Chapter 10

Metalloenzymes & Metalloporphyrins

The natural catalysts in biological systems are enzymes. The enzymes are basically macromolecules with the peptide chains. About 30% of the enzymes are metalloenzymes or metal activated enzymes. The biocatalysts have the main frame work built of proteins, but in metalloenzymes the activity depends on the presence of described metal ion. Thus, the metalloenzymes have two structural components. The protein portion called as apoenzyme and a small non-protein prosthetic group which may be a simple metal ion or a complexed metal ion. Such enzymes having both protein part and non-protein part are called holoenzymes. Protein chain largely regulates the enzymatic activity of metalloenzymes.

Functions of metals in biology:

- Electron transport/transfer.
- Small molecule/atom transport/transfer.
- Bind and activate substrates.
- Stablized a protein structure.

Functions of protein in metalloenzymes.

- Tunes the metal ions properties.
- Usually creates hydrophobic pockets
- Responsible for selectivity in substrate binding.
- Proteins can strain the metal ion geometry their transition state

Properties of metals in metalloenzymes.

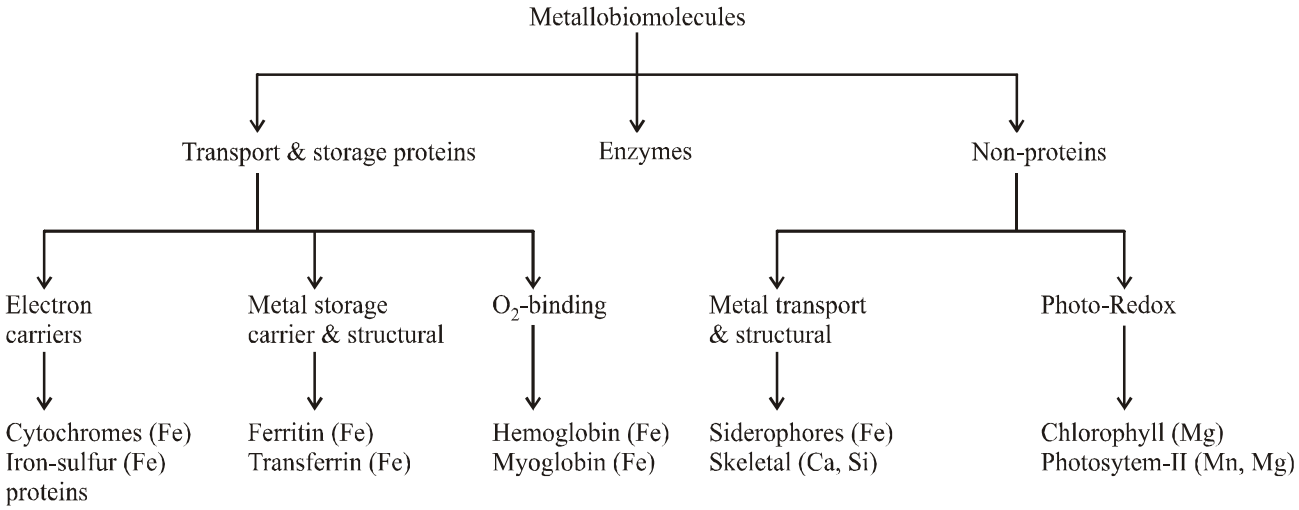
- Catalyses the reaction.
- Metal plus ligands of the active sites of the metalloenzymes.

Some important points regarding in metalloenzymes:

- The metals involve in metalloenzymes are usually light metals predominantly Ca, Mg and first row transitions metals, exceptions metal is Mo.
- These metals are surrounded by amino acid ligand. Normally these are carboxylate (carbonic acid, aspartic acid) Sulphide (cystein) and occasially methionene or nitrogen in histidene.

The enzymes are named after the name of the substrate on which they act. For example, the enzyme that catalyses the peptide hydrolysis is called peptidase. Similarly, ferredoxin reduction is catalysed by ferredoxin reductase.

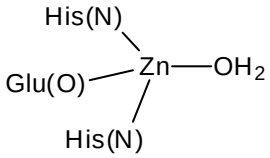
Classification of some Biomolecules containing metal ions



Some metalloenzymes and their functions

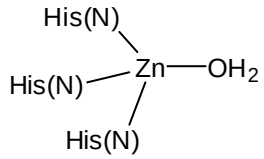
Metal	Enzyme	Biological Functions
Fe	Succinate dehydrogenase **Ribonucleotides reductase** Catalase Cytochrome P-450 Oxygenases	Aerobic oxidation of carbohydrates **DNA synthesis** Protection against H_2O_2 Hydroxylation Oxygen incorporation
Cu	Tyrosinase Amine oxidase Dopamine β hydroxylase Laccase Superoxide dismutase	Skin pigmentation oxidation of amine Hydroxylation of dopamine Oxidation of diphenols to quinones dismutation of superoxide
Zn	Carboxypeptidase Carbonic anhydrase Alcohol dehydrogenase Alkaline phosphatase	Protein digestion Hydration of CO_2 & dehydration of H_2CO_3 Alcohol metabolism Phosphate hydrolysis
Mn	Arginase Pyruvate carboxylase oxaloacetate decarboxylase	Urea formation pyruvate metabolism Decarboxylation
Co	Ribonucleotide reductase Glutamate mutase	DNA biosynthesis Amino acid metabolism
Mg	Hexokinase	Phosphate group transfer
Ca	ATP-ase	Hydrolysis of ATP
Ni	Urease	Urea hydrolysis
Fe and Cu	Cytochrome oxidase	Reduction of O_2 to H_2O
Fe and Mo	Nitrogenase	Nitrogen fixation

10.1. Active sites of metalloenzyme



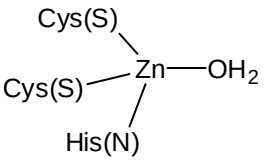
Peptide hydrolysis
(removes terminal amino
acids from proteins)

