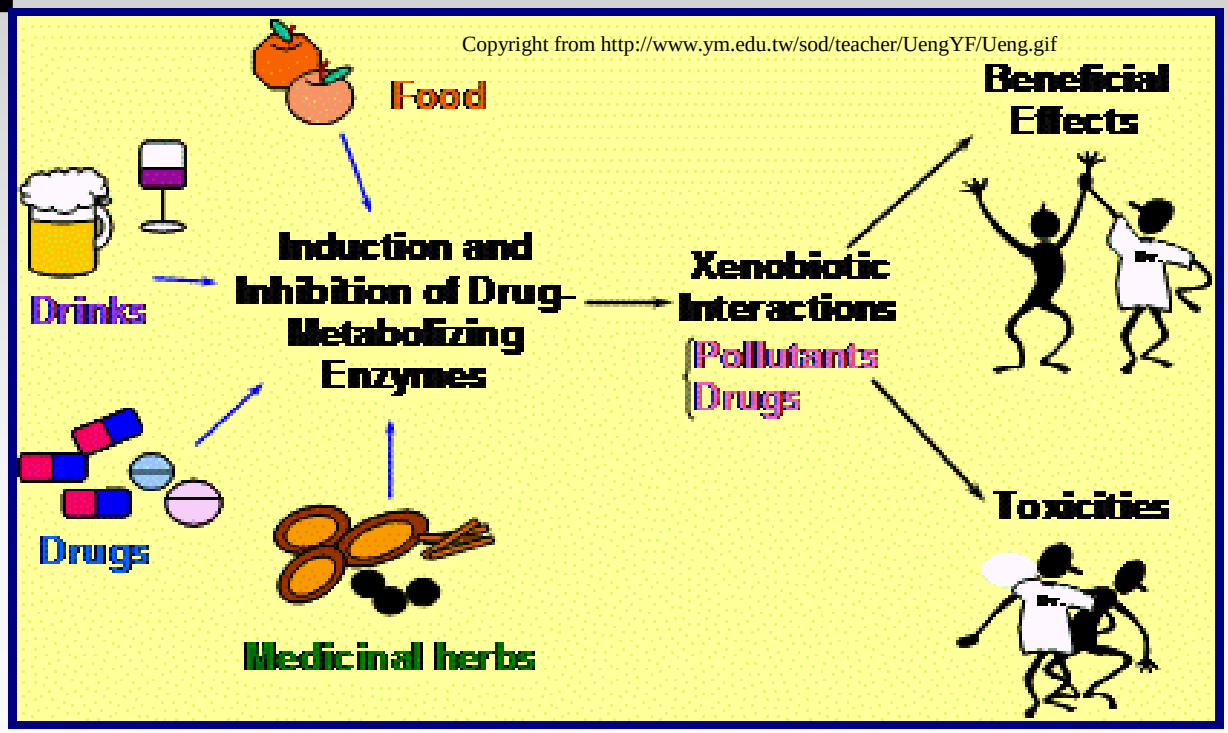
เมแทบอลิซึมจำเพาะ

(Specific Metabolism)



โดย อ. เอกวิทย์ ตรีเนตร ภาควิชาเคมี คณะวิทยาศาสตร์

วิชา คม 524

เมแทบอลิซึมจำเพาะ
(Principle of Metabolism)

1. ADME pathways:
Absorption, Distribution, Metabolism, and Excretion

2. Xenobiotic metabolism

ADME

Absorption, Distribution, Metabolism, and Excretion

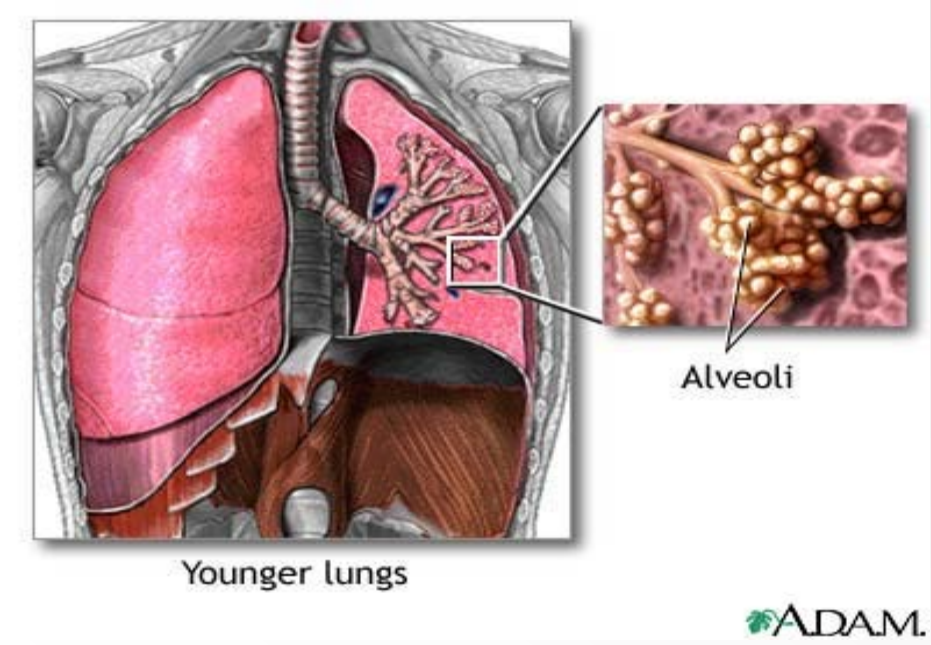
- pharmacokinetics and pharmacology

Absorption:
ability of a chemical to enter the blood (blood is in equilibrium with tissues)

- Inhalation
- Ingestion
- Dermal--absorption through epidermis

Absorption: Inhalation

- Gases into the blood stream
- via the alveoli



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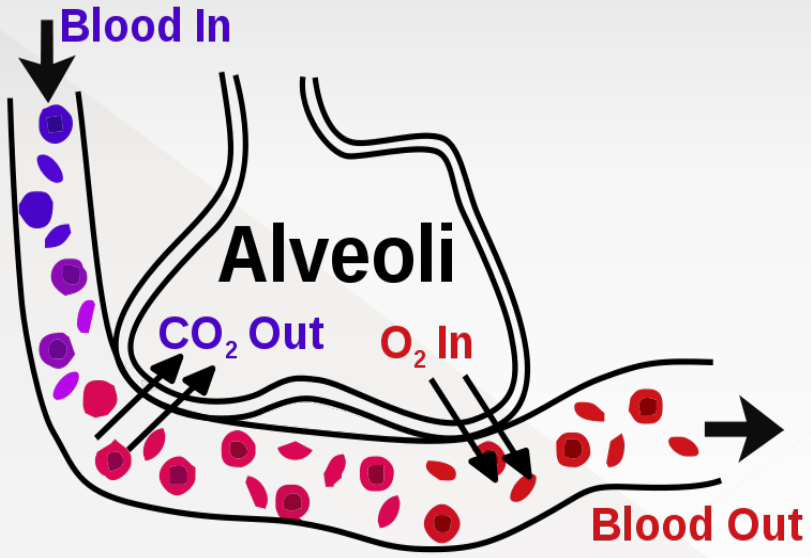

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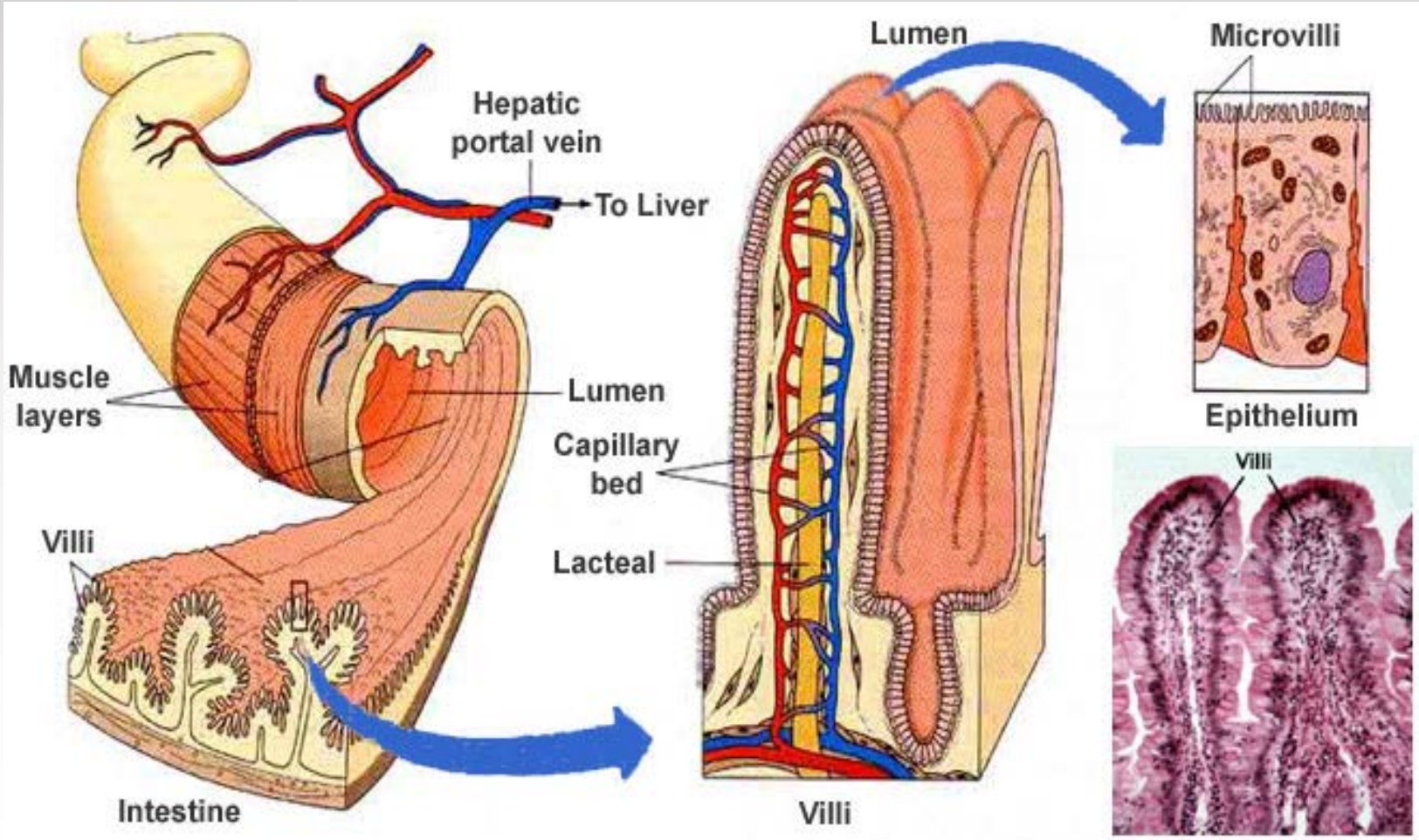
Absorption:Ingestion

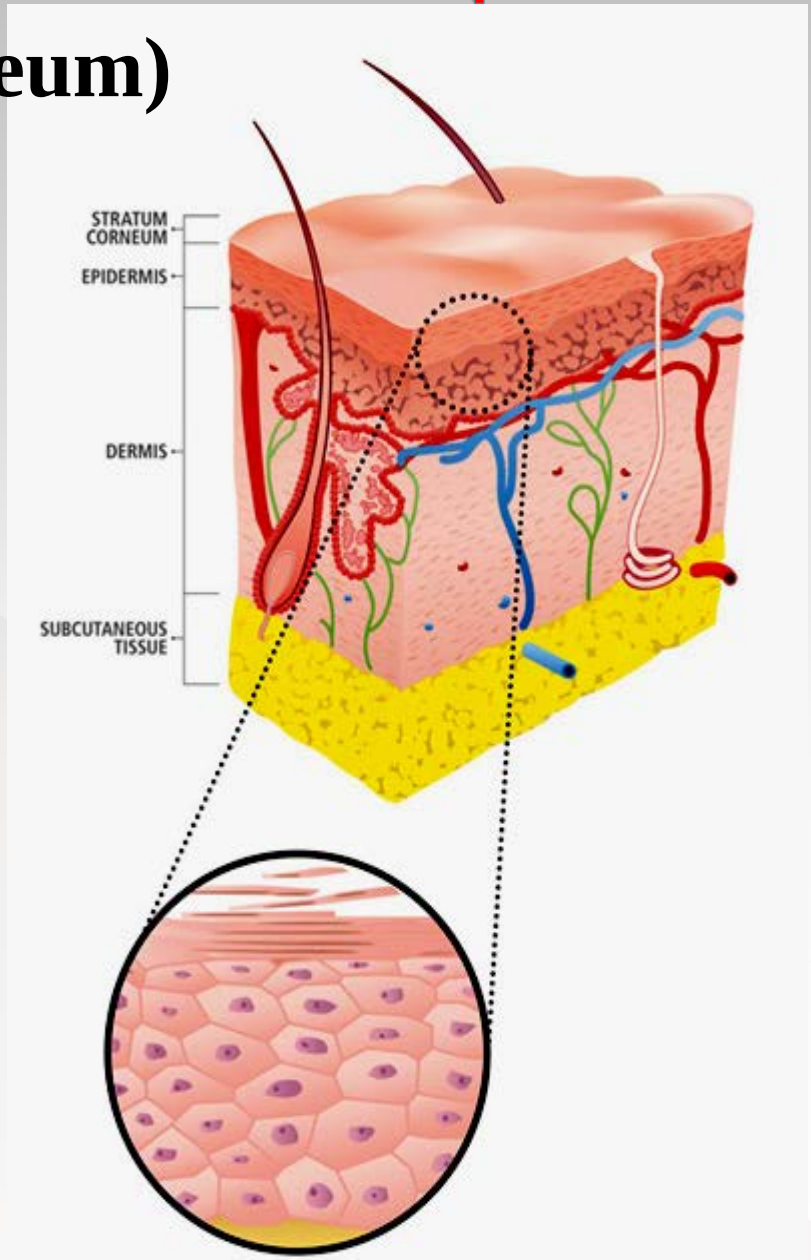
through GI tract

- stomach (acids)
- small intestine
- villi



Absorption: Dermal absorption

- **epidermis (stratum corneum)**
- **dermis**

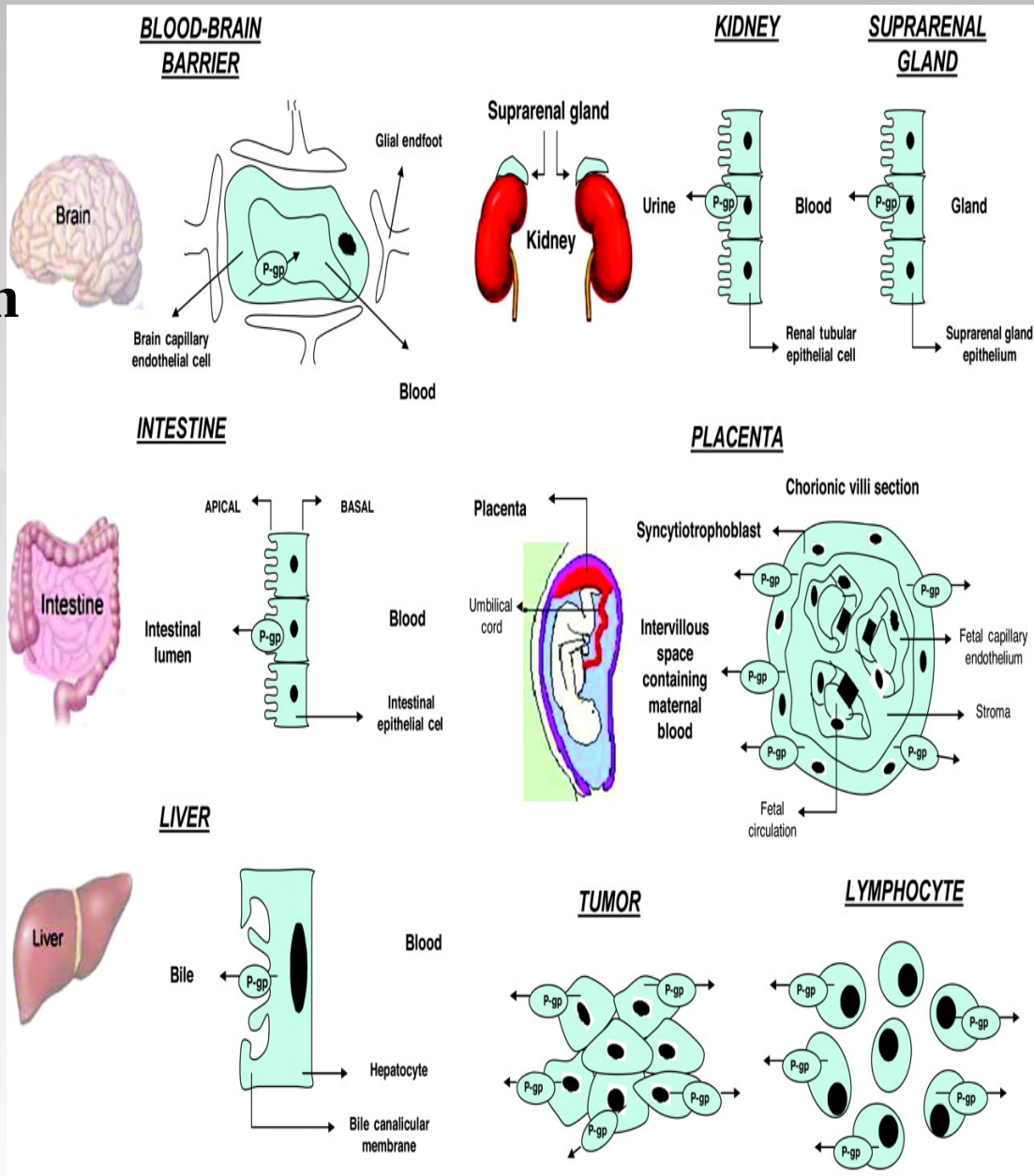


Distribution:
the process in which a chemical agent translocates throughout the body

- Blood
- Rate of distribution
- Storage and Binding

Distribution: Blood

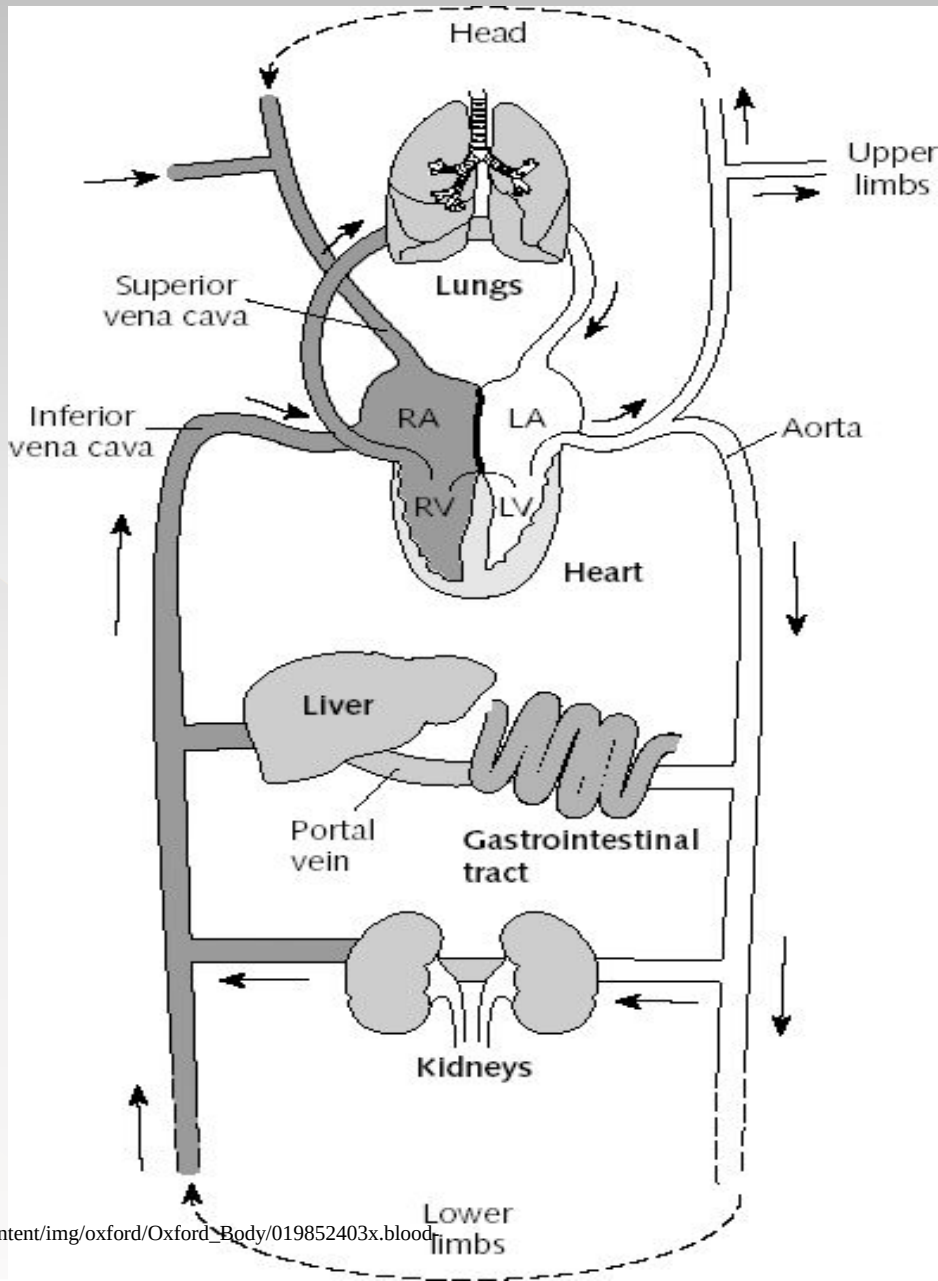
- Carries the agent
- Site of action
- - organs of storage depots
- - organs of transformation
- - organs of elimination



