Chapter

3

DISTRIBUTION OF DRUGS

Upon entering the systemic circulation, most drugs are simultaneously distributed throughout the body and eliminated. The fate of a drug after absorption is known as Drug Disposition.

This includes Drug Distribution and Elimination. Distribution usually occurs much more rapidly than elimination.

The rate of distribution to the tissue of each organ is determined by the blood flow through the organ and the ease with which the drug molecules cross the capillary bed and penetrate the cells of the particular tissue.

Drugs are eliminated from the body by metabolism and excretion. The liver is the major site of drug metabolism, but many other tissues contain metabolic enzymes and may contribute to the bio-transformation of certain drugs. The kidneys play a major role in the excretion of drugs and or their metabolites. Some drugs are excreted into the gastrointestinal tract by way of the bile and may be eliminated in the faeces.

Since the rates of drug distribution and elimination determine drug concentrations at receptor sites and often determines the frequency of drug administration.

Drug Distribution – is the reversible transfer of a drug between the blood and the extravascular fluids and tissues. It is a passive process and the driving force is the concentration gradient between blood and extravascular fluids and tissues.

Drug molecules are distributed throughout the body by means of the circulation of blood. The entire blood volume is pumped through the heart each minute, so that within moments after a drug enters the blood stream it is diluted into the total blood volume.

Almost all drugs can readily leave the capillaries and are rapidly diluted into a much larger volume. The permeability characteristics of capillaries, except in the brain, are like those of a rather "leaky" lipoid sieve. All drugs with molecular weights up to 500 or 600 that are free in the circulation readily penetrate the interstitial fluid bathing the cells.

Steps involved in drug distribution are,

- Permeation of free or unbound drug present in blood through the capillary walls and entry into the interstitial/extracellular fluid (ECF).

- Permeation of drug present in the ECF through the membrane of tissue cells and into the intracellular fluid. -this is the rate limiting steps and is dependent on,
  a) Rate of perfusion to the extracellular tissue
  b) Membrane permeability of the drug

Factors Influencing The Distribution of Drugs

1. Tissue permeability of the drug
   a) Physicochemical properties of the drug like molecular size pk_a and o/w partition coefficient
   b) Physiological barriers to diffusion of drugs
2. Organ/tissue size and perfusion rate
3. Binding of drugs to tissue components:
   a) Binding of drugs to blood components
   b) Binding of drugs to extravascular tissue proteins
4. Miscellaneous factors:
   a) Age
   b) Pregnancy
   c) Obesity
   d) Diet
   e) Disease states
   f) Drug interactions

1. Tissue permeability of the drug

a) Physicochemical Properties of the Drug

Important physicochemical properties of drug that influence its distribution are, molecular size, degree of ionization, partition coefficient and stereochemical nature.

Almost all drugs having molecular weight less than 500 to 600 Daltons easily cross the capillary membrane to diffuse into the extracellular interstitial fluids. Only small, water-soluble molecules and ions of size below 50 Daltons enter the cell through aqueous filled channels whereas those of larger size are restricted unless a specialized transport system exists for them.

The degree of ionization of a drug is an important determinant in its tissue penetrability. The pH of the blood and the extravascular fluid also play a role in the ionization and diffusion of drugs into cells. A drug that remains unionized at these pH values can permeate the cells relatively more rapidly. All drugs that ionize at plasma pH (i.e. polar, hydrophilic drugs), cannot penetrate the lipoidal cell membrane and tissue permeability is the rate-limiting step in the distribution of such drugs. Only unionized drugs which are generally lipophilic, rapidly cross the cell membrane.

Among the drugs that have same o/w partition coefficient but differ in the extent of ionisation at blood pH, the one that ionises to a lesser extent will have greater penetrability than that which ionises to a larger extent.

For eg: pentobarbital and salicylic acid have almost the same Ko/w but the former is more unionised at blood pH and therefore distributes rapidly.

The influence of drug pKa and Ko/w on distribution is illustrated by the example of thiopental, a nonpolar, lipophilic drug, largely unionised at plasma pH, readily diffuses into the brain whereas penicillin's which are water soluble and ionised at plasma pH do not cross the blood -brain barrier.

