. The structures of tetracycline antibiotic are given below.



	R_1	R_2	R_3	R_4
Tetracycline	H	OH	CH_3	H
Chlortetracycline	H	OH	CH_3	Cl
oxytetracycline	OH	OH	CH_3	H
Demeclocycline	H	OH	H	Cl

All tetracyclines are solid, bitter taste and weak water soluble antibiotics. Their aqueous solutions are unstable. Tetracycline are divided in three classes

Class 1	Class 2	Class 3
Tetracycline Chlortetracycline Oxytetracycline	Demeclocycline Methacycline	Doxycycline Minocycline

Mechanism of action:

Tetracyclines enter microorganisms in part by passive diffusion and in part by an energy-dependent process of active transport. Susceptible cells concentrate the drug intracellularly. Tetracyclines bind reversibly to the 30S subunit of the bacterial ribosome, blocking the binding of aminoacyl-tRNA to the acceptor site on the mRNA-ribosome complex. This prevents addition of amino acids to the growing peptide.

Uses:

Tetracycline possesses very wide bacteriostatic antibacterial activity. They are popular and low dose oral and topical therapy for acne. They are also used in urinary tract infection, upper respiratory tract infection, ophthalmic infections, sexually transmitted diseases, rickettsial infection, prophylaxis for malaria and prevention of traveler's diarrhea.

19.10 Summary

Antimicrobial agents are the greatest contribution of the 20th century to therapeutics. Antibiotics are produced by microorganisms and selectively suppress the growth or kill other microorganisms at very low concentration. Antimicrobial drugs produce action in many ways such as- inhibition of cell wall synthesis, inhibition of protein synthesis etc. Antibiotics which consists β-lactum ring in their chemical structure are called β-lactum antibiotics. The two major groups are penicillin's and cephalosporins. Cell wall is the outer covering of microorganisms which provide protection and stability to microorganisms. All β-lactum antibiotics interfere with the synthesis of bacterial cell wall. Broad spectrum antibiotics tetracycline, chlormphenical and macrolides erythromycin act by inhibiting bacterial protein synthesis.

19.11 Glossary

- Pyogenic infection: infection that result in pus production
- Inhibition zone: zone where no bacterial colony is present
- Pharmacophore: component in the structure that is responsible to activity
- Genetic code: set of rules by which information encoded within genetic material (DNA)
- Rocky mountain spotted fever: an infection disease caused by bacteriaum *Rickettsia* and transmitted by wood tick.

19.12 Review questions / Comprehensive Questions

1. Define antibiotics. Classify β-Lactum antibiotics.
2. Write a detail note on cell wall biosynthesis.
3. Write mechanism of action and clinical uses of β-Lactum antibiotics.
4. Write note on-
    a. Streptomycin
    b. Tetracycline

c. Chloramphenicol
d. Erythromycin

5. Write note on-
    a. Penicillin G
    b. Methicillin
    c. Cloxacillin
    d. Amphicillin

19.13 References and Suggested readings

- Principles of medicinal chemistry, volume II- Dr. S.S. Kadam, Dr. K.R. Mahadik, Dr. K.G. Bothara(Niraliprakashan) 2007.
- Foye's Principles of Medicinal Chemistry, David A. Williams, Thomas L. Lemke (Fifth edition) 2005, B.I. Publication and Pvt. Ltd.
- Pharmacology (5thed.)- H.P Rang and M.M Dale (Elsevier publisher) 2003.
- Basic & Clinical Pharmacology (ninth edition)- Bertram G.Katzung (Mc Graw Hill Publisher) 2004.

Unit - 20

Synthesis of Antibiotics

Structure of Unit:

20.1 Objective:

- In this unit we will discuss about the drugs which are used for bacterial infections and also their method of synthesis and uses.
- Here we will study about the drugs which show their action against Gram-positive bacteria and Gram-negative bacteria or both.

20.2 Introduction

The term antibiotic has been derived from the word antibiosis which according to biology concept means survival of fittest i.e. process in which one organism may destroy another to preserve itself it is define as chemical substance produced by living cells which is capable in small concentration inhibiting the life processes or even destroy the micro-organism. Penicillin was the first antibiotic introduced as a successful antimicrobial drug.

Historically the most common classification has been based on chemical structure and proposed mechanism of action, as follows:-

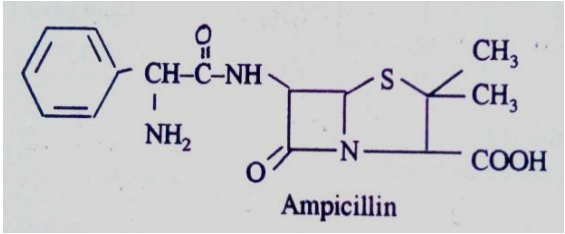
- Agents that inhibit the synthesis of bacterial cell walls, these include the penicillin and cephalosporin, which are structurally similar, and dissimilar agents such as cycloserine, Vancomycin bacitracin, and the imidazole antifungal agents.
- Agents that act directly on the cell membrane of the micro-organisms affecting permeability and leading to leakage of intracellular compounds. These include polymyxin, polyene antifungal agents, nystatin and anphotericin B, which bind to cell wall sterols.
- Agents that affect the function of 30S and 50S ribosomal subunits to cause reversible inhibition of protein synthesis; these include tetracyclines, erythromycins, chloramphenicol and clindamycin.
- Agents that bind to the 30S ribosomal subunit and alter protein synthesis, which eventually leads to cell death; these include amino glycosides.
- Agents that affect nucleic acid metabolism such as rifamycins, which inhibit DNA-dependent RNA polymerase.

20.3 Ampicillin:- (Omnipen)

It is broad spectrum penicillin, because it also acts against Gram- negative bacteria. It is susceptible to inactivation by enzyme, β-lactamase. Ampicillin can be prepared microbiologically by incubation of microbes (*pseudomonas speicies*) with 6-aminopenicillanic acid and α-phenylglycine via enzyme acylation of 6-APA (amino penicillanic acid).

