Drug Metabolism

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Introduction

- Biotransformation: Chemical alteration of the drug in body that converts nonpolar or lipid soluble compounds to polar or lipid insoluble compounds
- Consequences of biotransformation
  - Active drug → Inactive metabolite : Pentobarbitone, Morphine, Chloramphenicol
  - Active drug → Active metabolite: Phenacetin
  - Inactive drug → active metabolite: Levodopa

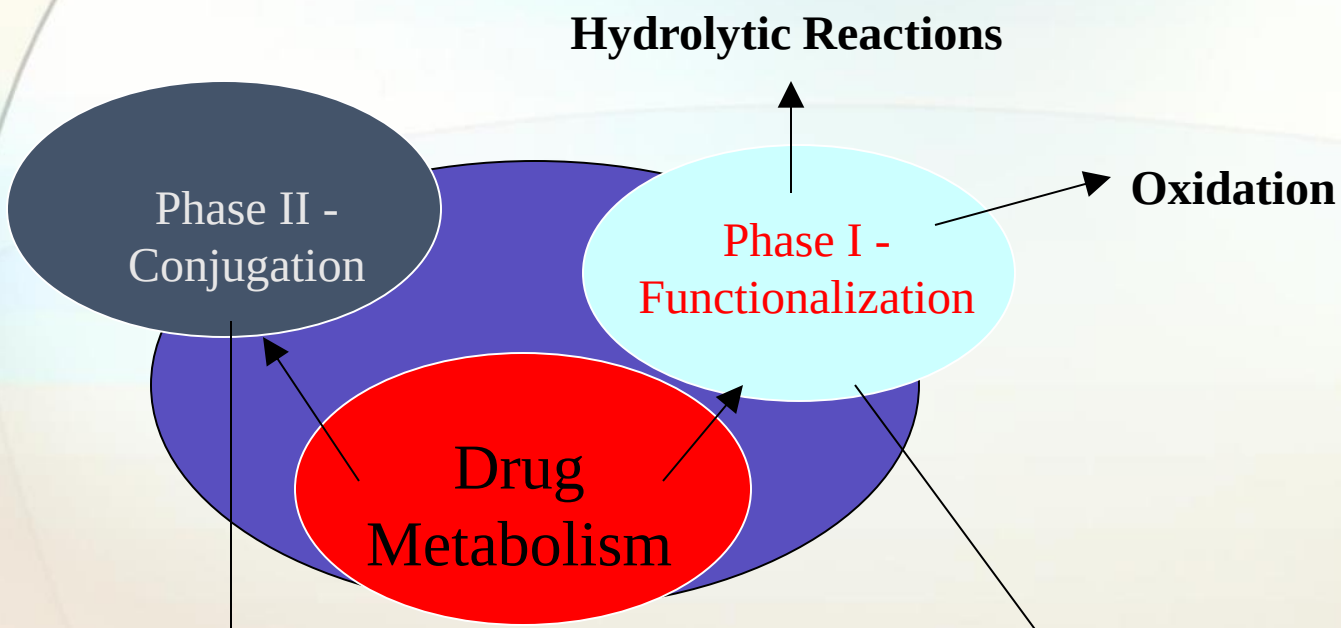
Prodrugs

- Inactive drug is converted to active metabolite
- Coined by Albert in 1958
- Advantages:
  - Increased absorption
  - Elimination of an unpleasant taste
  - Decreased toxicity
  - Decreased metabolic inactivation
  - Increased chemical stability
  - Prolonged or shortened action

Phases of Metabolism

- **Phase I**
  - Functionalization reactions
  - Converts the parent drug to a more polar metabolite by introducing or unmasking a functional group (-OH, -NH2, -SH).
- **Phase II**
  - Conjugation reactions
  - Subsequent reaction in which a covalent linkage is formed between a functional group on the parent compound or Phase I metabolite and an endogenous substrate such as glucuronic acid, sulfate, acetate, or an amino acid

Phases of Metabolism



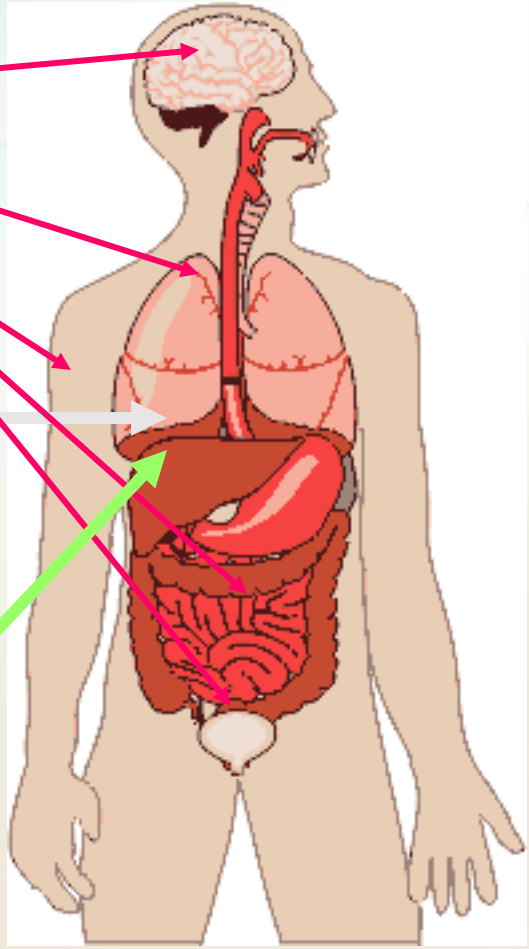
- **Conjugation**
  - Glucuronic acid
  - Sulfate, Glycine and other AA
  - Glutathione or mercapturic acid
  - Acetylation, Methylation

