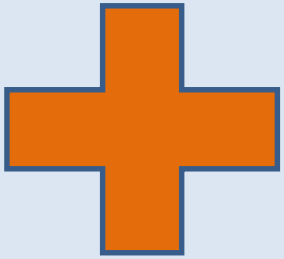
Role of hemoglobin in acid base balance

By

Dr. Walaa Bayoumie El Gazzar

Hemoglobin

Heme  Globin

Globin

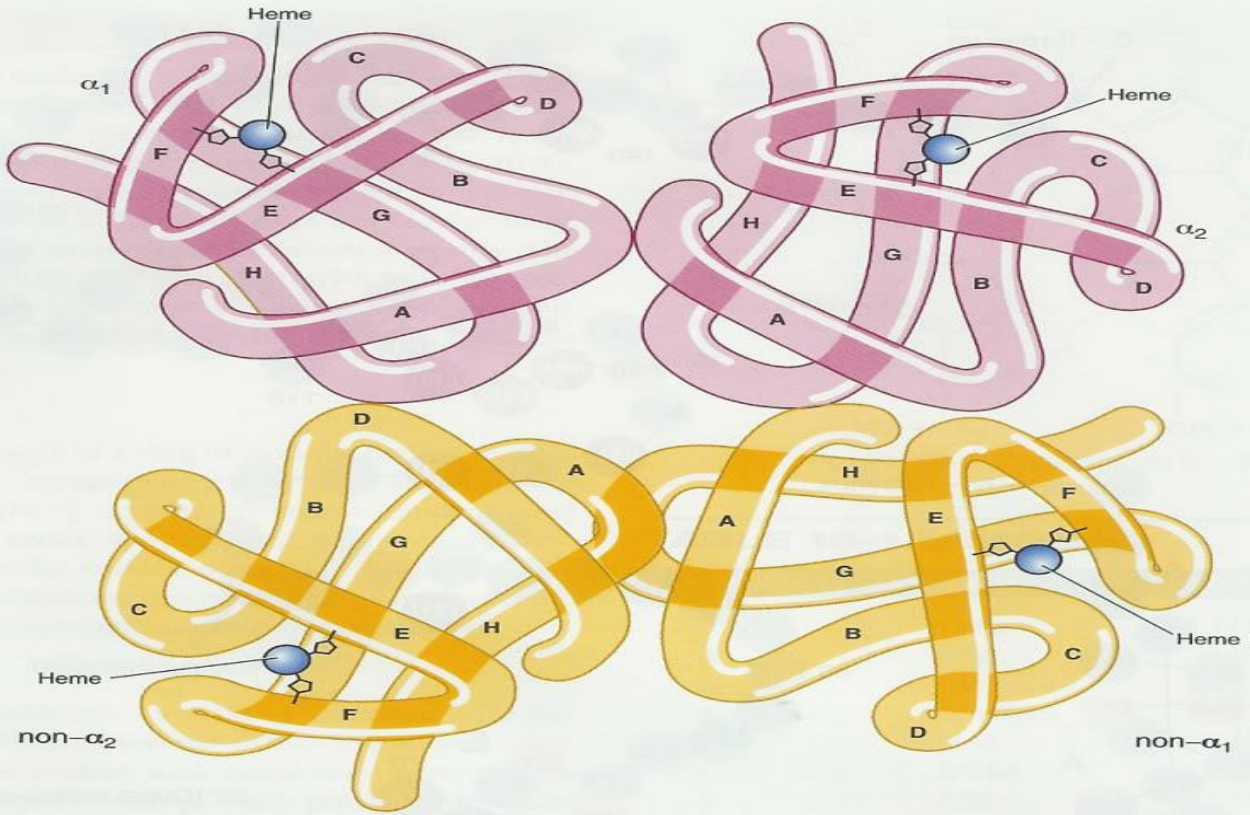


Figure 10-4 Complete Hb molecule. Heme is suspended between the E and F helices of the polypeptide chain. Pink represents α 1 (*left*) and α 2 (*right*); yellow represents non-α 2 (*left*) and non-α 1 (*right*).

Rodak, Hematology, third edtion

Globin

- **Tetramer : 4 polypeptide chains.**
- **Each polypeptide chain is formed of 7 or 8 helices which are termed A-B-C-D…….**
- **There are 4 types of the polypeptide chains that may enter in the formation of Hb (α-β-γ-δ).**

- α –chain → 141 amino acids

(α chain gene is on chromosome 16)

