Industrial Toxicology

Learning Objectives

- Know the assumptions of the dose-response and time-course curves
- Be able to define and label key points of a curve
- Know the difference between potency and efficacy
- Understand the basics of chemical interactions and how to determine their relationship

Learning Objectives

- Understand the properties of a chemical that influence absorption, metabolism, distribution, and excretion
- Understand the properties of biological systems that my influence the absorption, metabolism, distribution, and excretion of a chemical

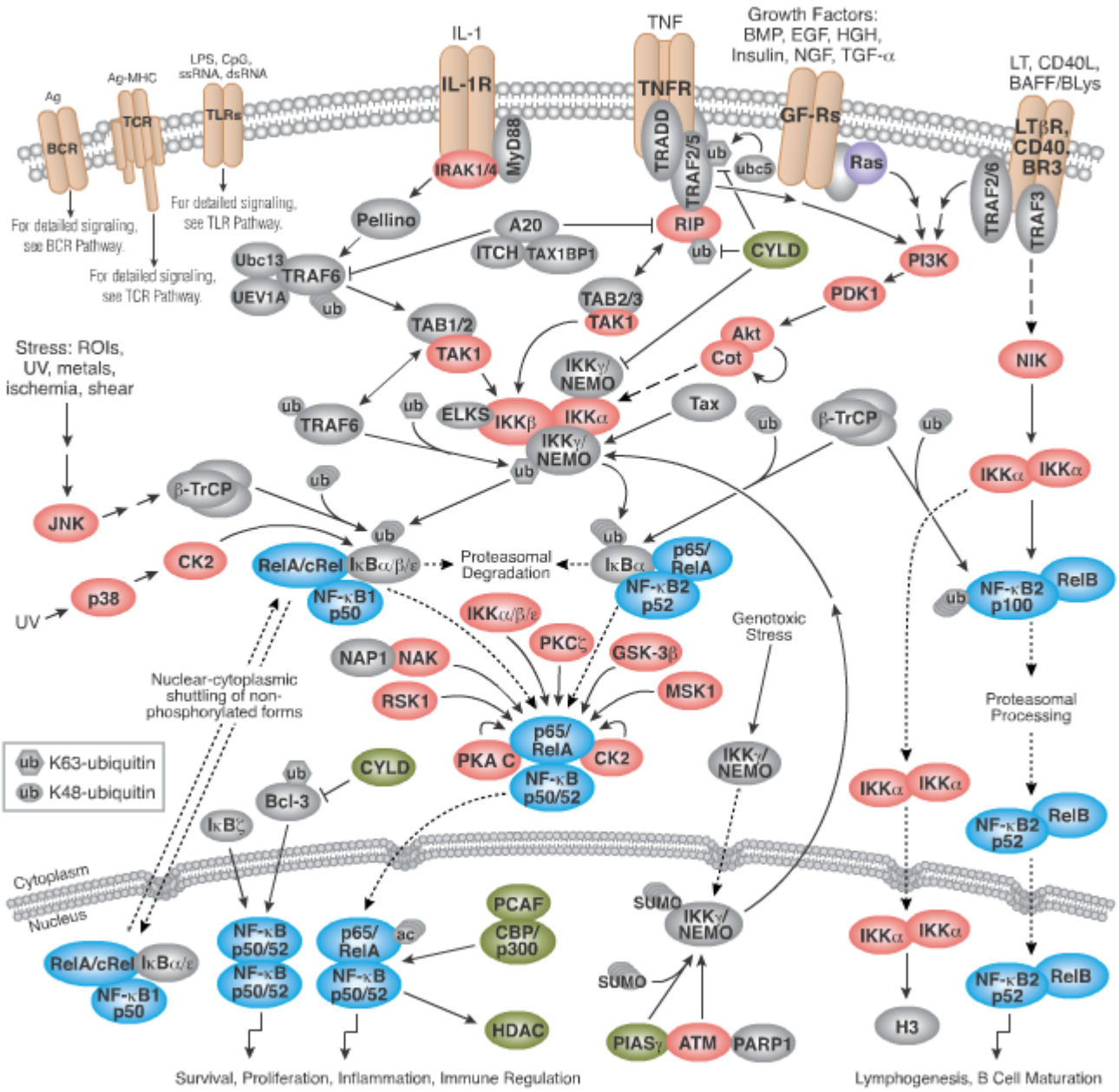
The Father of Toxicology

- Paracelsus (1493-1541)
  - *Bergsucht* (1533-1534)
    - Chronic arsenic, mercury, and miner's disease
    - "No substance is a poison by itself. It is the dose (the right amount of exposure) that makes a substance a poison"
    - "The right dose differentiates a poison and a remedy"

Assumptions of Dose-Response Relationships

1. relationship between chemical and response is causal.
2. magnitude of the response is related to dose.
   a. there is a receptor or other site that the chemical interacts with to produce an effect.
   b. the response and the magnitude of the response are related to the concentration of the agent at the site of action.
   c. the concentration at the site of action is related to the dose administered.
3. existence of a quantifiable, precise method of expressing and measuring the effect.

NF-κB Signaling

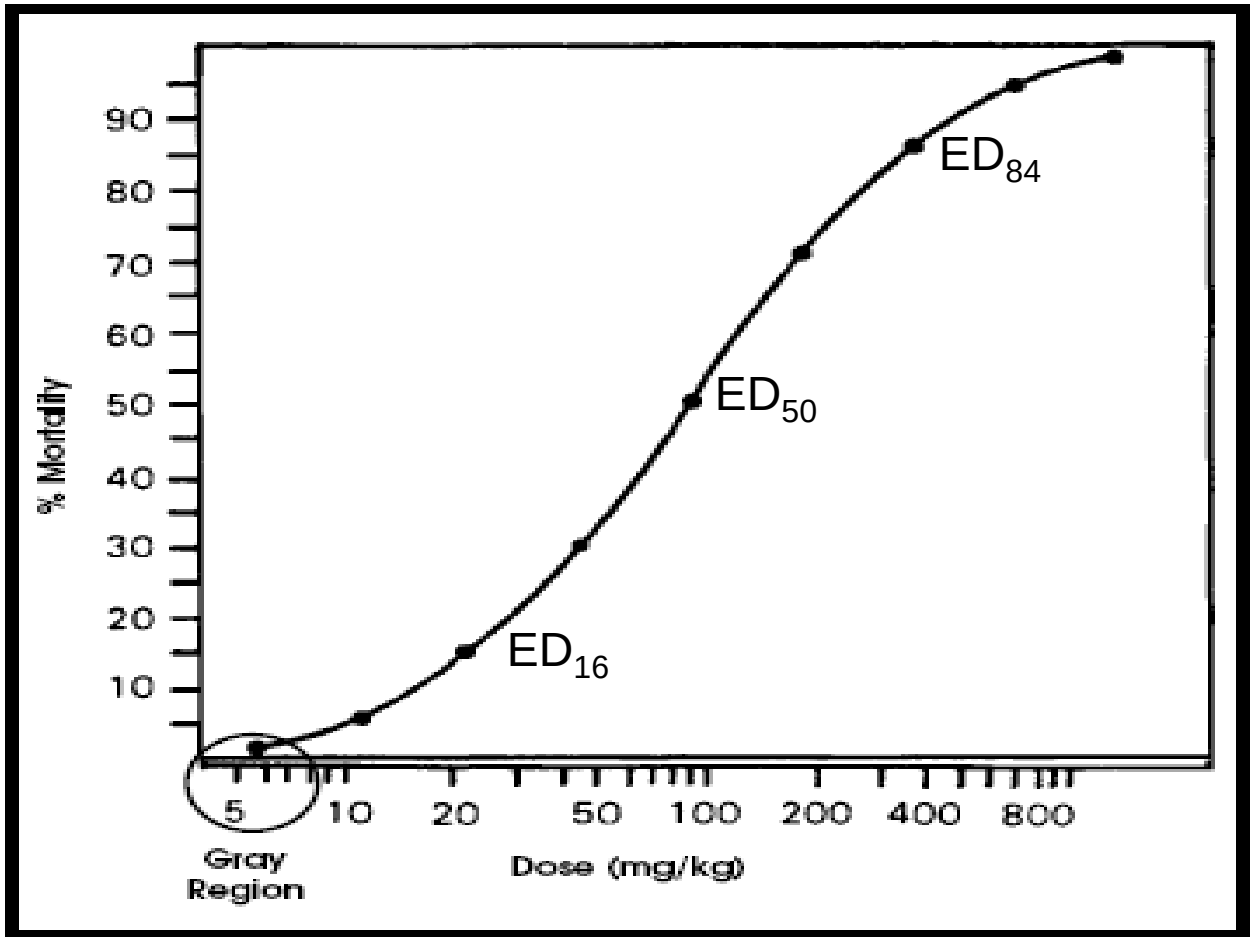


Point Estimates on the Dose-Response Curve

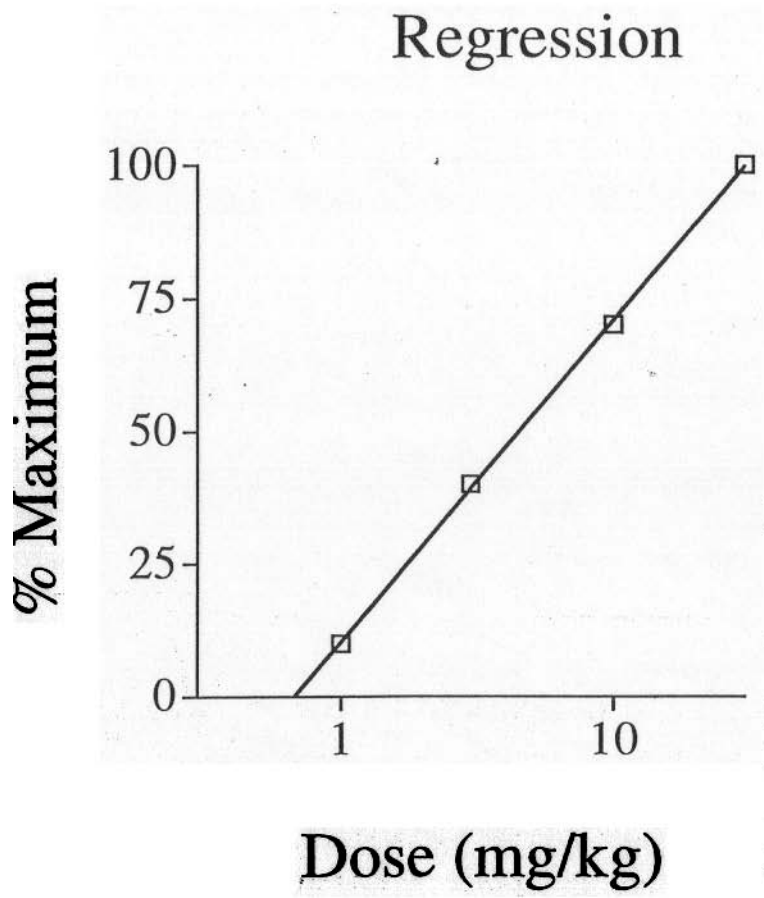
- ED_{50}: median effective dose
- LD_{50}: median lethal dose, used to assess acute toxicity

