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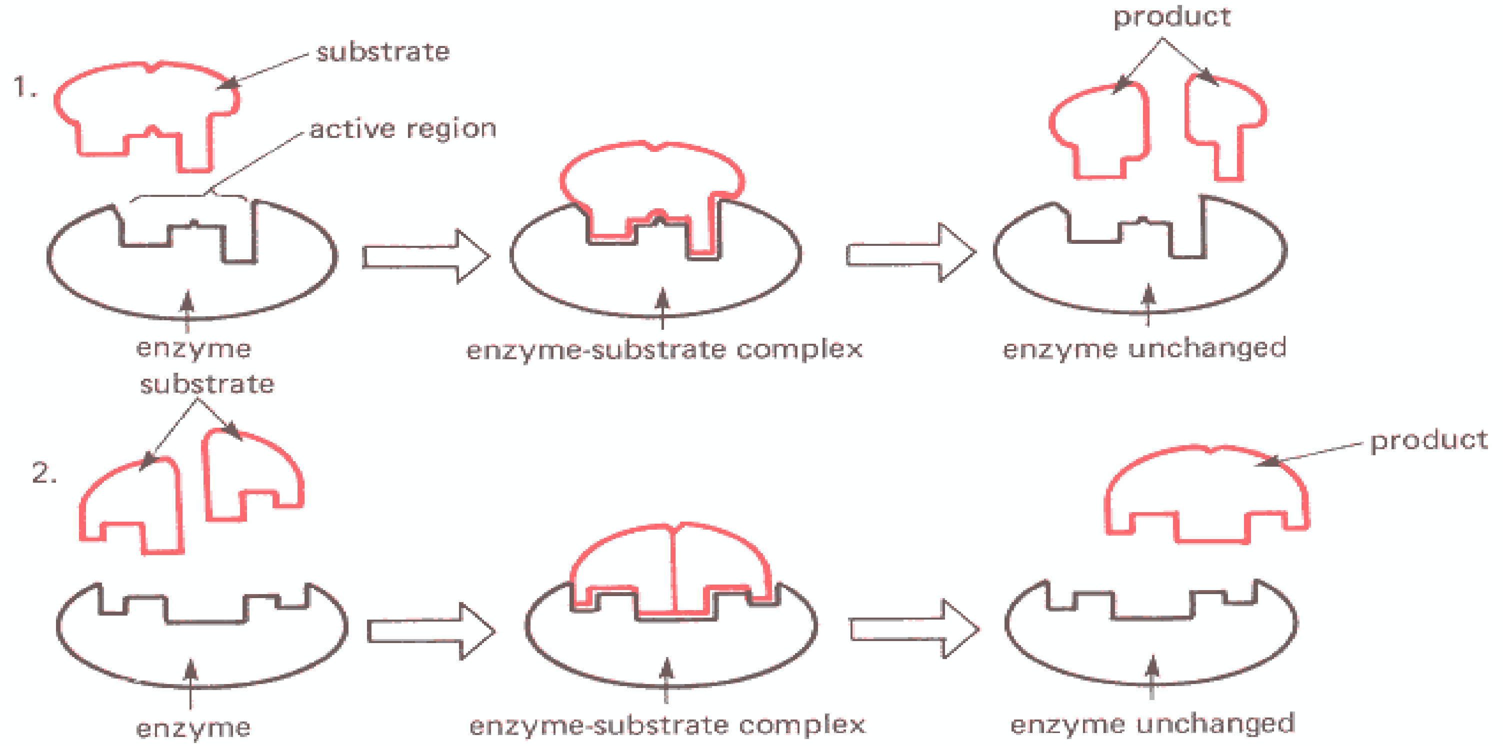
Enzyme Mechanisms

CLASSIFICATION OF ENZYMES

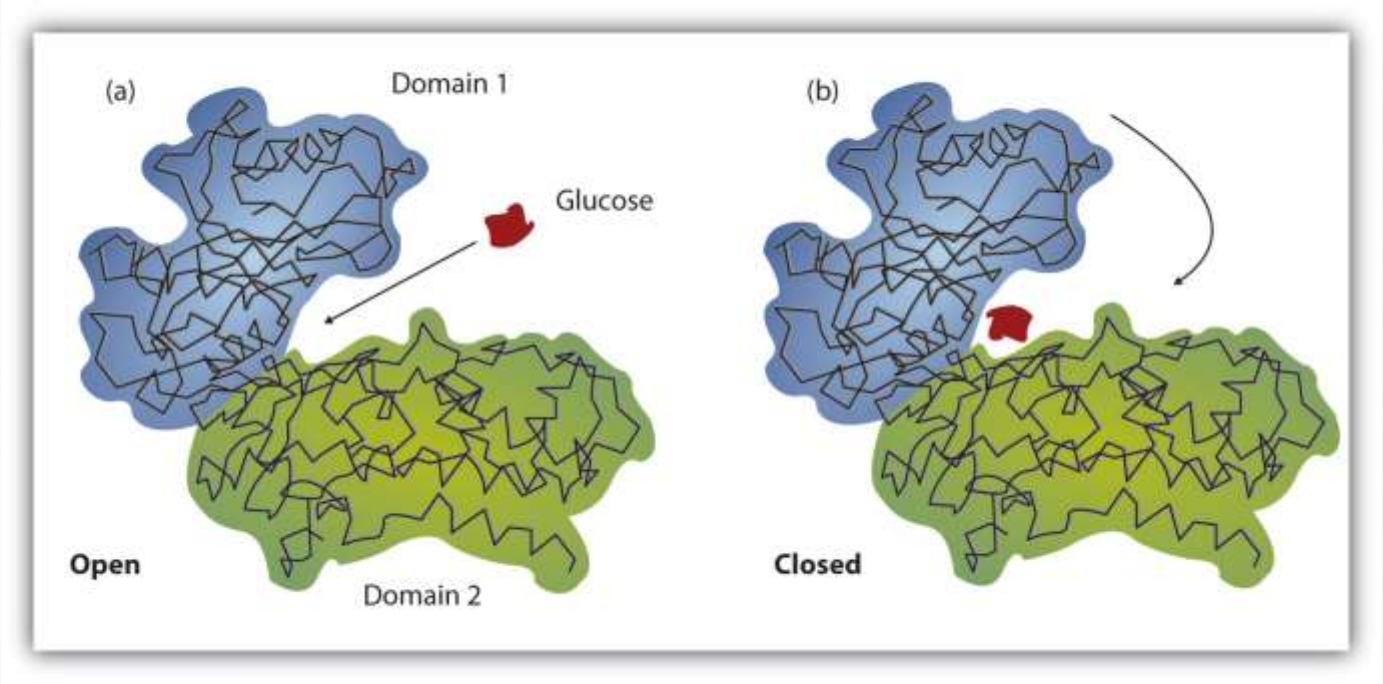
Group of Enzyme	Reaction Catalysed	Examples
1. Oxldoreductases	Transfer of hydrogen and oxygen atoms or electrons from one substrate to another.	Dehydrogenases Oxidases
2. Transferases	Transfer of a specific group (a phosphate or methyl etc.) from one substrate to another.	Transaminase Kinases
3. Hydrolases	Hydrolysis of a substrate.	Estrases Digestive enzymes
4. Isomerases	Change of the molecular form of the substrate.	Phospho hexo isomerase, Fumarase
5. Lyases	Nonhydrolytic removal of a group or addition of a group to a substrate.	Decarboxylases Aldolases
6. Ligases (Synthetases)	Joining of two molecules by the formation of new bonds.	Citric acid synthetase

Two Models for Enzyme-Substrate Interaction



1. a catabolic enzyme controlled reaction
2. an anabolic enzyme controlled reaction

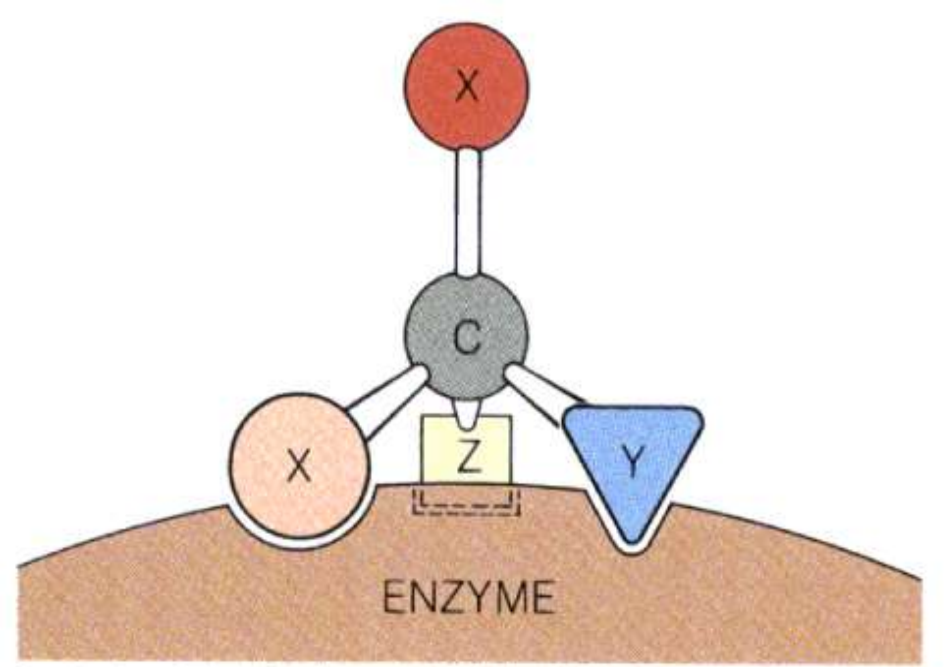
Induced Conformational Change in Hexokinase

