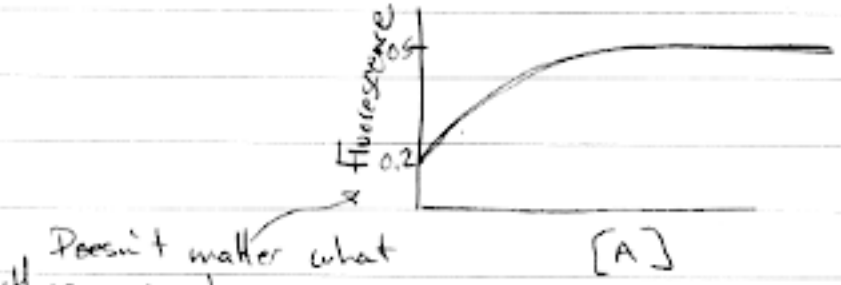
STOPPED HERE

Note that $\frac{\nu/N}{(1-\nu/N)} = K[A]$ (re-arrange first equation on previous page)

$$\frac{f}{1-f} = K[A]$$ f = fraction of (all) sites bound

f is often more readily measured.

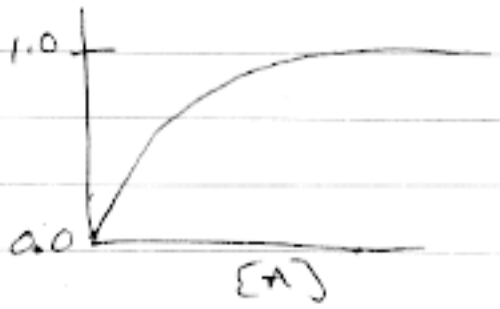
Example: If ligand binding results in the change in fluorescence of a Trp near the binding site, then



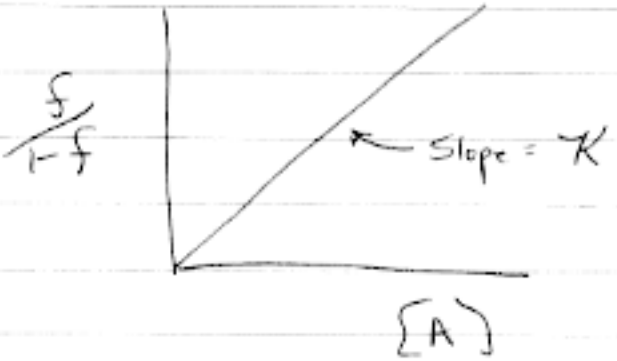
Doesn't matter what these numbers are

Renormalize to

Now that's $\frac{\nu}{N} = f$



Plot



OR BETTER YET, FIT DIRECTLY

IMPORTANT

If cooperativity, then empirically,

change to $\frac{f}{1-f} = K[A]^n$

"all-or-none"

n = Hill Coefficient ranges from 1 (no cooperativity) to N (infinitely cooperative)

N = # sites

K = a constant, but not K for 1 ligand

$$\underbrace{\log\left(\frac{f}{1-f}\right)}_{y} = \underbrace{n}_{m}\log\underbrace{[A]}_{x} + \underbrace{\log K}_{b}$$

Straight line plot ↗

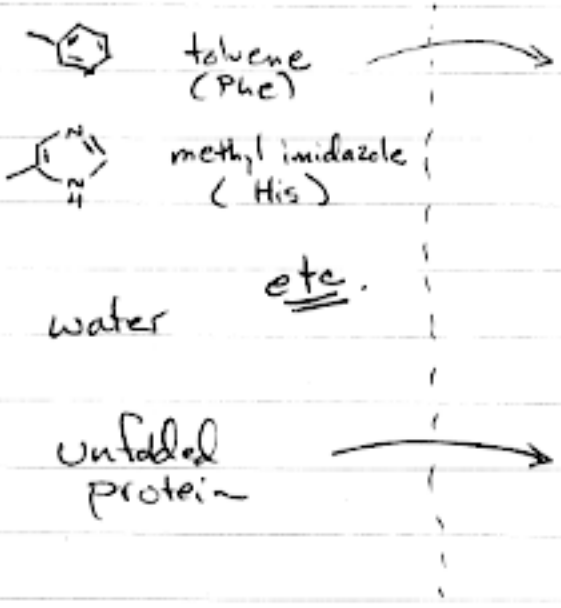
Free Energy of Transfer Between Phases

$\mu_A(s) = \mu_A(aq, sat.)$

$\Delta G = 0$

If we removed some sucrose, a bit of the solid would dissolve
If we added sucrose, a bit of the soln would precipitate

Relevance to protein folding



FROM SUCH MEASUREMENTS, AROSE HYDROPATHY INDEX

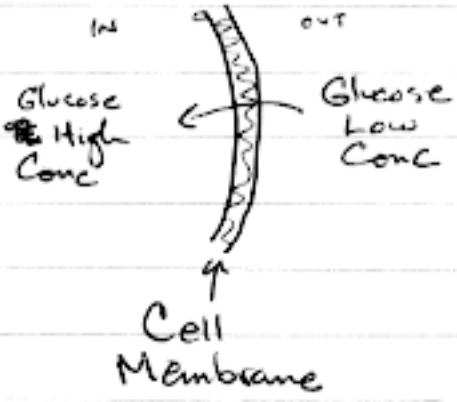
(HYDRO) (PATHOS)
water hate

His ⇒ −3.2 (Polar)
Phe ⇒ +2.8 (Hydrophobic)

Also transfer of a solute from one solvent to the same solvent, but different conc's.

Q:
A: Unfavorable as written

e.g.