- Within the linear range of a curve
  - Threshold dose: the lowest dose that elicits a response (ED_{16} or LD_{16})
  - Emax: The magnitude of the maximum response (ED_{84} or LD_{84})

Classic Dose-Response Curve



Determining the ED_{50} Value



y = a + bx
a = interecept
b = slope
x = log dose

y = 60.534LOG(x) + 10.292
r = 1.000

To obtain ED50:
y = 50
50 = 60.5Log(x) + 10.3
.656 = Log(x)
x = 4.5 mg/kg

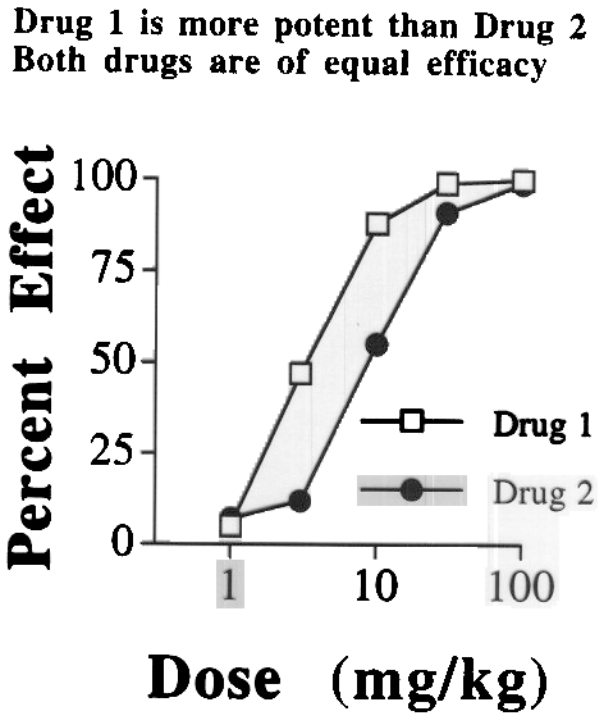
or ED84:
84 = 60.5Log(x) + 10.3
1.22 = Log(x)
x = 16 mg/kg

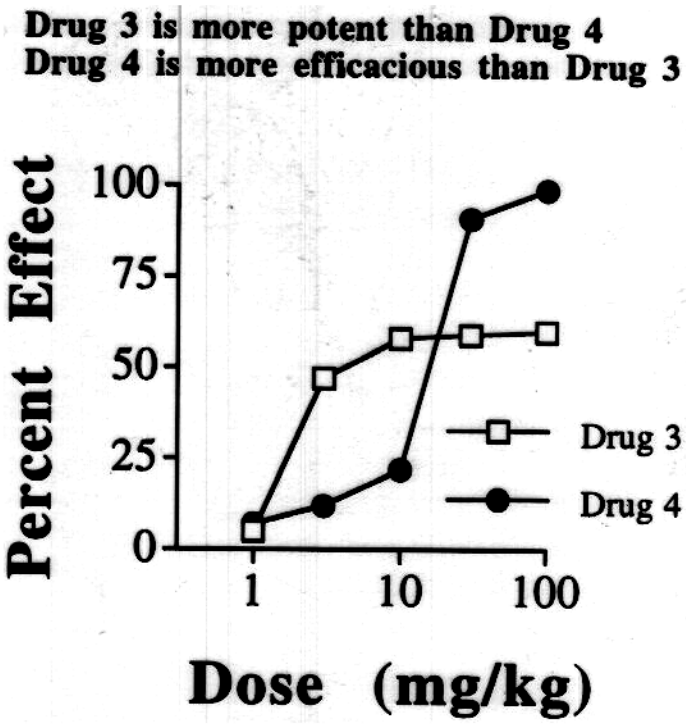
Comparison of Dose-responses

- Efficacy: the magnitude of the maximum response.
- Potency: Strength of a drug or chemical in producing a given effect.

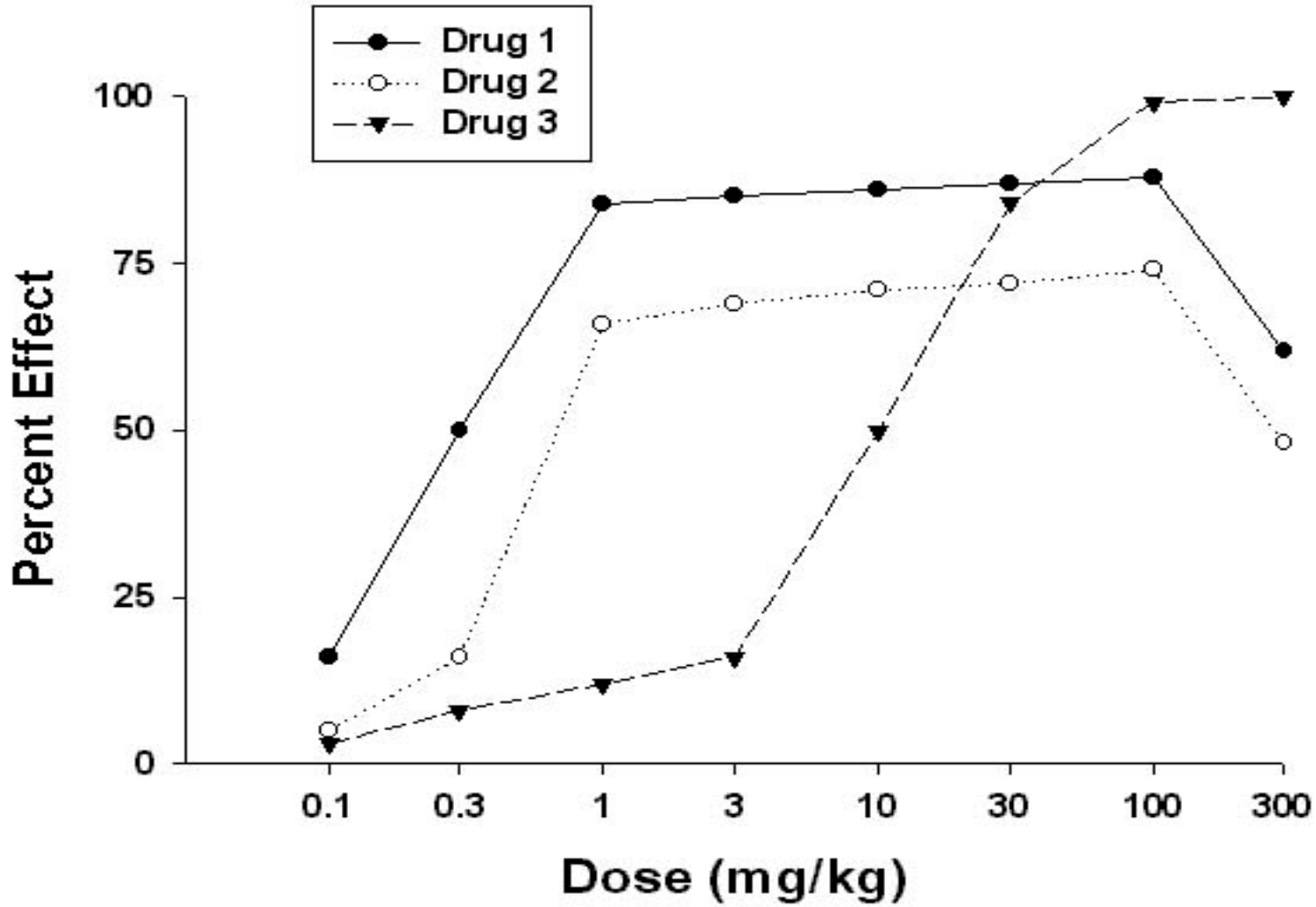
- Therapeutic Index: LD_{50} ED_{50}
- Margin of Safety: LD_{1} ED_{99}

Potency Versus Efficacy



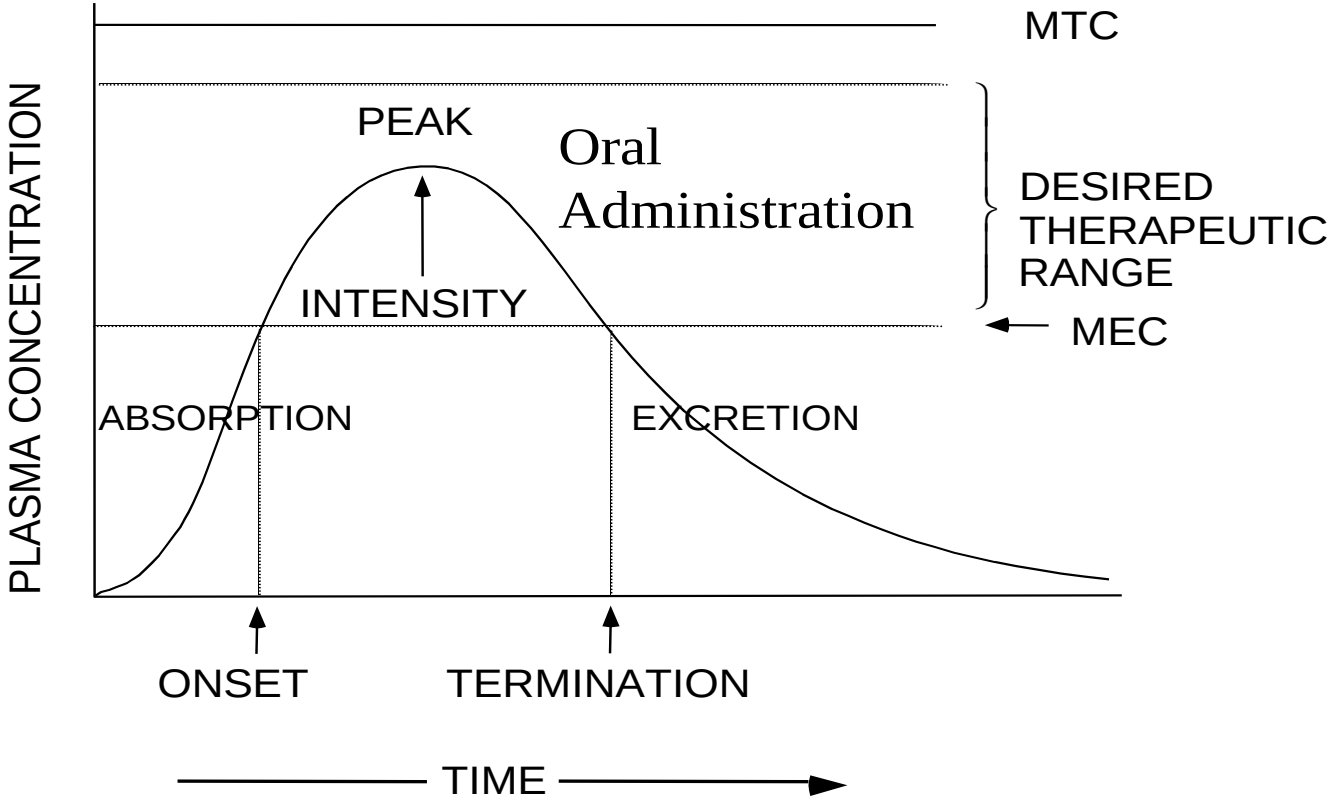


Which Drug Is the Most Potent? Which Drug Is the Most Efficacious?