Ampicillin has an amino group in its side chain. Ampicillin is not degraded by gastric acid. Its oral absorption is incomplete. Food interferes with absorption of ampicillin.

Structure of Ampicillin:-

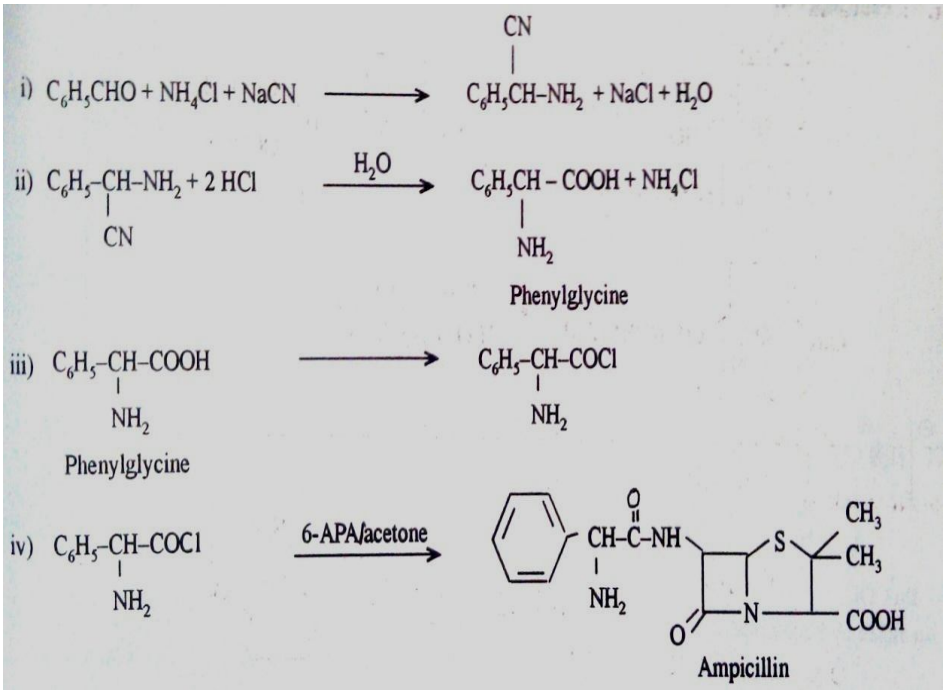


Ampicillin

IUPAC- (2S,5R,6R)-6-([(2R)-2-amino-2-phenylacetyl]amino)-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid

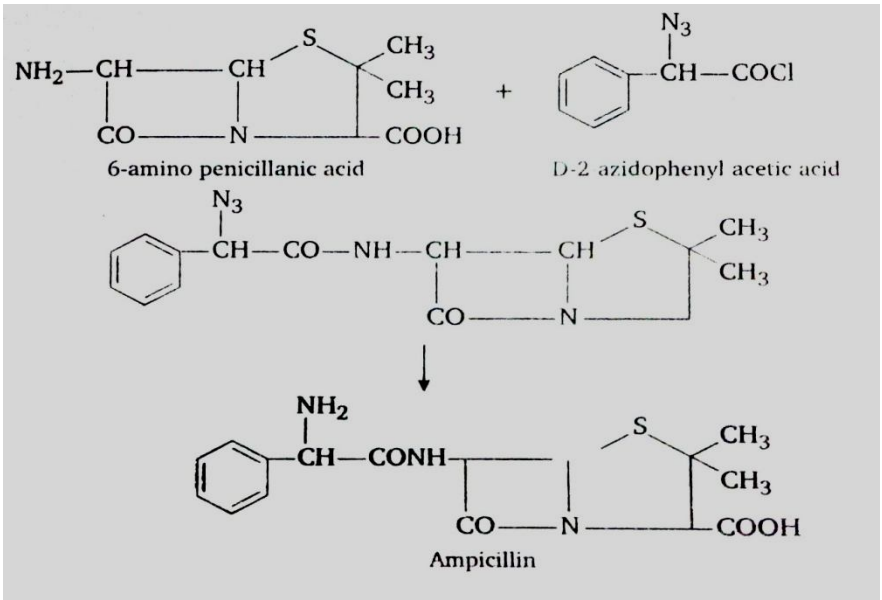
Synthesis of Ampicillin:-

Method-I



Method-II

It is prepared with the acylation of 6-aminopenicillanic acid (6-APA) with acid chloride from D-2-azidophenylacetic acid followed by a catalytic reduction.



Anpylrous ampicillin is a white odorless, crystalline powder, slightly soluble in water. The sodium salt has a bitter taste with hygroscopic properties. Ampicillin

is the drug of choice for gonococcal urethritis and upper respiratory tract infections.

Use:- Ampicillin is used for most acute infections of urinary tract. Respiratory tract infection including bronchitis, Otitis media, sinusitis etc. are generally treated with ampicillin. It is also a first line drug for meningitis and gonorrhea.

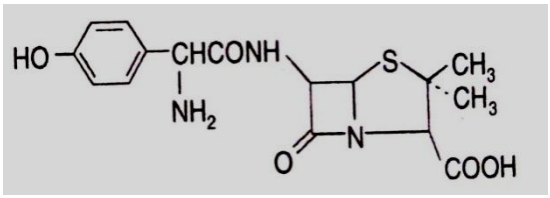
Averse effect:- Diarrhoea is frequent adverse effect after oral administration of ampicillin. It produce a high incidence of rashes, particularly, in AIDS, lymphatic leukemia and EB virus infections. Sometimes rashes may be of toxic nature.

20.4 Amoxicillin (Amoxil)

It is a moderate beta-lactam antibiotic used to treat baeterial infections caused by susceptible micro organisms. It is usually the drug of choice with the class because it is better absorbed following oral administration, than other beta-lactam antibiotics.

