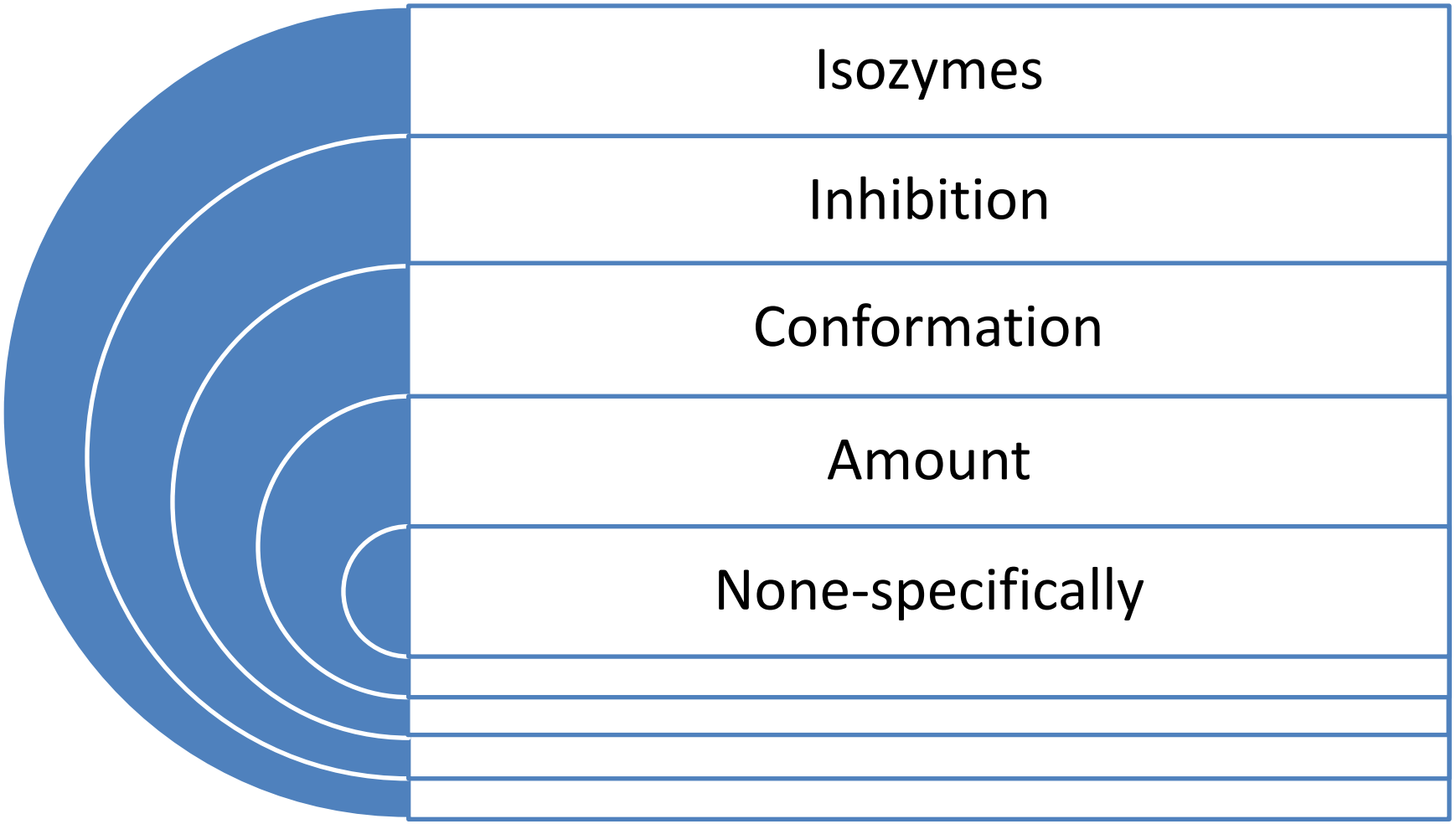
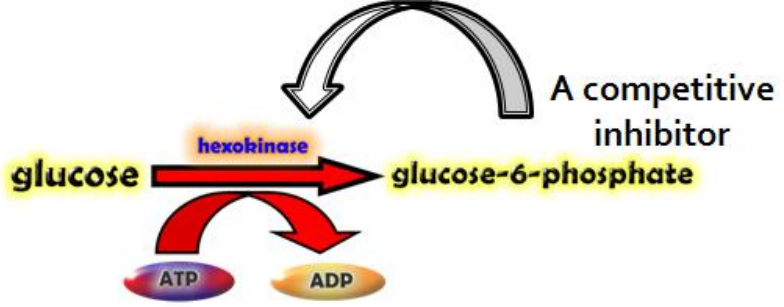
Enzymes Regulation