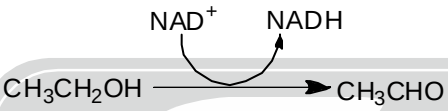
Carboxy peptidase



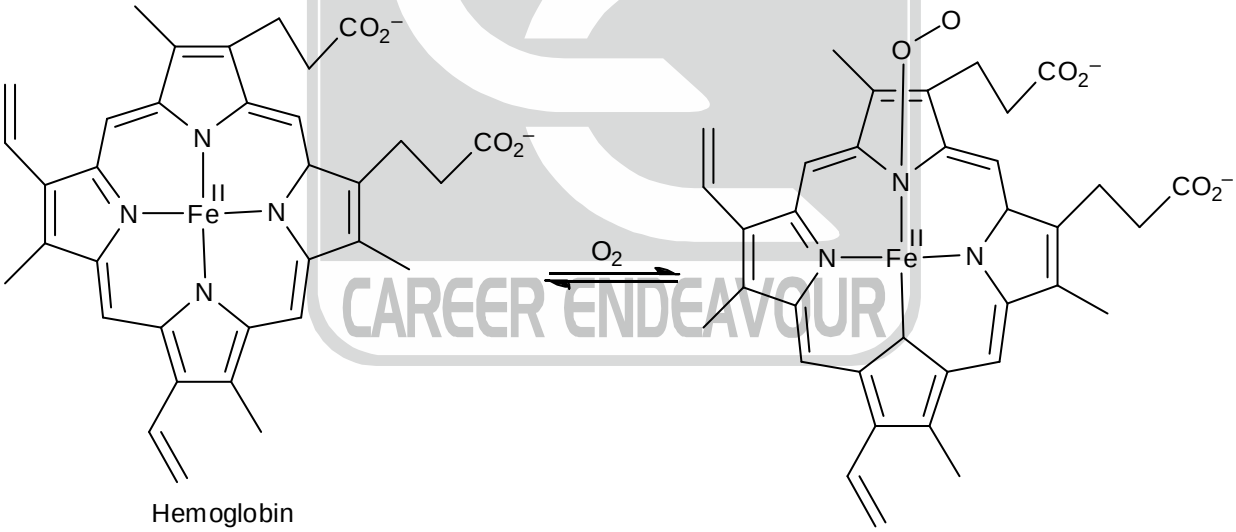
$H_2O + CO \longrightarrow H_2CO_3 \rightleftharpoons H^+ + HCO_3^-$

