Lecture 4

Enzymes

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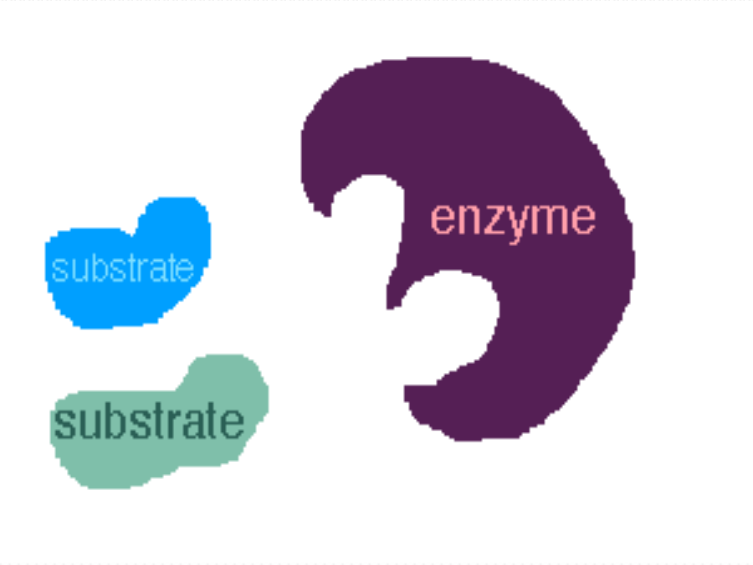
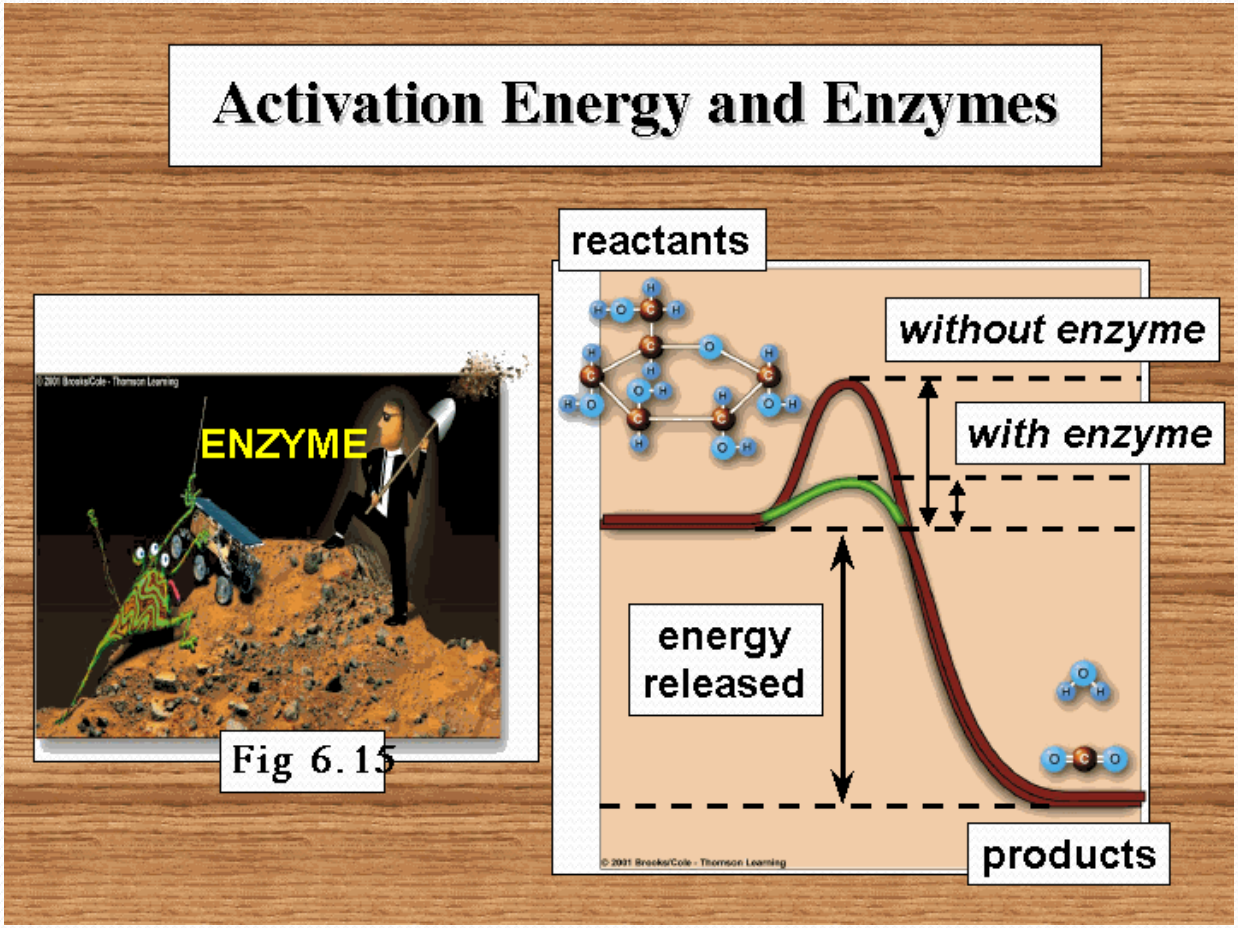
Defination



- **Enzymes** are proteins that increase the rate of reaction by lowering the energy of activation.
- Substrate that increase the velocity or rate of a chemical reaction without itself undergoing any change in the overall process
- Catalyst - Speeds up chemical reactions in living organisms by decreasing the energy needed to start the reaction (activation energy)

Notice that without the enzyme it takes a lot more energy for the reaction to occur. By lowering the activation energy you speed up the reaction.



Importance

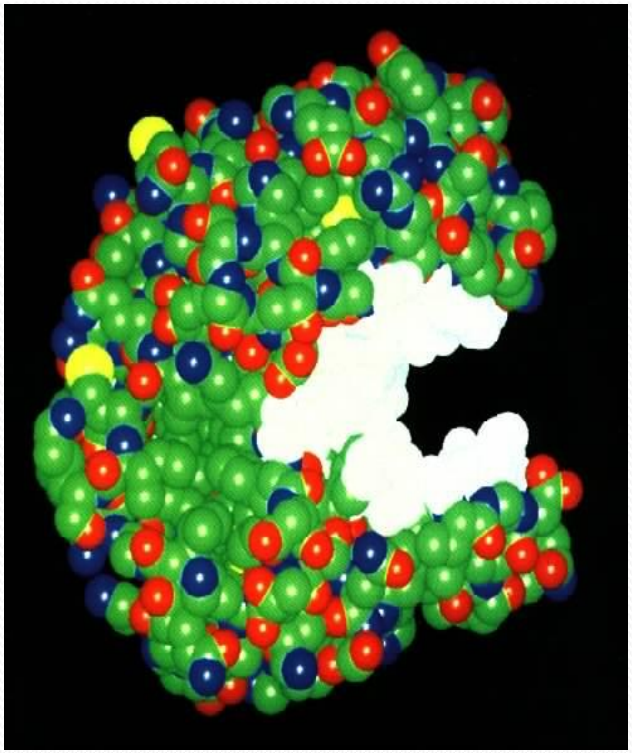
- Enzymes play an important role in Metabolism, Diagnosis, and Therapeutics.
- All biochemical reactions are enzyme catalyzed in the living organism.
- Level of enzyme in blood are of diagnostic importance e.g. it is a good indicator in disease such as myocardial infarction.
- Enzyme can be used therapeutically such as digestive enzymes.

Enzymes

- Enzymes are proteins that act as biological catalysts.
- Cells use enzymes to speed up chemical reactions that take place in cells.
- Enzyme speed up reactions by lowering the activation energies.
- Because a particular enzyme catalyzes only one reaction, there are thousands of different enzymes in a cell catalyzing thousands of different chemical reactions

Enzymes

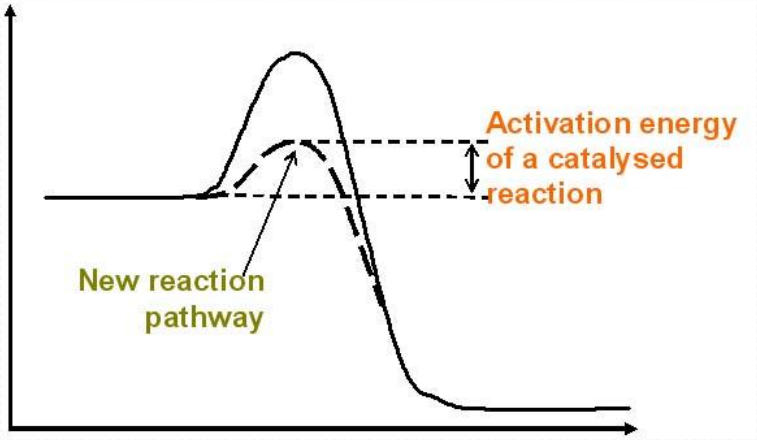
- Most enzymes are Proteins (tertiary and quaternary structures)
- Act as Catalyst to accelerates a reaction
- Not permanently changed in the process



Making reactions go faster

- Increasing the temperature make molecules move faster
- Biological systems are very sensitive to temperature changes.
- Enzymes can increase the rate of reactions without increasing the temperature.
- They do this by lowering the activation energy.
- They create **a new reaction pathway** "a short cut"

