GENESIS TUTORIALS

Institute for CSIR-UGC-NET/JRF, GATE & IIT-JAM

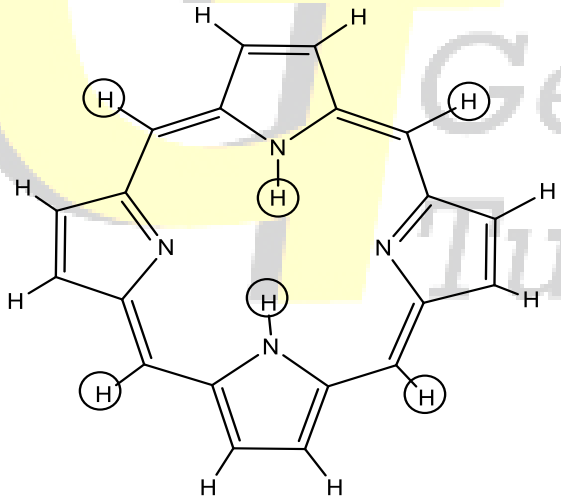
Bio-Inorganic Chemistry

INTRODUCTION

- Biological inorganic chemistry (bio-inorganic chemistry) is the study of inorganic elements as they are utilized in biology.
- The main focus is on metal ions, where we are interested in their interaction with with biological ligands and important chemical properties they are able to exhibit and impart to an organism. These properties include ligand binding, catalysis, regulation, sensing defence and structural support.
- Interdisciplinary field of chemical which largely focuses on the role of metal ions in living system.

Special Ligand

Porphine:- Derivatives porphyrin ring



Preparation:-

Properties:-

(1) Macrocyclic ligand

(2) Aromatic (Planar, π conjugated double bonds, follows Huckel Rule) (4n + 2 = 22, n = 5)

(3) If HCHO is replaced by PhCHO, then at meso position H is replaced by Ph.

(4) It is rigid due to delocalization of π electrons.

(5) Porphine molecule consists of unsubstituted tetrapyrrole connected by methylidyne (CH) bridges and these positions are labelled as $\alpha, \beta, \gamma, \delta$ or 5, 10, 15, 20 positions. (Numbering of carbon only)

(6) The porphyrin ring can accept two hydrogen ions to form the dication or donate two protons to form dianion. In metalloporphyrin complexes the inner H atoms are replaced as proton by dipositive metal ions, therefore metal free porphyrin ligand has –2 charges

(7) Spectral Properties :-

(a) NMR → 3 signal

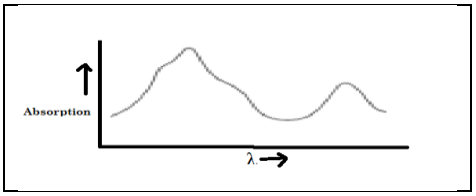
8H → attached to pyrrole (8.8 ppm)

2H → inner (attach to N) (–2 to –3 ppm)

4H → of methyledene bridges (4.4 ppm)

(b) IR → N–H stretching frequency (3300 cm^{-1})

(c) UV → Two broad bands are absorbed



NOTE:-

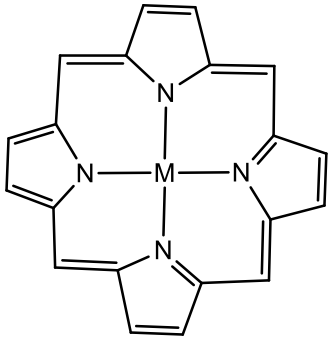
(1) The U-V visible spectrum of highly conjugated porphyrin ligand exhibit a strong absorption band at about 400 nm (Soret Band or B band) and several weaker bands (Q bands) at higher wavelengths (450 to 750nm).

(2) Both these bands arise from transition of electron from porphyrin π HOMO to the π^* LUMO

(3) It is the nature of metal centre and substituents on ring that affect the energies of these transitions and intensities of bands

(4) Metals ions (d^o, d^1, d^2, d^3 *or* d^{10}) in which dπ orbitals are relatively low in energy and do not form M → L π bonds and have little effect on porphyrin $\pi - \pi^*$ energy gap in absorption spectrum while metal ions d^n (n = 4–9) have filled dπ orbitals form metal to ligand π–bonds, this results in an increase in porphyrin π to π^* energy gap & cause hypsochromic (blue shift)

Metalloporphyrin:-



(Porphyrin have substituents at 8 pyrrole position)

(a) The size of the cavity in the centre of porphyrin ring is ideal for accommodation of metal ions of first transition series

(b) If the metal ion is too small the ring becomes ruffled to allow closer approach of nitrogen atoms to the metal ion, while if the metal ion is too large, it cannot fit into the cavity and occupies position above the ring which also becomes domed.

(c) Order of metal fitting inside the ring, according to size.

$$\underset{8}{Ni^{+2}} > \underset{9}{Cu^{+2}} > \underset{7}{Co^{+2}} > \underset{6}{Fe^{+2}} > \underset{10}{Zn^{+2}}$$

Spectral changes :-

- N–H stretching frequency disappears
- 2 signal in NMR
- Soret band becomes broad and Q band disappears

NOTE-

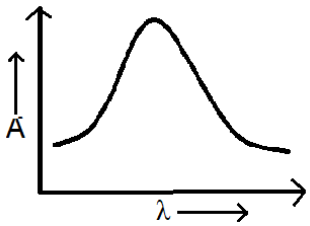
(a) $(S_0 \rightarrow S_2)$ is the soret bond

$(S_0 \rightarrow S_1)$ is the Q-band

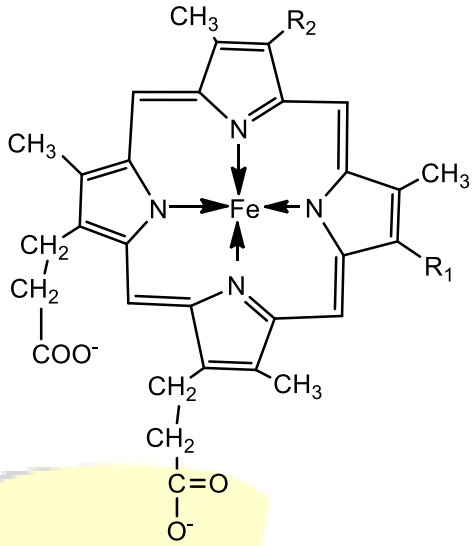
(b) An electronic transition to higher energy state S_2 is strongly allowed whereas an electronic transition to the lower energy mixed state S_1 is weakly allowed

(c) colour of metalloporphyrin (either oxidized or reduced form) is due to π (HOMO) to π^* (LUMO) transitions with in porphine ring

(d) Soret peak or soret band is an intense peak in blue wavelength region of visible spectrum.



HEME GROUP is a porphyrin ring with iron atom at center. The oxidation state may be +2 or +3.



Heme A: $R_1 = (-CH = CH_2)$, $R_2 = (C_{18}H_{30}OH)$

Heme B : $R_1 = R_2 = (-CH = CH_2)$

Heme C: $R_1 = R_2 = (-CH(CH_3)S - Protein)$

Chloroheme:- $R_1 = (-C(H) = O)$

$R_2 = (-CH = CH_2)$

NOTE:-

(1) Type A hemes are found in cytochrome a

(2) Type B hemes are found in hemoglobin, myoglobin, peroxidase and cytochrome b.

(3) Type C hemes are found in cytochrome c

(4) Chloroheme are found in chlorocruorin

All the biological uses of heme group are important, but the most important is the binding of dioxygen molecule.

Oxygen Transport System

- Heme group present → Hemoglobin
- Heme Group absent → Hemocyanin

Oxygen Storage System

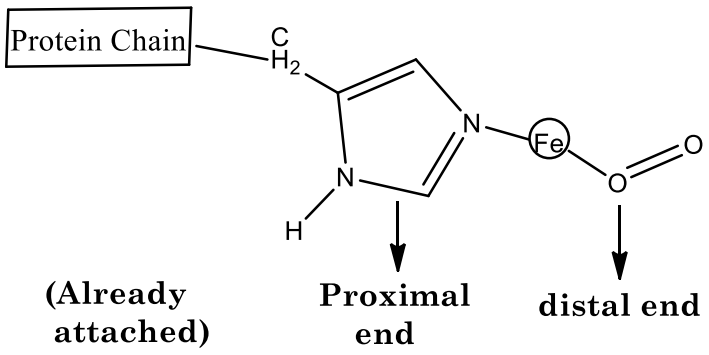
- Heme group present → Myoglobin
- Heme group absent → Hemerthyrin

(1)- MYOGLOBIN

- It is a Fe containing protein , Molecular wt. = 17000 Dalton.
- Co-ordinates O_2 reversibly and controls its concentration in tissue .
- It has a protein chain containing 153 amino acids residues folded about single heme

This restricts the access to the iron atom (by a second heme) and reduces the probability of formation of hematin-like (Fe III dimer)

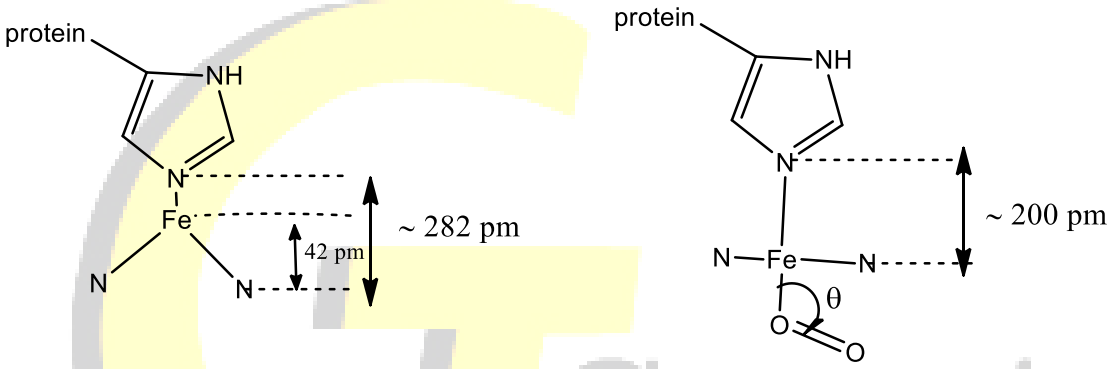
- In Mb, four co-ordination sites of Fe are attached to Nitrogen atom of four pyrrole ring, 5th co-ordination site is attached to nitrogen of imidazole ring of a proximal histidine of globin protein. The sixth vacant site trans to the imidazole nitrogen is vacant and reserved for dioxygen.
- Mb contains iron (II) in high spin state d^6 (HS).
- Iron (II) has a radius of approximately 92 pm in a pscudo octahedral environment (square pyramidal arrangement when O_2 is removed) and the iron atom will not fit into the hole of porphyrin ring and it lies 42 pm above the plane of the nitrogen atom in porphrin ring.



- When dioxygen molecule binds to Fe (II) it becomes low spin (d^6) → t_{2g}^6 (maximum CFSE → 2.4 Δo). The ionic radius of low spin iron (II) with coordination number 6 is only 75 pm.
- Fe (II) → High Spin→ $t_{2g}^4\ eg^2$

  Radius decreases

- Fe (II) → Low Spin → $t_{2g}^6\ eg^0$
- **Reason for radius decrease:-**We know that in octahedral complex, eg orbitals point towards ligands ,if they contain electron, then they will repel the ligands along co-ordinate axes, thus effective radius of iron (II) will be greater in high spin, but in low spin state all the electron are in t_{2g} so ligands approaches more closely hence ionic radius decrease.



Recent X-rays studies have shown that dioxygen is bound in a bent fashion with an Fe–O–O bond angle at 130°.

NOTE:-

(a) When oxygen or carbon-monooxide binds to 6th position, iron becomes coplanar with the porphyrin and resulting complex is diamagnetic.

(b) CO is a strong enough ligand to force spin pairing and result back π–bonding stablises the complex.

An alternative description is often considered in which bonding is expresses in terms of low spin Fe(III) co-ordinated by superoxide (O_2^-)

(2) HAEMOGLOBIN :-

- Hb consists of a tetramer of myoglobin sub units with four Fe sites that bind O_2 cooperatively.
- Molecular weight = 64500 Dalton
- Hb is found in red blood cells called erythrocytes and is responsible for their characterstic colour
- Hb picks up O_2 from lungs or gills and carries dioxygen in arterial blood to the muscles where the oxygen is transferred to another heme containing protein Mb which stores it until oxygen is required to decompose glucose to produce energy, CO_2 and water.
- Hb then uses certain amino acid group to bind CO_2 and carry it in various blood back to lungs.

Structure:

- Each Hb molecule is made up of 4 subunit, each of which consists of a globin protein in the form of folded helix or spiral .
- The globin proteins are of two types two arc α and two arc β

α globin protein consists of 141 amino acids.

β globin protein consists of 143 amino acids

- **Role of protein chain:-**

- In Hb, iron is formally Fe(II) and bonding to oxygen does not oxidise it to Fe(III), but when the protein is removed from heme group ,exposure to oxygen oxidises the iron quickly to a μ– oxo dimer containing 2 Fe(III) ions.
- The presence of hydrophilic protein around the heme, seems to present oxidation of Fe(II) ions in Hb but the presence of water alone allows oxidation of free heme.
- Thus each protein consists of one polar and one non polar group. Protein is attached to Fe(II) protoporphyrin through imidazole nitrogen of histidine residue in such a way that polar group of each protein are on the outside of the structure leaving a hydrophobic interior. Therefore, the heme group is held in a water resistant protein pocket.

Formation of Hematin (μ-oxo dimer)

- If free heme in aqucous solution is exposed to dioxygen, it is immediately converted into a stable μ-xo dimer known as Hematin.
- In hematin iron is in high spin Fe(III)
- Hematin is unable to Transport O_2

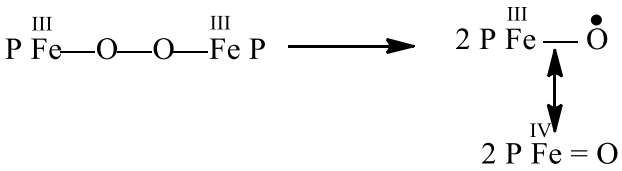
Ist step:- Is the binding of dioxygen molecule as in Hb

$$PFe^{II} + O_2 \rightleftharpoons PFe^{II}O_2 \longrightarrow PFe^{III}O_2^-$$

IInd step:- The bound dioxygen can now coordinate to a second heme forming a μ–peroxo dimer

$$PFe^{II}O_2 + Fe^{II}P \longrightarrow PFe^{III} - O - O - Fe^{III}P$$

IIIrd step:- Clevage of peroxo complexes result in two molecules of a ferryl complex with the iron in + 4 oxidation State.



Finally, the ferryl complex attracts another heme and results in formation of hematin

$$PFe^{IV} = O + PFe^{II} \longrightarrow PFe^{III} - O - Fe^{III}P$$

Hence, the globin part provides steric hindrance and prevents one oxoheme from attracting another heme.

NOTE:- (1) In vertebrates dioxygen enters the blood in the lungs or gills where the partial pressure of dioxygen is relatively high, it is then carried by red blood cells to the tissues where partial pressure is considerably lower.

(2) Hb has relatively high affinity for O_2 at high partial pressure of O_2 whereas myoglobin has relatively high affinity for O_2 at lower partial pressure

(3) Hb is saturated with O_2 in lungs, when Hb carries O_2 to muscle tissue, it experience a lower partial pressure of O_2 & its affinity for O_2 falls and in this situation affinity of Mb for O_2 is relative high therefore in muscle tissue O_2 is thermodynamically favoured transferred from Hb to Mb.

$$Hb + 4O_2 \longrightarrow Hb(O_2)_4 \qquad \text{(lungs)}$$

$$Hb(O_2)_4 + 4\,Mb \longrightarrow 4Mb(O_2) + Hb \qquad \text{(muscle tissues)}$$

This change in affinity can be attributed to there being 2 conformations.

(4) Tensed state (T) has a low affinity and Relaxed state (R) has a high affinity.

Deoxy-Hb is T, Fully loaded oxy–Hb is R.

Cooperative Effect

Binding of first O_2 molecule to the T–state is weak, but its addition, encourages the addition of second molecule to other heme subunit and this process continues till four O_2 binds to four sun units & this phenomenon is known as cooperative effect.