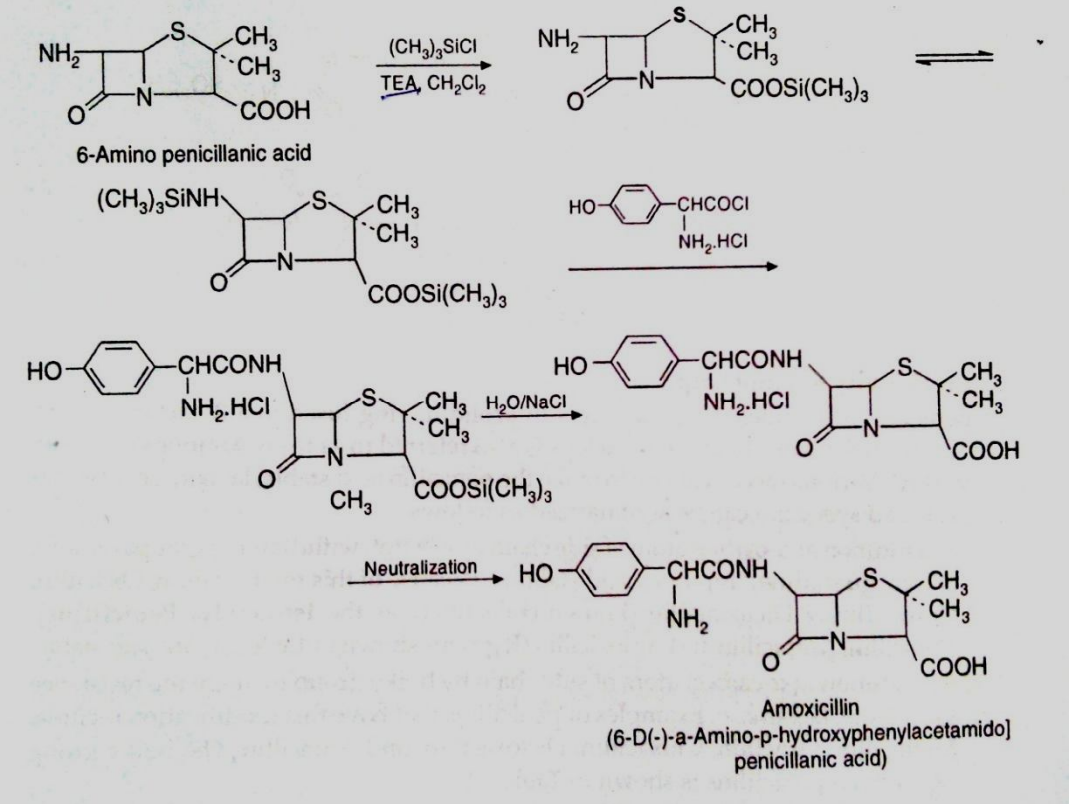
Amoxicillin is susceptible to degradation by beta-lactamase producing bacteria, and so may be given with clavulanic acid to decrease its susceptibility. It is commonly available by proprietary names such as "Augmentin".

Structure of Amoxicillin:-



IUPAC Name- [6-D (-)-α-amino-p-hydroxyphenylacetamido penicillanic acid]

Synthesis of Amoxicillin:-



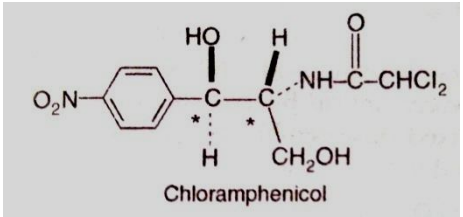
It is fine, white crystalline powder that is sparingly soluble in water. Its antibacterial spectrum is nearly identical to that of ampicillin.

20.5 Chloramphenicol (Chloromycetin, Mychel)

Chloramphenicol is a broad spectrum antibiotic. It is elaborated by *streptomyes venizulae*. It is now prepared synthetically. Chloramphenicol is a nitrobenzene derivative.