Time Course

Blood-Drug Concentration Time Curve

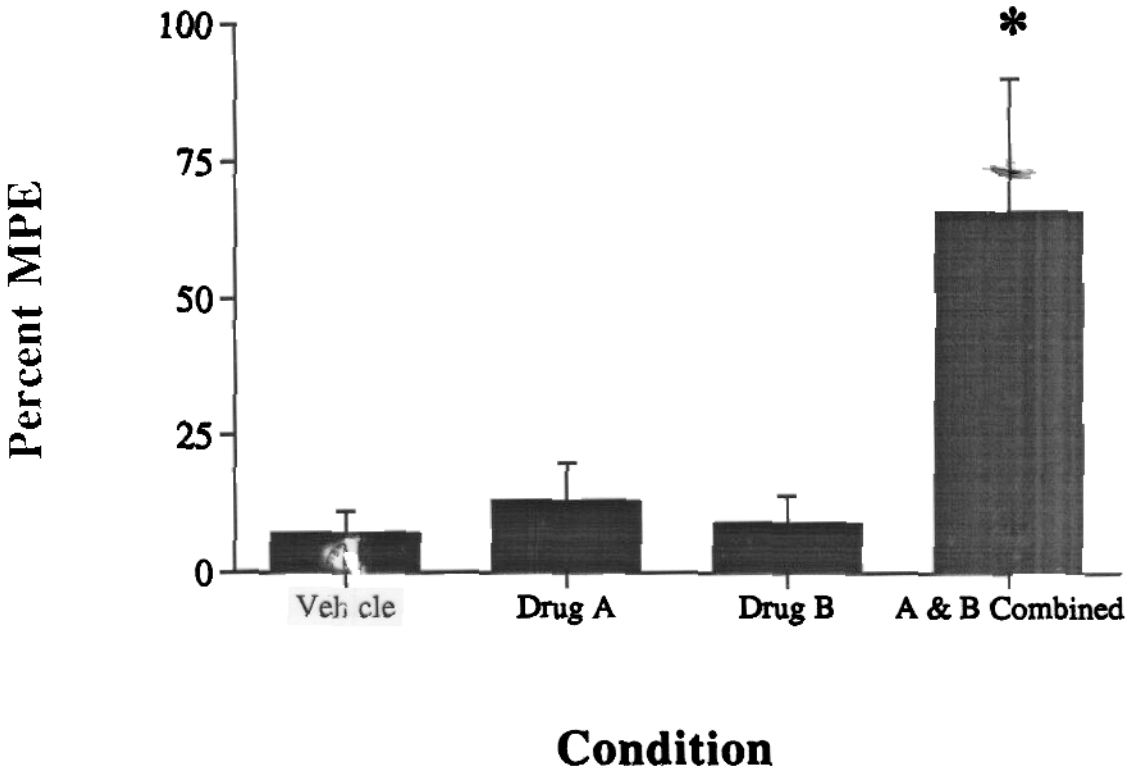


Drug effect is proportional to level of drug in plasma.

General Types of Chemical Interactions

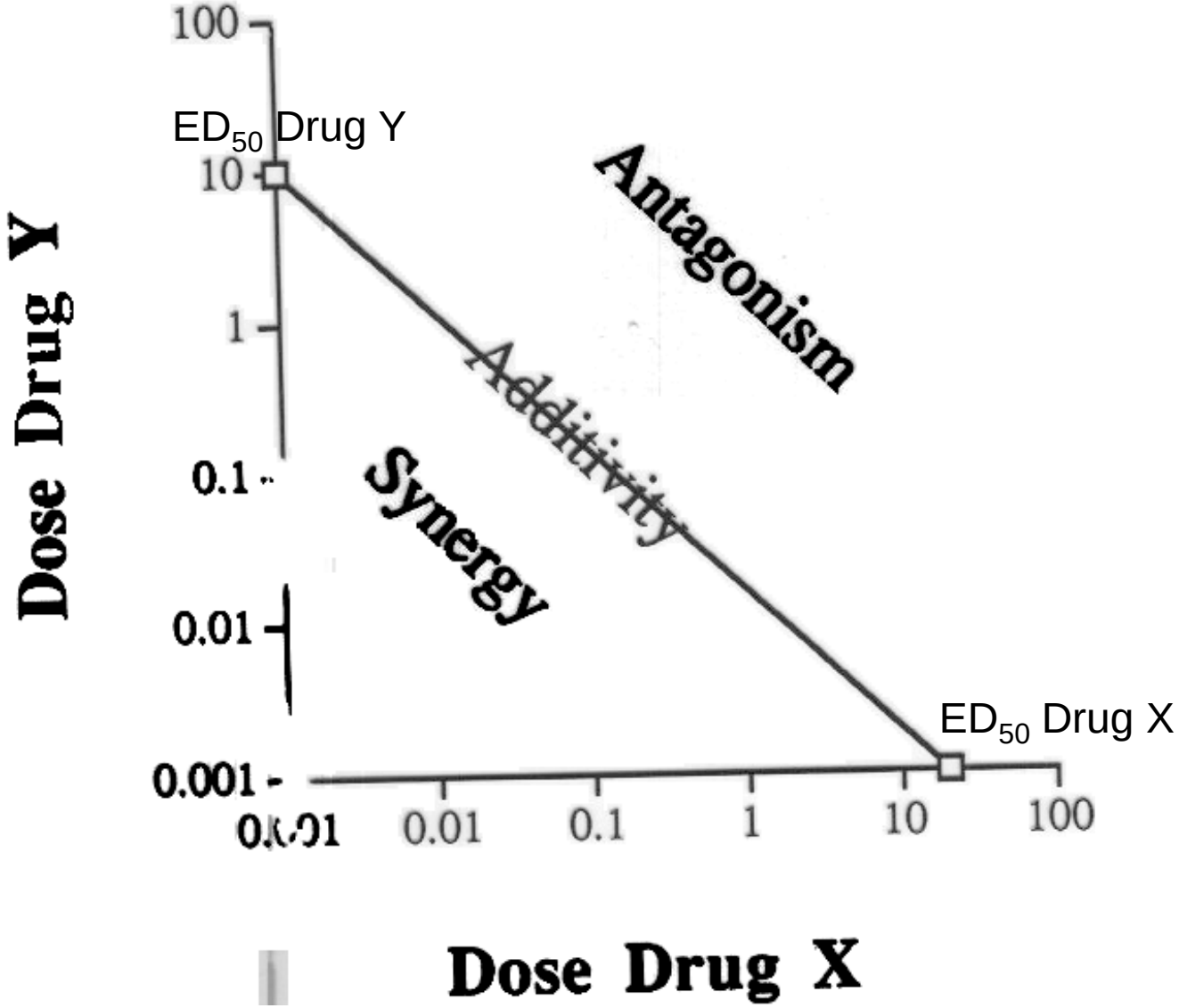
- 1) additive or zero interaction
  - Effect (A + B) = Effect A + Effect B
- 2) synergistic
  - Effect (A + B) > Effect A + Effect B
- 3) antagonistic
  - Effect (A + B) < Effect A + Effect B

What Type of Interaction?



*significantly different from all other groups ($p < 0.05$)

Isobolographic Analysis



Additive

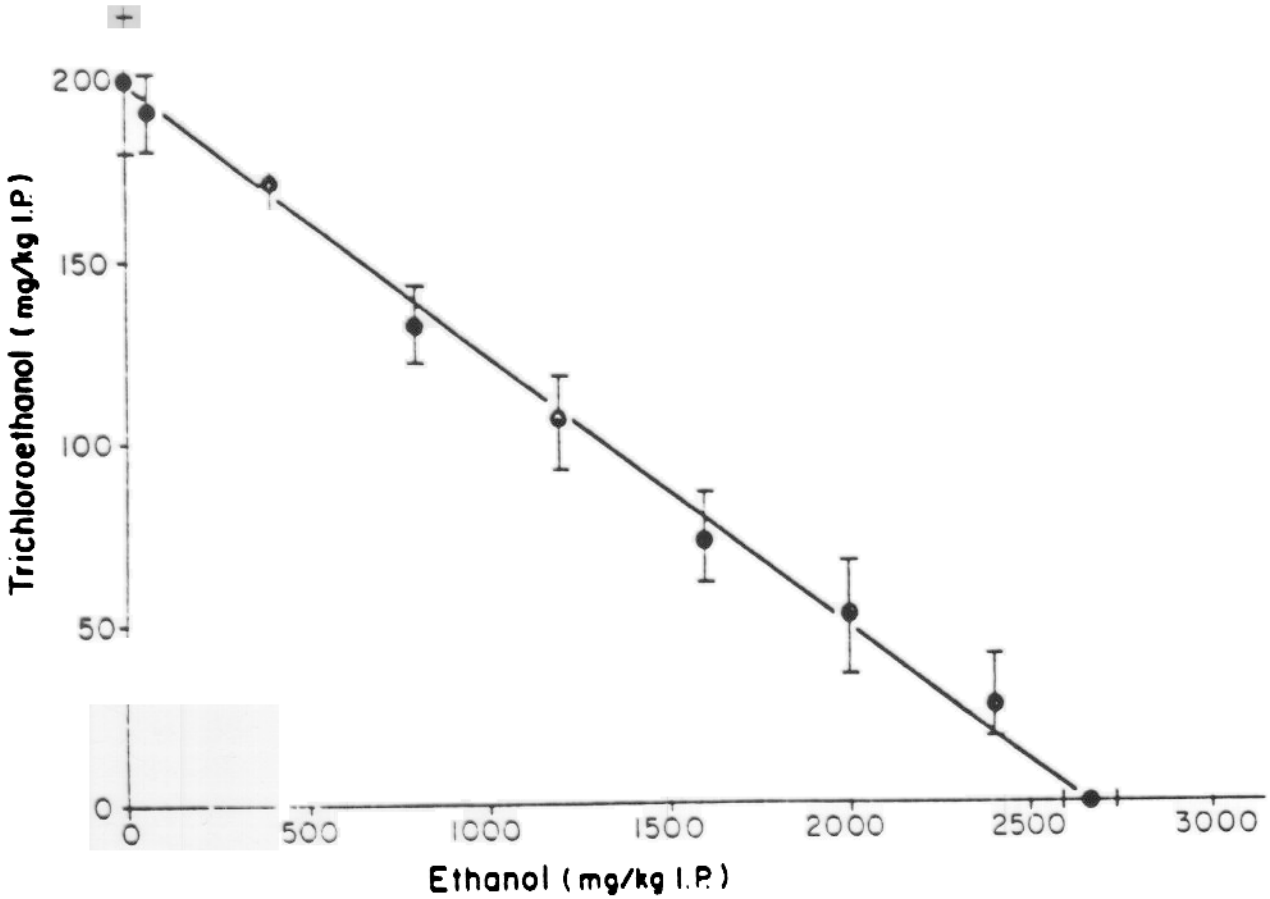


FIG. 2. Isobologram for the interaction of trichloroethanol and ethanol with respect to righting reflex loss in mice following their intraperitoneal administration. ED_{50} points are plotted with their 95% confidence limits. The expected location of the ED_{50} points, given the occurrence of a simple additive synergism, is given by the solid diagonal. Reprinted [illegible] Gessner and Cabana (1970).

