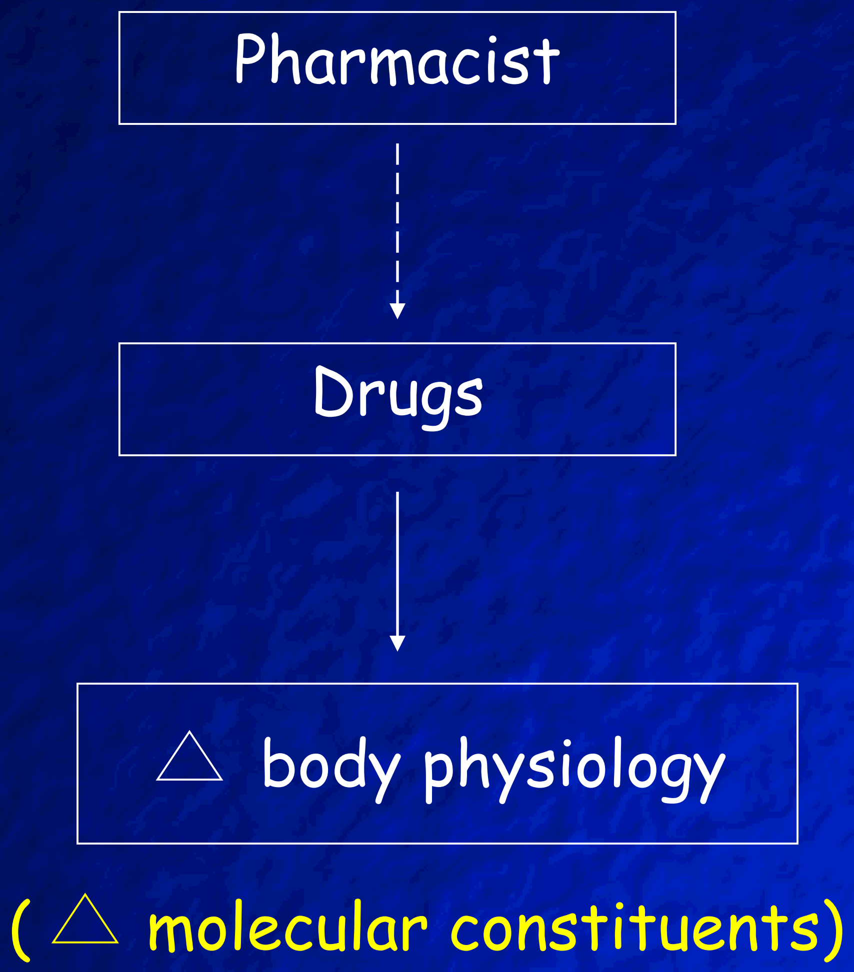
Why?


Pharmacist
Drugs
△ body physiology
(△ molecular constituents)

Mechanistic levels of response:

Altered patient response

physiologic systems

Vascular system

tissues / organs

blood, muscle, liver

cellular processes

erythrocytes

(proteins)

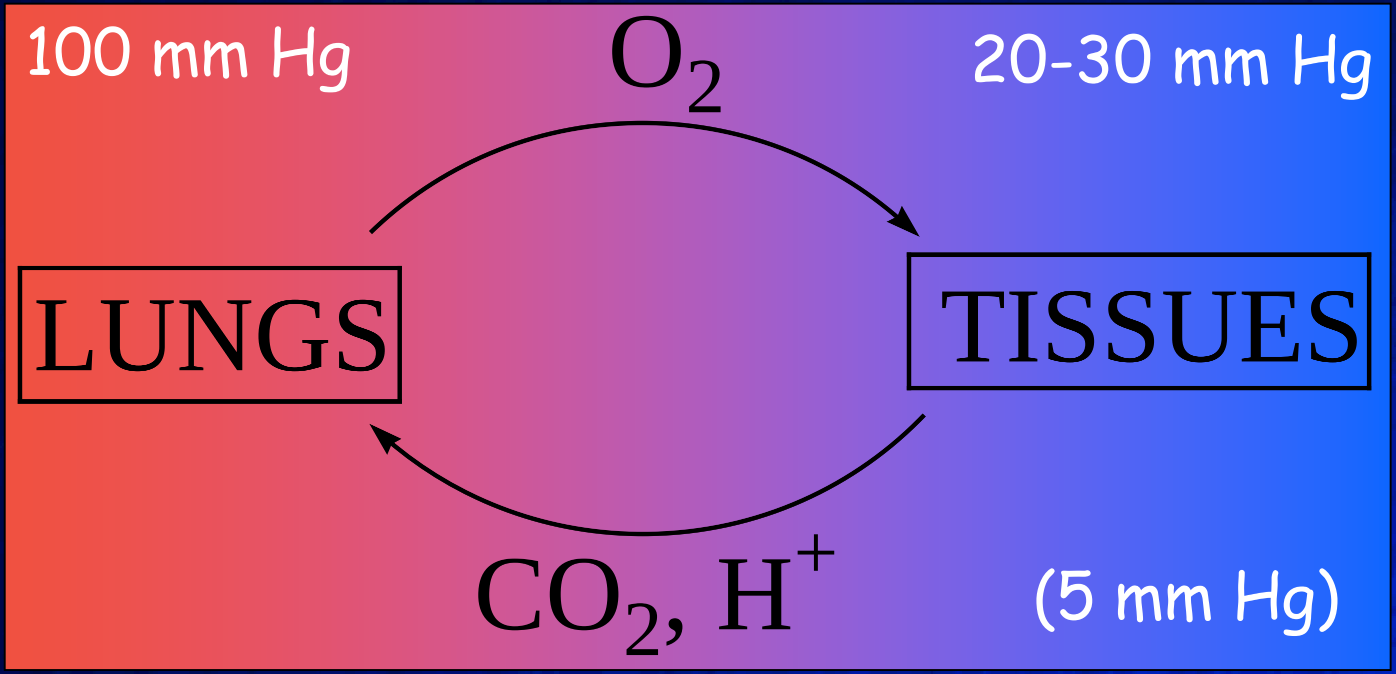
hemoglobin
myoglobin

UNIT OVERVIEW:
ERYTHROCYTES AND OXYGEN DELIVERY

1. Biology of erythrocytes / vasculature
2. Hemoglobin and Myoglobin function
3. Energy metabolism in erythrocytes
4. 2,3 Diphosphoglycerate
5. Drugs / toxins which affect erythrocyte function

Recommended reading:
(Devlin) pp. 393-410, (Stryer) pp. 269-274

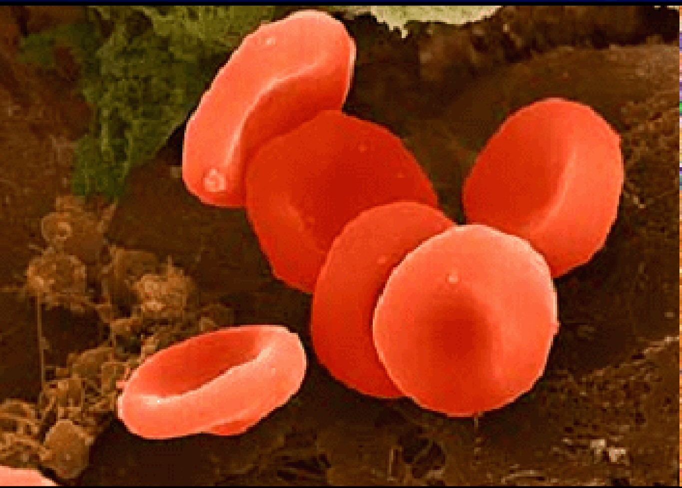
Primary RBC function: transport of O_2 / CO_2

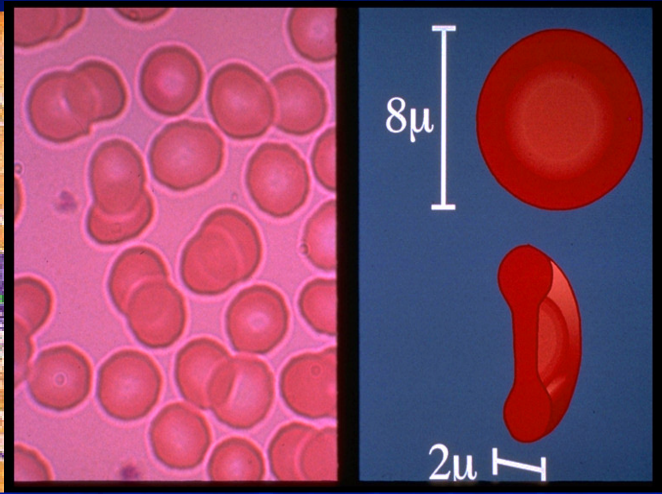


*Blood is a colloid, in addition to RBC's, blood also contains:

