Chem 431A-L17-F'07
admin: Quiz on Wednesday
Online quiz Chapt 4 due Monday, Nov.5
Last lecture:

1) look at α- and β-keratins, collagen, silk
2) Stability of protein: Anfinsen Expt
3) Thermodynamic factors in protein stability
4) directed folding, hydrophobic collapse
5) prions, molecular chaperones

Today:

1) Chou-Fasman Rules for 2° prediction
2) motifs, domains, 4° structure
3) oxygen-binding proteins (chapter 4)

*protein folding: not random conformational search but directed folding pathway, stability increases sharply as folding proceeds **molten globule*=result of *hydrophobic collapse* (likely scenario since proteins have hydrophobic cores) * prions (*pr*oteinaceous *i*nfectious *on*ly) – discuss creutsfeldt-jakob disease (mad cow) * molecular chaperones - shelters the hydrophobic sidechains of heat-denatured chaperonins. Hsp = *h*eat *s*hock *p*roteins. *Chaperonins* are more elaborate.	* look at ribbon representation of proteins(Fig 4-18) * *domains*, (Fig 4-19); stable globular units of proteins >200 aa ≈1000 unique domains **Motifs* (Fig 4-20) = common folding pattern of 2° structures also called supersecondary structure * go over the heating curves. Fig 4-26: shows cooperativity. Differentiate the curves. **Quaternary* structure = proteins >100 kD MW:composed of more than 1 polypeptide chain. *Multimers*. *Oligomers*. Spatial arrangement of these subunits = 4° struc

Prediction of 2, 3 struc: *Chou Fasman* rules for prediction: based on propensity of aa's to being in either α-helix or β-sheet. (must refer to table of P_α and P_β as a given) 1) Any segment of 6 or more residues with $\langle P_\alpha \rangle \geq 1.03$ as well as $\langle P_\alpha \rangle > \langle P_\beta \rangle$ and not including Pro is predicted to be α -helix. 2) Any segment of 5 or more residues with $\langle P_\beta \rangle \geq 1.05$ as well as $\langle P_\beta \rangle > \langle P_\alpha \rangle$ and not	including Pro is predicted to be β -sheet. (<P> means "average propensity") 4) Examine the sequence of tetrapeptides with $\langle P_\alpha \rangle < 0.9$, $\langle P_t \rangle > \langle P_\beta \rangle$. They have a good chance of being turns. Eg. $(Ala)_6$ will have $\langle P_\alpha \rangle = 1.42$ and $\langle P_\beta \rangle = 0.83$. fulfills (1). It's α-helix. Compare with Val. High propensity for β sheet structure

Oxygen binding proteins: Chapter 5 Introduction to protein structure and function: Often proteins bind *reversibly* to *ligands* at the *binding site* which is complementary to the shape fo the ligand. Concept of *lock & key* and *induced fit*. For	enzymes ligands are the *substrates* and the binding sites are the *active sites*. For this chapter we look at oxygen-binding proteins. We extend our knowledge to enzymes later.

The study of proteins took a big step forward with the determination of the first globular proteins. The first ones being Myoglobin (Mb), and then Hemoglobin (Hb). Both of these are important **Sperm Whale Mb** (Kendrew, 1959) -globular protein: 44x44x25Å. - 8 α -helices (labelled A - H) -has a single *heme* (a *porphyrin* ring system containing 4 *pyrrole* groups (there are 4 N atoms at the edge which ligand Fe^{2+} (ferrous). Fe^{2+} also liganded to a His side chain (His F8). O_2 can act as the 6th ligand. -heme is in a hydrophobic pocket between E and F chains.	when exposed to O_2 ISOLATED heme Fe^{2+} gets oxidized to Fe^{3+} irreversibly. but in the Mb, heme does not get oxidized to Fe^{3+}, but it changes electronic state, and the Mb goes from dark purple to bright red (like the color of oxygenated blood). Oxy vs deoxy Mb. Sometimes, oxygen can oxidize the Fe^{2+} to Fe^{3+} even in the Mb, it becomes metMb (*metmyoglobin*). This is a brown color of dried blood or old meat. In addition to O_2, other small molecs like NO, CO and H_2S can bind to heme in protein. CO has a 200 fold greater affinity for hemoglobin than O_2. It accounts for its toxicity.

-function is to facilitate O_2 transport in the muscles.(rapidly respiring tissue need O_2 fast at certain times and diffusion isn't fast enough. Mb acts like a bucket brigade. Of course, aquatic animals use it too as O_2 storage. Spermwhale has 10x more mb than terrestial animals. $Mb + O_2 <=> MbO_2$ $K= [MbO_2] / ([Mb][O_2])$ $(=> [MbO_2] =[Mb][O_2](K))$	K=affinity constant. saturation: q = fraction oxygenated = oxygenated/total = $[MbO_2]/\{[Mb]+[MbO_2]\}$ $=> \theta = [Mb][O_2](K) / \{[Mb] + [Mb][O_2](K)\} => \theta = [O_2]/\{(1/K)+[O_2]\}$ (sometimes, θ written as Ψ in other biochem texts) we note that we can simplify the equation further:

when $\theta = 1/2$ (i.e. 50% saturated),
then $0.5 = [O_2]_{1/2} / \{(1/K) + [O_2]_{1/2}\}$
this is true when $(1/K) = [O_2]_{1/2}$ (easily shown)
So, finally: $\theta = [O_2]/\{ [O_2]_{1/2} + [O_2]\}$; the relationship is unchanged if we express the $[O_2]$ in terms of "oxygen tension" (i.e oxygen partial pressure in the gas or the liquid). we have:
$\theta = P_{O2}/\{ P_{50} + P_{O2}\}$
It describes a hyperbola: in the form of $y = x/(a+x)$.
Inspect BINDING CURVE.

P_{50} for Mb is about 4 mm Hg.

For example. What is the saturation of Mb for the case when $pO_2 = 100$ mm Hg (as it is in the lungs; recall O2 = 20% of air, so pAir = 760, pO2 = (.20)(760)=150,)

$\theta = 100/(4+100)=0.96$ (96% saturated!).

In the tissues, what is it? $pO_2 = 30$ mm Hg :
$\theta = 30/(4+30)=0.88$ (88%).
It is binding the O_2less tightly than at the lungs.
(therefore can release some to the tissues).