

Chem 340 - Lecture Notes 9 – Fall 2013 – Phase equilibria (chap.4)

Our wide experience is that at high temperatures liquids boil and become gasses and at low temperatures they freeze to solid forms, we also know that reducing pressures can make them boil at lower temperatures, as in a vacuum distillation. Stability of phases of pure compounds depends on T & P, which can represent by chemical potential, $\mu(T,P)$

The chemical potential of each component in a system at equilibrium must be the same in all parts of the system, in any phase

Consider change in one component dn_1 from one phase to another dn_2

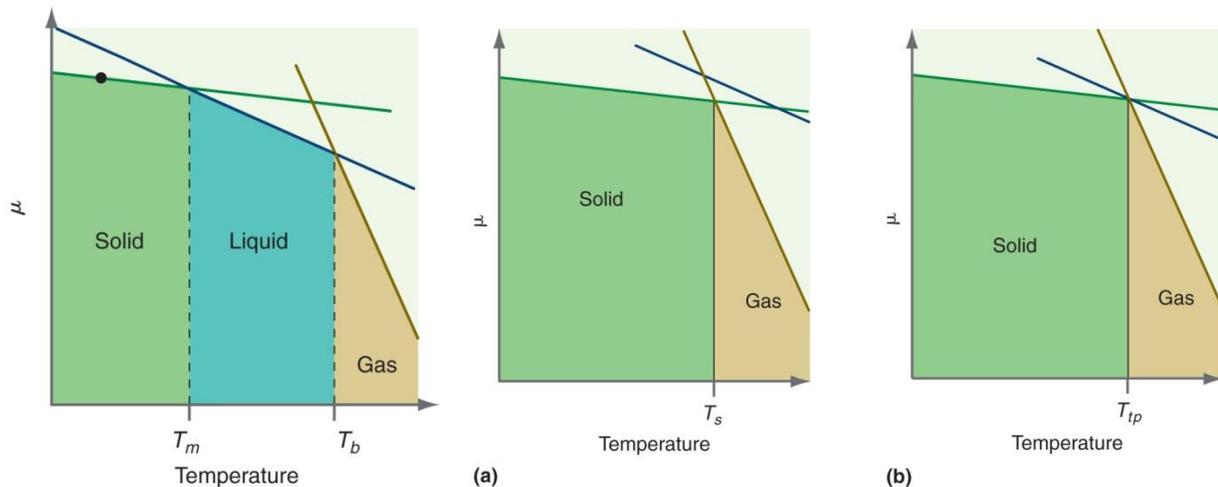
$dG = \mu_1 dn_1 + \mu_2 dn_2 = (\mu_1 - \mu_2) dn$ since amount lost from phase 1, gain in phase 2 at equilibrium, $dG = 0 = (\mu_1 - \mu_2)$ or $\mu_1 = \mu_2$ no change in chemical potential

If not at equilibrium, then system is driven to lower chemical potential, $\Delta G = \mu_1 - \mu_2 = (-)$

If change T, P can shift chemical potential, shift equilibrium and change phase, due to μ

From $d\mu = -S_m dT + V_m dP \rightarrow \left(\frac{\partial\mu}{\partial T}\right)_P = -S_m$ and $\left(\frac{\partial\mu}{\partial P}\right)_T = V_m$

μ always decreases with increasing T and increases with increasing P, $S_m^g > S_m^l > S_m^s$

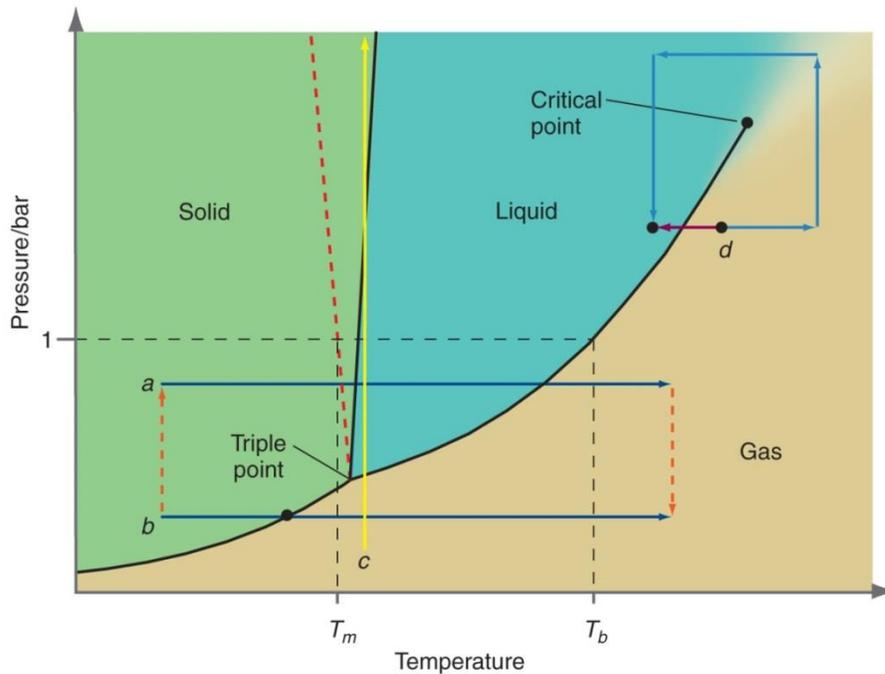


Increase T, solid chem. pot. becomes higher than liquid, **cross over \rightarrow phase change**, melting, then liquid higher than gas, so boil (vaporize). **Slopes vary - $S_m^g > S_m^l > S_m^s$**

As change pressure can move μ lines up, $\sim V$, boiling point (liquid-vapor) increase, alternatively, as decrease P, μ^{vapor} line moves furthest and vapor line crosses past liquid line, and **eventually solid, gas μ lines intersect below liquid \rightarrow sublimation** Liquid-Vapor intersection always moves up with P, T_b increase, but solid-liquid depends on relative density, T_m normally increase, but e.g. water decrease since $\rho_{\text{ice}} < \rho_{\text{water}}$ If they all intersect, called triple point, three phases in equilibrium

Note: these lines have curvature: $(\partial^2\mu/\partial T^2)_P = -(\partial S/\partial T) = -C_P/T$ [i.e. $dS=dq/T=(C_P/T)dT$]

Phase diagrams combine these behaviors in a T,P plot showing regions of stability



Consider **processes**:

