Distribution of Drugs

After the entry of drug into the systemic circulation, the drug is distributed to various sites throughout the body. Hence, distribution is the reversible transfer of a drug between compartments. Since the distribution process is carried out by the blood, one of the compartments is always the blood or plasma and the other compartments represents extravascular fluids and other body tissues.

→ Distribution is a passive process:- diffusion

→ Driving force is the concentration gradient between the blood and the extravascular tissues.

steps in drug distribution

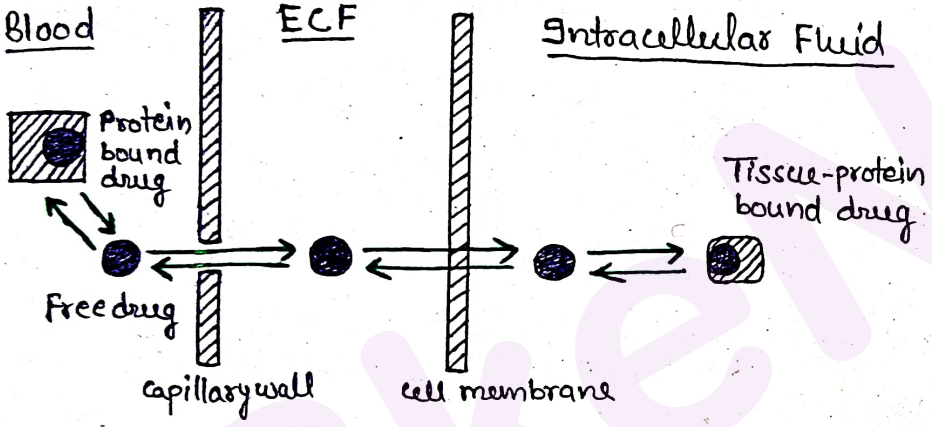
1) Permeation of free or unbound drug present in the blood through the capillary wall and entry into the interstitial / extracellular fluid (ECF)

2. Permeation of drug present in ECF through the membrane of tissue cells and into the

intracellular fluid. This step is rate-limiting and depends upon two major factors.

a) Rate of perfusion to the extracellular tissue.

b) Membrane permeability of drug.



Tissue Permeability of Drugs

Tissue permeability of a drug depends upon following two factors that restrict diffusion of drug into tissues;

1) Physicochemical properties

2) Physiological barriers

1) Physicochemical properties

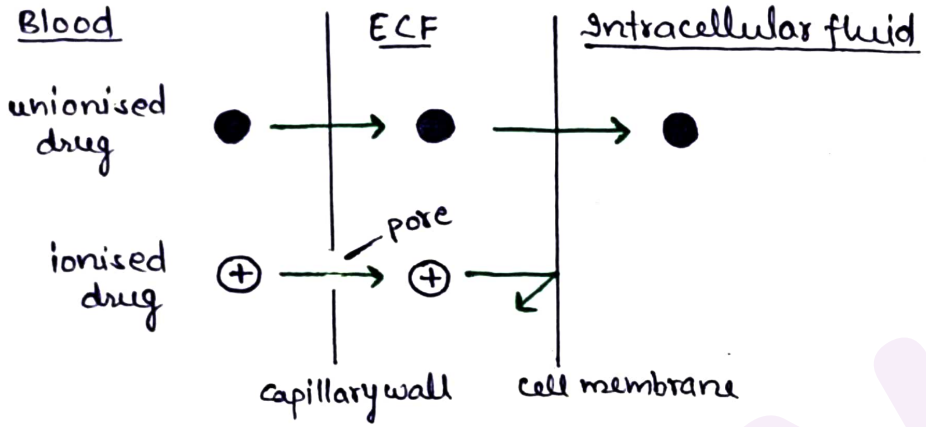
a) Molecular size

→ Molecular wt. less than 500 to 600 Dalton easily pass capillary membrane to extracellular fluid.