Sites of Drug Metabolism

Extrahepatic microsomal enzymes (oxidation, conjugation)

Hepatic microsomal enzymes (oxidation, conjugation)

Hepatic non-microsomal enzymes (acetylation, sulfation,GSH, alcohol/aldehyde dehydrogenase, hydrolysis, ox/red)



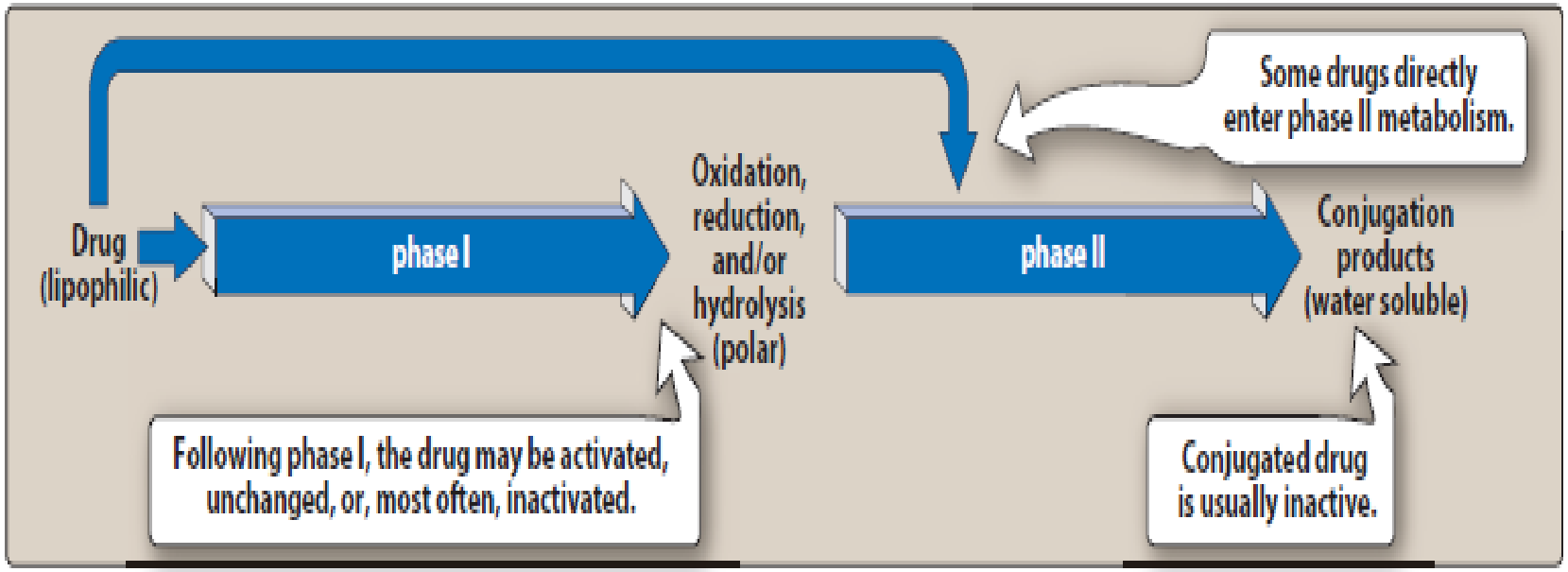


Figure 1.16

The biotransformation of drugs.

