Enzyme

Catalytic efficiency

Enzyme	Turnover number (molecules reacted per enzyme molecule per minute)
Carbonic anhydrase	36,000,000
Catalase	5,600,000
Phosphoglucose isomerase	1,240
Succinate dehydrogenase	1,150

♥Enzymes are usually very **specific** as to which reactions they catalyze and the substrate that are involved in these reactions.
♥Complementary shape, charge and hydrophilic/hydrophobic characteristics of enzyme and substrates are responsible for this specificity.
♥Enzymes **catalyze the forward and backward reactions equally**. They do not alter the equilibrium itself, but only the speed at which it is reached.

Catalytic efficiency (cont.)

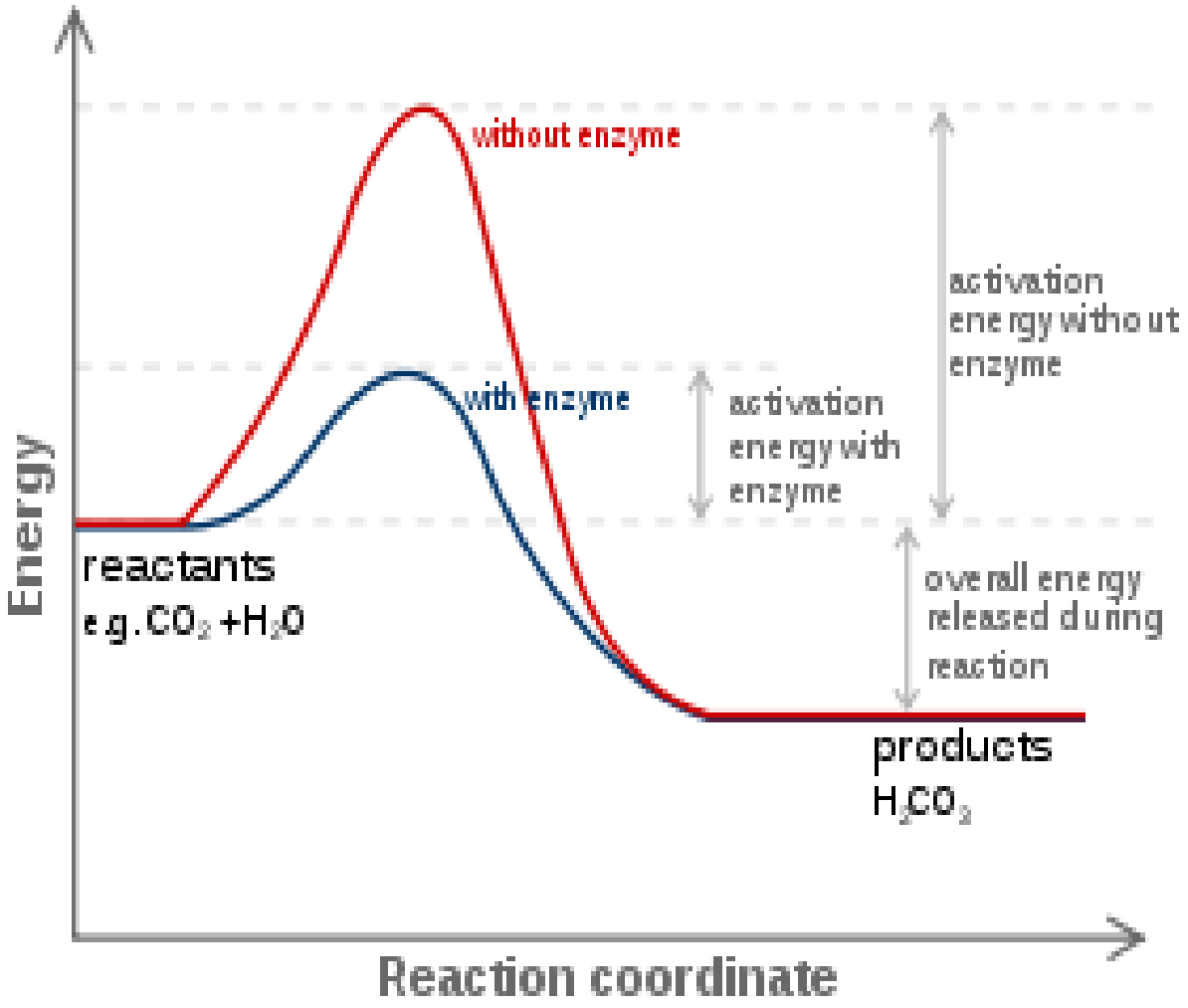
- For example, Carbonic anhydrase catalyzes its reaction in either direction depending on the concentration of its reactants.

$$CO_2 + H_2O \xrightarrow{\text{Carbonic anhydrase}} H_2CO_3$$

- (in tissues ,high carbon dioxide conc.)

$$H_2CO_3 \xrightarrow{\text{Carbonic anhydrase}} CO_2 + H_2O$$

- (in lungs, low carbon dioxide conc.)


without enzyme
with enzyme
activation energy without enzyme
activation energy with enzyme
overall energy released during reaction
reactants
e.g. $CO_2 + H_2O$
products
H_2CO_3
Energy
Reaction coordinate

The Lock and Key Hypothesis

- Fit between the substrate and the active site of the enzyme is exact
- Like a key fits into a lock very precisely
- The key is analogous to the enzyme and the substrate analogous to the lock.
- Only one substrate will fit into thee active site, just as one key fits a lock.
- Temporary structure called the enzyme-substrate complex formed
- Products have a different shape from the substrate

