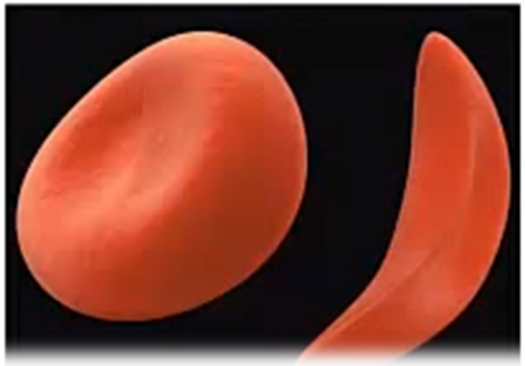
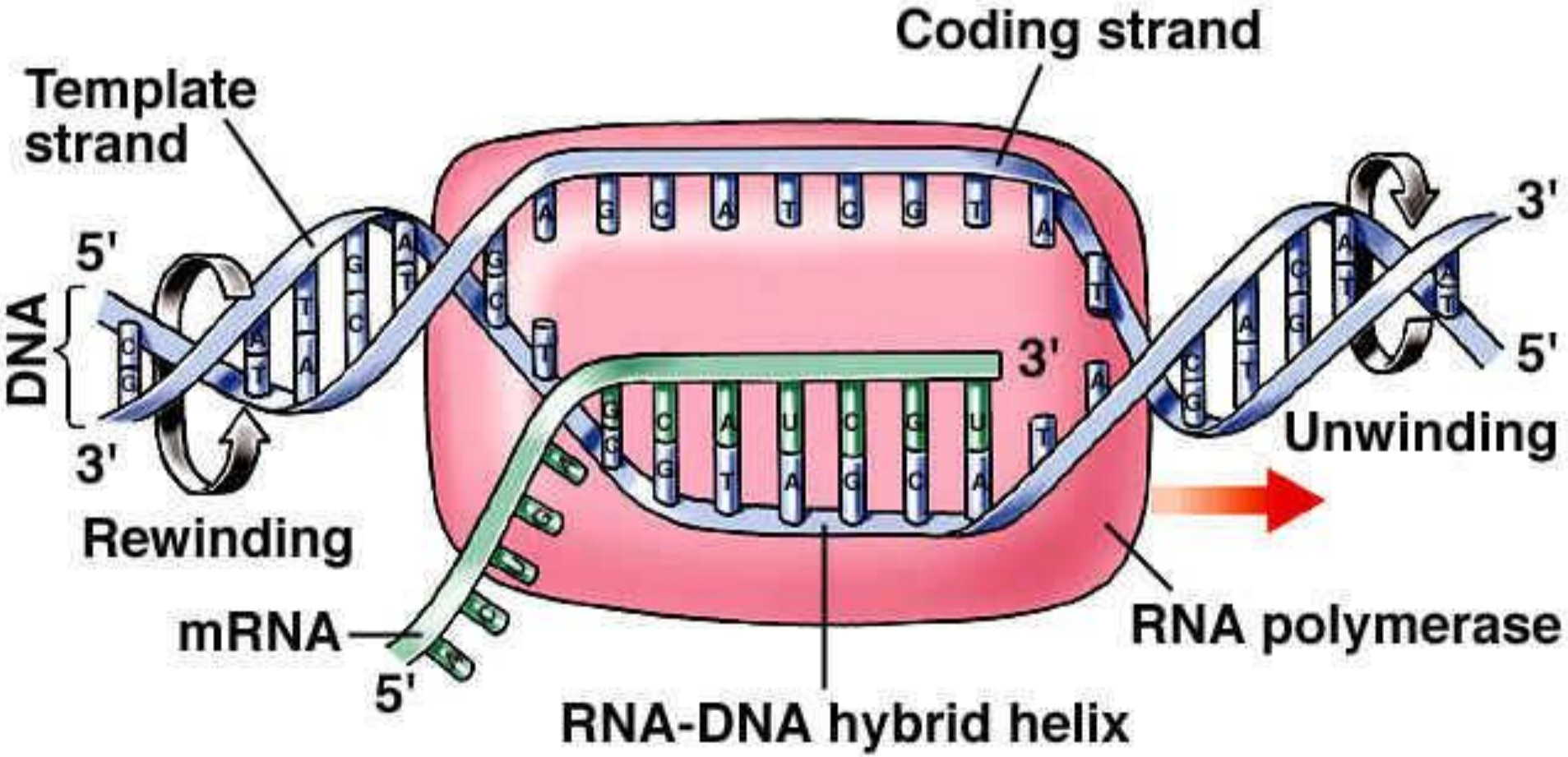
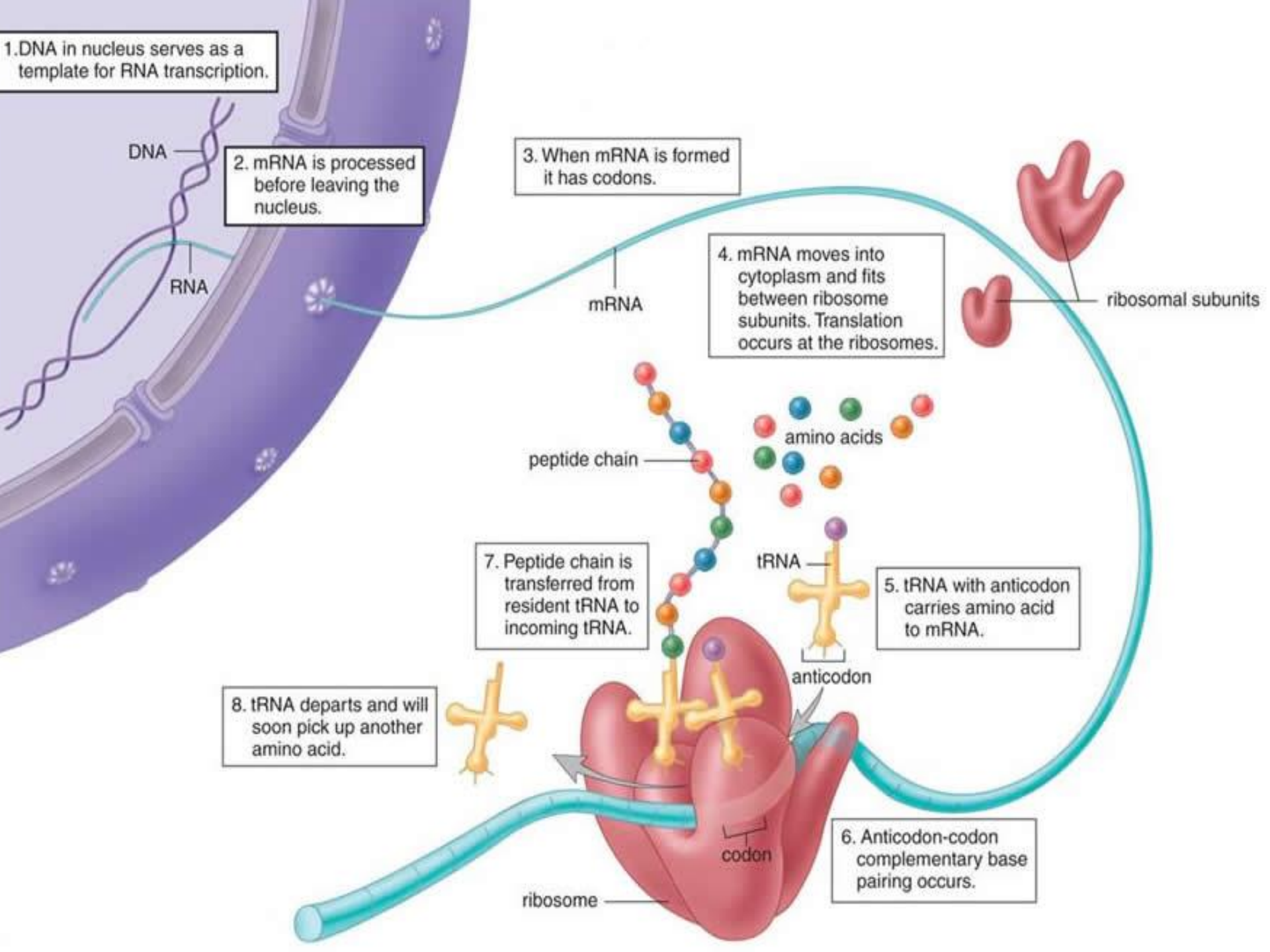
Studies of Human Hemoglobin Established That One Gene Encodes One Polypeptide

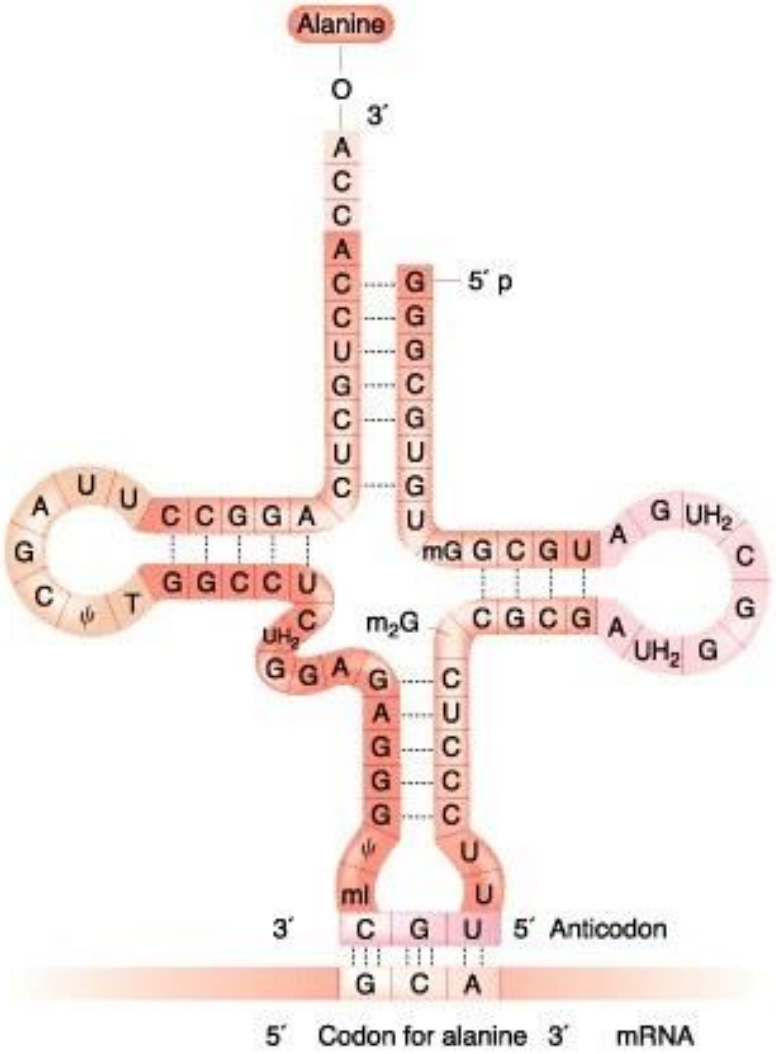
- Not all proteins are enzymes, and some proteins have more than one subunit
  - Because of this, the one-gene:one-enzyme hypothesis was modified to **one-gene:one-protein** and then to **one-gene:one-polypeptide chain**
- **Sickle-cell anemia** is a recessive genetic disease in which afflicted individuals are homozygous for the Hb^S hemoglobin allele
- Heterozygotes are carriers of the affected gene but are largely unaffected