Carboxy anhydrase

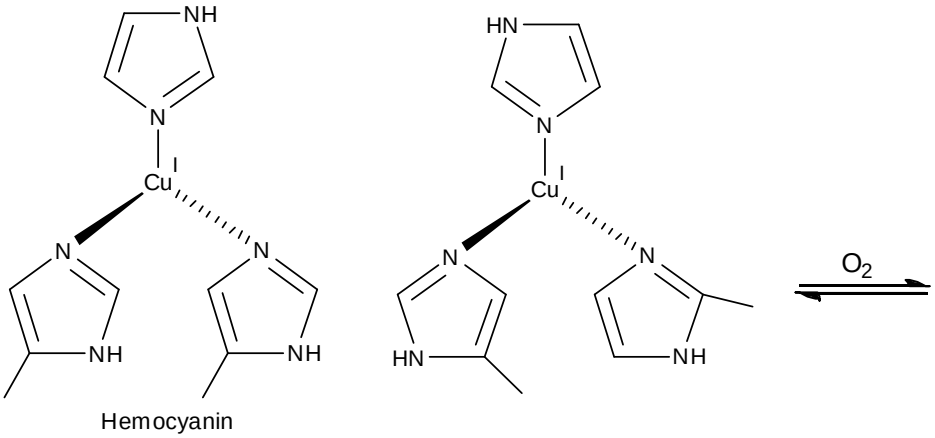




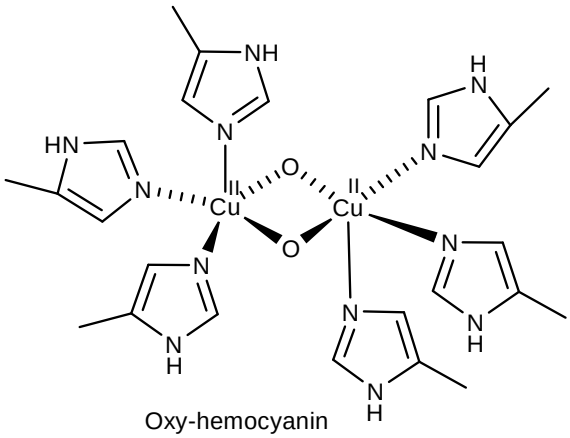
Liver alcohol dehydrogenase



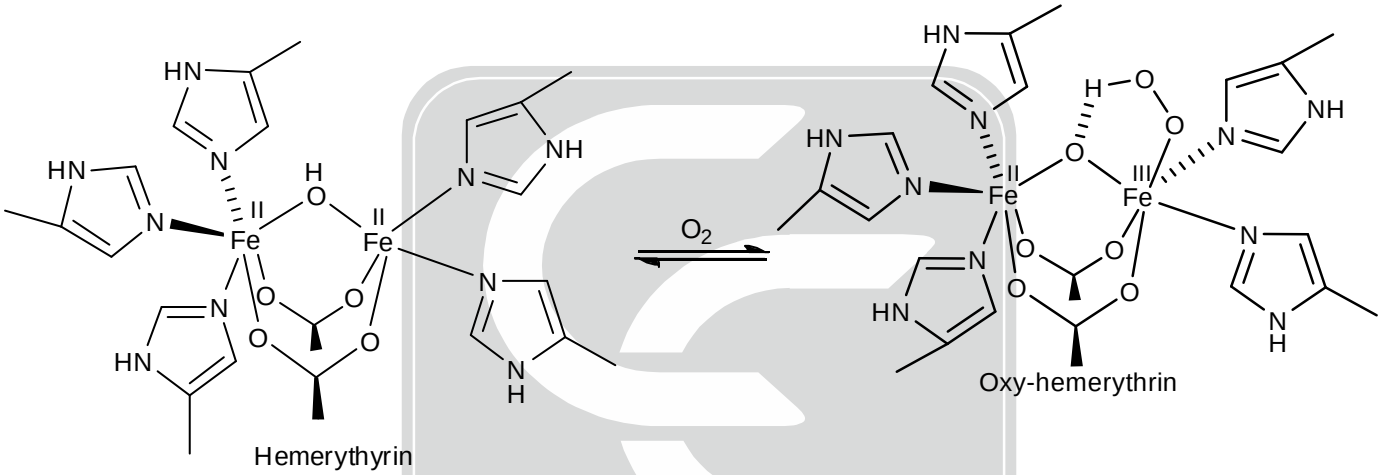
Hemoglobin



Hemocyanin

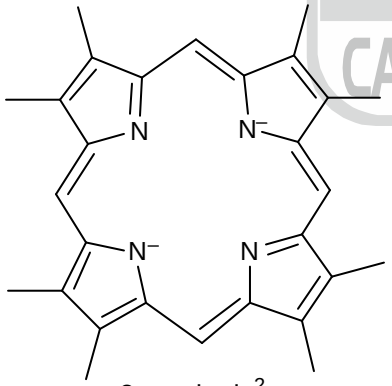


Oxy-hemocyanin



Hemerythyrin

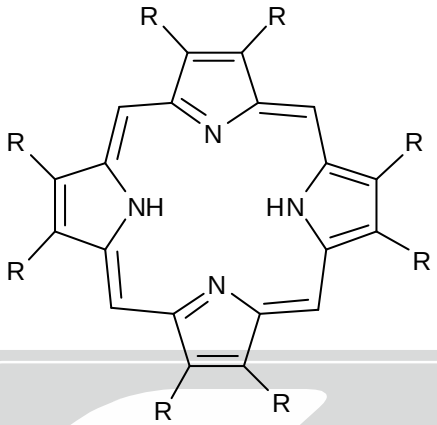
Oxy-hemerythrin



8 porphyrin^{2-}

10.2 PORPHYRINS

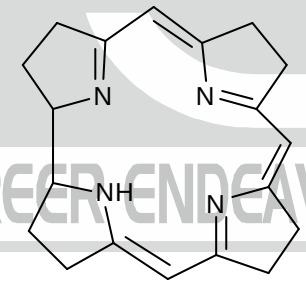
Porphyrins are tetrapyrrole macrocycles with conjugated double bonds and various groups attached to the perimeter. The porphyrins can accept two hydrogen ions to form +2 diacids or donate two protons to form –2 dianions.



(Porphyrin)

variation of substituents facilitates the tuning of electron donating and electron withdrawing ability of the ligand.

The porphyrin and corrin ring systems are of great biological importance. Four pyrrole units are linked by —CH═ bridges as shown in figure but in corrin ring one —CH═ group is less.



(corrin)

The corrin ring has 19 carbons, whereas porphyrins have 20. Also, the pyrrol like rings in corrin are fully saturated "edge-carbon" centre whereas porphyrins are highly conjugated. Because of the high number of sp^3 carbon centre, corrins are more flexible than porphyrins and are not as flat. Porphyrins is aromatic in nature.

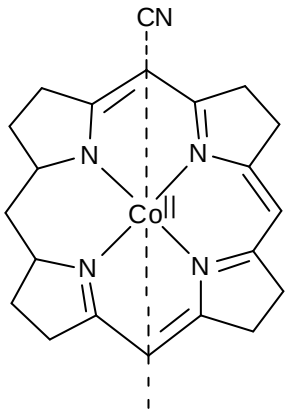
These rings are intensely colored.

Most importance of these rings in the bio-system can be illustrated as:

(i) Iron complex of the substituted porphin is Heme.

(ii) When magnesium lies at the centre of substituted porphin ring; the resulting complex is called chlorophyll.

(iii) If cobalt is the central metal atom of substituted corrin ring system, is called vitamin B_{12} (cobalamine) Cobalt is present in +3 oxidation state in vitamin B_{12}. Vitamin B_{12} is the well known naturally occurring organometallic compound. It is only vitamin known that contain metal. It may be reduced by one electron (vitamin B_{12} *r*) or two electron (vitamin B_{12} *s*) to form cobalt (II) and cobalt (I) oxidation state.



Vitamin B_{12} is the only known essential bio-molecule with a stable metal carbon bond.

$[\text{Vit. } B_{12}\ \text{Co(I)}] + CH_3I \longrightarrow [\text{Vit } B_{12}\ \text{Co(III)}CH_3]^+ I^-$

Characterization of porphyrins:

(i) UV-visible spectroscopy: The free base porphyrin usually have four absorption bands in the range of 500 to 650 nm. In addition to the four bands, there is a band in the range of 400 to 450 nm. This is known as the "soret band" and is a characteristic of all conjugated tetrapyrroles.

(ii) IR-spectroscopy: The NH stretching frequency band appears at 3300 cm^{-1}.

(iii) ^{1}H–NMR spectroscopy : In the ^{1}H–NMR spectroscopy the N–H proton appears at high field due to anisotropic effect. It is generally appears at –2 to –3 ppm.

The metalloporphyrin have no N–H proton signal appear in the NMR spectroscopy.

The UV-visible spectroscopy is one of the best tool for the characterisation of porphyrin.

Porphyrins are found in many metalloenzyme:

	Enzyme	**Function**
Fe-porphyrin	Cytochrome	Electron transfer
Fe-porphyrin	Hemoglobin, Myoglobin	Dioxygen carrier
Mg-porphyrin	Chlorophyll	Photosynthesis

10.3 OXYGEN CARRIER AND STORAGE DEVICES: Dioxygen (O_2) is biologically very much important. The most common mode of reaction of molecular oxygen with transition metal complexes is oxidation. The reaction of dioxygen with a complex so as to incorporate the dioxygen ligand without undergoing any reduction on oxygen is called oxygenation. This is contrast to oxidation reaction in which O_2 looses its identity during the reaction.