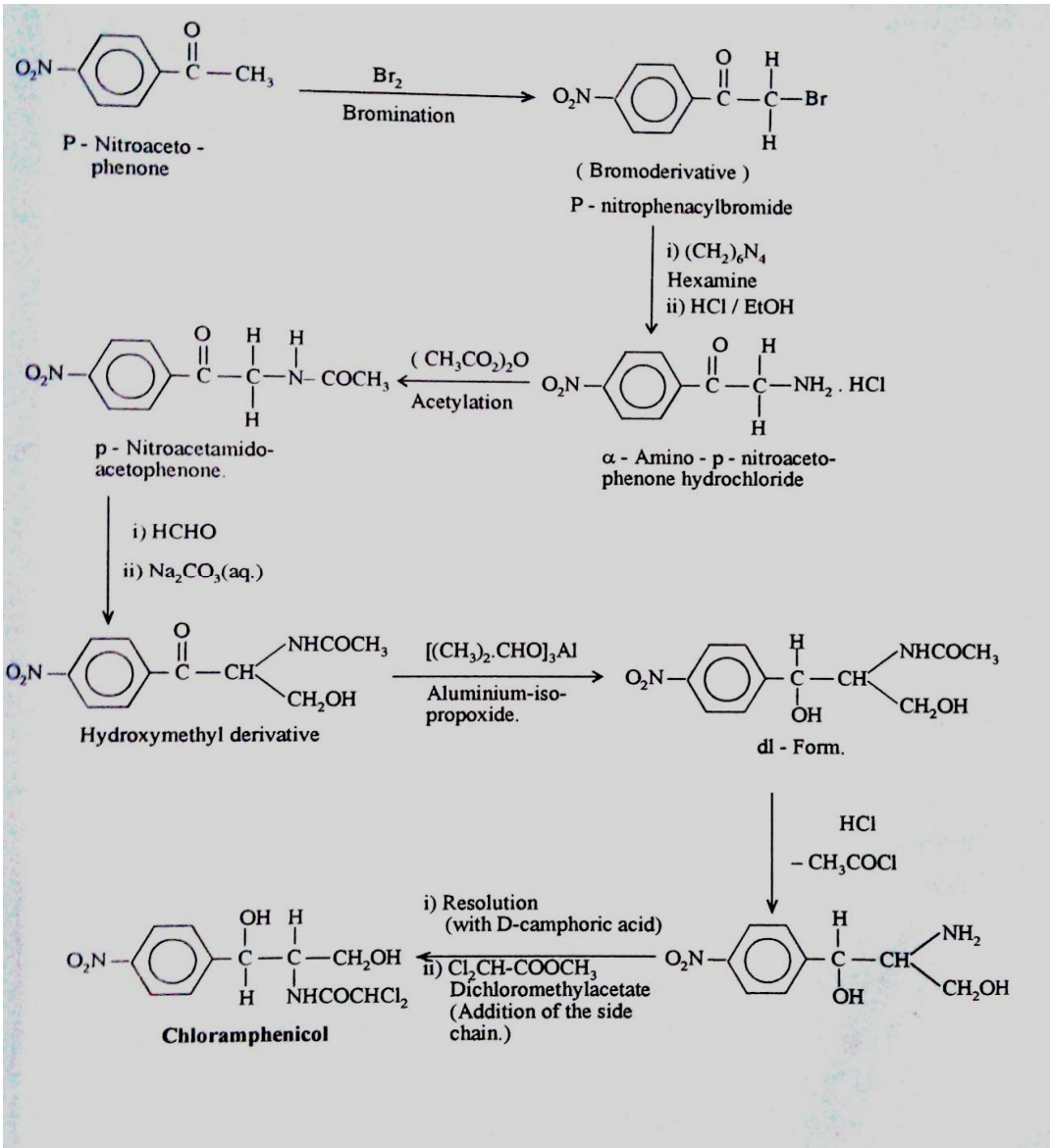
Chloramphenicol is bacteriostatice. It acts by inhibiting bacterial protein synthesis.

Structure of Chloramphenicol:-



IUPAC Name:- 2,2–Dichloro-N-[2-hydroxy-1-(hydroxylmethyl)-2-(4-nitrophenyl)ethyl]-acetamide.

Synthesis:-



Properties and Use: - It occurs as a fine yellowish white, needle like crystals. It is slightly soluble in water and has a bitter taste.

Chloramphenicol is an important drug for the treatment of typhoid fever caused by *salmonella typhi*, also give excellent result in haemophilus influenzae meningitis, Neisseria meningitis.

20.6 Cephalosporin

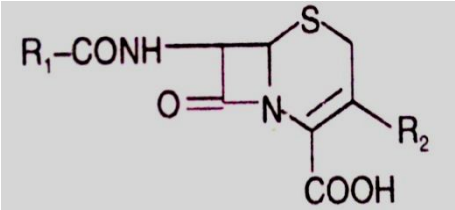
The cephalosporins are beta- lactum antibiotics isolated from cephalosporium Species and are prepared semi synthetically. These come under the class of 7-

amino cephalosporinic acid (7-ACA) derivatives and are much more acid stable than the corresponding 6-APA compounds.

The cephalosporin have a mechanism of action similar to that of penicillin's, mainly they inhibit the cross – linking of the peptidoglycane units in bacterial cell wall by inhibiting transpeptidase enzyme.

The cephalosporin nucleus, 7-amino cephalosporonic acid (7-ACA) was derived from cephalosporin C. Modification of the 7-ACA side chain resulted in the development of the useful antibiotic agents.

Basic Structure of Cephalosporin

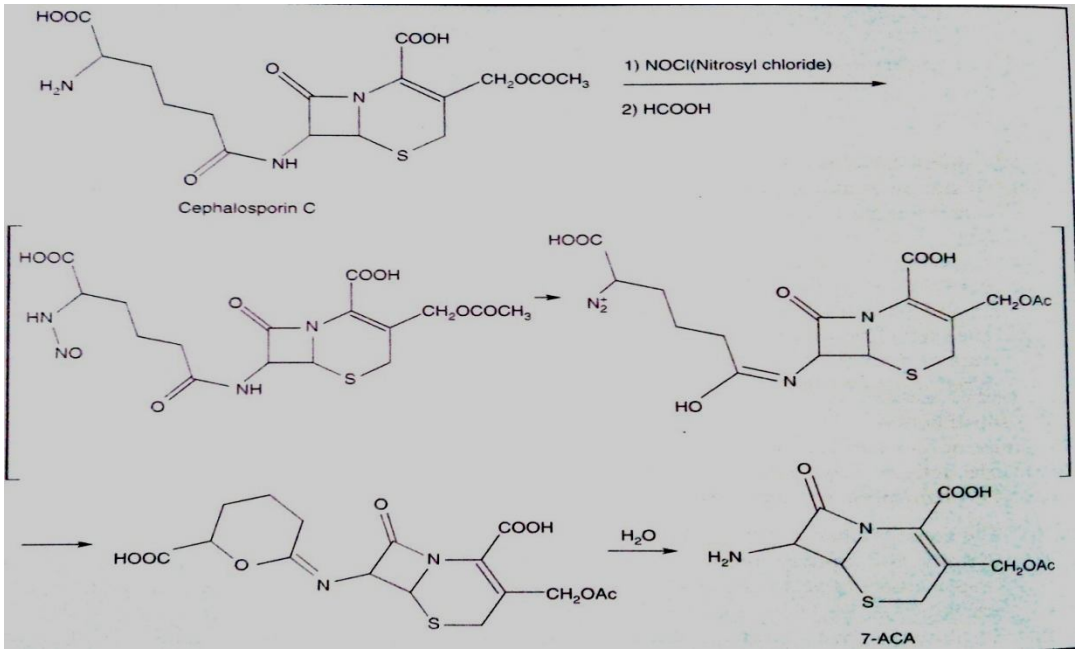


If-

	R_1	R_2
Cephalexin	C_6H_5-CH(NH_2)-	CH_3
Cefadroxil	HO-C_6H_4-CH(NH_2)-	CH_3

Synthesis of 7-amino cephalosporinic acid (7-ACA) from cephalosporin C:-

Cephalosporin C is isolated on an industrial scale by fermentation using "*Cephalosporium acremonium*"

