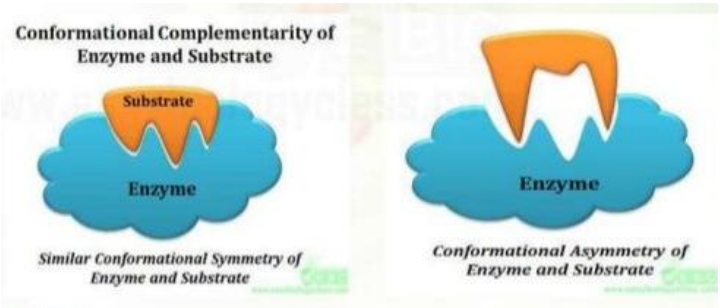
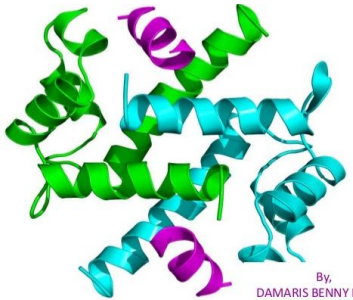
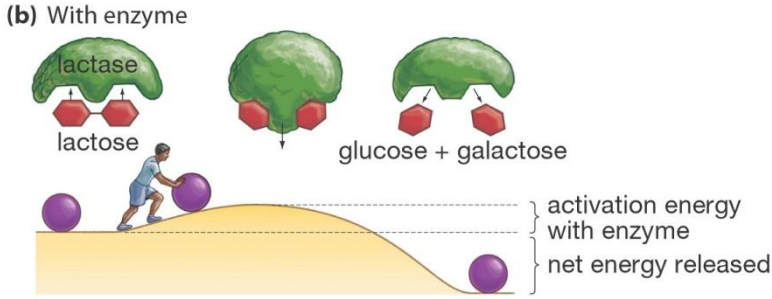
Enzyme Structure, Classification & Mechanism of Action





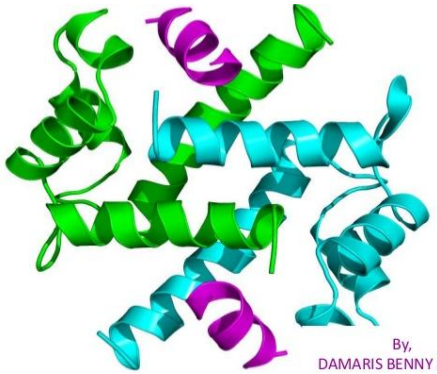


Enzymes: Fundamental Key To Life

There are two fundamental conditions for life.

1) The living entity must be able to self-replicate (Reproduction)

2) The organism must be able to catalyze biochemical reactions efficiently and selectively.



Enzyme: Biological Catalyst

- Enzymes are highly effective catalysts, commonly enhancing reaction rates by a factor of 10^5 to 10^{17}.

- Enzymes have extraordinary catalytic power, a **high degree of specificity for their substrates**, accelerate chemical reactions tremendously, and function in aqueous solutions under very mild conditions of temperature and pH.

TABLE 6–5 Some Rate Enhancements Produced by Enzymes

Cyclophilin	10^5
Carbonic anhydrase	10^7
Triose phosphate isomerase	10^9
Carboxypeptidase A	10^{11}
Phosphoglucomutase	10^{12}
Succinyl-CoA transferase	10^{13}
Urease	10^{14}
Orotidine monophosphate decarboxylase	10^{17}

Enzyme: Biological Catalyst

- ✓ **Enzymes are central to every biochemical process.**
- ✓ **Acting in organized sequences, they catalyze the hundreds of stepwise reactions that degrade nutrient molecules, conserve and transform chemical energy, and make biological macromolecules from simple precursors.**
- ✓ **Not altered or consumed during reaction.**
- ✓ **Reusable**

Catalyst

- **A *catalyst* is a chemical that speeds up the reaction but is not used up in the reaction**
  - **Lowers the activation energy needed to start a reaction**
  - **Is not used up during the reaction**
  - **Is unchanged after a reaction**

Early Research on Enzymes

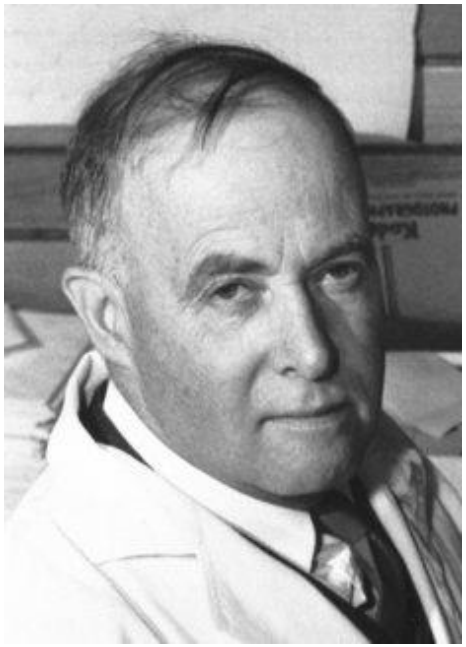
- **In 1850s, Louis Pasteur concluded that fermentation of sugar into alcohol by yeast is catalyzed by "ferments."**
- **In 1897, Eduard Buchner discovered that yeast extracts could ferment sugar to alcohol, proving that fermentation was promoted by molecules that continued to function when removed from cells.**
- **Frederick W. Kühne called these molecules enzymes.**

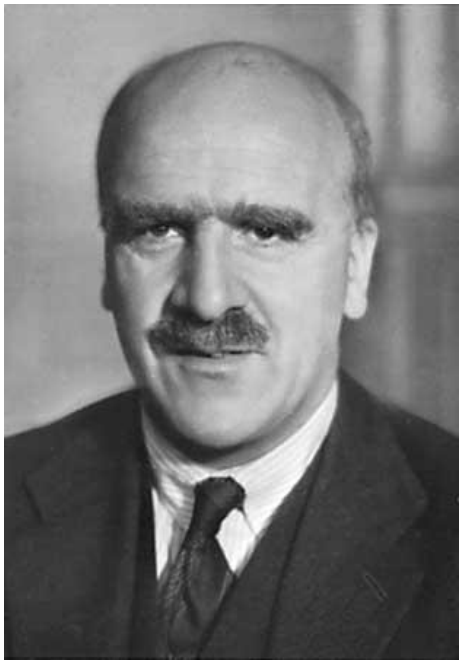
Eduard Buchner was German chemist and zymologist, awarded the 1907 Nobel Prize in chemistry for his work on fermentation

Early Research on Enzymes

- **The isolation and crystallization of urease by James Sumner in 1926. He postulated that all enzymes are proteins.**

- **J. B. S. Haldane made the remarkable suggestion that weak bonding interactions between an enzyme and its substrate might be used to catalyze a reaction.**





James B. Sumner was an American chemist. He discovered that enzymes are proteins and can be crystallized, for which he got Nobel Prize in 1946