Some transition metal complexes act as reversible carriers of molecular O_2 i.e., they take up and release O_2 reversibly as follows:

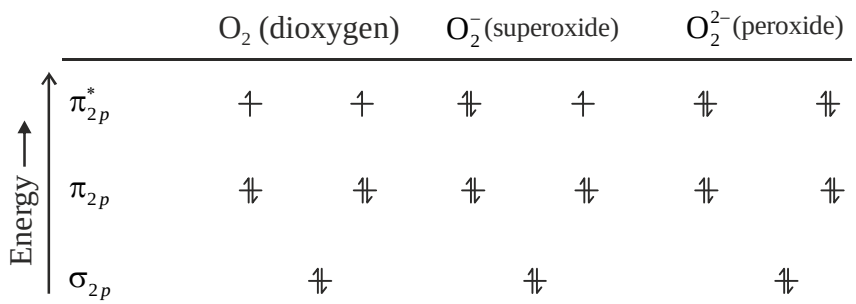
$$\text{Carrier} + O_2 \rightleftharpoons \text{Carrier}\ (O_2)$$

Molecular O_2 can be reduced by one electron process without O – O bond broken and each of these species can act as ligand towards transition metal.

$$\underset{\text{(dioxygen)}}{O_2} \overset{+e}{\underset{-e}{\rightleftharpoons}} \underset{\text{(superoxide)}}{O_2^-} \overset{+e}{\underset{-e}{\rightleftharpoons}} \underset{\text{(peroxide)}}{O_2^{2-}}$$

To understand about oxygen carriers we consider electronic structure of O_2 molecule.

$O_2 = \sigma_{1s^2}\ \sigma^*_{1s^2}\ \sigma_{2s^2}\ \sigma^*_{2s^2}\ \sigma_{2p_x^2}\ \pi_{2p_y^2}\ \pi^*_{2p_z^1}\ \sigma^*_{2p_x^0}\ \pi_{2p_y^2}\ \pi^*_{2p_z^1}$ [Taking *x* axis as internuclear axis]



Bond order	2.0	1.5	1.0
Bond length	1.21	1.28	1.49

IR stretching frequency:

$\nu(O-O)cm^{-1}$	1560	1150–1100	850–740

Iron Storage:

Protein	Ions	Proteins	Ions
Serum albumin	Cu^{+2}, Zn^{+2}	Ferritin	Fe^{3+}
Phosphoprotein	Ca^{+2}	Metallothiene	Cu^{+2}, Zn^{+2}
		Ceruloplasmin	Cu^{+2}, Zn^{+2}

Transport and storage of molecular oxygen is an essential physiological function

Example	Metal	Mole ratio (O_2/metal)	Function	Ligands
Hemoglobin (Hb)	Fe(II)	1/1	Carrier	Porphyrin
Myoglobin (Mb)	Fe(II)	1/1	Storage	Porphyrin
Hemerythrin	Fe(II)	1/2	Storage	Protein
Hemocyanin	Cu(I)	1/2	Carrier	Protein

Iron containing oxygen carrier are present inside the cells and copper containing oxygen carrier are found in extracellular fluids.

Both heme and non-heme iron protein are involved in oxygen transport and storage. Heme O_2 carrier are responsible for red colour of human blood, while the blue pigment of crab blood, hemocyanin. Some marine worms have a violet colour, which is due to presence of non-heme iron-protein hemerythrein.

10.4. Hemoglobin (Hb) and Myoglobin (Mb) in oxygen transport mechanism:

Hemoglobin occurs in all vertebrates (with certain exception) and in many invertebrates, it has also been found in certain strains of yeasts, mould etc.

Fe-porphyrin referred to as heme is the prosthetic group of hemoglobin (Hb) and Myoglobin (Mb). Each Hemoglobin (Hb) molecule has four heme groups bound to the globin on its surface. On each heme unit of Hemoglobin (Hb) the four square-planer coordination sites of the Fe(II) are occupied by porphyrin nitrogen atom. Whereas the fifth coordination site of iron occupied by the nitrogen of histidine (distal

histidine) ligand of the protein chain i.e. globin chain, and the sixth position of iron remains vacant or occupied by H_2O in their deoxy forms and this site is occupied by O_2 in their oxy forms. The distal histidine protects Hb and Mb-form CO poisoning. Myoglobin (Mb) is a monomer having only one heme unit. Hemoglobin and myoglobin are transport and storage of dioxygen respectively.

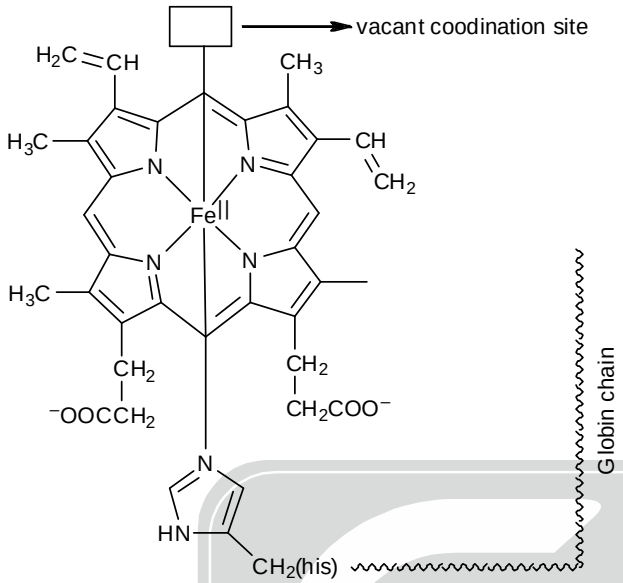


Fig. Structure of a heme unit in hemoglobin and myoglobin

The planer porphyrin ring of heme unit of Hb and Mb, due to the presence of conjugated double bonds in the porphyrin, stable π and low lying π^* orbital are available and these allow the characteristic charge transfer electronic transition to give the red colour of blood.

10.5. Function of Hb and Mb: Hemoglobin (Hb carries O_2 from lungs to tissues where it is transferred to myoglobin (Mb) and stored therein for metabolic requirements. To make this process thermodynamically possible, the oxygen affinity of Hb in lungs where oxygen concentration is high should be greater than that of Mb and reverse condition should arise in the tissue where oxygen concentration is less.