Modes of regulation

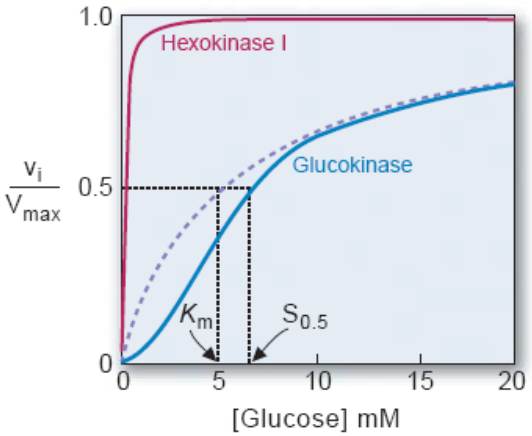


Isozymes (isoenzymes)
The Differential K_M Value
"Hexokinase"

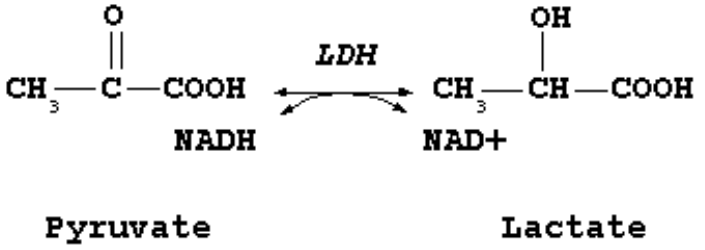


- **What are isozymes? Same substrate & product, different gene, <u>different localization</u>, <u>different parameters (K_m, V_{max}, k_{cat})</u>**
- **Hexokinase found in RBCs & in liver**
- **Catalyzes the first step in glucose metabolism**
- **Hexokinase I (RBCs): K_M (glucose) ≈ 0.1 mM**
- **Hexokinase I V (glucokinase, liver, pancreas) ≈ 10 mM**
- **RBCs: when blood glucose falls below its normal fasting level (≈ 5 mM), RBCs could still phosphorylate glucose at rates near V_{max}**

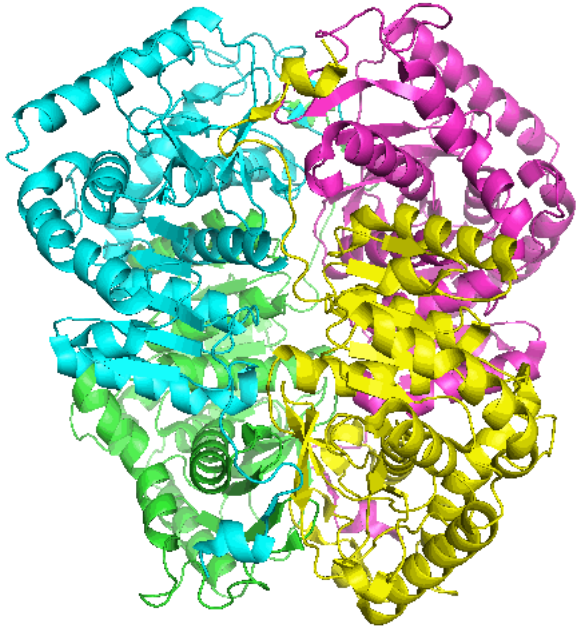
- **Liver: rate of phosphorylation increases above fasting levels (after a high-carbohydrate meal)**
  - **High K_M of hepatic glucokinase promotes storage of glucose**
- **Pancreas: works as a sensor**