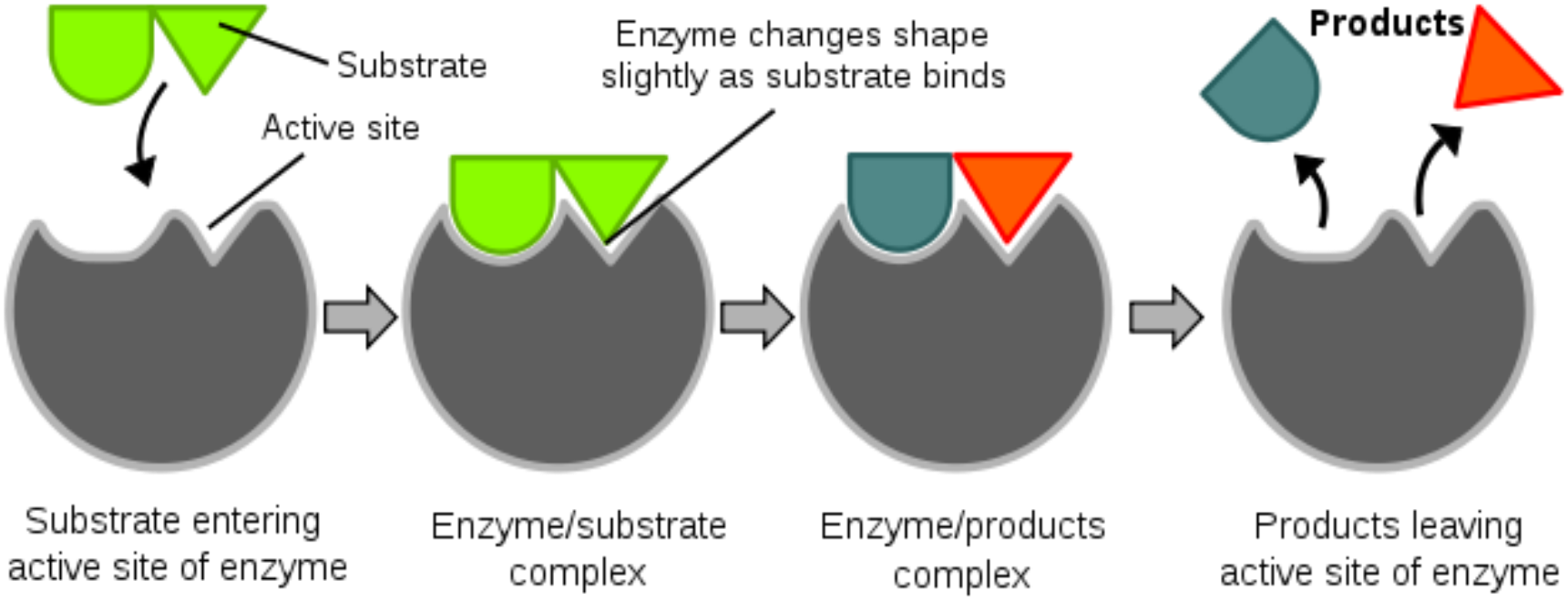
J. B. S. Haldane was a British-Indian scientist wrote a treatise entitled *Enzymes.*

Chemical Nature of enzymes

- **Most Enzymes Are Proteins (**with the exception of a small group of catalytic RNA molecules, all enzymes are proteins.)
- Their **catalytic activity depends on the integrity of their native protein conformation.**
- If an **enzyme is denatured or dissociated into its subunits, catalytic activity is usually lost.** Thus the primary, secondary, tertiary, and quaternary structures of protein enzymes are essential to their catalytic activity.
- Many require **non-protein coenzymes or cofactors** for their catalytic function.

ACTIVE SITES

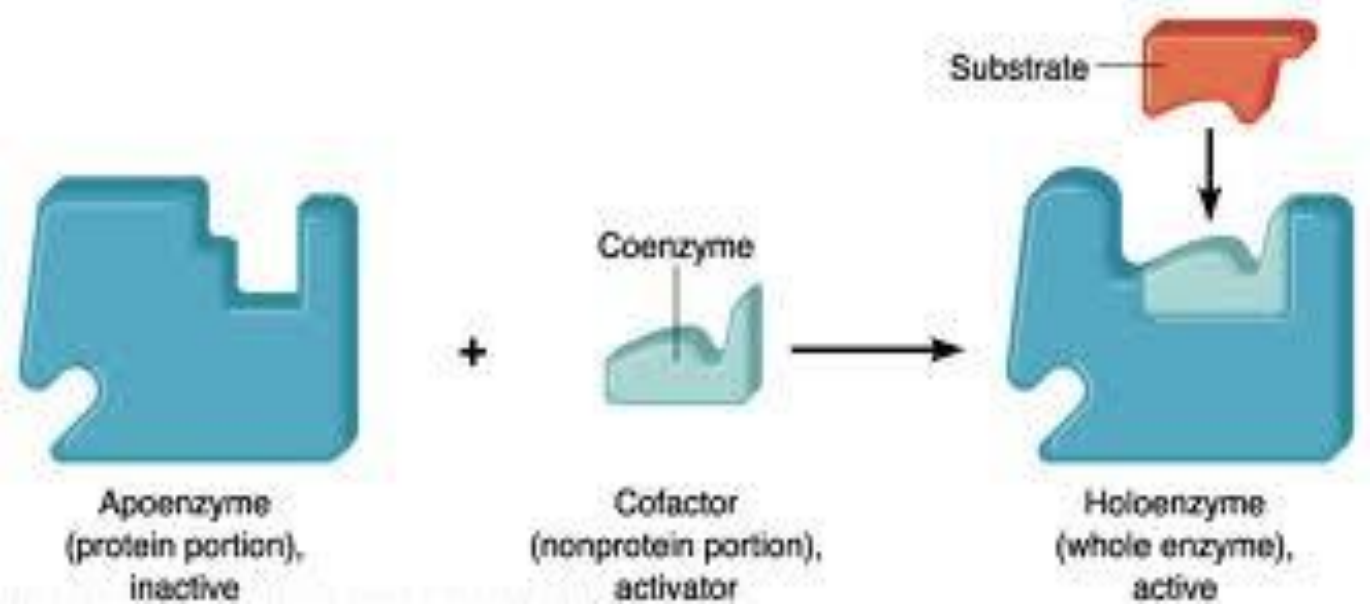
- **Enzyme molecules contain a special pocket or cleft called the active sites.**



APOENZYME and HOLOENZYME

- A complex enzyme **without its non protein moiety** is termed as **apoenzyme** and it is inactive.

- Holoenzyme is an active enzyme **with its non protein component.**



Prosthetic Group

- **Prosthetic Group:**
- **A prosthetic group is a non-protein chemical compound that are distinguished by their tight, stable incorporation into a protein's structure by covalent or non-covalent forces and is required for catalysis.**

Co-enzymes & Co-factors

Co-enzyme: **The non-protein component** (complex organic or metalloorganic molecule called), loosely bound to apoenzyme by non-covalent bond.

- **Examples : vitamins or compound derived from vitamins.**

- **Co-factor:** The non-protein component **(either one or more inorganic ions, such as Fe+2, Mg+2, Mn+2, or Zn+2),** bind in a transient, dissociable manner either to the enzyme **(metal activated enzymes)**

Some inorganic elements that serve as Co-enzymes for enzymes

Biocytin
Coenzyme A
5'-Deoxyadenosylcobalamin
(coenzyme B_{12})
Flavin adenine dinucleotide
Lipoate
Nicotinamide adenine dinucleotide
Pyridoxal phosphate
Tetrahydrofolate
Thiamine pyrophosphate