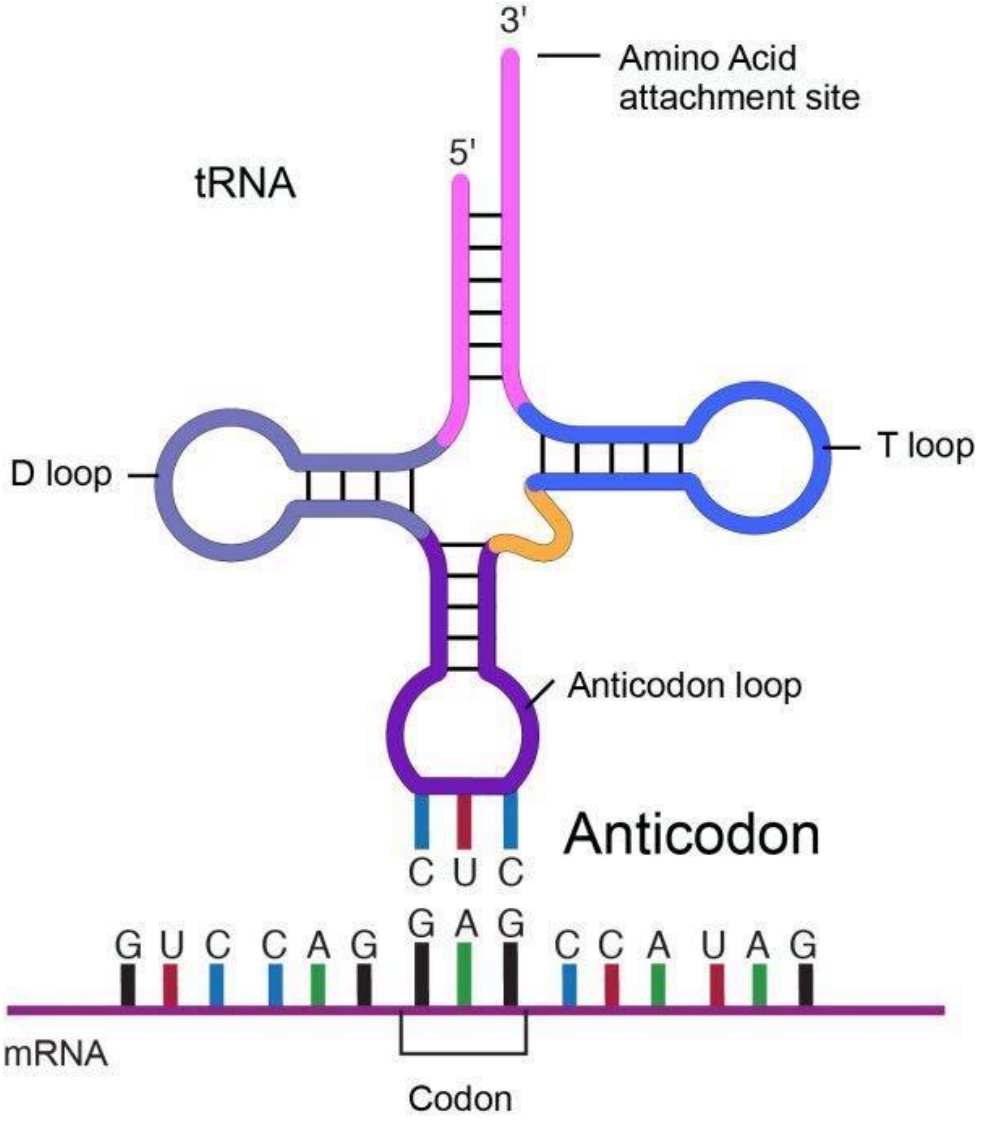


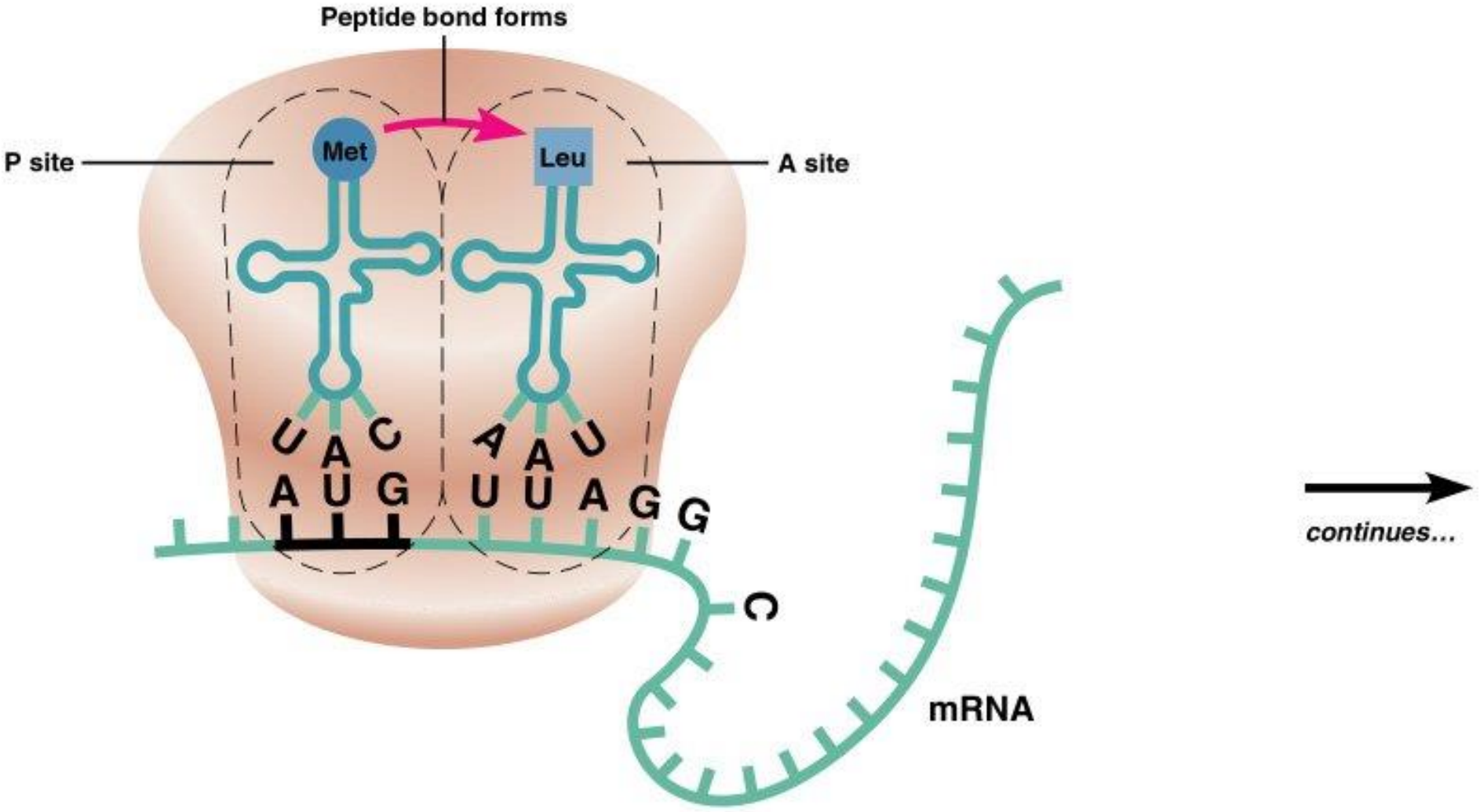
Transcription Bubble



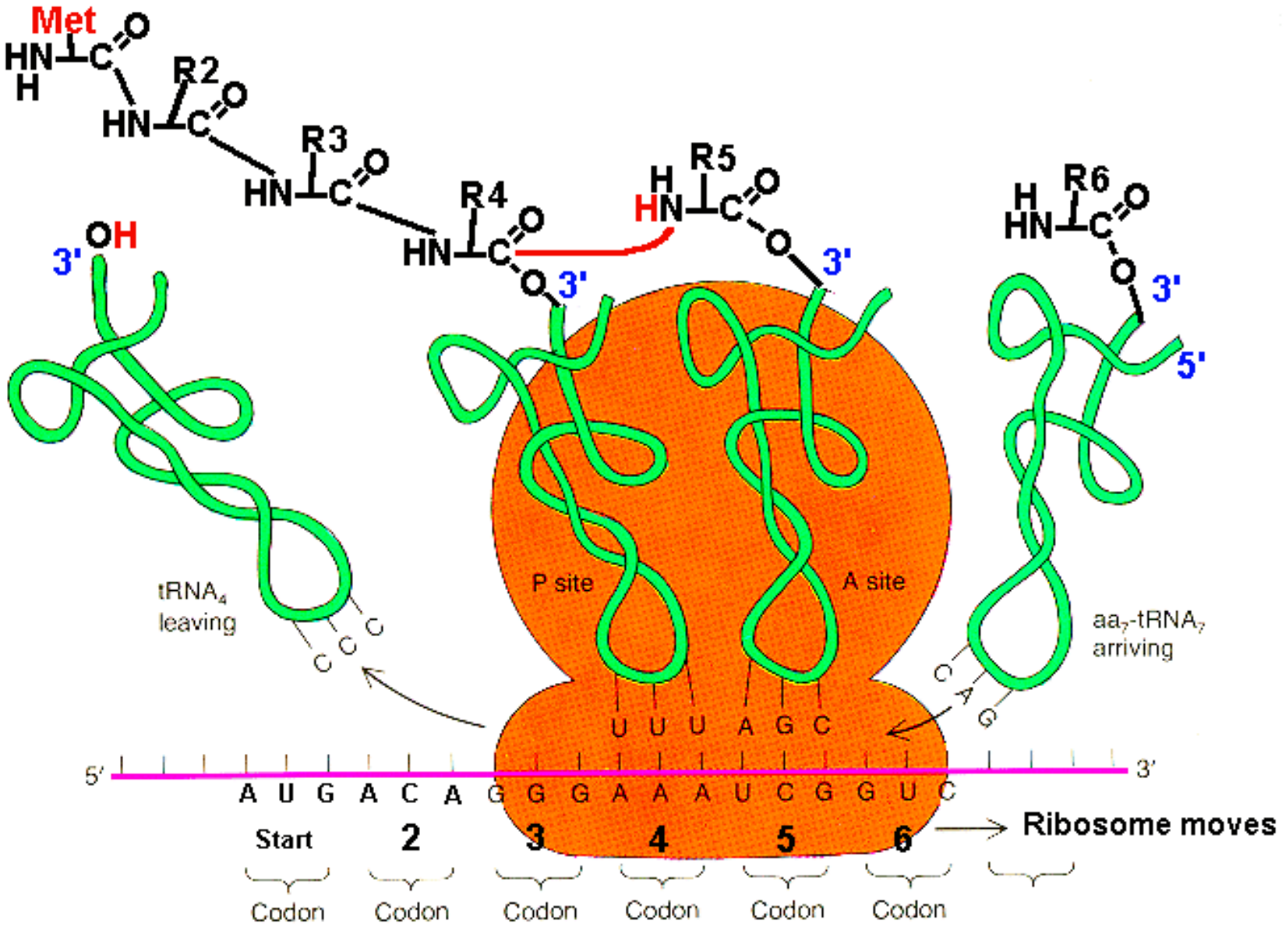

1. DNA in nucleus serves as a template for RNA transcription.
DNA
2. mRNA is processed before leaving the nucleus.
RNA
3. When mRNA is formed it has codons.
mRNA
4. mRNA moves into cytoplasm and fits between ribosome subunits. Translation occurs at the ribosomes.
ribosomal subunits
peptide chain
amino acids
tRNA
5. tRNA with anticodon carries amino acid to mRNA.
anticodon
7. Peptide chain is transferred from resident tRNA to incoming tRNA.
8. tRNA departs and will soon pick up another amino acid.
codon
6. Anticodon-codon complementary base pairing occurs.
ribosome


Alanine
3′
5′ p
mG
m2G
UH2
ml
3′
5′ Anticodon
5′ Codon for alanine 3′ mRNA


3′
Amino Acid attachment site
5′
tRNA
D loop
T loop
Anticodon loop
Anticodon
C U C
G A G
G U C C A G
C C A U A G
mRNA
Codon



3 The place on the ribosome where the first tRNA sits is called the P site. In the A site next to it, the second codon of the mRNA pairs with a tRNA carrying the second amino acid.



Modified from Griffiths et al., AN INTRODUCTION TO GENETIC ANALYSIS, 6th Ed., W.H. Freeman & Co., 1996.

1st base

2nd base	U		C		A		G		3rd base
U	UUU	Phenylalanine	UCU	Serine	UAU	Tyrosine	UGU	Cysteine	U
	UUC	Phenylalanine	UCC	Serine	UAC	Tyrosine	UGC	Cysteine	C
	UUA	Leucine	UCA	Serine	UAA	Stop	UGA	Stop	A
	UUG	Leucine	UCG	Serine	UAG	Stop	UGG	Tryptophan	G
