Classes 6 and 7

(Ch. 5, pp. 157-174)

Protein structure & function:

Myoglobin & Hemoglobin

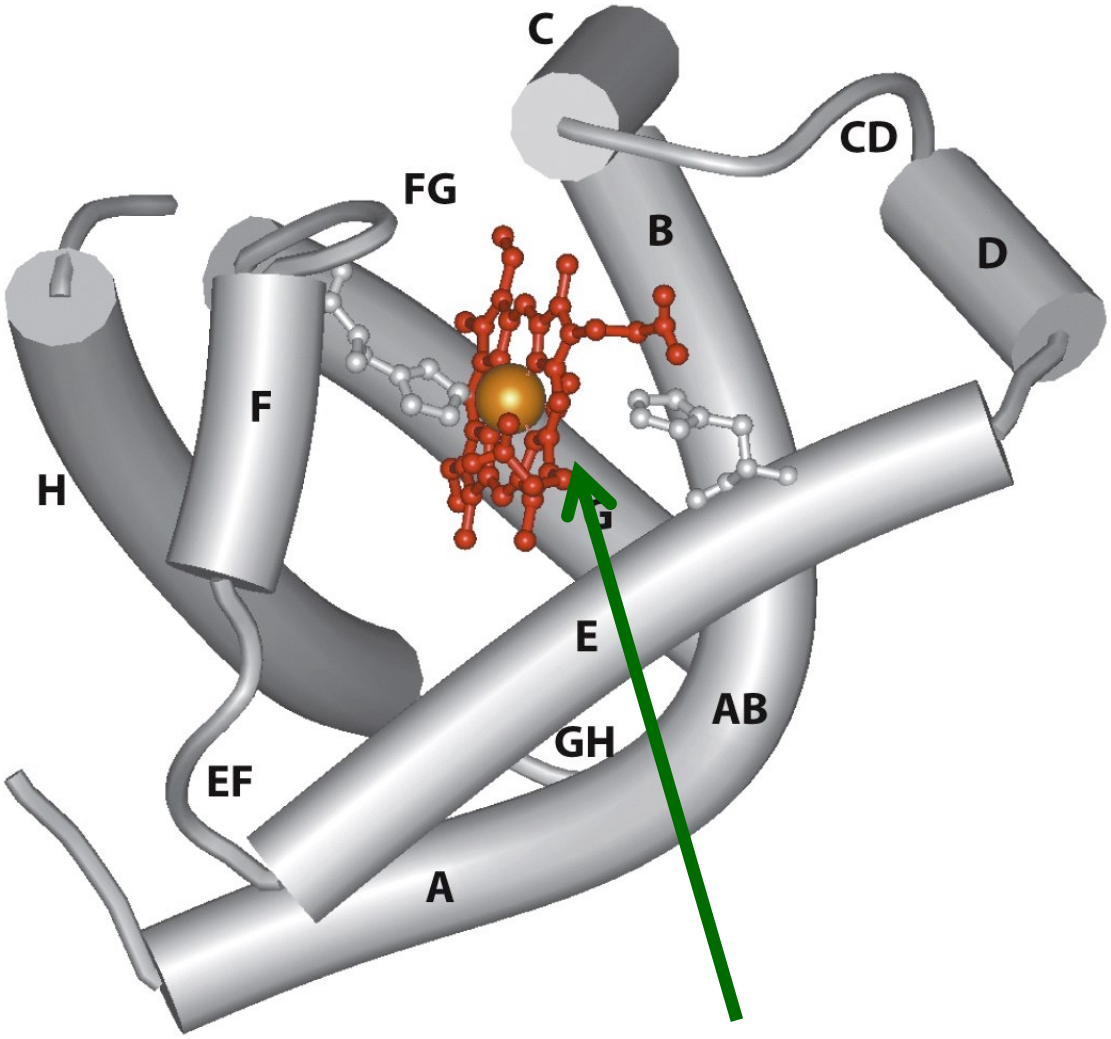
We will have a “structure” activity in lab next week which will complement the lecture material.

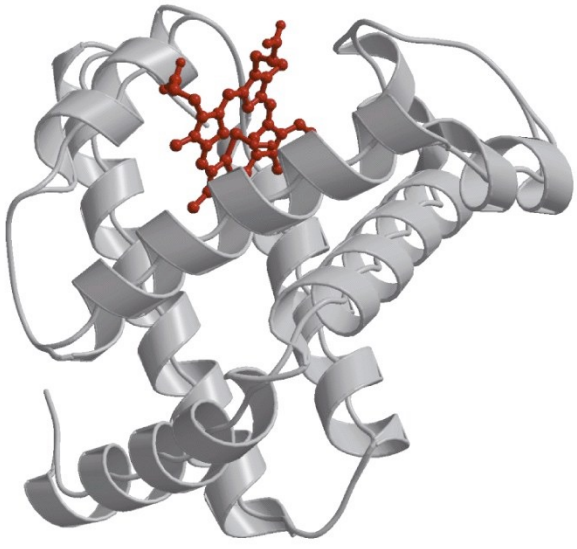
There is also a HW (#3) due Tues, Sept 29.

What do **aerobic** organisms absolutely need to generate energy during metabolism?

- Function of myoglobin?
- Function of hemoglobin?
- How do structural differences lead to different functions?
- Quantitatives of ligand-protein binding
- Physiological factors that affect O_2 binding by Hb
- Hemoglobin mutants

Myoglobin (monomer)





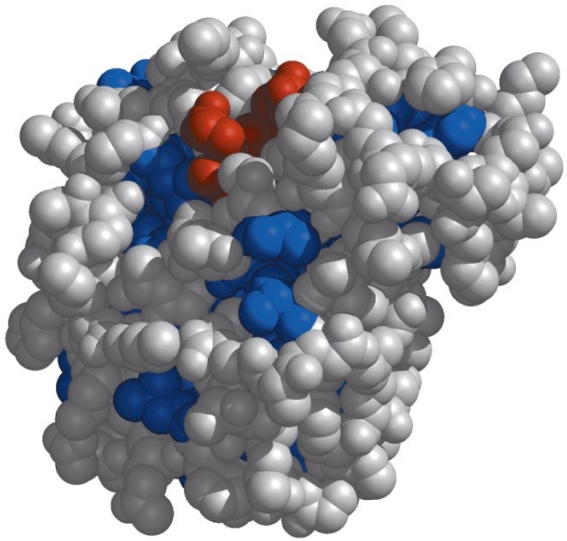


Figure 5-3

Heme **prosthetic** group:
Compound covalently attached to protein that contributes to protein's function.

Hemoglobin (tetramer)