- Additional cell types: leucocytes (WBC's), platelets (clotting)
- Free proteins: albumin, globulins (Ig), ferritins (transport), enzymes (clotting), hormones
- Other non-cellular components: electrolytes

Erythrocytes:





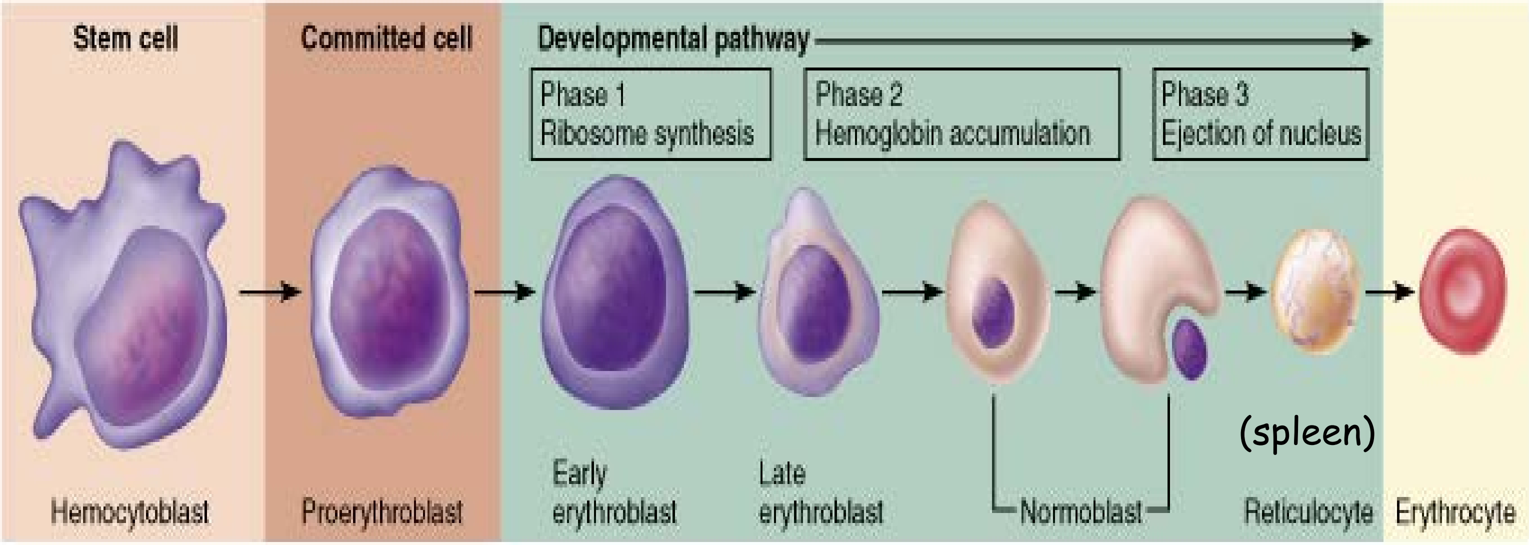
From Tina Carvalho (MicroAngela)

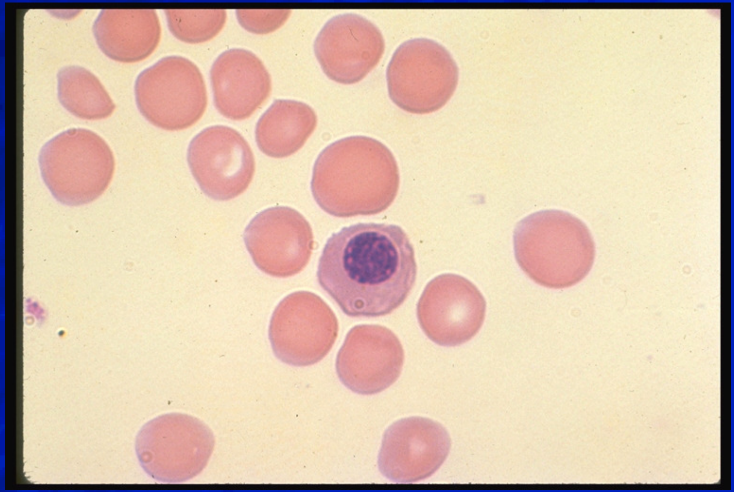
Key erythrocyte features:

- 95% of cellular protein is hemoglobin (35% by weight)
- humans: 300x 10^6 Hb molecules/RBC, 4.2-5.8 x 10^6 RBC's/cc blood
- hematocrit 35-50%, produce / destroy 2.5 x 10^6 RBC's/sec
- RBC's harbor variety of membrane transporters (glucose) on cell surface

RBC Parameters	Normal Values
Hematocrit	
Females	35-47%
Males	40-52%
Hemoglobin	
Females	12.0-16.0 gm/dl
Males	13.5-17.5 gm/dl
MCV	80-100 fl
Reticulocyte Count	0.2-2.0%

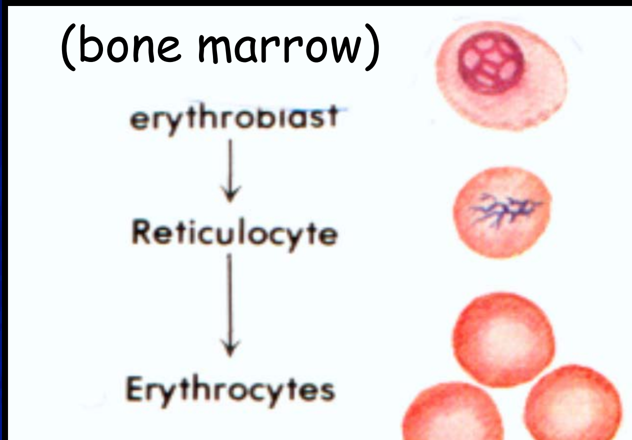
Erythrocyte development:





BIOLOGY OF ERYTHROCYTES/VASCULATURE

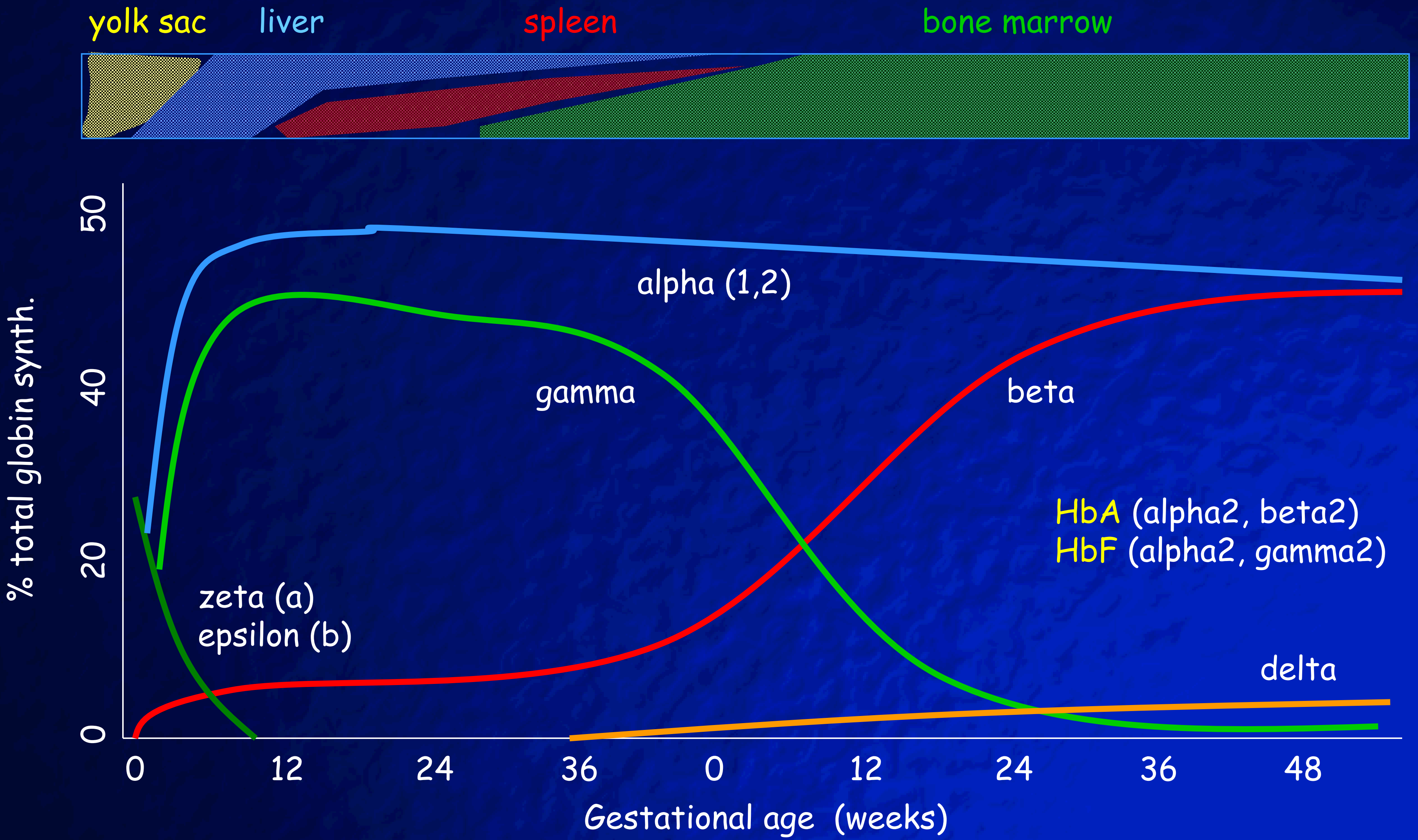
* Fetal development – RBC's produced in liver. Bone marrow production commences at 4 mo. in humans. Adult – erythrocyte production occurs only in bone marrow.



Phys. Mech. of Disease: 4th Ed. 1

* Mitochondria, nuclei and endoplasmic reticulum lost as reticulocytes mature into adult erythrocytes. Thus NO mito. Respiration.
* Therefore NO gene transcription or protein translation occurs in RBC's – all proteins within the erythrocyte must be produced at the time of genesis.
* No mitochondrial respiration, thus low ATP formation. Energy requirements of the cell must be met largely through GLYCOLYSIS.

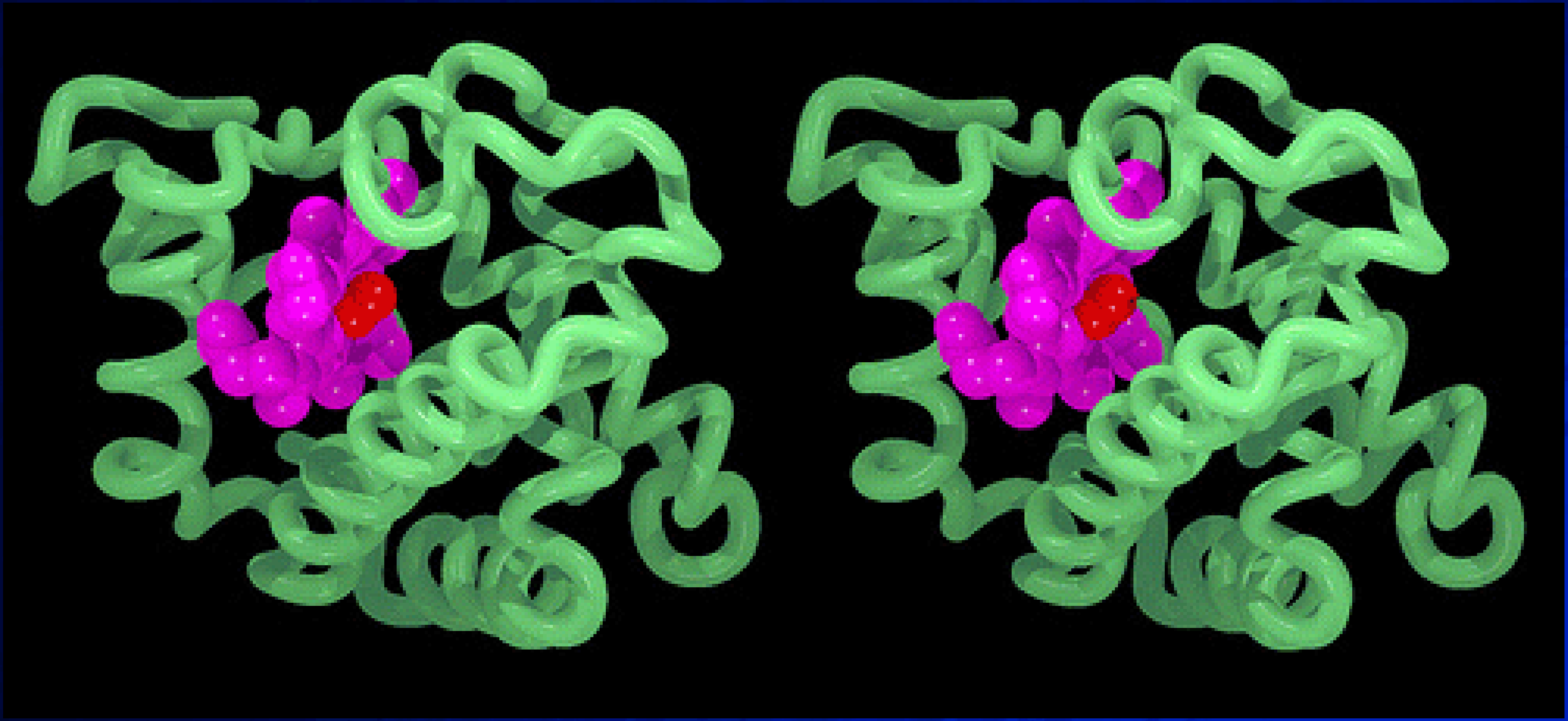
Globin synthesis during development:



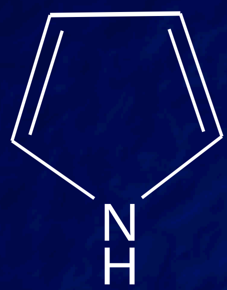
HEMOGLOBIN and MYOGLOBIN

- Physical structure of hemoglobin
- Developmental expression of globin genes
- Mechanisms of O_2 regulation
- Allosterism and important conformational changes
- Regulation by external agents

Structure of Myoglobin:



Hemoglobin / Myoglobin: Heme-containing proteins



pyrrole ring

The iron atom in heme can form 6 bonds.

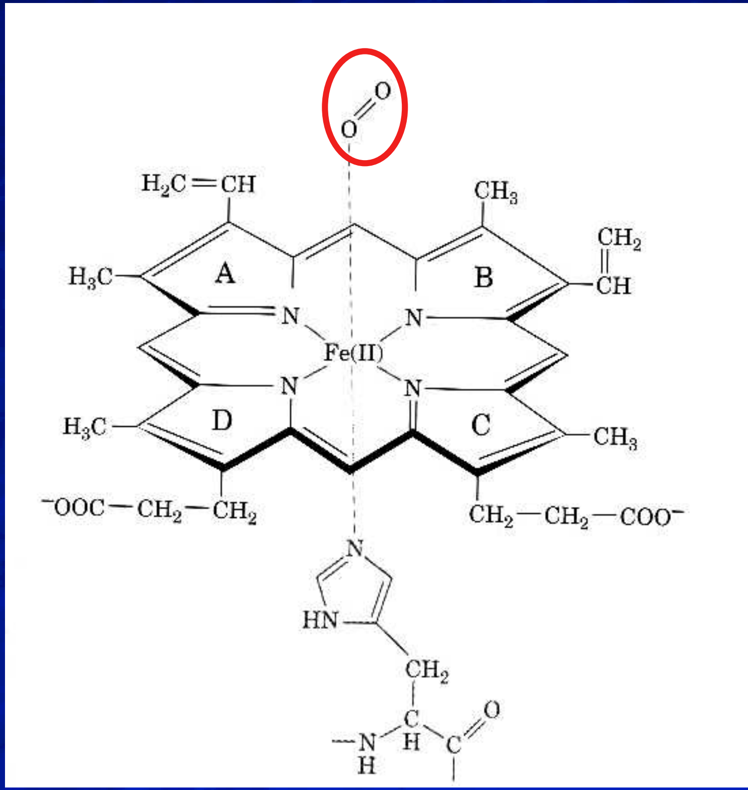
Catabolism:

Fe ⟶ reused (Tf)

Globin ⟶ peptidase (AA)

Heme ⟶ Bilirubin

Heme - a cyclic tetrapyrrole (Fe(II)-protoporphyrin IX)

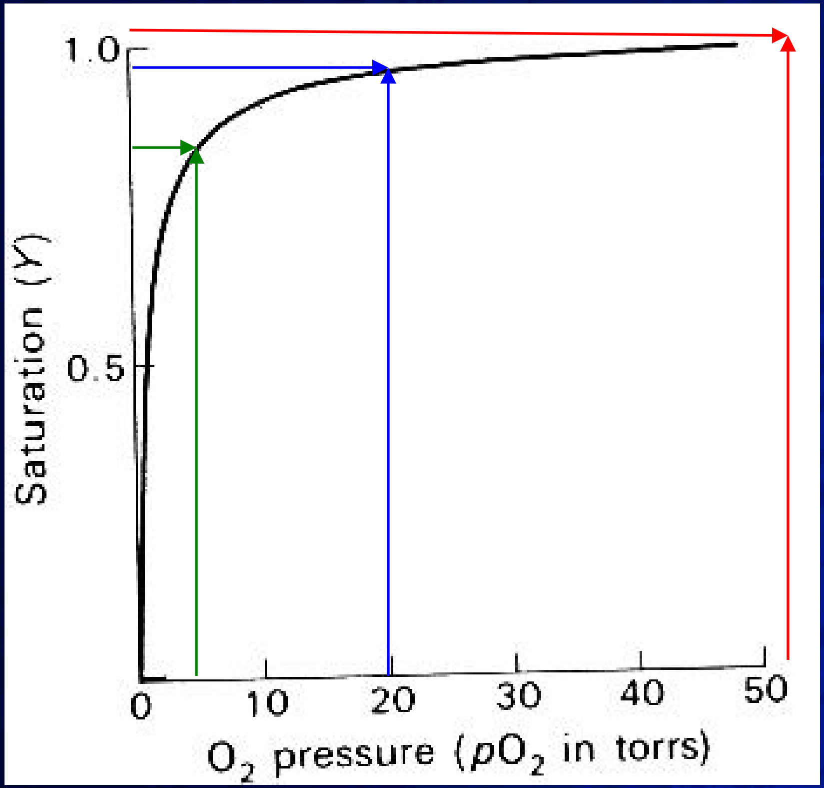


O_2 binding to heme of Mb or Hb is reversible

Devlin Fig. 9.31, 9.32, pyrrole - Harpers Fig. 7-1

Myoglobin (Mb) O_2 binding curve:

Myoglobin: single chain protein, 1 heme/protein, first x-ray solution structure solved. Because it contains only a single subunit, it does NOT display cooperativity or allosterism (hyperbolic O2 curve).



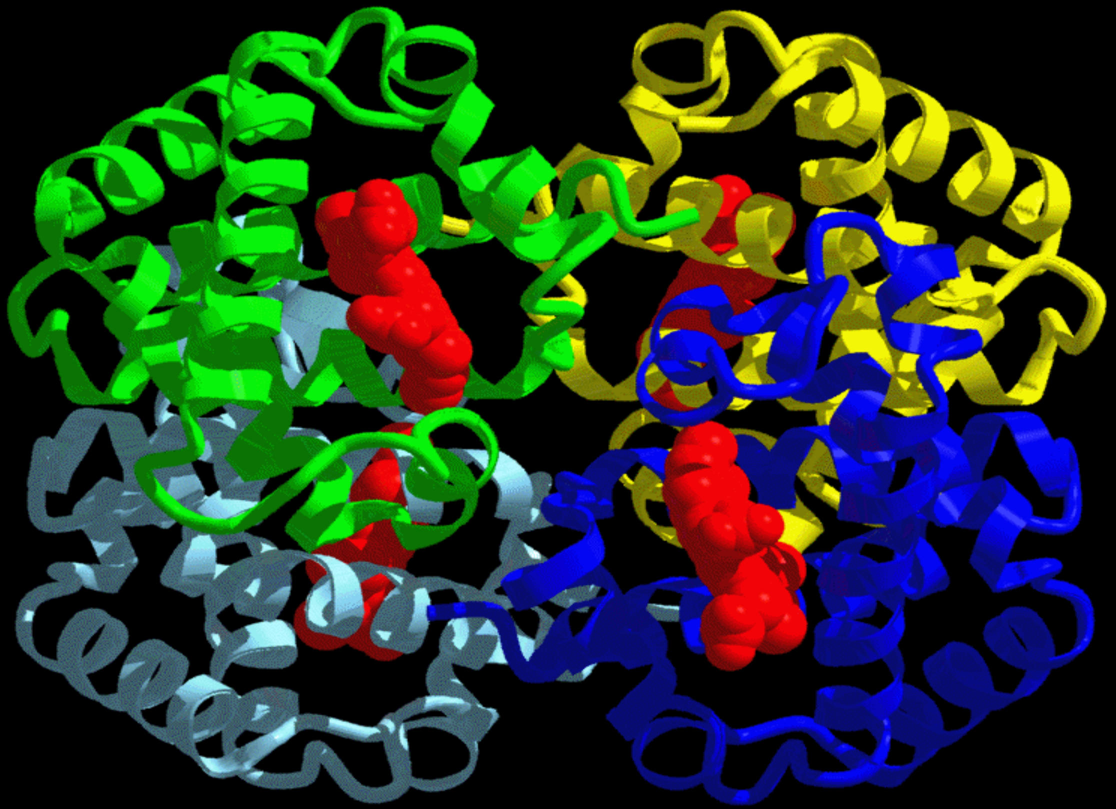
P_{50} Mb: 2.8 torr

low P_{50} = high O_2 affinity

pO_2 lungs: 100 mm Hg
tissues: 20 mm Hg
working muscle: 5 mm Hg
~1,100 m. years ago

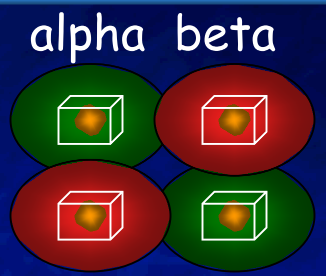
modified from Stryer Fig 10.17, see also Devlin 9.35

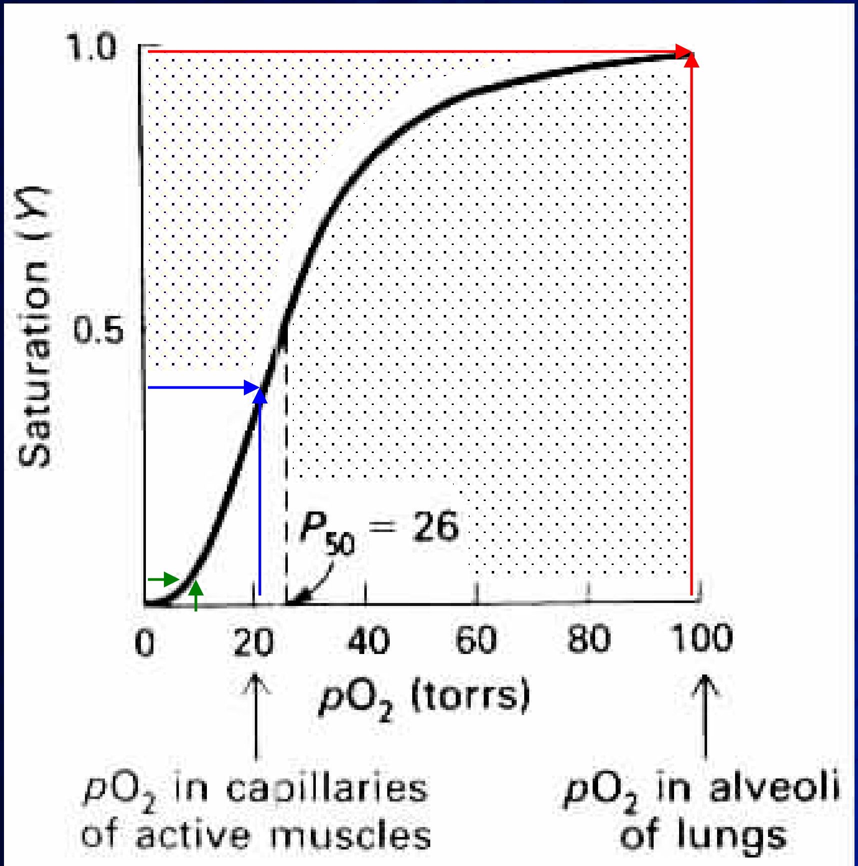
Hemoglobin Structure:



Hemoglobin (Hb) O_2 binding curve:

Hb: tetrameric protein (~2-500 m years)
4 subunits and heme groups/protein





Devlin 9.35

The tetrameric structure of Hb imparts it with several important properties:

Allosterism:
The binding of a ligand (O_2) at one site affects the binding of other ligands at distal sites. Thus Hb exhibits sigmoidal O_2 kinetics.