C	CUU	Leucine	CCU	Proline	CAU	Histidine	CGU	Arginine	U
	CUC	Leucine	CCC	Proline	CAC	Histidine	CGC	Arginine	C
	CUA	Leucine	CCA	Proline	CAA	Glutamine	CGA	Arginine	A
	CUG	Leucine	CCG	Proline	CAG	Glutamine	CGG	Arginine	G
A	AUU	Isoleucine	ACU	Threonine	AAU	Asparagine	AGU	Serine	U
	AUC	Isoleucine	ACC	Threonine	AAC	Asparagine	AGC	Serine	C
	AUA	Isoleucine	ACA	Threonine	AAA	Lysine	AGA	Arginine	A
	AUG	Methionine (Start)	ACG	Threonine	AAG	Lysine	AGG	Arginine	G
G	GUU	Valine	GCU	Alanine	GAU	Aspartic Acid	GGU	Glycine	U
	GUC	Valine	GCC	Alanine	GAC	Aspartic Acid	GGC	Glycine	C
	GUA	Valine	GCA	Alanine	GAA	Glutamic Acid	GGA	Glycine	A
	GUG	Valine	GCG	Alanine	GAG	Glutamic Acid	GGG	Glycine	G

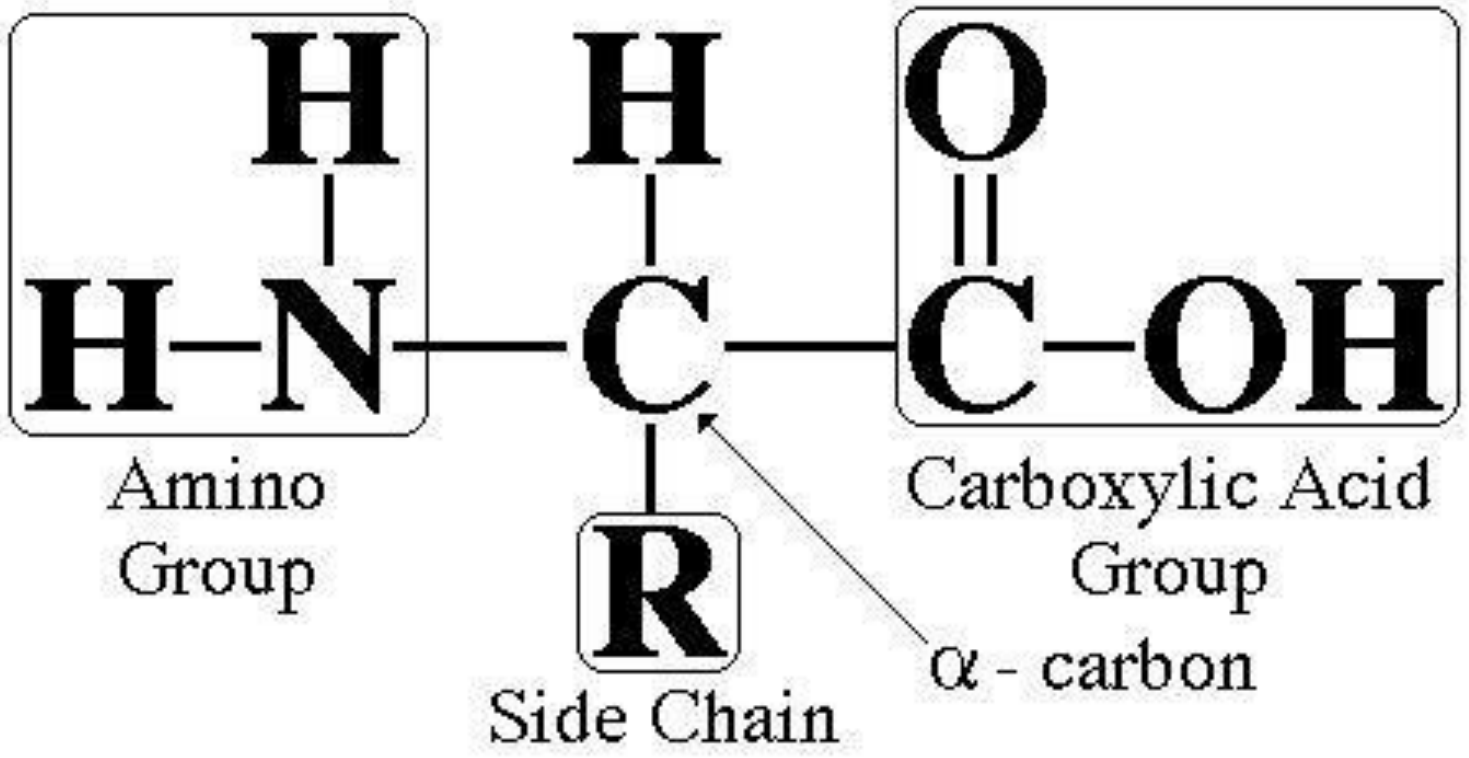
Nonpolar, aliphatic Polar, uncharged Aromatic Positively charged Negatively charged

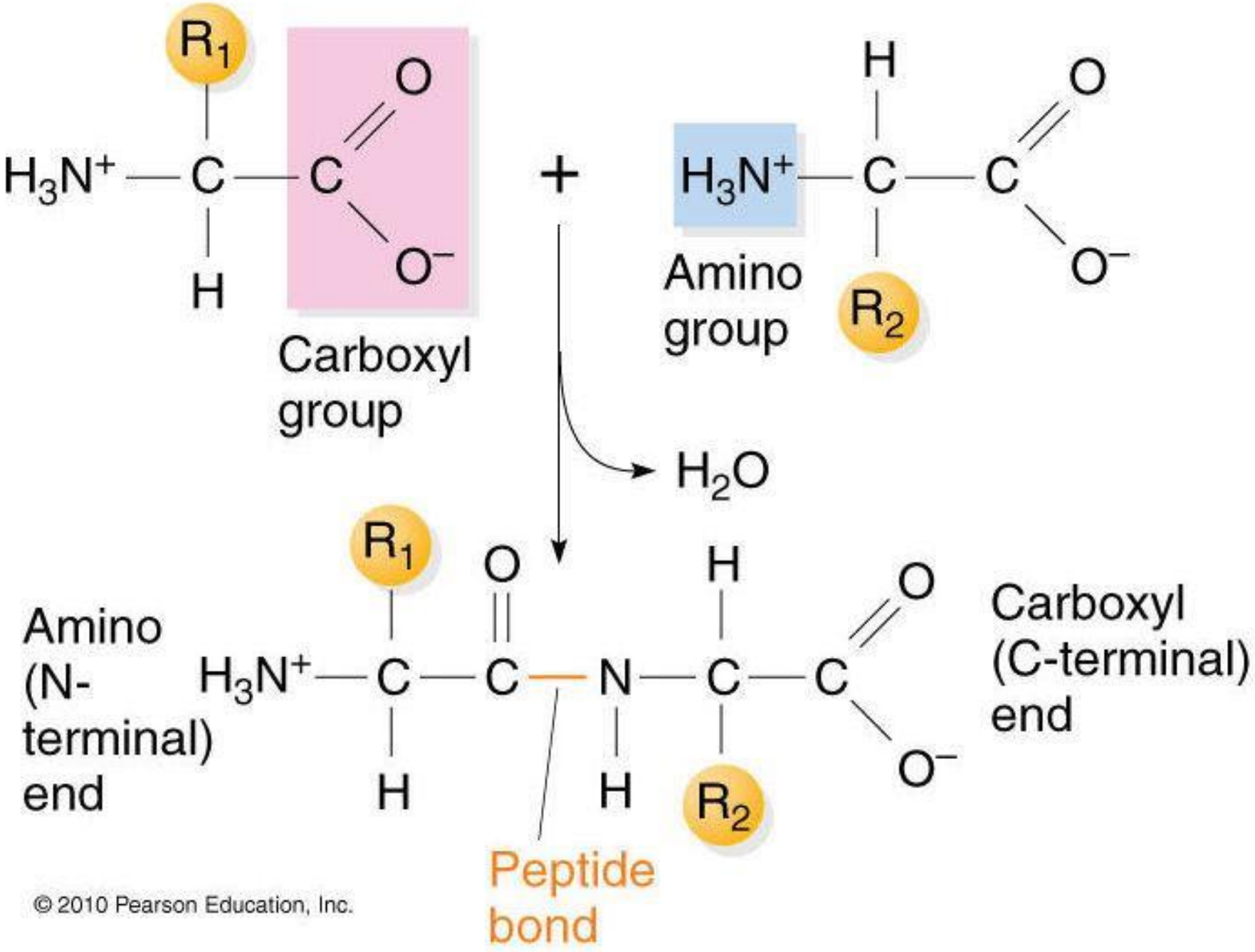
TABLE 6-1 Amino Acids

Proteins are made up of about 20 common amino acids. The first column lists the essential amino acids for human beings (those the body cannot make—that must be provided in the diet). The second column lists the nonessential amino acids. In special cases, some nonessential amino acids may become conditionally essential (see the text). In a newborn, for example, only five amino acids are truly nonessential; the other nonessential amino acids are conditionally essential until the metabolic pathways are developed enough to make those amino acids in adequate amounts.