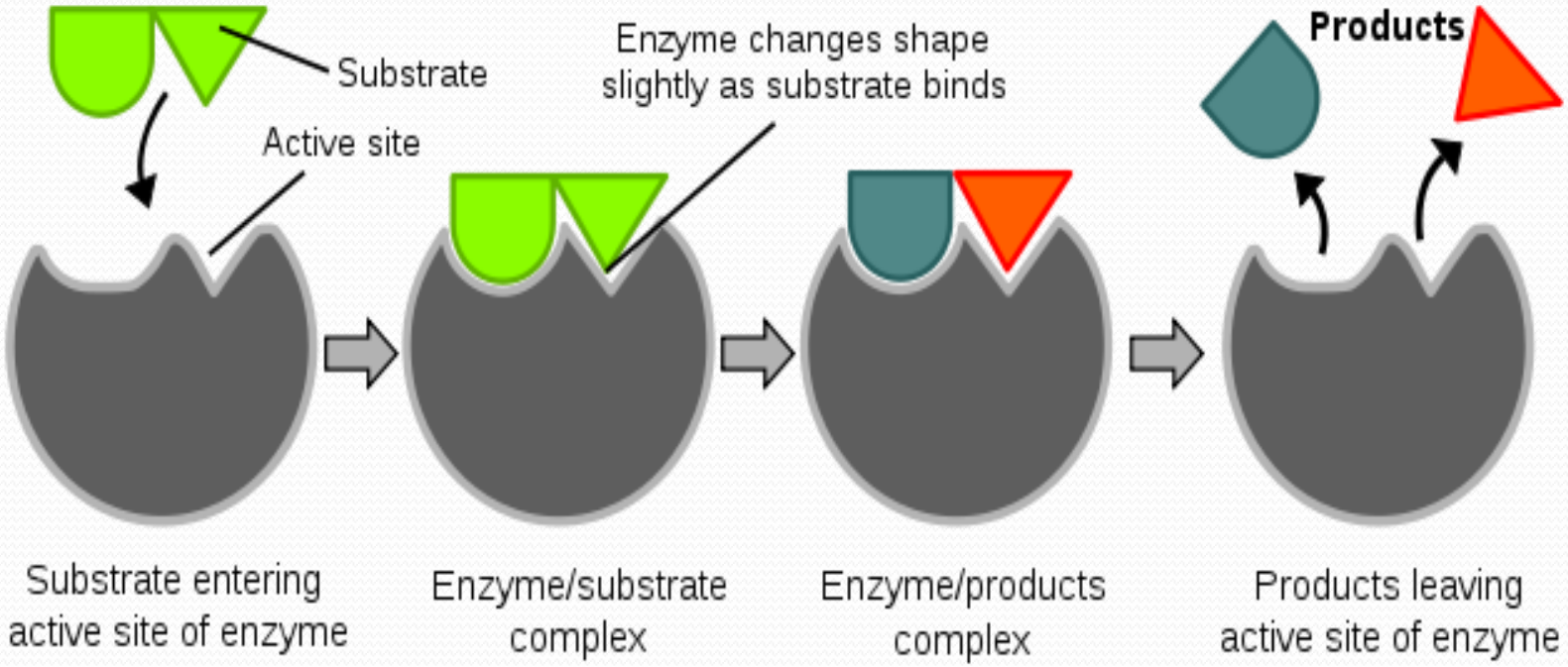
An enzyme controlled pathway

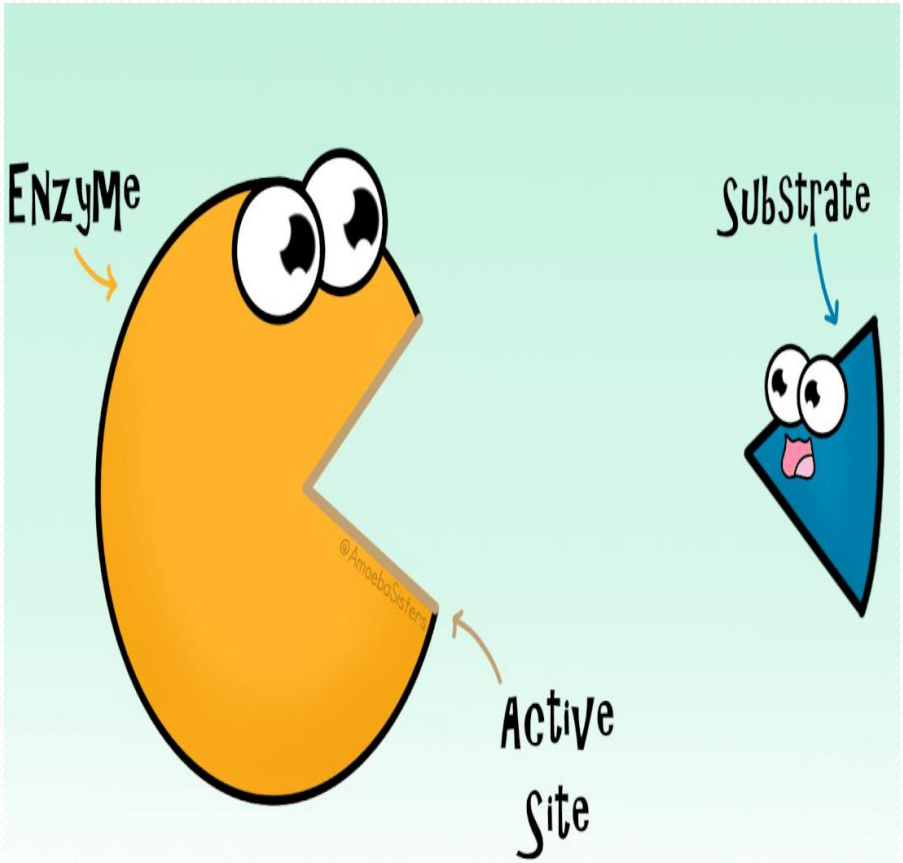


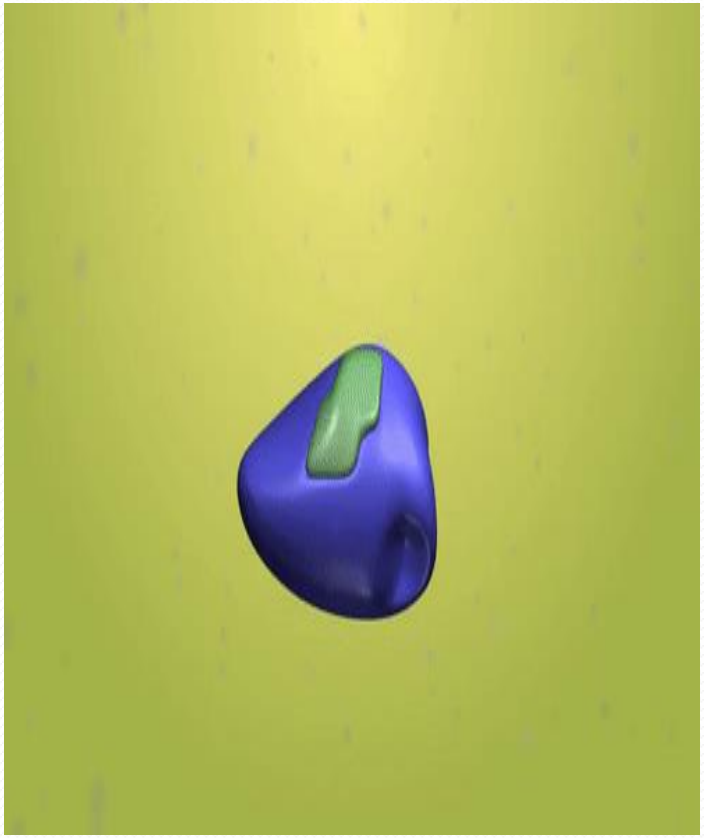
- Enzyme controlled reactions proceed 108 to 1011 times faster than corresponding non-enzymic reactions.

ACTIVE SITE

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

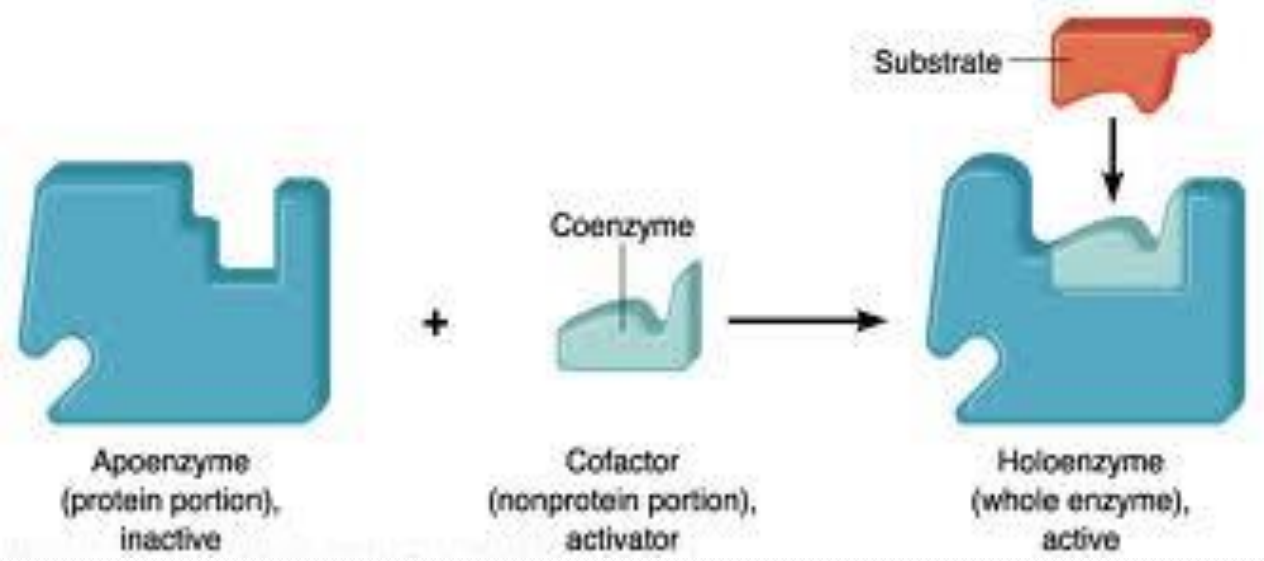



Enzyme
Substrate
Active
Site



APOENZYME and HOLOENZYME

- The enzyme without its non protein moiety is termed as apoenzyme and it is inactive.
- Holoenzyme is an active enzyme with its non protein component.



Important Terms to Understand Biochemical Nature And Activity of Enzymes

- **Cofactor:**
  - **A cofactor is a non-protein chemical compound that is bound (either tightly or loosely) to an enzyme and is required for catalysis.**
  - **Types of Cofactors:**
    - **Coenzymes.**
    - **Prosthetic groups.**

Types of Cofactors

- **Coenzyme:**
  The non-protein component, loosely bound to apoenzyme by non-covalent bond.
- **Examples : vitamins or compound derived from vitamins.**
- **Prosthetic group**
  The non-protein component, tightly bound to the apoenzyme by covalent bonds is called a Prosthetic group.