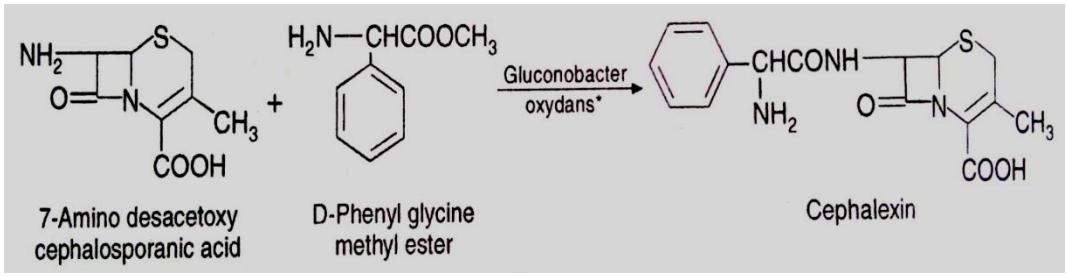


The key reaction based on a method for removing glutamate residue in cephalosporinic C, involves conversion of the primary amine in the molecule to a diazo function by reaction with nitrosyl chloride and formic acid. The diazo function can be displaced by oxygen from the enol form of the amide at the 7 position to form iminolactone. Hydrolysis of imine leads to 7-ACA.

Examples of cephalosporin derivatives:- cephalexin, cefadroxil, cefuroxime, ceftizoxime, cefazolin etc.

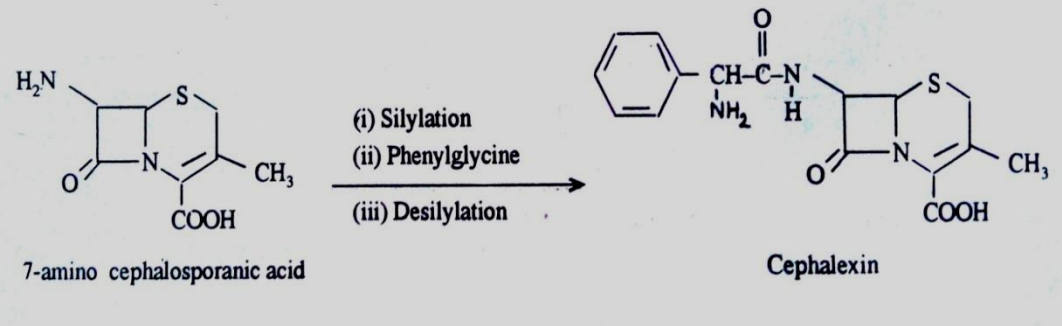
Synthesis of cephalexin:- (Keflex)

Method- I



In first method 7-aminodesacetoxy cephalospranic acid react with D-phenyl glycine methyl ester and give the final product i.e. Cephalexin

Method- II

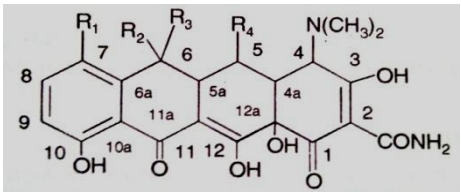


It is rapidly and completely absorbed from the GI tract and thus has become popular. It is acid stable used as the monohydrate, primarily for the treatment of respiratory, urinary, skin and soft tissue infections.

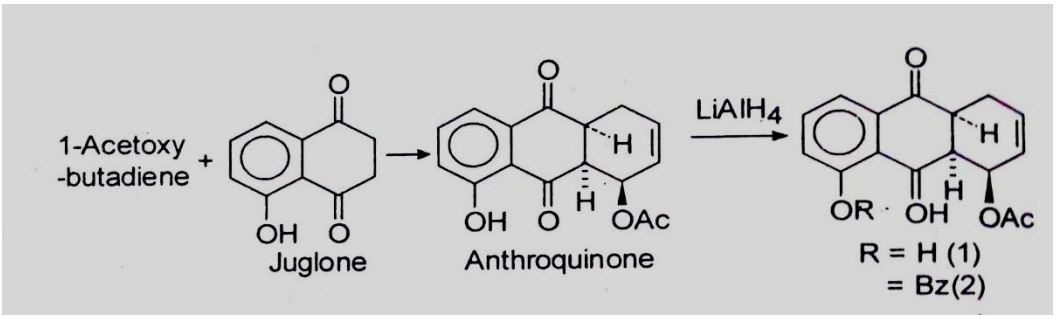
20.7 Tetracyclins

These are broad spectrum antibiotics. They are effective against gram positive organisms, actinomyces, rickettsiae and Chlamydia organisms. Chlortetracycline is obtained from "streptomyces aureofaciens"

