Biotransformacija zdravil

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Biotransformacija zdravil

- Živali – kompleksni sistemi za odstranjevanje telesu tujih snovi (ksenobiotiki) – strupene rastline
- Zdravila – poseben primer ksenobiotikov
- Distribucija – lipofilne snovi – kopičenje v tkivih
- Hidrofilne snovi se laže in hitreje izločajo.
- Pomen stereoselektivnosti (različne poti za razl. izomere)
- Biotransformacija zdravil – 2 fazi: I in II (običajno si sledita).

Rezultati biotransformacije

- Nastanek neaktivnega metabolita (nepolarna snov ⇒ polarna snov)
- Iz zdravila nastane aktiven metabolit – primeri:

Neaktivna oblika (predzdravilo)	Aktivno zdravilo	Aktivni metabolit	Toksični metabolit
Azathioprine	→	Merkaptopurin	
Kortizon	→	Hidrokortizon	
Prednizon	→	Prednizolon	
Enalapril	→	Enalaprilat	
Zidovudin	→	Zidovudin trisfosfat	
Ciklofosfamid	→	Fosforamidni derivat	akrolein
	Diazepam	Nordiazepam → Oxazepam	
	Morfin	Morfine 6-glukuronid	
	Halotan	→	Trifluoroocetna k.
	Paracetamol	→	N-Acetil-p-benzokvinonimin

Fazi biotransformacije

- Reakcije I faze so katabolne (npr. oksidacija, redukcija ali hidroliza) – produkti često kemično bolj reaktivni (lahko tudi bolj toksični)
- Reakcije II faze so sintetske (anabolne) – konjugacija → manj aktivni produkti (praviloma).
- I faza poveča reaktivnost molekule – priprava na konjugacijo.
- Obe fazi povečata hidrofilnost molekule.
- Mesta biotransformacije – jetra (gladki ER – mikrosomi), GIT (MAO), plazma (holinesteraza), pljuča

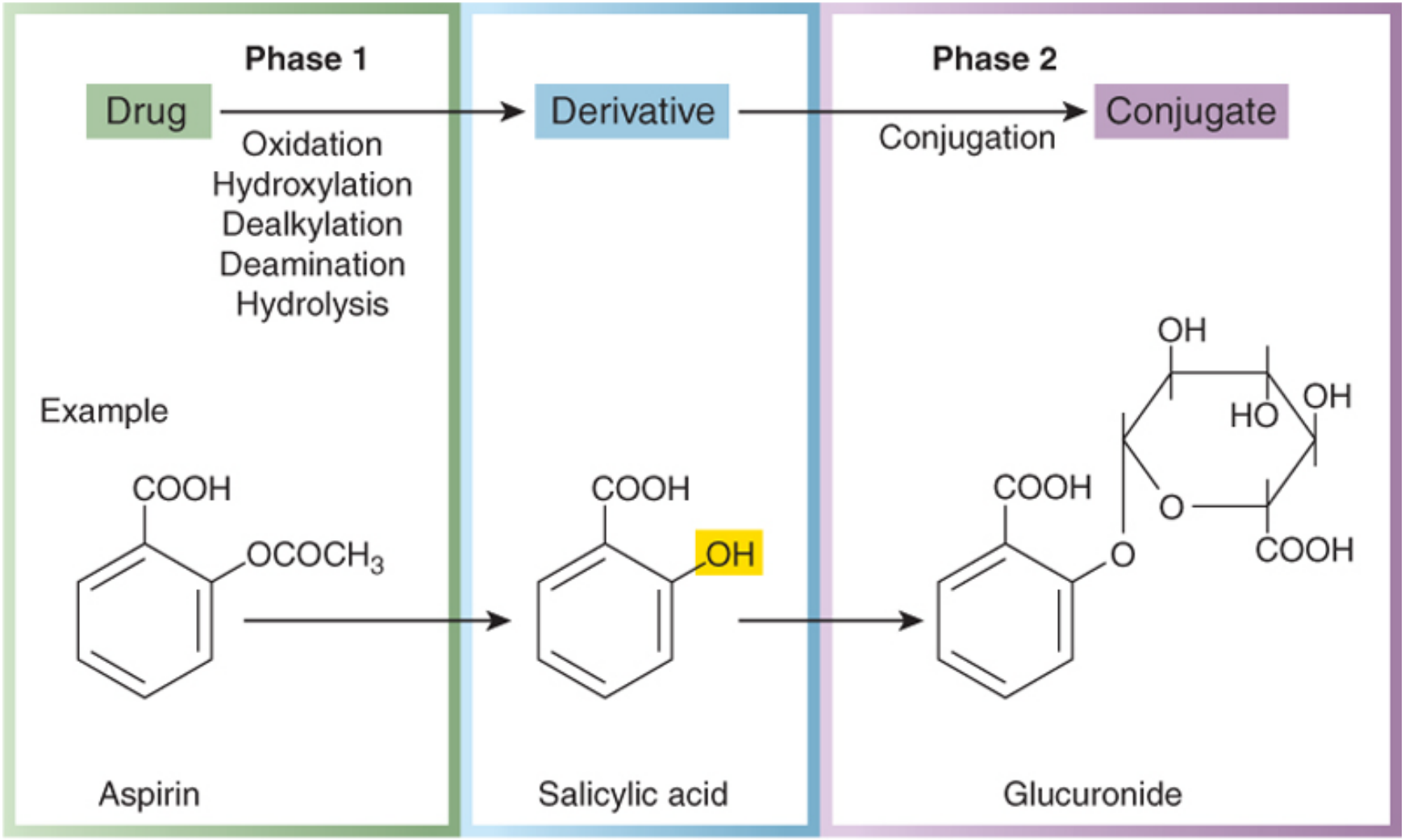


Figure 8-1 The two phases of drug metabolism.

Table 3–2 Major Reactions Involved in Drug Metabolism

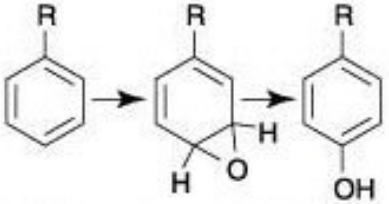
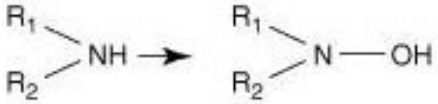
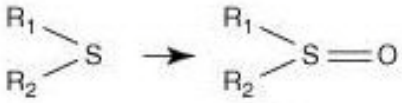
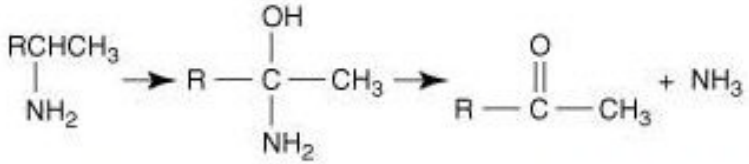
REACTION		EXAMPLES
I. Oxidative reactions		
N-Dealkylation	$RNHCH_3 \rightarrow RNH_2 + CH_2O$	Imipramine, diazepam, codeine, erythromycin, morphine, tamoxifen, theophylline, caffeine
O-Dealkylation	$ROCH_3 \rightarrow ROH + CH_2O$	Codeine, indomethacin, dextromethorphan
Aliphatic hydroxylation	$RCH_2CH_3 \rightarrow RCHOHCH_3$	Tolbutamide, ibuprofen, phenobarbital, meprobamate, cyclosporine, midazolam
Aromatic hydroxylation		Phenytoin, phenobarbital, propanolol, ethinyl estradiol, amphetamine, warfarin
N-Oxidation	$RNH_2 \rightarrow RNHOH$ 	Chlorpheniramine, dapsone, meperidine
S-Oxidation		Cimetidine, chlorpromazine, thioridazine, omeprazole
Deamination		Diazepam, amphetamine

