ملخص محاضرات في الكيمياء الحياتية

References

- Principles of Biochemistry by Lehninger
- Biochemistry by
  Donald Voet & Judith Voet
- Some Arabic Biochemistry Books by

د . النجفي سعيد طلال او فليح ال خوله

Cells, Water, and Buffers

Cells: The Bio of Biochemistry

What is Biochemistry?

It is the study of chemical processes within and relating to, living organisms.

Where?

Biochemistry happens inside organisms.

Water, Everywhere

- Vital for life, water is by far the most abundant component of every cell.

- Everything that happens in cells, reactions buried deep inside by enzymes, away from water, still are influenced by water.

Buffers Keep the Cellular Environment Stable

- Water can ionize to form H+ (proton) and OH- (hydroxide).
- We measure the proton concentration of a solution with pH.

$$pH = -Log[H+]$$

- Measure the hydroxide concentration with the pOH by the parallel equation,

$$pOH = -Log[OH-]$$

- In pure water, dissociation of a proton from it creates a hydroxide,
- So the pOH of pure water is 7, as well. This also means that

$$pH + pOH = 14$$

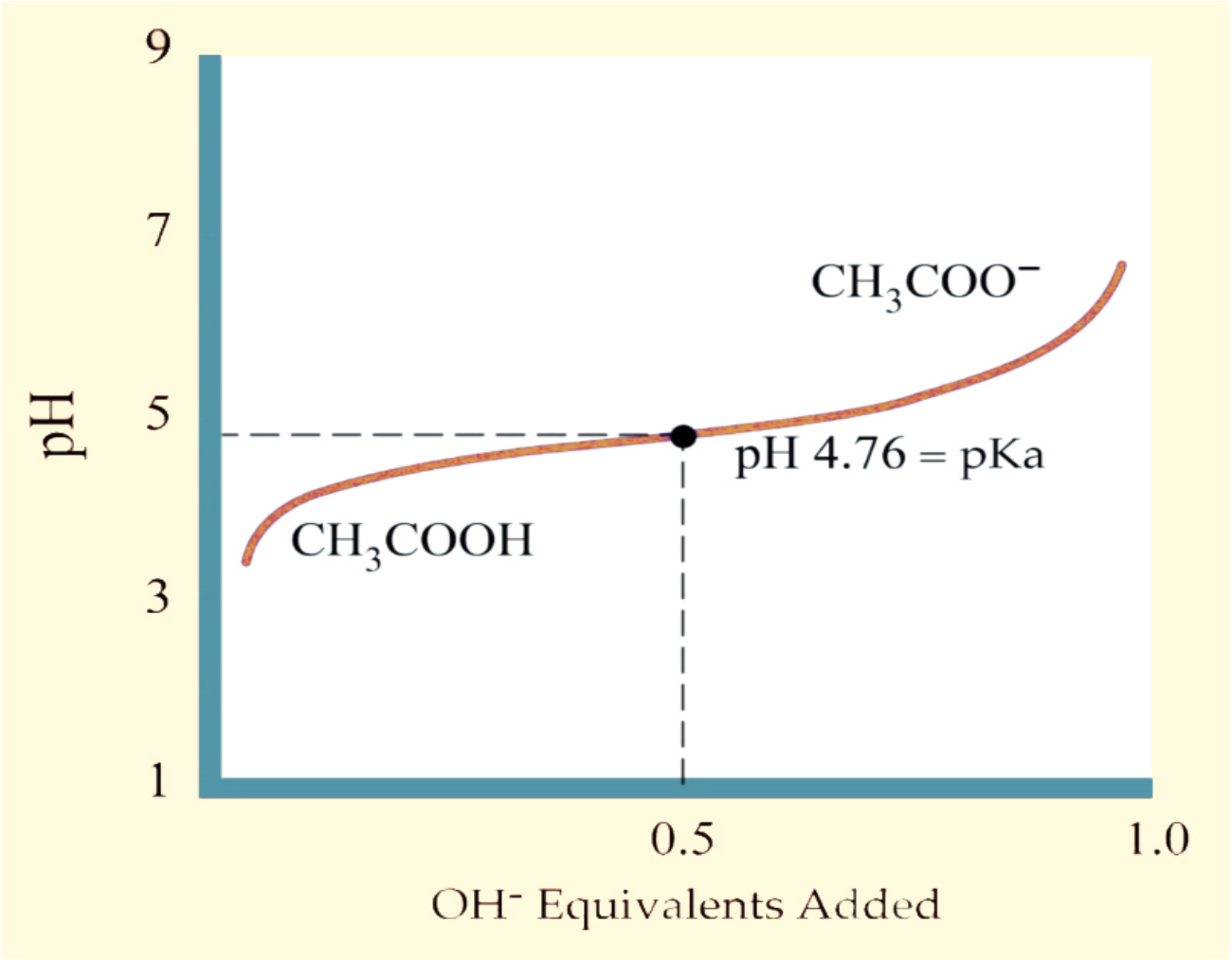
Henderson-Hasselbalch

- The Henderson-Hasselbalch equation defines the relationship between pH and the ratio of Ac and HAc. It is as follows

$$pH = pKa + \log([Ac^-]/[HAc])$$

- It is useful to be able to predict the response of the HAc system to changes in H+ concentration.

An example of buffer system - acetic acid/acetate (acetate buffer) titration curve.



This tells us is that the pH is not changing much.

Energy

Living organisms are made up of cells, and cells contain many biochemical components such as proteins, lipids, and carbohydrates.

Living cells are not random collections of these molecules. They are extraordinarily organized.

In the nonliving world, there is a universal tendency to increasing disorder.

Maintaining and creating order in cells takes the input of energy.

Where does energy come from?

- Without energy, life is not possible. It is therefore important that we consider energy first in our attempt to understand biochemistry.

- Photosynthetic organisms can capture energy from the sun, converting it to chemical forms usable by cells.

- Heterotrophic organisms like ourselves get our energy from the food we eat.

Definition of Gibbs free energy

- Gibbs free energy is a thermodynamic potential that measures the process-initiating work obtainable from a thermodynamic system at constant temperature and pressure (isothermal, isobaric).

- Mathematically, the Gibbs free energy is given as

$$G = H - TS$$

Where H is the enthalpy, T is the temperature in Kelvin, and S is the entropy.

Why do organisms need energy?

- If entropy always increased everywhere, you could not do this. However, with the input of energy, you overcame the disorder. The cost of fighting disorder is energy.

- There are, of course, other reasons that organisms need energy. Muscular contraction, synthesis of molecules, neurotransmission, signaling, thermoregulation, and subcellular movements are examples.

- In summary, energy is needed for cells to perform the functions
- That they must carry out in order to stay alive.
- At its most basic level, this means fighting a continual battle with entropy.

Carbohydrates

Carbohydrates

- Carbohydrates are the most abundant bimolecular on Earth. Each year, photosynthesis converts more than 100 billion metric tons of CO_2 and H_2O into cellulose and other plant products.

- Insoluble carbohydrate polymers serve as structural and protective elements in the cell walls of bacteria and plants and in the connective tissues of animals.

Major size classes of carbohydrates

There are three major size classes of carbohydrates:

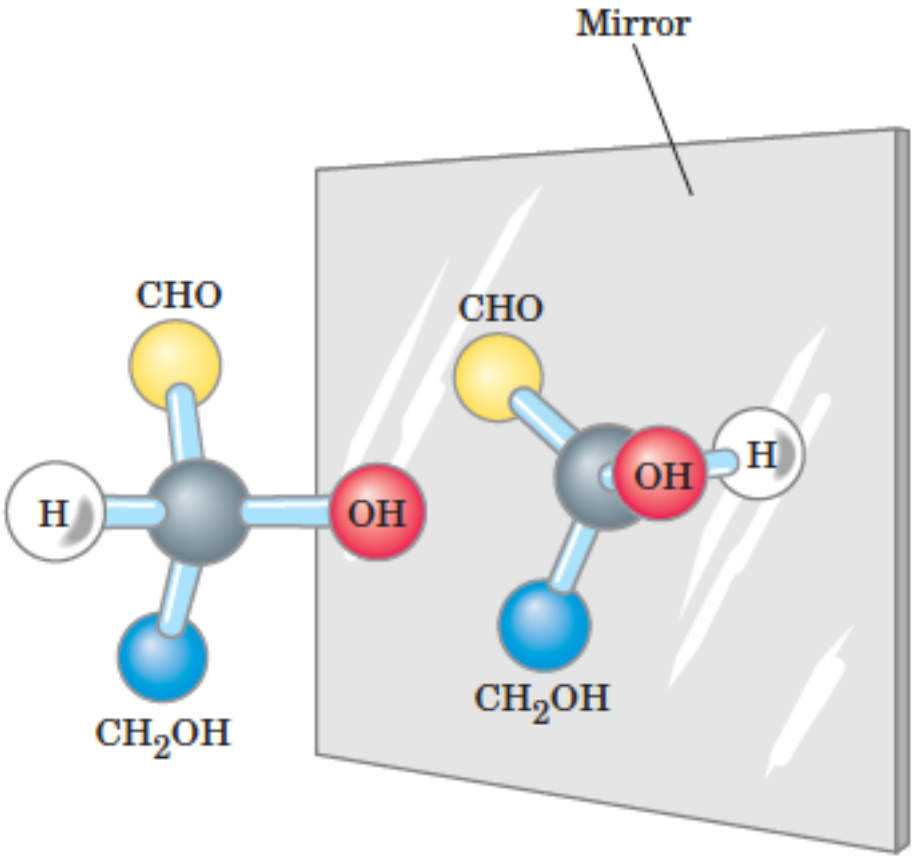
- Monosaccharides,
- Oligosaccharides, and
- Polysaccharides

The word “saccharide” is derived from the Greek sakcharon, meaning “sugar”.

Monosaccharides Have Asymmetric (Chiral) Centers

The stereoisomers are mirror images of each other.

All the monosaccharides except dihydroxyacetone contain one or more asymmetric (chiral) carbon atoms and thus occur in optically active isomeric forms.



