Lecture 8 Rate Equations (ch 15)

1. Turnover rate in propulsion (p577).

slides We saw how actin polymerization drives a cell across a surface:

i) Listeria moves $0.2\,\mu m/s = 200\,nm/s$

G-Actin monomer is 2.7 nm, elongation of chain

$\Rightarrow$ rate is $\frac{200}{2.7} \simeq 70$ monomers/s.

ii) EColi flagellum. 100 Hz rotation

~15 'poles' of motor protein

~1500 reaction steps/s.

slide 3.1 iii) Other rates (p88)

lysozyme turnover	0.5/s ~ 1 sec
carbonic anhydrase	600,000/s = 1 μs
side chain rotation	500 ps. = 2×10^9/s
H-bond proton relay in H_2O	10 ps
covalent bond vibration	10 fs.

2. Rate equation paradigm (p580).

Molecules in solution, concentration $c_j = [j]$, $j = 1 \ldots n$

$$\frac{dc_j(t)}{dt} = f(c_1 \cdots c_n, \underbrace{k_1 \cdots k_m}_{\text{rate constants}})$$

Rate $\propto$ concentrations (in dilute limit) of all species involved

Rate $\propto$ rate constant.

Chemistry notation:

1 species → 1st order reaction

2 " 2nd " " etc

$$\frac{dc_j(t)}{dt} = k\,[c_1]^{n_1}\,[c_2]^{n_2} \cdots$$

n_1: order wrt species 1 etc

units of k follow accordingly

3. First-order reactions (example on HW)

$$\frac{dc}{dt} = -kc \qquad c = c_0 e^{-kt} = c_0 e^{-t/\tau} \qquad \tau = 1/k$$

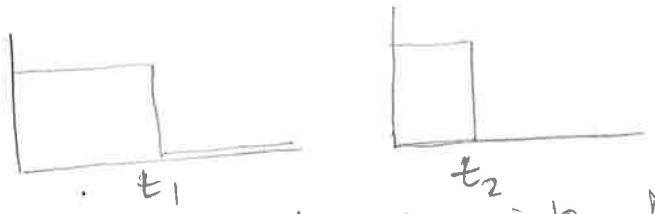
[could increase or decrease : bdy conditions]

exponential decay

Statistical mechanical picture of decay:

"Waiting time" distribution: state of i'th molecule

trajectory $\sigma_i(t) = \begin{cases} 1 & t < t_i \text{ before reacting} \\ 0 & \text{after} \end{cases}$



etc

Divide time domain into N steps of Δt

$t = N\Delta t$

Probability of decay within Δt is $k\Delta t$

Probability to survive N steps then decay is

$$p(t)\Delta t = (1 - k\Delta t)\ldots(1 - k\Delta t)\cdot k\Delta t$$
$$= (1 - k\Delta t)^N \cdot k\Delta t \simeq e^{-k\Delta t N} \cdot k\Delta t$$

$$p(t) = k e^{-kt} = \frac{1}{\tau} e^{-t/\tau}$$ same result as above.

Waiting time distribution

Example; retinal cis $\rightleftharpoons$ trans { isomerisation = like decay

associated with absorption/emission photon

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Central role in:

i) photosynthetic bacteria (purple membrane)

ii) vision.

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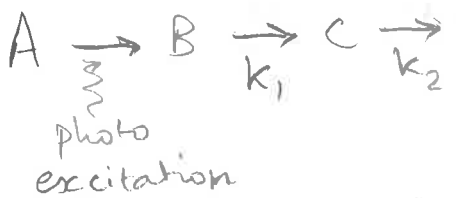
Bacteriorhodopsin. Photon → cis-trans isomerisation → electron transport across memb. This is used to drive ATP synthesis.

BR rate equations are all 1st order, involving a single species, but form a chain



Reaction rates are 'nested' with later ones orders of magnitude slower than the early ones. Result is complete conversion.
States are identified spectroscopically by the wavelength of peak adsorption.

$$E = h\nu = \hbar\omega = hc/\lambda$$

4. Photosynthesis. (p747, ch18)

movie slides

Similar story of photosynthetic reaction centre (PRC)
As in mitochondria, the chloroplasts drive a proton gradient across the membrane, which then runs an ATP synthase.

The multiple states of the reaction are all connected by electron transfers between different {pigments / chromophores} – coloured molecules that have optically excited charged states.

Reaction rates are governed by quantum mechanical overlap of these molecules. The electron transfers through a set of overlapping molecules. Fermi's "Golden Rule"

~100 chlorophyll molecules form a large antenna that funnels the electron to the PRC. eventually onto plastocyanin which moves from PSII over to PSI.