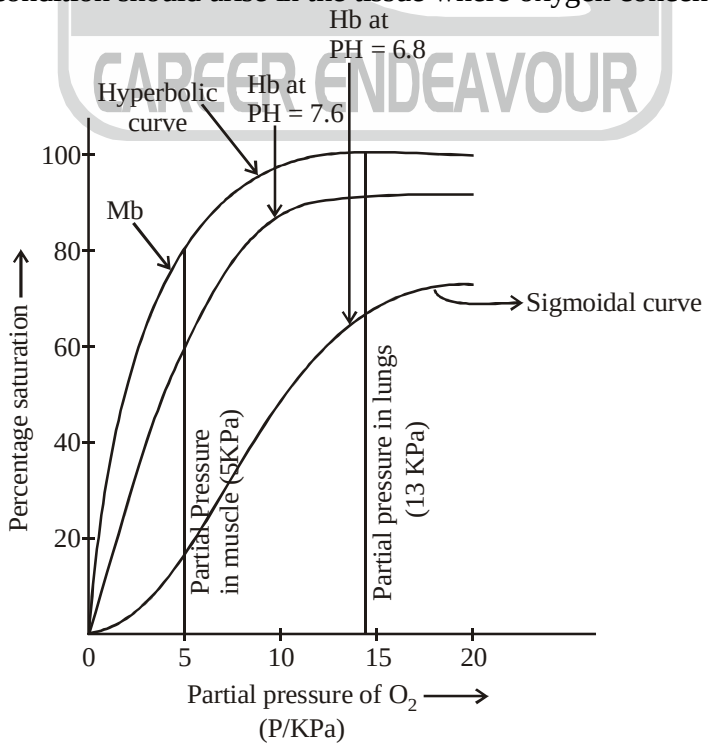


Fig. Oxygenation curves for Hb and Mb

Figure show that Hb is about as good on O_2 binder as Mb at high O_2 pressure. It is much poorer at the lower pressures prevailing in muscle and hence passes its oxygen on to Mb as required. Moreover, the need for O_2 will be greatest in tissues where O_2 is consumed followed by production of CO_2. The CO_2 lowers the pH, thus causing the Hb to release even more oxygen to Mb. The pH sensitivity (Bohr effect) as well as the progressive increase of O_2 binding constants in Hb are due to the interaction between the subunits, Mb behaves more simple because it consists of only one unit. Thus, each of the two is essential in the complete oxygen transport process.

10.5. Oxygenation equilibria of haemoglobin (Hb) and myoglobin (Mb)

Both Hb and Mb bind with oxygen but first one act as carrier and later one as storage device in human body. It raises interesting point why one act as carrier and other as storage device while both have same heme unit. The difference lies on structure of both Hb and Mb and on the environment of the human body where these have to show their function. Hb consists of four unit of heme group while Mb has single heme unit. Heme unit is embedded in hydrophobic proteins environment (Globin proteins) in both biological devices. The hydrophobic protein environment protects the oxidation of Fe^{+2} to Fe^{+3}. In case of Hb system, four heme units cooperate to each other and adjust their structure according to their comfort. This behaviour is probably due presence of α-helix of proteins which act like a spring and connecting to these heme units. In Hb as the one O_2 binds, the molecular shape changes such a way that those additional O_2 molecules could bind. This is a kind of communication among four heme units of Hb makes O_2 binding process very easier. It is interesting that the four equilibrium constants (K_1, K_2, K_3, K_4) respective to four step reaction equilibria become in increasing order ($K_1 < K_2 < K_3 < K_4$) while one expect in reverse order. Not only this, even K_1 (5 to 60) is much smaller than K_4 (3000 to 6000).

$$Hb + O_2 \rightleftharpoons HbO_2 \qquad K_1 = 5 \text{ to } 60$$

$$Hb(O_2) + O_2 \rightleftharpoons HbO_2$$

$$Hb(O_2)_2 + O_2 \rightleftharpoons Hb(O_2)_3$$

$$Hb(O_2)_3 + O_2 \rightleftharpoons Hb(O_2)_4 \qquad K_4 = 3000 \text{ to } 6000$$

In similar way as one O_2 get removed from Hb, it triggers the release of remainder O_2 molecules. This whole phenomenon is called as "cooperative effect." The removal of oxygen is also favoured by pH change. When CO_2 concentration in capillaries is enhanced, it supports the following reaction in forward direction which increase the acidity.

$$2H_2O + CO_2 \rightleftharpoons HCO_3^- + H_3O^+$$

This decreased in pH favours the release of O_2 from Hb. This effect is called as Bohr Effect. These are two important factors which make labile binding of O_2 and easier removal. In the Hb and supports the carrier activity of Hb. Myoglobin has one heme group per molecule and serves as storage in muscles but binding mechanism of O_2 is similar to Hb.

$$Mb + O_2 \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} MbO_2$$

$$K_1 = \frac{[MbO_2]}{[Mb][O_2]} \qquad \ldots (1)$$

Saturation, $$\theta = \frac{[MbO_2]}{[Mb]_{total}} \qquad \ldots (2)$$

$$[Mb]_{total} = [Mb] + [MbO_2]$$

$[Mb]_{total} = [Mb] + K_I[Mb][O_2]$ from equation (1) by puting the value of $[MbO_2]$

$$[Mb]_{total} = [Mb](1 + K_I[O_2])$$

Substituting the value of $[MbO_2]$ in equation (2).

$$\theta = \frac{K_I[Mb][O_2]}{[Mb](1 + K_I[O_2])}$$

$$\theta = \frac{K_I[O_2]}{1 + K_I[O_2]} \quad \text{... (3)}$$

$$1 - \theta = \frac{1}{1 + K_I[O_2]} \quad \text{... (4)}$$

Dividing (3) by equation (4)

$$\frac{\theta}{1-\theta} = K_I[O_2]$$

This equation of is for Mb which is containing only one heme unit. Oxygenation curve of Hb may be approximated as $\frac{\theta}{1-\theta} = K_I[O_2]^n$ with $n \sim 3$

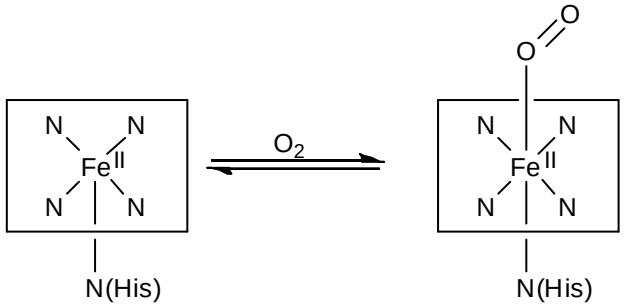
This equation is known as Hill equation and the exponent, n is called the Hill constant.

