

SB18

Sickle-Cell Disease

A Close Call for a Boy Scout

The Patient Is Admitted with a Painful Crisis

- A 14-year-old African American boy known to have sickle-cell disease was admitted to the hospital.
- Two days before admission, he was hiking with his scout troop but had no symptoms at that time.
- The day prior to admission he developed lower back pain and left hip pain.



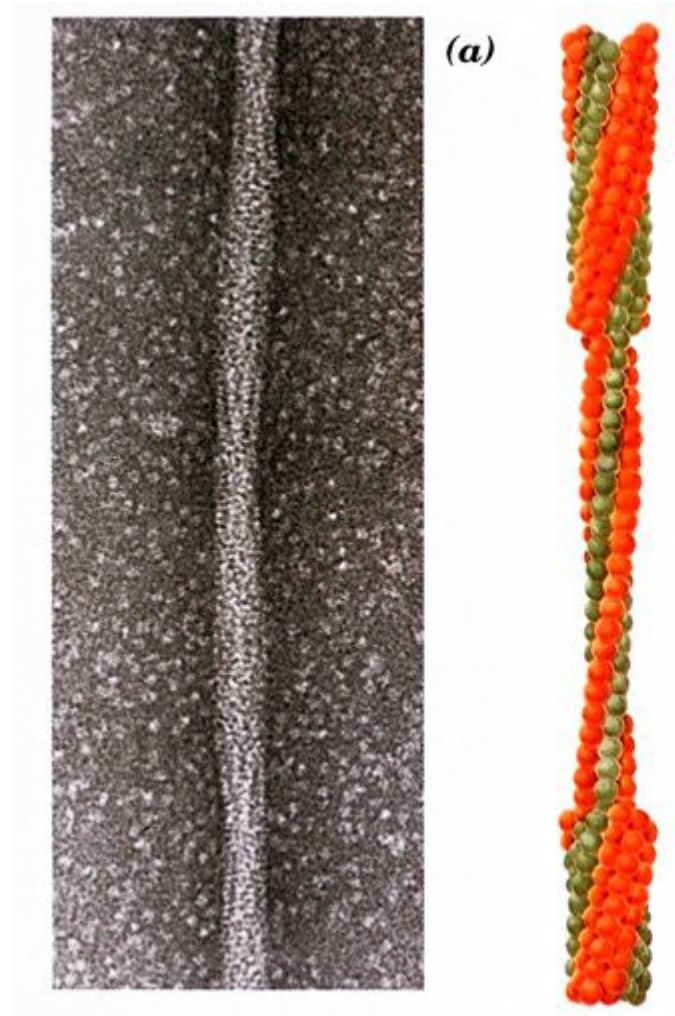
(a)



(b)

- The vaso-occlusive events common in those with sickle-cell disease cause “painful crises,”
- the most distressing symptom of the illness.
- Vaso-occlusion occurs due to fibers of hemoglobin within sickled red blood cells.

- The double strand of sickled hemoglobin serves as a building block for higher-order structures.
- The fibers within sickled cells actually consist of seven pairs of strands



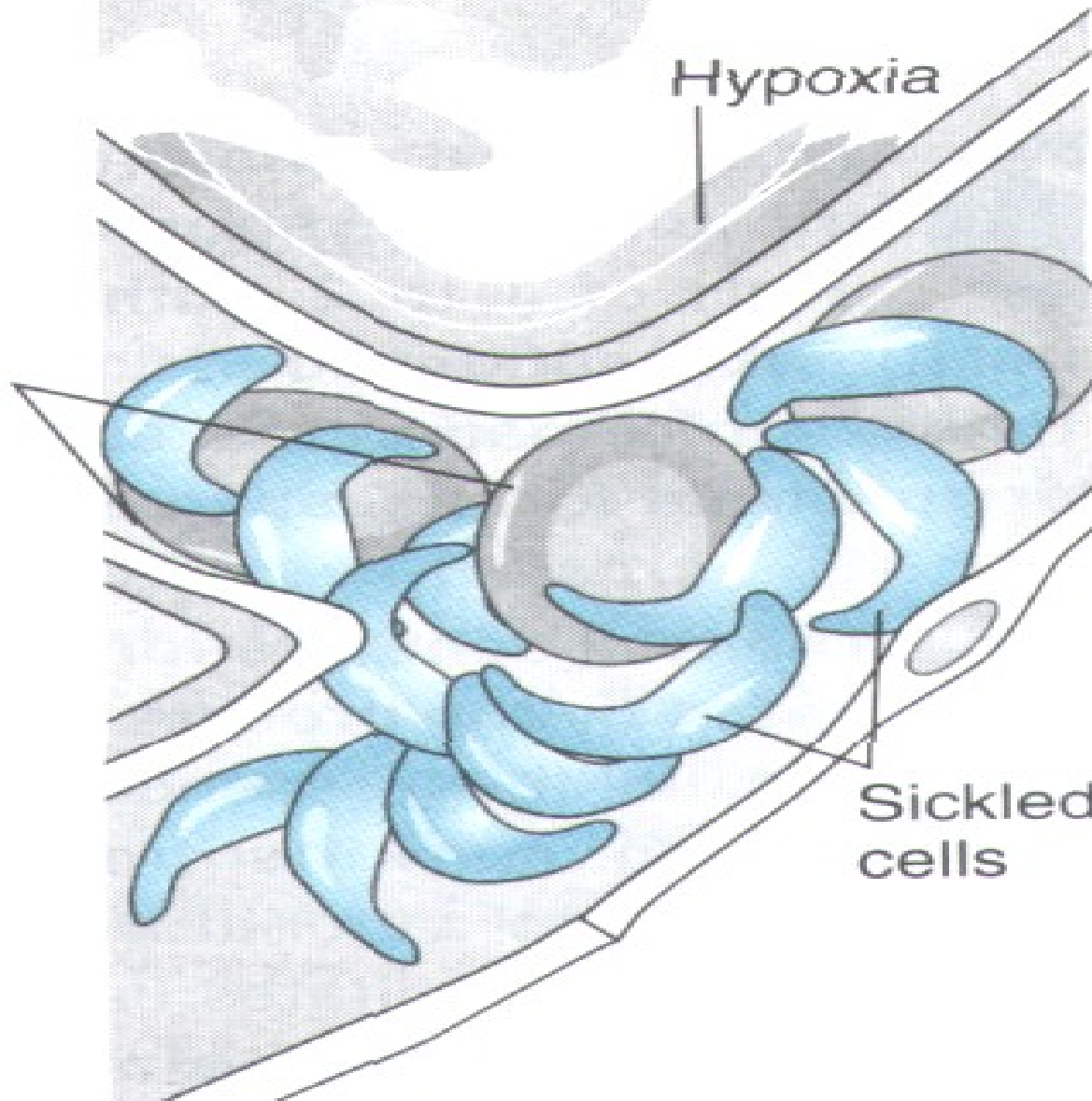
Vascular
occlusion

Necrosis

Normal
tissue

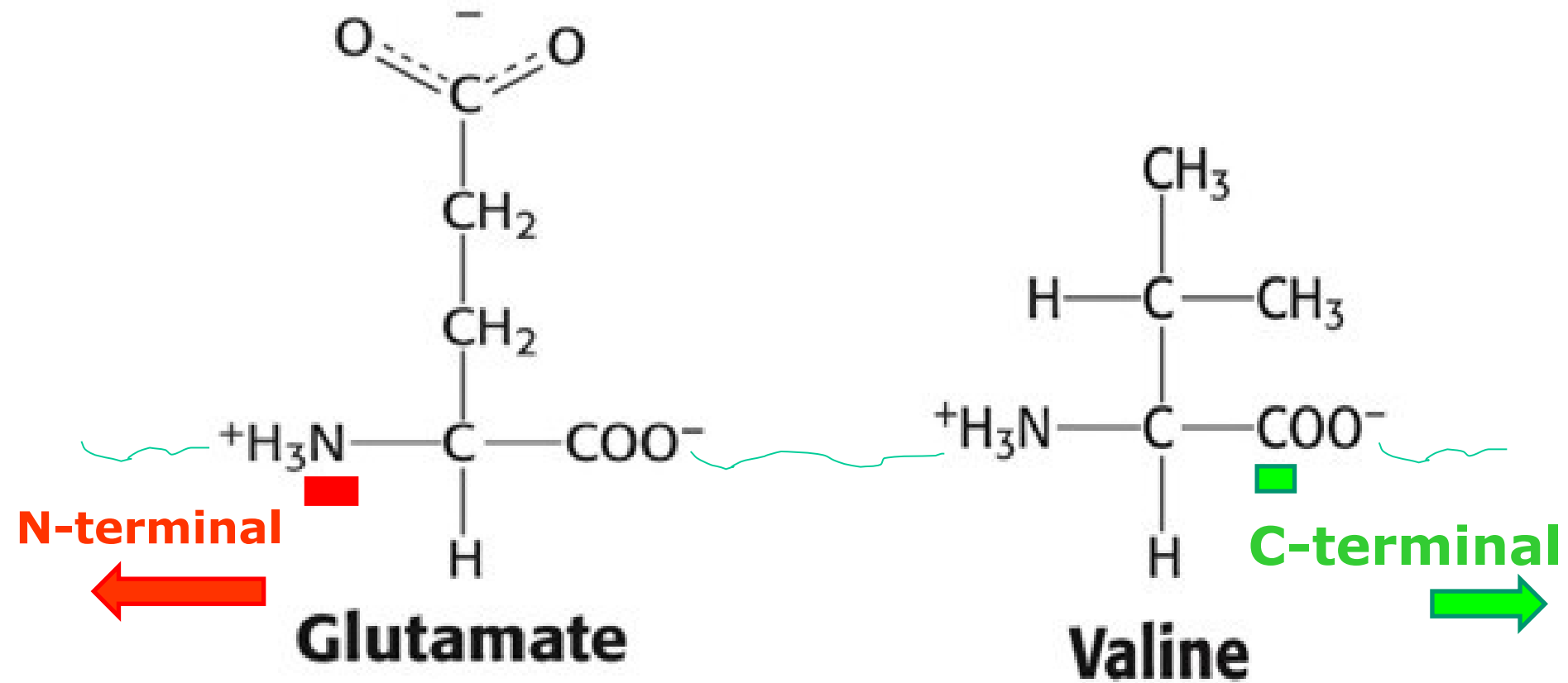
Hypoxia

Sickled
cells

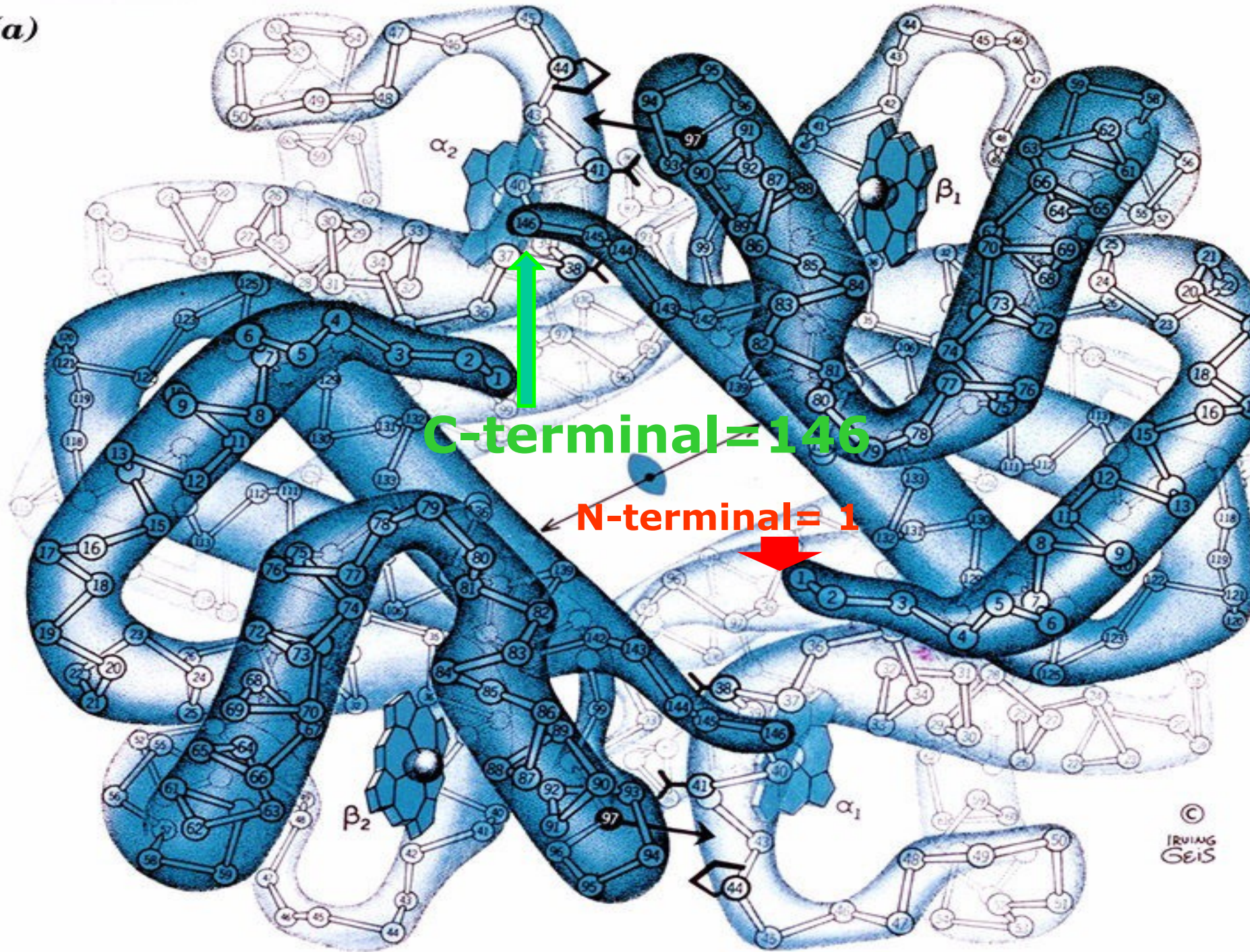


What is the difference in the structure of normal hemoglobin (HbA) and sickle cell hemoglobin (HbS)?

