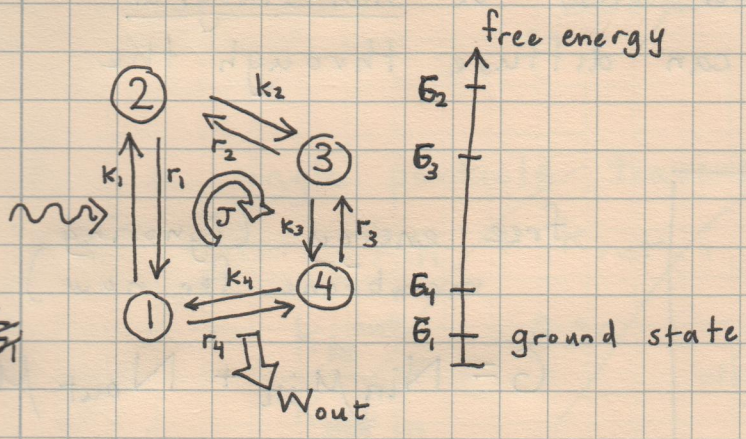
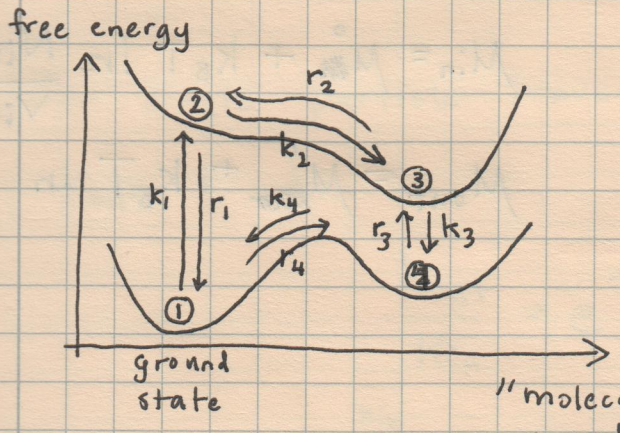
Dissecting a simplified cycle from a light sensitive protein





(schematic representation)

$$\frac{k_1}{r_1} = \frac{k_1^\circ + \alpha}{r_1^\circ + \gamma}$$

α = rate of absorbing photon $\propto$ incident flux

γ = rate of emitting photon (might also be dependent on incoming light b/c stimulated emission)

$$= \frac{k_1^\circ}{r_1^\circ}\,\frac{\left(1+\frac{\alpha}{k_1^\circ}\right)}{\left(1+\frac{\gamma}{r_1^\circ}\right)}$$

$$\equiv e^{-\beta(G_2-G_1)}\, e^{\beta W_{in}}$$

r_1 = spontaneous fluorescence back to ground $\sim$ (ps - ns timescales)$^{-1}$

k_2 = conformational rearrangement (must be fast to compete with r_1)

$\frac{k_2}{r_1} \sim$ "quantum yield" = 0.1 - 0.7 in most photoproteins

k_3 = decay due to phonon/photon emission (often IR) $\sim$ (ps - ns timescales)

state 4 = long-lived intermediate, can be harnessed to function (i.e. work from 4 → 1 transition)

$\Rightarrow$ have verified with frog vision experiments at different temperatures

$r_4 \approx$ spontaneous thermal excitation to "signaling" or "work" state 4 from 1, w/o absorbing a photon

$\Rightarrow$ represents an <u>error</u> in vision

depends on difference $G_4 - G_1$;