Synergism

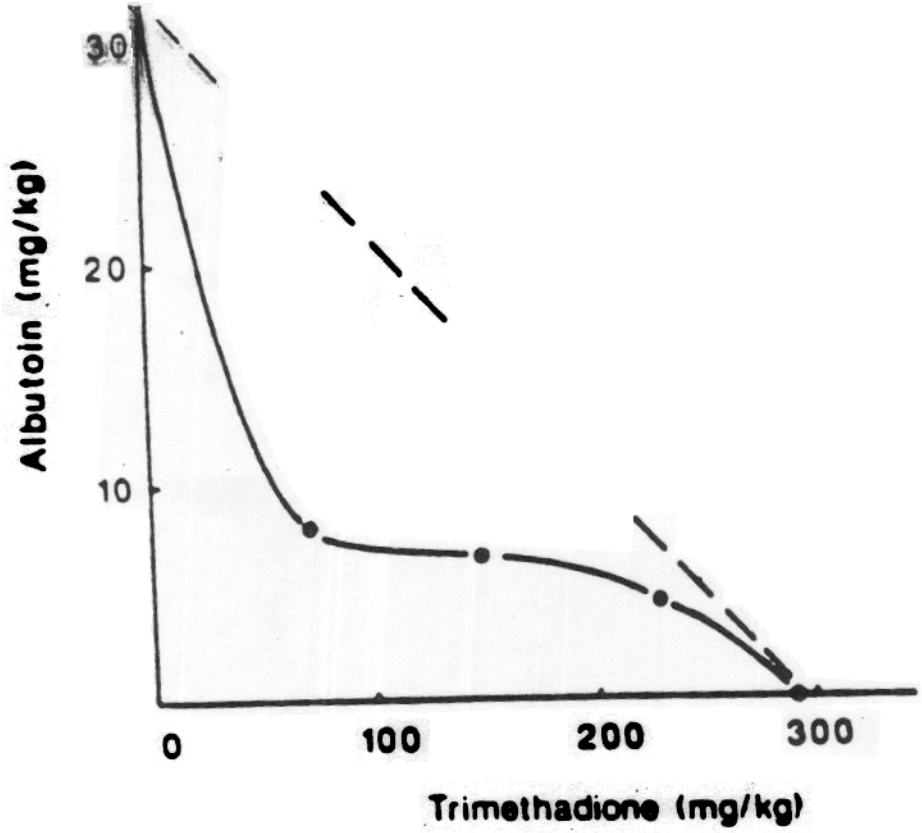


FIG. 2. Isobole showing synergy between trimethadione and albutoin in protecting mice against clonic seizures induced by pentylenetetrazol (redrawn from fig. 3 of Wallin et al. (524). The dashed line indicates the zero interaction isobole, i.e., the locus of all combinations that would have produced this effect if there had been no interaction. The three combinations (●) shown are all to the left and below this line, showing that less of the drugs is needed than expected, i.e., they are synergistic.

Antagonism

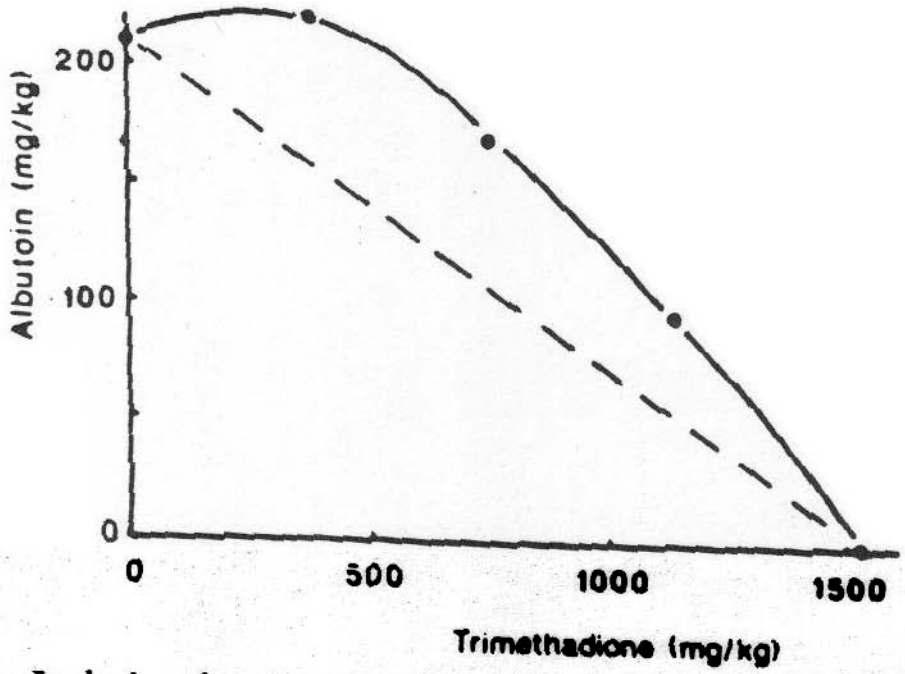


FIG. 3. Isobole showing antagonism between trimethadione and albutoin in protecting mice against electrically induced tonic extensor seizures (redrawn from fig. 3 of Wallin et al. (524)). The three combinations shown are all above and to the right of the zero interaction line, showing that the drugs are less effective in combination than expected.

Type of Interaction Is Dose-dependent

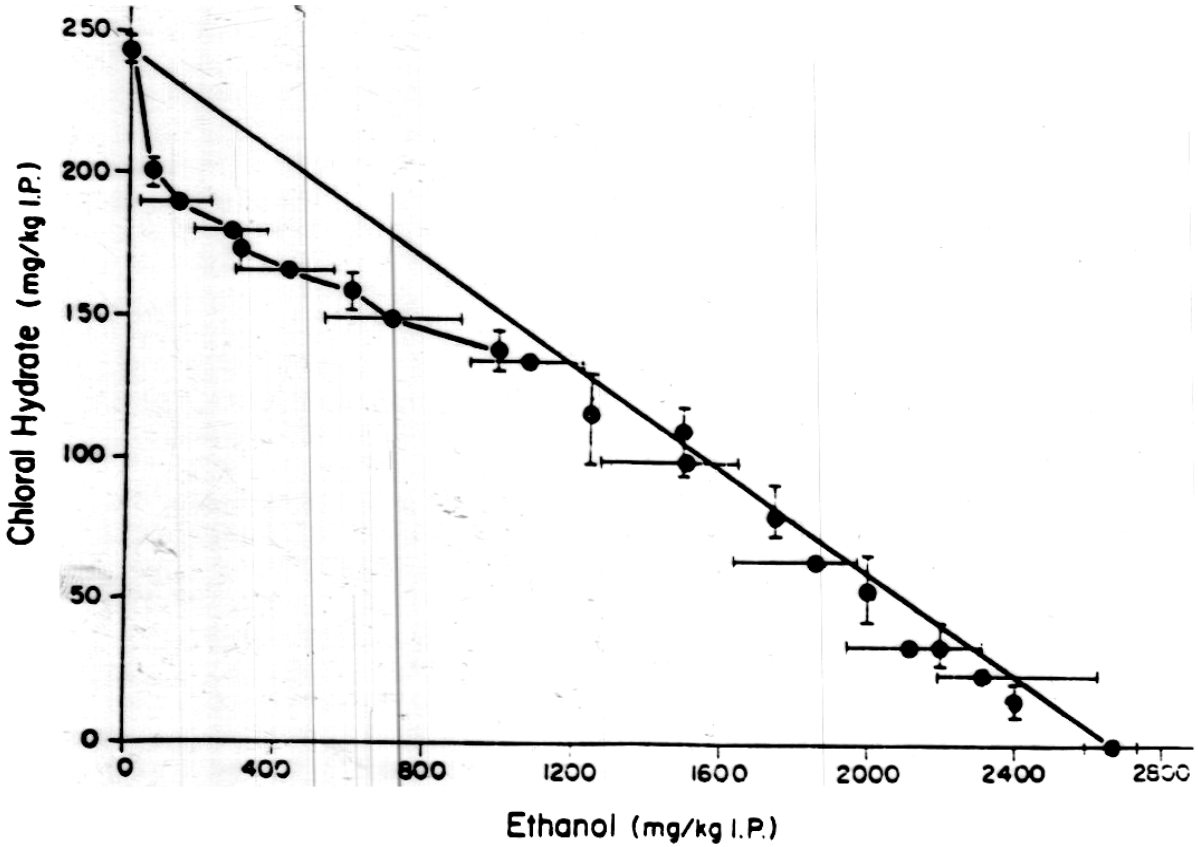


FIG. 3. Isobologram for the interaction of chloral hydrate and ethanol with respect to righting-reflex loss in mice following their intraperitoneal administration. ED_{50} points are plotted with their 95% confidence limits. The expected location of the ED_{50} points, given the occurrence of a simple additive synergism, is given together with the 95% confidence limits by the solid and dotted straight line diagonals, respectively. In the area of significant potentiation, the ED_{50} isobol has been generated by connecting adjacent ED_{50} points. Reprinted from Gessner and Cabana (1970).