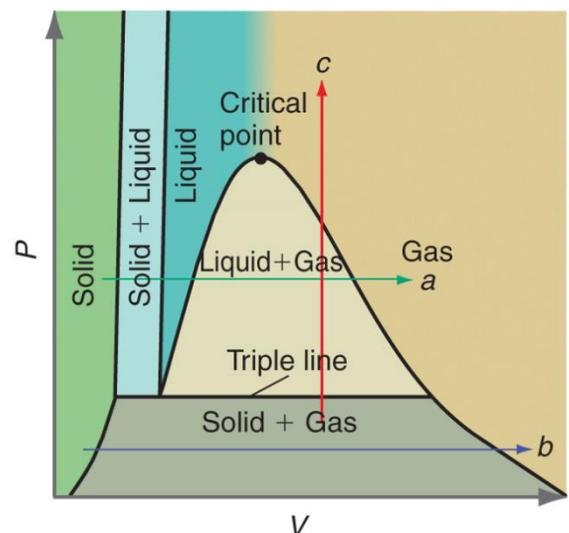
- a, green region solid, const. P, increase T, to 1st black line → solid-liquid equilibrium, then blue area liquid, to 2nd black line → liquid-vapor equilibrium, beige region – gas
- b, start solid (green) to black line solid-vapor equilibrium, then vapor (beige)
- c, const. T, increase P, start vapor, at line condense to liquid, (blue), then form solid

Special points – triple point, solid-liquid-vapor equilibrium
 Critical point, liquid-vapor indistinguishable, same density

Slope of solid-vapor and liquid-vapor positive since ΔV to vapor large and positive
 Slope of solid-liquid can be either depending on relative density, typical $\rho_{sol} > \rho_{liq}$, ex. H₂O

- Diagram can also be P-V
- Here areas are different phases, some mixed
- Green is solid (single phase)
- Light blue – solid-liquid equilibrium
- Blue-green is liquid (low V, high P)
- Tan is vapor (single phase), but
- Grey is solid-vapor and beige is liquid-vapor

So T-P phase diagrams indicate equilibria by lines, but P-V show regions of equilibria
 Critical point is top of liq-vap curve, indistinguishable
 Triple point intersect green-blue-grey (sol-liq-vap)



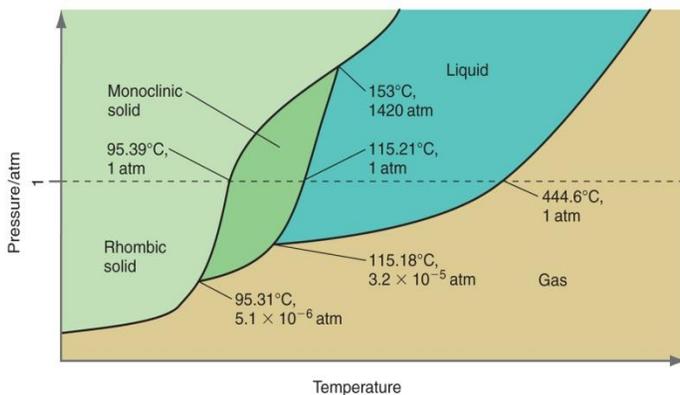
Equilibrium is on a line in T,P plot – see this from

- For isolated system: $dS = 0 = dS_\alpha + dS_\beta$ for two phases α and β in equilibrium so if push dq from $\alpha \rightarrow \beta$ then $dq_\alpha = -dq_\beta$ so this means: $0 = dq/T_\alpha - dq/T_\beta$ but this means $T_\alpha = T_\beta$, so phases in equilibrium same temperature
- If total volume and temperature constant, $dA = 0 = dA_\alpha + dA_\beta$
at const T, $dA = PdV$ (since $SdT = 0$), change one component $dV = dV_\alpha = -dV_\beta$
 $0 = P_\alpha dV - P_\beta dV$ or $P_\alpha = P_\beta$, equilibrium pressure same

Thus equilibria have phases with same T,P or on a line in T,P phase diagram

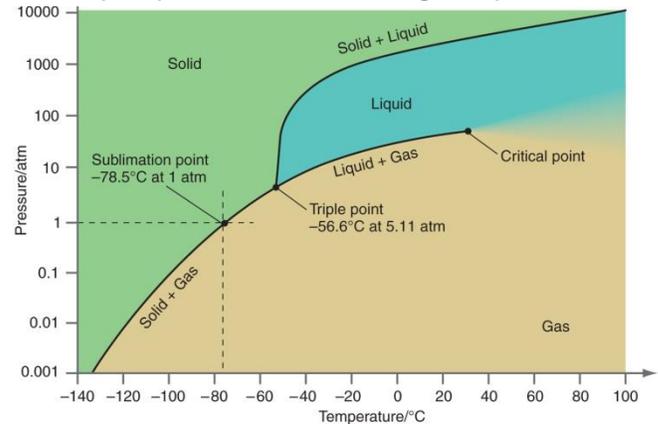
As shown above, const T,P $dG = 0$, *chemical potential the same*, $\mu_\alpha = \mu_\beta$, no net drive or no change, $dn = 0$, in amount of one phase or the other

Vapor phase unique, only one, but can have multiple solid phases (and maybe liquid) e.g. Sulphur has rhombic and monoclinic crystal (solid) phases



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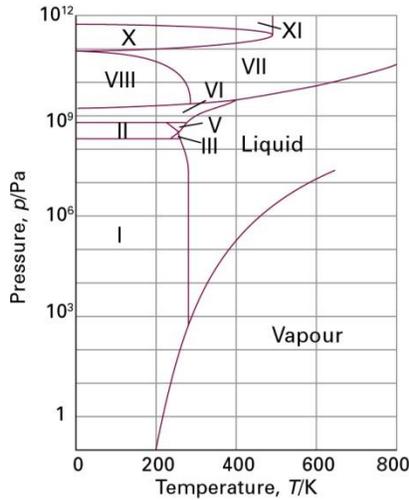
flip diagram, use log, for CO₂ at 1 atm sol-vap equil., increase P to get liquid



Phase Rule

When we drew the P,T phase diagram we saw a number of phases represented by areas, and saw them in equilibrium on a line or at a point (point is fixed, no freedom, line has 1 degree of freedom, move along it: $P = P(T)$, area – 2 degrees, any P,T) Question, how many phases can be in equilibrium at once? Depend on degree freedom Need to determine number of phases in system, #P, must be distinguishable and homogeneous, if multicomponent, the solutions must be miscible. Always true for vapor, not necessary for liquid (think oil and water), and often not for solids (crystal) Example: salt + water \rightarrow vapor, liquid (NaCl in water), salt, ice \rightarrow 4 phases or #P = 4 in a closed vessel, could have some in equilibrium, vapor above liquid (vapor pressure) or salt and liquid and vapor (saturate the solution and ppt out the salt), and at some temperature could form ice (crystallize out the salt) and be in equilibrium with liq, vap