The successive equilibrium for binding of O_2 to 4 Fe atoms are shown:

$$Hb + O_2 \overset{k_1}{\rightleftharpoons} Hb\,O_2$$

$$HbO_2 + O_2 \overset{k_2}{\rightleftharpoons} Hb\,(O_2)_2$$

$$Hb(O_2)_2 + O_2 \overset{k_3}{\rightleftharpoons} Hb\,(O_2)_3$$

$$Hb(O_2)_3 + O_2 \overset{k_4}{\rightleftharpoons} Hb\,(O_2)_4$$

Order:-

$$k_4 > k_3 > k_2 > k_1$$

Reason:-(1) The 4 subunit of Hb are linked with each other through salt bridges between four polypeptide chains and these are formed mainly due to the electrostate interaction between the $-NH_3^+$ and $-COO^-$ present on all of 4 polypeptide chain of Hb.

(2) The protein structures in Hb consists of peptide backbone with various side chains, these side chains consists of variety of non-polar (hydrocarbons), cationic (such as $-NH_3$) & anionic (such as $-COO^-$ group). These salt bridges between the polypeptide chains in Hb are believed to introduce strain in molecule. Therefore deoxy form of Hb is Tensed state (T).

(3) Now, the binding of the first O_2 molecule to T state is weak due to strain but as binds there is a decrease in size of Fe which allows it to move into the plane of porphyrin ring and this motion is particularly important for Hb because it pulls on the proximal histidine ligand, due to which helix structure moves.

(4) This results in breaking of some salt bridges which reduces strain in Hb molecules & therefore oxyform of Hb is called relaxed state (R state).

(5) The bonding of one dioxygen molecule to a sun unit of Hb reduces steric hindrance in other subunits & encourages the bonding of O_2 molecule to iron atom of the second

subunit which in turn encourage the third as well fourth subunits hence binding of 4^{th} is easier so, equilibrium constant increase.

Hemoglobin, cooperativity and Oxygen Dissociation curve:-

- We know that partial pressure inside the lungs is much greater than the partial pressure inside the muscle tissues hence there is a pressure gradient.
- Now, what happens when oxygen reach to the blood plasma of capillaries (muscle tissue), we call blood plasma a polar substance because it mainly consists of water. O_2 is a non-polar molecule, means it is hydrophobic and will not easily dissolve in blood plasma. Basically our blood carries a special type of carrier protein Hb. Hb picks up the O_2 molecule and protects them from hydrophilic environment.
- Basically, Hb consists of 4 poly peptide subunit, with in each subunit, hydrophobic sections are inside & there is heme group and each heme group, consists one Fe which binds single O_2.
- Molecule, so a maximum of $4O_2$ molecules can be binded by 4 heme with O_2.

A fully saturated Hb with O_2 is called oxy hemoglobin

But when it does not contain O_2 it is called Deoxy Hb.

POSITIVE COOPERATIVITY

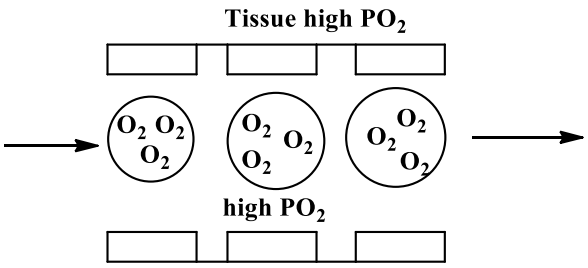
When one oxygen molecule binds to deoxy Hb, it causes a conformational change in the structure of Hb. Thus makes the other three heme groups much more likely to bind other O_2 molecules. Likewise when oxy Hb is saturated with four O_2 molecules and one of the O_2 molecules dissociates, it causes a conformational change that increases the likelihood that other three O_2 molecules will dissociate. This behavior is called Positive cooperativity.

What will make our Hb, actually release oxygen?

* O_2 naturally travels from high partial pressure of O_2 to lower partial pressure of O_2 (PO_2).

* As Hb carriers O_2 in blood, it protects the non-polar molecule from the polar plasma. Therefore, oxygen remains comfortably with the Hb until the Hb arrives at the tissues of body.

(Outside of protein is polar, while inside is non-polar)



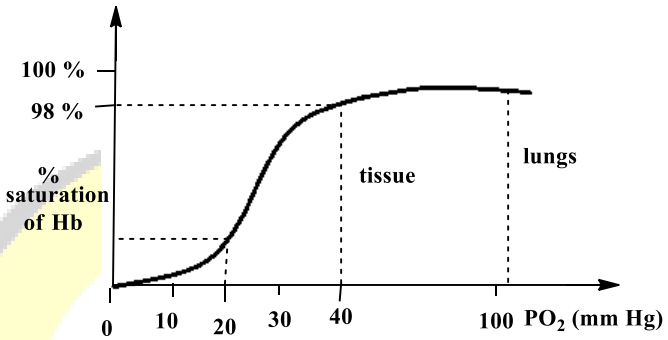
Why partial pressure is low inside tissues??

Ans:- Because cells insides tissues continously use O_2 to break glucose and generate energy hence O_2 has been used up so partial pressure of O_2 decrease.

Now, because O_2 will move naturally from high partial pressure to low partial pressure, the Hb will unload O_2 and as one molecule dissociate, it increases the tendency for further dissociation or enhance the further dissociation of O_2.

Now,

If we plot the % of Hb saturated with oxygen versus PO_2 we obtain the oxygen dissociation curve



This curve has 'S shape' ie. sigmoidal shape and the reason for this shape is positive cooperative activity.

Analysis of graph:-

⟹ At or around 100 atm pressure, about 98% of Hb is fully saturated with O_2.

⟹ Flat slope basically means, even if partial pressure decrease to 80mm of Hg inside lungs most of Hb is still be able to pick up O_2. Hence, the flat curve tells us that, even if the PO_2 in lungd drops, Hb will still pretty much saturated.

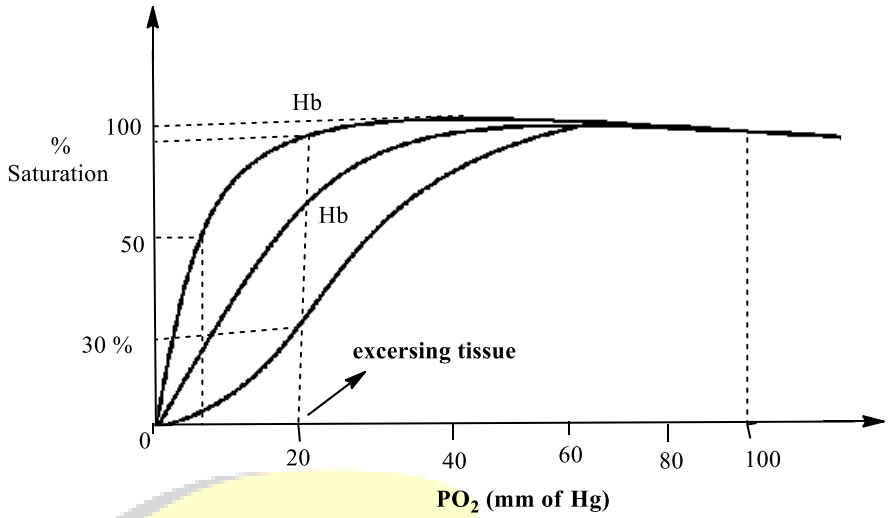
⟹ If we are not exercising, the average PO_2 will be around 40mm of Hg, the % saturated will be around 70% that means Hb will unload some oxygen but if we are exercising then PO_2 drops (say around 20 mm of Hg) means that below 20 mm of Hg a small drop in pressure basically unload all oxygen, but on right side we see that even small change in pressure (drop) the O_2 can still bind to Hb.

Myoglobin v/s Hemoglobin Dissociation curve:-

- Hb is the primary oxygen carrier within our blood, it delivers O_2 from lungs to tissues and we know that it consists of four individual polypeptide sub units and these can interact so Hb exhibits cooperativity.

  Means if we take a fully unsaturated Hb protein & one of the group accepts O_2 molecule then that will create a conformational change in entire structure and make other heme group to make them much more likely to accept O_2 molecules Likewise, if we have fully saturated oxy

Hb molecule & one of heme group unload O_2 then there will be change in conformation structure and it will make easy for other 2 heme gi to release oxygen.



→ Once O_2 is delivered to our muscle tissue, the tissue must be able to store the oxygen for later use. This is done by special protein called Myoglobin.

→ Mb, unlike Hb, consists of a single polypeptide subunit that contains single heme group. Mb can only bind a single O_2 molecule. In addition Mb does not display cooperativity and so does not create a sigmoidal curve. (Hyperbolic curve)

Physiological significance

⟹ Inside our lungs, the PO_2 is around 100 mm of Hg. According to curve, both Mb & Hb are about 98% saturated with oxygen

⟹ In exercising tissue, the partial pressure can drop to 20mm Hg so, according to the Hb curve, the % saturation of Hb in the tissues is around 32 % However, the % saturation of Mb at same partial pressure is about 9 %

$$\Delta\% \text{ of Hb} = 98\% - 32\% = 66\%$$

$$\Delta\% \text{ of Mb} = 98\% - 91\% = 7\%$$

⟹ This data tells us that Mb binds O_2 much more tightly than Hb and would not make a good carrier of O_2 in the blood because it would not unload the O_2 effectively to our tissue Mb can however store O_2 in our muscle tissue until very low partial pressure of O_2.

Hence 66% of Mb will unload oxygen

While 7 % of Hb will unload oxygen

Hence, Mb tightly bind oxygen

BOHR EFFECT

⟹ The Oxy-Hemoglobin dissociation curve describes the affinity for O_2. These are several factors that affects the affinity for O_2

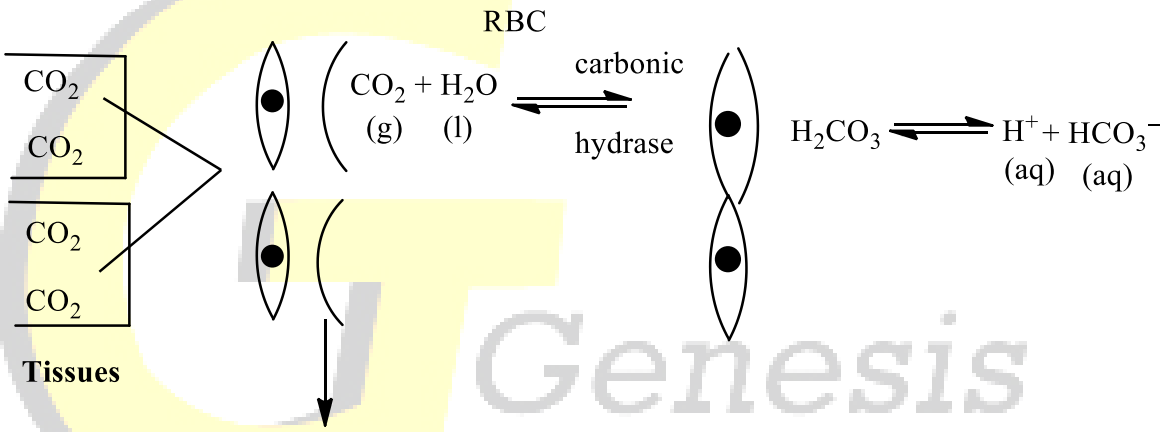
pH & Bohr effect

⟹ CO_2 is the major by-product of cellular metabolism, like O_2 (CO_2 is non-polar)

[Two polar bands O = C = O points in opposite directions dipole moment cancels out]

Question-How to solve this problem?

⟹ Our body converts CO_2 into a slightly different form to make it more soluble in our blood plasma, when our tissues are exercising, CO_2 is produced in large amount & they diffuse across the capillary tubes.



Diffuses along the capillary walls and enters RBC in blood plasma in RBC's there is a special type of catalytic enzyme called Carbonic hydras.

It combines CO_2 & H_2O to give H_2CO_3 (carbonic acid), it is a weak acid so it dissociates into H^+ & CO_3^-, because these two ions have charge so they are soluble in our blood plasma.

Hence exercising tissues produces a large number of CO_2, automatically mores to RBC's & convert into H^+ & HCO_3^-.

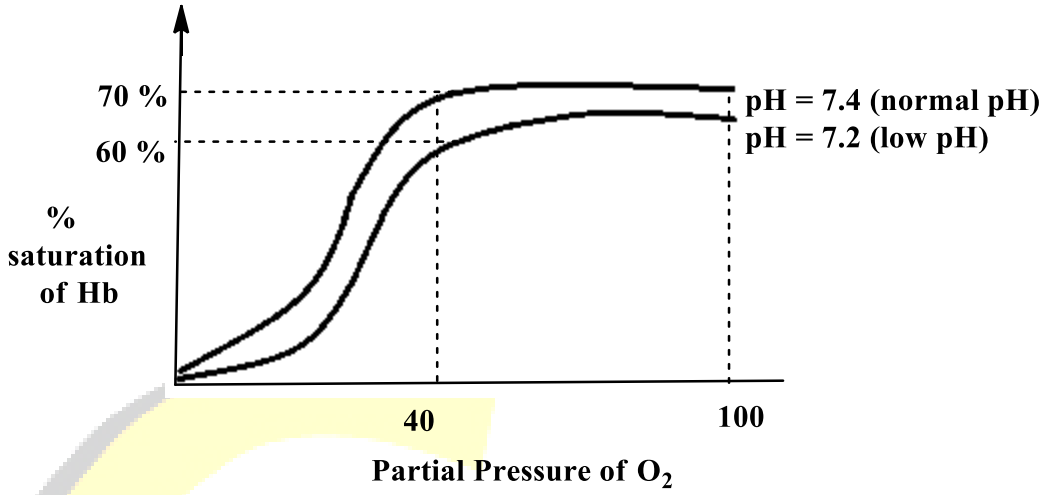
Now, concentration of H^+ ions → pH

More the H^+ ions, our blood plasma will be acidic and this effect is called Bohr effect.

NOTE As the concentration of H^+ inside blood increase, pH decrease, this in turn affects Hb ability to bind O_2. This effect is called **Bohr effect**.

Ques. How the presence of CO_2 & H^+ affects the affinity of O_2 to bind to Hb?

Ans H^+ ions (CO_2) bind to Hb at special allosteric sites and create conformational changes, they decrease the ability of Hb to bind oxygen. This shifts the entire dissociation curve to shift right.



Now, When tissues began to exercise, we produce more H^+ ions then the curve shifts right. This is called Bohr effects.

The average partial pressure of O_2 in our tissues is 40 mm Hg. At lower pH values (exercising tissue) shifting the curve to the right means the Hb is more likely to unload O_2 into our tissue.

→ At a pH of 7.4, 70% of Hb is saturated (30% is unloaded) while at a pH of 7.2, 60% is saturated (40% is unloaded)

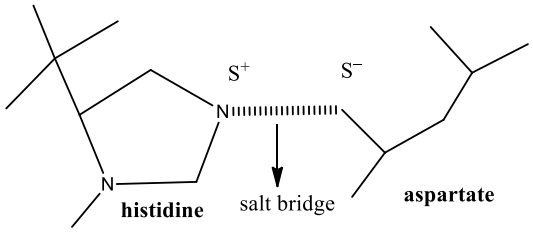
→ Notice that a change in pH does not really affect the affinity of Hb at high partial pressure values. Physiologically, this is beneficial because we do not want Hb to bind less oxygen in our lungs. We see that both curves shows that at a pressure of 100 mm of Hg, the % of Hb fully saturated with O_2 is about 98%

Hence CO_2 and H^+ ions, together effect the affinity (Bohr effect)

At higher concentration of CO_2 & a lower pH, shifts the curve to right thereby decrease the affinity of Hb for O_2 & allowing it to unload more O_2 to tissues.

Question How this actually takes place??

Ans In protein chain of Hb there are several molecules that can bind H^+ ions (the amino groups of the terminal residue of α –sub units and the histidine amino acids) and they participate in stabilizing salt bridge



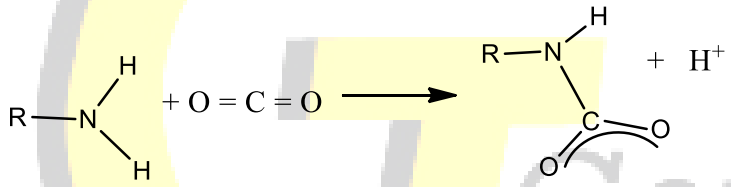
* As the H^+ ions concentration increase, the histidine chain becomes protonated. This allow it to form a salt bridge (strong electric interaction) with nearby aspartate residue. The salt bridge stabilizes the T–state of deoxy Hb, which lowers its affinity for oxygen.