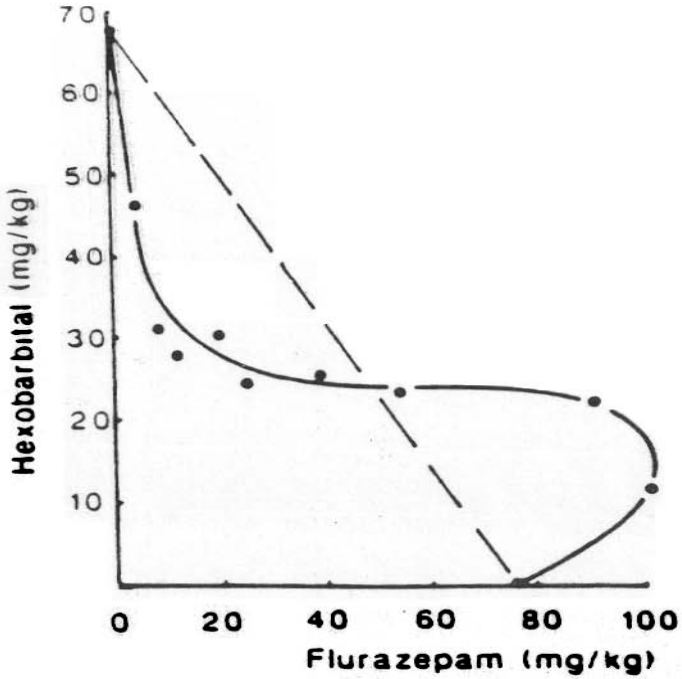
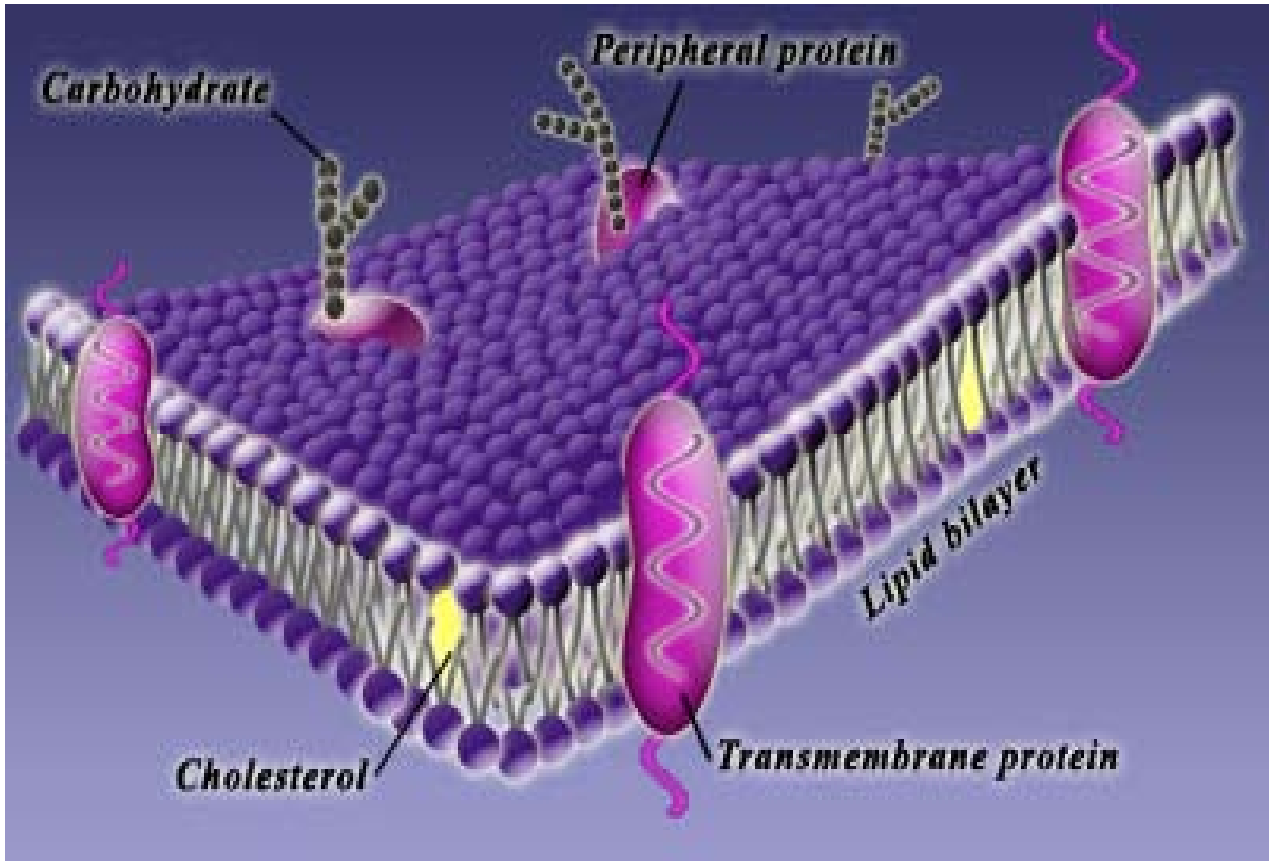


FIG. 8. Isobole for anaesthetic effects of fluorazepam and hexobarbital (redrawn from fig. 3 of Norberg and Wahlström (384)). The isobole is markedly inconsistent, with synergy in one region, antagonism in another, and zero interaction at one point.

ADME

- Absorption
- Distribution
- Metabolism
- Excretion

Absorption


Carbohydrate
Peripheral protein
Lipid bilayer
Cholesterol
Transmembrane protein

Ionization & Absorption

- Conditions for absorption:
  - ✓ *Lipid solubility*
  - ✓ *Some water solubility to get out of the membrane*
- Many chemicals are weak acids or weak bases
- They exist in equilibrium between:
  - ✓ *Ionized form: polar and water soluble*
  - ✓ *Un-ionized: non polar and lipid soluble*
  - ✓ *Ionization state is pH dependent*

Ionization & Absorption

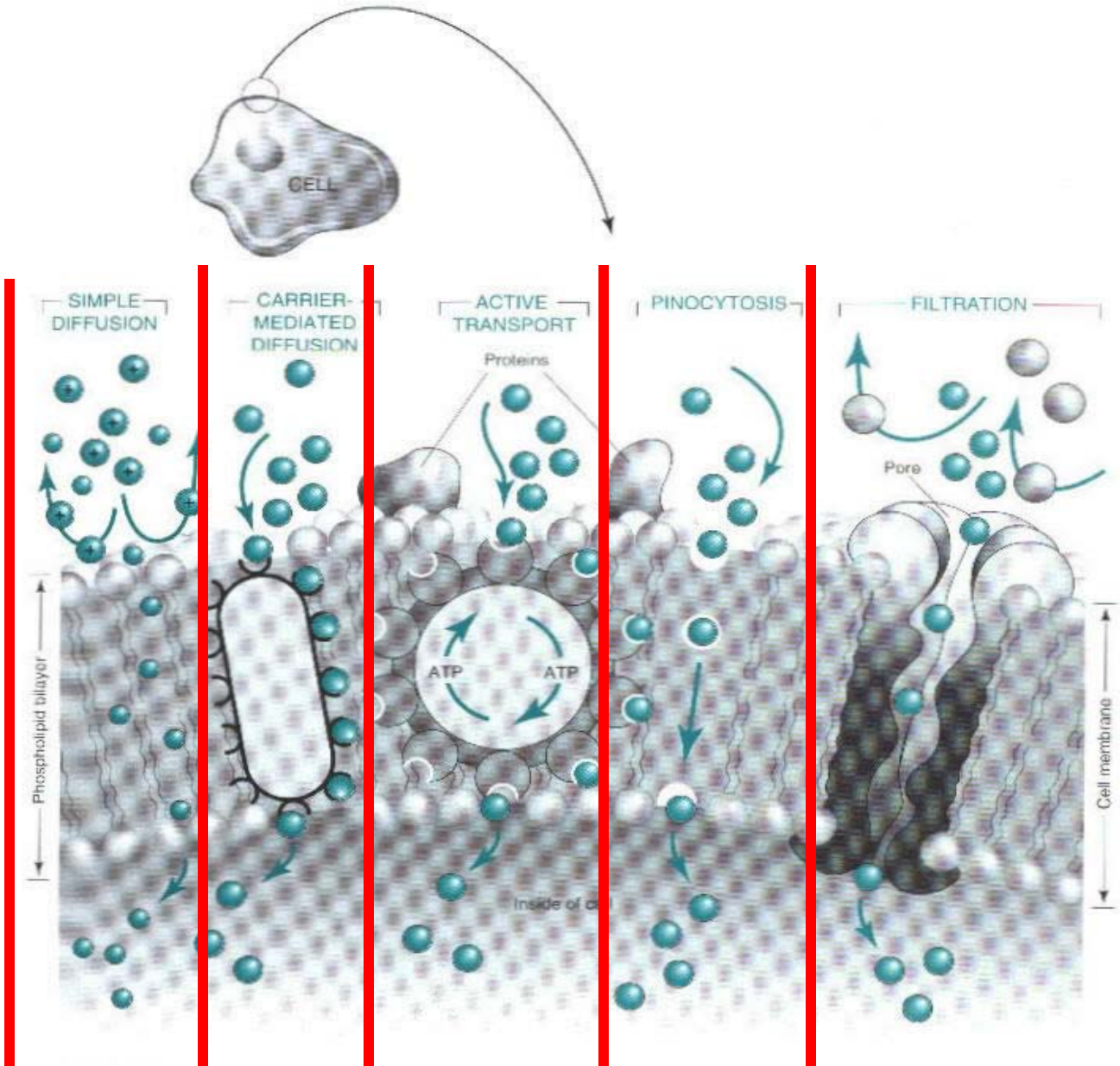
Un-ionized chemicals are more soluble in lipids (penetrate biological membranes) than ionized chemicals

non-polar chemicals penetrate biological membranes Easier than polar chemicals

Un-ionized, non-polar chemicals have the highest lipid solubility (penetrate biological membranes)

Acidic chemicals are ionized in basic environment; Basic chemicals are ionized in acidic environment

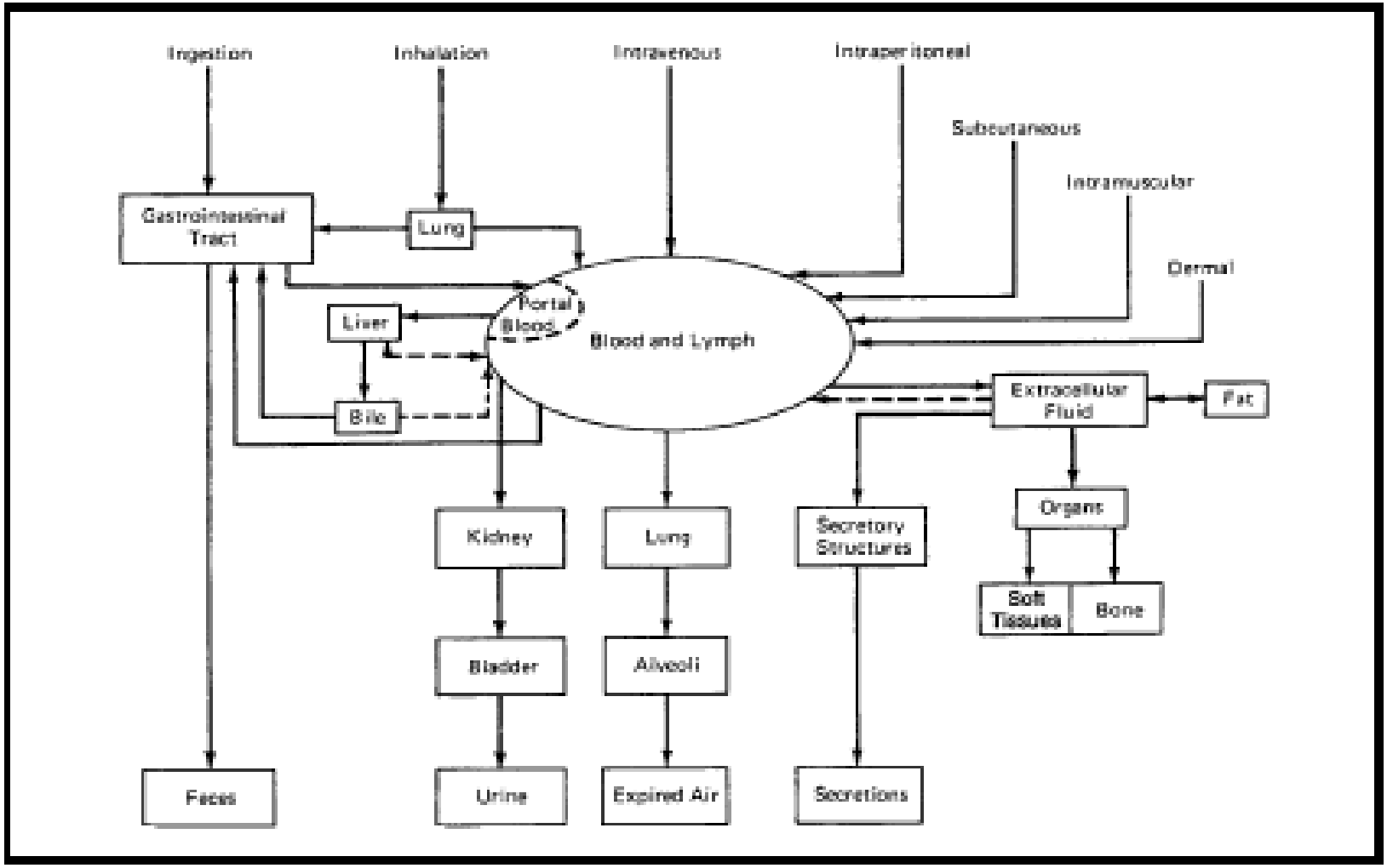
Chemical/Drug Transport Mechanisms



Route of Exposure

- In an industrial setting
  - Inhalation
    - Fastest route of exposure for a chemical to reach the blood stream
  - Dermal
    - Slowest route of exposure
  - Ingestion
    - Important when eating and drinking around chemicals
  - Injection (not necessarily intravenous)
    - Accidental injection of petroleum chemicals (or other) under the skin with a high pressure air system