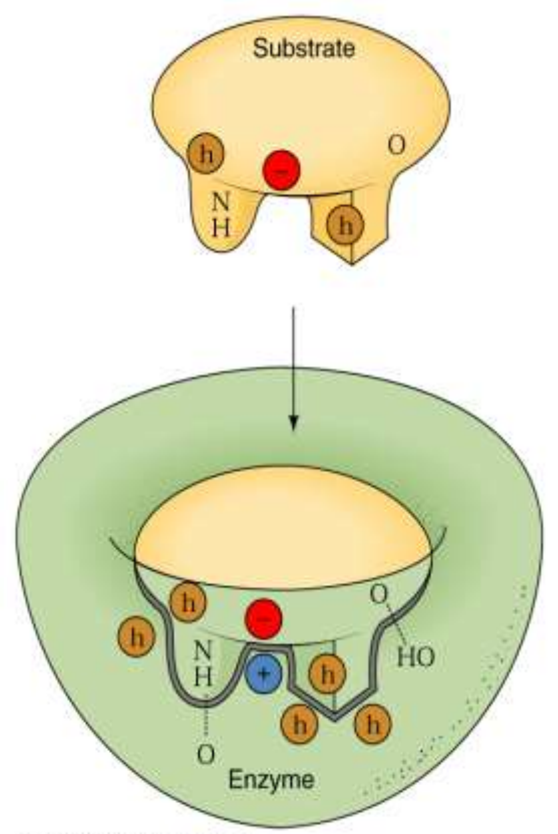


Coenzymes

Coenzyme	Examples of some chemical groups transferred	Dietary precursor in mammals
Thiamine pyrophosphate	Aldehydes	Thiamin (vitamin B_1)
Flavin adenine dinucleotide	Electrons	Riboflavin (vitamin B_2)
Nicotinamide adenine dinucleotide	Hydride ion ($:H^-$)	Nicotinic acid (niacin)
Coenzyme A	Acyl groups	Pantothenic acid, plus other molecules
Pyridoxal phosphate	Amino groups	Pyridoxine (vitamin B_6)
5′-Deoxyadenosylcobalamin (coenzyme B_{12})	H atoms and alkyl groups	Vitamin B_{12}
Biocytin	CO_2	Biotin
Tetrahydrofolate	One-carbon groups	Folate
Lipoate acid	Electrons and acyl groups	Not required in diet

Stereo specificity Conferred by an Enzyme

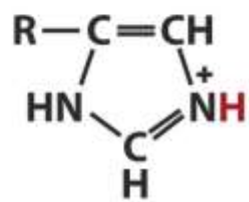
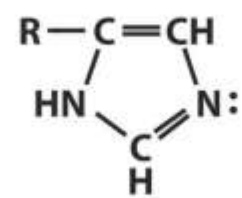
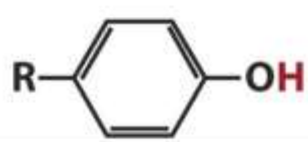




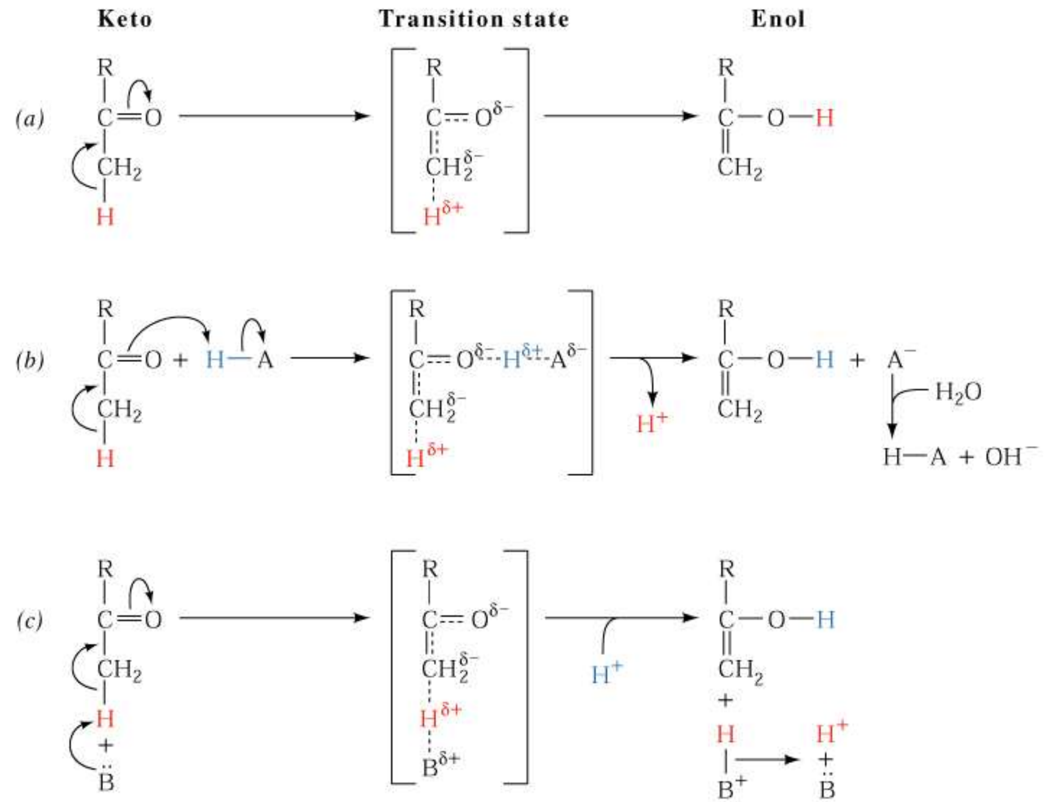
Catalytic Mechanisms

- Acid-base catalysis
- Covalent catalysis
- Metal ion catalysis
- Electrostatic catalysis
- Proximity and orientation effects
- Preferential binding to transition state (transition state stabilization)

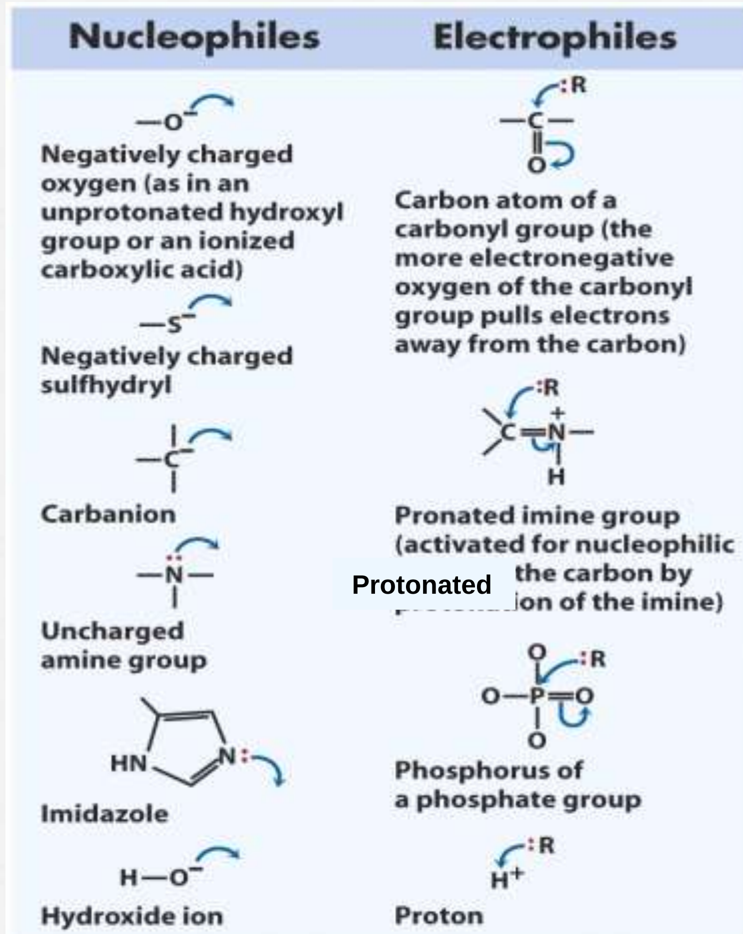
Acid-Base Catalysis

Amino acid residues	General acid form (proton donor)	General base form (proton acceptor)
Glu, Asp	$R-COOH$	$R-COO^-$
Lys, Arg	$R-\overset{+}{N}H_3$	$R-\ddot{N}H_2$
Cys	$R-SH$	$R-S^-$
His		
Ser	$R-OH$	$R-O^-$
Tyr		