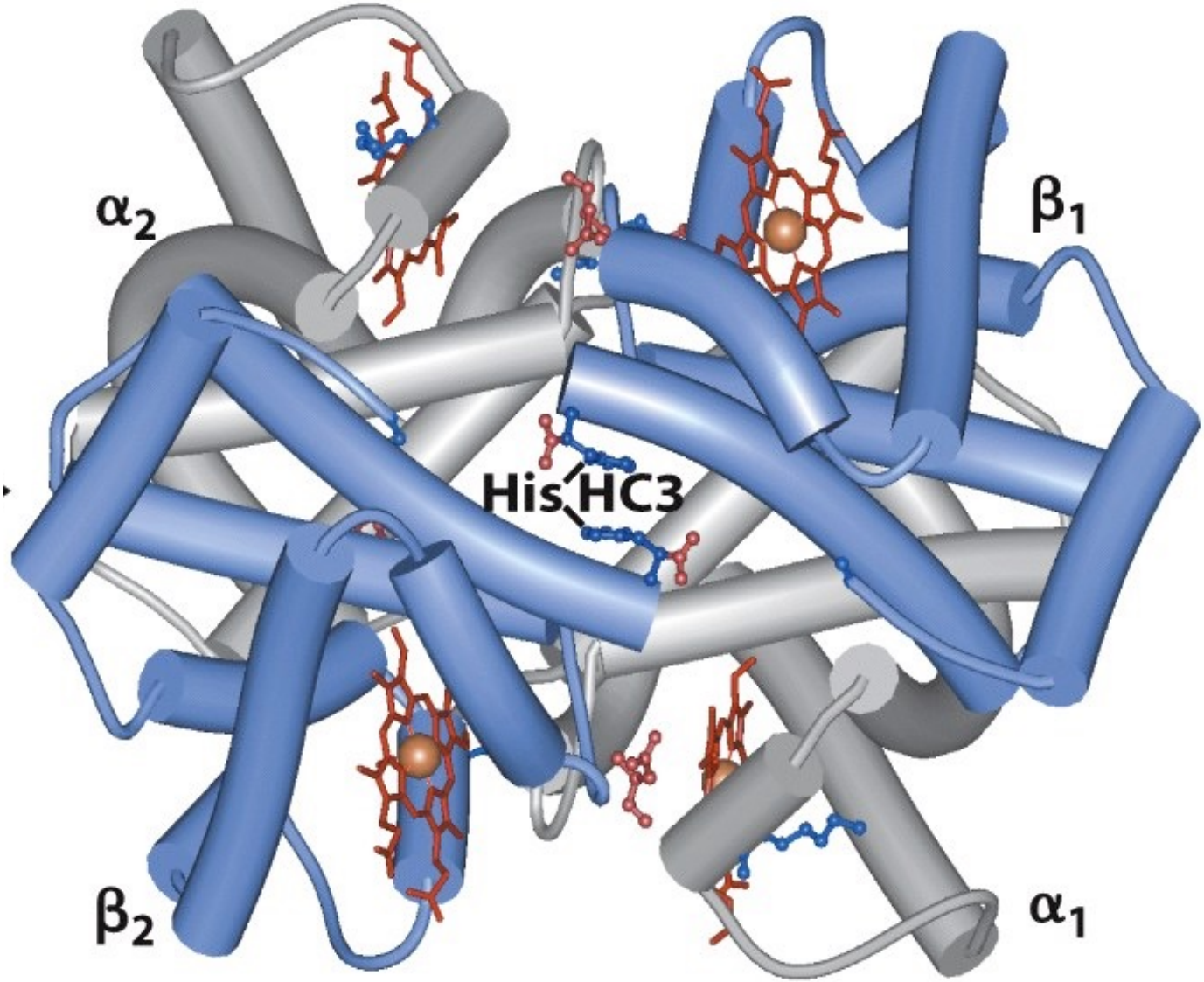


Figure 5-10

Comparison of Mb & Hb ß subunit

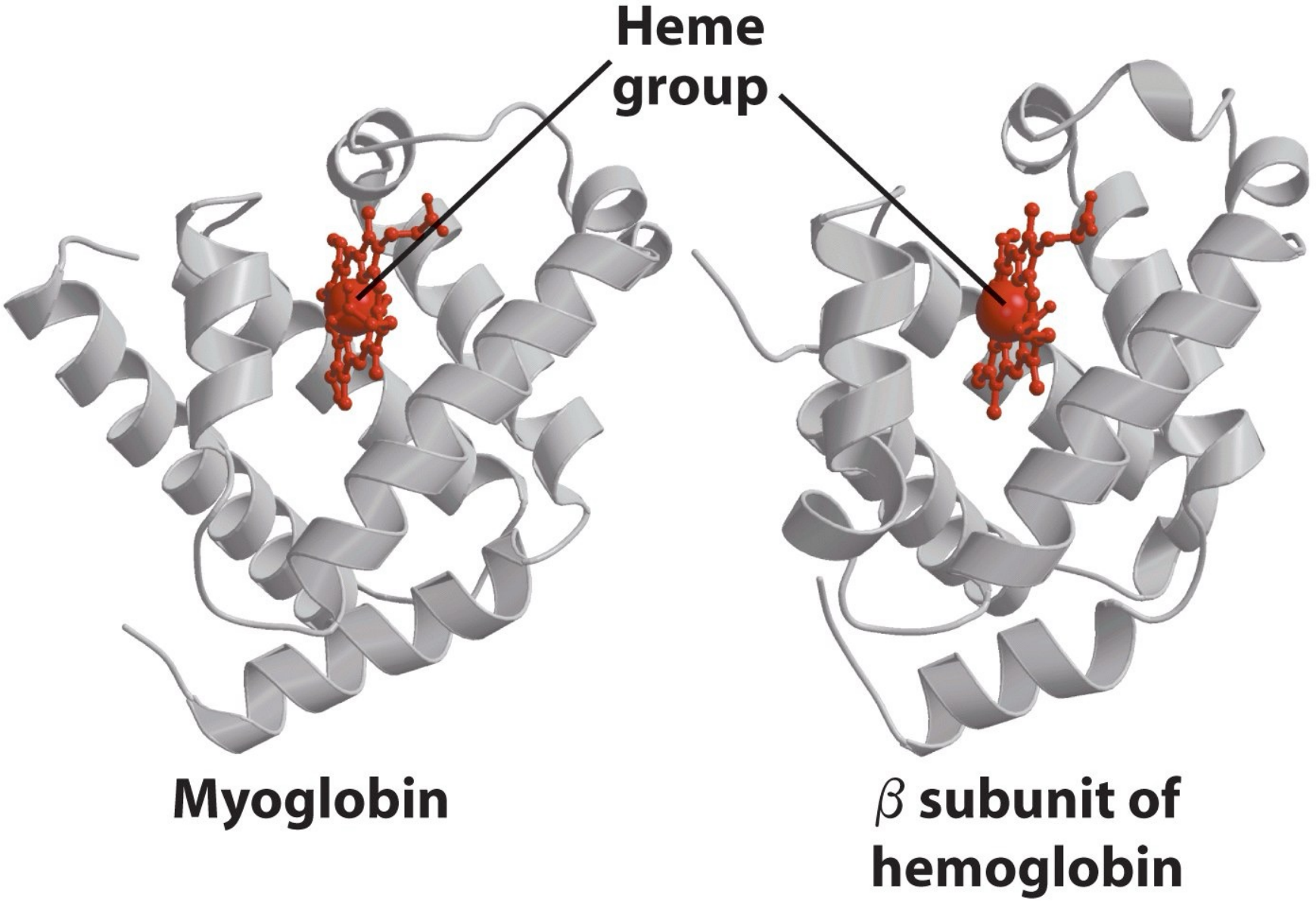
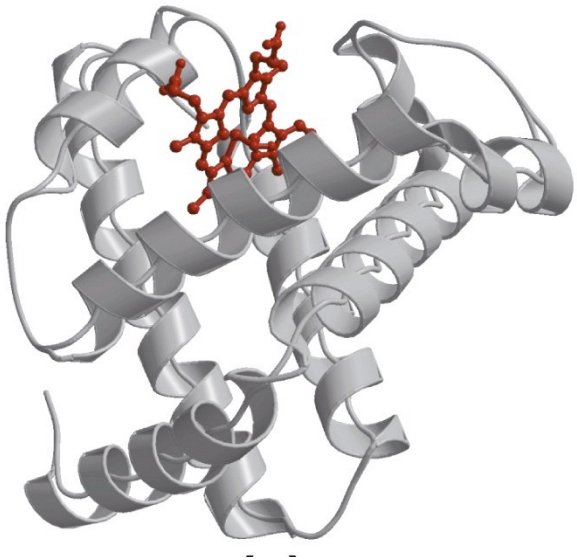
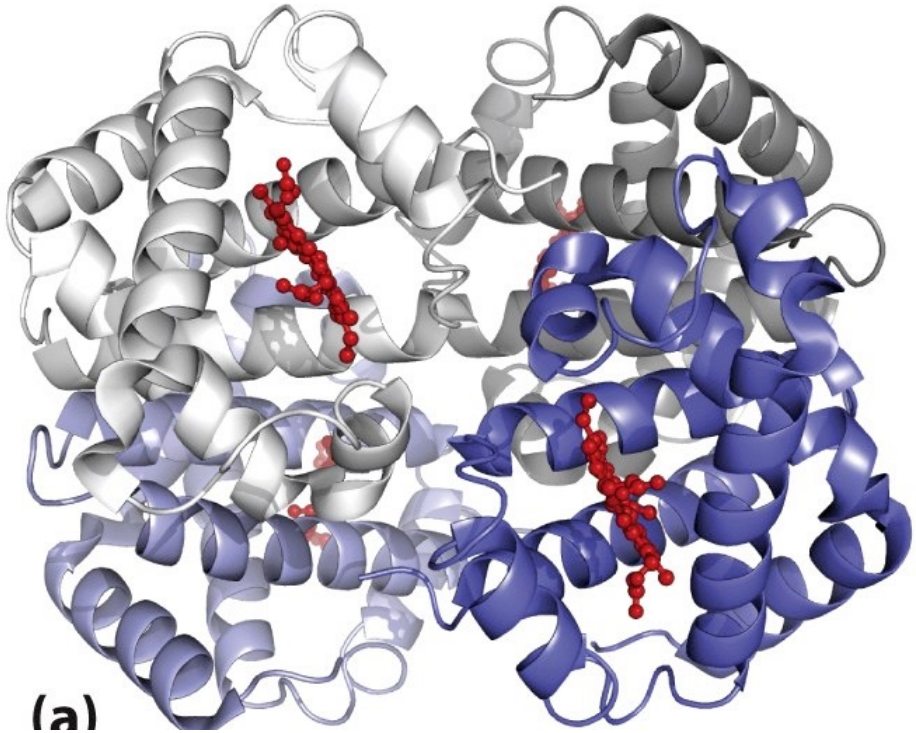


Figure 5-6

What is the difference between Mb and Hb?



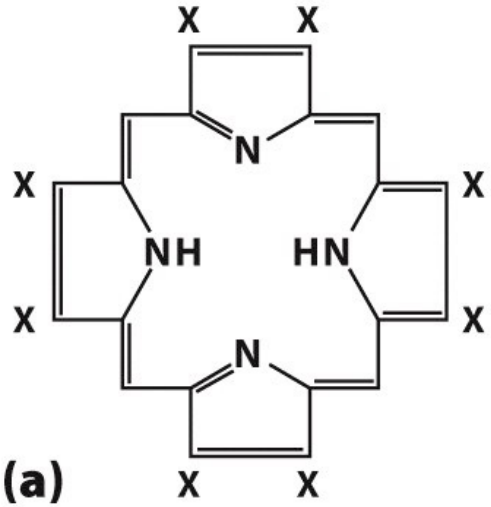
Myoglobin

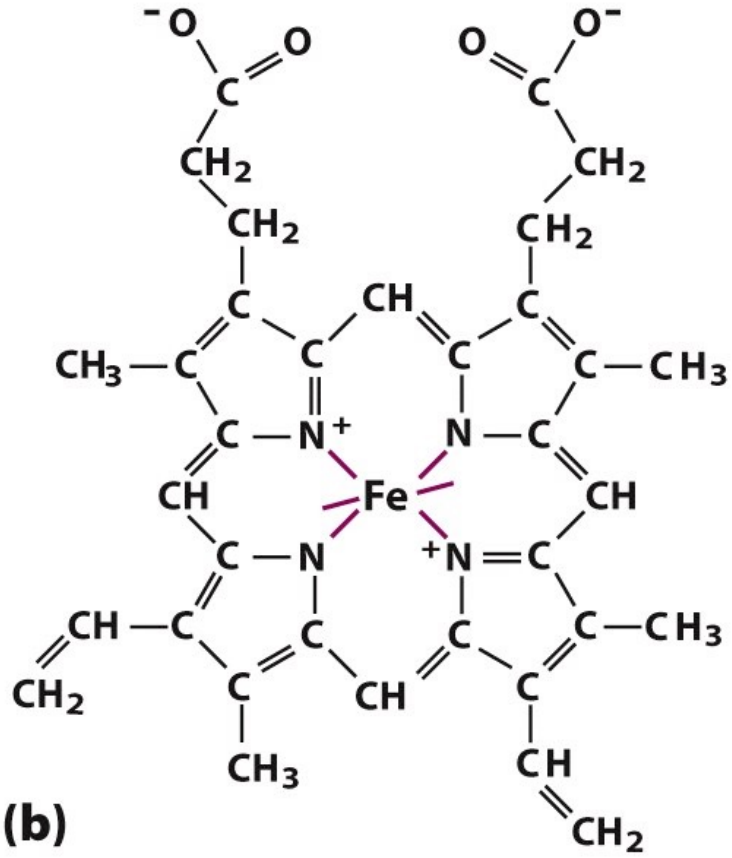