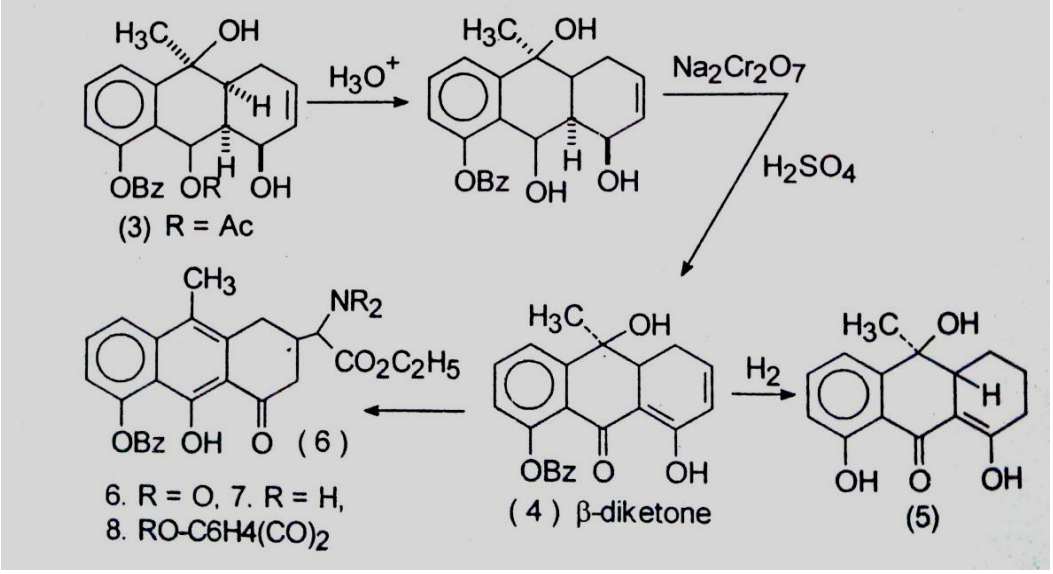
Basic Structure:-



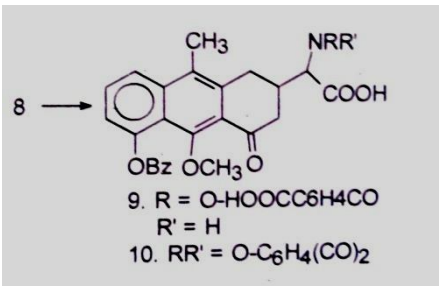
Synthesis:-



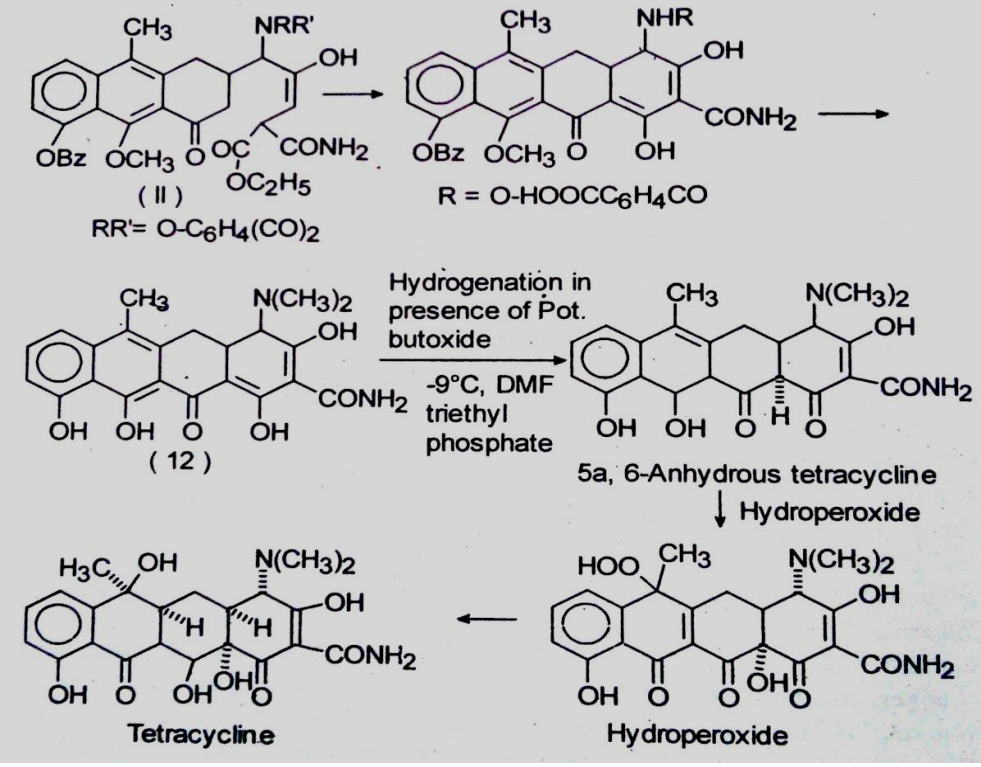
- The phenolic group was benzylated to the ether (2). This was converted to the methyl carbinol (3) with methyl magnesium iodide.



- The acetate rearranged during the later process, following hydrolysis of the acetate ester, the tris alcohol was oxidised with chronic acid in acetone to yield the enolised Beta-diketone (4).
- Hydrogenation yielded the tetracycline BCD-model compound (5).
- The beta ketone was condensed with ethyl nitroacetate then dehydrated to yield the nitro ester (6) reduced to the amino ester (7) with zinc dust in acetic acid and converted to the phthalimidoester (8) by reacting with carbethoxy phthalimide.
- Methylation with methyl iodide/silver oxide followed by saponification and recrystallization of intermediate phthalimide (9) from hot acid (10).



- Treatment of (10) with phosphorus pentachloride formamide followed by $C_2H_5OMgCH(CO_2C_2H_5)CONH_2$ gave the N-phthaloylgalylo malonate (11), which was cyclised by dicylanion in dimethyl sulfoxide to the hydronaphthalene (12), in turn hydrolysed with hydrogen bromide in acetic acid and methylated with methyl iodide in THF to ± (-12-deoxy-5a,6-anhydrotetracyclin) (12).