- β-γ-δ chains → 146 amino acids

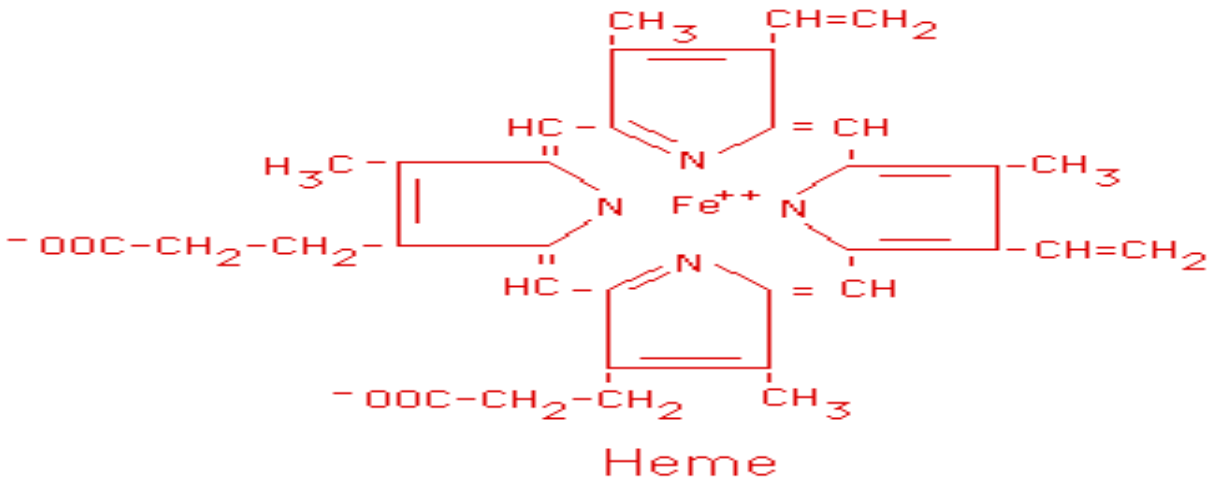
(β-γ-δ chain genes are on chromosome 11)

- **Hb is composed of 2 α and 2 either β , γ or δ**

- **Hemoglobin A, the major hemoglobin in adults, is composed of 2 α chains and 2 β chains.**

  (two identical dimers, (αβ)1 and (αβ)2)

Heme



Heme

- The iron atom of heme occupies the central position of the porphyrin ring.

- In Hb iron is in the ferrous state (Fe^{++})
- Ferrous iron (Fe^{++}) has 6 valencies.
- Iron carries oxygen.

