Pharmacokinetics (PK): Drug Metabolism/Biotransformation

1. Principles:

Drug biotransformation (Metabolism) is the biochemical modification of pharmaceutical compounds by **endogenous enzymes**. This process is essentially a defense mechanism designed to convert **lipophilic xenobiotics** (which are difficult to excrete and have high Vd) into **hydrophilic** metabolites that are more readily eliminated by the kidneys or biliary system.

2. The Two-Phase System

The standard model of biotransformation involves two distinct enzymatic sequences, primarily localized in the smooth endoplasmic reticulum (microsomes) and cytosol of hepatocytes.

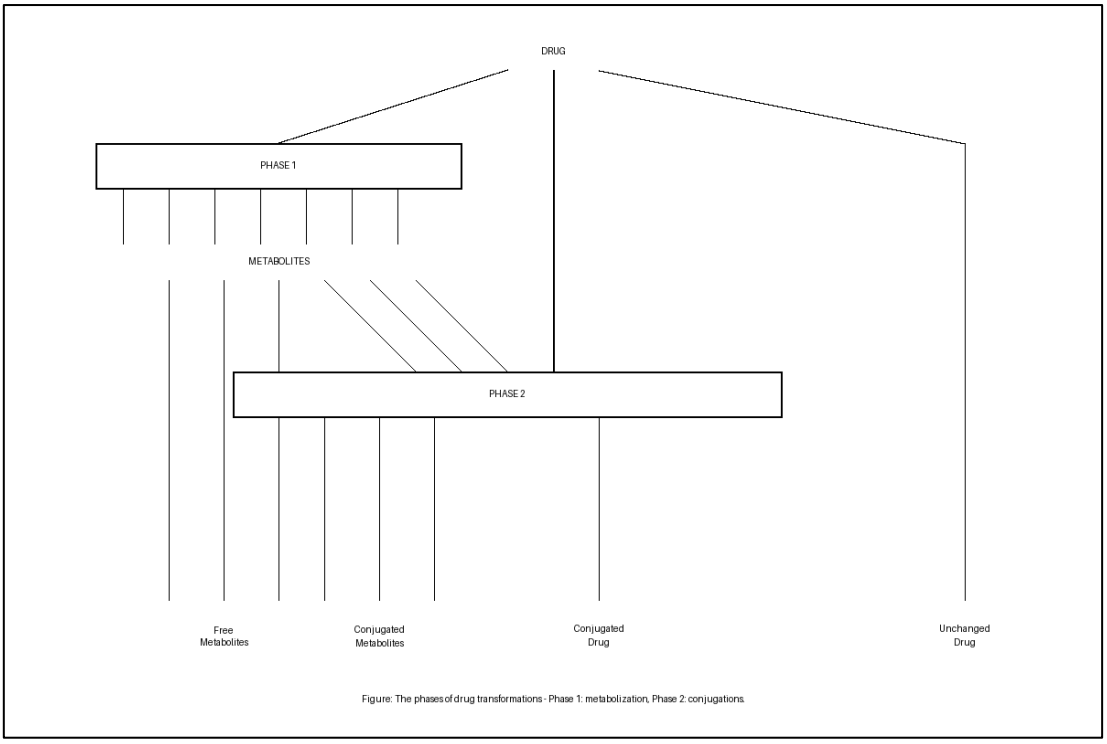

Figure: The phases of drug transformations - Phase 1: metabolization, Phase 2: conjugations.

Phase I: Functionalization

The objective of Phase I is to introduce or unmask a polar functional group (e.g., -OH, -NH_2, -SH, -COOH).

- **Primary Reactions:** Oxidation (most common), Reduction, and Hydrolysis.
- **Outcome:** The metabolite may be **inactivated**, **activated** (in the case of pro-drugs), or converted into an **active metabolite** with different pharmacologic properties. Sometimes it may elicit toxic byproduct.

- **Key Enzyme System:** The **Cytochrome P450 (CYP)** superfamily of heme-containing monooxygenases.The general equation for a CYP-mediated oxidation is:

$$\text{Drug} + O_2 + \text{NADPH} + H^+ \rightarrow \text{Drug-OH} + \text{NADP}^+ + H_2O$$

Phase II: Conjugation

Phase II reactions involve the covalent attachment of an endogenous, highly polar molecule to the Phase I metabolite (or the parent drug if it already possesses a functional group).