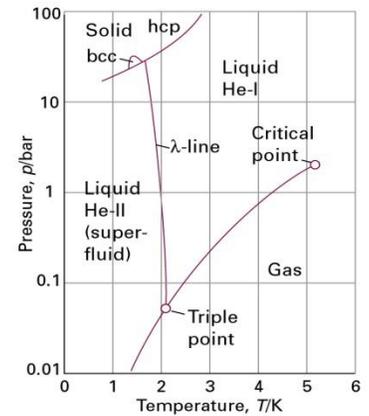
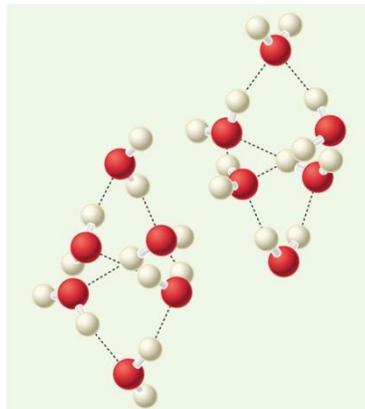
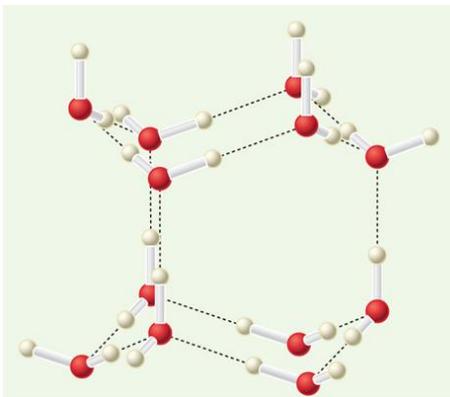
Gibbs phase rule puts a limit on this: $F = C - \#P + 2$ (justified: 4.1, p.139, text) F - degrees of freedom, how many parameters can vary independently, C – min. number of components, and #P – number of phases in equilibrium



Single component, can vary T,P, ($C=1$, $\#P=1$) but if 2 phases in equilibrium, $F = 1 - 2 + 2 = 1$, so can vary only one parameter, a line, e.g. vary T, $P=P(T)$, and if 3 in equilibrium, $F = 0$, or just a point, the **Triple point**

Consider the **water diagram**, left, multiple solid phases, each line an equilibrium of two phases, points where lines intersect have three phases (from text, p.141)

At $P = 1$ bar, ice I dominate, see hexagonal H-bonded structure, below left, but as raise pressure, gets more dense, ice VI (more compact center)



Liquids can also have phases, less common, but He-I and He-II are well known to separate at $T \sim 2.1$ K and reduced P (He-II super fluid, no viscosity, remarkably clear, bubble free) He-I condenses at $P = 1$ bar at $T \sim 4.2$ K

Other liquids normally one phase, but **liquid crystals and glasses are in-between** state

Water is the super important liquid phase for biology, and it is different from other liquids melting and boiling points are high for a small molecule, with a large liquid range, enthalpy of vaporization $\Delta H_{\text{vap}} = 41$ kJ/mol which is high, allow efficient evap. cooling heat capacity allows planet to keep its temperature more stable, support life

Volume contraction on cooling was noted before, ice floats, the planet does not solidify, planet can support marine life and eventually all life

Clapeyron-equation

We established that at equilibrium two phases, α, β have $T_\alpha = T_\beta$, $P_\alpha = P_\beta$, and $\mu_\alpha = \mu_\beta$,

So from this clearly at equilibrium: $\mu_\alpha(T,P) = \mu_\beta(T,P)$ so if change $P, T \rightarrow T+dP, T+dT$

Then $\mu_\alpha(T,P) + d\mu_\alpha = \mu_\beta(T,P) + d\mu_\beta$ which from above means $d\mu_\alpha = d\mu_\beta$

Re-express use fundamental equation: $d\mu_\alpha = -S_{m\alpha}dT + V_{m\alpha}dP$ and $d\mu_\beta = -S_{m\beta}dT + V_{m\beta}dP$

Then : $-S_{m\alpha}dT + V_{m\alpha}dP = -S_{m\beta}dT + V_{m\beta}dP \rightarrow (S_{m\beta} - S_{m\alpha})dT = (V_{m\beta} - V_{m\alpha})dP$

$$\frac{dP}{dT} = \frac{\Delta S_m}{\Delta V_m}$$

Rearrange to Clapeyron eqn:

Show: now for our P, T phase diagrams, we have a directive for the slopes of the lines

Consider **melting** $\Delta G_{fus} = \Delta H_{fus} - T\Delta S_{fus} = 0$ at equilibrium so $\Delta S_{fus} = \Delta H_{fus}/T$

Example: From tables one can see that values vary but an average : $\Delta S_{fus} \sim 22 \text{ J/molK}$

ΔV small for solid to liquid change, but mostly $\sim 4 \times 10^{-6} \text{ m}^3$, with most (+), but H_2O (-)

Plug in : $(dP/dT)_{fus} = \Delta S_{fus}/\Delta V_{fus} \sim 22 \text{ J/molK}/4 \times 10^{-6} \text{ m}^3 = 5.5 \times 10^6 \text{ Pa/K} = 55 \text{ bar/K}$

steep curve, 1° results in 55 bar, or invert, ~ 50 bar needed to change T_m by 1°

Vaporization same story, but ΔV_{vap} much bigger and about same for most substances,

molar volume $\sim 20 \text{ L/mol}$ for ideal gas where many liquids boil

Similarly ΔS_{vap} about the same, Trouton's rule, $\Delta S_{vap} = 90 \text{ J/molK}$

(except strong interact like water, ammonia – H-bonding)

Putting these together: $(dP/dT)_{vap} = \Delta S_{vap}/\Delta V_{vap} \sim 90 \text{ J/molK}/20 \times 10^{-3} \text{ m}^3$

$$= 5 \times 10^3 \text{ Pa/K} = 5 \times 10^{-2} \text{ bar/K}$$

More modest slope than solid-liquid curve, invert, see 1 bar shift T_b by $20^\circ \rightarrow$ big impact

Practical impact, pressure cooker can shift T_b with modest ΔP

Sublimation : state function means add vaporization to fusion: $\Delta S_{sub} = \Delta S_{fus} + \Delta S_{vap}$

Since all are positive $\Delta S_{sub} > \Delta S_{vap}$ and $\Delta V_{vap} \sim \Delta V_{fus} \rightarrow (dP/dT)_{sub} > (dP/dT)_{vap}$

So solid-vapor curve steeper slope than the liquid vapor curve (see figs, but close T_{tp})

Computing vapor pressure (**Clausius-Clapeyron equation**):