Table 3–2 Major Reactions Involved in Drug Metabolism

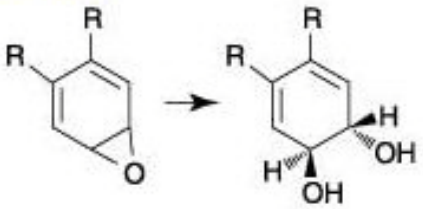
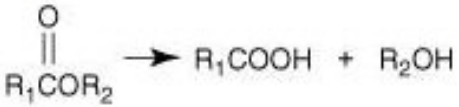
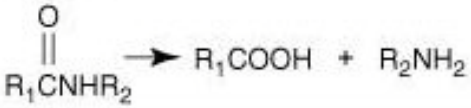
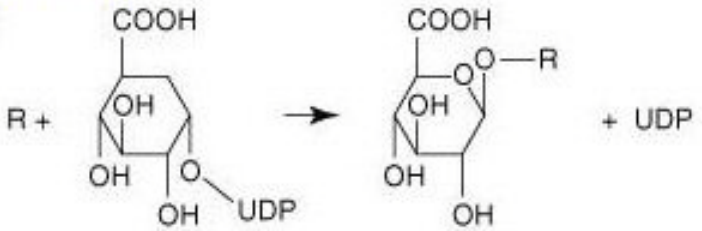
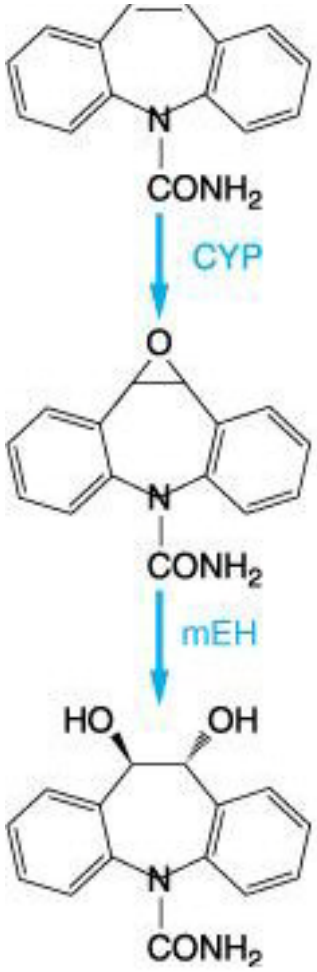
REACTION	EXAMPLES
II. *Hydrolysis reactions*	
	Carbamazepine
	Procaine, aspirin, clofibrate, meperidine, enalapril, cocaine
	Lidocaine, procainamide, indomethacin

Table 3–2 Major Reactions Involved in Drug Metabolism

REACTION		EXAMPLES
III. *Conjugation reactions*		
Glucuronidation	 UDP-glucuronic acid	Acetaminophen, morphine, oxazepam, lorazepam
Sulfation	$PAPS + ROH \rightarrow R-O-SO_2-OH + PAP$ 3'-phosphoadenosine-5' phosphosulfate → 3'-phosphoadenosine-5'-phosphate	Acetaminophen, steroids, methyldopa
Acetylation	$CoAS-CO-CH_3 + RNH_2 \rightarrow RNH-CO-CH_3 + CoA-SH$	Sulfonamides, isoniazid, dapsone, clonazepam (*see* Table 3–3)
Methylation	RO-, RS-, RN- + AdoMet → $RO-CH_3$ + AdoHomCys	L-Dopa, methyldopa, mercaptopurine, captopril
Glutathione conjugation	GSH + R → R-GSH	Adriamycin, fosfomycin, busulfan

Metabolism of carbamazepine by CYP and microsomal epoxide hydrolase (mEH).

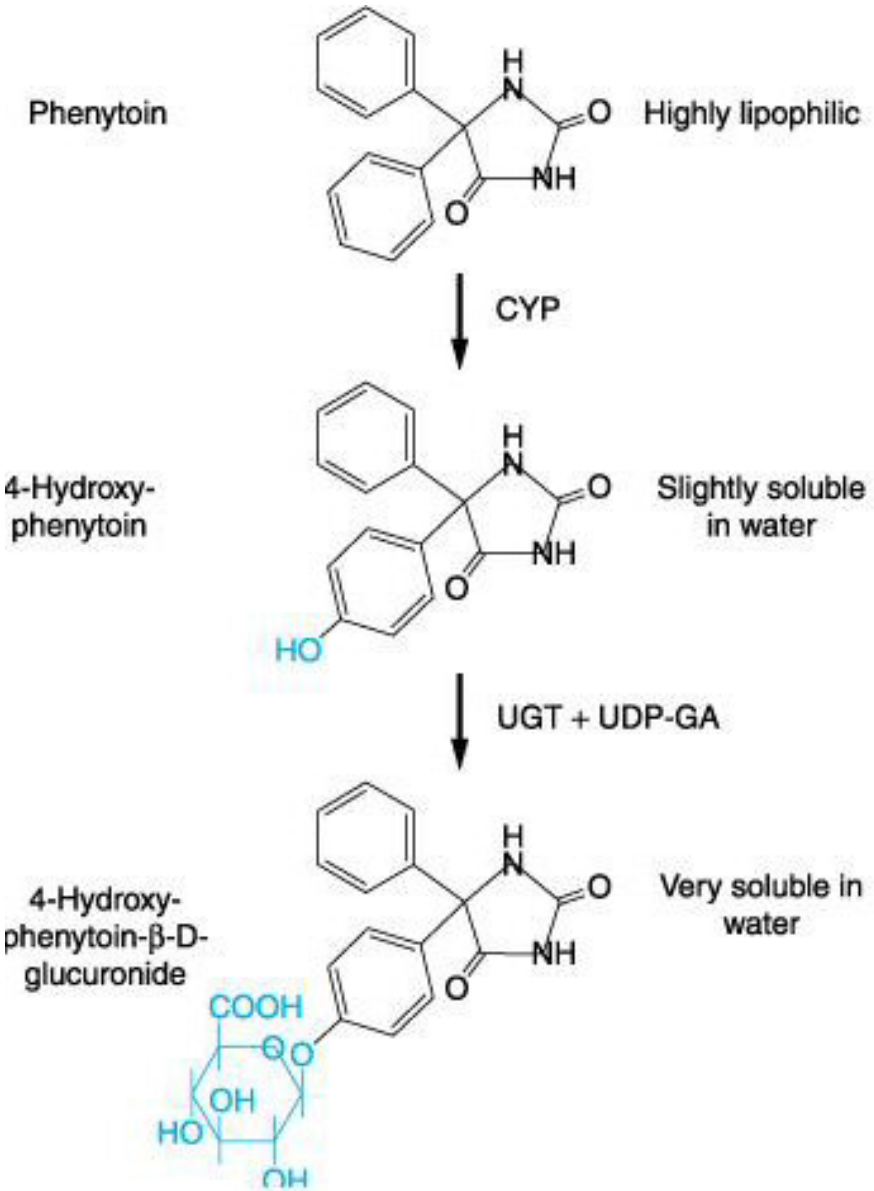
Carbamazepine is oxidized to the pharmacologically-active metabolite carbamazepine-10,11-epoxide by CYP. The epoxide is converted to a trans-dihydrodiol by mEH. This metabolite is biologically inactive and can be conjugated by phase 2 enzymes.



Source: Brunton LL, Lazo JS, Parker KL: *Goodman & Gilman's The Pharmacological Basis of Therapeutics*, 11th Edition: http://www.accessmedicine.com

Metabolism of phenytoin by phase 1 cytochrome P450 (CYP) and phase 2 uridine diphosphate-glucuronosyltransferase (UGT).