- **Objective:** To increase water solubility (decrease log P) to the point where the molecule is trapped in the renal tubule and excreted.
- **Primary Reactions:** * **Glucuronidation:** (UDP-Glucuronosyltransferases / UGT) — The most common Phase II reaction.
    - **Sulfation:** (Sulfotransferases / SULT).
    - **Glutathione Conjugation:** (GSH-transferases / GST) — Vital for neutralizing reactive electrophilic intermediates.
    - **Acetylation and Methylation.**

Feature	**Phase I (Functionalization)**	**Phase II (Conjugation)**
Reaction Types	Oxidation, Reduction, Hydrolysis	Glucuronidation, Sulfation, Acetylation
Enzymes	CYP450, FMO, Esterases	UGT, SULT, GST, NAT
Substrate Change	Addition/exposure of functional group	Attachment of a large polar group
Molecular Weight	Minimal change	Significant increase
Hydrophilicity	Moderate increase	Massive increase
Result	Active, Inactive, or Toxic metabolites	Usually inactive (highly polar)

3. Factors Influencing Biotransformation

- **Enzyme Induction:** Some drugs (e.g., Rifampin, Phenobarbital) increase the expression of CYP enzymes, leading to accelerated metabolism of co-administered drugs and potential therapeutic failure.
- **Enzyme Inhibition:** Drugs (e.g., Ketoconazole, Grapefruit juice) can bind and inhibit CYPs, leading to decreased clearance and potential toxicity of other drugs.
- **Genetic Polymorphism:** Variations in genes (e.g., *CYP2D6*) lead to "Poor Metabolizers" or "Ultra-rapid Metabolizers," necessitating personalized dosing.

4. Pro-drugs and Bioactivation

In some instances, the parent drug is pharmacologically inactive. Biotransformation is required to convert the pro-drug into its active form.

- **Example: Codeine** (pro-drug) is O-demethylated via **CYP2D6** into **Morphine** (active).
- **Toxicological Note:** Biotransformation can also produce reactive intermediates. For instance, **Paracetamol** (Acetaminophen) is converted into **N-acetyl-p-benzoquinone imine** (**NAPQI)**, a highly reactive intermediate that can cause hepatic necrosis if glutathione stores are depleted.
  NAPQI stands for **N-acetyl-p-benzoquinone imine**.
  In the context of drug biotransformation, it is a highly reactive and toxic intermediate metabolite of **Paracetamol** (Acetaminophen).

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1. Biochemical Formation

While most Paracetamol is metabolized via Phase 2 reactions (Glucuronidation and Sulfation), a small fraction (approximately 5–10%) undergoes **Phase 1 oxidation** by the Cytochrome P450 enzyme system, specifically the **CYP2E1** and **CYP3A4** isoenzymes. This oxidation produces NAPQI.

2. Detoxification and Toxicity

Under normal therapeutic conditions, NAPQI does not cause harm because it is immediately neutralized by **Phase 2 conjugation** with **Glutathione (GSH)**, catalyzed by Glutathione S-transferase. This forms a non-toxic mercapturic acid derivative which is excreted renally.

Pathological Scenario (Overdose):

- **Glutathione Depletion:** In an overdose, the primary Phase 2 pathways become saturated, and more drug is shunted toward the CYP450 pathway. This creates an excess of NAPQI that exhausts the liver's Glutathione stores.
- **Cellular Necrosis:** Once Glutathione is depleted, the "free" NAPQI binds covalently to vital cellular proteins and lipid bilayers of hepatocytes. This causes oxidative stress and mitochondrial dysfunction, leading to **Centrilobular Hepatic Necrosis**.

3. Clinical Antidote: N-acetylcysteine (NAC)

The standard treatment for Paracetamol toxicity is **N-acetylcysteine (NAC)**. It works through two mechanisms:

1. It acts as a precursor to **L-cysteine**, which is the rate-limiting substrate for Glutathione synthesis.
2. It can bind directly to NAPQI as a substitute for Glutathione.

5. Biotransformation of biologics

Biotransformation for **biologics** (large molecules like monoclonal antibodies, cytokines, and recombinant proteins) is fundamentally different from the "Small Molecule" (xenobiotic) pathways we just discussed. While small molecules rely on the liver's enzymatic "machinery" (CYP450 and Conjugation), biologics are essentially treated by the body as **nutrients** or **endogenous proteins**. They do not follow the Phase I/Phase II paradigm.

Feature	**Small Molecules (Xenobiotics)**	**Biologics (Large Proteins)**
Pathway	Phase I & II (CYP450, UGT)	**Proteolysis / Catabolism**
Main Site	Liver (Hepatocytes)	Plasma, Interstitium, Lysosomes
Metabolites	Polar derivatives (sometimes toxic)	**Amino Acids** (Non-toxic)
Recycling	None (Enterohepatic at most)	**FcRn Recycling** (for IgGs)
Excretion	Renal / Biliary	Minimal renal (too large for GFR)