Since the extent to which a drug exists in unionised form governs the distribution pattern, situations that result in alteration of blood pH affect such a pattern,

Eg: acidosis (metabolic or respiratory) results in decreased ionisation of acidic drugs and thus increased intracellular drug concentration and pharmacological action, whereas sodium bicarbonate induced alkalosis is sometimes useful in the treatment of barbiturate (and other acidic drugs) poisoning to drive the drug out and prevent further entry into the CNS and promote their urinary excretion by favouring ionisation.

In case of polar drugs where permeability is the rate-limiting step in the distribution, the driving force is the effective partition coefficient of drug. It is calculated by the following formula:

Effective Ko/w = (Fraction unionised at pH 7.4) (Ko/w of unionised drug)

b) Physiological barrier to distribution of drugs

Important physiological barriers are,

- Simple capillary endothelial barrier
- Simple cell membrane barrier
- Blood-brain barrier
- Blood-CSF barrier
- Blood-placental barrier
- Blood-testis barrier

Simple capillary endothelial barrier

All drugs, ionized or unionized, with a molecular size less than 600 Daltons, diffuse through the capillary endothelium and into the interstitial fund. Only drugs bound to the blood components are restricted because of the large molecular size of the complex.

Simple cell membrane

Once a drug diffuses from the capillary wall into the extracellular fluid, its further entry into cells of most tissues is limited by its permeability through the membrane that lines such cells. Such a simple cell membrane is similar to the lipoidal barrier in the GI absorption of drugs. **(Fig. 3.1)**

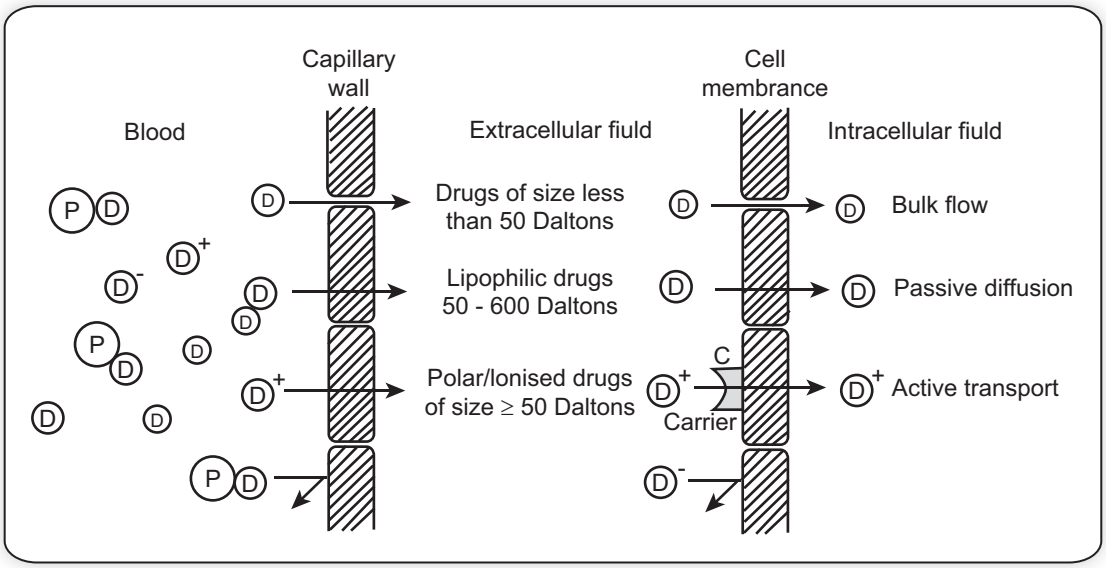

Fig. 3.1 : Plasma membrane barrier

Blood brain barrier (BBB)

The capillaries found in the brain are highly specialized and less permeable to water soluble drug. The brain capillaries consist of endothelial cells joined to one another by continuous tight intercellular junction comprising called as the blood brain barrier. The presence of special cells called as pericytes and astrocytes, which are the elements of the supporting tissue found at the base of endothelial membrane, form a solid envelope around the brain capillaries. As a result, the intercellular (paracellular) passage is blocked, so for a drug to enter from the capillary circulation into the brain, it has to pass through the cells (transcellular).