* Non polar O_2moves into RBC, where it is converted by carbonic anhyclrase into H^+ & HCO_3^- ions.

* A high concentration of CO_2 means more H^+ ions are produced. This decrease Hb affinity as a result of salt bridge formation.

* CO_2 can also bins to the amino terminal groups, group of subunits.

Ques. How CO_2 directly affects??



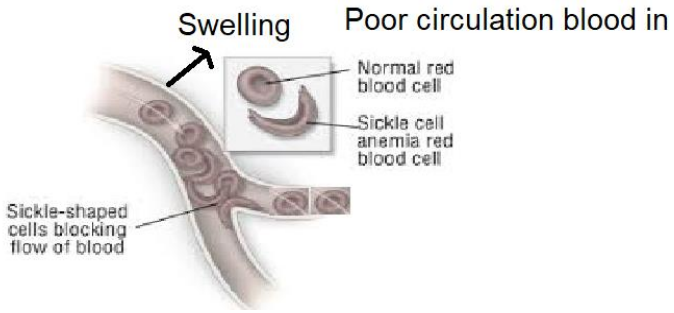
This will form–negatively changed carbonate groups which can participate in forming salt bridge that stabilises the deoxy Hb Terresed state

Myoglobin does not exhibit a sigmoid curve nor a Bohr effect

GENETIC DEFECTS

(1) Sickle cell Anemia

→ Individual sickle cell anemia have abnormal Hb molecules and the Hb molecules in their deoxygenated state begin to aggregate with one another to form long sickle shaped fibres, which converts the healthy biconcave shape of red bloods into sickle shape.



→ Normal healthy biconcave shape red blood cells, due to their biconcave shape they will easily squeeze and pass through capillaries and they will nor aggregate to one another bit when RBC transform their biconcave shape to sickle shape, they began to aggregate & cause clogging of these capillary, due to which pressure rise on other side & it leads to painful swelling

→ These sickle-shaped red blood cells can aggregate & clog tiny capillaries resulting in

(a) Painful swelling

(b) Impaired blood flow

(c) Increased risk of stroke (Poor blood flow to brain)

(d) Increase risk of pathogenic infection (poor blood circulation)

→ In addition, these abnormal red–blood cells have a shorter life span, this means they will die at a quicker rate and this can lead to a decrease in RBC count (anemia) a lower RBC count.

Question What causes the abnormal shape of Hb??

→ It is a genetic error that leads to a single amino acid substitution in the beta chains of Hb.

Glutamate 6 replaced by valine 6

→ Instead of synthesizing glutamate at 6^{th} position is valine at 6^{th} position

→ The problem is that there properties are different

glutamate ↓ polar molecule

valine ↓ non – polar molecule

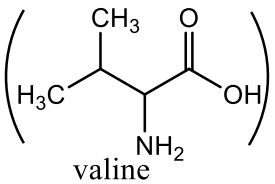
→ This will create a difference

So it will points to the surface outside of blood plasma

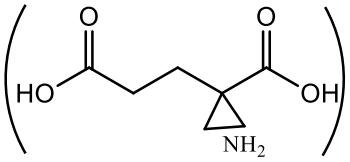
↓

Blood plasma consists of mainly water and it will interact with water molecules

We have 2 βchain in Hb & both of them have abnormalities



valine



cluster

Hence, the β –subunit of the normal Hb have a polar residue at the 6^{th} position that can interact with the polar water molecule found in bold plasma

In the abnormal Hb, the non-polar valine in the 6^{th} position would much rather interact with other nonpolar molecules rather than with H_2O

* i.e. if other deoxy Hb is in close proximity then they will aggregate and this process continues and they will have sickle cell shape and biconvular shape is deformed

* Aggregation only tales place in deoxy-Hb because in oxy Hb they lie inside

(2) Absence Methemoglobin Reductase

(a) Methemoglobin is a metalloprotein in which iron in the heme group is in +3 0.5 (not on + 2 of normal Hb)

(b) The size of Fe^{+3} ion is so small that it can fit into porphyrin ring of Hb without binding O_2 and therefore it prevents transfer of O_2. In human blood a trace amount (about 3%) of methemoglobin is normally produced spontaneously.

(c) Fe^{+3} has on increased affinity for O_2 binding and the binding of O_2 to met Hb results in an increased affinity of O_2 to other heme subunits that still contains Fe^{+2} ions with in same Ub molecule. This leads to an overall reduced ability of RB cells to release O_2 to muscle tissue.

(d) An enzyme met Hb Reductase converts the met Hb back to Hb and if there is absence of this enzyme, then there will be a higher level of met Hb which causes a disease Met hemoglo binemia.

→ bluish color

→ If it is present in excess, blood becomes abnormally dark bluish brown.

→ Normally 1 or 2% of person's Hb is MetHb.

1–2 % → Normal

< 10 % → No symptom

10–20% → skin discoloration

20–30% → Headache, Anxiety

30–50% → Fatigue, confusion, dizziness.

50–70% → coma

> 70% → High risk of death

Other Biological Dioxygen Carriers:-

(Non–heme)

Hemerthyrin ← → Hemocyanin

Hemerthyrin:-

(a) It is non-heme iron containing octameric protein found in marinc invertebrates (molluscs & arthapoda)

(b) Iron is present in +2 oxidation state and binds O_2 reverisibly

(c) It consists of 8 subunit each with 113 amino acid residences and a two Fe(II) active site

NOTE:-

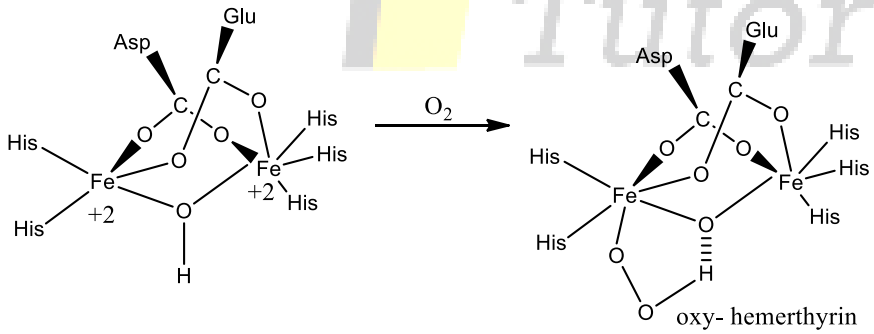
→ Major difference between Hb and hemerthyrin is the binding of O_2. In Hb one O_2 binds per Fe(II) ion whereas in hemerthyrin one O_2 binds to two Fe(II) ions.

→ Unlike Hb, hemerthyrin exhibits no cooperativity between the subunits during O_2 binding.

Structure:-

(1) Each subunit consist of 2 iron active sites connected by 3 bridging groups, two of which are carboxyl anions from glutamate and aspartate and other is bridged by oxygen atom (from water, hydroxo or oxo).

Deoxy hemerthyrin



Remaining ligand are 3 histidine unit on one iron atom & two histidine on other

→ One iron atom is Penta coordinated while the other is hexaco-ordinated

→ So the O_2 binds at a Penta coordinated Fe atom and converts to oxyhemethyrin.

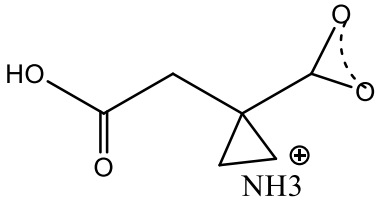
→ When O_2 binds to the vacant site two electron are transferred from two Fe(II) centre (one e^- from each Fe(II) subunit) to O_2 resulting in Fe(III) and peroxide (O_2^{-2}). The proton from the hydroxo bridge shifts to bound peroxide resulting in HO_2^- group.

→ The μ–oxo bridging group is associated with bound HO_2^- group H–bonding.

→ Mossbauer data indicates that the two Fe(III) atoms are in different environment in oxy hemerythrin.

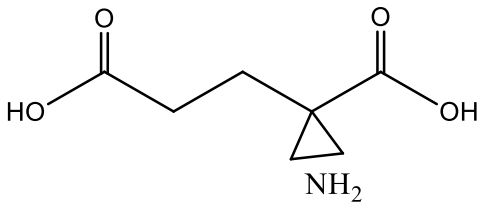
NOTE:

As partic acid



α-amino acid

Glutamic acid



α-amino acid

└ It ia non-essentail in humans because the body can synthesise it MSG (mono sod glutamate)

→ molecular. Wt = 108000

→ It is a non-heme protein

→ Transport O_2

→ Total unit = 8

→ Total Fe present = 16

→ One unit binds one O_2 so total hemerthyrin transfers eight O_2 molecules

→ Both Fe in +2 oxidation state

→ One Fe is hexa co-ordinated & other is Penta-coordinated.

→ Two Fe atom bridged by hydroxyl group & carboxylate chain of glutamic & aspartic acid.

Deoxy Form	**Oxy form**
→ The two Fe(II) center in deoxyhemerthyrin are strongly anti-Ferro magnetically coupled through Fe–O–Fe brodge	→ Antiferromagnetic coupling
→ So it is diamagnetic	→ diamagnetic

→ EPR inactive → EPR inactive

NOTE:-

(1) The structure of deoxy and oxy form of Hemerthyrin have been determined crystallographically.

(2) In deoxy form, a hydroxid–bridged $[Fe(II)]_2$ unit is present.

(3) The hydroxyl group (H atom) participates in O_2 binding, becoming part of $[HO_2]^-$ ligand.

(4) In both oxygenated and deoxygenated state it is diamagnetic.

Note:-	$\upsilon_O = 0$	1560 cm^{-1}	↓ stretching frequency Decrease
Superoxide	$\upsilon_O = O^-$	1100 cm^{-1}	
Peroxide	υO_2^{-2}	800 cm^{-1}	

NOTE:-

$Fe^{+2} - OH - Fe^{+2}$	deoxy (reduced)
$Fe^{+3} - OH - Fe^{+3}$	(semi-met)
$Fe^{+3} - OH - Fe^{+3} - OOH^-$	oxy (oxidised)
$Fe^{+3} - OH - Fe^{+3}$ – (any other ligand)	met (oxidised)
* Colourless	Voilet-Pink

Haemocyanin:-

(1) It contains neither the heme group nor the cyanide ion, the name simply means **blue blood.**

(2) Found in many species in Mollusca and Arthopoda.

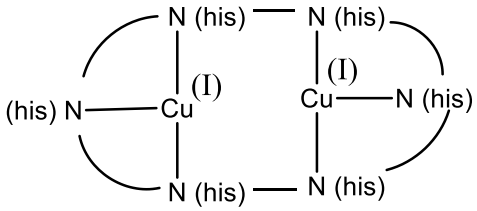
(3) It is a Cu containing protein which serves as O_2 carrier.

Mollusca → (snails)

Arthopoda → (crabs, lobsters)

Structure:-

Deoxy

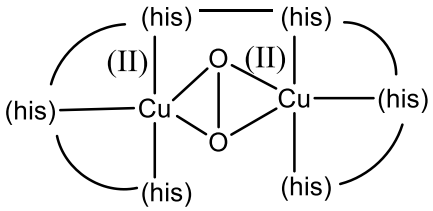


(Cu---Cu = 460 pm)

→ Cu (I) oxidation state

→ Colourless

oxy



(Cu ---- Cu = 360 pm)

→ Cu(II) oxidation state

→ Coloured (blue)

O^{-2} — Cu^{+2}

LMCT

→ Number of units are very large

→ Cu containing protein

Note → O_2 unit is bound in a bridging mode with an O–O bond length of 140 pm, typical ie found in peroxide complexes

→ O_2 binding is shown as:-

$$\boxed{Cu\ (II) - [O_2]^{-2} - Cu(II)}$$

ie e^- transfer accompanies O_2 binding

→ Resonance Raman spectroscopic data have shown that $\upsilon(O–O) \approx 750\ cm^{-1}$

→ The Cu(II) centres are strongly anti ferromagnetically coupled with $\mu[O_2]^{-2}$ ligand so oxy-Haenocyanin is diamagnetic.

→ Haenocyanin is oligomeric with each monomer containing Cu atom in close proximity

→ In deoxy state, each Cu atom is 3 Coordinate and bound in a pyramidal array by 3 histidine residues

NOTE:-

* The two Cu atoms are so far apart (460 pm) that there is no direct interaction between them. The low coordination number is typical of Cu (I) which is two or typical of four co-ordinated.

* Rapid and reversible co-ordination of O_2 occurs between two Cu atoms in a bridging dihapto manner. To accommodate the binding of O_2, the protein adjusts its conformation to bring two Cu atoms closer together and Cu sites becomes 5–coordinated.

Electron Transport System

In body different process needs electron ie. in different processes involvement of electron is there so electron transporters are required

electron transport system

Heme group present	Heme group absent
Cytochrome	→ Fe–S Protein
	→ Blue Cu protein

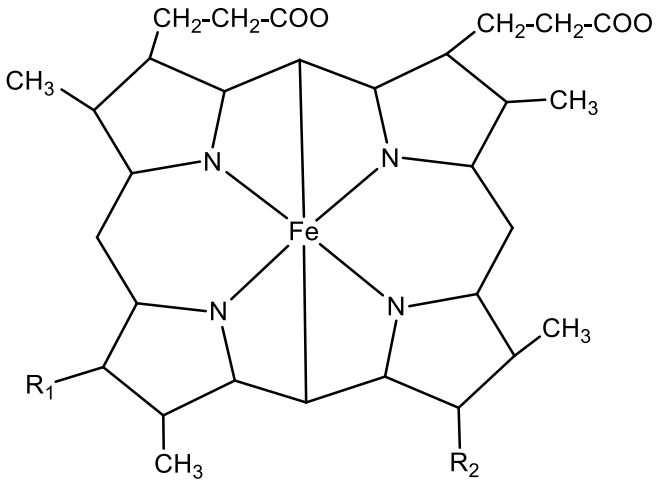
CYTOCHROMES

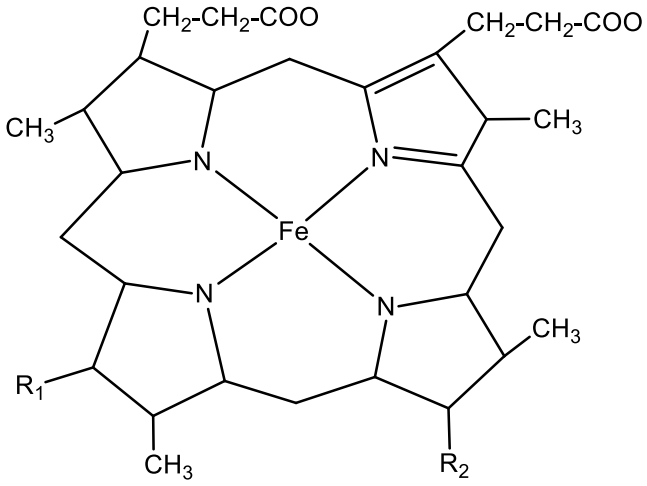
→ operate in potential region of –0.3 to 0.4V

(1) Found in both plants and animals and serve as electron carrier

(2) They contain heme group (Fe–porphyrin group)

(3) Three main types of cytochrome:-





NOTE:- 3 types
→ cytochrome a
→ cytochrome b
→ cytochrome c
They differe with respect to substituent on the periphery of heme. Many other cytochrome also exists ex. Cytoxidiese and cytP_{450}

Heme A:- $R_1 = CH = CH_2$

$R_2 = C_{18}H_{30}OH$

(found in Cytochrome - a)

Heme B:- $R_1 = R_2 = CH = CH_2$

(found in Hb, Mb, cytochrome b, peroxydase)

Heme C:- $R_1 = R_2 = CH(CH_3)$ S–protein

(found in cytochrome C)

(4) Here, the Fe(II) of heme group is attached to a N–atom of imidazole ring of histidine residue on the side of porphyrin plane, the sixth coordinated site of Fe(II) is occupied by tightly bound by S atom from a methionine residue of protein i.e. cytochromes are inert not only to oxygen but also for CO.