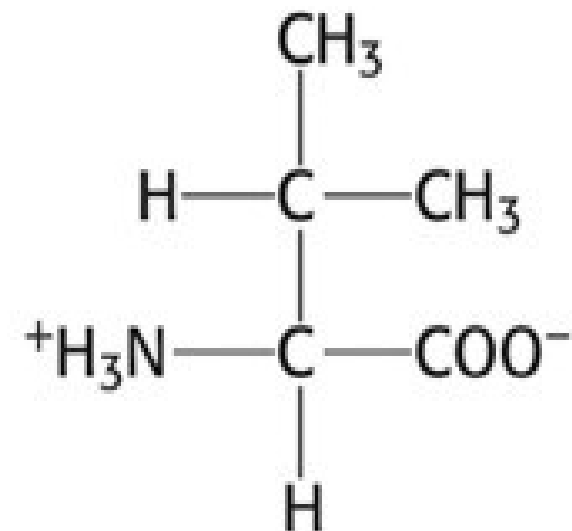
- What kind of behavior in relation to water show valine and glutamate?



(a)



- In HbS, the nonpolar amino acid valine has replaced the polar
- surface residue Glu6 of the β subunit, generating a hydrophobic
- **“sticky patch” on the surface of the β subunit of both oxyHbS and deoxyHbS**



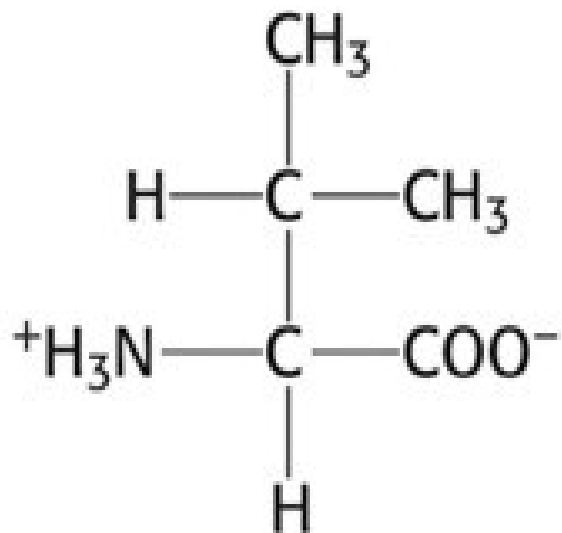
Valine

- Both HbA and HbS contain a complementary sticky patch on their surfaces
- that is exposed only in the deoxygenated T state.
- Thus, at low PO_2 , deoxyHbS can polymerize to form long, insoluble fibers.

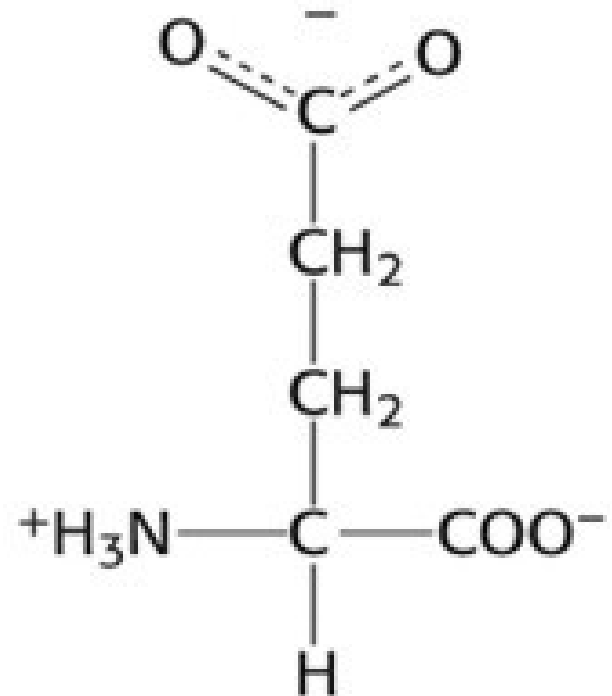
- Binding of deoxy-HbA terminates fiber polymerization,
- since HbA lacks the second sticky patch necessary to bind another Hb molecule.
- These twisted helical fibers distort the erythrocyte into a characteristic sickle shape,
- rendering it vulnerable to lysis in the interstices of the splenic sinusoids.

- A low P_{O_2} such as that at high altitudes exacerbates the tendency to polymerize.
- Emerging treatments for sickle cell disease include inducing HbF expression to inhibit the polymerization of HbS,
- stem cell transplantation, and, in the future, gene therapy.

HbS has less negative charge than the normal HbA



Valine



Glutamate

How decrease of the negative charge on the hemoglobin molecules affects water retention within the red blood cells (RBC)?

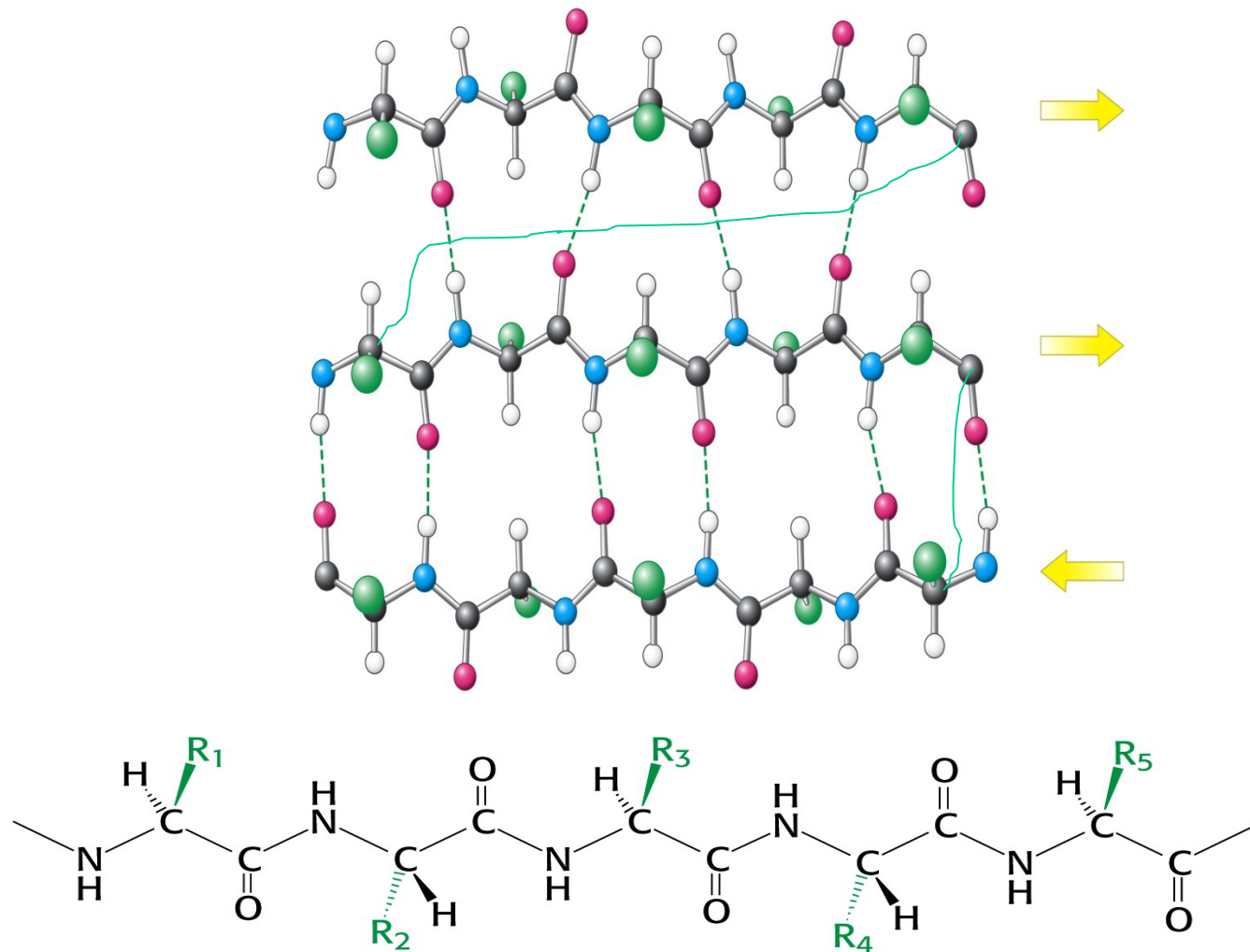
- In **healthy** individuals the **hemoglobin** molecules are **separated**.

Patients with **sickle cell** disease has an increase in hemoglobin concentration due to dehydration of erythrocytes.

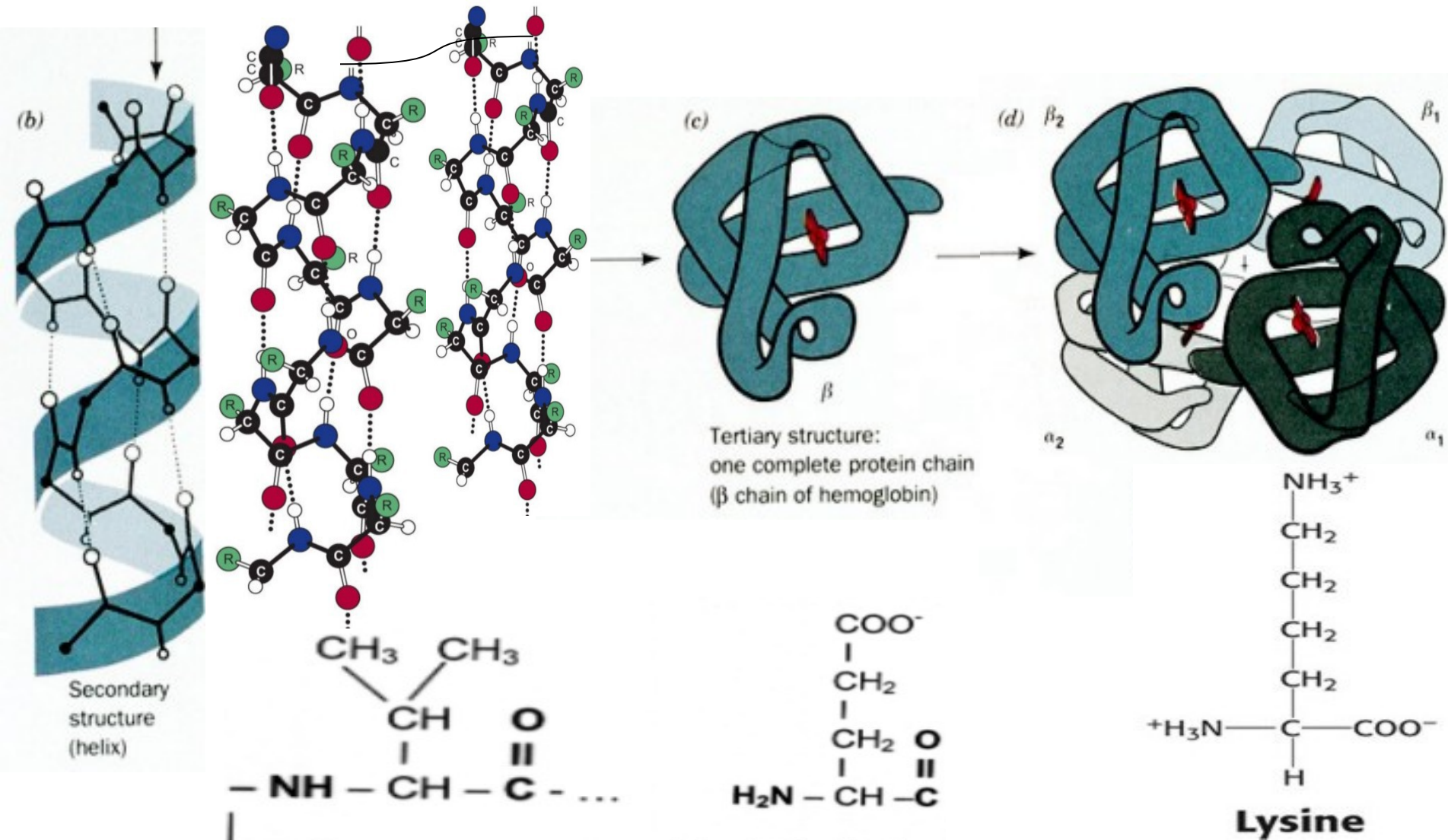
In this context, only one replaced valine in hemoglobin S (from Sickle RBC) is enough to induce self-association of proteins = precipitation or **coagulation**.

(a) - Lys - Ala - His - Gly - Lys - Lys - Val - Leu - Gly - Ala -
Primary structure (amino acid sequence in a polypeptide chain)

-folded sheet



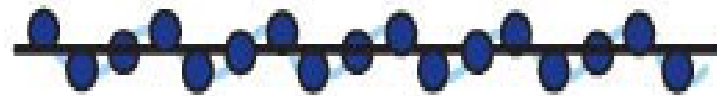
Which atoms or functional groups of amino acids are responsible for the formation of secondary, **tertiary** and quaternary structure?