A solute may thus gain access to brain via only one of the two pathways:

1. Passive diffusion through the lipoidal barrier: which is restricted to small molecules (with a molecular weight less than 700 Daltons) having high o/w partition coefficient.
2. Active transport of essential nutrients such as sugars and amino acids. Thus, structurally similar foreign molecules can also penetrate the BBB by the same mechanism.

The effective partition coefficient of thiopental, a highly lipid soluble drug is 50 times that of pentobarbital and crosses the BBB much more rapidly. Most antibiotics such as penicillin which are polar, water-soluble and ionised at plasma pH, do not cross the BBB under normal circumstances.

The selective permeability of lipid-soluble moieties through the BBB makes appropriate choice of a drug to treat CNS disorders.

Eg: Parkinsonism, a disease characterized by depletion of dopamine in the brain, is treated with levodopa, which can penetrate the CNS where it is metabolised to dopamine since dopamine as such does not cross the BBB.

Three different approaches to promote crossing the BBB by drugs:

i) Use of permeation enhancers such as dimethyl sulphoxide (DMSO).

ii) Osmotic disruption of the BBB by infusing internal carotid artery with mannitol

iii) Use of dihydropyridine redox system as drug carriers to the brain.

The lipid soluble dihydropyridine is linked as a carrier to the polar drug to form a prodrug that readily crosses the BBB. In the brain, the CNS enzymes oxidize the dihydropyridine moiety to the polar pyridinium ion form that cannot diffuse back out of the brain. As a result, the drug gets trapped in the CNS. Such a redox system has been used to deliver steroidal drugs to the brain.

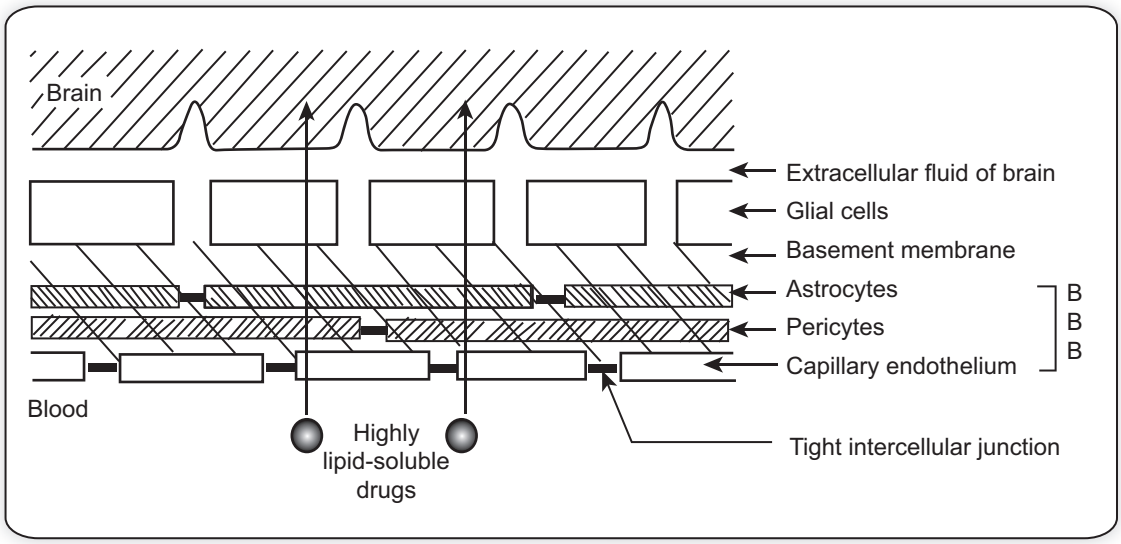

Fig. 3.2 : Blood Brain Barrier

Blood -cerebrospinal fluid barrier

The cerebrospinal fluid (CSF) is mainly produced by a special tissue called the choroid plexus, which is present in the lateral, third, and fourth ventricles of the brain. CSF is very similar in chemical composition to the brain's extracellular fluid (ECF).

In the choroid plexus, the blood capillaries are lined with cells that have small gaps (fenestrations). Through these gaps, many substances, including drugs, can move freely into the space between the capillary wall and the cells of the choroid plexus.