The data obtained from Hb between 10 and 90% oxygenation can be fitted to the Hill equation to give values of n ~ 3 for normal Hb. The form of oxygenation curve and that the fact if fits n>1 indicate that there is cooperative interaction between the subunits. The addition of oxygen to a subunit affects the oxygen affinities of other subunits, this is an example of Allosteric effect (Entatic effect) literally means "a stretched state or state of being under tension". n<4 indicates that the cooperative interaction between heme units is rather moderate.

The cooperative interaction where the binding of one molecule of a substance influences the binding of next molecule of the same kind is described to as a homotropic allosteric interaction.

Mechanism of homotropic allosteric effect of O_2 in Hb: On oxygenation, a trigger mechanism proposed by perutz operates through the heme-heme interaction to carry out the change, T-form to R-form.

To this change, researcher suggested that in deoxy-Hb, Fe(II) remains in high spin state, but on oxygenation Fe(II) attains the low spin state. This spin state change acts as the trigger.



Tensed form (T)
(Deoxyhemoglobin)
High spin, ESR active
t_{2g}^4 e_g^2 (paramagnetic)

Relaxed form (R)
(Oxyhemoglobin)
Low spin, ESR inactive
t_{2g}^6 e_g^0 (diamagnetic)

The Fe(II) – N bond length (high spin Fe(II) and Sp^2 N) is ~218pm. The e_g electrons directly interact with the ligands and thus the removal of e_g electrons in attaining the low spin state reduces the bond length. The size of porphyrin cavity allows the sitting of the metal having the M–N bond length ~200-205 pm. Thus the high spin Fe(II) is deoxy Hb cannot sit in the porphyrin cavity.

In fact, the size of Fe^{2+} increase by 28% on going from

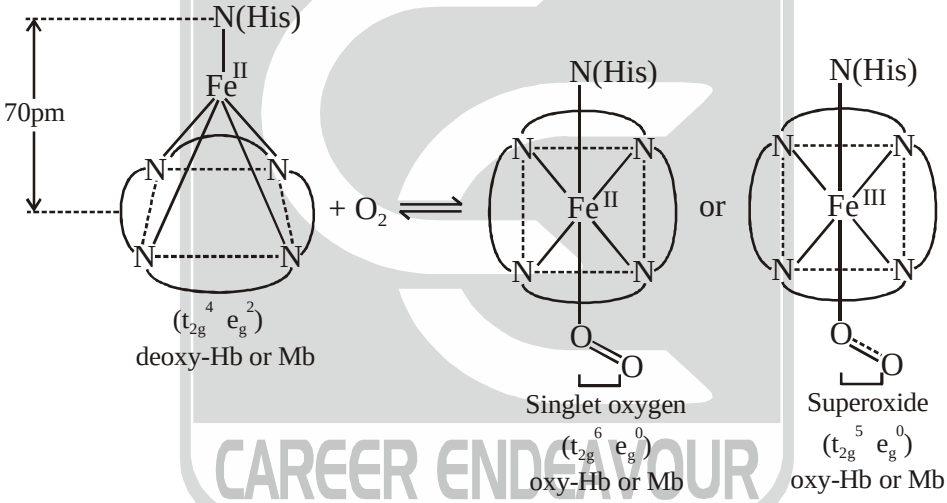
Low spin (oxyhemoglobin) (0.61Å)

↓ to

High spin (deoxy Hb) (0.78Å)

The globular protein prevents the irreversible oxidation of Fe(II) to Fe(III).

Here, it should be pointed out that there are evidences to support that the oxygenated form of Hb, iron exists as Fe(III) and O_2 as O_2^- (superoxide) because of the, O–O stretching frequency ~1106cm^{-1} is close to that of O_2^- ($\nu_{O-O} = 1097 cm^{-1}$). The Fe – O – O bond angle is close to 120°. In fact, Fe(II) is reversible oxidised to Fe(III) in this O_2^- uptake process and at the site of delivery of O_2, it again attains the Fe(II) state through deoxygenation.



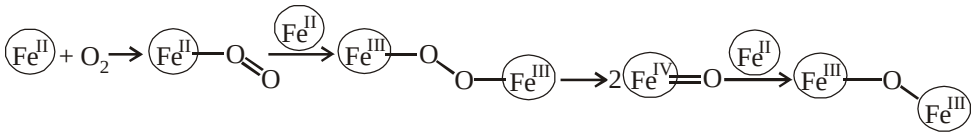
Here, it is important point out the fact that O_2 is not a very strong field ligand and consequently it is not expected to carry out the desired spin pairing on oxygenation of Hb. It is believed that the heme unit is properly tuned by its substituents to make the spin pairing easier. Thus, the oxy forms of Hb and Mb, O_2 remains as a singlet form. The singlet O_2 is a good π-acceptor and it can act as a fairly strong field ligand to induce the spin pairing in Fe(II).

10.6. Poisoning towards hemoglobin and myoglobin: Different π-acid ligands like CO, NO, PF_3 etc. which are electrically neutral and not much bulky can competitively replace O_2 from the sixth octahedral site of Hb and Mb. Consequently, the O_2 transport mechanism gets arrested and toxicity arises. CN^- may also bind the site, but the heme pocket surrounded by the hydrophobic environment does not welcome CN^- much. The σ-bonding ligands like NH_3 and amines may also block the oxygenation site.