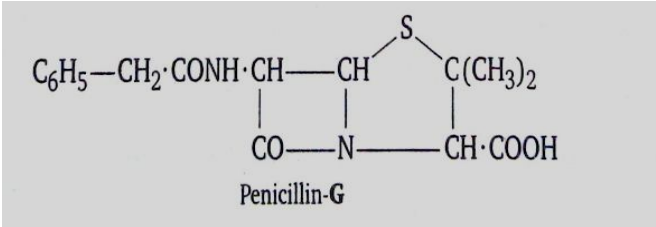


20.8 Penicillin G

Penicillin is a generic term which refers to a class of compounds of the molecular formula $CH_{11}N_2O_4SR$, produced by various strains of "*penicillium notatum*", "*penicillium chrysogenum*" and some other fermented moulds.

Penicillin G or benzyl penicillin is the most important commercial penicillin. It is a narrow spectrum antibiotic and its activity is limited primarily to gram – positive bacteria and few others

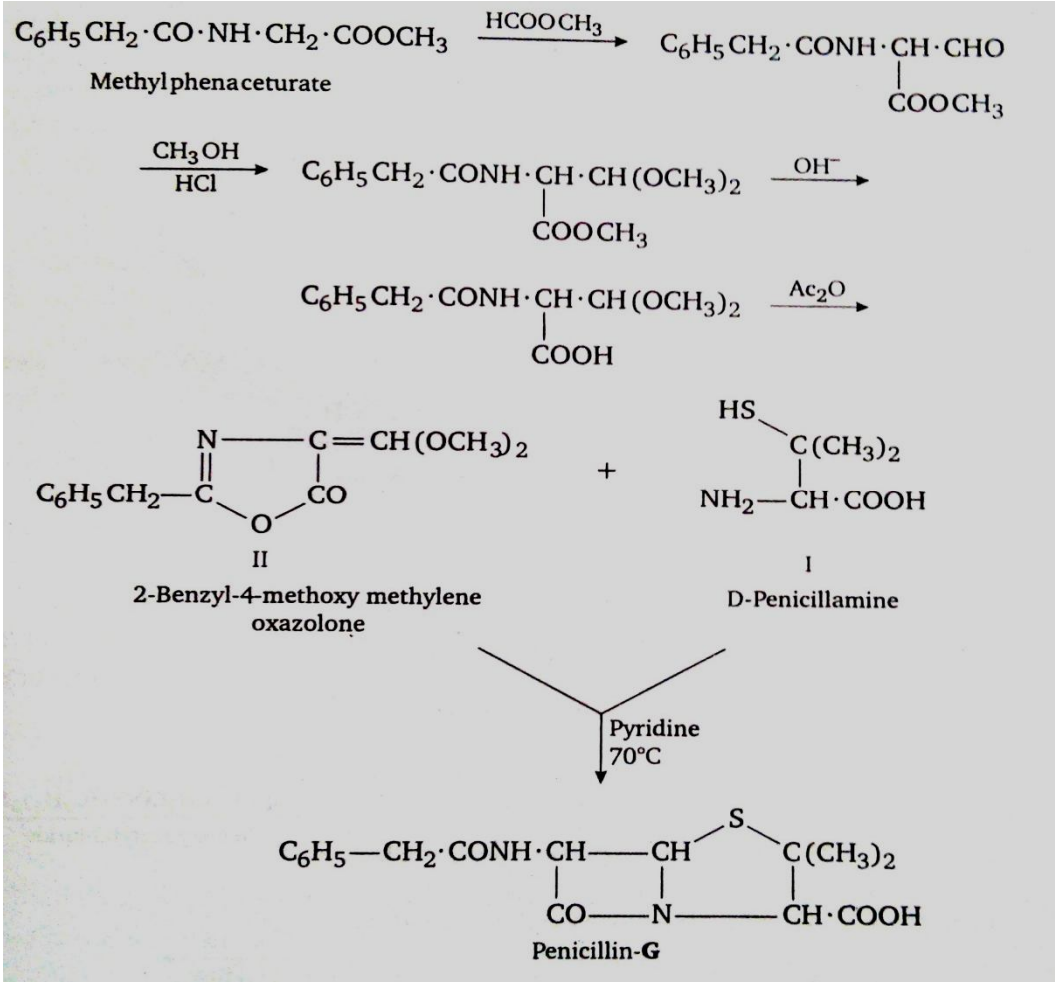
Structure of Penicillin G:-



IUPAC Name:- (2S,5R,6R)-3,3-dimethyl-7-oxo-6-[(2-phenylacetyl)amino]-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid

Synthesis:-

Penicillin G is obtained in small quantities by condensing D-penicillamine (I) with 2-benzyl-4-methoxy methylene oxazolone (II), in pyridine at 70°C. The benzyl penicillin was isolated as the crystalline triethyl amine salt. The starting material II was prepared by methylphenaceturate in the following way-



20.9 Streptomycin

It is a member of aminoglycoside antibiotics, obtained naturally and semisynthetically, having polybasic amino groups linked glcosidically to two or more amino sugars (streptidine, 2-deoxy streptamine, garosamine) residues.

Streptomycin was first discovered in 1944 by Wakesman et. at. from *streptomyces giseus.* Unlike penicillin (Which was a chance discovery) streptomycin was a product of deliberate search for drugs effective against Gram-negative bacteria. Neomycin was next drug (1949) to be isolated, but could not be used systemically.

All amino glycosides are produced from soil *actinomycetes* and have many common properties.

Streptomycin assumed great importance because this antibiotic is active against tubercle bacilli, but now practically restricted to treatment of tuberculosis.

