

Myoglobin and Hemoglobin

Myoglobin (Mb)

Myoglobin, an intracellular heme protein found in most cells that stores O₂ especially in heart and skeletal muscle. Its physiological function is to store O₂ in muscles and facilitate oxygen diffusion because O₂ solubility in aqueous solution is low.

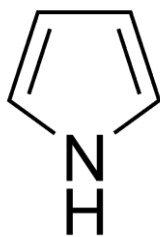
Myoglobin binds the O₂ released by hemoglobin which it stores to meet the demands of muscle contraction. As O₂ is used in the muscle cell for generation of ATP during contraction, it is released from myoglobin and picked up by cytochrome oxidase, a heme-containing enzyme in the electron transport chain that has an even higher affinity for oxygen than myoglobin.

Myoglobin consists of a single polypeptide chain of 153 amino acids attached to a single heme group. About 80% of myoglobin proteins are α helix. It consists of eight α helical segments; these are termed helices A–H. Each helical segment is terminated either by the presence of proline (whose five-membered ring cannot be accommodated in an α -helix) or by β -bends and loops. The eight α helical segments are folded into a globular structure, creating a cradle (box) and within this cradle lay a single heme group and the binding site of O₂. The heme of myoglobin lies between helices E and F.

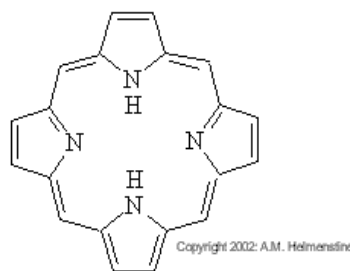
The polypeptide of myoglobin may be viewed as serving three critical functions: 1- it hold the heme group, 2- it protects the heme iron atom from oxidation, and 3- it provides a pocket into which the O₂ can fit.

Structure of heme in myoglobin

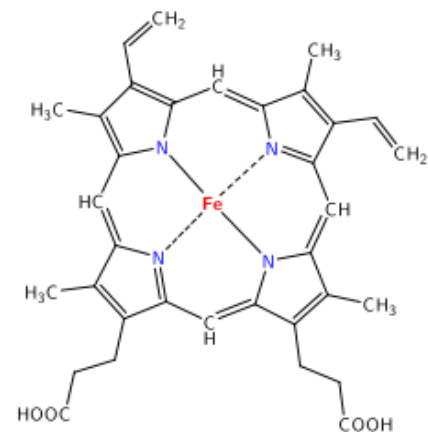
Both myoglobin and hemoglobin have heme. Heme is a complex of porphyrin and ferrous iron (Fe²⁺). Porphyrins are the pigment in red blood cells they are a group of organic compound that have four pyrrole subunits interconnected via α -methylene bridges (=CH-) and are readily bind metals (Fe). A pyrrole ring is a group of four carbon atoms and a nitrogen atom bonded together in a ring (see figure).



Pyrrole ring



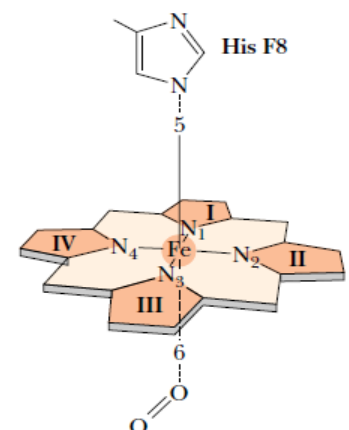
Porphyrin Ring



Heme

The iron is held in the centre of the porphyrin ring. Iron ions prefer to interact with six ligands

- A- Four of the ligands to this iron ion are provided by nitrogen atoms in the pyrrole ring system.
- B- The fifth ligand, on one side of the heme, is provided by a nitrogen atom from the imidazole group of His 93 (proximal histidine) (also known as His F8 the eighth residue of the 'F helix' of myoglobin).