Attachment of heme with globin chain

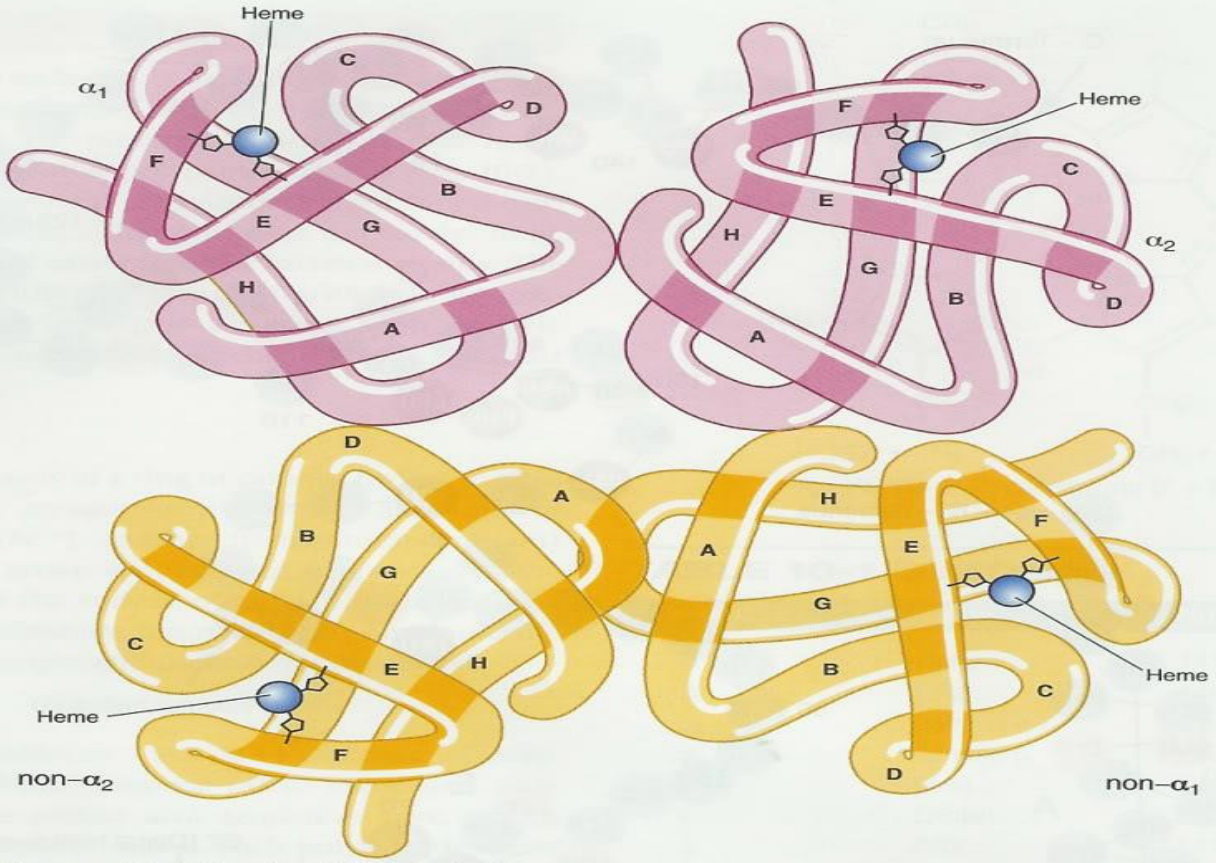
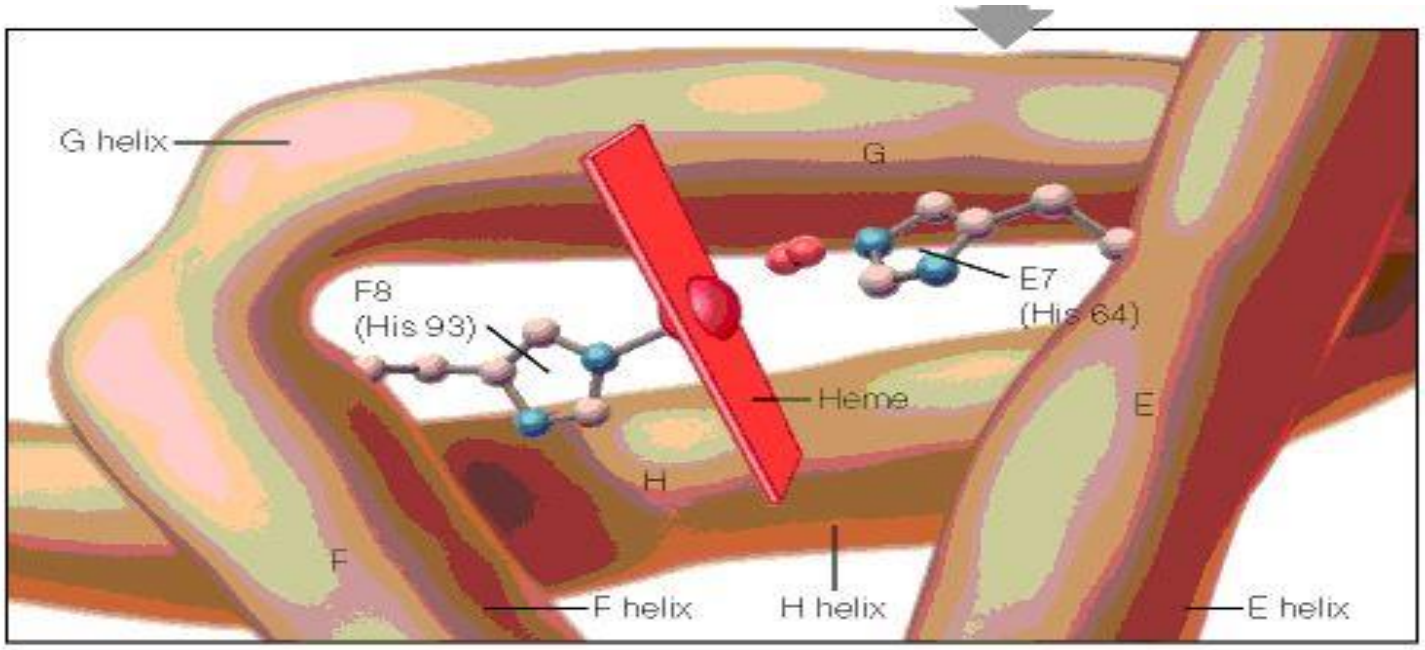