9.3.15

5. Bimolecular reaction.
Michaelis-Menten kinetics.

$$A + B \underset{k_-}{\overset{k_+}{\rightleftharpoons}} AB.$$ "on"

Forward rate = $k_+[A][B]$ bimolecular
Reverse rate = $k_-[AB]$ "off"

Equilibrium when rates are equal.

$$K_d = \frac{[A][B]}{[AB]} = \frac{k_-}{k_+} = \frac{k_{off}}{k_{on}}$$ [law of mass action

We met this before and saw that fraction of one of the species "bound" =

$$P_{B\text{-bound}} = \frac{[AB]}{[AB]+[B]} = \frac{[A]/K_d}{[A]/K_d + 1}$$

If we now consider the enzyme-substrate reaction rate:
E S

$$E + S \underset{k_-}{\overset{k_+}{\rightleftharpoons}} ES \xrightarrow{r} E + P.$$ ← product

$$\frac{d[E]}{dt} = -k_+[E][S] + k_-[ES] + r[ES]$$

$$\frac{d[S]}{dt} = -k_+[E][S] + k_-[ES]$$

$$\frac{d[ES]}{dt} = +k_+[E][S] - k_-[ES] - r[ES]$$

$$\frac{d[P]}{dt} = r[ES]$$

Want the steady state reaction rate generated by an excess of S and (slow) generation of P.

Define <u>steady state</u> as $\frac{d[ES]}{dt} = 0$

$$\Rightarrow \frac{[E][S]}{[ES]} = \frac{k_- + r}{k_+} = K_m \text{ another Em const.}$$

M-M result uses the maximum velocity of the reaction, generating product:

$$V_{max} = r[E_{tot}] = r([E] + [ES])$$

$$\Rightarrow \frac{d[P]}{dt} = r[ES] = r\frac{[E][S]}{K_m} = V_{max}\frac{[E][S]}{K_M([E]+[ES])}$$

$$= V_{max}\frac{[E][S]}{K_m[E] + [E][S]} = V_{max}\frac{[S]/K_m}{1 + [S]/K_m}$$

The formula is a titration curve just like that for P_{bound}:

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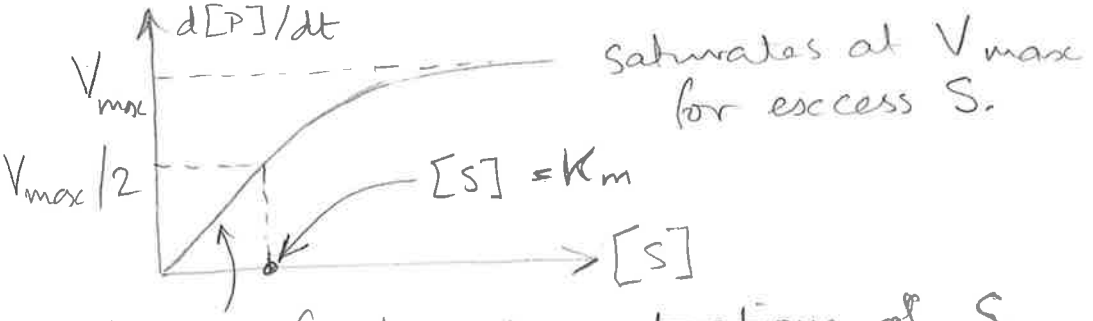

Convenient parameterisation of data in terms of K_m = conc at half rate max
V_{max} = maximum turnover

Assumption of steady state ignored the early time transients of the system, which we look at next.

6. Approach to steady state

$$A \underset{k_-}{\overset{k_+}{\rightleftharpoons}} B \xrightarrow{r} C$$

slightly simpler case. Omitting the [] for conc:

$$\frac{dA}{dt} = -k_+ A + k_- B$$

$$\frac{dB}{dt} = k_+ A - (k_- + r) B$$

$$\frac{dC}{dt} = rB$$

remove one variable and noting that only the first two equations are coupled:

$$\frac{d}{d\tau}\begin{pmatrix} A \\ B \end{pmatrix} = \begin{pmatrix} -k & 1 \\ k & -1-\varepsilon \end{pmatrix}\begin{pmatrix} A \\ B \end{pmatrix}$$

where $\tau = k_- t$ dimensionless time

$k = k_+/k_-$ $\varepsilon = r/k_-$

eigenvalue equation with solution:

$$\begin{pmatrix} A \\ B \end{pmatrix} = a_1 \begin{pmatrix} A_1 \\ B_1 \end{pmatrix} e^{\omega_1 \tau} + a_2 \begin{pmatrix} A_2 \\ B_2 \end{pmatrix} e^{\omega_2 \tau}$$