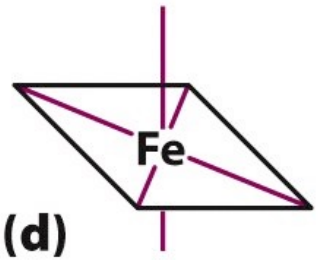
Hemoglobin

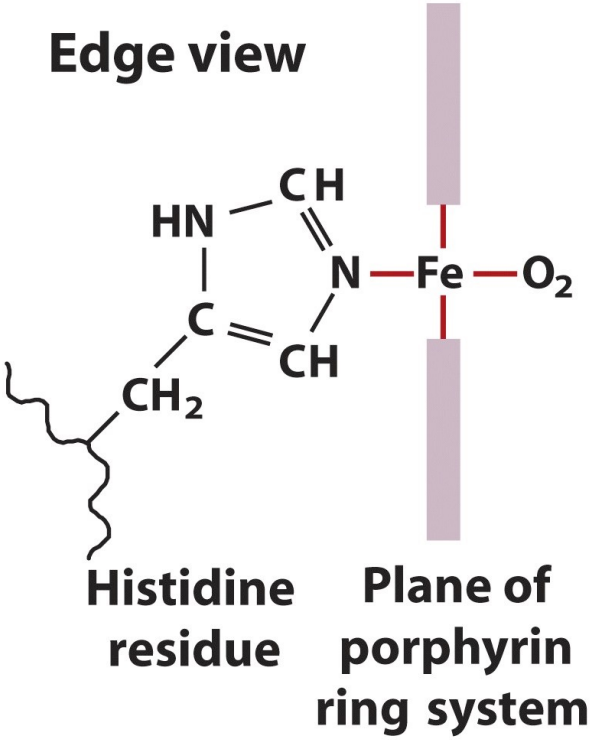
Need Hb to concentrate O_2 in our blood

Mb and Hb both bind O_2 via heme









Heme in myoglobin (similar in Hb)

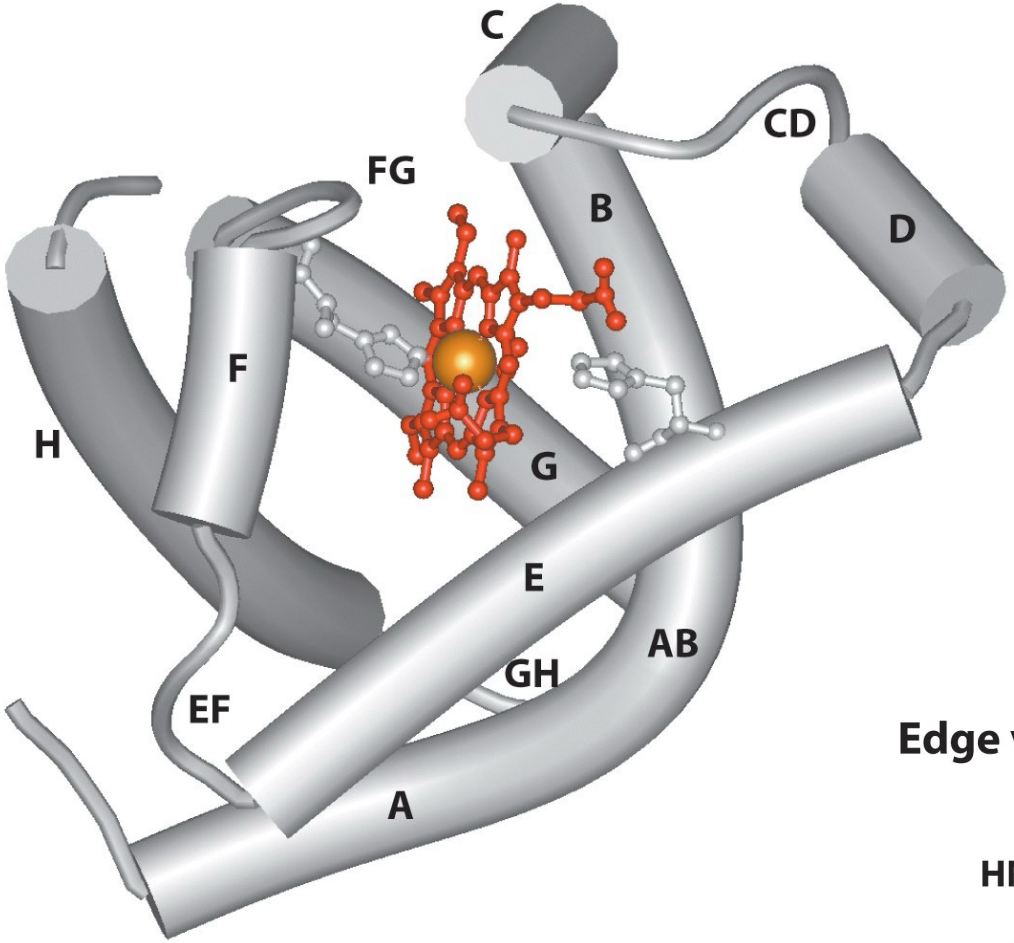
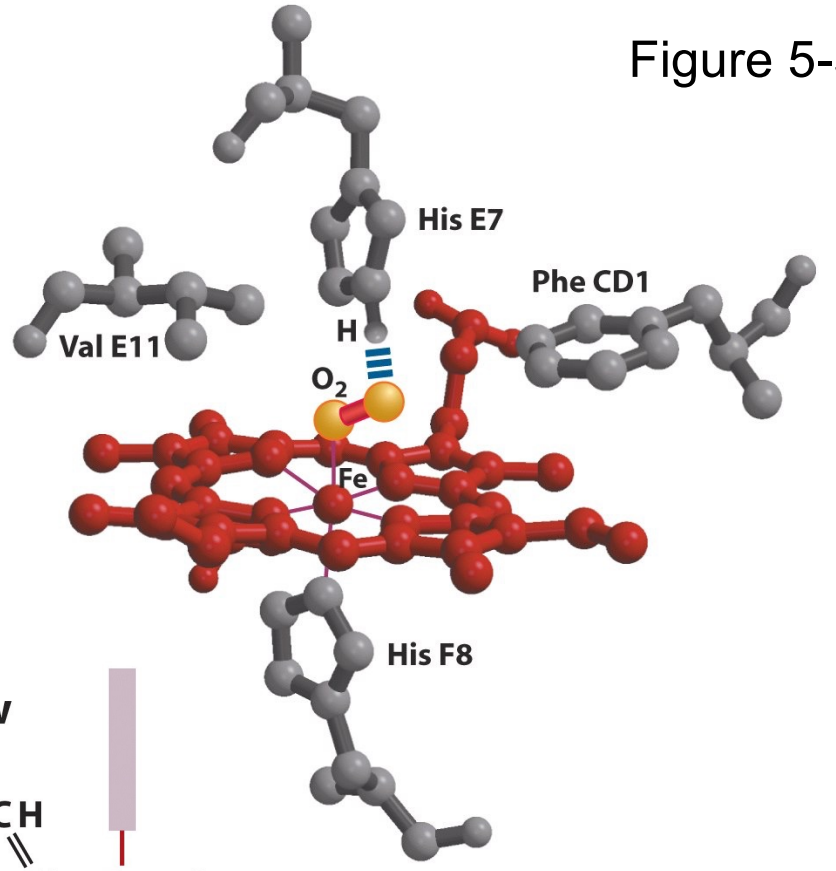
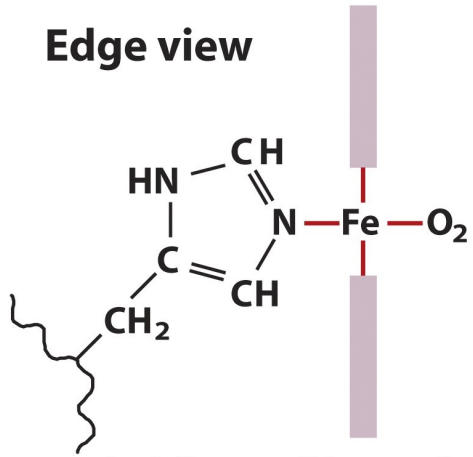