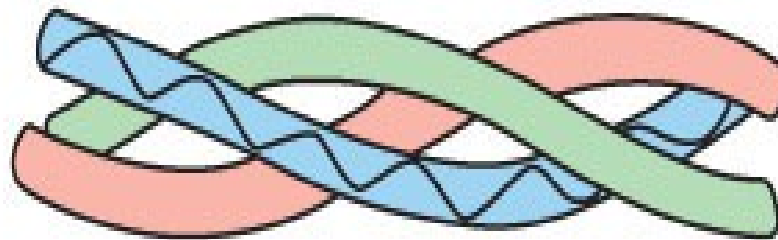
aminokiselinska
sekvenca

–Gly – X – Y – Gly – X – Y – Gly – X – Y –

sekundarna
struktura



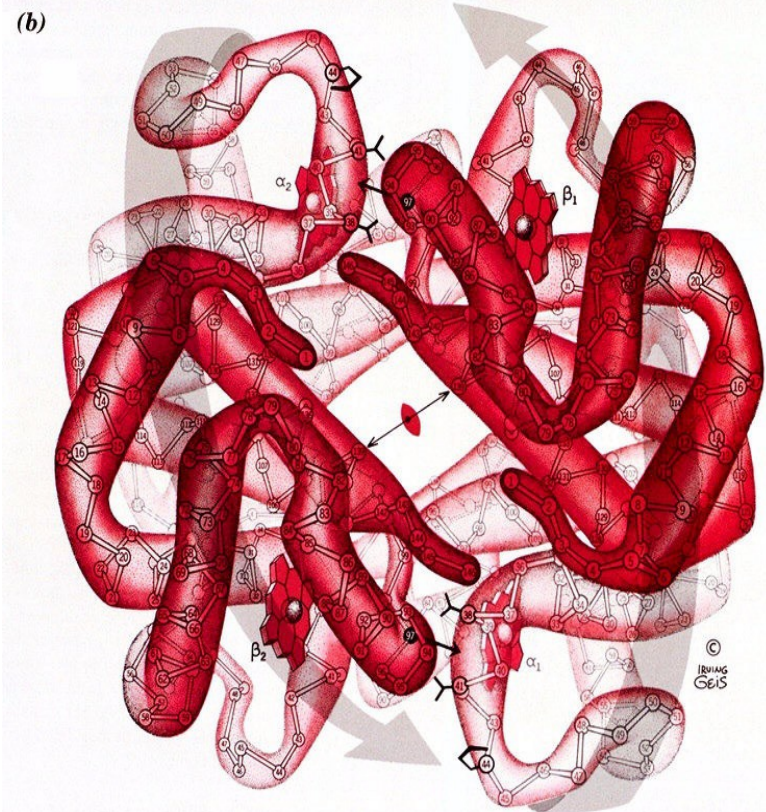
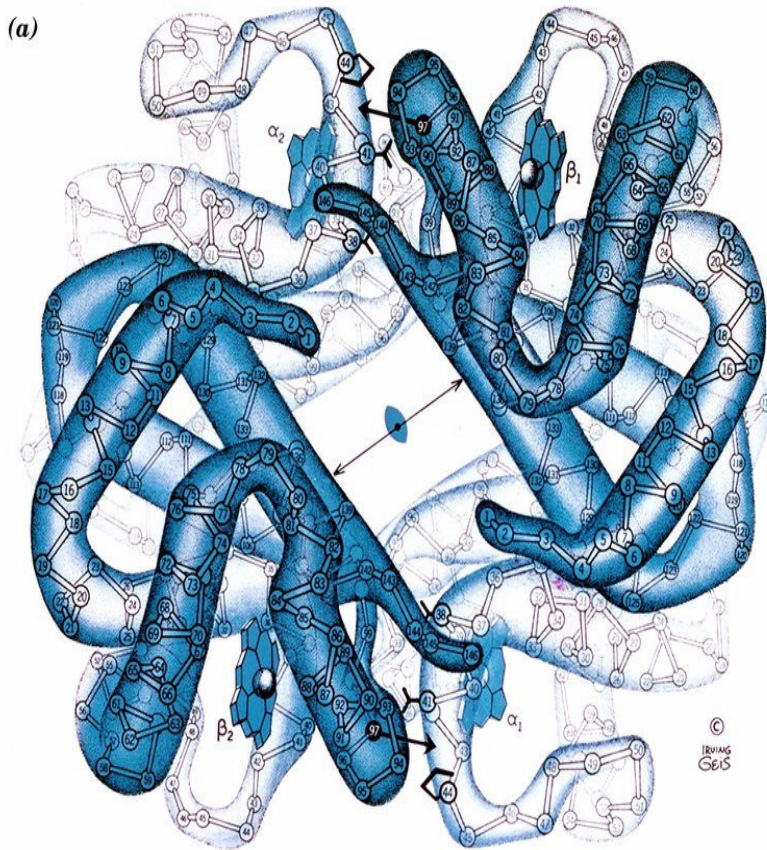
trostruka
uzvojnica



Slika 5-11. Primarna, sekundarna i kvaterna struktura kolagena.

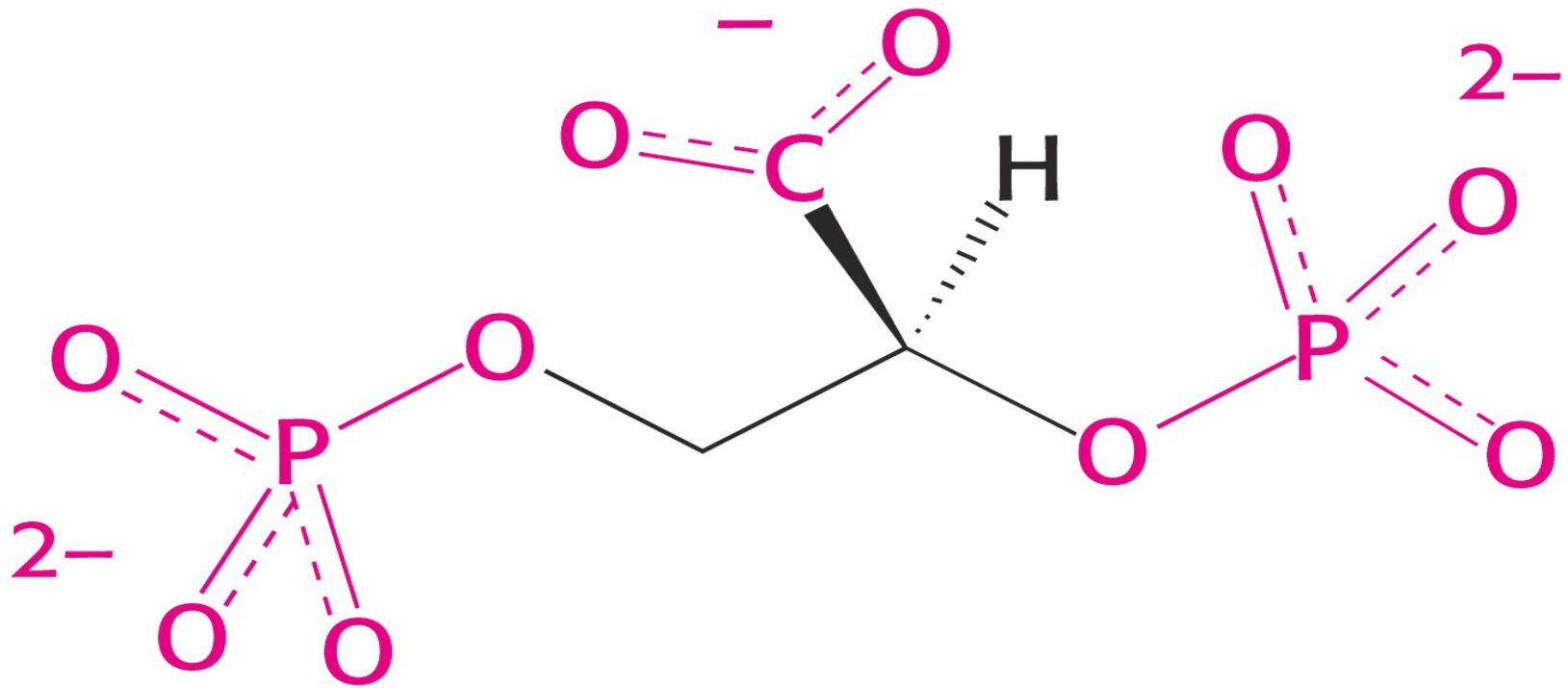
Is the diameter of the **central**
hemoglobin **cavity** larger
for deoxy (taut, tense, T state)
or for oxy (relax, R state)
hemoglobin molecule?

Deoxy- and oxy- hemoglobin

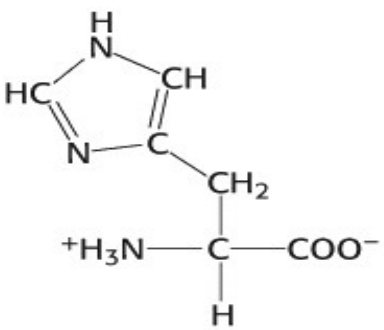


Which molecule binds within central hemoglobin cavity under conditions of low oxygen pressure?

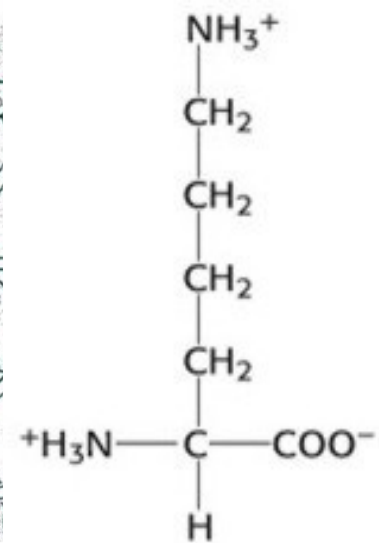
Explain the chemical bonds in the figure 6-12.



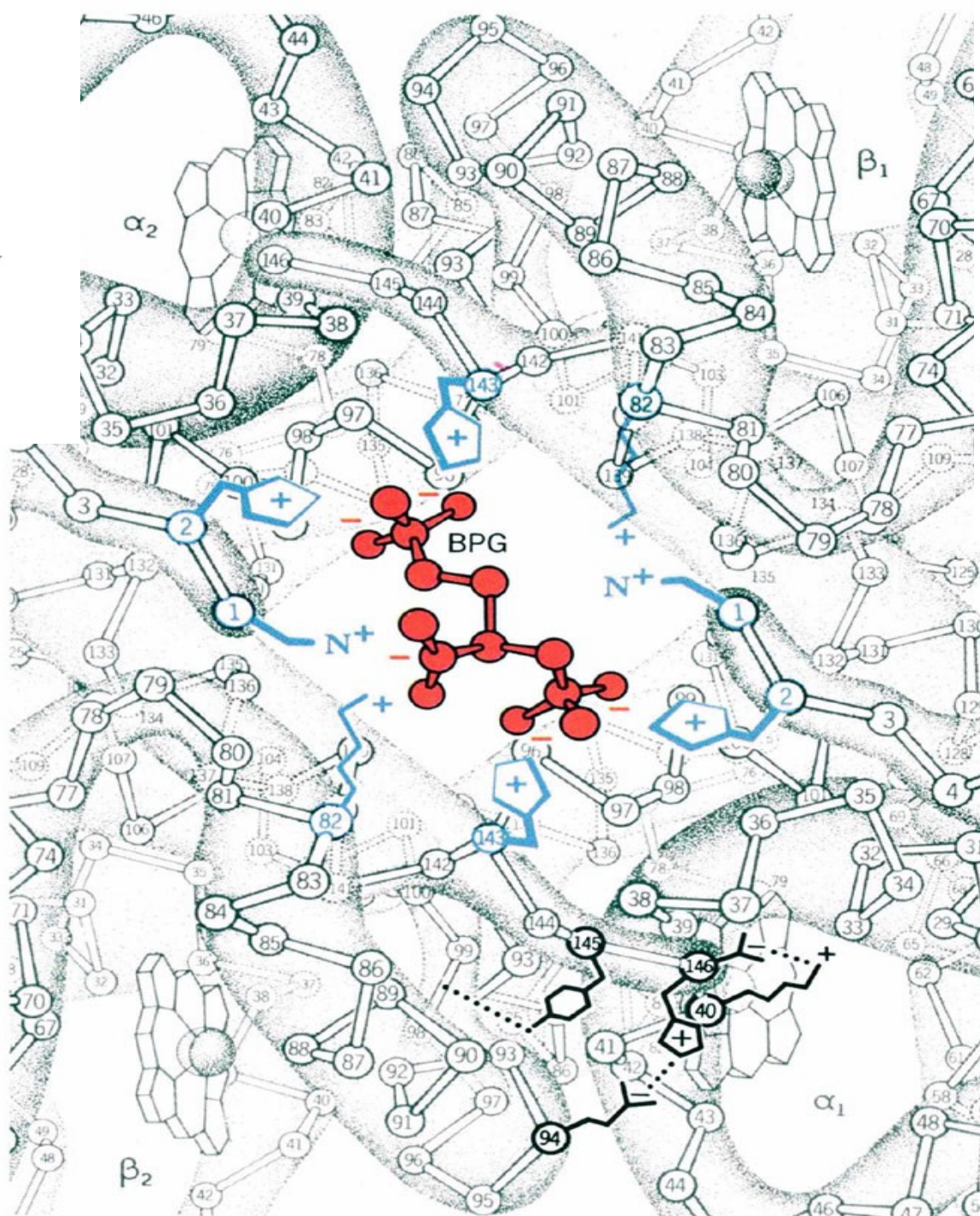
**2,3-Bisphosphoglycerate
(2,3-BPG)**



Histidine
(His, H)