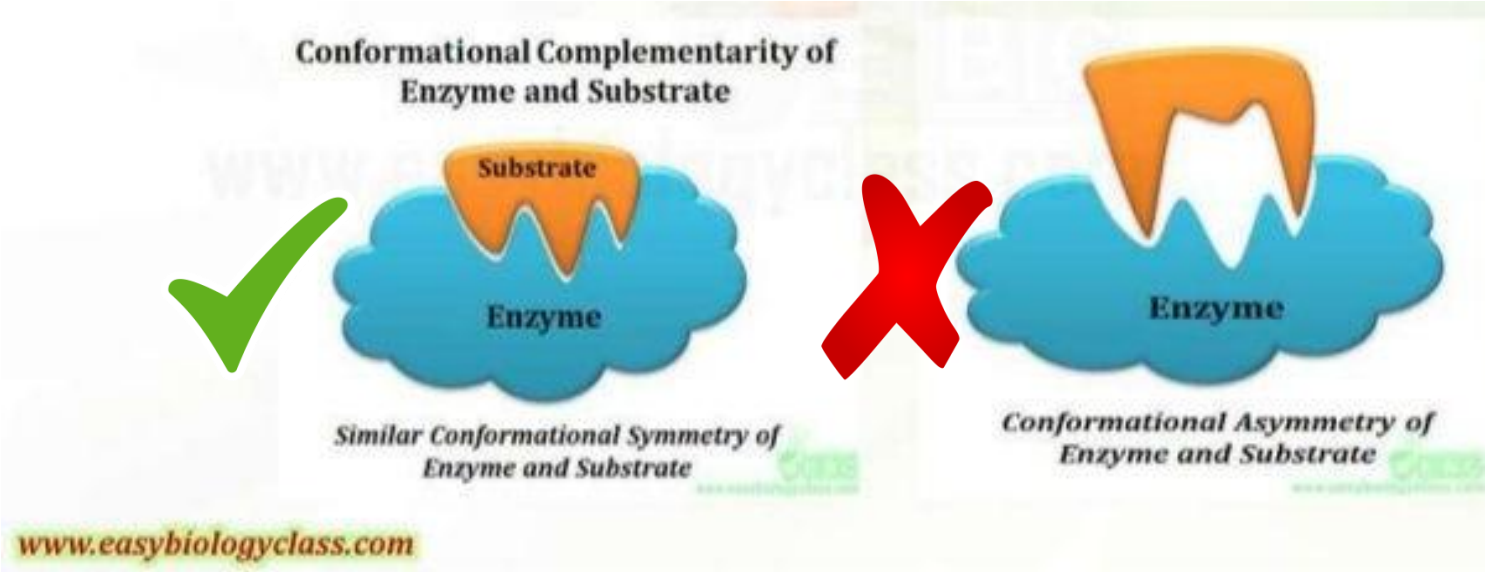
Some inorganic elements that serve as Co-factors for enzymes

Cu^{2+}	Cytochrome oxidase
Fe^{2+} or Fe^{3+}	Cytochrome oxidase, catalase, peroxidase
K^{+}	Pyruvate kinase
Mg^{2+}	Hexokinase, glucose 6-phosphatase, pyruvate kinase
Mn^{2+}	Arginase, ribonucleotide reductase
Mo	Dinitrogenase
Ni^{2+}	Urease
Se	Glutathione peroxidase
Zn^{2+}	Carbonic anhydrase, alcohol dehydrogenase, carboxypeptidases A and B

Enzyme Specificity

- **Specificity is the ability of an enzyme to choose exact substrate from a group of similar chemical molecules.**

- The specificity is actually a molecular recognition mechanism and it operates through the structural and conformational complementarity between enzyme and substrate



Degree of Enzyme Specificity

- Enzymes have varying degrees of **specificity** for substrates

- Enzymes may recognize and catalyze:
  - **a single substrate**
  - **a group of similar substrates**
  - **a particular type of bond**

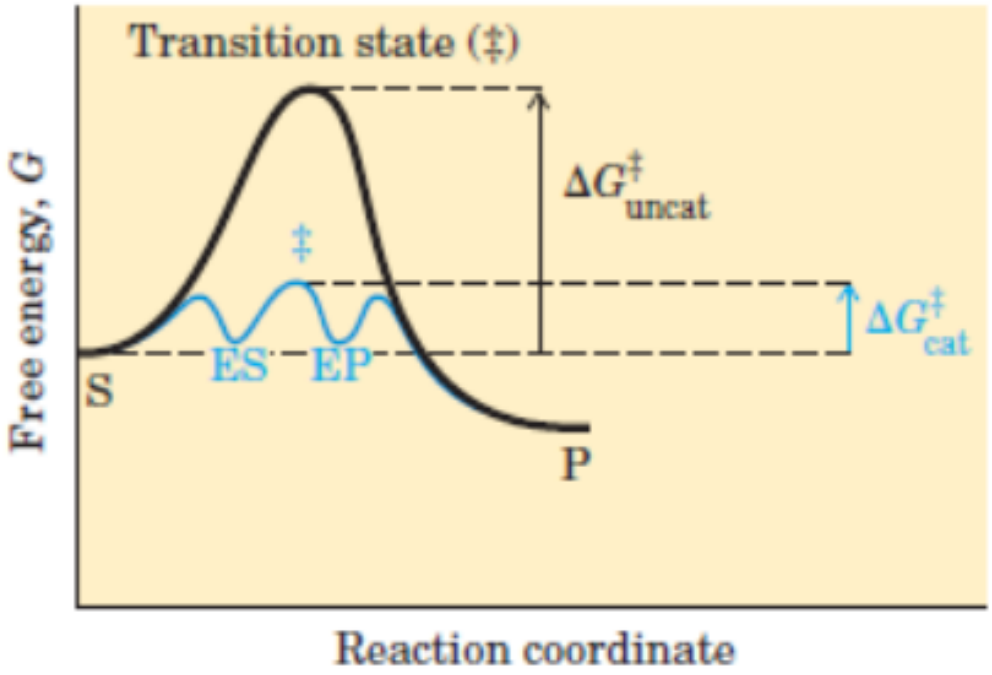
Table 21.2 Types of Enzyme Specificity

Type	Reaction Type	Example
Absolute	Catalyze one type of reaction for a single substrate	Urease catalyzes only the hydrolysis of urea
Group	Catalyze one type of reaction for similar substrates	Hexokinase adds a phosphate group to hexoses
Linkage	Catalyze one type of reaction for a specific type of bond	Chymotrypsin catalyzes the hydrolysis of peptide bonds

Activation energy or Energy of Activation

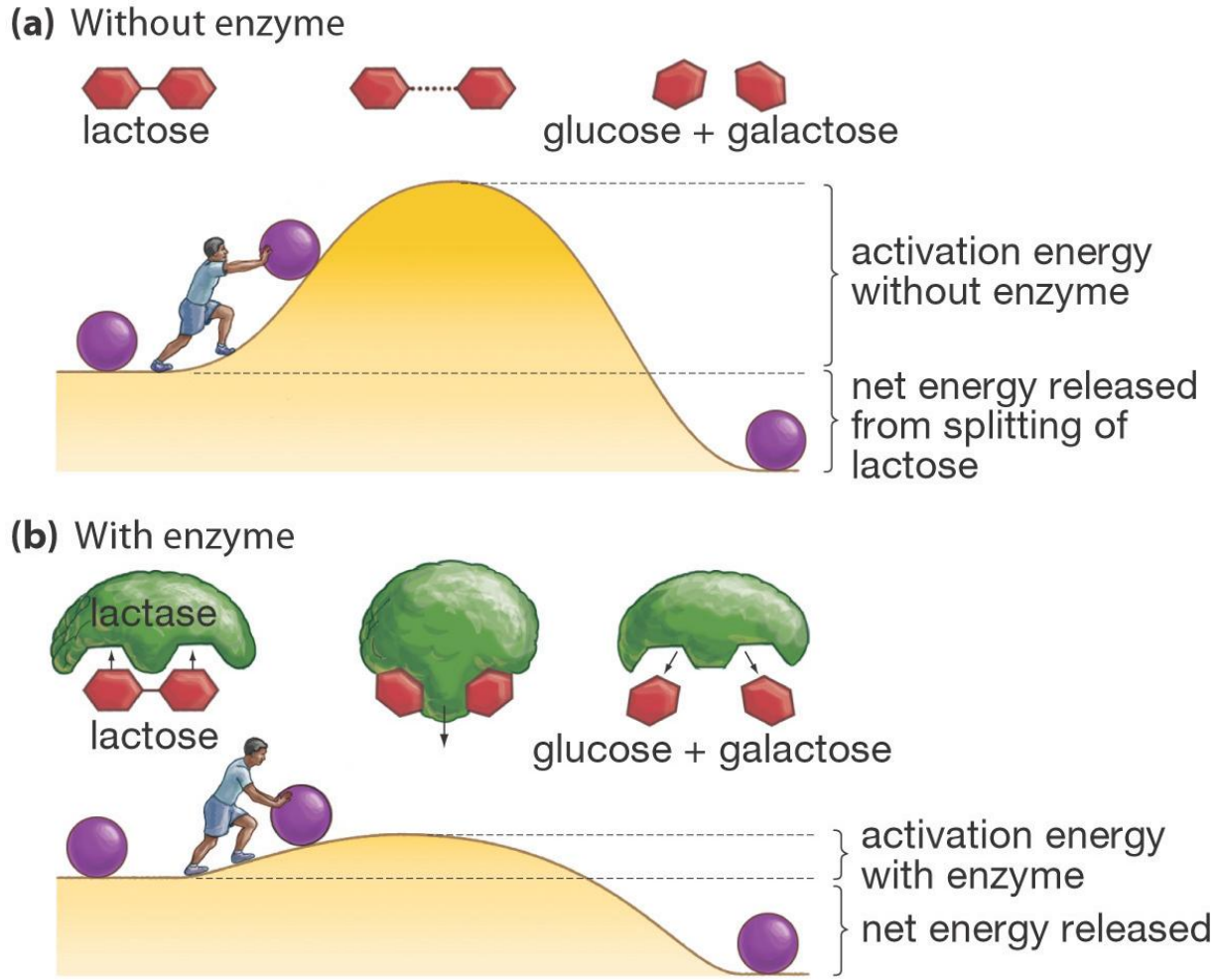
- **All chemical reactions require some amount of energy to get them started. This energy is called activation energy**