Figure 5-5



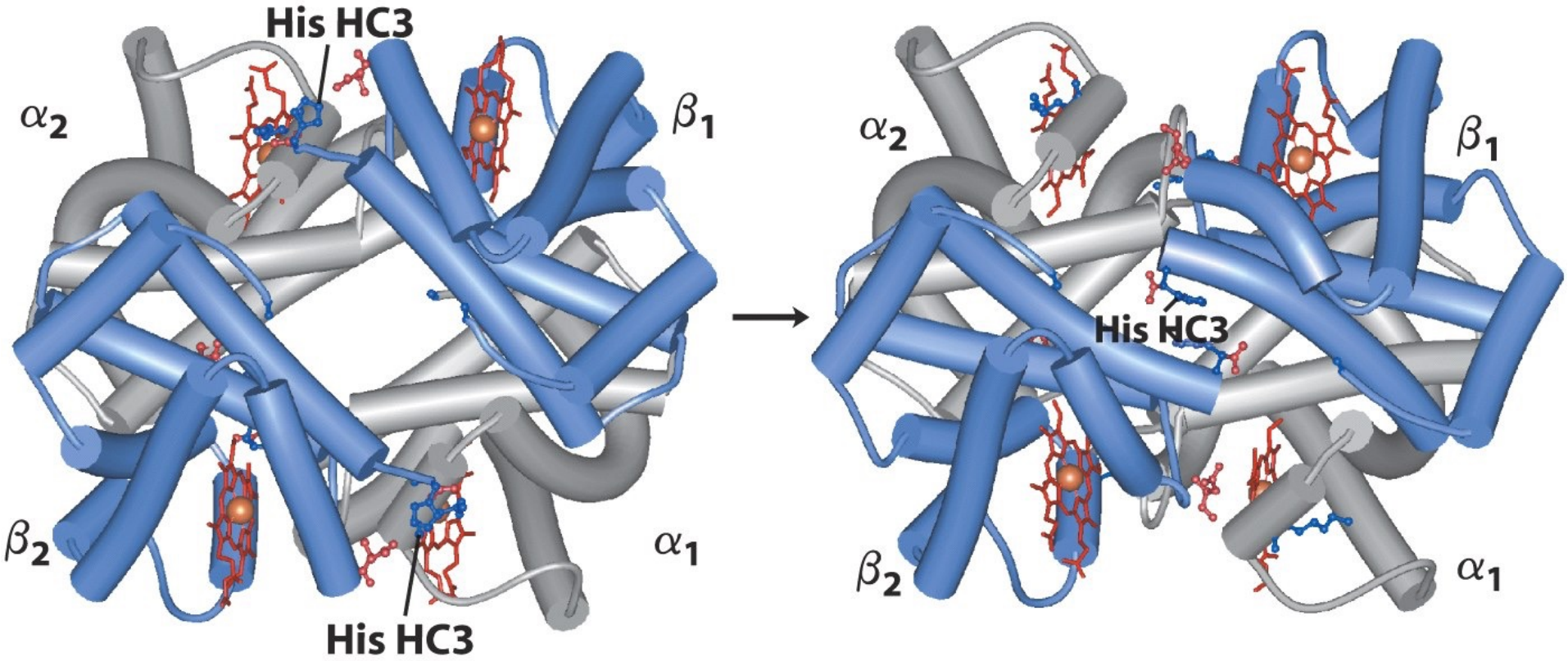


Histidine residue

Plane of porphyrin ring system

Figure 5-3

Mb does NOT undergo significant conformational change when it binds O_2, but Hb DOES!



Deoxy-Hb

Oxy-Hb

Tetrameric hemoglobin (dimer of dimers)

Packing contacts (horizontal) vs. sliding contacts (vertical)

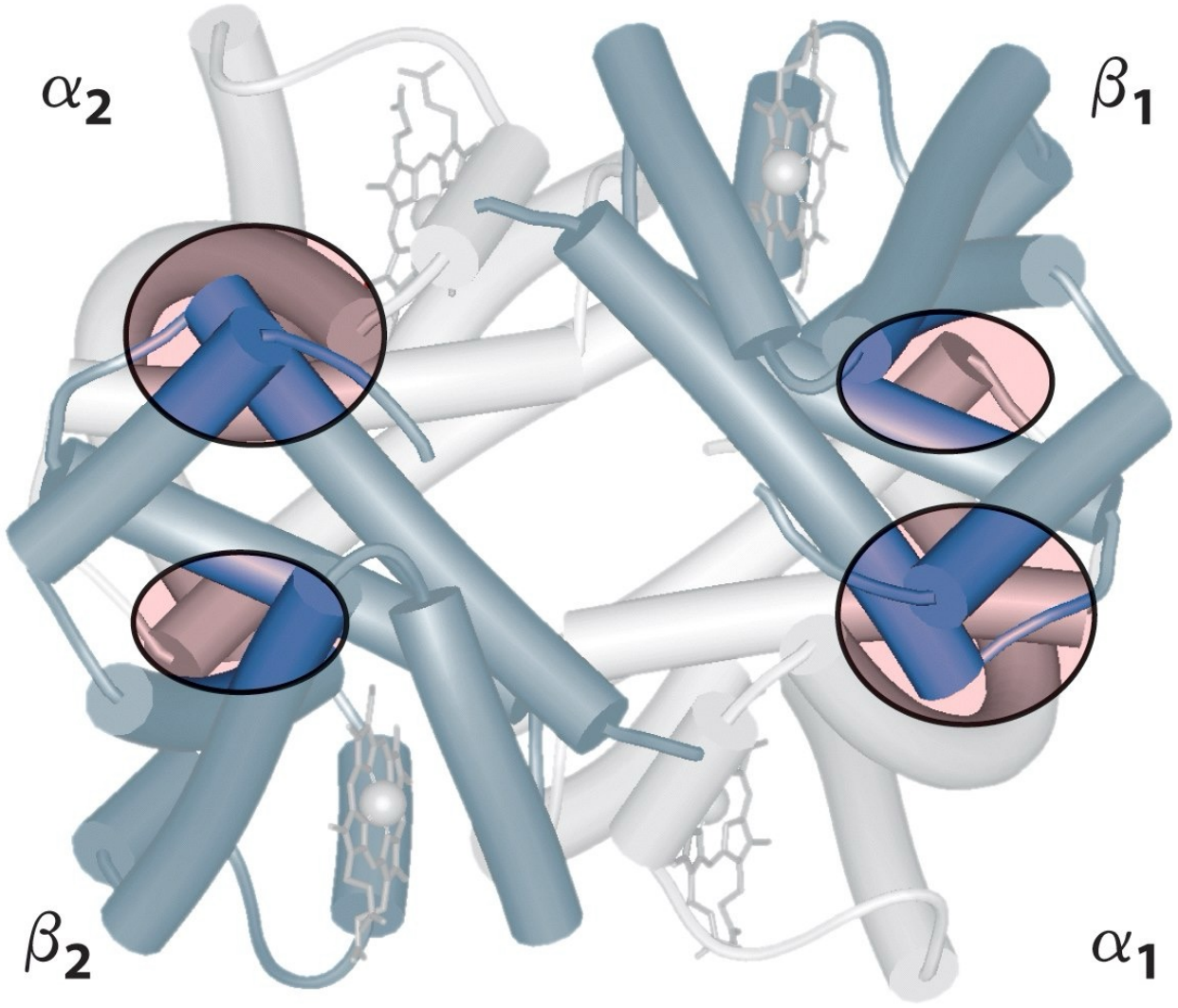
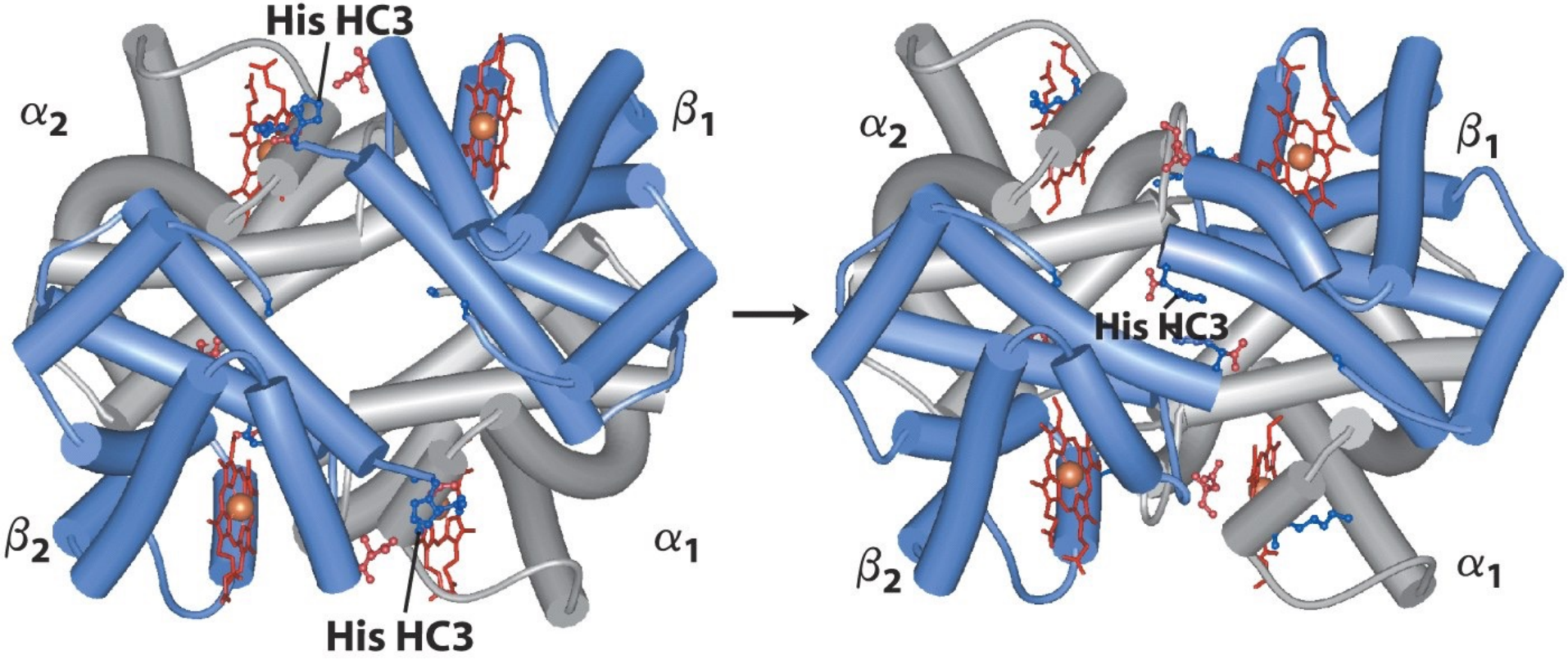