→ Penetration of drugs also depends on lipophilicity of drug. Only small, water-soluble molecules and ions of size below 50 Daltons enter the cell through aqueous filled channels (pore transport) whereas those of larger size are restricted.

b) Degree of ionisation

A drug that remains unionised at the pH of blood and extracellular fluid can permeate the cells relatively more rapidly. All drugs that ionise at plasma pH (i.e. polar, hydrophilic drugs), cannot penetrate the lipoidal cell membrane and tissue permeability is the rate-limiting step for such drugs.



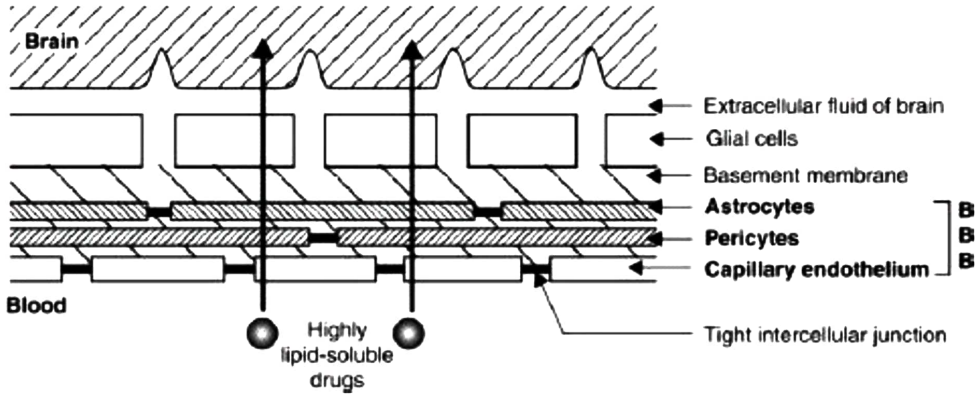
2) Physiological barriers

a) simple capillary endothelial barrier → All the drugs ionised or unionised with a molecular size less than 600 Daltons diffuse through the capillary endothelium. Only protein bound drugs are restricted because of large molecular size of the complex.

) Simple cell membrane barrier → once the drug diffuses from the capillary wall into the ECF, its further entry is limited by permeability of cell membrane.

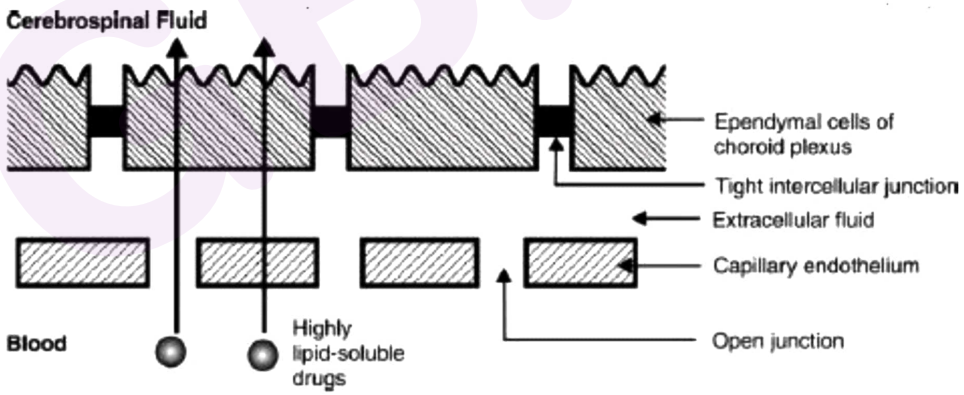
c) Blood-Brain Barrier (BBB) → The capillaries found in the brain are highly specialized and much less permeable to water-soluble drugs as compared to other capillaries in the body.