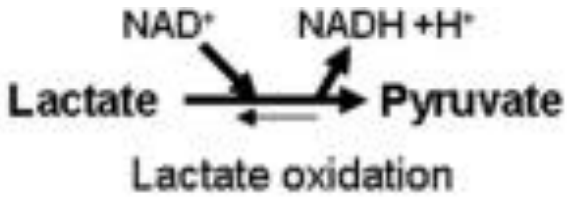
Lactate Dehydrogenase (LDH)

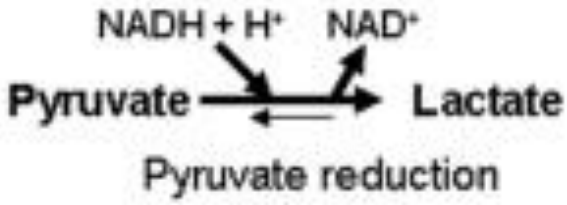


- **Aerobic vs. anaerobic**
- **Km: H4 >> M4**
- **Inhibition: H4 inhibited but not M4**

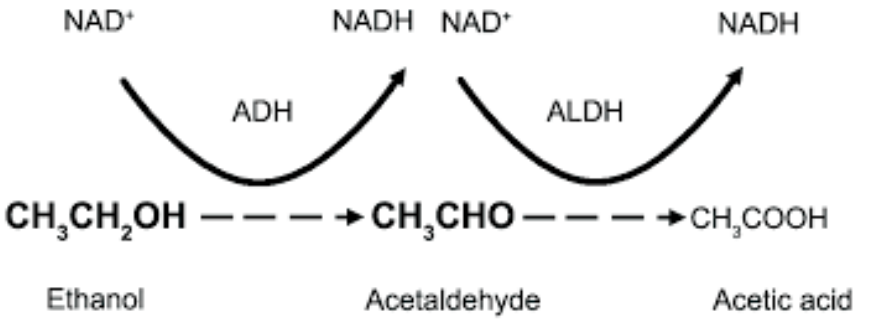


Intermediate activity





Aldehyde dehydrogenase (ALDH)



- Oxidation of acetaldehyde to acetate.
- Four tetrameric isozymes (I-IV)
- ALDH I (low Km; mitochondrial) and ALDH II (higher Km; cytosolic)
- ~50% of Japanese & Chinese are unable to produce ALDH I (not observed in Caucasian & Negroid populations)
  - Flushing response
  - Tachycardia

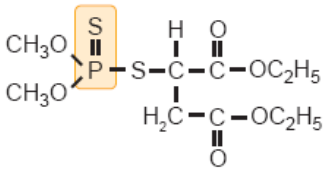
2. Inhbition

2.1 MECHANISM-BASED INHIBITORS

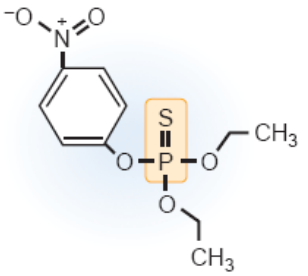
- Mechanism-based inhibitors mimic or participate in an intermediate step of the catalytic reaction
- The term includes:
- A. Covalent inhibitors
- B. Transition state analogs
- C. Heavy metals

- The kinetic effect of irreversible inhibitors is to <u>decrease the concentration of active enzyme</u>