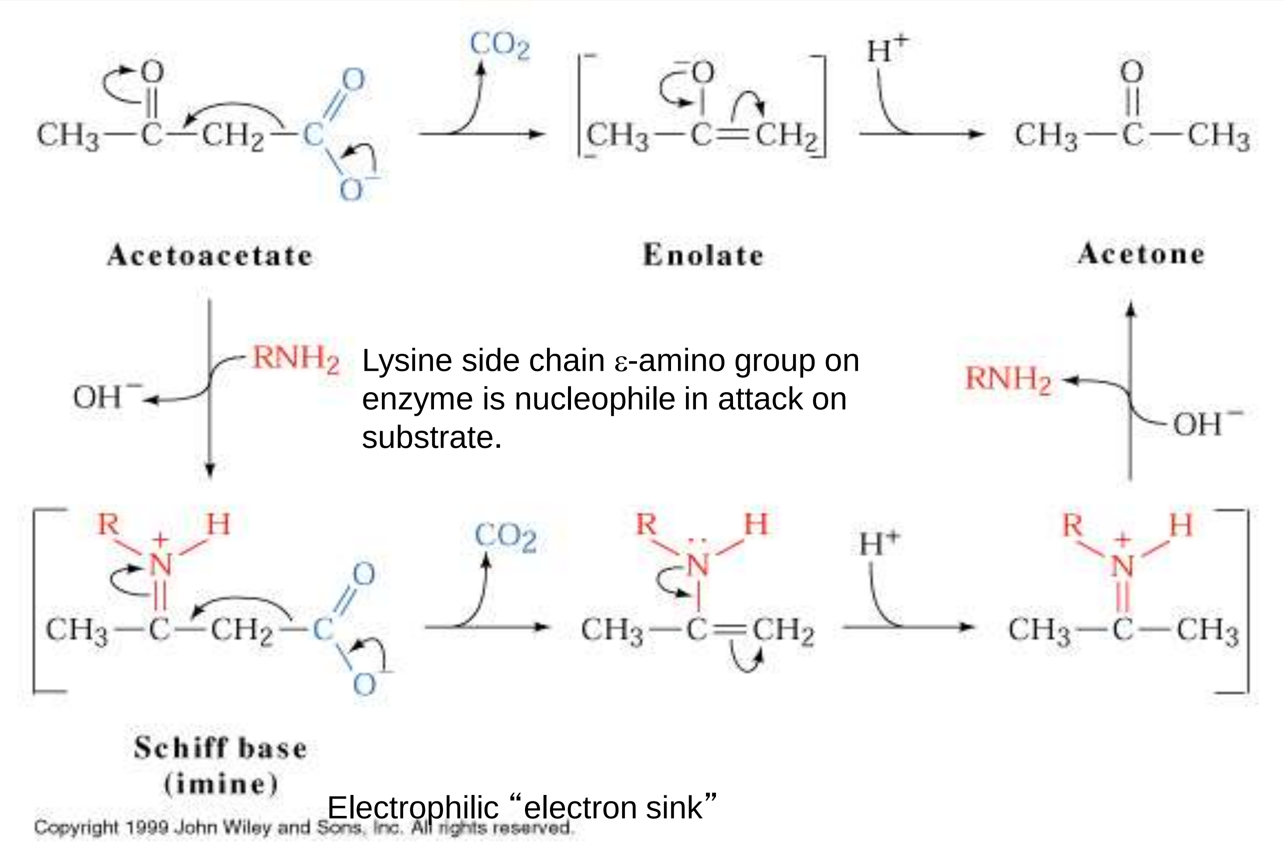
Keto-Enol Tautomerism: Uncatalyzed vs. Acid- or Base-Catalyzed



Covalent Catalysis: Nucleophiles and Electrophiles



Example of Covalent Catalysis: Decarboxylation of Acetoacetate



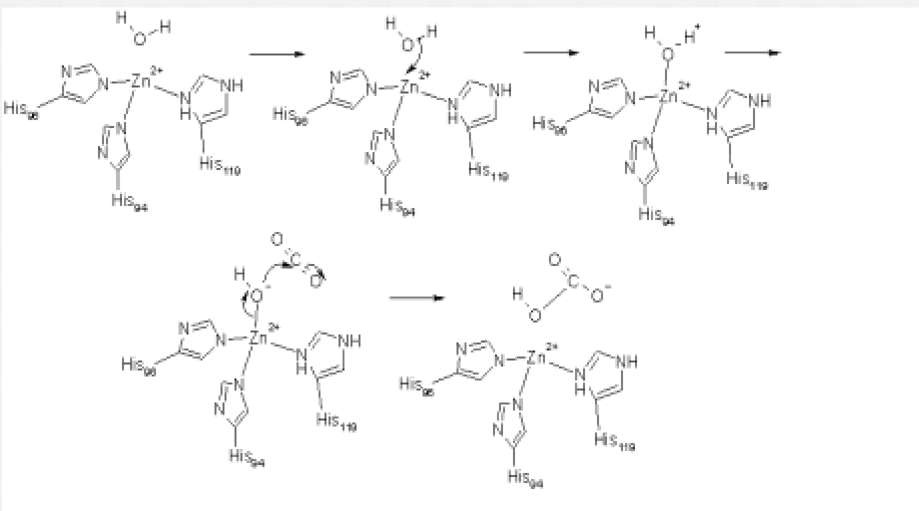
Lysine side chain ε-amino group on enzyme is nucleophile in attack on substrate.

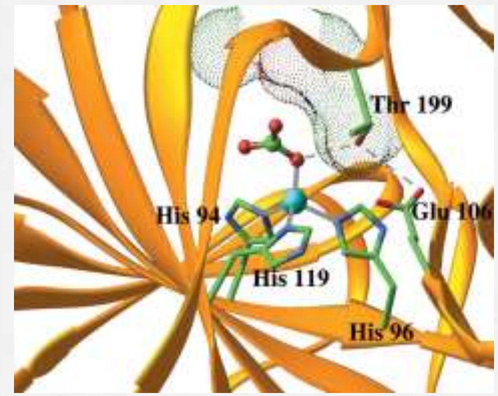
Electrophilic "electron sink"

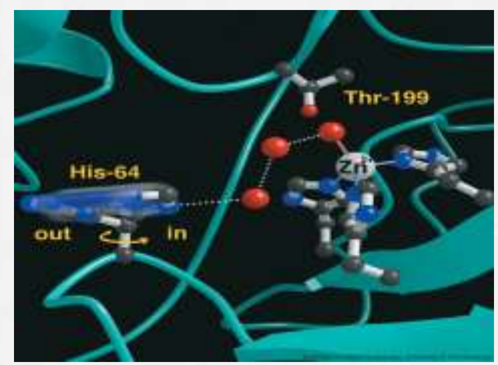
Example of Metal Ion Catalysis: Carbonic Anhydrase

Carbonic anhydrase catalyzes the reaction:

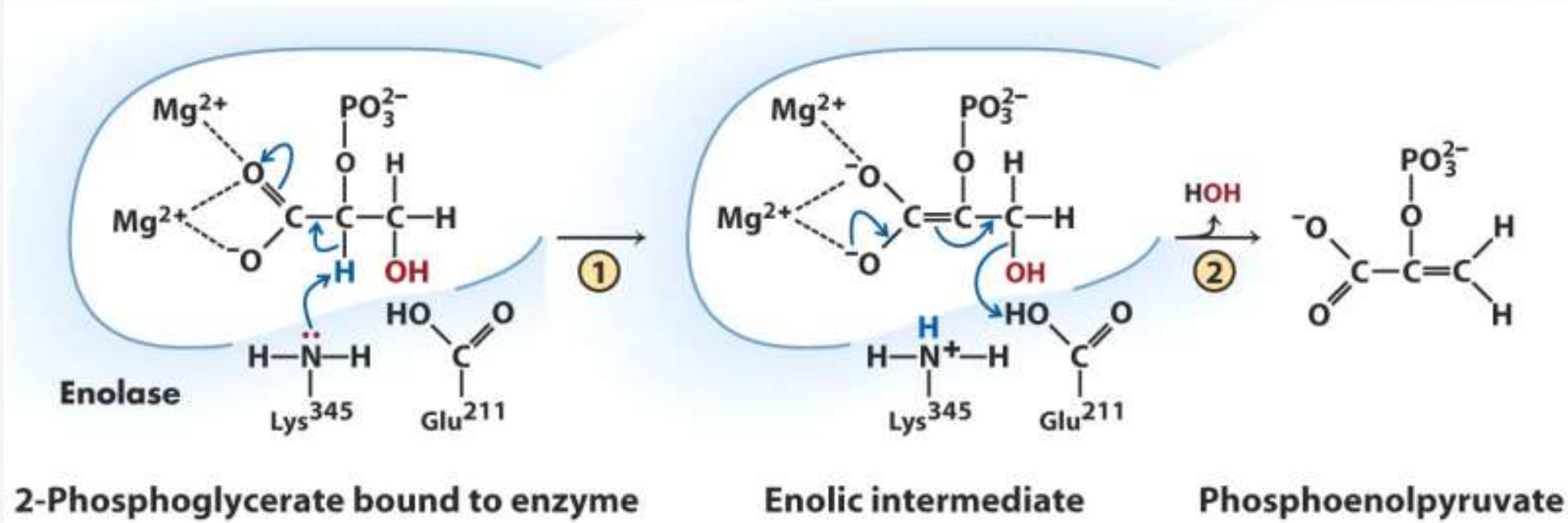
$CO_2 + H_2O \quad HCO_3^- + H^+$



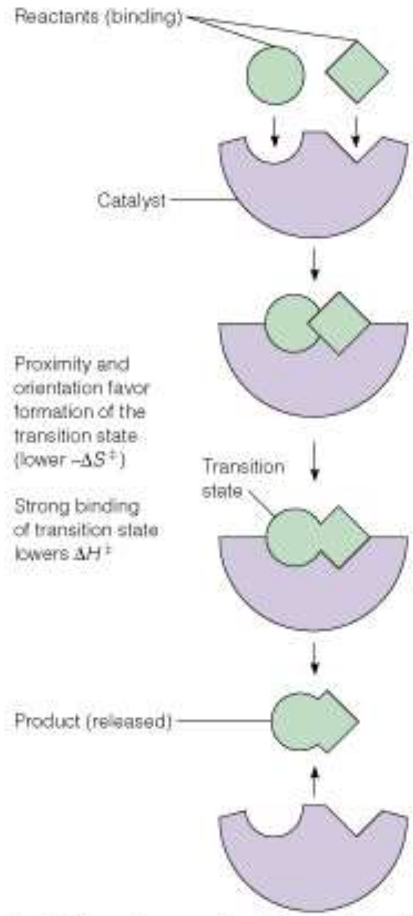




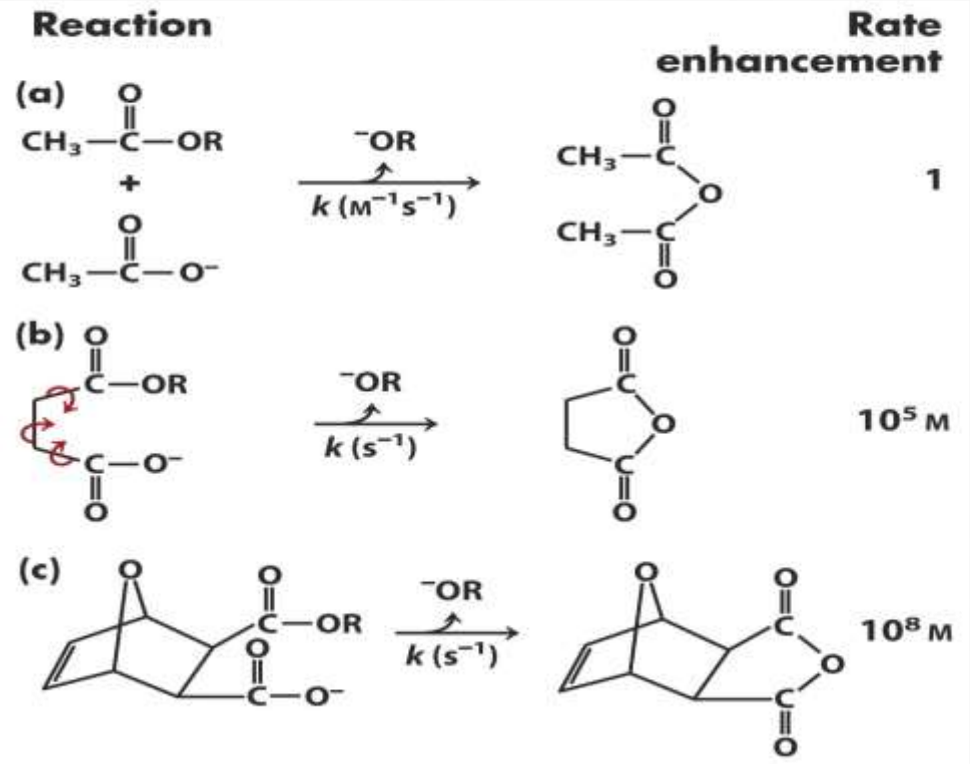
Enolase Mechanism



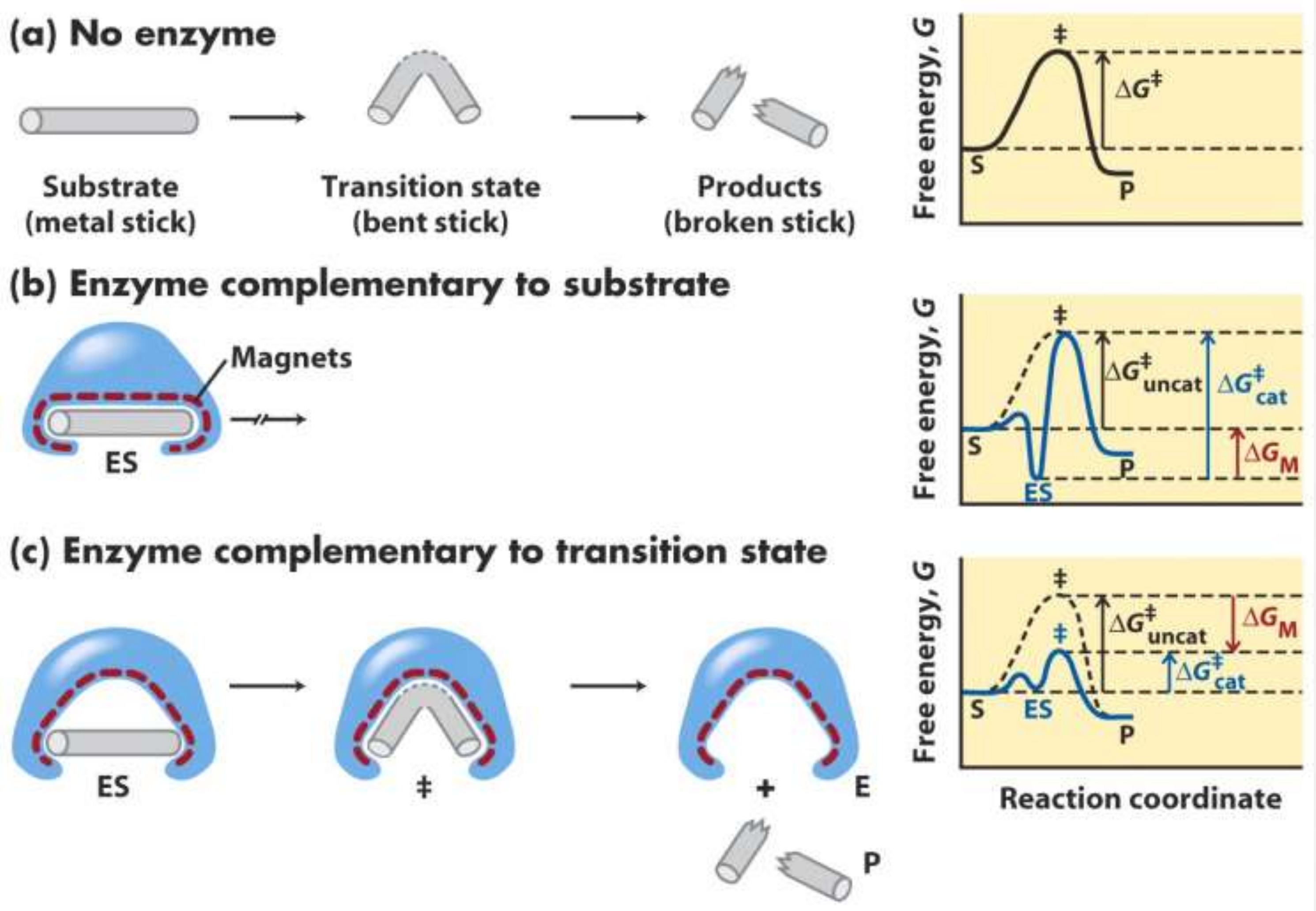
Entropic and Enthalpy Factors in Catalysis