Positive cooperativity:
The affinity of Hb for the 4^{th} O_2 is 100x greater than for the first, due to conformation changes in Hb.

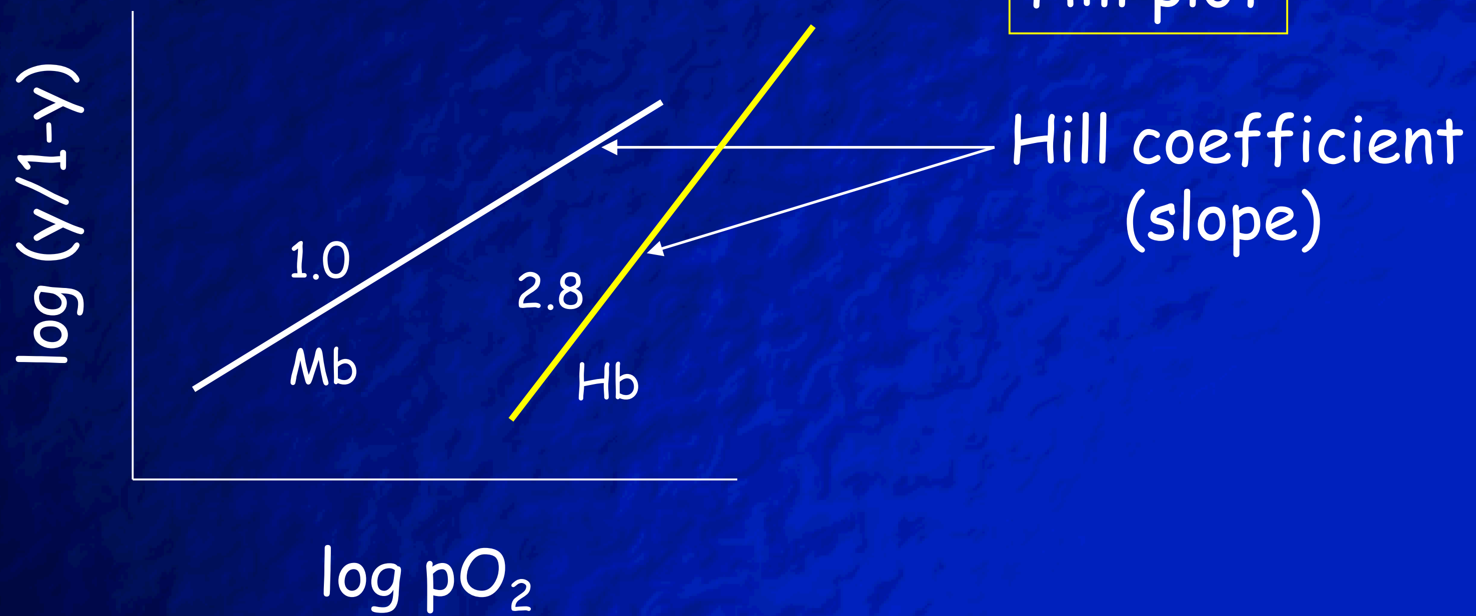
Measures of cooperativity, Hill plot:

$$Y = \frac{\text{number of binding sites occupied}}{\text{total number of binding sites}}$$

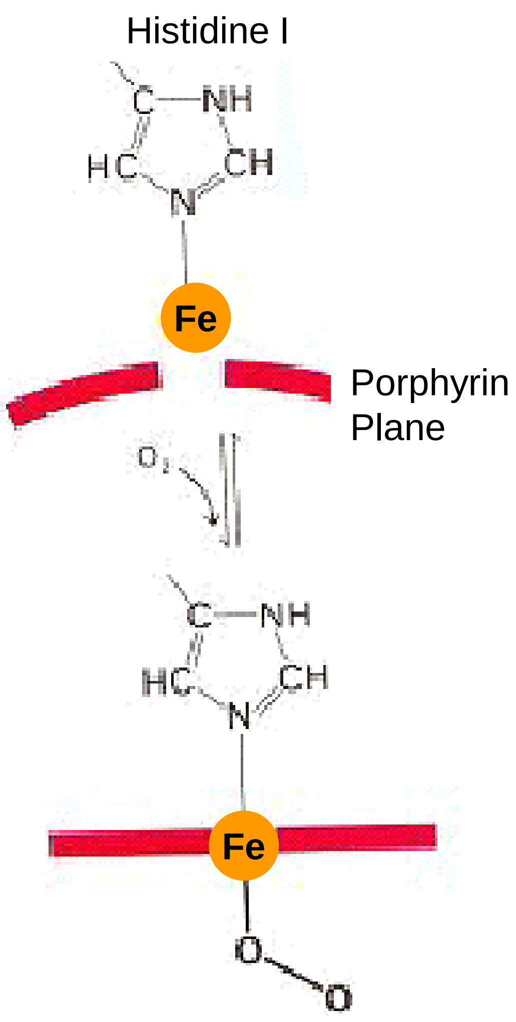
$$Y/1\text{-}Y = pO_2/\ pO2^{(50)}$$

$$\log Y/1\text{-}Y = \log pO2 - \log pO2^{(50)}$$

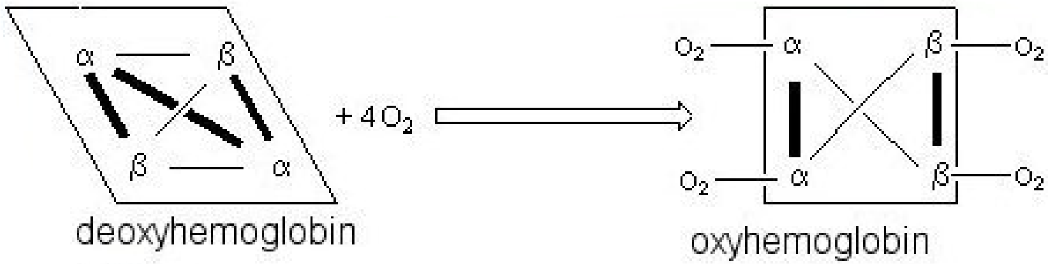
Hill plot



Perutz mechanism:



On the basis of the X-ray structure of oxy- and deoxyhemoglobin, Perutz formulated a mechanism for hemoglobin oxygenation. Perutz postulated that hemoglobin has 2 stable conformational states; the dexoy "T"-state, and the fully oxygenated "R"-state. The conformation of subunits in T-state hemoglobin differ from those in the R-state. O2 binding initiates a series of coordinated movements that result in a shift from the T to the R state in a few microseconds.



