Lecture Three

Two Models for Enzyme/Substrate Interactions:

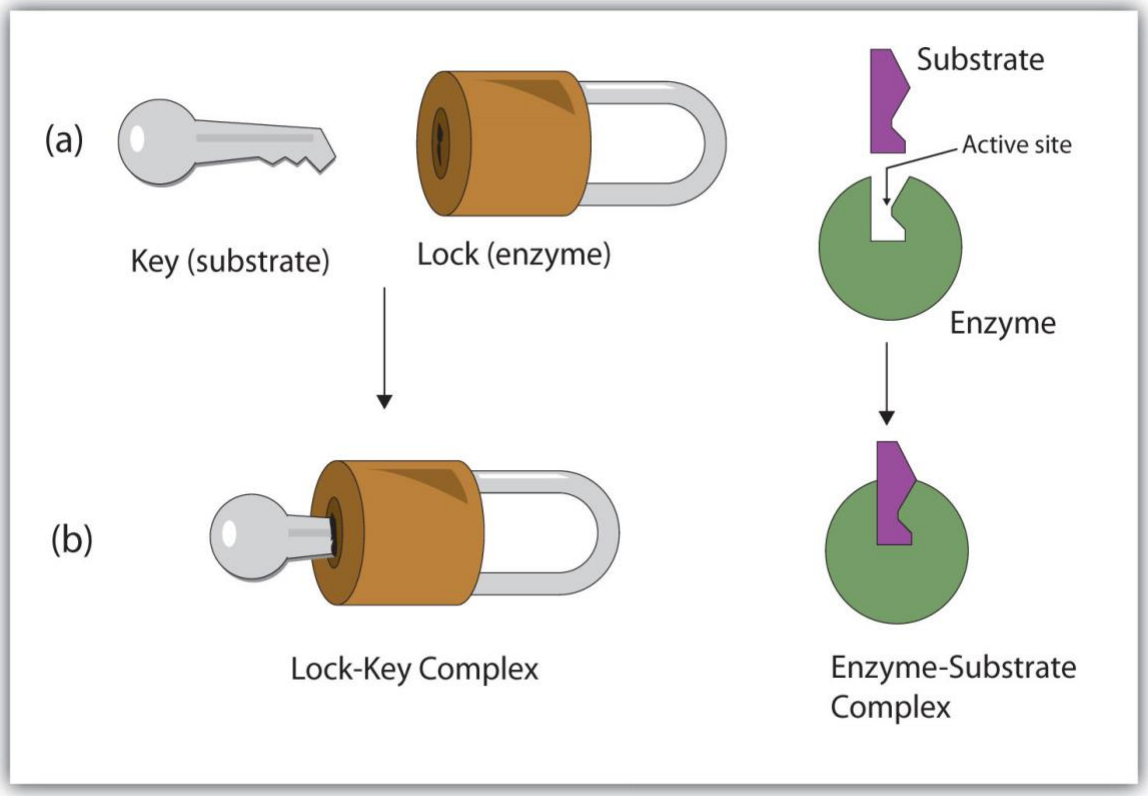
Active site:

1) During the enzyme action, there is a temporary combination between the enzyme and its substrate forming enzyme-substrate complex. **This occurs at active site of enzyme**.

2) This is followed by dissociation of this complex into enzyme again and products.

