KHATRA ADIBASI MAHAVIDYALAYA

E-Content

Department: Chemistry

Semester: VI (Honours)

Session: 2023-2024

Subject: Bio-inorganic Chemistry (CHEM/601/C-13)

Topic: Bio-inorganic Chemistry-1

Name of Teacher: Soumen Rakshit

Elements in the living systems

❖ Bulk / Constitutional elements

The elements that are required in relatively large amount; $1\text{-}10^4$ g mole in 75 kg adult human body.

Most abundant

H = 63%

O = 25.5%

C = 9.5%

N = 1.4%

Others

Ca

P

Na

K

S

Cl

Mg

Total = 11

Elements in the living systems

- **Essential elements**

The elements which have specific essential role to sustain life.

Elements:

Na K Mg Ca V Cr Mn

Fe Co Ni Cu Zn Mo Cd

Elements in the living systems

- **Beneficial elements**

The elements that are not required by all plants but can promote plants growth; and may be essential for a perticular taxa.

Elements:

Li Cr Ni Sn V W

Elements in the living systems

Trace elements

The elements that are present in trace amount ranging from 10^{-1} to 10^{-4} gm/molein 75 kg adult human body.

Elements:

Mo Mn Fe Co Cu Zn I

Ultra-trace elements

The elements that are present in trace amount ranging lower than 10^{-4} gm/mole in 75 kg adult human body.

Elements:

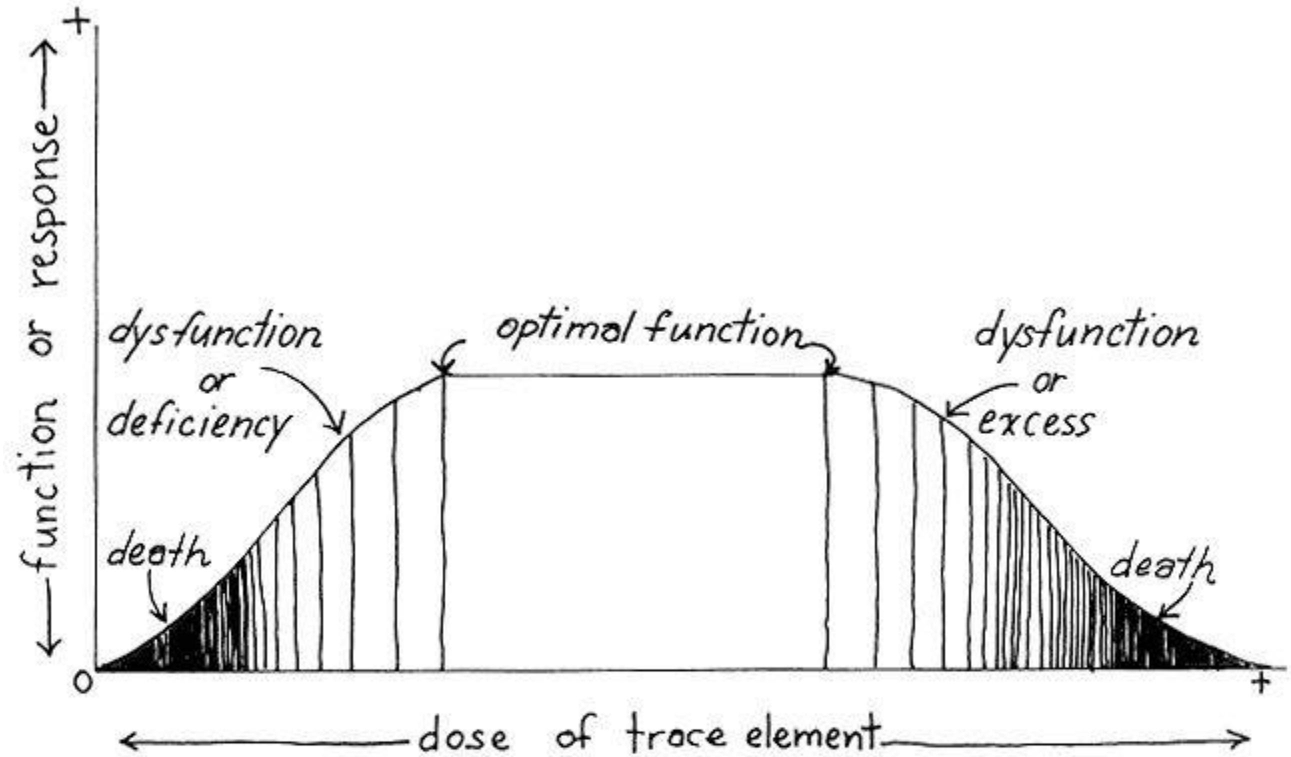
Li Cr Si Sn V W Br Se

Recommended daily dose for ~70 kg adult human body

Elements	Intake (per day)
Fe	10 mg (Male) 18 mg (Female)
Zn	15 mg
Mn	2.5-5 mg
Cu	2-3 mg
F	1.5-4 mg
Mo	150-500 µg
I	150 µg
Cr	50-200 µg
Se	50-200 µg

Deficiency or excess of these elements may give metabolic disorders that inhibit growth or may cause diseases.

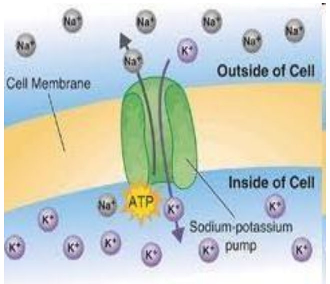
Variation of response with increasing dose of essential elements



Role of metals in biological systems

11
Na
22.98

19
K
39.09



Sodium potassium pump

(1/5th of all the ATP used)

Function

- Transmit nerve signal through electrical potential gradient.
- Regulate the correct osmotic pressure of blood

Deficiency

Na: Hyponatremia (lowering blood pressure & circulatory failure)

K: Epilepsy

20
Ca
40.08

Function

- Major constituent of bone and teeth.
- Blood coagulation

Deficiency

Ca: Rickets, steomalacia, Osteoporosis

12
Mg
24.31

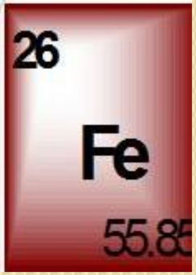
Function

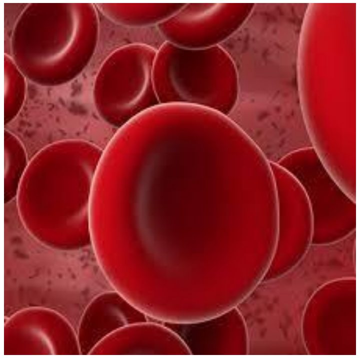
- Constitutional element as prosthetic group in chlorophyll.
- Activator of enzyme which utilize ATP
- Control muscle contraction, acts as co factor
- Intracelluler Mg2+ keeps ribosomes intact ; stabilizing DNA and RNA structure

Deficiency

Mg: Convulsion & neuromuacular irritation, Tremors and spasm

Role of metals in biological systems





Function

O2 transport/storage protein

Hb, Mb, Hemoerythrin

Iron storage protein

Ferritin

e transfer protein

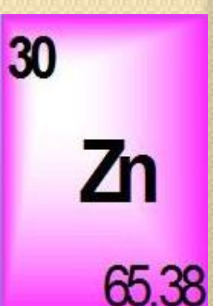
Ferredoxins, cytochrome

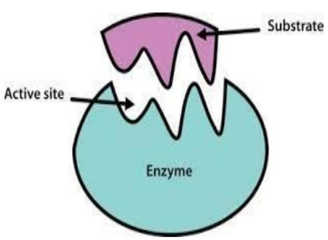
Enzyme

Cytocrome c oxidase, catalase, peroxidase, aconitase, nitrogenase

Deficiency

Fe: Anemia





Function

Enzyme

Carbonic anhydrases, carboxyl peptidases, alkaline phosphatage

Enzyme activator

Enolage, Peptidase, arginase, histidine deaminases

Deficiency

Zn: Dwarfism, hypogonadism

Role of metals in biological systems

27
Co
58.94

Function

- Vitamin B_{12} co-enzymes
- Required in enzyme as glutamate mutase, ribonucleotide reductase

Deficiency

Co: Pernicious Anemia

42
Mo
Molybdenum
95.94

Function

Enzyme

Nitrogenases, hydrogenase, Nitrate reductase, xanthine oxidase, sulphite oxidase

Deficiency

Mo: Plant growth disorder

29
Cu
63.55

Function

O2 transport/storage protein

Hemocyanin

Cu storage protein

Ceruloplasm

Enzyme

Cytocrome c oxidase, Ascorbic acid oxidase

Deficiency

Cu:Anemia, Menke's disease, demineralization of boanes, decolouration of skin & boan

Role of metals in biological systems

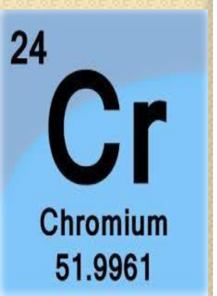
Function

☐Plant growth factor

☐O_2 transport in some lower form of life; hemovanadins

Deficiency

V: increase blood fat and colesterol level

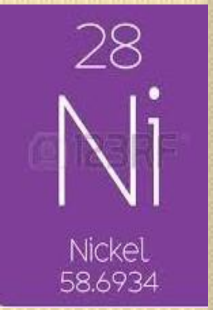


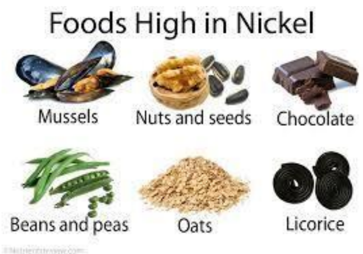
Function

☐Glucose tolerence factor

Deficiency

Cr: Suspected to cause Diabetes





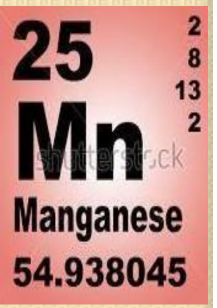
Function

Enzyme

Urease, methanogenic bacteria factor F430

Deficiency

Ni: whole leaf chlorosis along with necrotic leaf tips (accumulation of toxic levels of urea)



Function

Metalloenzyme

Pyruvate kinase, pyruvate carboxylase

Enzyme activator

Phosphoenol pyruvate, photosynthetic system

Deficiency

Mn: impaired ,reproductive function, skeletal abnormalities, impaired gluco tolerance

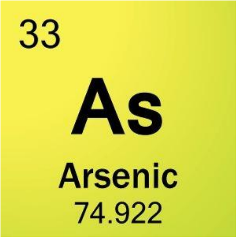
Toxic elements

- Most common toxic elements are
  Arsenic(As)
  Lead(Pb),
  Mercury(Hg),
  Cadmium(Cd)
  Copper (Cu)
  Beryllium (Be)
  Manganese (Mn)
  Selenium (Se)
  Tin(Sn)
  Antimony (Sb)

- Indoor concentration of heavy metals is generally less than their outdoor concentration

- They are mainly produced by industrial activities, and deposit slowly in the surrounding water and soil

Effect of toxic elements

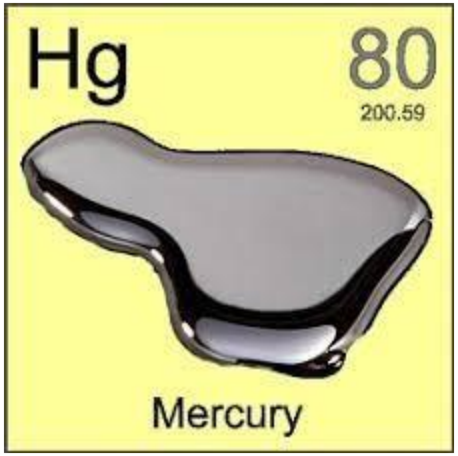


Toxic effects of As

- Birth defects
- Carcinogen:

  Lung cancer results from the inhalation of arsenic and probably also from its ingestion. Skin and liver cancer, and perhaps cancers of the bladder and kidneys, arise from ingested arsenic
- Gastrointestinal damage
- Severe vomiting
- Diarrhea
- Death

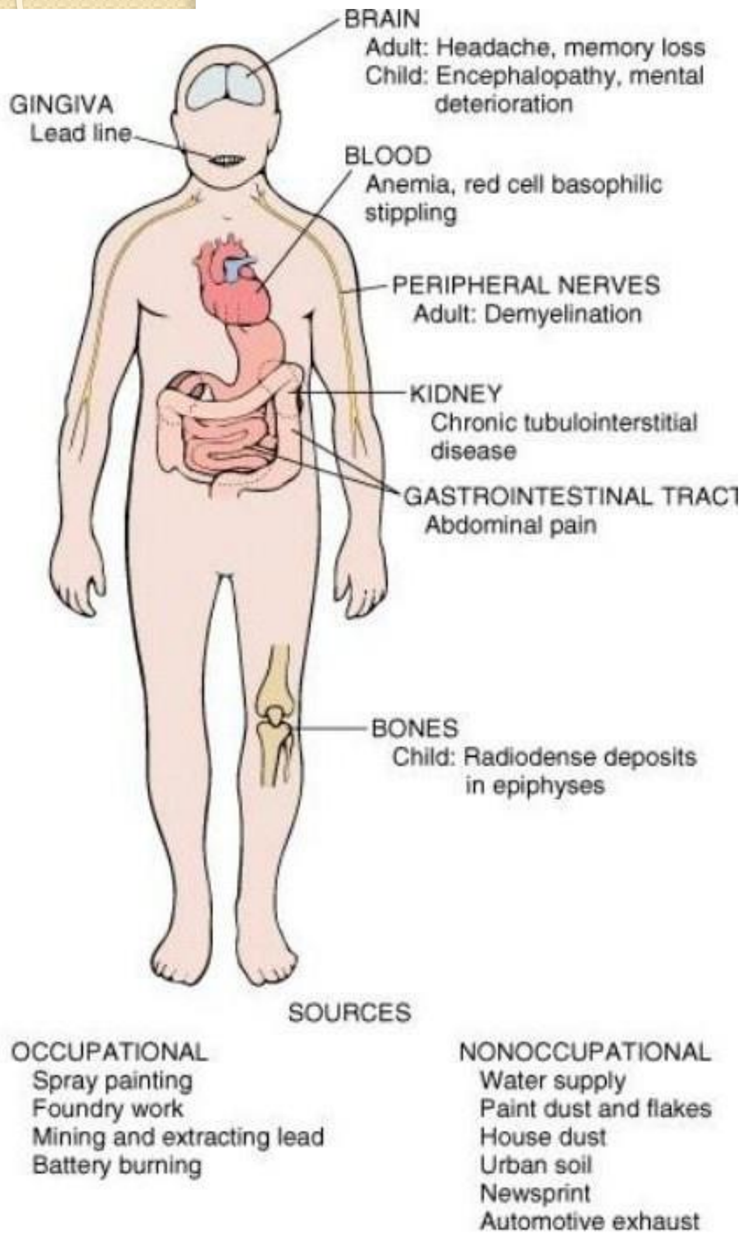
Toxic effects of Hg

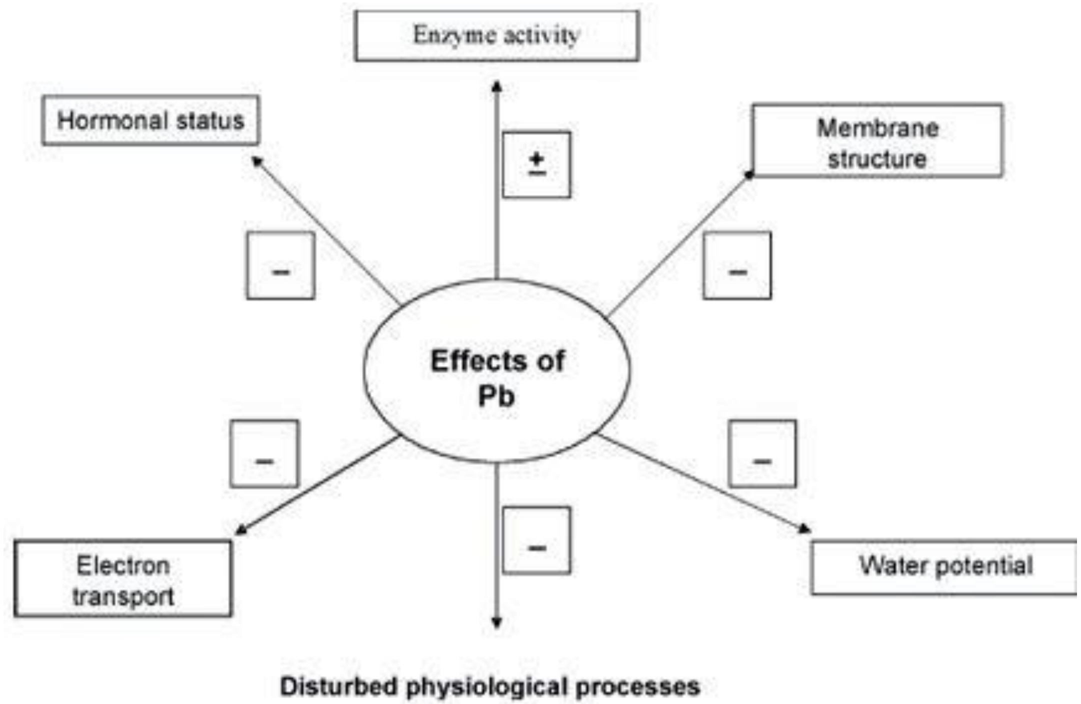


Victim of Minamata disease at Japan, in 1956

- Skin burns
- Irritation of nose and skin and rashes
- Excessive perspiration
- Damage to the kidneys, vision
- Minamata disease
- Dysfunctions of the central nervous system; severe brain damage and death
- Severe brain damage and death
- Loss of hearing and muscle coordination