CYP facilitates 4-hydroxylation of phenytoin to yield 5-(-4-hydroxyphenyl)-5-phenylhydantoin (HPPH). The hydroxy group serves as a substrate for UGT that conjugates a molecule of glucuronic acid using UDP-glucuronic acid (UDP-GA) as a cofactor. This converts a very hydrophobic molecule to a larger hydrophilic derivative that is eliminated via the bile.

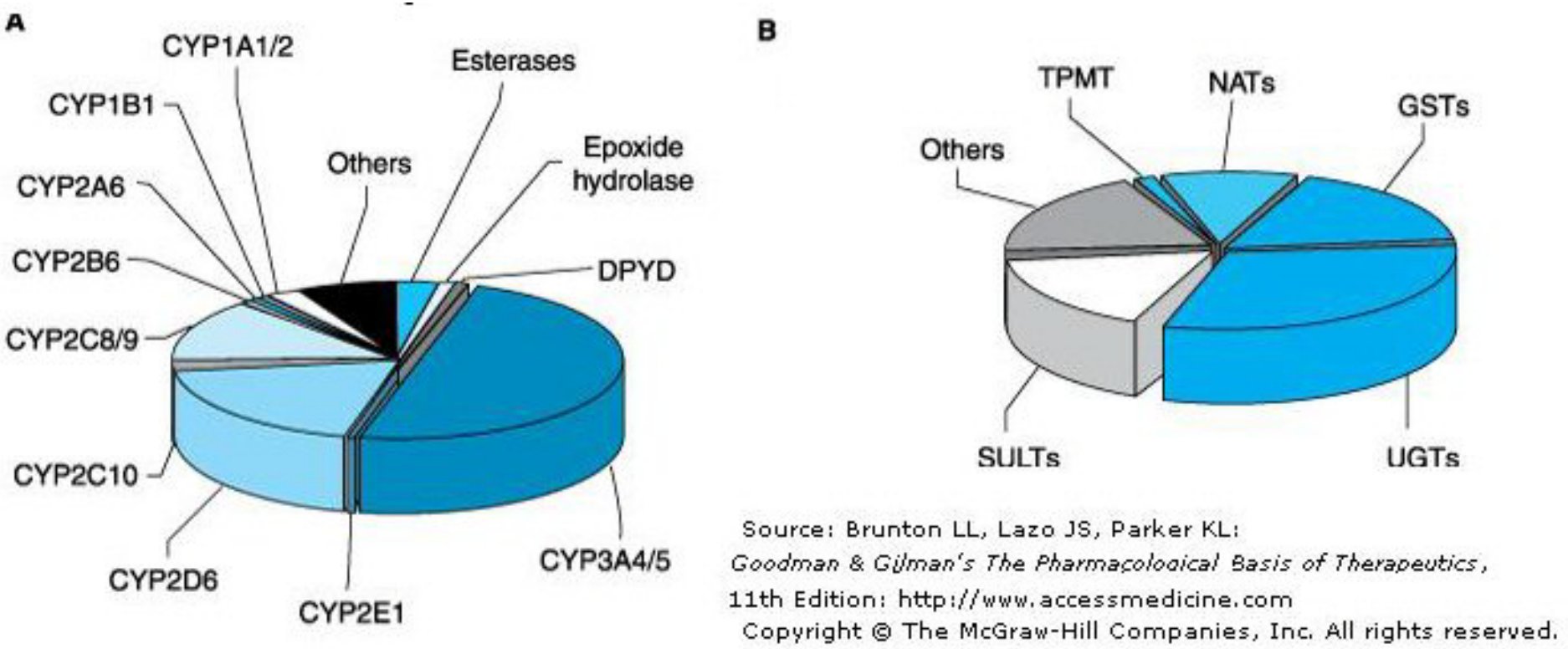


Source: Brunton LL, Lazo JS, Parker KL: *Goodman & Gilman's The Pharmacological Basis of Therapeutics*, 11th Edition: http://www.accessmedicine.com

Table 3–1 Xenobiotic Metabolizing Enzymes

ENZYMES	REACTIONS
Phase 1 "oxygenases"	
Cytochrome P450s (P450 or CYP)	C and O oxidation, dealkylation, others
Flavin-containing monooxygenases (FMO)	N, S, and P oxidation
Epoxide hydrolases (mEH, sEH)	Hydrolysis of epoxides
Phase 2 "transferases"	
Sulfotransferases (SULT)	Addition of sulfate
UDP-glucuronosyltransferases (UGT)	Addition of glucuronic acid
Glutathione-S-transferases (GST)	Addition of glutathione
N-acetyltransferases (NAT)	Addition of acetyl group
Methyltransferases (MT)	Addition of methyl group
Other enzymes	
Alcohol dehydrogenases	Reduction of alcohols
Aldehyde dehydrogenases	Reduction of aldehydes
NADPH-quinone oxidoreductase (NQO)	Reduction of quinones

mEH and sEH are microsomal and soluble epoxide hydrolase. UDP, uridine diphosphate; NADPH, reduced nicotinamide adenine dinucleotide phosphate.



The fraction of clinically used drugs metabolized by the major phase 1 and phase 2 enzymes.

The relative size of each pie section represents the estimated percentage of drugs metabolized by the major phase 1 (panel A) and phase 2 (panel B) enzymes, based on studies in the literature. In some cases, more than a single enzyme is responsible for metabolism of a single drug. CYP, cytochrome P450; DPYD, dihydropyrimidine dehydrogenase; GST, glutathione-S-transferase; NAT, N-acetyltransferase; SULT, sulfotransferase, TPMT, thiopurine methyltransferase; UGT, UDP-glucuronosyltransferase.

Citohrom P 450 – CYP

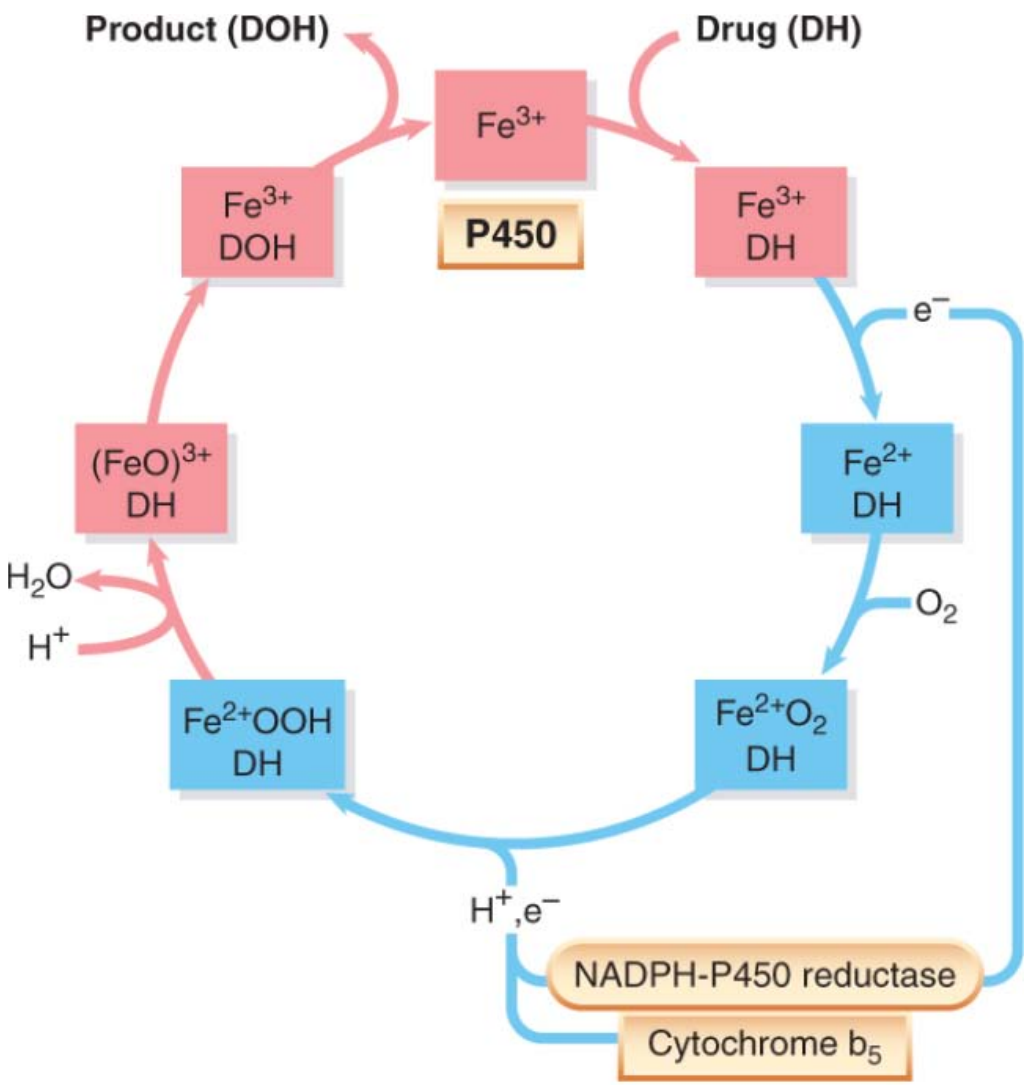
- Številni izoencimi (superfamily, človek > 50):
  - nomenklatura CYP1(2,3 – družina)A(B - E – poddružina)1(2 - 19 – oblika)
  - lokalizacija: predvsem jetra, druga tkiva (OŽ, ….)
- Udeleženost pri sintezi endogenih sestavin organizma (steroidi, žolčne kisline, eikozanoidi …) – selektivnost za substrat
- Udeleženost pri biotransformaciji ksenobiotikov – majhna hitrost reakcij – majhna substratna selektivnost → možnost kompeticije substratov
- Vir interakcij med zdravili
- Velika variabilnost porazdelitve izoencimov – polimorfizem genov → individualne razlike
- Možnost indukcije in inhibicije