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

Important Terms to Understand Biochemical Nature And Activity of Enzymes

Activation energy or Energy of Activation:

- **All chemical reactions require some amount of energy to get them started.**

OR

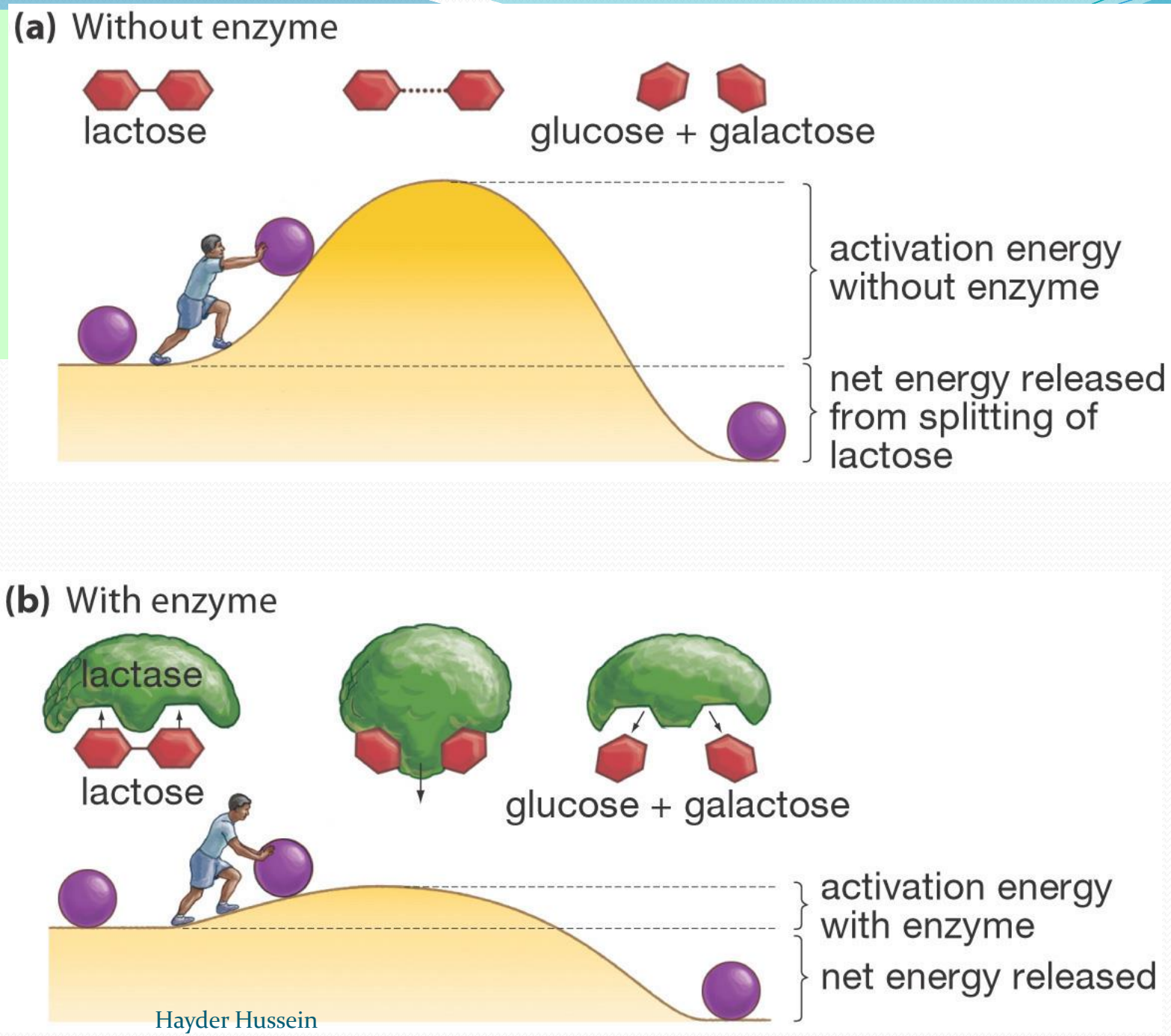
- **It is First push to start reaction.**

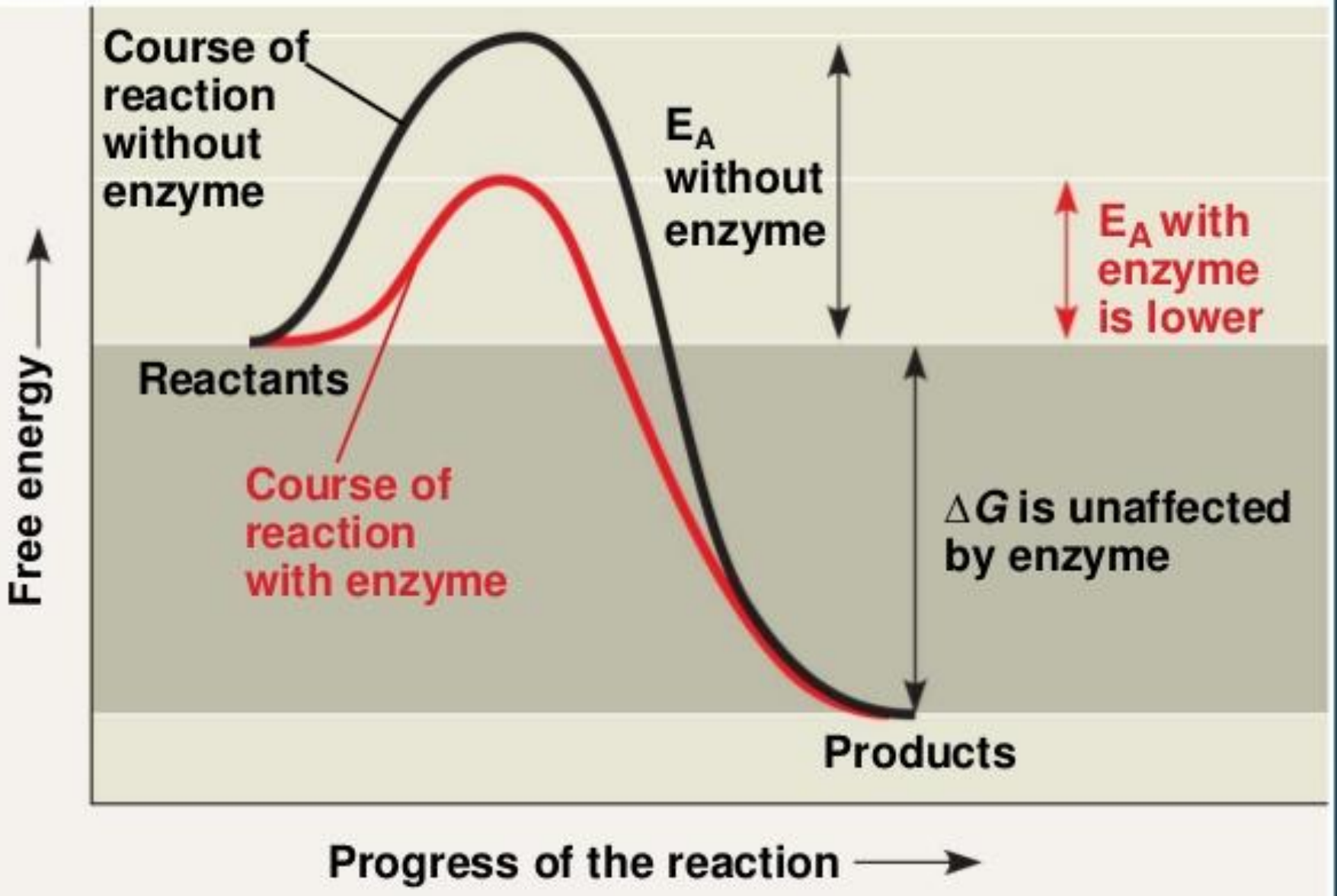
This energy is called activation energy.

Mechanism of Action of Enzymes

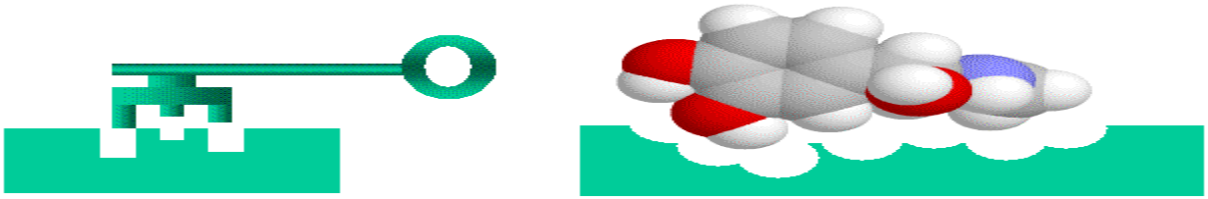
- **Enzymes increase reaction rates by decreasing the Activation energy:**
- **Enzyme-Substrate Interactions:**
  - **Formation of Enzyme substrate complex by:**
  - Lock-and-Key Model
  - Induced Fit Model

Enzymes Lower a Reaction's Activation Energy

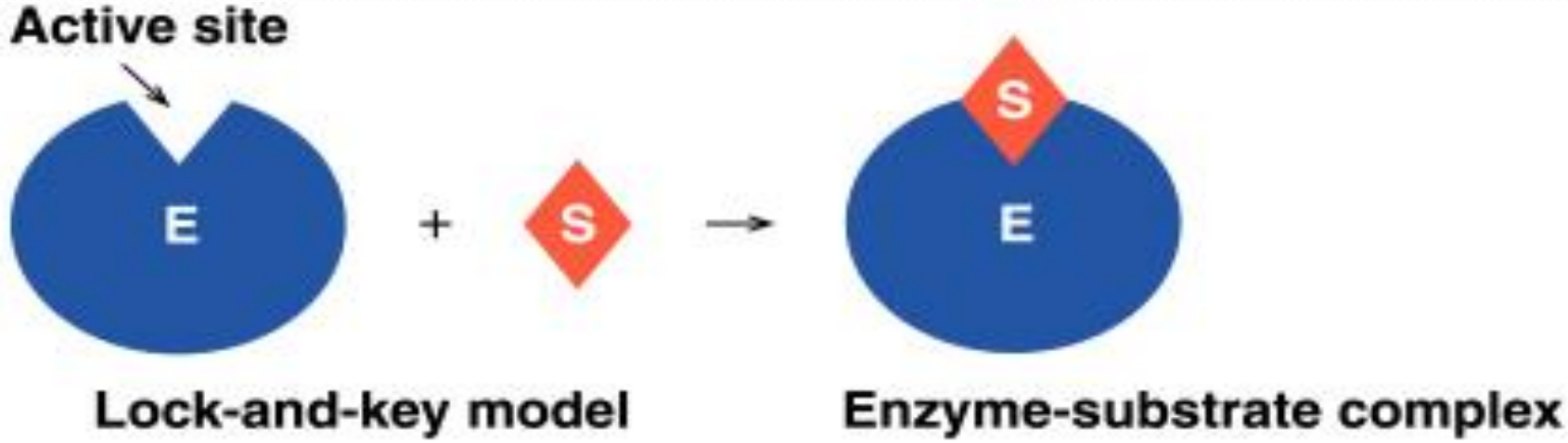



Course of reaction without enzyme
E_A without enzyme
E_A with enzyme is lower
Reactants
Course of reaction with enzyme
ΔG is unaffected by enzyme
Products
Free energy
Progress of the reaction