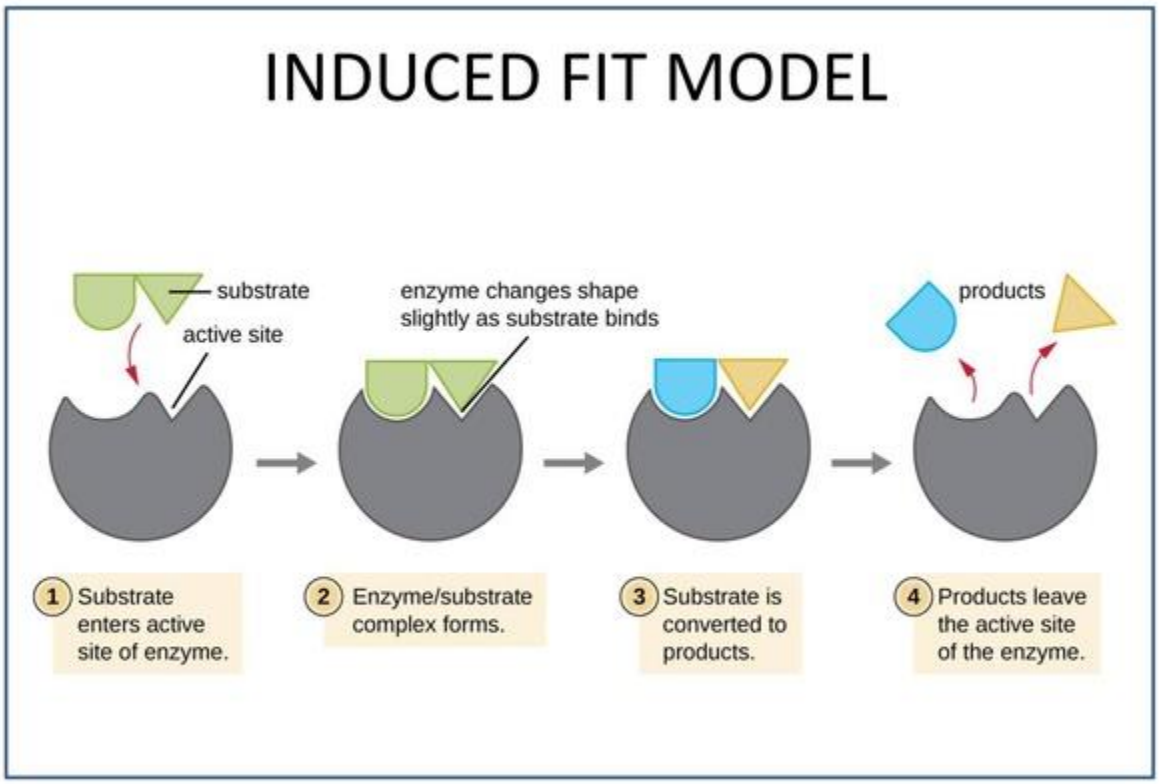
1-Lock and key model

- In this theory, the active site of the enzyme is complementary in conformation to the substrate so that enzyme and substrate "recognize" one another.



2- Induced Fit Model

- The enzyme changes shape upon binding the substrate, so that the conformation of substrate and enzyme protein are only complementary after the binding reaction.



FACTORS AFFECTING REACTION VELOCITY

Substrate concentration

The rate of an enzyme-catalyzed reaction increases with substrate concentration until a maximal velocity (V_{max}) is reached. The leveling off of the reaction rate at high substrate concentrations reflects the saturation with substrate of all available binding sites on the enzyme molecules present.

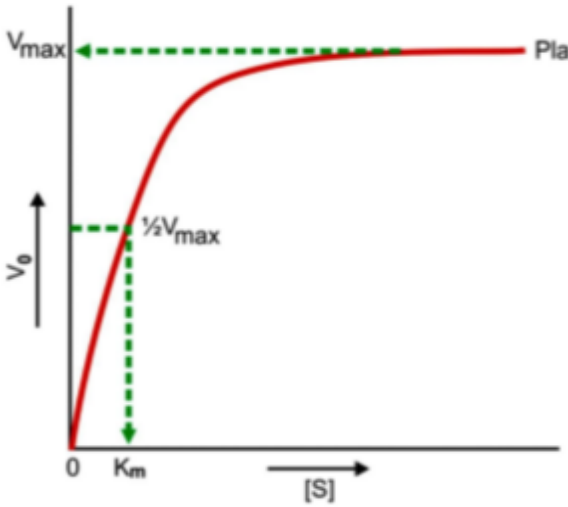


Figure 2.3: Effects of substrate concentration [S] on enzyme activity keeping enzyme concentration constant
where, V_0 : initial velocity
V_{max} : maximum velocity
K_m : 1/2 V_{max} = Michaelis Menten constant
[S] : substrate concentration

B. Temperature

- The optimal temperature for enzymatic activity in human body is **37 ċ**.i.e. the temperature of the cell.
- At **zero** temperature, the enzyme is **inactive**. The reaction velocity increases with increase of temperature until a maximum velocity is reached,
- Further elevation of the temperature results in a decrease in reaction velocity. At **55°C-60°C**, most enzymes are denaturated and become

permanently inactive.

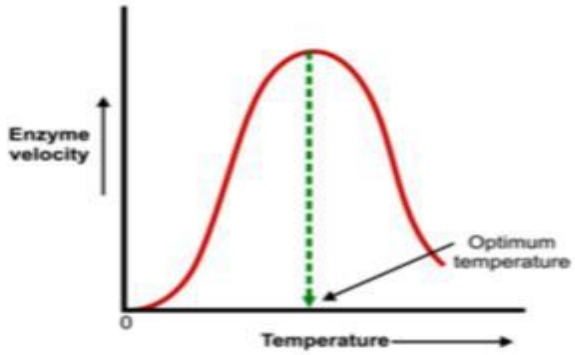


Figure 2.6: Effect of temperature on enzyme activity

➢ C. pH

Each enzyme has an opting on pH, i.e. a pH at which the enzyme activity is maximum. Below or above this pH, enzyme activity is decreased. The optimum pH differs from enzyme to enzyme, e.g. optimum for:

• Pepsin — I .2
• Trypsin — 8.0

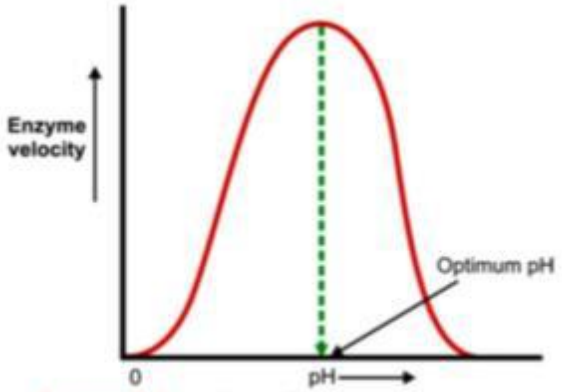


Figure 2.5: Effect of pH on enzyme activity

INHIBITION OF ENZYME ACTIVITY

➢ Enzyme Inhibition

Any substance that can diminish the velocity of an enzyme catalyzed reaction is called inhibitor. Two general classes of inhibitors are

1. Reversible inhibitor.
2. Irreversible inhibitor.

❖ Reversible Inhibitor

Reversible inhibitors bind to enzymes through noncovalent bonds. Different types of reversible inhibitors are:

A. competitive inhibition or substrate analogue inhibitor.

B. Noncompetitive inhibition

C. Un-Competitive Inhibitors

- **competitive inhibition or substrate analogue inhibitor.**

This type of inhibition occurs when the inhibitor binds reversibly to the same site that the substrate would normally occupy and, therefore, competes with the substrate for that site.

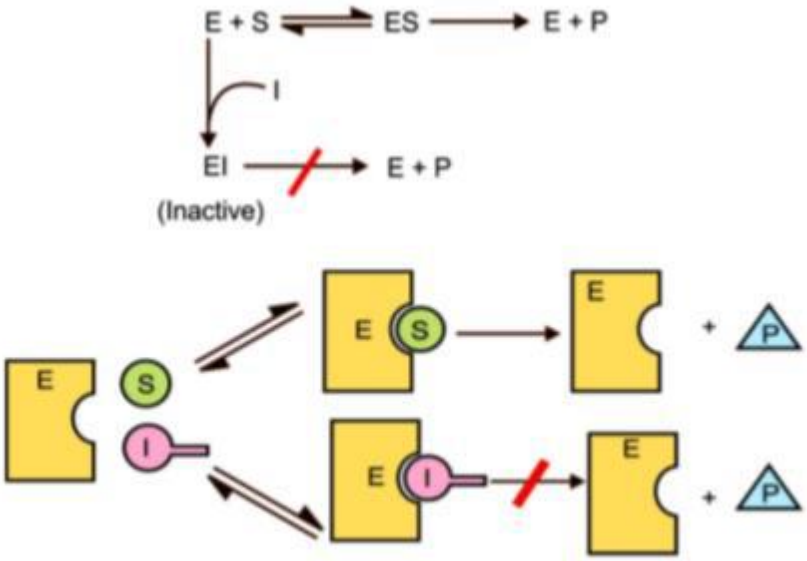
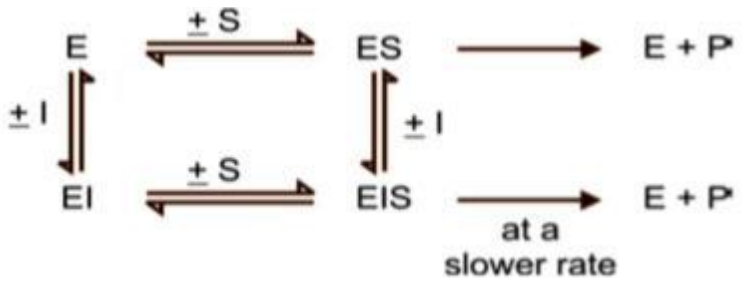


Figure 2.8: Diagrammatic representation of competitive inhibition where, E: Enzyme; S: Substrate; I: Competitive inhibitor; P: Product.

- **B. Noncompetitive inhibition**

Noncompetitive inhibition occurs when the inhibitor and substrate bind at different sites on the enzyme. The noncompetitive inhibitor can bind either free enzyme or the ES complex, thereby preventing the reaction from occurring.