2.1.A. Covalent Inhibitors



Malathion



Parathion

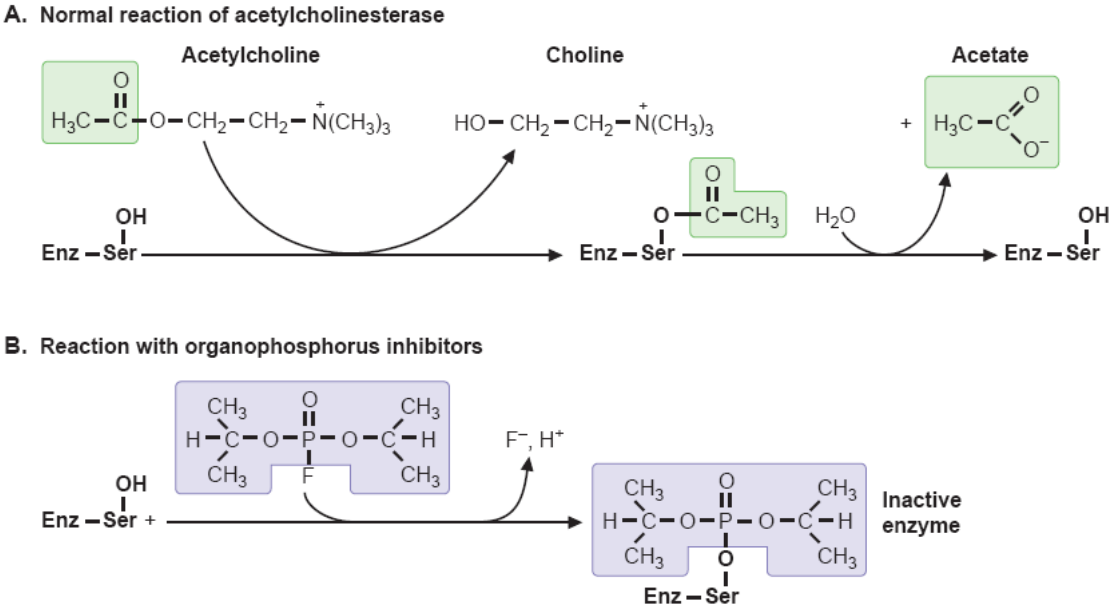
- Covalent or extremely tight bonds with active site amino acids
- Amino acids are targeted by drugs & toxins

- The lethal compound [DFP] is an organophosphorus compound that served as a prototype for:
  - The nerve gas sarin

Sarin

  - The insecticides malathion & parathion

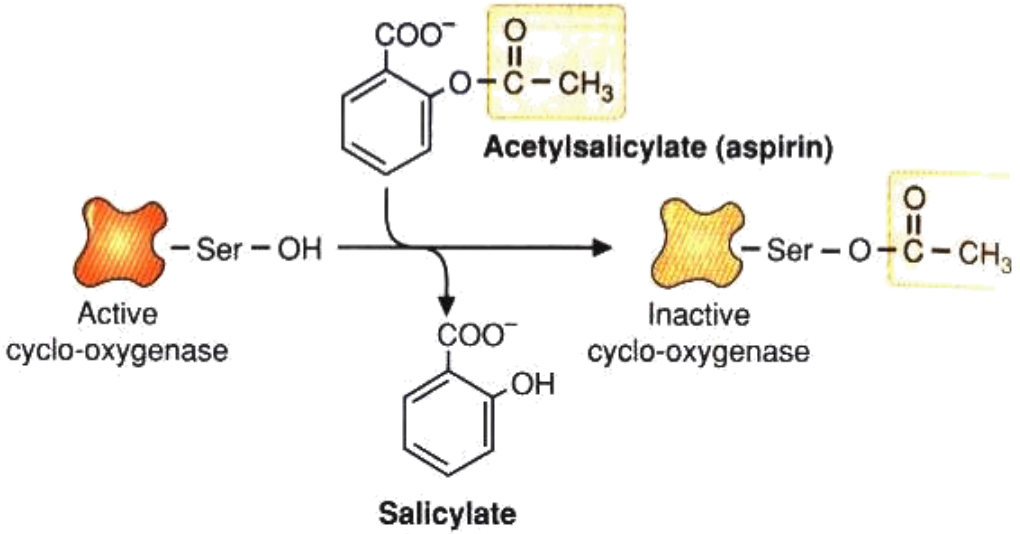
- DFP also inhibits other enzymes that use serine (ex. serine proteases), but the inhibition is not as lethal