(5) Nature of Metal:-

Reduced form : Fe (+2)
oxidised from: Fe (+3)
} low spin in both oxidation state

(6) Reduction potential

Cyt b (0.26V)

Cyt c (0.26V)

Cyt a (0.4V)

The difference in reduction potential for Fe(II) → Fe(III) oxidation result from changes in porphyrin substituents

(7) To see the capability of cytochromes for fast electron transfer we consider that the d–orbital of Fe (c rich) (t_{2g}) orbitals overlap with the orbital of porphyrin

Overlap between the t_{2g} orbital of Fe and low lying empty π^* orbitals on porphyrin effective extends the Fe orbitals out to the periphery of ring

(8) **Oxidized form** **Reduced form**

Fe (III)	Fe(II)
→ t_2g^5	→ t_2g^6 eg°
→ Paramagnetic	→ diamagnetic
→ ESR active	→ ESR inactive

(9) The potentials are such that e^- flow is (b → c → a → o_2)

NOTE:- Some of a type (cytochrome C-oxidase) are capable of binding O_2 molecules & reducing them, thus they are at last link in the respiratory chain of e^- and so they must be. 5 coordinated (in absence of O_2).

NOTE:- cytochromes are capable of performing, oxidation & reduction

Cytochrome c oxidase:-

→ It contain two heme group and 2 Cu atoms (Cu A & Cu B) of cytochrome type (a & a_3)

→ It is the terminal member of electron transfer chain and catalysis the reduction of O_2 to H_2O and contains four active metal centres i.e CuA, CuB, heme a & heme a_3

→ electron transfer involves the CuA & heme a sites i.e. e– being transferred from cytochrome C to CuA & then to heme a.

→ Heme a_3 & CuB provides the site for O_2 binding & O_2 to H_2O conversion and involved in pumping H^+ (four per O_2 molecule across mitrocondrial inner membrane)

Cytochrome P–450

(1) Cytochromes P–450 are metalloenzymes which function as monooxygenase and catalyse the insertion of oxygen into C–H bond of an aromatic and aliphatic hydrocarbon ie. the conversion of RH to ROH

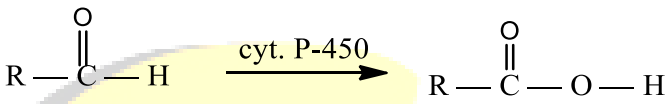
NOTE: Oxygenases are enzymes that insert oxygen into other molecules

a monooxygenare inserts 1 oxygen atom and dioxygenase insert 2 oxygen atom

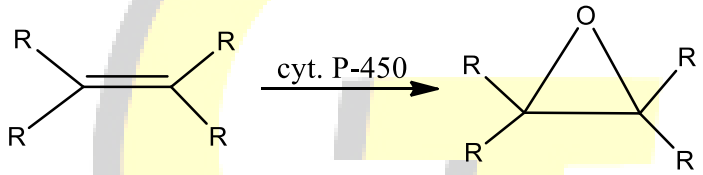
$$RH + O_2 + 2H^+ + 2e^- \xrightarrow{\text{cyt.P}-450} ROH + H_2O$$

→ Conversion of R.H (hydrocarbon) to ROH

→ conversion of an aldehyde to carboxylic acid



→ Conversion of alkene to epoxide:-

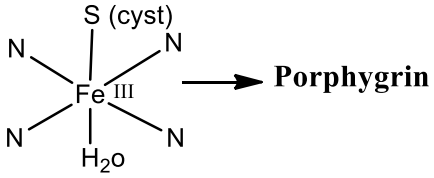


Hence, they facilitates the cleavage of O_2 and insert oxygen atom into substrates (C–H→ C–OH)

→ One oxygen atom is inserted into an organic substrate and one atom is reduces to H_2O.

Structure:-

(1) Active site in cytochrome P-450 is heme unit and here iron is present in +3 oxidation state



(2) The fifth co-ordinate site of iron is bound with the S-atom of cysteine

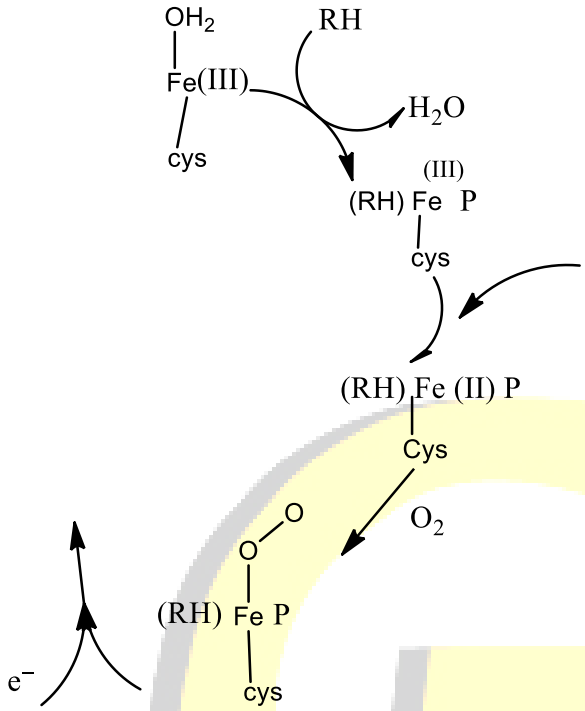
(3) In the rest state, it contains low spin Fe(III) centre.

NOTE:- Carbon–monoxide adducts of cytochromes P–450 absorb at 450 nm and this is the origin of name of enzyme,

Catalytic cycle

1st step→ binding of the organic substrate RH to the active site of metalloenzyme & loss of bound H_2O ligand

2nd 1–electron reduction of low spin Fe(III) to low spin Fe(II)



(3) → Binding of O_2 to give an adduct following by 1–electron transfer from iron to produce an Fe (III) peoxide complex.

(4) → acceptance of another electron to give {Fe (III)–O–O⁻} species which is protonated to {Fe(III)–O–OH}

(5) → Further protonation and loss of H_2O leaving an Fe(IV) = 0 species with a porphyrin ring formally a radical cation

(6) → Transfer of the oxido O atom to the bound R–H substrate and release of ROH with simultaneously binding of a H_2O ligand to active site of metalloenzymatic which once again contains Fe(III)

NOTE:- The insertion of O into the C–H bond of R–H is thought to involve a radical pathway

(2) **Iron–Sulphur Protein**

(1) They are non–heme iron proteins and are responsible for e^- transfer in plant & bacteria.

(2) Relatively low molar mass

(3) Contain 1, 2, 4 or 8 Fe atoms

(4) Generally, they operate at more negative potentials than cytochromes.

(5) They are composed of high spin Fe(III) or Fe(II) with sulfur ligands in tetrahedral environment.

(6) Thay have low range of reduction potentials (–0.50V to –0.5V), therefore they acts as a reducing agent in biochemical process

(7) Different types of these are involved in photosynthesis, nitrogen fixation

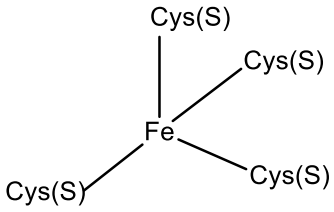
Ruberodoxin Ferredoxin

(1) <u>Ruberodoxin</u> → 1 electron transfer agent

⟹ Found in anaerobic bacteria (organism that does not require oxygen for growth), it patriciates in biological redox reaction

⟹ It is simplest non-heme iron protein and contains only one Fe atom

⟹ Fe atom is co-ordinated to 4 sulphur atom in distorted. Tetrahedral manner and they belong to amino acid system in protein chain. Iron is in + 3 oxidation state.



⟹ It has no labile Sulphur

(i.e inorganic Sulphur S^{-2} which can be liberated as H_2S by treatment with mineral acid, inorganic Sulphur atom are not a part of the protein instead they form bridges between Fe atom).

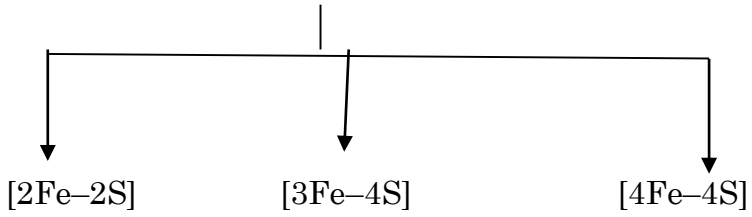
NOTE →

(●) Fe–S distance is 224 to 233 pm and S–Fe–S bond angle is 104 to 114°

(●) when Fe(III) is reduced to Fe(II), there is a slight increase in Fe–S distance but Both Fe(III) and Fe(II) are high spin in Tetrahedral geometry.

(2) Ferridoxins

Non-heme iron proteins with more than one iron atom



Note Natation:- [FenSn]

Atom in protein active site acid labile Sulphur

$$Fe_nS_n \xrightarrow{HCl} H_2S \xrightarrow{[O]} S \text{ (elemental form)}$$

(1) Fe_2S_2or [2Fe–2S] ferridoxins

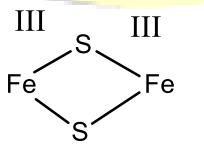
Called as plant ferredoxins and act as 1 electron transfer agents

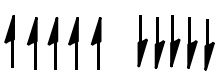
Very acidic proteins

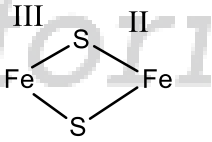
Acid labile Sulphur = 2

Acid non labile Sulphur = 4

Oxidized form **Reduced form**







S = 0 S = ½

Diamagnetic

EPR inactive EPR active Paramagnetic

Both Fe(III) are antiferromagnetically

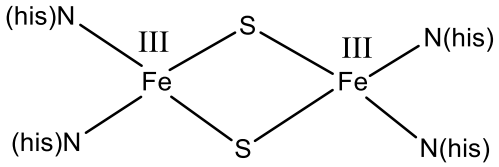
coupled via bridging atoms

⟹ [2Fe–2S] ferridoxins contain two Fe centers, bridged by two S^{-2} ligands with Tetradedral co-ordinated sphere each metal completed by 2 cysteine residue.

REISKE PROTEIN:- If two cysteine units are replaced by 2 histidine residues, then we obtain reiske protein.

⟹ It has a positive reduction potential in contrast to the negative values of [2Fe – 2S] ferridoxins and this difference is due to the His versus Cys co-ordination of one Fe–center

⟹ The reduction potential of Rieske FeS Centre is very pH dependent.



(2) [3Fe–4S] or Fe_3S_4 ferredoxin (one electron transfer agent)

⟹ Contains 3 Fe and four S^{-2} centres arranged in approximately cubic frame work with one corner vacant

→ Number of Fe = 3

→ Number of labile S = 4

→number of non-labile S = 3

Oxidised Form

Fe(III) Fe (III) Fe (III)

Antiferr O magnetic coupled

d^5

$S = {}^5/_2$ (Paramagnetic)

ESR active

Reduced Form

Fe (II) Fe (III) Fe(III)

Antiferr O magnetic coupled

d^6

S = 2

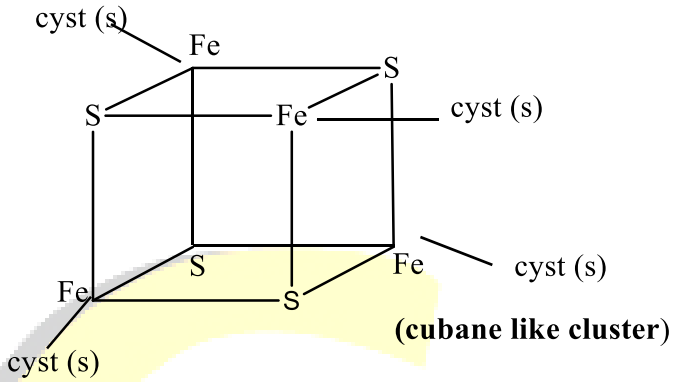
Paramagnetic ESR active

[4Fe–4S] or Fe_4S_4 ferredoxins:-

→ Most common and most stable ferredoxins

→ Found in bacteria and involve in anaerobic metabolism

→ These resembles [3Fe – 4S] ferredoxins but contain an additional Fe S(cyst) group which completes the approximately cubic cluster core.



(cubane like cluster)

→ Number of Fe atom = 4

→ Number of labile Sulfure = 4

→ number of non-labile S = 4

→ It is also 1 electron transfer agent

→ It contains 2 Fe(III) and two Fe(II) ions in oxidized form, one Fe (III) and 3 Fe(II) ions in reduced form.

→ In both forms Fe(II) and Fe(III) ions are high spin

→ **<u>Oxidised form</u>**

Fe(III)	Fe(III)	Fe(II)	Fe(II)
d^5(HS)	d^5(HS)	d^5(HS)	d^5(HS)

Cantiferromag netically coupled (S = 0)

→ diamagnetic

→ EPR inactive

→ **<u>Reduced form</u>**

Fe(III)	Fe(II)	Fe(II)	Fe(II)

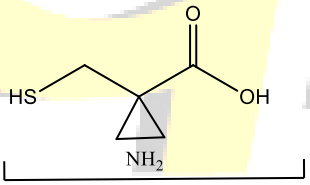
d^5(HS) d^6(HS) d^6(HS) d^6(HS)

S = ½ S = 0

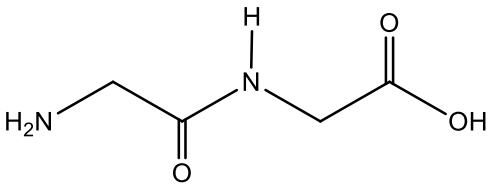
In the reduced form

Fe(III) and Fe(II) ions also couple antiferromagnetically resulting in (s = ½) therefore, it is paramagnetic & EPR active.

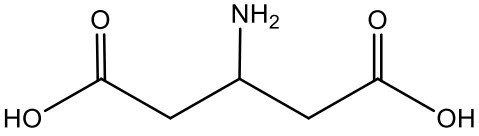
NOTE:- Cysteine –



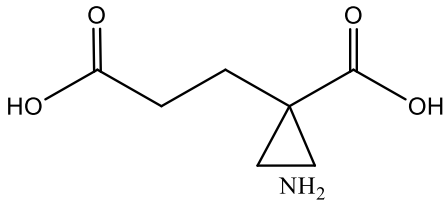
Methionine–



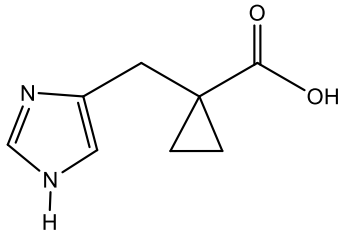
Glycine (gly)-



Aspartic acid



Histidine



Note:- 3 most important redox system

(1) high spin (Td) Fe(II) | Fe(III) in Rubredoxin, Ferredoxin

(2) Low spin octahedral Fe(II) | Fe (III) in Cytochromes

(3) Pscudotetrahedral (Cu(I)/Cu(II) in blue Cu protein such as Plastocyanin

Azurin and Stellacyanin

NOTE:- Gray has pointed out that these redox centers are ideally adopted for electron exchange, in that no change in spin state occurs and there is little or no movement of the ligand. The Frank Condon activation barrier will be small

Blue Cu protein:-

⟹ Combination of Cu and protein chain

⟹ Involved in electron transfer

⟹ Intense blue colour protein.

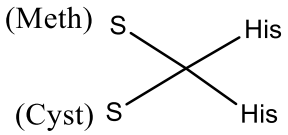
Oxidised form	**Reduced form**
Cu^{+2} (d^9)	Cu^{+1} (d^{10})

Plastocyanin:- mol wt = 10500 (between 97 and 104 amino acids)

⟹ Cu (I) so usually found in tetrahedral environment

⟹ Cu(II) d^9 and is usually octahedral co-ordinated with JTD (point towards square co-ordination)

→ In this case of plastocyanin, Cu is situated in a "flattened Tetrahedron" halfway between two geometries distorted Tetrahedron or pseudo Tetrahedron



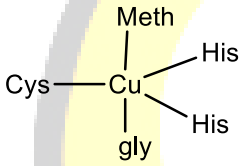
Plastocyanin is present in higher plants and blue–green algae cover all the plants which can be planted all trees, shrubs, flowering herbs etc).

→ And transport e^- between photosystems I and II

Azurin :-

Occurs in some bacteria and are involved in electron transport in the conversion of $[NO_3]^-$ to N_2 (mol.wt = 14600)

And protein chain contain 128 or 129 amino acid.



