Elementary reactions

stoichiometry = mechanism (Cl\textsuperscript{−} + H\textsubscript{2} → HCl + H\textsuperscript{·})

monomolecular reactions (decay: N\textsubscript{2}O\textsubscript{3} → 2 NO\textsubscript{2}; radioactive decay; some isomerisations)

\[
\text{ClCH}_3 + \text{CN}^- \rightarrow \text{Cl}^- + \text{CH}_3\text{CN}
\]

trimolecular reactions

O + O\textsubscript{2} + N\textsubscript{2} → O\textsubscript{3} + N\textsubscript{2}
(N\textsubscript{2} carries out the surplus energy)

Reaction mechanisms

A (general) reaction is a sequence of elementary reactions = reaction mechanism.

Example: \(2\text{H}_2 + \text{O}_2 \rightarrow 2\text{H}_2\text{O}\)

\[
\begin{align*}
\text{H}_2 + \text{O}_2 & \rightarrow \text{H}_2\text{O} + \text{H}^- \\
\text{H}_2\text{O} + \text{H}^- \rightarrow \text{H}_2\text{O}_2 + \text{H}^+ \\
\text{H}_2\text{O}_2 + \text{H}_2 & \rightarrow 2\text{H}_2\text{O} + \text{H}^+
\end{align*}
\]

radical A\textsuperscript{·}
activated molecule A\textsuperscript{·} (energy-rich, local energy minimum)
activated complex (transition state) AB\textsuperscript{·}, AB\textsuperscript{·}\textsuperscript{−} (saddle point)

Lindemann(-Hinshelwood) mechanism

\(4\text{H}_2 + \text{Cl}_2 \rightarrow 2\text{HCl}\)

initiation (typically free radicals are produced)
– heat
– chemical (peroxides)
– light (UV)

propagation (cyclic reaction with radical recovery)
– chain transfer (no branching)
– chain branching

termination
– recombination (of radicals)
– reaction (low-reactive radical—inhibition)
– deactivation at walls

Enzyme catalysis: Michaelis–Menten kinetics

Mechanism of Michaelis and Menten (Enzyme, Substrate, Product):

\[
\text{E} + S \rightarrow \text{ES} \rightarrow \text{E} + \text{P}
\]

stationary state (because \(c_E\cdot c_{ES} \ll c_S\)):

\[
dc_{ES} = k_{ES}c_E c_S - (k_{-1} + k_2)c_{ES} = 0
\]

balance: \(c_E + c_{ES} = c_{E0}\)

Eliminating \(c_E\) (\( \rightarrow c_{ES}\)) from \(\frac{dc_{ES}}{dt} = dc_E/dt = -dc_{ES}/dt = k_{1}c_E c_S - k_{-1}c_{ES}\)

\[
dc_{ES}/dt = k_{2}c_{ES} - k_{1}c_{ES}/c_{S} + 1 = \frac{v_{\text{max}}}{K_M + c_{S}}
\]

Michaelis–Menten kinetics II

Experimentally available: \(K_M\) and \(v_{\text{max}} = k_{2}c_{E0}\)

Integrated form

\[
\frac{dc_{S}}{dt} = -\frac{v_{\text{max}}}{K_M + c_{S}} + 1
\]

cannot solve for \(c_S(t)\) (using elem. functions) \(\rightarrow\) numerical solution
**Metabolism of alcohols**

Alcohol dehydrogenase, various types human:

In liver and the lining of the stomach

Further oxidation to acids and H2O + CO2

CH3CH2OH → CH3 CHO + Hangover
CH2OH CH2OH → (COOH)2 (kidney stones)
CH3OH → HCHO + H2O

**Example.** Calculate the time needed to metabolize cS = 1 wt. % of ethanol to cS = 0.1 % Data: vmax = 0.12 g L−1 h−1, K M = 0.06 g L−1.

Pblood = 1.06 g cm−3 → 1 wt. % = 1.06 g L−1

0th order: τ = cS − cS0 = \( \frac{(1 - 0.1) \times 1.06}{0.12} \) h = 7.7 h

More accurate: τ = \( \frac{K_M \ln \left( \frac{cS_0}{cS} \right)}{v_{max}} \) = 9.2 h

**Example—carbonic anhydrase**

**Michaelis–Menten kinetics III**

Rate:

\[ v = \frac{-dcS}{dT} = \frac{v_{max}}{K_M/cS + 1} \]

**Example—carbonic anhydrase**

\[ CO_2 + H_2O \rightarrow HCO_3^- + H^+ \]

<table>
<thead>
<tr>
<th>[CO2]/mmol dm−3</th>
<th>v/mol dm−3 s−1</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.25</td>
<td>2.78 × 10−5</td>
</tr>
<tr>
<td>2.5</td>
<td>5.00 × 10−5</td>
</tr>
<tr>
<td>5.0</td>
<td>8.33 × 10−5</td>
</tr>
<tr>
<td>20.0</td>
<td>16.7 × 10−5</td>
</tr>
</tbody>
</table>

1 s = 1000 ms

[according to DeVoe, Kistiakowski, JACS 83, 274 (1961)]

**Inhibition**

- reversible
- irreversible

**reversible inhibition:** the inhibitor is bound non-covalently (H-bonds, etc.), decreases the turnover

\[ E + S \xrightleftharpoons[k_{-1}^i]{k_i} ES \xrightleftharpoons[k_i]{k_{-i}} E + P \]

\[ + \quad + \]

\[ k_i \uparrow k_{-i} \quad k_i \uparrow k_{-i} \]