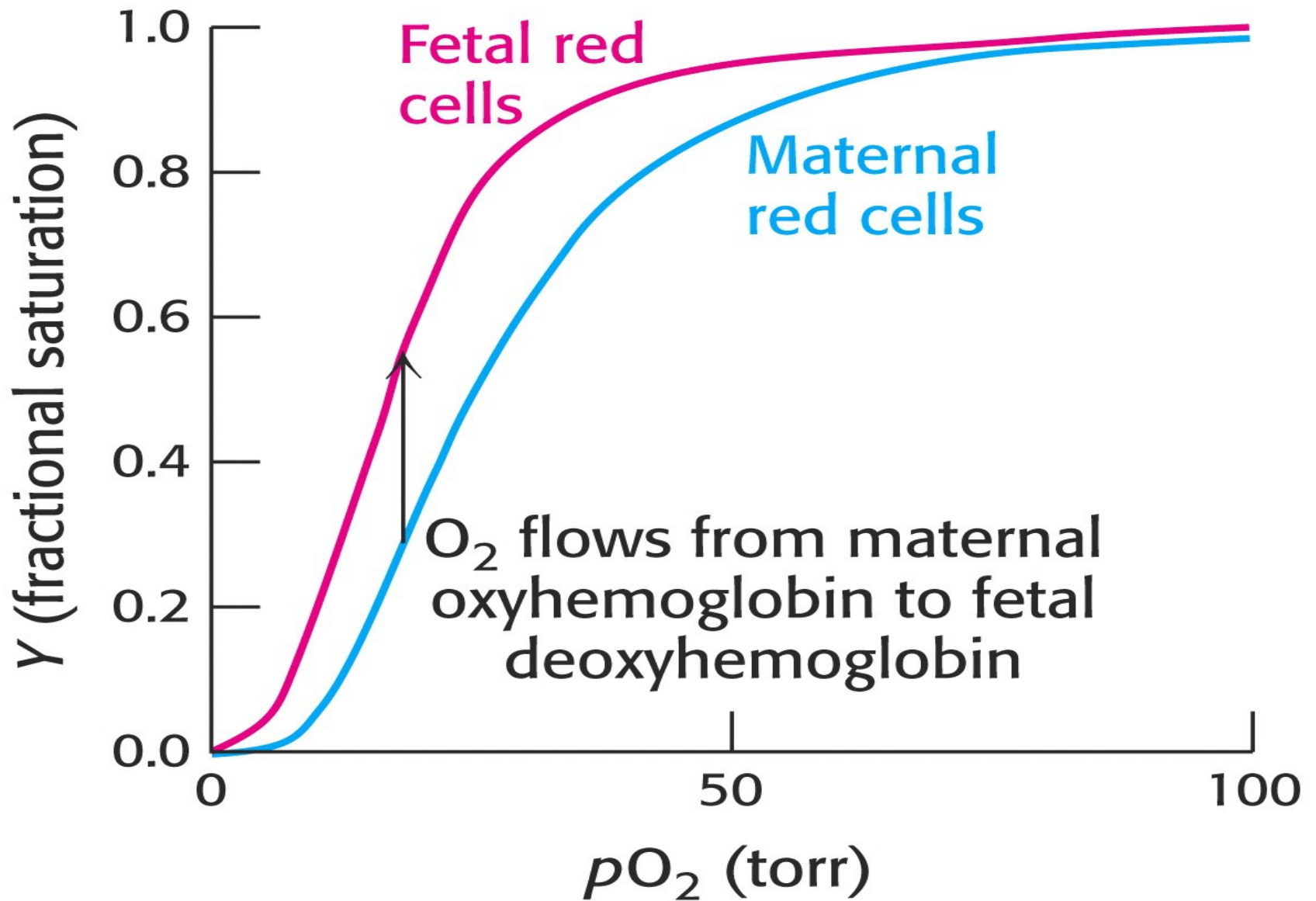


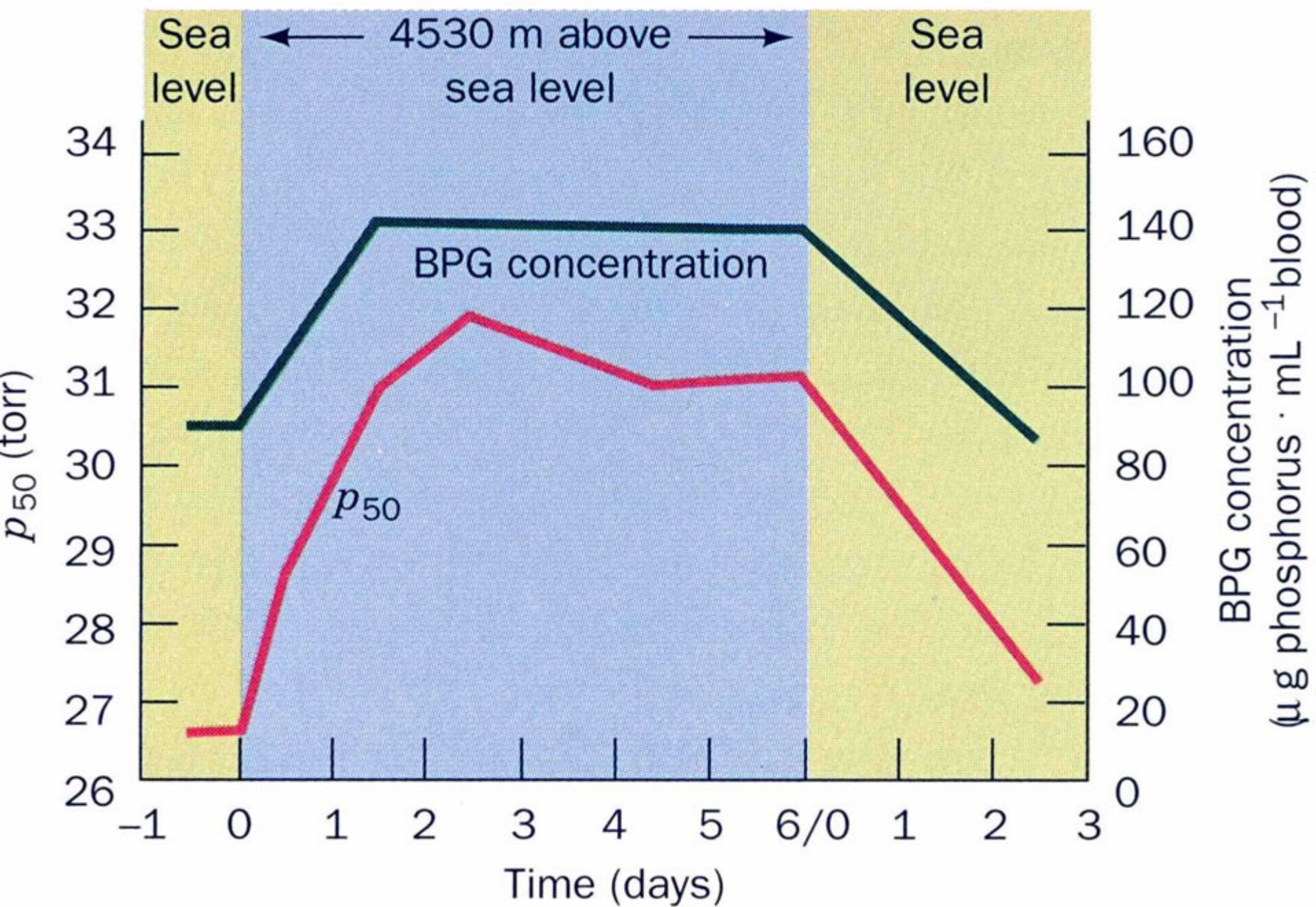
Lysine

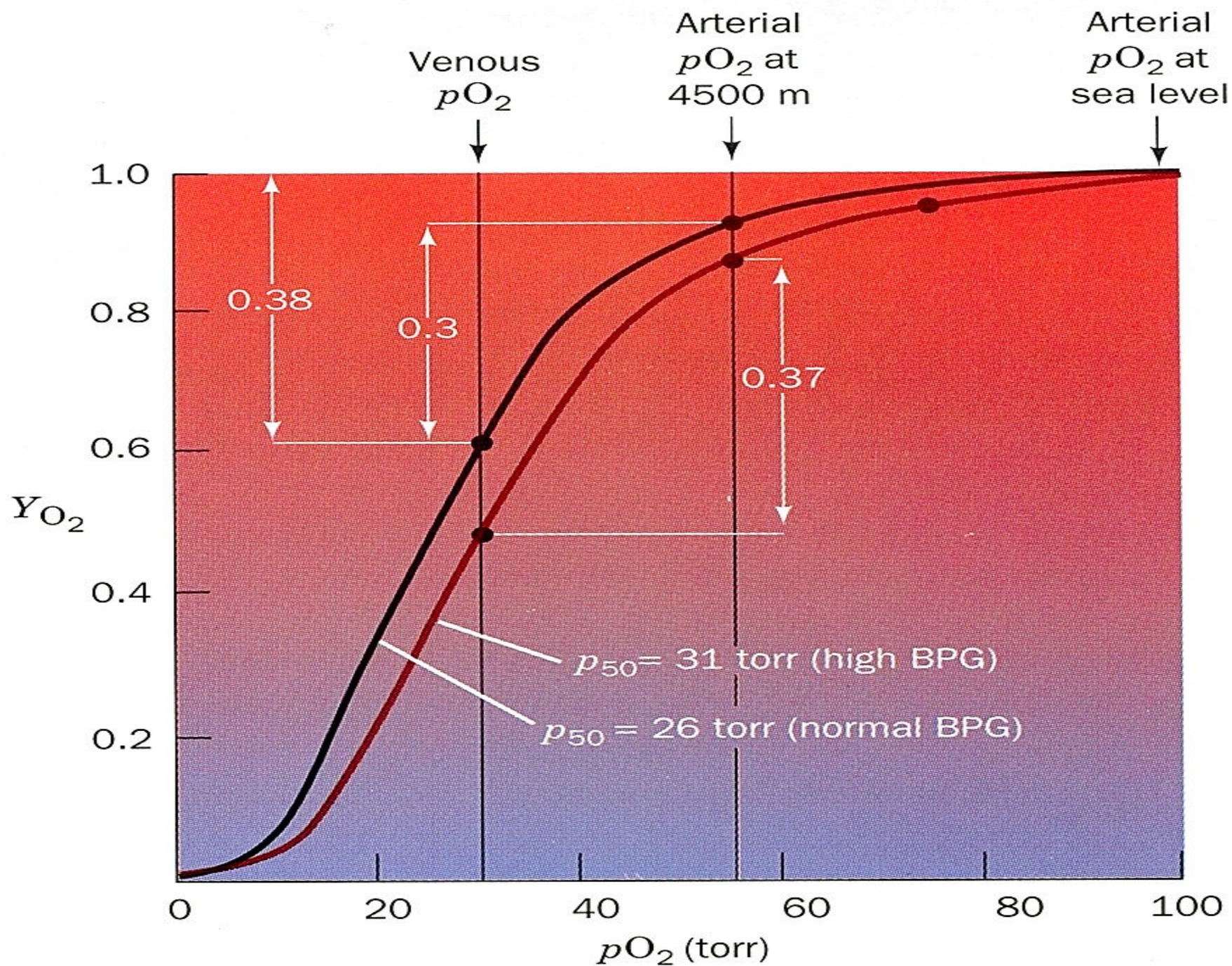


- What does P_{50} value represent?
- P_{50} values for HbA and fetal HbF are 26 and 20 mm Hg, respectively. Which Hb has a higher affinity for oxygen?

- Relative affinities of different hemoglobins for oxygen
- are expressed as P_{50} , the P_{O_2} that **half-saturates** them with O_2 .
- Hemoglobins saturate at the partial pressures
- of their respective respiratory organ, eg, the lung or placenta.