The difference between the energy levels of the ground state and the transition state is the **activation energy. The** rate of a reaction reflects this activation energy: a higher activation energy corresponds to a slower reaction.

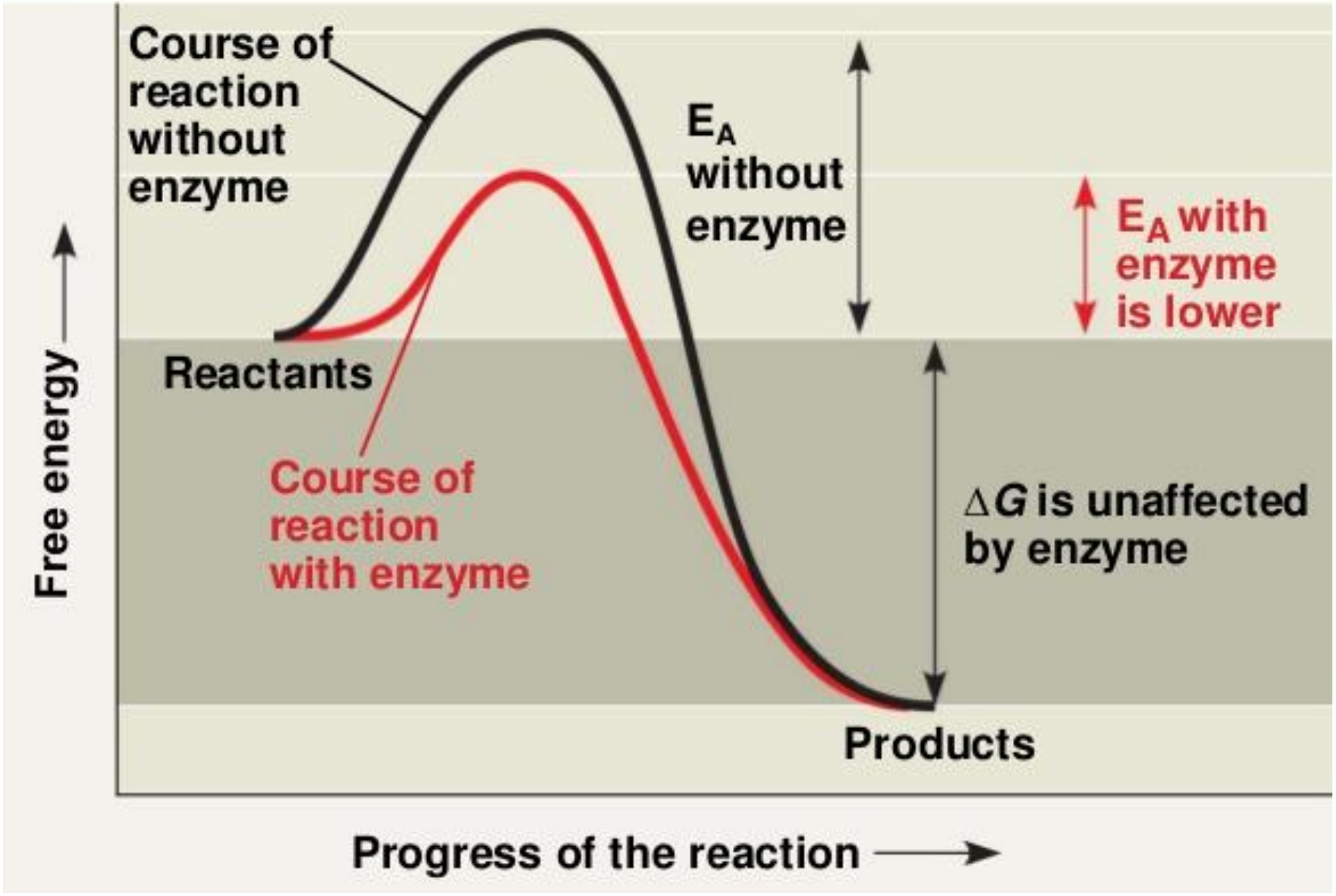


Mechanism of Action of Enzymes

- **Enzymes increase reaction rates by decreasing the Activation energy:**



Enzymes Lower a Reaction's Activation Energy

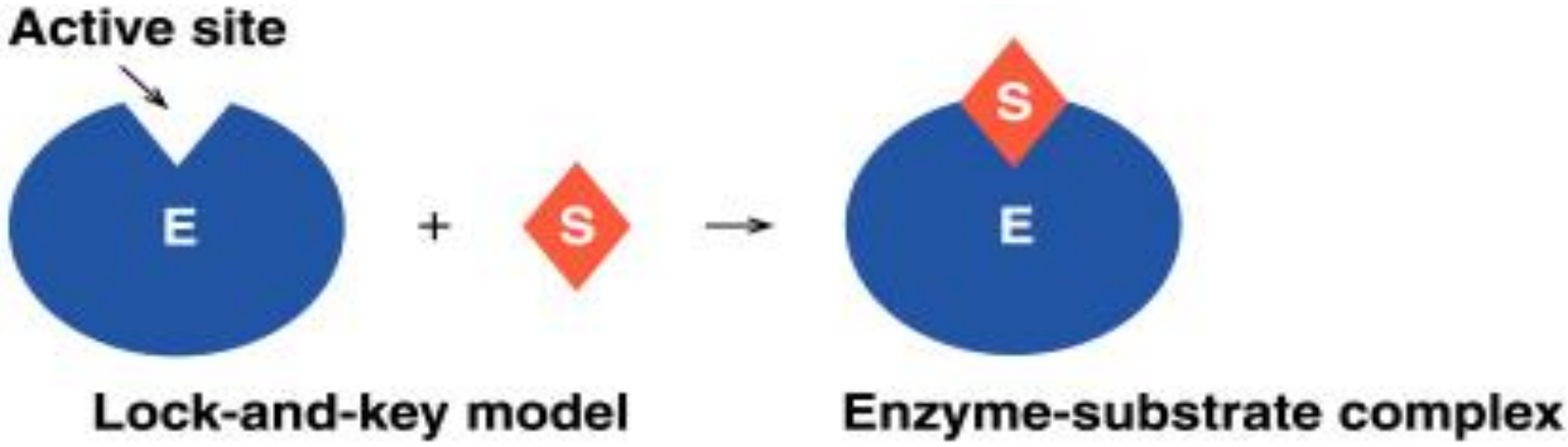

Course of reaction without enzyme
E_A without enzyme
E_A with enzyme is lower
Reactants
Course of reaction with enzyme
ΔG is unaffected by enzyme
Products
Free energy
Progress of the reaction