Lock-and-Key Model

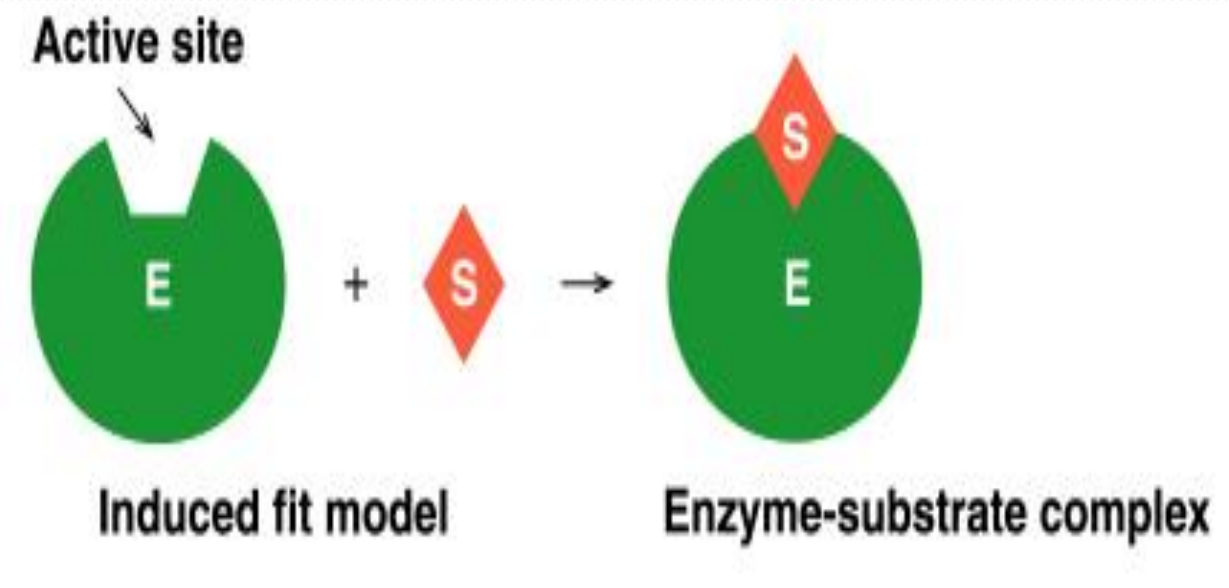


- In the **lock-and-key model** of enzyme action:
  - the active site has a rigid shape
  - only substrates with the matching shape can fit
  - the substrate is a key that fits the lock of the active site
- This is an older model, however, and does not work for all enzymes



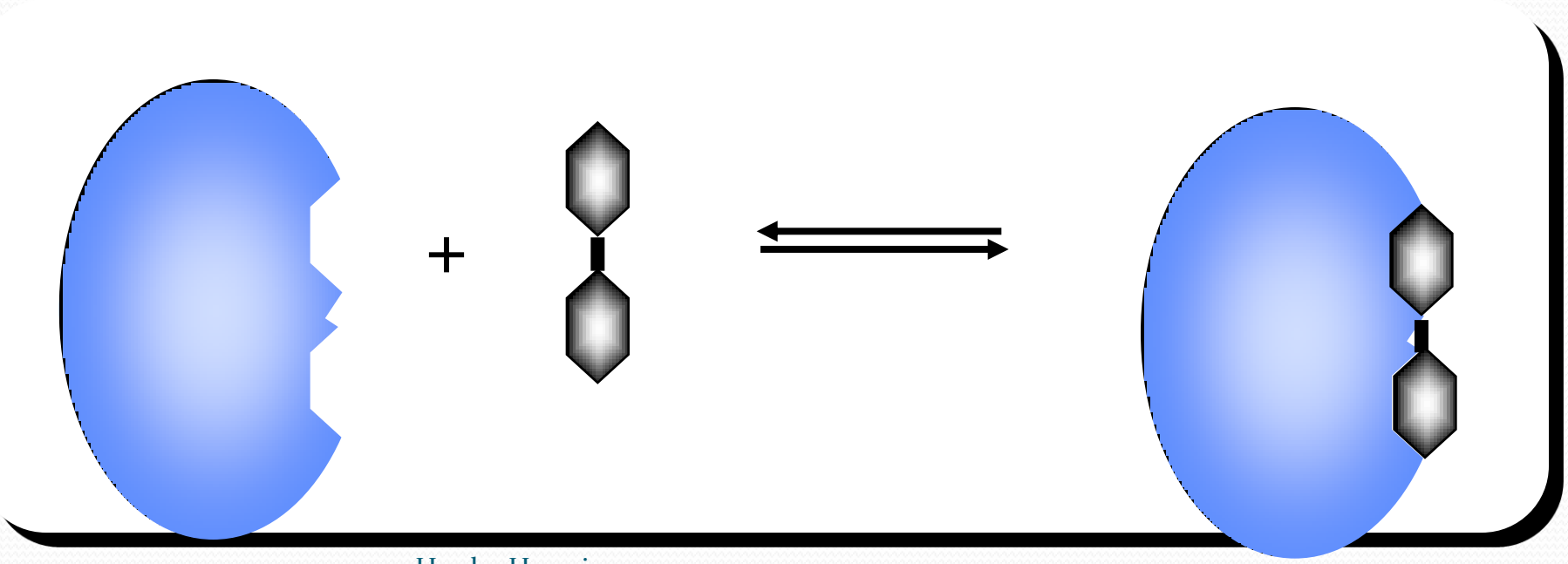
Induced Fit Model

- 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

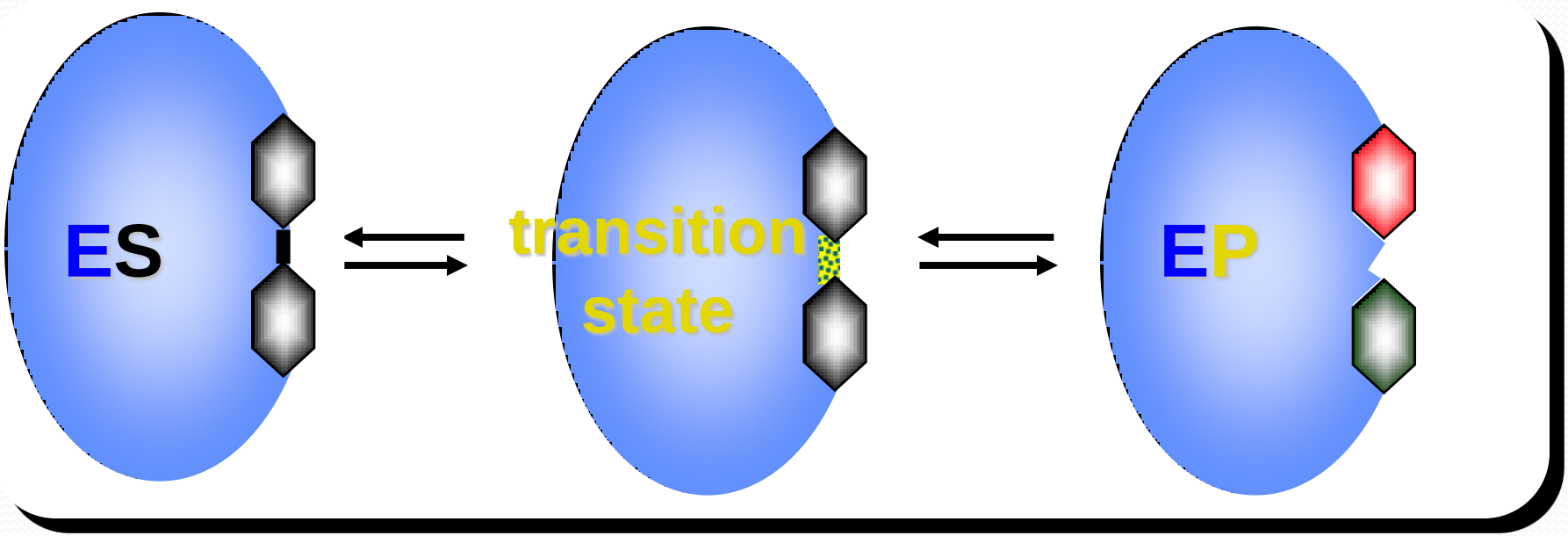
- Step 1:
- Enzyme and substrate combine to form complex
- E + S ⇌ ES
- Enzyme Substrate Complex