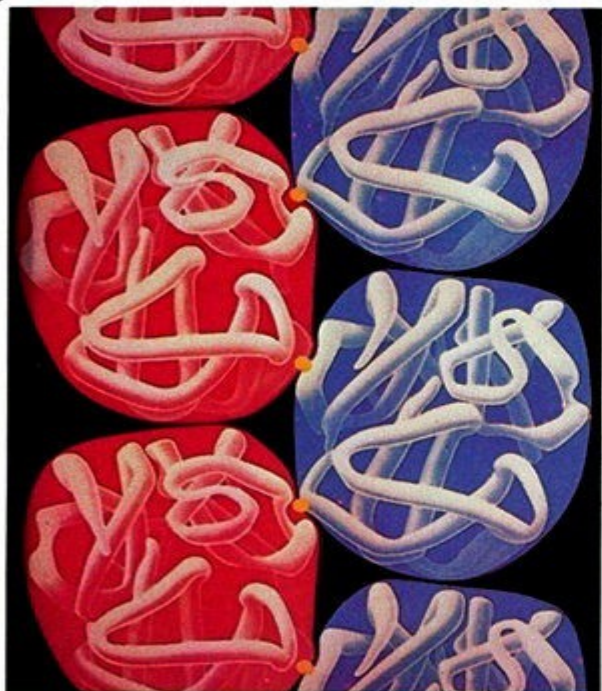




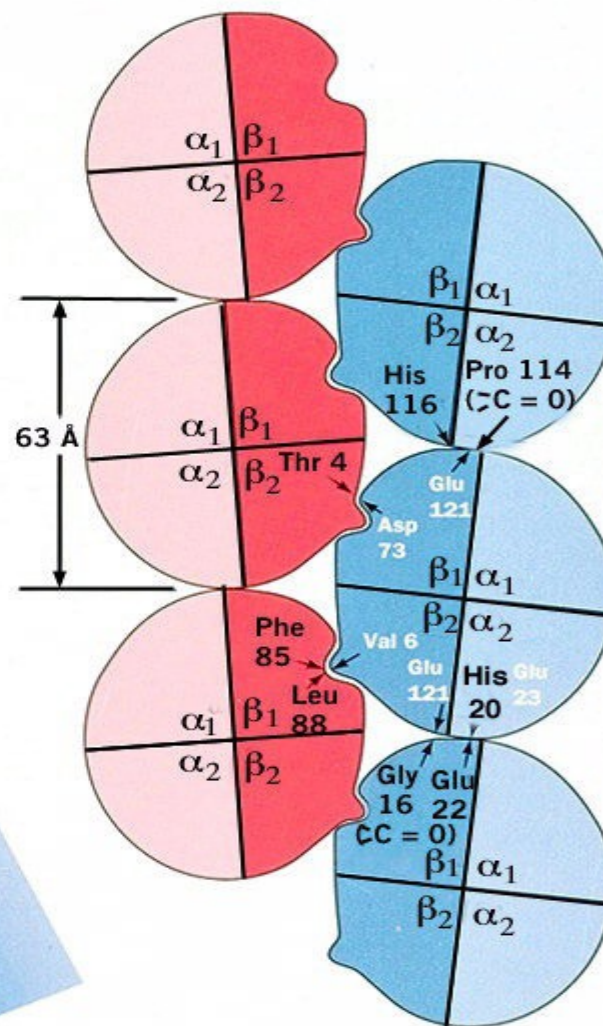
Why do RBC tend to sickle
under conditions of low oxygen pressure
(prevailing in the organs and muscles)?

- Both HbA and HbS contain a complementary sticky patch on their surfaces
- that is exposed only in the deoxygenated T state.
- Thus, at low PO_2 , deoxyHbS can polymerize to form long, insoluble fibers.

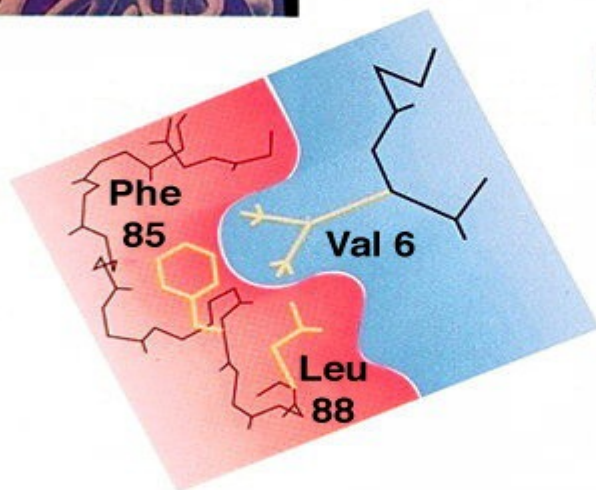
(a)



(b)



(c)



(a) Homogeneous nucleation



(b) Heterogeneous nucleation



Growth of
thermodynamically
unstable aggregates

Critical
nucleus

Increasing
stability and
rapid growth

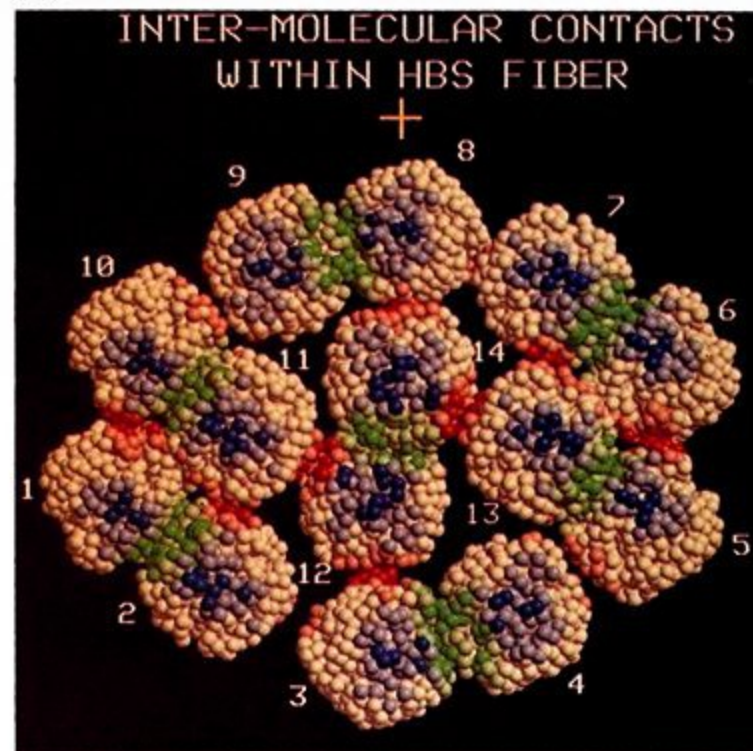




(a)



(b)



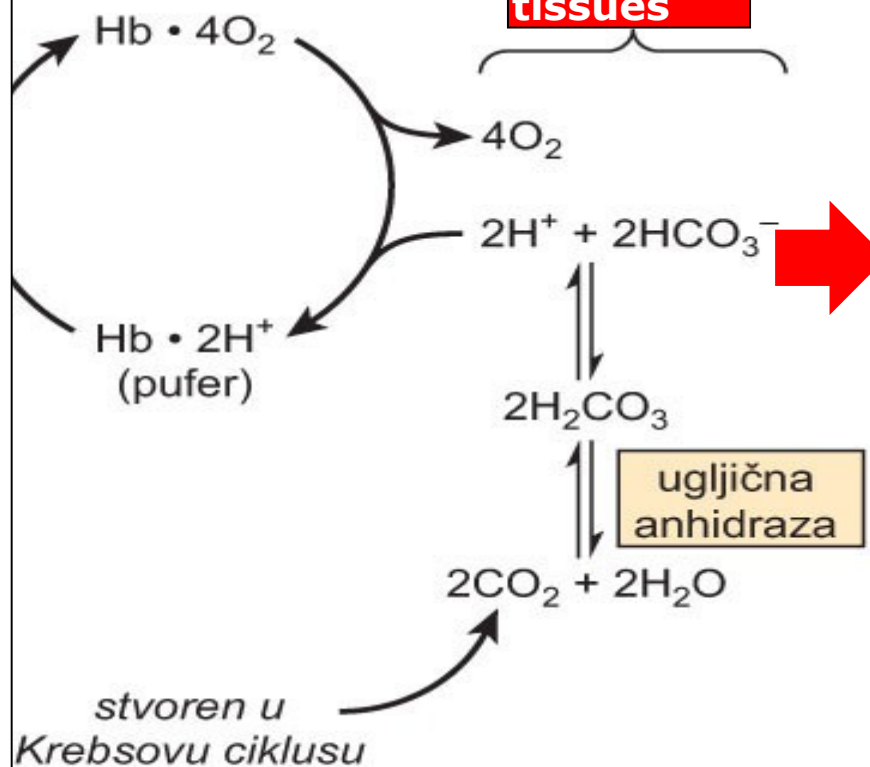


- Twisted HbS helical fibers distort the erythrocyte into a characteristic sickle shape,
- rendering it vulnerable to lysis in the interstices of the splenic sinusoids.

- Has H^+ higher affinity for a deoxy or oxy hemoglobin?
- In which form CO_2 is transferred from the all tissues to the lung? Describe figure 6-11.

anhidraza

peripheral
tissues



činak. Ugljikov dioksid iz perifernoga tkiva
natnu kiselinu, koja disocira na proton i hid-

The Bohr effect

peripheral
tissues

lungs

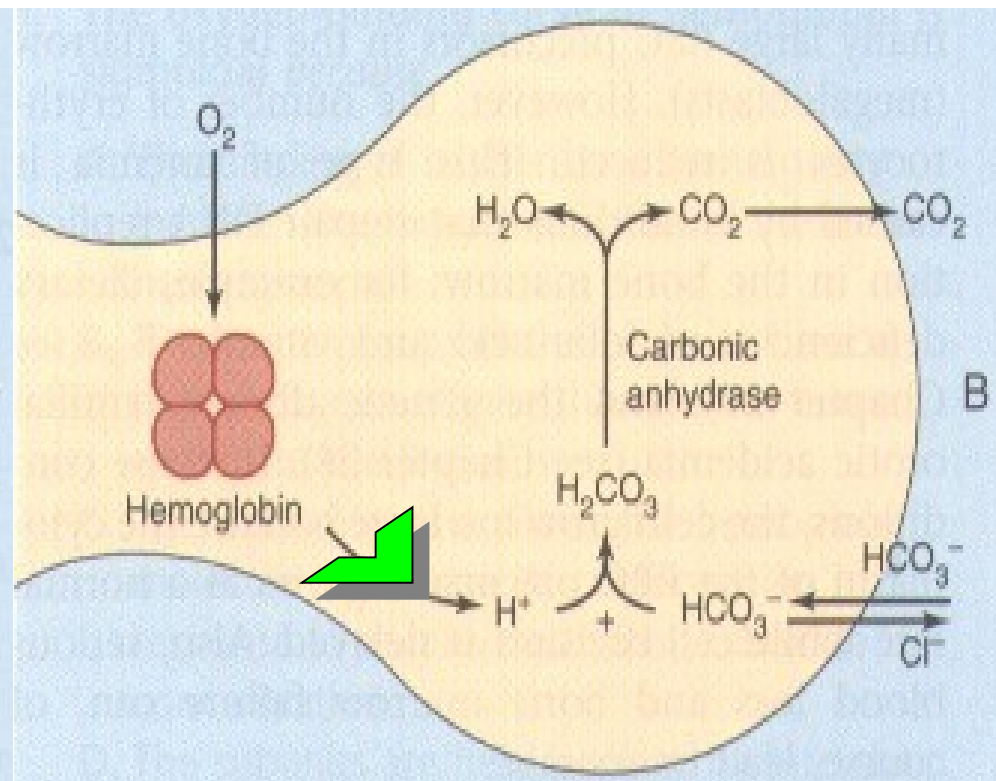
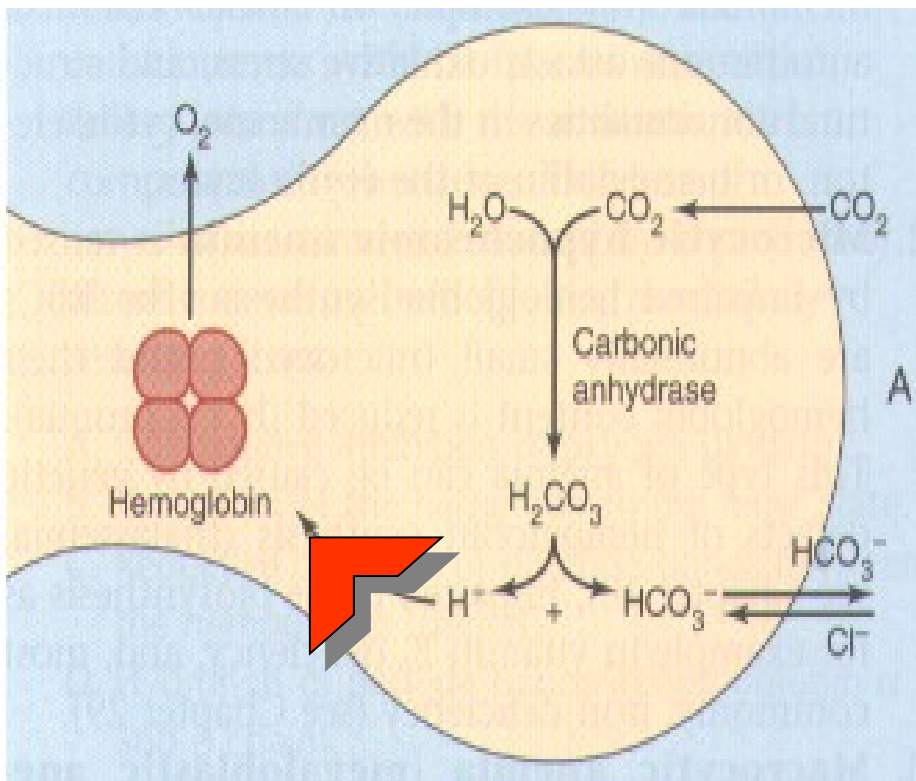
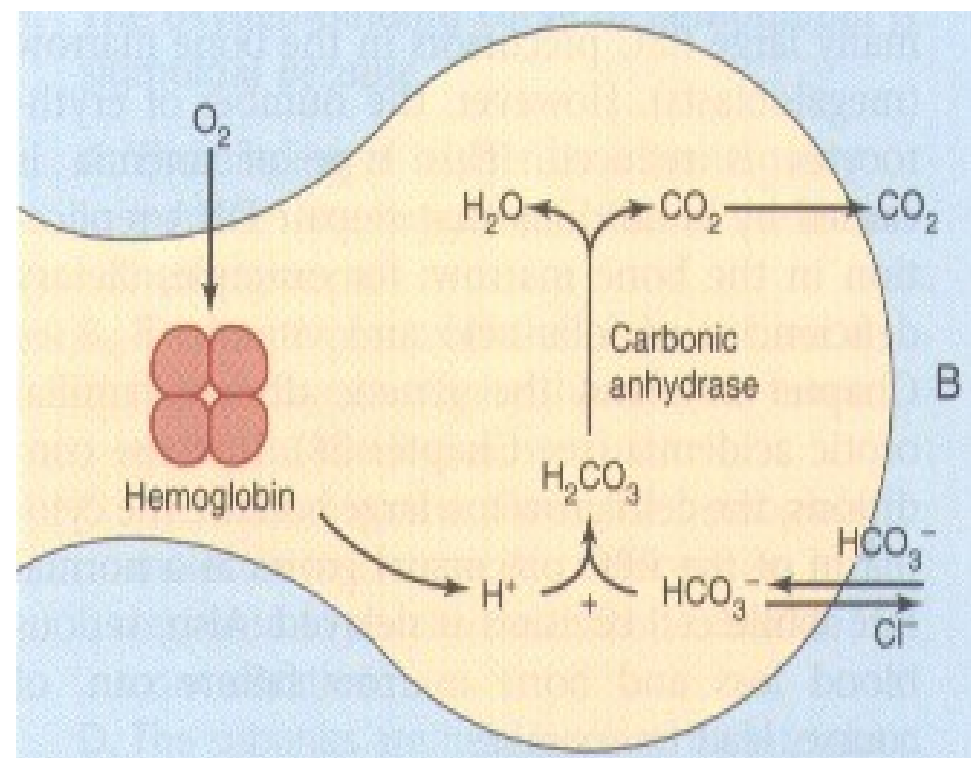
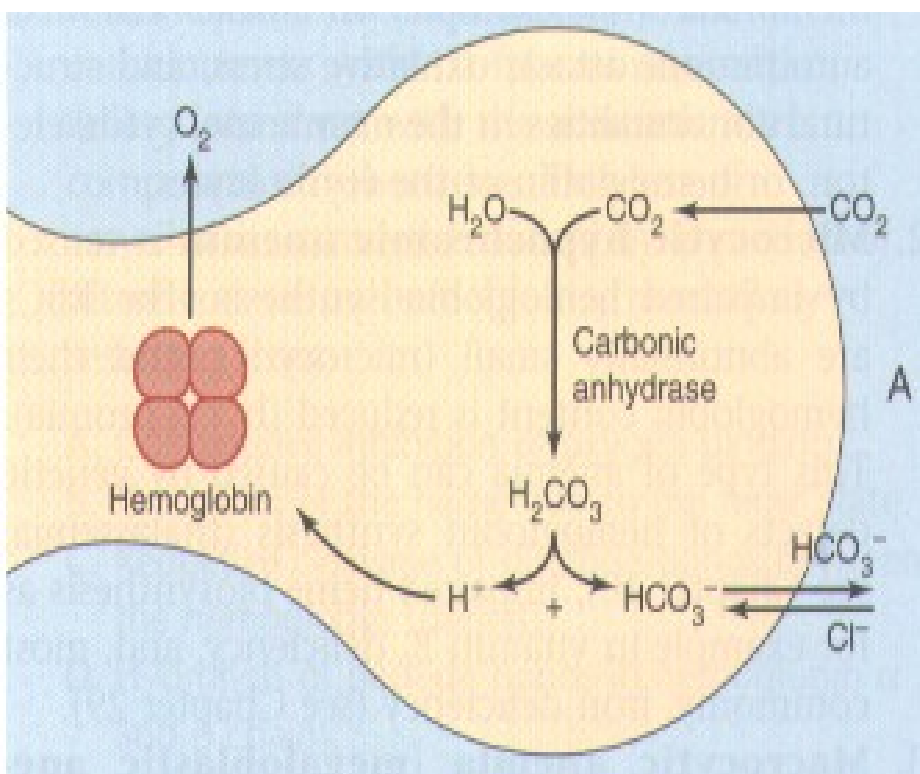