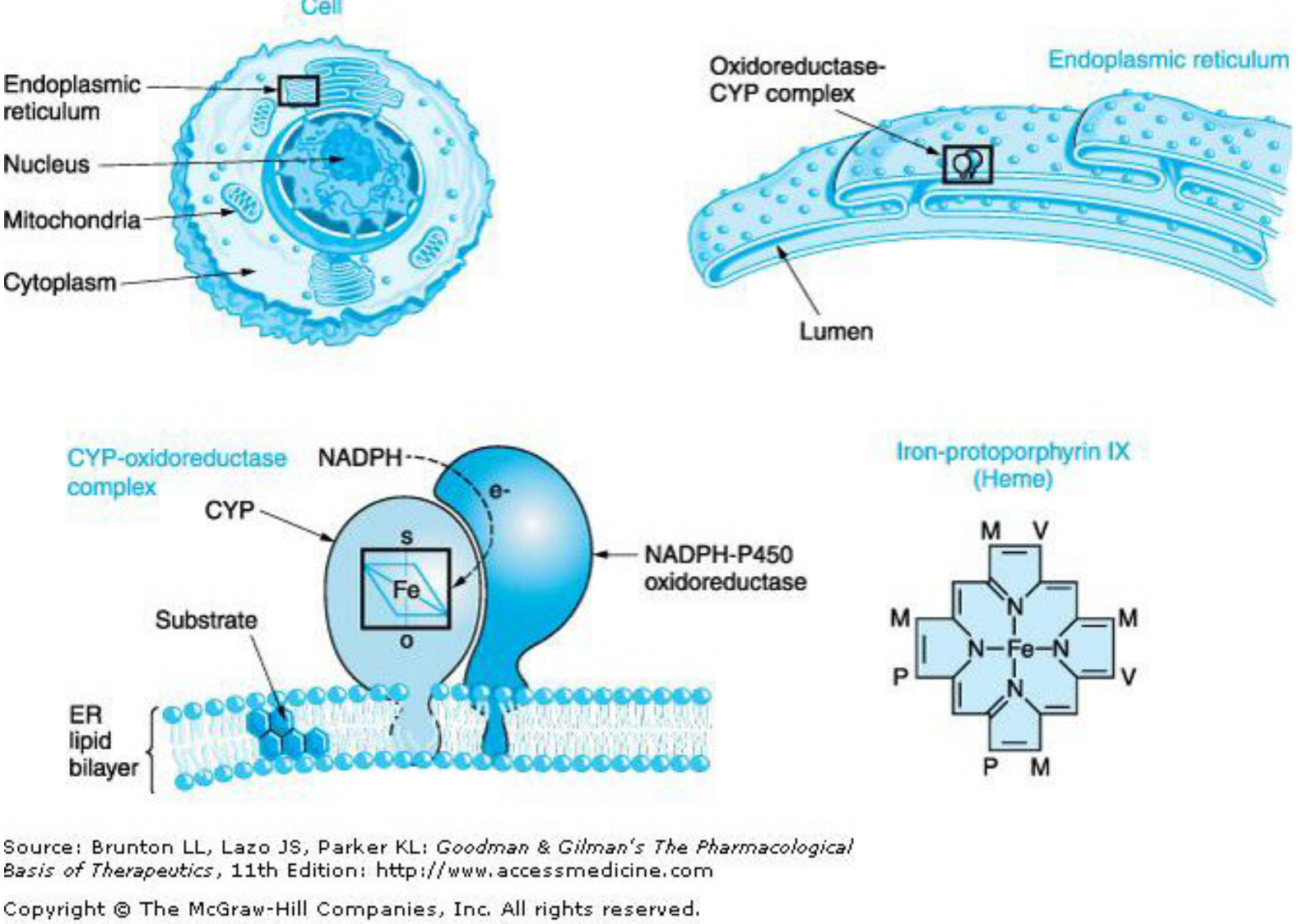
Examples of drugs that are substrates for P450 isoenzymes

Isoenzyme P450	Drug(s)
CYP1A2	Caffeine, paracetamol (→NAPQI; see p. XXX), tacrine, theophylline
CYP2B6	Cyclophosphamide, methadone
CYP2C8	Paclitaxel, repaglinide
CYP2C19	Omeprazole, phenytoin
CYP2C9	Ibuprofen, tolbutamide, warfarin
CYP2D6	Codeine, debrisoquine, ***S***-metoprolol
CYP2E1	Alcohol, paracetamol
CYP3A4, 5, 7	Ciclosporin, nifedipine, indinavir, simvastatin

(Adapted from http://medicine.iupui.edu/flockhart/table.htm.)

Figure 8-2 The monooxygenase P450 cycle. P450 containing ferric iron (Fe3+) combines with a molecule of drug ('DH'); receives an electron from NADPH-P450 reductase, which reduces the iron to Fe2+; combines with molecular oxygen, a proton and a second electron (either from NADPH-P450 reductase or from cytochrome b5) to form an Fe2+OOH-DH complex. This combines with another proton to yield water and a ferric oxene (FeO)3+-DH complex. (FeO)3+ extracts a hydrogen atom from DH, with the formation of a pair of short-lived free radicals (see text), liberation from the complex of oxidised drug ('DOH'), and regeneration of P450 enzyme.





Source: Brunton LL, Lazo JS, Parker KL: *Goodman & Gilman's The Pharmacological Basis of Therapeutics*, 11th Edition: http://www.accessmedicine.com

Location of CYPs in the cell.