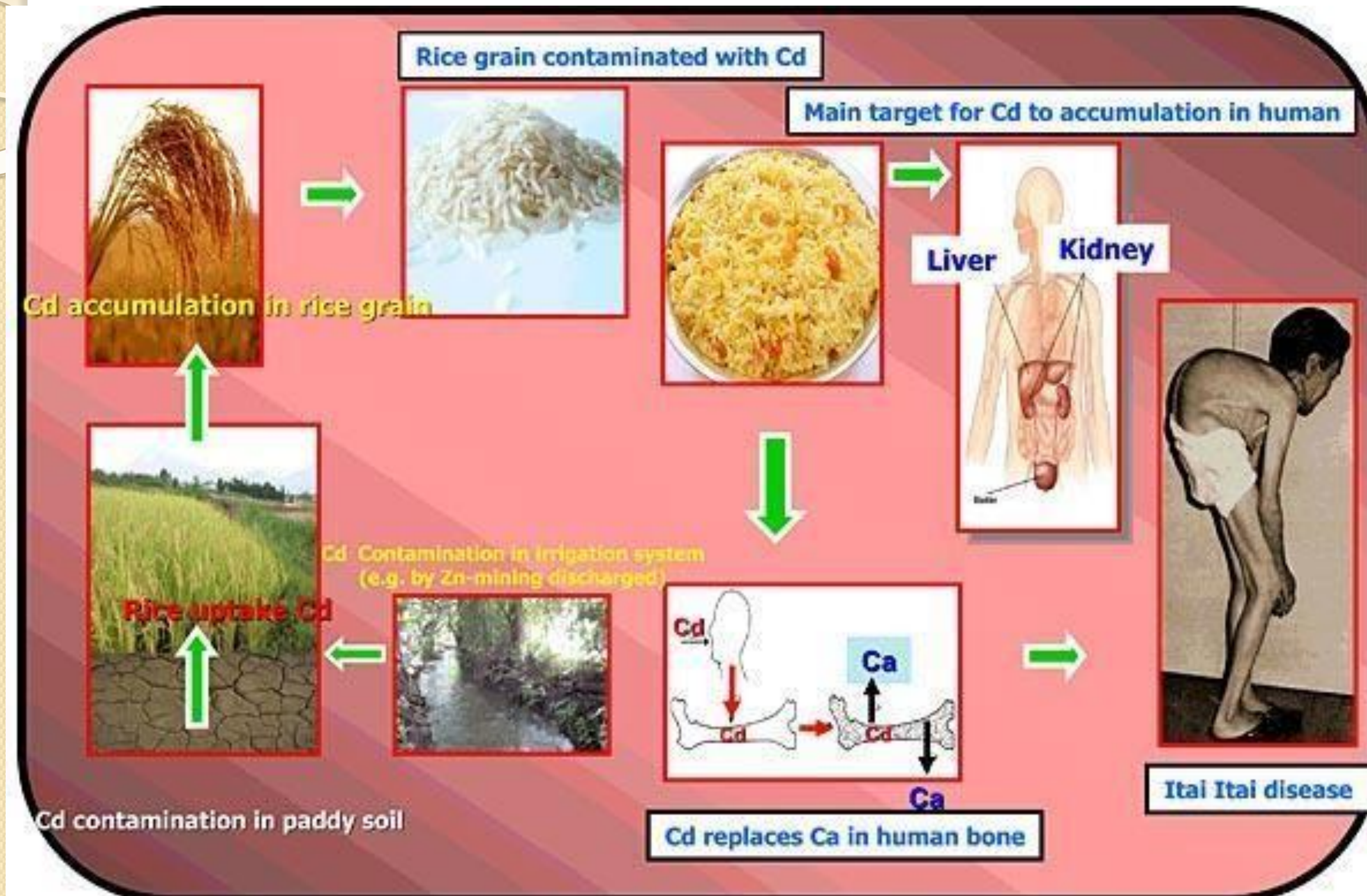
Toxic effects of Pb





- Lead breaks the blood-brain barrier and interferes with the normal development of brain in infants
- lower IQ levels in children

Toxic effects of Cd



Metallobiomolecules

Biomolecules

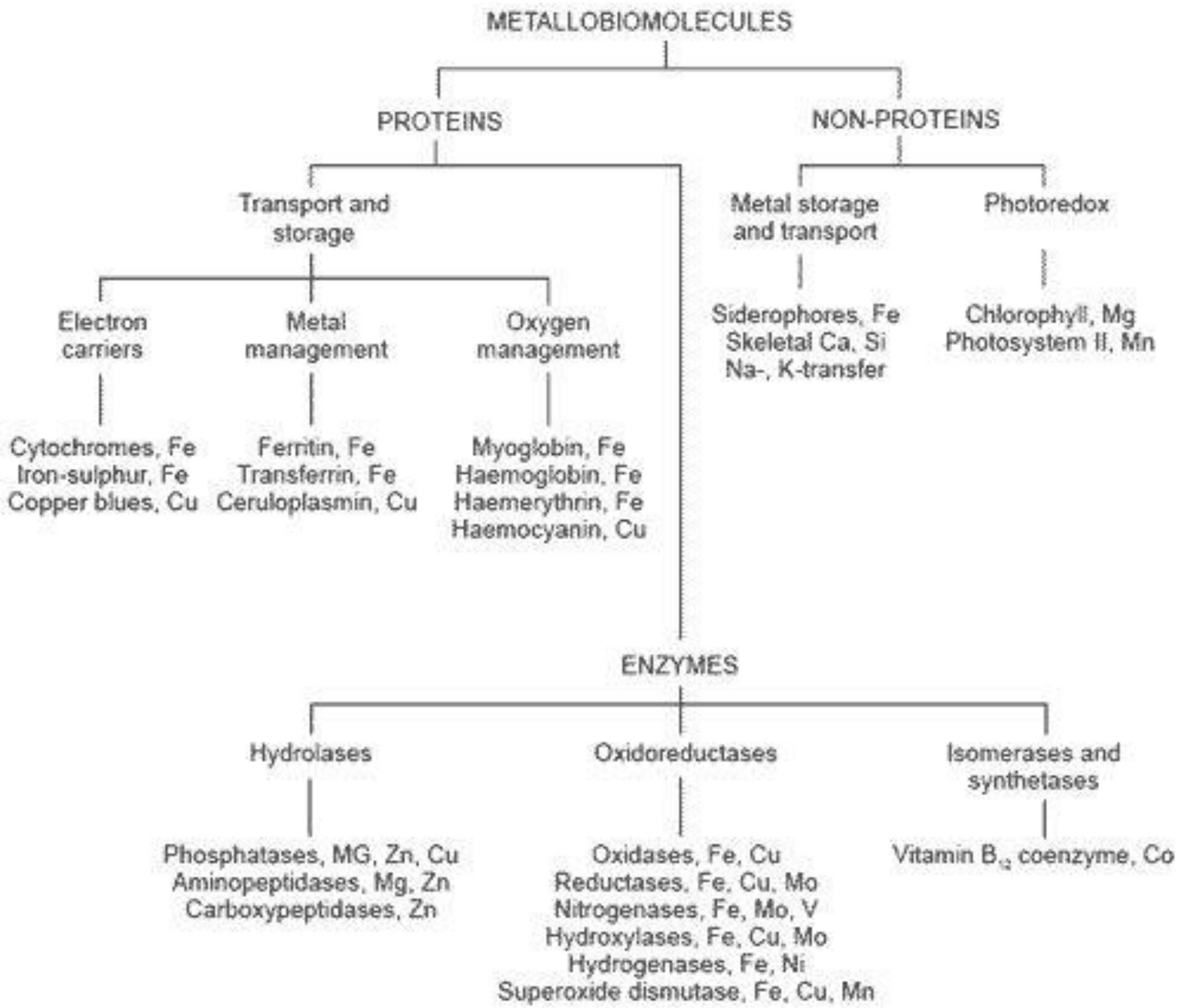
Biomolecules are molecules appear in biological systems to perform a specific function, like carbohydrates, proteins, lipids and nucleic acids.

Metallobiomolecules

Metallobiomolecules are molecules associated with metal ions which play a major role in regulating biological processes, like biomolecules do.

Characteristic feature of metallobiomolecule is as the name implies association of metal ion with molecular part.

Classification of metallobiomolecules



Structure of a metallo-protein :A metal complex perspective

Spiral - α helix form of protein

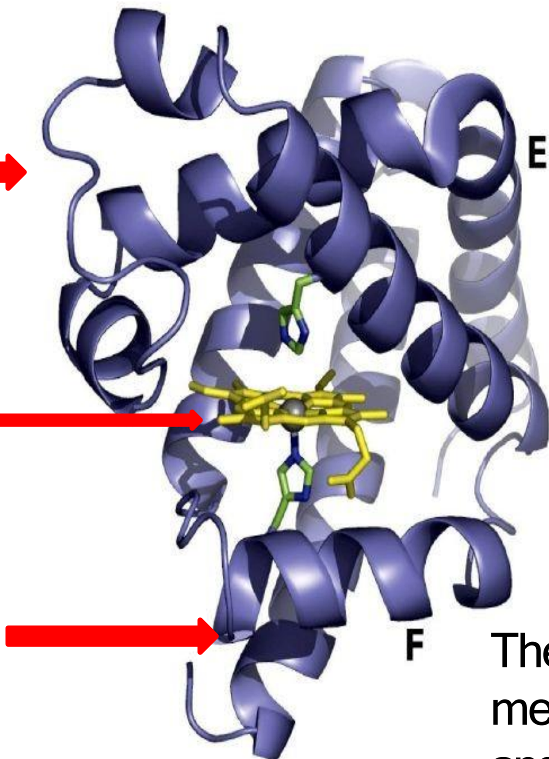
Tape - β Pleated sheet form of protein

Prosthetic groups – A metal complex positioned in a crevice. Some of the ligands for this complex or some times all of the ligands are provided by the side groups of the amino acid units.

βPleated βsheet

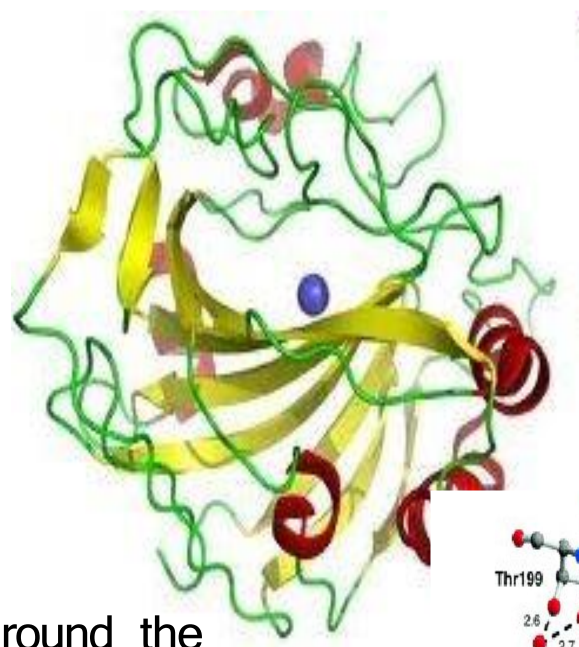
Prosthetic groups

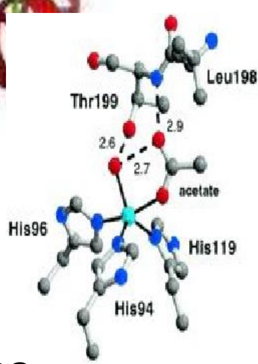
α helix



Myoglobin

The geometry around the metal and bond distances and angles are decided by the protein unit





Carbonic anhydrase

Metalloenzymes and Oxygen carriers = Protein + Cofactor

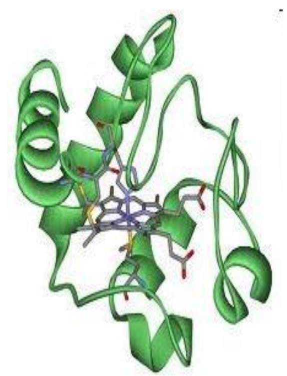
Cofactor

A **cofactor** is anon-protein chemical compound that is bound to aprotein and is required for the protein's biological activity.These proteins are commonly **enzymes**. Cofactors are either organic or inorganic.