Figure 3.7 The major mechanism of carbon dioxide transport. Note that all processes are reversible. Their direction is determined by the concentrations of the involved substances in the extrapulmonary tissues **(A)** and in the lung capillaries **(B)**.



- What is the structural difference between **methemoglobin** and normal hemoglobin A molecule?
- What is the structural difference between **hemoglobin M** and normal hemoglobin A molecule? Does hemoglobin M show Bohr effect?

Methemoglobinemia

- the heme iron is ferric (Fe^{3+}) rather than ferrous (Fe^{2+})
- methemoglobin thus can neither bind nor transport O_2
- Normally, the enzyme methemoglobin reductase
- reduces the Fe^{3+} of methemoglobin to Fe^{2+}

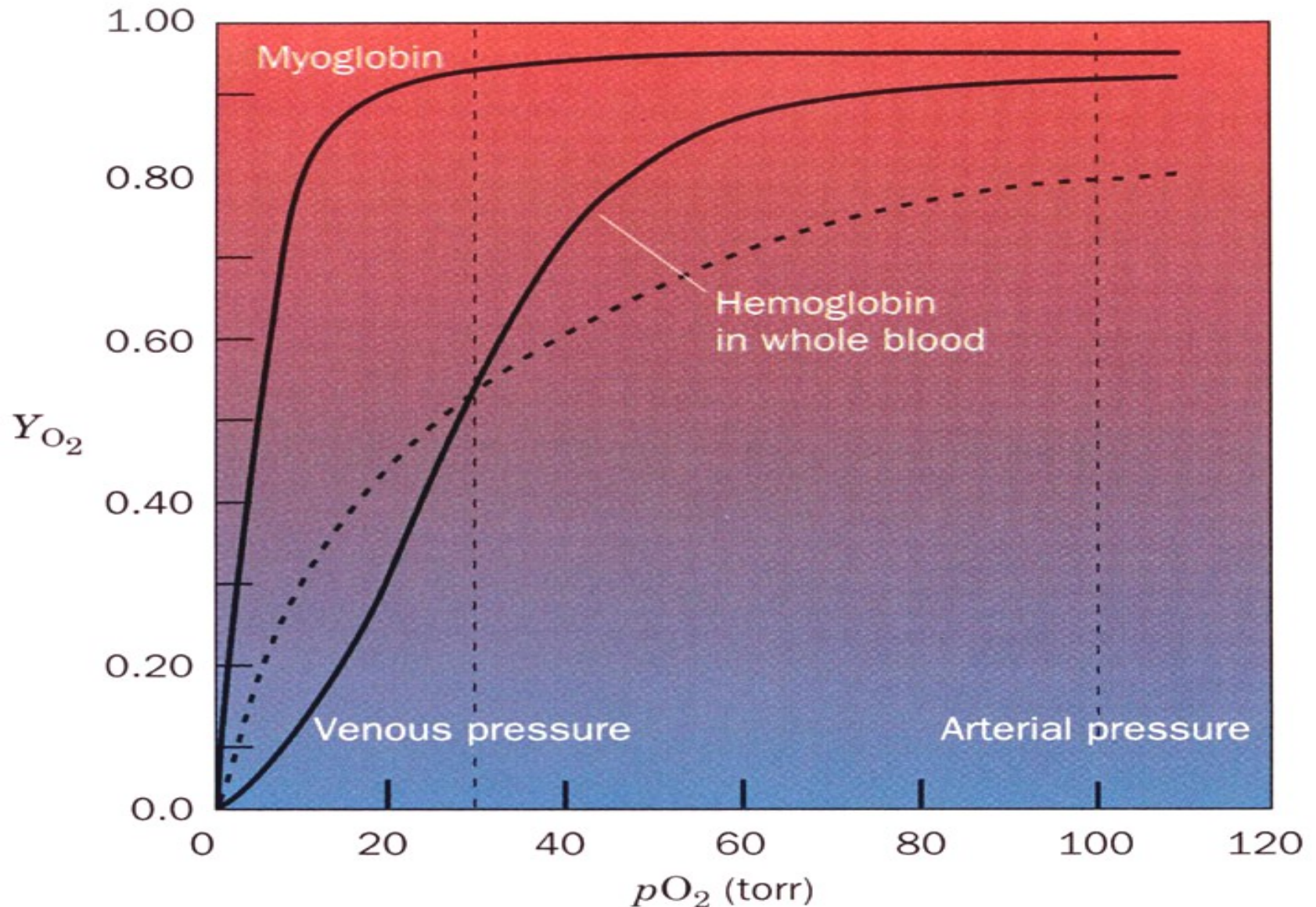
Methemoglobin can arise by oxidation of Fe^{2+} to Fe^{3+}

- as a side effect of agents such as sulfonamides,
- from hereditary hemoglobin M,
- or consequent to reduced activity of the enzyme methemoglobin reductase.

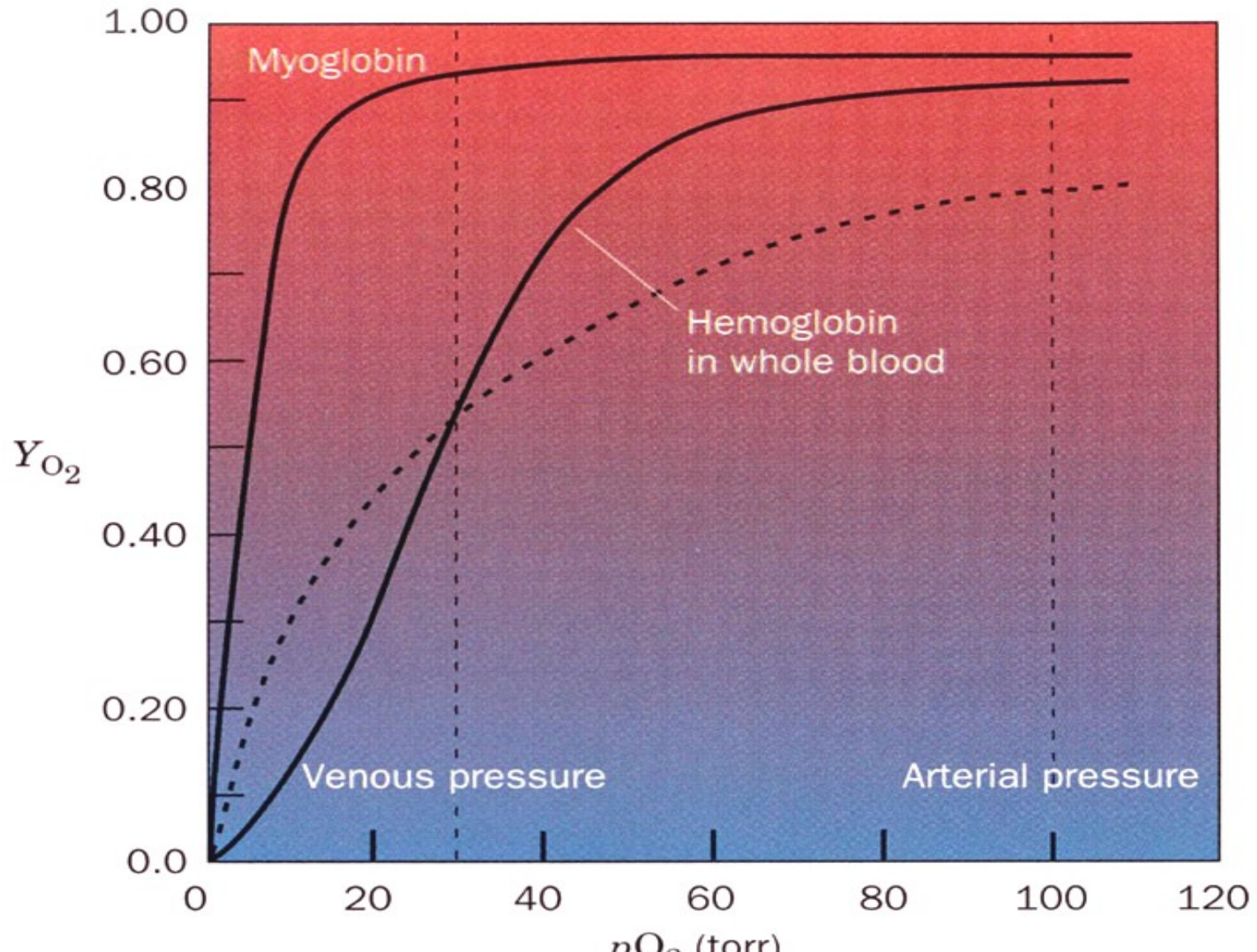
Hemoglobin M

- Histidine has been replaced by tyrosine that stabilizes the Fe^{3+} form.
 - chain hemoglobin M variants, favors the T state. Oxygen affinity is reduced, and the Bohr effect is absent.
- β -chain hemoglobin M variants exhibit R-T switching, and the Bohr effect is therefore present.