(Triogonal bipyramidal grometry)

NOTE:- Both have high reduction potential (measured at pH = 7)

+ 370 mv (plastocyanin)

+ 308 mv (azurin)

NOTE: Blue Cu protein

Name arises due to intense blue colour in oxidized state which arises due to ligand (thiolate) to metal charge transfer (LMCT)

METALLOENZYMES

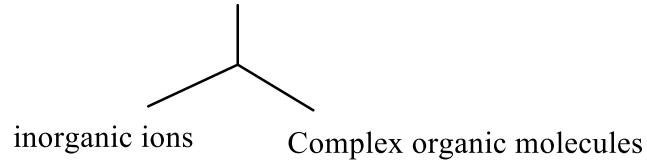
Enzymes:- Enzymes are larger protein molecules that catalyses a large number of biochemical reaction.

→ They generally increase the rate of reaction by lowering the activation energy

→ Basic structure of enzymes are built of proteins called **Apoenzyme** and a small prosthetic group (metal ion).

Example Heme is a prosthetic group in Hb

COFACTOR:- (non protein chemical compound or metallic ion, required for an enzyme activity i.e. they are helper molecules that assist in biochemical transformation)



Zinc Metalloenzymes

→ Carbonic Anhydrase II → Allkaline phosphatase

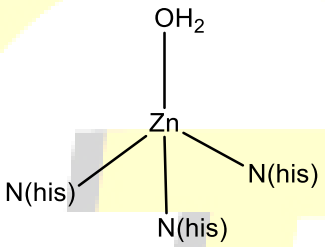
→ Carboxy Peptidase A & G2 → β – lactamase

Zn(II) is not a redox active centre and so cannot take part in electron transfer process. However it is hard metal centre and is ideally suited to co–ordination by N and O donors.

(1) Carbonic Anhydrase:- (CO_2 dehydratase / carbonate dehydratase)

→ Present in RBC (mol. Wt = 30000)

→ This metalloprotein consists of 260 amino acids and contains Zn^{+2} ion bond by 3 histidine residues and the tetrahedral co-ordination sphere is completed by hydroxide ion or water molecule



→ **Function**:- It catalyses the reversible hydration of CO_2

$$CO_2 + H_2O \leftrightharpoons [HCO_3]^- + `H^+$$

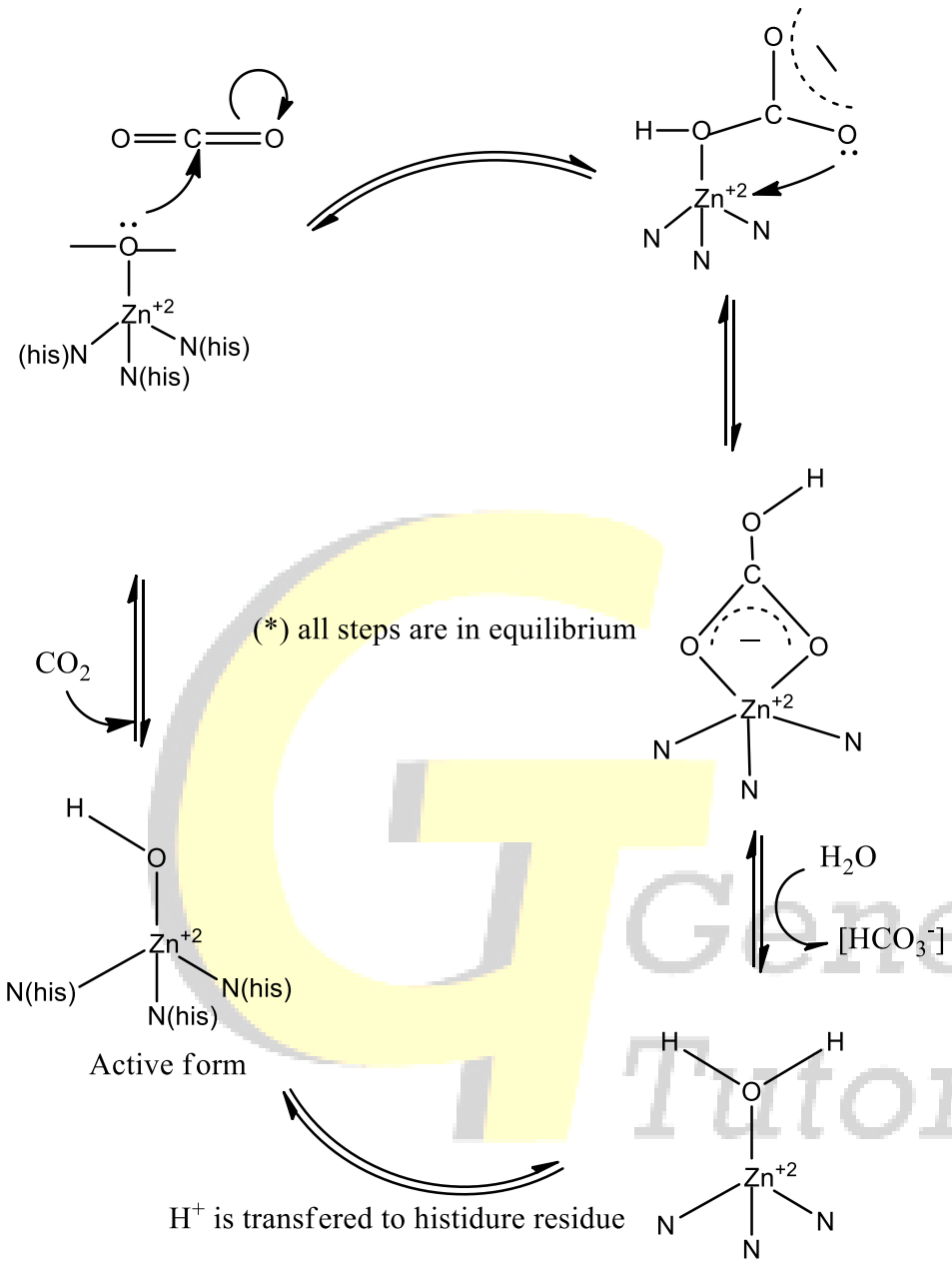
→The process is slow (K = 0.037 s^{-1}) because turnover of CO_2 by biological sustem is very high &such a rate is too slow to sustain aerobic life. Carbonic Anhydrase increases the rate of hydrolysis by a factor of $\approx 10^7$ at physiological pH.

→ Working:- the Zn^{+2} ion is more acidic in carbonic anhydrase then in carboxy peptidase. The presence of neutral or less basic histidine residue instead of glutamate residue contributes to greater acidity of Zn^{+2} ion.

→ In addition, the three histidine's are pulled back making Zn more electronegative & more acidic towards the 4^{th} position. Thus the coordinated water becomes polarized & losses H^+ ion to give Zn OH^-

Catalytic Cycle:-

NOTE:- The nu$^{\ominus}$ OH^- attacks on the carbon atom of CO_2 captured in hydrophobic pocket near Zn^{+2} ion, and a 5-coordinated Zn^{+2} is formed. In which a carbonate form HCO_3^- is replaced by H_2O



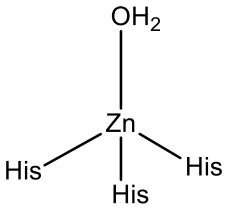
⇒ Protonation of H_2O *ligand* co-ordinated to Zn^{+2} regerate $Zn–OH^-$ which attacks another CO_2 with continuation of catalytic cycle.

CARBOXY PEPIIDASE (it is an exopeptidase)

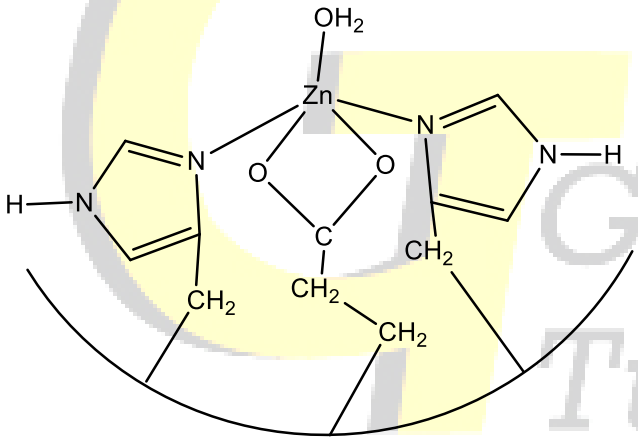
(1) It is a pancreatic metalloenzyme which hydrolysis the peptide bonds in protein during the process of digestion.

(2) The enzyme consists of a single protein chain of 307 amino acid & one Zn^{+2} (molar mass = 34800)

(3) **Structure:-**

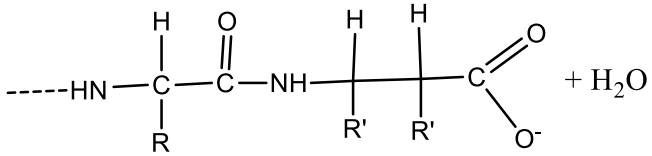


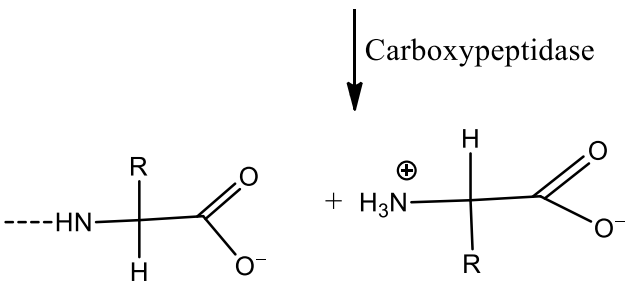
Or



The metal ion is co-ordinated to two N– atom of two histidine residues oxygen atom of a glutamate residue that act as a bidentate ligand and to a water molecule.

(4) The cavity has a hydrophobic pocket close to Zn^{+2} ion that can accommodate organic group of peptide undergoing hydrolysis





(5) It is involved in the degladation of peptide bond

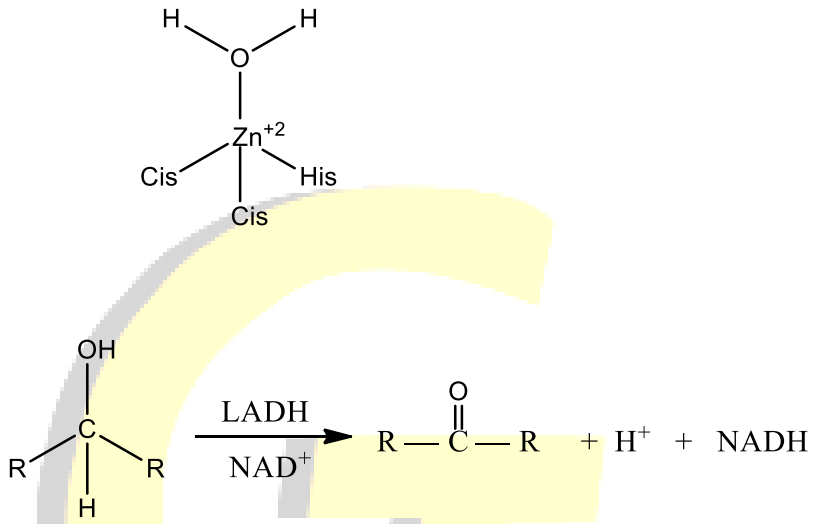
(3) Liver alcohol Dehydrogenase Enzyme:-

(1) These enzyme facilitate the interconversion between alcohols & aldehydes & ketones with the reduction of nicotinamide adenine dinucleotide (NAD^+ to NADH)

(2) In humans and may other animal they serve to break down alcohols that are toxic.

(3) Structure

(4)

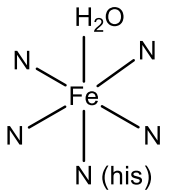


(4) Peroxidase Enzyme

(1) Peroxidase is a heme protein catalyzing the [Oxidation] of substrates by H_2O_2

(2) → Fe(III) → HS

→ Paramagnetic



(3) Peroxidase catalyse reduction of H_2O_2,

$$H_2O_2\,(aq) + 2e^- + 2H^+(aq) \rightarrow 2H_2O(l)$$

Catalase:-

→ It catalyse the disproportionation of H_2O_2 & organic peroxides. They also catalyze the [Oxidation] of substrates by H_2O_2

(5) Superoxide Dismutase:-

(1) In biochemistry dismutation means disproportionation reaction. This enzyme catalyses the dismutation (disproportionation) of superoxide ion into oxygen & H_2O_2)

$$2\,O_2^- + 2\,H^+ \rightarrow H_2O_2 + O_2$$

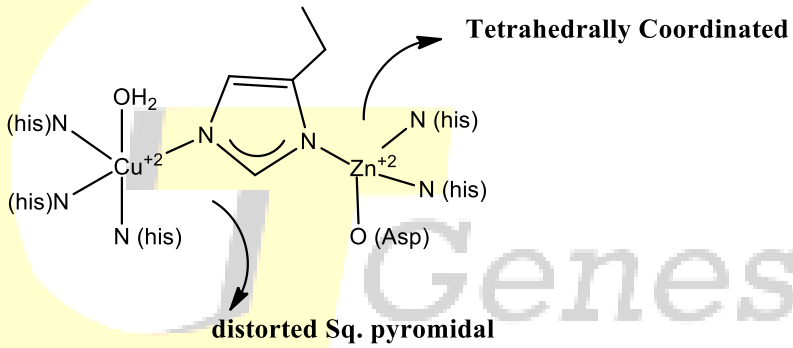
There are 3 types of superoxide dismutase:-

(1) Cu – Zn SOD → called as BOVINE SOD and found in Mitochondria of eukaryotic cells any organism whose cells have nucleus

(2) Mn – SOD

(3) Fe – SOD → are found in bacteria (pyrokaryotes)

NOTE:- Structure of Cu–Zn SoD



Cu^{+2} and Zn^{+2} are co-ordinated to imidazole of (His) residue

→ Here Cu^{+2} is functional unit whereas Zn^{+2} ion is a supportive that holds the bridge imidazole His residue in place & provides structural stability.

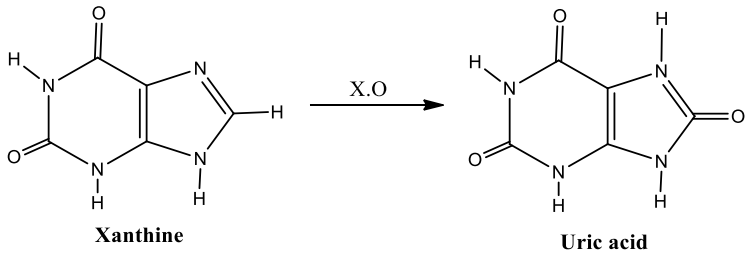
→ Superoxide is produced as a by-product of oxygen metabolism if it not regulated, it cause many types of cell damage

→ H_2O_2 is also damaging and is degraded by other enzyme such as catalase

(6) XANTHINE OXIDASE

(1) Contains two atoms of Mo, $4Fe_2$, $4Fe_2S_2$ and FAD (Flavin adenine dinucleotide) sites

(2) It catalysis the oxidation of xanthine to uric acid.



Xanthine **Uric acid**

(3) electron flow may be represented as:

$$\boxed{\text{Xanthine} \rightarrow \text{Mo} \rightarrow 2Fe_2S_2 \rightarrow \text{FAD} \rightarrow O_2}$$

NOTE:- An excess of uric acid accumulation leads to gout and it can be treated with inhibitor of Xanthine oxidase.

NOTES:-

(7) ALDEHYDE OXIDASE →

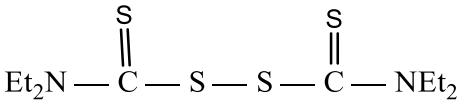
(1) It also contains 2 (Mo | $2Fe_2S_2$ | FAD) units with a mol.wt of 300000.

(2) It convert acetaldehyde to acetic acid via electron flow

Acetaldehyde → MO (VI) → $2Fe_2S_2$ → FAD → O_2

(3) When ethanol is consumed, the initial metabolic product is extremely poisonous acetaldehyde, which is kept in low concentration by the oxidase–catalyzed conversion to harmless acetic acid.

NOTE:- The drug **Antabuse**, used for treating alcoholism , is a Sulphur containing drug (disulfiram)



It prevents acetaldehyde dehydrogenase from converting acetaldehyde to acetic acid leading to a build up of acetaldehyde levels in blood

(8) Alcohol Dehydrogenase:- (ADH) (redox enzyme)

(1) When ethanol is consumed it is oxidized to extremely poisonous acetaldehyde. The oxidation of ethanol to acetaldehyde occurs in liver by alcohol dehydrogenase.

$$CH_3CH_2OH + NAD^+ \xrightarrow{\text{Alcohol dehydrogenase}} CH_3CHO + NADH + H^+$$

(here NAD^+ is reduced by alcohol)

(2) It is a dimer of 2 subunit, it has two Zn atoms per sub unit

NOTE:- Another minor route of oxidation of ethanol to acetaldehyde involves cytochrome P-450 (Which uses molecular oxygen) and NADPH

(9) VITAMIN B-12 and Coenzyme B-12

→ Are natural organometallic compound

→ It is observed that the deficiency of vit-B12 or B12 coenzyme causes pernicious Anemia in humans.