The CN^- actually blocks the cytochrome C oxidase involved in the respiratory chain. To remove the bound CN^- from cytochrome C oxidase, some methe-Hb (oxy-hemoglobin) are to be generated either by inhalation of amylnitrite vapour or by injection of $NaNO_2$ solution. Met-Hb bearing Fe can bind CN^- more strongly than the cytochrome C oxidase. Consequently, CN^- removed from the respiratory chain to regenerate the electron tunneling path.

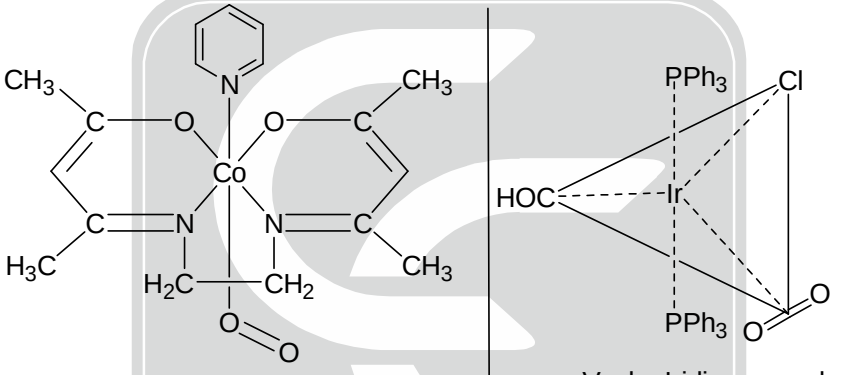
10.7. Importance of globulin (protein chain):

- Allosteric effect of O_2, H^+, CO_2, Cl^- and diphosphoglycerate (DPG) on O_2 affinity of Hb.
- Protection of Hb and Mb from irreversible oxidation by O_2
- Weakening the interaction of CO with the heme and stabilising the binding of O_2 by the histidine residue.
- σ-donor properties of the imidazole moitey of the proximal histidine stabilise the O_2 binding by using the π-acceptance property of O_2.
- Protonation and deprotonation sites in globin protein chain are important in maintaining the biological pH and CO_2 transport.
- Prevent the irreversible μ-oxo complex formation.



μ-oxo complex

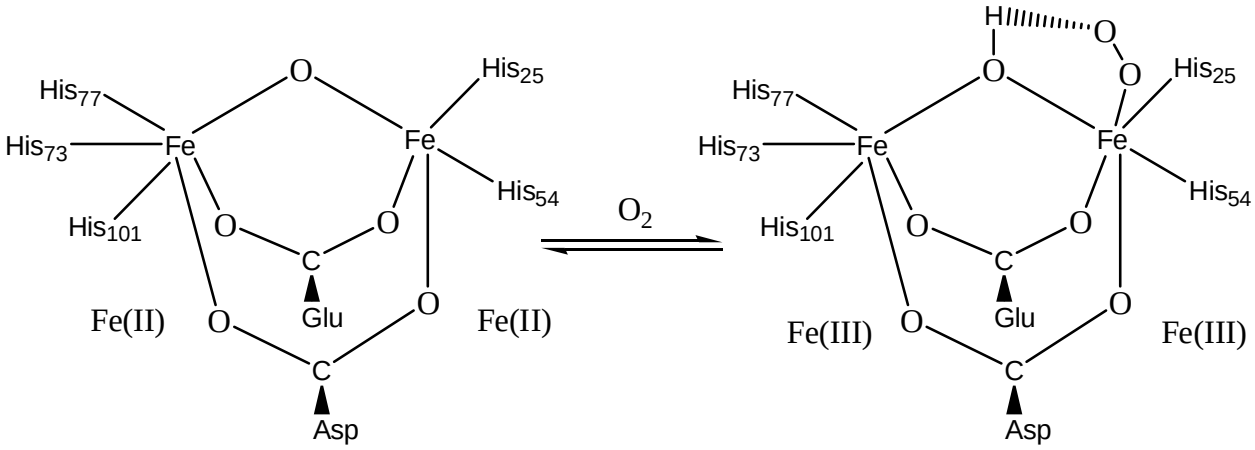
Some synthetic complexes as such Co (acacen) complex and (triphenylphosphine) iridium known as vaska's Iridium complex show direct oxygenation and help to understand oxygen binding in biological system.



Co(acacen) complex Vaska Iridium complex

10.8. Hemerythrin: (An oxygen uptake metalloprotein)

- Non heme iron protein.
- Fe(II) oxidation state.
- It consists of eight indentical units each containing two iron atom.
- Molecular weight 108,000.

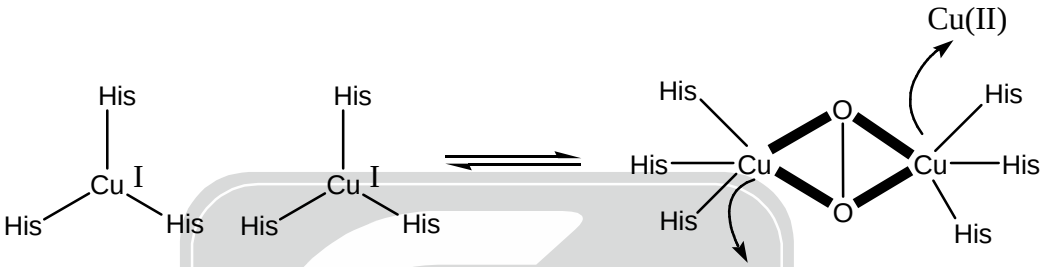


deoxy hemerythrin (Colourless)

oxy hemerythrin (Violet-red)

- Deoxyhemerythrin is paramagnetic and high spin.
- The environment of iron atom is pseudo octahedral.
- Each dioxygen binding site contains Fe(II) atom and the reaction takes place via redox reaction to form Fe(II) peroxide O_2^{2-}.
- The antiferromagnetic coupling of two Fe gives rise to diamagnetism of oxy hemerythrins at low temperature.
- The oxyhemerythrin has lower magnetic moment at room temperature and it becomes diamagnetic at 1.4 to 4.2 k.
- The Raman spectrum of oxy (^{16}O ^{18}O) hemerythrin, which shows the two oxygen atoms to be in nonequivalent positions.

