Semester I.
Seminar 8.

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Exam titles 15-16

15.

- First pass effect
- Synthesis, storage, release and elimination of acetylcholine (Ach). Demonstration of Dale's experiment
- Agents used in anemias

16.

- Drug elimination: I. Biotransformation
- Non-adrenergic, non-cholinergic (NANC) transmission
- Drugs used in coagulation disorders

Pharmacokinetics

- „Effect of the body on the drug”
- Fate of the drug is divided into 4 stages designated by the acronym ‘ADME’:

- • Absorption from the site of administration
- • Distribution within the body
- • **Metabolism**
- • Excretion

Invasion

Elimination

First pass effect

First pass effect

- The **first-pass effect** is a phenomenon of drug metabolism:
  - the concentration of a drug is greatly reduced before it reaches the systemic circulation („presystemic metabolism”)

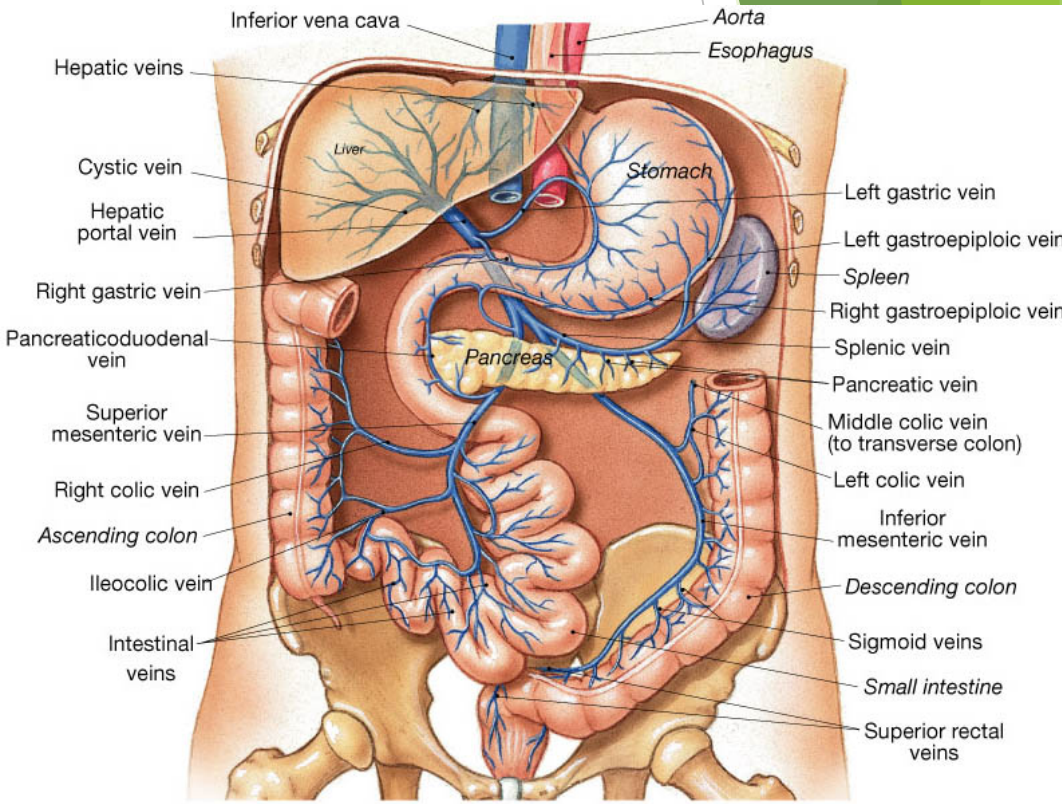
- Route: drug is swallowed ➔ absorbtion hepatic portal system (portal vein) ➔ drug enters liver ➔ The liver metabolizes many drugs. ➔ drug gets into systemic circulation

Other mechanisms take part as well:

- The four primary systems that affect the first pass effect of a drug are
  - the enzymes of the gastrointestinal lumen,
  - gut wall enzymes,
  - bacterial enzymes, and
  - hepatic enzymes

Consequences of First pass effect

- Bioavailability of a drug is low
- Higher dose is needed orally than parenterally
- Marked individual variations exist in the extent of first-pass metabolism



Examples: Substantial first-pass metabolism

Table 9.2 Examples of drugs that undergo substantial first-pass elimination

Aspirin	Metoprolol
Glyceryl trinitrate	Morphine
Isosorbide dinitrate	Propranolol
Levodopa	Salbutamol
Lidocaine	Verapamil

Rang and Dale 7th ed.

Drug elimination:
I. Biotransformation

Definition

"Biotransformation of drug is defined as the **conversion from one chemical form to another".**

- the term is used synonymously with ***metabolism***.