However, the cells of the choroid plexus themselves are tightly connected to each other by tight junctions. This creates a selective barrier called the blood–CSF barrier, which works somewhat like the blood–brain barrier (BBB).

The ability of drugs to cross this barrier depends on their properties:

- Highly lipid-soluble drugs can cross the blood–CSF barrier easily.
- Moderately lipid-soluble or partially ionized drugs cross more slowly, so their entry into the CSF is limited.

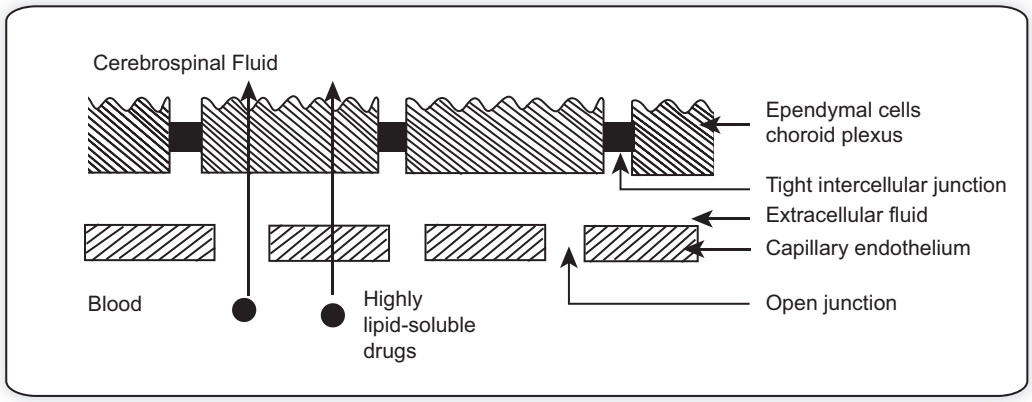

Fig. 3.3 : Blood-CSF barrier

Blood placental barrier

The placental barrier is the protective boundary between the mother's blood and the baby's (foetal) blood. It is made up of several tissue layers, mainly the trophoblast, the basement membrane, and the endothelium of foetal blood vessels. This barrier controls which substances can pass from the mother to the foetus.

Many drugs are small in size (molecular weight less than 1000 Daltons) and fat-soluble. Such drugs can cross the placental barrier quite easily by simple diffusion. Examples include alcohol (ethanol), sulphonamides, barbiturates, gaseous anaesthetics, steroids, narcotic painkillers, anticonvulsants, and some antibiotics.

Because these drugs can reach the foetus quickly, and their effects on the developing baby may sometimes be harmful or cause birth defects (teratogenic effects), it is safest to avoid the use of most drugs during pregnancy unless absolutely necessary. **(Fig. 3.4)**

Blood – testis barrier

Inside the testis, there is a special protective barrier called the blood–testis barrier. This barrier is formed by tight junctions between neighbouring Sertoli cells.

The main function of this barrier is to control what can enter and leave the area where sperm cells develop. Because of it, many substances, including certain drugs, cannot easily reach the developing sperm cells (spermatocytes and spermatids).

This protection is important because it helps maintain the right environment for sperm production and shields the developing sperm from harmful chemicals and immune system attack.

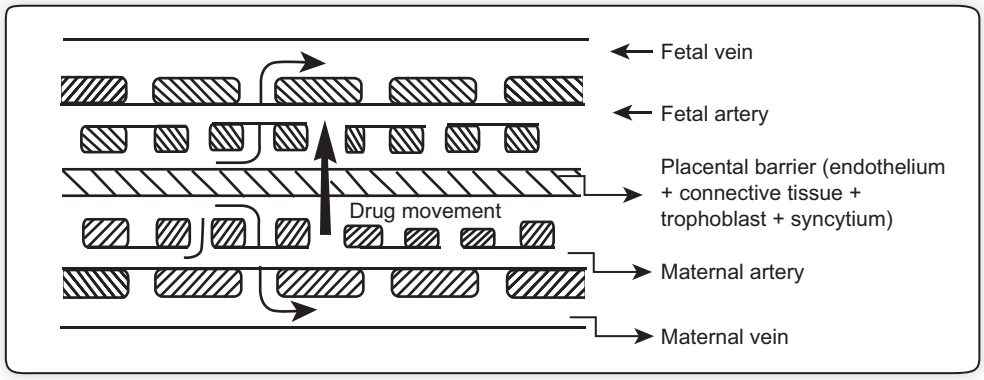

Fig. 3.4 : Blood- Placental barrier

2. Organ /Tissue size and perfusion rate

Drug distribution will be perfusion rate-limited if,

(i) The drug is highly lipophilic.