Distribution



Distribution

- **Passage of chemicals from bloodstream to various tissues**

- **Factors influencing distribution are:**
  - **Ability to cross biological barriers**
  - **Physicochemical properties of chemicals**
  - **Blood flow to tissue**
  - **Plasma proteins binding (ability of chemicals to exit vascular system)**

Perfusion-Rate Limitation

- **Biological membranes are no barrier to distribution:**
  - ✓ **Small & highly lipophilic chemicals (ethanol)**
  - ✓ **Loose membranes**
- **Perfusion rate of different tissues differs**
- **Direct correlation between tissue perfusion and distribution time**

Blood Flow to Various Tissues

Rapidly perfused (ml/100 g/min) tissues respond quickly

Brain (55)
Liver (20)
Kidney (450)

Less rapidly perfused tissues respond to chemical more slowly

Muscle (3)
Skin (5)

Poorly perfused tissues respond very slowly to chemical

Fat (1)

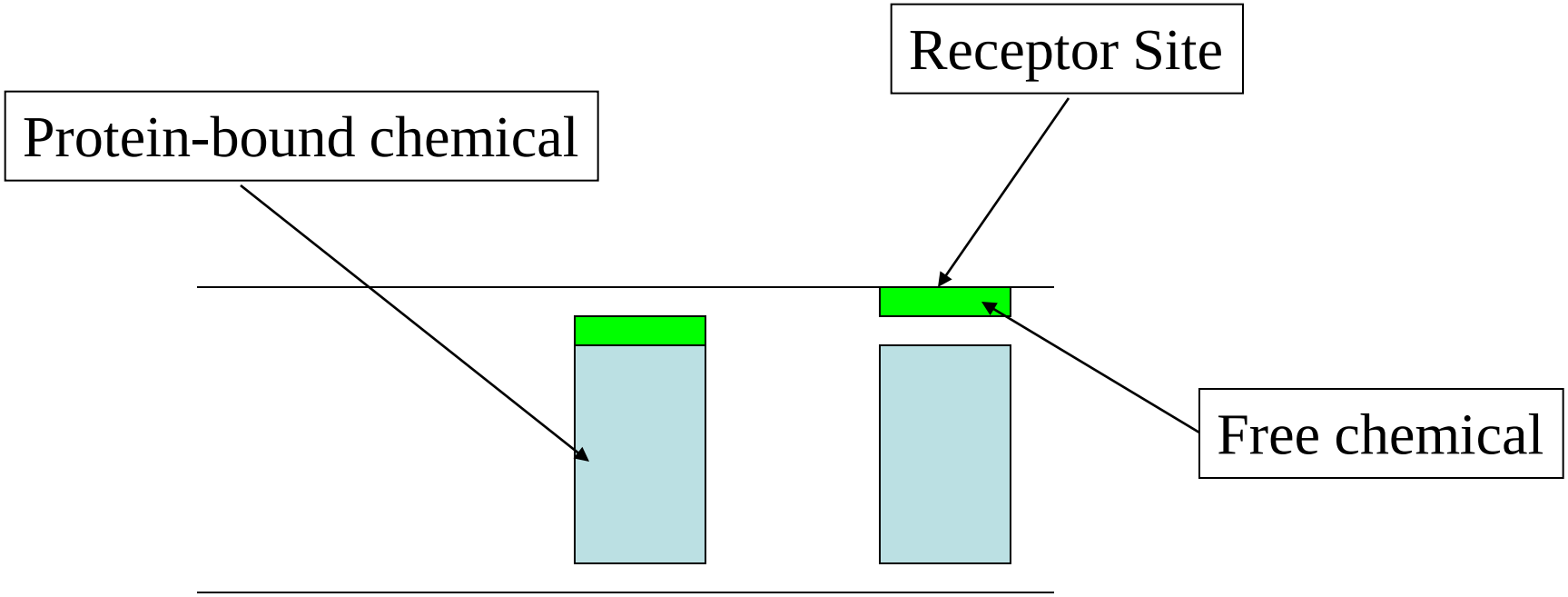
Permeability-Rate Limitation

- **Physicochemical properties of drugs:**
  - ✓ **Polar drugs**
  - ✓ **Size**
  - ✓ **Ionization: Ion trapping in a tissue**

- **Membrane permeability depends on tissue**
  - ✓ **Liver: porous capillary**
  - ✓ **Brain: blood brain barrier; high liposolubility**
  - ✓ **Kidney: high membrane permeability**

Plasma Protein Binding

- Many chemicals bound to circulating plasma proteins such as albumin
- Equilibrium between free and bound chemical
- Only free chemical can act at receptor site or site of action

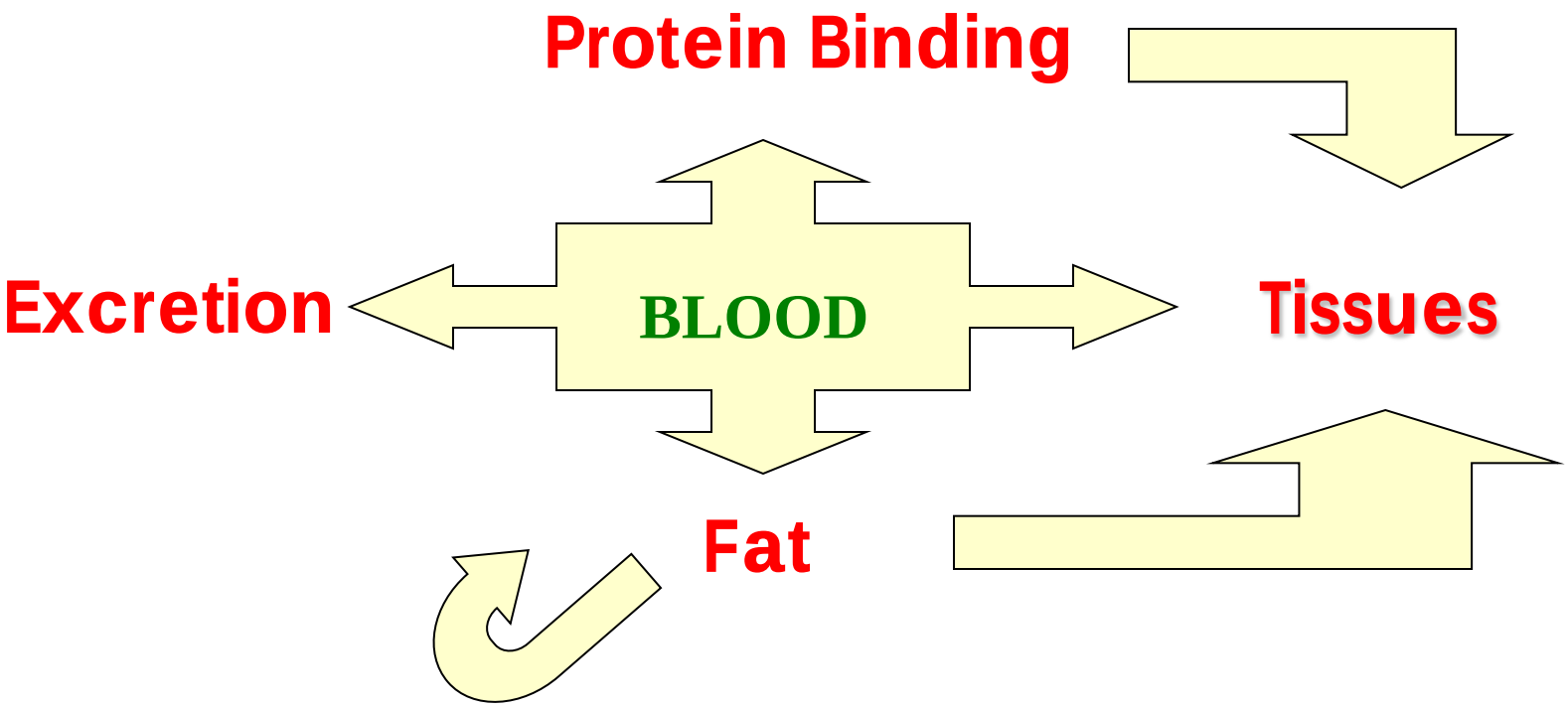


Clinical Significance of Protein Binding

- **Be aware of the medications that workers may be taking**
  - **May have interactions or toxic effects due to protein binding interactions between chemical toxicant and prescription drug**
- **Highly protein bound drugs**
  - Small change in binding results in large change in free drug concentrations
- **Drugs with low therapeutic window**
  - Toxicities due to increased free drug concentration
- **Drug/chemical toxicant interaction: displacement**

  - Displacement of drug from protein binding sites
- **Physical state such as renal failure, inflammation, fasting, malnutrition can have effect on plasma protein binding**

Tissue Distribution & Binding



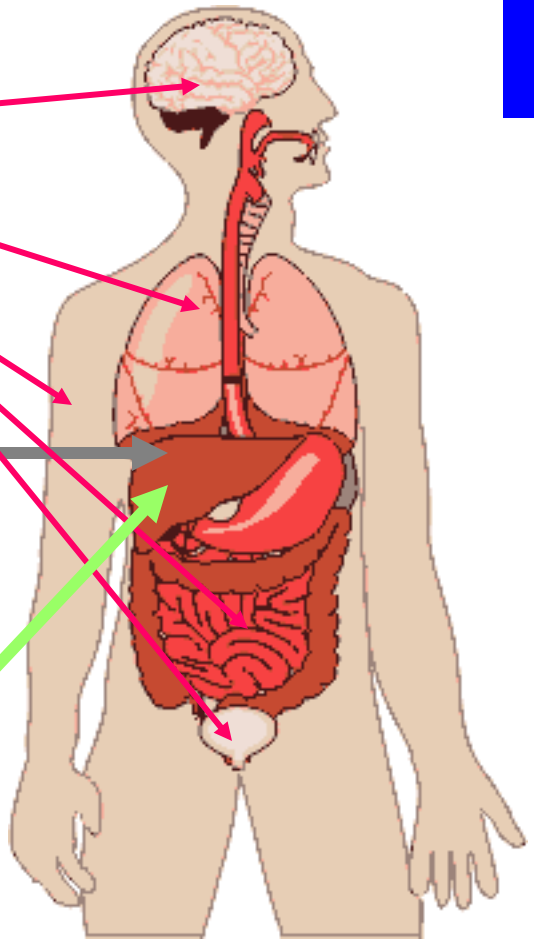
Metabolism

Drug Metabolism Sites

Extrahepatic microsomal enzymes
(oxidation, conjugation)