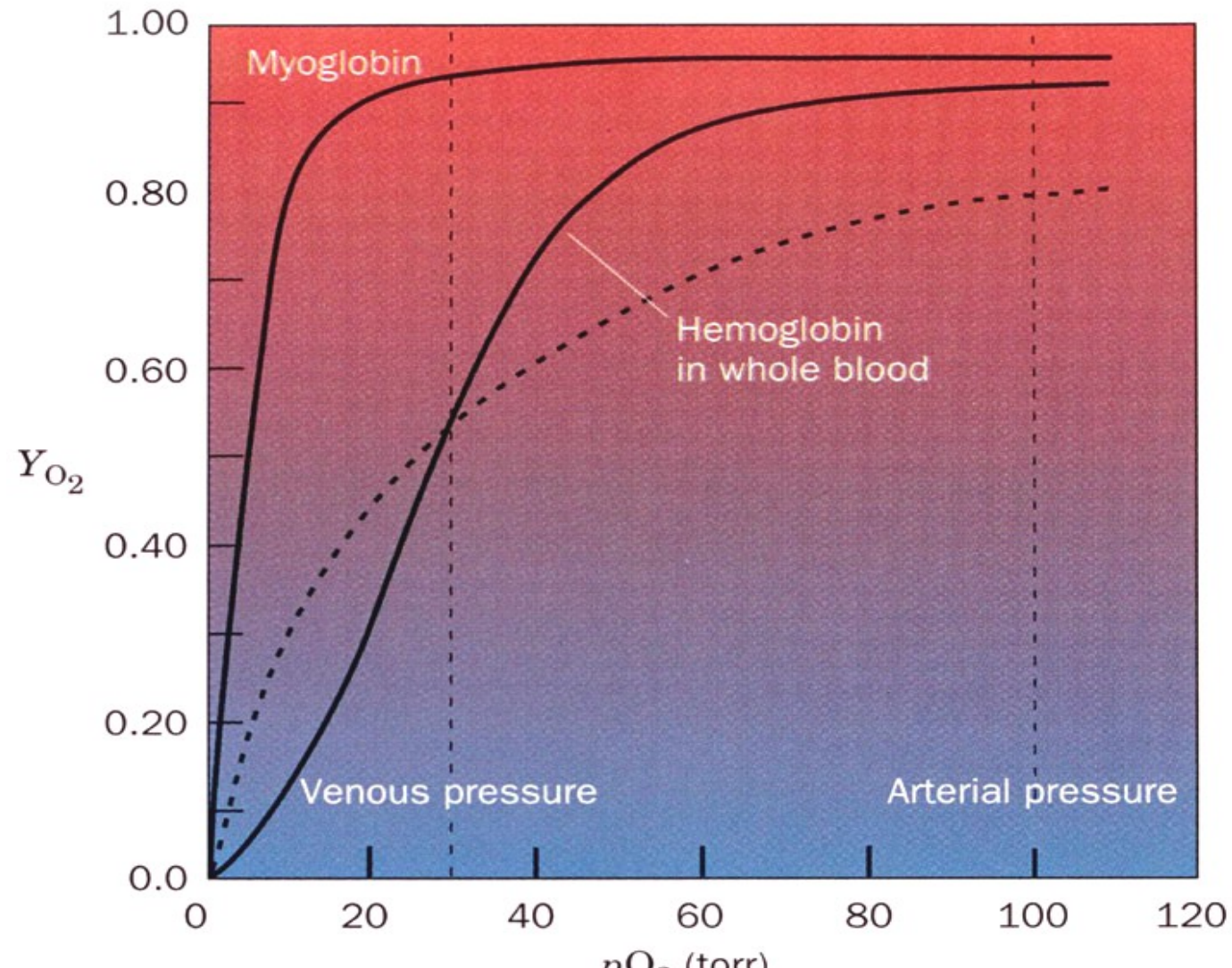
- Why is myoglobin unsuitable as an O_2 transfer protein, but very suitable for the storage of O_2 ?
- Describe figure 6-5.

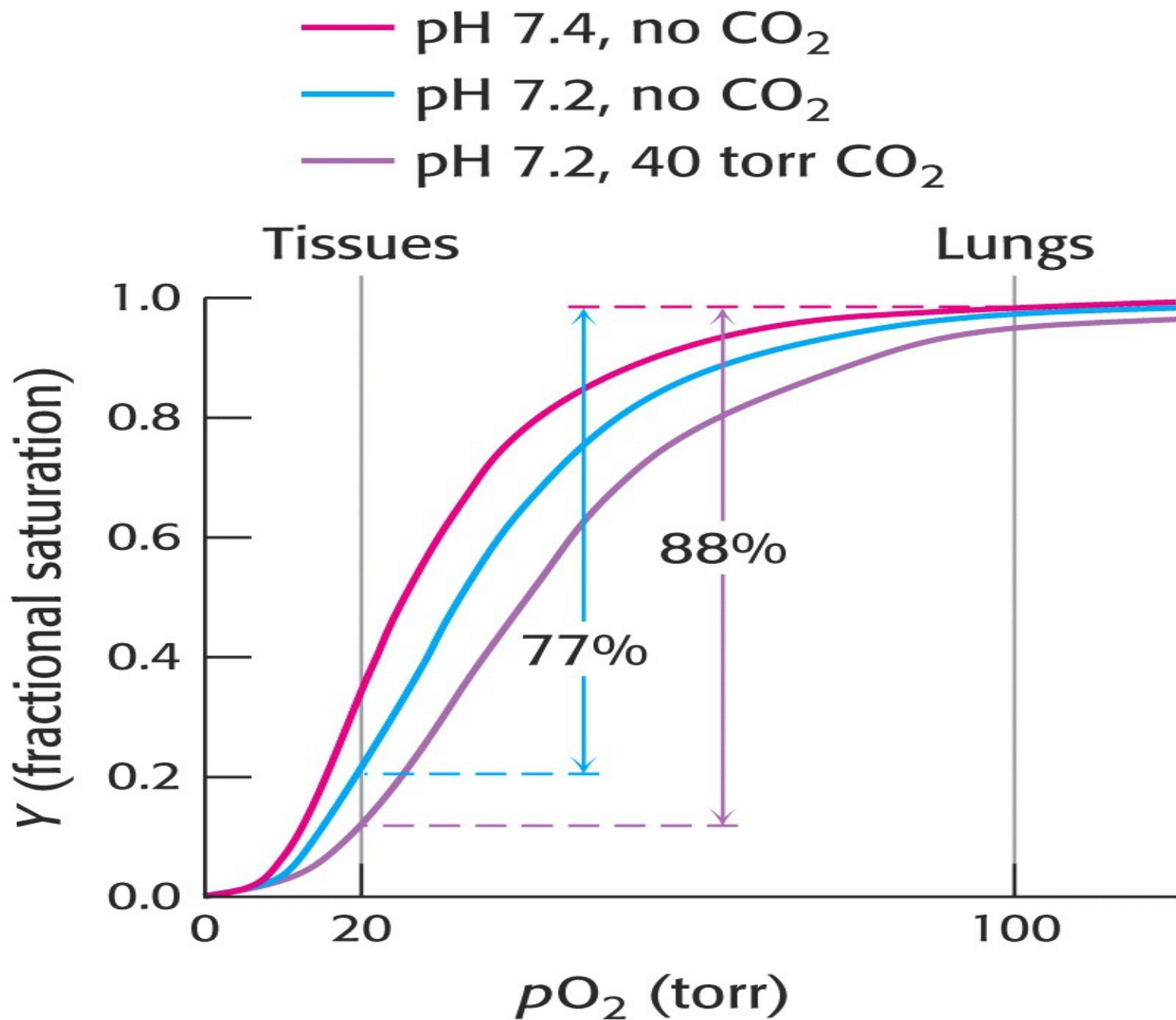


- The O_2 -binding curve for myoglobin is hyperbolic,
- but for hemoglobin it is sigmoidal,
- a consequence of cooperative interactions in the tetramer.



- Cooperativity maximizes
- the ability of hemoglobin both to **load** O_2 at the P_{O_2} of the **lungs**
- and to **deliver** O_2 at the P_{O_2} of the **tissues**.







SCURVY

- Jacques Cartier in 1536 gave a vivid description of this dietary deficiency disease, which afflicted his men as they were exploring the Saint Lawrence River:
- ...Others also had all their **skins** spotted, with **spots of blood** of a purple colour: then did it ascend up to their ankles, knees, thighs, shoulders, arms, and necks. Their mouths became stinking, their gums so rotten, that all the **flesh did fall of**, even to the roots of the teeth, which did also almost all fall out.



Explain the concept
of post-translational modifications.

- The maturation of proteins into their final structural state which involves the cleavage or formation (or both) of covalent bonds.

Why are many polypeptides initially synthesized as larger precursors called proproteins?

Collagen illustrates the role of posttranslational processing in protein maturation

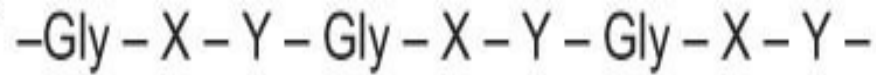
- Many polypeptides are initially synthesized as larger precursors called proproteins.
- The “extra” polypeptide segments in these proproteins often serve as leader sequence that target a polypeptide to particular organelle or facilitate its passage through a membrane.

- Other “extra” polypeptide segments ensure that the **potentially harmful activity** of a protein
- such as the **proteases trypsin and chymotrypsin** remains inhibited until these proteins reach their final destination
- Superfluous peptide regions are removed by selective **proetolysis**.

- Other covalent modifications may take place
- that add new chemical functionalities to a protein.
- The maturation of collagen illustrates all that processes.

- Which a small amino acid must be at every third position of the collagen alpha chain?

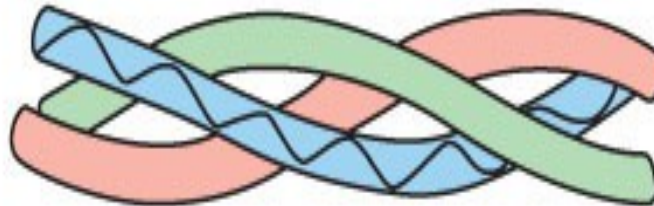
aminokiselinska
sekvenca



sekundarna
struktura

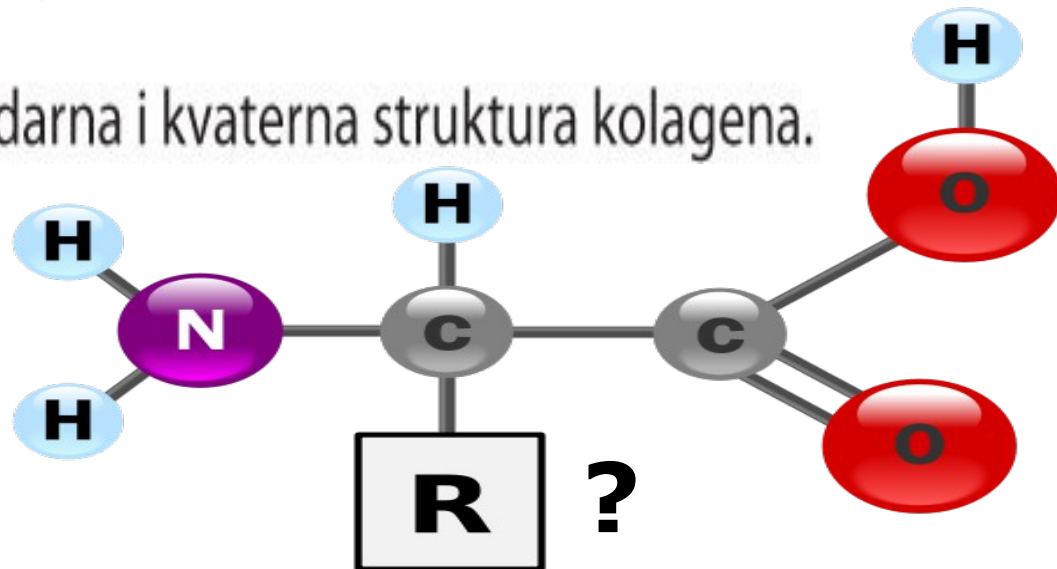
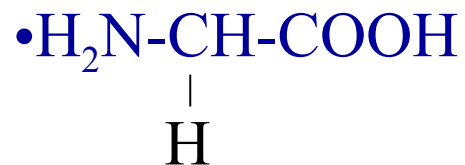


trostruka
uzvojnica



Slika 5-11. Primarna, sekundarna i kvaterna struktura kolagena.

- **glycine.**

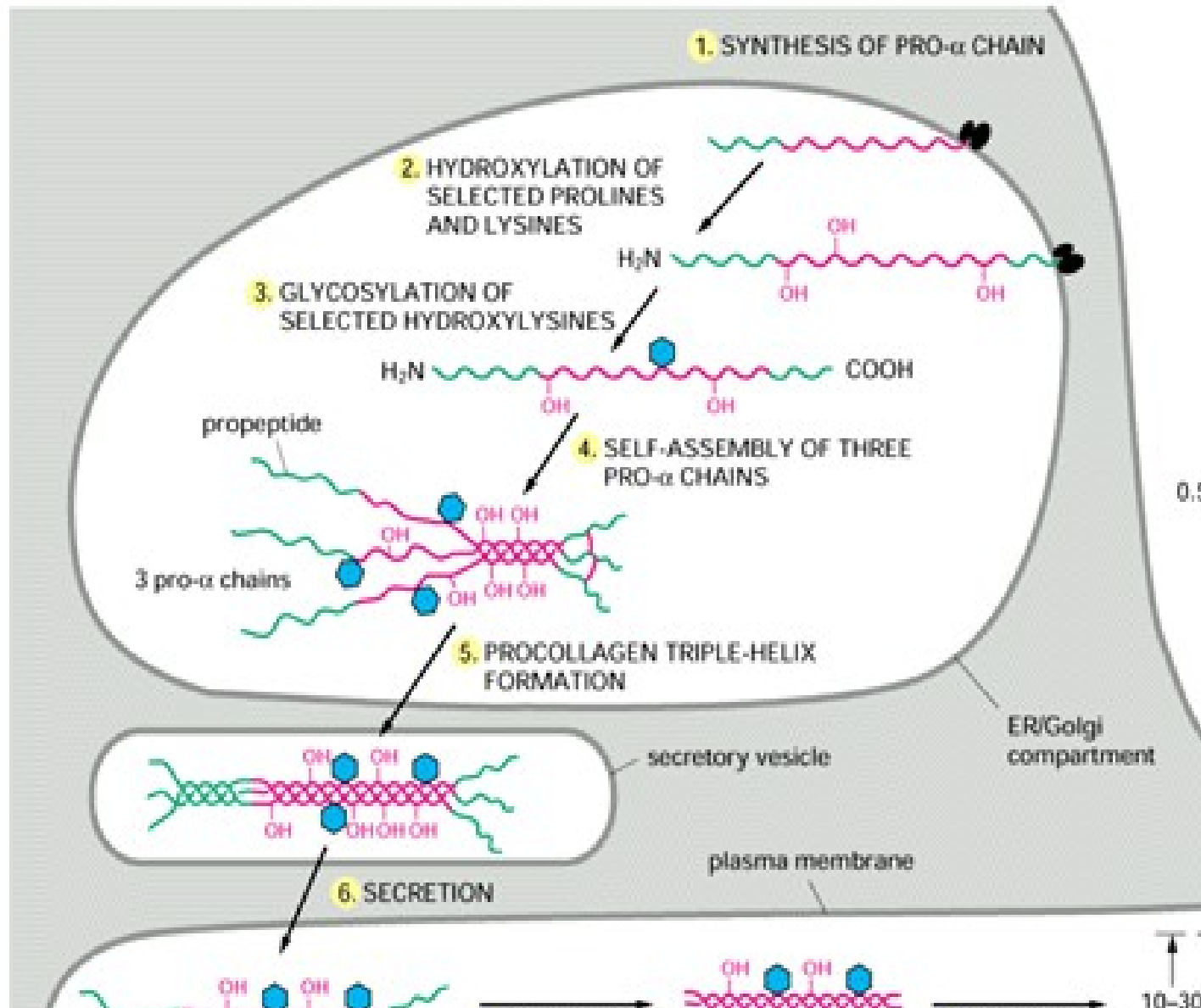


- Collagen contains approximately 1000 amino acids. Which amino acids are repeated in collagen about 100 times?
- Which enzymes catalyze the hydroxylation of these amino acids?