The brain capillaries consists of endothelial cells which are joined to one another by continuous tight intercellular junctions which is called as blood-brain barrier. The presence of special cells called as pericytes and astrocytes which are elements of the endothelial membrane, form a solid envelope around the brain capillaries. As a result the intercellular paracellular passage is blocked, so the drug has to pass through the cell rather than between them to reach the brain

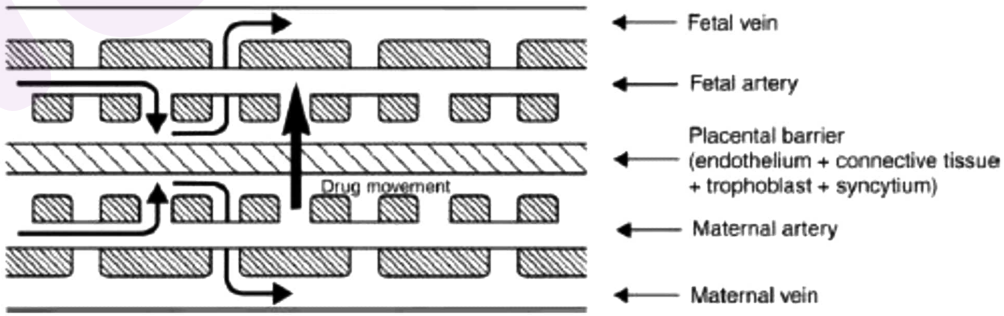


d) Blood-cerebrospinal fluid barrier → The cerebrospinal fluid (CSF) is formed mainly by the choroid plexus of the lateral, third and fourth ventricles and is similar in composition to the ECF of brain.

→ The capillary endothelium that lines the choroid plexus have open junctions or gaps and drugs can easily flow freely into the extracellular space between the capillary wall and the choroid cells. The choroid cells are joined to each other by tight junctions forming the blood-CSF barrier which has permeability characteristics similar to that of BBB.



e) Blood-Placental Barrier → The maternal and the foetal blood vessels are separated by a number of tissue layers made of foetal trophoblast basement membrane and the endothelium which together constitute the placental barrier. Blood-placental barrier is not as effective as BBB.



f) Blood-testes barrier → This barrier is not located at the capillary endothelium level but at sertoli-sertoli cell junction. This barrier restricts the passage of drugs to spermatocytes and spermatids.

Protein Binding of Drugs

A drug in the body can interact with several tissue components of which the two major categories are:

1) Blood, and
2) Extravascular tissues.

The interacting molecules are generally the macromolecules such as proteins, DNA or adipose. The proteins are particularly responsible for such an interaction.

Protein binding may be divided into:-

1) Intracellular binding → where the drug is bound to a cell protein which are generally drug receptors. This binding results into a pharmacological response.
2) Extracellular binding → where the drug binds to an extracellular protein but binding does not usually show a pharmacological response. The most important extracellular proteins are plasma proteins, particularly albumin.

Characteristics of a bound drug (protein-bound)

→ The protein bound drug is both pharmacokinetically and pharmacodynamically inert.
→ Also, it is pharmacologically not active.
→ A bound drug cannot undergo membrane transport because of its enormous size.

Binding of drugs to proteins is generally reversible which shows that it generally involves weak chemical bonds such as-

1) hydrogen bonds
2) hydrophobic bonds
3) Ionic bonds
4) van der Waal's forces

Binding of drugs falls into 2 classes:-

1) Binding of drugs to blood components like-
   a. plasma proteins
   b. blood cells
2) Binding of drugs to extravascular tissue proteins, fats, bones, etc.

Binding of drugs to blood components

Plasma protein - drug binding

After the entry of drug into systemic circulation, the main interaction of drug in the blood compartment is with the plasma proteins which are present in abundant amounts and in large varieties.

→ The binding is reversible

→ The order of binding of drugs to various plasma proteins is:

Albumin > α_1- acid glycoprotein > lipoproteins > Globulins

Albumin, as we can see is the protein to which most drugs bind. Albumin has a molecular weight of 65000 Daltons. Albumin is 59% of the total plasma protein.