Proximity and Orientation Effects

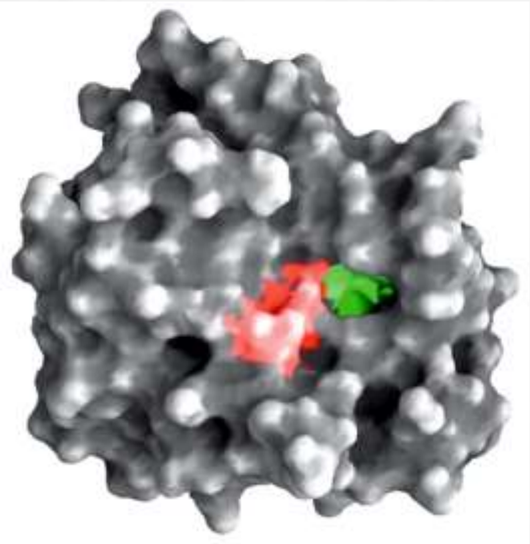
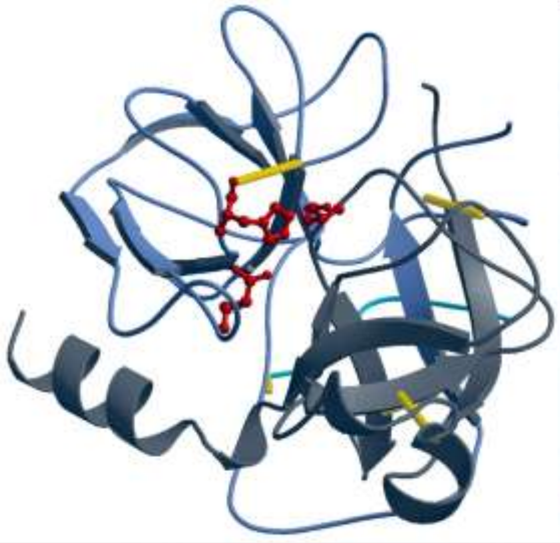
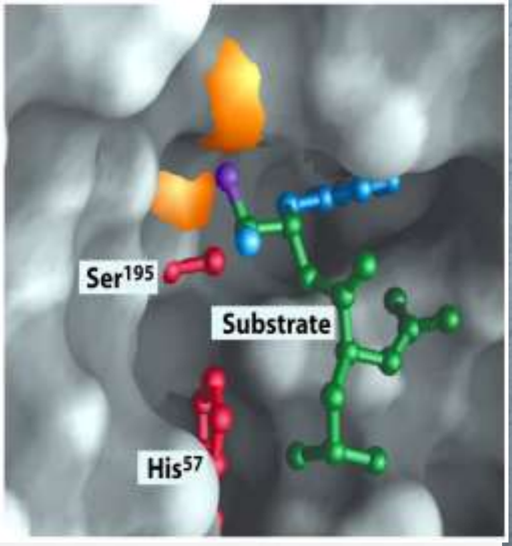


Enzymes Are Complementary to Transition State



Serine Protease Mechanism: Multiple Catalytic Mechanisms at Work

Structure of the Serine Protease Chymotrypsin



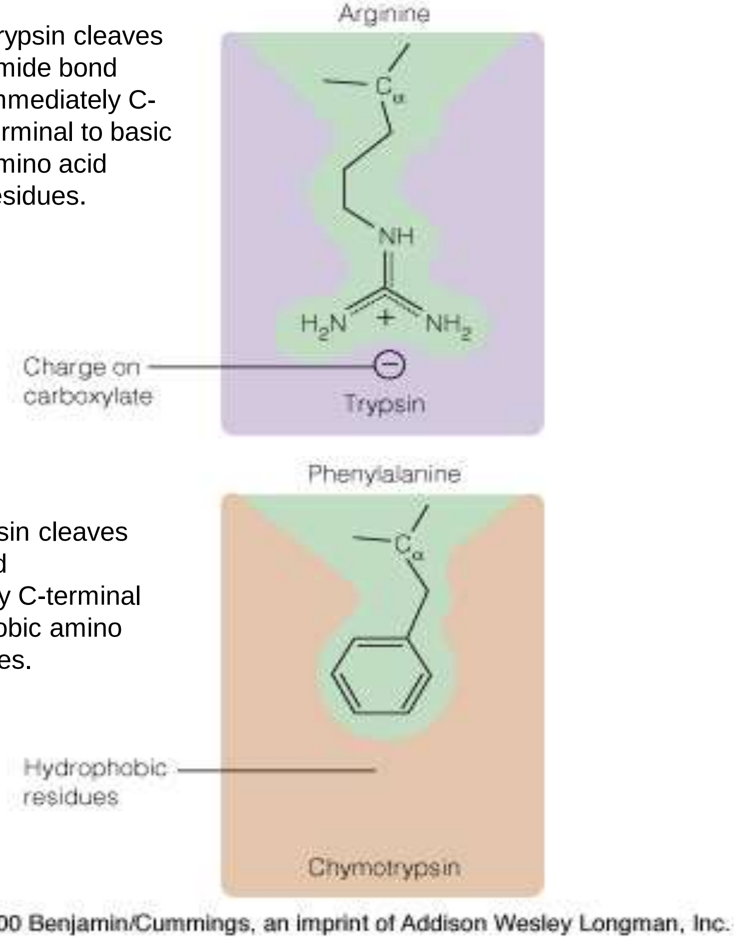
Serine Protease Substrate Specificity and Active-Site Pockets

Substrate specificity in serine proteases through active-site binding of side chain of amino acid residue adjacent to amide bond that will be cleaved.

$$R_1-\overset{\overset{\displaystyle O}{\|}}{C}-NH-R_2 + H_2O \xrightarrow{\text{trypsin}} R_1-\overset{\overset{\displaystyle O}{\|}}{C}-O^- + H_3\overset{+}{N}-R_2$$

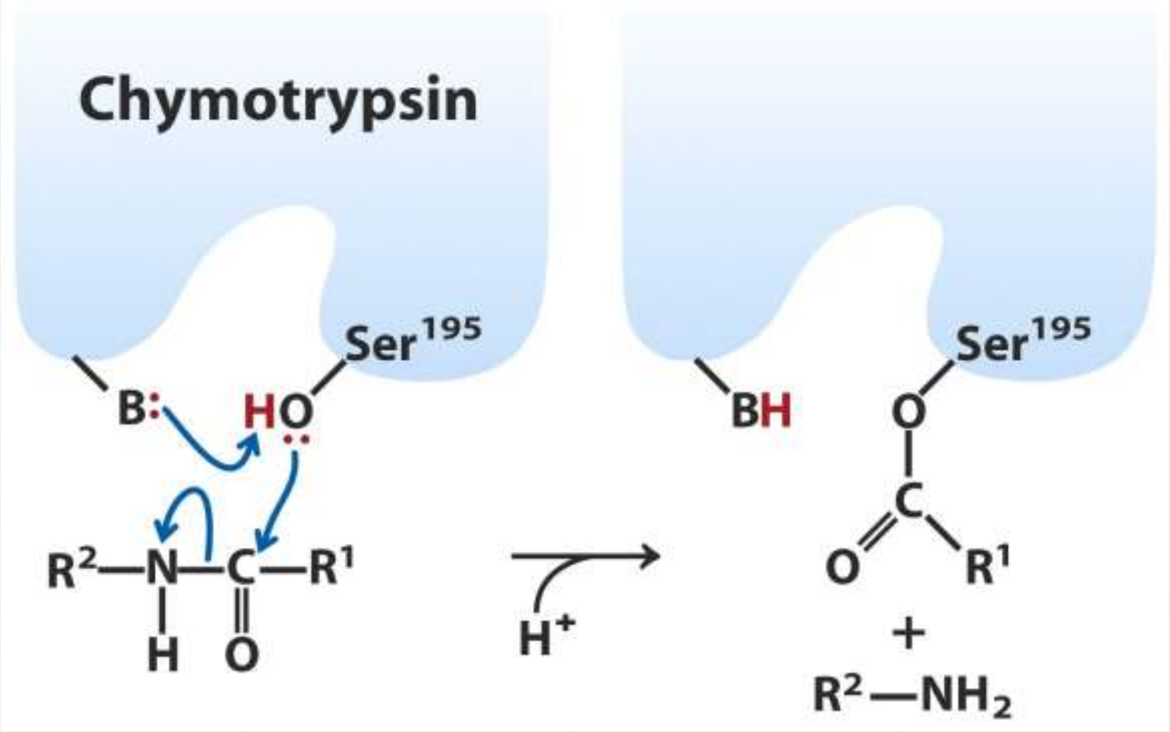
Polypeptide

Trypsin cleaves amide bond immediately C-terminal to basic amino acid residues.