(ii) The membrane across which the drug is supposed to diffuse is highly permeable such as those of the capillaries and the muscles.

Only highly lipophilic drugs such as thiopental can cross the most selective of the barriers like the BBB, highly permeable capillary wall permits passage of almost all drugs (except those bound to plasma proteins). In both these cases, the rate limiting step is the blood flow or perfusion to the tissues. More blood flow, faster is the distribution.

Perfusion rate is the volume of blood that flows per unit time per unit volume of the tissue. It is expressed in ml/min/ml of the tissue.

Highly perfused tissues such as lungs, kidney, liver, heart and brain are rapidly equilibrated with lipid soluble drug **(Table - 3.1).**

If $K_{t/b}$ is the tissue/blood partition coefficient of drug then the first order distribution rate constant, K_t is given by following equation

$$K_t = \frac{\text{Perfusion rate}}{K_{t/b}} \qquad (3.1)$$

The tissue distribution half- life is given by equation,

$$\text{Distribution half-life} = \frac{0.693}{K_t} = \frac{0.693K_{t/b}}{\text{perfusion rate}} \qquad (3.2)$$

The extent to which a drug is distributed in a particular tissue or organ depends upon the size of the tissue (i.e. tissue volume) and the tissue/blood partition co-efficient of the drug.

Table - 3.1 : Relative volume of different organs, blood flow and perfusion rate under basal condition assuming the total body volume to be 70 liters

Organ/Tissue	% Of body volume	Blood flow (ml/min)	% cardiac output	Perfusion rate (ml/min/ml)
1. Highly perfused				
Lungs	0.7	5000	100.0	10.2
Kidney	0.4	1250	25.0	4.5
Liver	2.3	1350	27.0	0.8
Heart	0.5	200	4.0	0.6
Brain	2.0	700	14.0	0.5
2. Moderatly perfused				
Muscles	42.0	1000	20.0	0.034
Skin	15.0	350	7.0	0.033
3. Poorly perfused				
Fat (Adipose)	10.0	200	4.0	0.03
Bone (Skeleton)	16.0	250	5.0	0.02

Eg: Thiopental a lipophilic drug has a high tissue/blood partition coefficient towards the brain and still higher for adipose tissue. Since the brain (site of action) is a highly perfused organ, following iv injection, thiopental readily diffuses into the brain showing a rapid onset of action. Adipose tissue being poorly perused, takes longer to get distributed with the same drug. But as the concentration of thiopental in the adipose proceeds towards equilibrium, the drug rapidly diffuses out of the brain and localizes in the adipose tissue whose volume is more than 5 times that of brain and has greater affinity for the drug. The result is rapid termination of action of thiopental due to such a tissue redistribution.

3. Binding of drugs to tissue components:(Discussed in Chapter Protein Binding)

a) Binding of drugs to blood components

b) Binding of drugs to extravascular tissue proteins

4. Miscellaneous factors

Age

Differences in distribution pattern of a drug in different age groups are mainly due to differences in:

(a) Total body water (both intracellular and extracellular)- is much greater in infants

(b) Fat content is also higher in infants and elderly

(c) Skeletal muscles are lesser in infants and in elderly

(d) Organ composition- the BBB is poorly developed in infants, the myelin content is low and cerebral blood flow is high, hence greater penetration of drugs in the brain

(e) Plasma protein content- low albumin content in both infants and in elderly

Obesity

The adipose tissue content is high in obese people, which leads to take up of a large fraction of lipophilic drugs even though the perfusion through it is low. The high fatty acid levels in obese persons alter the binding characteristics of acidic drugs.

Diet

A diet high in fats will increase the free fatty acid levels in circulation thereby affecting binding of acidic drugs such as NSAIDs to albumin.