A large variety of drugs ranging from weak acids, neutral compounds to weak bases bind to albumin.

Four different sites have been identified on albumin (or Human Serum Albumin, HSA) for drug-binding. They are:

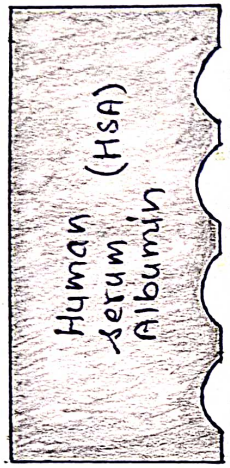
site-I :- Also called as warfarin and azapropazone binding site, it is the region to which large number of drugs bind.
e.g. NSAIDs, sulphonamides, phenytoin.

site-II :- Also called diazepam binding site.
Drugs bind to this region include benzodiazepines, medium chain fatty acids, ibuprofen, ketoprofen, tryptophan, cloxacillin, probenicid, etc.

* site-I and II are responsible for the binding of most drugs.

site-III :- Digitoxin-binding site

site-IV :- tamoxifen binding site



→ site-I (warfarin binding site)

→ site-II (diazepam binding site)

→ site-III (digitoxin binding site)

→ site-IV (tamoxifen binding site)

Binding of drugs to blood cells

Blood cells constitute >40% of blood. RBCs is the main major component constituting 95% of blood cells. RBC comprises of 3 components each of which can bind to drugs:

1) Haemoglobin → It has a molecular weight of 64500 (almost equal to HSA) but is almost 7 to 8 times the concentration of albumin in blood.

Drugs like phenytoin, pentobarbital and phenothiazines bind to haemoglobin.

2) Carbonic anhydrase → drugs that bind to it are acetazolamide and chlorthalidone.

3) cell membrane → Imipramine and chlorpromazine bind to RBC membrane.

Tissue binding of drugs

A drug can bind to one or more tissue components.

significance of tissue-drug binding:

1) Increases the apparent volume of distribution.

2) It results in localization of a drug at a specific site in the body.

Factors influencing localization of drugs in tissues include lipophilicity and structural features of the drug, perfusion rate, pH differences, etc.

For majority of drugs that bind to extravascular tissues, the order of binding is:

Liver > Kidney > Lung > Muscles

Several examples of tissues that bind to drugs:

1) Liver (paracetamol)
2) Lungs (imipramine)
3) Kidneys (lead, mercury, cadmium)
4) Skin (chloroquine, phenothiazines)
5) Eyes (chloroquine, phenothiazines)
6) Hairs (Arsenicals, chloroquine, phenothiazines)
7) Bones (Tetracycline)
8) Fats (lipophilic drugs like thiopental & DDT)
9) Nucleic acids (DNA interact with chloroquine and quinacrine).

Factors affecting Protein-drug binding

1. Drug-related Factors

a) Physicochemical characteristics of the drug:- An increase in lipophilicity increases the extent of binding.

b) Concentration of drug in the body:- Alteration in the concentration of drug and protein molecules brings alteration in the protein binding process.

c) Affinity of a drug for a particular binding component.

2. Protein/Tissue related factors

a) physicochemical characteristics of protein:- Lipoproteins and adipose tissue tend to bind liphophilic drugs by dissolving them in their lipid core.

b) concentration of protein:- Binding occurs predominantly with albumin, as it is present in high concentration in comparison to other plasma proteins.

3. Drug interactions

competition between drugs for binding sites

$$D_1 + P \xrightarrow{D_2} D_2 + P$$

D_1 = displaced drug; D_2 = displacer drug.

They are also called as displacement interactions.

4. Patient-related factors

a) Age:- neonates: low albumin content, more free drug.
young infants: high dose of digoxin due to large renal clearance.
elderly: low albumin, so more free drug

b) disease states

- renal failure (uremia): ↓ albumin content
- hepatic failure: ↓ albumine synthesis

Apparent volume of drug distribution

A drug in circulation distributes to various organs and tissues. when process of distribution is complete, different organs and tissues have different concentrations of drug which can be determined by the volume of tissues in which the drug is present. Since, different tissues have different concentrations of drug, the volume of distribution cannot be calculated by just knowing the volume of tissues.

