Drug Metabolism

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Biotransformation: the process whereby lipid-soluble drugs are converted to more water-soluble metabolites

a.k.a. drug metabolism

Implies a change in structure of the drug

Two types biotransformations: Phase 1 reactions are non-synthetic, Phase 2 reactions are synthetic

Drugs can undergo either type reaction alone or they can undergo combinations of Phase 1 and Phase 2 reactions

Conversion to water-soluble products occurs through metabolic transformations

Consequences of drug biotransformation

<table>
<thead>
<tr>
<th>Drug (D)</th>
<th>Enzyme (E)</th>
<th>Metabolites (DS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Active</td>
<td>MAJOR</td>
<td>Inactive</td>
</tr>
<tr>
<td>Active</td>
<td></td>
<td>More toxic</td>
</tr>
<tr>
<td>Inactive</td>
<td>MINOR</td>
<td>Active</td>
</tr>
<tr>
<td>Active</td>
<td></td>
<td>Equally active</td>
</tr>
</tbody>
</table>

Metabolism can be a “Double edged sword”

Consequences of drug biotransformation

Metabolism promotes drug excretion

Increased water solubility/ increased ionization, especially by phase 2 reactions decreases reabsorption in GI tract and kidney

Prepares drug for secretion into bile and urine

Decreases entry of drugs into cells (effects on Vd)

Sites of biotransformation: tissues

Occurs in many places in the body, but especially the liver, GI tract, and kidneys.

Other sites of metabolism include skin, lungs, brain, placenta,

Decreases entry of drugs into cells (effects on Vd)
Sites of biotransformation: cellular

In the liver, a major site of drug biotransformation is in the membrane of the smooth endoplasmic reticulum

Homogenized SER purified by centrifugation = “microsomes”

Enzymes localized to microsomes include the major types of Phase I and Phase 2 enzymes: the cytochrome P450s and the UDP-glucuronosyltransferases, respectively.

SER is not the only cellular or extracellular location of drug metabolizing enzymes

Types of non-synthetic Phase 1 reactions

- **Oxidation** – addition of oxygen, most common type of Phase 1 reaction
  - Two types: oxygen appears in drug metabolite structure oxidation occurs followed by molecular rearrangement
  - Reduction – removal of oxygen
  - Hydrolysis –splitting of an ester or amide bond by insertion of water-second most common type of phase 1 reaction
  - Phase 1 reactions generally “functionalize” a chemical (adds functional groups or unmasks functional groups such as –OH, COOH, amine group, etc.
  - Functional groups serve as “handles” for synthetic type (Phase 2 reactions)

Oxidation I

**Oxidative dealkylation**

- \( \text{RNH}_2 \rightarrow \text{RNHOH} \)
  - Oxidation / molecular rearrangement

- **N-Dealkylation**
  - Murphee, ethylmorphine, benzphetamine, amipryline, caffeine, theophylline.

- **O-Dealkylation**
  - Codeine, \( \text{p-nitroanisole} \).

- **S-Dealkylation**
  - \( 6\)-Methylthiopurine, methitural.

**Alkyl group equals a saturated carbon side chain (methyl or ethyl group)**

Oxidation II

**N-Oxidation**

- \( \text{RNH}_2 \rightarrow \text{RNHOH} \)
  - Oxygen in drug metabolite

- **Primary amines**
  - \( \text{RNH}_2 \rightarrow \text{RNHOH} \)
  - 2-Arylamine/fluorene, epoxides, etc.