Figure 5-8

Conformational changes in Hb along sliding contacts upon O_2-binding



Deoxy-Hb
Low O_2 affinity

Oxy-Hb
High O_2 affinity

Figure 5-10

Heme has high affinity for CO (CO poisoning)

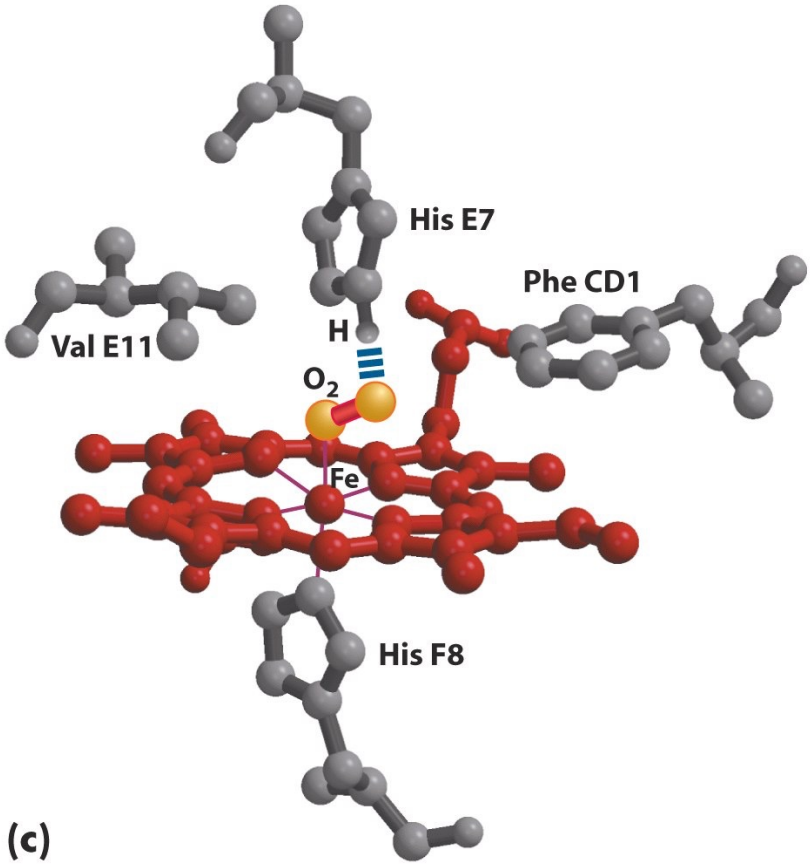


Figure 5-5

Hyperbolic O_2 binding curve for Mb

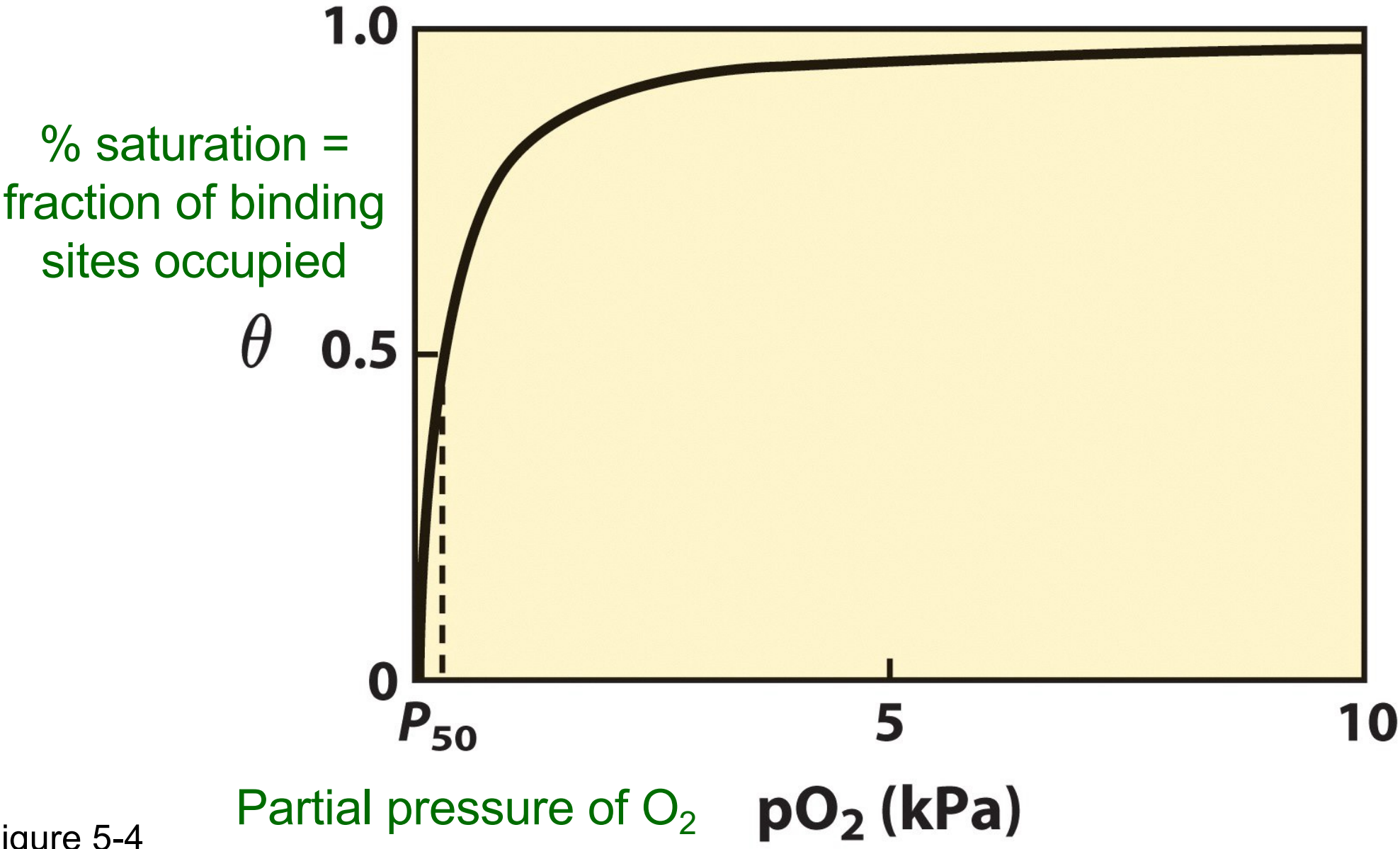


Figure 5-4

Sigmoidal O_2 binding curve for Hb

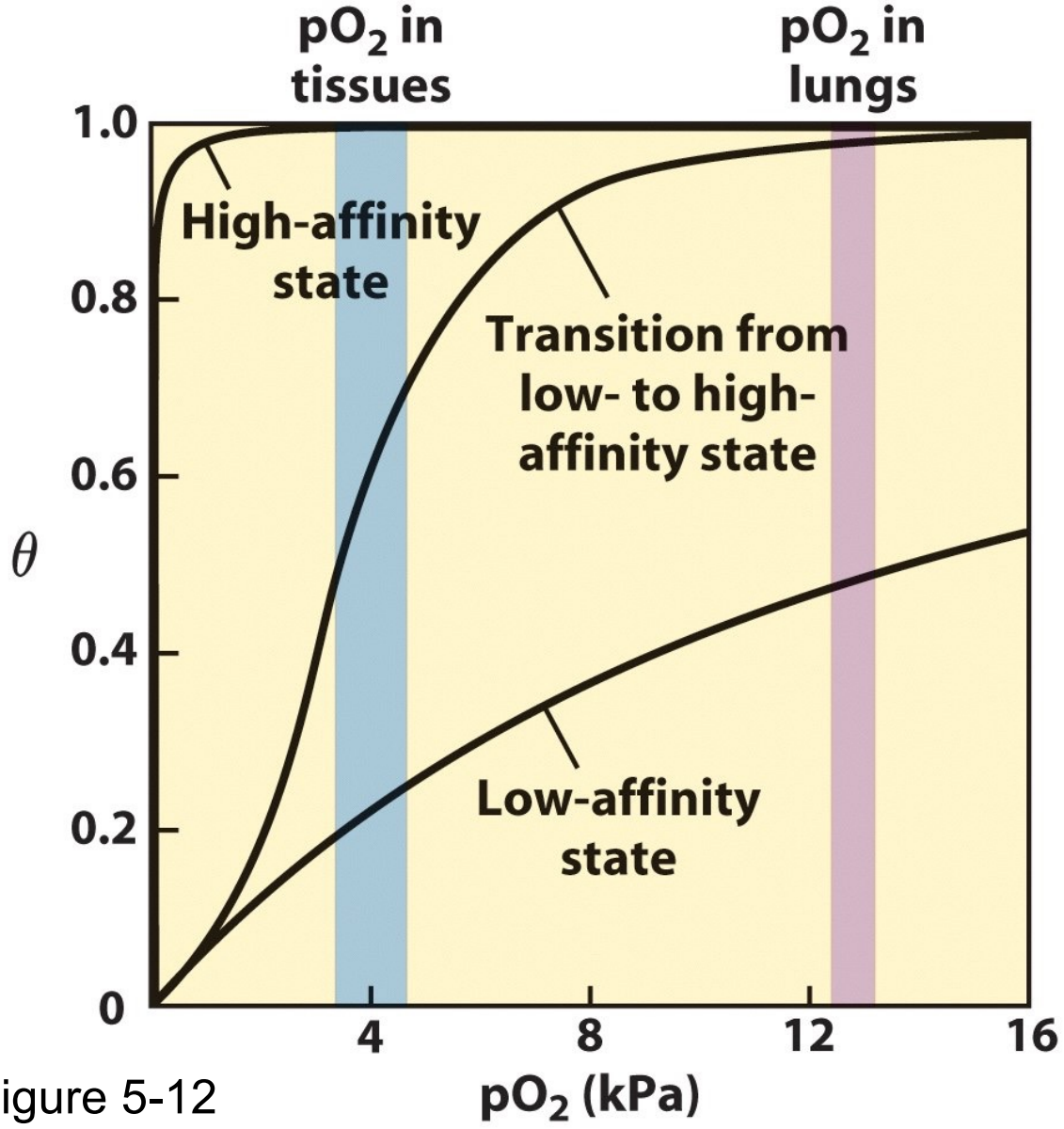


Figure 5-12

Sigmoidal curve == positive cooperativity

Binding of 1st O_2 is most difficult (low affinity)

Once 1st O_2 is bound, 2nd, 3rd and 4th O_2 bind with increasing ease (switch to hi affinity)

