3.6. Enzyme Catalysis

An enzyme is a protein or a protein like substance with catalytic properties. Enzymes show high specificity in their

catalytic activity toward one substrate; and they involve no complicating side reactions. An enzyme is named by adding '–ase' after the major part of the name of the substrate (or the reactant). For example, enzyme that catalyses decomposition of urea is called 'urease' and the enzyme that catalyses hydrolysis of 'sucrose' is called 'sucrase'. The catalytic effect of enzymes is, by far, startling. For example, compare the catalytic effect of hydrogen ions with that of the enzyme sucrase on hydrolysis of sucrose at the blood temperature (37°C). The activation energy of the hydrogen ion catalysed reaction is 107 kJ/mol, while that of the enzyme catalysed reaction is 36kJ/mol. These figures correspond to 10^{12} times rise in the reaction rate due to the presence of enzyme.

Following are general characteristics of the enzyme catalysed reactions.

1. At the end of the reaction, enzyme is neither consumed nor produced.
2. Enzymes catalyse only those reactions which in their absence will occur, but at much slower rate.
3. Enzymes are highly specific in their catalytic behaviour. It is rare that the same enzyme may catalyse two reactions with different substrates.
4. The enzyme catalysed reactions do not require extreme conditions of low or high temperature, instead they occur in a moderate temperature range.
5. Enzyme catalysed reactions involve no side reactions and therefore, they produce no undesired products.

There are three kinds of the enzyme catalysed reactions:

(I) soluble enzyme – soluble substrate type
(II) soluble enzyme – insoluble substrate type
(III) insoluble enzyme – soluble substrate type

Type (I) reactions are homogeneous and the other two are heterogeneous. Type (III) reactions have gained interest due to their growing applications in industry. But, the greatest

importance of the enzyme reactions is due to their vast applications in reactions in living cells. These reactions generally occur in homogeneous liquid phase. We shall restrict our theoretical discussion to the homogeneous enzyme catalysis. The results of these discussions may apply to the other two types of reactions also.

Steady-state Enzyme Kinetics

3.6.1. Michaelis – Menten Mechanism of Enzyme Reactions

Let us consider the catalytic action of enzyme E on substrate S. Michaelis and Menten proposed that an enzyme catalysed reaction involves the reversible formation of an enzyme – substrate complex ES, which hydrolyses to give the free enzyme E and the product P.

$$E + S \underset{k_2}{\overset{k_1}{\rightleftharpoons}} ES \qquad (3.52)$$

$$ES + H_2O \xrightarrow{k_3} P + E \qquad (3.53)$$

Rate of formation of ES = k_1 [E].[S] – k_2 [ES]

Rate of disappearance of ES = k_3 [ES].[H_2O]

Since the enzyme E is not consumed, the rates of formation and disappearance of ES must be equal. Therefore,

$$k_1 [E].[S] - k_2 [ES] = k_3 [ES].[H_2O] \qquad (3.54)$$

Let the total enzyme concentration be [E_t]. Then, at any instant during the progress of the reaction,

$$[E_t] = [E] + [ES] \qquad (3.55)$$

Combining equations (3 54) and (3.55),

$$[ES] = \frac{k_1 [E_t].[S]}{k_1 [S] + k_2 + k_3 [H_2O]} \qquad (3.56)$$

Considering the reaction steps (3.52) and (3.53), the rate of the reaction (–d[S]/dt) is given by

$$v = -\frac{d[S]}{dt} = k_1 [E].[S] - k_2 [ES] \qquad (3.57)$$

Substituting the value of [E] in terms of $[E_t]$ and [ES] from equation (3.55)

$$v = -\frac{d[S]}{dt} = k_1 [E_t].[S] - (k_1[S] + k_2) [ES]$$

Putting the value of [ES] from equation (3.56) in the above equation,

$$v = -\frac{d[S]}{dt} = k_1 [E_t].[S] - \frac{(k_1[S] + k_2)\, k_1 [E_t] [S]}{k_1 [S] + k_2 + k_3 [H_2O]}$$