Marchetti S et al. The Oncologist 2007;12:927-941

Distribution: Rate of distribution

- **blood flow**
- **characteristics of toxicant**

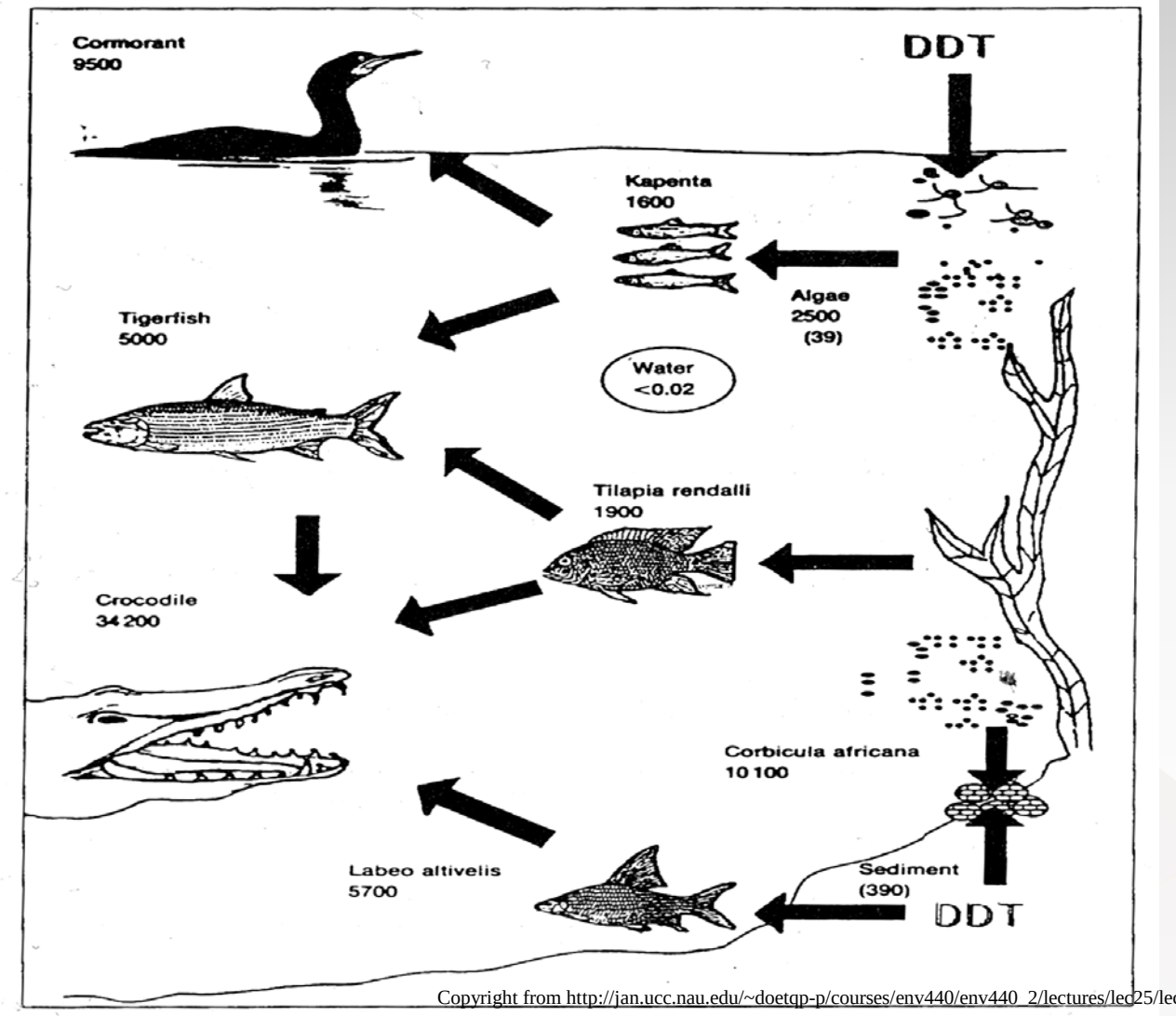


Distribution: Storage and Binding

- **Storage in Adipose tissue**
- **Storage in Bone**
- **Binding to Plasma proteins**

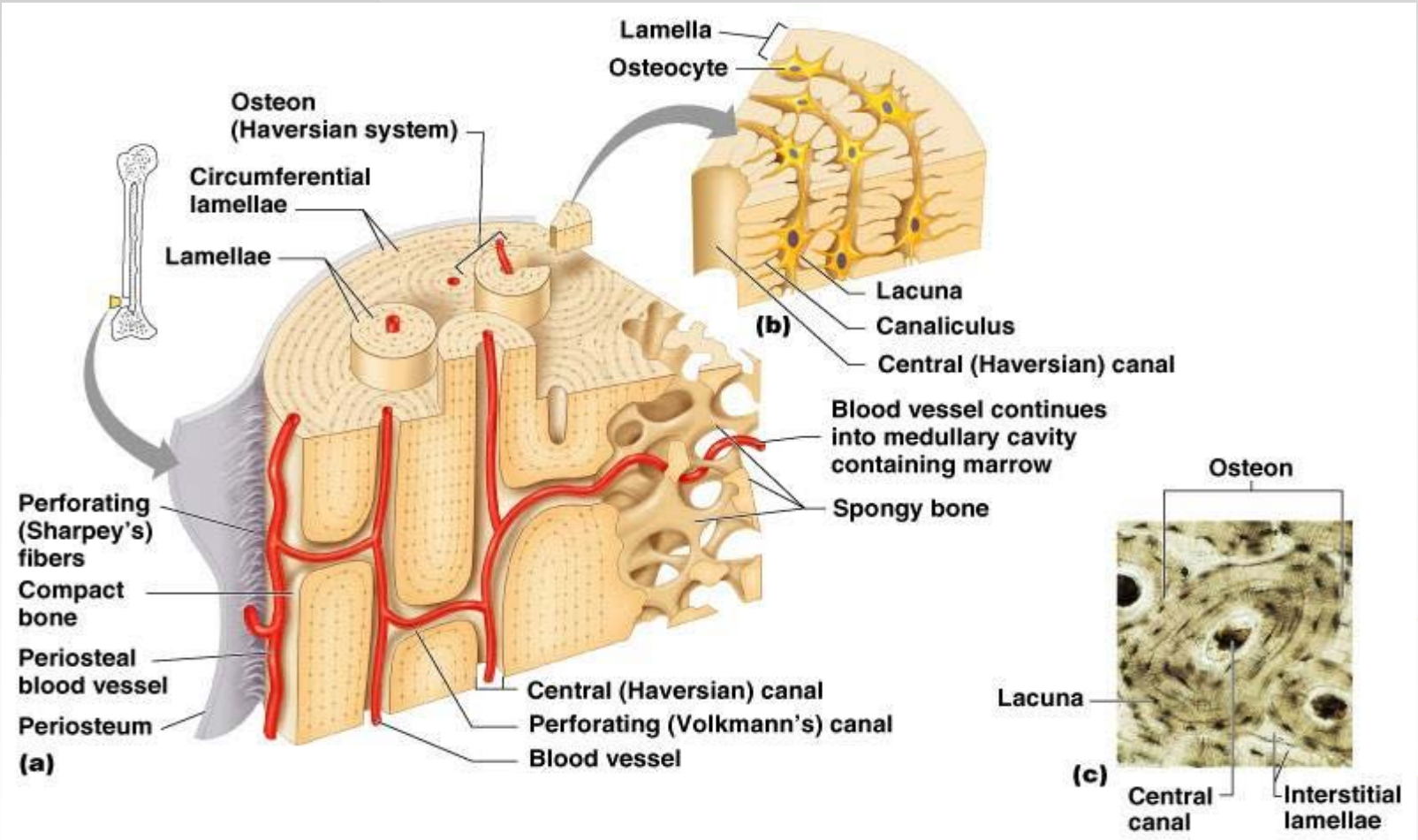
Distribution:
Storage and Binding: Adipose tissue

- lipophylic compounds (DDT)



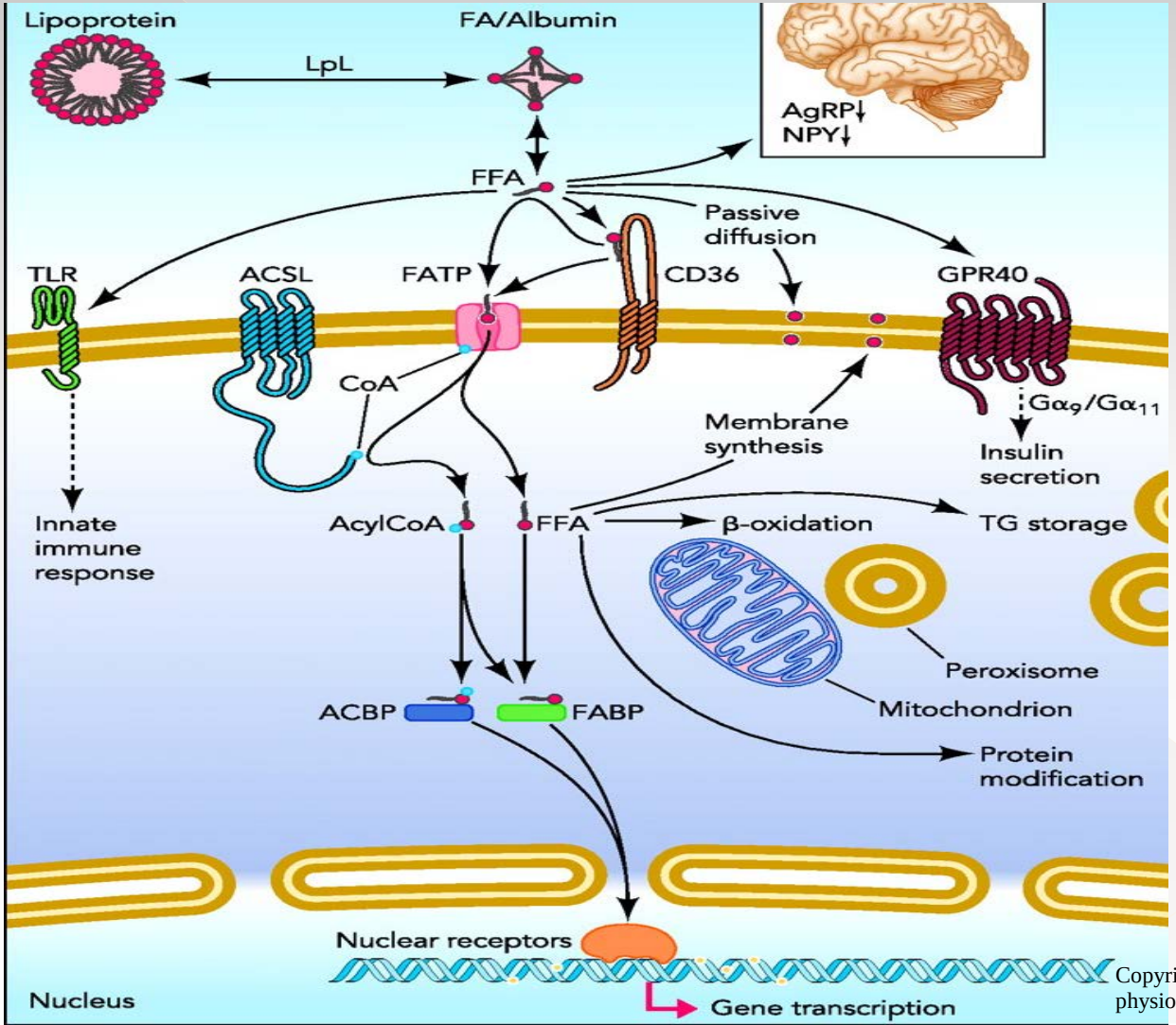
Distribution:
Storage and Binding: Bone

- **Chemicals analogous to Calcium**
- **- Fluoride, Lead, Strontium**



Distribution:
Storage and Binding: Plasma protein

- - Albumin

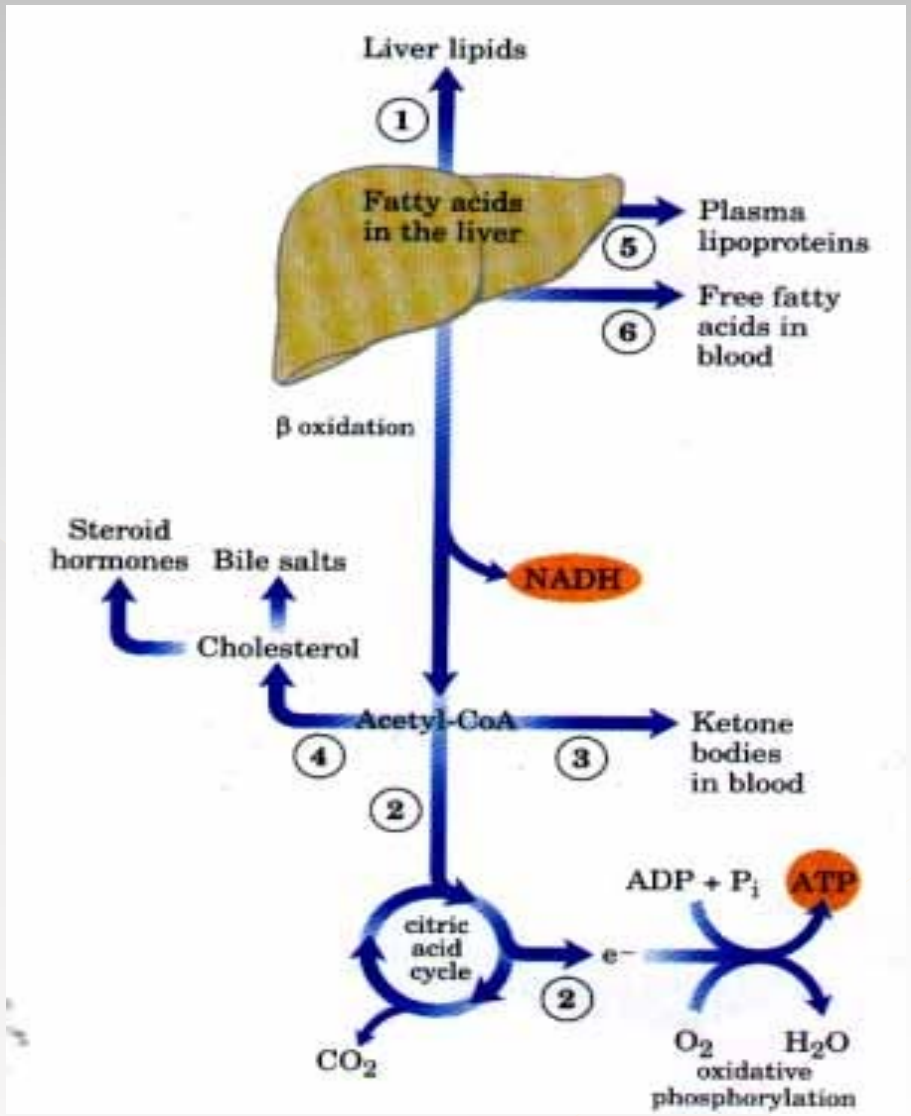


Target Organs

- Not all organs are affected equally
  - greater susceptibility of the target organ
  - higher concentration of active compound

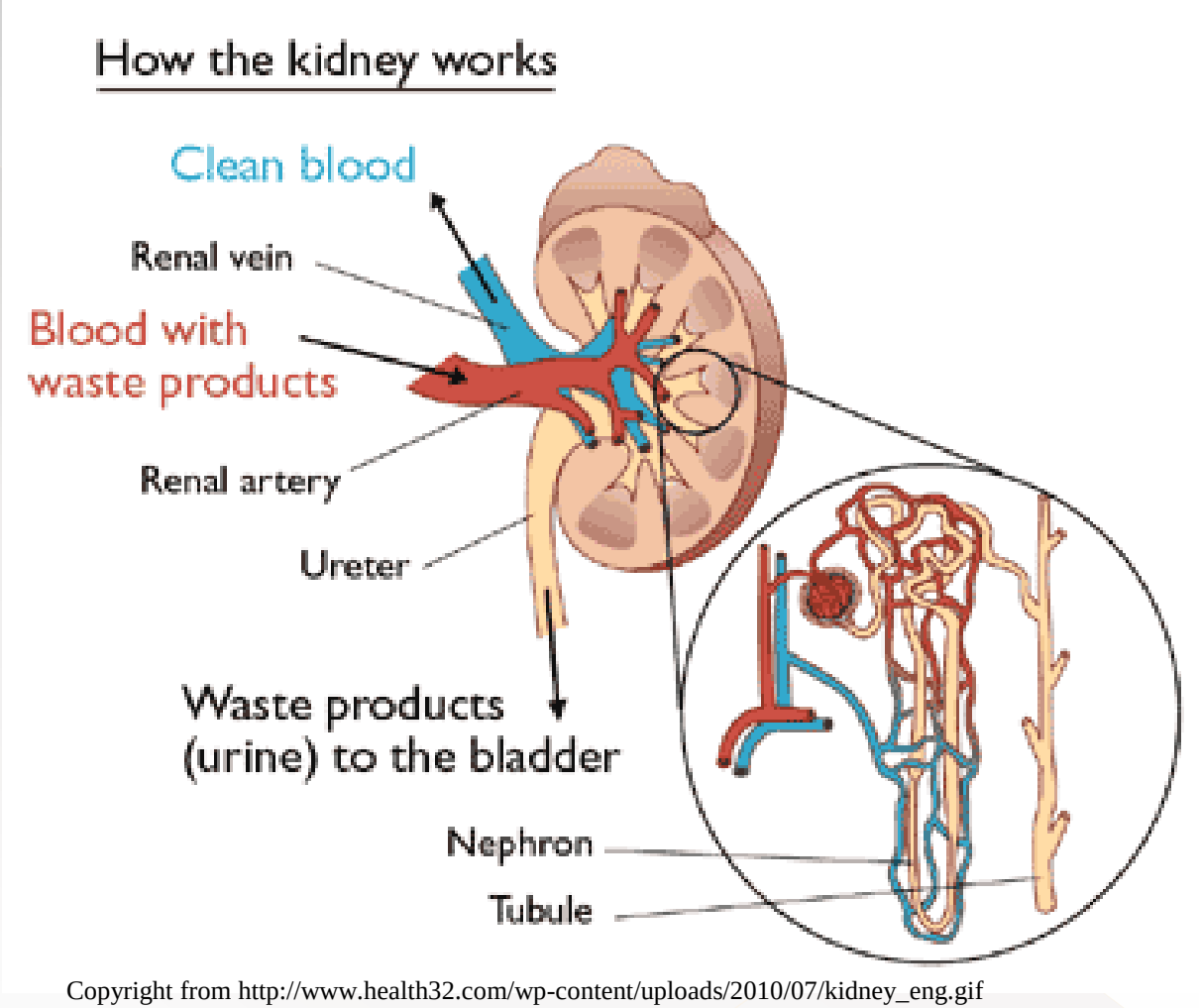
Target Organs: Liver

- oxidative reactions



Target Organs: Kidney

- **Concentrates chemicals**



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Target Organs: Lung

- **Site of exposure**

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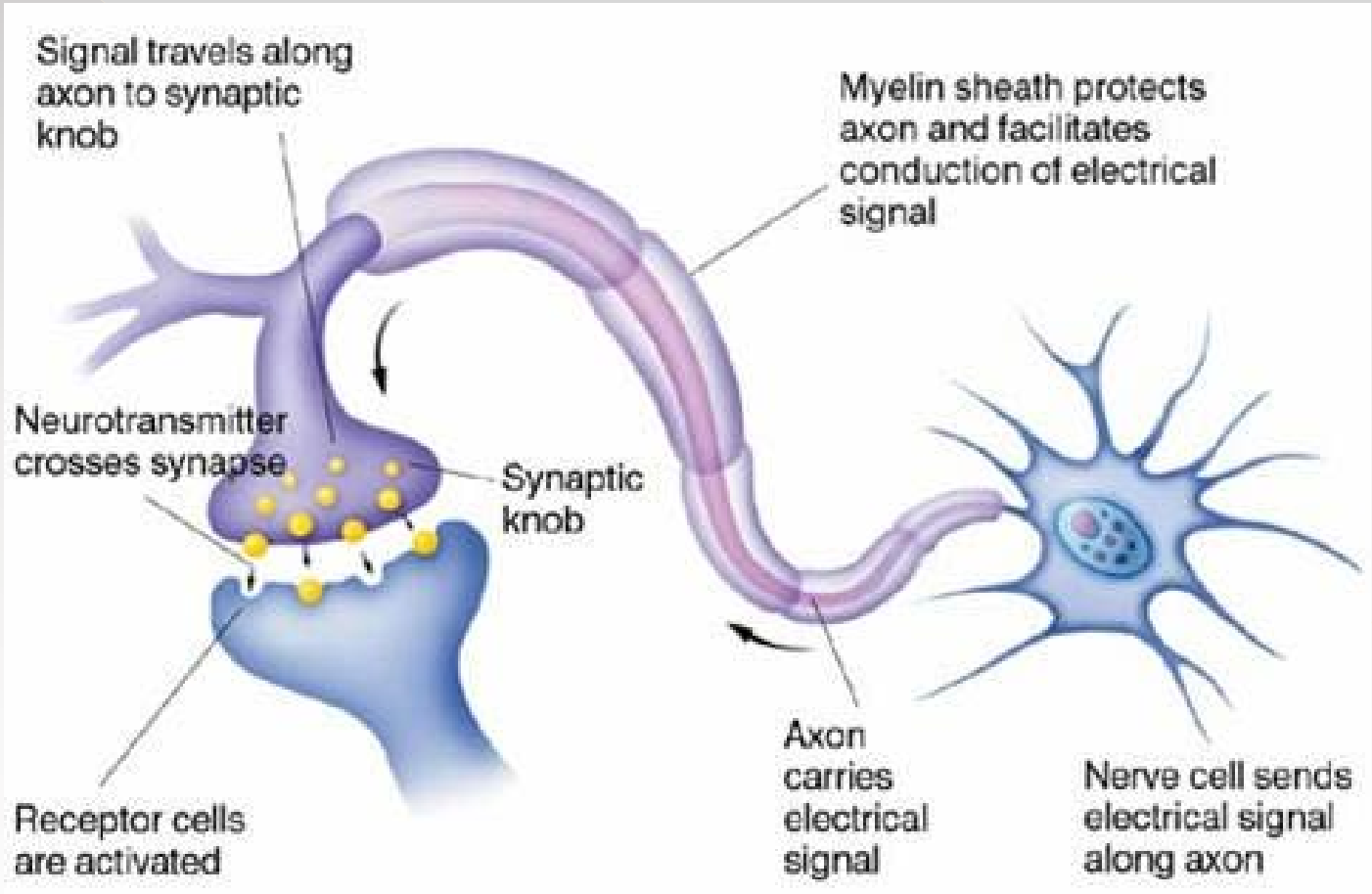
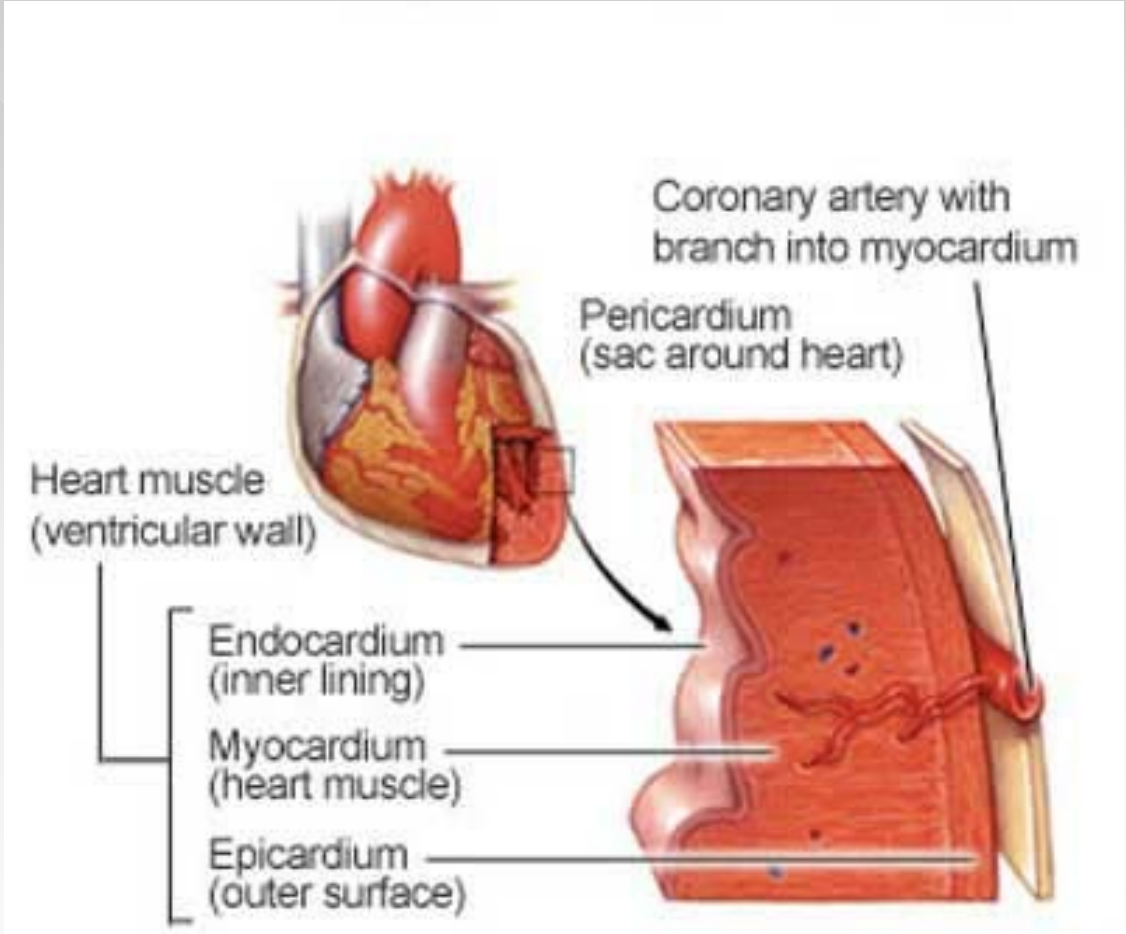
Target Organs: Neurons