2.1.A. Covalent Inhibitors

- **Aspirin (acetylsalicylic acid): covalent acetylation of an active site serine in the enzyme prostaglandin endoperoxide synthase (cyclooxygenase)**

- **Aspirin resembles a portion of the prostaglandin precursor that is a physiologic substrate for the enzyme**

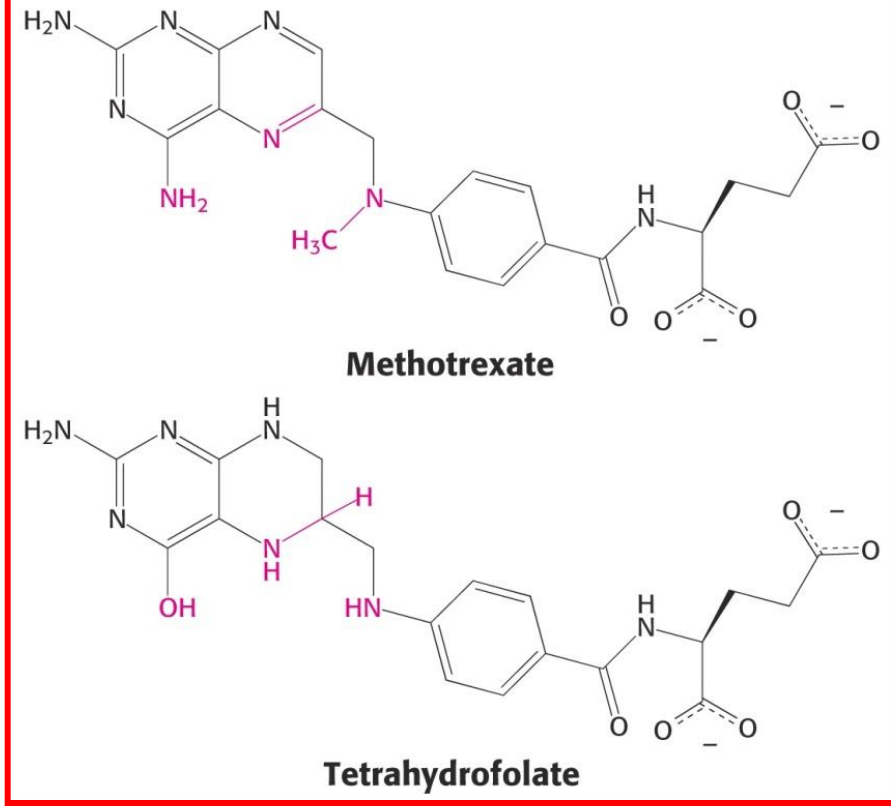


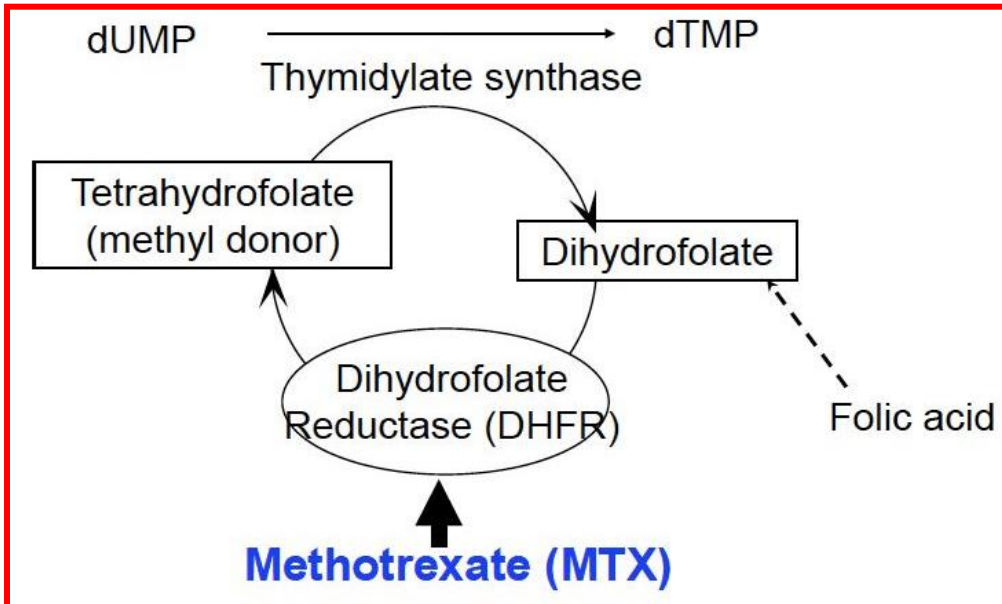
2.1.B. Transition-State Analogs & Compounds that Resemble Intermediate Stages of the Reaction