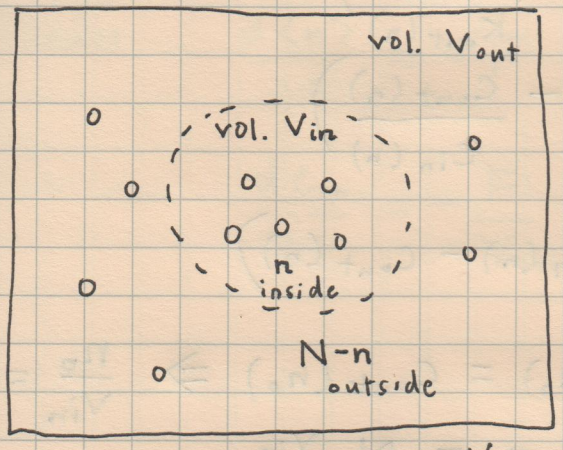
in equilibrium (no photons)

$$P_1^s = \frac{e^{-\beta G_1}}{Z} \qquad P_4^s = \frac{e^{-\beta G_4}}{Z}$$

$$\Rightarrow \frac{P_4^s}{P_1^s} = e^{-\beta(G_4-G_1)}$$

Energetics of transport across a membrane

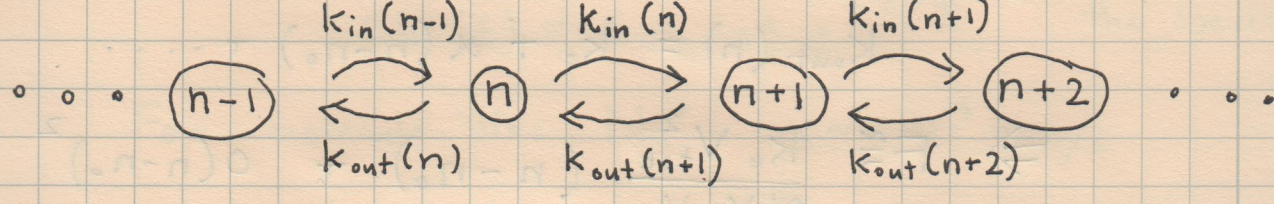
i) Consider first a non-charged chemical species that can diffuse through the membrane.



total $V_{tot} = V_{in} + V_{out}$

For simplicity, assume first V_{in}, V_{out} fixed, but n can change by diffusion thru membrane.

state model:



MR: $$\frac{k_{out}(n)}{k_{in}(n-1)} = e^{-\beta(\mu_{out}(n) - \mu_{in}(n))}$$

$$\mu_{out}(n) = \mu_0 + k_B T \ln \underbrace{\frac{N-n}{V_{out}}}_{C_{out}(n)}$$

$$\mu_{in}(n) = \mu_0 + k_B T \ln \underbrace{\frac{n}{V_{in}}}_{C_{in}(n)}$$

$$\mu_{out} - \mu_{in} = k_B T \ln \frac{C_{out}}{C_{in}}$$

driving a particle against a ten-fold conc. diff. betw. in and out $\Rightarrow 2.3\, k_B T$

$$\Rightarrow \frac{k_{out}(n)}{k_{in}(n-1)} = \frac{C_{in}(n)}{C_{out}(n)}$$

or $$\frac{k_{out}(n)}{k_{in}(n)} \approx \frac{C_{in}(n)}{C_{out}(n)} \quad \text{if } n \gg 1$$

since $k_{in}(n-1) \approx k_{in}(n)$

If we are in state n, net ~~flux~~ rate of particles leaving the cell:

$$F = k_{out}(n) - k_{in}(n)$$

$$= k_{out}(n)\left(1 - \frac{k_{in}(n)}{k_{out}(n)}\right)$$

$$= k_{out}(n)\left(1 - \frac{c_{out}(n)}{c_{in}(n)}\right)$$

$$= \frac{k_{out}(n)}{c_{in}(n)}\left(c_{in}(n) - c_{out}(n)\right)$$

Balance occurs when $c_{in}(n_0) = c_{out}(n_0) \Rightarrow \frac{n_0}{V_{in}} = \frac{N - n_0}{V_{out}}$

at value $n_0 = N\frac{V_{in}}{V_{tot}}$

Taylor expanded near n_0,

$$k_{out}(n) = k_0 + \alpha(n - n_0) + \cdots$$

$$\Rightarrow F \cong \frac{k_0 V_{tot}^2}{N V_{in} V_{out}}(n - n_0) + O(n - n_0)^2$$