- Oxygen dependent
- Irreversible damage



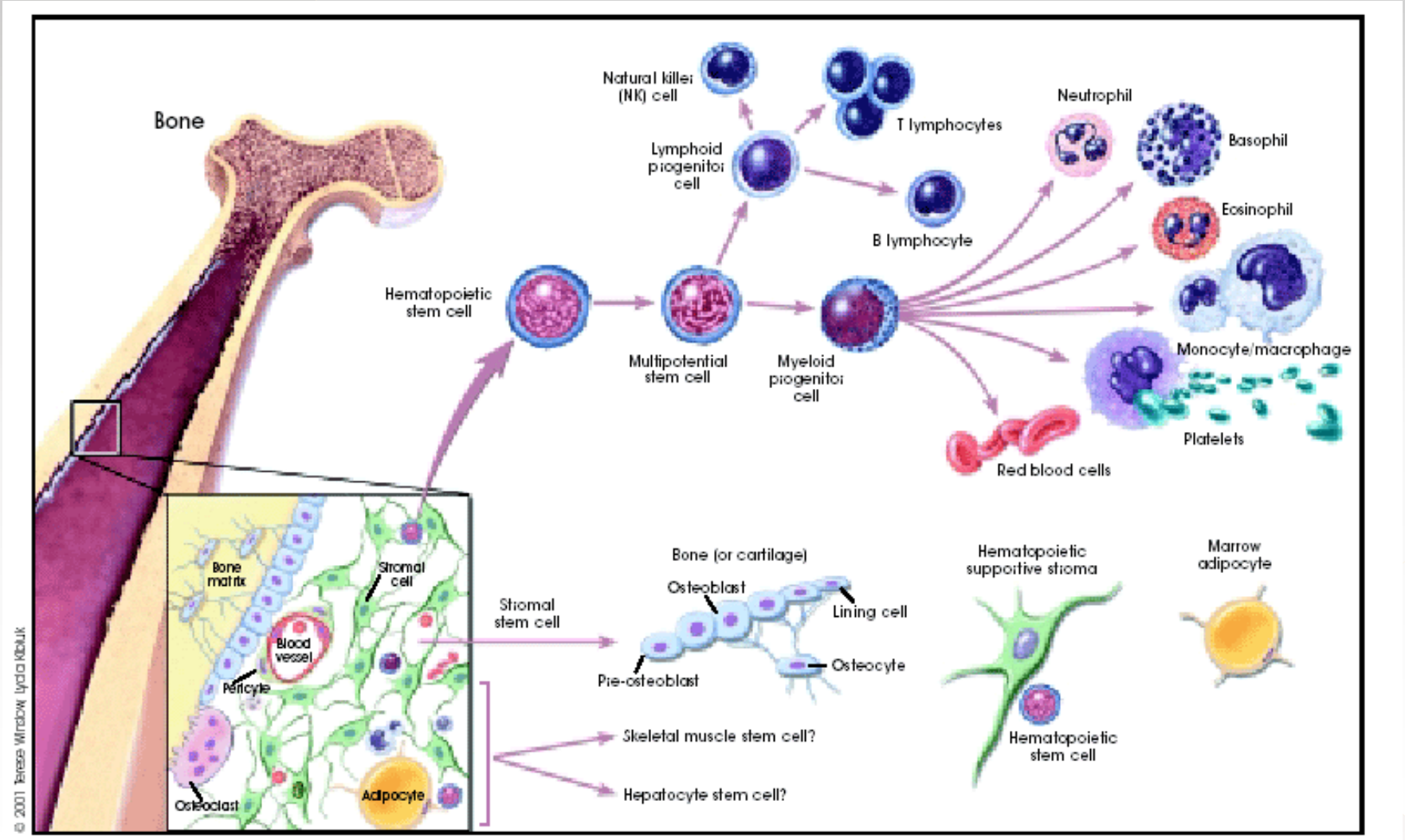
Target Organs: Myocardium

- **Myocardium--oxygen dependent**



Target Organs: Bone marrow / intestinal mucosa

- **Rapid divide**



Target Sites: Mechanisms of Action

- Molecular
- Cellular

Target Sites: Mechanisms of Action

- **Cellularly, chemical can**
  - interfere with **receptor-ligand binding**
  - interfere with **membrane function**
  - interfere with **cellular energy production**
  - bind to **biomolecules**
  - perturb **homeostasis (Ca)**

Target Sites: Mechanisms of Action

- Molecularly, chemical can interact with

  Proteins Lipids Carbohydrate DNA

Metabolism

- Chemical parent compounds are modified by enzymatic reactions.
- Objective--make more water soluble and easier to excrete
  - decrease lipid solubility
    decrease amount at target
  - increase ionization
    increase excretion rate --> decrease toxicity
- Bioactivation--Biotransformation can result in the formation of **reactive metabolites**

Biotransformation (Metabolism)

Compound	Without Metabolism	With Metabolism
Ethanol	4 weeks	10mL/hr
Phenobarbital	5 months	8hrs
DDT	infinity	Days to weeks

Biotransformation

- Main organs
  - High - LIVER
  - Medium - Lung, Kidney, Intestine
  - Low - Others

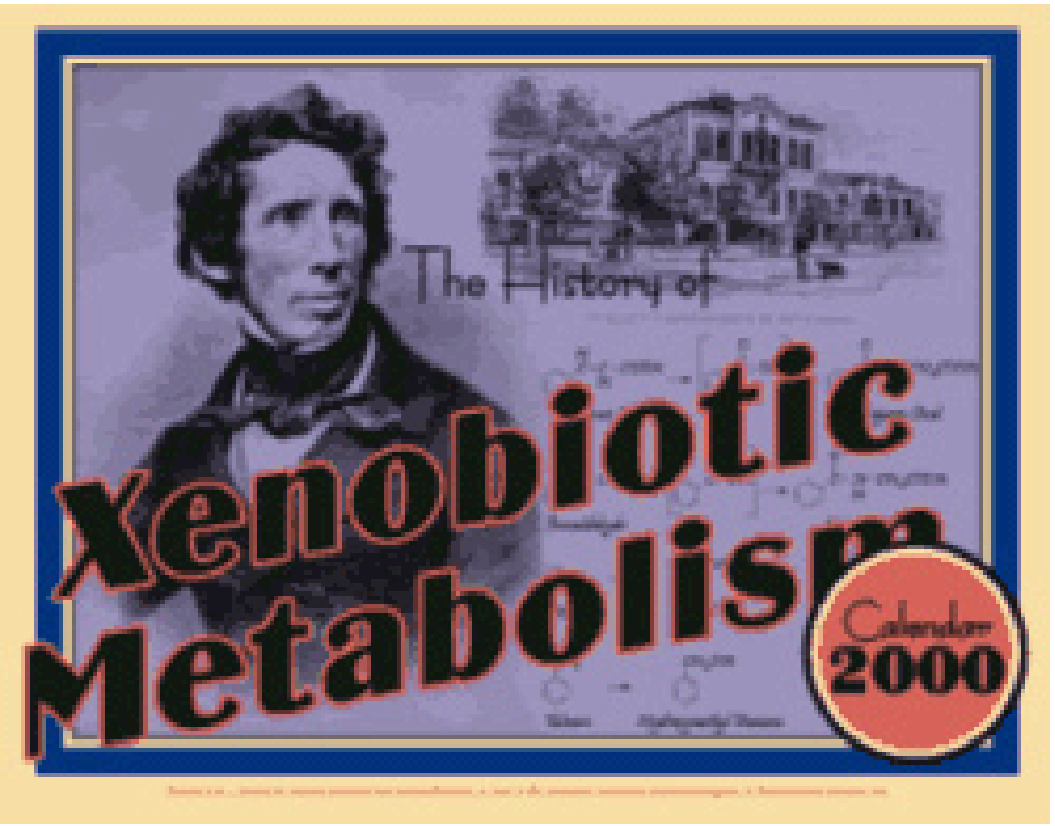
- Biotransformation Pathways
  - Phase I--Water soluble production
  - Phase II--Conjugation with a soluble endogenous agent

Excretion:
Toxicants are eliminated from the body

- Urinary excretion
- Exhalation
- Biliary Excretion via Fecal Excretion
- Milk Sweat Saliva

เมแทบอลิซึมของสารแปลกปลอม

(Xenobiotic Metabolism)



Xenos – “stranger”

Xenobiotic – compound that is foreign to body

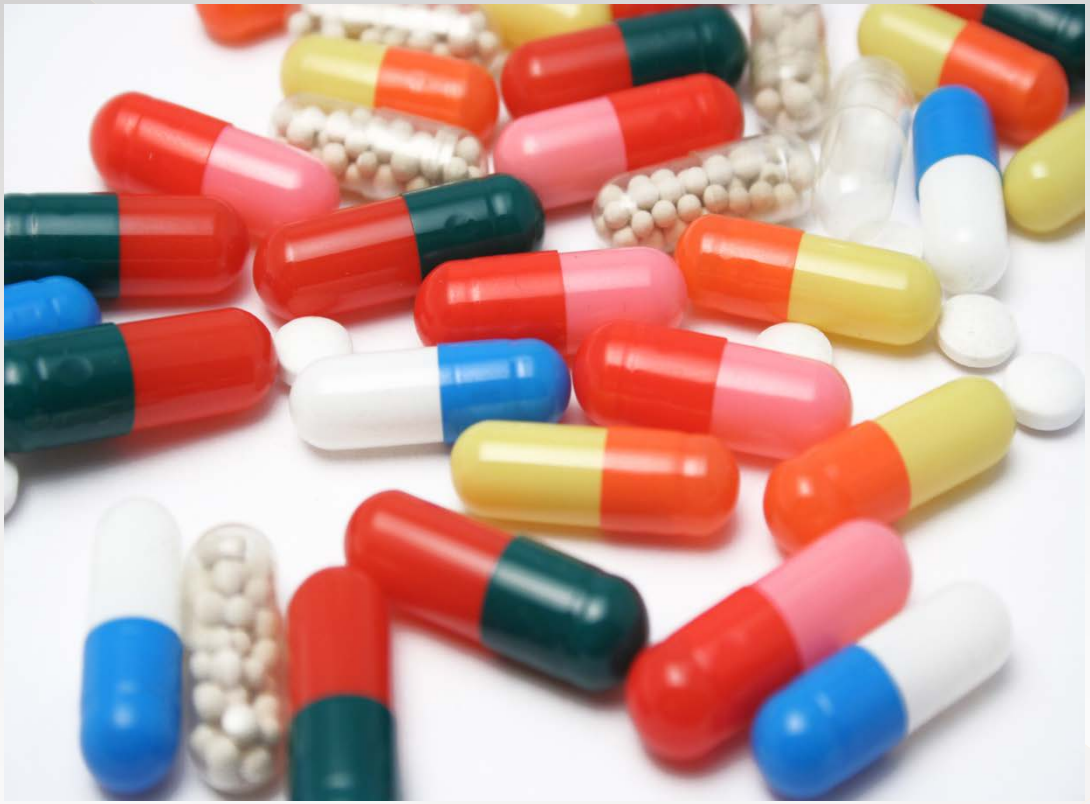
Types of Xenobiotic

-Drugs

- Carcionogens

- Environmental chemical or pollutants

Types of Xenobiotic -Drugs



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Types of Xenobiotic -Carcionogens

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Types of Xenobiotic -Environmental chemical or pollutants

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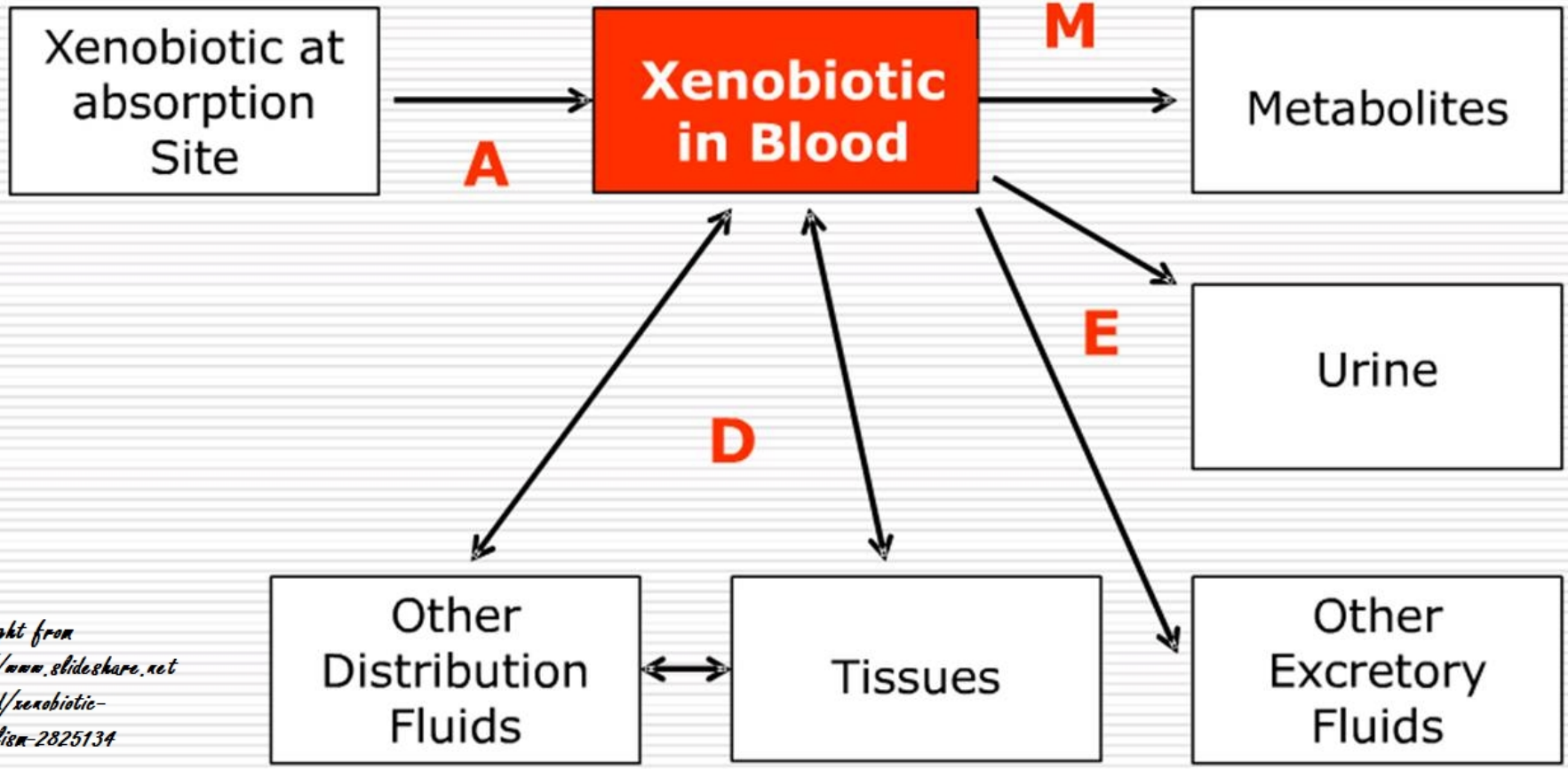
Xenobiotic ADME

Xenobiotic ADME



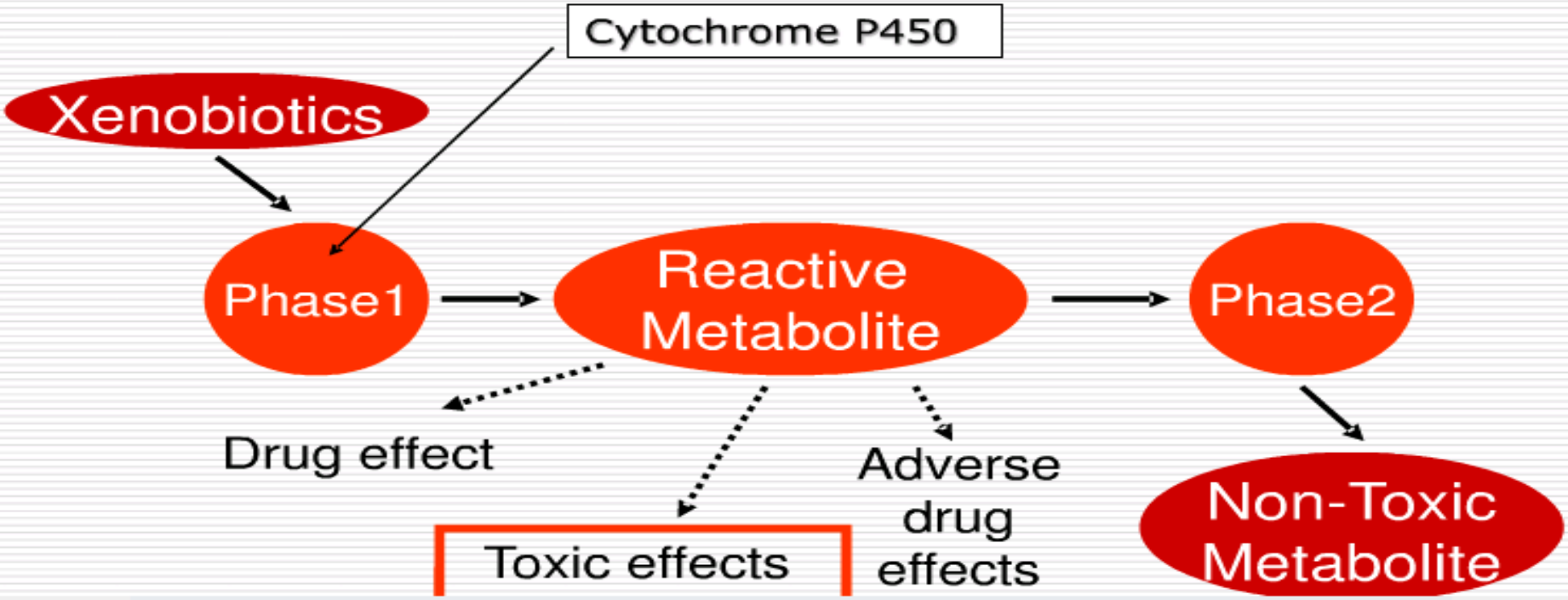
Phases of Xenobiotic Metabolism

Phase I – Modification

Phase II – Conjugation

Phase III – Further and Excretion

Cellular Response to Xenobiotics



Phases of Xenobiotic Metabolism

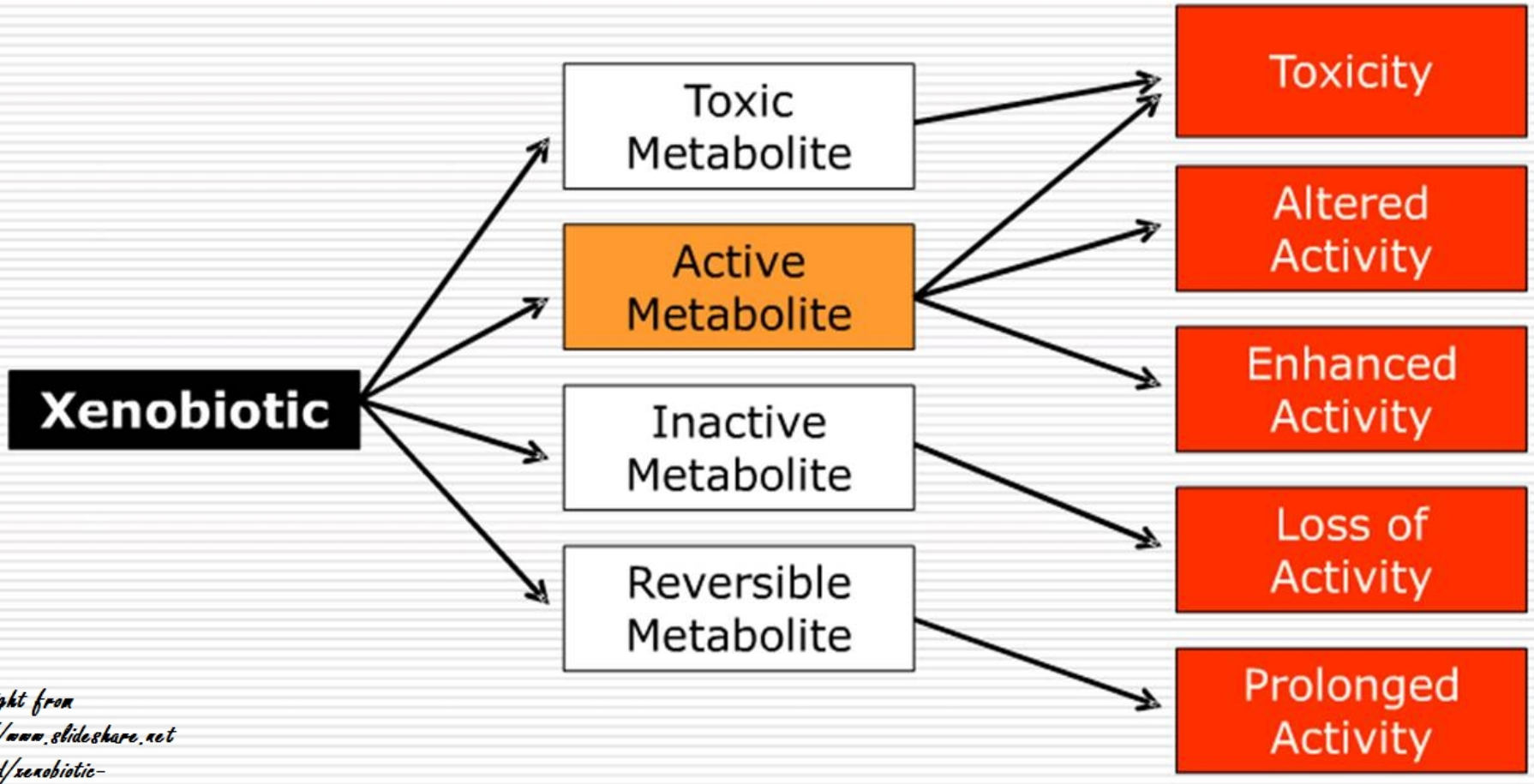
Phase I – Modification

- Change to biologic active compound (functionalization)