List some of the post-translational modifications of procollagen (fibrillar collagen precursors) that occur **within the cell** (Harper, Table 48-3).

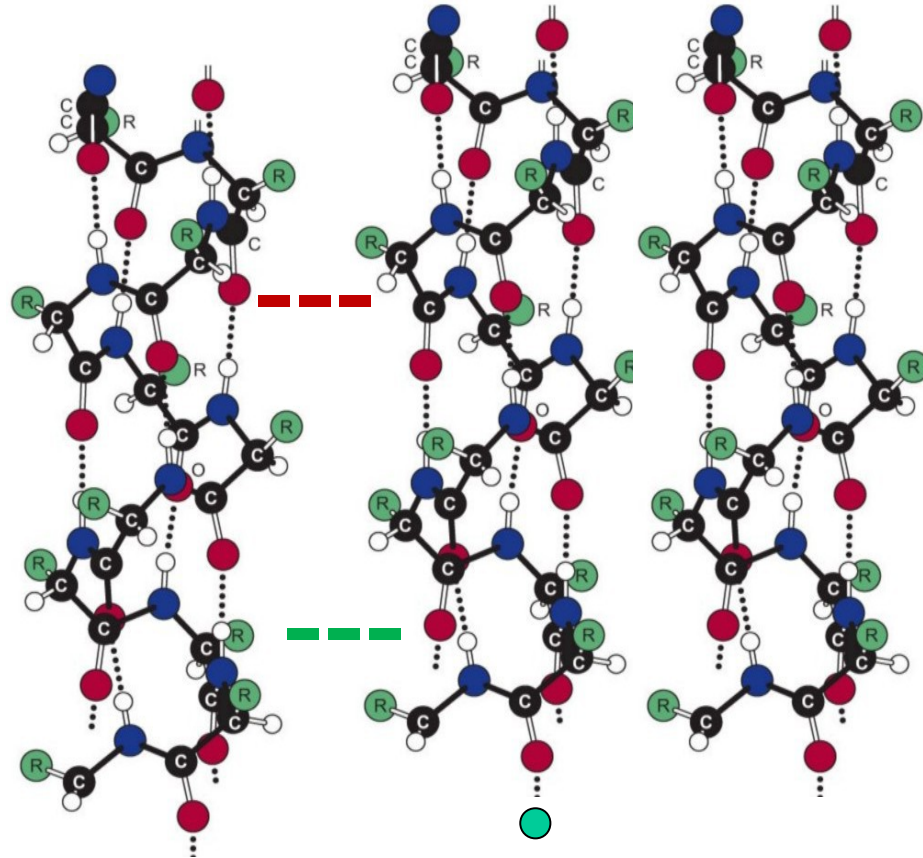
Intracellular processing of the collagen precursor

- Cleavage of signal peptide
- Hydroxylation of prolyl residues and some lysyl residues
- Formation of triple helix



- Do tRNAs for hydroxylated amino acids exist?

- How hydroxyl group of hydroxyproline contribute to the quaternary structure of collagen?

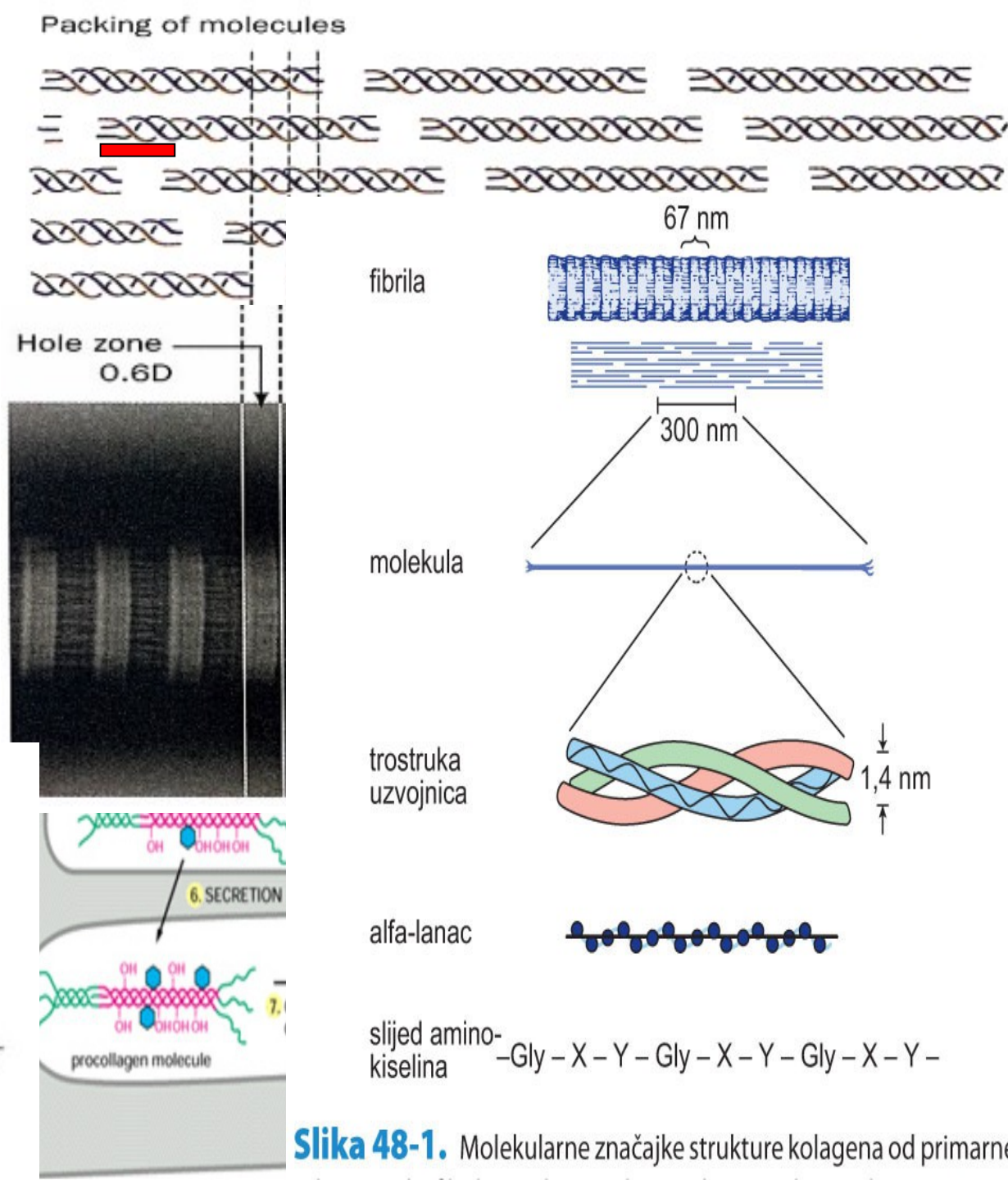
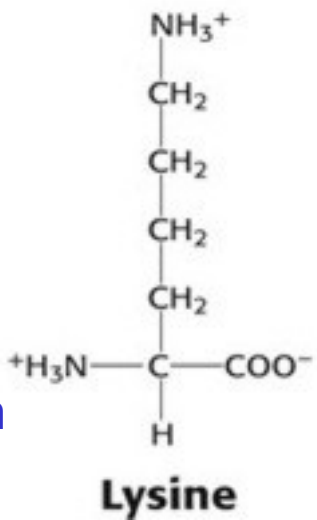


- List some of the procollagen post-translational modifications that occur **outside the cell.**

Extracellular Processing:

- Cleavage of amino and carboxyl terminal propeptides
- Assembly of collagen fibers in quarter-staggered alignment
- Oxidative deamination of -aminogroups of lysyl and hydroxylysyl residues to aldehydes

Formation of intra- and **interchain cross-links** via Schiff bases and aldol condensation products



Slika 48-1. Molekularne značajke strukture kolagena od primarne

- By which biochemical reaction vitamin C prevents scurvy pathogenesis?

Scurvy

PATHOBIOCHEMISTRY

- Prolyl hydroxylase enzyme, similar to the hemoglobin, contains atom of Fe (II), which is subject to oxidation to Fe (III).
- Reduction of Fe (III) to Fe (II) is conducted by vitamin C.
- Collagen synthesized during vitamin C deficiency is insufficiently hydroxylated, and therefore insufficient in strength.

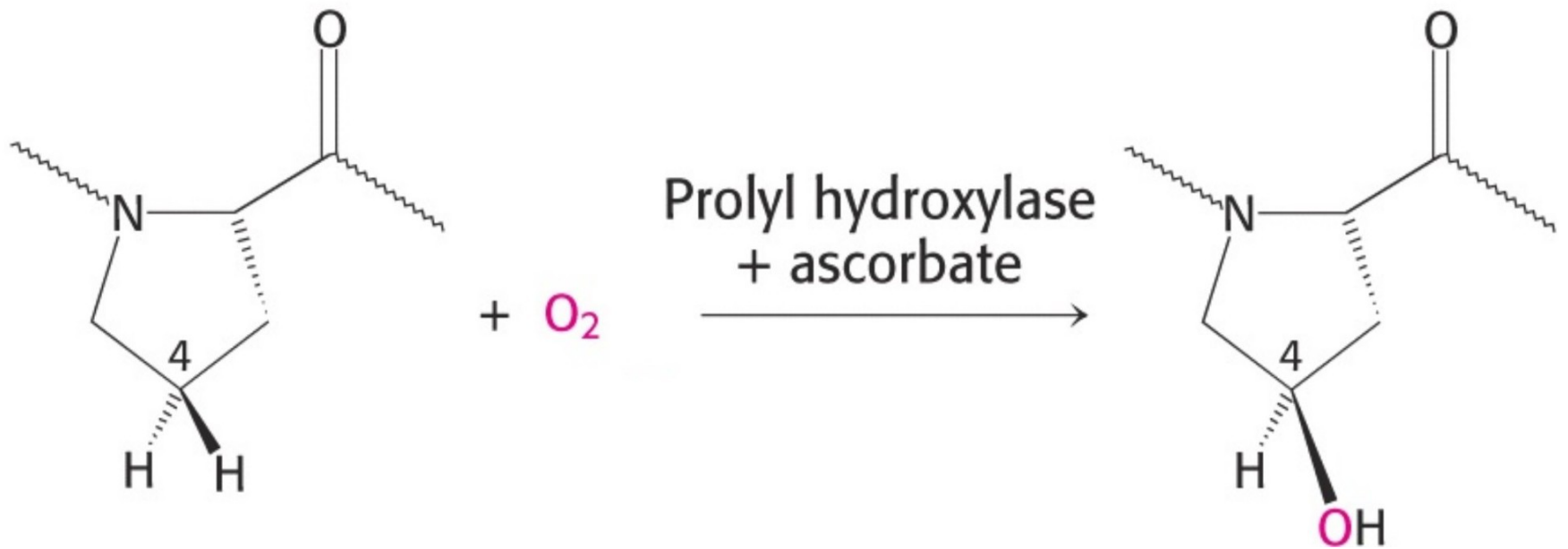


Table 48-1 (Harper)

Types of collagen and their genes

Typ	Genes	Tissues
I	COL1A1, COL1A2	Most connective tissues, including bone
II	COL2A1	Cartilage
III	COL3A1	Extensible connective tissues such as skin, lung, and the vascular system
And so on...		
cca 28		

Table 48-4. Diseases caused by mutations in **collagen genes** or by deficiencies in the activities of **posttranslational enzymes** involved in the biosynthesis of collagen

Gene or enzyme	Disease
COL1A1, COL1A2	Osteogenesis imperfecta , type 1
COL7A1	Epidermolysis bullosa
Lysyl hydroxylase	Ehlers-Danlos syndrome type VI
Procollagen N-proteinase	Ehlers-Danlos syndrome type VII