EI + S \xrightleftharpoons[k_i^j]{k_{-i}^j} ESI \xrightleftharpoons[k_i^j]{k_{-i}^j} E + P

**irreversible inhibition:** “catalyst poisoning”, usually covalently bound → inactive complex EI

**Competitive reversible inhibition**

\[ E + S \xrightleftharpoons[k_{-1}^i]{k_i} ES \xrightleftharpoons[k_i]{k_{-i}} E + P \]

\[ + \quad + \]

\[ k_i \uparrow k_{-i} \quad k_i \uparrow k_{-i} \]

EI

The inhibitor binds to the same site as the substrate ("competes" with the substrate)

**Uncompetitive reversible inhibition**

\[ E + S \xrightleftharpoons[k_{-1}^i]{k_i} ES \xrightleftharpoons[k_i]{k_{-i}} E + P \]

\[ + \quad + \]

\[ k_i \uparrow k_{-i} \quad k_i \uparrow k_{-i} \]

ESI \xrightleftharpoons[k_i^j]{k_{-i}^j} E + P

The inhibitor binds to the enzyme-substrate complex Also anti-competitive often partial (slows down the reaction)

**Mixed (non-competitive) reversible inhibition**

\[ E + S \xrightleftharpoons[k_{-1}^i]{k_i} ES \xrightleftharpoons[k_i]{k_{-i}} E + P \]

\[ + \quad + \]

\[ k_i \uparrow k_{-i} \quad k_i \uparrow k_{-i} \]

ESI \xrightleftharpoons[k_i^j]{k_{-i}^j} E + P

Mixed inhibition: the inhibitor bound both to E and ES (Pure) non-competitive inhibition: inhibitor affects a different part of the enzyme, k_i = k_i^j, k_{-i} = k_{-i}^j

**Mixed units**

- **Enzyme activity unit (amount of substance / time)**
  - SI: mol s−1 (katal)
  - more common: µmol/min ("enzyme unit", U)
- **Specific activity** (per kg of enzyme)
  - SI: mol s−1 kg−1
  - µmol min−1 mg−1
- **Turnover number** (per mole)
  - SI: mol s−1 mol−1 = s−1
  - often min−1 etc.
- **Molar mass** g mol−1 = Da (dalton) or 1 g mol−1/N A = \( \frac{1}{6.02 \times 10^{23}} \) = 1 u – 1.660539 × 10−27 kg = 1 Da

**Example:** 1 µg of enzyme (M = 40 kDa) in the excess of substrate provides the reaction rate of 6 µmol of substrate/min. What is the turnover number (in s−1)?
Reversible inhibition: some math

\[ E + S \xrightleftharpoons[k_{-1}]{k_i} ES \xrightarrow{k_2} E + P \]
\[ I \xrightarrow{k_i} EI \xrightarrow{k_{i-1}} E + I \]
\[ \frac{d[ES]}{dt} = k_i c_E c_S - (k_{-1} + k_2)c_ES - k_i' c_{ES} + k_{i-1}' c_{EI} = 0 \]

Stationary state:
\[ \frac{d[ES]}{dt} = k_i c_E c_S - (k_{-1} + k_2)c_ES - k_i' c_{ES} + k_{i-1}' c_{EI} = 0 \]

Pre-equilibrium:
\[ c_{ES} = k_i c_E c_S, \quad c_{EI} = k_{i-1}' c_{EI} \]

Balance: \( c_E + c_{ES} + c_{EI} + c_{ESI} = c_{E0} \)
we assume \( c_I \gg c_E \), \( \Rightarrow c_I \approx c_{I0} \) is known (no balance of I needed)

Reversible inhibition: Lineweaver–Burk

\[ \frac{1}{v} = \frac{aKM}{vmax} \frac{1}{c_S} + \frac{a'}{vmax} \]

Inhibitor: \( \alpha = 1 + \frac{k_i}{k_{-1}} c_I \), \( \alpha' = 1 + \frac{k_i'}{k_{i-1}} c_I \)

Stationary state: \( \frac{d[ES]}{dt} = k_i c_E c_S - (k_{-1} + k_2)c_ES - k_i' c_{ES} + k_{i-1}' c_{EI} = 0 \)

Pre-equilibrium: \( c_{ES} = k_i c_E c_S, \quad c_{EI} = k_{i-1}' c_{EI} \)

Balance: \( c_E + c_{ES} + c_{EI} + c_{ESI} = c_{E0} \)

we assume \( c_I \gg c_E \), \( \Rightarrow c_I \approx c_{I0} \) is known (no balance of I needed)

Reversible inhibition: Lineweaver–Burk

Time vs. substrate conc. Lineweaver–Burk

Chain reactions: \( \Phi > 1 \)

Example: How much HI decomposes by absorbing energy of 100 J in the form of light of wave length 254 nm?

\[ \Phi = \frac{\text{# of photons absorbed}}{\text{# of molecules transformed/decomposed/...}} \]

\[ \Phi > 1 \]

Example: How much HI decomposes by absorbing energy of 100 J in the form of light of wave length 254 nm?

0.42 mmol

Reversible inhibition – summary

Competitive: \( \alpha > 1 \)

 inhibitor binds to the free enzyme in the L-B diagram:
 greater \( K_M \) the same \( v_{max} \)

Uncompetitive: \( \alpha' > 1 \)

 inhibitor binds to the enzyme-substrate complex in the L-B diagram:
 smaller \( K_M \) smaller \( v_{max} \)

Mixed (non-competitive): \( \alpha, \alpha' > 1 \)

 inhibitor binds to both the free enzyme and enzyme-substrate complex in the L-B diagram:
 the same \( K_M \) smaller \( v_{max} \)