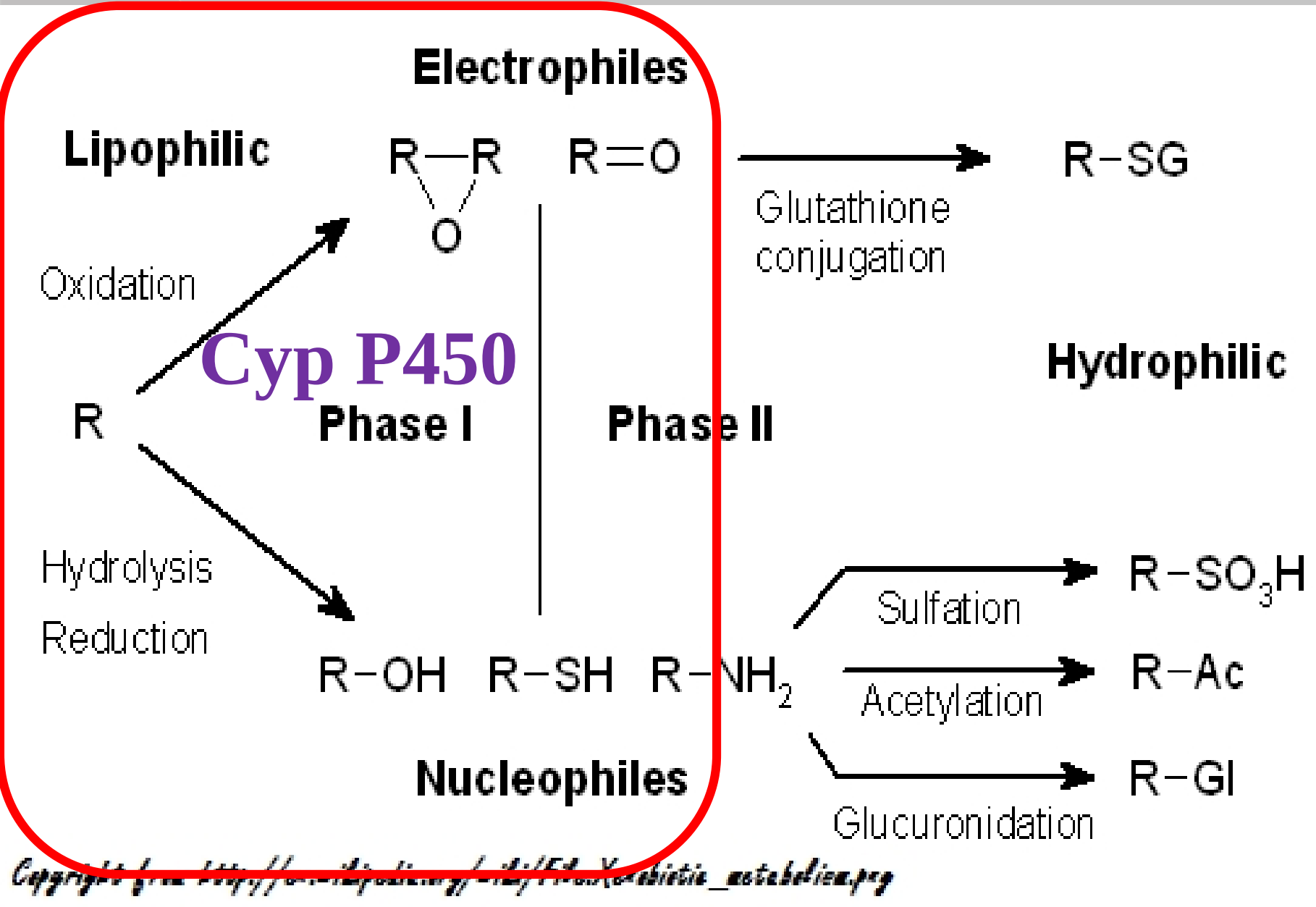
- Catalyzed by Cytochrome P450

Xenobiotic Biotranformation

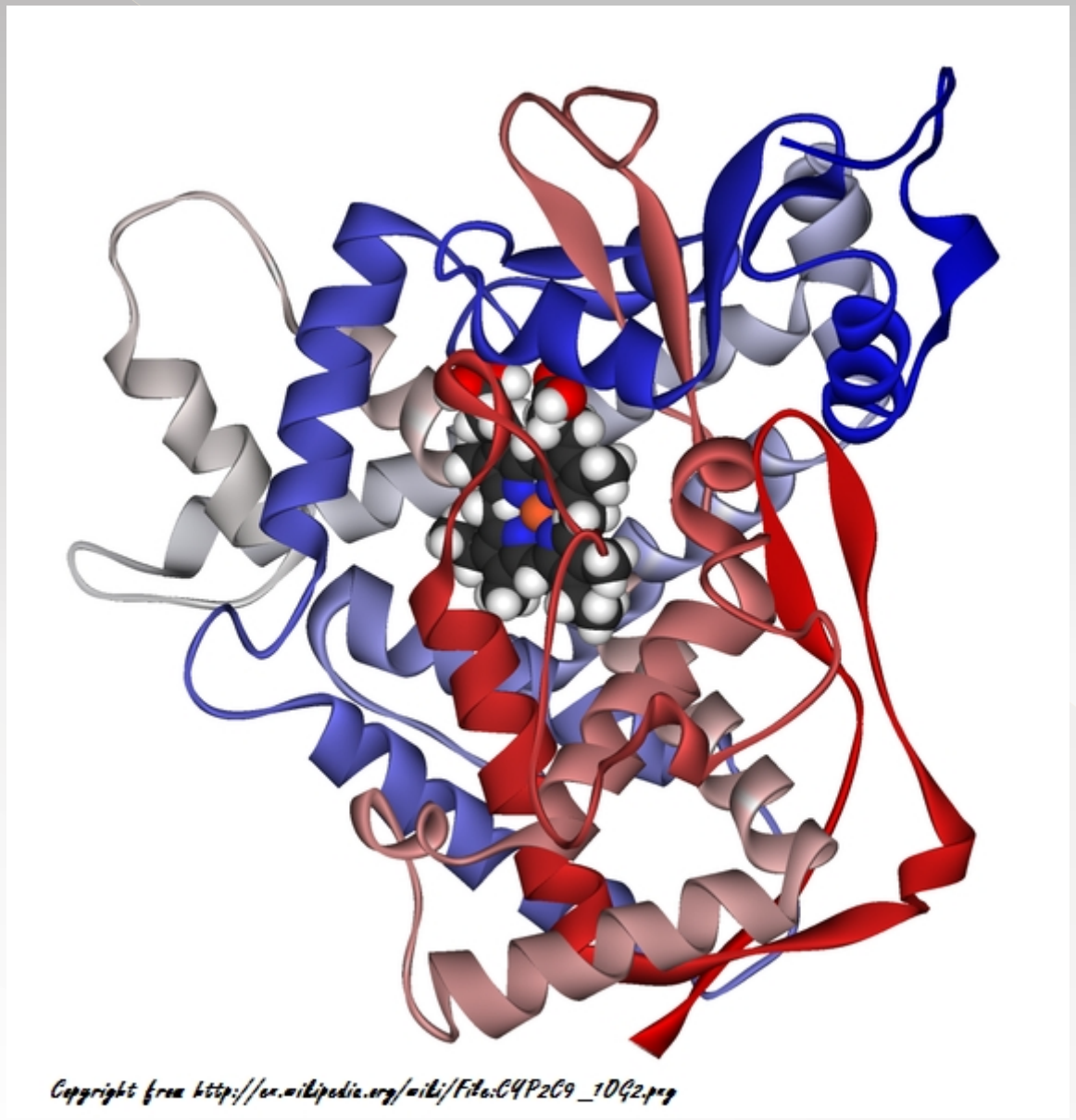
Xenobiotics: Fates



Phases I of Xenobiotic Metabolism

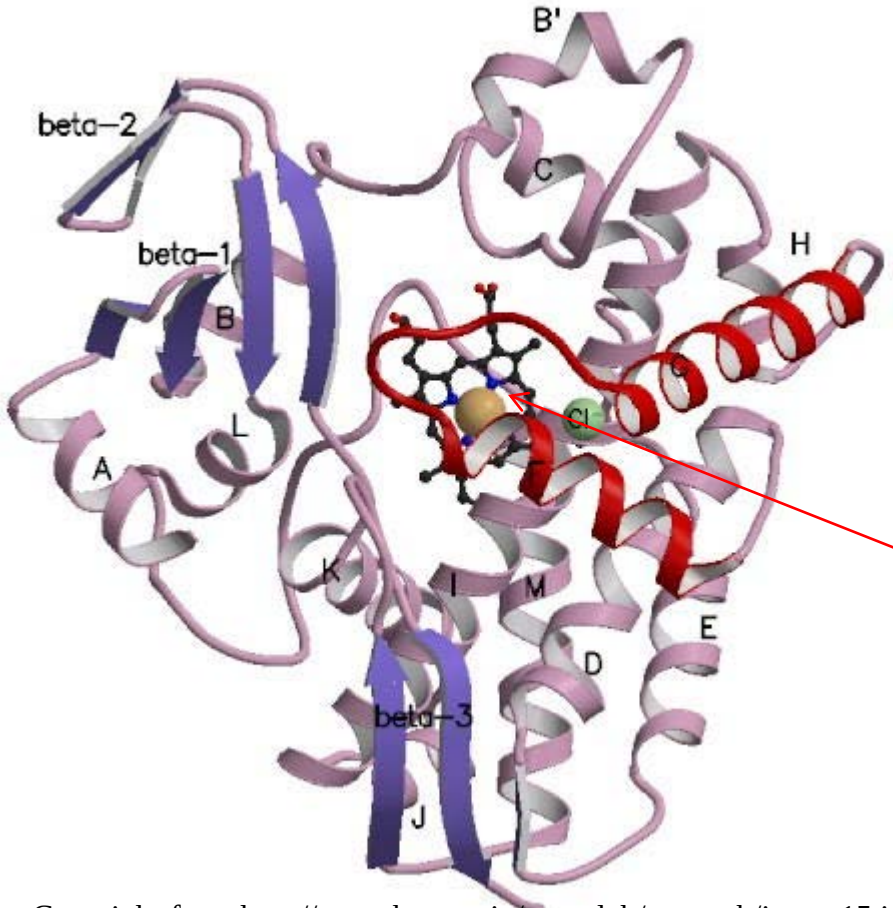


Phases I of Xenobiotic Metabolism



Cytochrome P450 oxidase are important enzymes in xenobiotic metabolism.

Cytochrome P450



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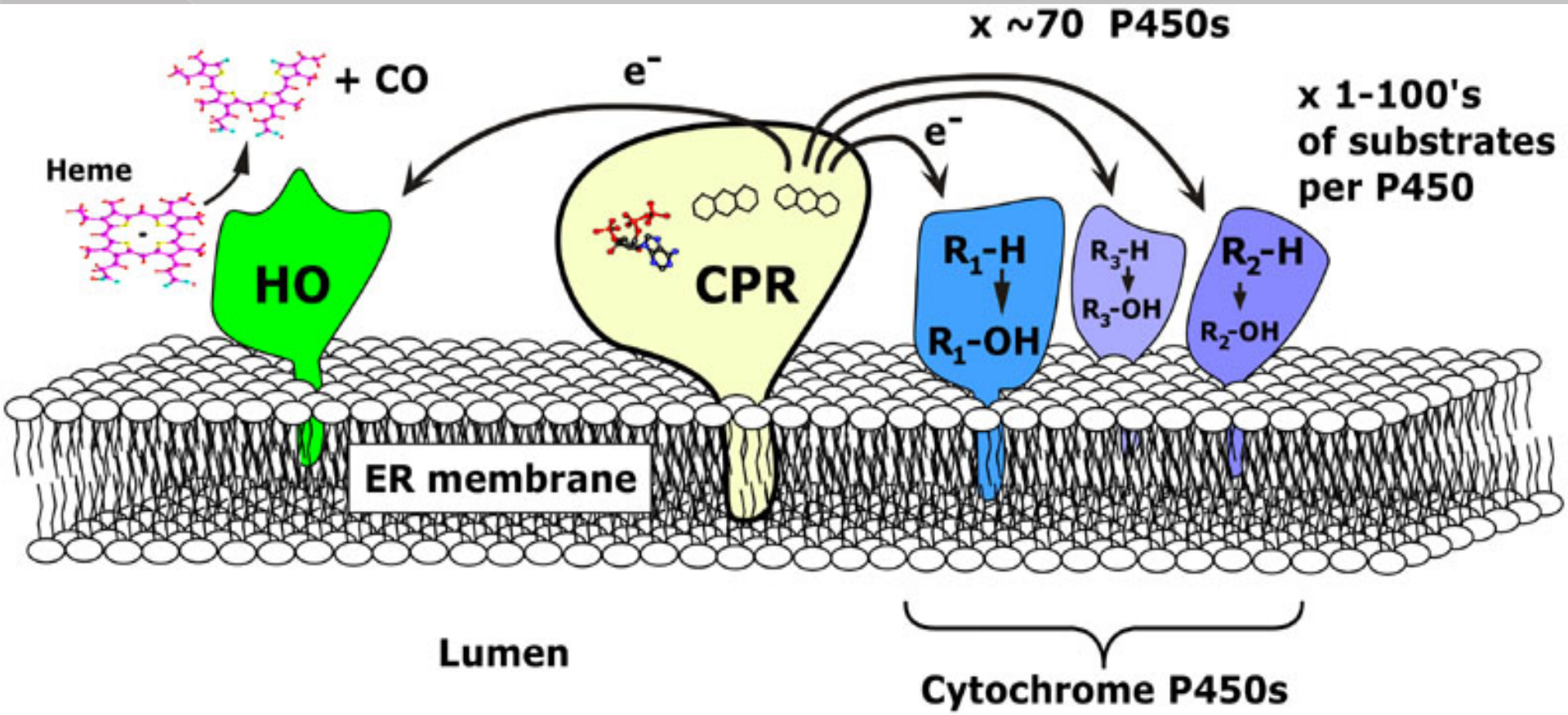
Iron ion (Active site)

Cytochrome P450

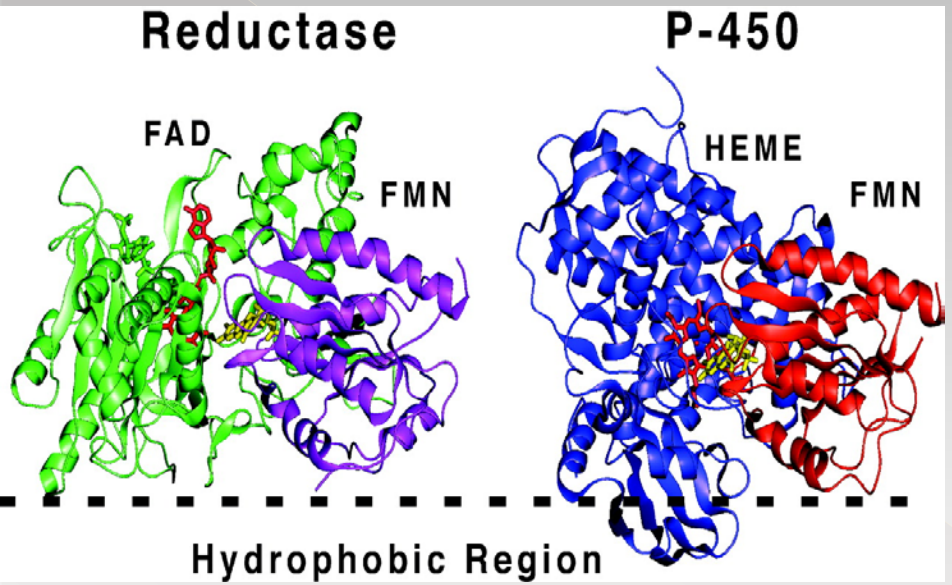


Heme oxygenase (*HO*)

C-Reactive protein (CRP)

Membrane bound proteins

Cytochrome P450



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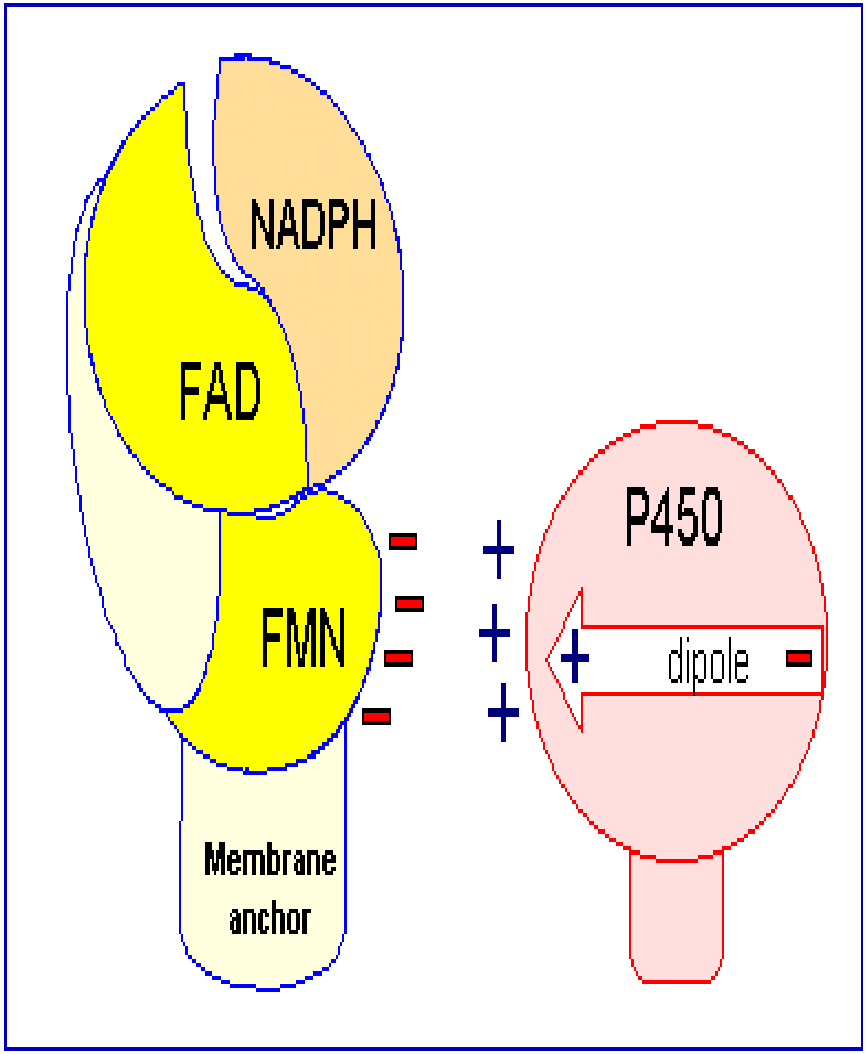

