Ch. 29: reaction mechanisms

go over pages 21-26 of my Chapter 19 lecture notes for Chem 1312 since they do a good job going over elementary reactions.

Note: the textbook, on page 1193, discusses in detail when the steady state approx is valid.

29-6: the Lindemann mechanism.

Consider isomerization or decomposition reactions in the gas phase.

ex: \[ \text{CH}_3\text{CN} \rightleftharpoons \text{CH}_3\text{CN} \]

ethyl fluoride \[ \rightleftharpoons \text{HF} + \text{ethene} \]

isopropyl ether \[ \rightleftharpoons \text{propene} + \text{isopropanol} \]

Only one reactant, so how does the reaction proceed? Experiments show that the rate law is 1st order at high concentration and 2nd order at low concentration of the reactant. Would like to explain this.

Lindemann proposed a 2nd order bimolecular collisional activation step.
Look at $A \leftrightarrow B$:

$$A + M \xrightarrow{k_1} A^* + M$$
$$A^* \xrightarrow{k_2} B$$

$A^*$ represents an energized reactant molecule.

$M$ collides with and transfers energy to $A$.

$M$ can be another reactant molecule or a product molecule,
or more likely an inert buffer gas molecule such as $N_2$ or $Ar$.

Rate of product formation is

$$\frac{d[B]}{dt} = k_2[A^*] \quad \text{(*)}$$

Invoke the steady state approach on $A^*$:

$$\frac{d[A^*]}{dt} = 0 = k_1[A][M] - k_{-1}[A^*][M] - k_2[A^*]$$

Solve for $[A^*] = \frac{k_1[M][A]}{k_2 + k_{-1}[M]}$ and sub. in (*)

$$\frac{d[B]}{dt} = -\frac{d[A]}{dt} = \frac{k_1k_2[M][A]}{k_2 + k_{-1}[M]} = \text{keff}[A]$$

where $\text{keff} = \frac{k_1k_2[M]}{k_2 + k_{-1}[M]}$ is pressure-dependent so it's not a proper rate constant.
Consider the limiting cases of high and low pressure.

**High Rate of Collisional Deactivation > Rate of Reaction**

Why? Lindemann postulated that there must be a time lag between the activating collision and the reaction, so at high concentration the \( \Lambda \) species can be deactivated before it has a chance to react.

This is equivalent to assuming a slow conversion to products from the activated state.

We have \( k_1 [M][\Lambda] > k_2 [\Lambda] \Rightarrow k_1 [M] > k_2 \)

\( \Rightarrow k_{eff} = \frac{kk_2 [M]}{k_1 [M]} = \frac{kk_2}{k_1} \quad \text{and the overall rate law is} \quad \frac{d[B]}{dt} = \frac{kk_2 [\Lambda]}{k_1} \quad \text{1st order} \)

**Low Rate of Conversion to Products > Deactivation Rate**

equivalent to assuming slow deactivation

\( k_2 > k_1 [M] \) and \( k_{eff} = \frac{kk_2 [M]}{k_2} = k_1 [M] \)

and the overall rate law is \( \frac{d[B]}{dt} = k_1 [M][\Lambda] \quad \text{2nd order} \)
Lindemann theory breaks down for 2 main reasons:

(i) the bimolecular step takes no account of the energy dependence of activation; the internal degrees of freedom of the molecule are completely neglected, and the theory consequently underestimates the rate of activation.

(ii) the unimolecular step \((A^* \rightarrow B)\) fails to take into account that a unimolecular reaction specifically involves one particular form of molecular motion, e.g., rotation about a double bond for cis-trans isomerization.

These are all fixed in RRKM theory.
Consider \( \text{H}_2(\text{g}) + \text{Br}_2(\text{g}) \rightleftharpoons 2\text{HBr}(\text{g}) \)

with experimentally determined rate law:

\[
\frac{d[\text{HBr}]}{dt} = \frac{k[\text{H}_2][\text{Br}_2]^{\frac{1}{2}}}{1 + k'[\text{HBr}][\text{Br}_2]}
\]

The product decreases the reaction rate as it accumulates. It is thus an inhibitor!

The proposed mechanism, in terms of elementary reactions, is:

\[
\begin{align*}
\text{Br}_2 + \text{M} & \xrightleftharpoons[k_1]{k_{-1}} 2\text{Br} + \text{M} \\
\text{Br} + \text{H}_2 & \xrightarrow{k_2} \text{HBr} + \text{H} \quad k_{-2} = \text{inhibition} \\
\text{H} + \text{Br}_2 & \xrightarrow{k_3} \text{HBr} + \text{Br} \quad k_{-3} = \text{inhibition}
\end{align*}
\]

The propagation steps are chain reactions involving free radicals.

This is also important in polymerization.
To obtain the experimental rate law from this mechanism, we need to eliminate \( M \), \( H \), and \( Br \) from the elementary reactions.

