4.1: Myoglobin, Hemoglobin, and their Ligands

Hemoglobin:

- 1st. protein whose molecular weight was determined
- 1st protein to be assigned a specific function - dioxygen transport
- has a prosthetic group (non-amino acid) heme group (protoporphyrin IX with a ferrous ion) covalently attached to the protein. The heme group binds dioxygen.
- 1st protein in which a point mutation (single base pair charge) causes a single amino acid change in the protein, marking the start of molecular medicine
- 1st protein with high resolution x-ray structure
- theory for dioxygen binding explain control of enzyme activity
- the binding of dioxygen is regulated by binding of H^+, CO_2, and bisphosphoglcyerate which bind to sites (allosteric) distant from oxygen binding site.
- crystals of deoxy-Hb shatter on binding dioxygen, indicating significant conformational changes on binding.

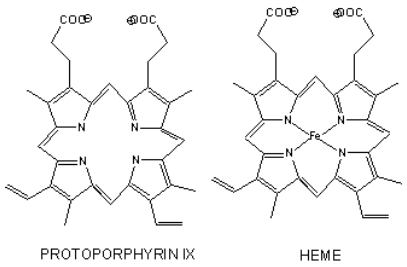

Figure 4.1.1: *The heme group contains protoporphyrin IX, with four tetrapyrrole rings linked by methene bridges. Attached to the tetrapyrrole structure are four methyl, two vinyl, and two proprionate groups. These can be arranged in 15 ways, only one (IX) occurs in biological systems.*

The heme fits into a hydrophobic crevice in the proteins with the proprionate groups exposed to solvent. The Fe^{2+} ion is coordinated to 4 N's on the 4 pyrrole rings, The 5th ligand is a supplied by proximal His (the 8th amino acid on helix F) of the protein. In the absence of dioxygen, the 6 ligand is missing. and the geometry of the complex is square pyramidal with the Fe above the plane of the heme ring. A distal His (E7) is on the opposite side of the heme ring, but too far to coordinate with the Fe. When dioxgen binds, it occupies the 6th coordination site and pulls the Fe into the plane of the ring, leading to octahedral geometry. CO, NO, and H_2S also bind to the 6th site, but with higher affinity than dioxygen, which can lead to CO poisoning. The distal His keeps these ligands (including dioxygen) bound in a bent, non-optimal geometry. This minimizes the chances of CO poisoning.

Myoglobin

- Myoglobin is a relatively small protein that contains 150 amino acids.
- Mb is extremely compact, and consists of 75% alpha helical structure.
- The interior amino acids are almost entirely nonpolar. The only polar amino acids found completely buried are the two His (proximal and distal) found at the active site of dioxygen binding.

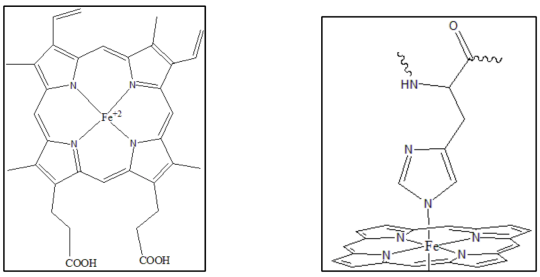

Figure 4.1.2: *The skeletal structure of the heme prosthetic group found within the structure of myoglobin. The porphyrin ring contains four pyrrole nitrogens bound to a ferrous (Fe(II)) ion center. There are six coordination sites in the Fe(II) ion; four are occupied by the pyrrole nitrogens, one is occupied by a proximal histidine, one site can be occupied by a dioxygen molecule (not shown).*

Difference between Hb and Mb

Hb is a tetramer of two α and two β subunits held together by IMF's (an example of quarternary protein structure), and 4 bound hemes, each of which can bind a dioxygen. In a fetus, two other subunits make up Hb (two zeta - ζand two epsilion - ε subunits - analogous to the two αand two β subunits, respectively). This changes in fetuses to two α and two γ subunits. Fetal Hb has higher affinity for dioxygen than adult Hb. Mb is a single polypeptide chains which has higher affinity for dioxygen than Hb.

The α and β chains are similar to Mb, which is unexpected since only 24 of 141 residues in the α and β chains of Hb are identical to amino acids in Mb. This suggests that different sequences can fold to similar structures. The globin fold of Mb and each chain of Hb is common to vertebrates and must be nature's design for dioxygen carriers. A comparison of the sequence of Hb from 60 species show much variability of amino acids, with only 9 identical amino founds. These must be important for structure/function. All internal changes are conservative (e.g. changing a nonpolar for a nonpolar amino acid). Not even Pro's are conserved, suggesting there are different ways to break helices. The two active site His are conserved, as is Gly B6 (required for a reverse turn).
