Metabolism

• Chemical transformation of xenobiotics
• Occurs in mostly in liver (enzymatic processes)
• Conversion into more hydrophil. subst. - excretion urine
• May convert procariogenics into cytotox., mutagenic compounds
• Different persons may have differences in metabolism (genetic diff., physiol. factors)
• Metabolism of one xenobiotic may influence metab. of another

Xenobiotics
• Drugs
• Other foreign non-essential compounds

Metabolism in non-hepatic tissue
• Intestine mucosa
• Kidney
• Lung
• Bacteria in GI-tract

First-pass metabolism:
Xenobiotic metabolized before reaching general circulation
First-pass metabolism:
Xenobiotic metabolized before reaching general circulation

A) Metab. lungs (inhaled subst)
   Intestine mucosa, GI bacteria

B) Absorb under tung
   Absorb from rectum
Pathways of metabolism

Phase 1: Biotransformation
Attachment of new functional groups, transformation of exist. funct. groups
oxidation, reduction, hydroxylation, hydrolysis etc.

Phase 2: Conjugation.
Masking of an exist. funct. group by for instance
acetylation, glycosylation, attachment amino acid etc

More hydrophilic drug

Renal excretion
Phase 1
Metabolism by cytochrome P450 enzyme system (CYP450)

• Located in endoplasmatic reticulum (liver and other cells)
• Electron transport system - oxidation, monooxygenase
• Heme protein + flavoprotein
• Capable of oxidation - many different xenobiotics
**CYP450 families and sub-families**

**Family 1:**

- **CYP1A1**
  - Aromatic hydrocarbon hydroxylase, metabol. PAH etc.

\[
\begin{align*}
&\begin{array}{c}
\text{CYP1A2} \\
\text{Ox of arylamines, nitrosamines, aromatic hydrocarbons}
\end{array}
\end{align*}
\]

**Family 2:**

- **CYP2A6**
- **CYP2B6**
- **CYP2C**
- **CYP2D6:** Often enantioselective, lipophil. amines
- **CYP2E1:** Halogenated hydrocarbons, other org solvents

**Family 3:**

- **CYP3A4**
CYP450 / Mechanisms of metabolic transformations

Hydroxylation of alkane
Dehydrogenation of alkane

R-CH₃ \xrightarrow{\text{CYP450}} \text{R-CH}_2\text{OH} \xrightarrow{\text{Other enzymes}} \text{R-CO}_2\text{H}
Proposed mech. ox of alkenes

Alkene, aromatic π-complex

Allylic alchol

Radical σ-complex

1,2 migration

alkynes

Proposed mechanism of oxidation of alkenes and alkynes using iron(III) complexes.
Proposed mech. ox of aromatics
Proposed mech. react. on heteroatom cont. compounds

- Fe$^{5+}$O
- Fe$^{4+}$OH
- Fe$^{3+}$
- HX$^\cdot$R
- i.e. hemiacetal
- i.e. N oxide
- N, O, S dealkylation
- cleav. of small alkylgroups (Me)
- Dehalogenation: HX + carbonyl comp.
Proposed mech. react. on heteroatom cont. compounds

\[ 
\begin{align*}
\text{Fe}^{2+} &= \text{O} \\
\text{Fe}^{4+} &= \text{OH} \\
\text{Fe}^{4+} &= \text{OH} \\
\text{Fe}^{4+} &= \text{OH} \\
\text{N-oxide} \\
\text{sulfide ox, see FMO} \\
\text{Stable if R, R', R'' \neq H} \\
\text{hydroxylamine} \\
\text{hemiaminal} \\
\text{nitroso comp.} \\
\end{align*}
\]
CYP450  Induction / inhibition by xenobiotics

Xenobiotics may enhance metabol. of them selves as well as other comp. taken at the same time
Induce transcript CYP450 mRNA - Synth. CYP450 enzymes (enzyme induction)
• Drugs
• Ethanol
• Organic solvents
• Components in cig. smoke

St. Johns Worth (Johannesurt, prikkperikum)
CYP450 Inhibitors
Reversible CYP enzyme inhibitors: Several drugs ex. antimycotic azoles
CYP450 Inhibitors

Complexation inhibitors
ex. metabolites from alkylamines

\[
\text{R-NMe}_2 \stackrel{\text{CYP450}}{\longrightarrow} \text{R-N}=\text{O}
\]