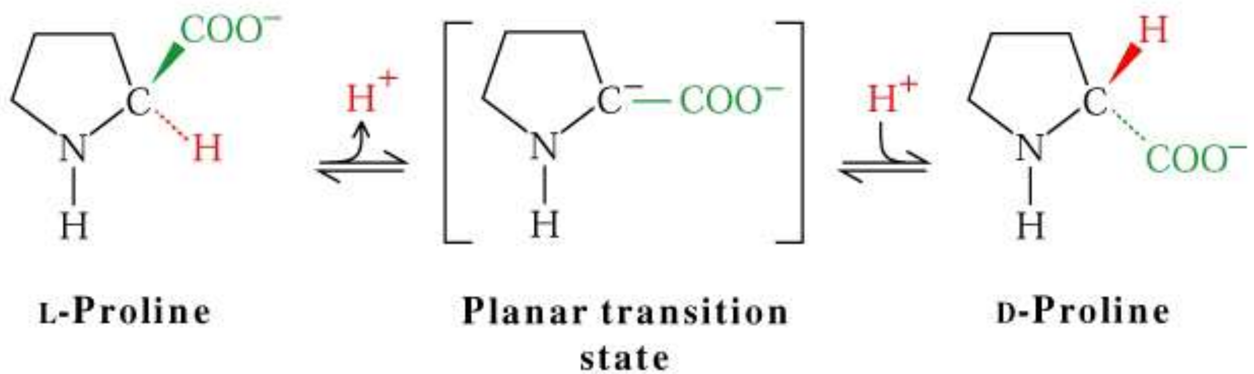


Chymotrypsin cleaves amide bond immediately C-terminal to hydrophobic amino acid residues.

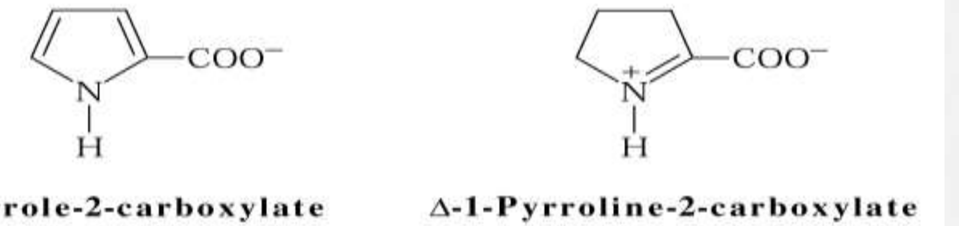
Serine Nucleophile in Serine Proteases



Transition State in Proline Racemase Reaction and Transition State Analogs



Proline racemase preferentially binds transition state, stabilizing it, and is potently inhibited by transition state analogs.

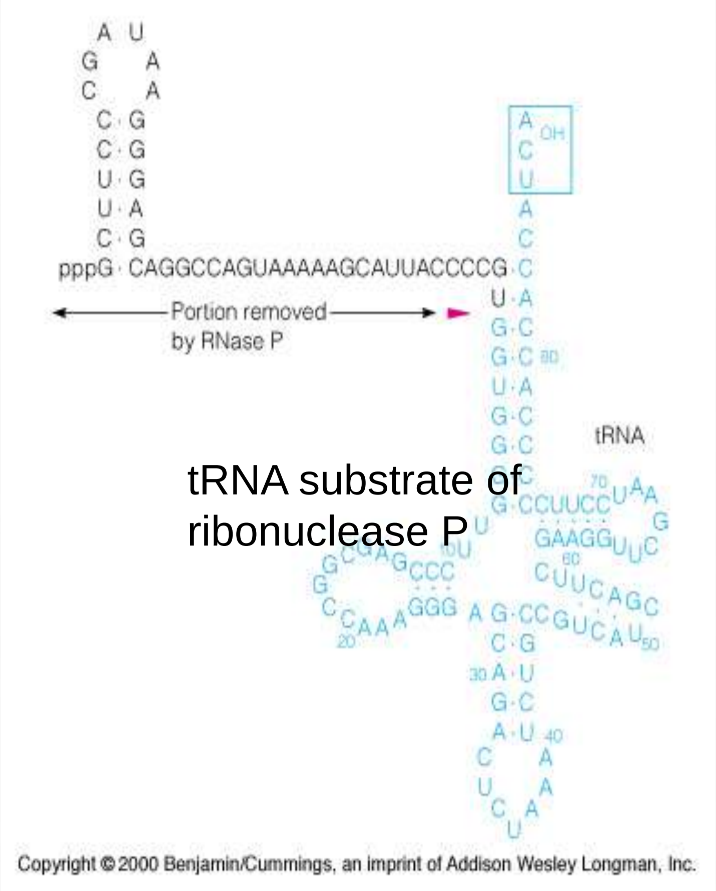


RNA-Based Catalysts (Ribozymes)

Cleavage of a Typical Pre-tRNA by Ribonuclease P

Ribonuclease P is a ribonucleoprotein (RNA- and protein-containing complex), and the catalytic component is RNA.

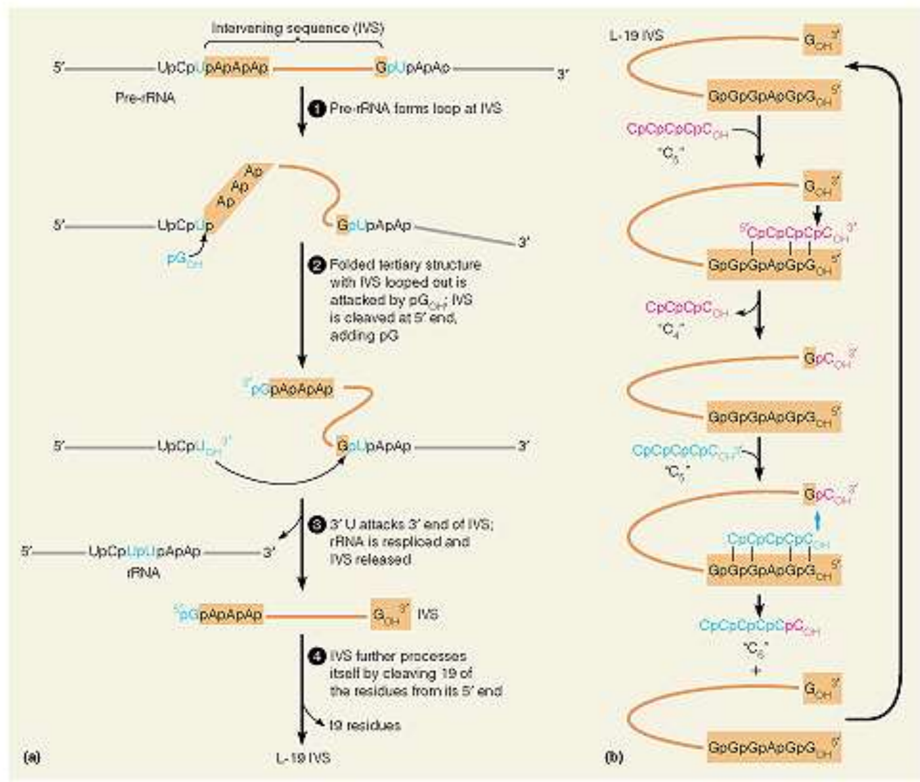
An even more complex example of an RNA- and protein-containing enzyme system is the ribosome. The central catalytic activity of the ribosome (peptide bond formation) is catalyzed by an RNA component.