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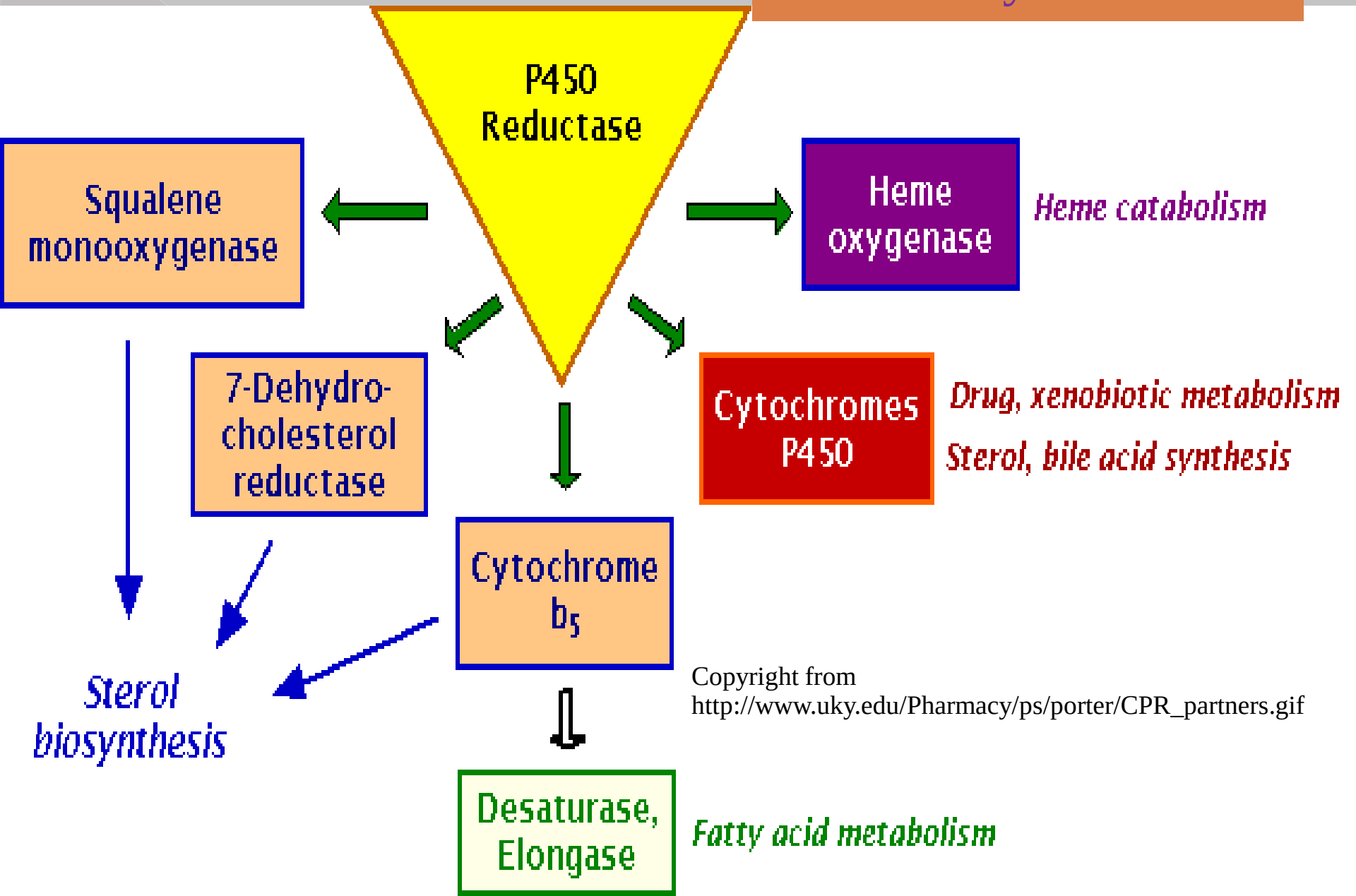
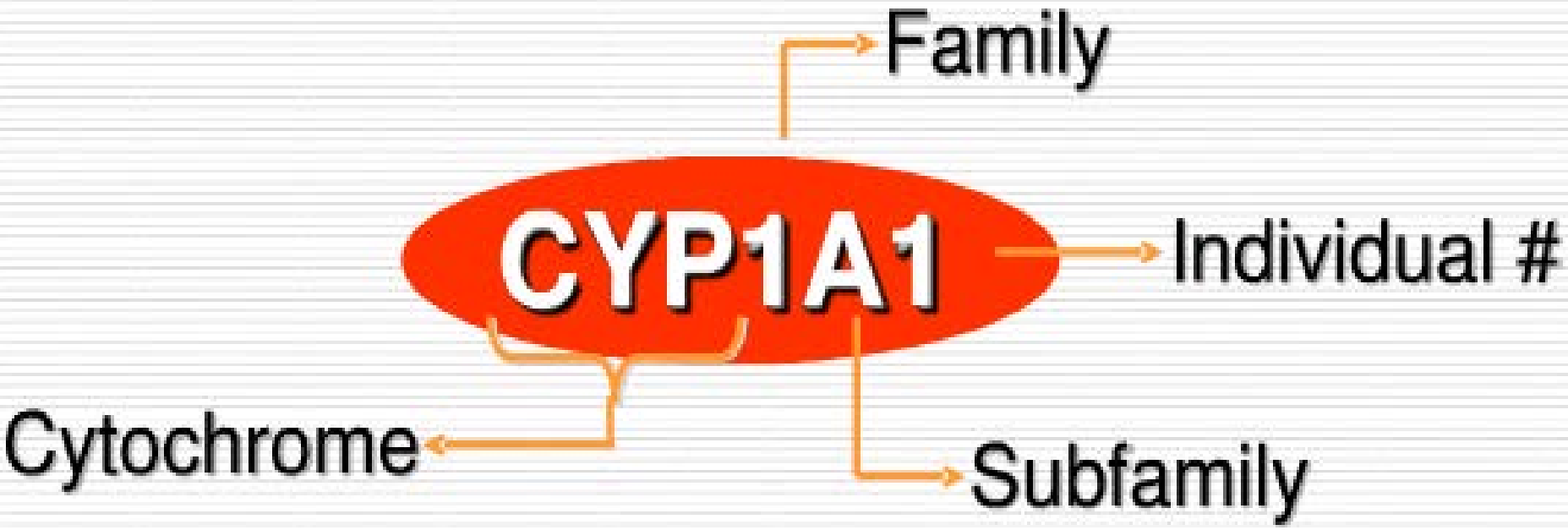
Cytochrome P450

Oxygenase enzyme family in ER



Characterization of Cytochrom P450

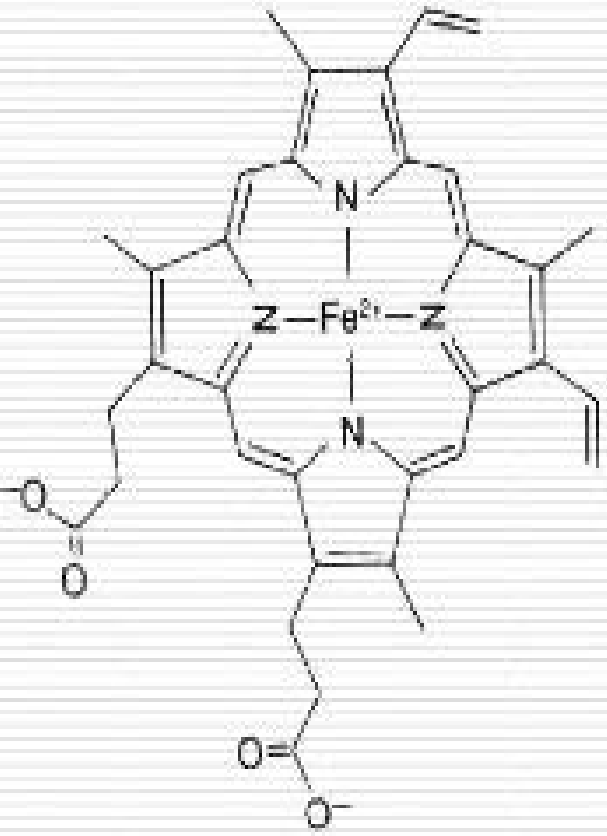
1. large # of Isoforms (150): Nomenclature

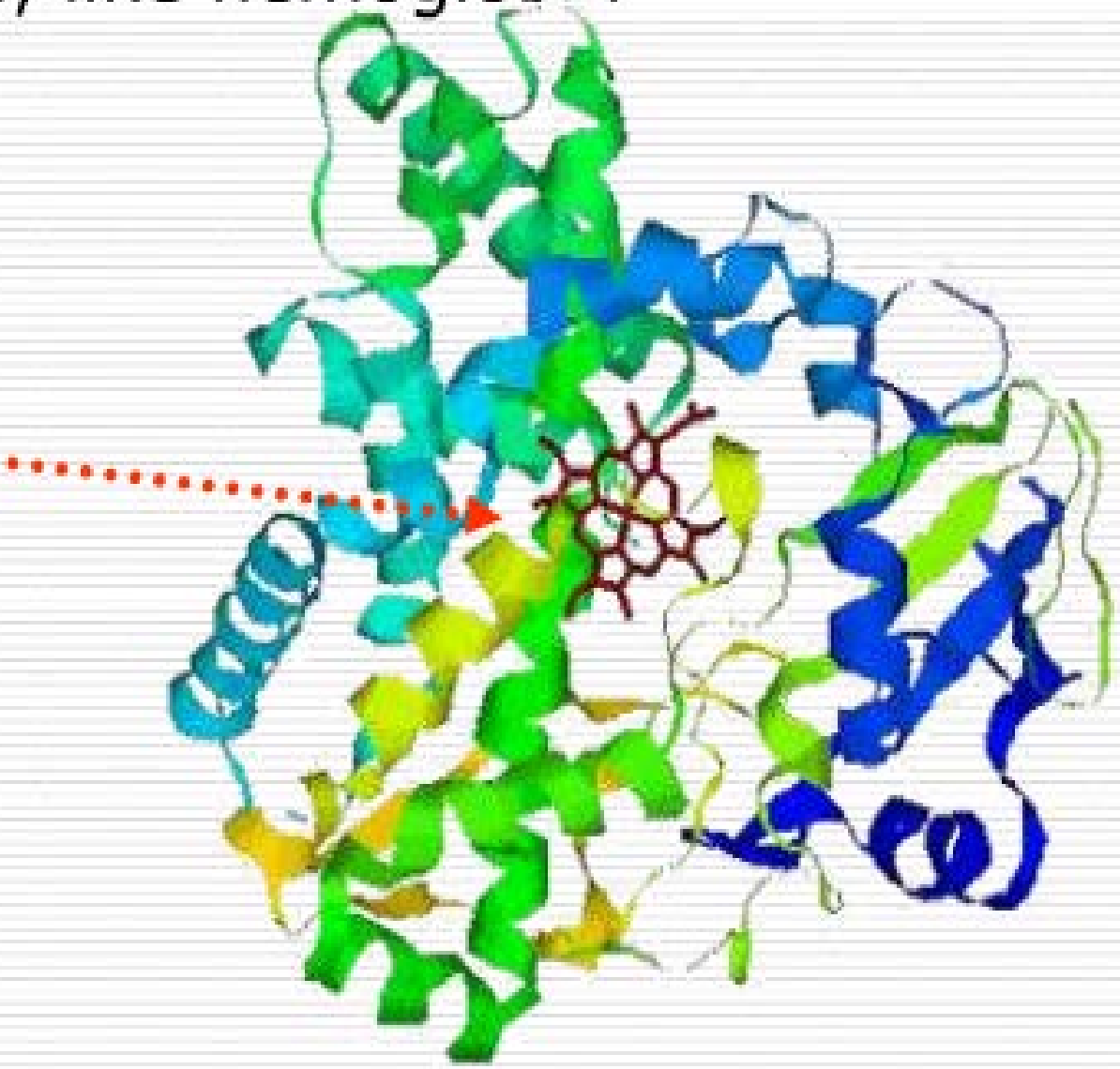


CYP1A1 - Gene encoding for CYP1A1

Characterization of Cytochrom P450

2. hemoproteins, like hemoglobin



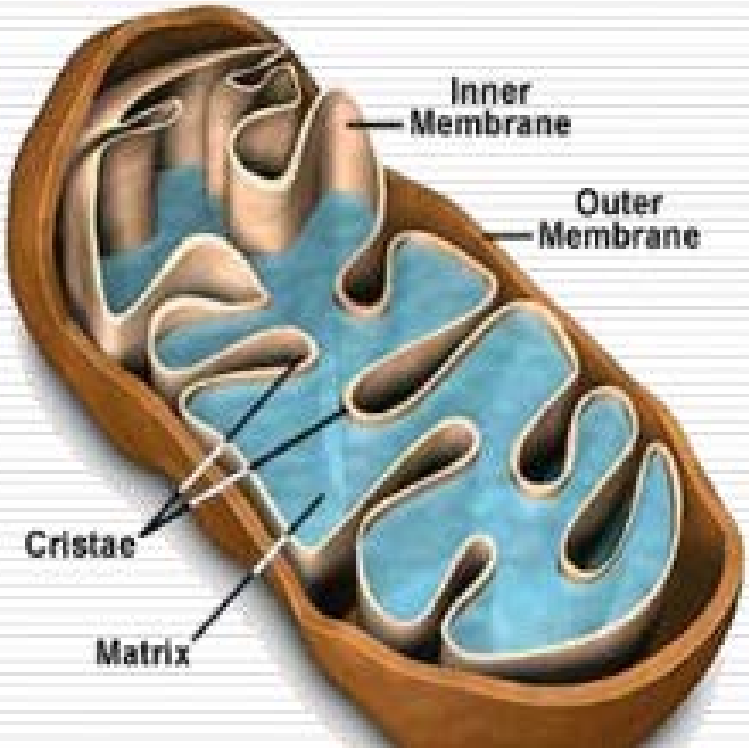


Characterization of Cytochrom P450

3. widely distributed among species

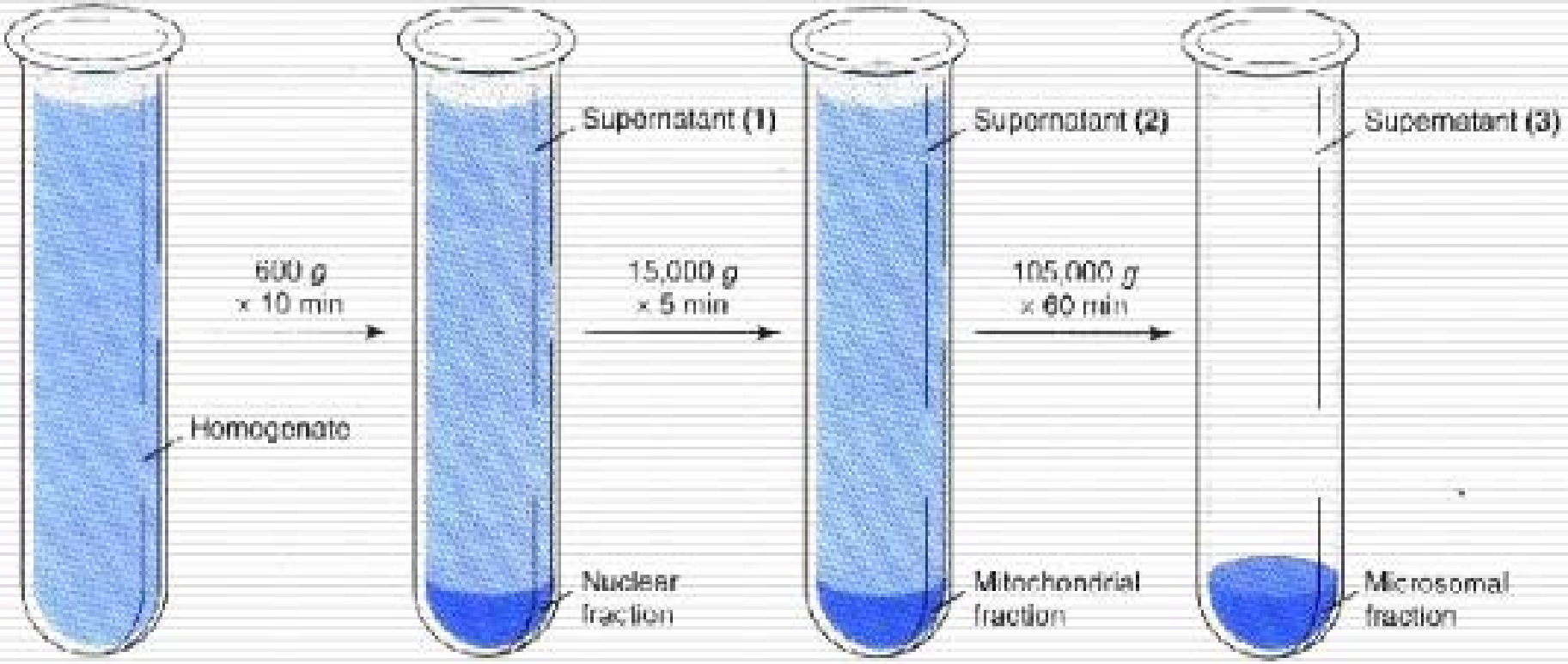
P450-containing systems fall into two major classes:

- bacterial/mitochondrial (type I)
- microsomal (type II)



Characterization of Cytochrom P450

4. Microsomal - membranes of the smooth endoplasmic reticulum, mostly in liver



Hepatic microsomes - cytochrome P450 comprise nearly 20% of total proteins

Characterization of Cytochrom P450

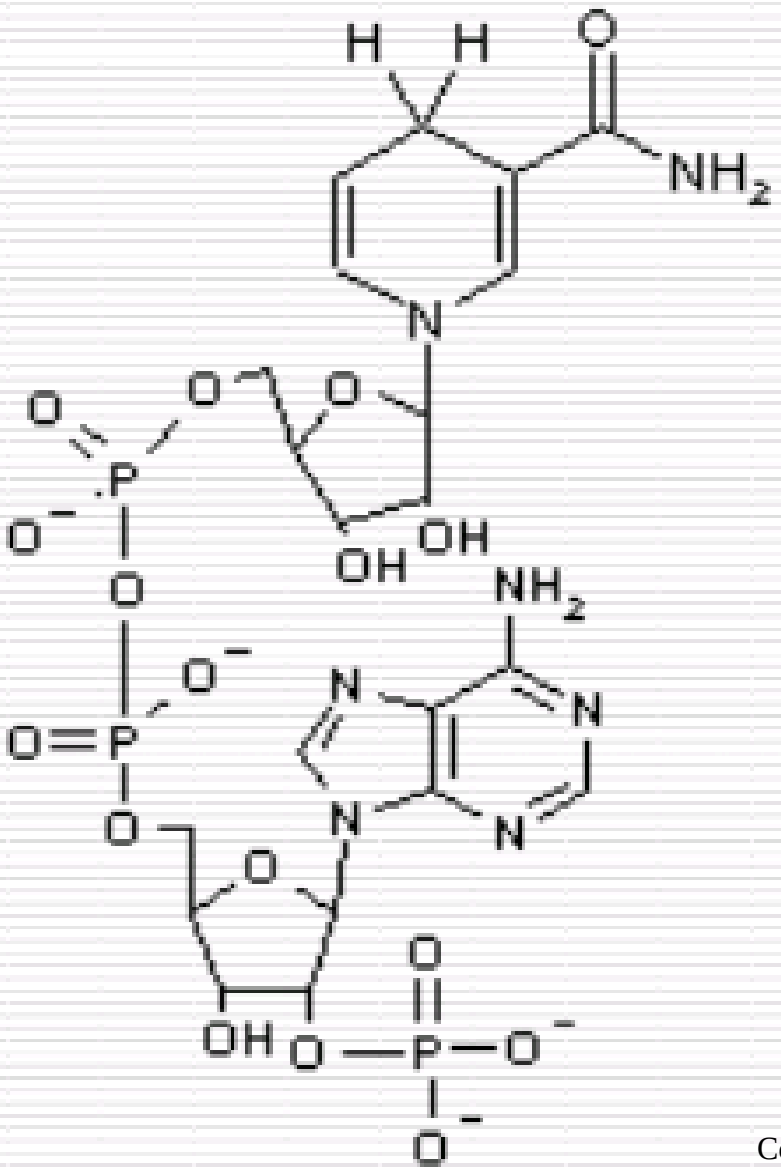
5. Multiple isoforms with wide and overlapping substrate specificities
   - xenobiotics
   - endogenous compounds - steroids, eicosanoids, fatty acids, retinoids, etc.

Isoenzymes of Cytochrom P450

Family	Function	Members	Names
CYP1	drug and steroid (especially estrogen) metabolism	3 subfamilies, 3 genes, 1 pseudogene	CYP1A1, CYP1A2, CYP1B1
CYP2	drug and steroid metabolism	13 subfamilies, 16 genes, 16 pseudogenes	CYP2A6, CYP2A7, CYP2A13, CYP2B6, CYP2C8, CYP2C9, CYP2C18, CYP2C19, CYP2D6, CYP2E1, CYP2F1, CYP2J2, CYP2R1, CYP2S1, CYP2U1, CYP2W1
CYP3	drug and steroid (including testosterone) metabolism	1 subfamily, 4 genes, 2 pseudogenes	CYP3A4, CYP3A5, CYP3A7, CYP3A43

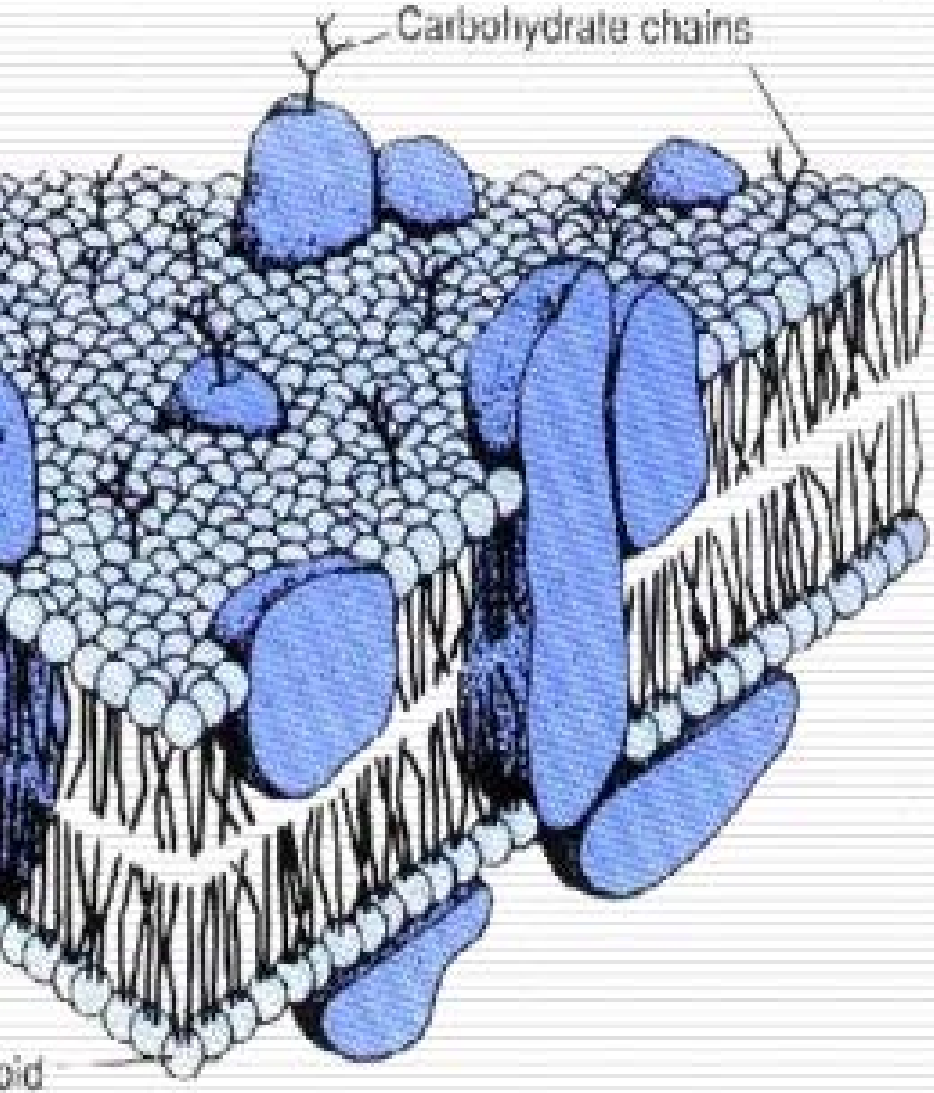
Enzyme	Substrates
CYP1A2	Amitriptyline, Betaxolol, Caffeine, Clomipramine, Clozapine, Chlorpromazine, Fluvoxamine, Haloperidol, Imipramine, Olanzapine, Ondansetron, Propranolol, Tacrine, Theophylline, Verapamil, (R)-Warfarin
CYP2A6	Coumarin, Betadiene, Nicotine
CYP2C9	Amitriptyline, Diclofenac, Demadex, Fluoxetine, Ibuprofen, Losartan, Naproxen, Phenytoin, Piroxicam, Tolbutamide, (S)-Warfarin
CYP2C19	Amitriptyline, Citalopram, Clomipramine, Diazepam, Imipramine, Omeprazole

