Pharmacokinetics of peptides and proteins

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Introduction:

- Pharmacokinetics describes the time course of the concentration of a drug in body fluid that results from the administration of a certain dosage regimen.
- It comprises all processes affecting drug absorption, distribution, metabolism and excretion.
- It has the property of **what the body does to the drug.**

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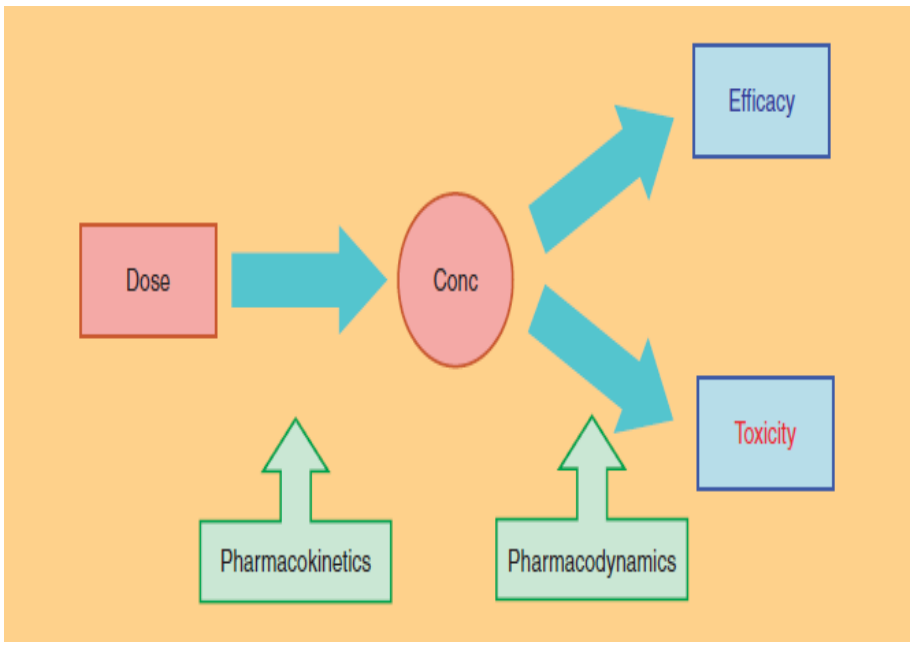

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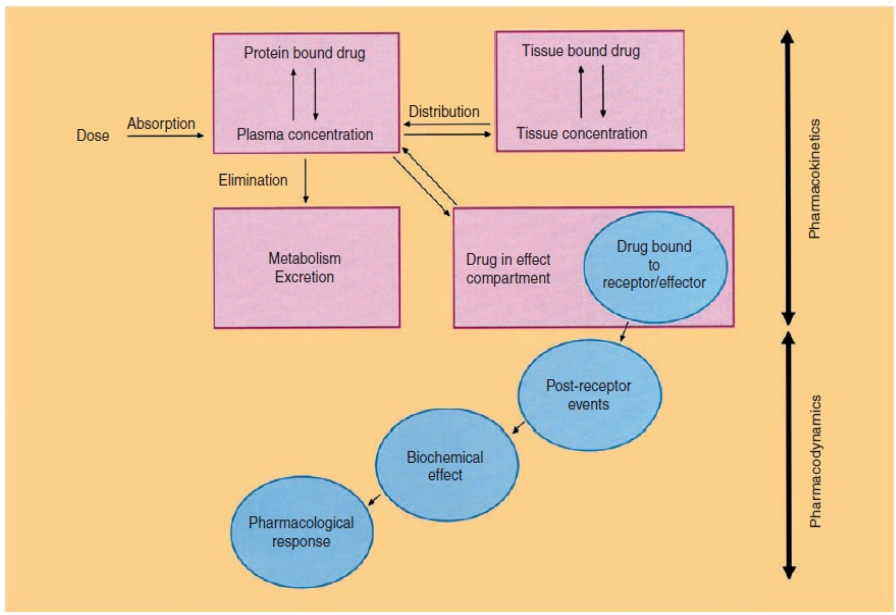

Figure 2 Physiological scheme of pharmacokinetic and pharmacodynamic processes.