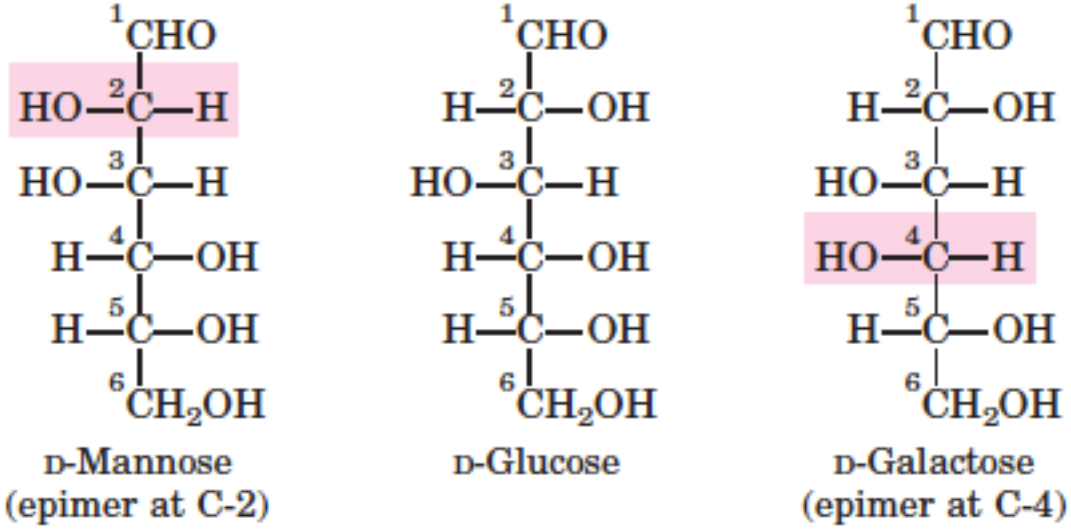
Ball-and-stick models

Stereoisomers:

- By convention, one of these two forms is designated the D isomer, the other the L isomer.

- In general, a molecule with n chiral centers can have 2^n stereoisomers. Glyceraldehyde has 2^1 = 2; the aldohexoses, with four chiral centers, have 2^4 = 16 stereoisomers.

Epimers:

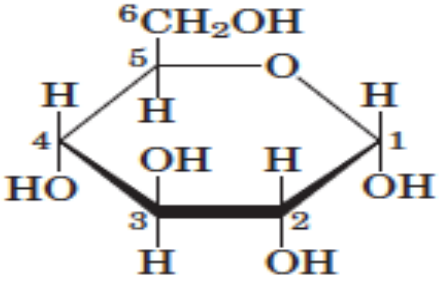


D-Glucose and two of its epimers are shown as projection formulas. Each epimer differs from D-glucose in the configuration at one chiral center (shaded red).

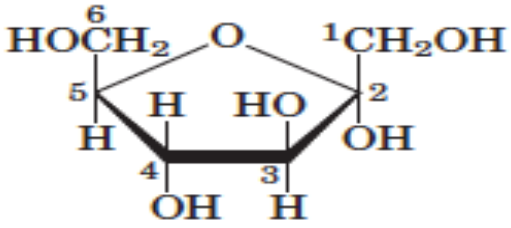
Anomers:

- Aldohexoses also exist in cyclic forms having five-membered rings.
- They resemble the five-membered ring compound furan, are called furanoses.
- However, the six-membered aldopyranose ring is much more stable than the aldofuranose ring and predominates in aldohexose solutions.
- Isomeric forms of monosaccharides that differ only in their configuration about the hemiacetal or hemiketal carbon atom are called anomers.

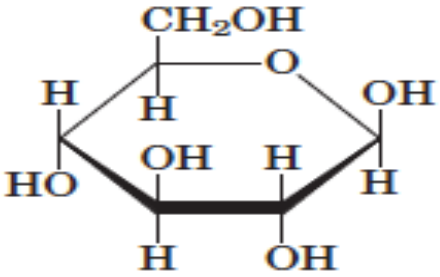
Pyranoses and furanoses (Haworth perspective formulas)



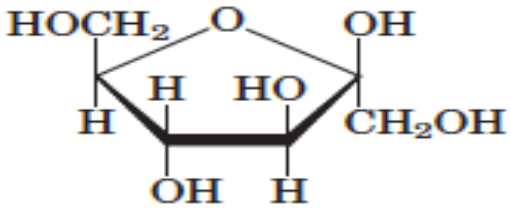
α-D-Glucopyranose



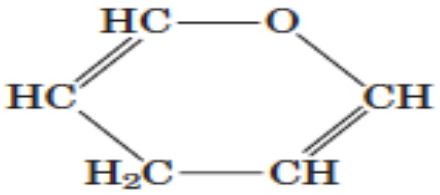
α-D-Fructofuranose



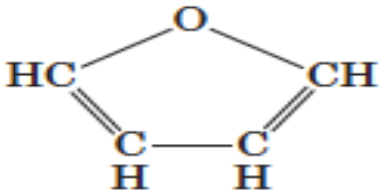
β-D-Glucopyranose



β-D-Fructofuranose



Pyran



Furan

Monosaccharides Are Reducing Agents

- Monosaccharides can be oxidized by relatively mild oxidizing agents such as ferric (Fe^3+) or cupric (Cu^2+) ion. Carbonyl carbon is oxidized to a carboxyl group.

- Glucose and other sugars capable of reducing ferric or cupric ion are called reducing sugars.

- This property is the basis of Fehling's reaction, a qualitative test for the presence of reducing sugar. By measuring the amount of oxidizing agent reduced by a solution of a sugar, it is also possible to estimate the concentration of that sugar.

Oligosaccharides

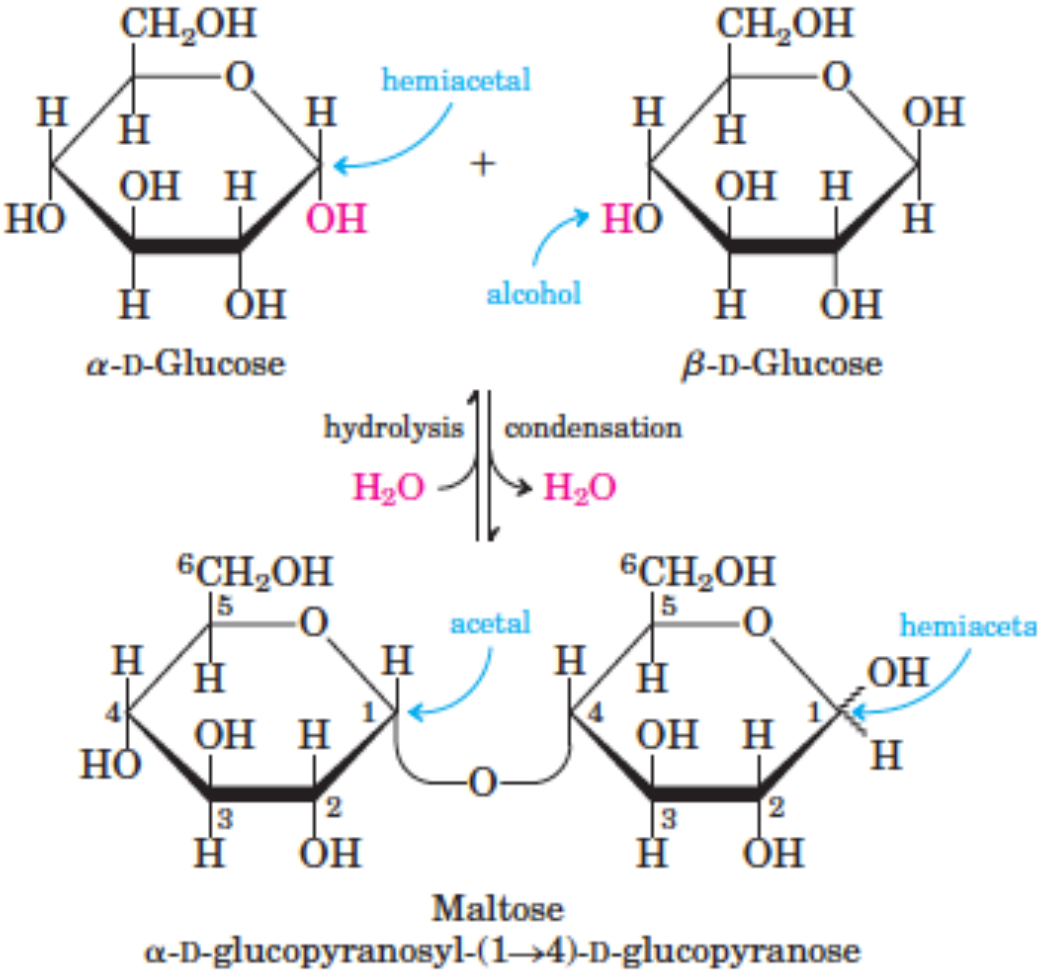
- Oligosaccharides consist of short chains of monosaccharide units, or residues, joined by characteristic linkages called glycosidic bonds.

- The most abundant are the disaccharides, with two monosaccharide units.

- *The two monosaccharides joined covalently by an O -glycosidic bond*, which is formed when a hydroxyl group of one sugar reacts with the anomeric carbon of the other.

- This reaction represents the formation of an acetal from a hemiacetal.

Formation of maltose



Glc(1- 4)Glc

The maltose molecule retains a reducing hemiacetal at the C-1 not involved in the glycosidic bond. maltose is a reducing sugar. The glucose residue with the free anomeric carbon.

Polysaccharides

- The polysaccharides are sugar polymers containing more than 20 or so monosaccharide units, and some have hundreds or thousands of units.

- Some polysaccharides, such as cellulose, are linear chains; others, such as glycogen, are branched.

- Both glycogen and cellulose consist of recurring units of D-glucose.