Phase I / Non Synthetic Reactions

Oxidation

- Addition of oxygen/ negatively charged radical or removal of hydrogen/ positively charged radical.
- Reactions are carried out by group of mono-oxygenases in the liver.
- Final step: Involves cytochrome P-450 haemoprotein, NADPH, cytochrome P-450 reductase and O2

Cytochrome P450 enzymes

- Monooxygenase enzyme family
- Major catalyst: Drug and endogenous compound oxidations in liver, kidney, G.I. tract, skin and lungs
- Oxidative reactions require: CYP heme protein, the reductase, NADPH, phosphatidylcholine and molecular oxygen
- Location: smooth endoplasmic reticulum in close association with NADPH-CYP reductase in 10/1 ratio
- The reductase serves as the electron source for the oxidative reaction cycle

Cytochrome P family

- Multiple CYP gene families have been identified in humans, and the categorized based on protein sequence homology
- Most of the drug metabolizing enzymes are in CYP 1, 2, & 3 families .
- Frequently, two or more enzymes can catalyze the same type of oxidation, indicating redundant and broad substrate specificity.
- CYP3A4 is very common to the metabolism of many drugs; its presence in the GI tract is responsible for poor oral bioavailability of many drugs

Cytochromes: Metabolism of Drugs

CYP Enzyme	Examples of substrates
1A1	Caffeine, Testosterone, **R-Warfarin**
1A2	Acetaminophen, Caffeine, Phenacetin, **R-Warfarin**
2A6	17β-Estradiol, Testosterone
2B6	Cyclophosphamide, Erythromycin, Testosterone
2C-family	Acetaminophen, Tolbutamide (2C9); Hexobarbital, S-Warfarin (2C9,19); Phenytoin, Testosterone, **R- Warfarin**, Zidovudine (2C8,9,19);
2E1	Acetaminophen, Caffeine, Chlorzoxazone, Halothane
2D6	Acetaminophen, Codeine, Debrisoquine
3A4	Acetaminophen, Caffeine, Carbamazepine, Codeine, Cortisol, Erythromycin, Cyclophosphamide, S- and **R-Warfarin**, Phenytoin, Testosterone, Halothane, Zidovudine

Adapted from: *S. Rendic Drug Metab Rev 34: 83-448, 2002*

Non-CYP Drug Oxidations

- Monoamine Oxidase (MAO), Diamine Oxidase (DAO)
  - MAO (mitochondrial) oxidatively deaminates endogenous substrates including neurotransmitters
  - Dopamine, serotonin, norepinephrine, epinephrine
- Alcohol & Aldehyde Dehydrogenase
  - Non-specific enzymes found in soluble fraction of liver
  - Ethanol metabolism
- Flavin Monooxygenases
  - Require molecular oxygen, NADPH, flavin adenosine dinucleotide (FAD)

Reduction

- Converse of oxidation
- Drugs primarily reduced are chloralhydrate, chloramphenicol, halothane.

Hydrolysis

- Cleavage of drug molecule by taking up a molecule of water.
- Sites: Liver, intestines, plasma and other tissues
- Examples: Choline esters, Procaine, Isoniazid, pethidine, oxytocin.

Cyclization and Decyclization

- **Cyclization**
  - Formation of ring structure from a straight chain compound
  - E.g. Proguanil
- **Decyclization**
  - Opening up of ring structure of the cyclic drug molecule
  - E.g. Barbiturates, Phenytoin.

Phase II reactions

- Conjugation of the drug or its phase I metabolite with an endogenous substrate to form a polar highly ionized organic acid

- Types of phase II reactions
  - Glucuronide conjugation
  - Acetylation, Methylation
  - Sulfate conjugation, Glycine conjugation
  - Glutathione conjugation
  - Ribonucleoside/ nucleotide synthesis

Glucuronide Conjugation

- Conjugation to α-d-glucuronic acid
- Quantitatively the most important phase II pathway for drugs and endogenous compounds
- Products are often excreted in the bile
- Requires enzyme UDP-glucuronosyltransferase (UGT)
- Compounds with a hydroxyl or carboxylic acid group are easily conjugated with glucuronic acid which is derived from glucose

Glucuronide Conjugation Continued..

- Enterohepatic recycling may occur due to gut glucuronidases
- Drug glucuronides excreted in bile can be hydrolysed by bacteria in gut and reabsorbed and undergoes same fate.
- This recycling of the drug prolongs its action e.g.Phenolphtalein, Oral contraceptives
  - Examples: Chloramphenicol, aspirin, phenacetin, morphine, metronidazole

Acetylation

- Common reaction for aromatic amines and sulfonamides
- Requires co-factor acetyl-CoA
- Responsible enzyme is N-acetyltransferase
- E.g. Sulfonamides, isoniazid, Hydralazine.

Sulfate Conjugation

- Major pathway for phenols but also occurs for alcohols, amines and thiols
- Sulfate conjugates can be hydrolyzed back to the parent compound by various sulfatases
- Examples include: a-methyldopa, albuterol, terbutaline, acetaminophen, phenacetin

Amino Acid Conjugation:

- ATP-dependent acid: CoA ligase forms active CoA-amino acid conjugates which then react with drugs by N-Acetylation:
  - Usual amino acids involved are:
    - Glycine. Glutamine, Ornithine, Arginine

Glutathione Conjugation:

- Glutathione is a protective factor for removal of potentially toxic compounds

- Conjugated compounds can subsequently be attacked by g-glutamyltranspeptidase and a peptidase to yield the cysteine conjugate => product can be further acetylated to N-acetylcysteine conjugate

  E.g. Paracetamol

Hofmann elimination

Inactivation of the drug in the body fluids by spontaneous molecular re arrangement without the agency of any enzyme

e.g. Atracurium.