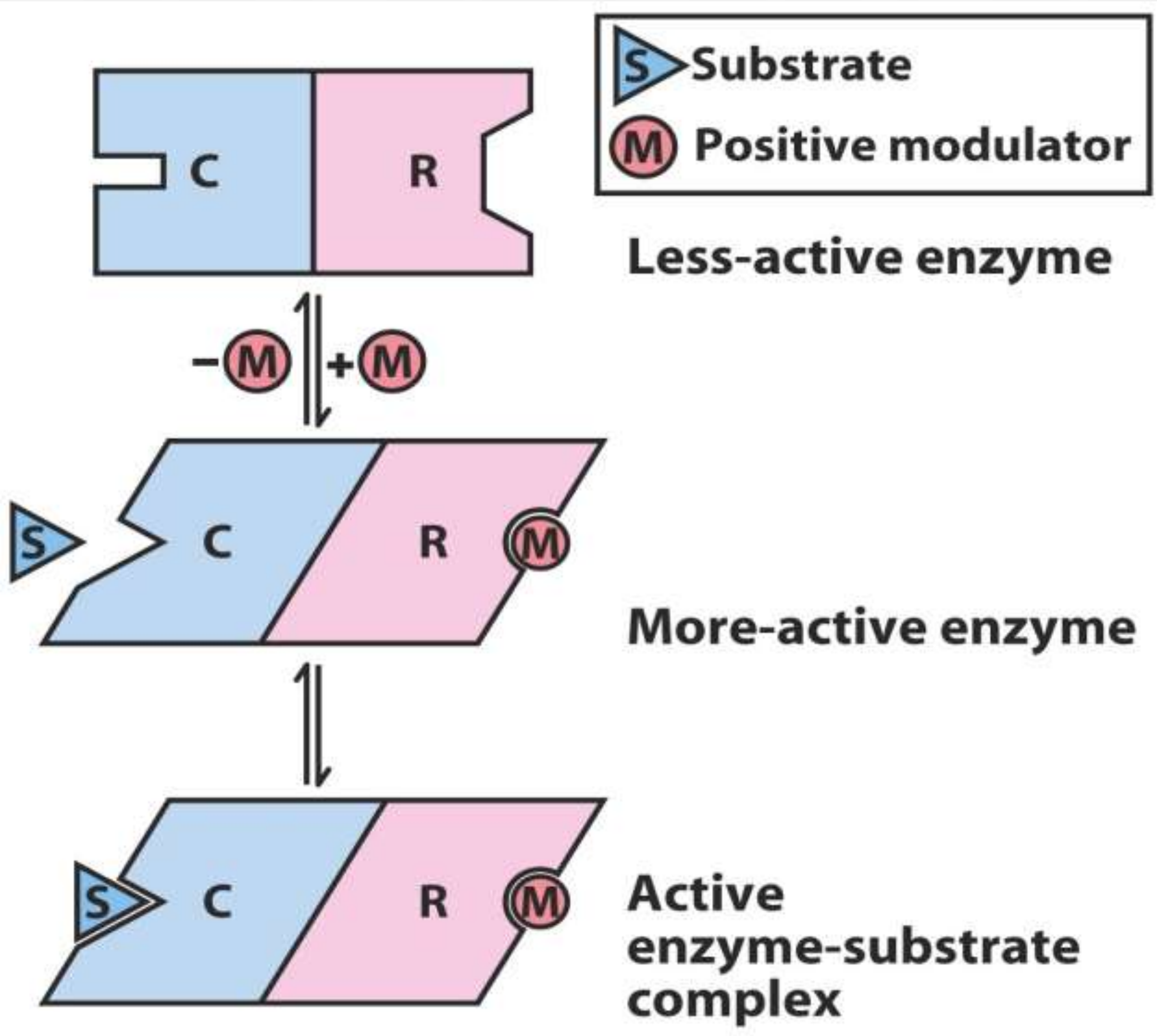
tRNA substrate of ribonuclease P

Catalysis by the Intervening Sequence in *Tetrahymena* Preribosomal RNA

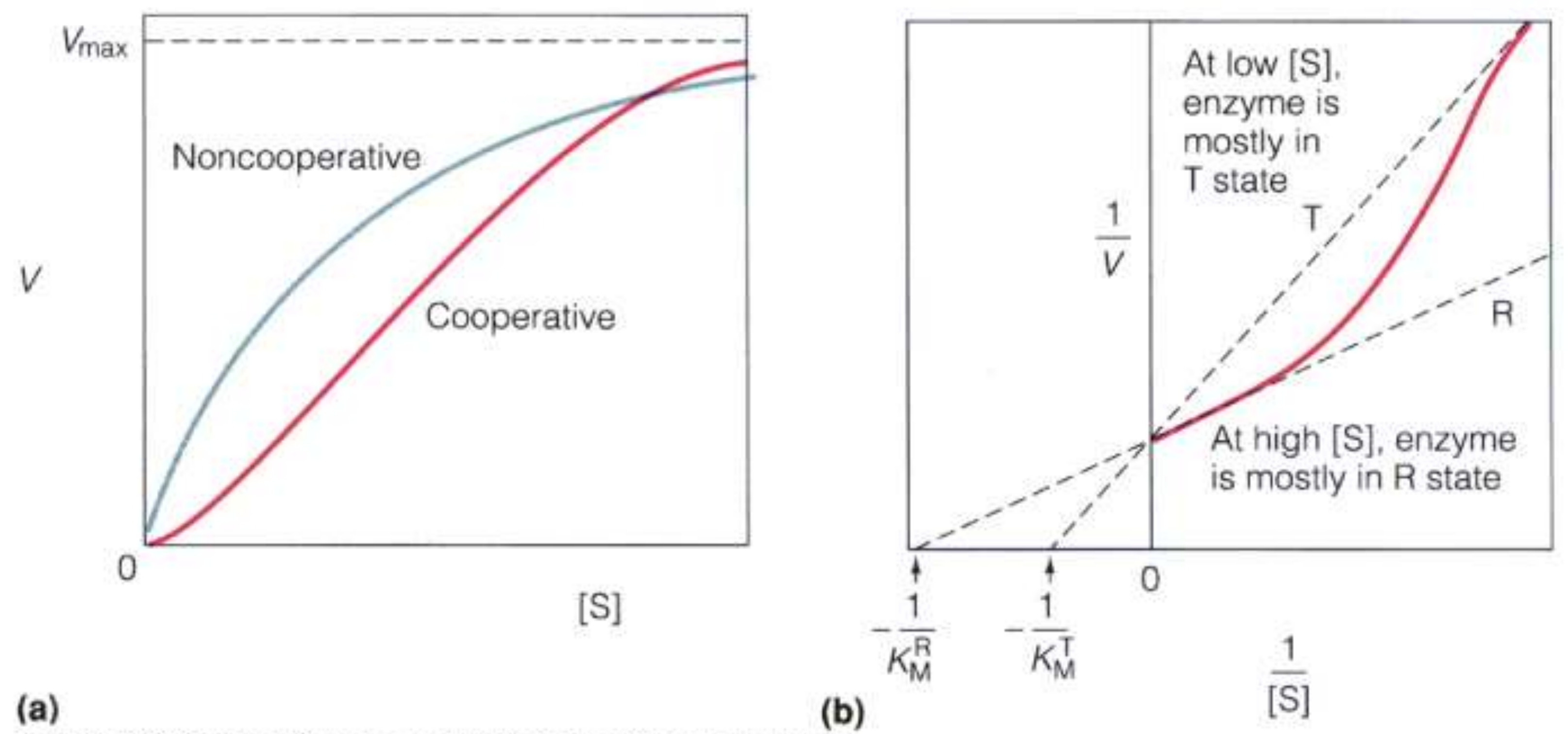
RNA by itself without any protein can be catalytic.



Enzyme Regulation


S Substrate
M Positive modulator
C
R
Less-active enzyme
−M
+M
S
C
R
M
More-active enzyme
S
C
R
M
Active
enzyme-substrate
complex

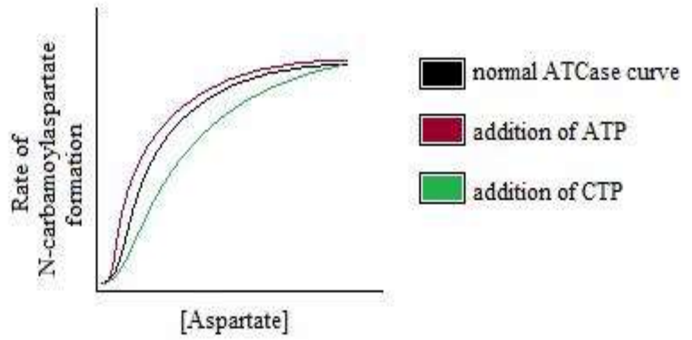
Effect of Cooperative Substrate Binding on Enzyme Kinetics



Cooperative enzymes do not obey simple Michaelis-Menten kinetics.