$$= \frac{k_0}{c_0}\left(c_{in}(n) - c_{out}(n)\right) + \cdots$$

where $c_0 = \frac{N}{V_{tot}}$

$$\text{flux} = \frac{\text{rate}}{\text{~~unit~~ area of cell surface}} = \frac{F}{A} = \underbrace{\frac{k_0}{c_0 A}}_{\text{units: } \frac{s^{-1}}{nm^{-3}\,nm^2} = \frac{nm}{s}}\left(c_{in}(n) - c_{out}(n)\right)$$

$\frac{k_0}{c_0 A} \equiv P$ permeability of membrane to the species (can be small but never exactly zero)

If $V_{out} \to \infty$, $c_{out} = 0$ (infinitely dilute external liquid)

then $n_0 = 0$

rate at which c_{in} decreases:

estimate $$\frac{d\langle n \rangle}{dt} \approx -F = \rho A\, c_{in}(n)$$

$$\frac{dc_{in}}{dt} = -\frac{F}{V_{in}} = -\underbrace{\frac{\rho A}{V_{in}}}\, c_{in}$$

$$= \rho \frac{4\pi R^2}{\frac{4}{3}\pi R^3} \quad \text{for sphere}$$

$$= \frac{3\rho}{R} \equiv \tau^{-1} \quad \text{inverse timescale to leak the species out}$$

$$c_{in}(t) = c_{in}(0)\, e^{-t/\tau}$$

For molecule like Na^+, $\rho \sim 10^{-5}$ nm/s

typical bacteria $R \sim 1000$ nm

$\Rightarrow \tau \sim 3 \times 10^7$ s > 1 yr

effectively impermeable

but for H_2O or NH_3, $\rho \sim 10^5$ nm/s

$\Rightarrow \tau \sim 3 \times 10^{-3}$ s

essentially completely permeable

Now imagine the case where membrane is essentially impermeable to a molecule, but V_{in} can change (water can go in + out); assume no cost to stretching membrane for now

total free energy $G = n\mu_{in}(n) + (N-n)\mu_{out}(n)$

where n is fixed

$$G = n\mu_0 + (N-n)\mu_0 + k_B T\left[n \ln \frac{n}{V_{in}} + (N-n)\ln \frac{N-n}{V_{out}}\right]$$

($V_{out} = V_{tot} - V_{in}$)

