

# 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  $k_2$

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