First pass Metabolism

- Metabolism of a drug during its passage from the site of absorption into the systemic circulation.
- Extent of first pass metabolism differs in different drugs

Extent of first pass metabolism of important drugs

Low	Intermediate	High – not given orally	High oral dose
Phenobarbitone	Aspirin	Isoprenaline	propranolol
Phenylbutazone	Quinidine	Lidocaine	Alprenolol
Tolbutamide	Desipramine	Hydrocortisone	Verapamil
Pindolol	Nortriptyline	Testosterone	Salbutamol

Attributes of drugs with high first pass metabolism

- Oral dose is considerably higher then sublingual or parenteral dose
- Marked individual variation in the oral dose due to differences in the extent of first pass metabolism
- Oral bioavailability is apparently increased in patients with severe liver disease
- Oral bioavailability of a drug is increased if another drug competing with it.

E.G. Chloropromazine and Propranolol

Inhibition of Microsomal enzymes

- Competitively inhibit the metabolism of another drug if it utilizes the same enzyme or co factors.
- A drug may inhibit one isoenzyme while being itself a substrate of another isoenzyme

e.g. quinidine is metabolized by CYP3A4 but inhibits CYP2D6

- Inhibition of drug metabolism occurs in a dose related manner and can precipitate toxicity of the object drug.
- Blood flow limited metabolism

e.g. Propranolol reduces rate of lidocaine metabolism by decreasing hepatic blood flow.

Microsomal Enzyme Induction

- Certain drugs, insecticides and carcinogens increase the synthesis of microsomal enzyme protein.
- Different inducers are relatively selective for certain cytochrome P-450 enzyme families e.g.
  - Phenobarbitone , rifampin, glucorticoids induce CYP3A isoenzymes
  - Isoniazid and chronic alochol consumption induce CYP2E1
- Induction takes 4-14 days to reach its peak and is maintained till the inducing agent is present.

Consequences of Induction

- Decreased intensity or Increased Intensity of action of drug
- Tolerance- autoinduction
- Interfere with adjustment of dose of another drug
- Interference with chronic toxicity

- Some foods, other drugs, herbs, environmental agents, inhibit/induce CYP's → change in metabolism drugs → change drug activity
  - Grapefruit juice, St. John's wort inhibit drug metab by inhib'n CYP enz's
  - Brusssels sprouts, cigarettes induce P450 enz's

Role of Metabolism in pediatric and elderly

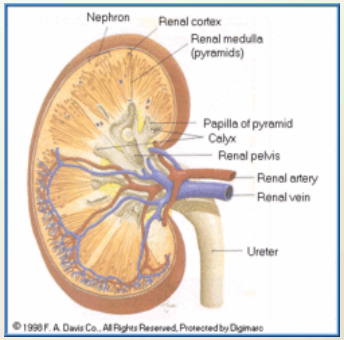
- New born has low GFR and tubular transport is immature, so the t1/2 of the drug like streptomycin and penicillin is prolonged
- Hepatic drug metabolising system is inadequate in new borns e.g. chloramphenicol can produce gray baby syndrome
- In elderly the renal function progressively declines
- Reduction of hepatic microsomal activity and liver blood flow
- Incidence of adverse drug reactions is much higher in elderly

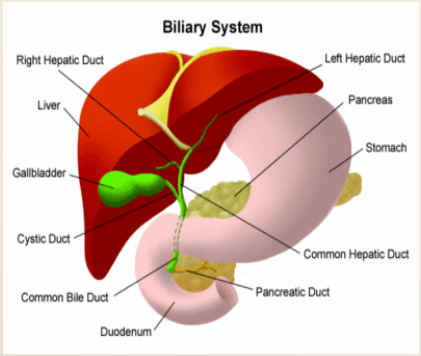
EXCRETION or ELIMINATION OF DRUGS

- Excretion is a process whereby drugs are transferred from the internal to the external environment
- Despite the reduction in activity that occurs as a drug leaves its site of action, it may remain in the body for a considerable period, especially if it is strongly bound to tissue components.

- Excretion, along with metabolism and tissue redistribution, is important in determining both the duration of drug action and the rate of drug elimination.

- Principal organs involved
  - Kidneys,
  - Lungs,
  - Biliary system
  - Intestines
  - Saliva
  - Milk.