Enzyme-Substrate Interactions

- **Formation of Enzyme substrate complex by:**
- Lock-and-Key Model
- Induced Fit Model

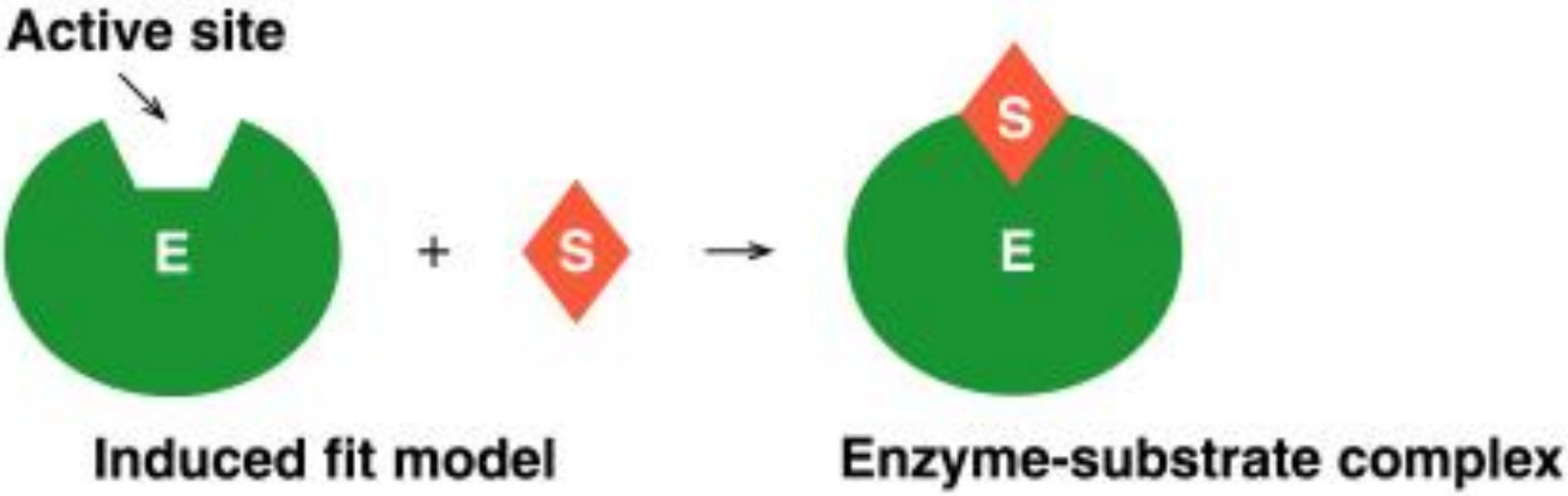
Lock-and-Key Model

- In the **lock-and-key model** of enzyme action:
  - the active site has a rigid shape
  - only substrates with the matching shape can fit
  - the substrate is a key that fits the lock of the active site
- This is an older model, however, and does not work for all enzymes



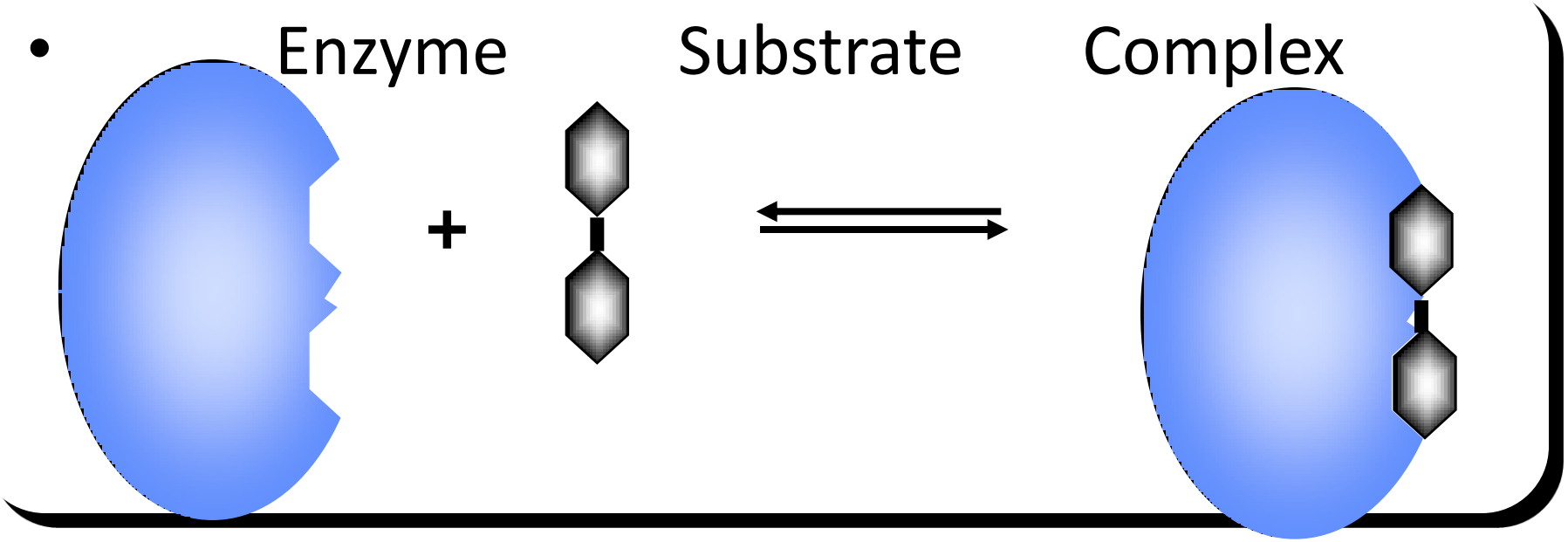
Induced Fit Model

- In the **induced-fit model** of enzyme action:
  - the active site is flexible, not rigid
  - the shapes of the enzyme, active site, and substrate adjust to maximumize the fit, which improves catalysis
  - there is a greater range of substrate specificity
- This model is more consistent with a wider range of enzymes