•Products have a different shape from the substrate

Once formed, they are released from the active site,

Leaving it free to become attached to another substrate

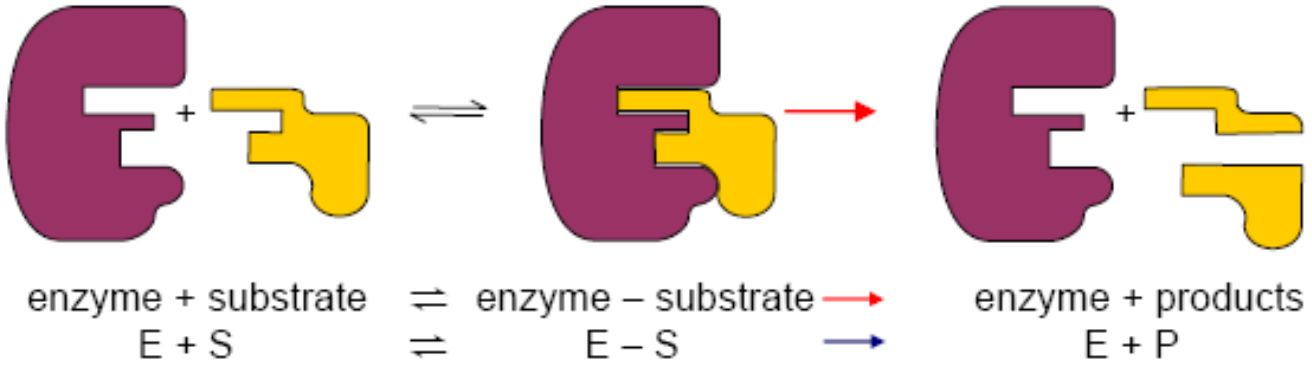
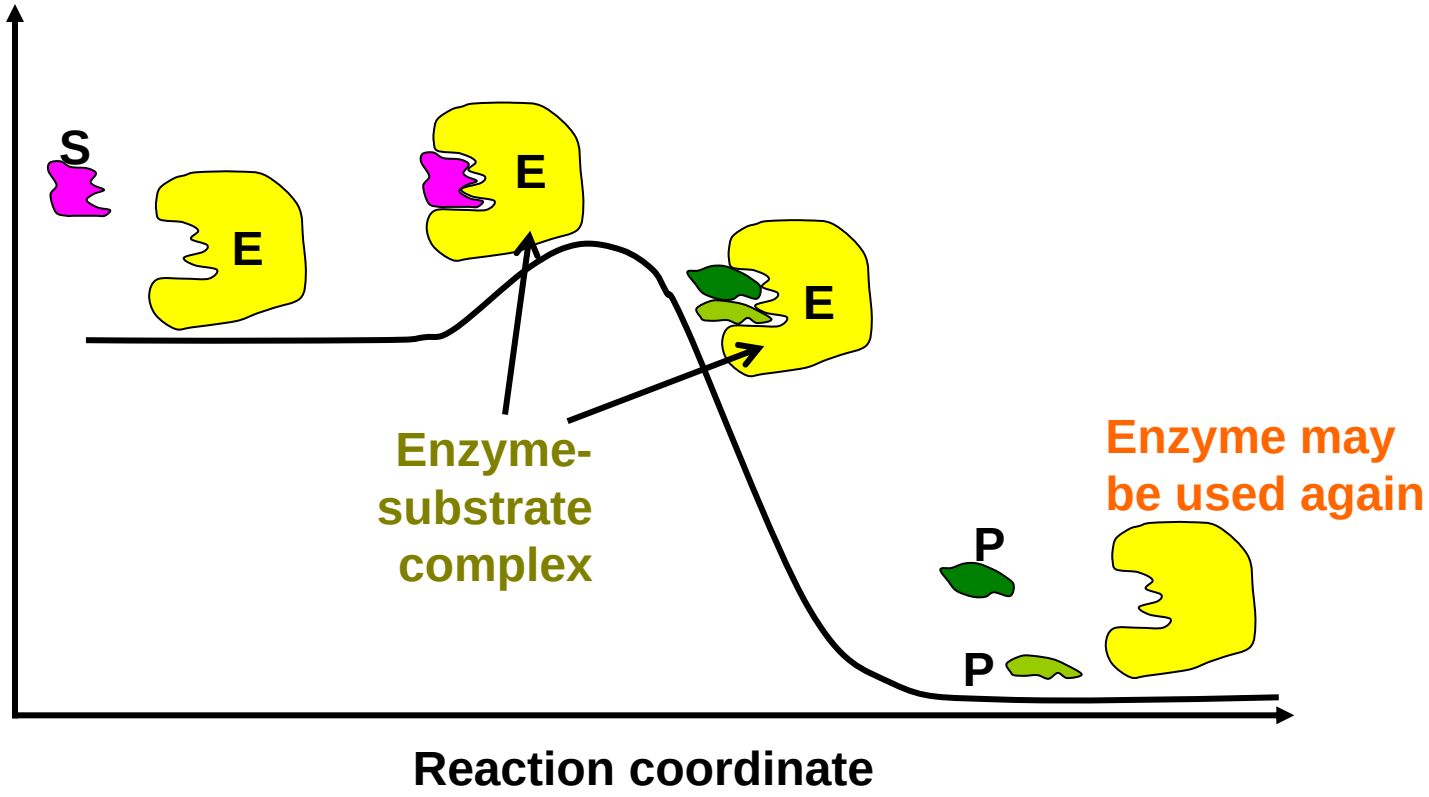


Figure 1.18 – the 'lock and key' mechanism



- First stage & second stage of reaction is reversible if the available energy is not greater than the E_a
- Enzymes function by providing alternative reaction pathway that required a lower activation energy (E_a)
- Once the products are formed, they leave the active site of the enzymes and are free to combine with a new substrate molecule.