- C- The sixth ligand to iron is provided by molecular oxygen, which binds to the heme group (see figure).
- D- On the oxygen-binding side of the heme lays another histidine residue, His E7 (also known as distal histidine and His 64). While its imidazole function lies too far away to interact with the Fe atom, it is close enough to contact the O₂. His E7 hydrogen bond with O₂. Two hydrophobic side chains on the O₂-binding side of the heme, valine E11 and Phenylalanine CD1 (the first residue in the segment between helices C and D), help hold the heme in place and they are flexible allowing O₂ to enter and exit. Therefore, the O₂- binding site is a sterically hindered region. It helps stabilize the binding of oxygen to the ferrous iron through creating a special microenvironment for the heme.

As said above at the centre of the heme group is the iron +2 metal ion. The nitrogen atoms and oxygen bind to the iron ion through what are called coordinate covalent bonds (dipolar bond). This means that, unlike normal covalent bonds where each atom contributes one electron for the bond, the nitrogen contributes both electrons for the coordinate covalent bond. The oxygen molecule will ultimately bind to this iron ion also using a coordinate covalent bond.

The folding pattern puts hydrophilic amino acid side chains on the outside and buries hydrophobic side chains in the interior, making the protein highly water soluble. The hydrophobic environment in the interior of myoglobin or hemoglobin prevents the oxidation of iron.

The interior contains only nonpolar residues such as Leu, Val, Phe, and Met. The exceptions are His E7 and His F8 that found in the interior which has a function in O₂ binding.

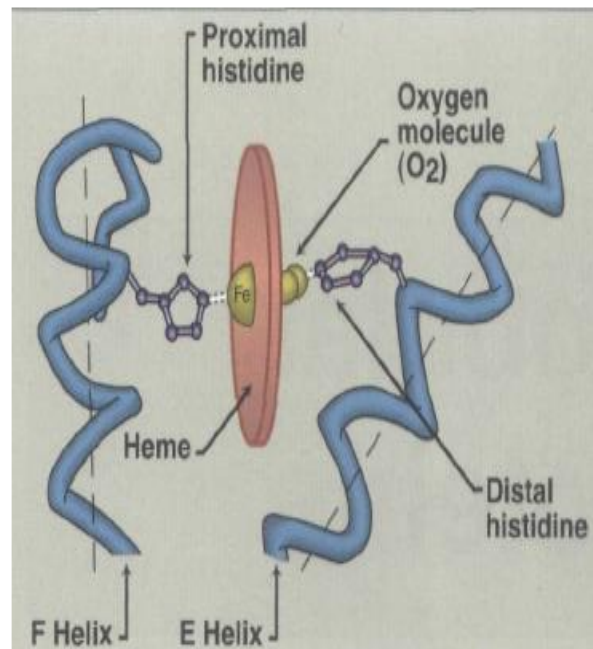
Hemoglobin (Hb)

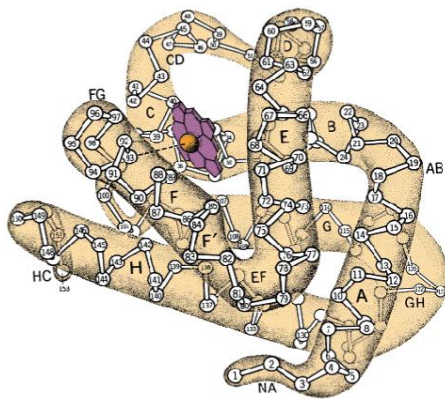
Hemoglobin, the intracellular protein that gives red blood cells their colour. Hemoglobin is responsible for binding oxygen in the lung and transporting the bound oxygen throughout the body where it is used in aerobic metabolic pathways. When the Hemoglobin is carrying an oxygen molecule it is call oxyhemoglobin, when it is not carrying an oxygen molecule it is called deoxyhemoglobin.