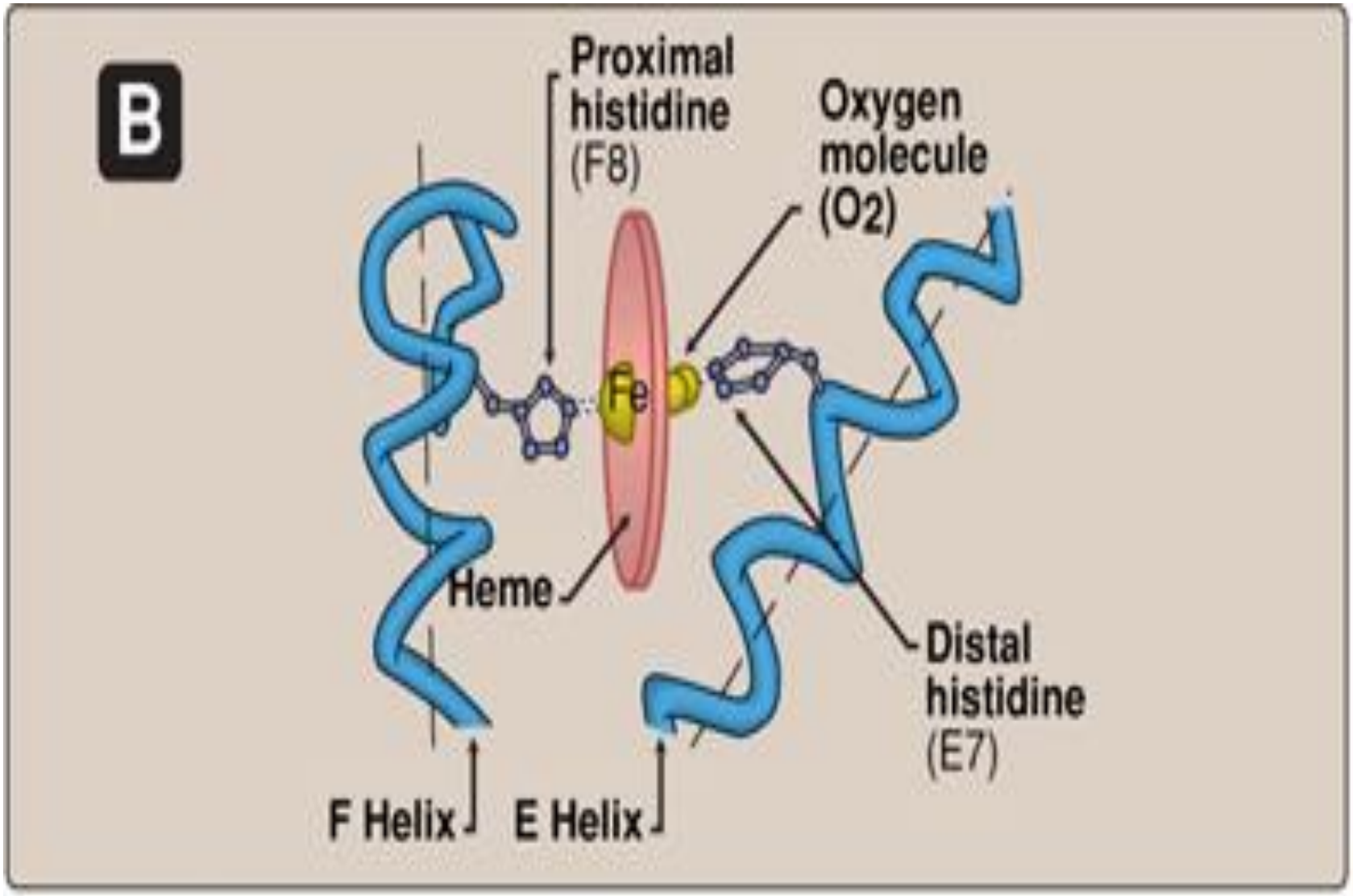
Figure 10-4 Complete Hb molecule. Heme is suspended between the E and F helices of the polypeptide chain. Pink represents α 1 (*left*) and α 2 (*right*); yellow represents non-α 2 (*left*) and non-α 1 (*right*).

Rodak, Hematology, third edtion

- **Each polypeptide binds a heme molecule at its center. (4 heme residues per Hb molecule)**



- **Heme lies in a pocket (hdrophobic cleft) between E and F helices**


B
Proximal histidine (F8)
Oxygen molecule (O_2)
Fe
Heme
Distal histidine (E7)
F Helix
E Helix