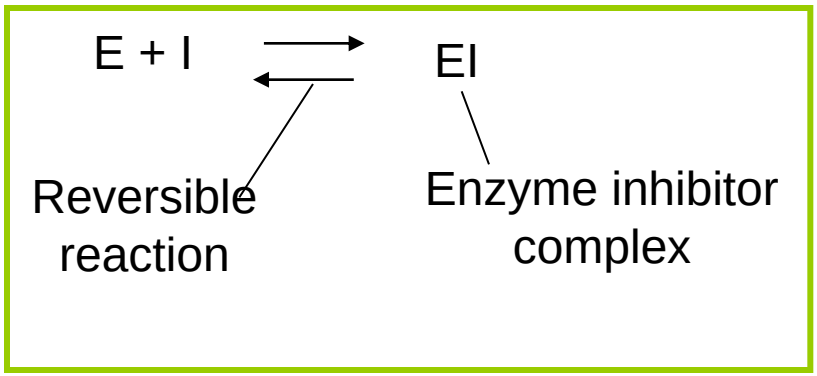
Inhibitors

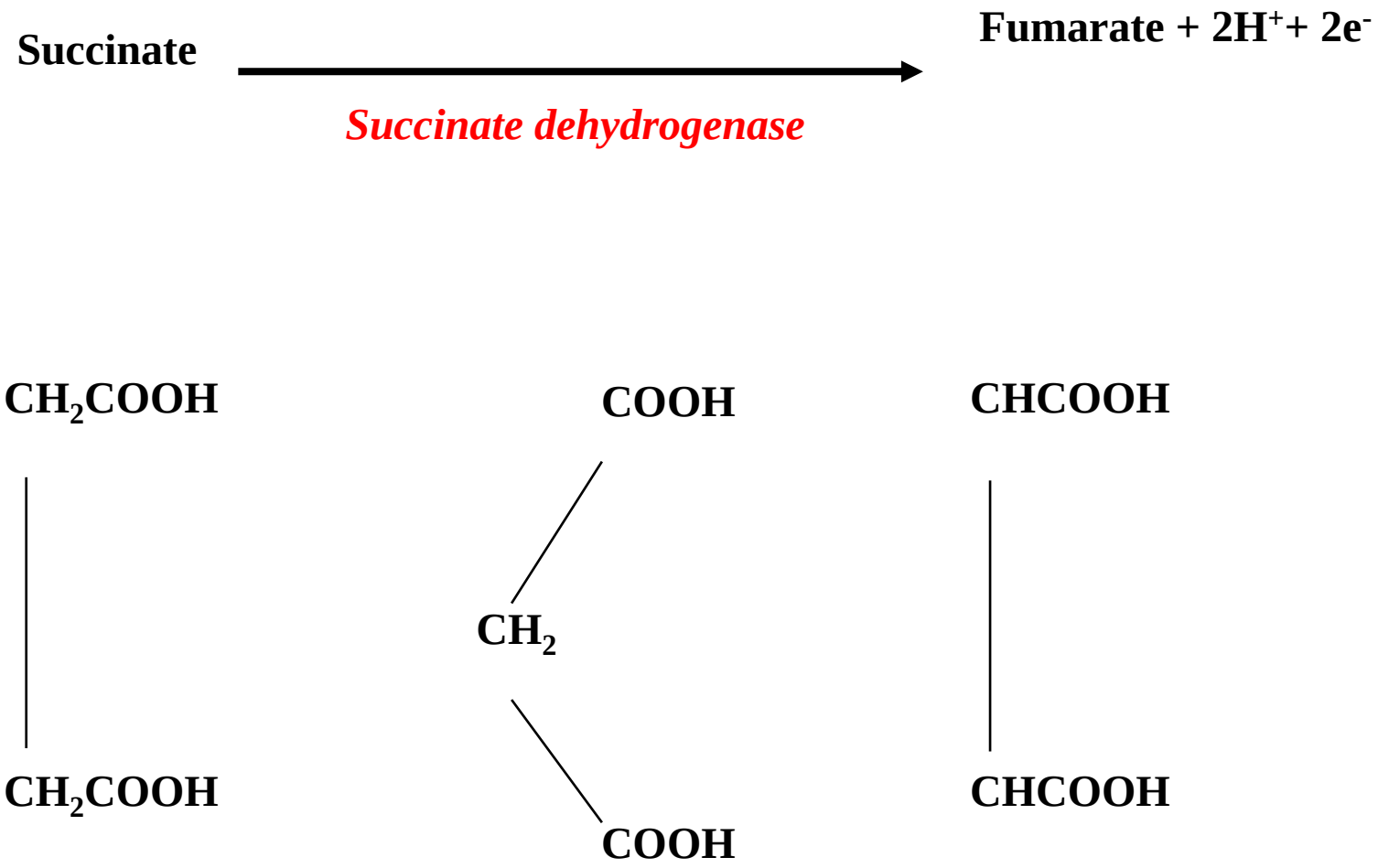
- Inhibitors are chemicals that reduce the rate of enzymic reactions.
- Many drugs and poisons are inhibitors of enzymes in the nervous system.

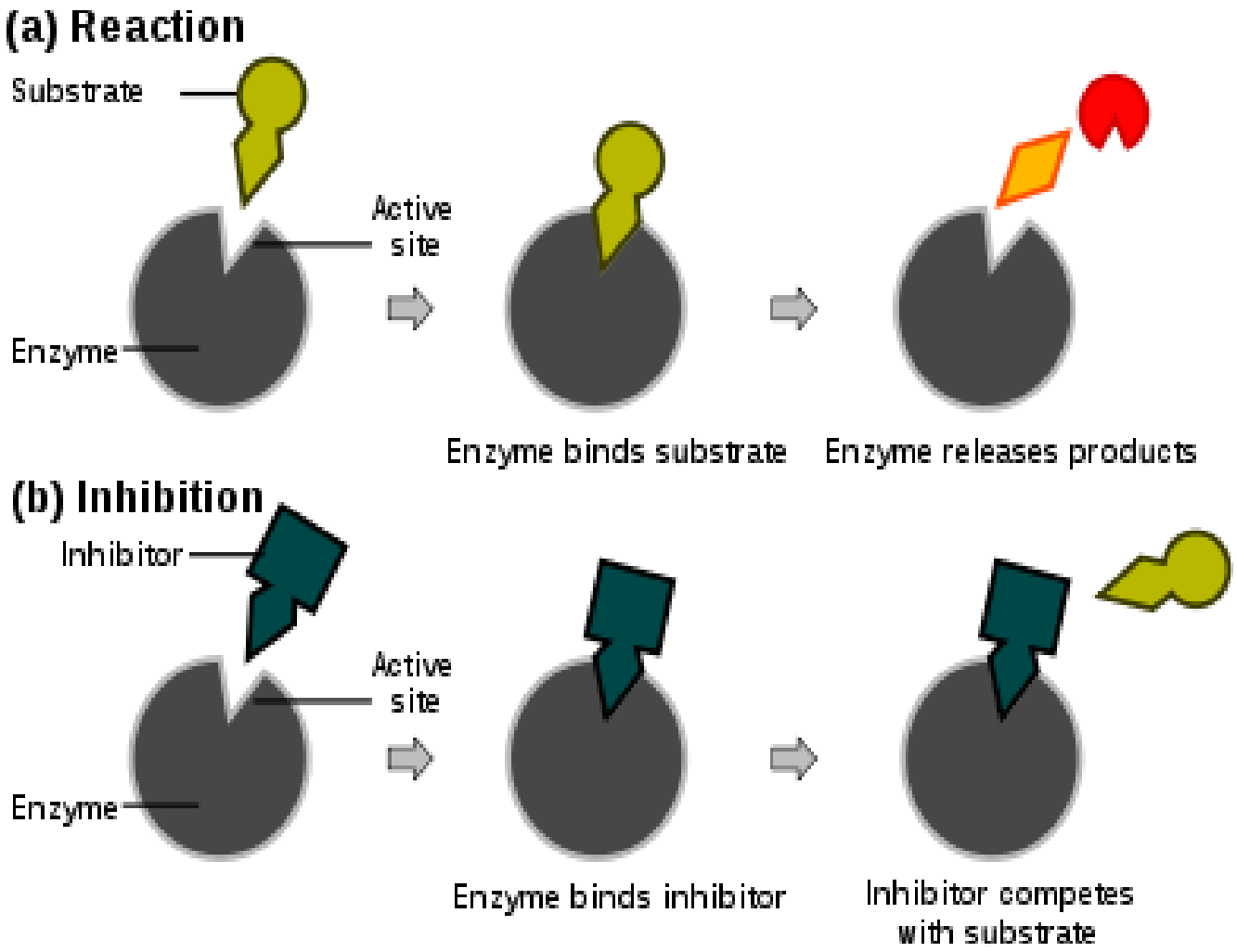
Reversible inhibitors

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.
- Are able to bind to the active site but do not participate in catalysed reactions.
- Reaction is reversible by an increase in substrate concentration.