Hemoglobin (Hb) is a compact globular protein. It consists of four polypeptide chains, 2 α chains and 2 of β chains, each of which is very similar structurally to the myoglobin polypeptide chain, and each bears a heme group. The α and β subunits differ in primary structure (i.e., they have different sequences of amino acids and are encoded by different genes) and are related to myoglobin. Only about 18% of the residues are identical in myoglobin and in α and β subunit in Hb but the three polypeptides have similar tertiary structures.

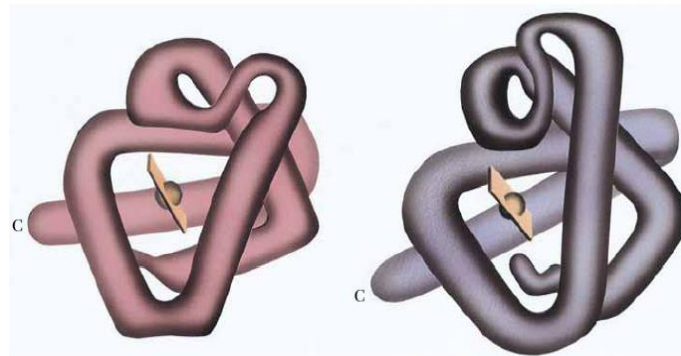
The β chain at 146 amino acid residues is shorter than the myoglobin chain (153 residues), mainly because its final helical segment, the H helix, is shorter.

The α -chain (141 residues) also has a shortened H helix and lacks the D helix (see figure below).





Myoglobin



α- globin in Hb

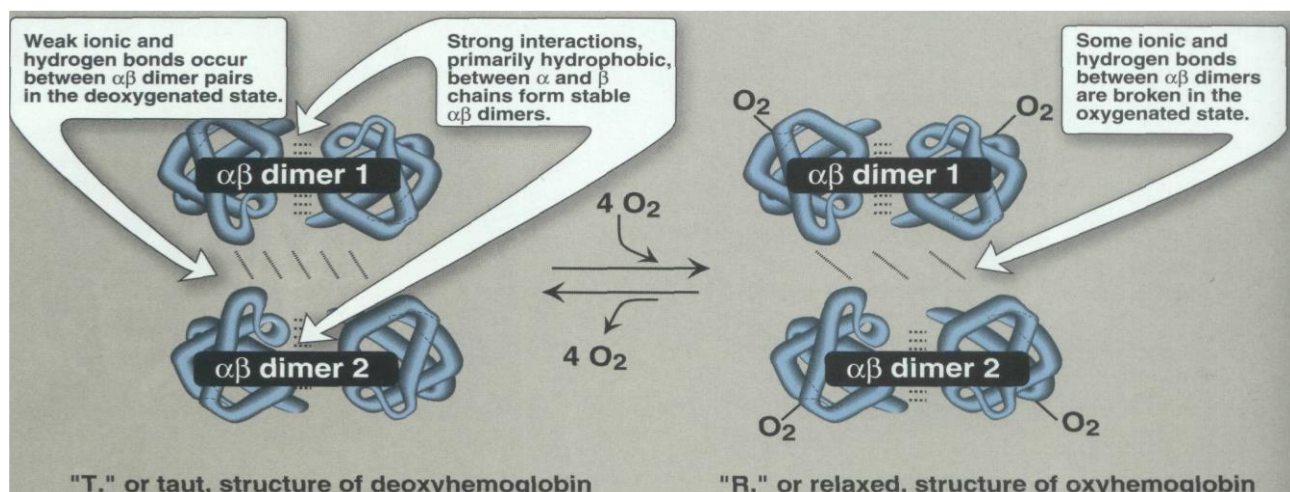
β-globin in Hb

Quaternary structure of hemoglobin: The subunit interactions are between dissimilar chains; $\alpha 1$ associate with $\beta 1$ subunit ($\alpha 1\beta 1$) and $\alpha 2$ associate with $\beta 2$ ($\alpha 2\beta 2$). The hemoglobin tetramer can be visualized as being composed of two identical dimmers, dimer one ($\alpha 1\beta 1$) and dimer two ($\alpha 2\beta 2$). The two polypeptide chains within each dimer are held tightly together, primarily by hydrophobic interactions. While dimer 1 and 2 interact through hydrogen bond and weak ionic bond (called salt bridge).