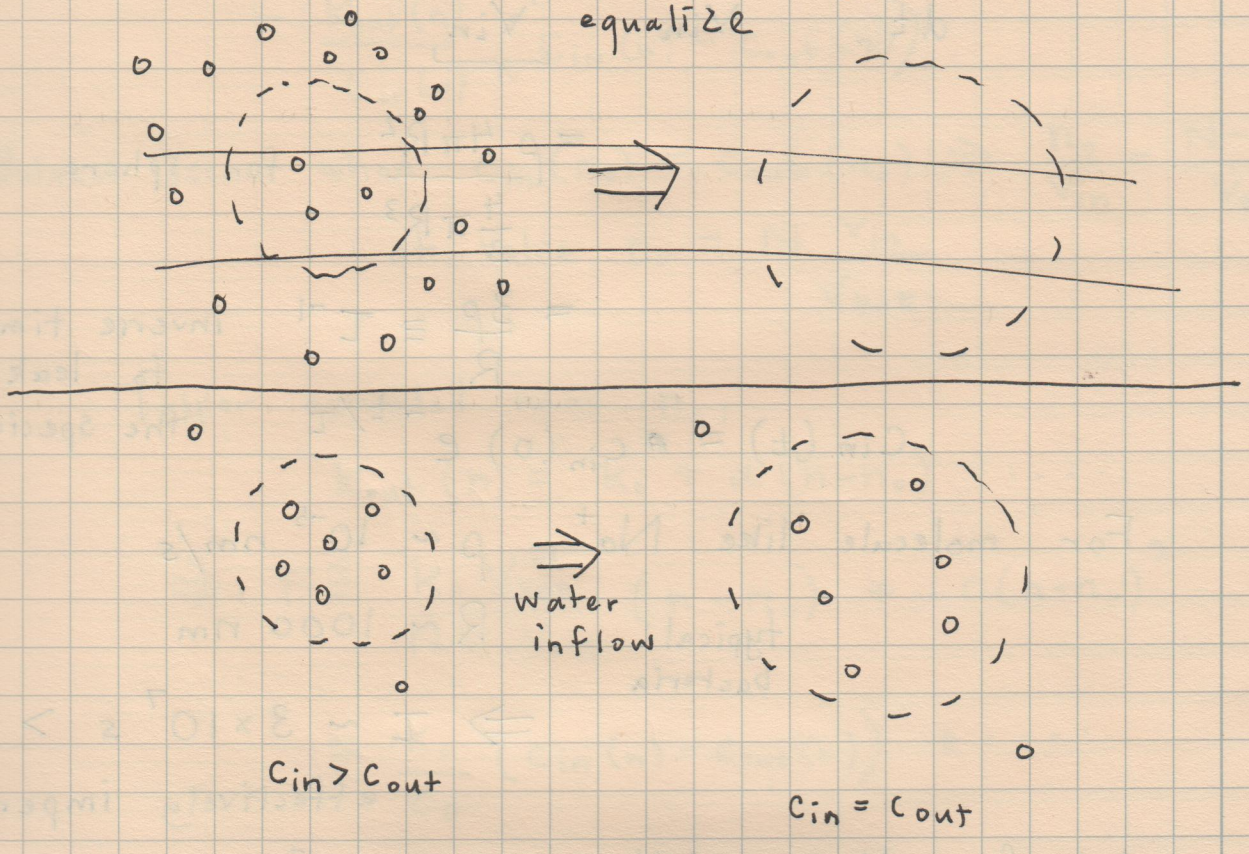
as system goes to equilibrium, $G \to$ minimum

$$\frac{\partial G}{\partial V_{in}} = k_B T\left[-\frac{n}{V_{in}} + \frac{N-n}{V_{tot}-V_{in}}\right] = 0$$

($\frac{n}{V_{in}} = c_{in}$, $\frac{N-n}{V_{tot}-V_{in}} = c_{out}$)

at minimum $c_{in} = c_{out}$

$\Rightarrow$ V_{in} will change until concentrations equalize



This flow of water, expanding or contracting membrane leads to osmotic pressure or turgor pressure. You can counteract it by pumping, or by rigid external cell wall to keep cell from expanding (i.e. plants, bacteria).

$\Rightarrow$ It keeps plants upright (wilting is the lack of this pressure).

Easily generalized to many species (all impermeable):

$$G = \sum_i \left(n_i \mu_{i,in}^{***} + (N_i - n_i)\mu_{i,out} \right)$$

$$= \sum_i \left[N_i \mu_0^{(i)} + k_B T \left[n_i \ln \frac{n_i}{V_{in}} + (N_i - n_i) \ln \frac{N_i - n_i}{V_{out}} \right] \right]$$

$$\frac{\partial G}{\partial V_{in}} = 0 = k_B T \left[\sum_i \left(-\frac{n_i}{V_{in}} + \frac{(N_i - n_i)}{V_{out}} \right) \right]$$

$$\text{so} \quad \sum_i c_{i,in} = \sum_i c_{i,out} \quad \text{in equilibrium}$$

A cell like yeast can balance osmotic shock of extra salt outside by producing glycerol inside, so long as concentrations in total balance.

⇒ For earliest life, particle transporters across membrane necessary "valves" to relieve osmotic pressure, suggesting possible development first in some non-biological "frame", like pores of rock in hydrothermal vent (we will return to this idea later).