RENAL EXCRETION

- Kidney is the primary organ of removal for most drugs especially for those that are water soluble and not volatile.
- The three principal processes that determine the urinary excretion of a drug

1. glomerular filtration,
2. tubular secretion, and
3. tubular reabsorption (mostly passive back-diffusion)

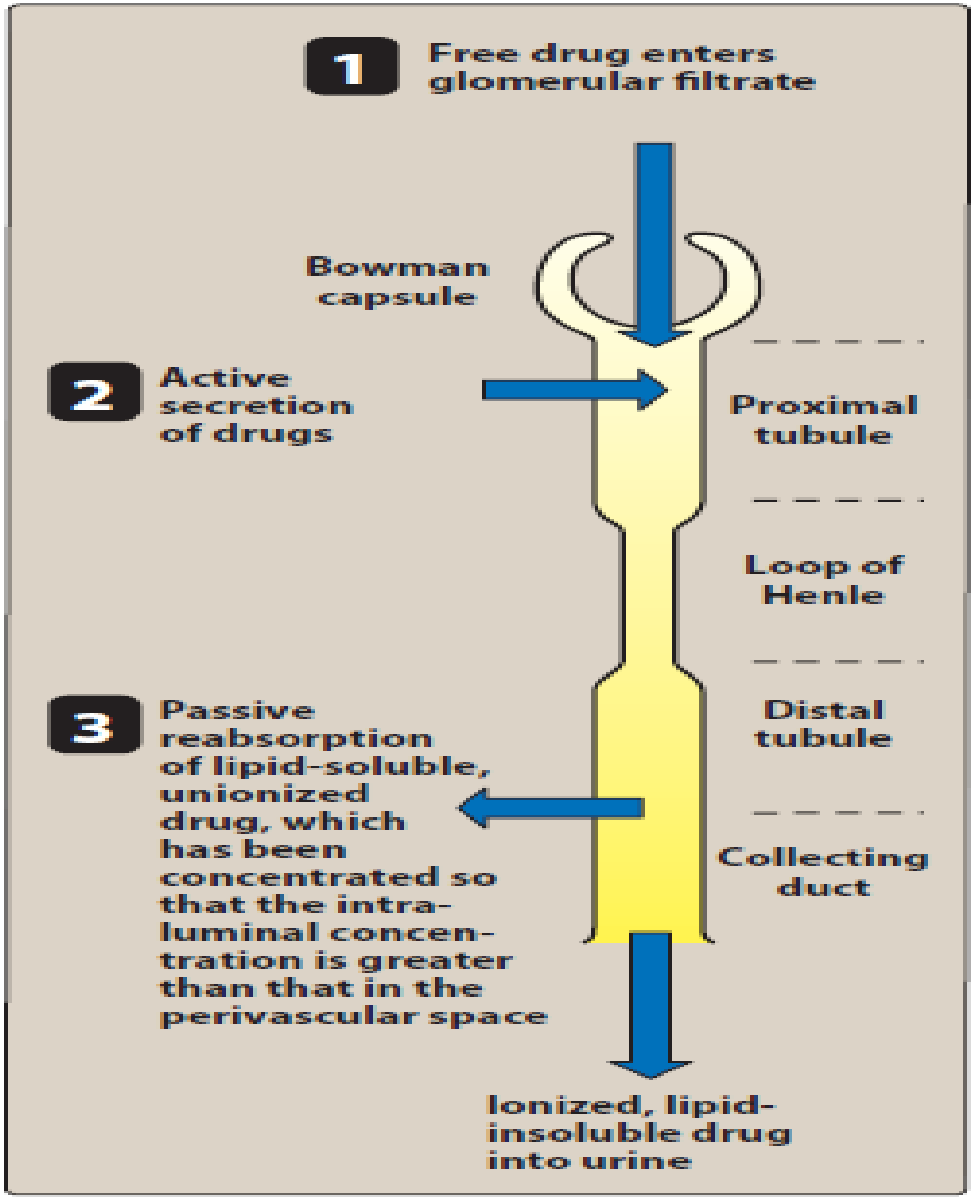


Figure 1.19
Drug elimination by the kidney.

Passive Diffusion

- Urinary excretion of drugs (i.e., weak electrolytes) is the extent to which substances diffuse back across the tubular membranes and reenter the circulation.

- The concentration gradient thus established will facilitate movement of the drug out of the tubular lumen, given that the lipid solubility and ionization of the drug are appropriate.
- The pH of the urine (usually between 4.5 and 8) can markedly affect the rate of passive back-diffusion.
- The back-diffusion occurs primarily in the distal tubules and collecting ducts, (most of the urine acidification takes place)
- **Acidification increases reabsorption (or decreases elimination) of weak acids**, such as salicylates, and **decreases reabsorption (or promotes elimination) of weak bases, such as amphetamines.**

- Effects of pH on urinary drug elimination may have important applications, especially in cases of overdose.
- Eg: enhance the elimination of a barbiturate (a weak acid) by administiring bicarbonate to the patient.
- This procedure alkalinizes the urine and thus promotes the excretion of more completely ionized drug.
- **Excretion of bases can be increased by making the urine more acidic through the use of an acidifying salt, such as ammonium chloride.**

Active Tubular Secretion

- A number of drugs can serve as substrates for the two active secretory systems in the PCT
- Actively transfer drugs from blood to luminal fluid, are independent of each other; one secretes organic anions, and the other secretes organic cations.
- The secretory capacity of both the organic anion and organic cation secretory systems can be saturated at high drug concentrations.