- Most carbohydrates found in nature occur as polysaccharides; polysaccharides, also called glycans, which are divided to two classes

1. Homopolysaccharides contain only a single type of monomer;

2. Heteropolysaccharides contain two or more different kinds.

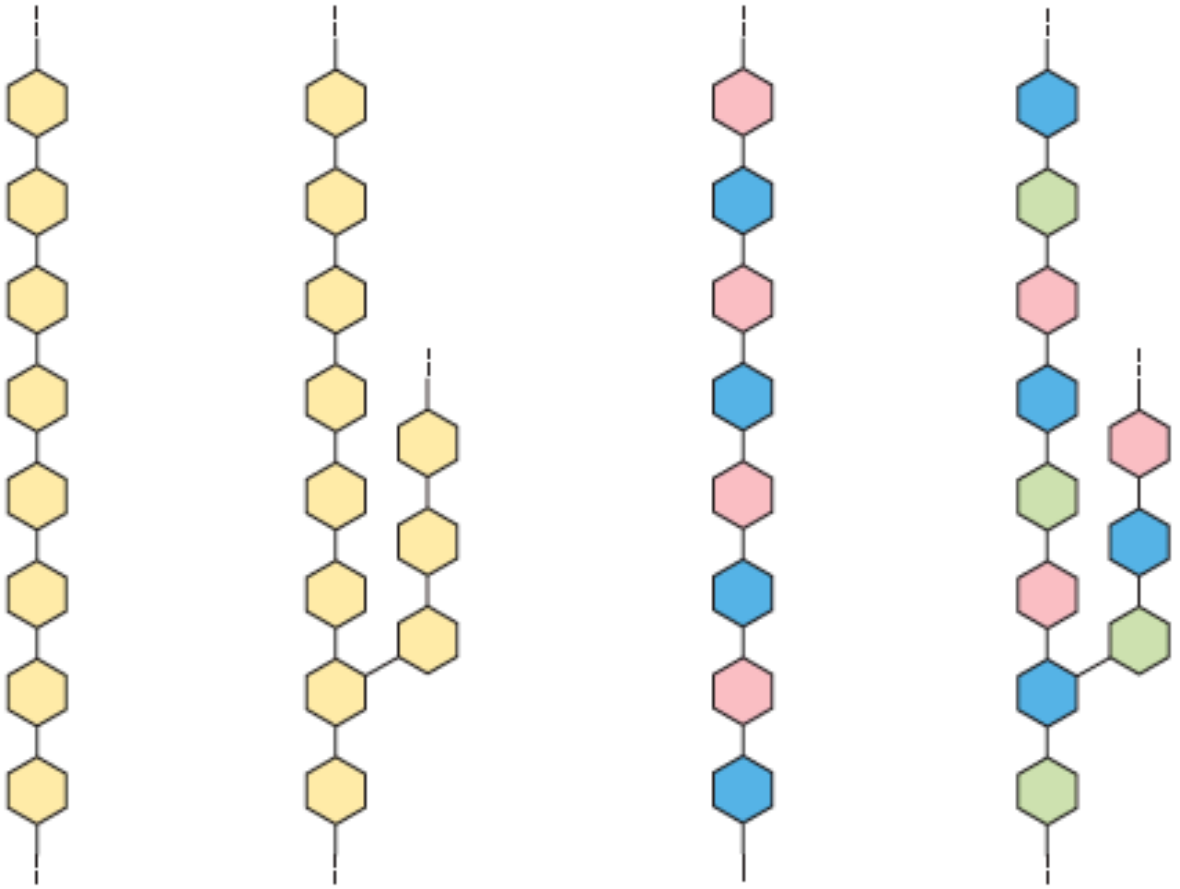
Homopolysaccharides

Unbranched Branched

Heteropolysaccharides

Two monomer types, unbranched

Multiple monomer types, branched



Why not store glucose in its monomeric form?

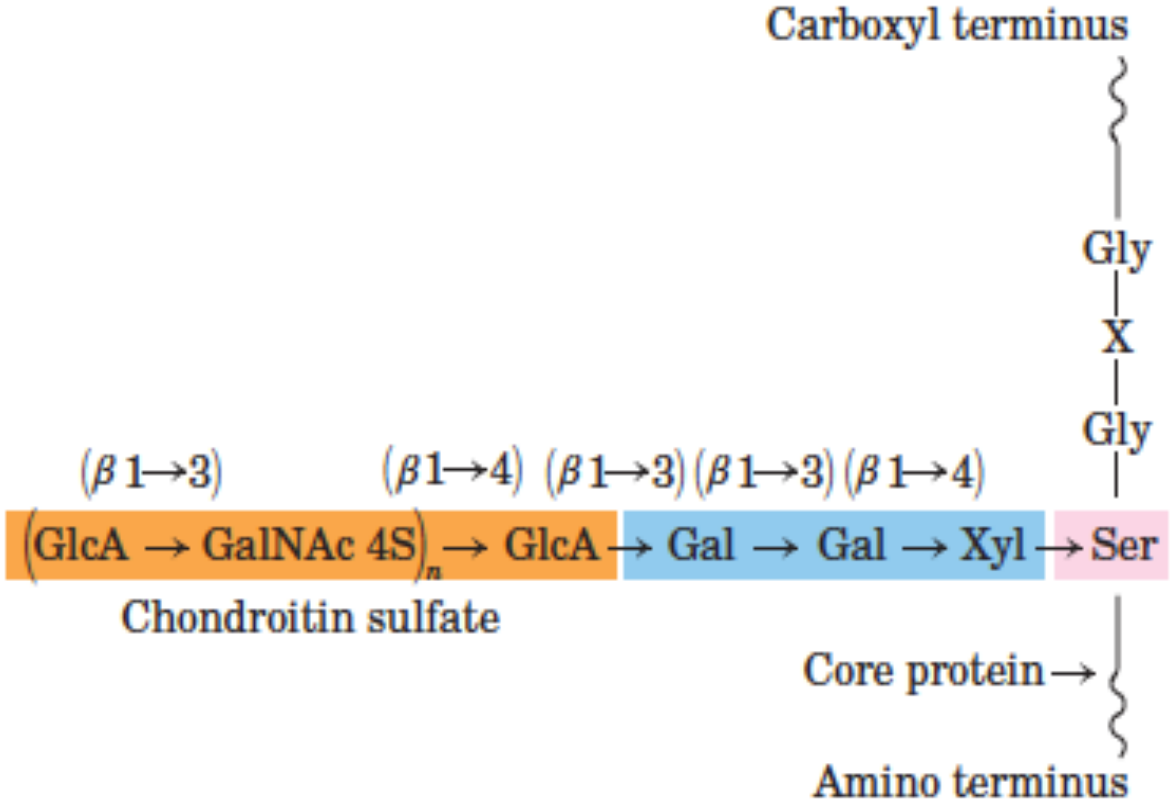
- It has been calculated that hepatocytes (a cell of main tissue of the liver make up to 70-85% of liver mass) store glycogen equivalent to a glucose concentration of 0.4 M.

- The actual concentration of glycogen, which is insoluble and contributes little to the osmolarity of the cytosol, is about 0.01 M.

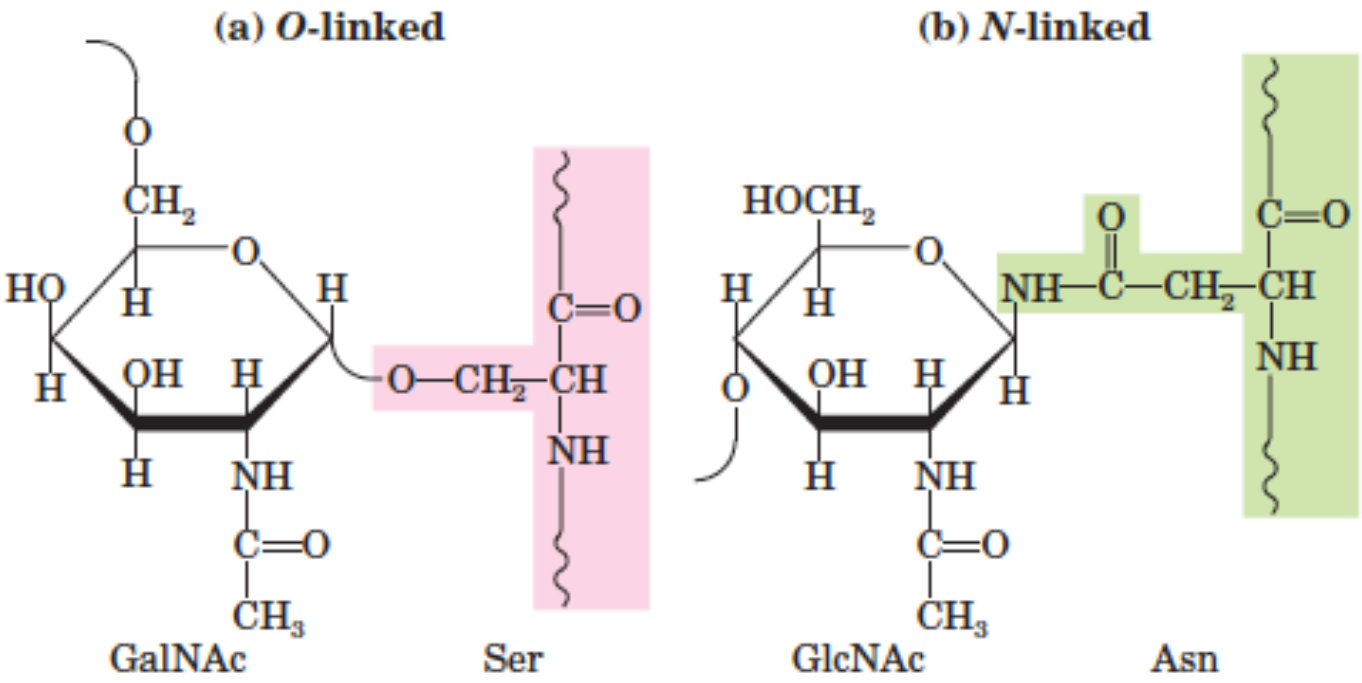
- *If the cytosol contained 0.4 M glucose, the osmolarity would be threateningly elevated, leading to osmotic entry of water that might rupture the cell.*

Proteoglycans

Proteoglycans are macromolecules of the cell surface or extracellular matrix in which one or more glycosaminoglycan chains are joined covalently to a membrane protein or a secreted protein.