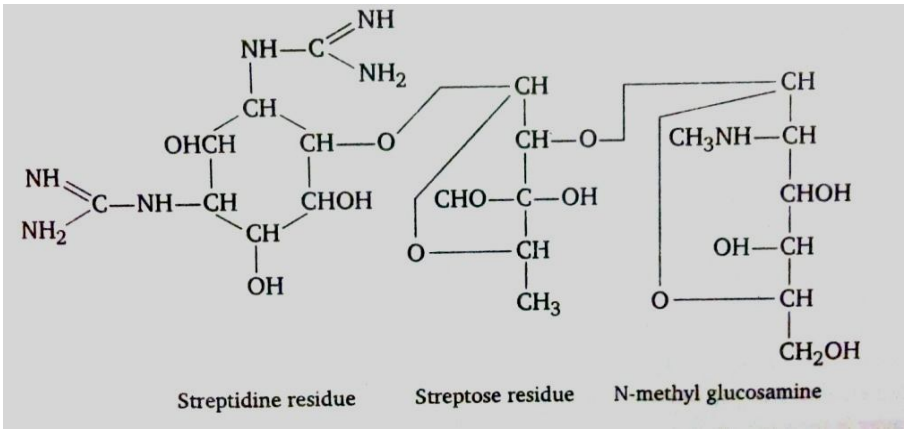
The mode of action of this antibiotic is in 2 steps:

- Transport of the streptomycin through the bacterial cell wall and cytoplasmic membrane.
- Binding to ribosomes resulting in inhibition of protein synthesis.

Adverse effects:- By streptomycin, rashes, eosinophilia, fever, exofoliative dermatitis, amphylaxis, scotoma, parethesias, pain at injection site, auditory disturbances are found as adverse effects.

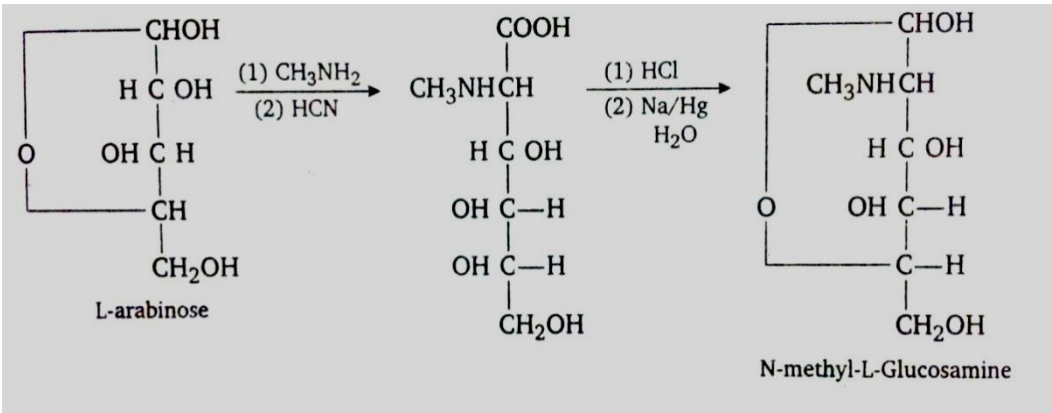
Uses:- It is used in tuberculosis, urinary tract infections, peritonitis, septicaemia, subacute endocarditis, tularemia and plague.

Structure:-



Synthesis:-

Synthesis of N-methyl glucosamine: It is prepared from L-arabinose.



20.10 Summary

Antibiotics or antibacterials are a type of antimicrobial used specifically against bacteria; antibiotics are used to treat various infections. and are often used in medical treatment of bacterial infections. They may either kill or inhibit the growth of bacteria.

With advances in medicinal chemistry, most modern antibacterials are semi synthetic modifications of various natural compounds. These include, for example, the beta-lactam antibiotics, which include the penicillins (produced by fungi in the genus Penicillium), the cephalosporins, and the carbapenems. Compounds that are still isolated from living organisms are the aminoglycosides, whereas other antibacterials—for example, the sulfonamides, the quinolones, and the oxazolidinones—are produced solely by chemical synthesis.

Side-effects range from mild to very serious depending on the antibiotics used, the microbial organisms targeted, and the individual patient.

20.11 Glossary

- **Amphylaxis:-** This is a serious allergic reaction that is rapid in onset and may cause death. It typically causes a number of symptoms including an itchy rash, throat swelling, and low blood pressure. Common causes include insect bites and stings, foods, and medications.
- **Antifungal Agents:-** is a pharmaceutical fungicide used to treat and prevent mycoses such as athlete's foot, ringworm, candidiasis (thrush), serious systemic infections such as cryptococcal meningitis, and others.
- **Hygroscopic:-** Describing the ability or tendency of a material to take up moisture readily from the surrounding air or other moist materials.
- **Gonococcal Urethritis:-** In men, purulent discharge usually indicates a urethritis of gonococcal nature, while clear discharge indicates urethritis of non-gonococcal nature.
- **Scotoma:-** This is a defect of vision in a defined area of the visual field in one or both eyes.
- **Septicaemia:-** Septicemia is also known as bacteremia or blood poisoning. Septicemia occurs when a bacterial infection enters the bloodstream. Untreated septicemia can quickly progress to sepsis, which

is a serious complication of an infection characterized by inflammation throughout the body. This inflammation can cause blood clots and block oxygen from reaching vital organs, resulting in organ failure and death in some cases.

- **Tularemia:-** This is a serious infectious disease caused by the bacterium *Francisella tularensis*. The bacteria can penetrate into the body through damaged skin and mucous membranes, or through inhalation. Humans are most often infected by tick bite or through handling an infected animal.

20.12 Review questions /comprehensive questions

1. What are antibiotics? Discuss in detail with the synthesis of any one antibiotic drug.
2. Give the synthesis of any two-
    - Ampicillin
    - Amoxicillin
    - Cephalosporin.
3. Give the basic structure and basic method for the synthesis of antibiotics.
4. Give the synthesis of Penicillin G.
5. Give the Synthesis of tetracycline.
6. How 7-amino cephalosporinic acid (7-ACA) Synthesize from cephalosporin C?
7. Discuss about streptomycin. Give structure and synthesis of N-methyl glucosamine.

20.13 eferences and suggested readings

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