Biotransformation may lead to:

Pharmacologic Inactivation of Drug

Active Drug → **Inactive Drug**

Ex: Salicylic Acid → Salicyluric Acid

Active Metabolite From An Inactive Drug

Inactive(Prodrug) → **Active**

Ex. Enalapril → Enalaprilate

No Change in Pharmacological Activity

Active → **Active Drug**

Ex. Codeine → Morphine

Drug metabolising organs

- Liver is the heart of metabolism
  - Because of its relative richness of enzymes in large amount.

- Schematic chart of metabolizing organs (decreasing order):

- Liver > Lungs > Kidney > Intestine > Placenta > Skin > Brain > Testes > Muscle > Spleen

Fundamental concepts in drug biotransformation

- Lipid soluble drugs are poorly excreted in the urine.
- They tend to store in fat and/or circulate until they are converted
  - to more water soluble metabolites (**phase I biotransformation**) or
  - to metabolites conjugated with water soluble substances (**phase II biotransformation**).
- Water soluble drugs are more readily excreted in the urine.
  - (They may be metabolized, but generally not by the CYP enzyme systems.)

Phase I reactions (catabolic)

1. **Microsomal (P450-dependent) oxidations**
2. **Non microsomal oxidations**
3. **Reduction**
4. **Hydrolysis**
5. **Hydratation**
6. **Isomerisation**
7. **Mixed reactions**

- Phase 1 reactions often produce a reactive group (='functionalisation')
- This group then serves as the point of attack for the Phase 2 reactions.
- The products are often more chemically reactive and hence, paradoxically, sometimes more toxic or carcinogenic than the parent drug.

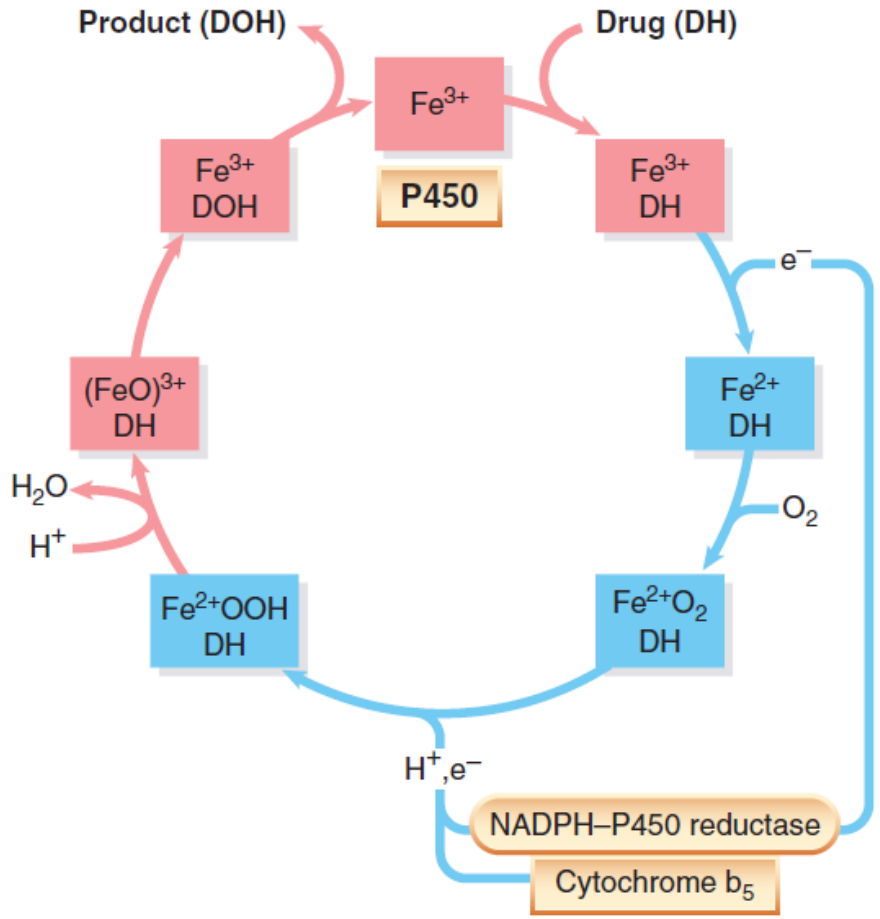
Phase I reactions

1. Microsomal oxidation.

- Takes place in the ER
- The enzymes are part of an electron-transport-chain consisting of
  - Flavoprotein, or NADPH:P450 reductase and
  - Hemoprotein or **cytochrome P450**
    (these are the mixed function oxidases)
- Reduced NADPH and O_2 is necessary for its functioning