(Is a types of Anemia in which not enough RBC are present due to lack of Vit B-12)

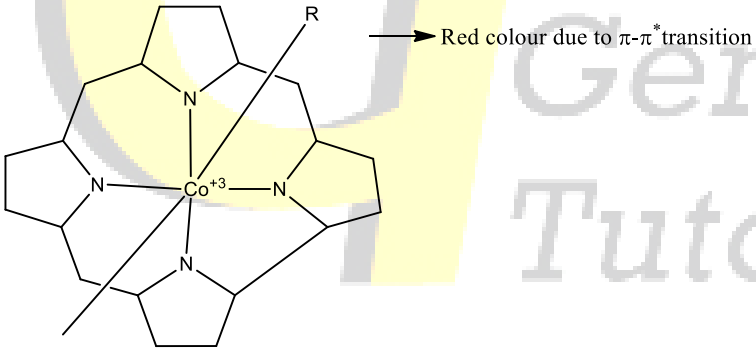
Symptom:- Tired feeling, shortness of breath, pale skin depression

Structure:-

(a) Co(III) ion is coordinated to 4N atoms of a Corrin ring. (It is a modified ring which has one less = CH–bridge b/w two pyrole rings than porphyrin ring. Therefore Corrin ring is less symmetric and less unsaturated than Porphyrin ring.)

(b) The 5th and 6th co-ordination site on the Co are filled by a N atom from the imidazole ring and cyanide ion.

Note:- In vivo the cyanide ion is not present and 6th position is occupied by loosely bonded water molecule.



R = CN $\boxed{\text{Vit B} - 12 \text{ or cyano cobalamin}}$

R = $\frac{5'-\text{deoxy adenosyl cobalamin}}{(\text{coenzyme B 12})}$

Active Site:- Co^{+3} (Oh) → low spin ($t_{2g}^6\, eg^0$)

→ Diamagnetic

→ ESR in active

NOTE:- Vit B_{12} may be reduced by 1 electron or two electron to form Co(II) or Co(I) complexes. Co(I) is strongly Nucleophilic and readily undergoes alkylation via oxidative addition

$$[B_{12}(Co^{I})] + CH_3I \rightarrow [B_{12}Co^{III} - CH_3]^+I^-$$

Other

NOTE:- Vit B(12) Co^{+3}

(Reduction)

$1e^-$	$2e^-$
Co^{+2}	Co^{+1}
↓	↓
Vit $B_{12}r$	Vit $B_{12}s$ blue-green color
↓	↓
Brown	Nu^-
due $\pi - \pi^*$	and it undergoes
transition in corrin ring	alkylation reaction
$t_{2g}^6\ eq^1$	t_{2g}^6
↓	↓
epr active due to the prsence of 1 unpaired e^-	epr active due to two unpaired e^-

* **Coenzyme B-12**

→ Reduction of –CH(OH) group to $–CH_2–$ group

→ Reduction of Ribonucleic acid (RNA) to Deoxy Ribonucleic acid (DNA)

* Iron Storage and Transport

(1) Transferrin:-

(a) Human and other animals absorb iron as Fe(II) from food in their digestive system as it passes from stomach (which is acidic) into blood, it oxidise to Fe(III) in a process catalyzed by Cu metalloenzyme cerruloplasmin.

(500 billion blood cells per day)

(b) Fe (III) then binds with a transferrin protein & transported to bone marrow where it is released from transferrin protein after the reduction of Fe(III) to Fe(II) because Fe(II) binds less effectively to transferrin.

(c) Fe(II) is used to synthesize other iron compound such as Hb, Mb & Cytochrome

(d) When a RBC become aged after an average of 16 weeks the Hb is decomposed & iron is recovered is by transferrin after [O] to Fe(III).

Iron Transport Protein

Transferrin — Si derophores

→ In higher animals in blood stream → aerobic microrganism

NOTE:- These iron-binding protein are responsible for the transport of iron to the side of synthesis of other iron containg compound (Hb, Mb) and insertion via enzymes into porphyrin ring.

→ Transpot Fe (III)

→ mol wt = 80,000

→ Higher animals:- animals of relatively advanced or developed characterstics such as mammals & other vertebrates.

→ Transport of Fe(III)

→ low mol wt = (500 – 1000)

→ Depending upon their molecular structure, they are classified as

ferrichromes ferrioxamines entrobactins

Note:- These molecules are polyclentate with many potential ligating atoms to form chelates. They form extremely stable octahedral complexes with high spin Fe(III)

* Although the complexes are very stable which is extremely important to their biological function, they are labile which allowes the iron to be transported & treansferred within the bacteria.

* Hb, Mb, cytochromes → Fe(II) siderophores, Transferrin → FeIII

Storage of Iron

→ Ferritin is a iron storage protein

→ In humans & other higher animals

→ It is compared of a protein and an iron core.

→ Protein consists of 24 perptide chains with about 175 amino acid and forms a hollow sphere of about 100 Å in diameter. It contains 45000 Fe (III) ions and some hydroxo as well as oxo & phosphate ligands.

→ Ferritin is found in liver & bone marrow

→ Water soluble crystalline substance

→ Contains hydrophilic & hydrophobic channels.

→ Other iron storage protein is Hemosiderin and it is water insoluble

NOTE:- Mosr abundant element in earth crust → Oxygen (46.6%)

(Si) 2nd most abundant (27.7%)

(Al) 3rd most abundant (8.1%)

(Fe) 4th most abundant (5%)

NITROGEN FIXATION

(1) On earth Nitrogen is found in elemental form N_2 but we are unable to utilize it.

Nitrogen fixation is the process that converts atmospheric Nitrogen into NH_3

Two main process are:-

(a) Biological process (By Bacteria) – In vivo

(b) Industrial process (By Haber-Bosch Process) In vitro

In both process N_2 is converted to NH_3 by breaking $N \equiv N$ bond (highest dissociation energy = 945 KJ/mol)

In vitro (Industrial process → HABER's process)

400–500°C

$$N_2 + 3H_2 \xrightarrow[200\ atm]{400-500°C} 2NH_3$$

Fe/Mo is catalyst (this process is very expensive)

In vivo

(1) Blue green algae is a wide variety of bacteria that fix N_2 (natural process) at surrounding temperature only.

(2) These bacterias are free living or from symbiotic association with plants or other organisms

Important bacteria which fix N_2 are:-

→ Clostridium

→ Azotobacter

→ Rhizobium → best known which is found in the root nodules of leguminious plants (beans, peas & soya)

The ammonia so formed is used in amino acids & protein synthesis by plants.

(3) Types of Nitrogenase Enzyme:-

(a) Vanadium Nitrogenase

(b) Iron Nitrogenase

(c) Molybedenum nitrogenase

(4) Nirogenases consists of 2 metalloproteins

⟹ Fe protein

⟹ MFe (M = Mo, V, Fe) protein as Cofactor (non-Protein chemical compound or metallic ion i.e. required for an enzyme's activity)

Note:- Vanadium Nitrogenase (containing Fe protein, 4Fe-4S-ferridoxin & FeV cofactor)

→ It has less activity than Mo nitrogenase

→ The presence of Mo is necessary component of most nitrogenase

MOLYDENUM NITROGENASE:-

(a) consist of 2 different protein

Fe | MoFe Protein

→ Molar mass = 60,000

→ Called P-cluster

→ has 2 Fe_4S_4 units connected by 2 cys ligand bridge and these 2 units are also linked by S–S bond

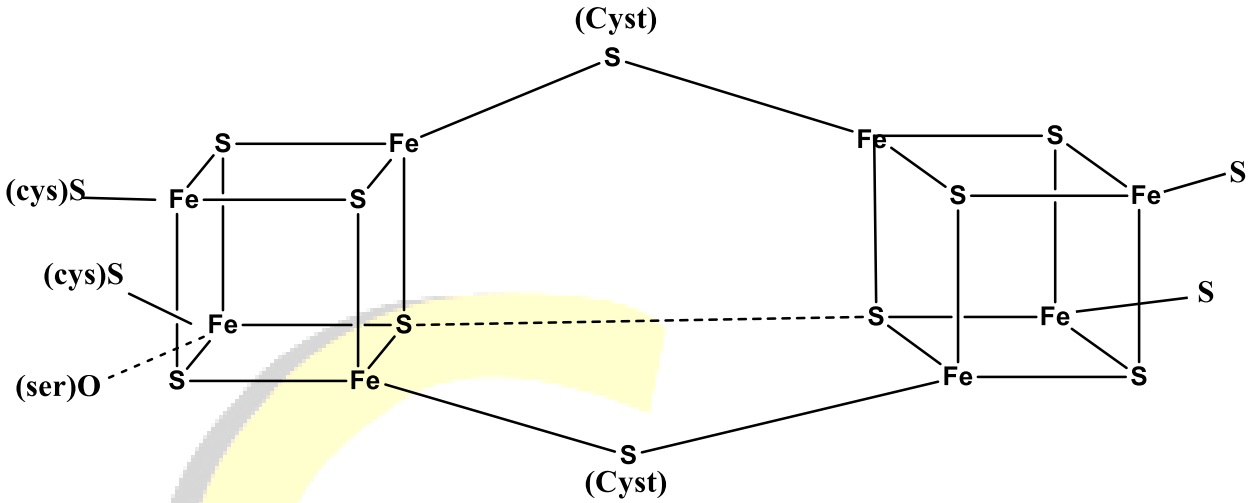
NOTE:- In one Fe_4S_4 unit there is a serine ligand attaché to one iron atom in add to cysteine ligand

→ Molar mass = 220000 to 24000

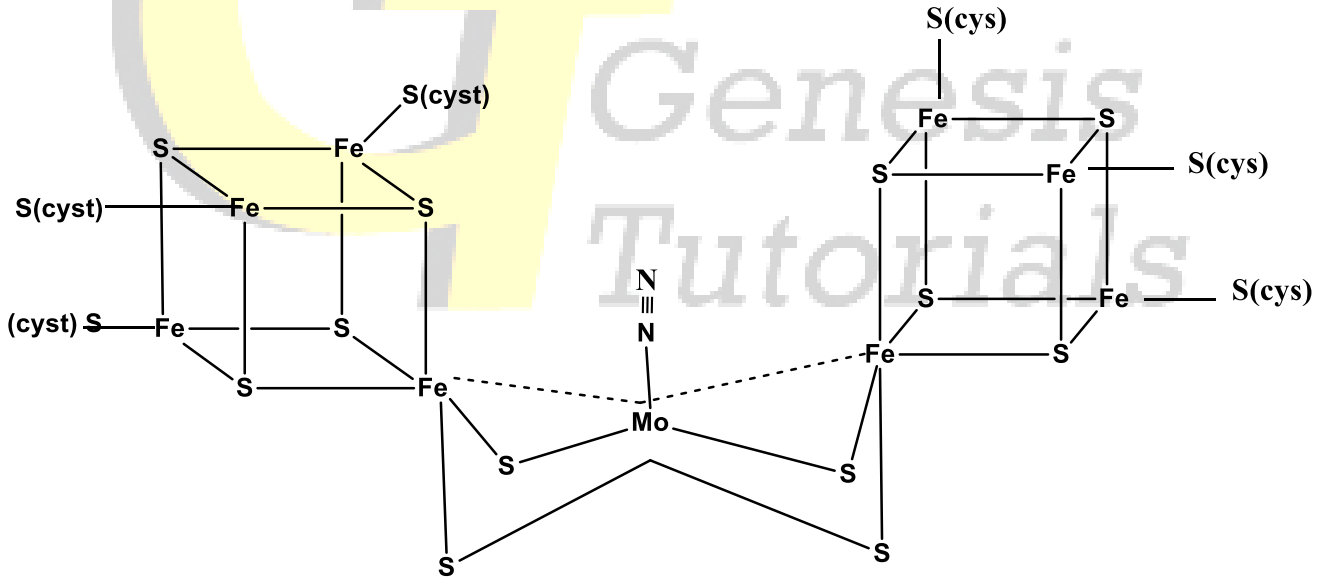
→ Mo atom is linked to two Fe_4S_4 unit by 2 sulphide group & Mo-Fe bond

→ Reduction of N_2 occurs at Mo site of enzyme

(a) Structure of P–cluster in Nitrogenase:-



(b) Structure of active site of Mo-Fe protein Nitrigenase:-



The enzyme nitrogenase in various bacteria catalyses the activated of reduction of N_2 to NH_3 according to equation

$$N_2 + 16\ Mg\ ATP + 8H^+ + 8e^- \rightarrow 2NH_3 + 16\ Mg\ ADP + 16\ Pi + H_2$$

Pi is inorganic phosphorous

NOTE:- (a) The e^- required in reduction of N_2 are transferred by reduced form of ferredoxins & feavidoxins (e^- transfer proteins)

(b) The e^- are first transferred to smaller protein (P–Cluster). The reduced fe protein transfer its reducing e^- to Mo Fe protein & then to N_2.

e^- flow

$$\boxed{\text{Fe duster} \rightarrow \text{Mo Fe protein} \rightarrow N_2}$$

$$Mo\text{–}N \equiv N \xrightarrow[2H^+]{2e^-} Mo - N - NH_2 \xrightarrow[2H^+]{2e^-}$$

The energy for this process is provided

by hydrolysis of ATP to ADP + Pi

$$\boxed{\begin{array}{c} Mo \text{—} NH \text{—} NH_3 \\ \downarrow \ 2e,\ 2H^+ \\ Mo + 2NH_3 \end{array}}$$

PHOTOSTNTHESIS

(a) It is a redox process by which green plants convert water & atmospheric CO_2 into carbohydrate & release O_2 by absorbing sunlight

$$6CO_2 + 6H_2O \xrightarrow{h\nu} C_6H_{12}O_6 + 6O_2$$

(b) It involves reduction of CO_2 & oxidation of H_2O to O_2.

2 step reaction:

$\rightarrow 2H_2O \rightarrow 4H^+ + O_2$

$\rightarrow 6CO_2 + 12H^+ \rightarrow C_6H_{12}O_6 + 3O_2$

(c) Photosynthesis occurs in chloroplast of the cells of green plants.

* In this sunlight energy is converted to chemical energy in form of ATP & NADPH.