O_2 loading/unloading by Hb

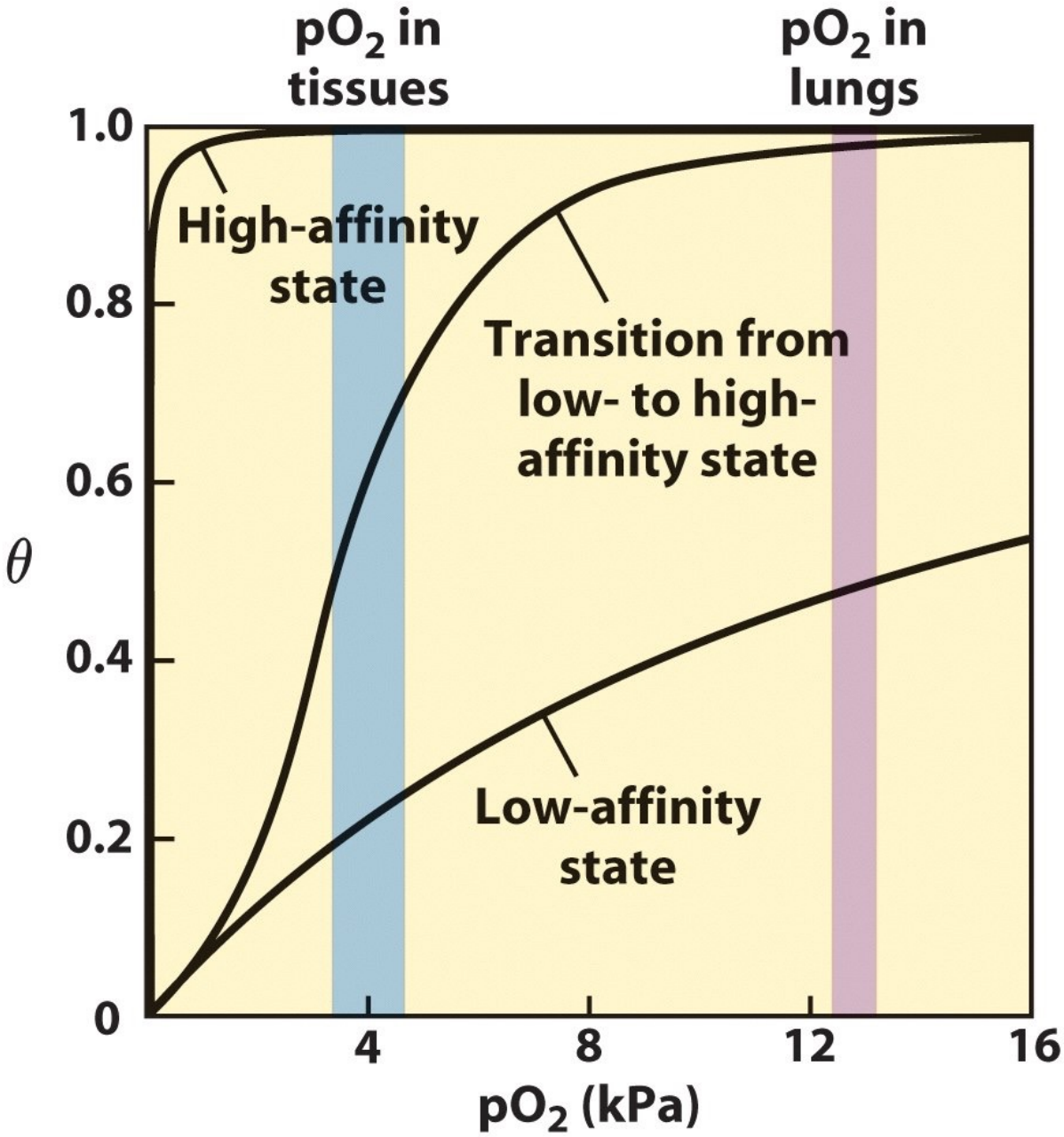


Figure 5-12

Mb cannot substitute for Hb

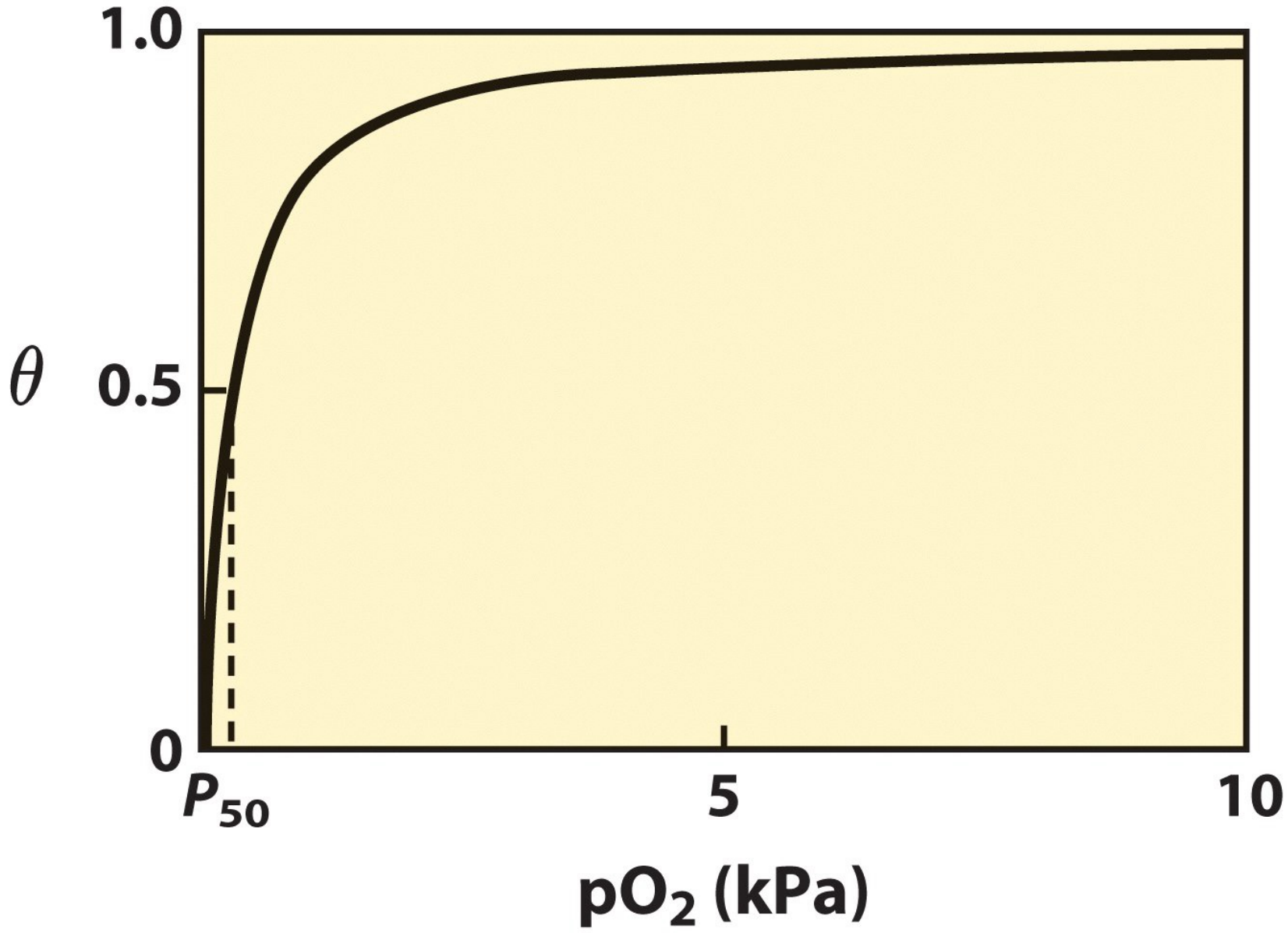


Figure 5-4

Mathematical equation for Hb O_2 binding curve

Hill's coefficient (n) – indicates cooperativity

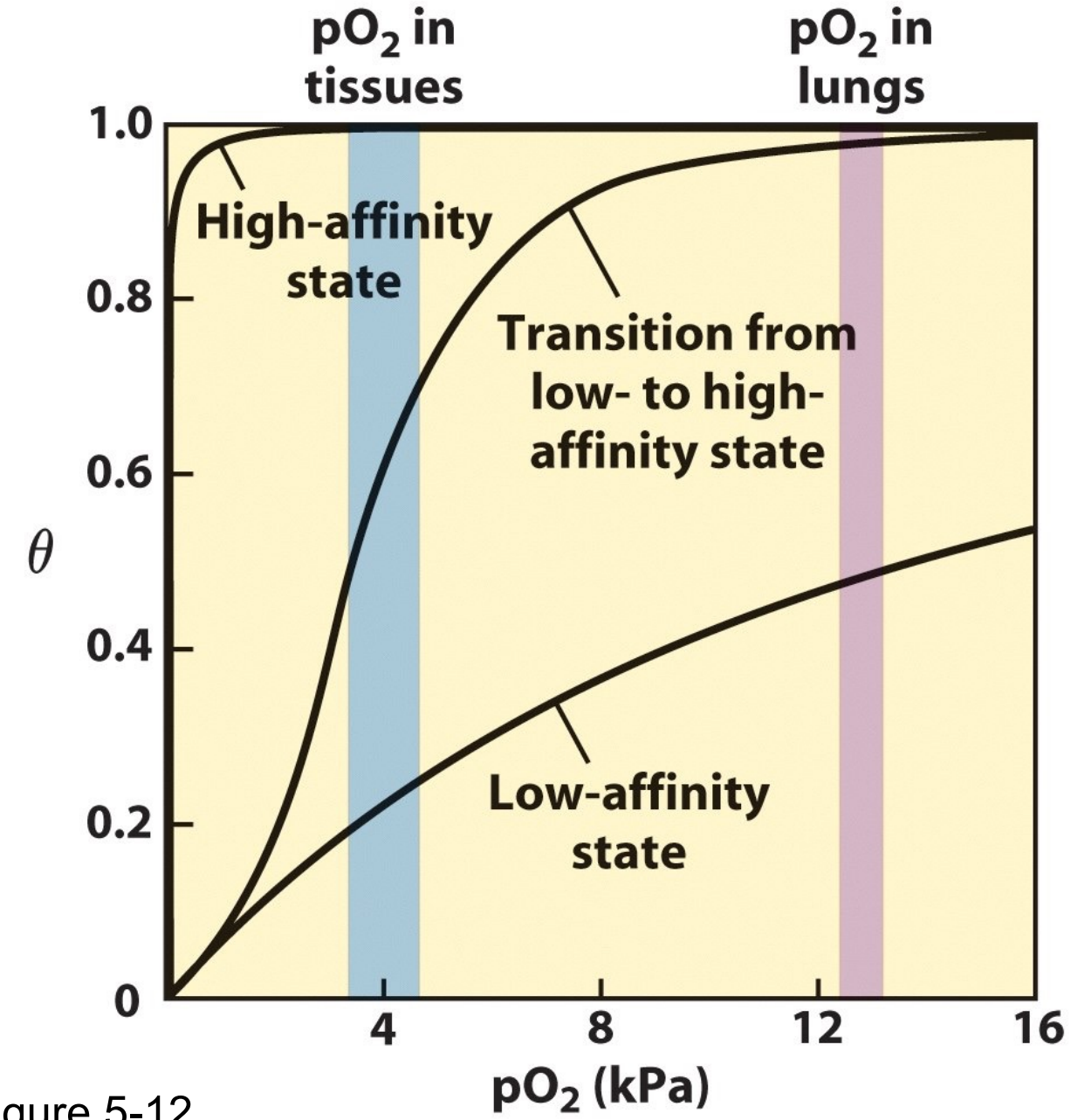


Figure 5-12

Conformational change upon O_2 binding

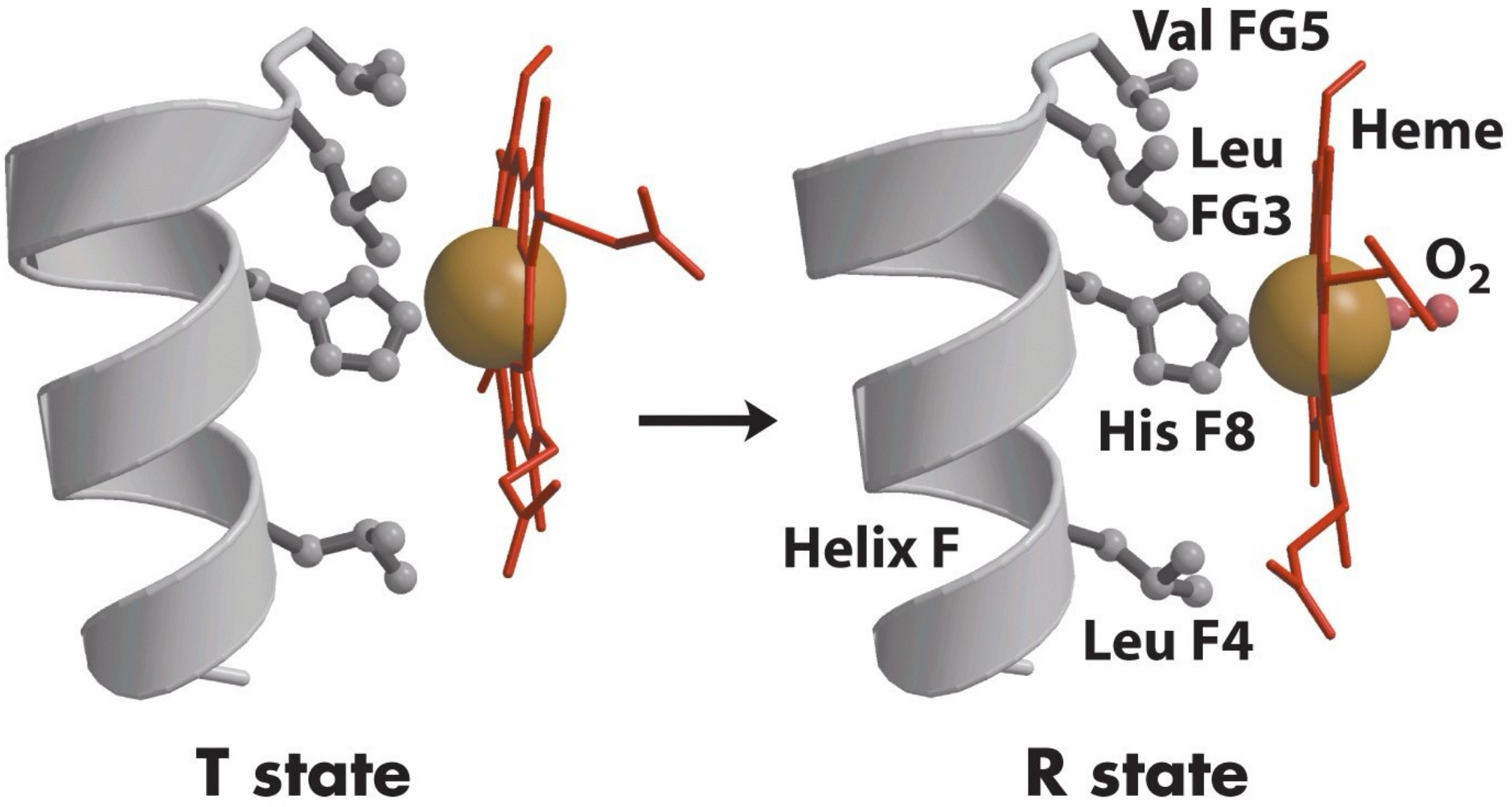
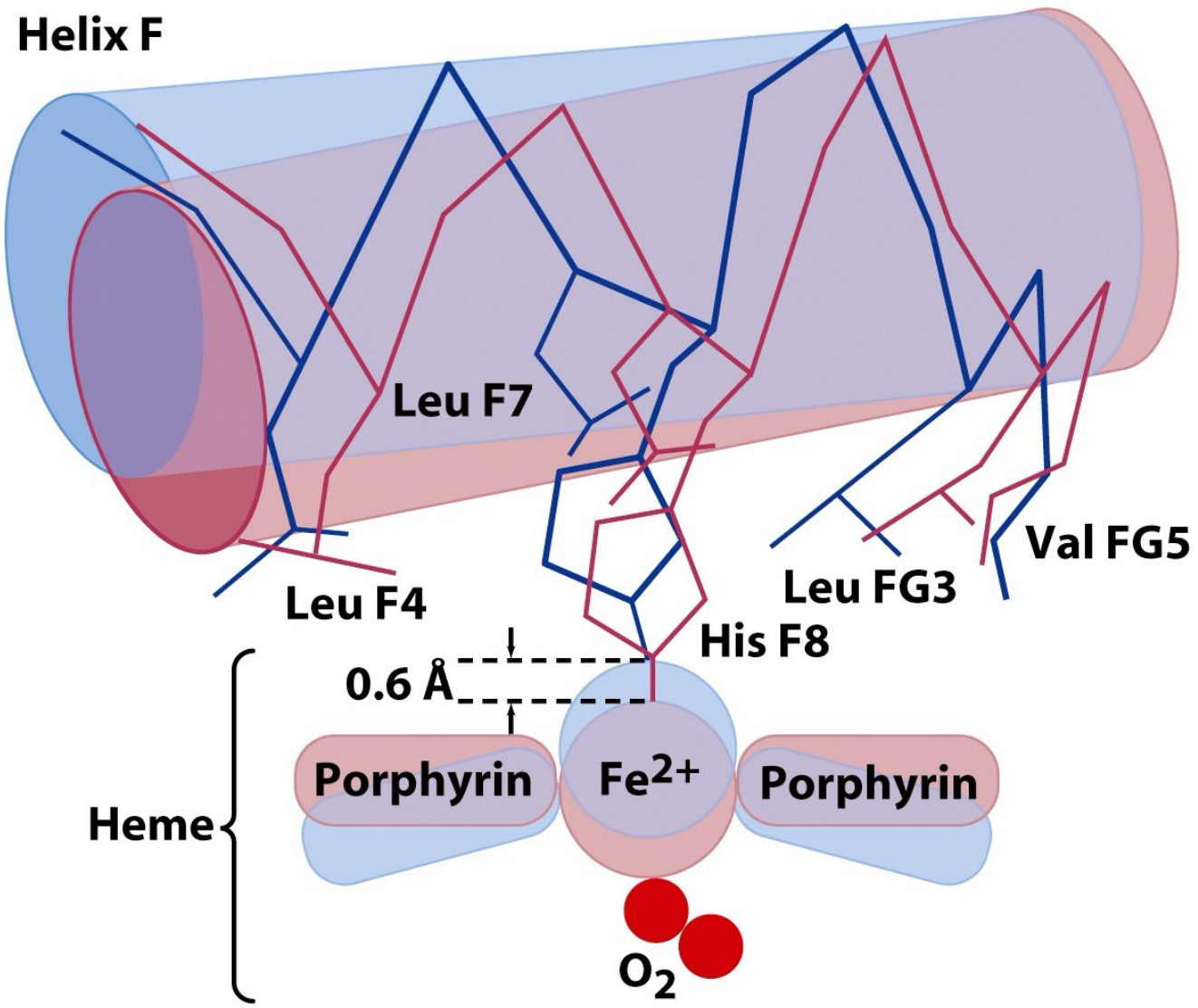


Figure 5-11

Conformational change upon oxygen binding



Small molecule effectors of O_2 binding

- CO_2, H^+ (i.e. pH) and 2,3-bisphosphoglycerate (BPG)
- When these molecules bind to Hb, they preferentially stabilize **deoxy-Hb** conformation, so they **decrease** affinity for O_2 **(allosteric inhibitors).**
- These small ligands bind, not at the heme, but at a different (characteristic) site on Hb.

Bohr effect (pH)