However, there exists a constant relationship between the concentration of drug in plasma (C) and the amount of drug in the body (X).

$$X \propto C$$

$\boxed{X = V_d C}$ ⇒ total drug amount in body

where V_d is proportionality constant 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.

$$V_d = \frac{X}{C}$$

we can write,

amount of drug in plasma = $V_p C$

and amount of drug in extravascular tissues

$= V_t C_t$

Hence, total amount of drug in the body is the sum of amount of drug in plasma and the amount of drug in extravascular tissues.

Thus, $\boxed{V_d C = V_p C + V_t C_t}$

where, V_d = apparent volume
V_p = volume of plasma
V_t = volume of extravascular tissues
C_t = drug concentration in tissue
C = drug concentration in plasma

Kinetics of protein binding

If P represents proteins and D represents the drug, then applying law of mass action to reversible protein binding,

$$P + D \rightleftharpoons PD \quad \text{— (1)}$$

At equillibrium,

$$K_a = \frac{[PD]}{[P]\,[D]} \quad \text{— (2)}$$

$$[PD] = K_a [P] [D] \quad \text{— (3)}$$

where, $[P]$ = concentration of free protein
$[D]$ = concentration of free drug
$[PD]$ = concentration of protein-drug complex
K_a = association rate constant

If P_t is the total concentration of protein present (bound and unbound), then:

$$P_t = [PD] + [P] \quad \text{— (4)}$$

If r is the number of moles of drug bound to total moles of protein, then;

$$r = \frac{[PD]}{[P_t]} = \frac{[PD]}{[PD]+[P]} \quad \text{— (5)}$$

substituting the value of $[PD]$ from equation-3 in equation 5, we get

$$r = \frac{K_a [P][D]}{K_a [P][D] + [P]} = \frac{K_a [D]}{K_a [D] + 1} \quad \text{— (6)}$$

Equation-(6) is true only when there is only one binding site on the protein.
If more than one or N number of binding sites are available per mole of the protein, then:

$$r = \frac{N K_a [D]}{K_a [D] + 1} \quad \text{— (7)}$$

The value of K_a and number of binding sites, N can be obtained by plotting equation-⑦ in four different ways:

① Direct plot → is made by plotting r versus [D]. when all the binding sites are occupied, plateau is reached.

plateau → saturation of binding sites at high drug concentration

At the plateau,

$$r = N$$

when $r = N/2$, $[D] = 1/K_a$

② Scatchard plot → is made by transforming equation-⑦ into a linear form.

$$r = \frac{NK_a[D]}{K_a[D]+1}$$

$$r + rK_a[D] = NK_a[D]$$

$$r = NK_a[D] - rK_a[D]$$

$$\frac{r}{[D]} = NK_a - rK_a \quad \text{—— ⑧}$$

A plot of $r/[D]$ versus r yields a straight line. slope of the line $= -K_a$, y-intercept $= NK_a$ and x-intercept $= N$.

3. Klotz Plot/ Double Reciprocal Plot

The reciprocal of equation-⑦ yields:

$$\frac{1}{r} = \frac{K_a[D]+1}{NK_a[D]} = \frac{1}{N} + \frac{1}{NK_a[D]} \quad \text{—— ⑨}$$

A plot of $1/r$ versus $1/[D]$ yields a straight line with slope $\frac{1}{NK_a}$ and y-intercept $1/N$.