Disease States

In disease states the of drug distribution characteristics is altered due to

a) Altered albumin and other drug-binding protein concentration.

b) Altered or reduced perfusion to organs or tissues.

c) Altered tissue pH.

An example of altered permeability of the physiological barriers is that of BBB. In meningitis and encephalitis, the BBB becomes more permeable and thus polar antibiotics such as penicillin G and ampicillin which do not normally cross it, gain access to the brain. In a patient suffering from congestive cardiac failure (CCF), the perfusion rate to the entire body decreases affecting distribution of all drugs.

Drug Interactions

Drug interactions that affect distribution are mainly due to differences in plasma protein or tissue binding of drugs.

Pregnancy

During pregnancy, the growth of uterus, placenta and foetus increases the volume available for distribution of drugs. The foetus represents a separate compartment in which a drug can distribute. The plasma and the ECF volume also increase but there is a fall in albumin content.

Distribution Volumes

A drug in blood circulation distributes to various organs and tissues. When distribution equilibrium is reached, different organs and tissues containing varying concentrations of drug can be determined by the volume of tissues in which the drug is present.

There exists a constant relationship between the concentration of drug in plasma, C, and the amount of drug in the body, X.

$$X \text{ a } C \qquad (3.3)$$

$$X = V_d C \qquad (3.4)$$

V_d=proportionality constant having the unit volume and called as **apparent volume of distribution.** It is defined as the hypothetical volume of body fluid into which a drug is dissolved or distributed . it is called as apparent volume because all parts of the body equilibrated with the drug do not have equal concentration.

Hence V_d is

$$\frac{\text{Amount of drug in the body (X)}}{\text{Plasma drug concentration (C)}} \qquad (3.5)$$

$$\text{Or} \quad V_d = \frac{X}{C} \qquad (3.6)$$

There is usually a considerable difference between the apparent volume of distribution of a drug and the real volume in which it distributes. The apparent volume of distribution is simply a proportionality constant relating the plasma concentration to the total amount of drug in the body. The apparent volume of distribution of a drug may vary in man from 0.04 l/kg (plasma volume) to 20.l/kg or more.

The real volume of distribution has direct physiological meaning and is related to the body water. The real distribution volume of a drug is related to body water and can never exceed total body water, that is, 58 per cent of body weight or about 41 in a normal 70-kg man.

The plasma volume can be determined by use of substance of high molecular weight or substance that is totally bound to plasma albumin. Certain dyes such as Evans blue are essentially confined to the circulating plasma and can be used to determine plasma volume.

The extracellular fluid (ECF) volume can be determined by substance that easily penetrate the capillary membrane and rapidly distribute throughout the ECF but do not cross the cell membrane. Certain substances such as chloride and bromide ions distribute rapidly into the interstitial fluid but do not readily cross cell membranes so they may be used to estimate extracellular water. The volume of total body water may be approximated by means of heavy water (D_2O) or certain lipid-soluble but poorly tissue-bound substances such as antipyrine. **(Table - 3.2)**

Table - 3.2 : Markers used to measure the volume of real physiological compartment

Physiological fluid compartment	Markers used	Approximate volume (litres)
Plasma	Evans blue, indocyanine green, albumin	3
Erythrocytes	C_r -51	2
Extracellular fluid	Non-metabolizable saccharides like insulin, mannitol & radioisotopes of selected ions Na^+, Cl^-, Br^-	15
Total body water	Deuterium oxide, antipyrine	42

Since the markers are not bound or negligibly bound to plasma or tissue proteins, their apparent volume of distribution is same as their true volume of distribution. The situation is different with most drugs which bind to plasma proteins or extravascular tissues or both. Certain generalizations can be made regarding the apparent volume of distribution of such drugs:

- Drugs which bind selectively to plasma proteins or other blood components, eg: warfarin (i.e. those that are less bound to extravascular tissues), have apparent volume of distribution smaller than their true volume of distribution. The V_d of such drugs lies between blood volume and total blood volume (i.e. between 6 to 42 litres); for eg: warfarin has a V_d of about 10 litres.
- Drugs which bind selectively to extravascular tissues, eg. chloroquine (i.e. those that are less bound to blood components), have apparent volume of distribution larger than their real volume of distribution. The V_d of such drugs is always greater than 42 litres or total blood volume for eg: chloroquine has a V_d of approximately 15,000 litres. Such drugs leave the body slowly and are generally more toxic than drugs that do not distribute deeply into body tissue.