They can also be classified

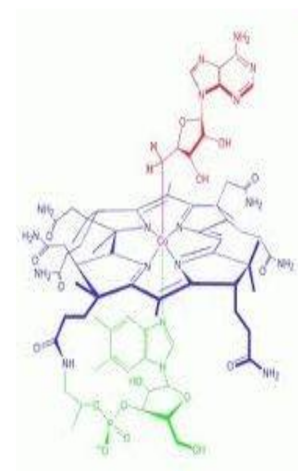
a) loosely-bound or protein-free cofactors termed **coenzymes**

b) tightly-bound cofactors termed **prosthetic groups.**

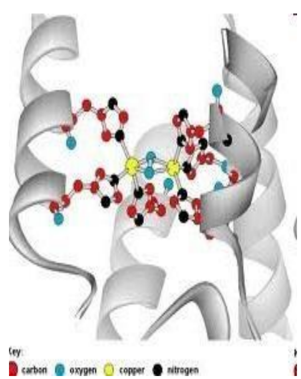


Cytochrome C

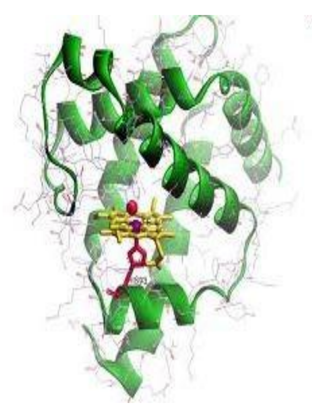
Porphyrins with different metals at its centre are a common prosthetic group in bioinorganic chemistry



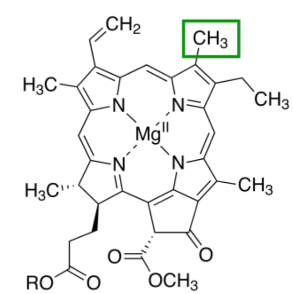
Coenzyme B12



Hemocyanin

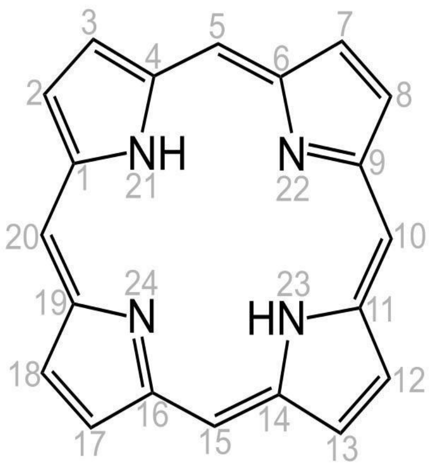


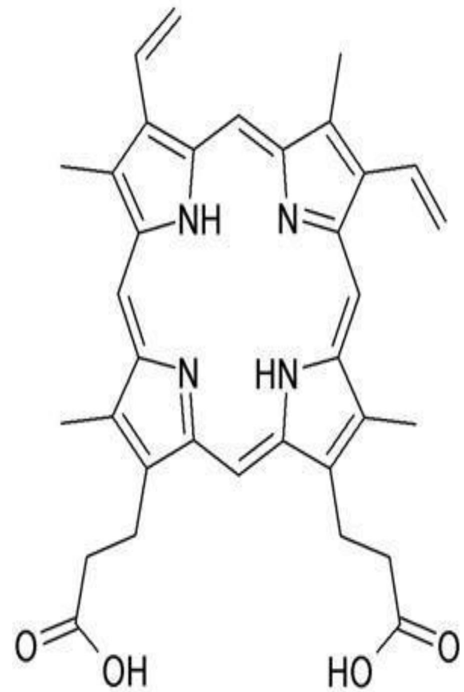
Myoglobin

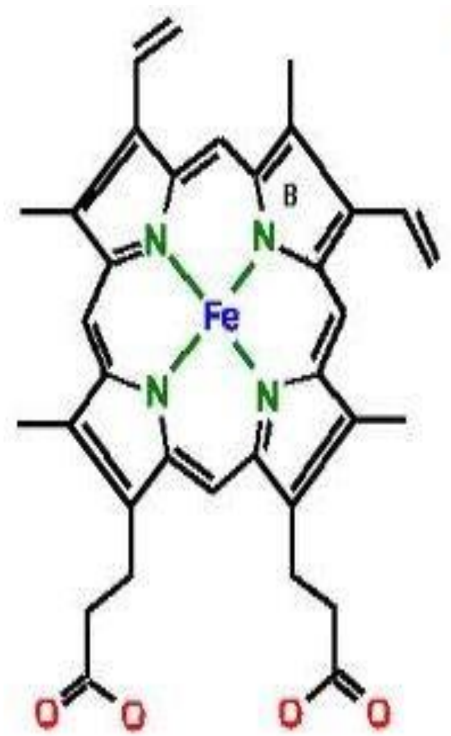


Chlorophyll

Protoporphyrin IX and Heme

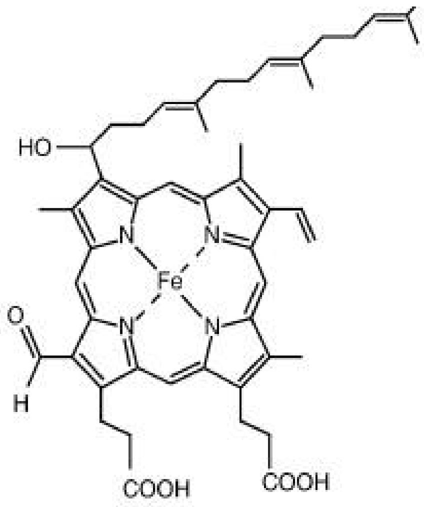




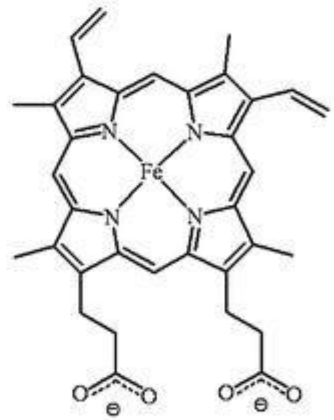


15 different ways to arrange the substituents around the porphyrin. Only one isomer protopophyrin IX is found in the living system. Porphyrins are planar and aromatic

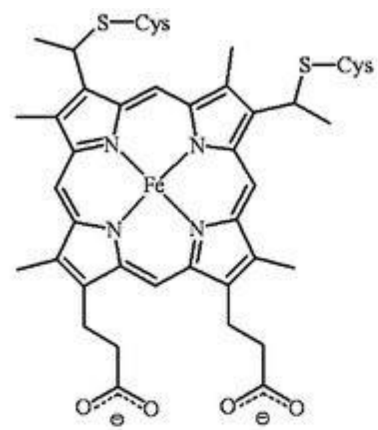
Heme & Hemoglobin



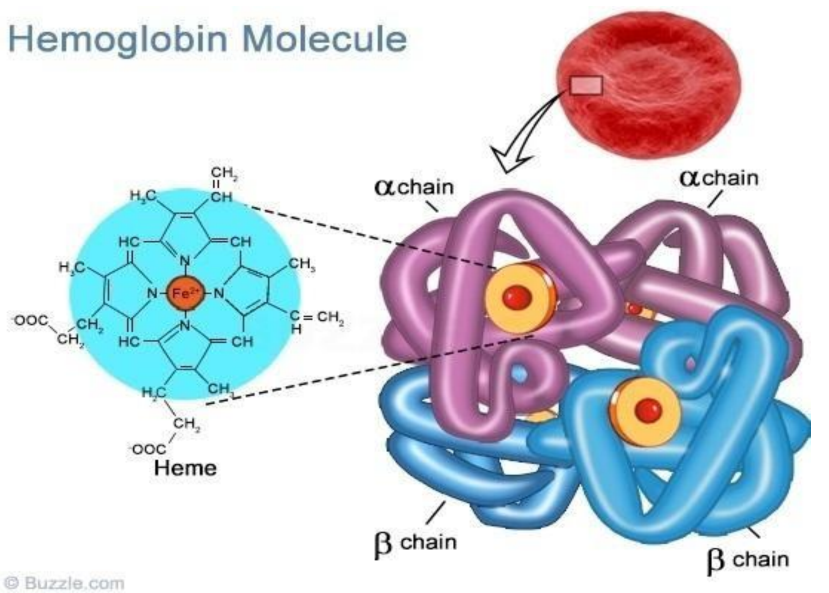
Heme A



heme *b*



heme *c*

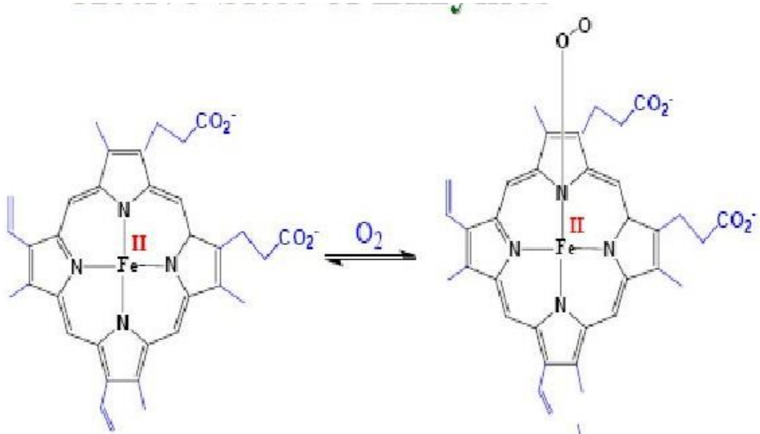


O_2 - uptake proteins:

Hemoglobin
Myoglobin
Hemocyanin
Hemerythrin

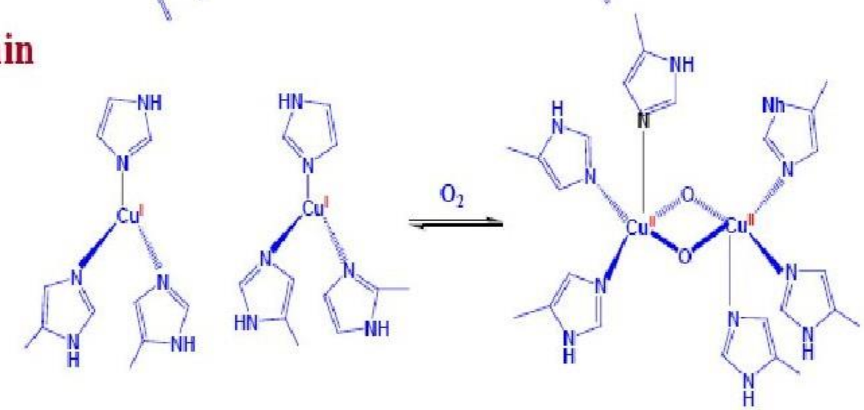
Inorganic Prosthetic group of three well known oxygen carriers

Hemoglobin



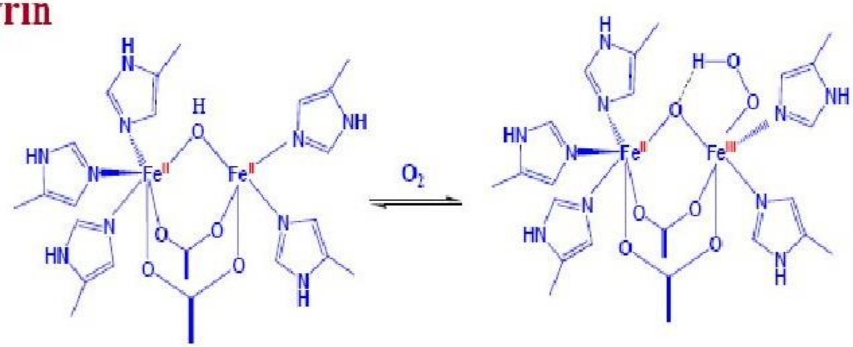
Present in Vertebrates

Hemocyanin



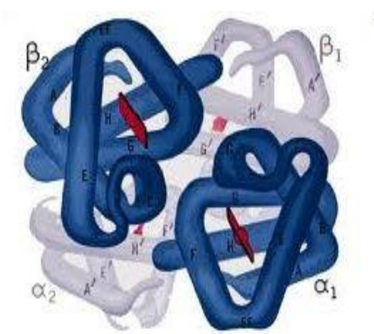
Present in molluscs

Hemerythyrin

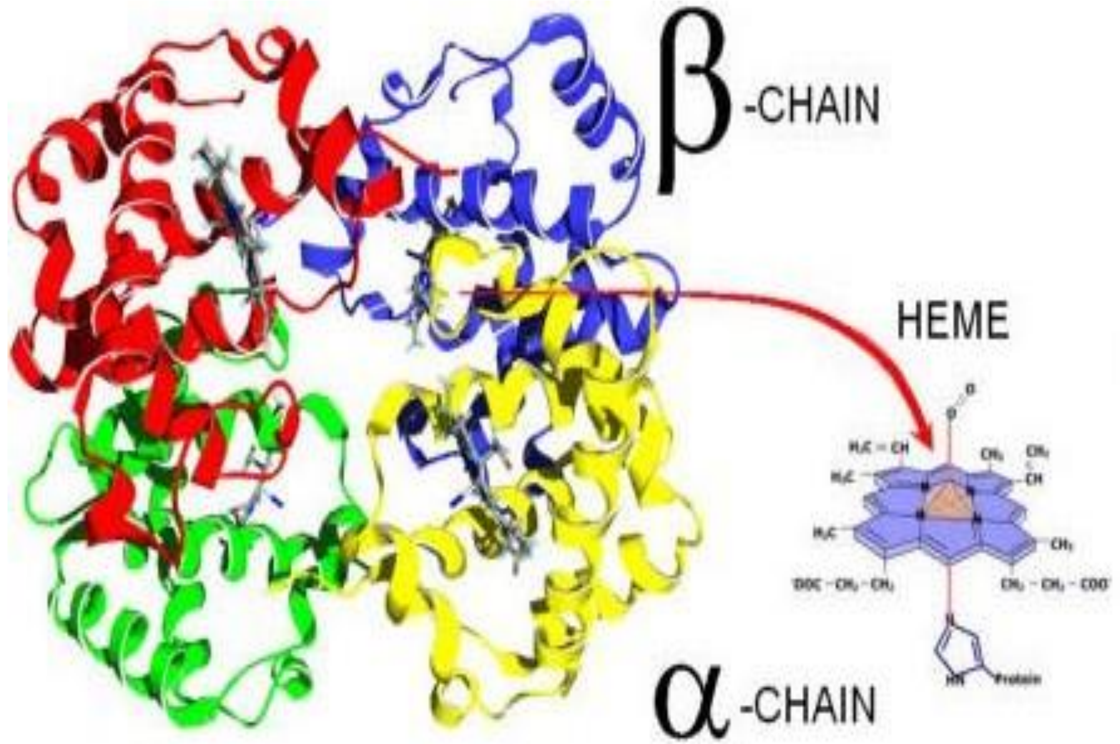


Present in some sea worms

Hemoglobin Hb



Four units of Hb



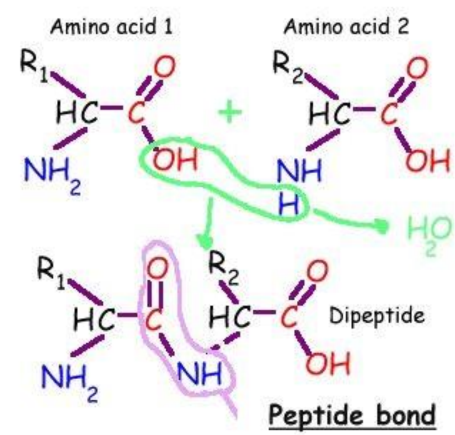
<u>3 major types of Hb</u>

Hb A (Adult)

Hb F (Fetal)

Hb S (Sickle cell)

Hb is tetramaric protein
M.W = 64500 Dalton

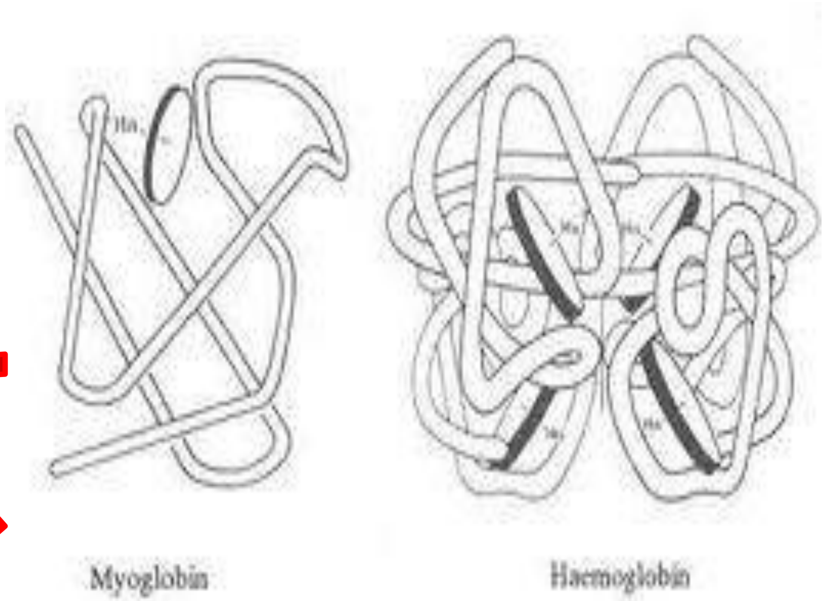


α Chain = 141 amino acid
β Chain = 146 amino acid

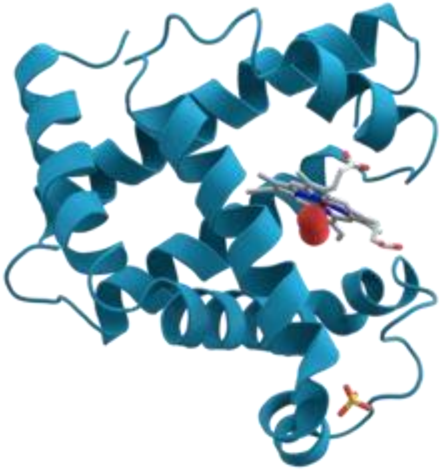
2α and 2β peptide chain are interlinked through H bonded interaction (COO^-…..NH)