Characterization of Cytochrom P450



6. NADPH, *not* NADH, is involved in the reaction mechanism of cytochrome P450

Characterization of Cytochrom P450



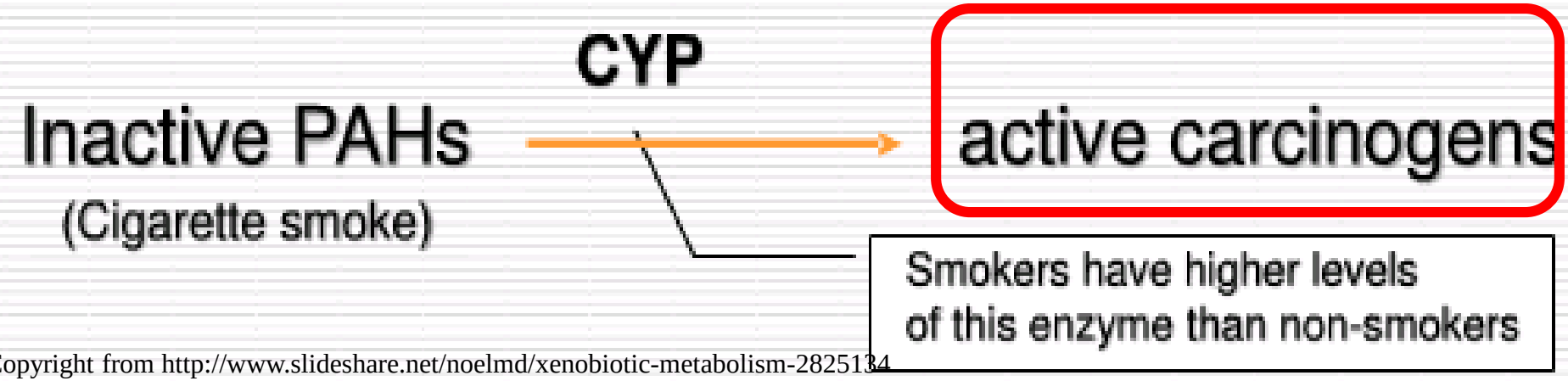
7. Lipids are important components of cytochrome P450
   - Phosphatidylcholine, major lipid found in membranes of ER, is the preferred lipid

Characterization of Cytochrom P450

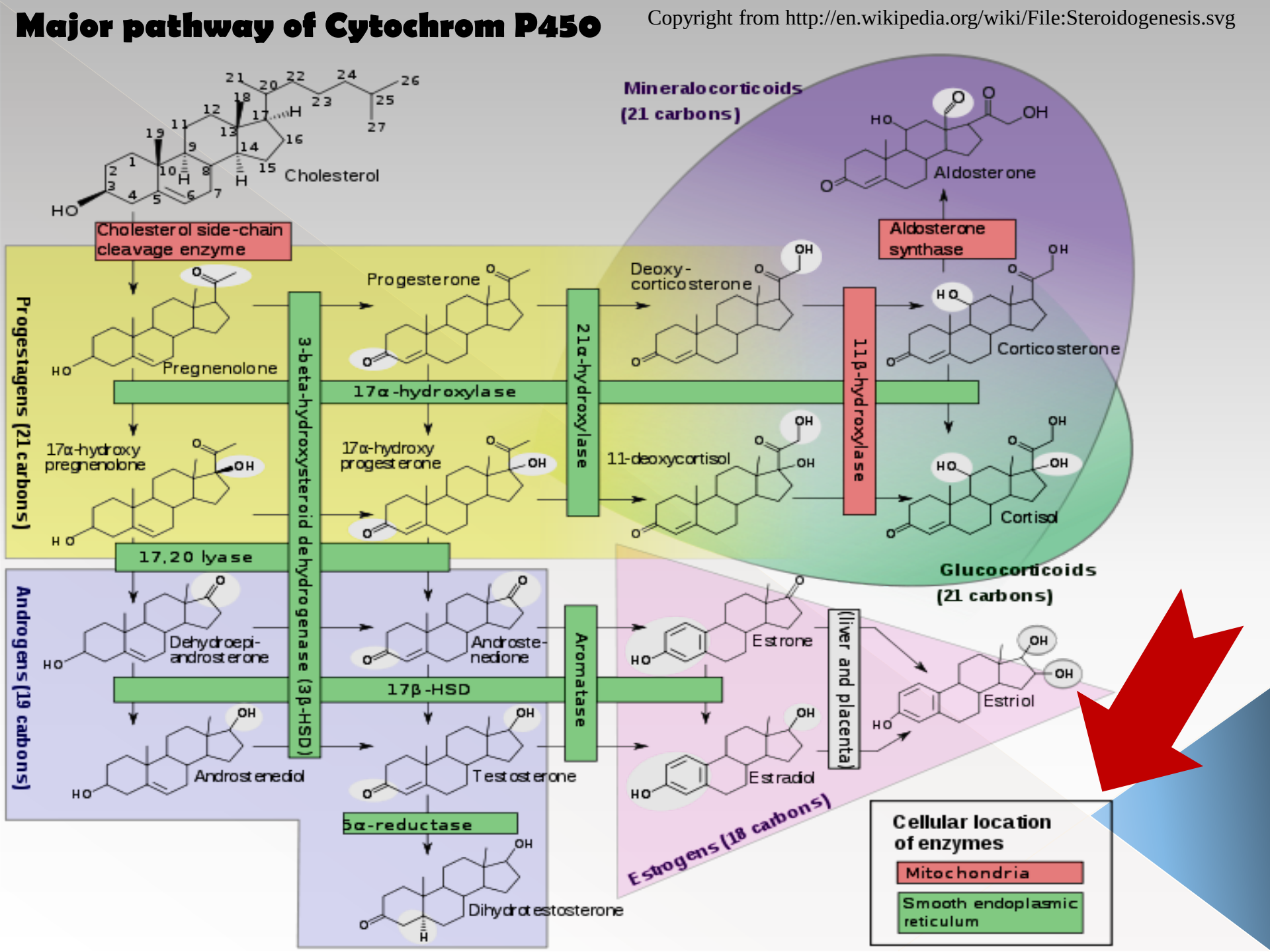
8. Most isoforms of cytochrome P450 are *inducible*
   - example 1: Phenobarbital induces hypertrophy of SER → 3-4 fold increase in cytochrome P450 → ↑metabolism of warfarin → inadequate dosage
   - example 2: Ethanol induces CYP2E1 which metabolizes chemicals found in tobacco smoke → carcinogenicity → ↑risk of cancer from smoking

Characterization of Cytochrom P450

9. Isoforms of cytochrome P450 are particularly involved in the metabolism of *Polycyclic Aromatic Hydrocarbons (PAHs)*
   - formerly called *Aromatic Hydrocarbon Hydroxylases*
   - involved in the metabolism of PAHs and in carcinogenesis produced by PAHs



Major pathway of Cytochrom P450



Characterization of Cytochrom P450

10. Polymorphism

- exist in different forms, some of which exhibit higher or lower catalytic activity
- responsible for the variations of drug responses noted among many patients

Inducer / Inhibitor of Cytochrom P450

Enzyme	Inducers	Inhibitors
CYP1A2	Cigarette Smoke, Phenobarbital, Ritonavir, Carbamazepine, Charbroiled Foods, Vegetables, Omeprazole	Enoxacin, Ciprofloxacin, Grepafloxacin, Fluvoxamine, Fluoxetine, Nefazodone
CYP2A6	Barbiturates	
CYP2C9	Rifampin, Carbamazepine, Ethanol, Phenytoin	Amiodarone, Fluvastatin, Fluvoxamine, Fluoxetine, Fluconazole, Miconazole, Metronidazole, Ritonavir, Sulfamethoxazole
CYP2C19	Rifampin	Fluvoxamine, Fluoxetine, Ticlopidine, Ritonavir
CYP2D6	Pregnancy	Quinidine, Fluoxetine, Paroxetine, Sertraline, Thioridazine, Cimetidine, Diphenhydramine, Haloperidol, Ticlopidine (Ticlid), Ritonavir

Phases of Xenobiotic Metabolism

Phase I – Modification
Phase II – Conjugation
Phase III – Further and Excretion

Phases of Xenobiotic Metabolism

Phase II – Conjugation

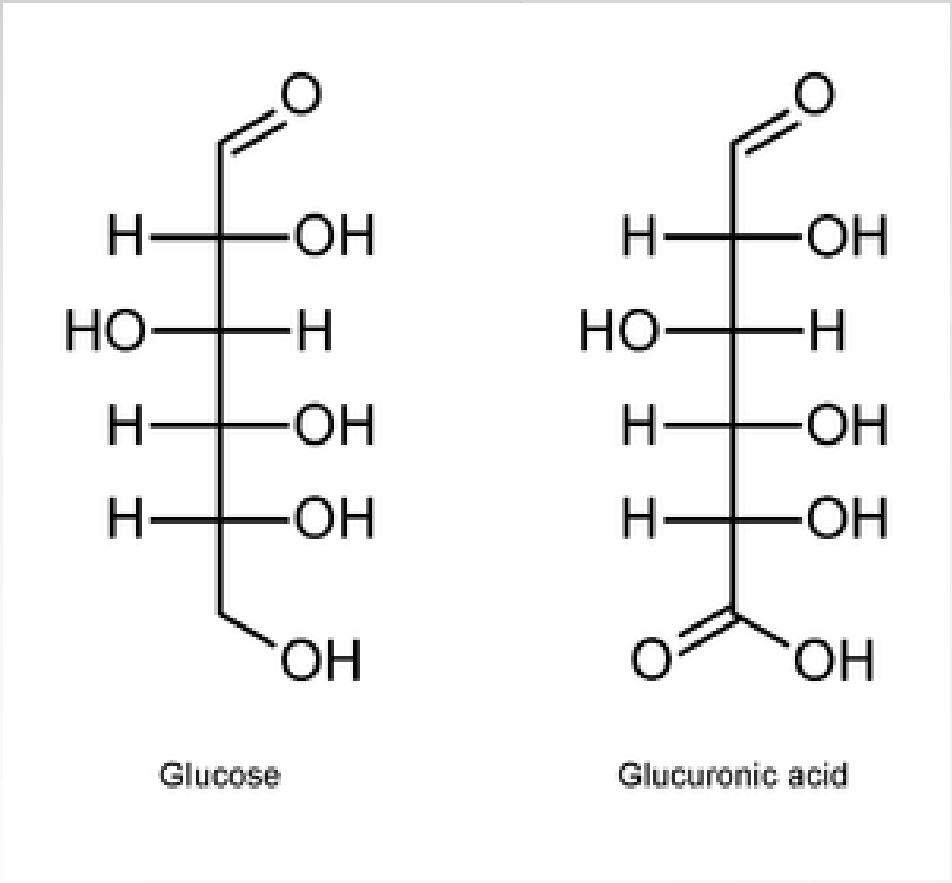
Conjugated with charged compounds

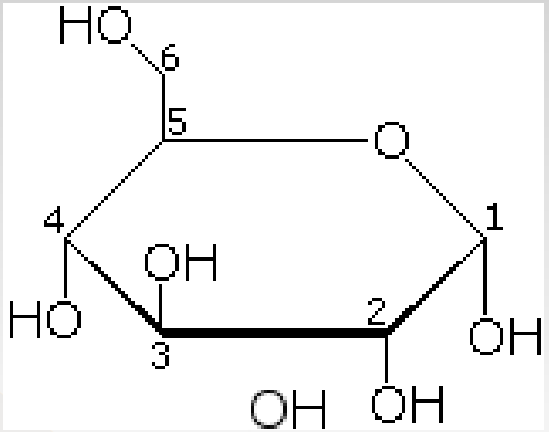
- **Glucuronic acid * glucuronidation**
- **Glutathione (GSH) * GSH conjugation**
- **Sulfate* sulfation**
- **Acety-CoA* acetylation**