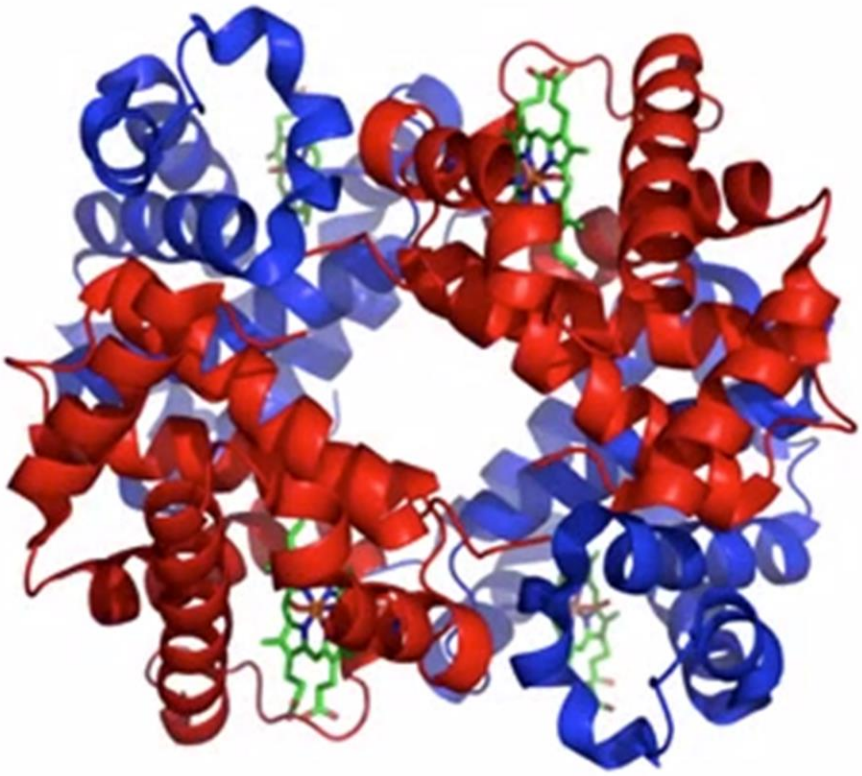
Essential Amino Acids		Nonessential Amino Acids	
Histidine	(HISS-tuh-deen)	Alanine	(AL-ah-neen)
Isoleucine	(eye-so-LOO-seen)	Arginine	(ARJ-ih-neen)
Leucine	(LOO-seen)	Asparagine	(ah-SPAR-ah-geen)
Lysine	(LYE-seen)	Aspartic acid	(ah-SPAR-tic acid)
Methionine	(meh-THIGH-oh-neen)	Cysteine	(SIS-teh-een)
Phenylalanine	(fen-il-AL-ah-neen)	Glutamic acid	(GLU-tam-ic acid)
Threonine	(THREE-oh-neen)	Glutamine	(GLU-tah-meen)
Tryptophan	(TRIP-toe-fan,	Glycine	(GLY-seen)
	TRIP-toe-fane)	Proline	(PRO-leen)
Valine	(VAY-leen)	Serine	(SEER-een)
		Tyrosine	(TIE-roe-seen)

Amino Acid Structure



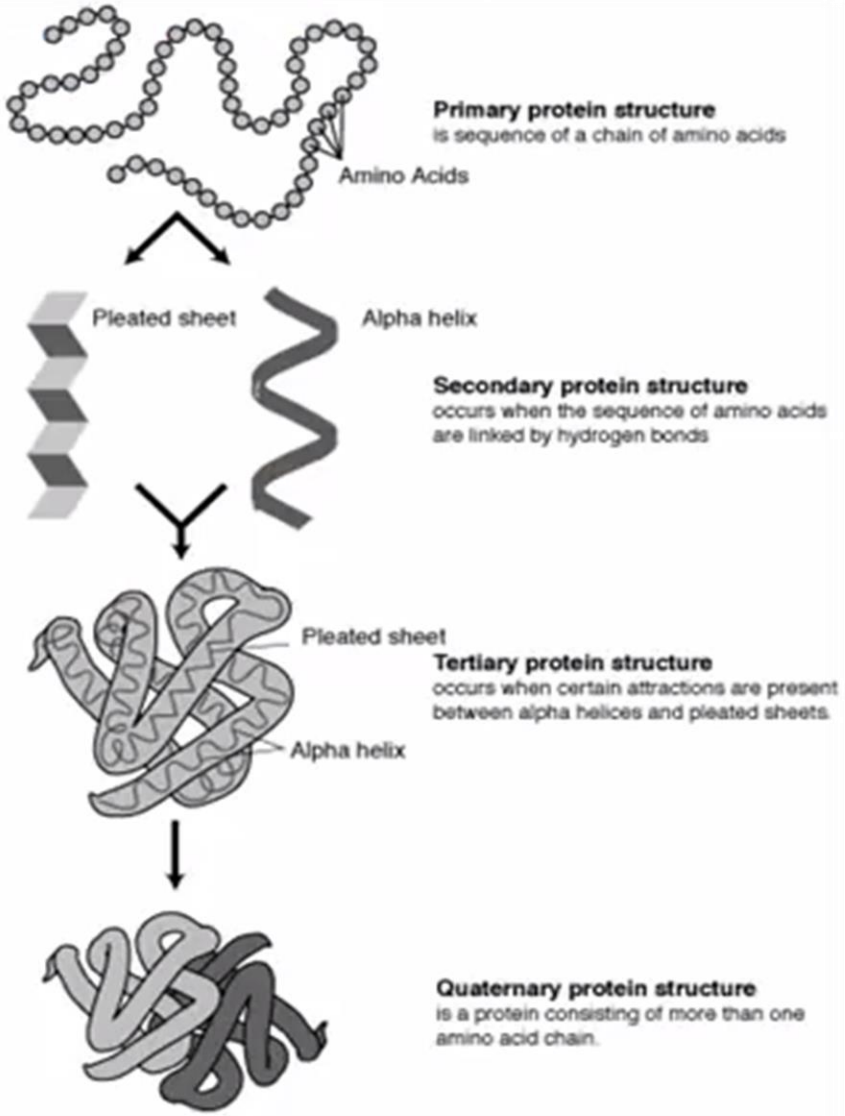


4 levels of structure are used to describe proteins



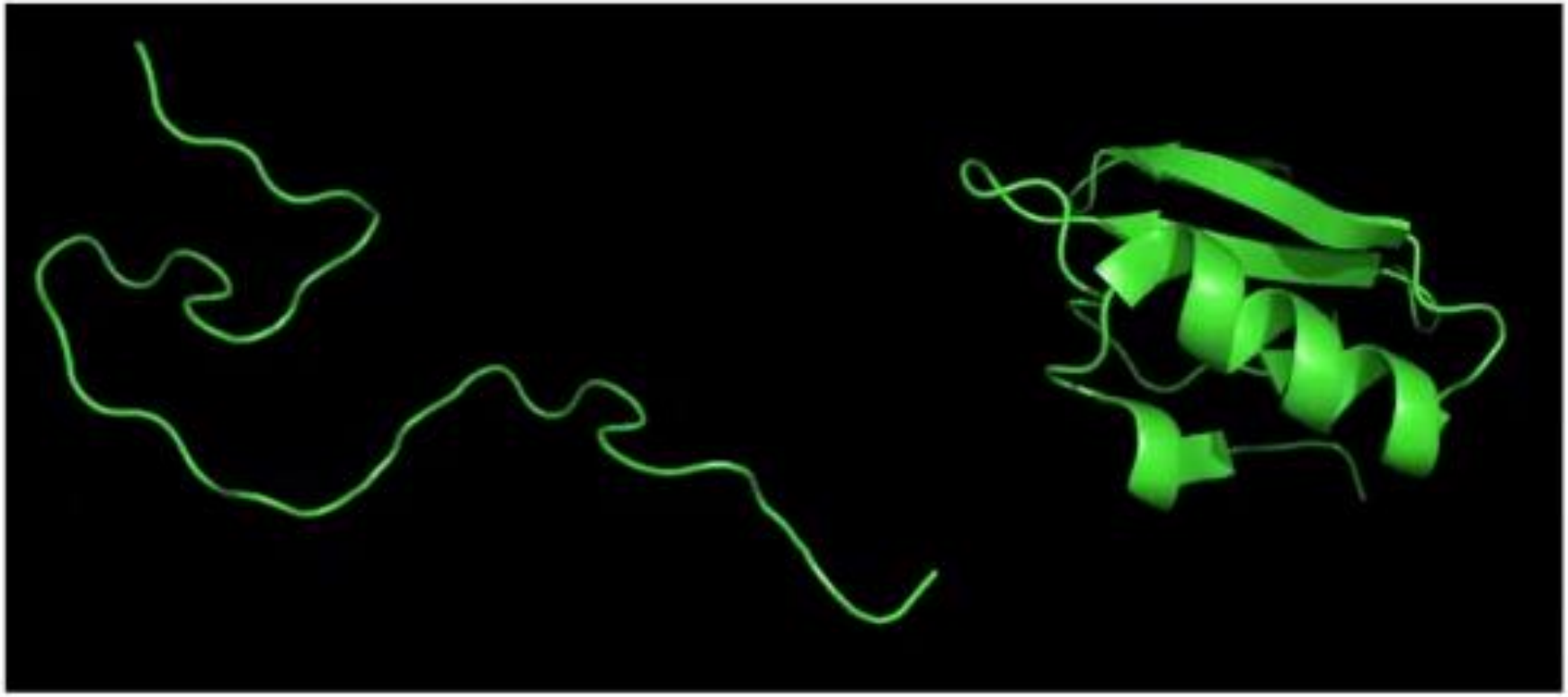
The bonds that hold protein structure together