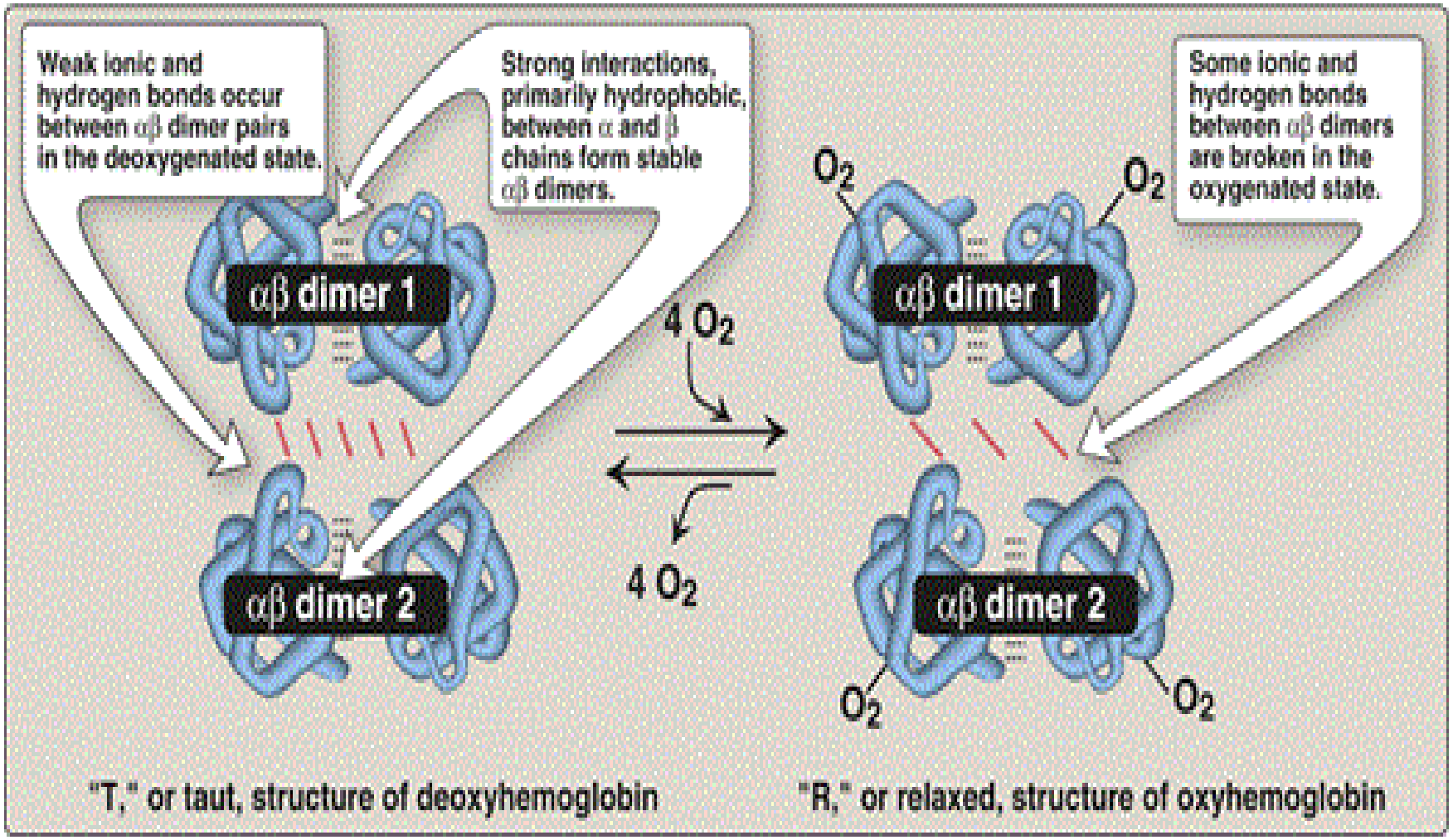
- The iron of heme is coordinated with the nitrogen of the imidazole ring of one histidine in the F helix this histidine is called the **proximal histidine**

- The other histidine residue in the E helix is called the **distal histidine** (lies near the heme but is not bonded to it). It stabilizes binding of oxygen to heme and destabilizes binding of carbon monoxide.

- The 2 polypeptide chains of each dimer are **tightly** held together, mostly by **hydrophobic bonds.**
- Each dimer is held relatively **loosely** to the other dimer, mostly by **ionic and hydrogen bonds.** This allows movement of the 2 dimers relative to each other, a process that occurs during oxygenation and deoxygenation.
- Thus 2 forms of Hb can be recognized:

1- The “T” form (Hb)

2- The “R” form (Hb)


Weak ionic and hydrogen bonds occur between αβ dimer pairs in the deoxygenated state.
Strong interactions, primarily hydrophobic, between α and β chains form stable αβ dimers.
Some ionic and hydrogen bonds between αβ dimers are broken in the oxygenated state.
αβ dimer 1
αβ dimer 2
4 O2
4 O2
O2
O2
αβ dimer 1
αβ dimer 2
O2
O2
"T," or taut, structure of deoxyhemoglobin
"R," or relaxed, structure of oxyhemoglobin

Binding of oxygen to Hb

- The ease with which an oxygen molecule binds to a certain Hb molecule depends upon whether other oxygen molecules are bound to it.
- Binding of oxygen to Hb is facilitated by previous binding of other oxygen molecules **(cooperative binding kinetics).**
- The affinity of Hb for the last oxygen molecule is about 300 times greater than for the first oxygen molecule.

- This pulls the proximal histidine towards the porphyrin ring and is accompanied by deprotonation of the imidazole ring of histidine and of N-terminal amino groups in the peptide chain.
- This leads to rupture of salt bonds between globin chains, and Hb changes from the T to R state, increasing its affinity for oxygen.
- This is sometimes called **heme-heme interaction.**

- **The "T" form (Hb):**

  - This is the taut (tense) form of hemoglobin. The polypeptide chains are difficult to move relative to each other because of the presence of more ionic bonds between the 2 dimers.

  -It is stabilized by protonation due to increased number of charged groups, increasing ionic bonds.

  -This occurs when CO2 is added to the blood by the tissues increasing $[H^+]$ in red cells. It is stabilized by deoxygenation, which leads to protonation of Hb.

  -It has a lower affinity for oxygen.

- **The “R” form (Hb):**

-This is the relaxed form of Hb. There are less ionic bonds between the two dimers, and the polypeptide chains are more free to move relative to each other.

-It is stabilized by deprotonation due to decreased number of charged groups, decreasing ionic bonds.

-This occurs at the lungs when CO2 is lost from the blood. It is stabilized by oxygenation, which leads to deprotonation of Hb.