The figure shows increasingly microscopic levels of detail, sequentially expanding the areas within the black boxes. CYPs are embedded in the phospholipid bilayer of the endoplasmic reticulum (ER). Most of the enzyme is located on the cytosolic surface of the ER. A second enzyme, NADPH-cytochrome P450 oxidoreductase, transfers electrons to the CYP where it can, in the presence of O2, oxidize xenobiotic substrates, many of which are hydrophobic and dissolved in the ER. A single NADPH-CYP oxidoreductase species transfers electrons to all CYP isoforms in the ER. Each CYP contains a molecule of iron-protoporphyrin IX that functions to bind and activate O2. Substituents on the porphyrin ring are methyl (M), propionyl (P), and vinyl (V) groups.

Monooksigenaze , ki vsebujejo flavin (FMO)

- FMO – izražene v jetrih, vezava na ER
- 6 družin FMO, največ FMO3 (jetra). FMO3 – metabolizem: nikotin, H2-antagonisti (cimetidin and ranitidin), antipsihotiki (klozapin), antiemetiki (itoprid)
- FMO – manjši prispevek k metabolizmu zdravil – benigni metaboliti.
- FMO niso podvrženi indukciji, inhibicija redka → za razliko od CYP, FMO niso udeleženi pri interakcijah med zdravili.

Hidrolitični encimi

- Epoksidna hidrolaza – hidroliza epoksidov, nastalih pod vplivom CYP.
- Karboksilesteraze – superdružina encimov – kataliza hidrolize snovi z estersko in/ali amidno vezjo (lokalni anestetiki).

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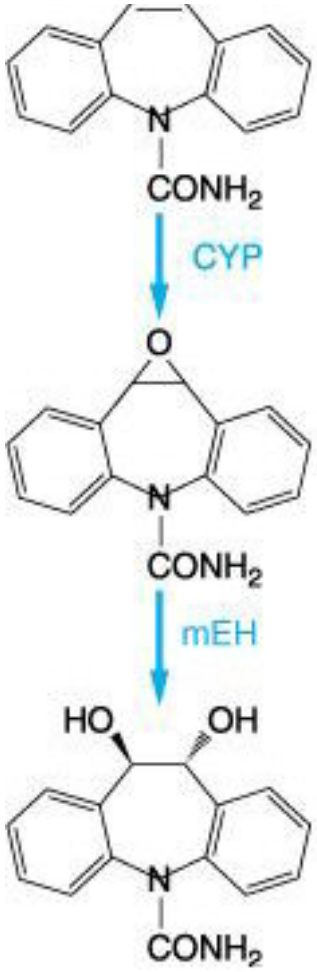

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Reakcije II faze - konjugacija

- Pogosto – nadaljevanje I faze
- Konjugacija:
  - z glukuronsko kislino (UGT, UDP-glukuronosiltransferaza)
  - z glutationom (GST, glutation-S-transferaza)
  - z žvepleno kislino (SULT, sulfotransferaza)
  - z ocetno kislino – acetiliranje (NAT, N-acetiltransferaza) genski polimorfizem (NAT1, NAT2)
  - z metilno skupino – metiliranje (TPMT, tiopurin metiltransferaza)
- Produkti bolj vodotopni → lažje izločanje

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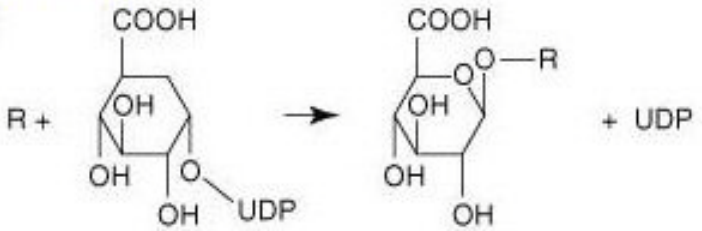
Glucuronidation	 UDP-glucuronic acid	Acetaminophen, morphine, oxazepam, lorazepam
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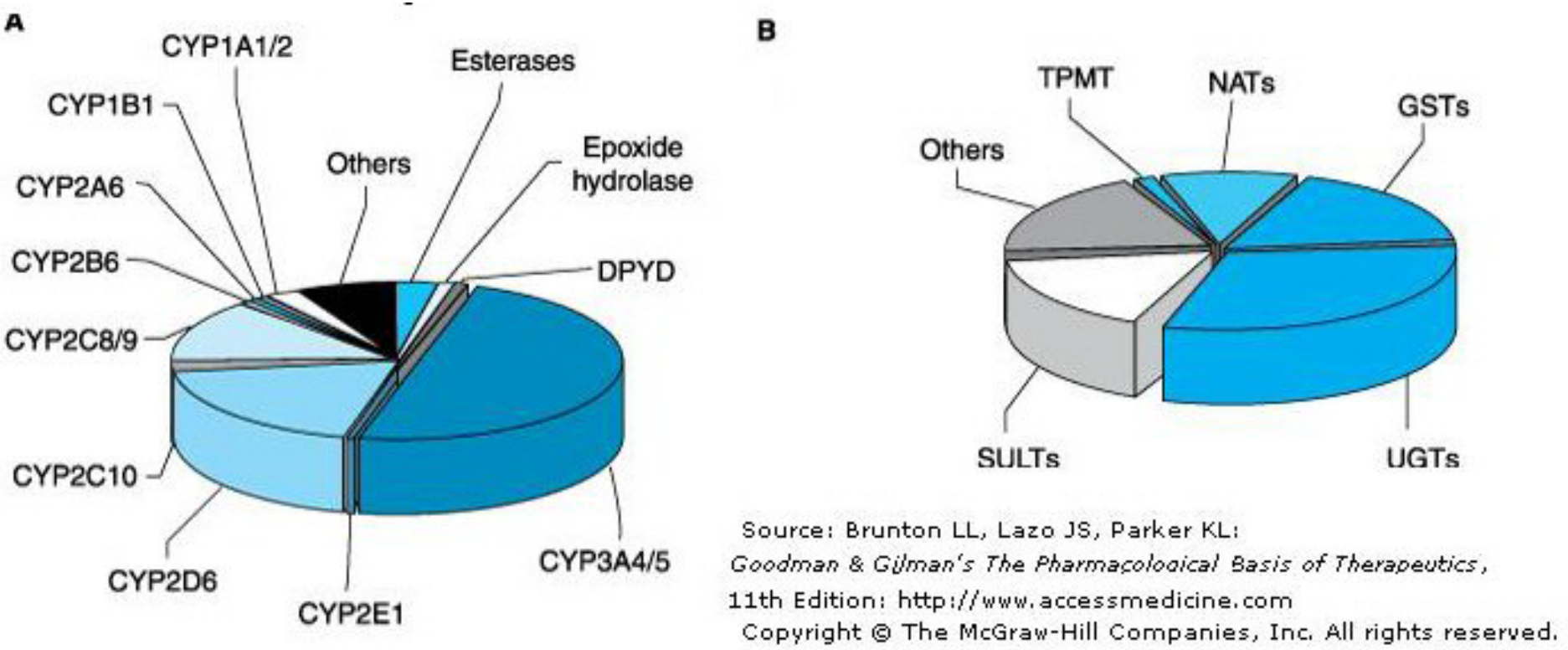
Acetylation	$CoAS-CO-CH_3 + RNH_2 \rightarrow RNH-CO-CH_3 + CoA-SH$	Sulfonamides, isoniazid, dapsone, clonazepam (*see* Table 3–3)
Methylation	RO-, RS-, RN- + AdoMet → $RO-CH_3$ + AdoHomCys	L-Dopa, methyldopa, mercaptopurine, captopril
Glutathione conjugation	GSH + R → R-GSH	Adriamycin, fosfomycin, busulfan