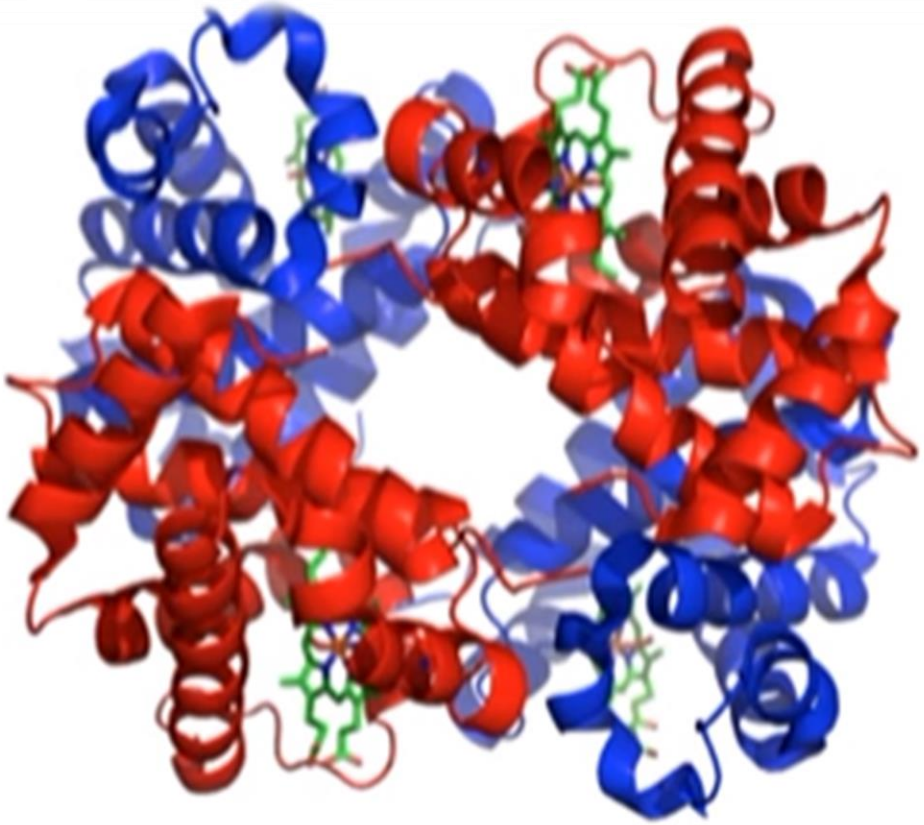
- Primary structure:
  - peptide bonds
- Secondary structure:
  - hydrogen bonds
- Tertiary structure:
  - hydrogen bonds
  - ionic bonds
  - van der Waals interactions
  - disulfide bridges



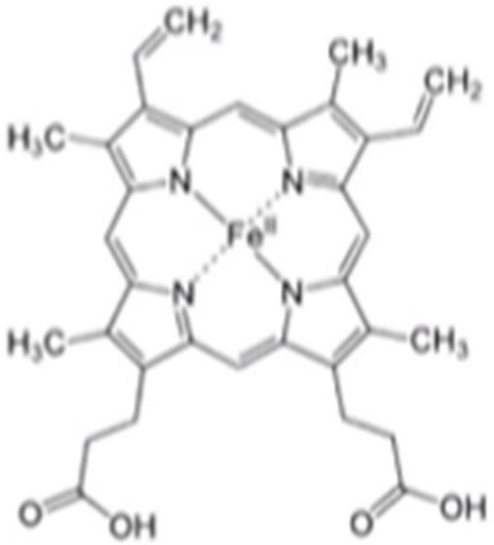
Polypeptide

Protein



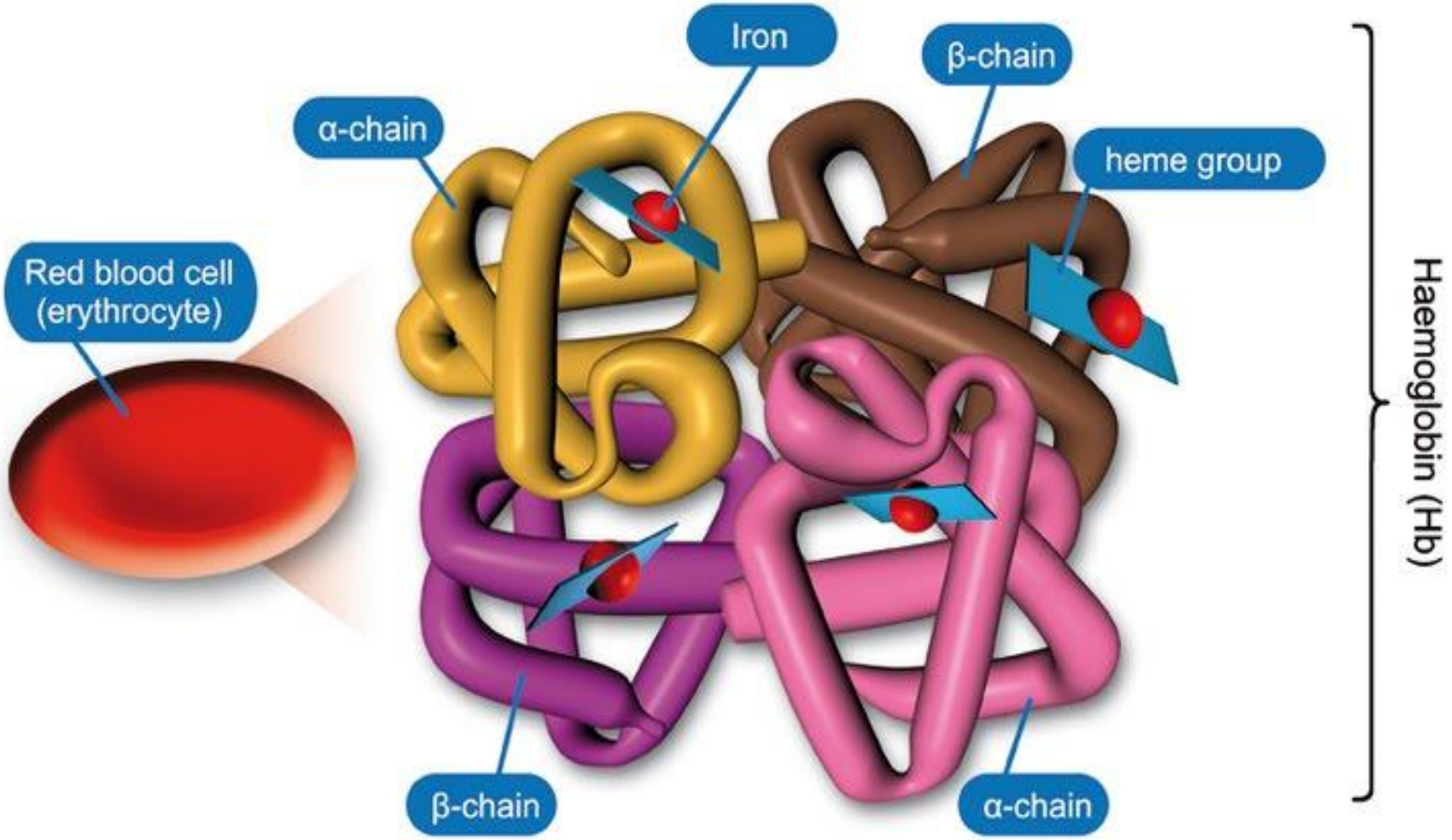


Quaternary structure of hemoglobin



Heme group

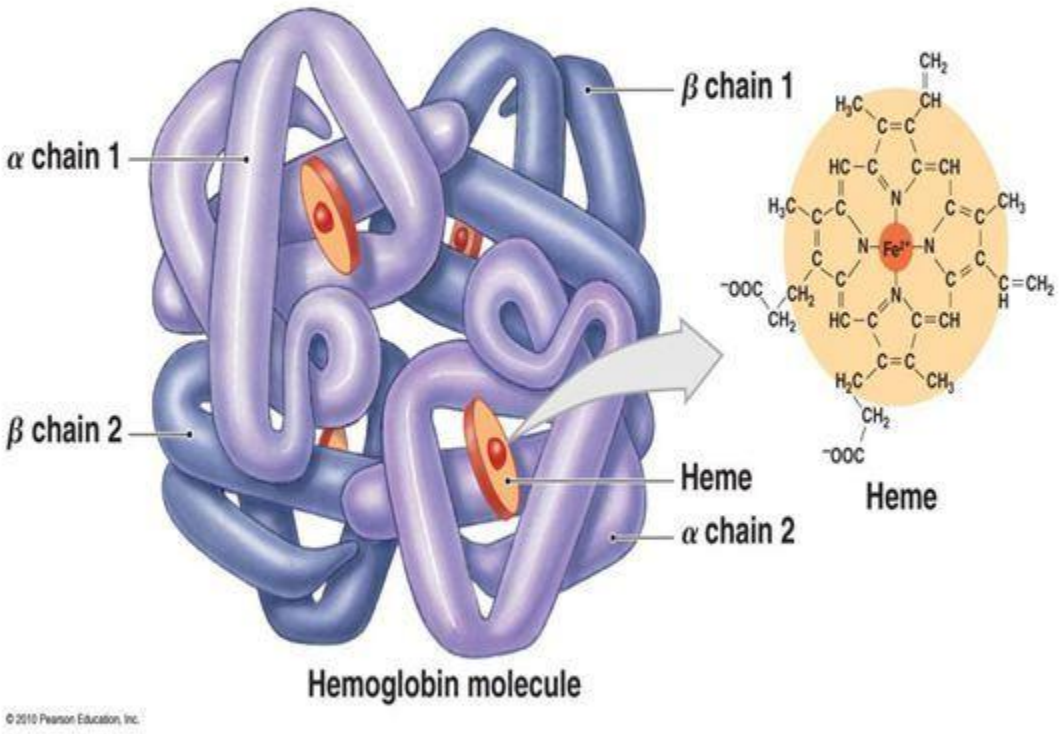
Structure of haemoglobin



Each erythrocyte (RBC) contains ~270 million haemoglobin molecules

Hemoglobin structure

- 4 polypeptide chains (2 alpha and 2 beta chains)
- Each chain has an associated heme group
- Each heme group has a central iron atom
  - Serves as the binding site for oxygen molecule