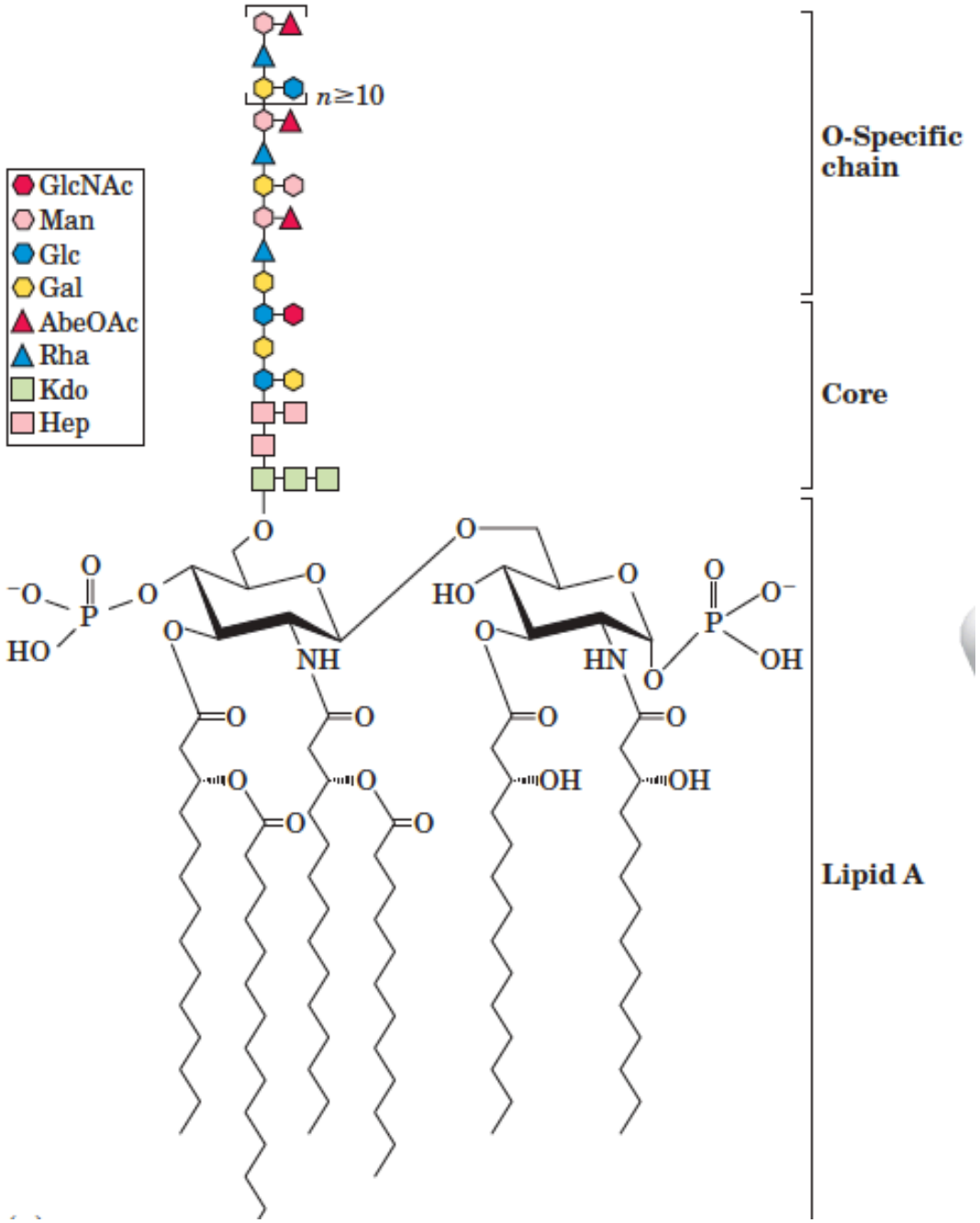
Glycoproteins



Glycoproteins have one or several oligosaccharides of varying complexity joined covalently to a protein. They are found *on the outer face of the plasma membrane*, in the extracellular matrix, and in the blood.

Glycolipids

Glycolipids are membrane lipids in which the hydrophilic head groups are oligosaccharides, which, as in glycoproteins, act as specific sites for recognition by carbohydrate-binding proteins.



Amino acids, peptides and proteins

Charged:

- Arginine - Arg - R
- Lysine - Lys - K
- Aspartic acid - Asp - D
- Glutamic acid - Glu - E

Polar (may participate in hydrogen bonds):

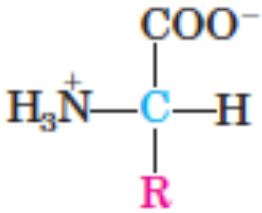
- Glutamine - Gln - Q
- Asparagine - Asn - N
- Histidine - His - H
- Serine - Ser - S
- Threonine - Thr - T
- Tyrosine - Tyr - Y
- Cysteine - Cys - C
- Methionine - Met - M
- Tryptophan - Trp - W

Hydrophobic (normally buried inside the protein core):

- Alanine - Ala - A
- Isoleucine - Ile - I
- Leucine - Leu - L
- Phenylalanine - Phe - F
- Valine - Val - V
- Proline - Pro - P
- Glycine - Gly - G

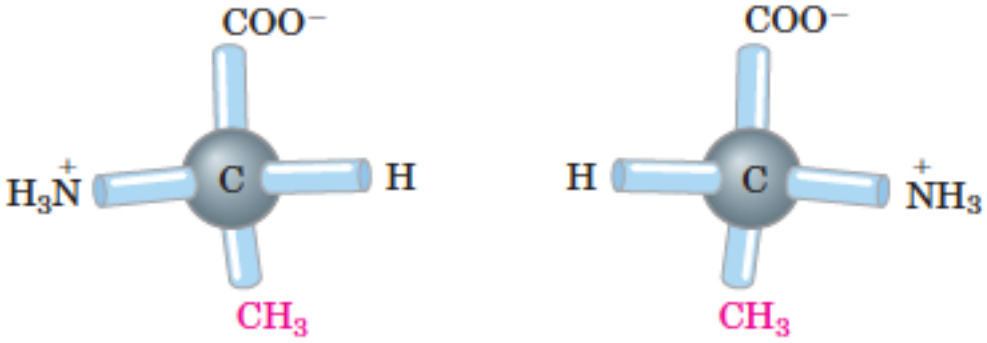
General structure of an amino acid

All 20 of the common amino acids are α-amino acids. They have a carboxyl group and an amino group bonded to the same carbon atom. They differ from each other in their side chains, or R groups;



which vary in structure, size, and electric charge, and which influence the solubility of the amino acids in water.

Stereoisomerism in amino acids



(a) L-Alanine D-Alanine

(b) L-Alanine D-Alanine

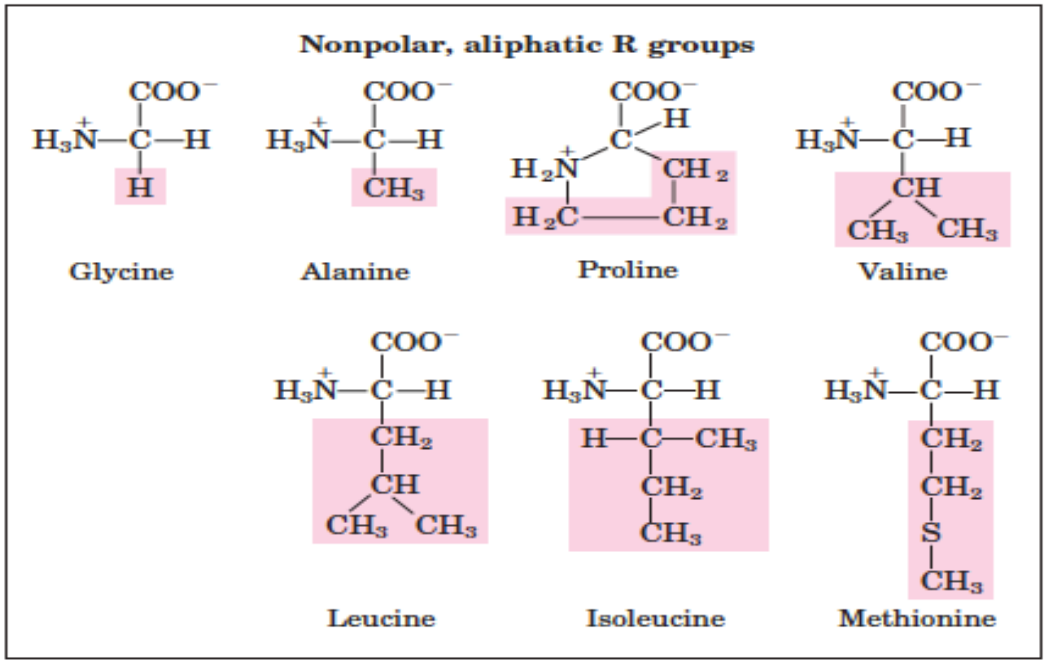
(c) L-Alanine D-Alanine

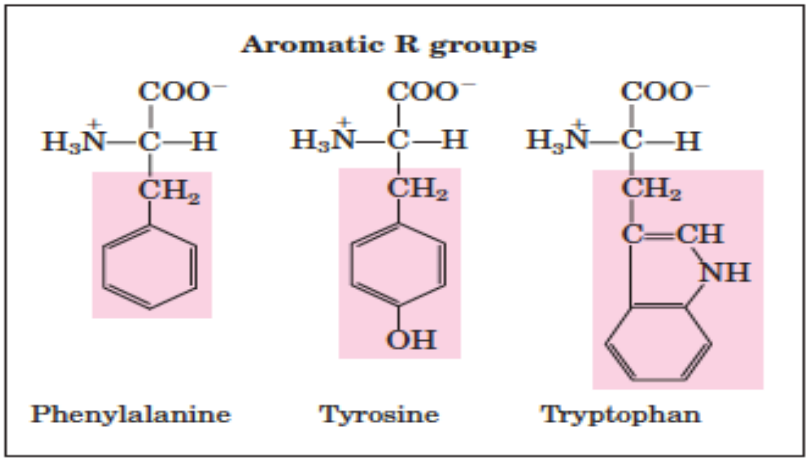
Amino Acids Can Be Classified by R Group

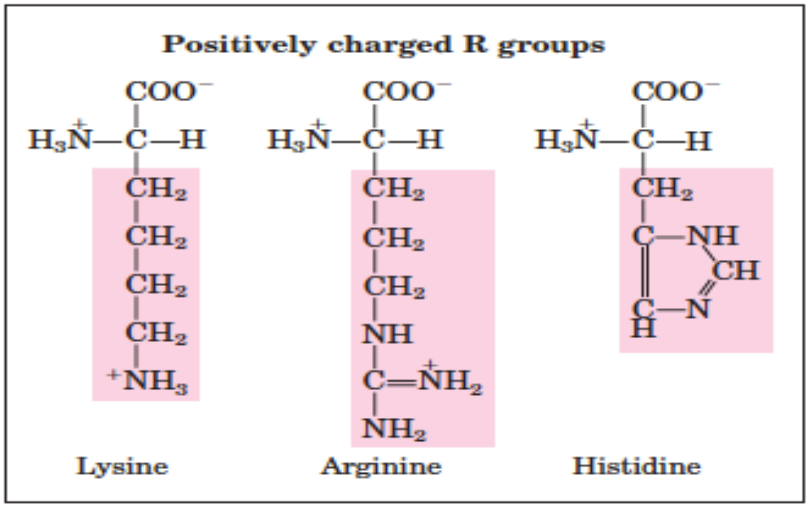
- The topic can be simplified by grouping the amino acids into five main classes based on the properties of their R groups.