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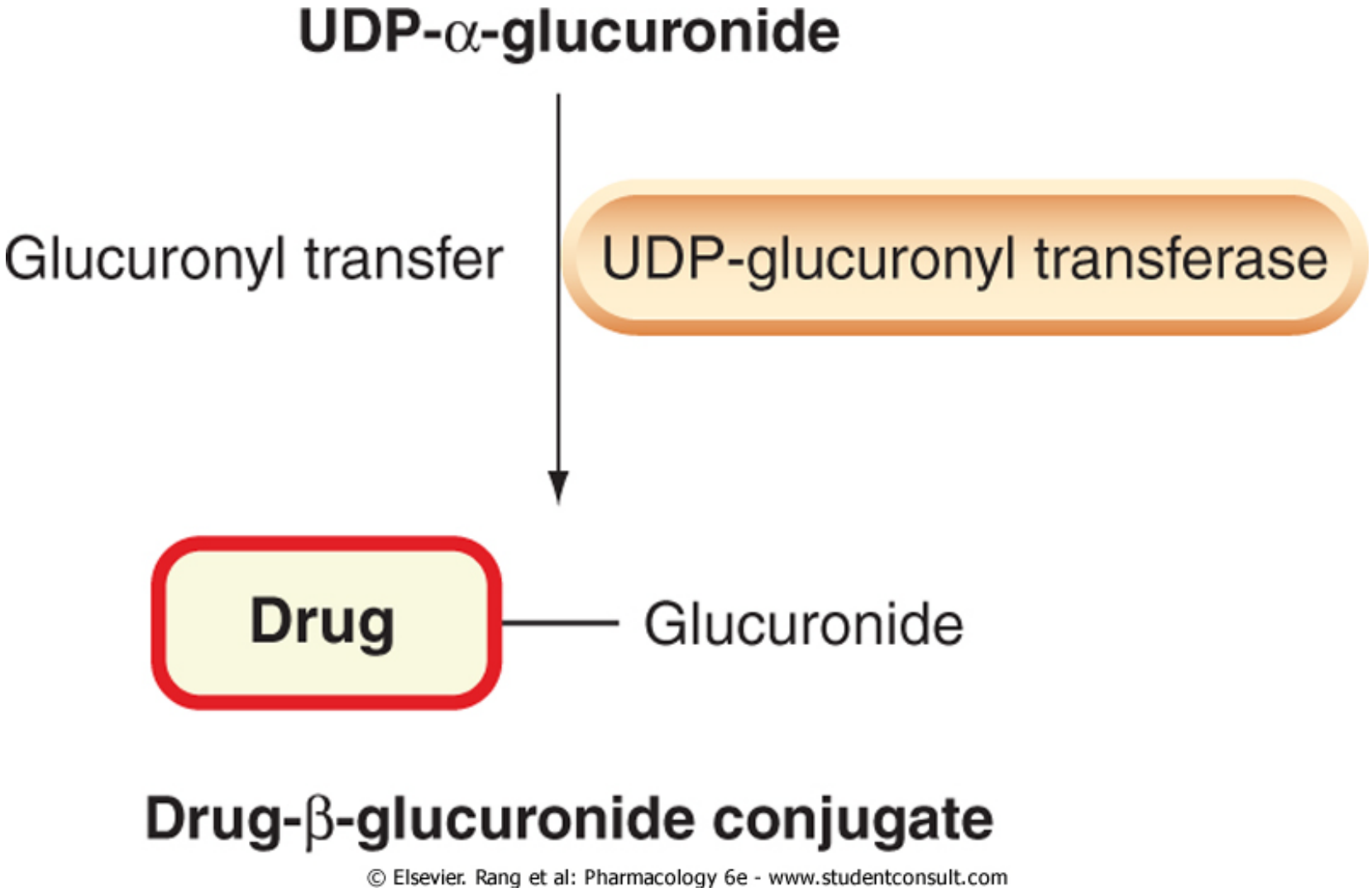
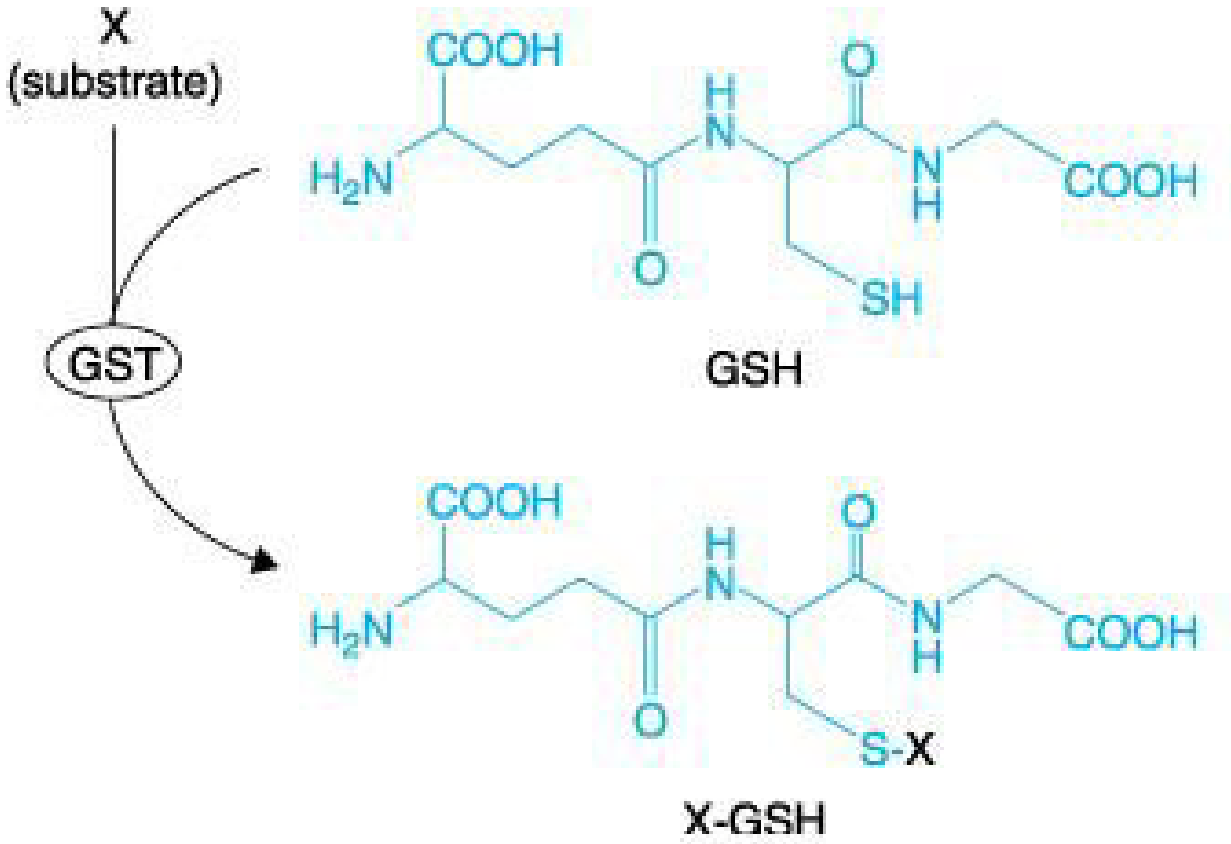
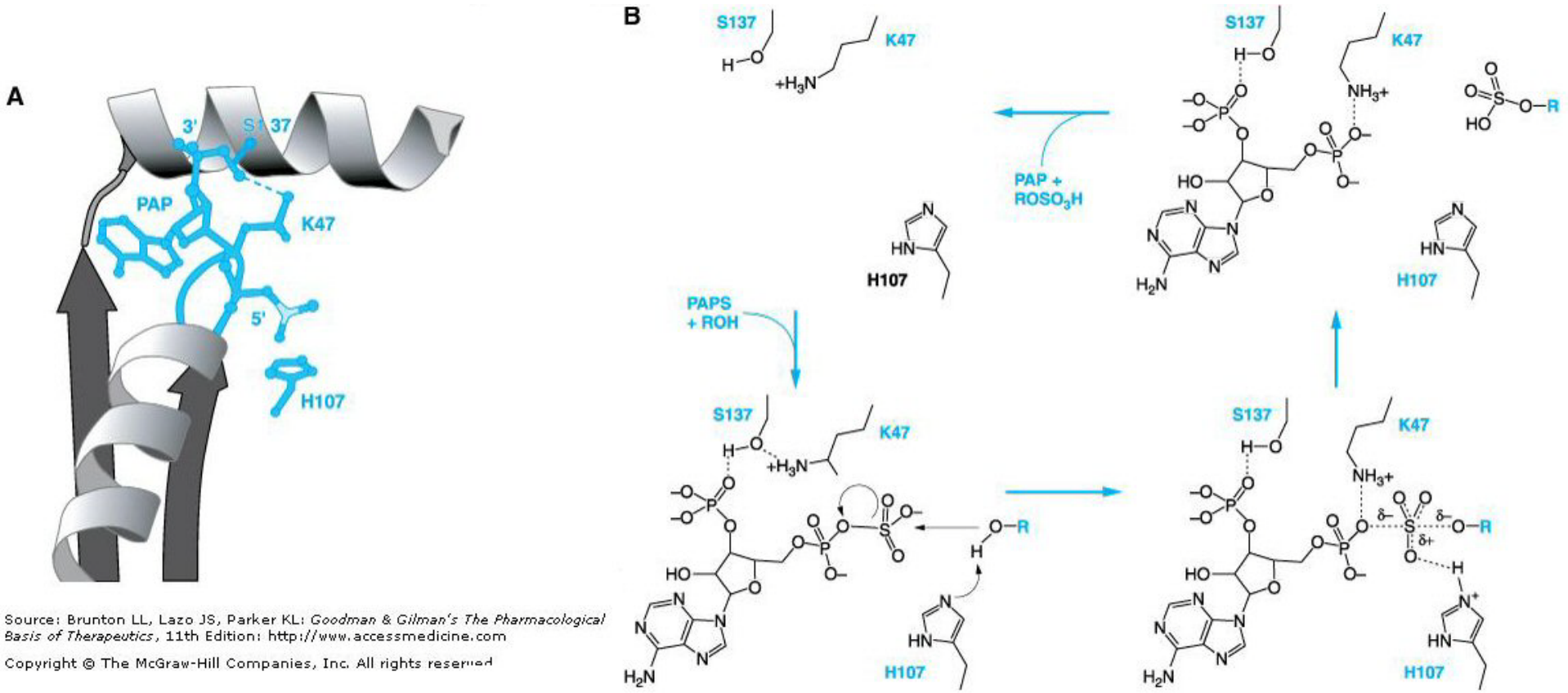