Hemoglobin exists in to form T & R forms:

a) T form: The deoxy form of hemoglobin is called the "T" or taut (**tense**) form. In the T form, the two $\alpha\beta$ dimmers interact through a network of ionic bonds and hydrogen bonds that constrain the movement of the polypeptide chains. The T form is the **low oxygen-affinity form** of hemoglobin.

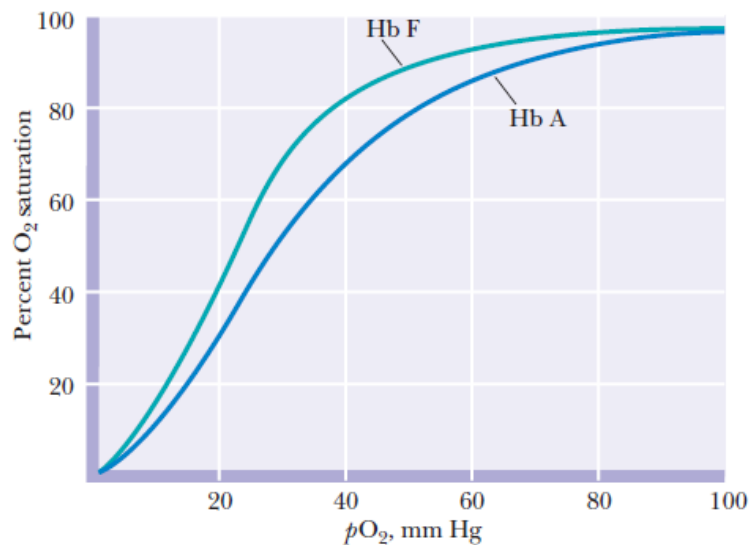
b) R form: The binding of oxygen to hemoglobin causes the rupture of some of the ionic bonds and hydrogen bonds between the $\alpha\beta$ dimmers. This leads to a structure called the "**R,**" or **relaxed** form, in which the polypeptide chains have more freedom of movement. The R form is the **high oxygen-affinity form** of hemoglobin.



RBCs are typically shaped as biconcave disks: flattened and depressed in the centre. Biconcave shape gives them a much greater surface area with which to exchange oxygen and carbon dioxide in the lungs and with body cells besides giving them flexibility to squeeze through tiny capillaries. Red blood cells are able to carry oxygen so efficiently because of haemoglobin.

Fetal Hemoglobin Has a Higher Affinity for O₂ Because It Has a Lower Affinity for BPG

The fetus depends on its mother for an adequate supply of oxygen, but its circulatory system is entirely independent. Gas exchange takes place across the placenta. Ideally fetal Hb should be able to absorb O₂ better than maternal Hb so that an effective transfer of oxygen can occur. Fetal Hb differs from adult Hb in that the β -chains are replaced by very similar, but not identical, 146-residue subunits called γ chains (gamma chains). Fetal Hb is thus $\alpha_2\gamma_2$. 2,3-BPG binds less effectively with the γ chains of fetal Hb (also called Hb F). (Fetal γ chains have Serine (uncharged) instead of Histidine at position 143, and thus lack two of the positive charges in the central BPG-binding cavity). Figure below compares the relative affinities of adult Hb (also known as Hb A) and Hb F for O₂ under similar conditions of pH and [2,3-BPG]. Note that Hb F binds O₂ at pO₂ values where most of the oxygen has dissociated from Hb A. Much of the difference can be attributed to the low capacity of Hb F to bind 2,3-BPG; Hb F thus has an intrinsically greater affinity for O₂, and oxygen transfer from mother to fetus is ensured.



Comparison of the oxygen saturation curves of Hb A and Hb F under similar conditions of pH and 2,3-BPG