Important for delivery of O_2 to actively metabolizing tissues

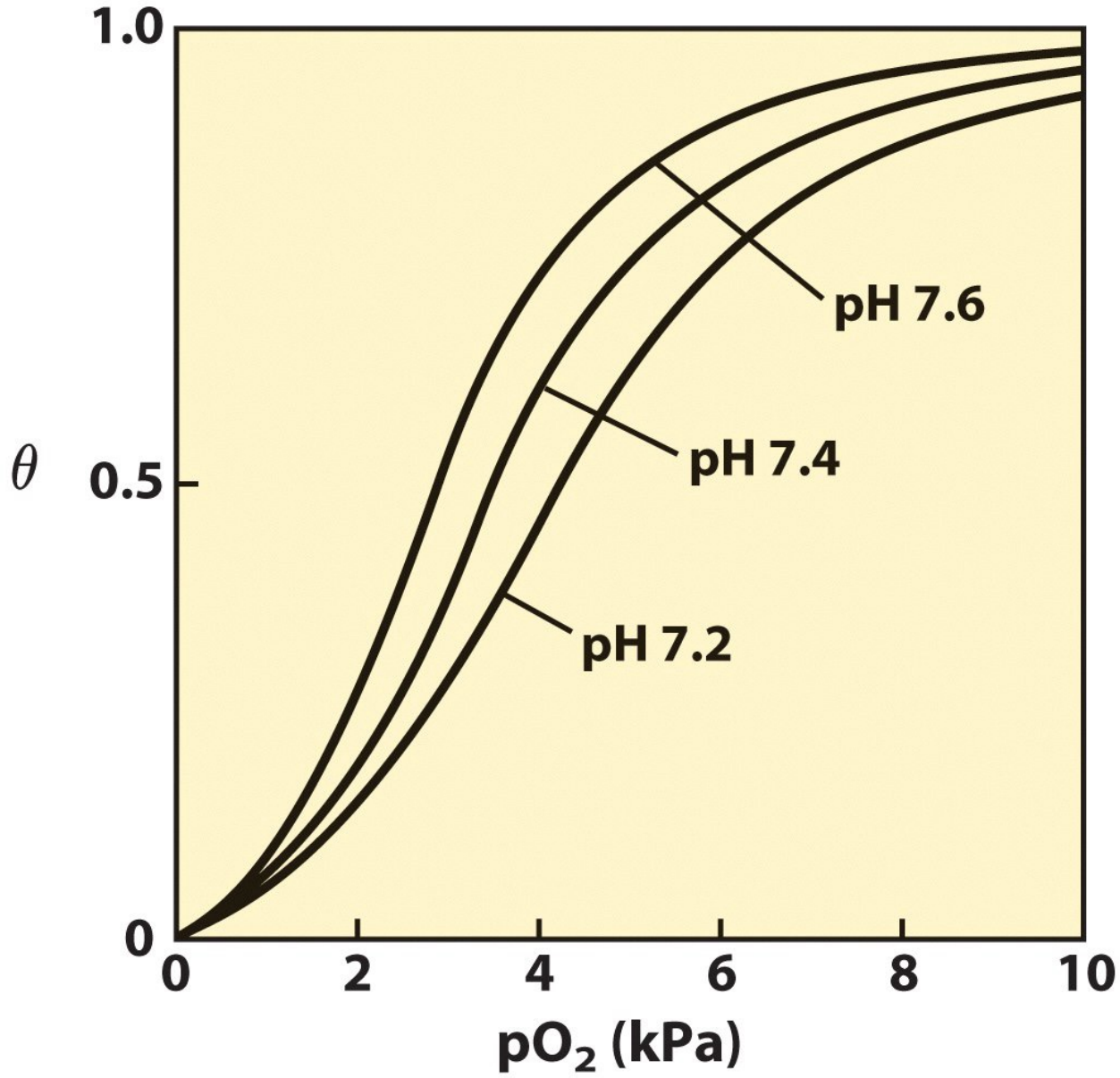


Figure 5-16

BPG stabilizes deoxy-hemoglobin

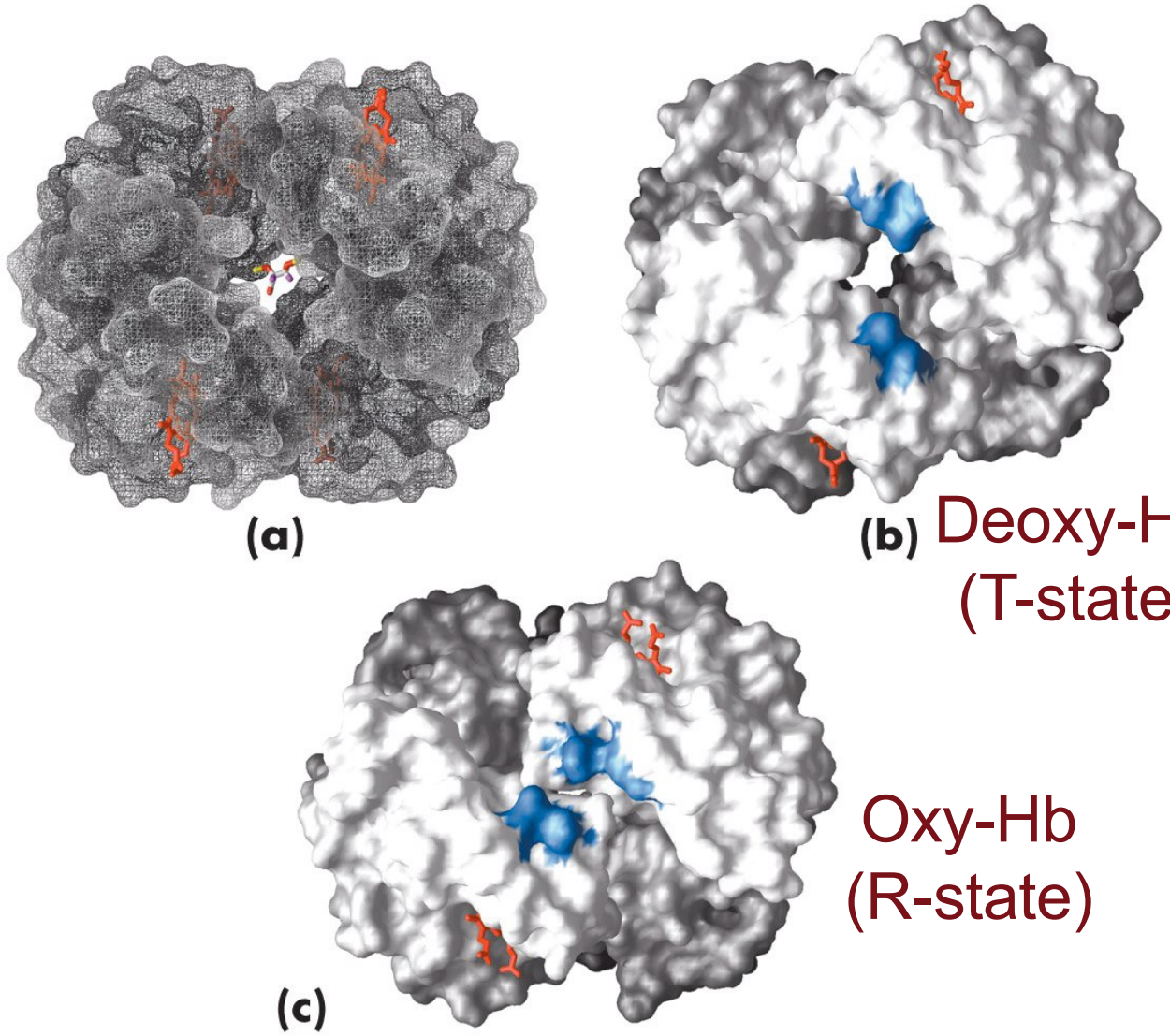


Figure 5-18

Effect of BPG on O_2 binding

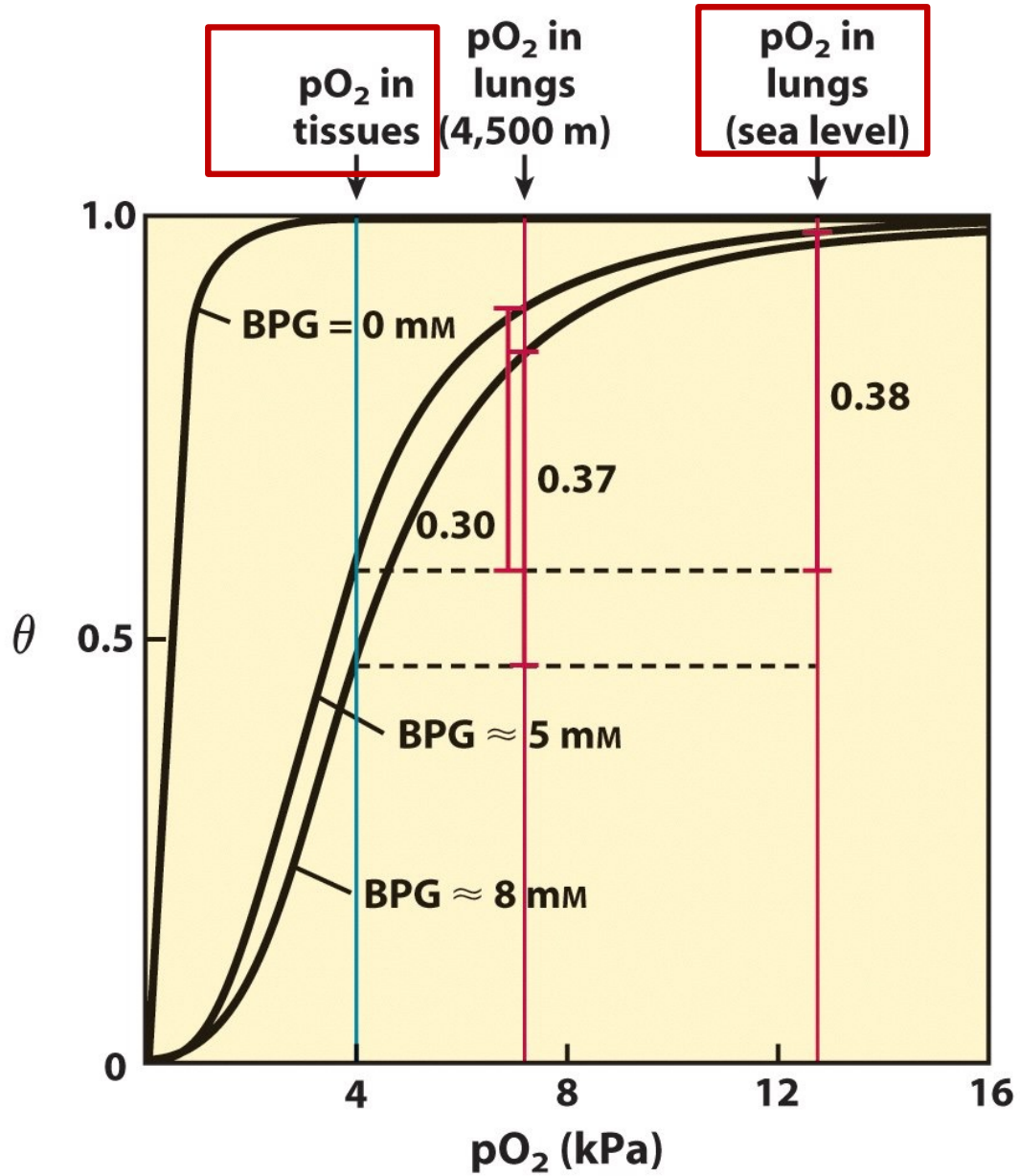
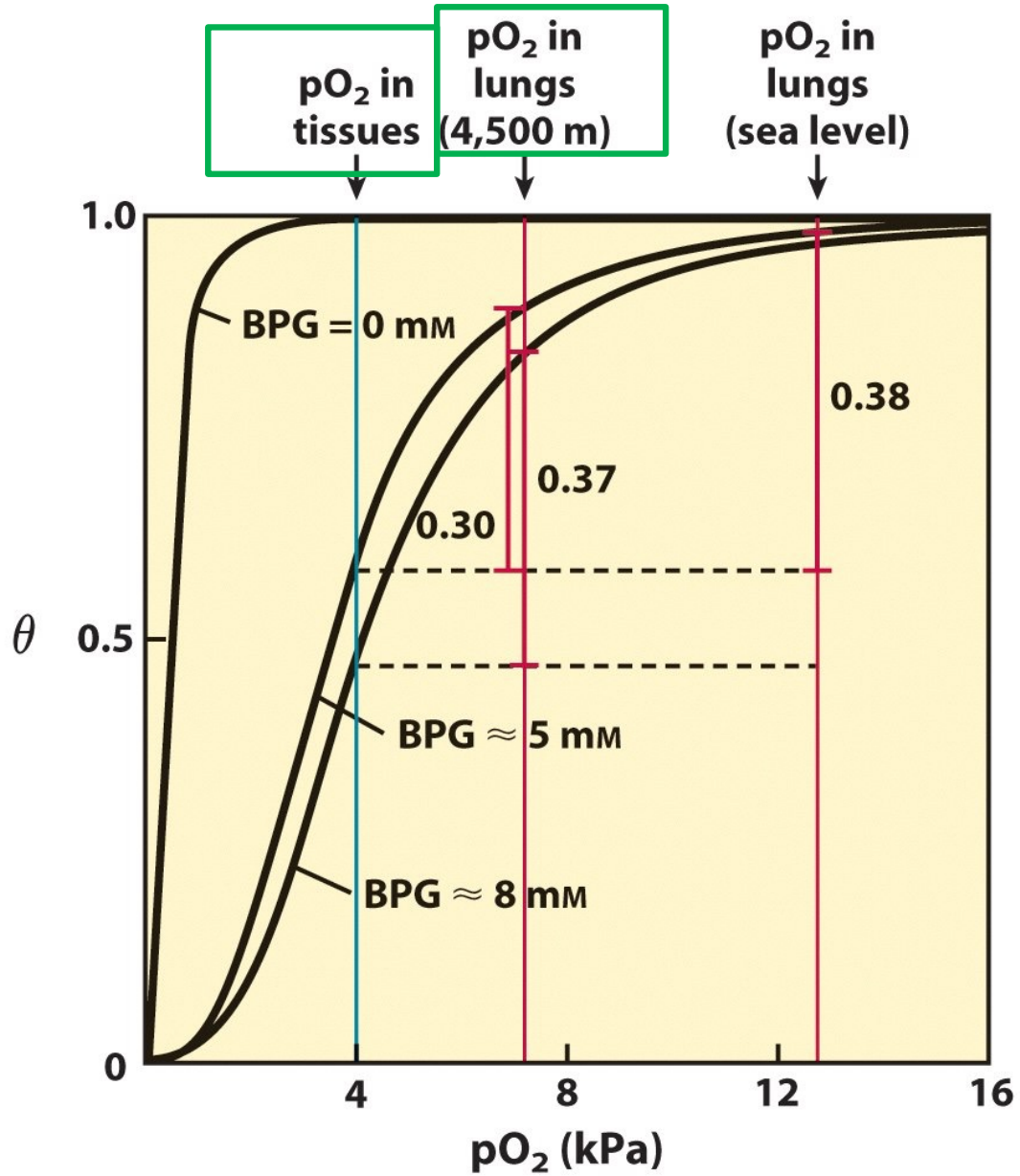


Figure 5-17

BPG and high altitude adaptation



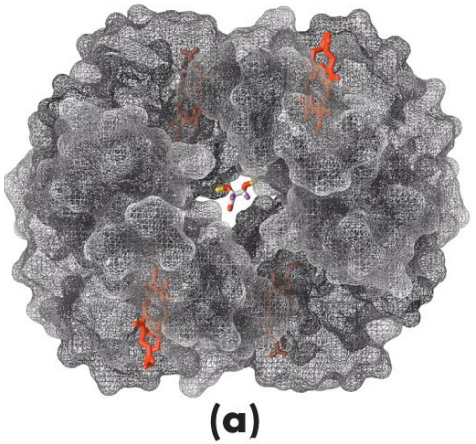
High altitude adaptation – increase [BPG] to 8 mM

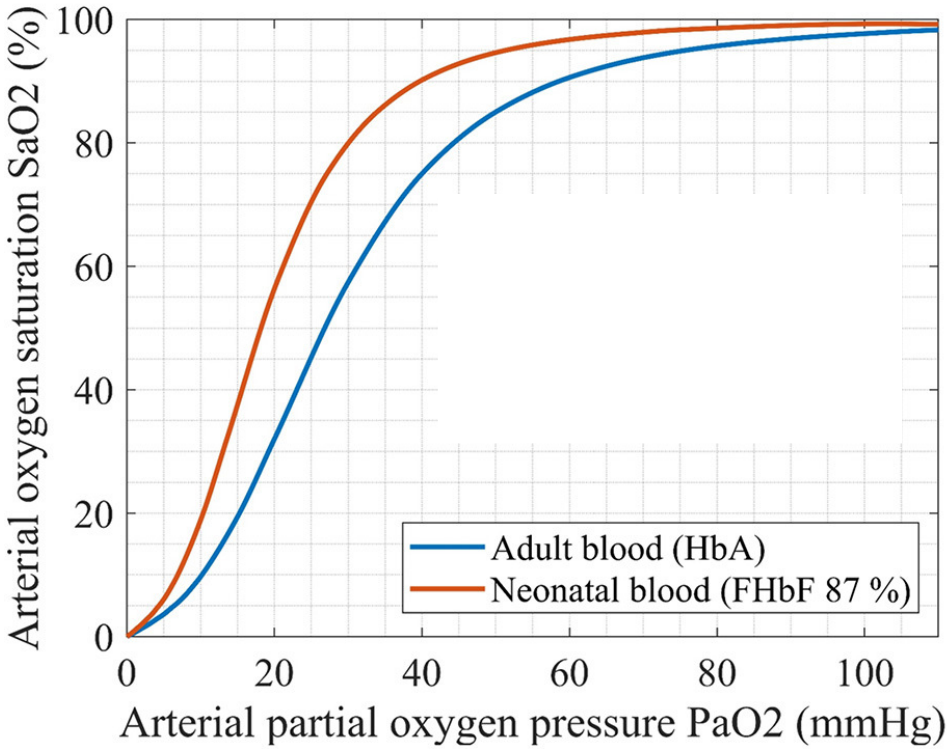
Figure 5-17

BPG and fetal development

- Fetal Hb (α2γ2) binds BPG with lower affinity than adult Hb (α2β2) – so binds O_2 with higher affinity

Adult Hb β subunit has His143 in central cavity
Fetal Hb γ subunit has Ser143 instead