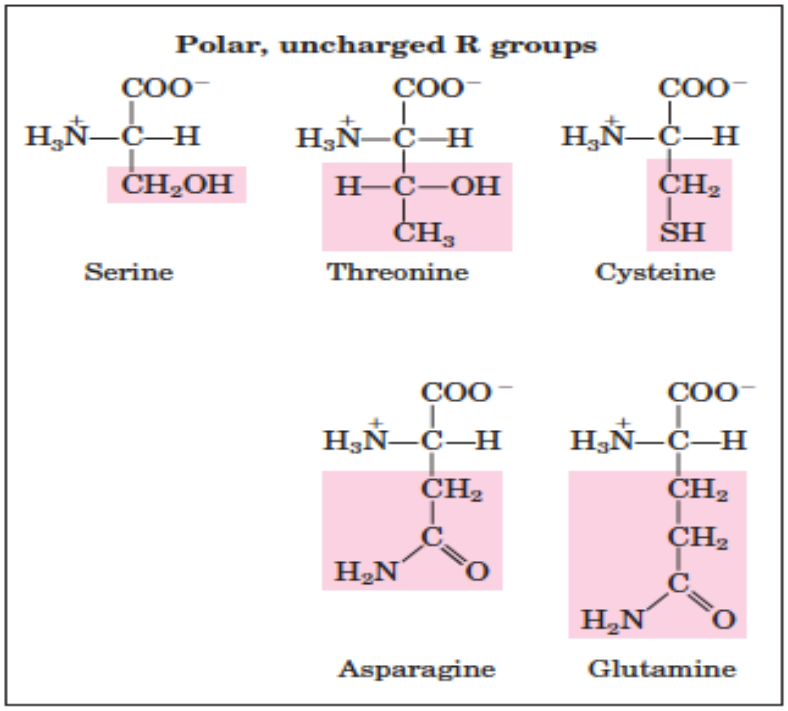
1. Nonpolar, aliphatic R groups
2. Polar, uncharged R groups
3. Aromatic R groups
4. Positively charged R groups
5. Negatively charged R groups

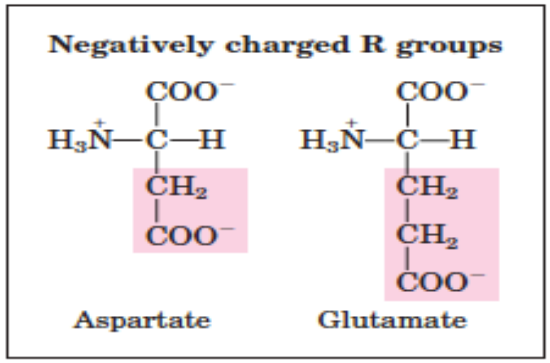
Common AAs of proteins





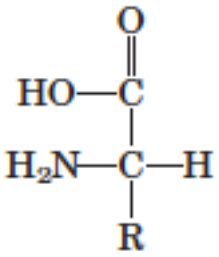




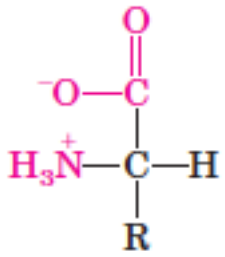


Amino Acids Can Act as Acids and Bases

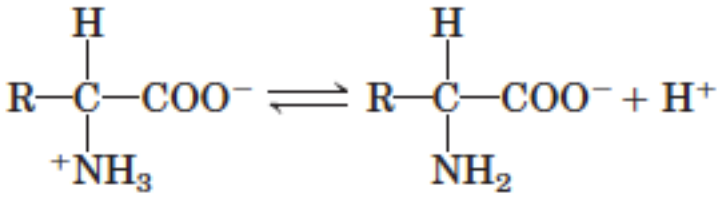
When an amino acid is dissolved in water, it exists in solution as the dipolar ion, or zwitterion.



Nonionic form

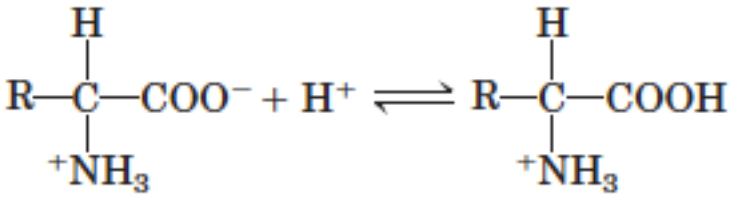


Zwitterionic form



Zwitterion

acid (proton donor)

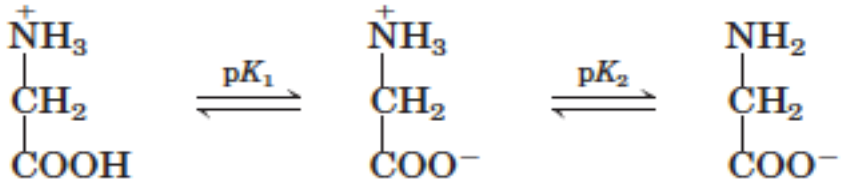


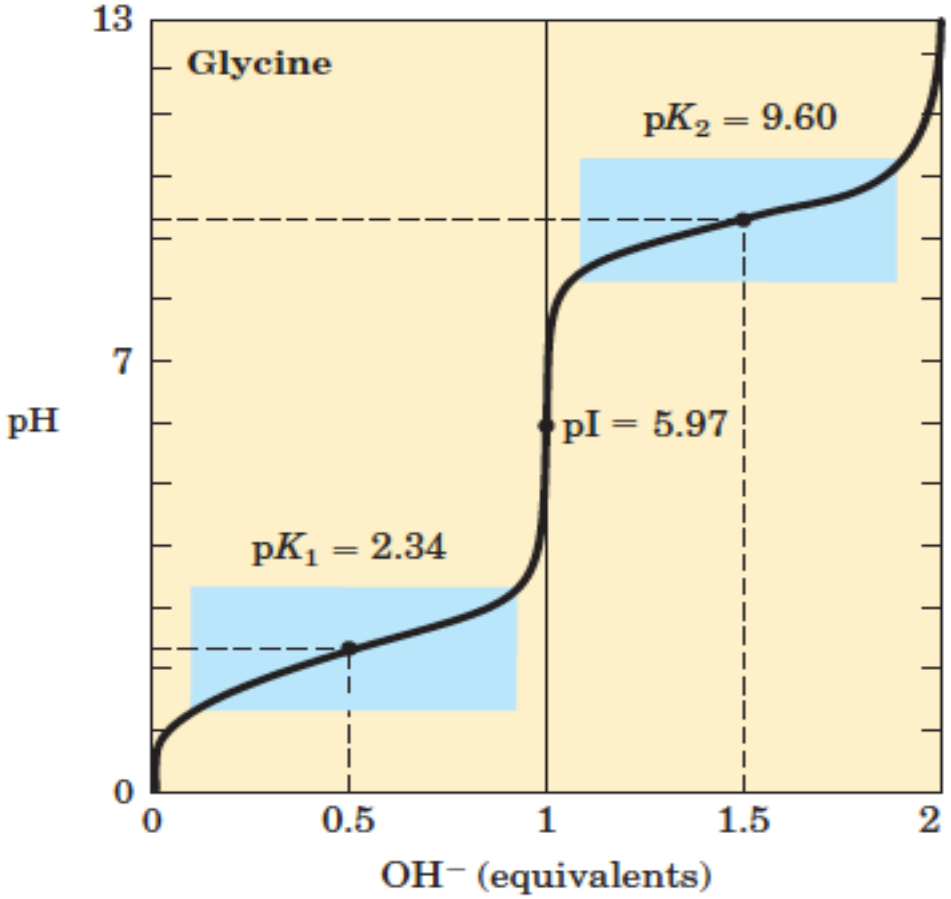
Zwitterion

base (proton acceptor)