Hemoglobin A (HbA), also known as adult hemoglobin, **hemoglobin A1 or α2β2,** is the most common human hemoglobin tetramer, comprising over 97% of the total red blood cell hemoglobin. It consists of **two alpha chains and two beta chains**

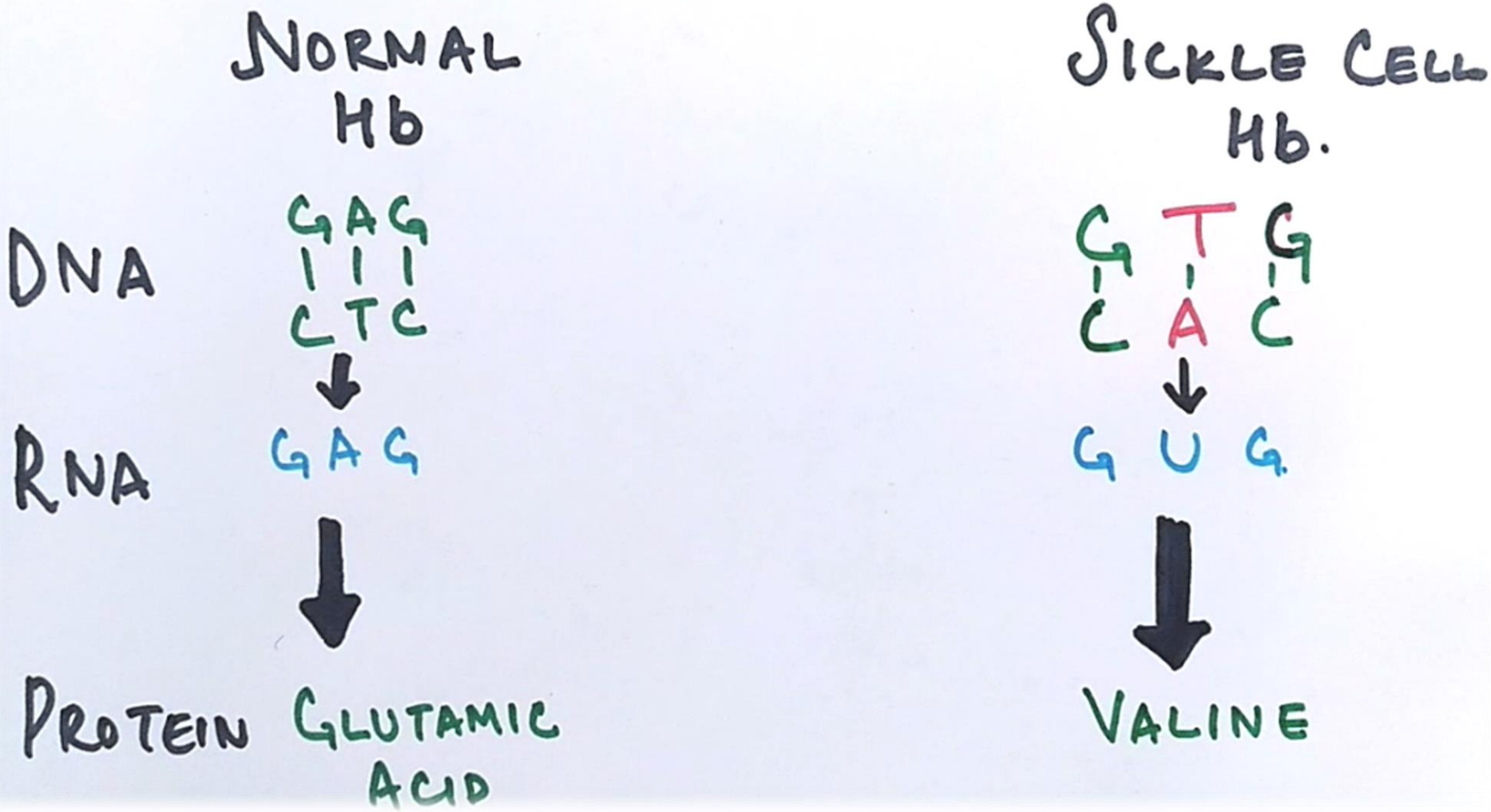
Hemoglobin variants:

Hb A1C
Hemoglobin A2
Hemoglobin C
Hemoglobin F

Hemoglobin protein subunits (genes):

Alpha globin 1
Alpha globin 2
Beta globin
Delta globin

MOLECULAR LEVEL



Primary structure (amino acid sequence)

The haemoglobin molecule consists of 4 polypeptide (globin) chains. In adults there are 2 alpha chains and 2 beta chains.

alpha chain (141 amino acid residues):

val - leu ser pro ala asp lys thr asn val lys ala ala try gly lys val gly ala his ala gly glu tyr gly ala glu ala leu glu arg met phe leu ser phe pro thr thr lys thr tyr phe pro his phe - asp leu ser his gly ser ala - - - - - gln val lys gly his gly lys lys val ala asp ala leu thr asn ala val ala his val asp asp met pro asn ala leu ser ala leu ser asp leu his ala his lys leu arg val asp pro val asp phe lys leu leu ser his cys leu leu val thr leu ala ala his leu pro ala glu phe thr pro ala val his ala ser leu asp lys phe leu ala ser val ser thr val leu thr ser lys tyr arg

beta chain (146 amino acid residues)

val his leu thr pro glu glu lys ser ala val thr ala leu try gly lys val asn - - val asp glu val gly gly glu ala leu gly arg leu leu val val tyr pro try thr gln arg phe phe glu ser phe gly asp leu ser thr pro asp ala val met gly asn pro lys val lys ala his gly lys lys val leu gly ala phe ser asp gly leu ala his leu asp asn leu lys gly thr phe ala thr leu ser glu leu his cys asp lys leu his val asp pro glu asn phe arg leu leu gly asn val leu val cys val leu ala his his phe gly lys glu phe thr pro pro val gln ala ala tyr gln lys val val ala gly val ala asp ala leu ala his lys tyr his

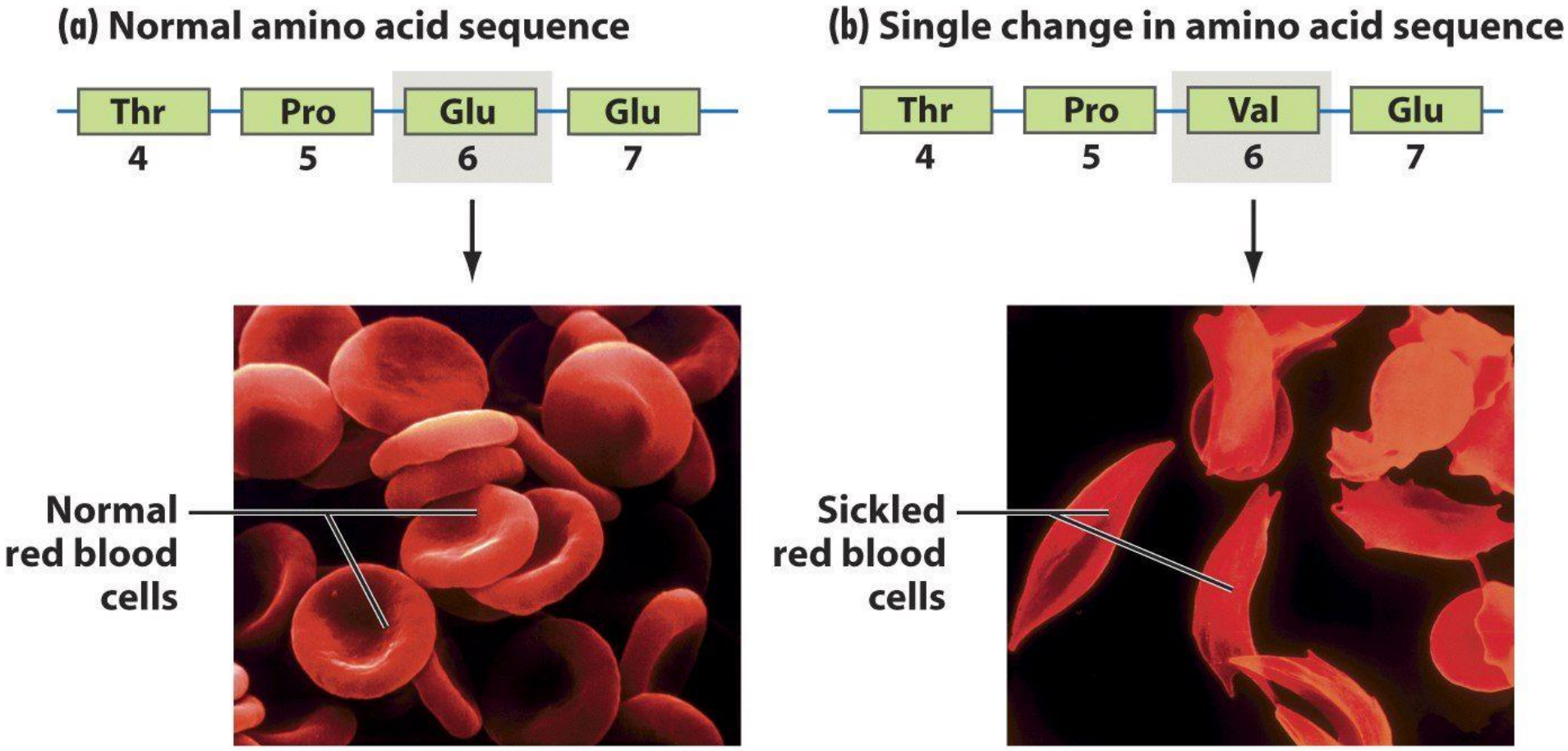
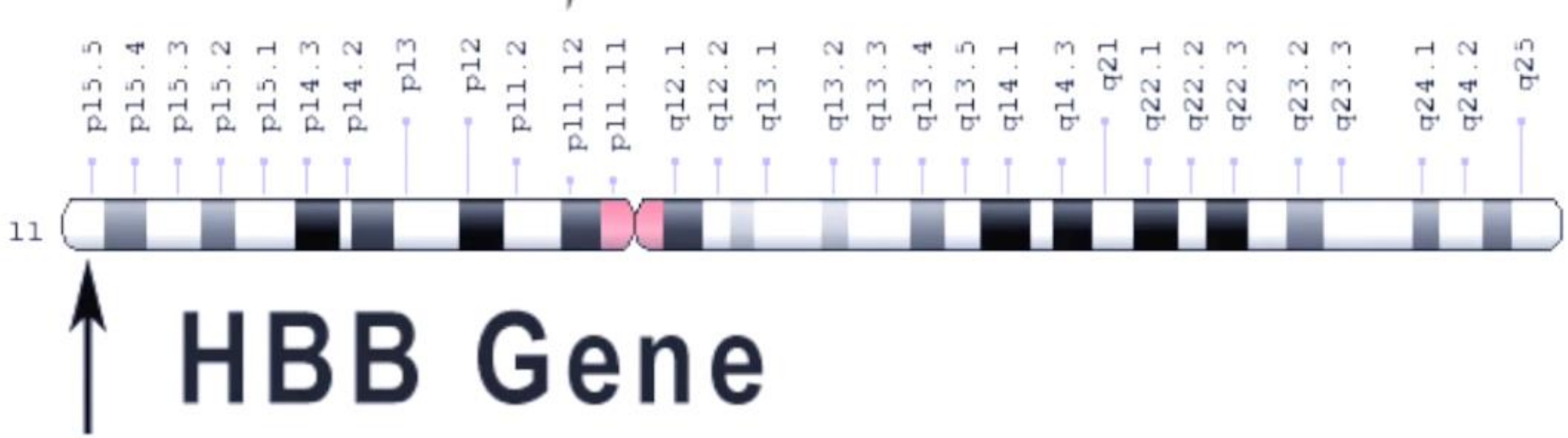