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- Pharmacokinetics for proteins may be different from that for conventional drugs.(Why?)

This related to:

1) The structural similarity to some endogenous compounds.
2) With regulatory feedback mechanisms.
3) Difficulties in analysis (interferences).
4) Their large molecular weights.

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Absorption of protein therapeutics

- Poorly absorbed orally. (as taken before).
- Mainly administered parenterally (I.V, I.M, and S.C.) depending on type of protein.
- One of the potential limitations of SC and IM administrations, however, are the pre-systemic degradation processes, local blood flow, injection trauma and the capillaries sizes.

then **Ka = F . K app**

Ka= the true abs. rate constant

F= The bioavailability compared to IV adm.

Kapp= apparent abs. rate constant for IM,SC adm.

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Distribution of protein therapeutics

- The rate and extent of protein distribution are largely determined by their size and Mol.wt, physicochemical properties (like charge, lipophilicity), protein binding, and their dependency on active transport.
- The lymphatic system play important role in distribution of proteins depending on size.
- Protein charge is important for electrostatic attraction of +vely charged proteins with –vely charged cell membranes (containing glycoaminoglycans).
- I.V. administered proteins may follow one or two-compartmental model or may be non-compartmental (with rapid elimination rates) in distribution.

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- Then for distribution characterization :

1) Biopsy or necropsy for determination of actual proteins concentrations in the tissue.

2) Bio-distribution studies with radiolabeled compound and/or imaging techniques.

- The binding to endogenous protein structures (specific) can affect the distribution, pharmacodynamics (PD) and disposition properties of proteins.
- The binding may be non specific to plasma proteins (albumin and lipoproteins).
- Site-specific receptor mediated uptake can also substantially influence and contribute to the distribution, elimination and PD of proteins.

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Elimination of protein therapeutics

- The exogenous proteins are subjected to the same catabolic pathways as endogenous ones.
- The end products of protein metabolism are thus amino acids that are reutilized in the endogenous amino acids pool for synthesis of endogenous proteins.
- The elimination pathways includes:

1) **Proteolysis**
2) **GIT protein metabolism**
3) **Renal protein metabolism and excretion**
4) **Hepatic protein metabolism**
5) **Receptor-mediated protein metabolism.**

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Molecular weight	Elimination site	Predominant elimination mechanisms	Major determinant
<500	Blood, liver	Extracellular hydrolysis Passive lipoid diffusion	Structure, lipophilicity
500–1,000	Liver	Carrier-mediated uptake Passive lipoid diffusion	Structure, lipophilicity
1,000–50,000	Kidney	Glomerular filtration and subsequent degradation processes (see Fig. 4)	Molecular weight
50,000–200,000	Kidney, liver	Receptor-mediated endocytosis	Sugar, charge
200,000–400,000		Opsonization	α_2-macroglobulin, IgG
>400,000		Phagocytosis	Particle aggregation

Note: Other determining factors are size, charge, lipophilicity, functional groups, sugar recognition, vulnerability for proteases, aggregation to particles, formation of complexes with opsonization factors, etc. Mechanisms may overlap and endocytosis may occur at any molecular weight range.
Source: After Meijer and Ziegler, 1993.

Table 1 Molecular weight as major determinant of the elimination mechanisms of peptides and proteins.

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Proteolysis:

- The metabolic rate for protein degradation generally increases with decreasing mol.wt. from large to small proteins to peptides, but is also dependent on other factors such as size, charge, lipophilicity, functional groups and molecular properties (glycosylation, 2° or 3° structures).
- The clearance may include metabolism and cellular uptake.
- Proteolytic degradation can occur unspecifically nearly everywhere in the body (within blood) or can be limited to a specific organ or tissue.
- As enzymes, proteases and peptidases are found extra and intra-cellularly.

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GIT protein metabolism:

- As before, mainly for orally administered proteins and some parenterally administered proteins in the intestinal mucosa following intestinal secretion (as for endogenous albumin).