- The most common microsomal oxidation reaction catalyzed by cytochrome P450 enzymes is **a monooxygenase reaction**
  (cytochrome P450-system=monooxigenase system)
- Other reactions include: aromatic, aliphatic oxidation, epoxide forming, N-, O- S- dealkylization, oxidative desamination, S-, N- oxidation, dehalogenization, alcohol-oxidation

The monooxygenase reaction



overall net effect of the reaction is quite simple = the addition of one atom of oxygen (from molecular oxygen) to the drug to form a hydroxyl group (product, 'DOH'), the other atom of oxygen being converted to water

DH + O_2 + NADPH + H^+ → **DOH** + H_2O + $NADP^+$

The CYP-s

- CYP = CYtochrome P450
  - Cyto = cell; chrome = colorful; P = pink ➔ 450= 450nm spectral absorption
- They are hemoproteins = contain heme cofactor (including an iron atom; $Fe^{2/3+}$)
- The CYP **play a key role in the metabolism**
  - of endogenous substrates
    - (e.g. fatty acids, eicosanoids, sterols and steroids, bile acids, vitamin D, retinoids and uroporphyrogens)
    - e.g. enzymes of steroid synthesis are ALL cytochrome P450s
  - of foreign chemicals/drugs (= detoxification)
- Numbers:
  - More than 21000 distinct CYP proteins are known
  - ***Humans have 18 families of CYP-s***
- Most important in drug metabolism are the CYP 1, 2 and 3 subfamilies:

CYP3A4 - Participate in the 60% of the metabolism of the drugs.
CYP2D6 - (20-25%)
CYP2C19 - (Smaller percent but some significant interactions)
CYP1A2 – chronic smoking induces it
CYP2E1 – chronic alcohol consumption induces it

Phase I reactions

2. Non-microsomal oxidations.
   - Performed by enzymes with NAD-cofactor found in mitochondrium, and in cytoplasm.
   - (alcoholdehydrogenase, aldehyde-oxidase, aromatase, amin-oxidase)
3. Reductional tranformations.
   - In microsomes, cytoplasm or by bacteria in the intestines.
   - (azo-reductase, nitro-reductase, epoxid reduction, reduction of heterocyclic compounds)
4. Hydrolysis.
   - Performed by esterases.
   - (ester-, amid-, azide-hydrolysis)
5. Hydratation
   - Incorporation of H2O into molecules
6. Isomerisation
   - Transforming from one isomer to another
7. Mixed reactions
   - closing of rings, opening of rings, N-carboxylation, dimerisation, transamidation, decarboxylation

Phase II reactions (anabolic)

1. Glucuronide Conjugation
2. Methylation
3. Acetylation
4. Sulfate Conjugation
5. Conjugation With Amino Acids (mainly Glycine)
6. Glutathione Conjugation
7. Cyanide Conjugation

- Phase 2 reactions combine functional group of compound with endogenous substance (=’Conjugation’)
  - This often happens on groups formed in the phase I reactions.
- The final compounds always have a larger molecular weight.
- Products are usually very hydrophilic➔watersolubility increases➔excretion increases
- As opposed to phase I, drug effect decreases almost always in phase II reactions.
- Sometimes conjugates may transform further (III. phase)