To do this, we apply the steady state approximations to \( H \) and \( Br \), and \( M \) ends up canceling out.

Also, we set \( k_3 = 0 \) because it is known this reaction is very slow.

\[
\frac{d[H]}{dt} = 0 = k_2[Br][H_2] - k_{-2}[HBr][H] - k_3[H][Br_2]
\]

\[
\frac{d[Br]}{dt} = 0 = 2k_1[Br_2][M] - 2k_{-1}[Br]^2[M] - k_2[Br][H_2] + k_{-2}[HBr][H] + k_3[H][Br_2]
\]

Add these: \( 0 = k_1[Br_2][M] - k_{-1}[Br]^2[M] \)

\[
[H] = \left( \frac{k_1}{k_{-1}} \right)^{1/2} [Br_2]^{1/2} = K_{i}^{1/2} [Br_2]^{1/2}
\]

Substitute this into the \( \frac{d[H]}{dt} = 0 \) expression to get

\[
[H] = \frac{k_2[Br][H_2]}{k_{-2}[HBr] + k_3[Br_2]} = \frac{k_2K_{i}^{1/2} [Br_2]^{1/2} [H_2]}{k_{-2}[HBr] + k_3[Br_2]}
\]

Then we use \( \frac{d[HBr]}{dt} = k_2[Br][H_2] - k_{-2}[HBr][H] + k_3[H][Br_2] \)

with the \([H]\) and \([Br]\) steady state expressions to obtain the observed rate law with \( k = k_2K_{i}^{1/2} \); \( k' = k_{-2}/k_3 \).
29-9: Michaelis-Menten Mechanism for Enzyme Catalysis

\[ \frac{E + S}{k_1} \xrightarrow{k_{-1}} ES \xrightarrow{k_2} E + P \]

\( E = \text{enzyme} \); \( S = \text{substrate} \); \( P = \text{product} \); \( ES = \text{enzyme-substrate complex} \)

Rate: \( \frac{d[P]}{dt} = k_2 [ES] \) during the initial stages of the reaction, before \( [P] \) is significant.

Apply the steady state approx to \([ES]\):

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

\[ \Rightarrow [ES] = \frac{k_1 [E][S]}{k_{-1} + k_2} \]

\([E]_o = [E] + [ES] = \text{total amount of } E \text{ added}

\([S]_o = [S] + [ES] \approx [S] = \text{substrate in large excess relative to the enzyme.} \)

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

\[ \Rightarrow [ES] + \frac{k_1 [ES][S]}{k_{-1} + k_2} = \frac{k_1 [E]_o [ES]}{k_{-1} + k_2} \]

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

\[ \Rightarrow [ES] = \frac{[E]_o}{1 + \frac{k_{-1} + k_2}{k_1} [S]} \]
Thus the rate is $k_2[E_S]$

$$\frac{k_2[E_I_0]}{1 + \frac{K_m}{[S]}} = \frac{k_2[E_I][S]}{K_m + [S]}$$

$$K_m = \frac{k_i + k_2}{k_i}$$  Michaelis constant

$$V_{\text{max}} = k_2[E_I_0] = \text{maximum rate}$$

Three special cases:

1. Low $[S]: \frac{K_m}{[S]} \gg 1 \Rightarrow \text{rate} = \frac{V_{\text{max}}}{K_m} \quad [S] < [S]$

2. High $[S]: \frac{K_m}{[S]} < 1 \Rightarrow \text{rate} = V_{\text{max}}$

3. $K_m = [S]: \text{rate} = \frac{V_{\text{max}}}{2}$

- substrate conc. at which the reaction rate is half its maximal value.

$$\frac{1}{\text{rate}} = \frac{1}{V_{\text{max}}} + \frac{K_m}{V_{\text{max}} \cdot [S]}$$

So plot $\frac{1}{\text{rate}}$ vs $\frac{1}{[S]}$, straight line with:

- slope $\frac{K_m}{V_{\text{max}}}$
- y-intercept $\frac{1}{V_{\text{max}}}$
- x-intercept $-\frac{1}{K_m}$

Lineweaver-Burk plot
<table>
<thead>
<tr>
<th>Enzyme</th>
<th>$k_{\text{cat}}/K_M$ (s$^{-1}$M$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetylcholinesterase</td>
<td>$1.6 \times 10^8$</td>
</tr>
<tr>
<td>Carbonic anhydrase</td>
<td>$8.3 \times 10^7$</td>
</tr>
<tr>
<td>Catalase</td>
<td>$4 \times 10^7$</td>
</tr>
<tr>
<td>Crotonase</td>
<td>$2.8 \times 10^8$</td>
</tr>
<tr>
<td>Fumarase</td>
<td>$1.6 \times 10^8$</td>
</tr>
<tr>
<td>Triose phosphate isomerase</td>
<td>$2.4 \times 10^8$</td>
</tr>
<tr>
<td>$\beta$-Lactamase</td>
<td>$1 \times 10^8$</td>
</tr>
<tr>
<td>Superoxide dismutase</td>
<td>$7 \times 10^9$</td>
</tr>
</tbody>
</table>
Table 8.7  Substrate preferences of chymotrypsin