10.9. Hemocyanin: (An oxygen uptake protein)



Cu(I) distorted. T-shape inormous strain at the oxygen site oxyhemocyanin.

Oxy-hemocyanin

- Oxygenation provides copper with normal four co-ordinated structure and remove a strain.
- Deoxyhemocyanin is diamagnetic, colourless, in which both the Cu atom (+1) acid state.
- Oxyhemocyanin is blue.
- Oxyforms may have following resonating structure.

$$Cu^{+}O_2Cu^{+} \longleftrightarrow Cu^{2+}O_2^{-}Cu^{+} \longleftrightarrow Cu^{+}O_2^{-}Cu^{2+} \longleftrightarrow Cu^{2+}O_2^{2-}Cu^{2+}$$

- Raman spectrum of oxyhemocyanin indicated a $Cu^{2+}O_2^{2-}Cu^{2+}$. This one structure of the oxygenated active site and diamagnetism might be due to anti-ferromagnetic interaction between the Cu(II).
- Both d-d transition and charge transfer responsible for colour spectra (blue)
- The copper is in the +1 oxidation state in the deoxy form and +2 in the oxy form.

PROBLEMS

1. Storage of iron in the body is carried out by
   (a) Hemoglobin (b) Ferritin
   (c) Cytochrome P450 (d) Myoglobin
2. The metal present at the active site of the protein carboxypeptidase A is
   (a) zinc (b) Molybdenum
   (c) Magnesium (d) Cobalt
3. Of the following metal that is present in the respiratory protein hemocyanin is
   (a) V (b) Fe (c) Mo (d) Cu
4. The reaction catalysed by superoxide dismutase is
   (a) hydrogenation (b) disproportationation
   (c) deiodination (d) bromination

5. Match the dioxygen binding protein in column-I with the appropriate species bound to them listed in column-II

Column-I	**Column-II**
P : Myoglobin	1: $[O_2]^{2-}$
Q: Hemocyanin	2: $[HO_2]^-$
R: Hemerythrin	3: $[O_2^-]$

   (a) P–1, Q–2, R–3 (b) P–3, Q–1, R–2 (c) P–2, Q–3, R–1 (d) P–3, Q–2, R–1
6. The metal present at the active site of the protein carbonic anhydrase is
   (a) Mo (b) Co (c) Zn (d) Pt
7. Non-heme-iron-sulfur proteins are involved in
   (a) Electron transfer (b) Proton transfer
   (c) Both (a) and (b) (d) Oxygen transfer
8. Which one of the following statements for hemoglobin is NOT correct
   (a) The binding with O_2 is weaker in comparison with myoglobin
   (b) Iron is 5-coordinated
   (c) Iron is coplaner with the porphyrin ring in the absence of oxygen
   (d) The oxidation state of iron is +2
9. The metalloenzyme involved in the key step of the skin pigment melanin formation is
   (a) superoxida dismutase (b) Myoglobin
   (c) Tyrosinase (d) Nitrogenase
10. Hemocyanin contains
   (a) a dinuclear copper core and binds dioxygen in the cupric state
   (b) a dinuclear copper core and binds dioxygen in the cuprous state
   (c) a mononuclear copper core and binds dioxygen in the cuprous state
   (d) a mononuclear copper core and binds dioxygen in the cupric state
11. The biological role of cytochrome P-450 is
   (a) Nitrogen fixation (b) Oxidation of RH to ROH
   (c) Amide hydrolysis (d) Oxidation of amino acid

12. Which of the following metalloprotein does not have iron in the active site?
(a) Hemoglobin (b) Hemerythrin (c) Hemocyanin (d) Cytochrome-C

13. Storage of iron in the body is carried out by
(a) hemoglobin (b) ferritin
(c) cytochrome P-450 (d) Myoglobin

14. Myoglobin is a
(a) catalyst for epoxidation reaction (b) component in photosynthetic system
(c) Nitrogen fixation enzyme (d) Di-oxygen binding metalloprotein

15. The quaternary structure of human hemoglobin is best described as a
(a) Dimer of two myoglobin dimers (b) Tetramer of identical subunits
(c) Tetramer of two different subunits (d) Tetramer of four different subunits

16. The role of Cu in hemocyanin is
(a) CO_2 fixation (b) N_2 fixation
(c) O_2- binding and transport (d) Epoxidation of olefins

17. In the transformation of oxyhemoglobin to deoxyhemoglobin
(a) Fe^{2+} in the low spin state changes to Fe^{+2} in the high spin state
(b) Fe^{+2} in the low spin state changes to Fe^{+3} in the low spin state
(c) Fe^{+2} in the high spin state changes to Fe^{+2} in the low spin state
(d) Fe^{+2} in the high spin state changes to Fe^{3+} in the high spin state

18. Hemerythrin belong to the group of
(a) non-heme iron protein (b) binuclear copper protein
(c) heme-iron protein (d) non-heme-non iron protein

19. Which of the following metalloproteins converts oxygen to water
(a) catalase (b) cytochrome oxidase
(c) haloperoxidase (d) hemoglobin

20. Heme is a versatile molecule present in a large number of metalloproteins and enzymes. The absorption spectrum of heme is characterized by sharp band at near 400nm and weaker band at 520-550 nm. The origin of these absorption bonds are
(a) MLCT-Transition (b) LMCT-Transition
(c) *d–d* Transition (d) $\pi - \pi^*$ Transition

ANSWER KEY

1. (b)	**2.** (a)	**3.** (d)	**4.** (b)	**5.** (c)
6. (c)	**7.** (a)	**8.** (c)	**9.** (c)	**10.** (a)
11. (b)	**12.** (c)	**13.** (b)	**14.** (d)	**15.** (b)
16. (c)	**17.** (a)	**18.** (a)	**19.** (b)	**20.** (a)