$$\begin{vmatrix} -k-\omega & 1 \\ k & -1-\varepsilon-\omega \end{vmatrix} = (k+\omega)(1+\varepsilon+\omega) - k = 0$$

$$\omega^2 + (1+\varepsilon+k)\omega + (k + k\varepsilon - k) = 0$$

$$\omega_{1,2} = -\frac{1}{2}(1+\varepsilon+k) \pm \frac{1}{2}\sqrt{(1+\varepsilon+k)^2 - 4k\varepsilon}$$

both roots are negative as expected for $e^{\omega\tau}$

Interesting limit of slow generation of product, also relevant to M-M: $\boxed{\varepsilon \ll 1}$

$$\sqrt{\quad} = \sqrt{(1+k)^2 - 4\varepsilon k}$$

$$\simeq (1+k)\left(1 - \frac{1}{2}\frac{4\varepsilon k}{(1+k)^2}\right) = (1+k) - \frac{2\varepsilon k}{1+k}$$

$$\omega_2 \simeq -(1+k) \qquad \omega_1 \simeq -\varepsilon\frac{k}{1+k}$$

fast slow.

If, in addition $\boxed{k \simeq 1}$ for equal interconversion of A and B: $k_+ \simeq k_-$, then eigenvectors

$$\begin{pmatrix} 1 & 1 \\ 1 & 1 \end{pmatrix}\begin{pmatrix} A_2 \\ B_2 \end{pmatrix} = 0 \rightarrow \begin{pmatrix} 1 \\ -1 \end{pmatrix} \text{ fast } \begin{matrix} A\downarrow \\ B\uparrow \end{matrix}$$

$$\begin{pmatrix} -1 & 1 \\ 1 & -1 \end{pmatrix}\begin{pmatrix} A_1 \\ B_1 \end{pmatrix} = 0 \rightarrow \begin{pmatrix} 1 \\ 1 \end{pmatrix} \text{ slow. } A, B\downarrow$$

Start with $B = C = 0$ $\quad A = A_0$ initial conc.

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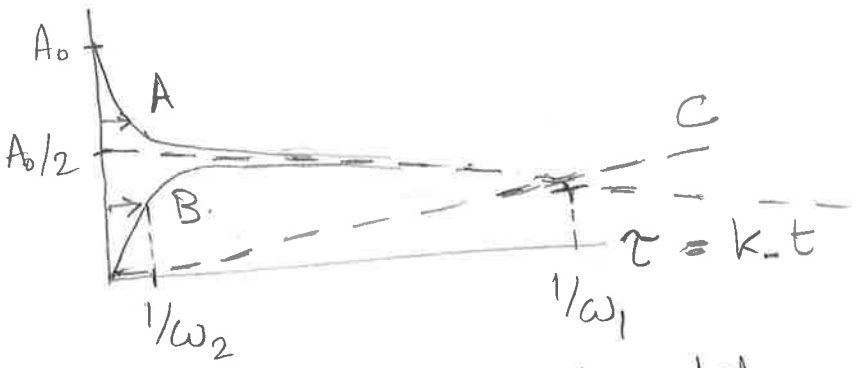
$k = 1$
$\varepsilon = 0.01$



$C = r\int B\,dt$ has small initial delay, then steady rise, then saturation as B runs out.

i) Separation of time scales
fast equilibration of A & B.
slow conversion of A, B → C
} allowed when rates very different

ii) Application to M-M when we assumed

$$\frac{d[ES]}{dt} = 0 \Rightarrow \text{initial equilibration is over.}$$

ok except for transient

7. Actin polymerization

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Important roles of actin and microtubules in powering mitosis (cell division)

i) microtubules connect centromeres to the spindle poles. Motor action pulls the sets of chromosomes apart.

movie

ii) once complete, an actin waist is formed which constricts off the membranes of the two daughter cells.

Experiments show conc. dependent polymerization rates for actin, 10× faster on the "barbed" end than the "pointed" end.

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Pointed end binds ATP-actin more slow
Barbed end binds ADP-actin more fast

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4-levels of heirarchy of models, later ones considering 2× ends + ATP/ADP roles.

i) equilibrium models don't work:
all-or-nothing behaviour like a phase transition.

ii) good models have equilibrium with a population of monomers, c_0, which gets depleted:

Assume M initial nuclei in volume V. FIXED

$$\frac{dn}{dt} = k_{on}\left(c_0 - \frac{M n(t)}{V}\right) - k_{off}$$

conc. tied up in polymers

Fixed conc. of polymers is omitted because it is constant.

