Chapter 5 → 1, 2, 3

10/12/01

Phase Relationships

Raoult's Law for ideal solutions ($\gamma_A = 1$)

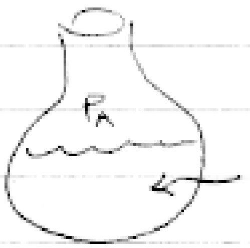
$$P_A = X_A P_A^{\bullet}$$

vapor pressure of A (EtOH) (or H_2O)

Mole fraction of A in solvent (ethanol in water) (water in ethanol)

vapor pressure of pure A (EtOH) (or H_2O)

$$\frac{\text{moles EtOH}}{(\text{moles EtOH}) + (\text{moles } H_2O)}$$



water w/ dissolved ethanol

Henry's Law – what about things like gases, etc

$$P_B = k_A X_A$$

vapor pressure of solute B

Henry's Law constant for solute B in solvent A

Mole fraction of B in sol'n

Depends on temperature and on the particular solvent

Implications: Vapor pressure lowering can be used to measure solute concentrations

Protein ⇒ $X_p = 10^{-8}$ $X_{H_2O} = 1 - 10^{-8}$ vapor pressure lowering

At equilibrium:

$$\mu_{O_2}(aq) = \mu_{O_2}(g)$$

$$\mu_{H_2O}(aq) = \mu_{H_2O}(g)$$

$a_{O_2}(aq) = a_{O_2}(g)$ etc.

We saw before that this lead to Raoult's Law

In air: O₂ is about 20%

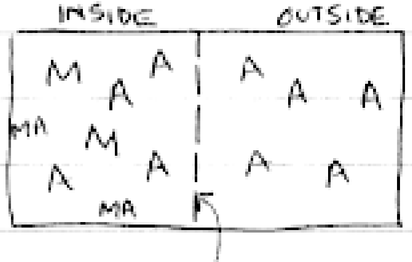
∴ $P_{O_2} = 0.2(1$

$P_{O_2} \approx 0.2$ atm

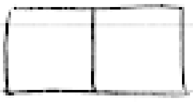
$X_{O_2} = 4.7 \times 10^{-6}$ (dissolved)

Equilibrium Dialysis

Classic



Membrane Permeable to ligand A but not to macromolecule M



Two separate containers
Two sides are NOT at equilib

M ≡ macromolecule is NOT at equilibrium between the left and right sides. (Similarly MA is NOT

A ≡ ligand will equilibrate

but also have $M + A \overset{K}{\rightleftharpoons} MA$ $\quad K = \frac{[MA]}{[M][A]}$

$$\mu_A^{IN} = \mu_A^{OUT}$$

$$a_A^{IN} = a_A^{OUT}$$

& $C_A^{IN} \approx C_A^{OUT}$

$C_M^{ORIG} = C_M + C_{MA} = [M] + [MA]_{IN}$

&

$C_A^{\substack{INSIDE \\ TOTAL}} = [A]_{IN} + [MA]_{IN}$

$C_A^{INSIDE} = C_A^{OUTSIDE}$

Measurable Quantities

$C_M = [MA] + [M]$ You know this because you set it up that way

$C_A^{OUTSIDE} = [A]_{OUT}$ } Measure (radioactivity)

$C_A^{\substack{INSIDE\\TOTAL}} = [A] + [MA]$

then

$$C_A^{BOUND} = [MA] = C_A^{\substack{INSIDE\\TOTAL}} - C_A^{INSIDE} = C_A^{\substack{INSIDE\\TOTAL}} - C_A^{OUTSIDE}$$

Finally,

$$K = \frac{[MA]}{[M][A]} = \frac{C_A^{\substack{INSIDE\\TOTAL}} - C_A^{OUTSIDE}}{\underbrace{\{C_M - (C_A^{\substack{INSIDE\\TOTAL}} - C_A^{OUTSIDE})\} C_A^{OUTSIDE}}_{\text{ugly, but doable}}}$$

MORE COMMONLY

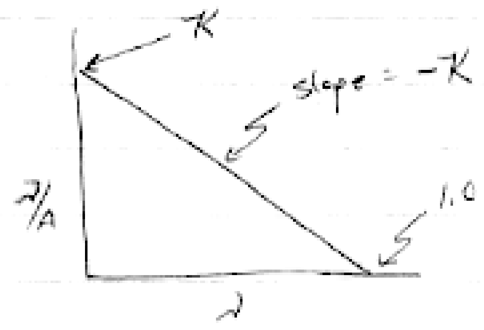
$$\nu = \frac{C_A^{BOUND}}{C_M} = \underset{\uparrow}{\frac{[MA]}{[M]_{TOT}}} = \frac{[MA]}{[MA]+[M]} = \frac{\#\text{ A bound to M}}{\text{total \# of M}}$$

$$= \text{"fraction bound"} = \frac{C_A^{\substack{INSIDE\\TOTAL}} - C_A^{OUTSIDE}}{C_M}$$

then

$$K = \frac{\nu}{(1-\nu)C_A^{OUTSIDE}} = \frac{\nu}{(1-\nu)[A]}$$

$$\frac{\nu}{[A]} = K(1-\nu) = K - \nu K$$



That all assumed $M + A \rightleftharpoons MA$

ν goes from 0 to 1

~~[illegible]~~

What if more than 1 A can bind to M?
(MULTIPLE BINDING SITES)

IF N sites, then ν goes 0 to N

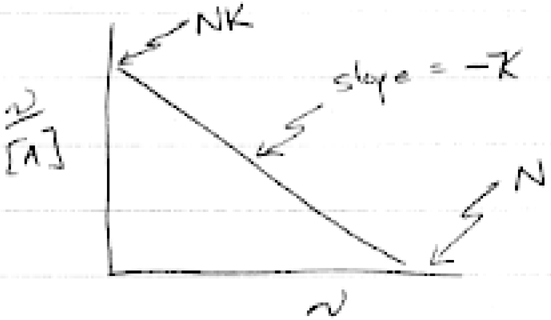
Replacing ν by ν/N

$$\frac{(\nu/N)}{[A]} = K - K\frac{\nu}{N}$$

$$\Downarrow$$

$$\frac{\nu}{[A]} = \underset{\substack{\uparrow \\ \text{y-intercept}}}{KN} - \underset{\substack{\uparrow \\ \text{slope}}}{K}\nu$$

SCATCHARD EQUATION



SCATCHARD PLOT

Valuable, particularly when you don't know how many sites there are.

If NOT a straight line, then the model is too simple (usually need cooperativity etc)

Reminder

ν = average # A's bound per macromolecule
N = Number of binding sites per macromolecule

ν/N goes from $0 \rightarrow 1$ (always)