Integrate Clapeyron: $\int dP = \int (\Delta S_{fus}/\Delta V_{fus})dT = \int (\Delta H_{fus}/\Delta V_{fus})(dT/T) \sim (\Delta H_{fus}/\Delta V_{fus})\int dT/T$

Evaluate: $P_f - P_i = (\Delta H_{fus}/\Delta V_{fus})\ln(T_f/T_i) = (\Delta H_{fus}/\Delta V_{fus})\ln[(T_i+\Delta T)/T_i] \sim (\Delta H_{fus}/\Delta V_{fus})[\Delta T/T_i]$

(last step uses $\ln(1-x) \sim -x$ for small x) \rightarrow see ΔP vary linearly with ΔT ($\Delta S/\Delta V \sim \text{const}$)

For liquid-vapor, $\Delta V \sim V_{vap}$ use ideal gas:

$$(dP/dT)_{vap} = \Delta S_{vap}/\Delta V_{vap}$$

$$\sim \Delta H_{vap}/TV_{vap} \sim P\Delta H_{vap}/RT^2$$

$$dP/P = [\Delta H_{vap}/RT^2]dT \text{ if } \Delta H_{vap} \sim \text{const.};$$

$$\int_{P_i}^{P_f} \frac{dP}{P} = \frac{\Delta H_{vaporization}}{R} \times \int_{T_i}^{T_f} \frac{dT}{T^2}$$

$$\ln \frac{P_f}{P_i} = -\frac{\Delta H_{vaporization}}{R} \times \left(\frac{1}{T_f} - \frac{1}{T_i} \right)$$

Same story with sublimation, use ΔH_{sub} :

Example: Consider Hg spill, if $\Delta H_{\text{vap}} = 60.9 \text{ kJ/mol}$ and $T_b = 630 \text{ K}$, what is the vapor pressure of Hg at room temperature ($T = 298 \text{ K}$)?

$$\ln(P_f/P_i) = -\Delta H_{\text{vap}}/R(1/T_f - 1/T_i) = -60.9 \times 10^3 / 8.314 (1/298 - 1/630) = -13.0$$

$$P_f = 2.4 \times 10^{-6} \text{ bar} \quad \text{use } P_i \sim 1 \text{ bar, boiling point}$$

Note: safe health limit (OSHA 8 hr total) is 0.1 mg/m^3 -- how does this compare?

$$m/V = n(\text{MW})/V = (\text{MW}) P/RT$$

$$= (201 \text{ g/mol})(2.4 \times 10^{-6} \text{ bar}) / (0.082 \text{ L}\cdot\text{bar/mol}\cdot\text{K} \cdot 298 \text{ K}) = 19.7 \times 10^{-6} \text{ g/L}$$

$$m/V = 19.7 \text{ mg/m}^3 \quad (\text{unit change: } 10^3 \text{ mg/g} \times 10^3 \text{ L/m}^3 = 10^6)$$

Well over the allowable limit—this is why we must clean up Hg spills

More complex example from Engel:

EXAMPLE PROBLEM 7.2

The normal boiling temperature of benzene is 353.24 K , and the vapor pressure of liquid benzene is $1.00 \times 10^4 \text{ Pa}$ at 20.0°C . The enthalpy of fusion is 9.95 kJ mol^{-1} , and the vapor pressure of solid benzene is 88.0 Pa at -44.3°C . Calculate the following:

- $\Delta H_m^{\text{vaporization}}$
- $\Delta S_m^{\text{vaporization}}$
- Triple point temperature and pressure

Solution

- We can calculate $\Delta H_m^{\text{vaporization}}$ using the Clapeyron equation because we know the vapor pressure at two different temperatures:

$$\ln \frac{P_f}{P_i} = -\frac{\Delta H_m^{\text{vaporization}}}{R} \left(\frac{1}{T_f} - \frac{1}{T_i} \right)$$

$$\begin{aligned} \Delta H_m^{\text{vaporization}} &= \frac{R \ln \frac{P_f}{P_i}}{\left(\frac{1}{T_f} - \frac{1}{T_i} \right)} = \frac{8.314 \text{ J mol}^{-1} \text{ K}^{-1} \times \ln \frac{101,325 \text{ Pa}}{1.00 \times 10^4 \text{ Pa}}}{\left(\frac{1}{353.24 \text{ K}} - \frac{1}{273.15 + 20.0 \text{ K}} \right)} \\ &= 33.2 \text{ kJ mol}^{-1} \end{aligned}$$

$$\text{b. } \Delta S_m^{\text{vaporization}} = \frac{\Delta H_m^{\text{vaporization}}}{T_b} = \frac{33.2 \times 10^3 \text{ J mol}^{-1}}{353.24 \text{ K}} = 93.9 \text{ J mol}^{-1} \text{ K}^{-1}$$

c. At the triple point, the vapor pressures of the solid and liquid are equal:

$$\ln \frac{P_{tp}^{liquid}}{P^\circ} = \ln \frac{P_i^{liquid}}{P^\circ} - \frac{\Delta H_m^{vaporization}}{R} \left(\frac{1}{T_{tp}} - \frac{1}{T_i^{liquid}} \right)$$

$$\ln \frac{P_{tp}^{solid}}{P^\circ} = \ln \frac{P_i^{solid}}{P^\circ} - \frac{\Delta H_m^{sublimation}}{R} \left(\frac{1}{T_{tp}} - \frac{1}{T_i^{solid}} \right)$$

$$\ln \frac{P_i^{liquid}}{P^\circ} - \ln \frac{P_i^{solid}}{P^\circ} - \frac{\Delta H_m^{sublimation}}{RT_i^{solid}} + \frac{\Delta H_m^{vaporization}}{RT_i^{liquid}} = \frac{(\Delta H_m^{vaporization} - \Delta H_m^{sublimation})}{RT_{tp}}$$

$$T_{tp} = \frac{(\Delta H_m^{vaporization} - \Delta H_m^{sublimation})}{R \left(\ln \frac{P_i^{liquid}}{P^\circ} - \ln \frac{P_i^{solid}}{P^\circ} - \frac{\Delta H_m^{sublimation}}{RT_i^{solid}} + \frac{\Delta H_m^{vaporization}}{RT_i^{liquid}} \right)}$$

$$= \frac{9.95 \times 10^3 \text{ J mol}^{-1}}{8.314 \text{ J K}^{-1} \text{ mol}^{-1} \times \left(\ln \frac{10,000 \text{ Pa}}{1 \text{ Pa}} - \ln \frac{88.0 \text{ Pa}}{1 \text{ Pa}} - \frac{(33.2 \times 10^3 + 9.95 \times 10^3) \text{ J mol}^{-1}}{8.314 \text{ J K}^{-1} \text{ mol}^{-1} \times 228.9 \text{ K}} + \frac{33.2 \times 10^3 \text{ J mol}^{-1}}{8.314 \text{ J K}^{-1} \text{ mol}^{-1} \times 293.15 \text{ K}} \right)}$$