- Each drug will have its own characteristic maximum rate of secretion (transport **maximum,Tm)**.

- Some drugs that are not candidates for active tubular secretion.
- Eg: Metabolites that are formed as a result of conjugative reactions.

- **Any drug known to be largely excreted by the kidney that has a body half-life of less than 2 hours is probably** eliminated, at least in part, by tubular secretion.

- Some drugs can be secreted and **have long half-lives, however, because of extensive passive reabsorption in distal segments** of the nephron.
- These tubular transport mechanisms are not as well developed in the neonate as in the adult.
- Functional capacity may be diminished in the elderly.
- Compounds that undergo active tubular secretion also are filtered at the glomerulus (assuming protein binding is minimal).

Organic Anion Transport	Organic Cation Transport
Acetazolamide	Acetylcholine
Bile salts	Atropine
Hydrochlorothiazide	Cimetidine
Furosemide	Dopamine
Indomethacin	Epinephrine
Penicillin G	Morphine
Prostaglandins	Neostigmine
Salicylate	Quinine

Active Tubular Reabsorption

- Some substances filtered at the glomerulus are reabsorbed by active transport systems found primarily in the proximal tubules.

- Active reabsorption is particularly important for endogenous substances, such as ions, glucose, and amino acids, although a small number of drugs also may be actively reabsorbed.

- The probable location of the active transport system is on the luminal side of the proximal cell membrane.

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

- Changes in any of these factors may result in clinically important alterations in drug action.

- In the final analysis, the amount of drug that finally appears in the urine will represent a balance of filtered, reabsorbed (passively and actively), and secreted drug.

- For many drugs, the duration and intensity of pharmacological effect will be influenced by the status of renal function, because of the major role played by the kidneys in drug and metabolite elimination.

Biliary Excretion

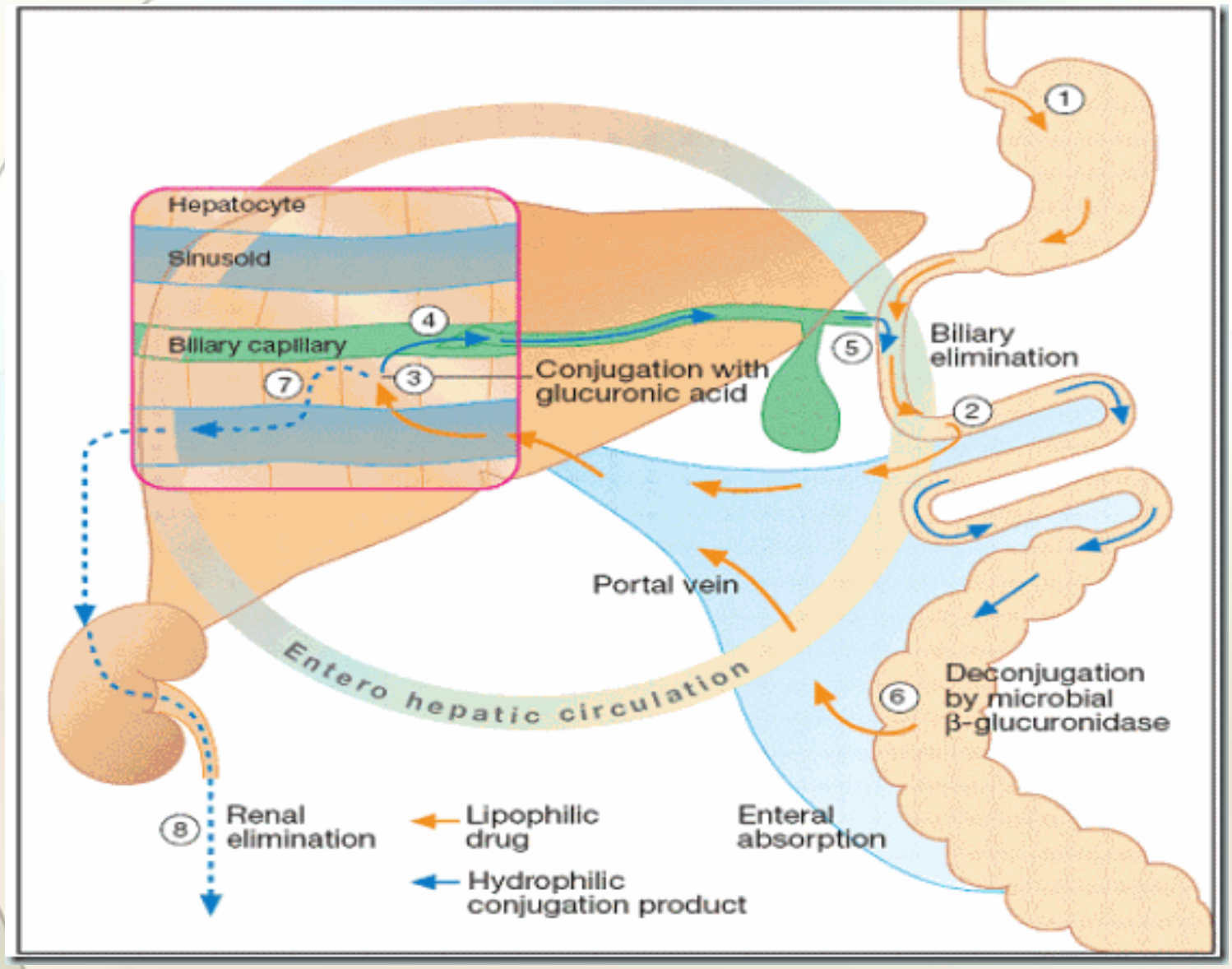
- Only small amounts of most drugs reach the bile by diffusion.
- However- Biliary excretion plays a major role (5–95% of the administered dose) in drug removal for some anions, cations, and certain un-ionized molecules, such as cardiac glycosides.
- Important for the excretion of some heavy metals.
- The ability of certain compounds to be actively secreted into bile accounts for the large quantity of these drugs removed from the body by way of the feces.