Enzyme-product complex

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

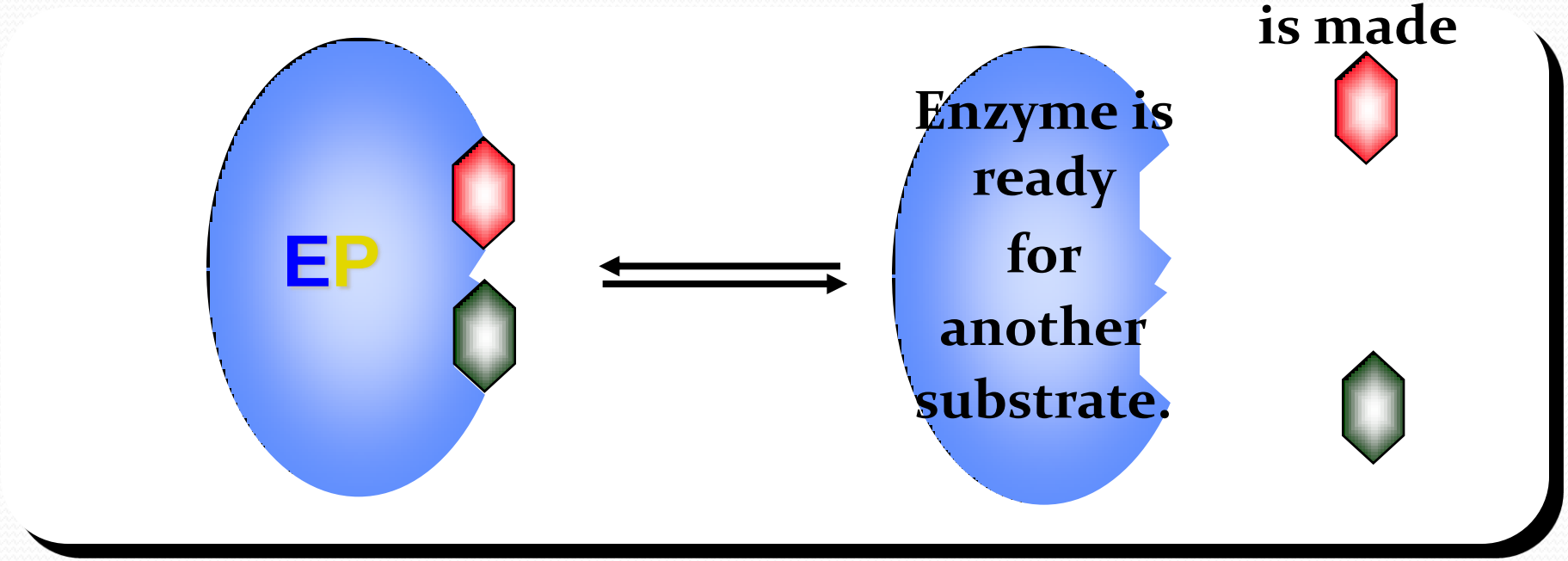
- ES ⇌ EP

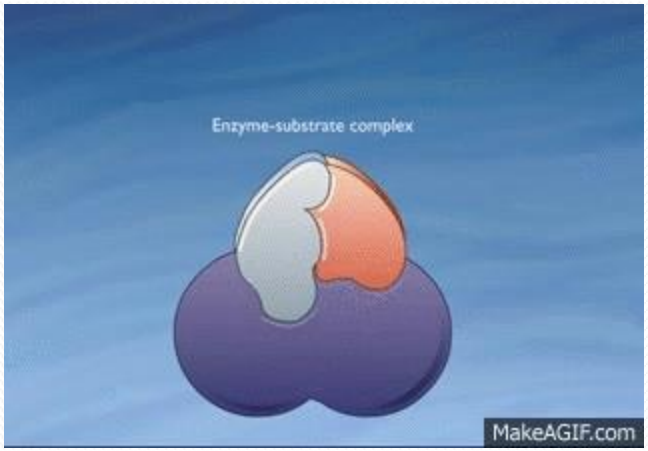


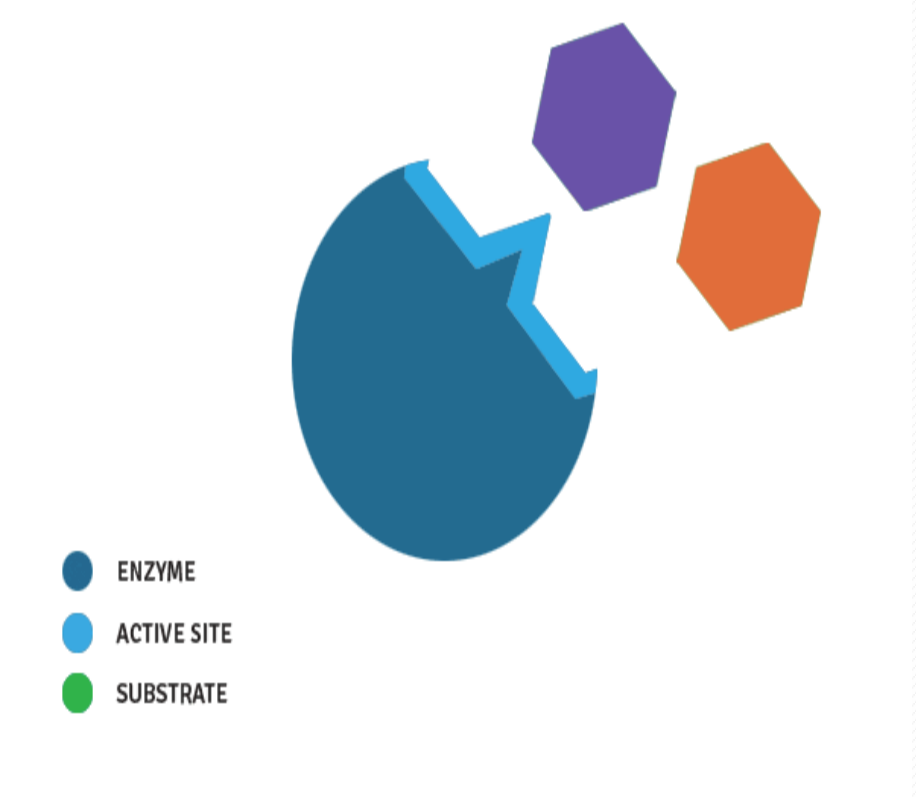
Product

- The enzyme and product separate

- EP ⇌ E + P




Enzyme-substrate complex
MakeAGIF.com


ENZYME
ACTIVE SITE
SUBSTRATE

Naming Enzymes

- **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**

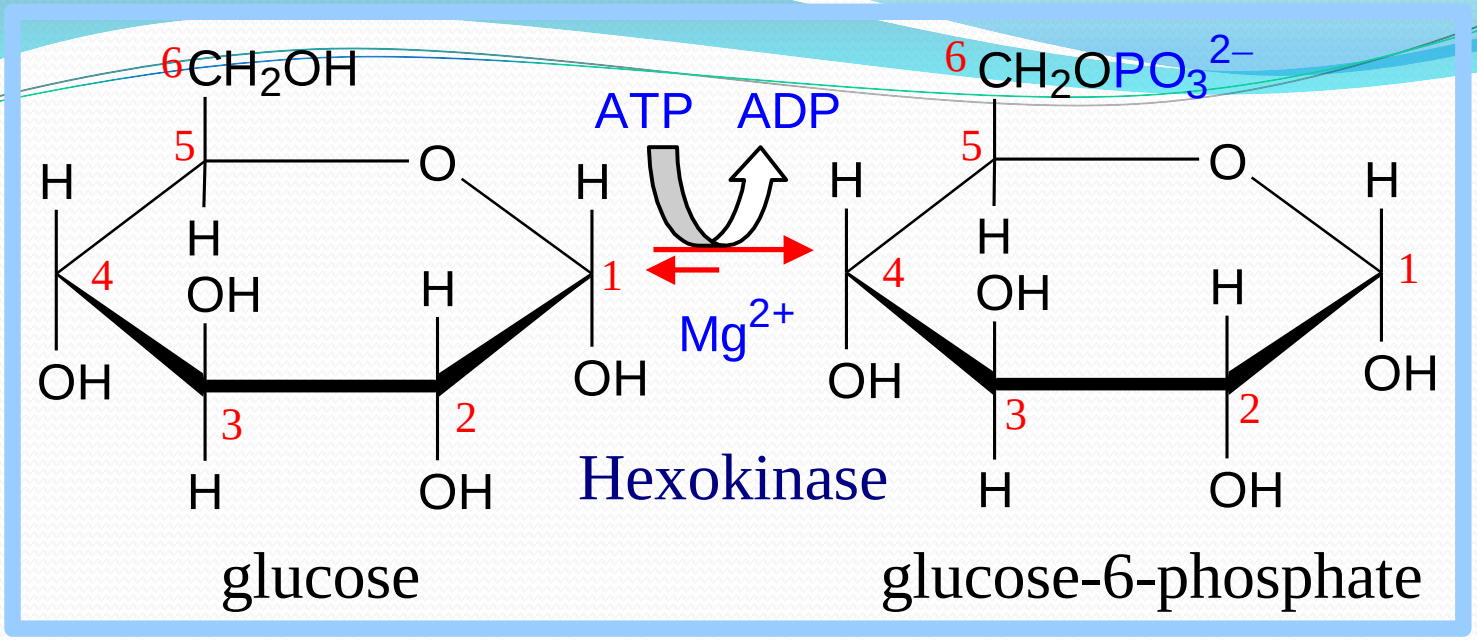
- **EC: (2.7.1.1) these components indicate the following groups of enzymes:**

- **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

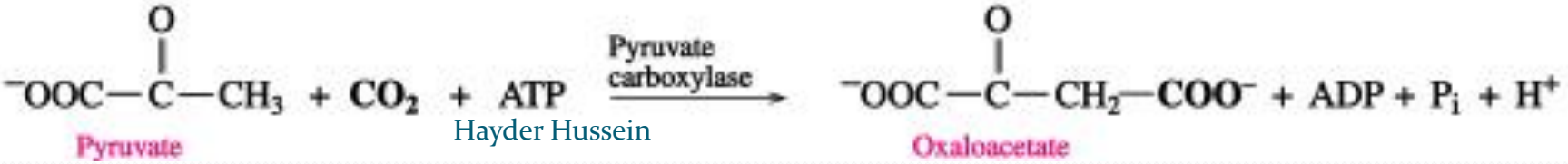
Oxidoreductases, Transferases and Hydrolases

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



Factors affecting Enzymes

- Concentration of enzyme
- substrate concentration
- pH
- temperature
- inhibitors

Hayder Hussein

Concentration of enzyme

As the concentration of the enzyme is

increased, the velocity of the reaction

proportionately increases In fact, this property of enzyme is made use in determining the serum enzymes for the diagnosis of diseases. By using a known volume of serum, and keeping all the other factors (substrate, pH, temperature etc.) at the optimum level,