R = relaxed = oxy state, T = tense = deoxy state

Oxygen Binding Site of Hemoglobin:

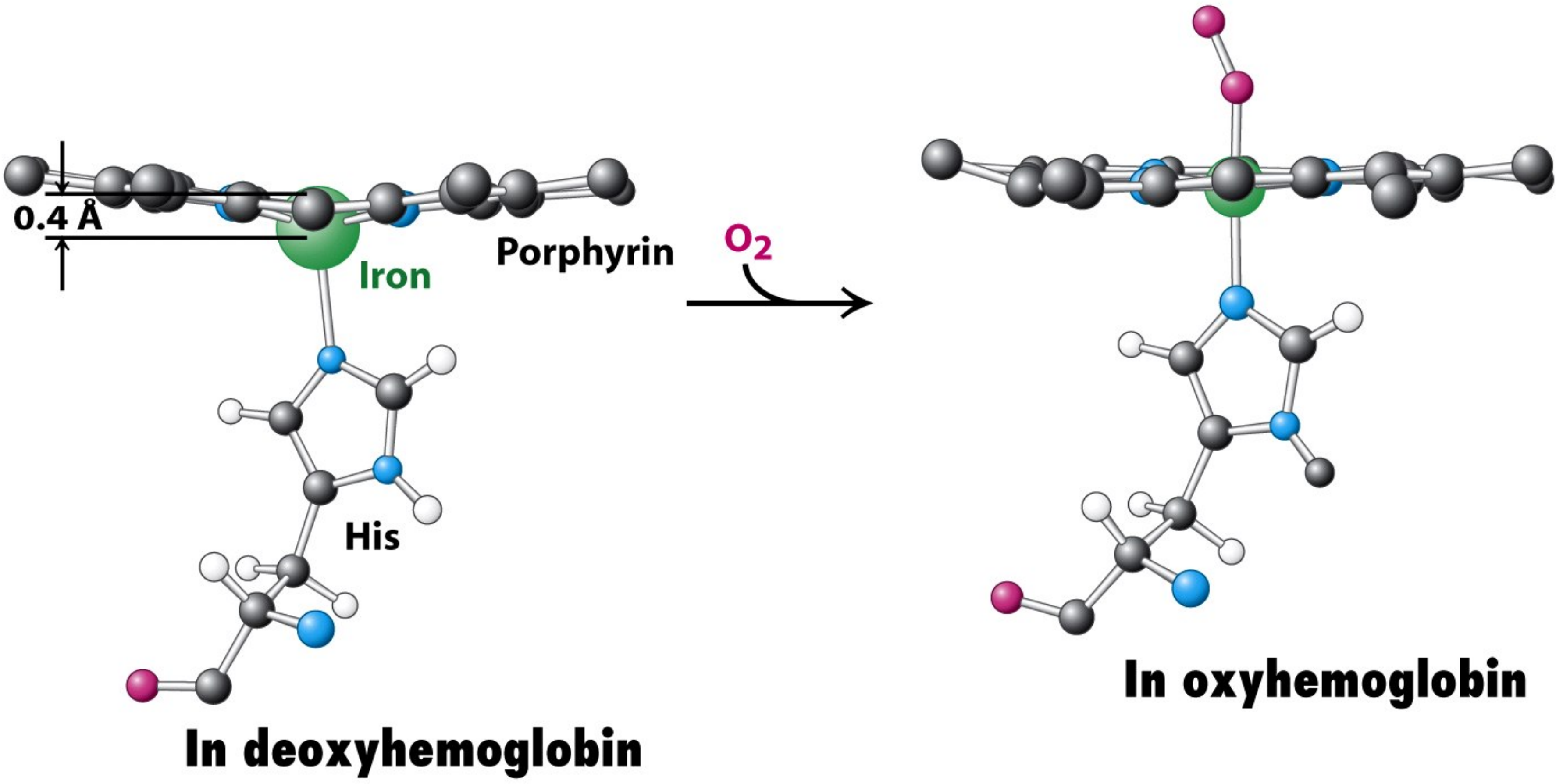


Figure 7-2
Biochemistry, Sixth Edition

Oxygen Binding Site of Hemoglobin:

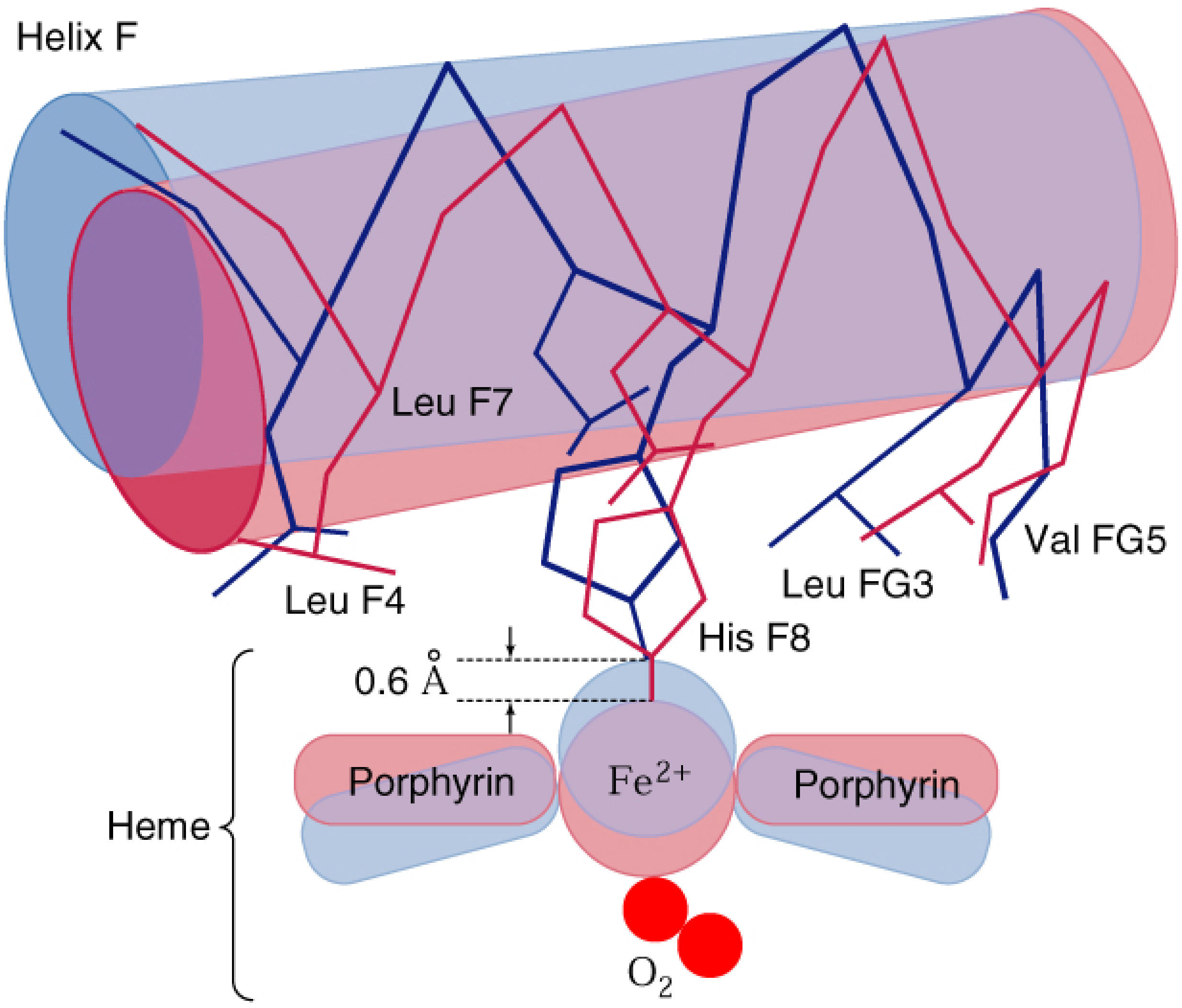
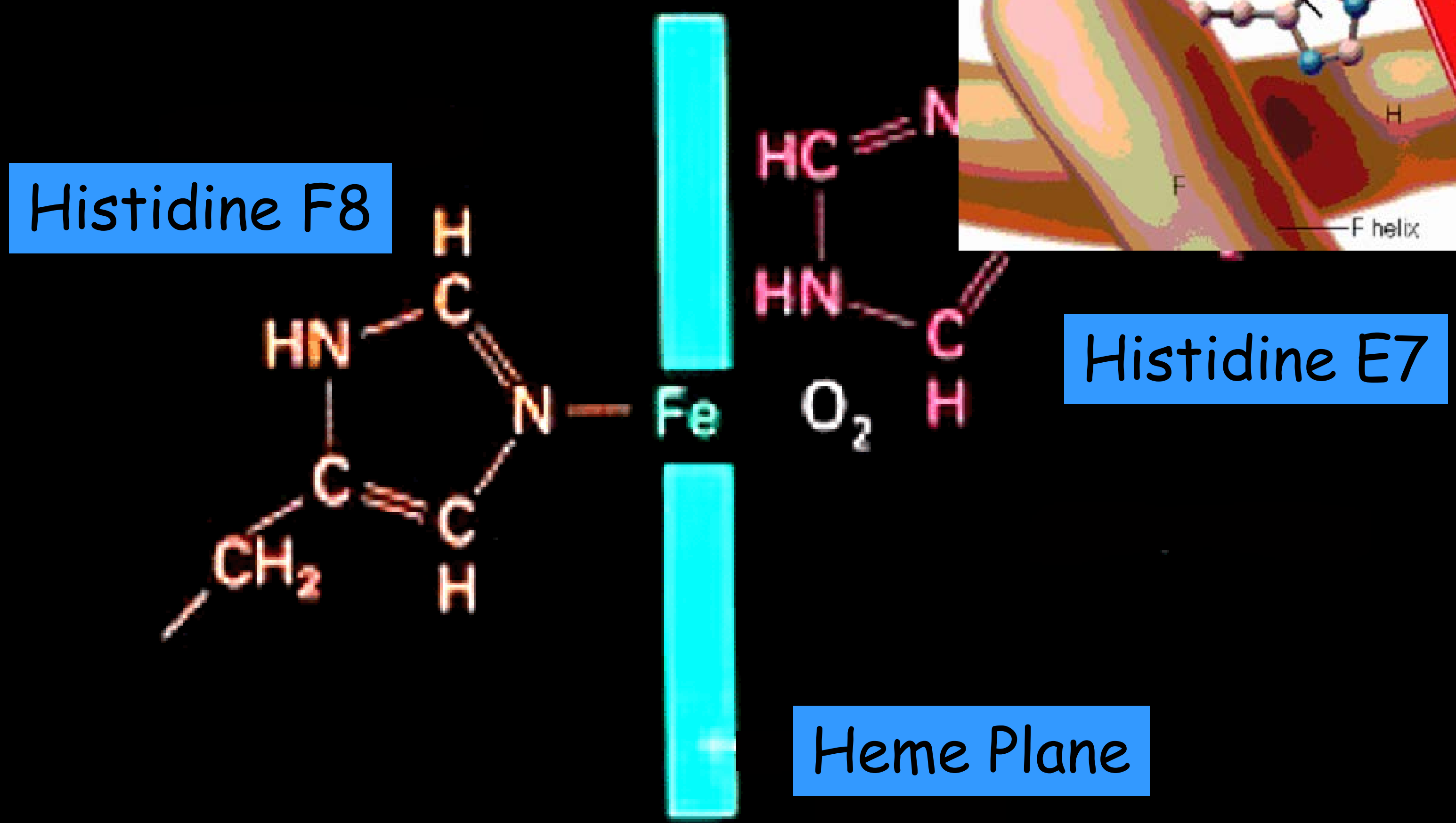
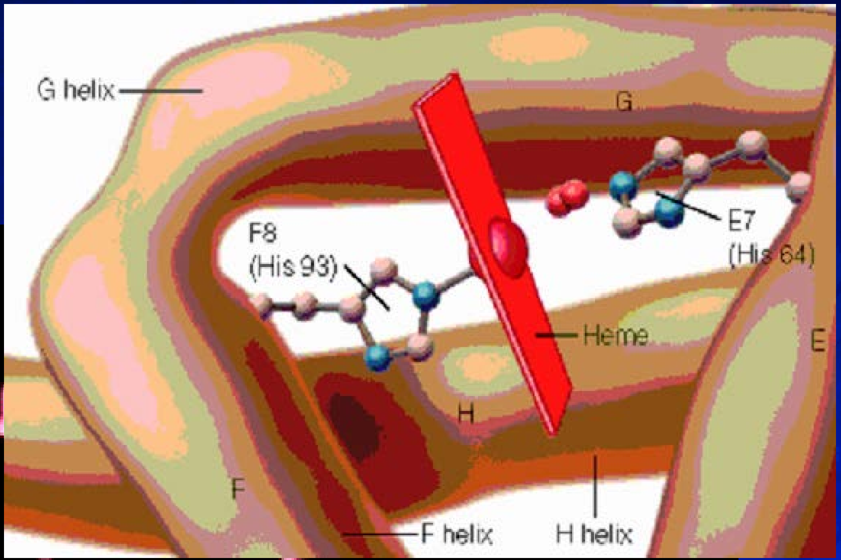