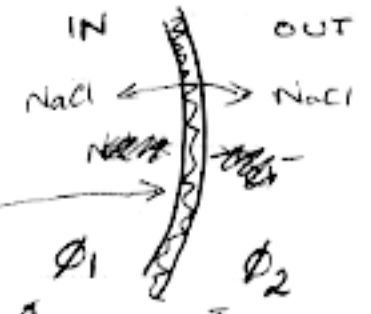


IF $C_A^{IN} > C_A^{OUT}$

$$\Delta\mu = \mu_A^\circ + RT \ln a_A^{IN} - \left(\mu_A^\circ + RT \ln a_A^{OUT}\right) = RT \ln \frac{C_A^{IN}}{C_A^{OUT}} > 0$$

Complication: Charged molecule / membrane potential

New TERM.

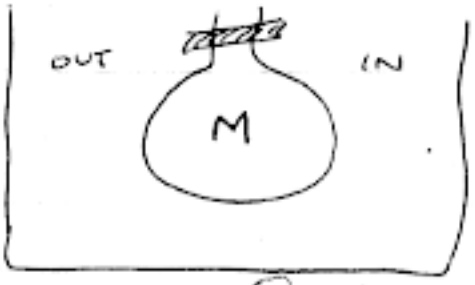


V = Voltage $= \phi_2 - \phi_1$

This is what's important

~~No potential~~

(assume positive charge)



M = macromolecule
⇒ at conc C_M
⇒ w/ charge z_M

Electrical Neutrality Require (1) $C^{OUT}_{Na^+} = C^{OUT}_{Cl^-} = C^{OUT}_{NaCl}$

(2) $C^{IN}_{Cl^-} = C^{IN}_{Na^+} + z_M C_M$

$\mu^{OUT}_{NaCl} = \mu^{IN}_{NaCl}$ (at equilib, membr permeable to ~~for small ions~~)

$\mu^{IN/OUT}_{NaCl} = \mu^{\circ}_{NaCl} + RT \ln a_{NaCl} = RT \ln a_{Na^+} + RT \ln a_{Cl^-}$

$RT \ln a^{OUT}_{Na^+} a^{OUT}_{Cl^-} = RT \ln a^{IN}_{Na^+} a^{IN}_{Cl^-}$

$C^{OUT}_{Na^+} C^{OUT}_{Cl^-} = C^{IN}_{Na^+} C^{IN}_{Cl^-}$

(2) $C^2_{NaCl\ OUT} = C^{IN}_{Na^+} \left(C^{IN}_{Na^+} + z_M C_M \right)$

$C^2_{NaCl\ OUT} = \left(C^{IN}_{Na^+} \right)^2 + z_M C_M C^{IN}_{Na^+}$

$C = x^2 + Bx$

$$C_{Na^+_{INSIDE}} = \frac{-z_M C_M + \sqrt{(z_M C_M)^2 + 4C^2_{NaCl\ OUT}}}{2}$$

$$\frac{C_{Na^+_{INSIDE}}}{C_{Na^+_{OUTSIDE}}} = \underbrace{\frac{-z_M C_M}{2C_{Na^+_{OUT}}}} + \underbrace{\sqrt{\left(\frac{z_M C_M}{2C_{Na^+_{OUT}}}\right)^2 + 1}}$$

Generally, for a protein

$C_M \ll C_{Na^+_{OUT}}$

C_M (protein)

So $\frac{C_{Na^+_{INSIDE}}}{C_{Na^+_{OUT}}} \approx 1 - \frac{z_M C_M}{2C_{Na^+_{OUT}}} < 1$

STOPPED

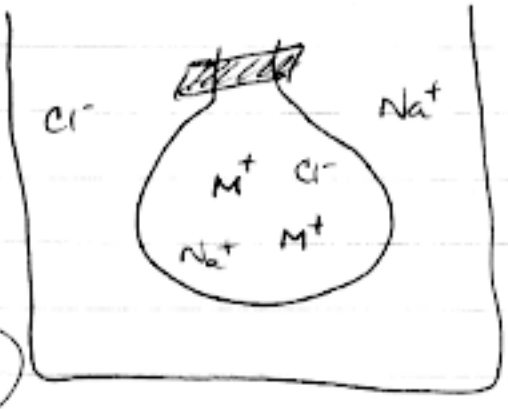
$\therefore$

$C_{Na^+_{INSIDE}} < C_{Na^+_{OUTSIDE}}$

CAN ALSO SHOW THAT

(because $C^{OUT}_{Na^+} C^{OUT}_{Cl^-} = C^{IN}_{Na^+} C^{IN}_{Cl^-}$)

$C_{Cl^-_{INSIDE}} > C_{Cl^-_{OUTSIDE}}$



Net +10 charge

$z_M = +10$

FOR Protein at 1.0 mM (high!) dialyzed against 100mM NaCl

$C_{Na^+_{INSIDE}} = 0.95\ C_{Na^+_{OUTSIDE}}$

$C_{Na^+_{INSIDE}} = 95\text{mM}$

$C_{Cl^-_{INSIDE}} = 105\text{mM}$ — To balance protein charge

At equilibrium

$$RT \ln \frac{C^{IN}_{Na^+}}{C^{OUT}_{Na^+}} + ZFV = 0$$

$$\therefore \quad V = -\frac{RT}{ZF} \ln \frac{C^{IN}_{Na^+}}{C^{OUT}_{Na}} = +1.3 \text{ mV}$$

at 298K

IN THIS CASE, THE ABOVE EQUATION REFERS TO Na^+, SO $Z = 1$

DONNAN POTENTIAL

SHOULD GET SAME NUMBER (AND SIGN) IF WE CALCULATED IT WITH $C^{IN}_{Cl^-}$ and $C^{OUT}_{Cl^-}$