Myoglobin Mb

153 amino acids



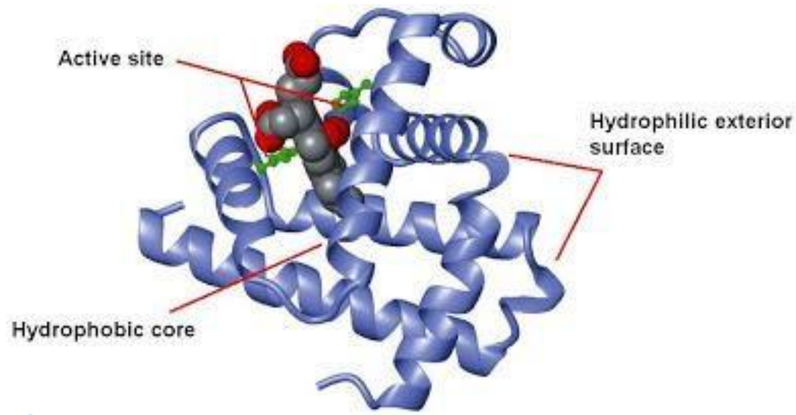
Hb is not an exact tetramer of Mb

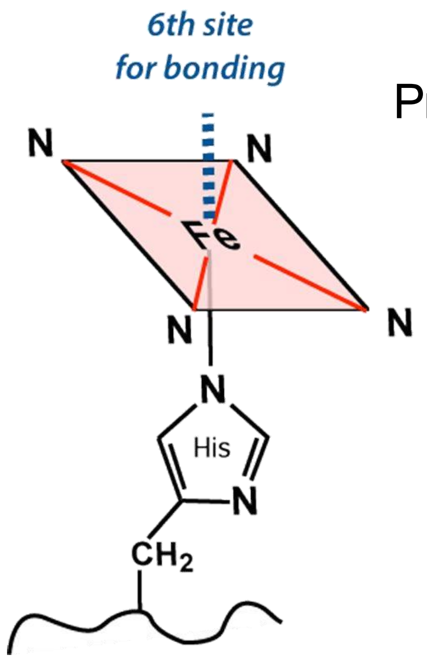


Mb is monomaric Protein with single polypeptide chain through non conductive self association

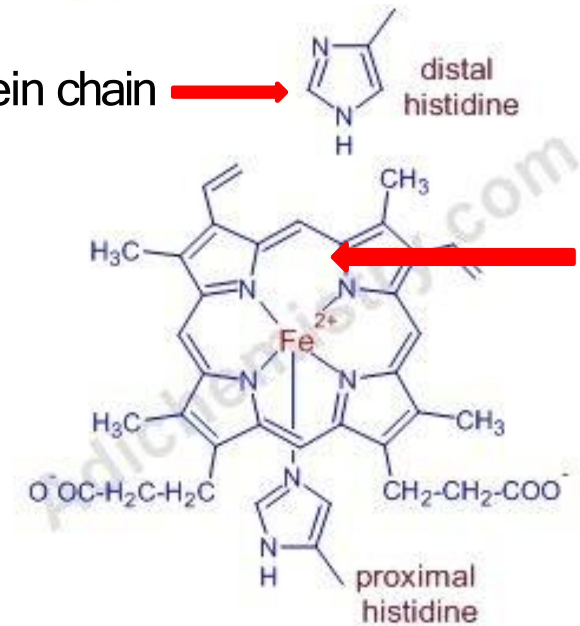
Mw = 17000 Daltons

Active Sites of Hemoglobin & Myoglobin





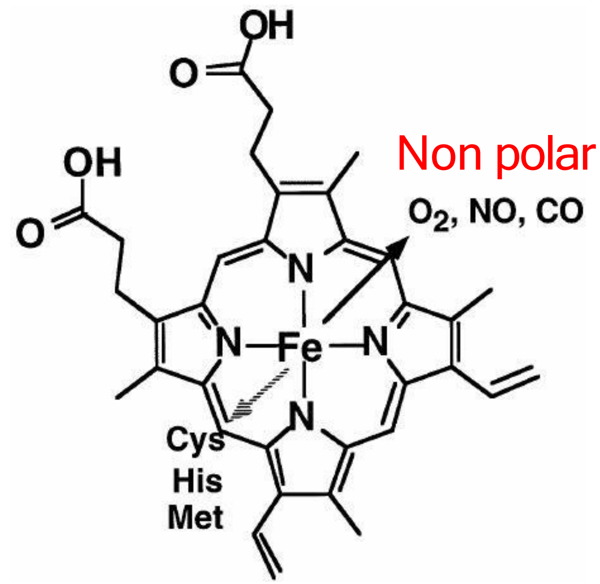
Protein chain

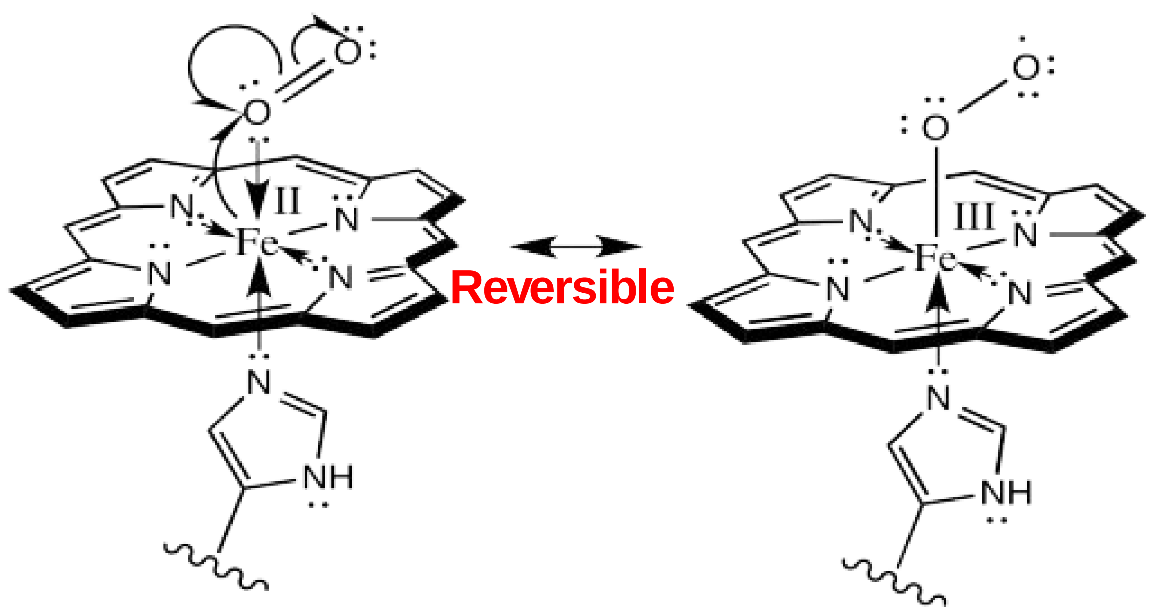


hydrophobically shield

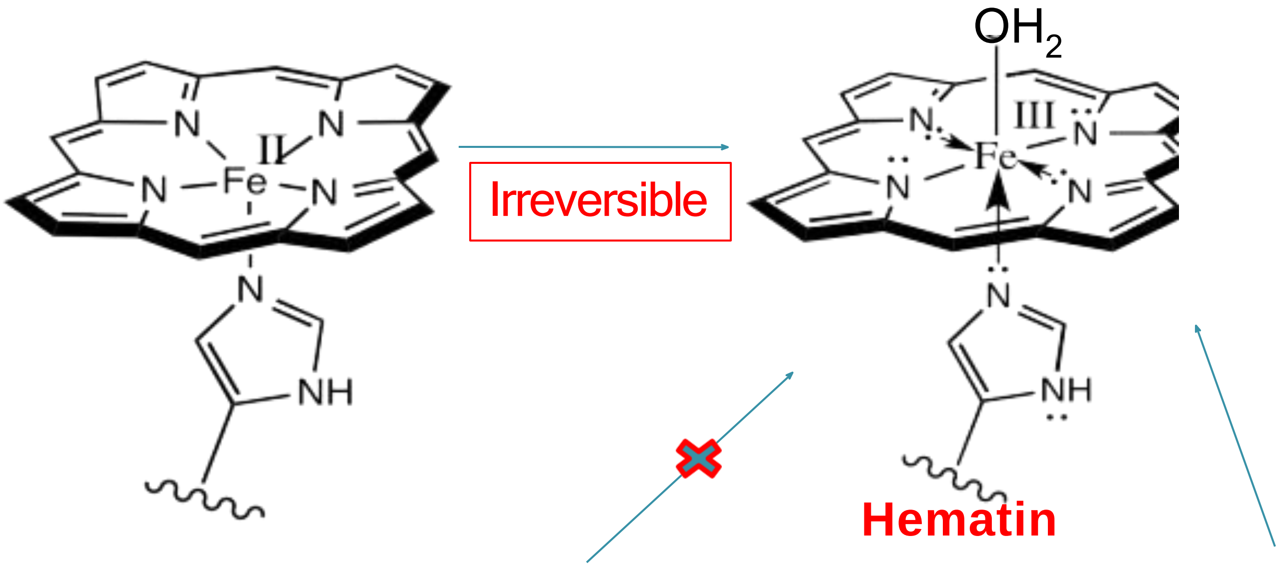
In heme group Fe (II) coordinated by 4 N atom of pophyrine ring
5th site is occupied by N atom of histidin protein chain (globin)
6th site is vacant ; but hydrophobically shield by protein chain

Active Sites of Hemoglobin & Myoglobin





Active Sites of Hemoglobin & Myoglobin

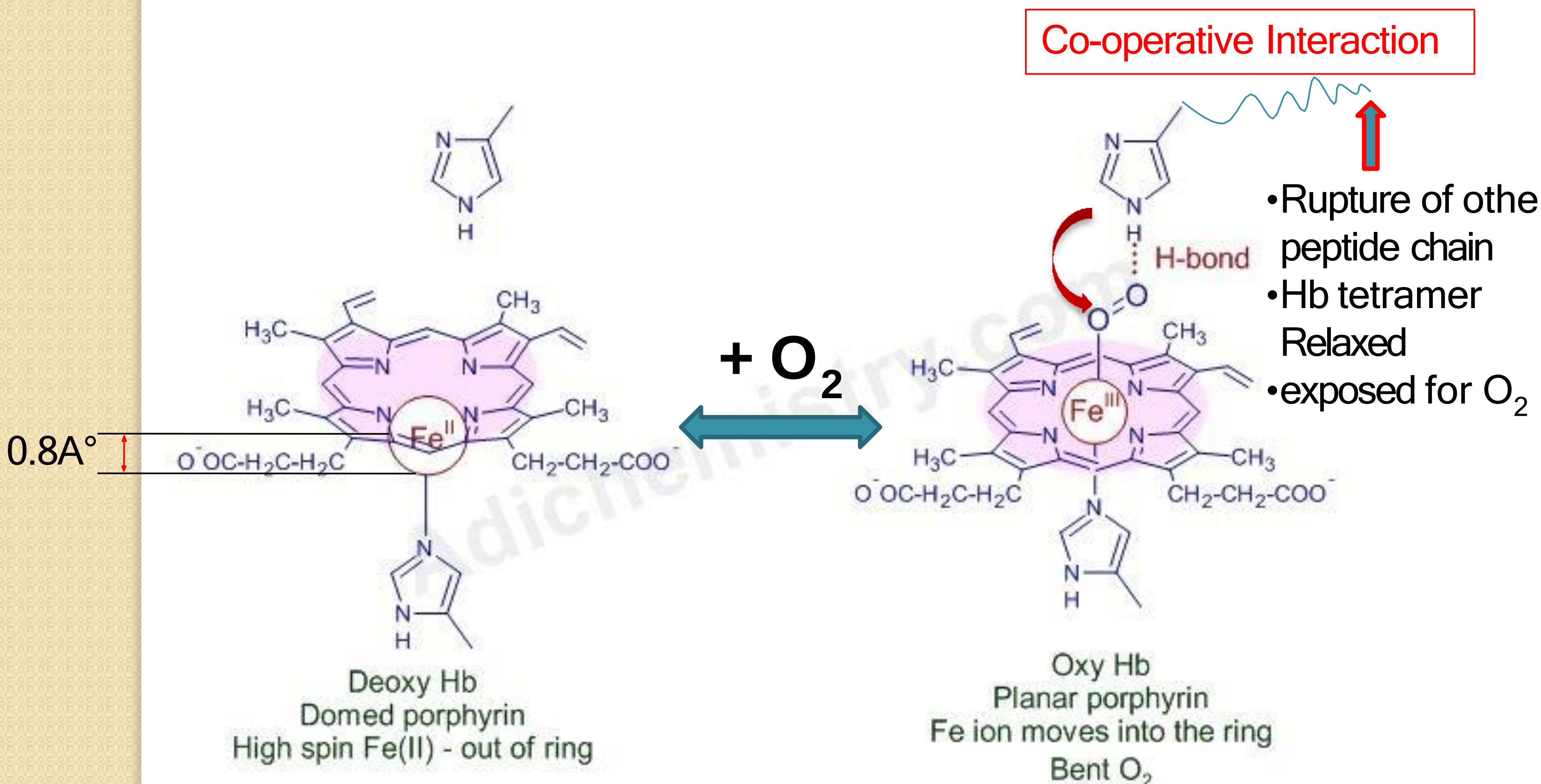


Non polar
O_2,CO etc

Readily binds with
polar Cl^-, CN^-, S^{2-} etc

Formation of Hematin is one kind of disease; that inhibits oxygenation

Binding of O_2 in Hb & Mb



Fe (II) radius = 0.92A°

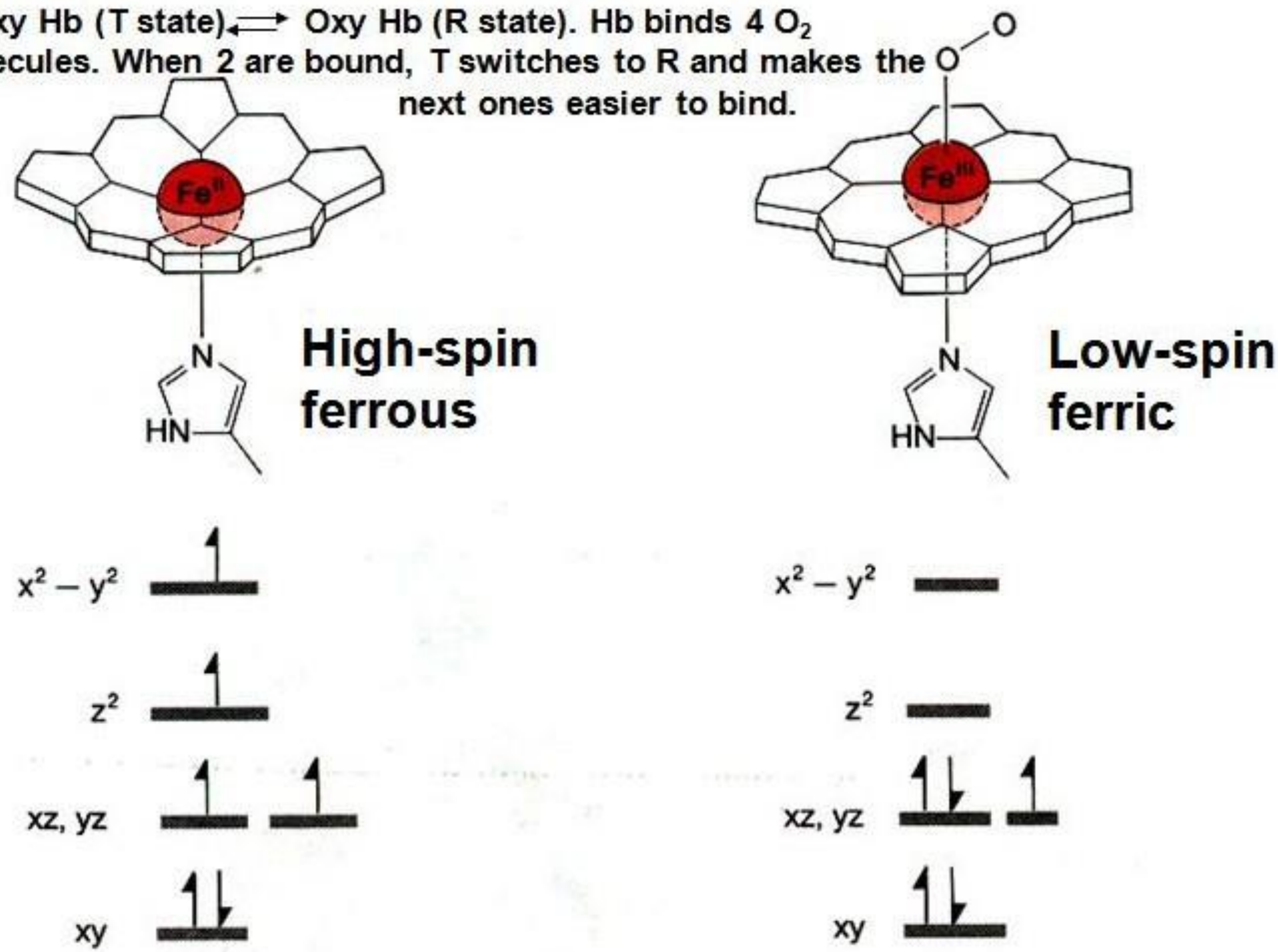
Fe (III) radius = 0.75A°

Tensed State (T)