Hepatic microsomal enzymes
(oxidation, conjugation)

Hepatic non-microsomal enzymes
(acetylation, sulfation,GSH, alcohol/aldehyde dehydrogenase, hydrolysis, ox/red)



LIVER

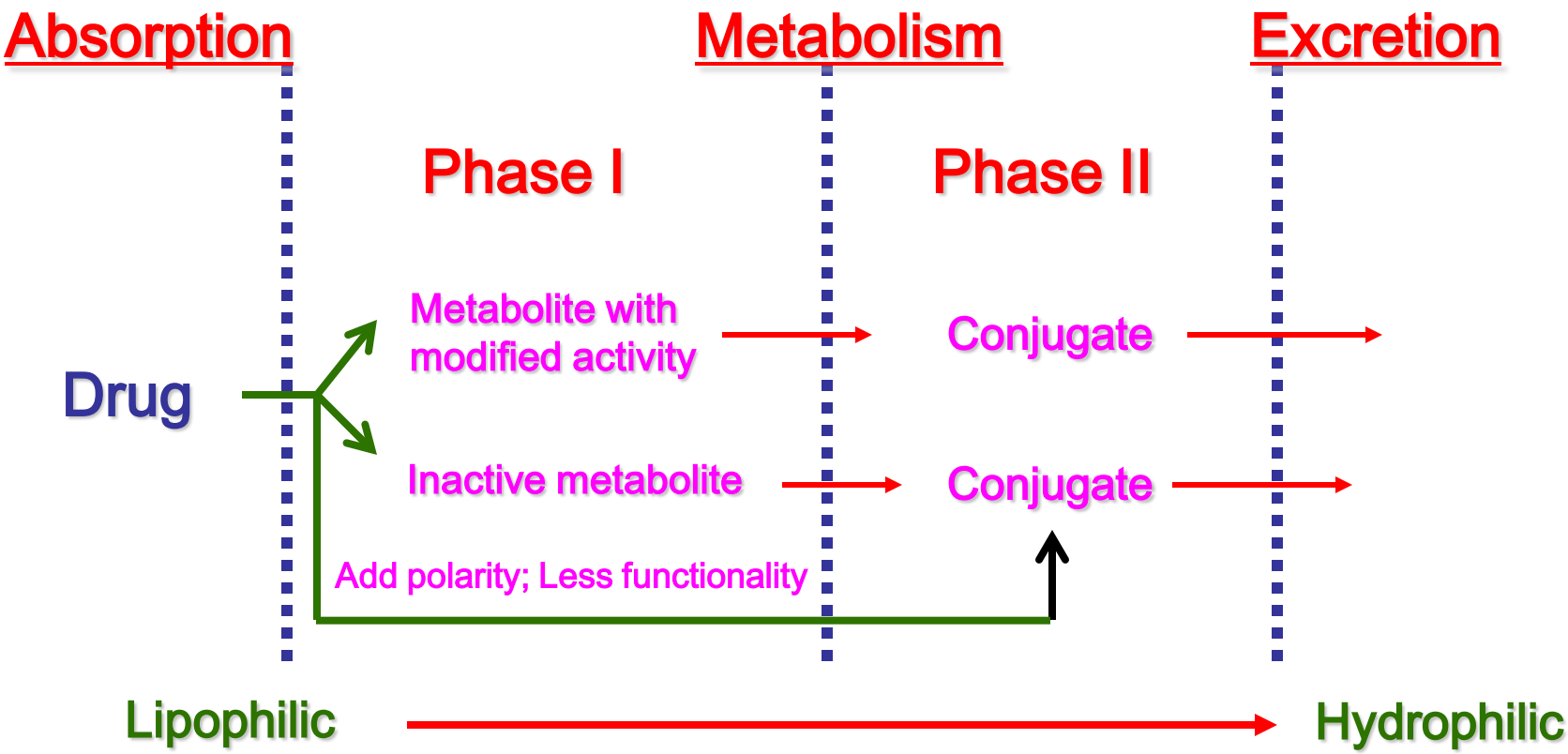
The liver is ideally placed to intercept natural ingested toxins (bypassed by injections etc) and has a major role in metabolism

Biotransformation

- **<u>Usually</u> inactivates the chemical/drug**
- **<u>Can</u> generate active metabolite; toxic metabolite**
- **<u>Often</u> generates polar, highly ionized metabolites.**
- **Some chemicals/drugs are eliminated unchanged**

Phase I and Phase II Metabolic Reactions

Biotransformation



Metabolic Reactions

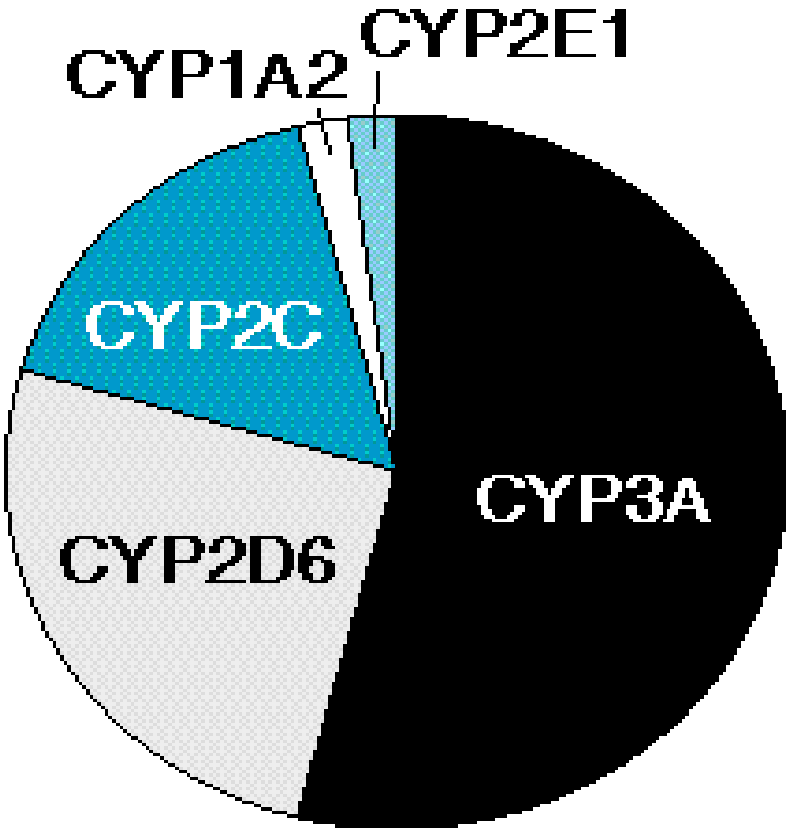
- Phase I Reactions
  - Adds polarity; less functionality (metabolite compared to parent compound)
    - Oxidation (P450 system)
    - Hydrolysis
    - Reduction
- Phase II Reactions
  - Adds endogenous conjugate to the chemical
    - Glucuronide conjugation (major reaction)
    - Sulfate conjugation

OXIDATION

- **P450-Dependent Drug Oxidation involves a loss of electron or gain of oxygen**

- **Non-specific system associated with endoplasmic reticulum**

- **Multiple forms of CYP-P450**

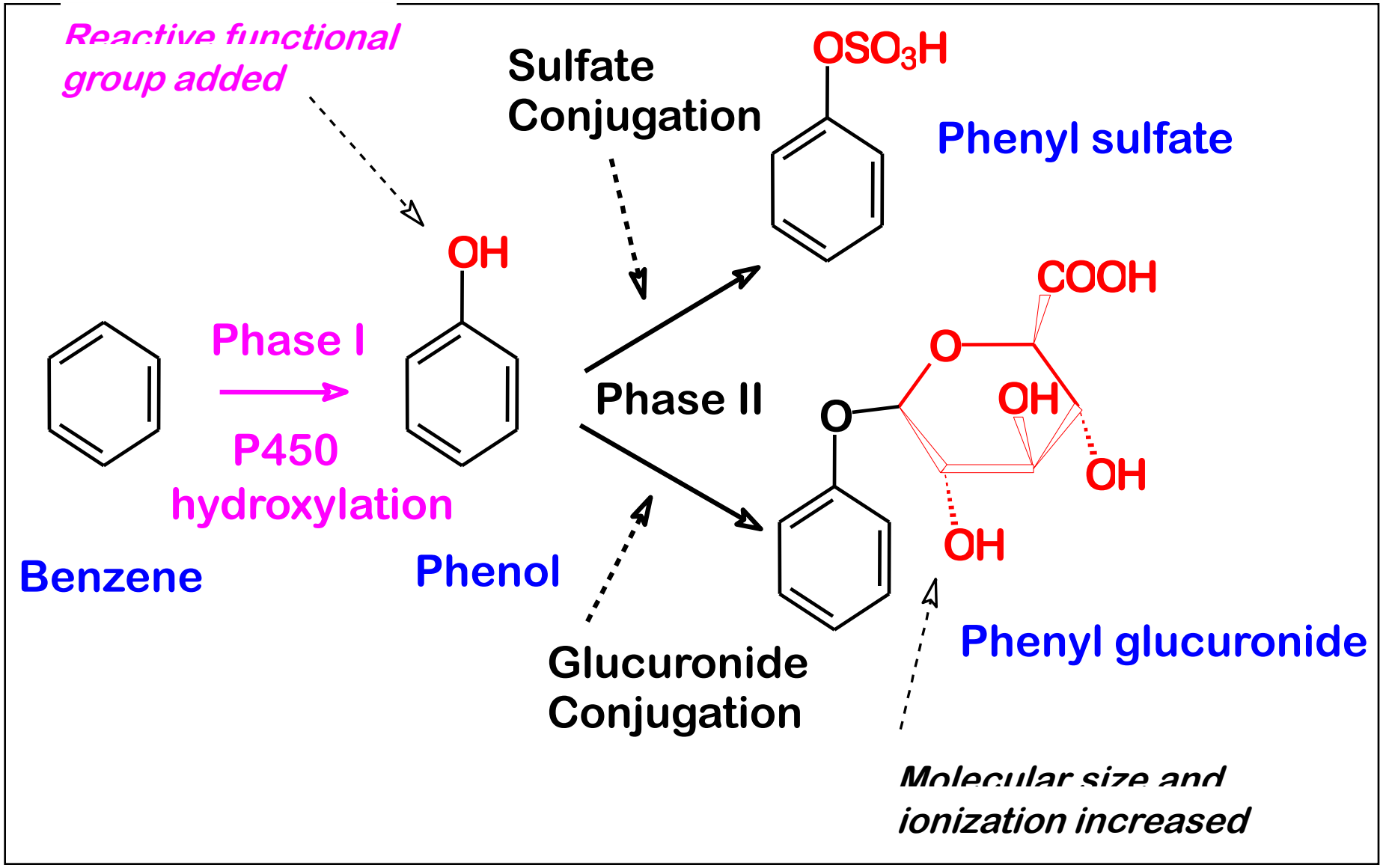


Relative contribution of the major P450 isoforms to human chemical/drug metabolism