- On the other hand, most drugs that are secreted by the liver into the bile and then into the small intestine are not eliminated through the feces.

ENTEROHEPATIC CIRCULATION

- The physicochemical properties of most drugs are sufficiently favorable for passive intestinal absorption that the compound will reenter the blood that perfuses the intestine and again be carried to the liver.
- Such recycling may continue (*enterohepatic cycle or circulation) until the drug either undergoes* metabolic changes in the liver, is excreted by the kidneys, or both.

- This process permits the conservation of such important endogenous substances as the bile acids, vitamins D3 and B12, folic acid, and estrogens


Hepatocyte
Sinusoid
Biliary capillary
4
3
7
Conjugation with glucuronic acid
5
Biliary elimination
1
2
Portal vein
Entero hepatic circulation
6
Deconjugation by microbial β-glucuronidase
8
Renal elimination
Lipophilic drug
Hydrophilic conjugation product
Enteral absorption

Drugs that Undergo Enterohepatic Circulation	
Adriamycin	Methadone
Amphetamine	Metronidazole
Chlordecone	Morphine
1,25-Dihydroxyvitamin D3	Phenytoin
Estradiol	Polar Glucuronic Acid Conjugates
Indomethacin	Polar Sulfate Conjugates
Mestranol	Sulindac

- Extensive enterohepatic cycling may be partly responsible for a drug's long persistence in the body.

- Orally administered activated charcoal and/or anion exchange resins have been used clinically to interrupt enterohepatic cycling and trap drugs in the gastrointestinal tract.

- Frequently, **when a compound is secreted into the intestine through the bile, it is in the form of a conjugate.**

- **Conjugated drugs will not be reabsorbed readily from the gastrointestinal tract unless the conjugate is hydrolyzed by gut enzymes such as -glucuronidase**.

- Such a continuous recirculation may lead to the appearance of drug-induced toxicity.

- Liver disease or injury may impair bile secretion and thereby lead to accumulation of certain drugs
- *Example* probenecid, digoxin, and diethylstilbestrol.

- Impairment of liver function can lead to decreased rates of both drug metabolism and secretion of drugs into bile.

- These effects may be brought about through an alteration in one or more of the following factors:
  - hepatic blood flow
  - uptake into hepatocytes
  - rate of biotransformation
  - transport into bile
  - rate of bile formation.
- In addition, antibiotics may alter the intestinal flora in such a manner as to diminish the presence of sulfatase and glucuronidase-containing bacteria.
- This would result in a persistence of the conjugated form of the drug and hence a decrease in its enterohepatic recirculation.

PULMONARY EXCRETION

- Any volatile material, irrespective of its route of administration, has the potential for pulmonary excretion.
- Certainly, gases and other volatile substances that enter the body primarily through the respiratory tract can be expected to be excreted by this route.
- *No* specialized transport systems are involved in the loss of substances in expired air; simple diffusion across cell membranes is predominant.
- The rate of loss of gases is not constant; it depends on the rate of respiration and pulmonary blood flow.
- The degree of solubility of a gas in blood also will affect the rate of gas loss.
- Gases such as nitrous oxide, which are not very soluble in blood, will be excreted rapidly, that is, almost at the rate at which the blood delivers the drug to the lungs.