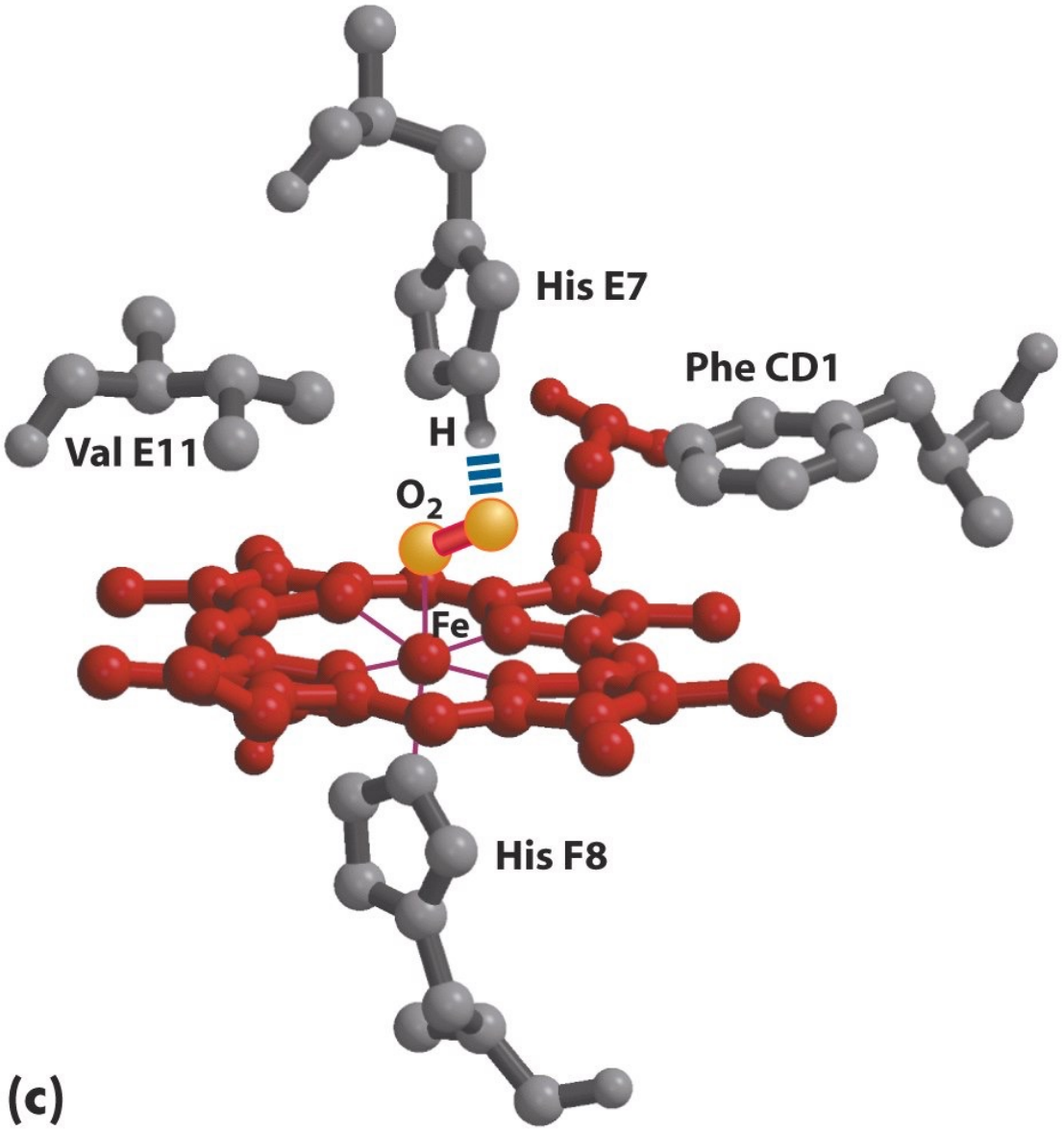
Mutant Hbs

Three types of defects:

- Change in surface residues (ex. HbE [GluB8(26)ß->Lys])

- Change in internal residues (ex. Hb Hammersmith)
- Change along α1-β2 sliding contacts (ex. Hb Kansas)

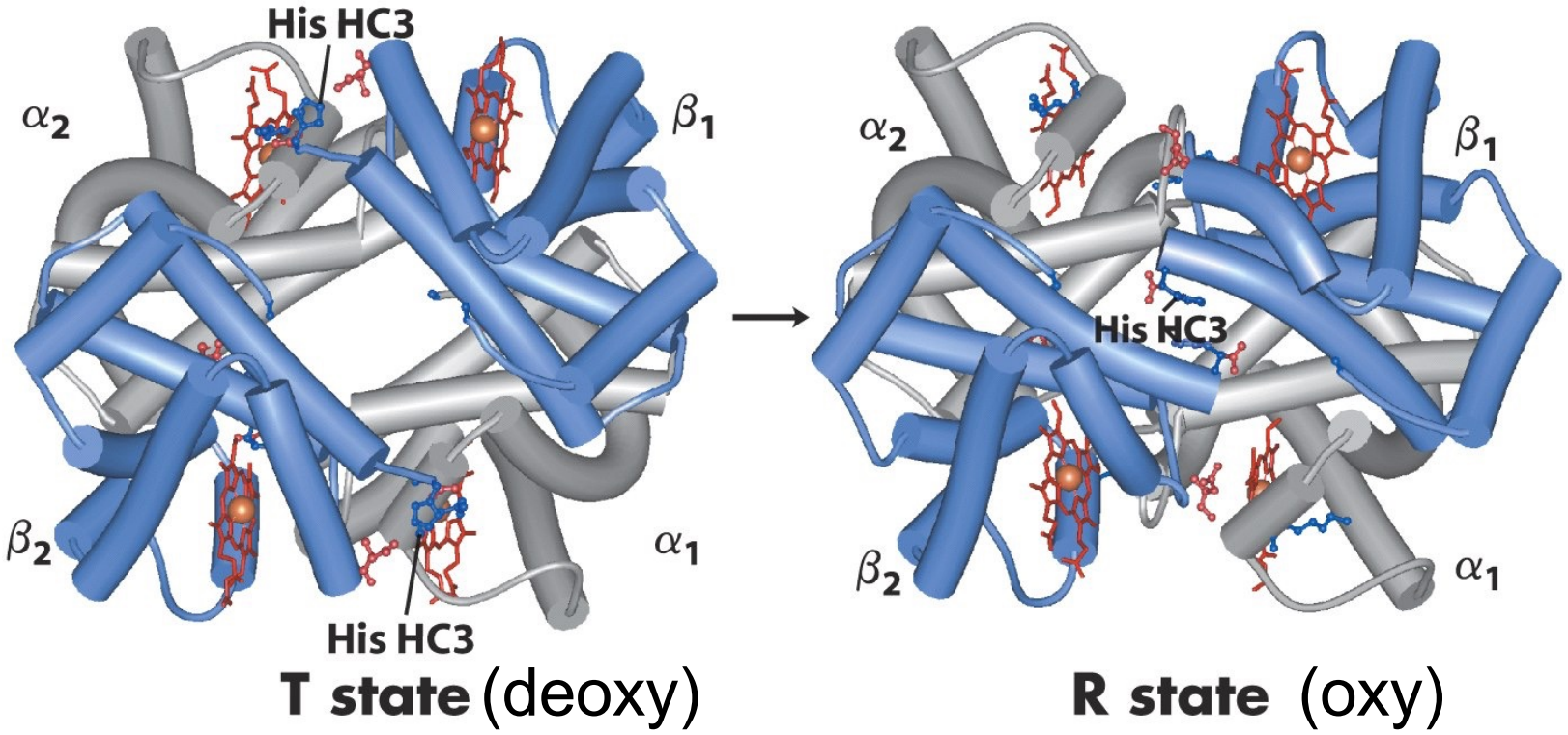
Hb hammersmith
Phe CD1 (42) ß --> Ser



PheCD1 and HisF8 are the only 2 invariant residues in all known Hbs.

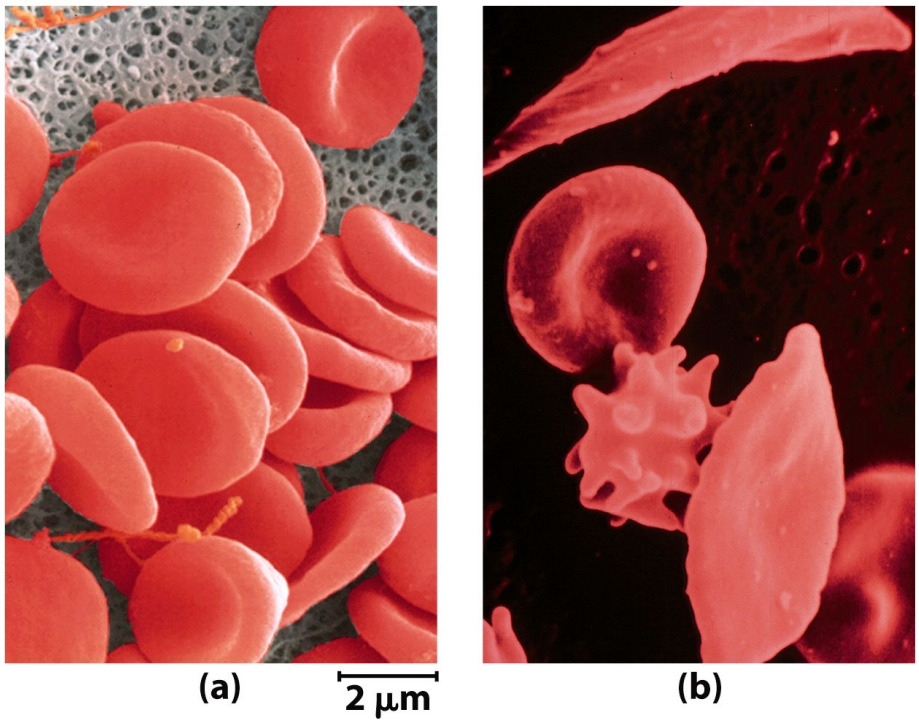
Figure 5-5

Hb Kansas (altered sliding contacts)
Asn G4 (102) ß --> Thr



Eliminates H-bond that stabilizes R-state. Mutant preferentially stays in T-state (low O_2 affinity; n=1.3)

Sickle cell anemia
HbS [Glu(6)ß --> Val]



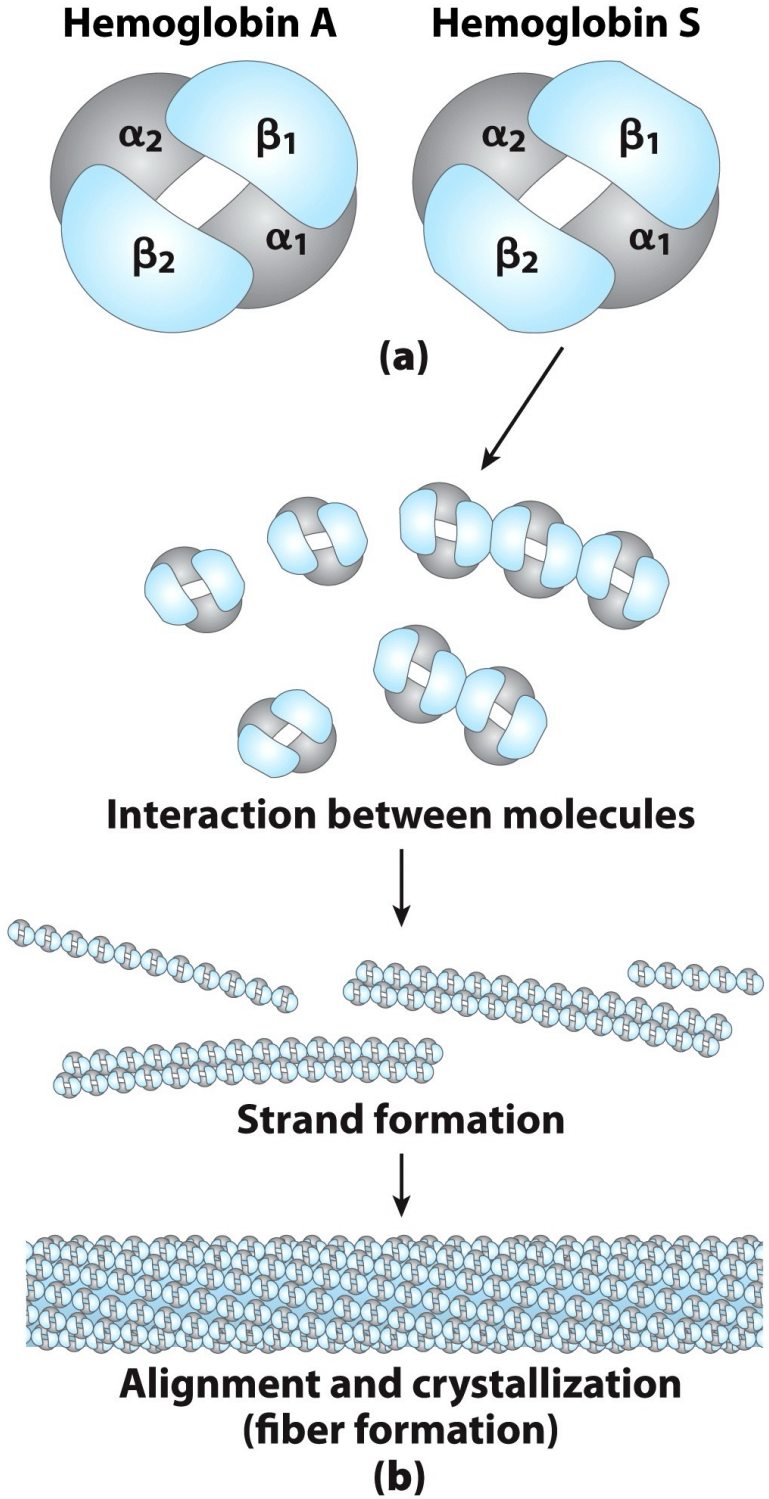


Figure 5-19 & 5-20

HbS heterozygotes are resistant to malaria

- Intracellular parasite lives in RBCs
- Lowers RBC pH by 0.4 units which favors deoxy-Hb (Bohr's effect)
- Increased sickling of RBCs with Hb^s which are removed by spleen early in infection
- Parasite eliminated!

***Plasmodium falciparum* infects red blood cells**