Succinate
Fumarate + 2H+ + 2e-
Succinate dehydrogenase
CH2COOH
CH2COOH
COOH
CH2
COOH
CHCOOH
CHCOOH
Malonate


(a) Reaction
Substrate
Active site
Enzyme
Enzyme binds substrate
Enzyme releases products
(b) Inhibition
Inhibitor
Active site
Enzyme
Enzyme binds inhibitor
Inhibitor competes with substrate

2. **Non-competitive:** Molecules binds to regions of the enzymes other than the active site and affect enzyme activity.

☺ These are not influenced by the concentration of the substrate. It inhibits by binding irreversibly to the enzyme but **not at the active site. Active site changes shape so substrate cannot bind.**

☺ **Cannot be overcome by adding substrate**

☺ **Effect**

- reduce the number of active enzyme molecules available for reaction

- max rate of reaction is lowered

☺ **Inhibition is reversible**

☺ **- when concentration of inhibitor falls**

- enzyme-inhibitor complex falls apart and the functional shape of the enzyme is restored.

Non-competitive inhibitors

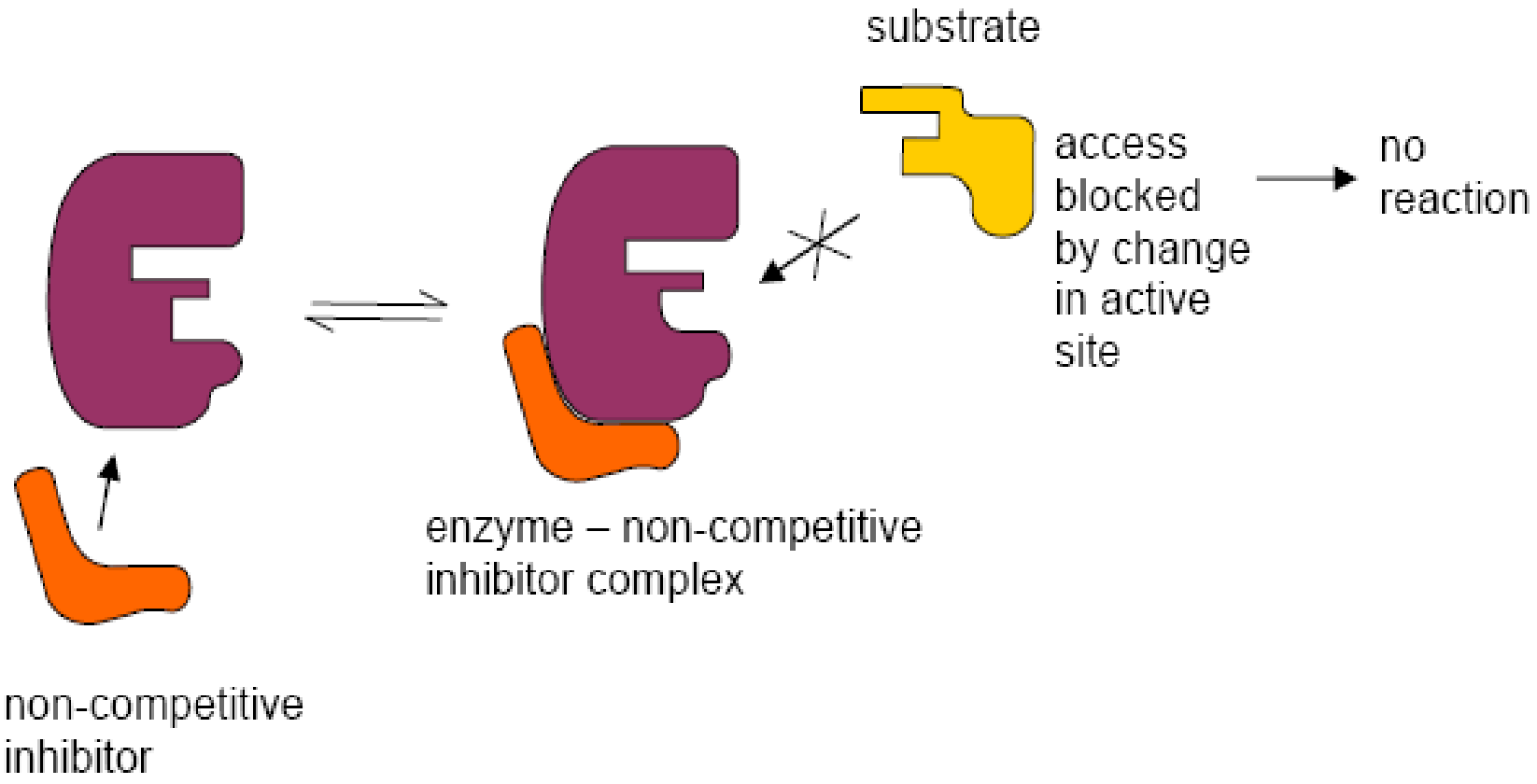
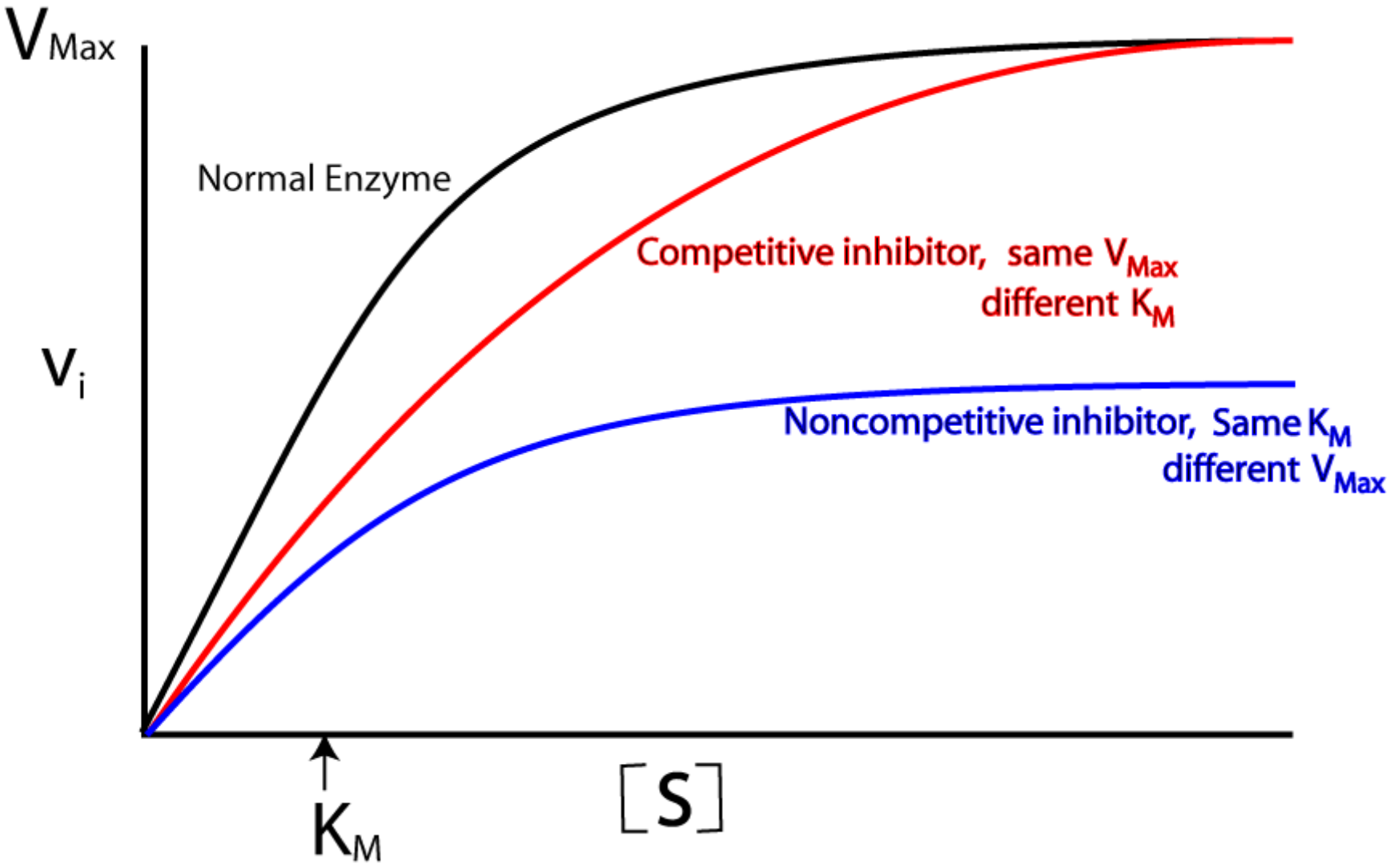