* Initiateed by capture of light energy in photoreceptors of chlorophycl found in chloroplaste of green leaf. (overall process is enclothermic)

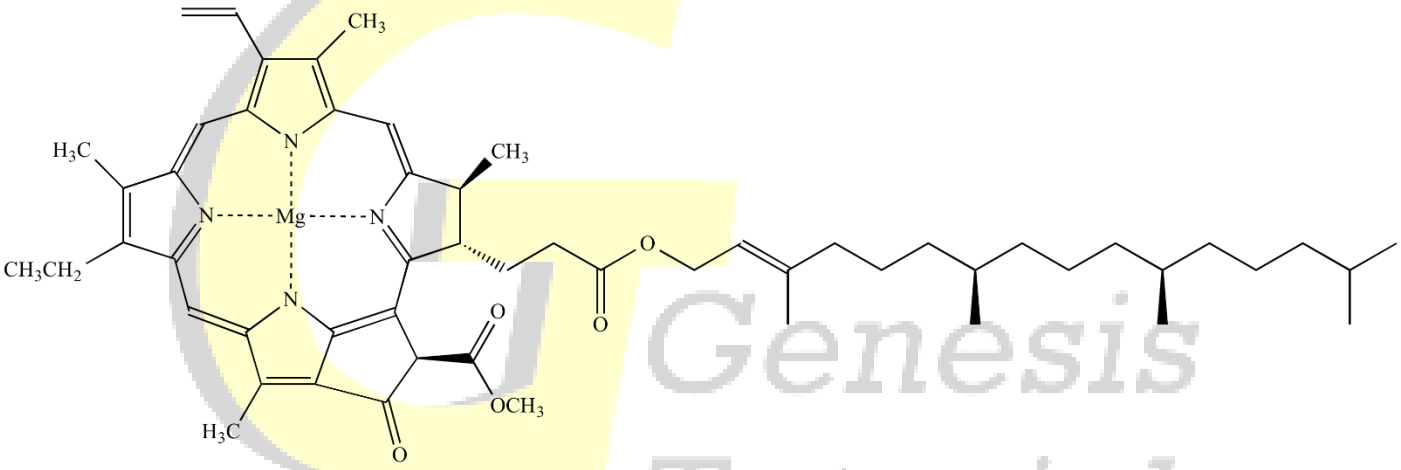
CHLOROPHYLL

(a) are complexes of mg^{+2} with porphyrin ring in which one pyrrole ring has been reduced called CHLORIN

Porphyrin ring with one = bond reduced

Hence. Chlorophyll are the complexes of mg^{+2} with substituted chlorine and a fused pentanone ring is also present

Structure:-



R = CH_3 (chlorophyll a)
R = CHO (cholorophyll b)

(b) In green plants there are two types of chlorophylls (a + b) difering in side group

(c) Chlorophylls is present in the chloroplaste of plants leaf and absorb light in the red region of visible spectrum (near 680–700 nm) therefore appears green

(d) Two bonds are abserved in theabsorption spectrum of chlorophyll a

* strong absorption band in red region (Q bond)

* Comparitively weak band in blue or near UV (soret band)

Reason:- $\pi - \pi^*$ Transition in porphyrin ring

PHOTOSYNTHESIS → light reaction
PHOTOSYNTHESIS → Dark reaction

$\Longrightarrow$ synthesis of food in presence of light is called photosynthesis

$\Longrightarrow$ Green plants has ability to capture light and convert into chemical energy

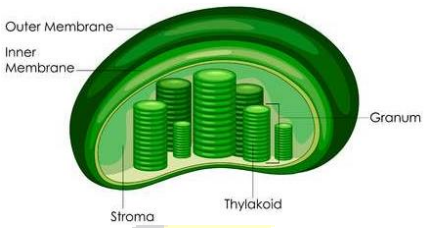
$\Longrightarrow$ Food (glucose) $\rightarrow C_6H_{12}O_6$

CO_2 (atmosphere) $\quad H_2O$

$\Longrightarrow$ Photosynthesis source of glucose as well as O_2 $\rightarrow$ byproduct

$\Longrightarrow$ takes place in Chloroplast

Green colour [Plastical] → Stroge of food
→ Photosyntheis



(P) Light reaction $\rightarrow$ always take place in presence of light

Dark reaction $\rightarrow$ it can also occur in absence of light

Light reaction to absorb light

Production of assimilatory power

$\downarrow$ $\Longrightarrow$ This process also called Photophosphorylation

[ATP + NADPH + H^+]

Supply energy — Reducing agent

$\Longrightarrow$ Dark reaction reducing CO_2 (reduction process)

Prime object is to make food

$CO_2 \rightarrow C_6H_{12}O_6$

In maximum O.S — R.A NAD PH + H^+

ATP $\rightarrow$ energy source here

* Here no role of light is there because we have assimilatory power here, no evolution of O_2

PHOTOSYSTEM II

⟹ There are 2 photochemical reaction centres

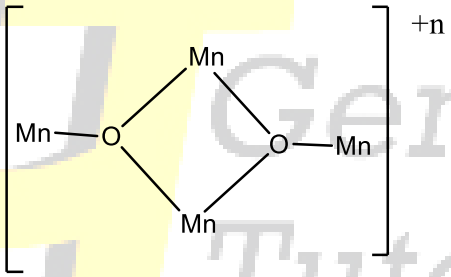
Photosystem – I (PS-I) And

Photosystem – II (PS–II)

⟹ Initiating P–II is called P_{680} (chlorophyll a2)

⟹ The chlorophyll a (chl-a_2) site of PSCII) absorbs quanta of higher energy from sunlight and it becomes excited and it has s strong tendency to transfer its excited e^- it is therefore, a strong reducing agent and it reduces the pheophtin (a chl-like pigment lacking mg^{+2}) of the PS-II e^- transfer chain

⟹ The loss of an e^- causes PS–II to becomes positively charge and it attracts e^- from magnesia protein (Mn_4 cluster)



⟹ The mangenese protein contains 3 Mn(II) and one Mn(III). The Oxidised manganese protein in turn attracts e^- from water & catalyses the oxidation of H_2O to O_2

⟹ The e^- from pheophytin is then transferred through various redox system like platoquinone, cytocromes, iron-sulphur protein & plastocyanin. At this point it leaves PS-II & moves on to PS-I

PS-I

(1) PS-I involves chlorophyll – a abd b when PS-I absorb energy from chlorophycles, the anterma pigmants (other pigments present in leaves to hearvest somelight & transfer energy to reaction centre).

The e^- received from PS II, is therefore further excited & becomes a reducing agent

(2) It parses on its e^- to short chain of e^- carriers which reduces ferredoxin & the reduced form of ferredoxin reduces $NADP^+$ by following reaction catalyzed by FAD enzyme called ferredoxin NADP reductase

$$2(\text{Ferridoxin})\ red + NADP^+ + 2H^+$$

$$\downarrow$$

$$2\ (\text{ferredoxin})ox + NADPH + H^+$$

Net reaction of PS–II & PS–I is:-

$$2H_2O + 2NADP^+ + 8\ \text{Photon} \rightarrow 2NADPH^+ + 2H^+ + O_2$$

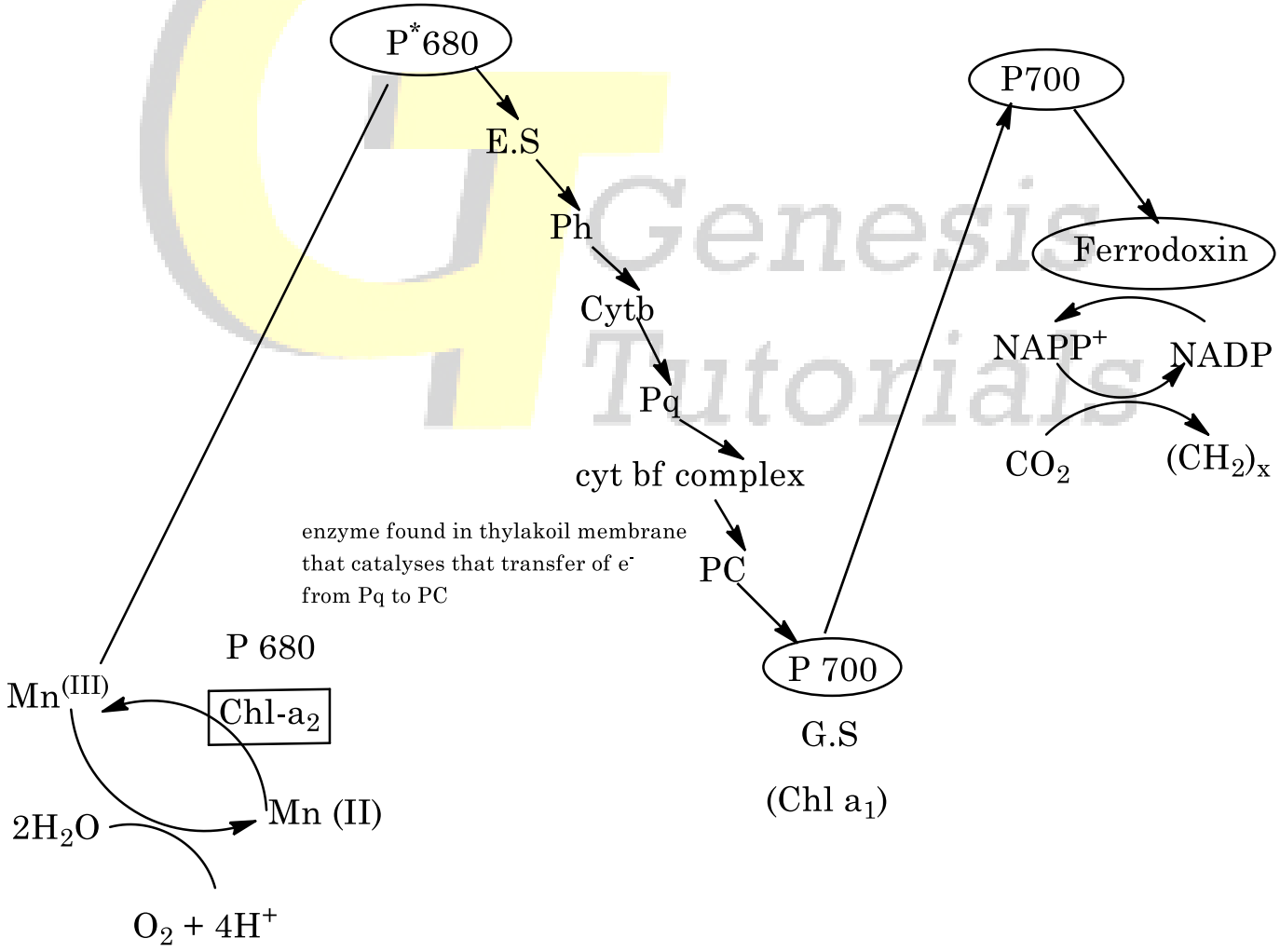
NADPH & ATP involves in the conversion of CO_2 into glucose by following net reaction:-

$$6CO_2 + 18ATP + 12\ NADPH + 12\ H^+ + 12H_2O$$

$$\downarrow$$

$$C_6H_{12}O_6 + 18\ ADP + 12\ 2NADP^+ + 18\ pi$$

Ph → pheophytin



NOTE:- NADPH → Nicotinamide adenine dinucleotide phosphate is a cofactor used in anabolic reactions (such as nucleic acid synthesis) which requires NABPM as a reducing agent

* CYT bf complex (plastoquinol plastocyanin reductase) is an enzyme found in thylakoid membrane on chloroplasts of plants that catalysis the transfer of e^- from Pq to Pc.

Elements

Essential	Trace	Ultratrace
Required in large quantities (Macronutrient)	Requried in small amount (Micronutrient)	
→ Na, K, Mg, Ca, P, S, Cl Along with 4 most abundant element (C, H, N, O)	→ First row TM (except Sc & Ti) Mo % W (from 1st & 2nd row T.N)	
	⟹ non metals (B, Si, Se, F & I)	

* **Ultratrace elements:-** Ni, Cd, Pd & As, concentration level (< 1 ppm) these elements are toxic at concentration above.

(1) Metal Complexes in Medicine

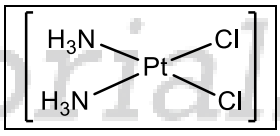
* Anti Cancer Drug:-

⟹ Discovered by B.Rosenberg & Cowerker (1969)

⟹ Sq. planar complex

Cis-diamminedichloro platinum (II)

⟹ ($Pt(NH_3)_2Q_2$)

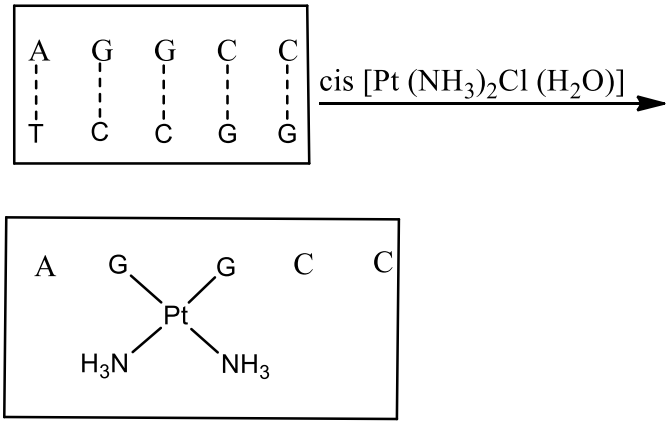


⟹ This compound is used as chemotherapeutic agent to inhibit rapid division of tumor cells.

⟹Proton NMR studies have suggested that Pt binds to N–7 position of guanine bases.

The most important interaction is intrastand linking of two agjacent guanine bases on DNA chain by Pt atom. The binding of cisplatin to DNA would seriously interfere with the ability of guanine bases to undergo Watson-(risk paining thus a portion of DNA chain) (self-complementary aligomer) reacts with cis–isomer and Watson crick pairing is disrupted

$Cis\text{–}Pt(NH_3)_2\,Cl_2 + H_2O \leftrightharpoons cis\,[Pt(NH_3)_2Cl(H_2O)]^+ + Cl^-$

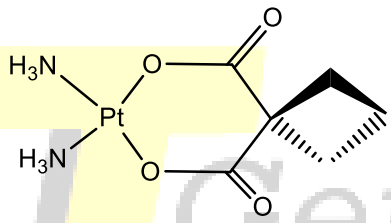


→ Binding of Pt distorts the local DNA structure and inhibits the cell division in cancer cells

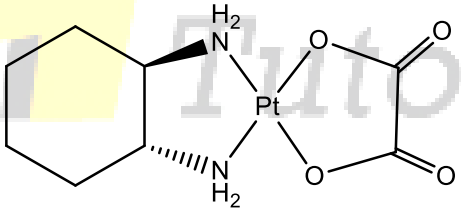
→ Cisplatin is most effective against testicular cancer (cancer of tests of males)

Cisplatin has negative side effects in kidney and neurotoxicity (Kidney → nephrotoxicity)

In order to avoid these side effects, alternative Pt–compound 'carboplatin' is used in which the Cis–Chloride ligands are replaced by O–Chelate, cyclobutanedicarboxylate



Structure of Carboplatin



Oxaliplatin

*** Wilson Disease:-**

⟹ It is a genetic disease caused due to overload of Cu on body

⟹ Patients suffering from Wilson disease have low levels of Cu storage protein Ceruloplasmin

⟹ Wilson disease is responsible for liver disease neurological damage and brown & green rings in cornea of eyes.

⟹ Many chelating ligands can be used to remove the excess copper but best is **D–penicillamine**

⟹ This chelating ligand forms a complex with Cu ions

⟹ The sulphydryl group of D–pencillamine effects removal of copper as Cu(I) complex

Anti-Arthritis

⟹ Complexes of Au(I) has been successfully used for the treatment of arthritic disorder in human (Rheumatic arthritis)

⟹ These complexes included

$Na_3[Au[S_2O_3)_2]$ Called Sanochrysin (Sodium auriothio sulphate)

⟹ Sodium salt of thiomalate called Hyochrysin

⟹ Auranofin

Hypercalcemia

⟹ Hypercalcemia is a disease which causes the rapid loss of calcium from bones of cancer patients

⟹ Gallium nitrate, $Ga(No_3)_3$ has been found to be most effective for treatment of hypercalcemia

Magnetic Resonance Imaging (MRI)

⟹ MRI uses a powerful magnetic field, radio waves & computer to produce detailed picture of inside our body

⟹ It may be used to help diagnose or monitor treatment for a variety of conditions within chest, abdomen etc.

⟹ The useful metal ions for magnetic resonance imaging in humans are Gd(III), Fe(III), Mn(II) ions

⟹ MRI is a medical imaging technique used in radiology to form picture of auatomy and physiological process of body

⟹ MRI scanner uses strong magnetic fields, electric field gradient & radio waves to generate images of organ in body

Siderosis Disease:-

⟹ An excessive intake of iron causes various problems called siderosis.

⟹ Chelation therapy is used to treat excess of iron. The patients who suffer from deposits of iron to failure of these organs

Chelating ligand used:-

Desferrioxamine—B - It is a polypeptide having strong affinity for Fe(III) but not for other.