Figure 3-13 Biological Science, 2/e



Beta globin (also referred to as **HBB**, β-globin, haemoglobin beta, hemoglobin beta, or preferably haemoglobin subunit beta) is a globin protein, which along with **alpha globin (HBA),** makes up the most common form of haemoglobin in adult humans, the **HbA.** It is 146 amino acids long and has a molecular weight of 15,867 Da. Normal adult human HbA is a heterotetramer consisting of two alpha chains and two beta chains.

HBB is encoded by the HBB gene on human chromosome 11. Mutations in the gene produce several variants of the proteins which are implicated with genetic disorders such as **sickle-cell disease**

Sickle cell disease

- More than a thousand naturally occurring HBB variants have been discovered. The most common is **HbS, which causes sickle cell disease.**

- HbS is produced by a **point mutation in HBB** in which the codon **GAG is replaced by GTG.**

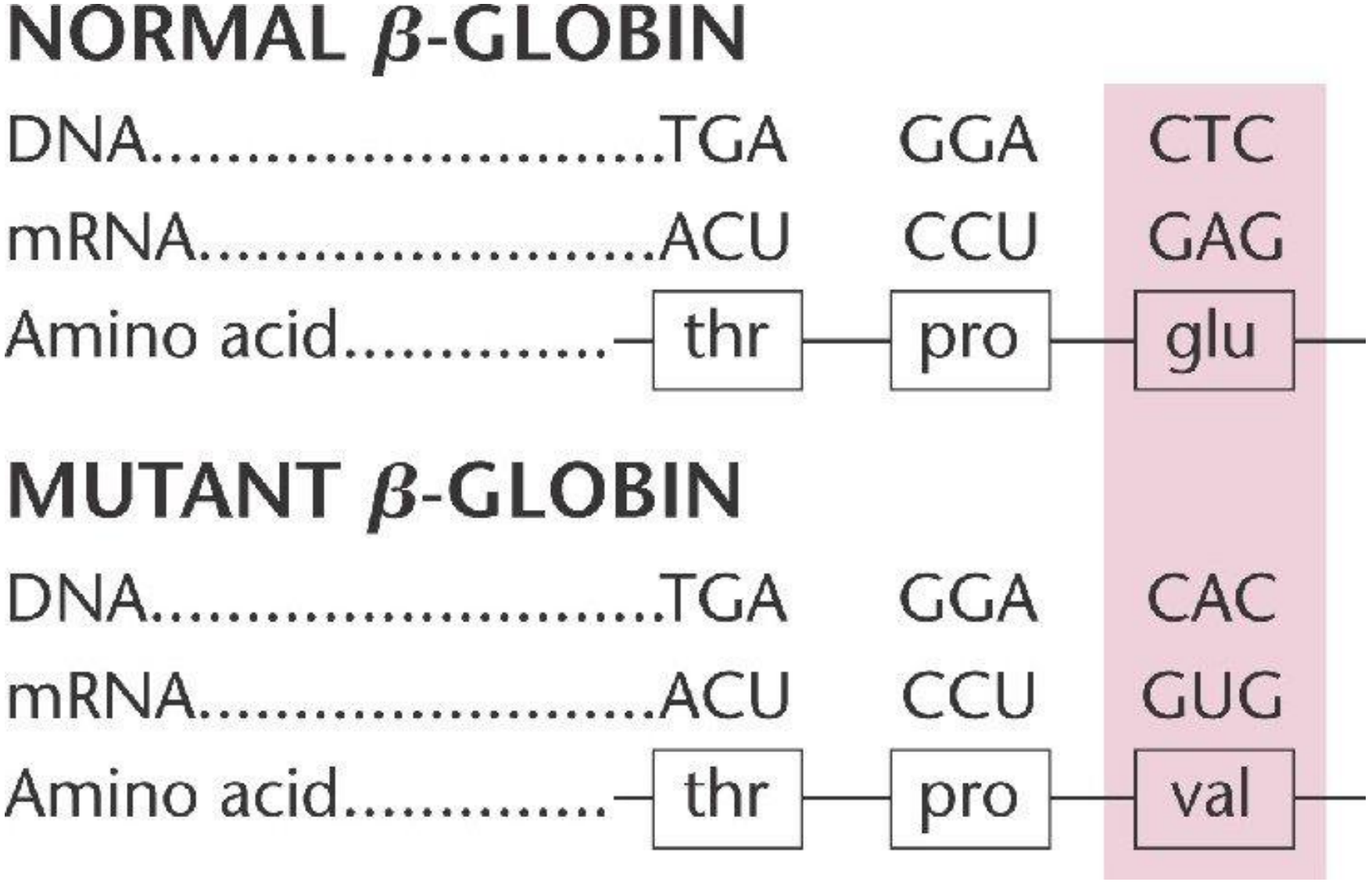
- This results in the replacement of **hydrophilic amino acid glutamic acid with the hydrophobic amino acid valine at the sixth position (β6Glu→Val).**

- This substitution creates a hydrophobic spot on the outside of the protein that sticks to the hydrophobic region of an adjacent haemoglobin molecule's beta chain.

- This further causes clumping of HbS molecules into rigid fibers, causing "sickling" of the entire red blood cells in the homozygous **(HbS/HbS)** condition.

- The homozygous allele has become one of the deadliest genetic factors.

- Whereas, people heterozygous for the mutant allele **(HbS/HbA)** are resistant to malaria and develop minimal effects of the anaemia.


NORMAL β-GLOBIN
DNA.....TGA GGA CTC
mRNA.....ACU CCU GAG
Amino acid.....
thr
pro
glu
MUTANT β-GLOBIN
DNA.....TGA GGA CAC
mRNA.....ACU CCU GUG
Amino acid.....
thr
pro
val

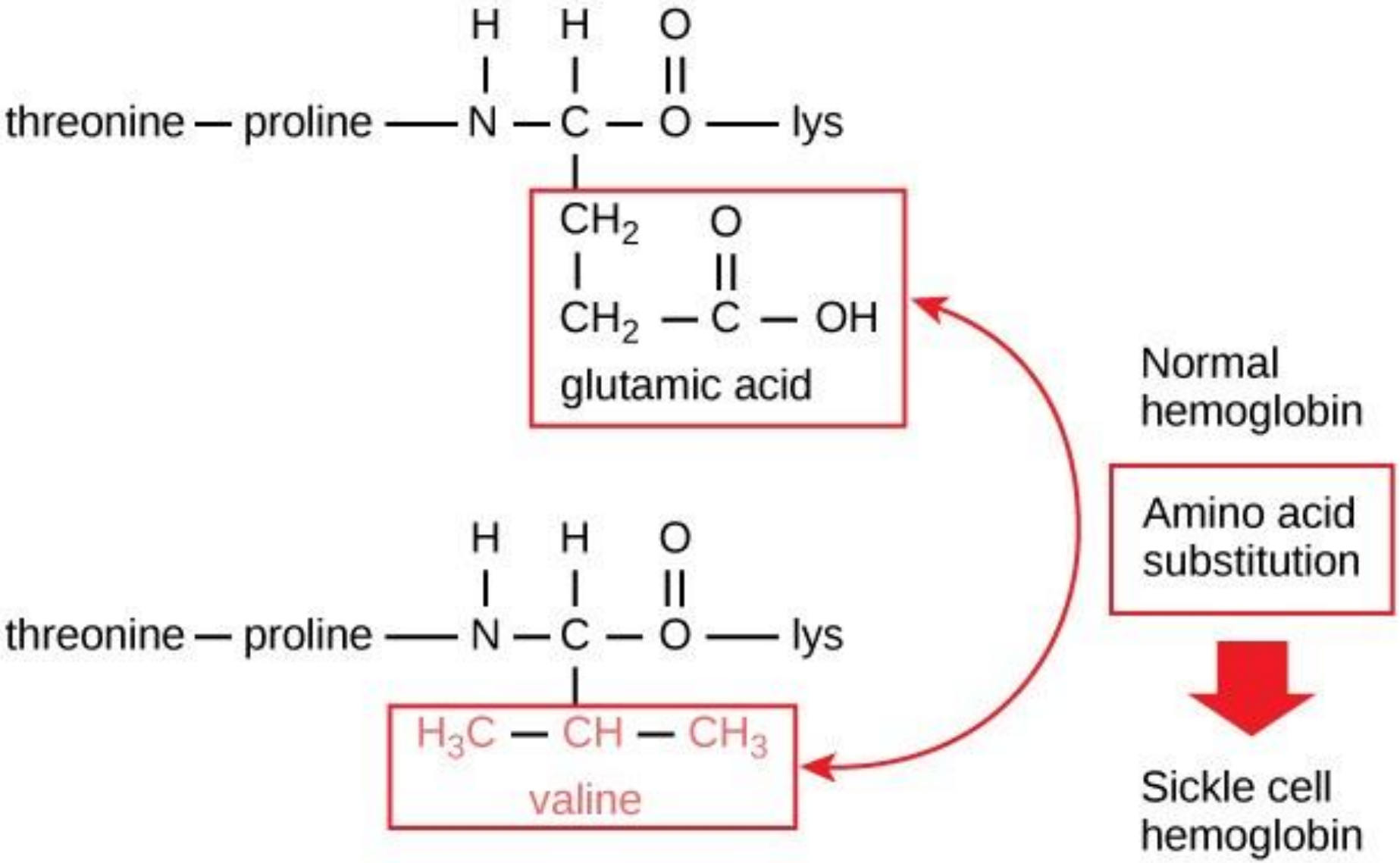
1. MUTATION IN HBB GENE.