➢ C. Un-Competitive Inhibitors

Inhibitor binds to a site other than the active site, but only when substrate is bound (Binds to ES complex). distorts active site; prevents reaction from occurring

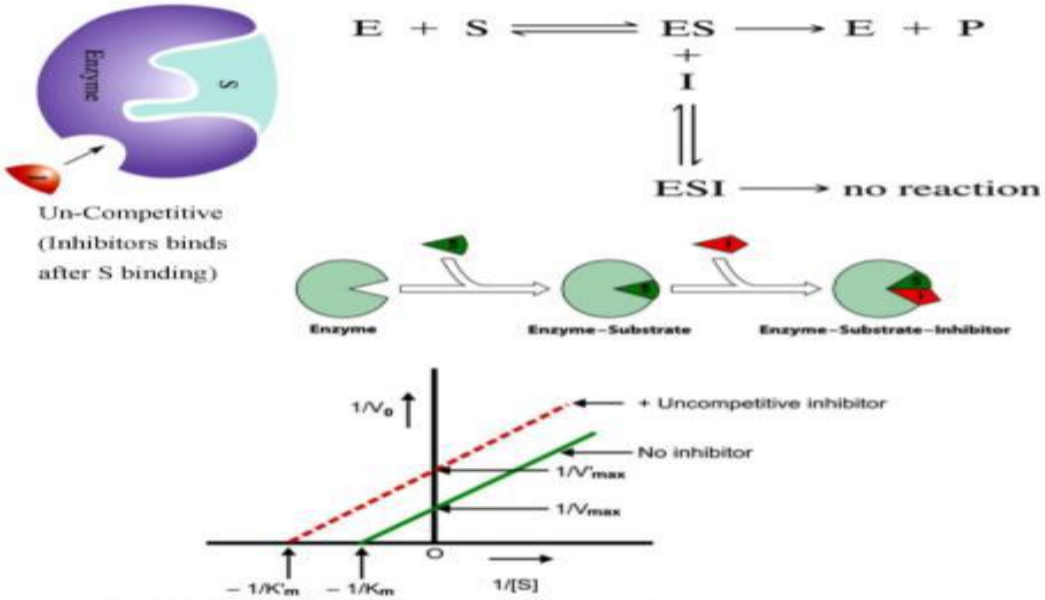


Figure 2.11: Effect of uncompetitive inhibitor on the double reciprocal plot; it shows parallel lines with decrease in both Vmax and Km

❖ Irreversible Inhibitor

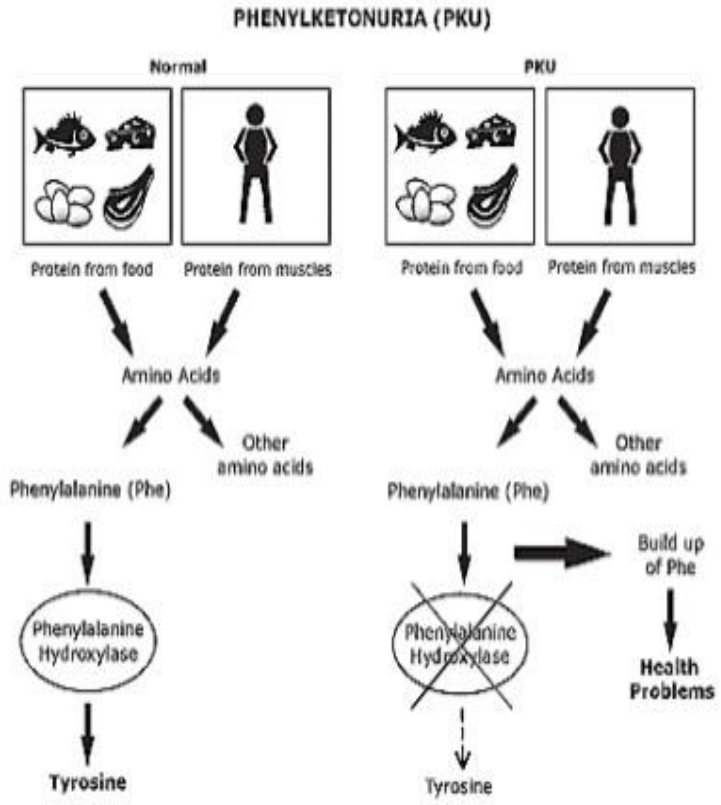
An irreversible inhibitor binds with an enzyme tightly covalently and forms a stable complex.

Type of inhibitor	Km	V_{max}
Irreversible	No effect	Decreased
Reversible competitive	Increased	No effect
Reversible noncompetitive	No effect	Decreased
Reversibe uncompetitive	Decreased	Decreased

Phenylketonuria (PKU)

• It is an inborn error of phenylalanine metabolism resulting from deficiency of phenylalanine hydroxylase (PAH), associated with the inability to convert phenylalanine to tyrosine

• In phenylketonuria, there is an accumulation of phenylalanine in tissues and blood and results in its increased excretion in urine


PHENYLKETONURIA (PKU)
Normal
PKU
Protein from food
Protein from muscles
Protein from food
Protein from muscles
Amino Acids
Amino Acids
Other amino acids
Other amino acids
Phenylalanine (Phe)
Phenylalanine (Phe)
Build up of Phe
Phenylalanine Hydroxylase
Phenylalanine Hydroxylase
Health Problems
Tyrosine
Tyrosine