- *Increasing cardiac output has the greatest effect on the removal of poorly soluble gases; for example, doubling the cardiac output nearly* doubles the rates of loss.

- Agents with high blood and tissue solubility, on the other hand, are only slowly transferred from pulmonary capillary blood to the alveoli.

EXCRETION IN OTHER BODY FLUIDS

Sweat and Saliva

- Minor importance for most drugs.

- Mainly depends on the diffusion of the un-ionized lipid-soluble form of the drug across the epithelial cells of the glands.

Milk

- The ultimate concentration of the individual compound in milk will depend on many factors, including the amount of drug in the maternal blood, its lipid solubility, its degree of ionization, and the extent of its active excretion.

- The physicochemical properties that govern the excretion of drugs into saliva and sweat also apply to the passage of drugs into milk.
- Since milk is more acidic (pH 6.5) than plasma, basic compounds (e.g., alkaloids, such as morphine and codeine) may be somewhat more concentrated in this fluid.

- In general, a **high maternal plasma protein binding of drug will be associated with a low milk** concentration.

- **A highly lipid-soluble drug sho**uld accumulate in milk fat.

First-order kinetics

- the rate of drug metabolism and elimination is directly proportional to the concentration of free drug, and first-order kinetics is observed. This means that a constant fraction of drug is metabolized per unit of time (that is, with each half-life, the concentration decreases by 50%).
- First-order kinetics is also referred to as linear kinetics.

Zero-order kinetics

- The rate of elimination is constant and does not depend on the drug concentration.
- The enzyme is saturated by a high free drug concentration, and the rate of metabolism remains constant over time.
- This is called zero-order kinetics (also called nonlinear kinetics).
- A constant amount of drug is metabolized per unit of time.

Total body clearance and drug half-life are important measures of drug clearance that are used to optimize drug therapy and minimize toxicity

Total body clearance

The total body (systemic) clearance, CLtotal, is the sum of all clearances from the drug-metabolizing and drug-eliminating organs. The kidney is often the major organ of elimination. The liver also contributes to drug clearance through metabolism and/or excretion into the bile. Total clearance is calculated using the following equation:

Cltotal= CLhepatic + Clrenal +Clpulmonary+CLother

where are CLhepatic + Clrenal typically the most important.

Clinical situations resulting in changes in drug half-life

When a patient has an abnormality that alters the half-life of a drug, adjustment in dosage is required.

Patients who may have an **increase in drug half-life** include those with

1) diminished renal or hepatic blood flow, for example, in cardiogenic shock, heart failure, or hemorrhage;

2) decreased ability to extract drug from plasma, for example, in renal disease; and

3) decreased metabolism, for example, when a concomitant drug inhibits metabolism or in hepatic insufficiency, as with cirrhosis.

These patients may require a decrease in dosage or less frequent dosing intervals.

In contrast, the **half-life of a drug may be decreased** by increased hepatic blood flow, decreased protein binding, or increased metabolism. **This may necessitate higher doses or more frequent dosing intervals.**

Optimization of dose

Drug regimens are administered as a maintenance dose and may require a loading dose if rapid effects are warranted.

Maintenance dose: Drugs are generally administered to maintain a Css within the therapeutic window. It takes four to five half-lives for a drug to achieve Css. To achieve a given concentration, the rate of administration and the rate of elimination of the drug are important.

Loading dose

Sometimes rapid obtainment of desired plasma levels is needed (for example, in serious infections or arrhythmias). Therefore, a “loading dose” of drug is administered to achieve the desired plasma level rapidly, followed by a maintenance dose to maintain the steady state. In general, the loading dose can be calculated as

Loading dose = (Vd) × (desired steady-state plasma concentration)/F

For IV infusion, the bioavailability is 100%, and the equation becomes

Loading dose = (Vd) × (desired steady-state plasma concentration)

Loading doses can be given as a single dose or a series of doses.
Disadvantages of loading doses include

- increased risk of drug toxicity
- a longer time for the plasma concentration to fall if excess levels occur.

A loading dose is most useful for drugs that have a relatively long half-life.

Without an initial loading dose, these drugs would take a long time to reach a therapeutic value that corresponds to the steady-state level

Which of the following phase II metabolic reactions makes phase I metabolites readily excretable in urine?

- A. Oxidation.
- B. Reduction.
- C. Glucuronidation.
- D. Hydrolysis.
- E. Alcohol dehydrogenation.

Alkalization of urine by giving bicarbonate is used to treat patients presenting with phenobarbital (weak acid) overdose. Which of the following best describes the rationale for alkalization of urine in this setting?

- A. To reduce tubular reabsorption of phenobarbital.
- B. To decrease ionization of phenobarbital.
- C. To increase glomerular filtration of phenobarbital.
- D. To decrease proximal tubular secretion.
- E. To increase tubular reabsorption of phenobarbital.

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