$$= 277 \text{ K}$$

We calculate the triple point pressure using the Clapeyron equation:

$$\ln \frac{P_f}{P_i} = -\frac{\Delta H_m^{vaporization}}{R} \left(\frac{1}{T_f} - \frac{1}{T_i} \right)$$

$$\ln \frac{P_{tp}}{101,325} = -\frac{33.2 \times 10^3 \text{ J mol}^{-1}}{8.314 \text{ J mol}^{-1} \text{ K}^{-1}} \times \left(\frac{1}{277 \text{ K}} - \frac{1}{353.24 \text{ K}} \right)$$

$$\ln \frac{P_{tp}}{P^\circ} = 8.41465$$

$$P_{tp} = 4.51 \times 10^3 \text{ Pa}$$

Surface tension

Droplets assume spherical shape in absence of other forces, maximize volume:surface if distort surface, must do **work**, at const V, T:

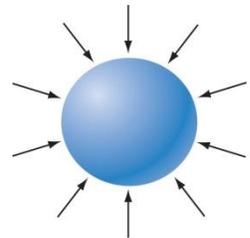
$dA = \gamma d\sigma$ Helmholtz free energy change depend on γ – surface tension, $d\sigma$ - area, \rightarrow to have $dA < 0$ **spontaneous**, want to **minimize area**, to keep it spherical need a **force**

consider: $d\sigma = 4\pi(r+dr)^2 - 4\pi r^2 = 4\pi(r^2+2rdr+dr^2) - 4\pi r^2 \sim 8\pi r dr$

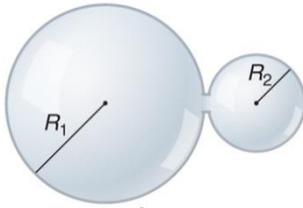
so if $dA = dw = \gamma d\sigma = 8\pi\gamma r dr$ then the force is

$F = 8\pi\gamma r$ from $w = F \cdot r$ that relates to total P in balance

$4\pi r^2 P_{in} = 4\pi r^2 P_{out} + 8\pi\gamma r$ - when drop stable, forces balance or $P_{in} = P_{out} + 2\gamma/r$



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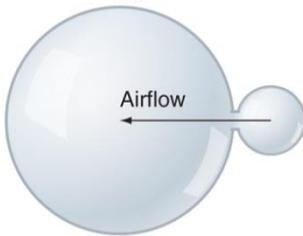


So as diameter $\rightarrow \infty$, $P_{in} \rightarrow P_{out}$ ($8\pi\gamma r$ small part)

Bring two bubbles together with different diameters:

$$P_1 - P_2 = 2\gamma/R_1 - 2\gamma/R_2 = 2\gamma(1/R_1 - 1/R_2)$$

P in smaller bubble greater ($P_1 < P_2$), if contact, air flows into larger one, they coalesce and bubble gets larger – **coarsening**



In lungs **alveoli** are small air sacs, water lined, and if they coarsen you would lose ability to breathe – collapse of lungs.

A **surfactant** in the lung surface lowers surface tension to stabilize alveoli, so they do not coarsen. Smoking, pollution can cause loss of surfactant ability to lower γ



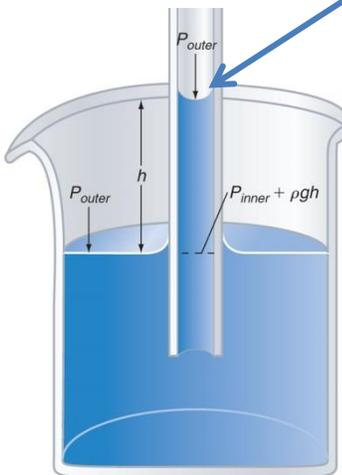
Vapor pressure of drop also depends on radius, can show:

$$\ln(P/P^0) = 2\gamma M/r\rho RT \quad M\text{-molar mass, } \rho\text{-density}$$

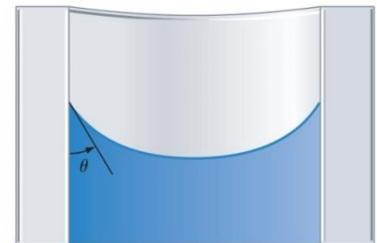
Small droplets evaporate faster than large ones and vapor condenses on large ones at expense of small ones, forming droplets in fog or clouds (relatively small difference)

Capillary rise is another example of the difference in P_{in} & P_{out}
Can view difference as force of gravity on mass raised in tube

$$P_{out}^{cap} = P_{in} + 2\gamma/r \rightarrow 2\gamma/r = \rho hg \quad (\text{note: density } \rightarrow P \sim F/\text{area})$$



Angle of contact, θ = contact angle depend on surface “wetting” $\theta = 0^\circ$, complete, $\theta = 180^\circ$, nonwetting



$$\text{Use as } P_{out} = P_{in} + 2\gamma(\cos\theta)/r \quad \text{or} \quad h = 2\gamma(\cos\theta)/\rho g r$$

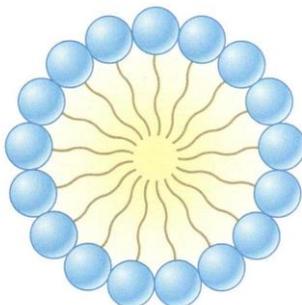
Measure contact angle, determine γ at solid-liquid interface why bugs can walk on water, surface tension balance weight

Amphiphilic Molecules also take advantage of the differences at surfaces, and can create distinct interfaces between phases

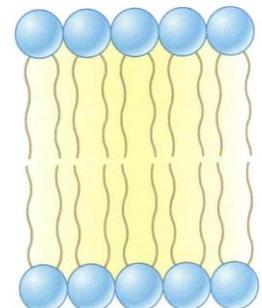
Detergents, (soap), fatty acids, surfactants and especially lipids

Create micelles and bilayers due to polar head groups (out to water), aliphatic tails (CH_2)_n to each other (hydrophobic effect).