-It has a higher affinity for oxygen.

Allosteric properties of Hb

- The ability of Hb to reversibly bind oxygen is affected by:

1-The pO2 (through heme-heme interactions as described before)

2-The pH of the environment

3-The pCO2

4-The availability of 2,3-bisphosphoglycerate.

- These are collectively called allosteric effectors

  (allos=other, steros=site) because their interaction at one site on the Hb molecule affects the binding of oxygen to heme groups at other locations on the molecule.

Carbon dioxide

- Some of the CO_2 (15%) delivered by the tissues to the blood is carried by the terminal amino groups of Hb in the form of carbamate.

$$Hb\text{-}NH_2 + CO_2 \longrightarrow Hb\text{-}NH\text{-}COO^- + H^+$$

This gives Hb a negative charge, increases the formation of ionic bonds, which stabilizes the T-form. The affinity of Hb for oxygen decreases, helping delivery of oxygen to the tissues.

pH (The Bohr Effect):

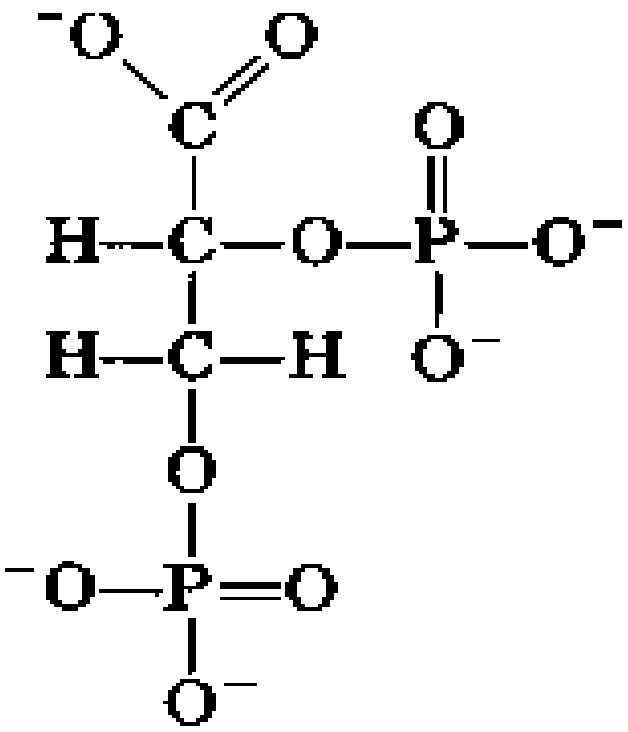
- Most of the CO2 delivered by the tissues to the blood is converted to H2CO3 in the red blood cells. H2CO3 liberates hydrogen ions, which protonate the N-terminal amino groups of the α-subunits and the C-terminal histidine of the β-subunits, stabilizing the T-form.
- The affinity of Hb for oxygen decreases, helping delivery of oxygen to the tissues. The reverse occurs at the lungs.

$$Hb\text{-}O_2 + H^+ \longrightarrow Hb\text{-}H^+ + O_2$$

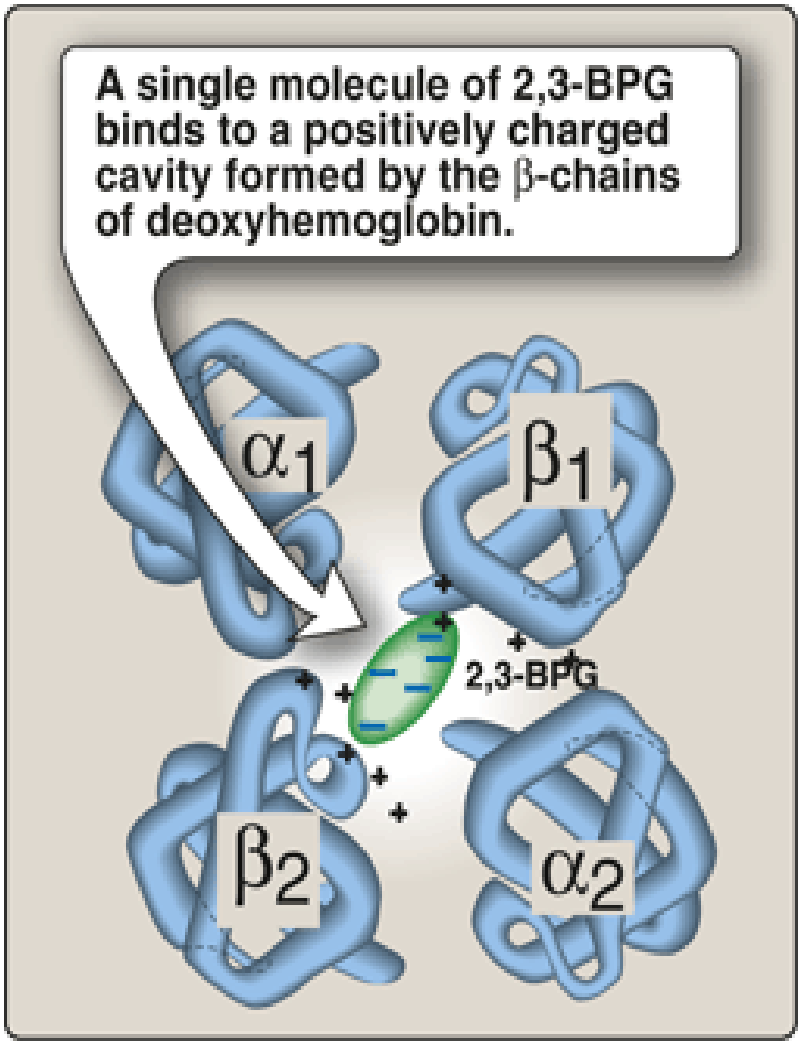
- The influence of pH and pCO_2 to facilitate oxygenation of Hb in the lungs and deoxygenation at the tissues is known as the **Bohr effect.**