Amino Acids Have Characteristic Titration Curves

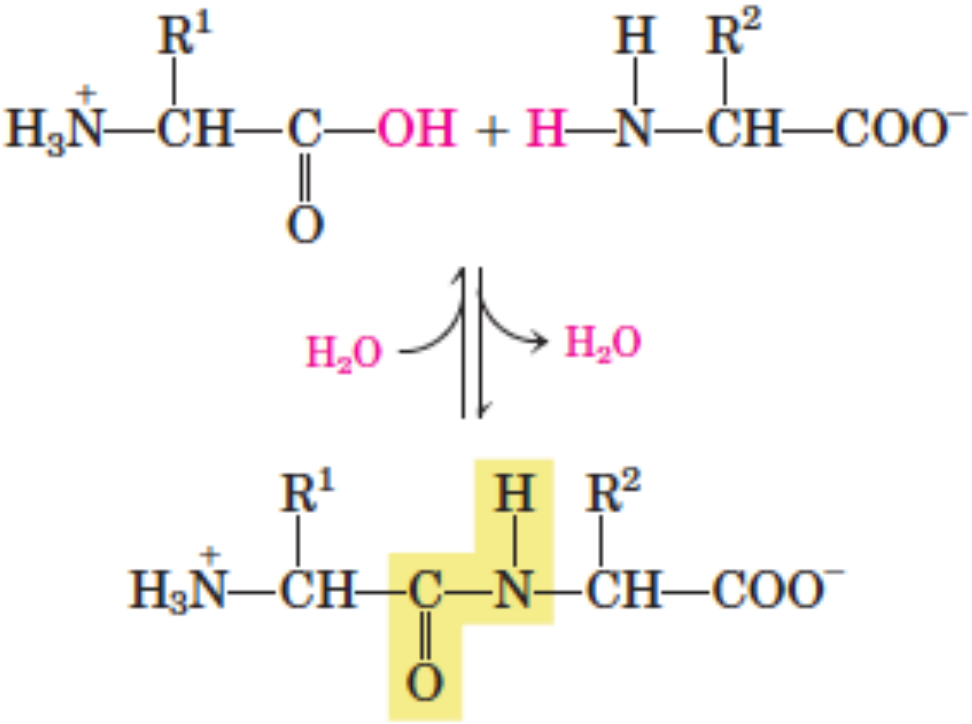
Glycine



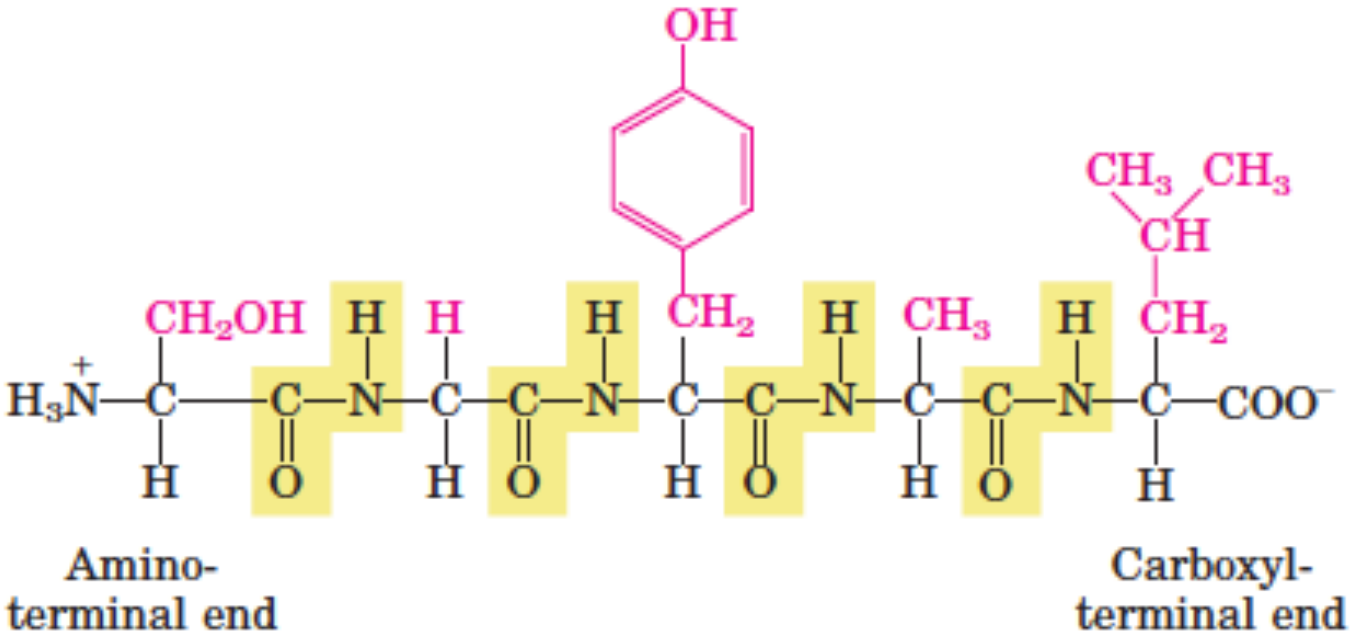


Another important piece of information derived from the titration curve of an amino acid is the relationship between its net electric charge and the pH of the solution.

Formation of a peptide bond by condensation



The pentapeptide serylglycyltyrosylalanylleucine, or Ser–Gly–Tyr–Ala–Leu.

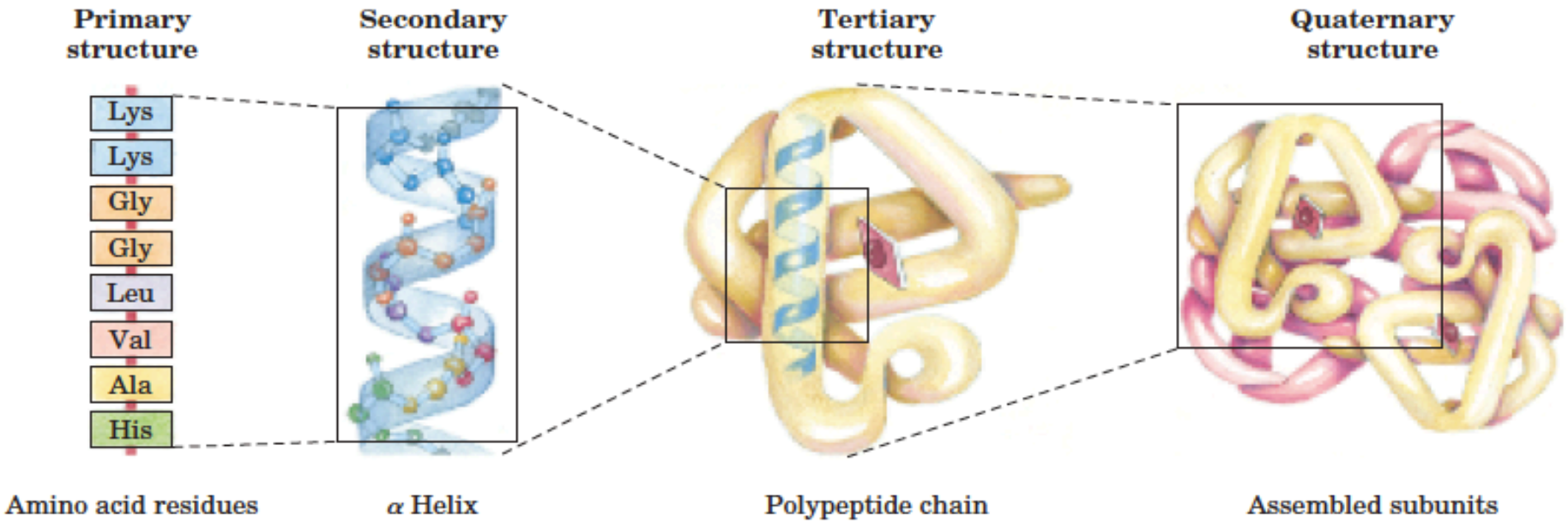


Conjugated Proteins

TABLE 3–4 Conjugated Proteins

Class	*Prosthetic group*	*Example*
Lipoproteins	Lipids	β_1-Lipoprotein of blood
Glycoproteins	Carbohydrates	Immunoglobulin G
Phosphoproteins	Phosphate groups	Casein of milk
Hemoproteins	Heme (iron porphyrin)	Hemoglobin
Flavoproteins	Flavin nucleotides	Succinate dehydrogenase
Metalloproteins	Iron	Ferritin
	Zinc	Alcohol dehydrogenase
	Calcium	Calmodulin
	Molybdenum	Dinitrogenase
	Copper	Plastocyanin