Method for studying drug distribution pattern

It is essential to understand the drug distribution pattern in drug discovery so as to provide evidence of the specificity and selectivity of drugs. Studies for determining drug distribution pattern includes,

1. **Micro dialysis:**

   This technique involves insertion of very fine probes into the tissues of the living body, mostly into fluid spaces, such as the CSF, and other extracellular fluids where possible. The probes consist of at least two concentric tubes, and a semipermeable membrane separating them, positioned such that an artificial extracellular dialysis fluid can be slowly infused through the probe and past the membrane. Unbound drug molecules in the tissues surrounding the probe diffuse into the flowing dialysate, which is then collected for analysis. The limitation of this technique is its invasive nature.

2. **Matrix-assisted laser desorption ionization-mass spectrometry imaging (MALDI-MSI):**

   It is used to study drug localization within microenvironmental tissue compartments. Thin sections of animal tissues are exposed to pharmaceutical drugs by either spotting or submerging. MALDI-MSI is then performed on the tissue to localize drug-distribution patterns.

3. **Autoradiography:**

   In this approach, the radioactively labelled drug substance under investigation is administered to test animals, usually mice or rats, which are killed at suitable time intervals, and frozen sections of tissues or of the whole body prepared. An image of the radiation is obtained by placing the sections next to a photographic emulsion. Thus, if a whole-body section is used, and the radioactivity is specifically localized in highly perfused organs such as the lungs and liver, the image will reveal this. Comparison with a normal photograph of the slice is used to identify which tissues contain the radioactivity. Densitometry can be used to quantify the relative amounts of radioactivity. This technique has been used to show highly-specific localization of drugs in tissues.

4. Positron emission tomography (PET):

Synthetic radioactive isotopes (e.g. ^{11}C, ^{13}N,^{15}O and ^{18}F) with atomic masses less than the naturally occurring stable isotopes have half-life values of 2-110 minutes and emit positrons that interact with electrons to emit gamma radiation that can be detected outside the body. The isotopes are generated in a cyclotron and incorporated into the drug molecule immediately before the administration of the drug. PET scanning permits the production of images of live organisms including humans.

Drug Binding in Plasma

Binding to plasma proteins may have a marked effect on the distribution and pharmacologic effects of a drug and on the rate at which it is eliminated from the body. The high molecular weight of the plasma proteins prevents passage across the capillary walls and assures retention in the vascular space.

The fraction of drug in the plasma bound to plasma protein is not immediately available for distribution into the extravascular space or for certain modes of elimination. The interaction of a drug with plasma proteins is a rapidly reversible process. As free (unbound) drug diffuses from the capillaries, bound drug dissociates from the protein until drug in the extravascular water equilibrates with free drug in the plasma.

Drug binding to plasma proteins may involve, ionic, van der Waals, hydrogen or hydrophobic bonds.

The most important contribution to drug binding in the plasma is made by albumin which comprises about one half of the total plasma proteins. In healthy individuals. albumin concentration in the plasma is about 4 g/100 ml. Lower levels are found during pregnancy (about 3.5 g/100 ml during the last trimester) in many disease states.

Albumin strongly binds a wide variety of drugs and is usually the only protein in the plasma to interact significantly with drug molecule.

The drug-protein interaction may be described by the law of mass action,

$$D_F + \text{Free sites} \; k_1 \rightleftharpoons k_2 D_B \qquad (3.7)$$

D_F, D_B are the free and bound drug

k_1 and $k_2 \rightleftharpoons$ are the rate constants for association and dissociation respectively, thus,

$$K = \frac{k_1}{k_2} = \frac{[D_B]}{[D_F]\,[\text{Free sites}]} = \frac{[D_B]}{[D_F]\,(n[P] - [D_B])} \qquad (3.8)$$

where, K is the equilibrium association constant, n is the number of binding sites per mole of protein, and D_F, D_B and *P* are the molar concentration of free and bound drug and protein respectively.