Relaxed State (R)

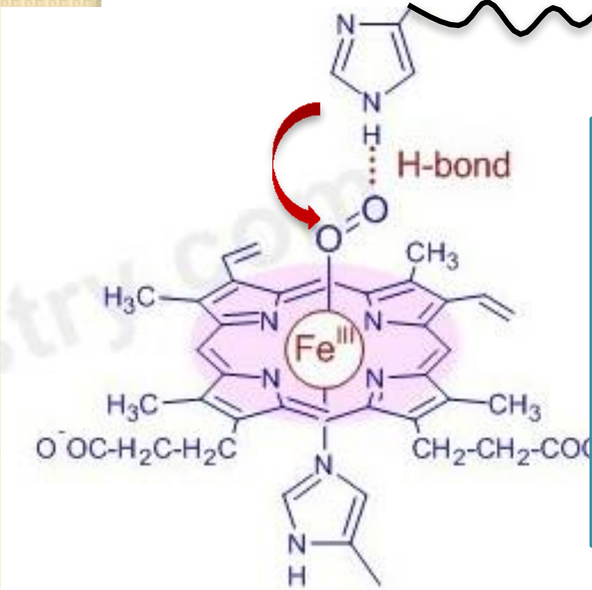
Binding of O_2 in Hb & Mb

Deoxy Hb (T state) ⇄ Oxy Hb (R state). Hb binds 4 O_2 molecules. When 2 are bound, T switches to R and makes the next ones easier to bind.



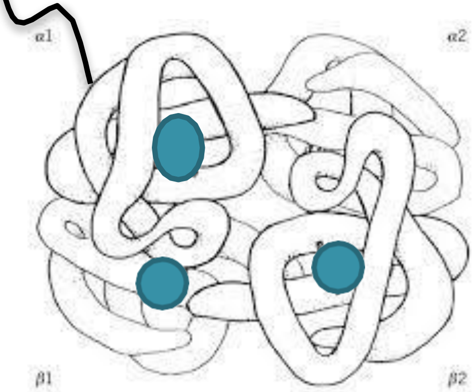
Cooperative Interaction

Co-operative Interaction



Oxy Hb
Planar porphyrin
Fe ion moves into the ring
Bent O_2

- Rupture of other peptide chain
- Hb tetramer Relaxed
- exposed 4 Fe (II) for O_2



$$Hb + O_2 \overset{K_1}{\longleftrightarrow} HbO_2$$

$$HbO_2 + O_2 \overset{K_2}{\longleftrightarrow} (HbO_2)_2$$

$$Hb(O_2)_2 + O_2 \overset{K_3}{\longleftrightarrow} (HbO_2)_3$$

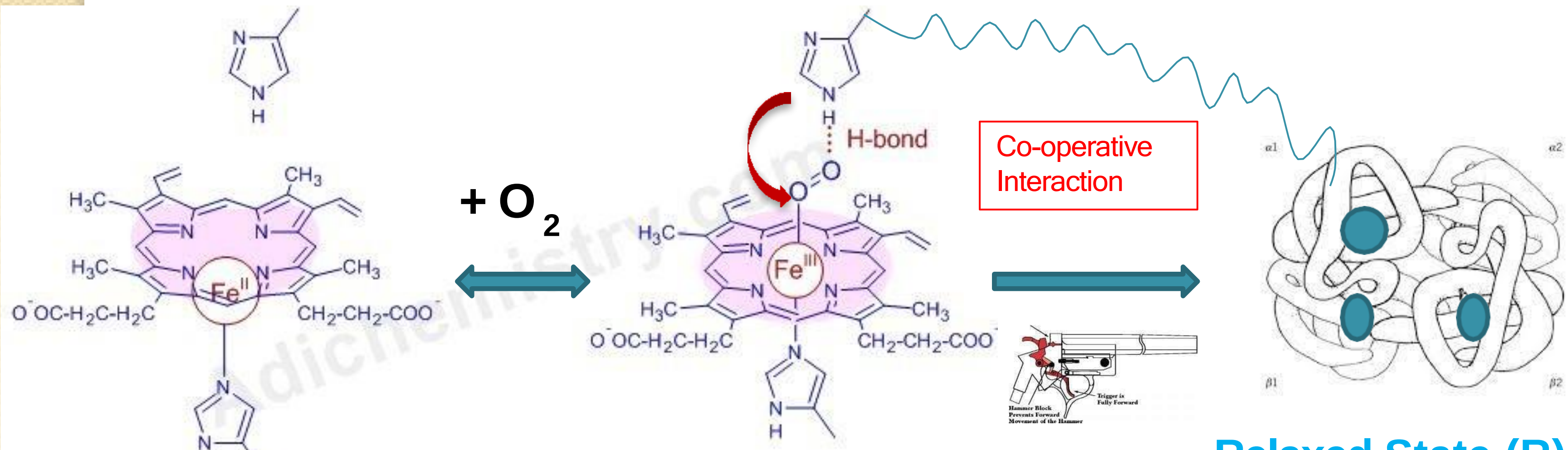
$$Hb(O_2)_3 + O_2 \overset{K_4}{\longleftrightarrow} (HbO_2)_4$$

Mb has no Co-operative interaction

K_4 (Hb) ~ K (Mb)

$K_1 < K_2 < K_3 < K_4$

Trigger Mechanism



Tensed State (T)

Low affinity form

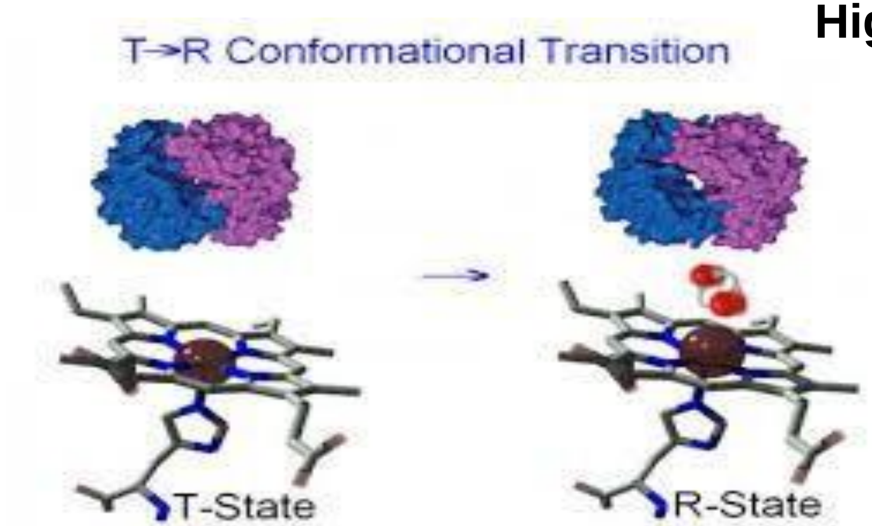
Relaxed State (R)

One Unit

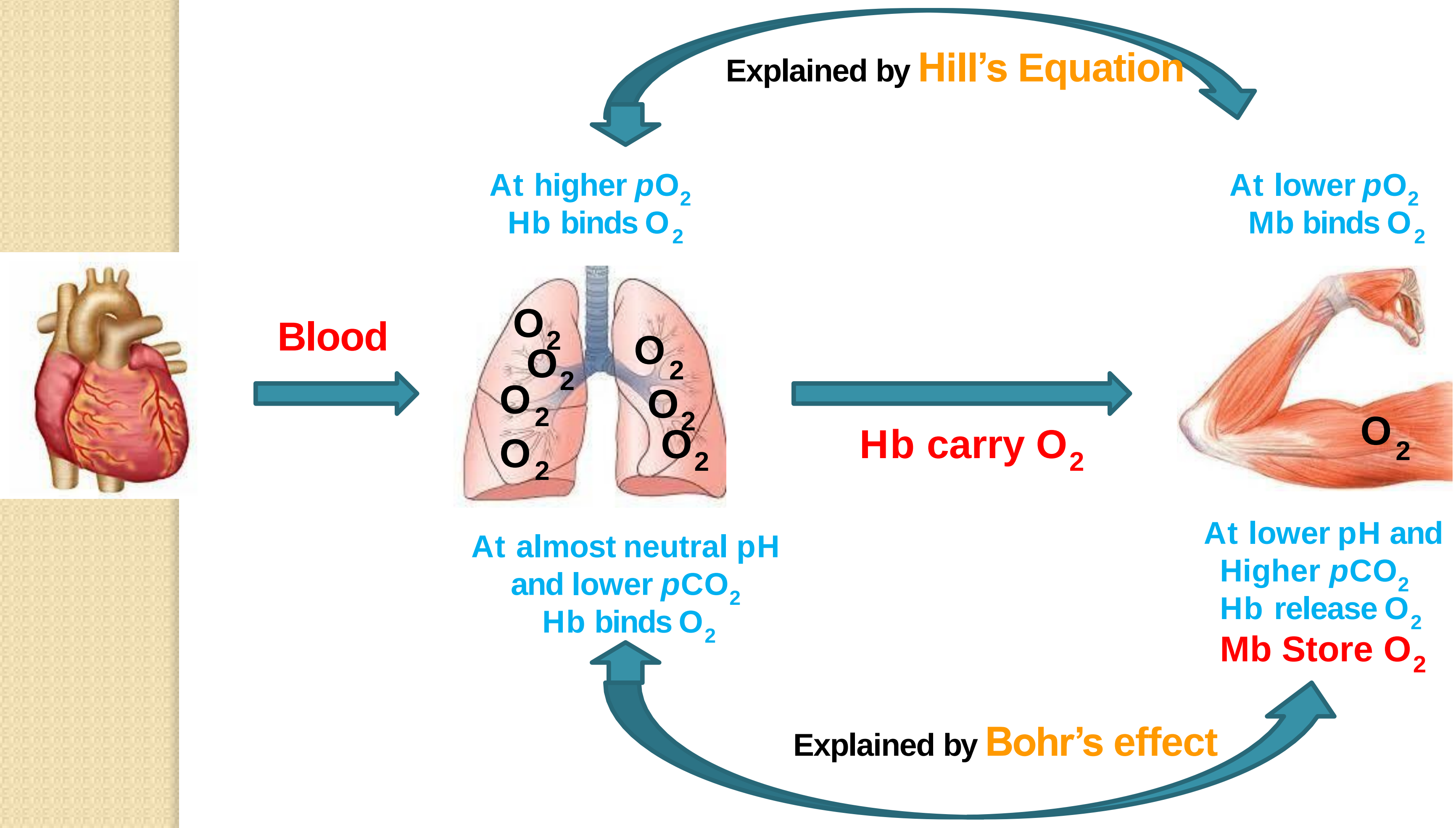
Relaxed State (R)

Rest Three Unit

High affinity form



Function of Hemoglobin and Myoglobin



Hill's Equation for Myoglobin

In lungs

$Hb + 4O_2 \longrightarrow Hb(O_2)_4$

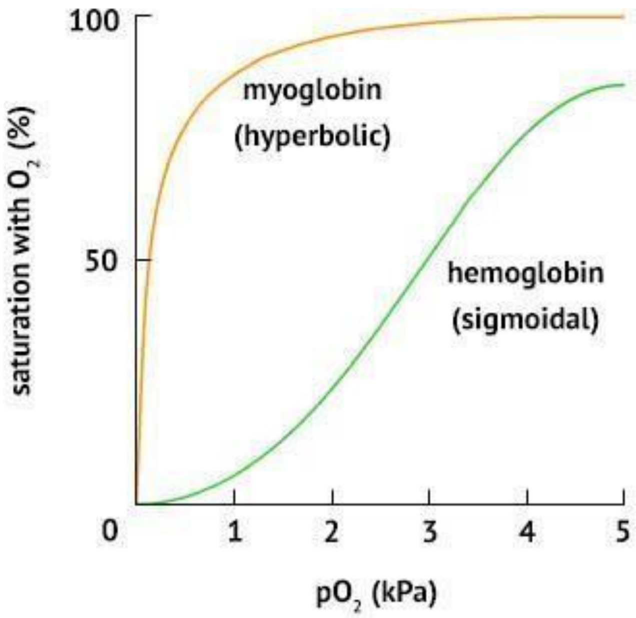
In muscle

$Hb(O_2)_4 + 4Mb \longrightarrow 4\,Mb(O_2) + Hb$

The oxygenation equilibrium for Mb is ---

$Mb + O_2 \rightleftharpoons Mb(O_2)$

$$K_{Mb} = \frac{[Mb(O_2)]}{[Mb][O_2]}$$



f = fraction of Mb bearing O_2 and $\boldsymbol{pO_2}$ = equilibrium partial pressure of O_2

$$K = \frac{f}{(1-f)\,pO_2} \qquad f = \frac{KpO_2}{1+KpO_2}$$

This is the equation for the **Hyperbolic curve** for Myoglobin

Hill plot for Hemoglobin

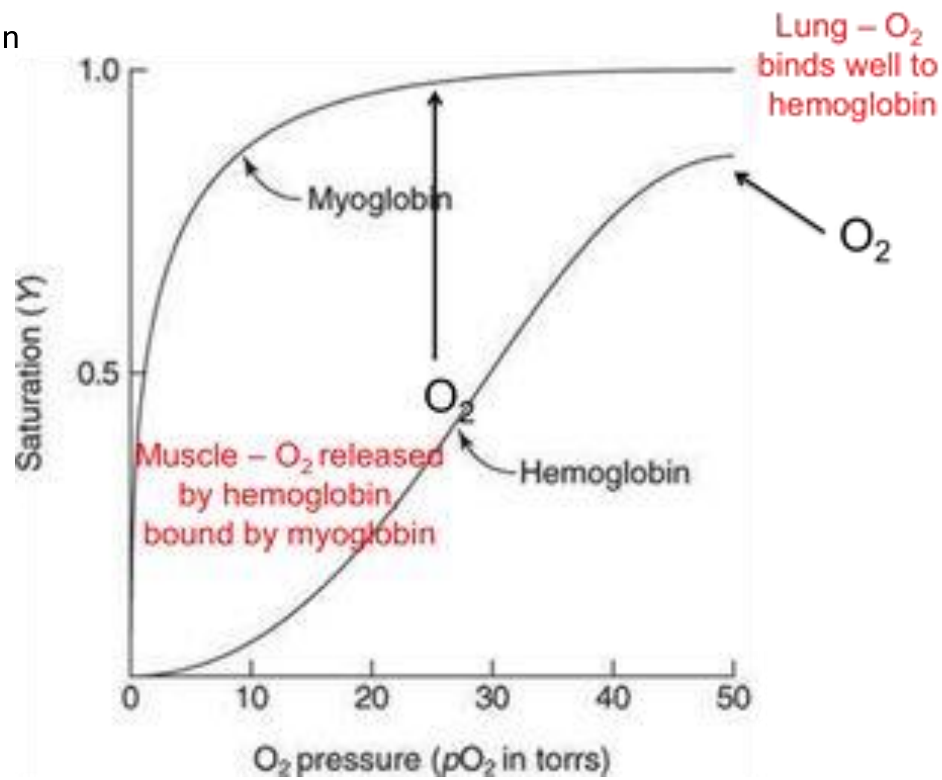
Due to tetrameric nature and cooperative interaction, oxygenation of Hb4 can be expressed as