- **Secondary amines**
  - \( \text{RNH}_2 \rightarrow \text{RNHOH} \)
  - Oxygen in drug metabolite

- **Tertiary amines**
  - \( \text{RNH}_2 \rightarrow \text{RNHOH} \)
  - N-Nitro, methylnitrogen

- **Aminos in methylnitrogen**
  - Oxygen in drug metabolite

Oxidation III

**S-Oxidation**

- \( \text{RCH}_2 \text{OH} \rightarrow \text{RCH}_2 \text{O} \)
  - Oxidation / molecular rearrangement

- **Deamination**
  - \( \text{NH}_2 \rightarrow \text{NH}_2 \)
  - \( \text{OH}_{2} \rightarrow \text{OH}_{2} \)

- **S-Oxidation**
  - \( \text{RSH} \rightarrow \text{RSH} \)
  - Oxidation / molecular rearrangement

**Deaminolysis**

- \( \text{CH}_2 \text{OH} \rightarrow \text{CH}_2 \text{OH} \)
  - Oxidation / molecular rearrangement

- **Deamination**
  - \( \text{NH}_2 \rightarrow \text{NH}_2 \)
  - \( \text{OH}_{2} \rightarrow \text{OH}_{2} \)

**Deamination**

- \( \text{NH}_2 \rightarrow \text{NH}_2 \)
  - Oxidation / molecular rearrangement

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**Deamination**

- \( \text{NH}_2 \rightarrow \text{NH}_2 \)
  - Oxidation / molecular rearrangement
Phenobarbital hydroxylation

Oxidation IV

<table>
<thead>
<tr>
<th>Reaction</th>
<th>Phenobarbital</th>
</tr>
</thead>
<tbody>
<tr>
<td>R-H → R-OH</td>
<td></td>
</tr>
<tr>
<td>Oxygen in drug metabolite</td>
<td></td>
</tr>
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</table>

Cytochrome P450

P450s are a major family of enzymes that catalyze oxidation reactions. Often referred to as Microsomal Mixed Function Oxidases because of the splitting of oxygen that occurs in the reaction. 

NADPH + O₂ + H⁺ + Drug → NADP⁺ + H₂O + Drug-OH

The ability to bind and utilize oxygen as a cosubstrate in the reaction is due to the presence of a ferrous ion containing heme group in the active P450 enzyme.

Oxidation V the P450 reaction cycle

1- Substrate Binding
2- Substrate Reduction
3- Substrate Oxygenation
4- Substrate Rearrangement
5- Product Dissociation

P450 collectively are able to oxidize thousands of drugs. Their versatility is due in part to the presence of multiple families of enzymes, each family containing multiple family members. Four P450s that are major contributors to drug oxidation are CYP1A2 (12% of all drugs), CYP2B6 (20%), CYP2E1 (6%), and CYP3A4 (28%). Others include CYP2A6, CYP2C9, CYP2C19 and CYP2D6.

Versatility also due to broad and overlapping substrate specificity of each P450. Drug drug interactions stemming from competition between drugs for P450 metabolism, especially CYP3A4 mediated metabolism, is common.

Regulation of P450 oxidases and effect on drug metabolism

The levels of certain P450s are higher in individuals who drink alcohol, smoke, or use certain therapeutic drugs (higher rates of metabolism → give more drug)

The levels of P450s are lower in the elderly and in infants (lower rates of metabolism → give less drug)

Other types of phase 1 reactions: ester hydrolysis

Hydrolysis of Aspirin

\[ \text{O} \quad \text{E} = \text{plasma esterase} \]

\[ \text{R} - \text{C} \quad \text{E} \quad \text{OR} \quad \text{E} \quad \text{R} - \text{COOH} + \text{RCH} \quad \text{acid} + \text{alcohol} \]

\[ \text{COOH} \quad \text{O} \quad \text{E} \quad \text{OH} \quad \text{OH} \quad \text{E} \quad \text{R} - \text{C} \quad \text{acetic} \]

\[ \text{aspirin} \quad \text{salicylic} \quad \text{acetic} \]
**Ester hydrolysis II**

Short-acting local anesthetic, injected into tissue (local effect), metabolized rapidly after absorption into blood