Renal protein metabolism and excretion:

- The kidneys are a major site of protein metabolism for smaller sized proteins that undergo glomerular filtration (GF).
- The size- selective cut-off for GF is approx. 60kD, or is most efficient for proteins smaller than 30kD and the rate falls off sharply for larger than 30kD.

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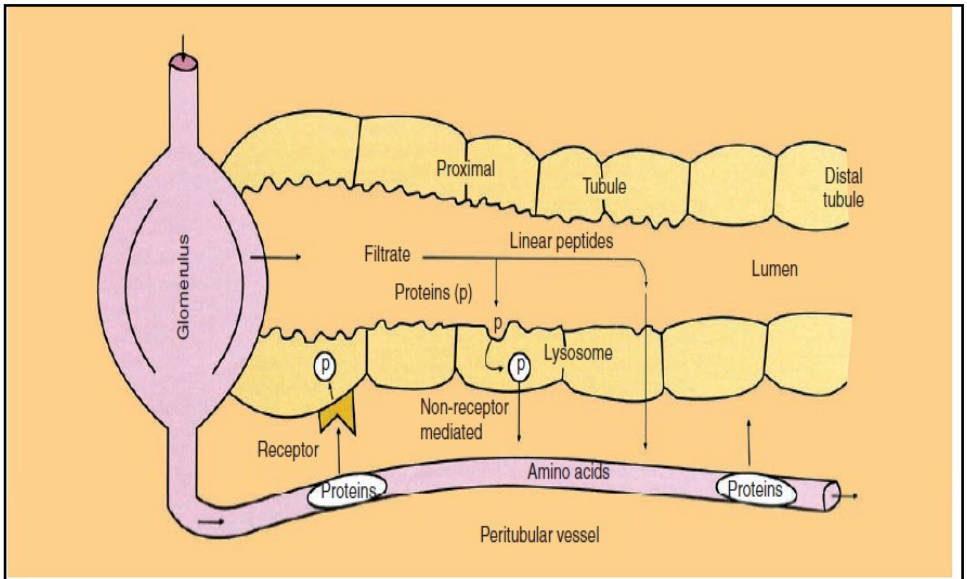

Figure 4 Pathways of renal metabolism of peptides and proteins: Glomerular filtration followed by either (a) intraluminal metabolism or (b) tubular reabsorption with intracellular lysosomal metabolism, and (c) peritubular extraction with intracellular lysosomal metabolism. *Source*: Modified from Maack et al., 1985.

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- Charge selectivity is important here, where anionic compounds (Ex. TNF- alpha) pass less readily than neutral ones which in turn pass through less readily than cationic ones, due to the –ve charge of GF (presence of abundance of glycosaminoglycan).
- Renal metabolism of peptides and small proteins is mediated through three highly effective processes (as in the figure), so only trace amounts of intact protein are detectable in urine.
- **The first mechanism,** involves GF of larger, complex peptides and proteins followed by reabsorption into endocytic vesicles in the proximal tubule and subsequent hydrolysis into small fragments and a.a.s. as for IL-2, GH and insulin.

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- **The second mechanism,** GF followed by intraluminal metabolism, predominantly by exo-peptidases in the luminal brush border membrane of the proximal tubule, the resulting peptide fragments and a.a.s are reabsorbed into the systemic circulation. As for glucagon and LH-RH (small linear peptides).
- **The third mechanism,** peri-tubular extraction of peptides and proteins from post-glomerular capillaries with subsequent intracellular metabolism. As for insulin and IL-2.

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Hepatic protein metabolism:

- The rate of hepatic metabolism is largely dependent on the specific a.a. sequence of the protein (endopeptidases or exopeptidases action).
- Mechanisms of hepatic uptake for proteins depend on the size and hydrophobicity.
- There are different hepatic cells for this uptake mechanisms like:

1) **Hepatocytes**
2) **Kupffer cells (specialized macrophages)**
3) **Endothelial cells**
4) **Fat-storing cells**

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The uptake mechanisms are:

1) Simple passive diffusion for small peptides with sufficient hydrophobicity (in hepatocytes). As for cyclosporine (cyclic peptides) which metabolized by microsomal enzymes in cytosol.