$Hb + nO_2 \rightleftharpoons Hb_4(O_2)_n$

$$K = \frac{[Hb(O_2)_4]}{[Hb][O_2]_n}$$

$$K = \frac{f}{(1-f)\,pO_2{}^n}$$

$$f = \frac{KpO_2{}^n}{1+KpO_2{}^n}$$



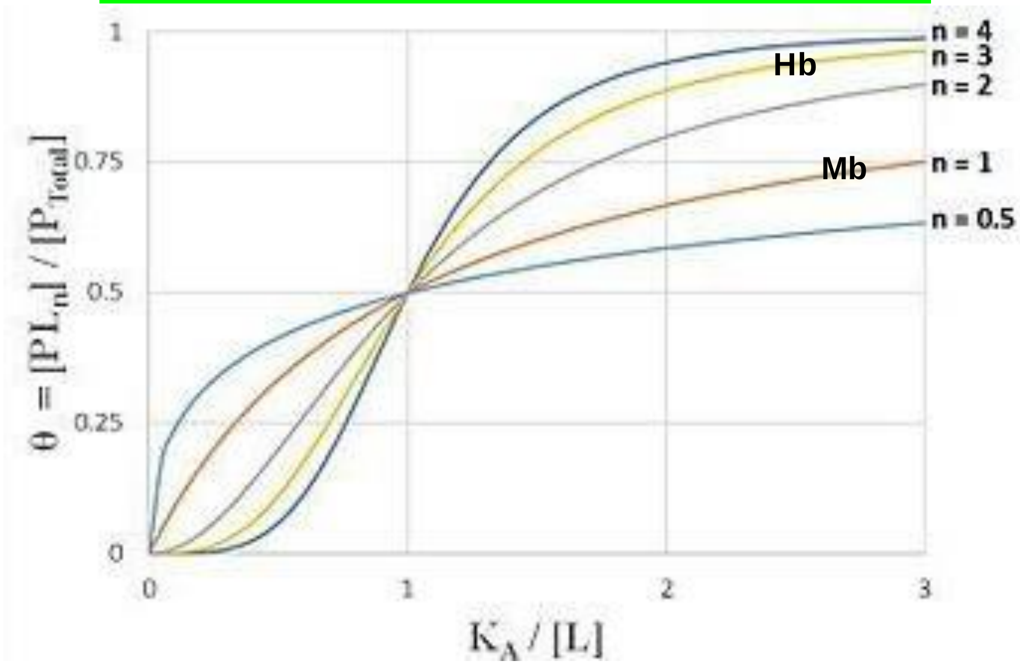
f = fraction of Hb bearing O_2

***p*O₂** = equilibrium partial pressure of O_2

n = Hill exponent

This is the equation for the **Sigmoid** curve for Hemoglobin

Hill Exponent



n = 1; shows **Hyperbolic curve**

- Here n ~3 (2.8), indicating 3 mole of O_2 is binding simultaneously
- That means, in presence of one or more bound O_2 further oxygenation occurs, instead of dissociation of $Hb_4(O_2)_n$; that confirms cooperative interaction.
- So Hb is more oxygenated at higher O_2 Pressure at Lungs or Gill.

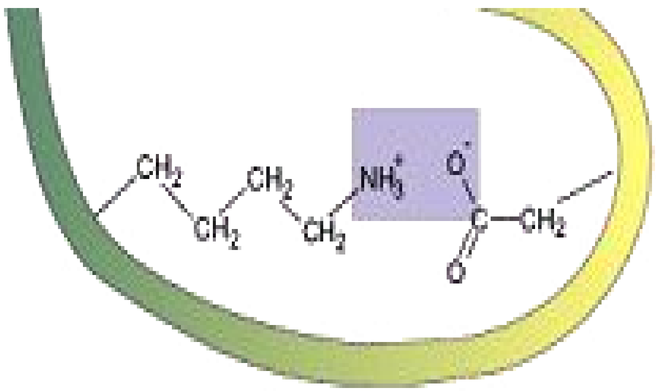
n = 3; shows **Sigmoid curve**

- Here n = 1, indicate binding of one mole of O_2
- There is no cooperative interaction
- So Mb uptakes O_2 in 1: 1 ratio

  $Mb + O_2 = Mb(O_2)$
- So Mb is oxygenated at lower O_2 Pressure at cell of muscle.

The Bohr Effect

As oxygenation of Hb is involving breakdown of (COO^- .... NH_4^+) salt bridge between and within sub-units. (cooperative interaction). So oxygenation of Hb is pH dependent. This effect is called Bohr's Effect. Christian Bohr, father of Niels Bohr discovered this effect.



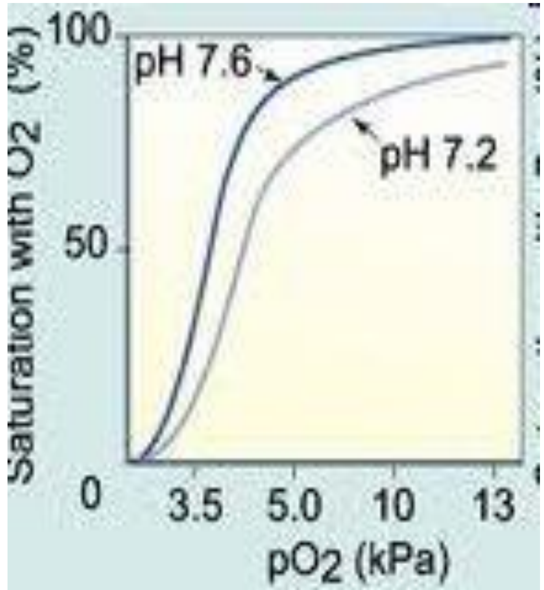
A salt bridge (weak) electrostatic interaction)

The chemical basis for the Bohr effect is due to the **formation of two salt bridges of the quaternary structure**.

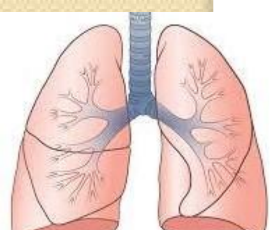
1. One of the salt bridges is formed by the interaction between Histidine 146 and Lysine 40.
2. The second bridge is formed with the aid of an additional proton on the histidine residue.

Below a pH of 6, the imidazole ring of histidine is mostly protonated thus favoring salt bridge formation

The Bohr Effect

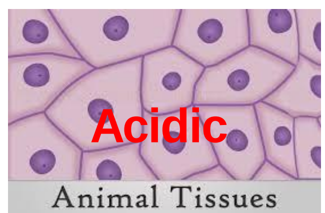


Total transport



$Hb + O_2 = Hb(O_2)_4$

Hb carry O_2



produce H_2CO_3, CO_2, lactic acid etc

Acidic condition

$Hb(O_2)_4 = Hb + O_2$

+ Mb

pH independe

$Mb(O_2)$

Store for respiration

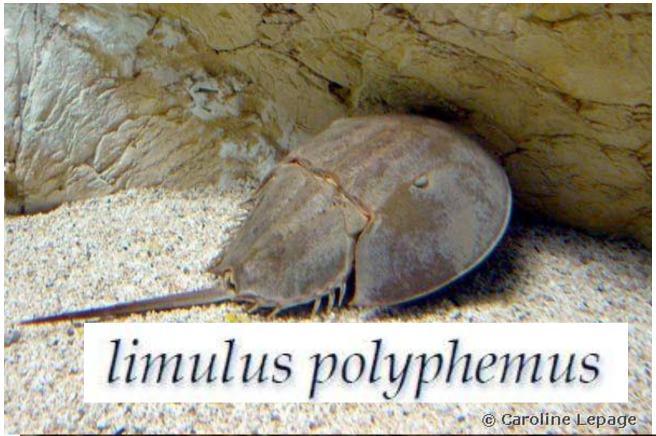
Hb reverse back to lungs with CO_2

At high O_2 pressure
High O2 affinity of Hb
Hb release CO_2

Acidic condition

CO_2 transported to lungs

Hemocyanin (Hc)

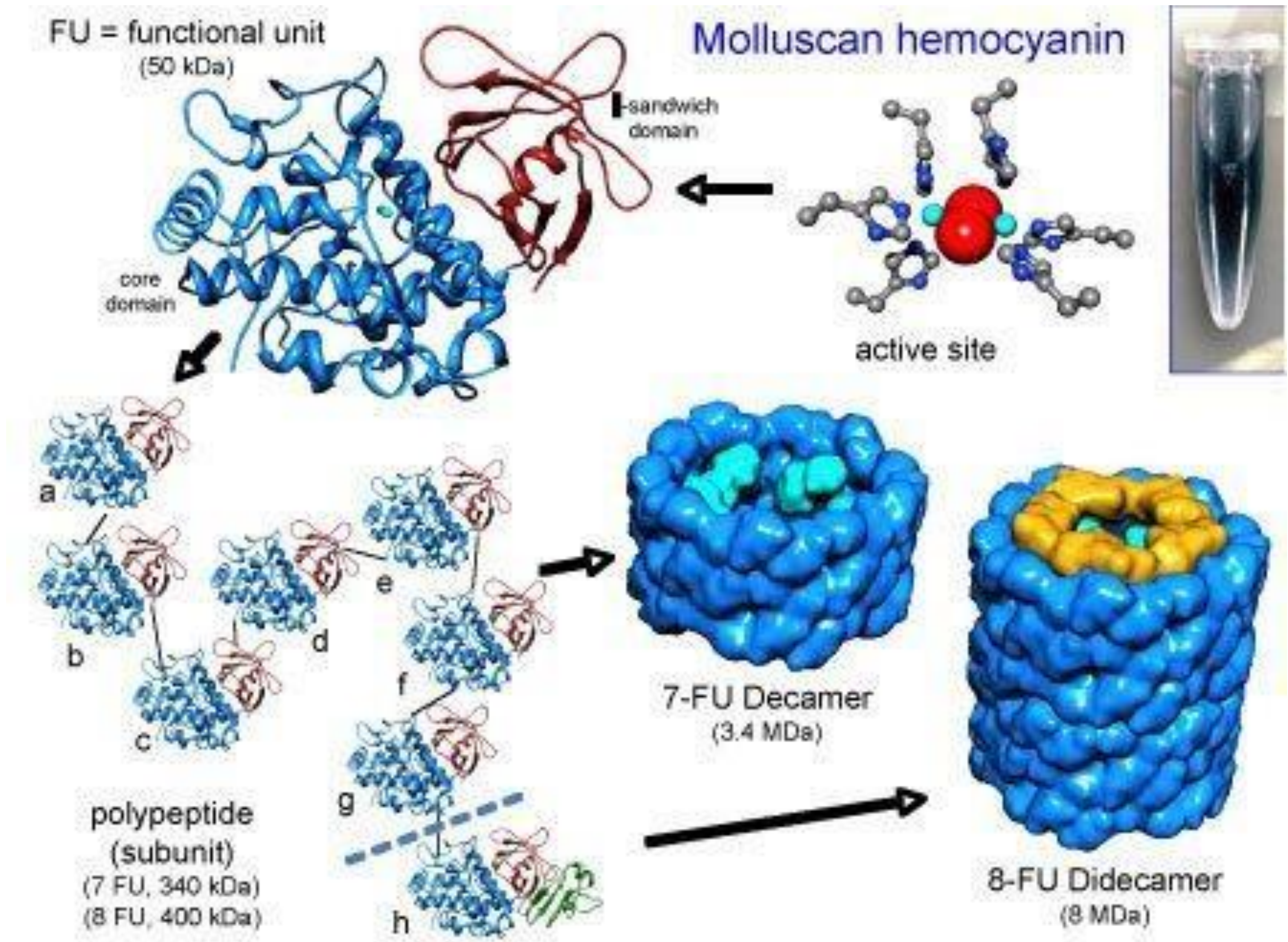


Cu containing O_2 transport protein

Structure

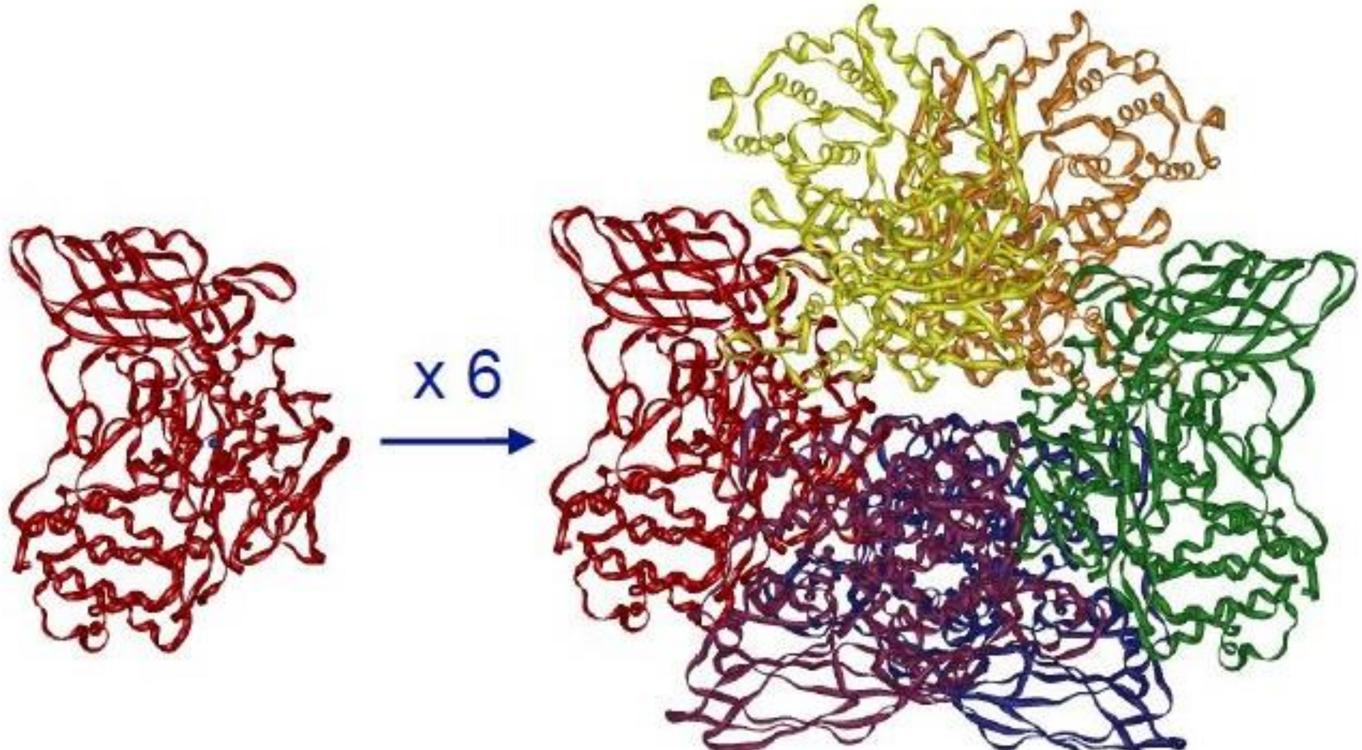
- Hc carry O_2 in the blood of most molluscs and some arthropods.
- Hc is made of individual subunit proteins which contains 1 active site of Cu.
- Each subunit weighs about 75 kDa. Subunits are arranged in chains or bundles in weights exceeding 1500 kDa.