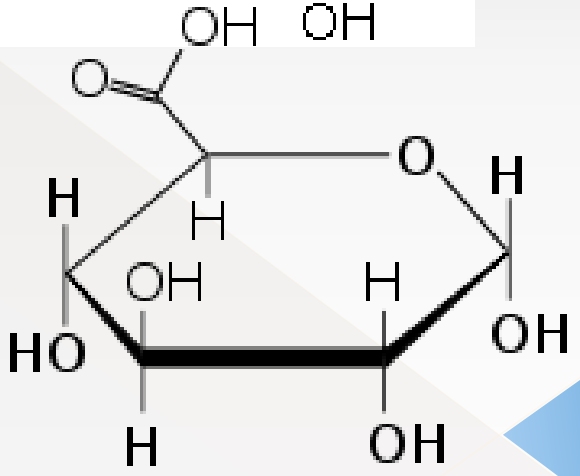
Phase II – Conjugation

1. Glucuronic acid * glucuronidation

glucuronyl donor : UDP- glucuronic acide



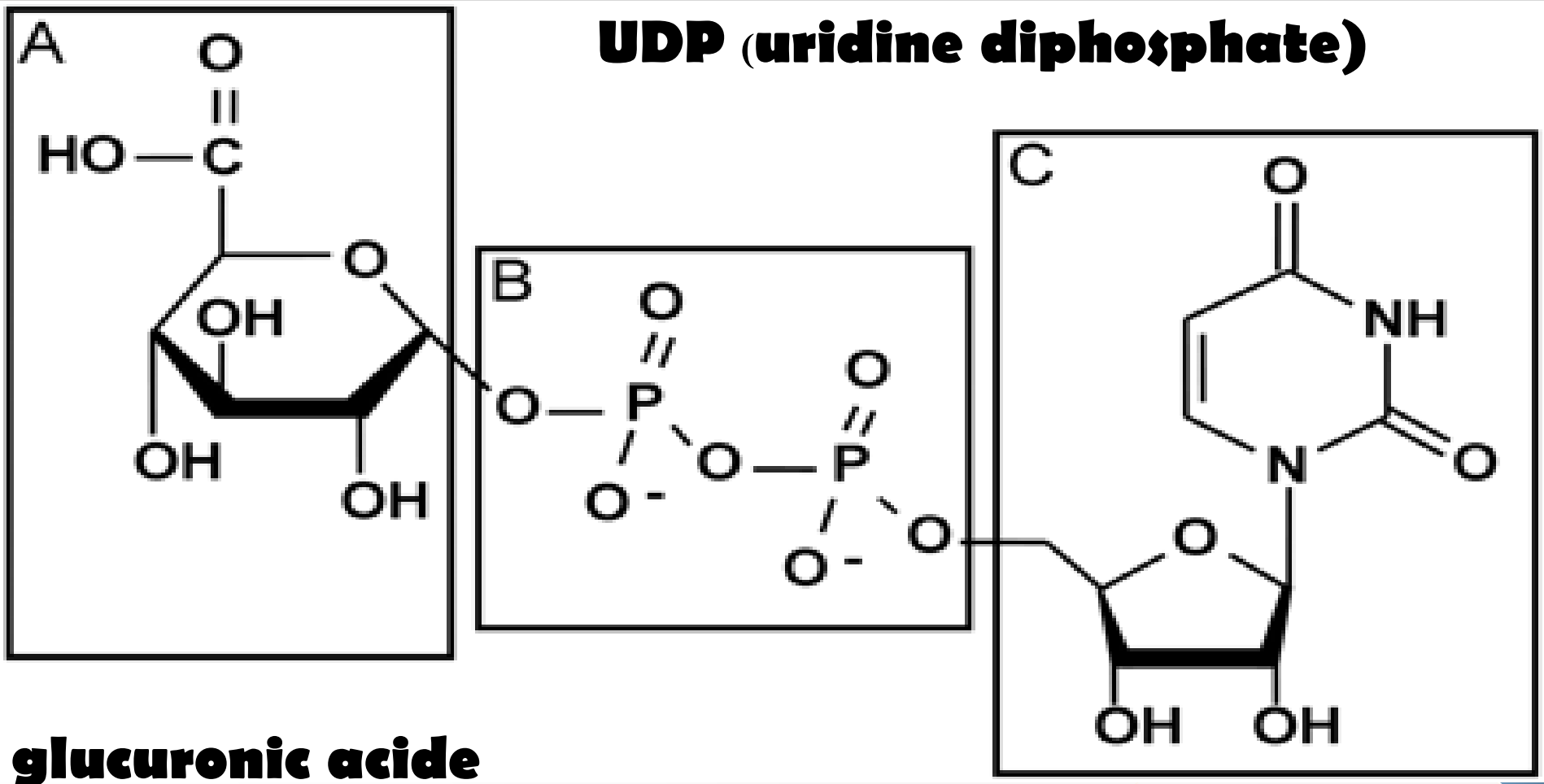




Phase II – Conjugation

1. Glucuronic acid * glucuronidation

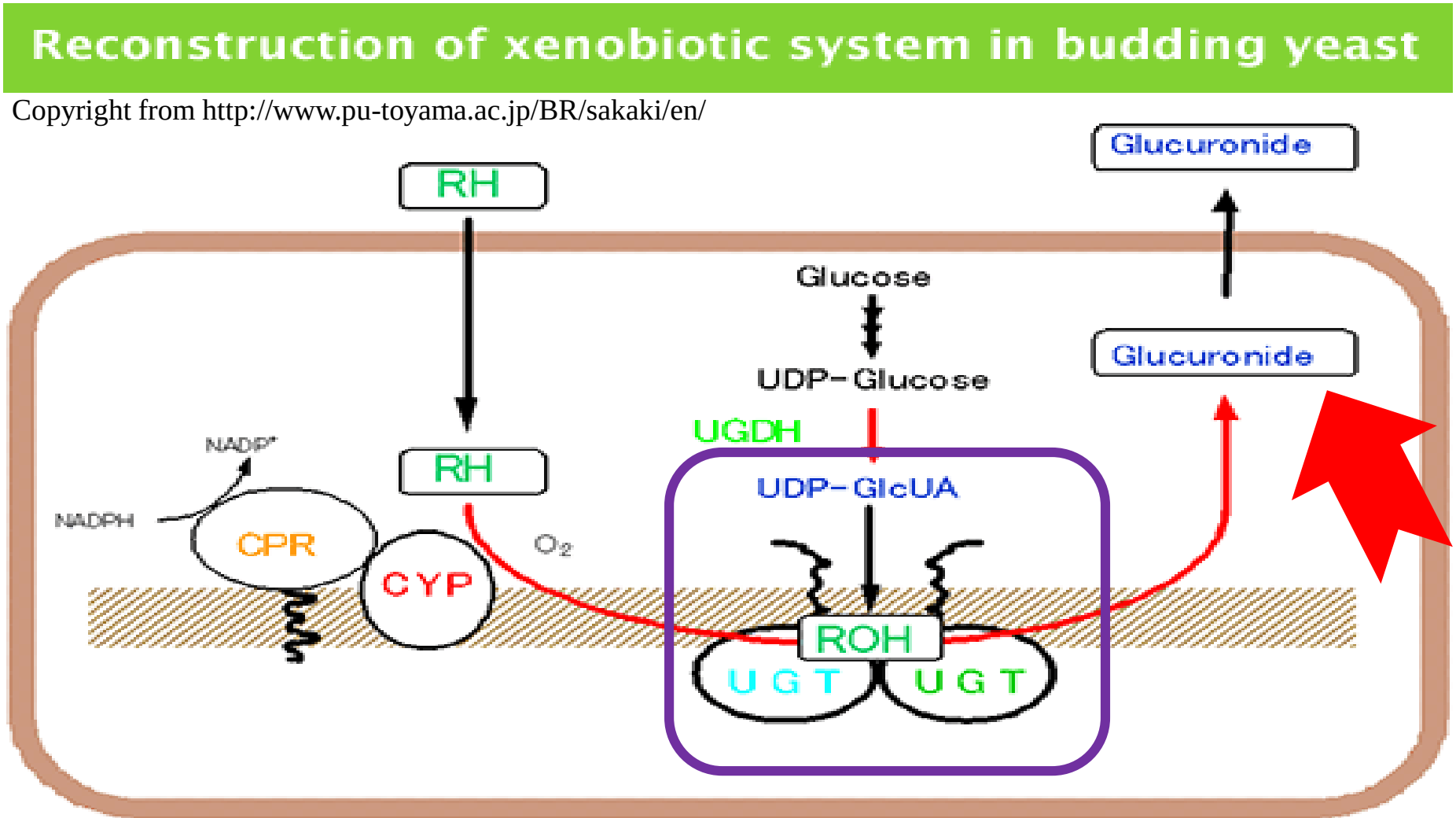
glucuronyl donor: UDP- glucuronic acid



Phase II – Conjugation

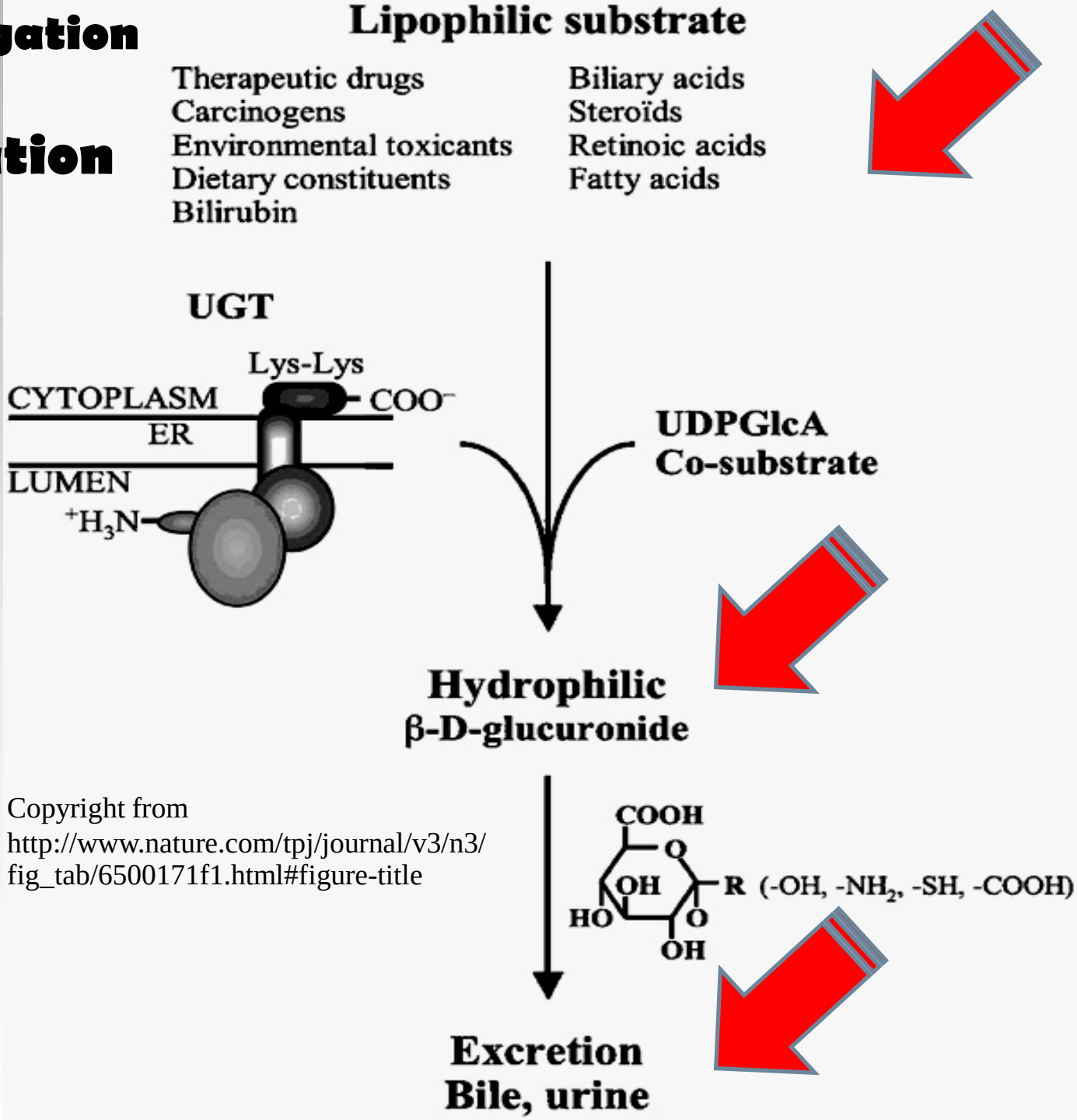
1. glucuronidation

glucuronyl donor : UDP- glucuronic acide



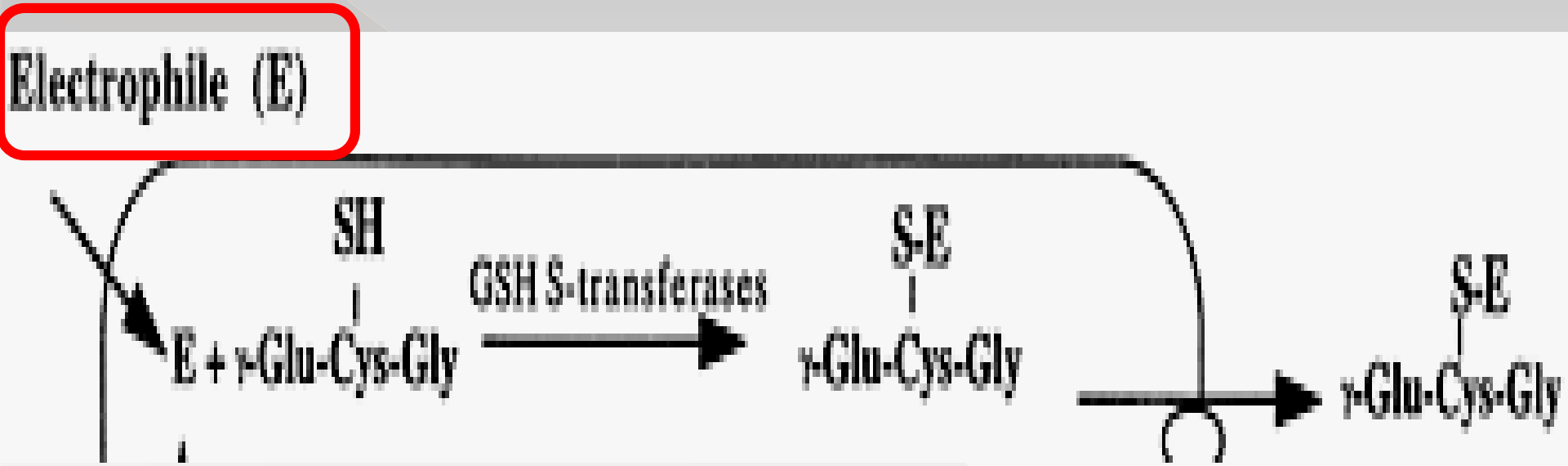
Phase II – Conjugation

1. glucuronidation



Phase II – Conjugation

2. Glutathione (GSH) * GSH conjugation

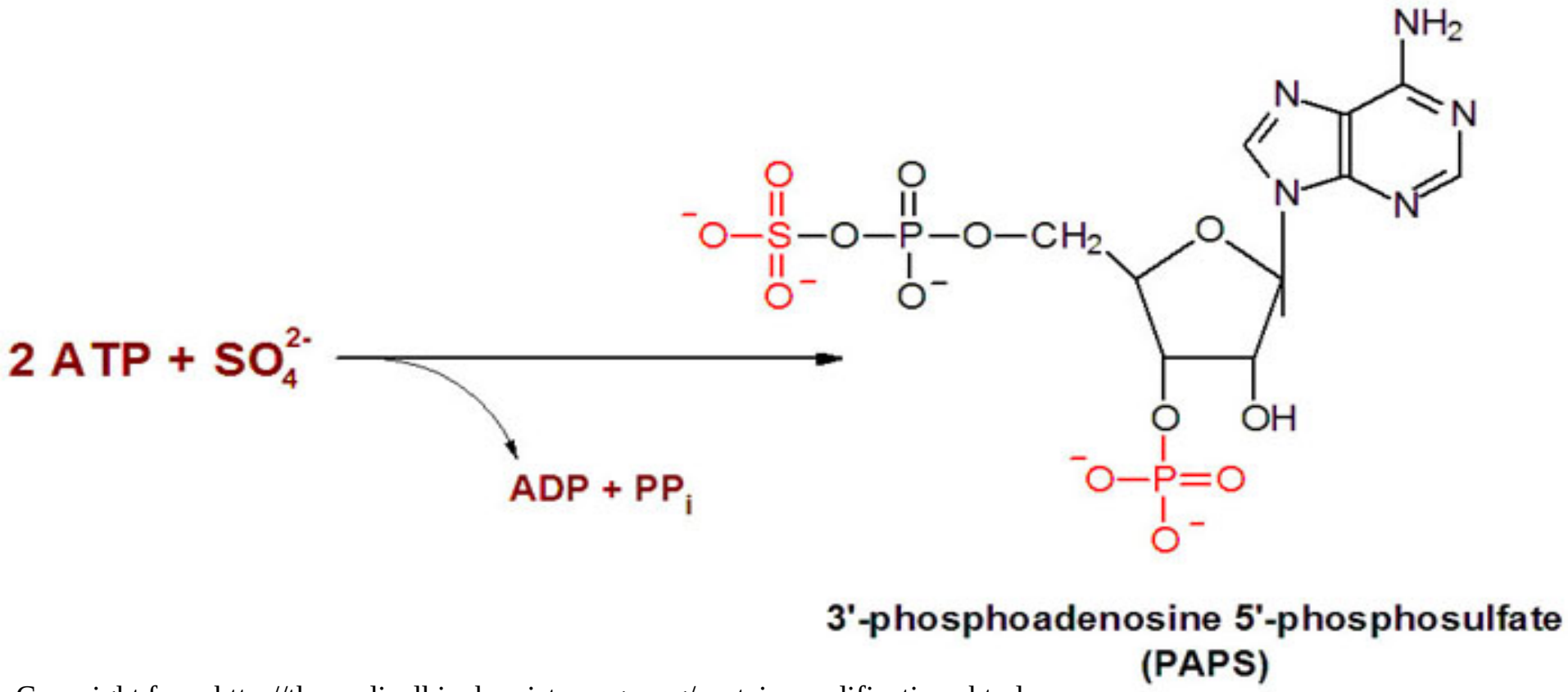


- Specific in electrophilic xenobiotics

Phase II – Conjugation

3. Sulfate* sulfation

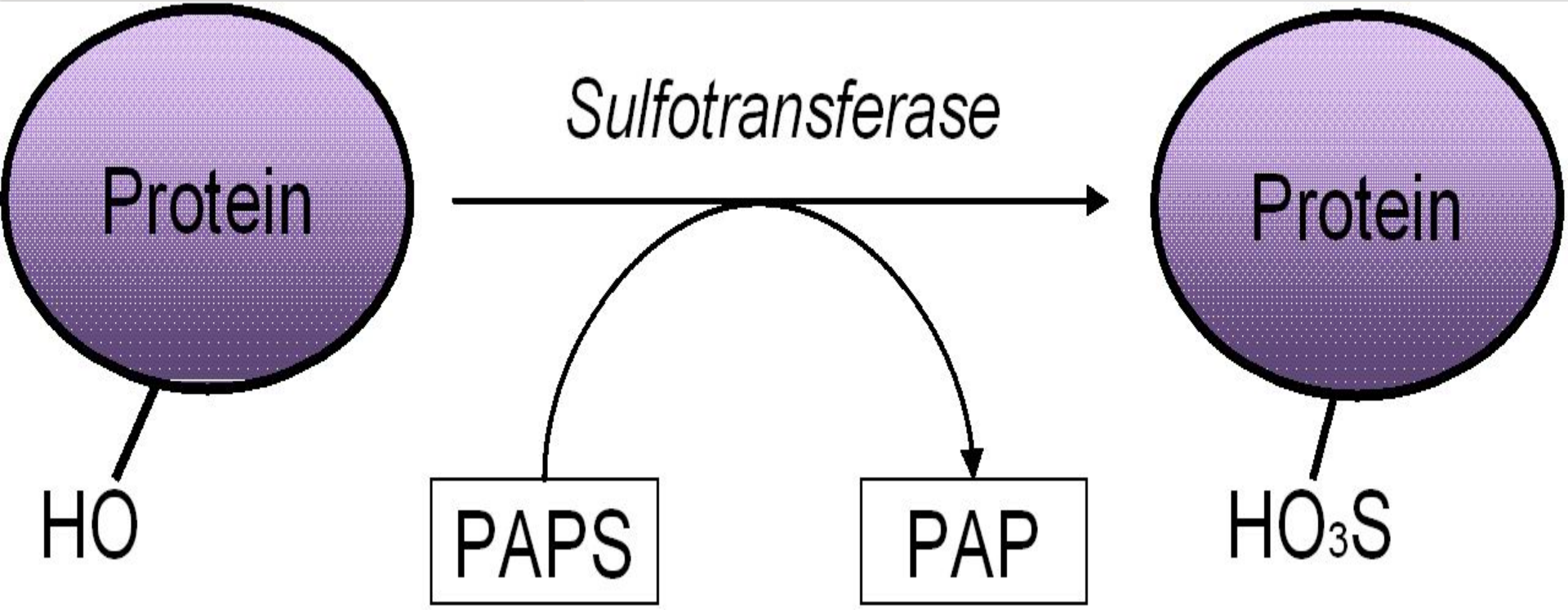
-Sufate donor : PAPS (adenosine 3' -phophate-5-phosphosulfate)



Phase II – Conjugation

3. Sulfate* sulfation

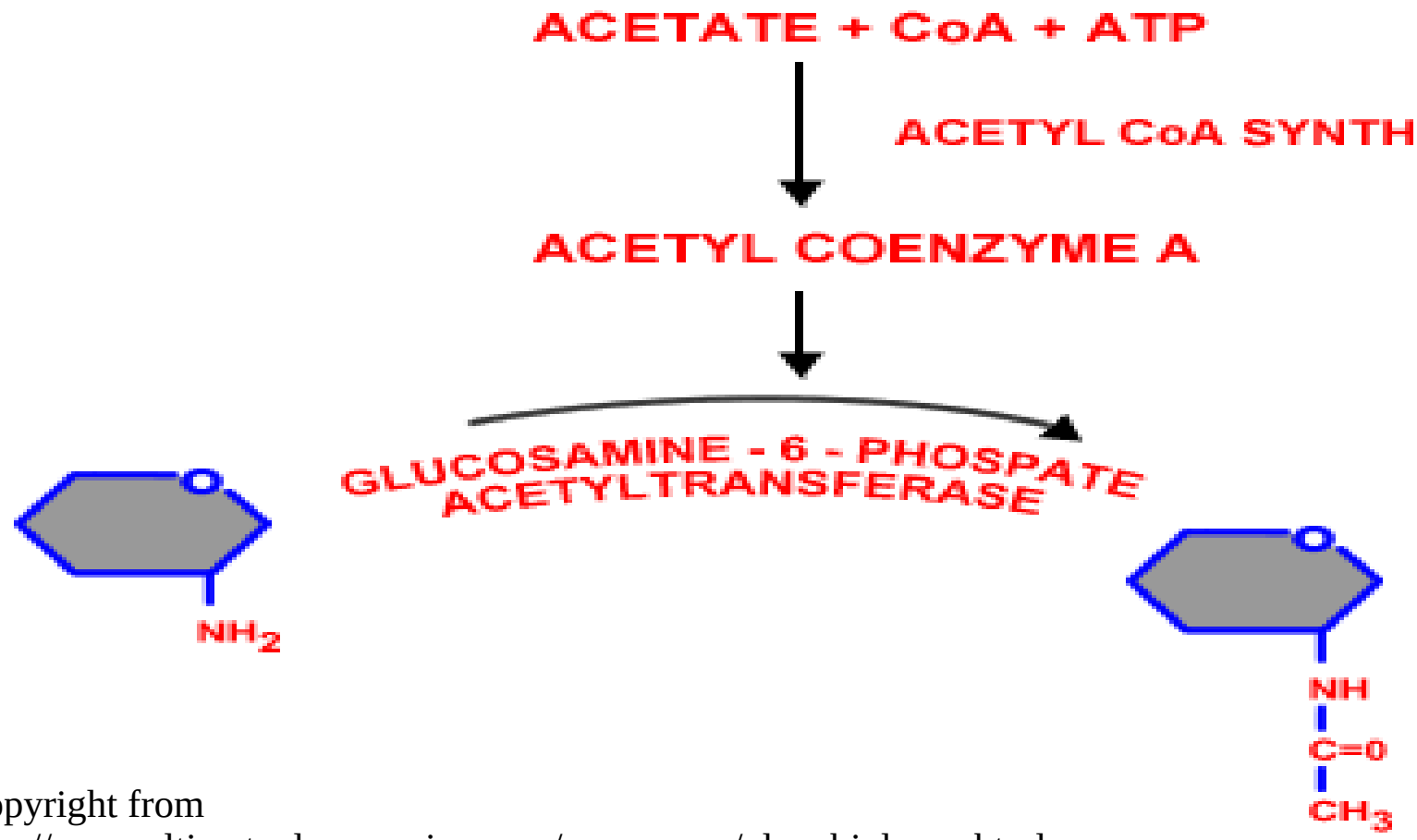
-Sufate donor : PAPS (adenosine 3' -phophate-5-phosphosulfate)



Phase II – Conjugation

4. Acety-CoA* acetylation

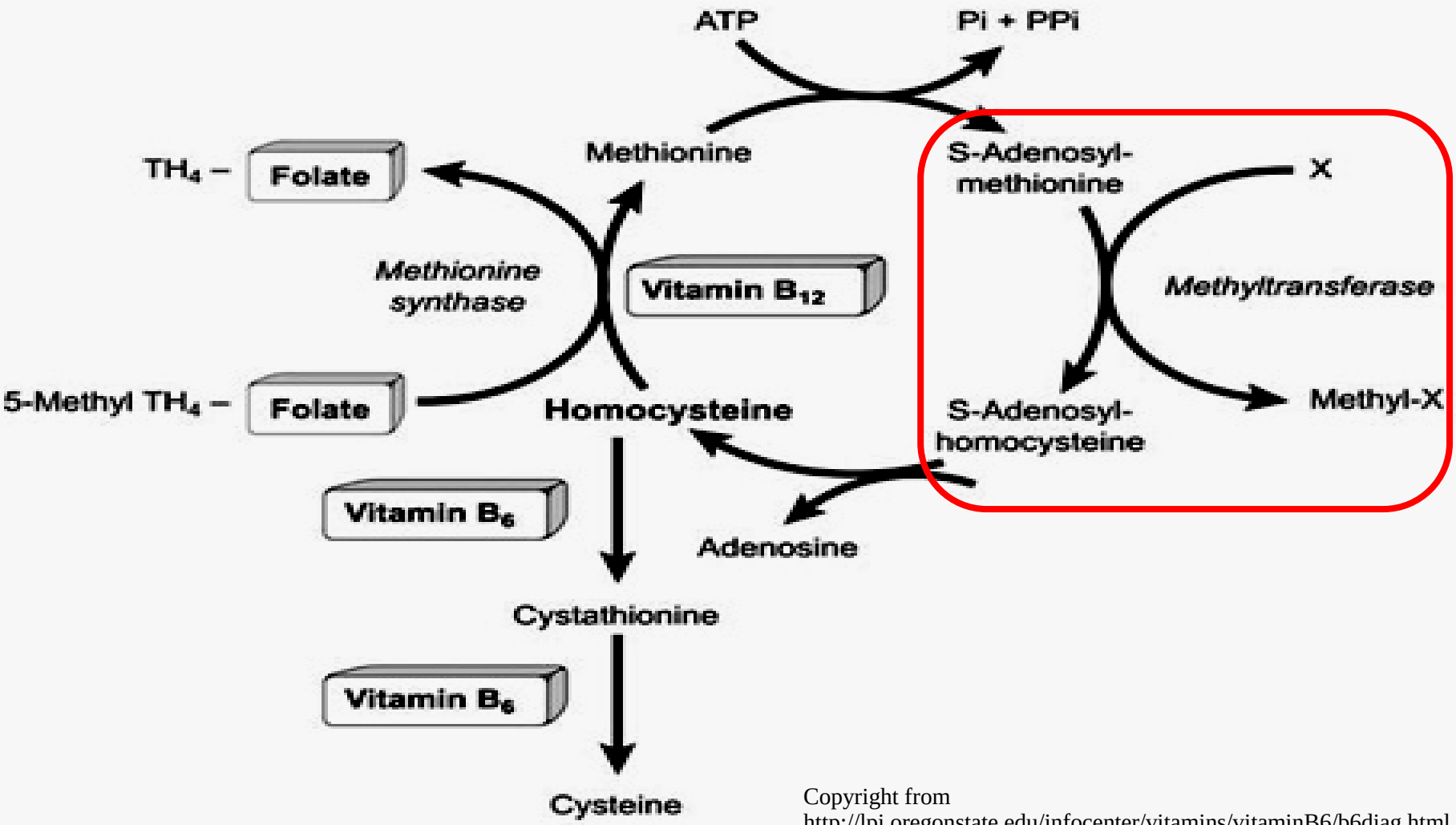
X + AcetylCoA → Acetyl – X + CoA



Phase II – Conjugation

5. Methyl* metylation

Methyl donor : S-adenosylmethionine



Phases of Xenobiotic Metabolism

Phase III – Further and Excretion

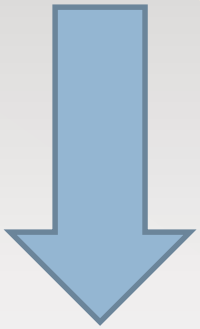
Gamma-glutamyltransferase or **gamma-glutamyl transpeptidase** (also **γ-glutamyltransferase**, **GGT**, **GGTP**, **gamma-GT**) (EC 2.3.2.2)

- Transfer gamma-glutamyl functional groups across the cellular membrane

- A diagnostic marker in medicine as same as alkaline phosphatase (ALP)