Some Chemicals Stimulate and Some Inhibit the P450 System

- **Stimulation is via <u>induction</u>, which means an increase in enzyme <u>synthesis</u>.**
- **Inhibition is via a <u>decrease in the activity of existing enzyme</u>.**
- **This can impact the actions of other chemicals or prescription drugs**

Phase II Reactions



Elimination of drugs from the body

MAJOR

KIDNEY
filtration
secretion
(reabsorption)

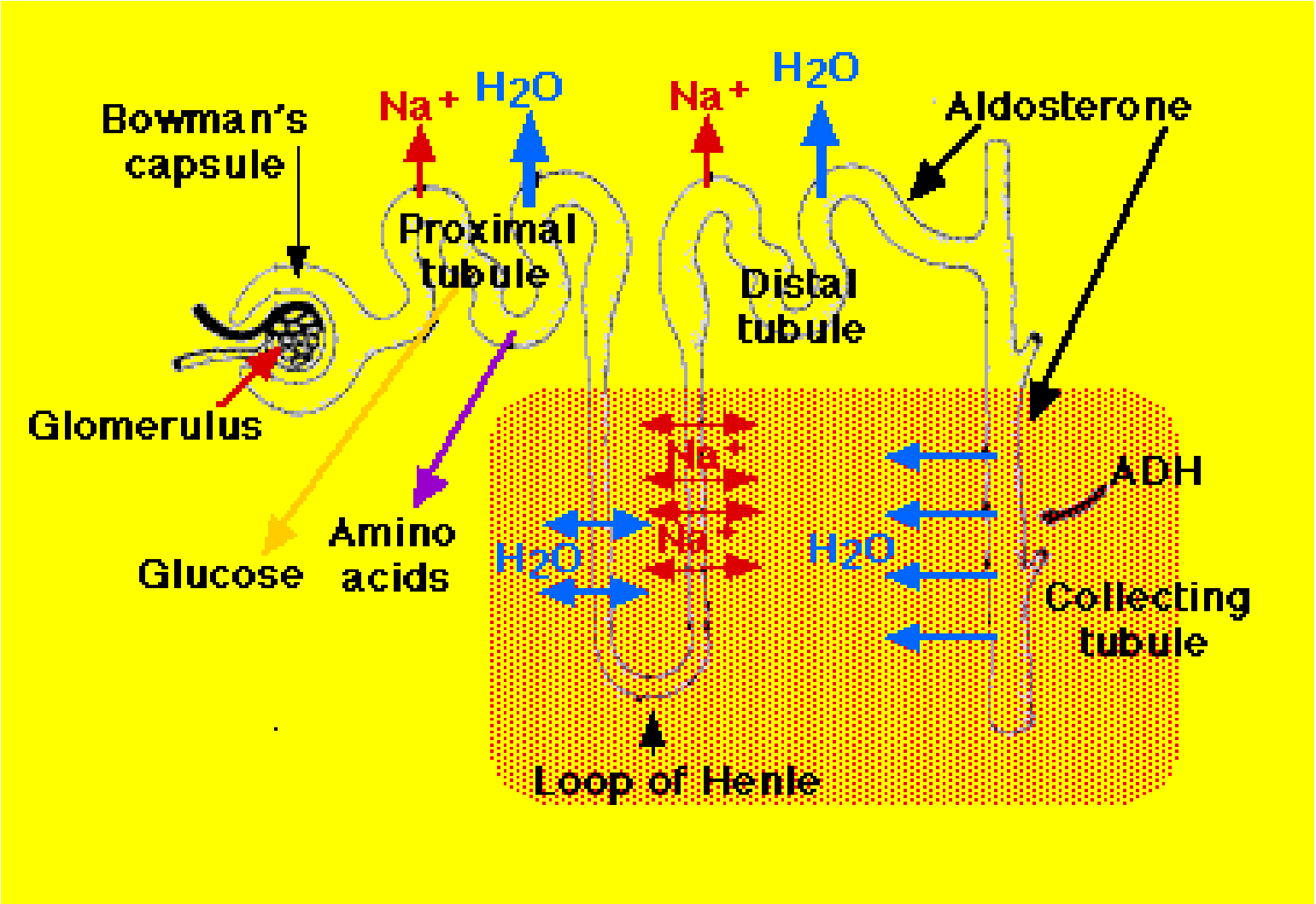
LIVER
metabolism
secretion
Enterohepatic recirculation;
Bile excretion for > 300 mol wt.

MINOR

LUNGS
exhalation

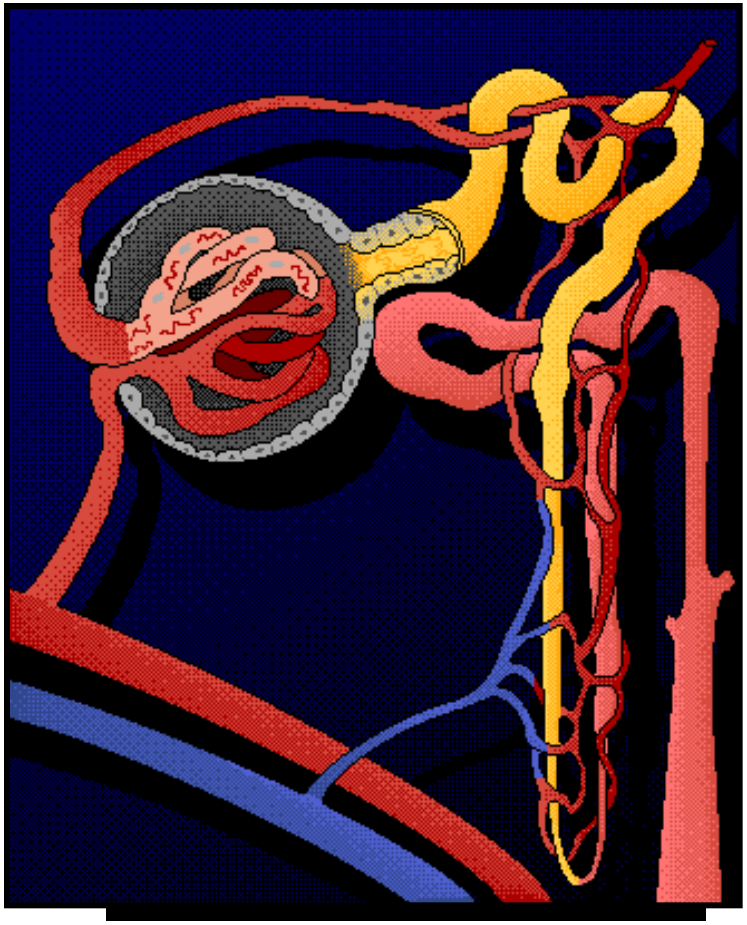
OTHERS
mother's milk
sweat, saliva etc.

Nephron Functional Structure



Chemical Elimination By The Kidney

- Primary site of drug elimination
- Three mechanisms of elimination:
  - ✓Glomerular Filtration
  - ✓Tubular Reabsorption
  - ✓Tubular Secretion

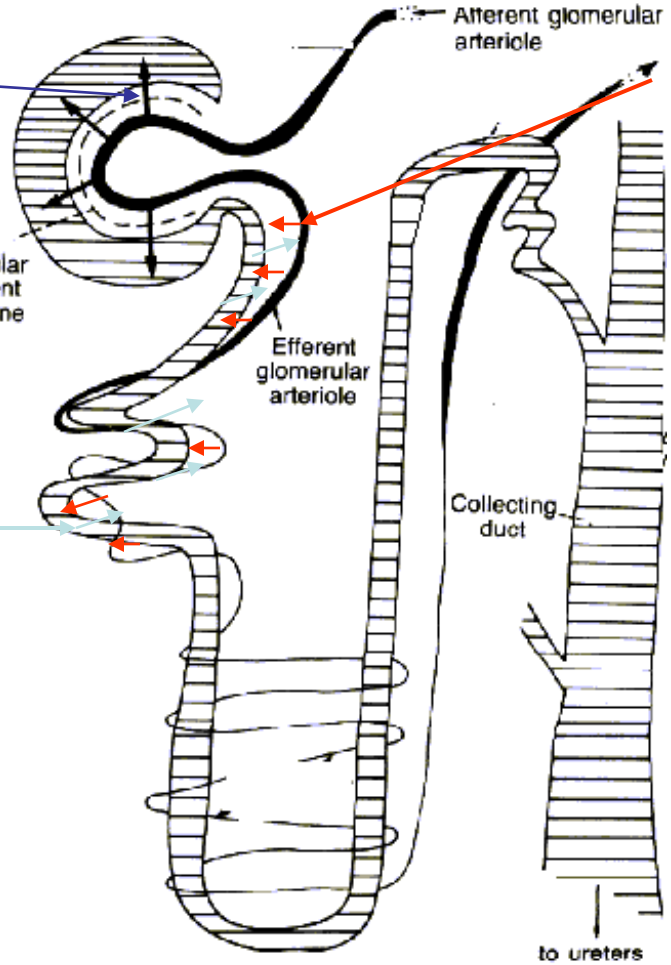


Chemical Elimination By The Kidney

chemicals filtered

some chemicals actively secreted

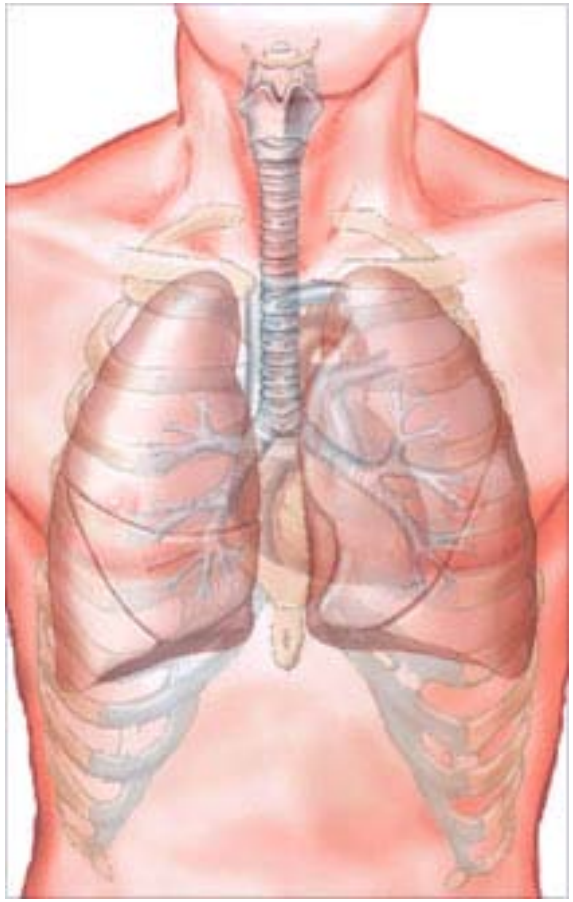
some chemicals reabsorbed



pH of urine can be a factor in elimination

Pulmonary Elimination

- **Elimination via the lungs & breath**
- **Major route for gaseous & volatiles substances**



Questions?