Figure 7-9. Movements of the heme and the F helix during the T → R transition in hemoglobin.

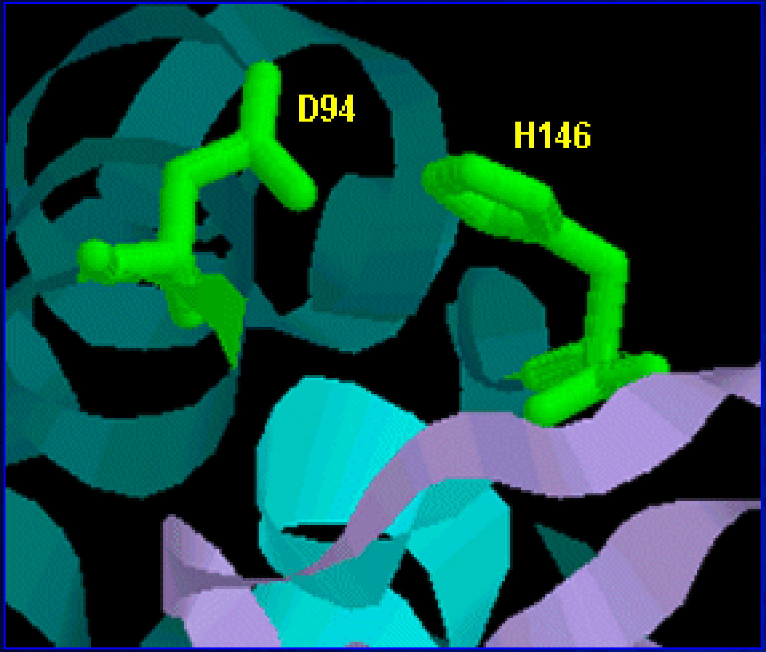
Oxygen Binding Site of Hemoglobin:

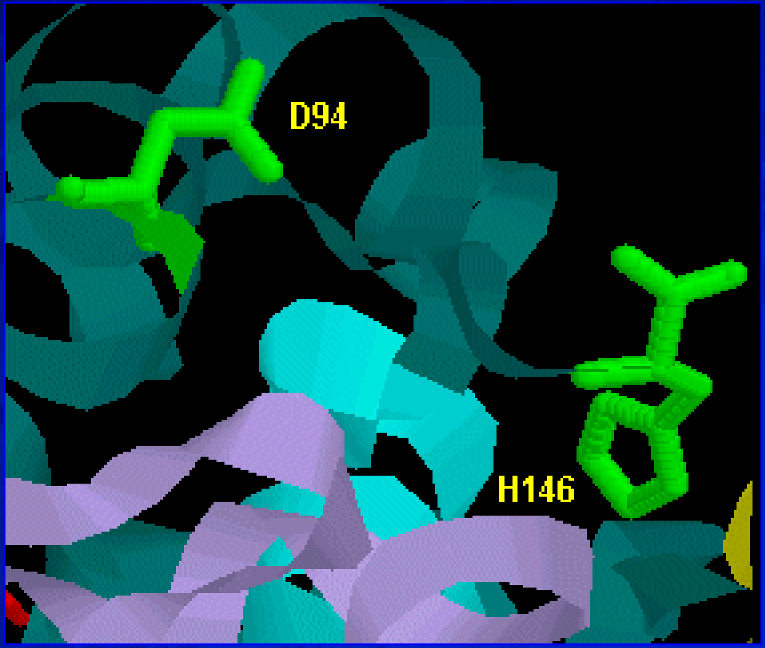




Structural states of Hb:

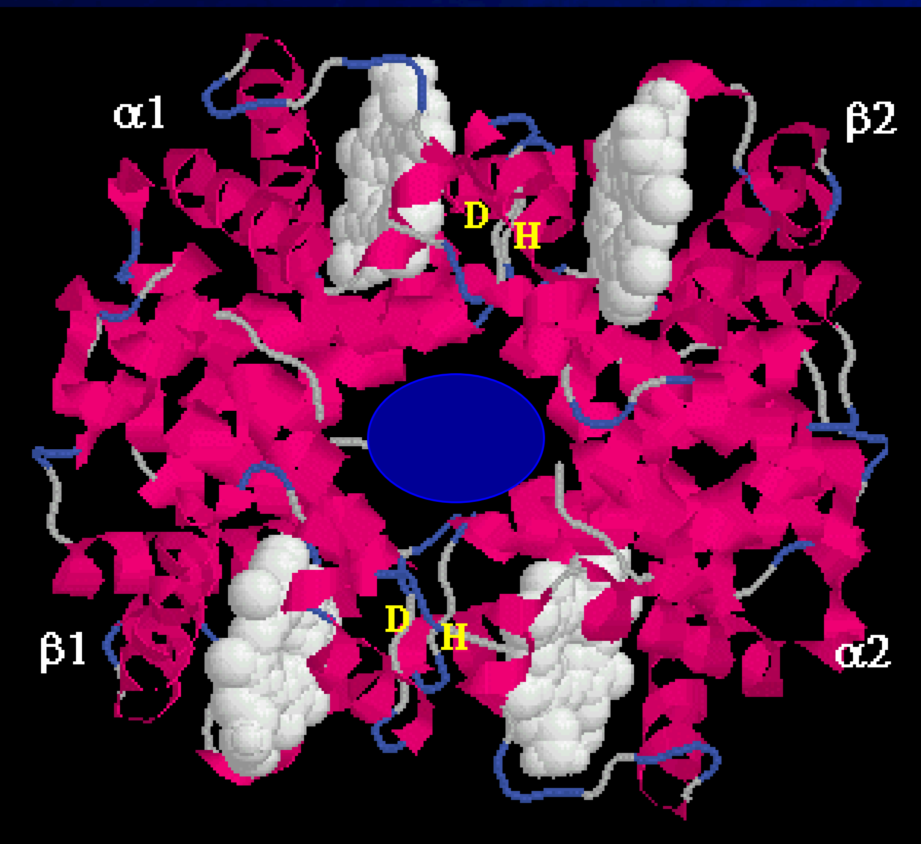
Deoxy Hb (T state) ⇄ Oxy Hb (R state)

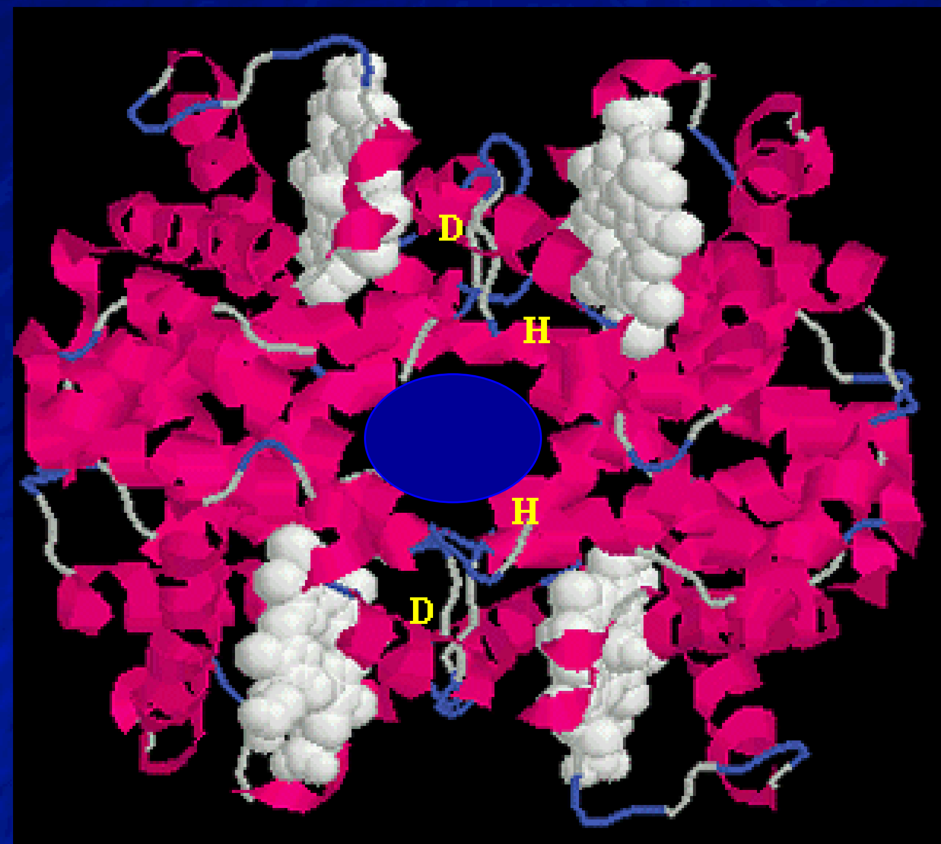




Structural states of Hb:

Deoxy Hb (T state) ⇄ Oxy Hb (R state)





The Bohr effect:

Blood pH: 7.35-7.5

$CO_2 + H_2O$

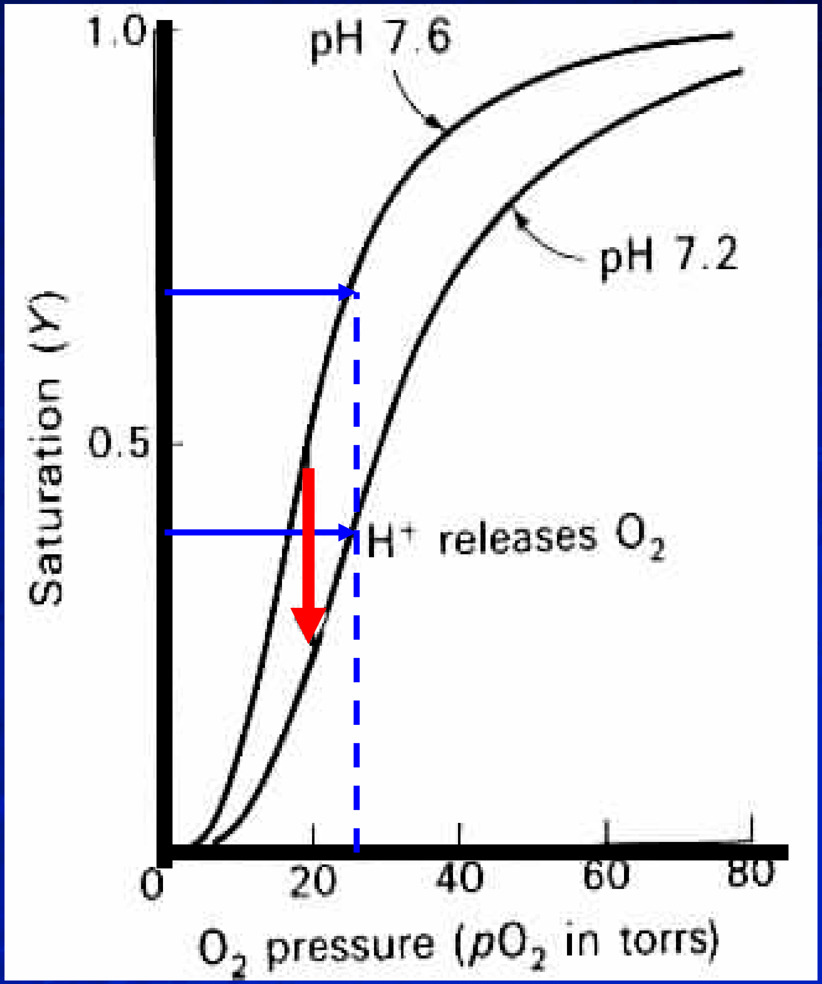
↓ carbonic anhydrase

T-Hb ↖ H_2CO_3 Carbonic acid

↓

$H^+ + HCO_3^-$ Bicarbonic ion

CO_2 and H^+ produced during metabolism causes ↓ pH in RBCs, resulting in protonation of some amino acid groups in Hb. These effects decrease the affinity of Hb for O_2 in RBCs (protons bind to the T form of hemoglobin thus stabilizing it).



See Devlin 9.42, and Stryer Fig. 10.25