2,3- Bisphosphoglycerate

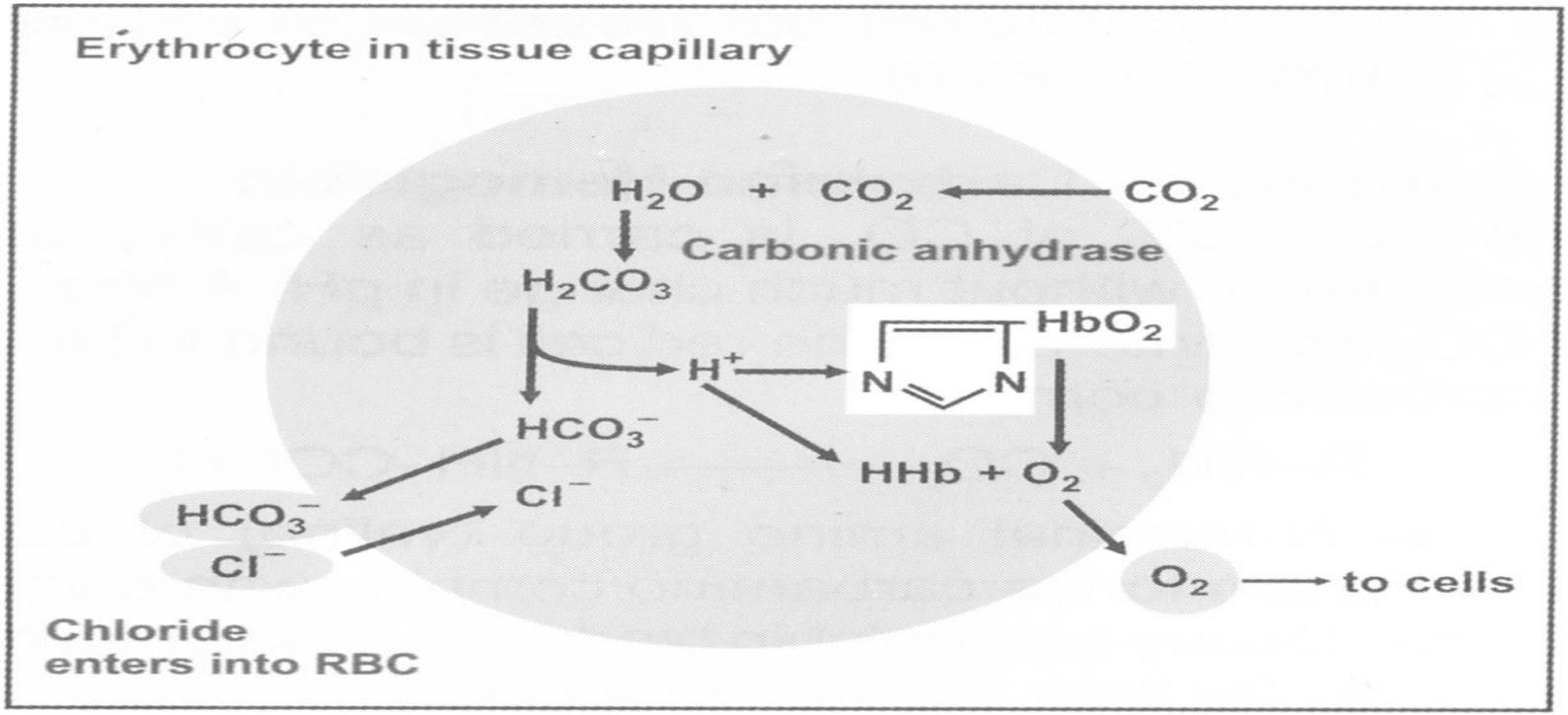
- The 2,3-bisphosphoglycerate (BPG) molecule carries 5 negative charges and is derived from oxidation of glucose (glycolysis) in red cells.
- It binds to a positively charged pocket in Hb between the 2 β chains (small cavity in the center of the four Hb subunits)
- Binding favors the T- form of Hb, reducing affinity for oxygen and helping delivery of oxygen to tissues.
- BPG increases in red blood cells in cases of chronic anemia and in hypoxia. This helps delivery of oxygen to tissues.