2) A carrier-mediated transport for cyclic and linear peptides of small size and hydrophobic nature (containing aromatic a.a.) in hepatocytes like cholecystokinin-8 (CCK-8) which metabolized by cytosolic peptidases and then excreted into bile via active export transporters.

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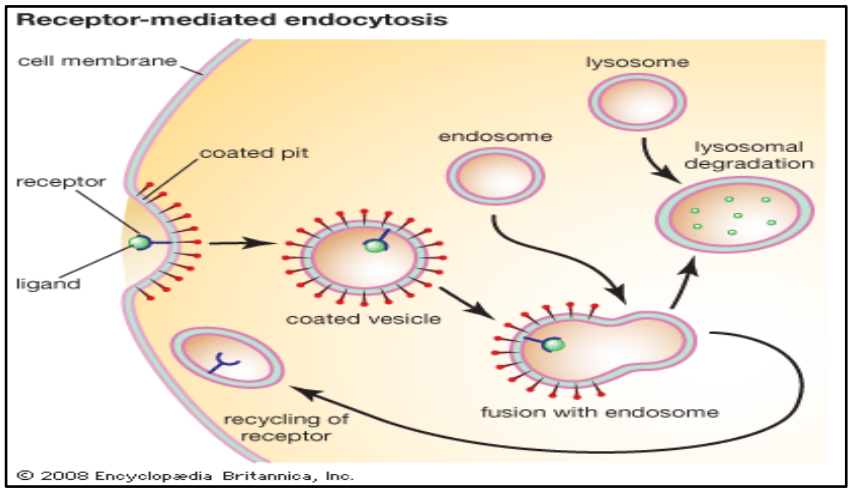
3) Receptor-mediated endocytosis (RME), in which the circulating proteins are recognized by specific hepatic receptor proteins (glycoproteins), as for insulin and epidermal growth factor (large peptides).



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- Here, recycling of receptors occurred, so depending on type of protein receptor, sometimes degradation may be occurred leading to a decrease in the concentration of receptors on the cell surfaces (receptor down-regulation) as for interferon and insulin.
- For glycoproteins, if a critical number of exposed sugar groups (mannose, galactose, fucose……etc) is exceeded, RME being an efficient hepatic uptake mechanism in hepatocytes (asialoglycoprotein Rc.), Kupffer and liver endothelial cells (mannose Rc.).
- Low density lipoprotein receptor-related protein (LRP) is a member of the LDL receptor family responsible for endocytosis of several lipoproteins, proteases and protease inhibitor complexes in the liver.

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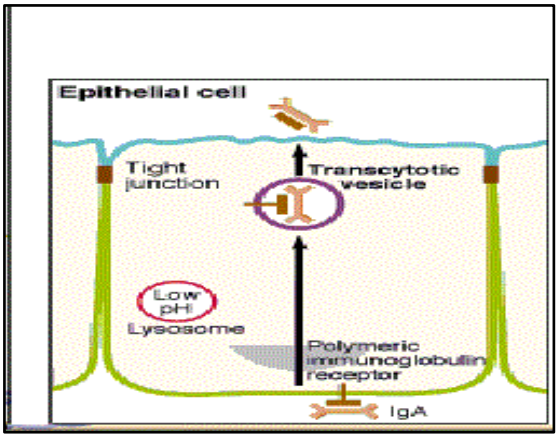
4) The transcytotic pathway, in which the endocytotic vesicle formed at the cell surface traverses the cell, degradation, and exocytosis into bile, as for polymeric immunoglobulin A.



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Receptor-mediated protein metabolism

- Occurred for proteins that bind with high affinity to membrane-associated receptors on the cell surface.
- Includes endocytosis and subsequent intracellular lysosomal metabolism.
- It is not constant (dose-dependent), decreases with increasing the dose.
- It is not limited for a specific organ or tissue type, but depend on number of protein drug receptors.
- Example metabolism of some proteins by linking to a receptor- mediated uptake into macrophages.

The end

good luck

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