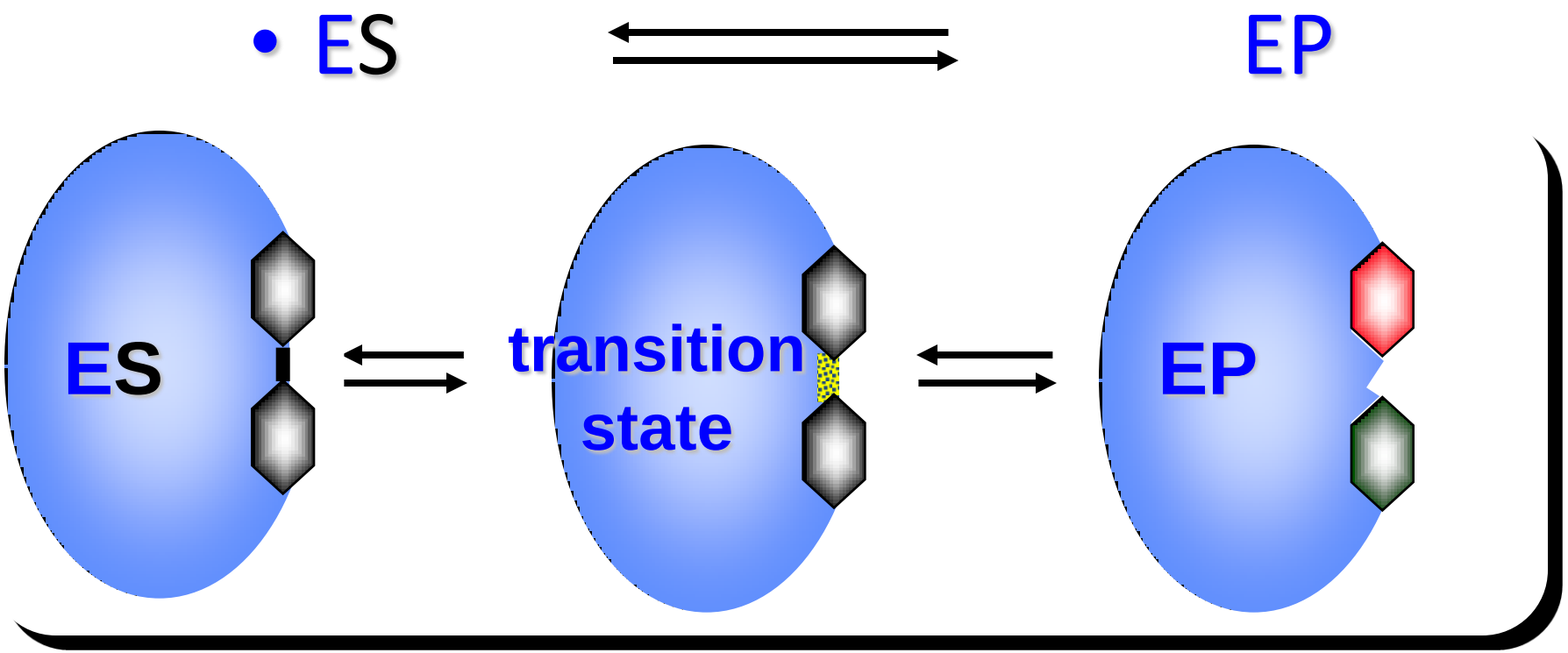
Enzyme-substrate complex

- Step 1:
- Enzyme and substrate combine to form complex
- E + S ⇌ ES
- Enzyme Substrate Complex



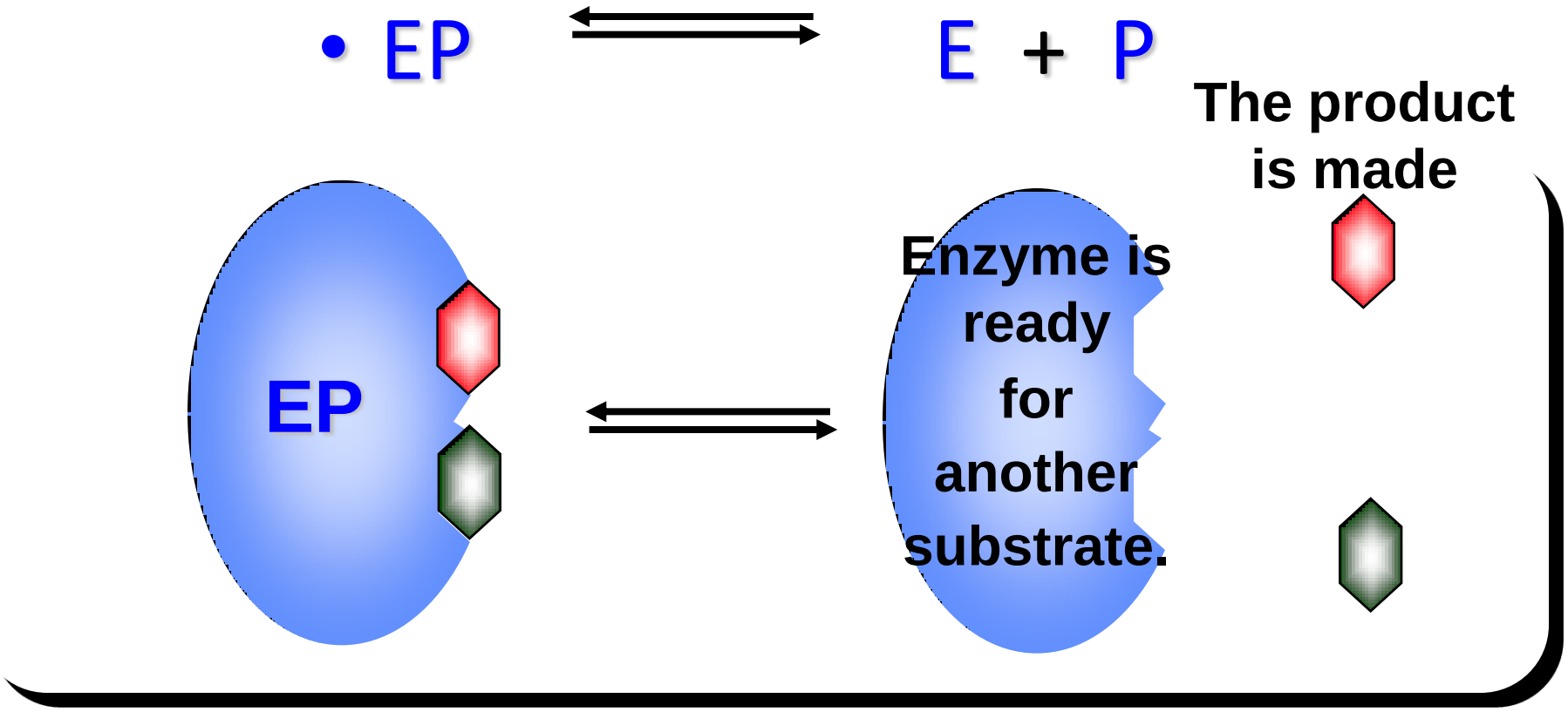
Enzyme-product complex

- Step 2:
- An enzyme-product complex is formed.



Product

- The enzyme and product separate



What Affects Enzyme Activity?

- **Three factors:**
  1. **Environmental Conditions**
  2. **Cofactors and Coenzymes**
  3. **Enzyme Inhibitors**