4. Hitchcock Plot → is made by rewriting equation-⑨-

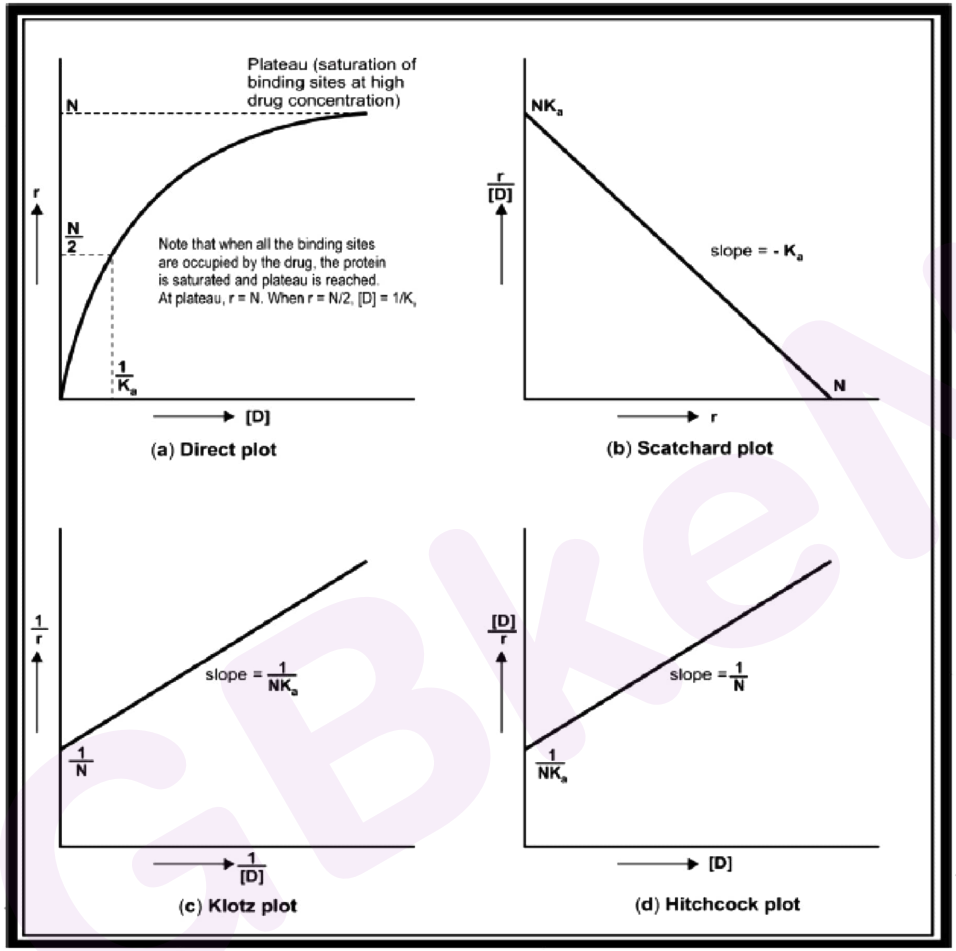
$$\frac{1}{r} = \frac{K_a[D]+1}{NK_a[D]}$$

$$\Rightarrow \frac{NK_a[D]}{r} = 1 + K_a[D] \quad \text{—— ⑩}$$

Dividing both sides by NK_a gives-

$$\frac{[D]}{r} = \frac{1}{NK_a} + \frac{[D]}{N} \quad \text{—— ⑪}$$

A plot of $[D]/r$ versus $[D]$ yields a straight line.



Significance of protein binding of drugs

1) Absorption → The concentration of free drug at the site of administration and systemic circulation is equal (i.e. in equillibrium). However, binding of absorbed drug to plasma proteins decreases the free drug concentration and disturbs such an equillibrium.

2) Distribution → Plasma protein binding restricts the entry of drugs in certain tissues. This prevents accumulation of large fraction of drug in such tissues and hence prevent toxic reactions

3) Metabolism → Protein binding decreases the metabolism of drugs and enhances the biological half-life. Only unbound fraction of drug gets metabolized.

4) Excretion → Only the unbound drug is capable of being excreted. Protein binding prevent the entry of drug to glomerular filtration.