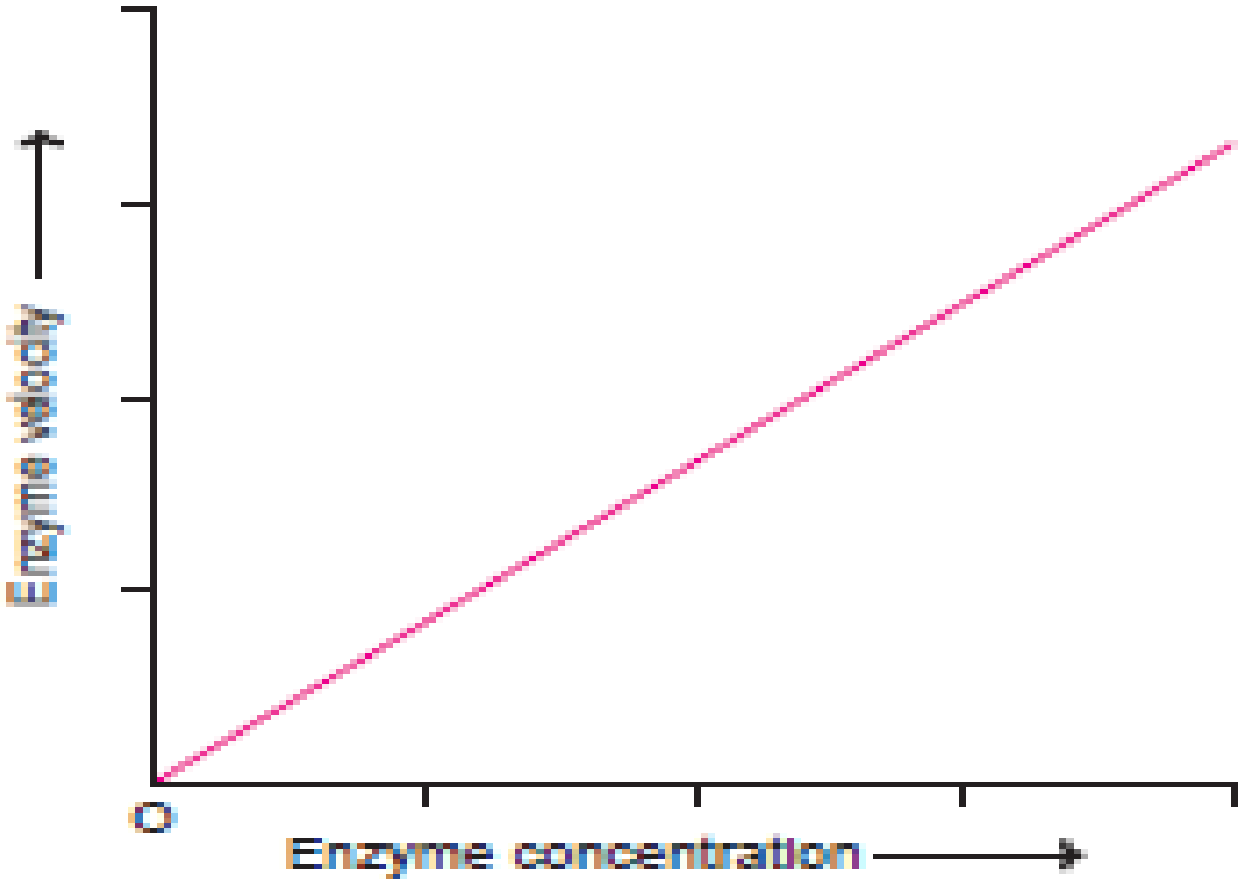
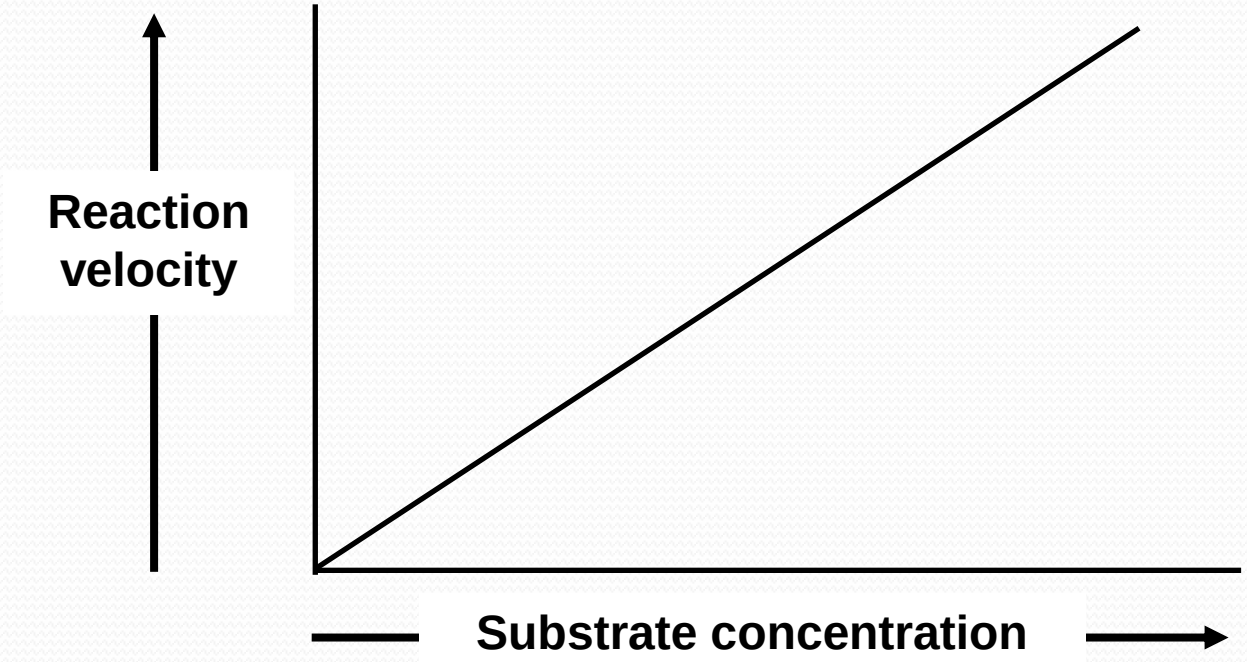


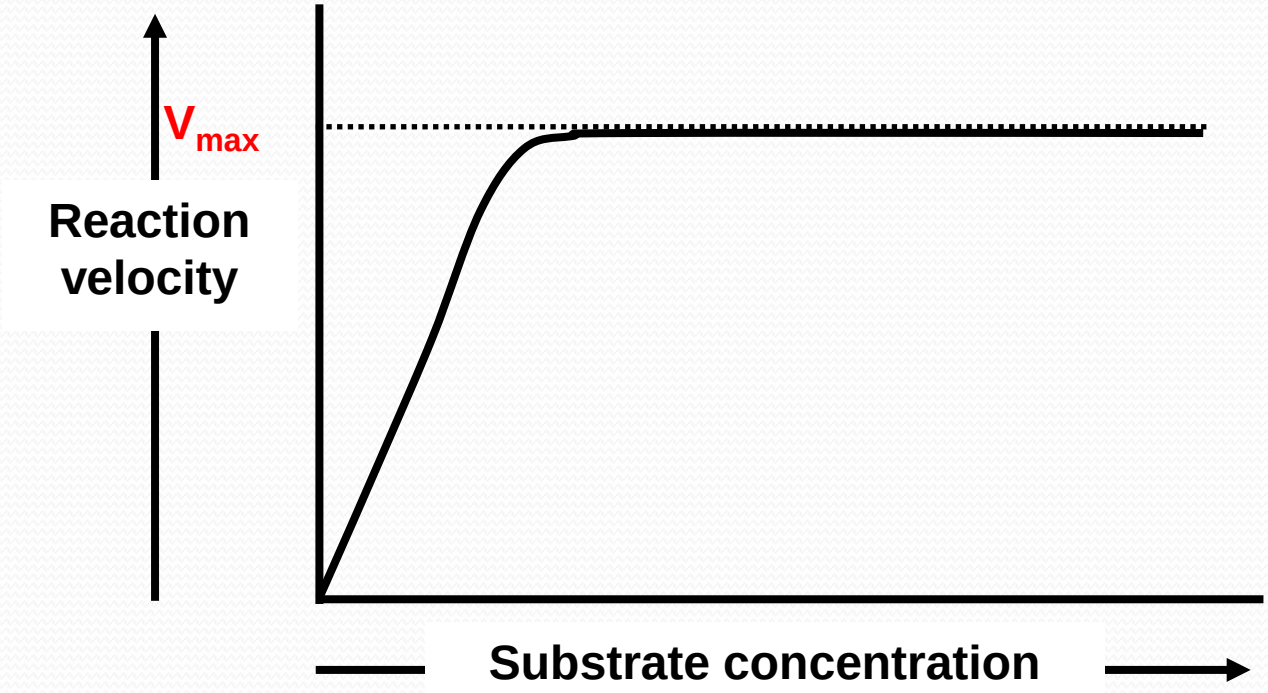
Fig. 6.1 : Effect of enzyme concentration on enzyme velocity.

1-Substrate concentration: Non-enzymic reactions



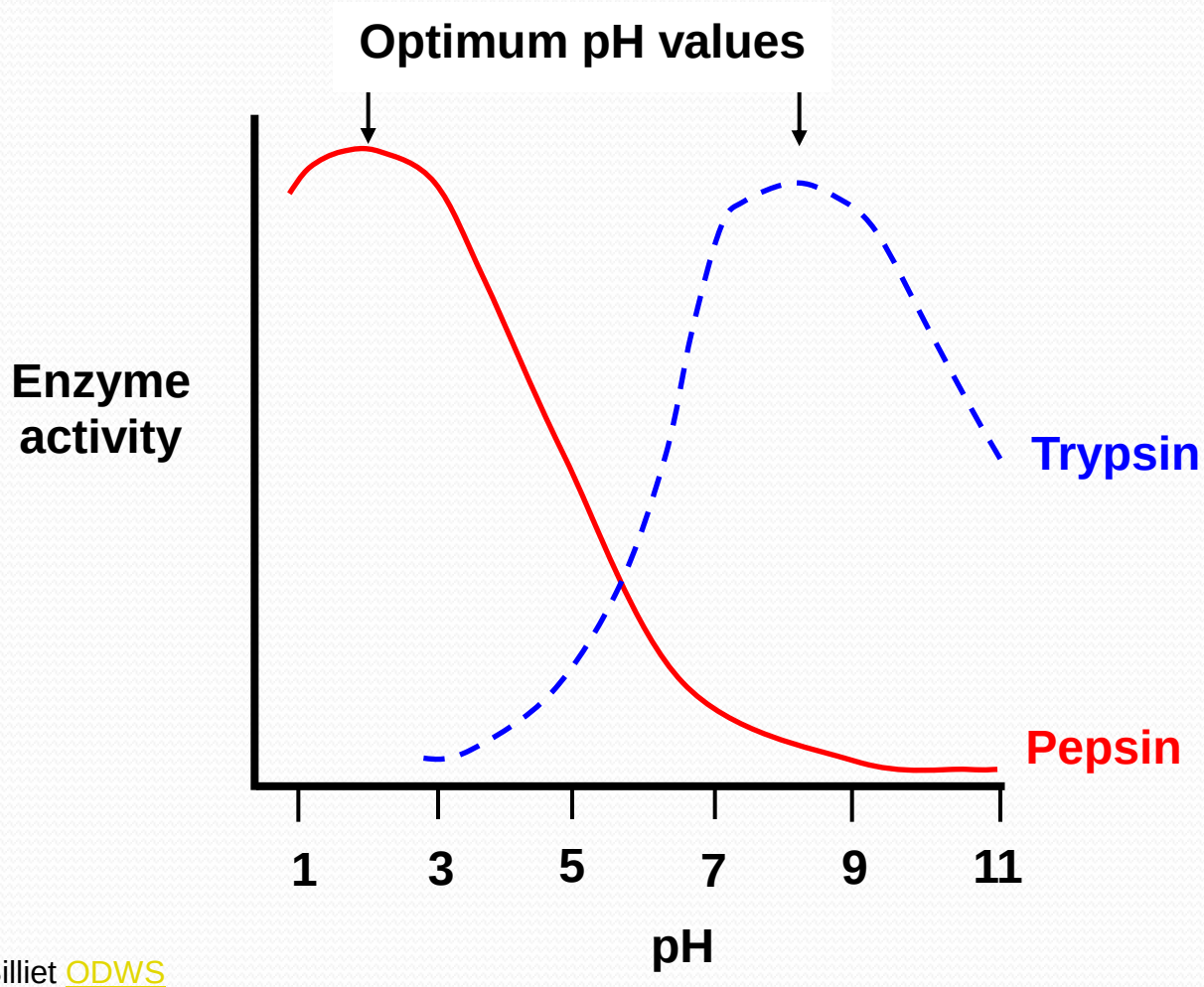
- The increase in velocity is proportional to the substrate concentration

1-Substrate concentration: Enzymic reactions



- Faster reaction but it reaches a saturation point when all the enzyme molecules are occupied.
- If you alter the concentration of the **enzyme** then $\mathbf{V_{max}}$ will change too.