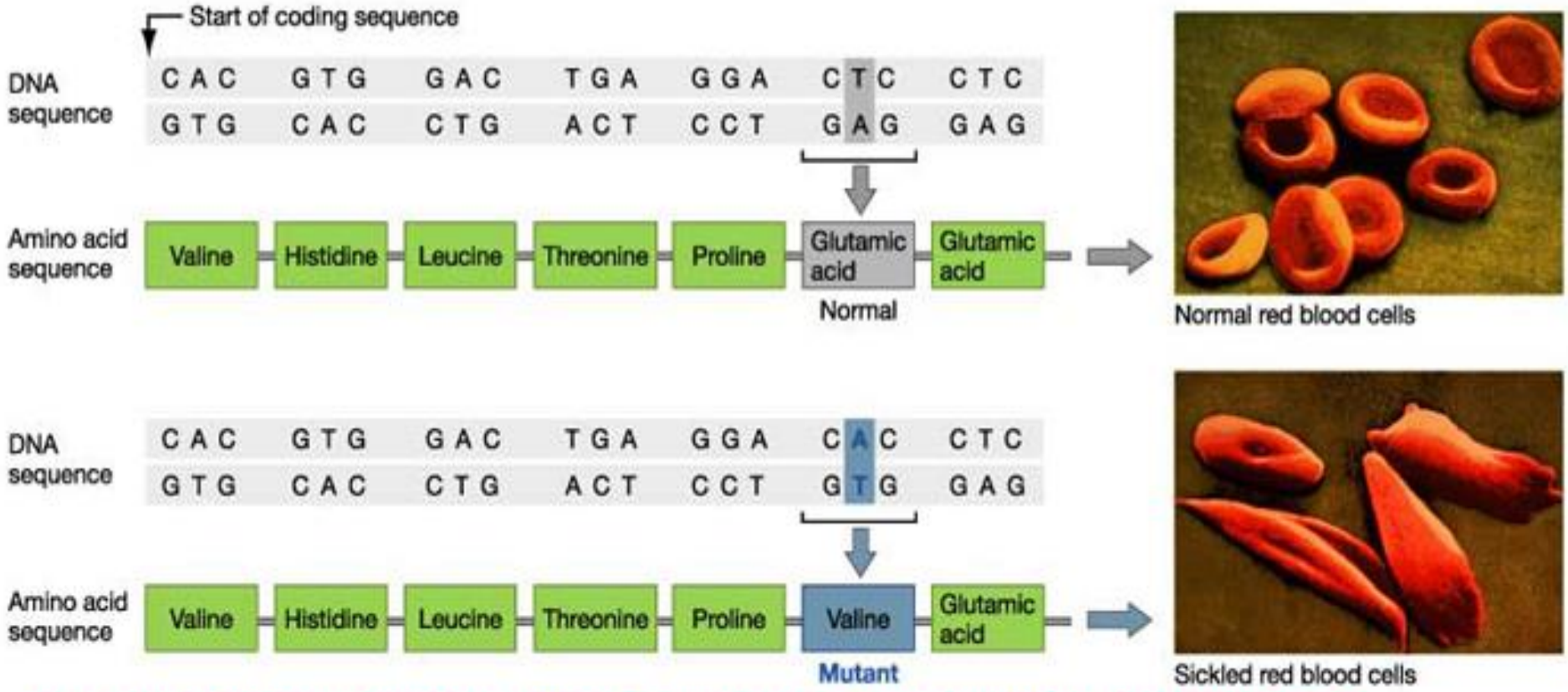
2. POINT MUTATION.

NORMAL HB. (HBN)

DEFECTIVE HB (HBS)

- VALINE : HYDROPHOBIC


threonine — proline — N — C — O — lys
CH2
O
CH2 — C — OH
glutamic acid
Normal hemoglobin
Amino acid substitution
threonine — proline — N — C — O — lys
H3C — CH — CH3
valine
Sickle cell hemoglobin



The change in amino acid sequence causes hemoglobin molecules to crystallize when oxygen levels in the blood are low. As a result, red blood cells sickle and get stuck in small blood vessels.

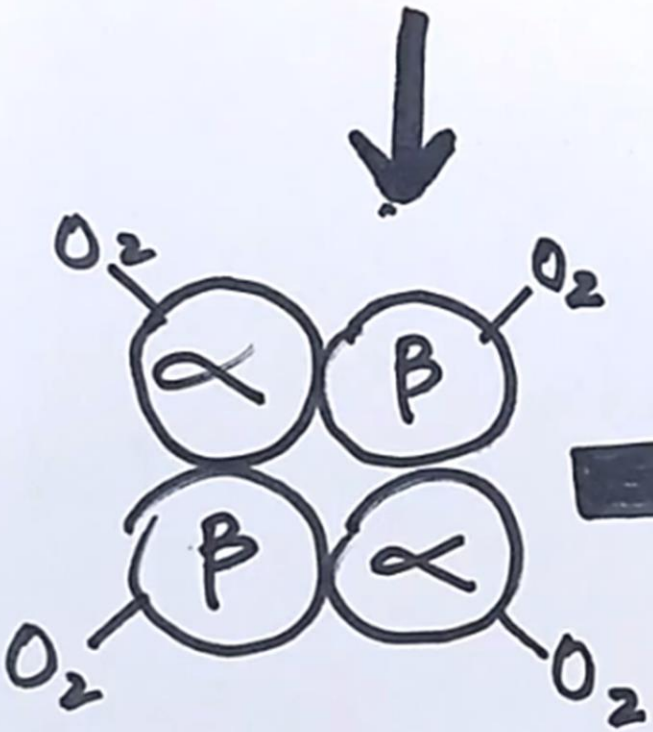
CHROMOSOME NO. 11.

SHORT ARM POSITION 15.5.

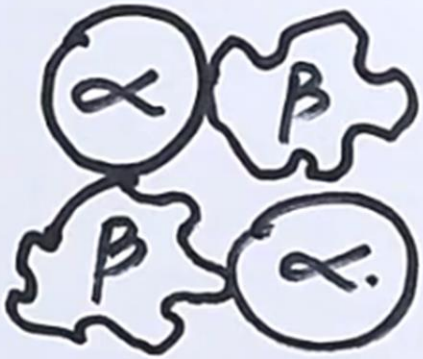
β-GLOBIN LOCUS ON IT.

PRODUCES HBB-PROTEIN.

TRANSPORTS O_2 VERY WELL



RELEASES O_2



TACTOIDS.

TACTOIDS

↓

CRYSTALIZATION OF HbS

↓

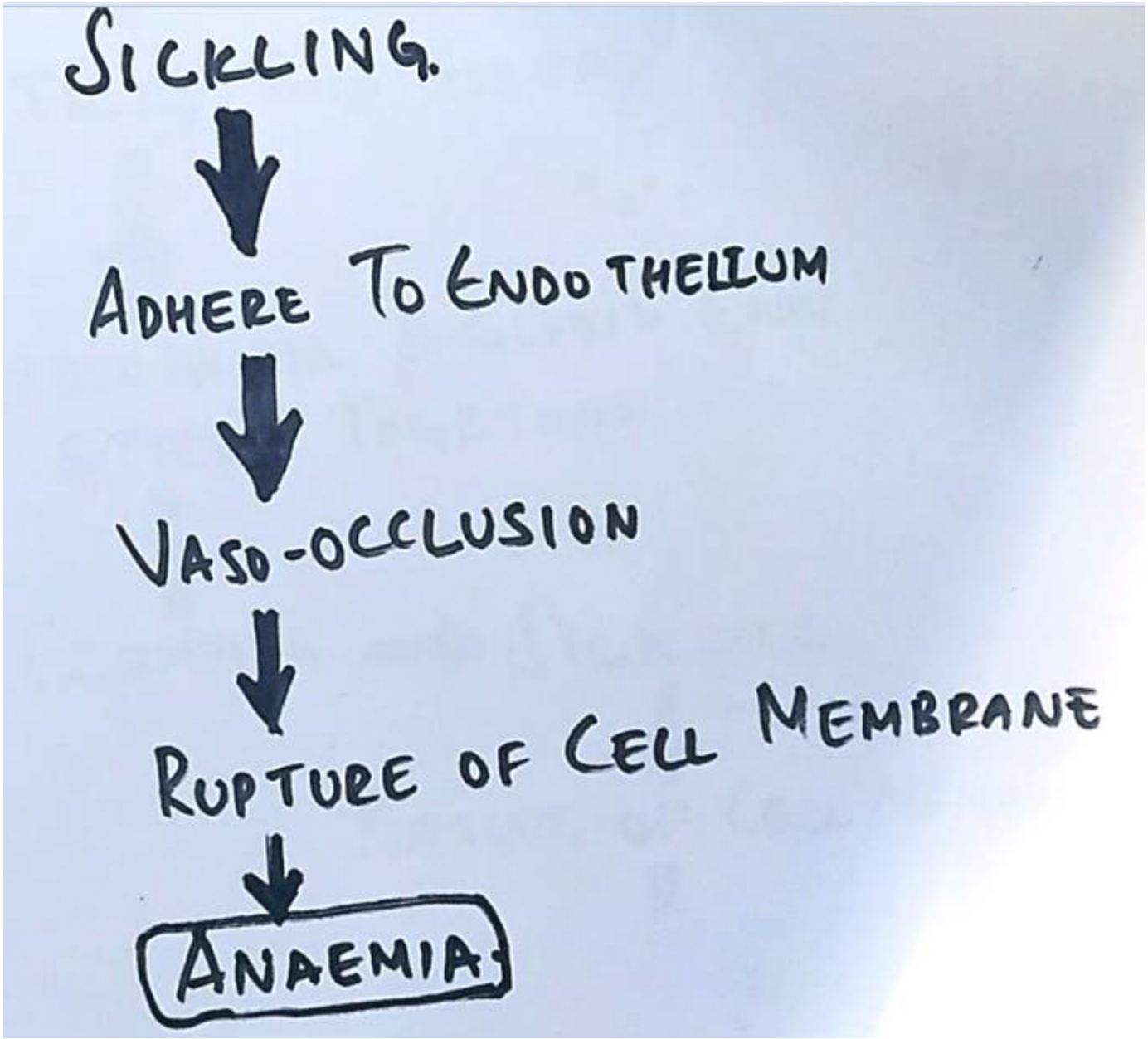
STICKY ENDS

↓

HYDROPHOBIC β-GLOBIN CHAINS WITH OTHER HbS MOLECULES

↓

CLUMPING → SICKLING.


SICKLING.
ADHERE TO ENDOTHELIUM
VASO-OCCLUSION
RUPTURE OF CELL MEMBRANE
ANAEMIA

http://www.biotopics.co.uk/as/haemoglobinproteinstructure.html