Figure 1.23 – scheme for non-competitive inhibition

Graph of Competitive Inhibitor and Non Competitive Inhibitor



Differences between competitive and non-competitive inhibitors?

	Competitive inhibitors	non competitive inhibitors
Similarity	Affect the rate of enzyme activities	
	bind to the enzymes and are reversible	
Differences	bind to the active site	bind to places other than the active site
	Affected by the concentration of substrate	not affected by the concentration of the substrates

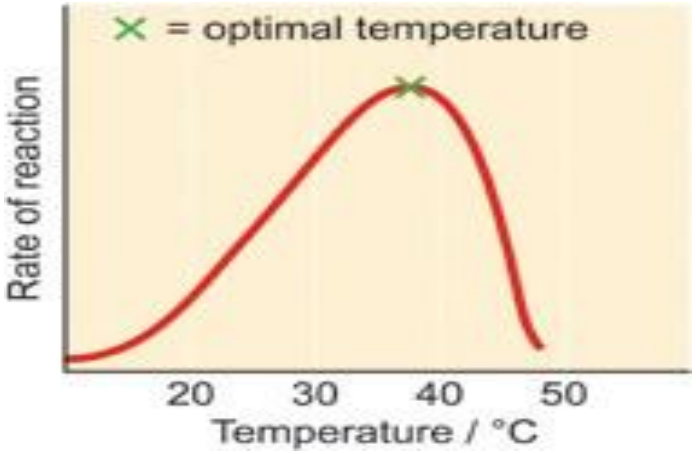
Applications of inhibitors

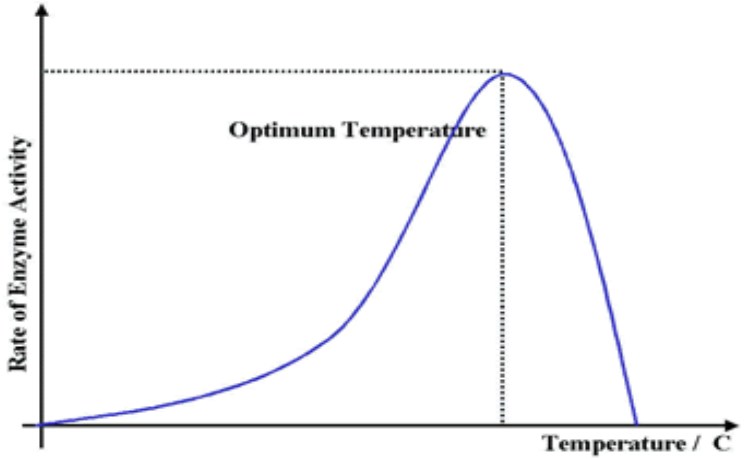
- **Negative feedback**: end point or end product inhibition
- **Poisons** snake bite, plant alkaloids and nerve gases.
- **Medicine** antibiotics, sulphonamides, sedatives and stimulants

Factors affecting enzyme activity

- Temperature
- pH
- Substrate concentration
- Enzyme concentration
- Inhibitors
- Concentration of Salts
- Effects of Heavy Metal

Temperature

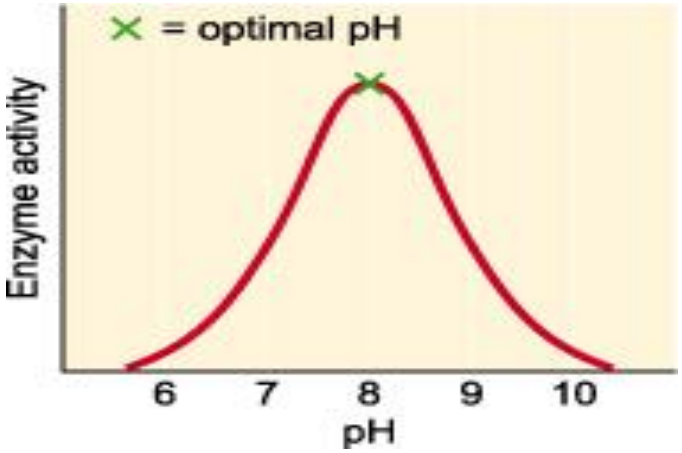


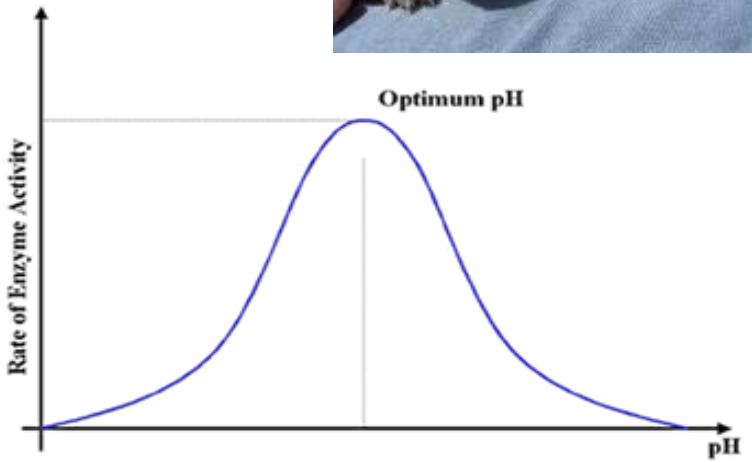


- Enzyme is inactive at low temperature. This is because the substrate molecules are moving at a relatively slow rate.
- They have low kinetic energy most of them do not possess the minimum energy required for reaction to occur.