Alkylamine
No inhib. activity

Nitrosoalkane
Irreversible complexation with Fe(II) heme in CYP sub families

Mechanism based inhibitors (suicide inhib)
ex. alkynes

\[\text{R} \rightarrow \text{FeO}^3 \text{CYP450} \rightarrow \text{R} \rightarrow \text{H}_2\text{O} \rightarrow \text{R-COOH} \rightarrow \text{R-CONH}\text{Enzyme} \]

Ethynyl estradiol
Phase 1 react. not involving CYP450

Other microsomal enzymes

Azoreductase

\[ \text{Prontocil} \xrightarrow{\text{Nitroreductase}} \text{Sulfanilamide} \]

Nitroreductase

\[ R-\text{NO}_2 \xrightarrow{\text{R-NH}_2} \]

Flavinmonooxygenase-FMO (N and S-ox.)

Peroxidases

**microsome**: Artefactual spherical particle, not present in the living cell, derived from pieces of the endoplasmic reticulum present in homogenates of tissues or cells: microsomes sediment from such homogenates when centrifuged at 100 000 g and higher: the microsomal fraction obtained in this way is often used as a source of mono-oxygenase enzymes.
Flavinmonooxygenase-FMO
Cont. FAD

Flavin adenine dinucleotide

\[ \text{FAD} \quad \Rightarrow \quad \text{FADH}_2 \]

Flavin mononucleotide

\[ \text{FMN} \quad \Rightarrow \quad \text{FMNH}_2 \]

Nicotinamid adenine dinucleotide

\[ \text{R=H} \quad \text{NAD}^{\oplus} \]

\[ \text{R=Phosphate: NADP}^{\ast}, \text{NADPH} \]

Riboflavin (Vit B2)

Adenine (Vit. B4)

Nicotinic acid / Niacin (Vit. B3)
Flavinmonooxygenase-FMO

Ox of soft Nu

• Amine: ox. to N-oxide / hydroxylamine
• Sulfide: ox to sulfoxide, further to sulfone

• Thiol:  
  \[ R-SH \rightarrow R-S-S-R \rightarrow R-S-S-R \]
Non-microsomal enzymes

• Enzymes in mitokondria
• Enzymes in soubile tissue fractions

R-OH \xleftrightarrow{\text{Alcohol dehydrogenase}} \text{aldehyde / ketone} \xrightarrow{\text{Aldehyde dehydrogenase}} R-\text{CHO}

CH_3OH \xrightarrow{\text{Alcohol dehydrogenase}} \text{CH}_2\text{O} \xrightarrow{\text{Aldehyde dehydrogenase}} \text{HCO}_2\text{H} \quad \text{Tox. effects: Acidosis, Blindness}

Dietary folic acid

\text{Tetrahydrofolate} \xrightarrow{\text{HCO}_2\text{H}} \text{10-Formyl-THF dehydrogenase} \xrightarrow{} \text{CO}_2
Non-microsomal enzymes (Phase 1)

Molybdenum Hydroxylases

- Aldehyde oxidase
- Xantine oxidase
- Xantine dehydrogenase

Cont. Mo in cat. site
Cont FAD and 2 Fe/s clusters
Use H₂O not O₂

Xanthine oxidase
Electron transfer: \( \text{FAD} - \text{Fe}_{2}\text{S}_2\text{I} - \text{Fe}_{2}\text{S}_2\text{II} - \text{Moco} - \text{Substrate} \)

Active form
• **Aldehyde oxidase**

  ![Chemical Reaction Diagram](image)

  - **Aza heterocycle**
  - **Famciclovir**
    - Antiviral (Herpes etc)
    - Pro-drug
  - **Penciclovir**

• **Xantine oxidase**

• **Xantine dehydrogenase**
  (requires NAD⁺)

  ![Chemical Reaction Diagram](image)

  - **Hypoxanthine**
  - **Xanthine**
  - **Uric acid**

  - **Treatment of gout (podagra)**
  - **Allopurinol**
    - **Alloxanthine**
    - Inhib. enzyme
Non-microsomal enzymes (Phase 1)

Oxidative deamination of amines

• Monoamine oxidase (MAO)
• Diamine oxidase (DAO)

\[
R-\text{CH}_2\text{-NH}_2 + O_2 \rightarrow [R-\text{CH}≡\text{NH}] \rightarrow R-\text{CHO} + \text{NH}_4^+
\]

Serotonine:
• Neurotransmitter; temp. control, mood
• Depression: Low serotonin activity
• MAO Inhibitors - Older antidepressants (low selectivity)

Other MAO substrates:

Noradrenaline

Dopamine

Low dopamine conc. ≈ Parkinson

Moklobemid
Aurorix® Moklobemid®

Not MAO substrates (subst at α-C):

(S)-amfetamin
Active transport re-uptake transmitter (not Acetylcholine)

Non-selective monoamine re-uptake inhib.
Tricyclic antidepressants

SSRI (selective serotonin re-uptake inhib.)
“Lykkepiller” Prozac etc (Fontex)
Non-microsomal enzymes (Phase 1)

Oxidative deamination of amines

• Monoamine oxidase (MAO)
• Diamine oxidase (DAO)

Oxidize diaminos, histamine

MAO like enzymes in plants

BAP  CYtokinin (Plant growth hormone)

CK analogs, Anders Bråthe
Non-microsomal enzymes (Phase 1)

Miscellaneous react.

Reductions

\[ R-S-S-R \rightarrow R-SH \]
\[ R-SO-R \rightarrow R-S-R \text{ (i.e. DMSO)} \]
\[ R-CO-R \rightarrow R-CHOH-R \]
\[ R-NO-R \rightarrow R-NH-R \]
\[ R-CH=CH-R \rightarrow R-CH_2-CH_2-R \]
\[ R-OH \rightarrow R-H \]
\[ Ar-OH \rightarrow Ar-H \]

\[ \beta\text{-Oxidation} \]

\[ R-CH_2-CH_2-CO_2H \rightarrow R-CH_2CH_2-CO-S-CoA \rightarrow R-CO_2H \rightarrow R-CO-S-CoA \]

Hydrolysis - Esterases

\[ R-O\cdot R' \rightarrow R\text{COH} \]

Esters as pro-drugs

Acetyl-CoA
Pathways of metabolism

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More hydrophilic drug

Renal excretion

Morphine

Demethylation Phase 1

Conjugation glucuronic acid Phase 2

![Chemical structures](image-url)
Phase 2: Conjugation

Most comp. excreted as conjugates, ionic, hydrophilic groups added, most common glucuronation

• Glucuronic acid conjugation
• Sulfate conjugation
• Conjugation with amino acids
• Acetylation
• Glutathione conjugation
• Methylation
Phase 2: Conjugation

• Glucuronic acid conjugation

Substrates: RXH: Xenobiotic / Phase 1 metabolite

• Alcohols
• Phenols
• Amines
• Sulfides
• Carboxylic acids
• 1,3-Dicarbonyls

Entero-hepatic recycling
Important for many hormones etc
UDP-Glucuronate

Relatively labile

Hydrolysis

Acyl migr.

Altered protein (hapten)
Unwanted immune response / allergic react.

ex. NSAIDs

Ar-NH₂ → Ar-NH-OH → Ar-NH-OH-Glu → Ar-NH-OH → Ar-N₂

Nitrene (c.f. carbene)
Bladder cancer
Phase 2: Conjugation

• Sulfate conjugation: Phenols, (alcohols, N-compounds)

\[
\text{ATP} \rightarrow \underset{\text{O-S-O-P}}{\text{PAPS}} \rightarrow \text{Sulfate conjugate}
\]

• Conjugation with amino acids (Most often Gly): Carboxylic acids

\[
\text{R-CO}_2\text{H} \rightarrow \text{R-S-CoA} \rightarrow \text{R-NH-CO}_2\text{H}
\]

No tox. conjugates known
Phase 2: Conjugation

• Acetylation: N-compounds

\[ \text{Acetyl-CoA} \rightarrow \text{Acetyl-N-compound} \]

• Glutathione conjugation: Electrophilic species

\[ \text{Glutathione conjugate} \rightarrow \text{Mercapturic acid der.} \]

\[ \text{R-X} + \text{HS-} \rightarrow \text{R-S-} \]

• Alkylhalides
• Epoxides
• Michael acceptors etc

May otherwise alkylate biomolecules

Paracetamol

Liver damage

R-X + S-R → R-S-R

:Nu (i.e. DNA)
Phase 2: Conjugation

- Methylation (O and N-compd)
- Prod. may be more lipophilic
- React. mainly aimed at converting endogenic compounds

O-Metylation by COMT (catecol O-methyl transferase)

SAM may also methylate N-comp.
Noradrenaline (Norepinephrine) → MAO → Aldehyde Dehydrogenase → Ephedrine Adrenerg agonist

COMT → MAO → Aldehyde Dehydrogenase → Ephedrine Adrenerg agonist