$$= \frac{k_1[E_t] [S]\ k_3 [H_2O]}{k_1 [S] + k_2 + k_3 [H_2O]} \qquad (3.58)$$

Since H_2O is in large excess, $[H_2O]$ does not change during the reaction. Therefore, we may write,

$$k_3 [H_2O] = k_3' \qquad (3.59)$$

Then, equation (3.58) changes to

$$v = -\frac{d[S]}{dt} = \frac{k_3'.[E_t] [S]}{[S] + (k_2 + k_3')/k_1} \qquad (3.60)$$

or,
$$v = -\frac{d[S]}{dt} = \frac{k_3'.[E_t] [S]}{[S] + K_m} \qquad (3.61)$$

where,
$$K_m = (k_2 + k_3')/k_1 \qquad (3.62)$$

K_m is called *Michaelis constant*. We can see that K_m is dependent on temperature and it is independent of concentration terms.

For a typical enzyme-catalysed reaction, the rate of the reaction rises with increasing substrate concentration [S], and at sufficiently high concentration it reaches a constant maximum value V_{max} (Figure 3.3). At a given temperature, V_{max} is the characteristic of the reaction.

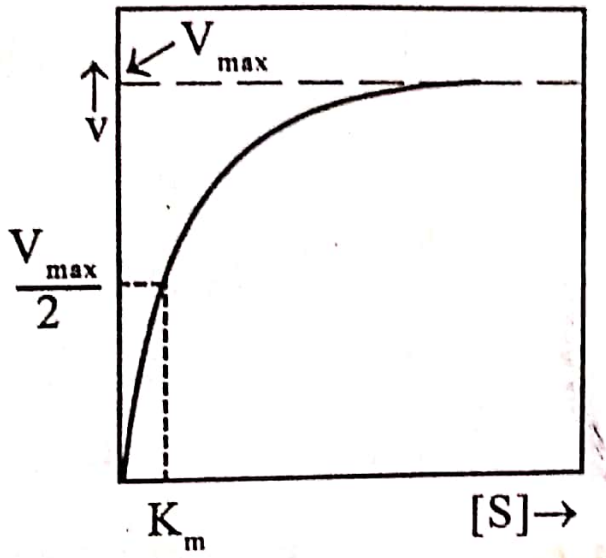

Figure 3.3. Concentration dependence of a typical enzyme - catalysed reaction

Corresponding to very high substrate concentration, $[S] >> K_m$ and $v = V_{max}$. Then from equation (3.61),

$$V_{max} = k_3'.[E_t] \qquad (3.63)$$

Combining equations (3.61) and (3.63),

$$v = -\frac{d[S]}{dt} = \frac{V_{max}[S]}{[S] + K_m} \qquad (3.64)$$

This is called the *Michaelis – Menten equation*. Consider the following special cases of this equation.

(i) *Very low concentration of the substrate*: In this case, $[S] << K_m$ and equation (3.64) reduces to

$$v = \frac{V_{max}}{K_m} [S] \propto [S] \qquad (3.65)$$

So, at low concentrations of the substrate, the reaction obeys first order kinetics.

(ii) *High concentration of the substrate:* We have already seen that at high concentration of the substrate, $[S] >> K_m$ and the rate of the reaction equals V_{max}. Thus,

$$v = V_{max} = \text{constant} \qquad (3.66)$$

So, at high concentration of the substrate, the rate of the reaction is independent of the concentration term corresponds to zero order kinetics.

(iii) *Significance of K_m*: If K_m is set equal to [S], equation (3.64) yields $v = ½ V_m$. Therefore, K_m is numerically equal to concentration of the substrate at which the reaction rate is half maximal.

$$\text{When } v = ½ V_{max}, \quad [S] = K_m. \qquad (3.67)$$

This property can be used for calculating K_m. See Figure 3.3.