

Basic enzyme kinetics

Initial Rate Studies

Two assumptions

1) Rapid equilibrium

i.e. formation of ES complex is very much faster than its breakdown to release product.

2) Steady state assumption (Briggs-Haldane)

1) Rapid equilibrium



i.e. $k_2 \ll k_1 \text{ \& } k_{-1}$

$$\therefore \frac{[E][S]}{[ES]} = \frac{k_{-1}}{k_1} = K_s$$

So K_s is a true dissociation constant.

2) Steady state assumption (Briggs-Haldane)

$$\frac{d[ES]}{dt} = 0$$

$$k_1[E][S] = (k_{-1} + k_2)[ES]$$

$$\frac{[E][S]}{[ES]} = \frac{(k_{-1} + k_2)}{k_1} = K_m$$

K_m is not a simple dissociation constant.

- The steady state assumption is more generally applicable because it does not make assumptions regarding the magnitudes of the individual rate constants.
- However for the assumption of S.S. to be valid the rate must be determined under conditions where $[ES]$ is unchanging.
- The only point where the reaction mixture is totally defined is at time zero as the reactants are mixed – hence if we determine the rate at time zero (tangent to the product-time curve at $t=0$) we know exactly what the conditions are.

Derivation of Michaelis Menten equation

- Mass Balance

$$[E_{total}] = [E] + [ES]$$

$$K_m \text{ or } K_s = \frac{[E][S]}{[ES]} \therefore [E] = \frac{[K_m][ES]}{[S]}$$

$$\therefore [E_{total}] = [ES] \left(1 + \frac{K_m}{[S]} \right)$$

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$$[ES] = \frac{[E_{total}]}{\left(1 + \frac{K_m}{[S]} \right)}$$

Multiplying by k_2

$$k_2[ES] = \frac{k_2[E_{total}]}{\frac{K_m}{[S]} + 1} = v = \frac{V_m}{\frac{K_m}{[S]} + 1} = \frac{V_m [S]}{K_m + [S]}$$