2-The effect of pH



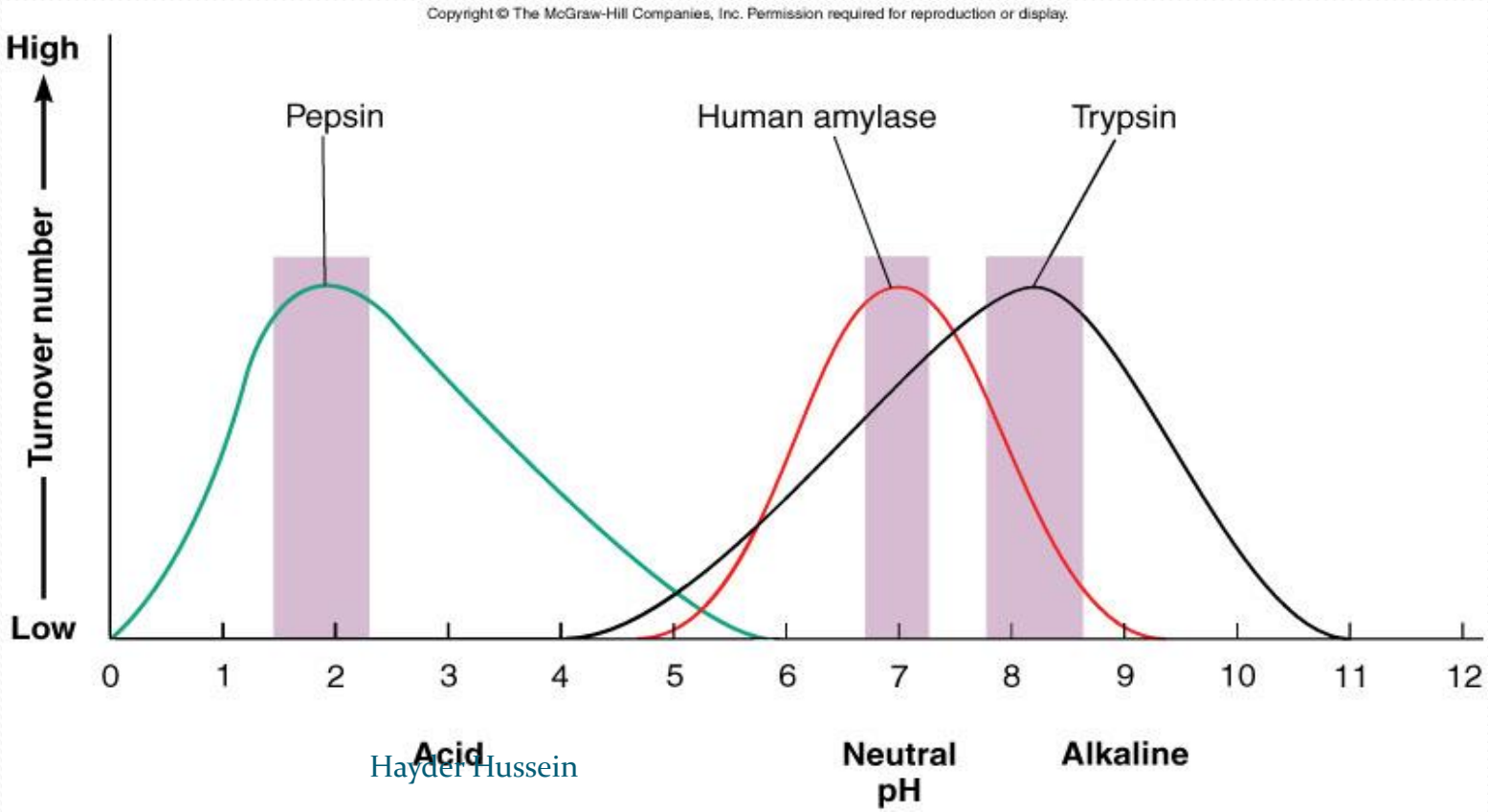
2-The effect of pH

- Extreme pH levels will produce **denaturation**
- The structure of the enzyme is changed
- The active site is distorted and the substrate molecules will no longer fit in it
- At pH values slightly different from the enzyme's optimum value, small changes in the charges of the enzyme and it's substrate molecules will occur
- This change in ionisation will affect the binding of the substrate with the active site.

The effect of pH

Most enzymes have an optimum pH of around 7 (neutral)

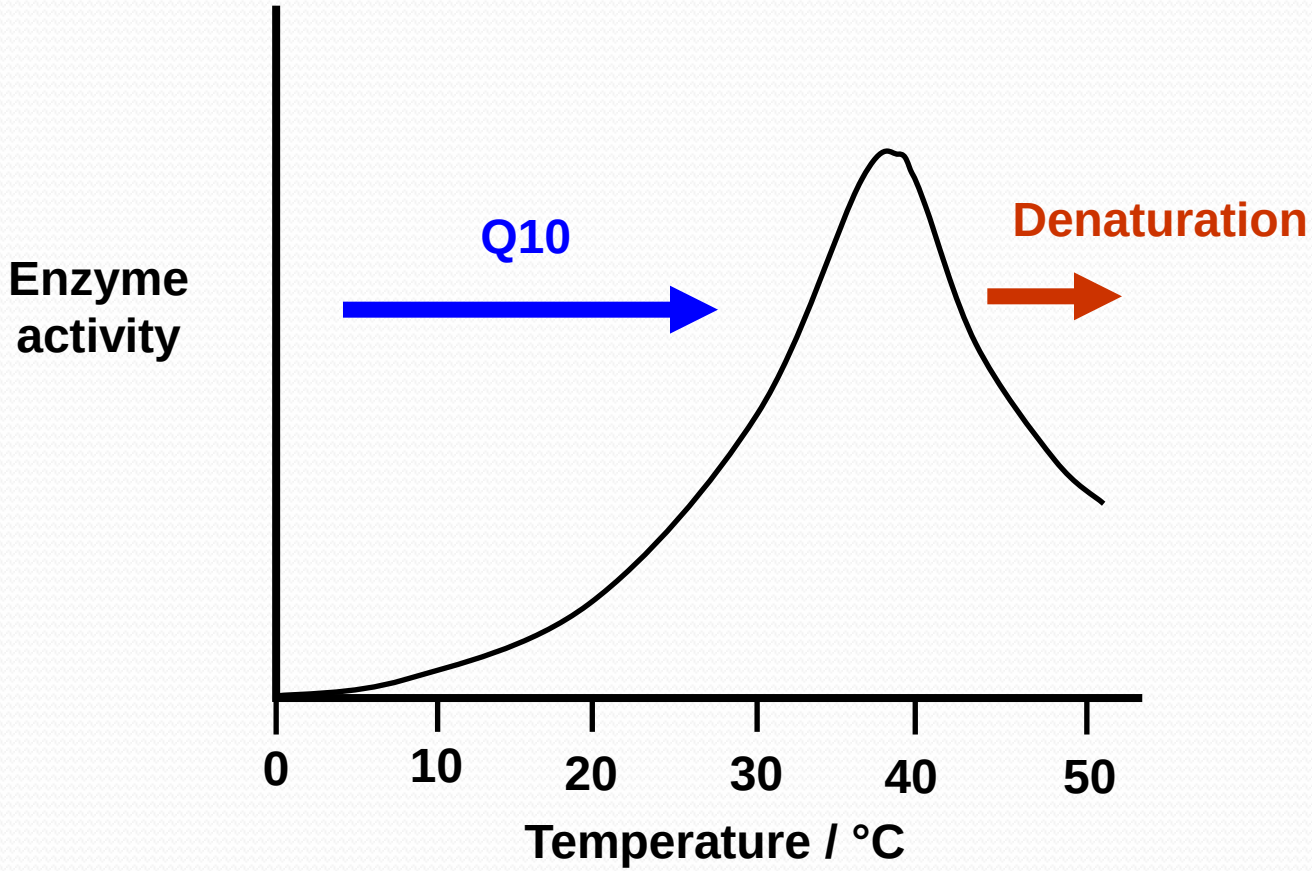
- However, some prefer acidic or basic conditions



Affects of temperature on an enzyme

- If temp to high or to low the enzyme will not fit. No reaction will occur.
- For most enzymes the optimum temperature is about 40
- Many are a lot lower, cold water fish will die at 40 because their enzymes denature
- A few bacteria have enzymes that can withstand very high temperatures up to 100°C
- Most enzymes however are fully denatured at 70°C

3-The effect of temperature



3-The effect of temperature

Temperature coefficient or **Q10** is defined as increase in enzyme velocity when th temperature is increased by 10°C. For a majority of enzymes, Q10 is 2 between 0°C and 40°C. Increase in temperature results in higher activation energy of the molecules and more molecular (enzyme and substrate) collision and interaction for the reaction to proceed faster.

4-Inhibitors

- Inhibitors are chemicals that reduce the rate of enzymic reactions.
- The are usually specific and they work at low concentrations.
- They block the enzyme but they do not usually destroy it.
- Many drugs and poisons are inhibitors of enzymes in the nervous system.

4-The effect of enzyme inhibition

- **Irreversible inhibitors:** Combine with the functional groups of the amino acids in the active site, irreversibly.

Examples: nerve gases and pesticides, containing organophosphorus, combine with serine residues in the enzyme acetylcholine esterase.