Figure 8-3 The glucuronide conjugation reaction.



Glutathione as a co-substrate in the conjugation of a drug or xenobiotic (X) by glutathione-S-transferase (GST).



The proposed reaction mechanism of sulfuryl transfer catalyzed by the sulfotransferases (SULTs).

A. Shown in this figure are the conserved strand-loop-helix and strand-turn-helix structure of the catalytic core of all SULTs where PAPS and xenobiotics bind. Shown is the hydrogen bonding interaction of PAPS with Lys47 and Ser137 with His107 which complexes with substrate (xenobiotic).

B. The proposed reaction mechanism shows the transfer of the sulfuryl group from PAPS to the OH-group on the substrate and the interactions of the conserved SULT residues in this reaction. (For additional information seeNegishi et al., 2001.)

Table 3–3 Indications and Unwanted Side Effects of Drugs Metabolized by N-Acetyltransferases

DRUG	INDICATION	MAJOR SIDE EFFECTS
Acebutolol	Arrhythmias, hypertension	Drowsiness, weakness, insomnia
Amantadine	Influenza A, parkinsonism	Appetite loss, dizziness, headache, nightmares
Aminobenzoic acid	Skin disorders, sunscreens	Stomach upset, contact sensitization
Aminoglutethimide	Adrenal cortex carcinoma, breast cancer	Clumsiness, nausea, dizziness, agranulocytosis
Aminosalicylic acid	Ulcerative colitis	Allergic fever, itching, leukopenia
Amonafide	Prostate cancer	Myelosuppression
Amrinone	Advanced heart failure	Thrombocytopenia, arrhythmias
Benzocaine	Local anesthesia	Dermatitis, itching, rash, methemoglobinemia
Caffeine	Neonatal respiratory distress syndrome	Dizziness, insomnia, tachycardia
Clonazepam	Epilepsy	Ataxia, dizziness, slurred speech
Dapsone	Dermatitis, leprosy, AIDS-related complex	Nausea, vomiting, hyperexcitability, methemoglobinemia, dermatitis
Dipyrone, metamizole	Analgesic	Agranulocytosis
Hydralazine	Hypertension	Hypotension, tachycardia, flush, headache
Isoniazid	Tuberculosis	Peripheral neuritis, hepatotoxicity
Nitrazepam	Insomnia	Dizziness, somnolence
Phenelzine	Depression	CNS excitation, insomnia, orthostatic hypotension, hepatotoxicity
Procainamide	Ventricular tachyarrhythmia	Hypotension, systemic lupus erythematosus
Sulfonamides	Antibacterial agents	Hypersensitivity, hemolytic anemia, fever, lupus-like syndromes

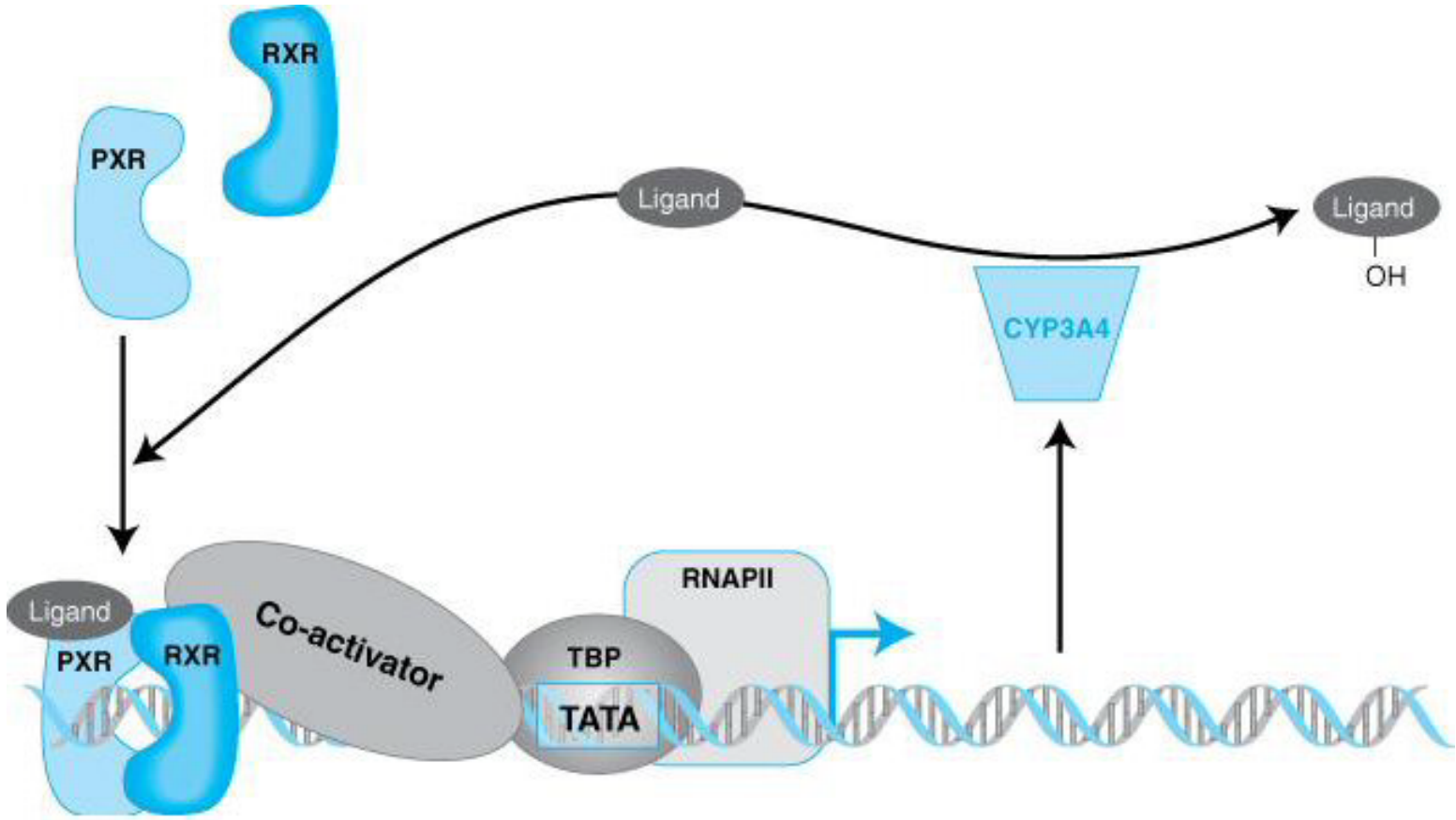
For details, *see* Meisel, 2002.

