Handout 4

Michaelis-Menten enzyme kinetics

\[ E + S \overset{k_1}{\underset{k_{-1}}{\rightleftharpoons}} ES \overset{k_2}{\rightarrow} E + P \]

E = enzyme  \quad S = substrate  \quad P = product

Full reaction kinetics for this system:

\[
\frac{d[E]}{dt} = -k_1[E][S] + k_{-1}[ES] + k_2[ES] \quad \frac{d[ES]}{dt} = k_1[E][S] - k_{-1}[ES] - k_2[ES] \\
\frac{d[S]}{dt} = -k_1[E][S] + k_{-1}[ES] \quad \frac{d[P]}{dt} = k_2[ES]
\]

Approach 1: quasi steady-state approximation for ES

\[
\frac{d[ES]}{dt} \approx 0 = k_1[E][S] - k_{-1}[ES] - k_2[ES]
\]

Using

\[
[E] + [ES] = [E]_0 = const \quad \rightarrow \quad [E] = [E]_0 - [ES]
\]

Substituting in above for [E] and solving for [ES],

\[
[ES] = \frac{k_1[E]_0[S]}{k_1[S] + k_{-1} + k_2} = \frac{[E]_0[S]}{[S] + (k_{-1} + k_2)/k_1} = \frac{[E]_0[S]}{[S] + K_M}
\]

where we have made the definition:

\[
K_M = \frac{k_{-1} + k_2}{k_1}
\]

Now compute the rate of product formation:

\[
v = \frac{dP}{dt} = \frac{k_2[ES]}{k_2[E]_0[S]} = \frac{[S]_0[S]}{[S] + K_M}
\]
Make the definition:

\[ v_{\text{max}} \equiv k_2 [E]_0 \]

Then the rate is given by,

\[ v = v_{\text{max}} \frac{[S]}{[S] + K_M} \]

**Approach 2: rapid equilibrium approximation for \( E + S \leftrightarrow ES \)**

Assume that substrate binding is in equilibrium with the reactants. Then,

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

Solving for \([E]\),

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

Combining with the mass balance equation \([E] + [ES] = [E]_0\),

\[ \frac{k_{-1} [ES]}{k_1 [S]} + [ES] = [E]_0 \]

Solving for \([ES]\),

\[ [ES] = \frac{[E]_0 [S]}{[S] + \frac{k_{-1}}{k_1}} \]

If we now define

\[ K_M \equiv \frac{k_{-1}}{k_1} \]

Then,

\[ [ES] = \frac{[E]_0 [S]}{[S] + K_M} \]

And the remainder of the derivation proceeds as before.

Therefore the quasi steady-state and equilibrium approximations lead to slightly different expressions for the Michaelis constant \( K_M \). For reactions that are rate-limited in the formation of the product, \( k_2 \ll k_{-1} \), the two approaches are roughly equivalent.