Michaelis-Menten equation

1. Michaelis-Menten equation describes how reaction velocity (V) varies with substrate concentration [S].

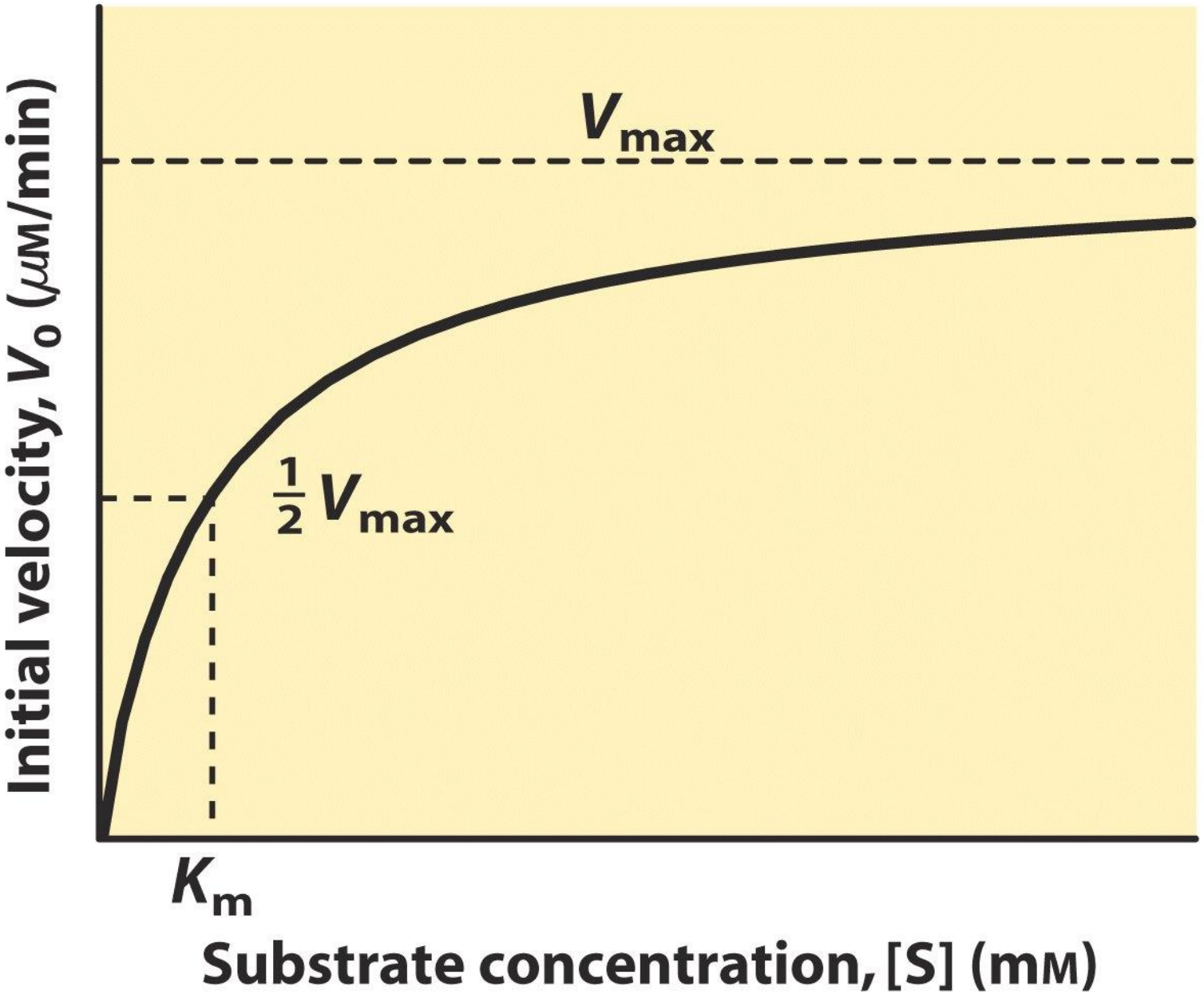
- The following equation is obtained after suitable algebraic manipulation.

$$V = V_{max} \frac{[S]}{[S] + K_M}$$

Note: V means V_0

K_m: Michaelis constant

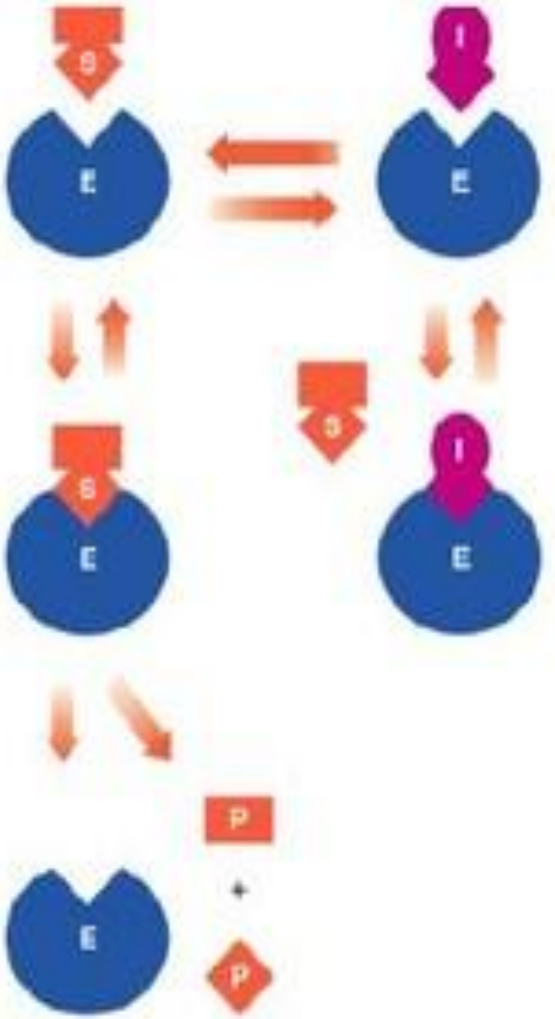
Michaelis Menten Curve



Reversible Competitive Inhibition

A **competitive inhibitor**:

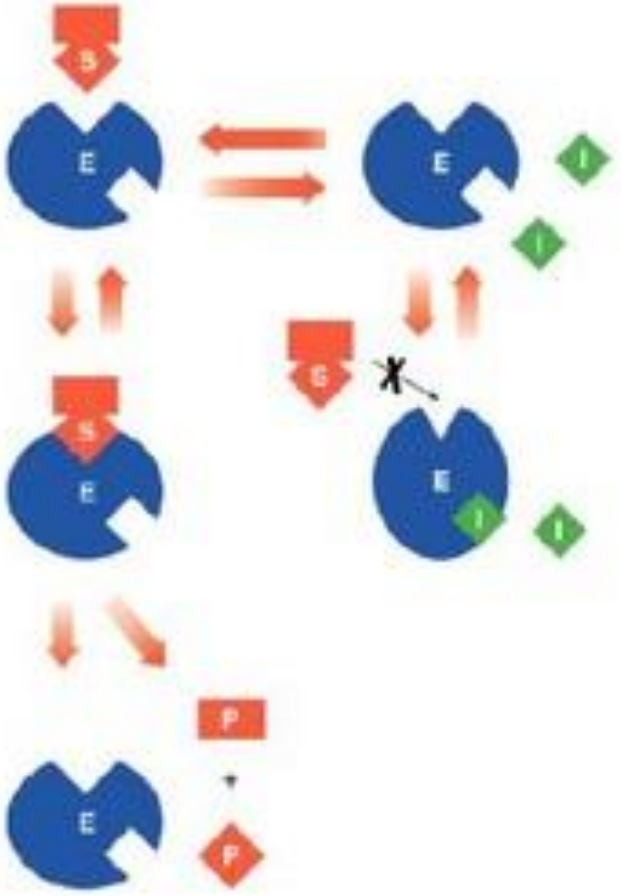
- Has a structure like the substrate.
- Competes with the substrate for the active site.
- Has its effect reversed by increasing substrate concentration.



Noncompetitive Inhibition

A **noncompetitive inhibitor**:

- Has a structure different than the substrate.
- Distorts the shape of the enzyme, which alters the shape of the active site.
- Prevents the binding of the substrate.
- Cannot have its effect reversed by adding more substrate.