- Transition-state analogs: extremely potent inhibitors (bind more tightly)
- Drugs cannot be designed that precisely mimic the transition state! (highly unstable structure)
- Substrate analogs: bind more tightly than substrates
- Known as suicide inhibitors

Methotrexate

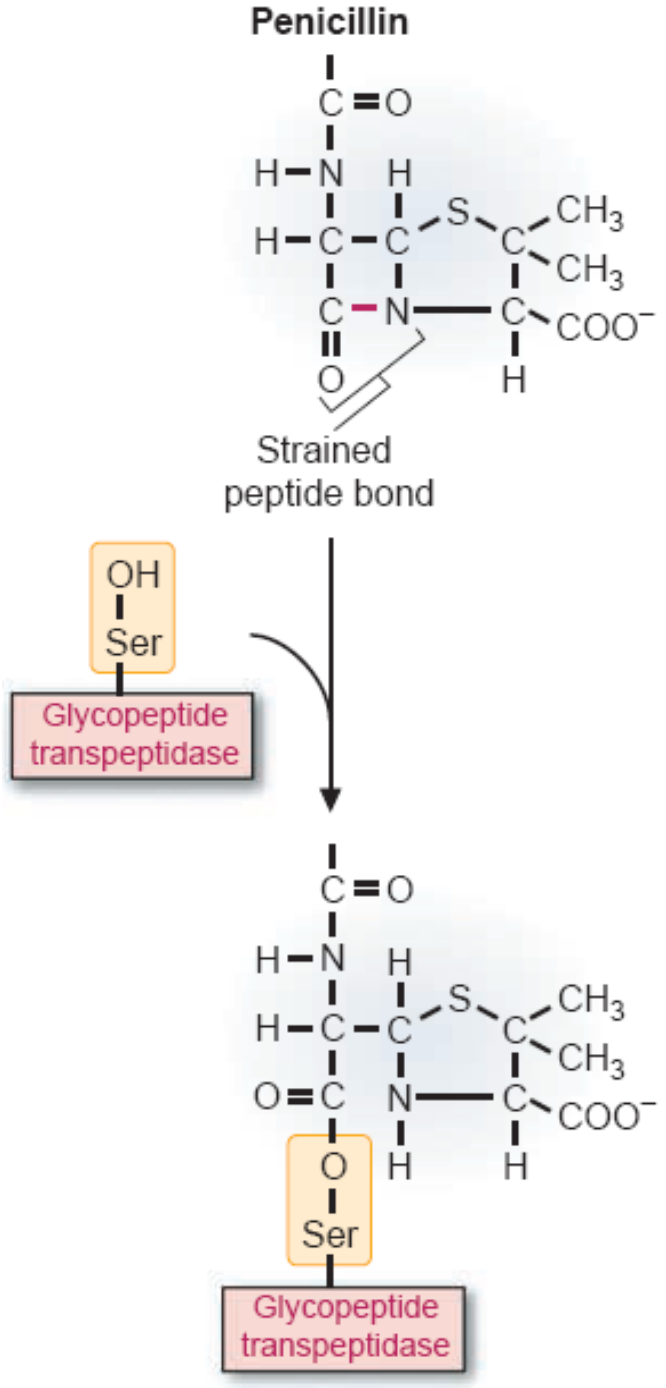
- Synthetic inhibitor
- Anticancerous
- Analog of tetrahydrofolate
- Binds to enzyme a 1000-fold more tightly
- Inhibits nucleotide base synthesis