- As the temperature rises, its activity increases as indicated by the increase in the rate of reaction since the molecules involved have greater kinetic energy.
- The more frequent collision between molecules also increase the chances of them coming into contact.
- The enzyme is twice as active for every 10°C rise in temperature until the optimum temperature is reached.
- Beyond the optimum temperature, any increase in temperature causes the rate of reaction to decrease sharply.
- Increased thermal motion of the polypeptide chain is causing disruption of the forces maintaining the three-dimensional shape of enzyme and eventually destroy the active sites.
- The enzyme is said to be denatured (irreversible).

pH





- Extreme changes in the acidity or alkalinity of solutions denature the enzymes.
- Changes in pH affect the the hydrogen and ionic bonding and ionization of amino acid side-chains in active site.

- The substrates are unable to bind to the active sites of the enzyme and reaction cannot take place.
- Optimum pH is the pH at which the rate of enzymatic reaction is at its fastest.
- Not all enzyme has the same pH optimum.
- Unlike the effects if heat on enzymes, the effects of pH are normally reversible.
- When the pH of the solution reverts to the optimum level for the enzyme, the ionic charges on the active sites are restored, thus enabling the enzyme to resume its normal function.

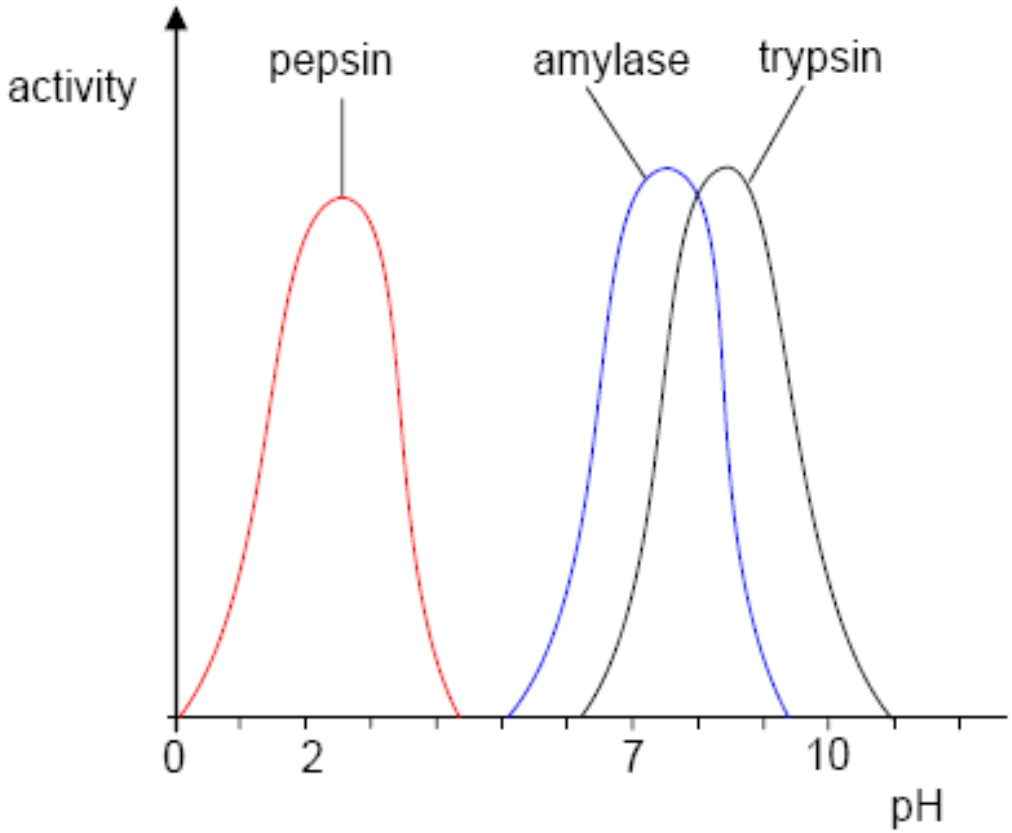
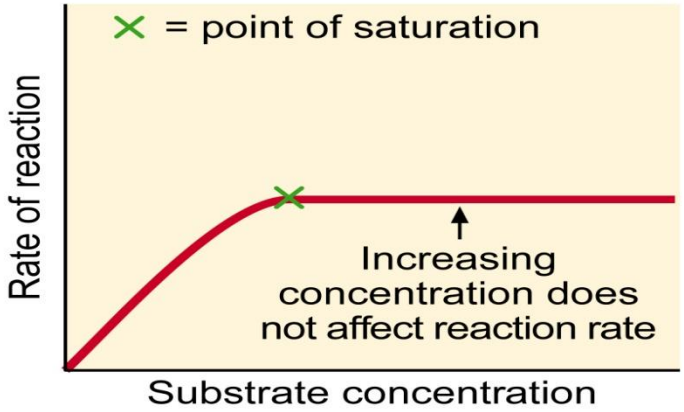
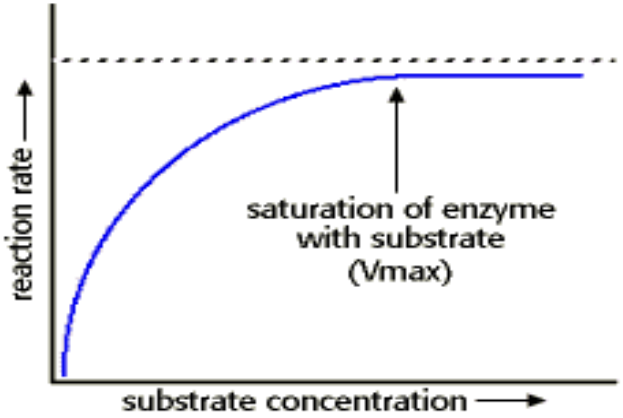