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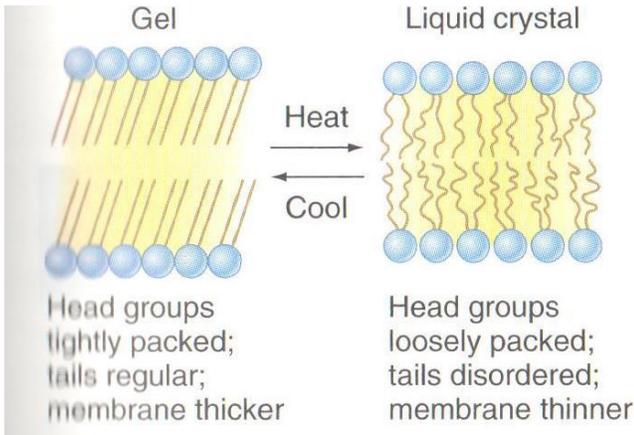


(a) Micelle

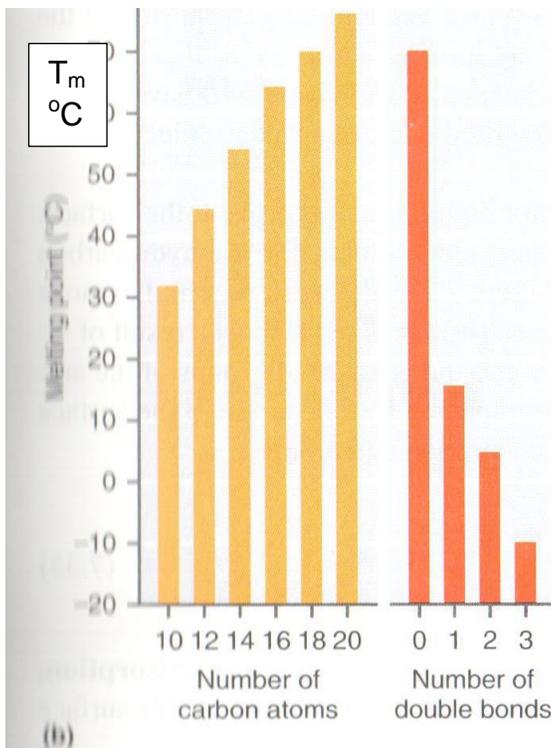


(b) Bilayer

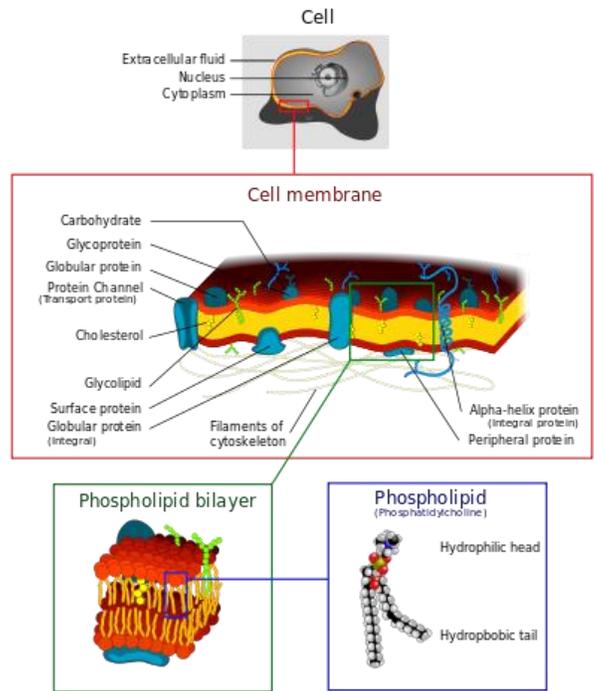
These lipid bilayers then form **structures in cells** e.g. nucleus, organelles (mitochondria) Form a barrier, also **transport mechanism**, since wall contains many protein channels **Phase transition**, **gel phase** – ordered tails → **liquid crystal** – less order, fluid motion



Phase transition T_m increase with length of tails and decrease with double bonds



and force on plate perimeter (p) is $F/p = \gamma \cos\theta$

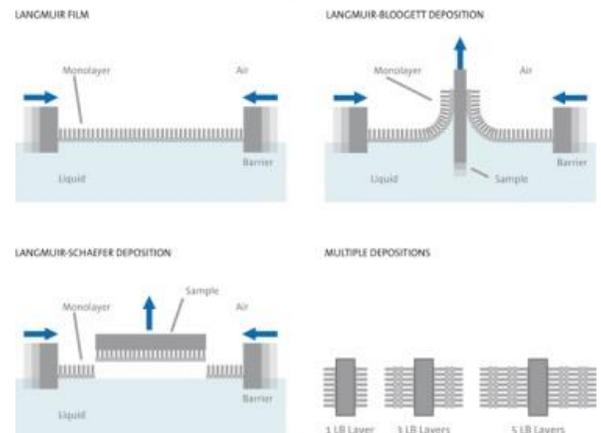


E 7.6 Some Common Phosphoglycerides Found in Biological Membranes

Head Group Negative at pH 7	
Lipid Name	X
Phosphatidic acid	-H
Phosphatidylserine	$-\text{CH}_2\text{CH}(\text{NH}_3^+)\text{COO}^-$
Phosphatidylinositol	$-\text{C}_6\text{H}_6(\text{OH})_5$
Phosphatidylglycerol	$-\text{CH}_2\text{CH}(\text{OH})\text{CH}_2\text{OH}$
Head Group Zwitterionic at pH 7	
Lipid Name	X
Phosphatidylethanolamine	$-\text{CH}_2\text{CH}_2\text{NH}_3^+$
Phosphatidylcholine	$-\text{CH}_2\text{CH}_2\text{N}(\text{CH}_3)_3^+$

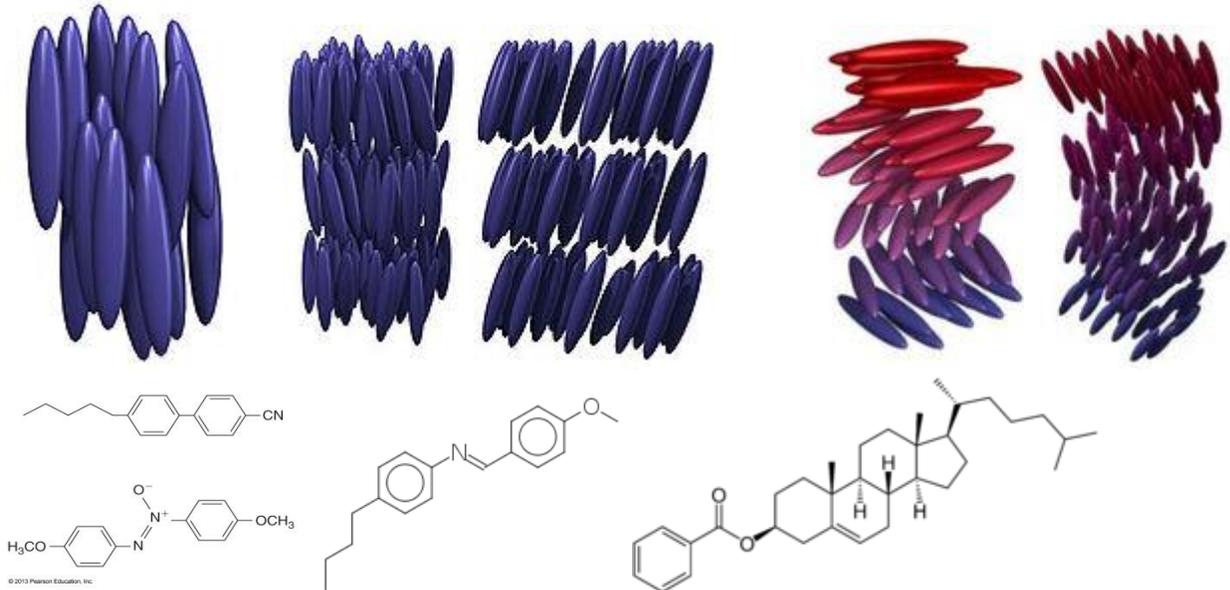
lipid surfaces on water can be studied directly using Langmuir Blodgett trough, or by transfer to a plate. Surface pressure control with movable barrier and measured with Wilhelmy balance (with blade entering surface)