Levels of structure in proteins



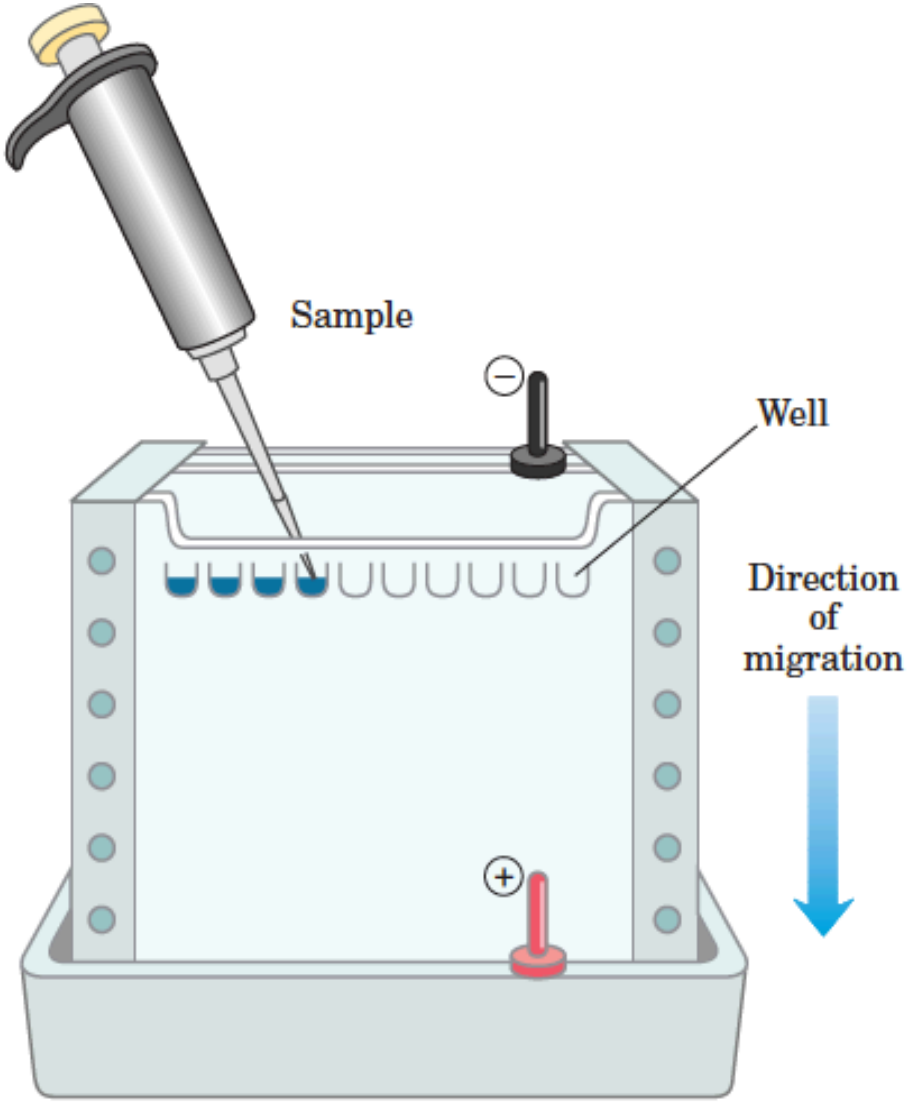
Protein separation

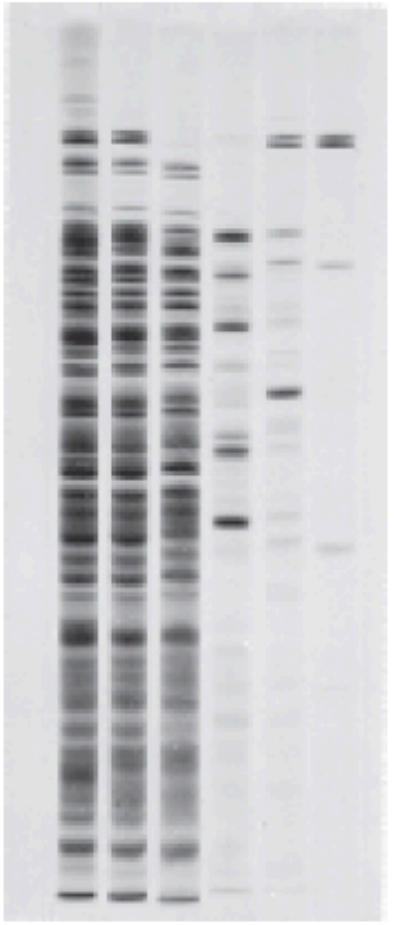
- Break cells to release the protein. This is called crude extract.
- Fractionation by ammonium sulfate.
- Dialysis separates proteins from solvents by a semi-permeable membrane.
- Column chromatography.
  1. Size-exclusion chromatography.
  2. Ion-exchange chromatography.
  3. Affinity chromatography.
  4. High-performance liquid chromatography (HPLC).

- ***Activity***: 1 unit (U) is the amount if enzyme that catalyzes the reaction of 1 µmol of substrate per minute under standard conditions.

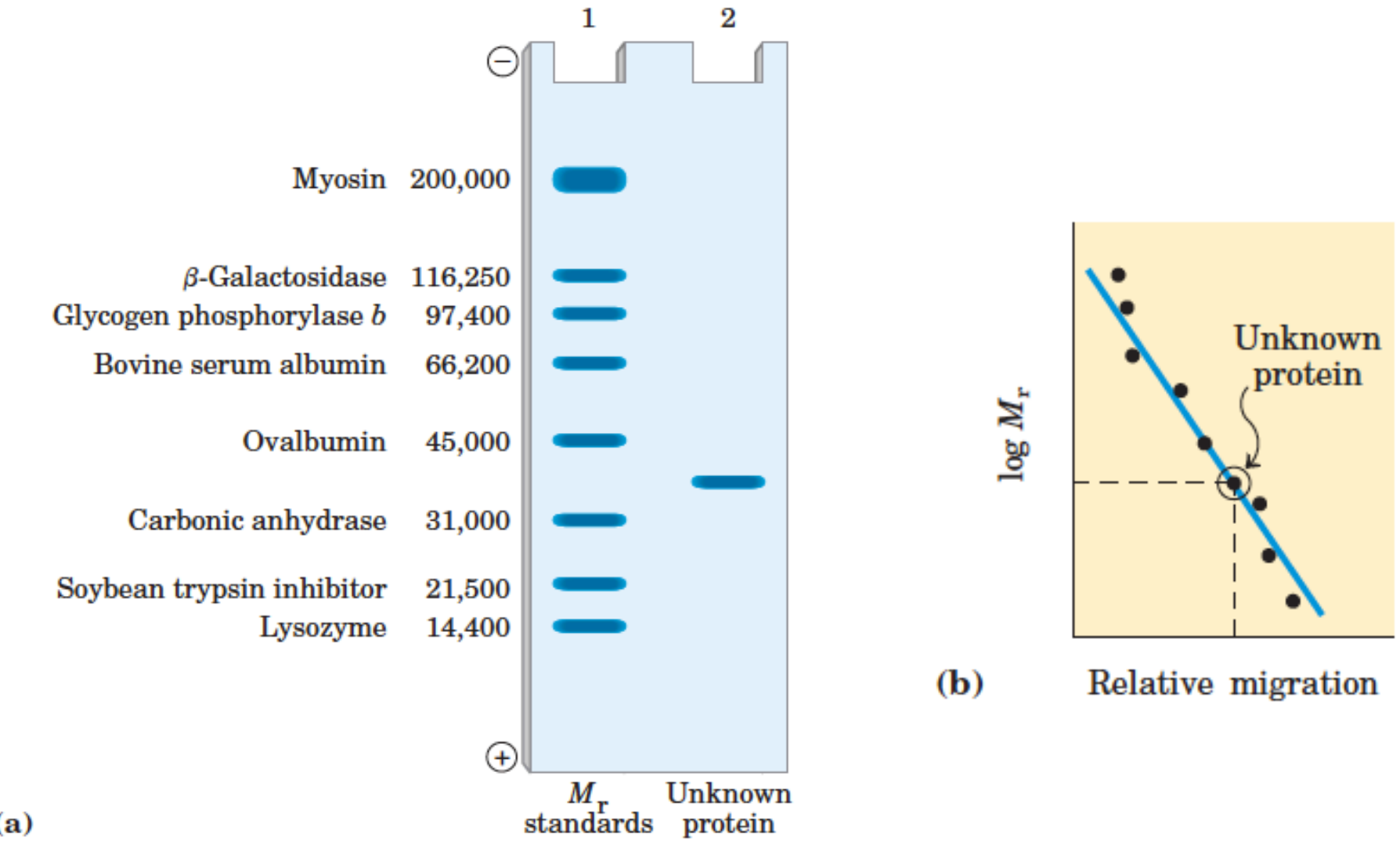
- ***Specific Activity***: is the number of enzyme units per ml divided by the concentration of protein in mg/ml.

Electrophoresis of proteins is generally carried out in gels made up of the cross-linked polymer polyacrylamide