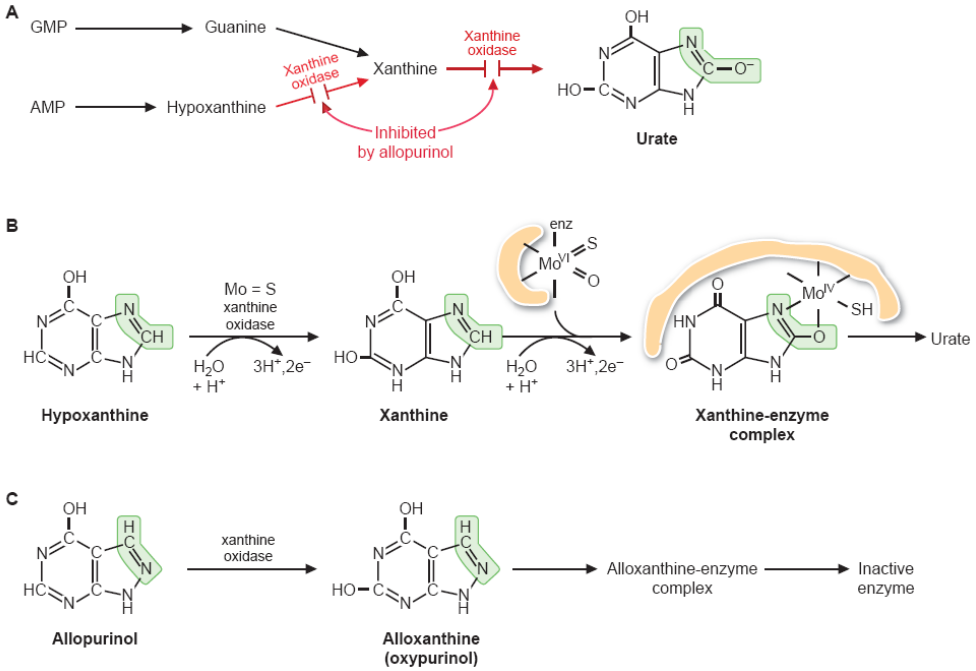
2.1.B.1 PENICILLIN

- **A transition-state analog to *glycopeptidyl transferase or transpeptidase***
- **Required by bacteria for synthesis of the cell wall**
- **The reaction is favored by the strong resemblance between the peptide bond in the β-lactam ring of penicillin & the transition-state complex of the natural transpeptidation reaction**
- **Inhibitors that undergo partial reaction to form irreversible inhibitors in the active site are sometimes termed *suicide inhibitors***



2.1.B.2 ALLOPURINOL

- **A drug used to treat gout**
- **Decreases urate production by inhibiting xanthine oxidase**
- **The enzyme commits suicide by converting the drug to a transition-state analog**
  - **The enzyme contains a molybdenum–sulfide (Mo-S) complex that binds the substrates and transfers the electrons required for the oxidation reactions**
  - **Xanthine oxidase oxidizes the drug allopurinol to oxypurinol, a compound that binds very tightly to a molybdenum–sulfide complex in the active site**



2.1.C. Heavy Metals

- **Tight binding of a metal to a functional group in an enzyme**
- **Mercury (Hg), lead (Pb), aluminum (Al), or iron (Fe)**
- **Relatively nonspecific for the enzymes they inhibit, particularly if the metal is associated with high-dose toxicity**
- **Mercury: binds to so many enzymes, often at reactive sulfhydryl groups in the active site**
  - **It has been difficult to determine which of the inhibited enzymes is responsible for mercury toxicity**
- **Lead provides an example of a metal that inhibits through replacing the normal functional metal in an enzyme, such as calcium, iron, or zinc**
  - **Its developmental & neurologic toxicity may be caused by its ability to replace Ca^{+2} in several regulatory proteins that are important in the central nervous system and other tissues**