Faktorji, ki vplivajo na metabolizem zdravil

- Inhibicija metabolnih encimov (nekatera zdravila, sok grenivke → sprememba učinkov antihipertenzivov, imunosupresivov, antidepresivov, antihistaminikov, statinov. Naringin in furanokumarini – inhib. CYP3A4).
- Genski polimorfizem – individualne razlike (NAT)
- Indukcija encimov (CYP):
  - Avtoindukcija
  - Heteroindukcija

Table 3–4 Nuclear Receptors That Induce Drug Metabolism

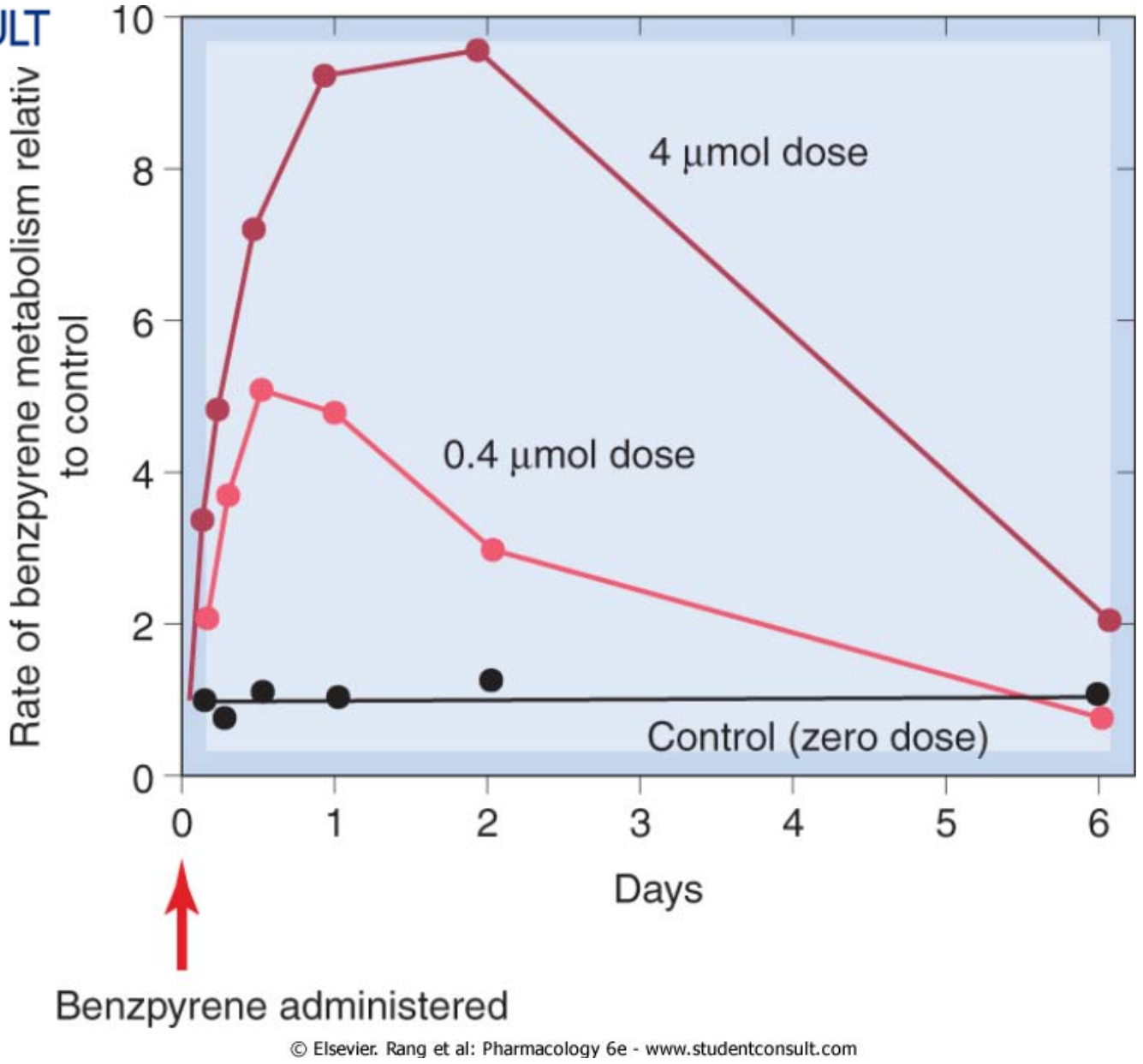
RECEPTOR	LIGANDS
Aryl hydrocarbon receptor (AHR)	Omeprazole
Constitutive androstane receptor (CAR)	Phenobarbital
Pregnane X receptor (PXR)	Rifampin
Farnesoid X receptor (FXR)	Bile acids
Vitamin D receptor	Vitamin D
Peroxisome proliferator activated receptor (PPAR)	Fibrates
Retinoic acid receptor (RAR)	*all-trans*-Retinoic acid
Retinoid X receptor (RXR)	9-*cis*-Retinoic acid



Induction of drug metabolism by nuclear receptor-mediated signal transduction.

When a drug such as atorvastatin (Ligand) enters the cell, it can bind to a nuclear receptor such as the pregnane X receptor (PXR). PXR then forms a complex with the retinoid X receptor (RXR), binds to DNA upstream of target genes, recruits coactivator (which binds to the TATA box binding protein, TBP), and activates transcription. Among PXR target genes are CYP3A4, which can metabolize the atorvastatin and decrease its cellular concentration. Thus, atorvastatin induces its own metabolism. Atorvastatin undergoes both ortho and para hydroxylation. (See Handschin and Meyer, 2003.)

Figure 8-4 Stimulation of hepatic metabolism of benzpyrene. Young rats were given benzpyrene (intraperitoneally) in the doses shown, and the benzpyrene-metabolising activity of liver homogenates was measured at times up to 6 days. (From Conney A H et al. 1957 J Biol Chem 228: 753.)



Metabolizem 1. prehoda

- Intenzivna razgradnja ob prvem prehodu skozi jetra
- Manjša biološka uporabnost zdravila
- Manjše koncentracije zdravila v plazmi
- Primeri:

Aspirin	Metoprolol
Gliceril trinitrat	Morfin
Izosorbid dinitrat	Propranolol
Levodopa	Salbutamol
Lidokain	Verapamil