Phase III – Further and Excretion

(5-L-glutamyl)-peptide + an amino acid

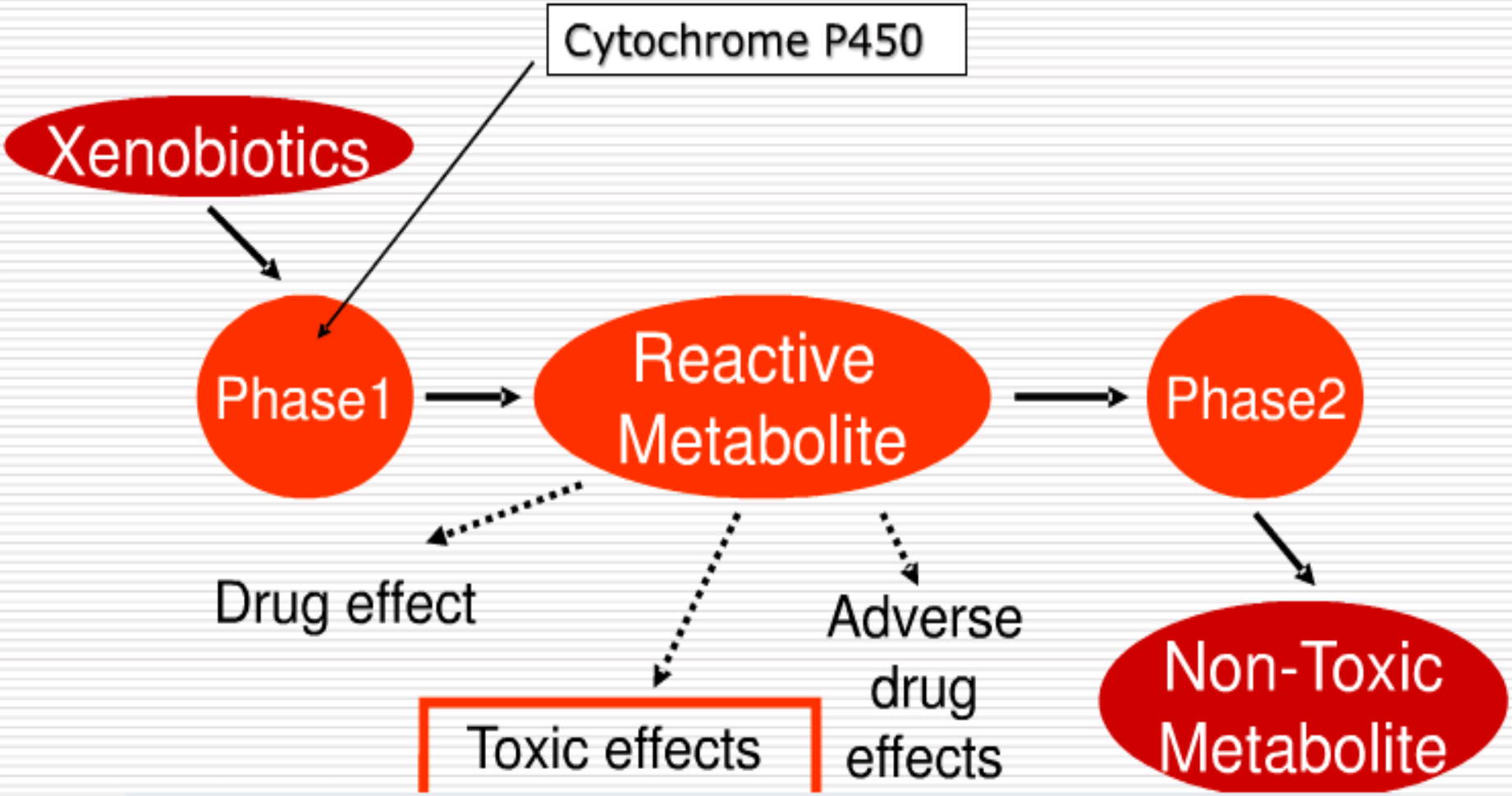


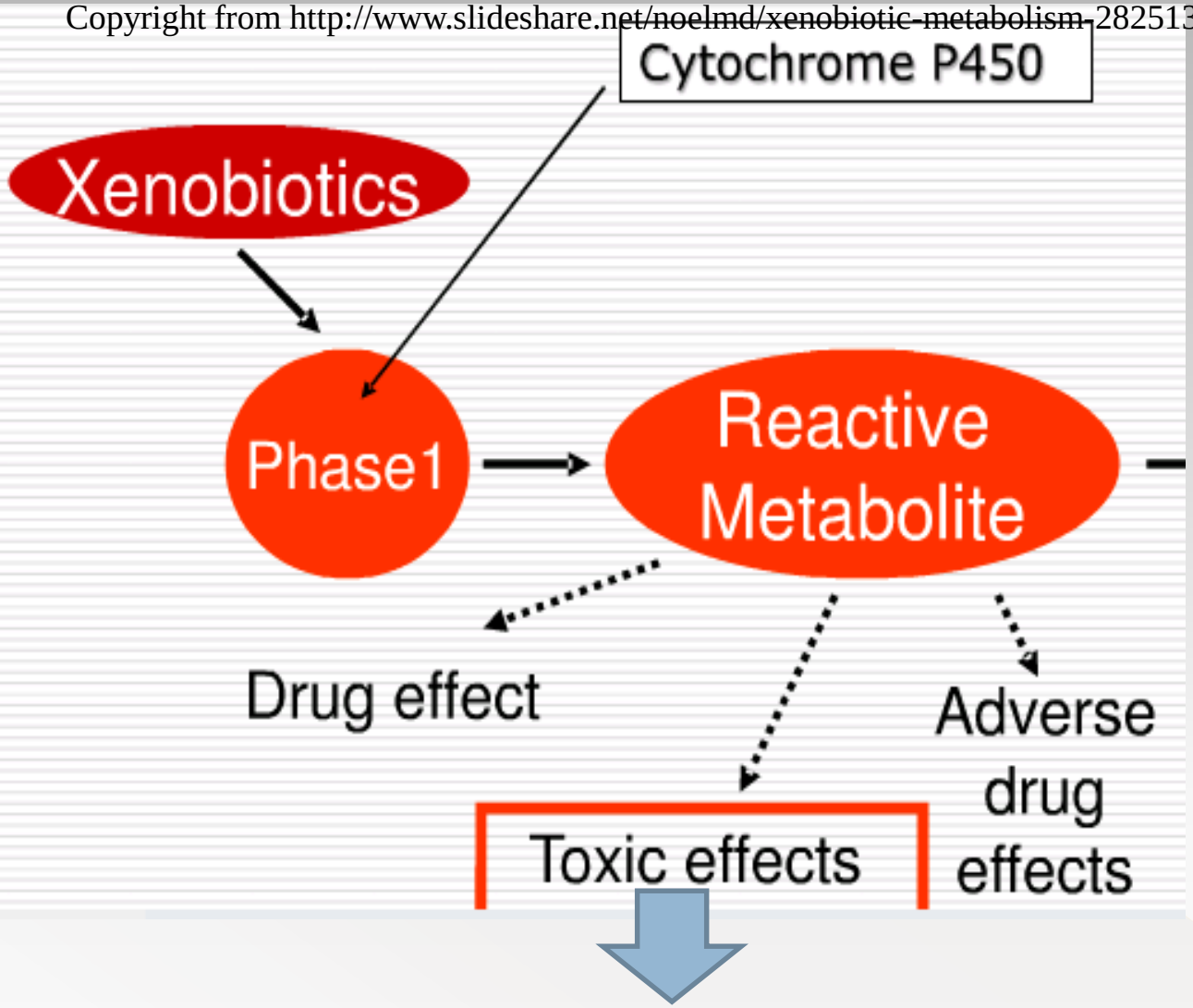
Gamma-glutamyltransferase
(γ-glutamyltransferase, GGT)

peptide + 5-L-glutamyl amino acid

Phases of Xenobiotic Metabolism)

Cellular Response to Xenobiotics





Covalent binding to macromolecule

1. Cytotoxicity
2. Immunologic damage
3. Mutation

Covalent binding to macromolecule

1. Cytotoxicity

A. Directly cytotoxic

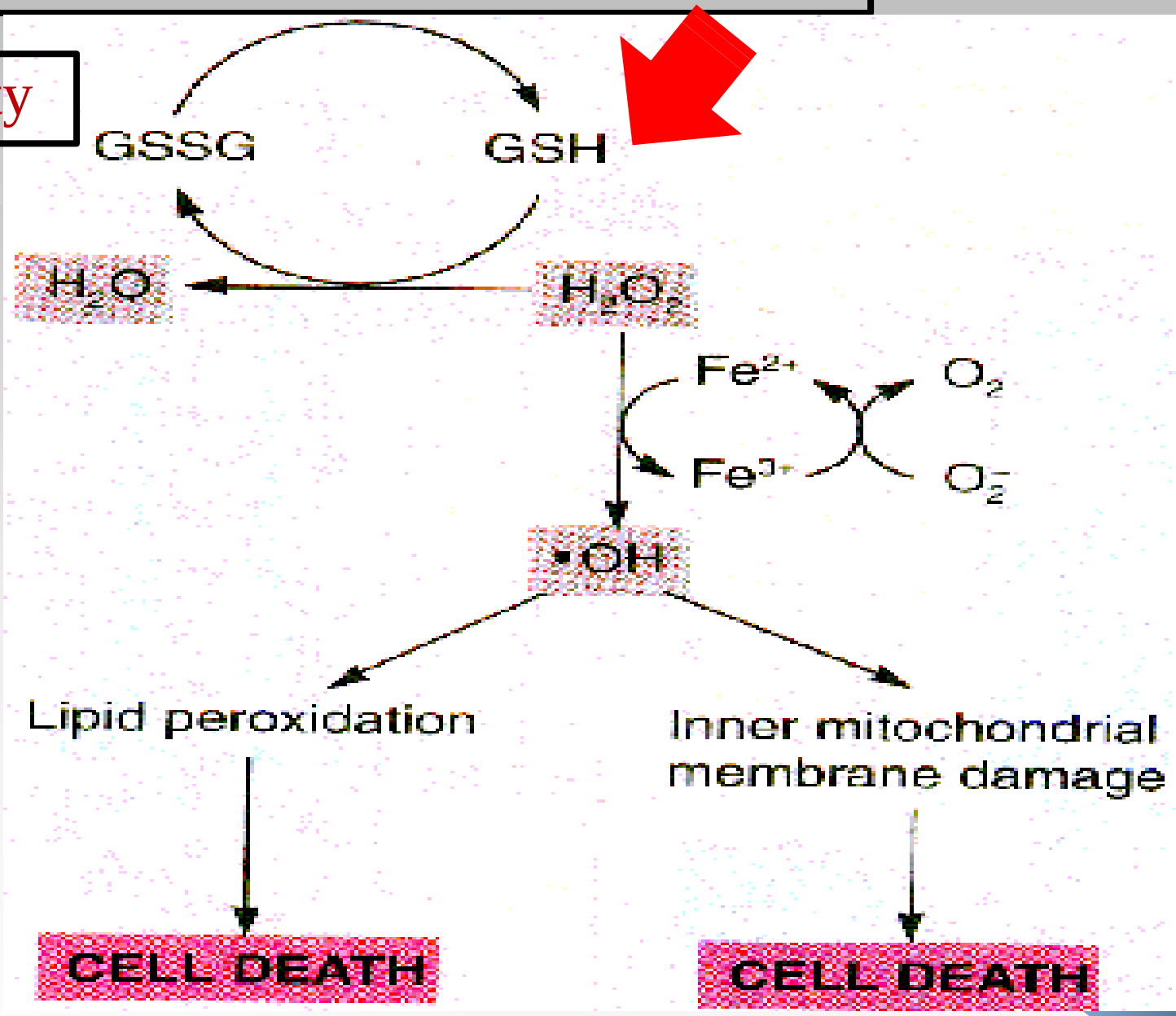
e.g. Mercury, lead, iron, cancer chemotherapeutic agents

- directly interacting with plasma membrane

- interacting with glutathione and weakening antioxidant defences causing plasma membrane injury

Covalent binding to macromolecule

1. Cytotoxicity



Covalent binding to macromolecule

1. Cytotoxicity

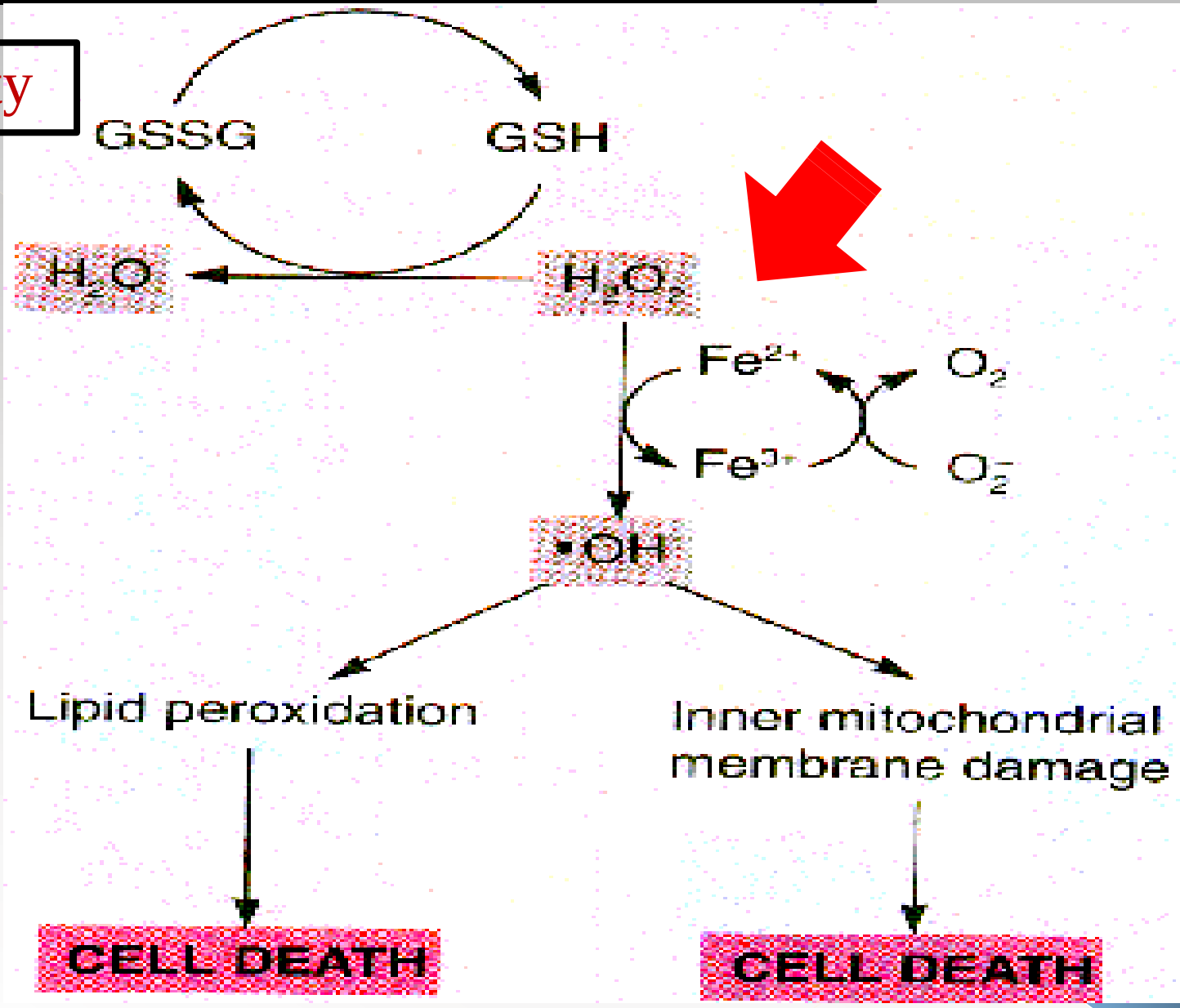
B. Indirectly Cytotoxic
- toxic metabolites
e.g. Acetaminophen, carbon tetrachloride, bromobezene

-metabolised by the mixed function oxidase system of SER

-membrane damage is initiated by metabolites or by activated oxygen species formed during the metabolism of the toxin.

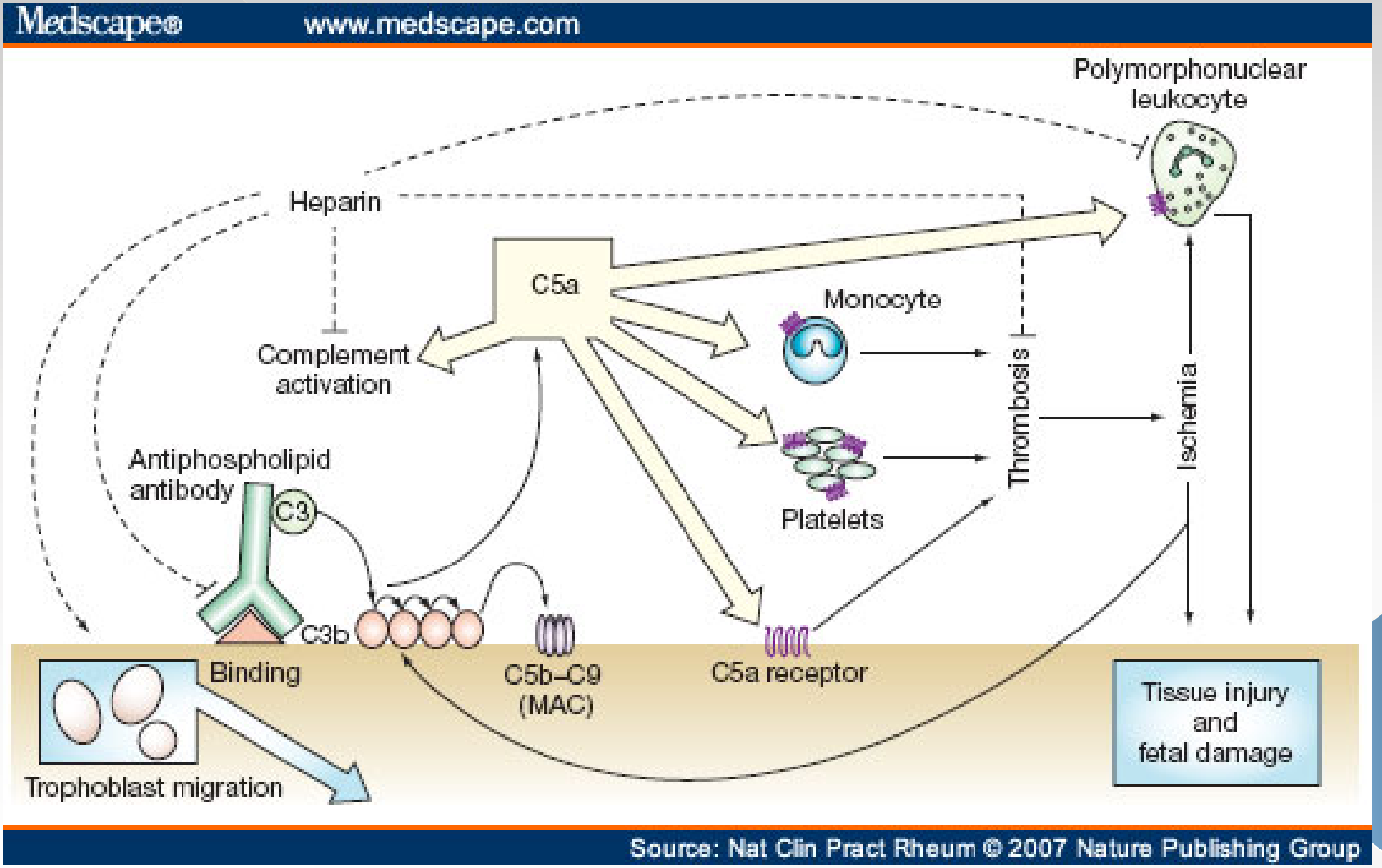
Covalent binding to macromolecule

1. Cytotoxicity



Covalent binding to macromolecule

2. Immunologic damage



Covalent binding to macromolecule

3. Mutation

GENETIC (DNA) ALTERATION

Causes of DNA abnormalities

-Acquired genetic abnormalities i.e. acquired during life due to external factors

Effects of DNA abnormalities

- Failure of synthesis of structural proteins

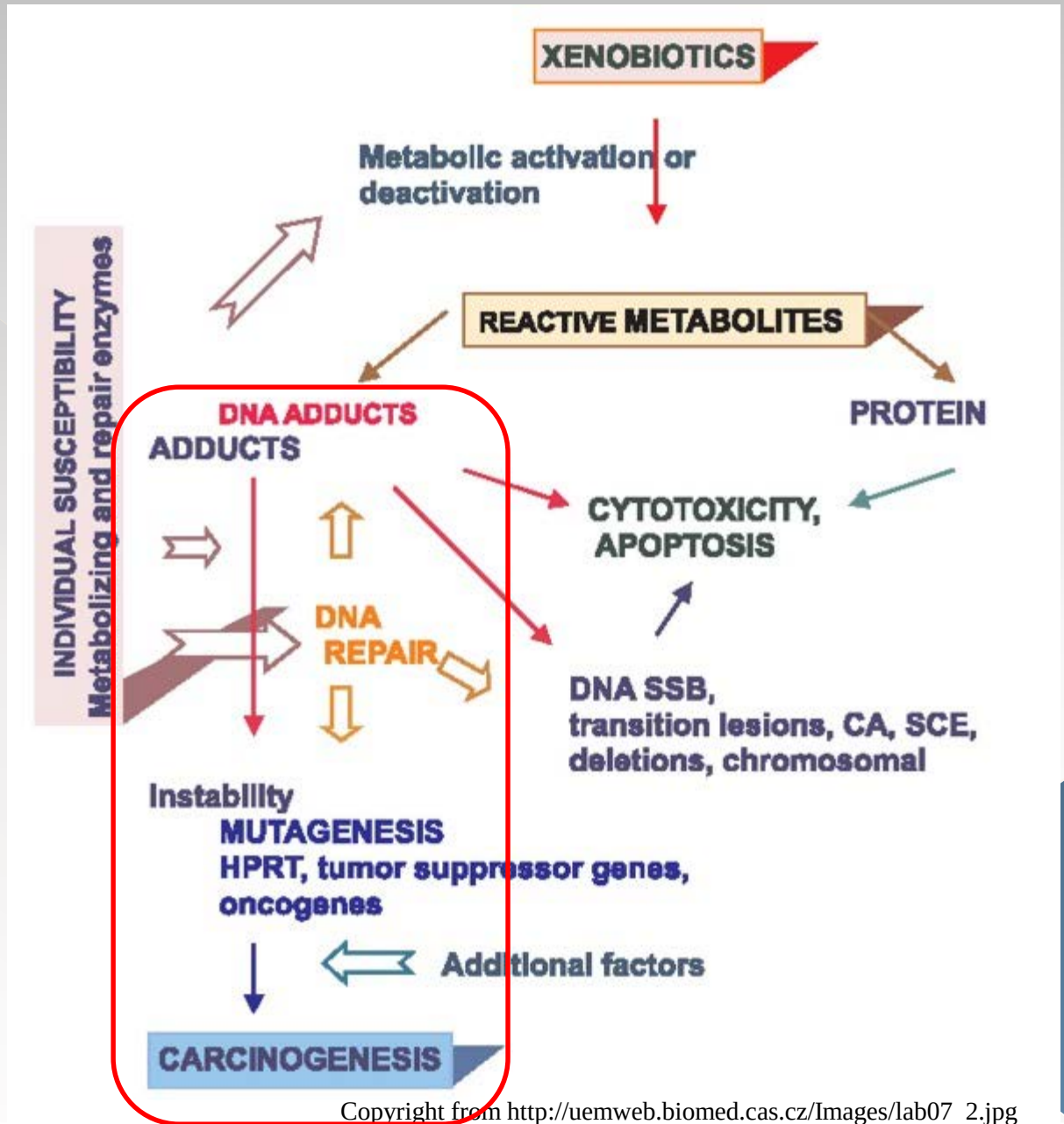
- Failure of mitosis

- Failure of growth-regulating proteins

- Failure of enzyme synthesis

Covalent binding to macromolecule

3. Mutation



Individual Susceptibility

Genetics-species, strain variation, interindividual variations (yet still can extrapolate between mammals--similar biological mechanisms)

Gender (gasoline nephrotox in male mice only)

Age--young (old too)

underdeveloped excretory mechanisms
underdeveloped biotransformation enzymes
underdeveloped blood-brain barrier
changes in excretion and metabolism rates, body fat

Individual Susceptibility

- Nutritional status
- Health conditions
- Previous or Concurrent Exposures
  - additive --antagonistic
  - synergistic

Lucky in Final Biochemical Examination