NOTE:-

* Calmodulin:- (CaM) → abbreviation for calcium modulates protein is a multifunctional intermediate calcium binding messenger protein found in eukaryotic cells

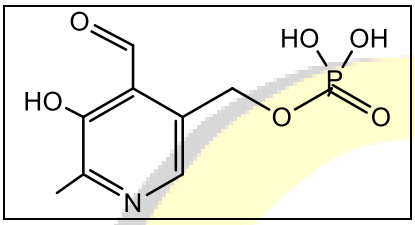
* Sulphite oxidase:- is an enzyme in mitochondria of all eukaryotes. It oxidise sulfite to sulohate & via cyt C transfers the e^- produced to e^- transport chain and allows generation of ATP

→ active site Mo(VI) center

↓

Contains molybdopteric cofactor

*** Vitamin B–6** → is a part of vit B group of essential nutrients



Its active form pyridoxal S'– phosphate works as coenzyme in many enzyme in amino acid, glucose & lipid metabolism

*** Biotin:-** water soluble B vitamin also called vitamin B_7

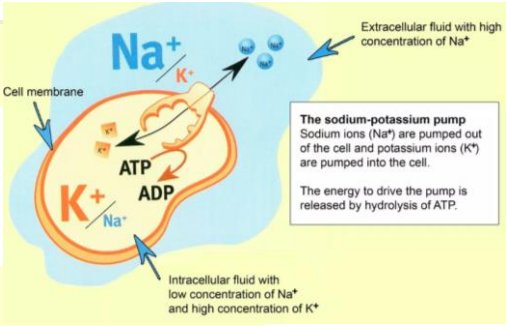
$C_{10}H_{16}N_2O_3S$

Folic acid:- $C_{19}H_{19}N_7O_6$

*** Order of conjugation:-**

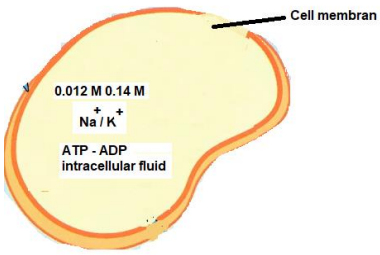
Porphyrin ring > Chlorin > Chorin

Sodium Potassium Pump:-



(1) The concentration gradient across cell membrane produces an electrical potential difference which is essential for functioning of nerve & muscle cells

Diffusion → movement of molecules from a region of high concentration region of low concentration



⟹ Lipid soluble substance diffuse through the cell membrane without the need of carrier & this process is called passive diffusion

⟹ But the movement of water soluble solutes & ions required specific transmembrane proteins which facilitates this transfer & this process is known as facilitated difference.

⟹ In this there is a movement of solute against the concentration gradient & this required an input of energy which is provided by the hydrolysis of ATP.

⟹ This facilitated diffusion required energy input & it is known as active membrane transport.

⟹ The eq if active transport is Na^+/K^+ pump present in animal cell.

⟹ In intracellular fluid of cells there is low concentration of Na^+ & high concentration of K^+ to extracellular fluid which contains high concentration of Na^+ & low concentration of K^+.

⟹ The carrier by the hydrolysis of ATP is sufficient to pump 3 Na^+ out of the cell & 2 K^+ and 1 H^+ion into the cell

⟹ Export of the Na^+ from the cell provides the driving force for many secondary active transport i.e import glucose, amino acids & other nutrient into the cell by the use of Na gradient

⟹ K^+ is required in the cell for glucose metabolism, protein synthesis & activation of may enzymes

⟹ The mechanism for ion transport involves natural polyether's which are called Ionohores (ions bearing) for eg Nonactin & valiomycin, thus have o donor atoms.

⟹ This ioniophores are maerecydic ligands & are lipid solution & specific for particular ions

⟹ Valinomycin is a natural lipid soluble molecule that kinds K^+ & facilitates their treanfer across the cell membrane.

⟹ Both gave selectivity for K^+ over Na^+ ion.

NOTES:- Ionomycin selective for Ca^{+2}

NOTES:- Alkaline phosphate (AP) →

⟹ Contains more than 1 Zn atom

⟹ It catalyses the hydrolysis of phosphate monoesters

⟹ $R–O–PO_3^{-2} + H_2O \rightarrow ROH + HOPO_3^{-2}$

⟹ATP catalyse the breakdown of organic phosphate including ATP, to provide phosphate required for growth of bones.

⟹ As it name implies, its optimum pH is the mild alkali region.

⟹ Active site contains two Zn atom located only about 0.4 nm apart (here mg is also present), here, the phosphate ion bridges the two Zn atoms.

*** β –lactamase**

Are enzyme produced by bacteria that provide multi resistance to β –lactam antibiotics such as penicillin

*** peroxidase:-**

⟹ Enzymes that catalyse the oxidation of a particular substance by H_2O_2

NOTE:- H_2O_2 → simplest proxide $(\overline{O} - \overline{O})$

⟹ Used as an oxisizer, bleaching agent, antiseptic

⟹ H_2O_2 is formed in human & animal as a short lived product

Penicilline:-

⟹ is a group of antibiotics. Those antibioties here among the first medications to e effective against many bacterial inspections

⟹ All pencillins are β- lactam antibiotic.

Chelation Therapy

(1) Fe overloading → caused by the faults in the regulation of Fe levels by ferritin or transferrin production and these are treated by chelation therapy

⟹ **Desferrioxamine** (desferral 55) is a ligand that is similar to siderophores and it is a very successful agent for ion overloaded.

(2) Chelation therapy is also used for the individuals who have been contaminated with Pu due to the exposure to nuclear weapons

- In its common 0.5, Pu(IV) & Pu(III) have similar charge densities to Fe(III) & Fe(II). So siderophores like chelating agents are used (3,4,3-LIMACC) → it contains 4 catechol groups.

Imaging Agents:-

*** Radio Pharmaceuticals:-** are used in the field of nuclear medicine as tracer in the diagnosis & treatment of many diseases.

⟹ ^{123}I → used in thyroid imaging

⟹ ^{99}Tc → Tc chnetinum is an artificial element prepared by nuclear reaction. Its metastable form ^{99}m Tc is a ribonucleotide useful in medical diagnostic useful in medical diagnostic technique SPECET (single photon emission computed tomography) as a myocardial imaging agent

^{99m} Tc → candiac imaging agent (allows gama rays to take images of heart)

* Oxygen atom transfer by Mo enzymes

(1) MO is used to catalyse o transfer in which o atom is provided by a water molecule

(2) Mo & W are only heavier element known so for to have specific function on Biology

(3) Mo enzymes catalyse the [O] and [R] of small molecules particularly inorganic species.

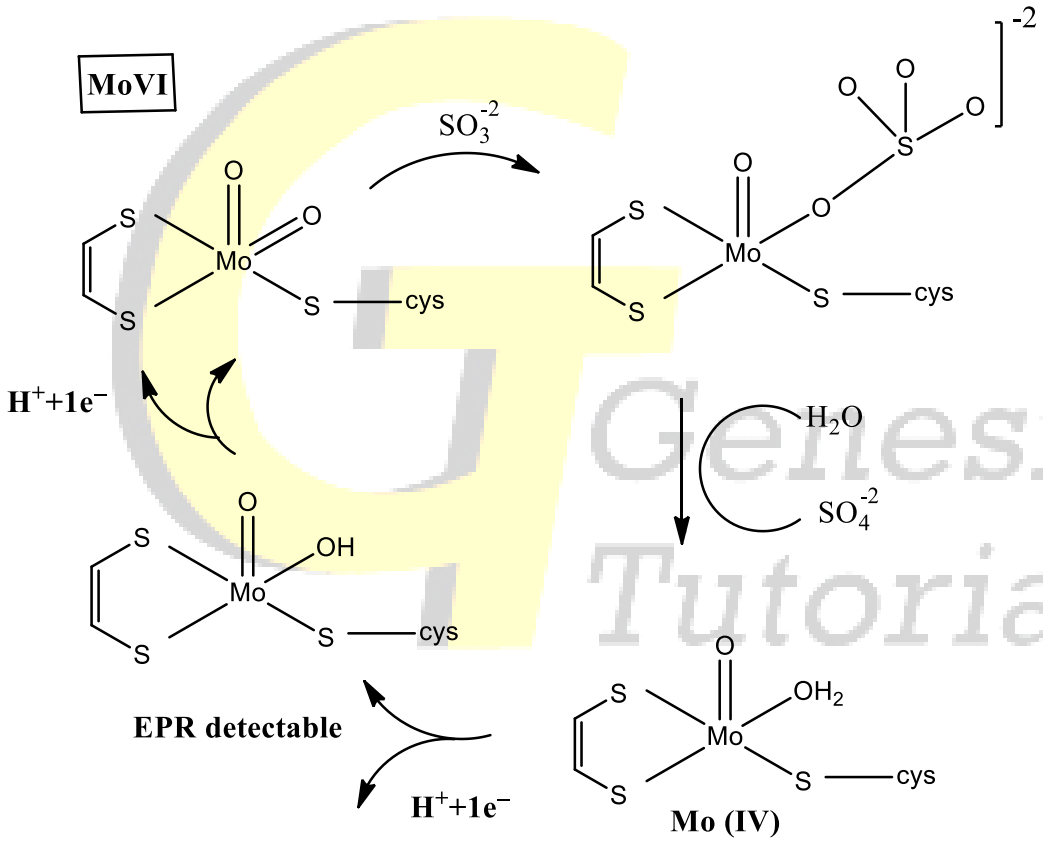
Reactions included oxidation of sulfite, assenite, xanthine, aldehydes & Co and reduction of nitrate and dimethyl sulfoxide (DMSO) .

(4) MO is suited for its rolle because it provides 3 stable oxidation states Mo(IV), Mo(V), Mo(VI)

⇒ **Sulphite oxidase** → Mo containing enzyme used for the oxidation of sulfite to sulphate

(●) The S atom of sulfite ion attach an e⁻ defined O atom Co-ordinated to Mo(VI), leading to Mo–O bond cleavage and formation of Mo (IV) and dissociation of SO_4^{-2}

Reoxidation back to Mo(VI) during which a transferable during which a transferable O atom is regained.



⇒ This kind of oxygenation reaction can be distinguished from that of Fe and Cu enzymes because with Mo enzymes the oxides group i.e. is transferred is not derived from molecular 0_2 but from water

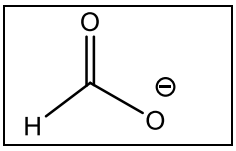
The Mo (VI) = O unit can transfer an O atom either directly or indirectly to reducing substrates such as SO_3^{-2} but cannot oxygenate C–H bonds & Mo(IV) is able to extract O atom from nitrate.

NOTE:-

Cr
MO
W

Format dehydrogenases → contains tungsten (IV) which catalyses the reduction of CO_2 to format

Are reducing agents



Bio alkylation

(1) Methyl mercury (CH_3Hg^+) is a form of mercury that is most easily bio accumulated in organism

(2) Mercury bio methylation is the transformation of divalent inorganic mercury Hg(II) to CH_3Hg^+ and it is carried out bt sulphate reducing bacteria that live in anoxic environment (low dissolved oxygen)

(3) several forms of mercury occur in environment

***** Elemental mercury (Hg^o) is a shiny silver-white, colorless commonly used in thermometers

(4) Methyl mercury cation ($MeHg^+$) is a neurotoxin that can adversely affect the development of the brain and nervous system, especially in children

The symptoms include:- Vision, speech and hearing impairment memory loss, lack of Co-ordination of movement disturbance in sensation wood swing & skin rashes

* Bio alkylation is a process in which direct linkage of an alkyl group to metals to form the metal–alkyl bond occurs in living organisms. The methyl group is the most common alkyl group that can be transferred & this process is called Bio methylation.

(5) Initially the large amount of mercury found in fish in matamata bay as thought to be due to inorganic mercury released as inorganic mercury releases as industrial waste to **bay** (castal body of water that directly connects to a larger main body of water (Ocean lake)). Later it was shown that mercury was present almost entirely as methyl mercury ($MeHg^+$)

The question is that now does inorganic mercury gets converted to methyl mercury & get accumulated in fish??

ANS:- Bacteria converts the inorganic mercury into methyl mercury through bio methylation

Fish absorb this methyl mercury through their gills and coking also does not remove appreciable amounts of the toxin. The FDA has recommended a level of 1ppve as the threshold for safe human consumption.

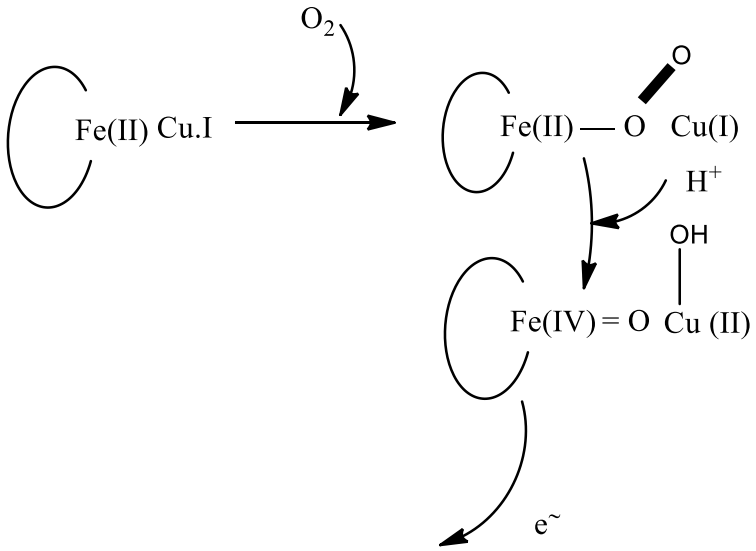
Oxidase

(1) are enzymes that catalyses the reduction of O_2 to water or H_2O_2 without incorporation of o atoms into the oxidizable substrates and they include cyt. C oxidase.

(2) $O_2(g) + 4e^- + 8H^+ \rightarrow 2H_2O(l) + 4H^+$ i.e. $4H^+$ are consumed chemically while $4H^+$ are pumped across the membrane against a concentration gradient such enzymes are called electro genic ion pump (or protonpump)

(3) In eukaryotes, cyt oxidase is located in the inner membrane of mitrochondria and has many subunits

(4) Here, the active site is Fe(II)-Cu(I) & O_2 binds to give an intermediate that resembles oxy Mb, unlike oxy Mb, this intermediate takes up other e^- i.e. immediately available producing a proxy species



* NOTE:- Acid Phosphatases contain a dinuclear metal site containing Fe(III)

Catalyse the hydrolysis of phosphate esters and they work under mild acidic conditions

The pink or purple colours of acid phosphatases are due to tyrosinate → Fe(III) charge transfer transition at 510–550nm

The active site contain two Fe atoms linked by ligands similar to haemerthyrin.

They are inactive in the oxidized {Fe(III) Fe(III)} state in which they are aften isolated.

In active state, one Fe is reduced to Fe(II)

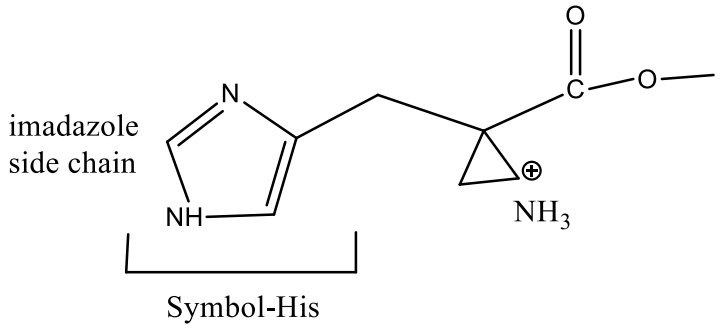
Acid phosphatases also occur in plants & in these enzymes the reducible Fe is replaced by Zn or Mn

NOTE:- Fe Zn centre is also found in an important enzyme called calcineurin which catalysis the phosphorylation of serine of threonine residue on certain protein surfaces.

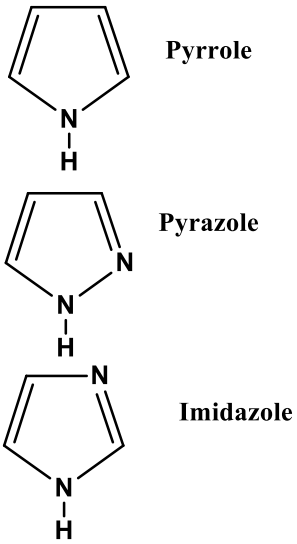
Calcineurin is activated by Ca^{+2} binding through calmodulin

Ca binding messenger protein found in eurkaryotic cells.

Histidine (α-amino acid)



Symbol-His



NOTE:-

(1) Haem → Porphyrin – Fe

(2) Cyt P_{450} → Haem – Enzymes

(3) Chlorophyll → Porphyin coordinated to Mg^{+2}

(4) Corroles (vit B-12) → One carbon shorter analogue

(5) Corpins (cofactor F_{430}) → Ni → Actire site in methyl coenzyme M-reductare

⇓ ⇓

Macroylind ligand required in methane producing bacteria

⇓ ⇓

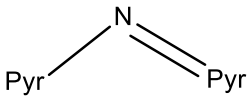
Highly reduced porphyrin last step for production of <u>methane</u>

⇓

Four pyrrole unit are connected (in <u>plane</u>)

(6) Phthalocyanine — Nitrogen

Substituted Porphyrin



(methyle connector replaced)