PABA → allergic reactions

**Amide hydrolysis**

**Amide hydrolysis III**

Longer acting local anesthetic Anti-arrhythmic

**Phase 2 reactions: glucuronidation**

<table>
<thead>
<tr>
<th>Reaction</th>
<th>Compound</th>
<th>Metabolite</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>salicylic acid (SA)</td>
<td>ester glucuronide of SA</td>
</tr>
<tr>
<td>10%</td>
<td></td>
<td>5%</td>
</tr>
</tbody>
</table>

**Phase 2 reactions: acetylation**

Acetylation is a major type of conjugation reaction for drugs with amine groups (the antibacterial sulfanilamide)

**Phase 2 reactions: glycine conjugation**

Conjugation with the amino acid glycine is another type of conjugation reaction.

75% major metabolite of Aspirin
Phase 2 reactions: methylation

- epinephrine
  - O-, N-methyltransferase (OMT & NMT)
  - S-Adenosylmethionine (SAM)
- metanephrine
- norepinephrine

Phase 2 reactions: glutathione conjugation

- ACETAMINOPHEN
- PAPS UDPGA
- HNCOCH₃
- SULFATE
- 45 - 50%
- HNCOCH₃
- OXIDATIVE STRESS (•OH, O₂•)
- POSTULATED INTERMEDIATES
- HIGH DOSE (>10g)
- NUCLEOPHILIC CELL MACROMOLECULES
- GLUCURONIDE
- 45 - 50%
- HNCOCH₃
- NAcetylcycteine
- ALCOHOLIC

Factors Influencing Drug Metabolism I

- Enzyme Induction (slow) increases drug clearance
  - Diseases: Hyperthyroidism
  - Drugs [many]: PB, Rifampin, Phenytoin, etc.
  - Conditions: smoking, alcoholism
  - Higher doses of drugs are required
  - Only one induction period then stable level

Factors Influencing Drug Metabolism II

- Enzyme Inhibition (fast) reduces drug clearance
  - Diseases: Hypothyroidism, Liver Disease
  - Drugs (many): Chloramphenicol, Cimetidine, Disulfiram, Ethanol (acute), etc.
  - Dietary factors: e.g., Grapefruit juice
  - Conditions: Pregnancy, Aging, Newborn

Effect of substrate (drug) concentration on rate of metabolism

- First order:
  - Rate of metabolism proportional to substrate concentration
  - Reaction rate $\propto \frac{1}{K_m}$

- Zero order kinetics:
  - Rate of metabolism independent of substrate concentration (constant)
  - Maximum velocity $V_{max}$

Factors Influencing Drug Metabolism I

- Enzyme Induction (slow) increases drug clearance

CYP1A2 is induced by cigarette smoking and consumption of charbroiled foods (active constituents are 3-methylcholanthrene and benzo[a]pyrene)

CYP2B6 and CYP3A4 are inducible by therapeutic drugs--phenobarbital, rifampin, and phenytoin.

CYP2E1 is induced by isoniazid (drug for tuberculosis) and by ethanol.

Factors Influencing Drug Metabolism II

- Enzyme Inhibition (fast) reduces drug clearance

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P450 induction

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Factors Influencing Drug Metabolism III

Age: low metabolism in elderly and newborn
start low and go slow with drug dose

Nutrition: high metabolism with chronic intake of alcohol
and charcoal cooked food and lower with high acute alcohol intake.

Factors Influencing Drug Metabolism IV

Genetic Variations:
Isoniazid [prophylaxis of tuberculosis] produces liver injury in slow acetylators (variation in N-acetyltransferase gene 2 gene or NAT2).

Succinylcholine [surgical muscle relaxant] produces prolonged respiratory depression (apnea) in patients with abnormal plasma cholinesterase which reduces the hydrolysis of succinylcholine.

Codeine (narcotic analgesic)-10-15% is converted to morphine by CYP2D6-mediated O-demethylation (dealkylation) CYP2D6 is polymorphic in the human population