Hemocyanin (Hc)



Crystal structure of hexameric Hc

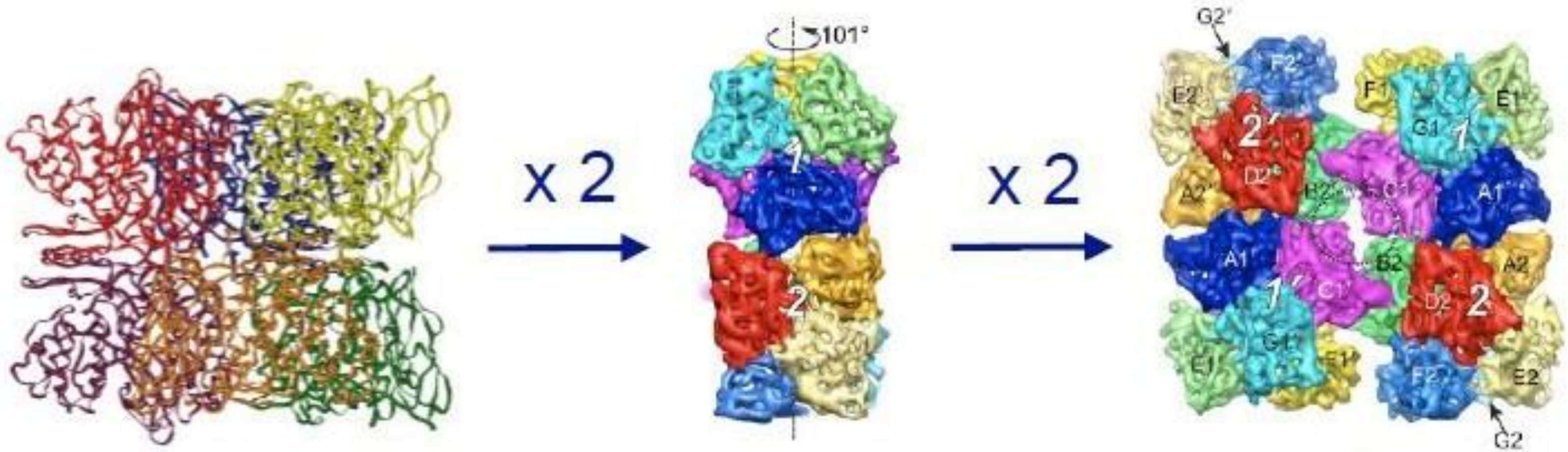
Panulirus interruptus (3.2 Å resolution)



Volbeda, A., Hol, W. G. *et al.* (1989) *J. Mol. Biol.* 209: 249-279

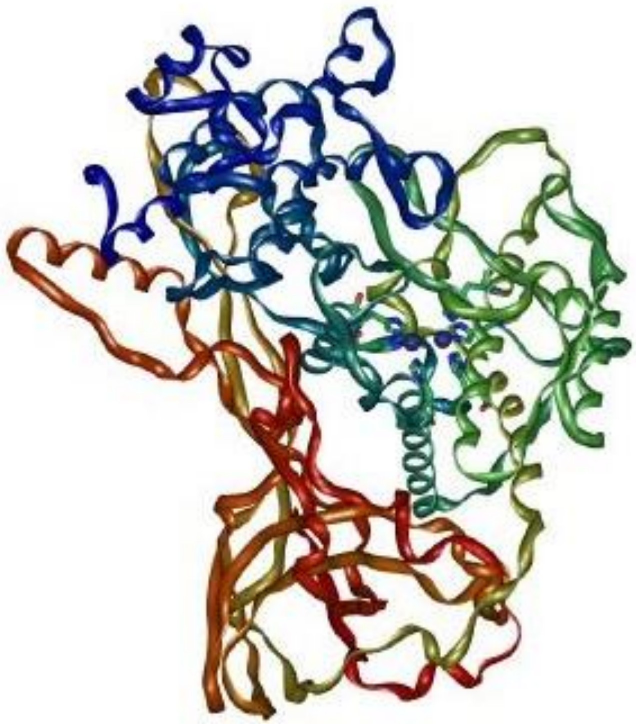
Oligomeric forms of Hc

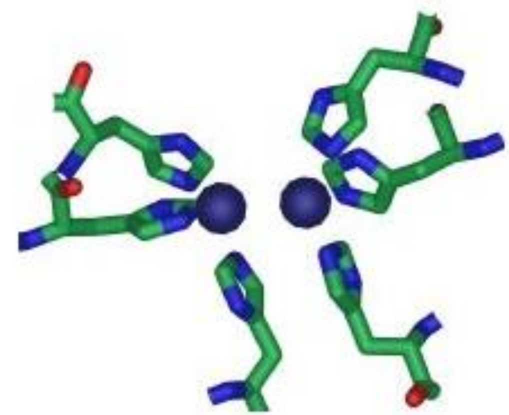
6 sub-u 12 sub-u 24 sub-u



Decker H. *et al.* (2009) *Structure* 17(5): 749-758

Hc active site

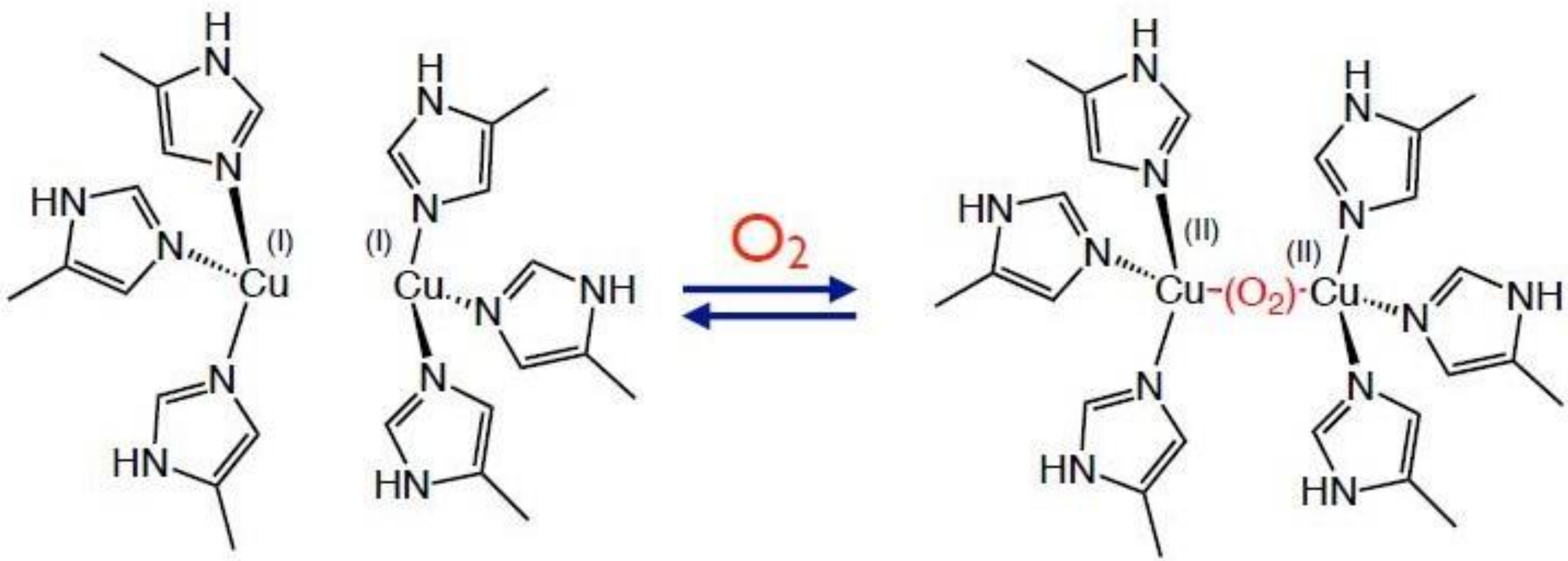




d_{Cu-Cu} = 3.8 Å

Volbeda, A., Hol, W. G. *et al.* (1989) *J. Mol. Biol.* 209: 249-279

Oxy form of Hemocyanin (oxy-Hc)



X-ray: $d_{Cu\text{-}Cu}$ = 3.8 Å

EPR: Silent

EXAFS: $d_{Cu\text{-}Cu}$ = 3.67 Å

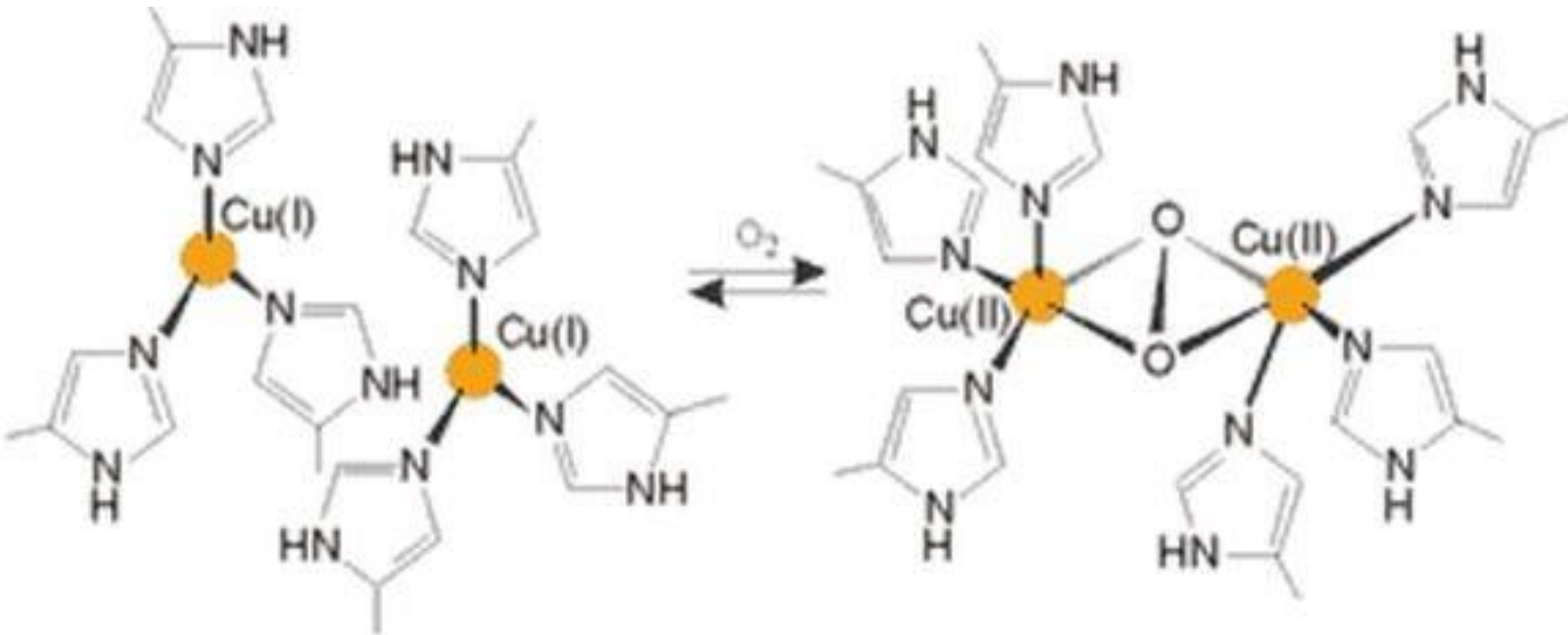
Raman: ν(O-O) = 749 cm^{-1}

UV-vis.: 345 nm (ε = 20 000 M^{-1} cm^{-1})

550 nm (ε = 1000 M^{-1} cm^{-1})

EPR: Silent

Mode of binding of O_2 to Cu (II) centre



Function of Hc

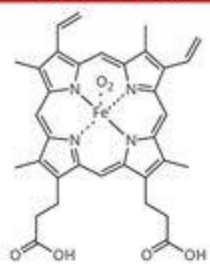
- Hc are respiratory proteins containing 2 Cu atoms that reversibly bind a single O_2.
- Oxygenation causes a color change between the colorless Cu(I) deoxygenated form and the blue Cu(II) oxygenated form.
- Because of the large size of Hc, it is found free-floating in the blood, unlike Hb, which must be contained in cells because its small size would lead it to clog and damage blood filtering organs such as the kidneys.
- This free floating nature allows for higher densities of Hc in the blood (as compared to Hb), and helps offset its low efficiency.

THE CHEMISTRY OF THE DIFFERENT COLOURS OF BLOOD

Red

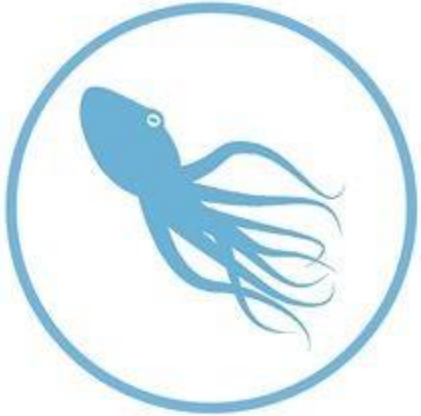
HUMANS AND THE MAJORITY OF OTHER VERTEBRATES

HAEMOGLOBIN



HAEMOGLOBIN
(oxygenated form)

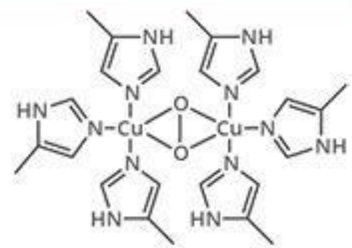
Haemoglobin is a protein found in blood, built up from subunits called 'haems'. These subunits contain iron, and their structure gives blood its red colour when oxygenated. Deoxygenated blood is a deep red colour - not blue!



Blue

SPIDERS, CRUSTACEANS, SOME MOLLUSCS, OCTOPUSES & SQUID

HAEMOCYANIN



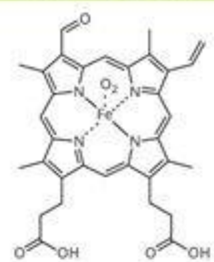
HAEMOCYANIN
(oxygenated form)

Unlike haemoglobin, which is bound to red blood cells, haemocyanin floats free in the blood. Haemocyanin contains copper instead of iron. When deoxygenated, the blood is colourless, but when oxygenated, it gives a blue colouration.

Green

SOME SEGMENTED WORMS, SOME LEECHES, & SOME MARINE WORMS

CHLOROCRUORIN



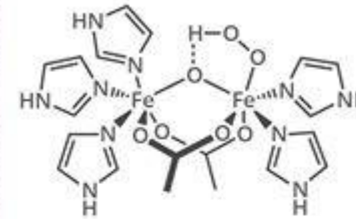
CHLOROCRUORIN
(deoxygenated form)

Chemically similar to haemoglobin; the blood of some species contains both haemoglobin & chlorocruorin. Light green when deoxygenated, it is green when oxygenated; although when more concentrated it appears light red.