<table>
<thead>
<tr>
<th>Amino acid in ester</th>
<th>Amino acid side chain</th>
<th>$k_{cat}/K_M \text{ (s}^{-1} \text{ M}^{-1})$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glycine</td>
<td>$-\text{H}$</td>
<td>$1.3 \times 10^{-1}$</td>
</tr>
<tr>
<td>Valine</td>
<td>$-\text{CH}$</td>
<td>$2.0$</td>
</tr>
<tr>
<td></td>
<td>$\text{CH}_3$</td>
<td></td>
</tr>
<tr>
<td>Norvaline</td>
<td>$-\text{CH}_2\text{CH}_2\text{CH}_3$</td>
<td>$3.6 \times 10^{2}$</td>
</tr>
<tr>
<td>Norleucine</td>
<td>$-\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_3$</td>
<td>$3.0 \times 10^{3}$</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>$-\text{CH}_2\text{C}_6\text{H}_4\text{CH}_3$</td>
<td>$1.0 \times 10^{5}$</td>
</tr>
<tr>
<td>Enzyme</td>
<td>Turnover number (per second)</td>
<td></td>
</tr>
<tr>
<td>----------------------------</td>
<td>------------------------------</td>
<td></td>
</tr>
<tr>
<td>Carbonic anhydrase</td>
<td>600,000</td>
<td></td>
</tr>
<tr>
<td>3-Ketosteroid isomerase</td>
<td>280,000</td>
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</tr>
<tr>
<td>Acetylcholinesterase</td>
<td>25,000</td>
<td></td>
</tr>
<tr>
<td>Penicillinase</td>
<td>2,000</td>
<td></td>
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<tr>
<td>Lactate dehydrogenase</td>
<td>1,000</td>
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<td>Chymotrypsin</td>
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<td>DNA polymerase I</td>
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<td>Tryptophan synthetase</td>
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<tr>
<td>Lysozyme</td>
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<tr>
<td>Enzyme</td>
<td>Substrate</td>
<td>$K_M$ (μM)</td>
</tr>
<tr>
<td>--------------------------------</td>
<td>----------------------------------</td>
<td>------------</td>
</tr>
<tr>
<td>Chymotrypsin</td>
<td>Acetyl-$L$-tryptophanamide</td>
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</tr>
<tr>
<td>Lysozyme</td>
<td>Hexa-$N$-acetylglucosamine</td>
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<td>β-Galactosidase</td>
<td>Lactose</td>
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<td>Threonine deaminase</td>
<td>Threonine</td>
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<tr>
<td>Carbonic anhydrase</td>
<td>$CO_2$</td>
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<td>Penicillinase</td>
<td>Benzylpenicillin</td>
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<td>Pyruvate carboxylase</td>
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<tr>
<td></td>
<td>$HCO_3^-$</td>
<td>1000</td>
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<td></td>
<td>ATP</td>
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</tr>
<tr>
<td>Arginine-tRNA synthetase</td>
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<td>3</td>
</tr>
<tr>
<td></td>
<td>tRNA</td>
<td>0.4</td>
</tr>
<tr>
<td></td>
<td>ATP</td>
<td>300</td>
</tr>
</tbody>
</table>
A biological catalyst (E = enzyme) lowers the activation energy for a chemical reaction.
Km is the concentration of substrate at which half the active sites are filled. Thus Km provides a measure of the substrate concentration required for significant catalysis to occur.

When Km is known, the fraction of sites filled, \( f_{ES} \), is

\[
 f_{ES} = \frac{[S]}{[S] + K_m}
\]

If we know \( V_{max} \) and \([E]_0\) (concentration of active sites), then we can compute \( k_2 = \frac{V_{max}}{[E]_0} \), the "turnover" number.

\( V_{max} \) is the number of substrate molecules converted into product per unit time when the enzyme is fully saturated with substrate.

Each catalyzed reaction takes place in time \( 1/k_2 \).

However, most enzymes are not normally saturated with substrate. This is a special case; the rate is given by \( V_{max} = \frac{K_m [S]}{K_m + [S]} = \frac{k_2 [E]_0}{K_m} [S] \).

So \( \frac{k_2}{K_m} \) governs the kinetics and gives us a measure of the catalytic efficiency.

\[
 k_2 = \frac{k_2}{K_m} \cdot \frac{k}{k_2+k_1} \quad \text{If} \quad k_2 \gg k_1, \quad k_2 = k_1, \quad \text{and} \quad k_1 = \frac{K_m}{k_2+k_1}
\]

Some enzymes reach this value and are "kinetically perfect."