Figure 1.27 - curves showing pH optima for several enzymes

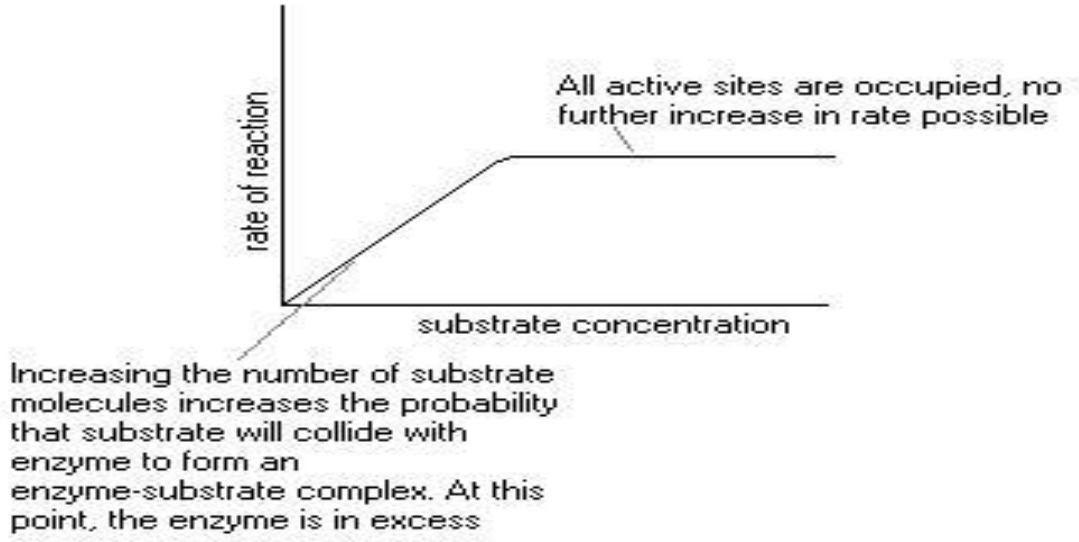
Substrate Concentration



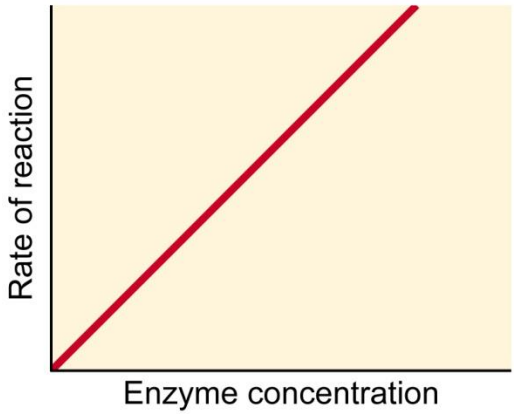


- At low substrate concentrations, few substrates are present and lots of active sites available.
- An increase in substrate concentration means more chances of collision between substrates and enzymes for a catalytic reaction to take place.

- As more substrates fill the active sites, more products are formed per unit time.
- The increase in substrate concentration will only lead to an increase in the rate if there are enough enzyme available to catalyze the additional substrates.
- When it reaches the point of saturation, the rate of reaction will not increase further and become constant.
- At this point, the enzyme concentration becomes a limiting factor.



Enzyme Concentration



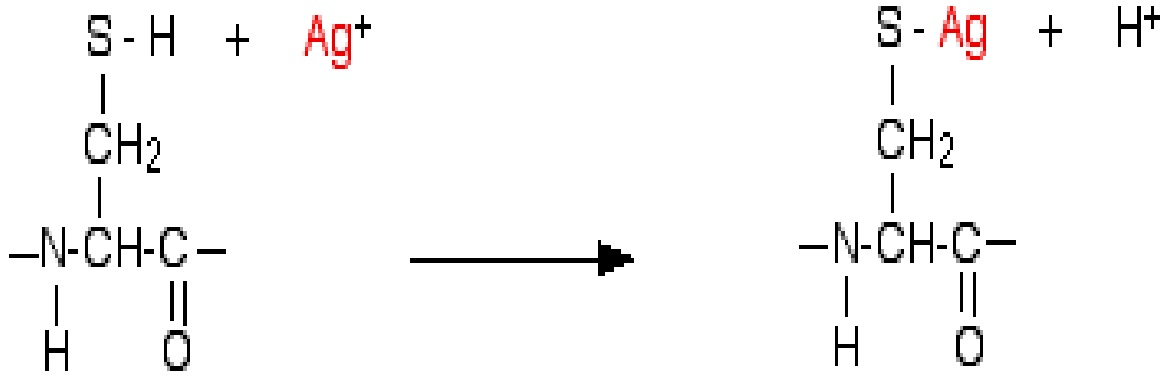
- The rate of reaction increases when enzyme concentration is increased, as long as no other factors limiting the rate and plenty of substrates are available.
- When the enzyme concentration is doubled, the rate of reaction will be doubled as long as substrates are present in excess.

Concentration of Salts

- High salt concentration changes ionic environment of an enzyme, disruption ionic interactions between different regions of the chain.
- Urea denatures proteins by disrupting the hydrogen bonds that maintain the secondary and tertiary structure of proteins
- Certain chemical inhibitors totally inactive enzymes; their effect are irreversible.

Effects of Heavy Metal

- **Heavy metals such as Ag+, Hg2+, Pb2+ have strong affinities for -SH groups.**
- Silver ions react with -SH groups in the side groups of cysteine residues in the protein chain:



cysteine residue in protein chain

- If the cysteine residue is somewhere on the protein chain which affects the way it folds into its tertiary structure, then altering this group could have an effect on the shape of the active site, and so stop the enzyme from working.

Questions

❶ **Factors affecting enzyme activities?**

- ✓ temperature
- ✓ concentration of enzymes
- ✓ concentration of substrate
- ✓ pH values
- ✓ presents of co-enzyme or cofactors
- ✓ presence of inhibitors

② **Explain why enzymes lose their catalytic effectiveness above 40 degree.**

- ✓ high kinetic energy
- ✓ bond broken
- ✓ lose their secondary and tertiary structure
- ✓ enzyme denatured

❸Explain how non competitive inhibitor affect the enzymes activities.