Enzyme

- **The name of an enzyme in many cases end in *–ase***
- **For example, *sucrase* catalyzes the hydrolysis of sucrose**

- **The name describes the function of the enzyme**
  For example, *oxidases* catalyze oxidation reactions

- **Sometimes common names are used, particularly for the digestion enzymes such as *pepsin* and *trypsin***

- **Some names describe both the substrate and the function**
- **For example, *alcohol dehydrogenase* oxides ethanol**

Enzymes Are Classified into six functional Classes (EC number Classification) by the International Union of Biochemists (I.U.B.). on the Basis of the Types of Reactions That They Catalyze

- **EC 1. Oxidoreductases**
- **EC 2. Transferases**
- **EC 3. Hydrolases**
- **EC 4. Lyases**
- **EC 5. Isomerases**
- **EC 6. Ligases**

Principle of the international classification

Each enzyme has **classification number**

consisting of four digits:

Example, **EC:** (**2.7.1.1**) **HEXOKINASE**

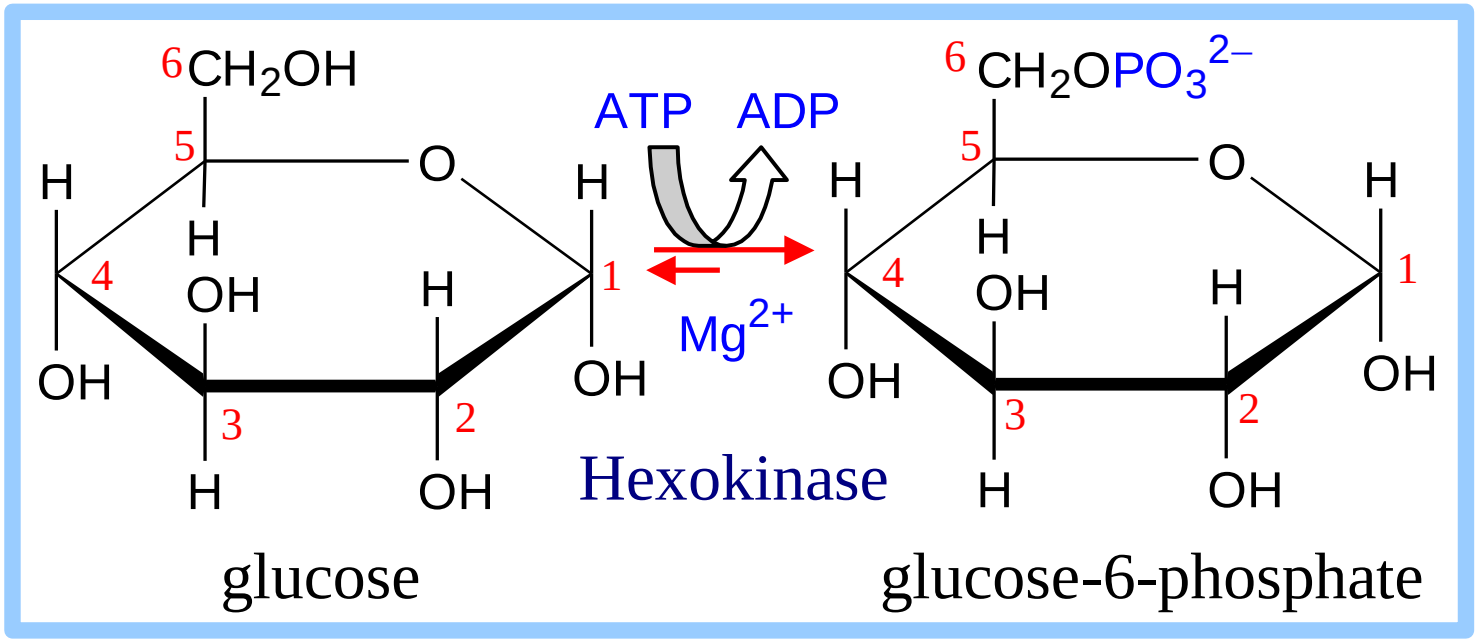
- **EC: (2.7.1.1) these components indicate the following groups of enzymes:**

- **2. IS CLASS (TRANSFERASE)**

- **7. IS SUBCLASS (TRANSFER OF PHOSPHATE)**

- **1. IS SUB-SUB CLASS (ALCOHOL IS PHOSPHATE ACCEPTOR)**
- **1. SPECIFIC NAME**

  ATP,D-HEXOSE-6-PHOSPHOTRANSFERASE (Hexokinase)

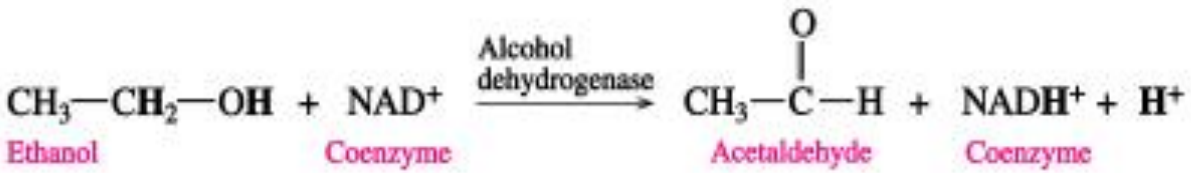


1. Hexokinase catalyzes:

Glucose + ATP → glucose-6-P + ADP

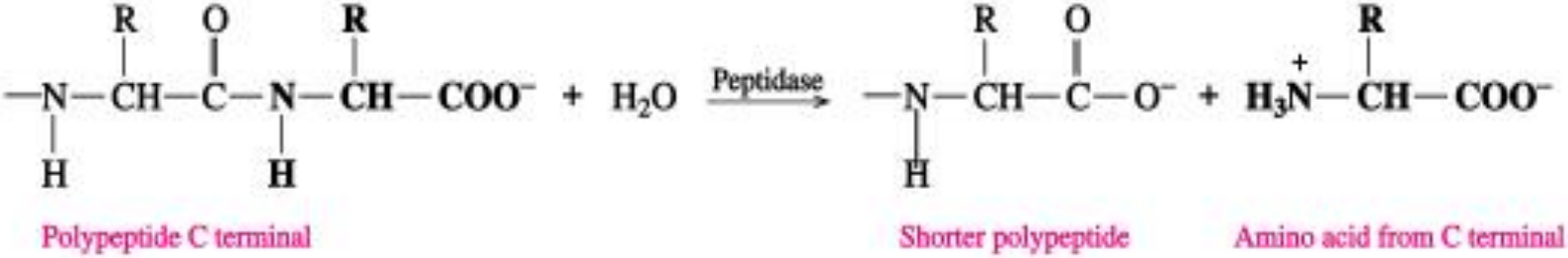
Oxidoreductases, Transferases and Hydrolases

Class	General Reactions Catalyzed	Typical Subclasses	Function
1. Oxidoreductases	Oxidation–reduction reactions	Oxidases Reductases Dehydrogenases	Oxidation Reduction Remove 2H to form double bonds



2. Transferases	Transfer of functional groups	Transaminases Kinases	Transfer amino groups Transfer phosphate groups

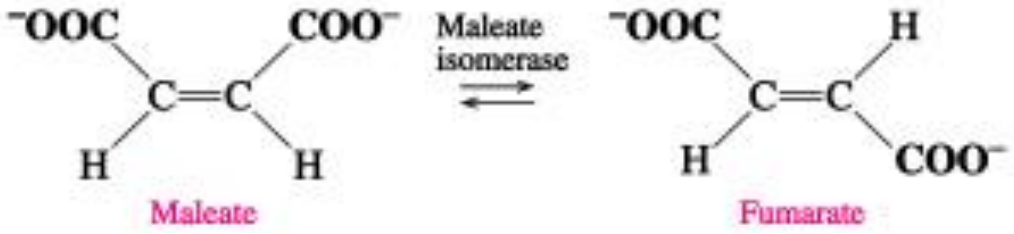
3. Hydrolases	Hydrolysis reactions	Peptidases Lipases Amylases	Hydrolyze peptide bonds Hydrolyze ester bonds in lipids Hydrolyze 1,4-glycosidic bonds in amylose



Lyases, Isomerases and Ligases

Class	General Reactions Catalyzed	Typical Subclasses	Function
4. Lyases	Addition of a group to a double bond or removal of a group from a double bond without hydrolysis or oxidation	Decarboxylases Dehydrases Deaminases	Remove CO_2 Remove H_2O Remove NH_3

5. Isomerases	Rearrangement of atoms to form isomers	Isomerases Epimerases	Convert cis and trans Convert D and L isomers



6. Ligases	Bonding of molecules using ATP energy	Synthetases Carboxylases	Combine molecules Add CO_2