2,3-Bisphosphoglycerate

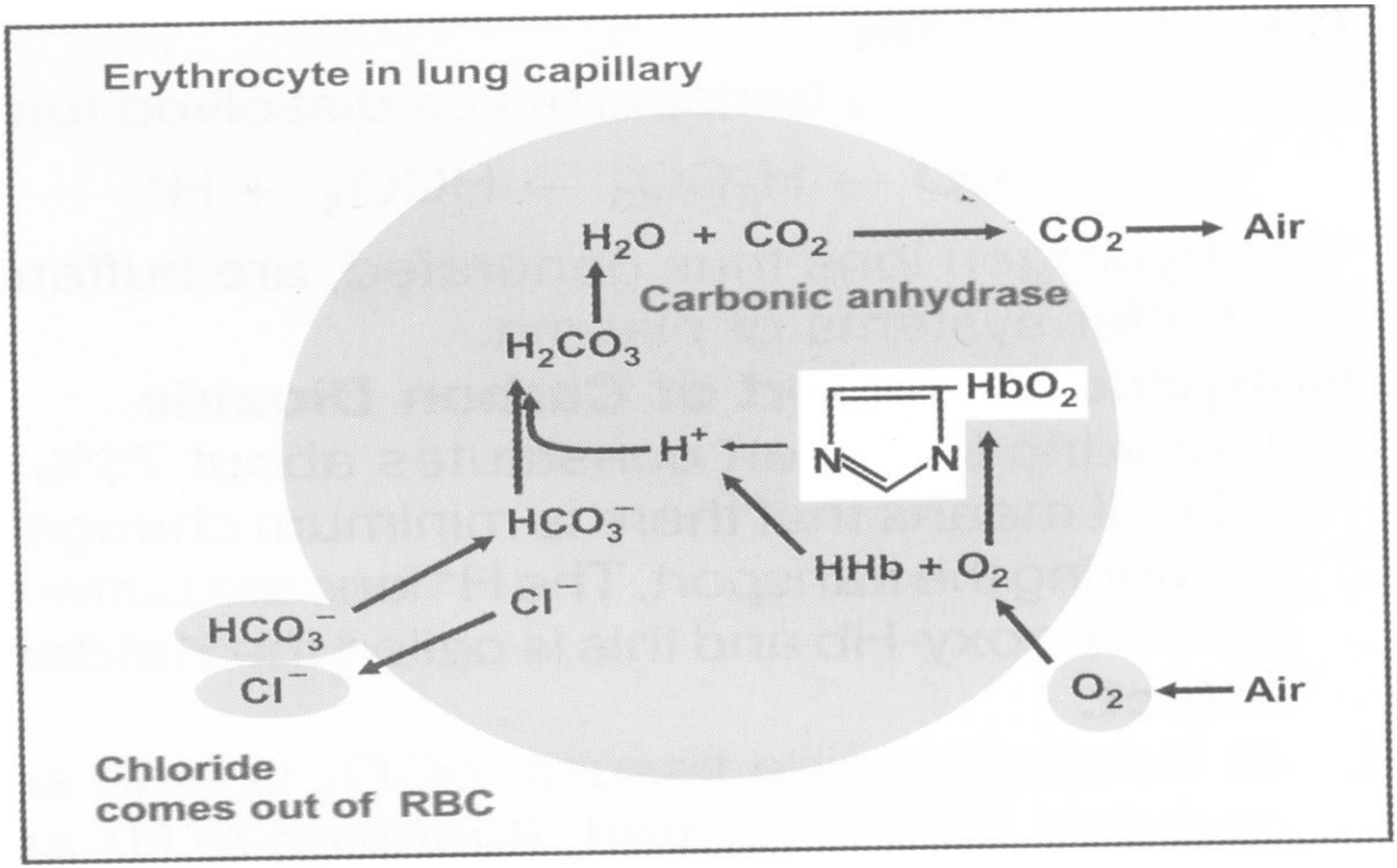


- BPG decreases in red blood cells upon storage of blood, reducing delivery of oxygen to tissues.
- This can be prevented by adding inosine to the blood collection medium, which keeps the level of BPG normal in red blood cells.



The chloride shift

When CO2 is taken up, the HCO3- concentration within the cell increases. This would diffuse out into the plasma. Simultaneously, chloride ions from the plasma would enter in the cell to establish electrical neutrality. This is called chloride shift or Hamburger effect.


Erythrocyte in lung capillary
$H_2O + CO_2$
CO_2
Air
Carbonic anhydrase
H_2CO_3
HbO_2
H^+
N
N
HCO_3^-
$HHb + O_2$
Cl^-
HCO_3^-
Cl^-
O_2
Air
Chloride
comes out of RBC