Pressure is difference in surface tension due to surfactant $\Pi = \gamma^o - \gamma$

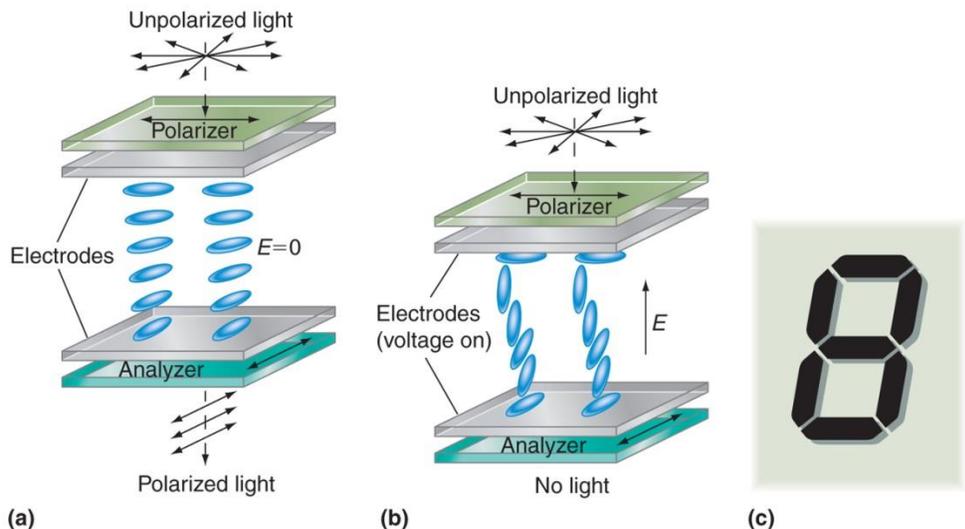


Liquid crystals: typically form from polar molecules with high aspect ratio (long), which favors self orientation, pack together better (hydrophobic parts), dipole select orient thermotropic, organic, have phase transition with temperature from solid to liquid crystal lyotropic, phase transition with T and concentration (in solvent)–biological (lipid, protein)

Can be parallel in a row or ordered in a sheet, sheets can be stacked or slide past
Phase transition between these various states , like solid phases but can have fluidity
Nematic, molecules Smectic have layers as well can also twist – (left) chiral nematic or cholesteric, (right) smectic C*



Also can orient stacks with a twist, which can polarize light and switch under electric field between nematic and cholesteric, phase transition takes energy from battery

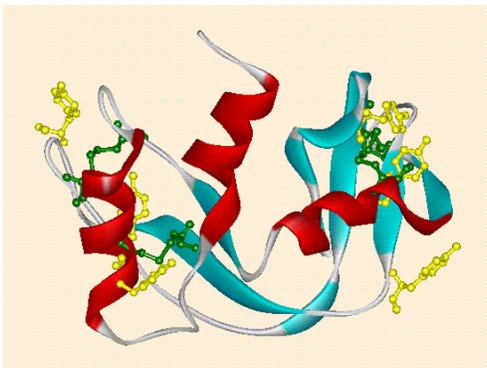


(backwards?)

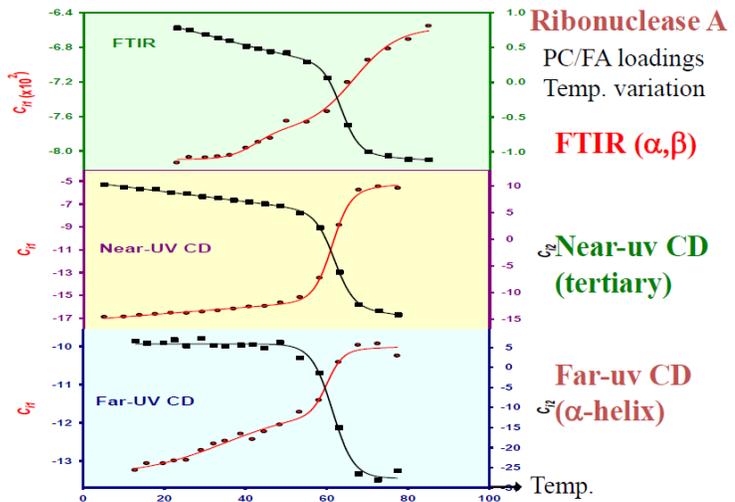
Used for digital displays in computers, televisions, phones etc.

Phase equilibrium usually about a system or substance in a system, like water → ice, or vapor pressure above a liquid. Then we looked further and asked about organization of like molecules (self-assembled) for liquid crystal or lipids - here the system becomes more microscopic, a cell or domain

Going further we can use the phase equilibrium idea to talk about macromolecules and their ordered or disordered structures – e.g. **protein or (poly) nucleic acid folding**
 This is a change in order with no change in chemical bonding, so like a phase transition
 But could view like a reaction form $P_f \rightarrow P_u$ folded to unfolded, in **equilibrium**



As RNase unfolds, helices and sheets lose H-bonds and become disordered, H-bond H₂O
 See that whole protein unfolds in single step (fluorescence, near-uv CD) secondary structure in 2 steps (far-uv CD, IR)



Pre-transition evident in far-uv CD and FTIR, not near-uv CD
 secondary structure in 2 steps (far-uv CD, IR)

Balance of forces leads to folding protein (simplified)