The effect of enzyme inhibition

- **Reversible inhibitors:** These can be washed out of the solution of enzyme by dialysis.

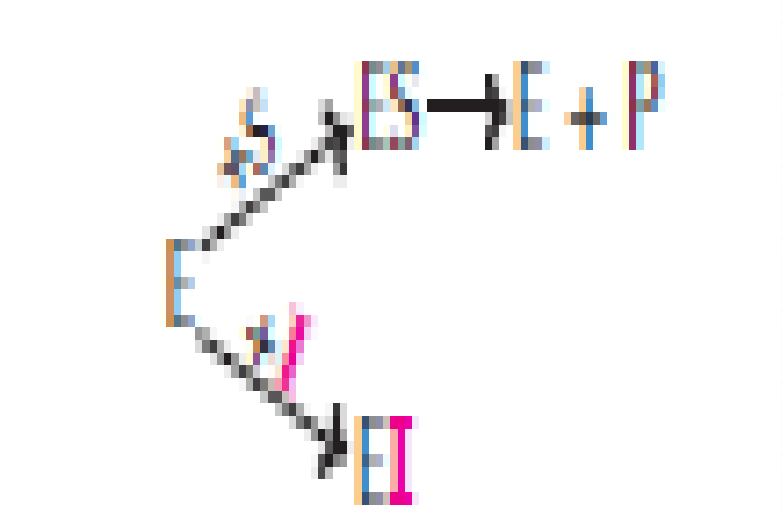
There are two categories.

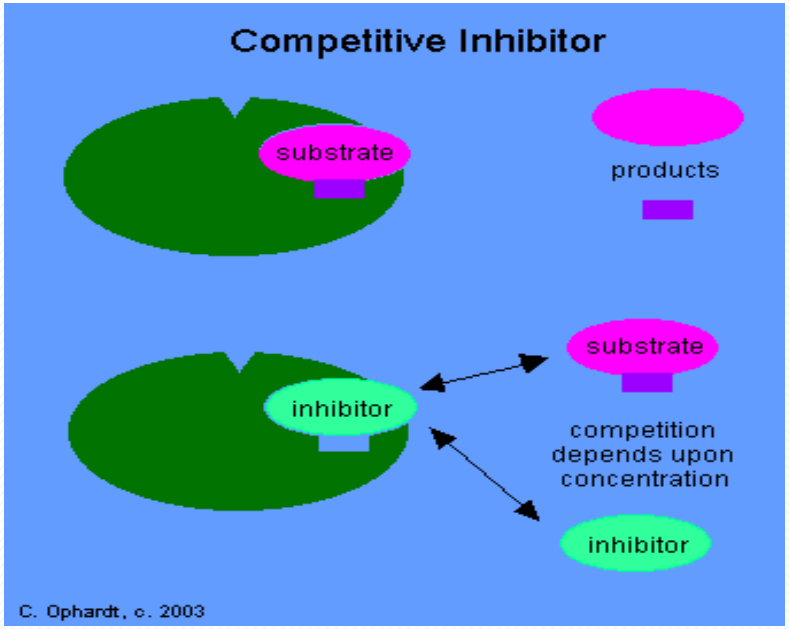
The effect of enzyme inhibition

1. **Competitive:** These compete with the substrate molecules for the active site.

The inhibitor's action is proportional to its concentration.

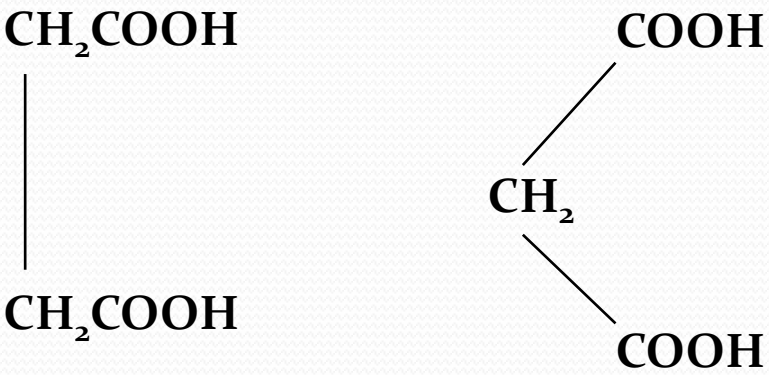
Resembles the substrate's structure closely.



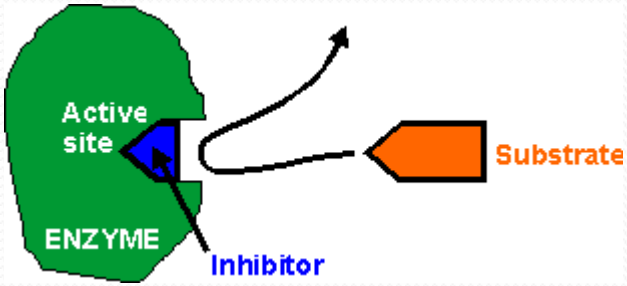


The effect of enzyme inhibition

Succinate → *Succinate dehydrogenase* → Fumarate + $2H^+ + 2e^-$



Malonate



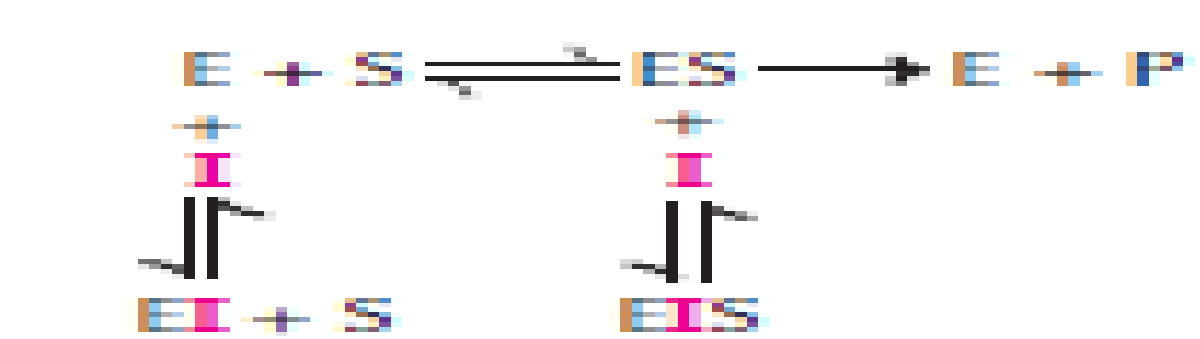
The effect of enzyme inhibition

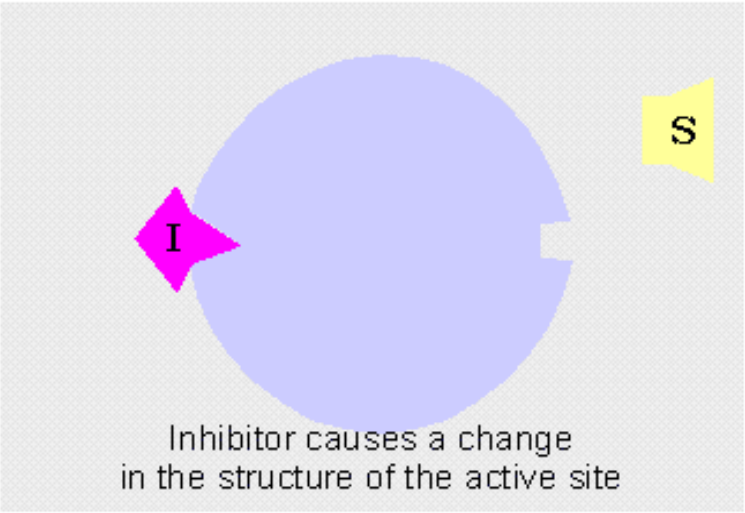
2. **Non-competitive:** These are not influenced by the concentration of the substrate. It inhibits by binding irreversibly to the enzyme but **not at the active site.**

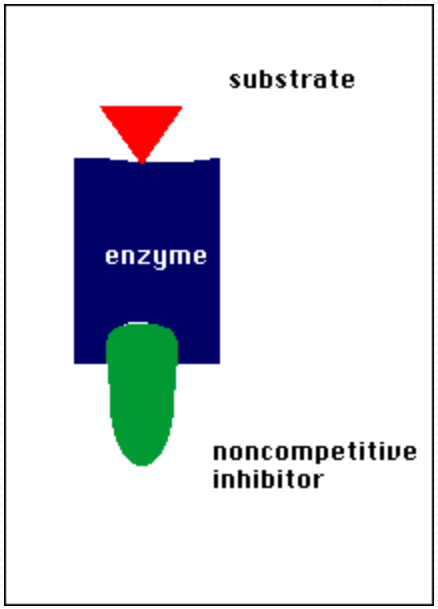
Examples

- Cyanide combines with the Iron in the enzymes cytochrome oxidase.
- Heavy metals, **Ag** or **Hg**, combine with **–SH** groups.

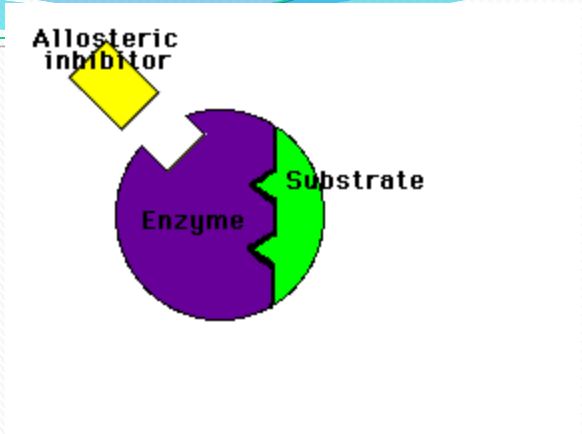
These can be removed by using a chelating agent such as EDTA.




S
I
Inhibitor causes a change
in the structure of the active site


substrate
enzyme
noncompetitive
inhibitor

3-Allosteric effectors :



- **3-Allosteric effectors :**
- Certain substances referred to as allosteric modulators (effectors or modifiers) **bind at the allosteric site and regulate the enzyme activity**. The enzyme activity is increased when a positive (+) allosteric effector binds at the allosteric site known as activator site. On the other hand,a negative (–) allosteric effector binds at the allosteric site called inhibitor site and inhibits the enzyme activity.

Applications of inhibitors

- **Negative feedback**: end point or end product inhibition
- **Poisons** snake bite, plant alkaloids and nerve gases.
- **Medicine** antibiotics, sedatives and stimulants

- Home work

What are applications of enzyme in médicine and in the life