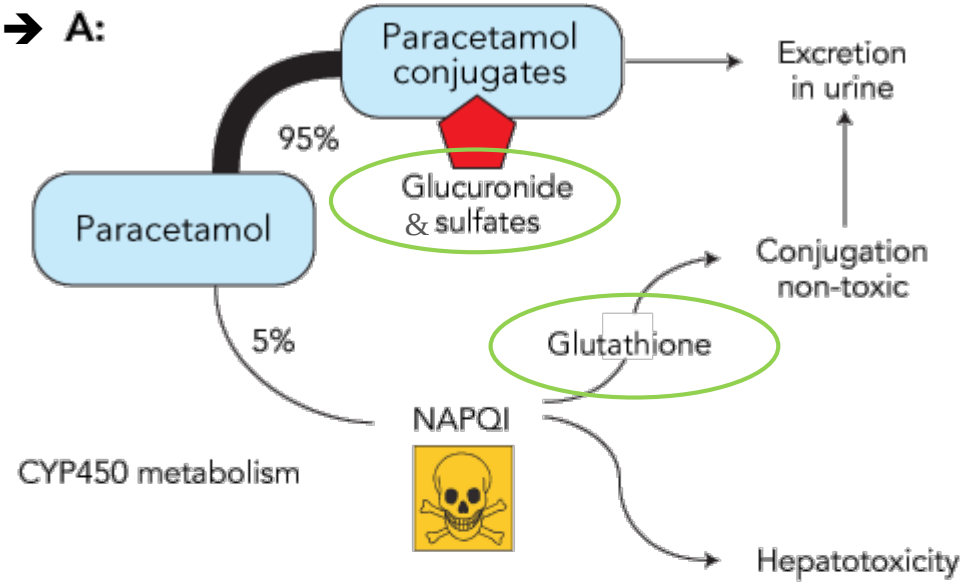
Phase II reactions

1. Glucuronide conjugation (=glucuronidation)
   - Performed by UDP-glucuronyl-transferase (UGT); UDP-glucuronic acid is needed
   - the enzyme beta-glucuronidase works against it
     - found in gut (see enterohepatic circle next seminar),
     - and in breast milk (which contributes to neonatal jaundice)
   - molecular weight may increase so much that product cannot be secreted through the glomerule ➔ it gets into the enterohepatic circle.
   - Endogenous substances:
     - adrenal corticosteroids
     - bilirubin
       - A deficiency in the bilirubin specific form of glucuronyl-transferase is thought to be the cause of **Gilbert's syndrome**, which is characterized by unconjugated hyperbilirubinemia.
       - It is also associated with **Crigler-Najjar syndrome**, a more serious disorder where the enzyme's activity is either completely absent (Crigler-Najjar syndrome type I) or less than 10% of normal (type II).
   - Exogenous substances:
     - Famously, UGT enzymes are not present in the genus Felis, and this accounts for a number of unusual **toxicities in the cat** family.
     - Infants may have a developmental deficiency in UDP-glucuronyl transferase, and are unable to hepatically metabolize the antibiotic drug **chloramphenicol** which requires glucuronidation. This leads to a condition known as **gray baby syndrome**

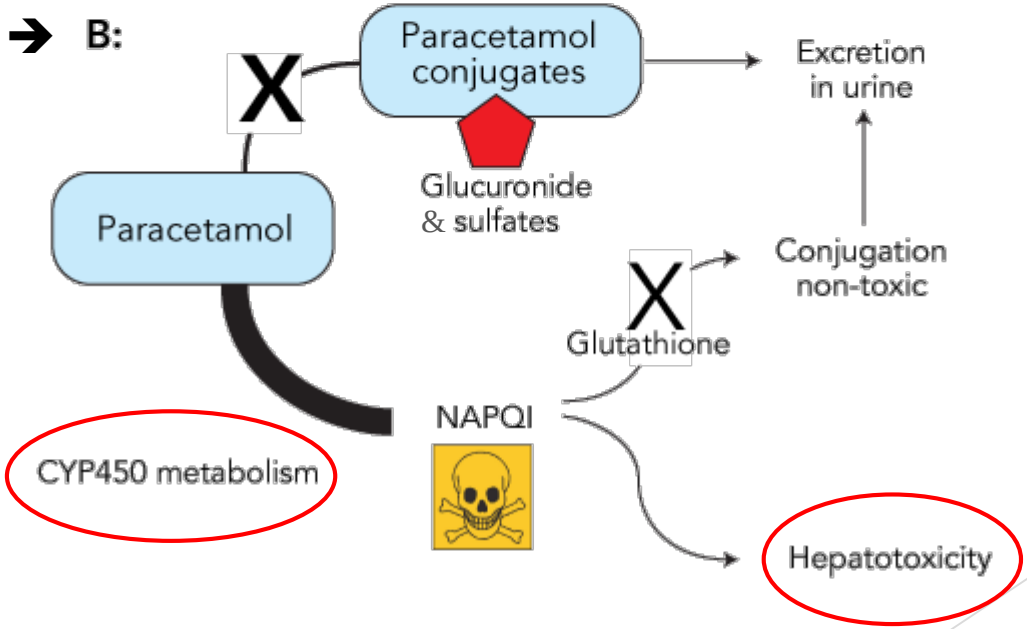
Phase II reactions

2. Metylation.
   - Performed by Metyltransferases (in ER and cytosol),
   - their metyl-donor is S-adenosyl-methionin (SAM)
   - Lungs, kidneys.
3. Acetylation.
   - N-acetyltransferase,
   - AcCoA is the cofactor.
   - Liver, lungs, kidneys, intestines.
4. Sulphate conjugation.
   - Performed by the sulphotransferase enzyme (in the cytosol),
   - its cofactor is the 3'-phosphoadenosine-5'-phosphosulphate (PAPS).
   - Very watersoluble, rapidly excreted substances are formed.
   - The process is saturable.
5. Aminoacid-conjugation (mainly Glycine).
   - Glycin is the most common, but also taurin-, glutamin-, ornitinconjugates are formed as well.
6. Glutathione-conjugation.
   - Through glutathion S-transferase enzymes.

Normal dose ➔ **A:**



Overdose ➔ **B:**



NAPQI = *N*-acetyl-*p*-benzoquinone imine