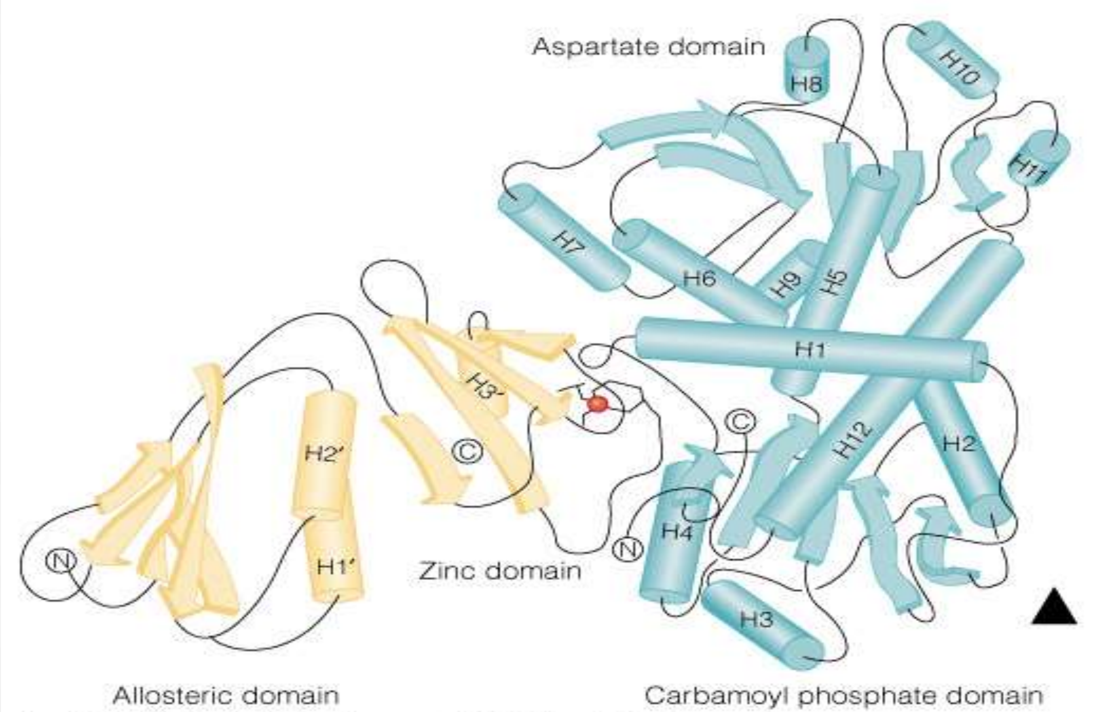
Regulation of ATCase by ATP and CTP

ATP is a positive heterotropic allosteric effector of ATCase, while CTP is a negative heterotropic allosteric effector.

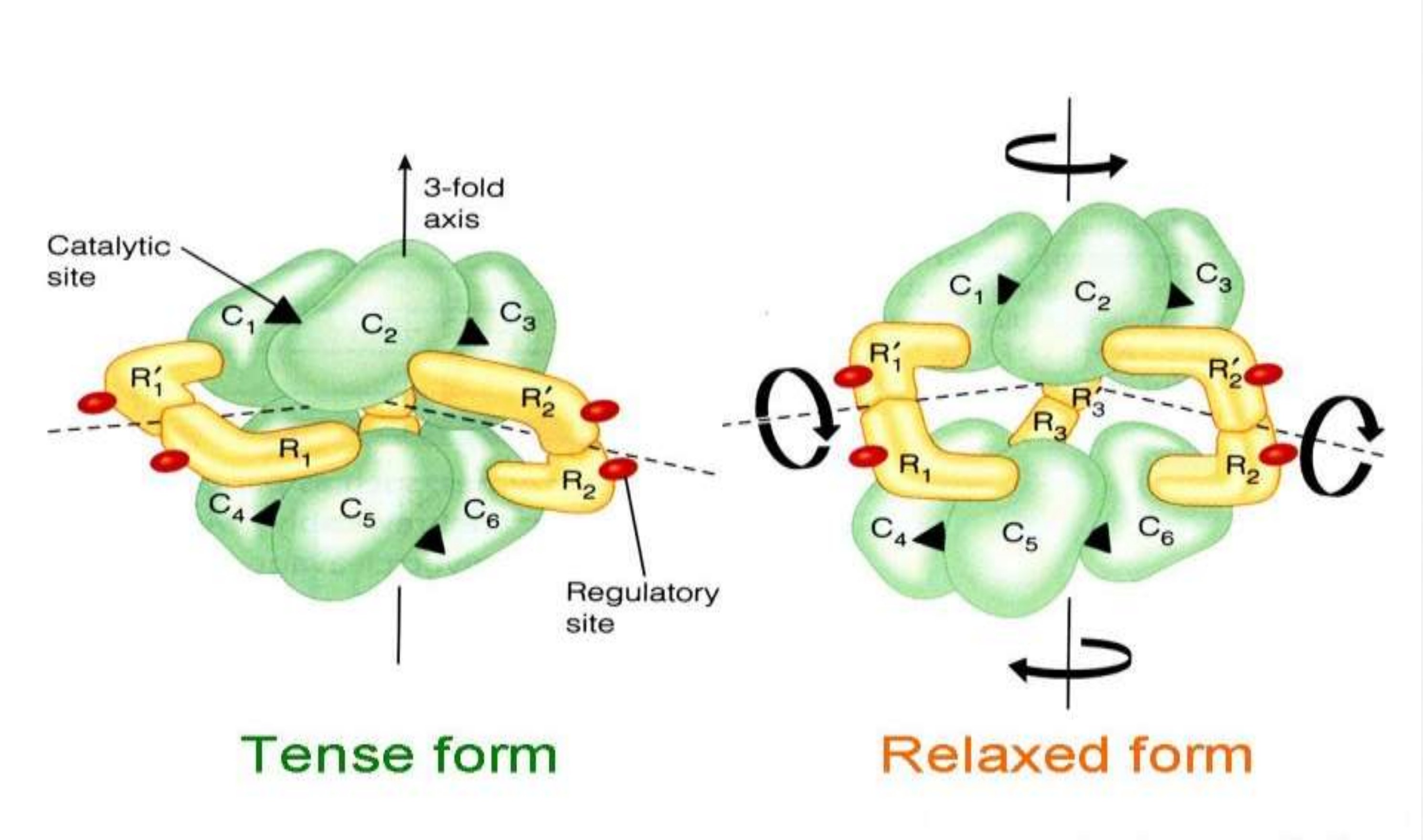


ATP and CTP effect the responses of ATCase to the substrate.

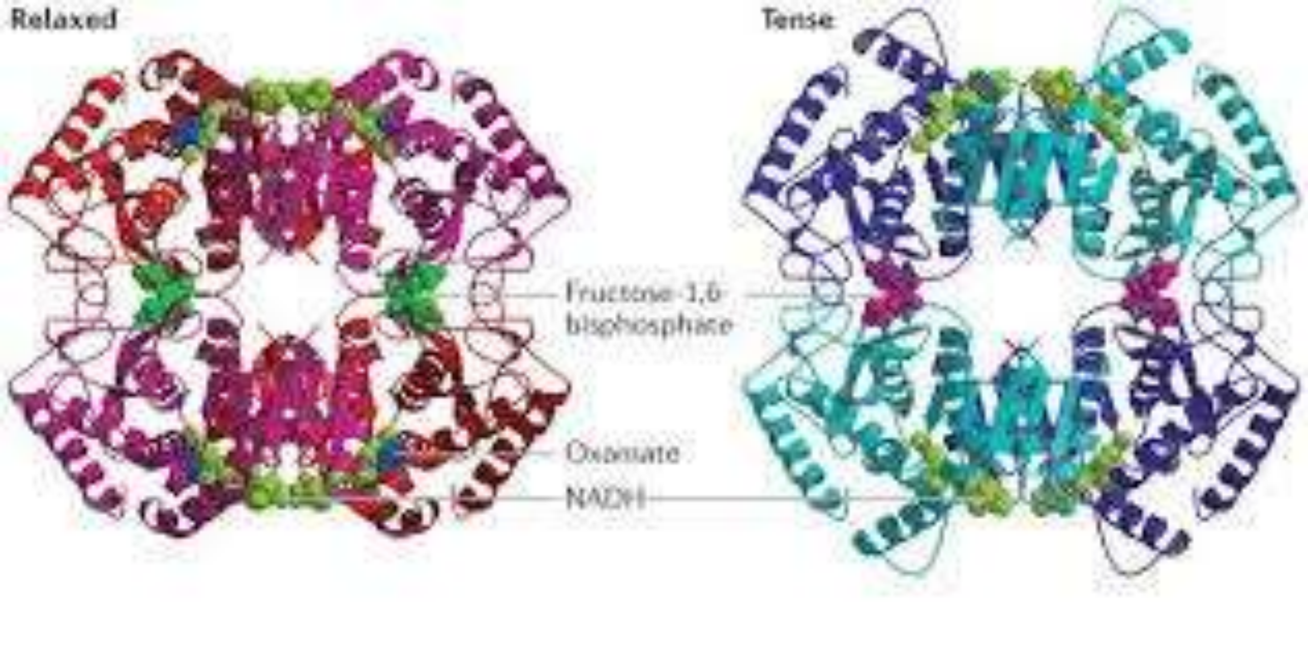
Detailed Structure of One Catalytic Subunit and Adjacent Regulatory Subunit of ATCase



Quaternary Structure of ATCase in T State and R State



X-Ray Structure of Aspartate Transcarbamoylase



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