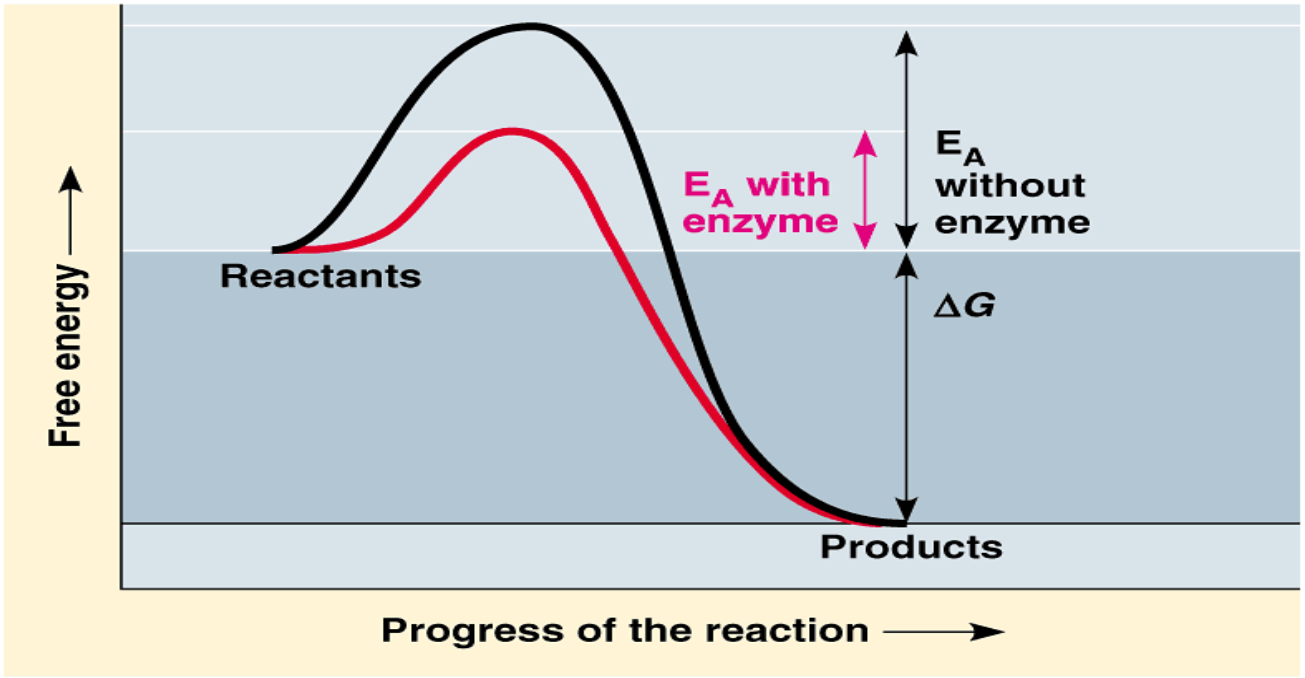
- ✓ inhibitors bind to the place other than active site
- ✓ alter the shape of the active site
- ✓ substrate unable to bind to the enzyme. no enzyme substrate complex.
- ✓ stops normal enzyme functions.

④ (a) Sketch the energy profile of an uncatalysed exothermic reaction, showing :

(I) the activation energy (Ea), and

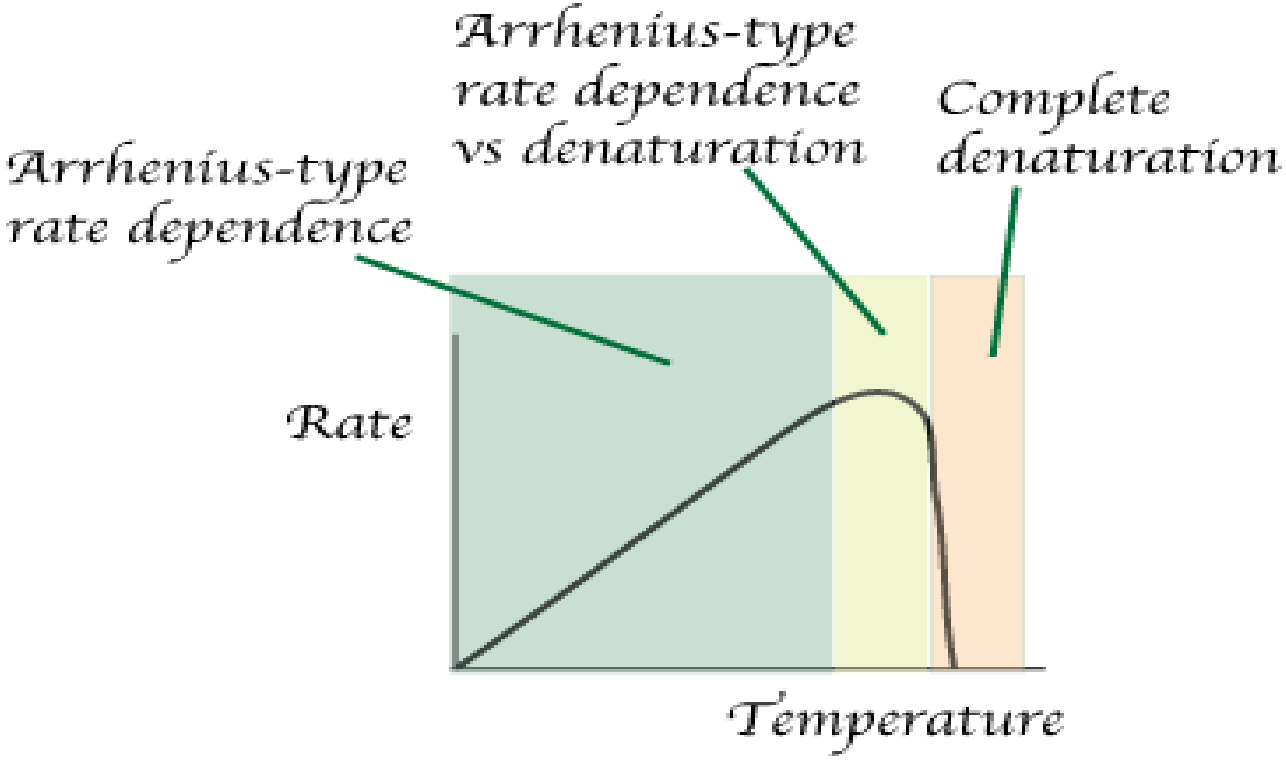
(ii) the enthalpy change of reaction (ΔHr).

(b) Sketch a similar energy profile for the above reaction when it is catalyzed by an enzyme.



❺ (a) Sketch a graph to show how the activity of an enzyme varies with temperature.

(b) Explain the shape of the graph in terms of kinetic theory and the effect of temperature on the integrity of the enzyme's structure.



- ✓ Between 0°C and approximately 40°C the rate of enzyme activity increases almost linearly
- ✓ the molecules are moving more quickly, increasing the frequency of collision
- ✓ a greater proportion of the collisions involve molecules with energy greater than the activation energy for the catalysed reaction.
- ✓ the rate of reaction starts to decrease above 40°C.
- ✓ Increased thermal motion of the polypeptide chain is causing disruption of the forces maintaining the shape of the enzyme molecules.
- ✓ The enzyme molecules are progressively denatured, causing the shape of the active site to change.
- ✓ Above 65°C the enzymes are completely heat denatured.

THE END