Violet

MARINE WORMS INCLUDING PEANUT WORMS, PENIS WORMS & BRACHIOPODS

HAEMORYTHRIN



HAEMORYTHRIN
(oxygenated form)

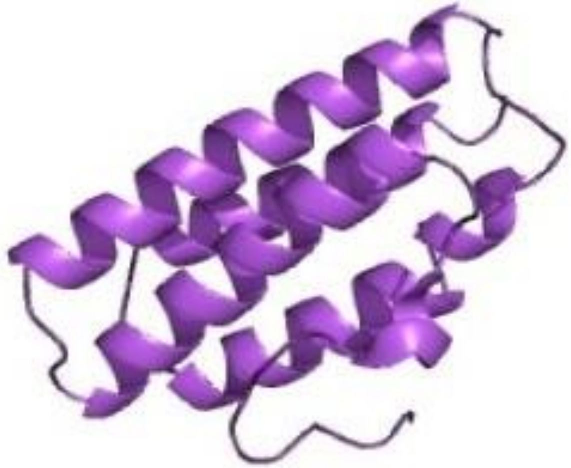
Haemorythrin is only 1/4 as efficient at oxygen transport when compared to haemoglobin. In the deoxygenated state, haemorythtin is colourless, but it imparts a violet-pink colour when oxygenated.

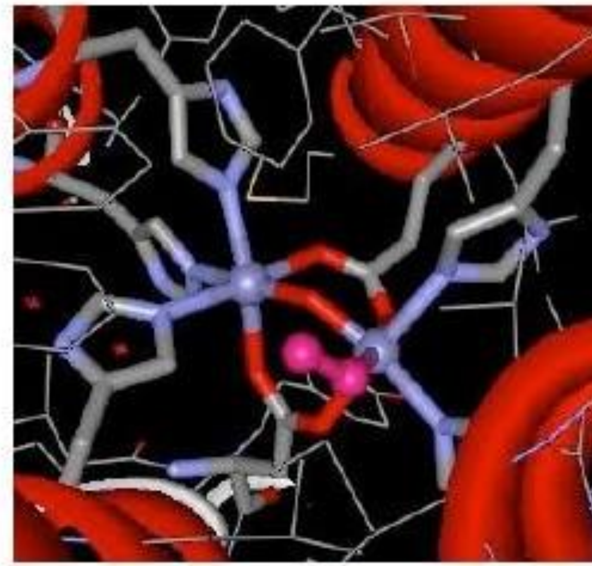
Hemerithrin (Hr)

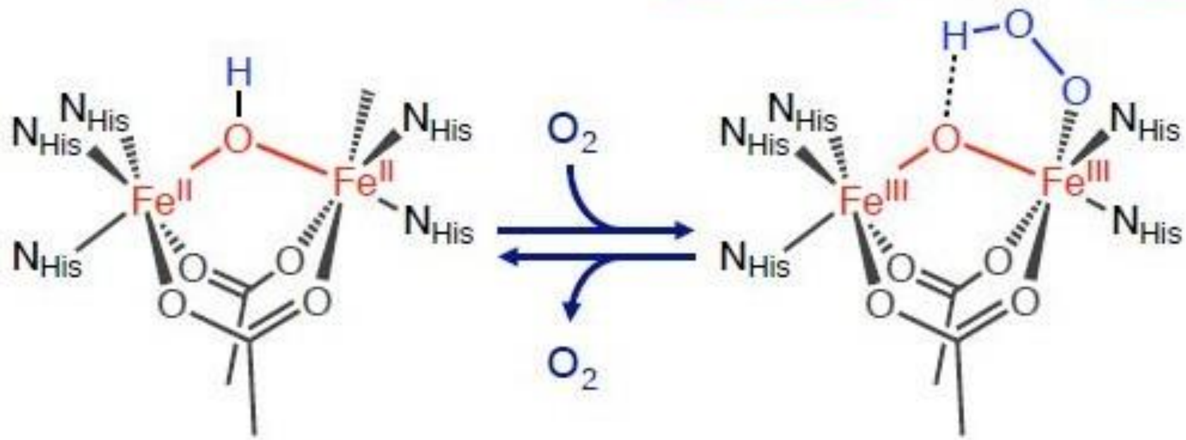


Fu containing O_2 transport protein

Hemerythrin (Hr)







deoxy-Hr oxy-Hr

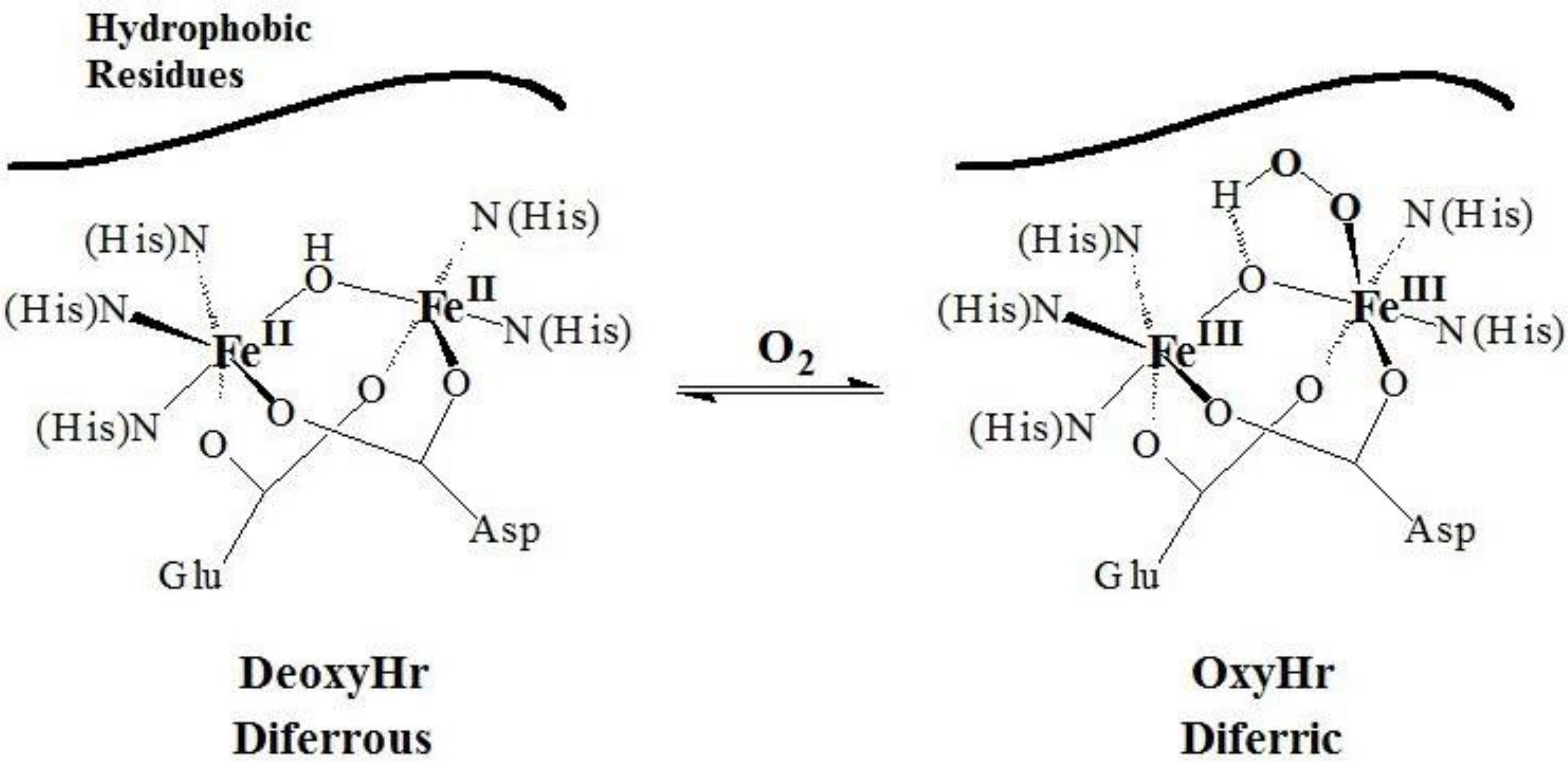
Structure of Hemerithrin (Hr)

□Non heme iron containing protein
□Molecular weight 108000 Dalton
□Consist of 8 sub units
□Each sub unit has 113 amino acids residue and 2 Fe(II) ions
□2Fe(II) ions connected through three bridge groups

a) carboxilate anion from glutamate
b) carboxilate anion from asparatate
c) by OH-

□One Fe (II) is octahedrally co-ordinated Another is five coordinated.
□One centre has three imidazole N atoms of histidin residue. Other have two to fill the five coordination.
□2Fe(II) strongly antiferromagnetically coupled through Fe-O-Fe bridge
□No cooperative interaction

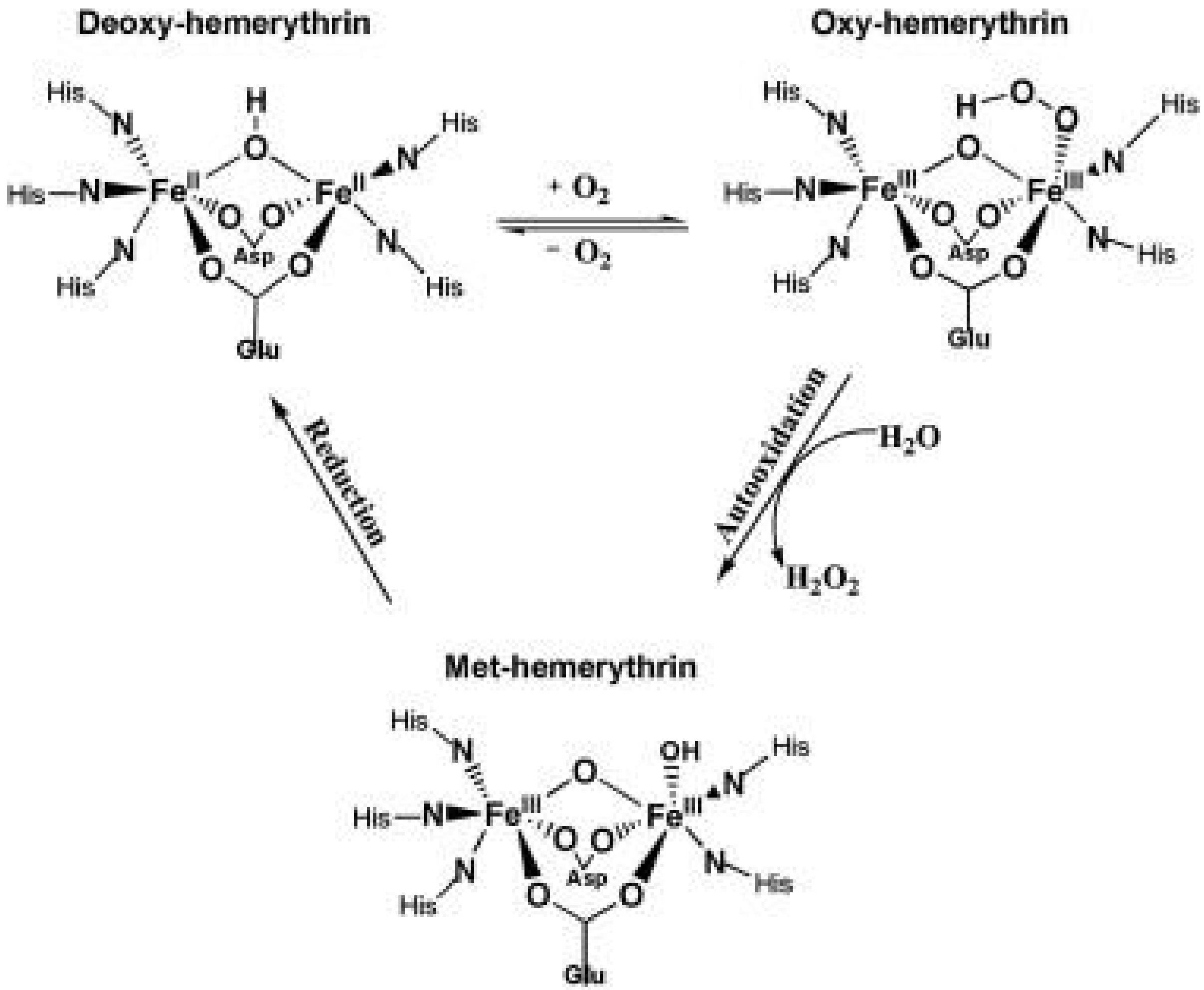
Chemistry at the Active Site of Hemerythrin (Hr)



Note proton-coupled electron transfer

Hemerythrin holds the O_2 as a hydroperoxide (HO_2, or -OOH$^-$)

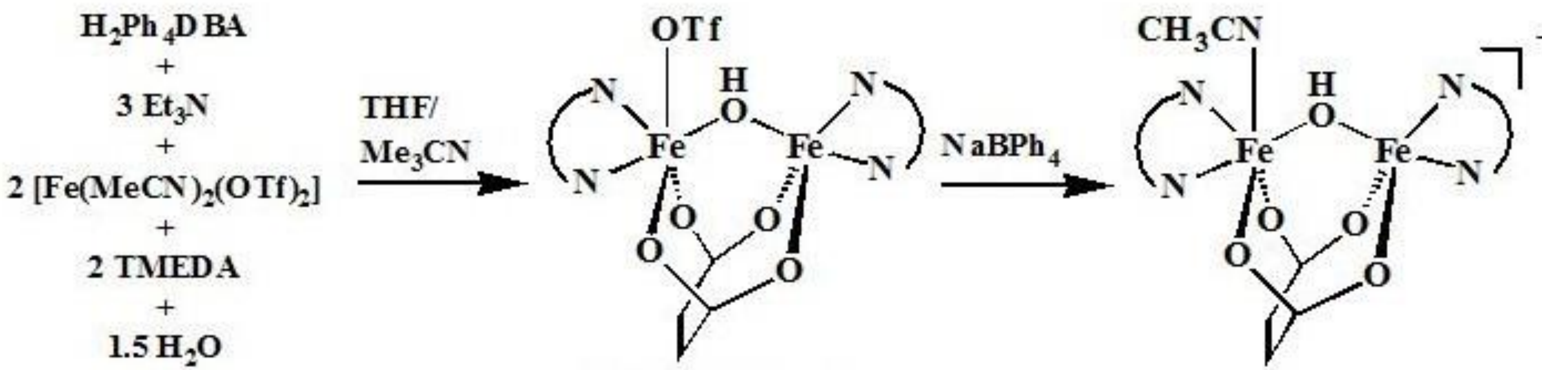
O_2 Uptake mechanism of Hr

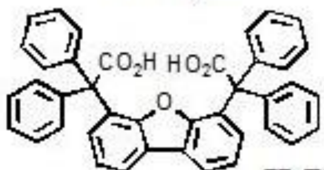


metaquohemerythrin

The uptake of O_2 by hemerythrin is accompanied by two-electron oxidation of the di ferrous centre to produce a hydroperoxide (OOH⁻) complex.

Functionally Relevant Models of DeoxyHr

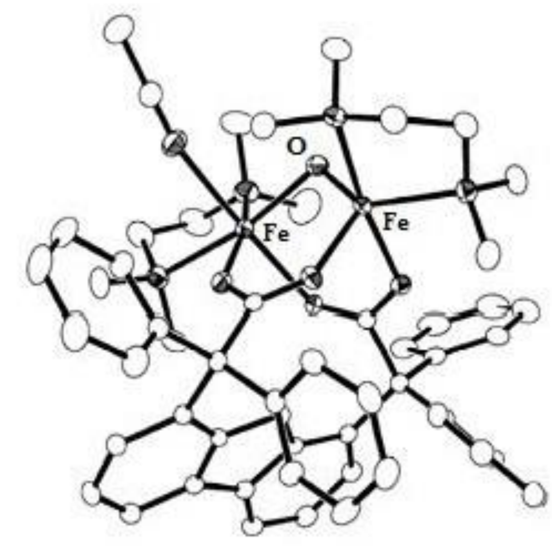




H_2Ph_4DBA

Mössbauer Parameters Identical to DeoxyHr
$\delta = 1.19$ mm s^{-1}
$\Delta E_Q = 2.81$ mm s^{-1}

Fe······Fe	3.1768(5) Å
Fe—O(H)	1.952(2), 2.034(2) Å
Fe–O(H)–Fe	105.65(8)°



$[Fe_2(\mu\text{-}OH)(\mu\text{-}Ph_4DBA)(TMEDA)_2(CH_3CN)]^+$

Thank you