$\frac{M n(t)}{V}$ is the concentration of monomers already polymerised, reducing the available concentration of monomers, C_0, for further reaction.

i) Steady state solution when $dn/dt = 0$

$$k_{on}\frac{M n(t)}{V} = k_{on}C_0 - k_{off} \Rightarrow n(t) = \frac{V}{M k_{on}}(k_{on}C_0 - k_{off})$$

ii) Transient solution to reach that state

$$n \sim n_0 e^{-t/\tau} \quad \tau = V/k_{on}M$$

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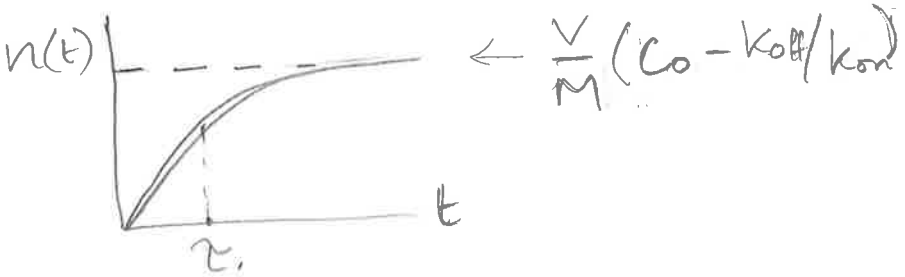

8. Heirarchy of models

Extend to the cases of pointed / barbed ends and ADP + ATP bound monomers.

Let $c = C_0 - \frac{M}{V}n$ be the actual local concentration of monomers. Rate equation now

$$\frac{dn}{dt} = k_{on}c - k_{off}$$

$$= 0 \text{ when } c = c^* = k_{off}/k_{on}$$

Length of polymer = no. monomers × spacing a.

$$\frac{d\langle L\rangle}{dt} = (k_{on}c - k_{off})a.$$

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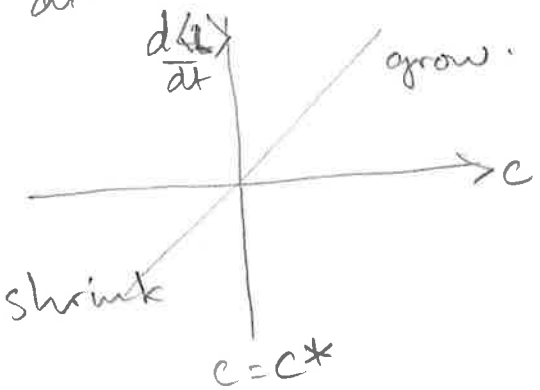

i) Consider different k_{on} and k_{off} for the ends

$$\frac{dn_+}{dt} = k_{on}^{+} c - k_{off}^{+} \quad \text{one end}$$

$$\frac{dn_-}{dt} = k_{on}^{-} c - k_{off}^{-} \quad \text{other end.}$$

But the ratios have to be the same because the monomers are the same

$$\frac{k_{off}}{k_{on}} = \frac{1}{V} e^{\Delta G / k_B T}$$ is the same for both

(ΔG vs concentration) idea from before

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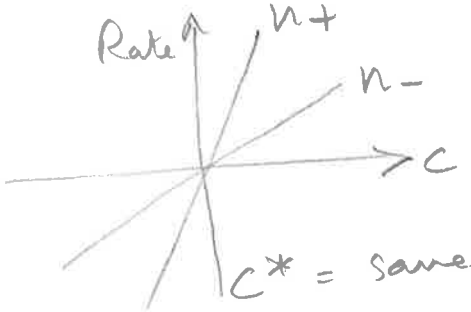

→ both ends shrink or grow together

ii) Different monomers at each end

Barbed ends — only ATP-monomers

Pointed ends — only ADP-monomers

ΔG no longer the same.

"Ideal" treadmilling occurs when

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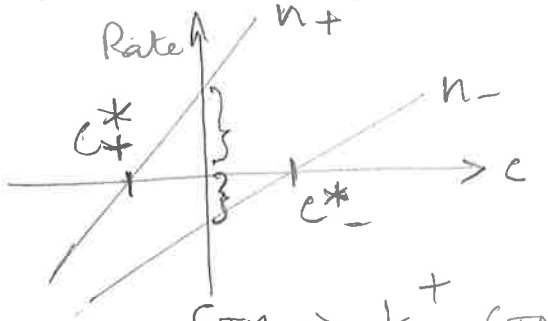

$$\frac{dn^+(c_{TM})}{dt} = -\frac{dn^-(c_{TM})}{dt}$$

$$\Rightarrow k_{on}^{+} c_{TM} - k_{off}^{+} = -\left(k_{on}^{-} c_{TM} - k_{off}^{-}\right)$$

$$\Rightarrow c_{TM} = \frac{k_{off}^{-} + k_{off}^{+}}{k_{on}^{-} + k_{on}^{+}}$$