- Entropy** favors unfolded state, many configurations (states) to occupy, fold just one
- Enthalpy** favors folded state, intern H-bonds and interactions of side chains
- Hydrophobic effect** favor fold, non-polar side chains away from water (elim.cages)

Unfold with temperature (entropy → ΔS+) also with pH, denaturant: e.g. urea, GuHCl since enthalpy also positive (ΔH+), ΔG (+) low temp, (-) at high

$$\Delta G = \Delta H - T\Delta S \text{ for denaturation reaction, } N \rightarrow D, \text{ native to denatured structure}$$

$$K = C_D/C_N = f_D C / f_N C = f_D / f_N = f_D / (1 - f_D) \text{ where } C \text{ is total protein concentration}$$

So usually modify measured parameter (like spectral response) to fraction denatured: f_D

$$\text{Melting temperature (transition) is } T_m \text{ where } f_D = 0.5 \text{ and } K = 1 \rightarrow \Delta G = 0$$

Two-state folder, means no intermediate in process, transition is simple sigmoidal curve (like near-UV above), DSC narrow C_p . If not say multistate (e.g. CD or IR above)

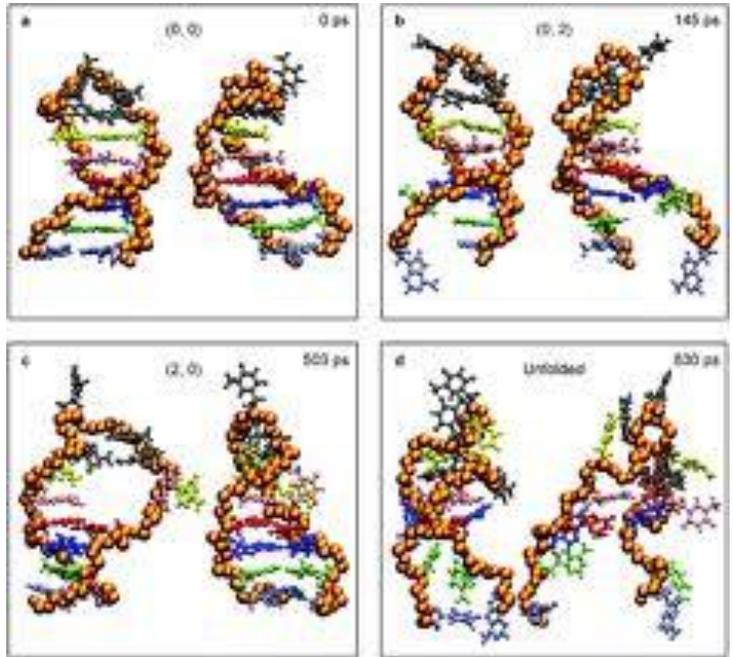
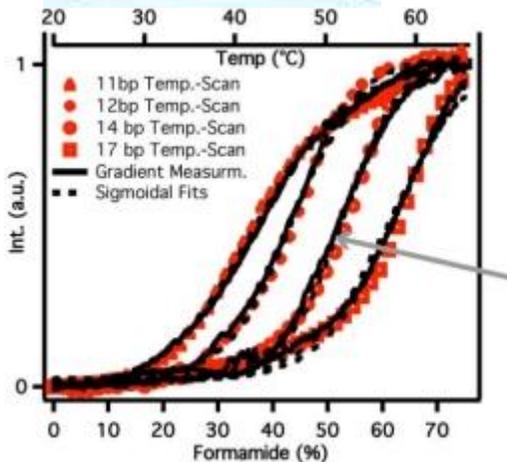
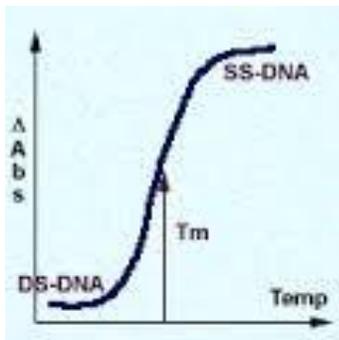
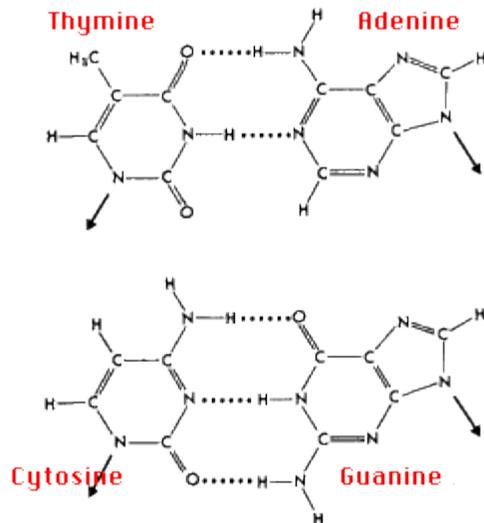
Van't Hoff relation: $\Delta H = RT^2(\partial \ln K / \partial T) \rightarrow \ln K(T) = \ln K^0 - (\Delta H/R)(1/T - 1/T^0)$ – any meas. K

Plot $\ln K$ vs $1/T$ and if $\Delta H \sim \text{const}$, get linear relationship **slope = $-\Delta H/R$ (2 state)**

DSC measures all enthalpy contributions, **if 2-state $\Delta H_{DSC} = \Delta H_{VH}$ if not $\Delta H_{DSC} > \Delta H_{VH}$**

Nucleic acids can also be unfolded with temperature change and added ligands (salts, solvent)

Interactions differ: H-bonds between base pairs and stacking of base π -systems both favor double helix form, but entropy again favors unfolded form



melting curves again measured with spectral response, often UV absorbance or CD, ds \rightarrow ss oligomers more gradual sigmoidal, but long duplex can be sharp. If duplex, ds, to single strand, ss

ds \rightarrow 2 ss then let C = total conc. strands, f = fraction ds, $C_{ds} = fC/2$, $C_{ss} = (1-f)C$

$$K = \frac{C_{ss}^2}{C_{ds}} = \frac{C^2(1-f)^2}{(fC/2)} = 2C \cdot \frac{(1-f)^2}{f}$$

Melting, at T_M $f=0.5$: $\Delta G^\circ = \Delta H^\circ - T_m \Delta S^\circ = -RT \ln C$

$$\text{from } K(T_m) = \frac{2C(1-0.5)^2}{0.5} = C \cdot \frac{2 \cdot 0.25}{0.5} = C$$

$$\text{so that } T_m = -R \ln C / \Delta H^\circ + \Delta S^\circ / \Delta H^\circ$$

plot $1/T_m$ vs $\ln C$ and get slope = $-R/\Delta H^\circ$

intercept of $+\Delta S^\circ / \Delta H^\circ$