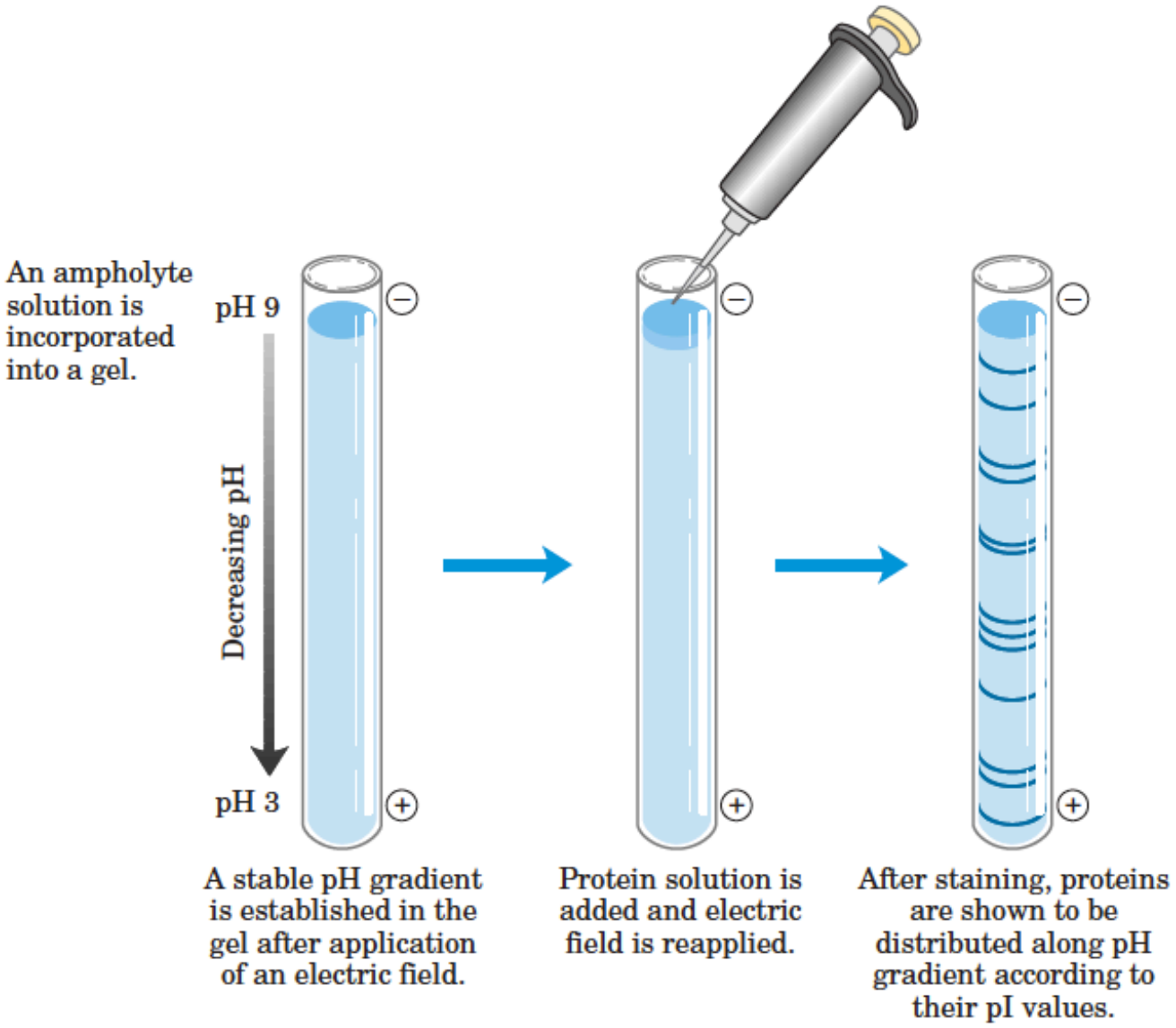


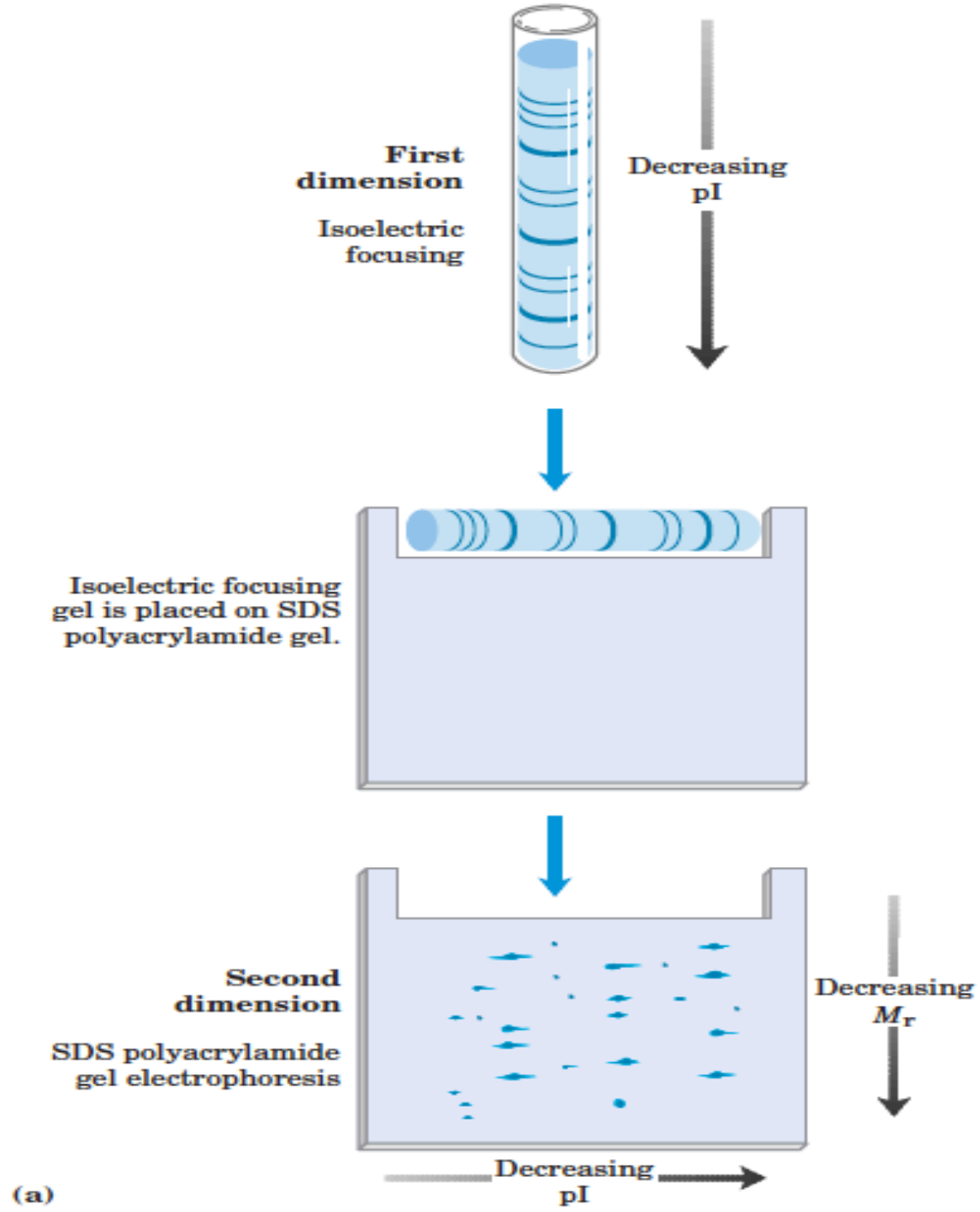


Estimating the molecular weight of a protein



Isoelectric focusing




First dimension
Isoelectric focusing
Decreasing pI
Isoelectric focusing gel is placed on SDS polyacrylamide gel.
Second dimension
SDS polyacrylamide gel electrophoresis
Decreasing M_r
Decreasing pI
(a)

Amino acid sequence of protein

- What is it that makes one protein an enzyme, another a hormone, another a structural protein, and still another an antibody? How do they differ chemically?

- The differences in primary structure can be especially informative. Each protein has a distinctive number and sequence of amino acid residues.

- *It quickly became evident that the nucleotide sequence in DNA and the amino acid sequence in proteins were somehow related.*

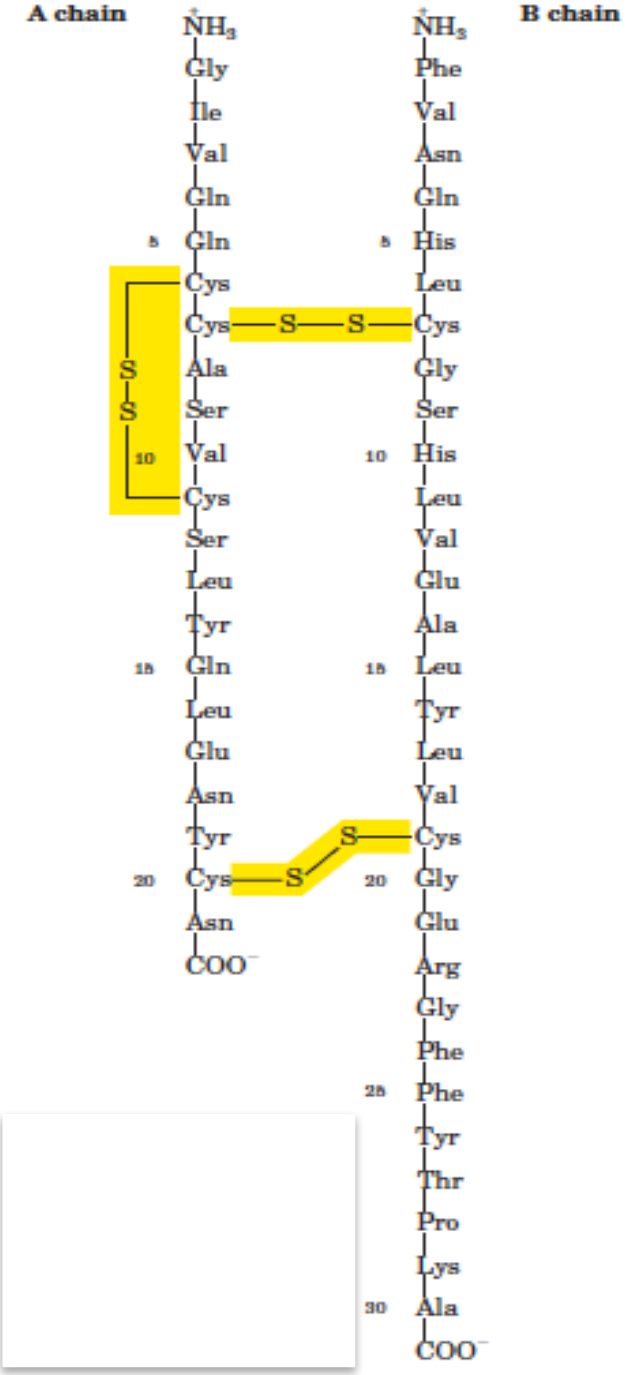
- Enzymes called proteases catalyze the hydrolytic cleavage of peptide bonds. Some proteases cleave only the peptide bond adjacent to particular amino acid residues

Amino acid sequence of bovine insulin.

The two polypeptide chains are joined by disulfide crosslinkages.

The A chain is identical in human, pig, dog, rabbit, and sperm whale insulins.

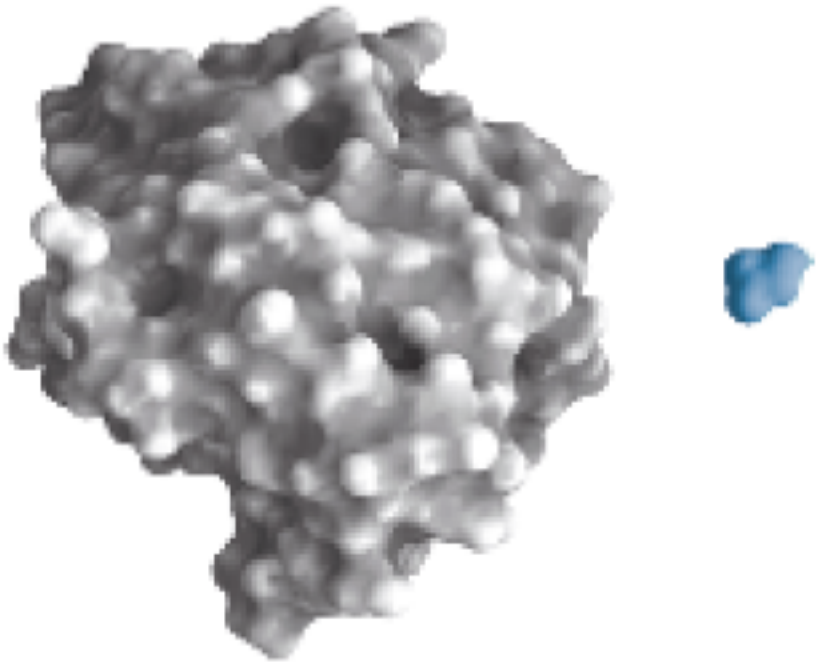
The B chains of the cow, pig, dog, goat, and horse are identical.



Structure of the enzyme chymotrypsin, a globular protein

Proteins are large molecules and, as we shall see, each has a unique structure.

A molecule of glycine (blue) is shown for size comparison.



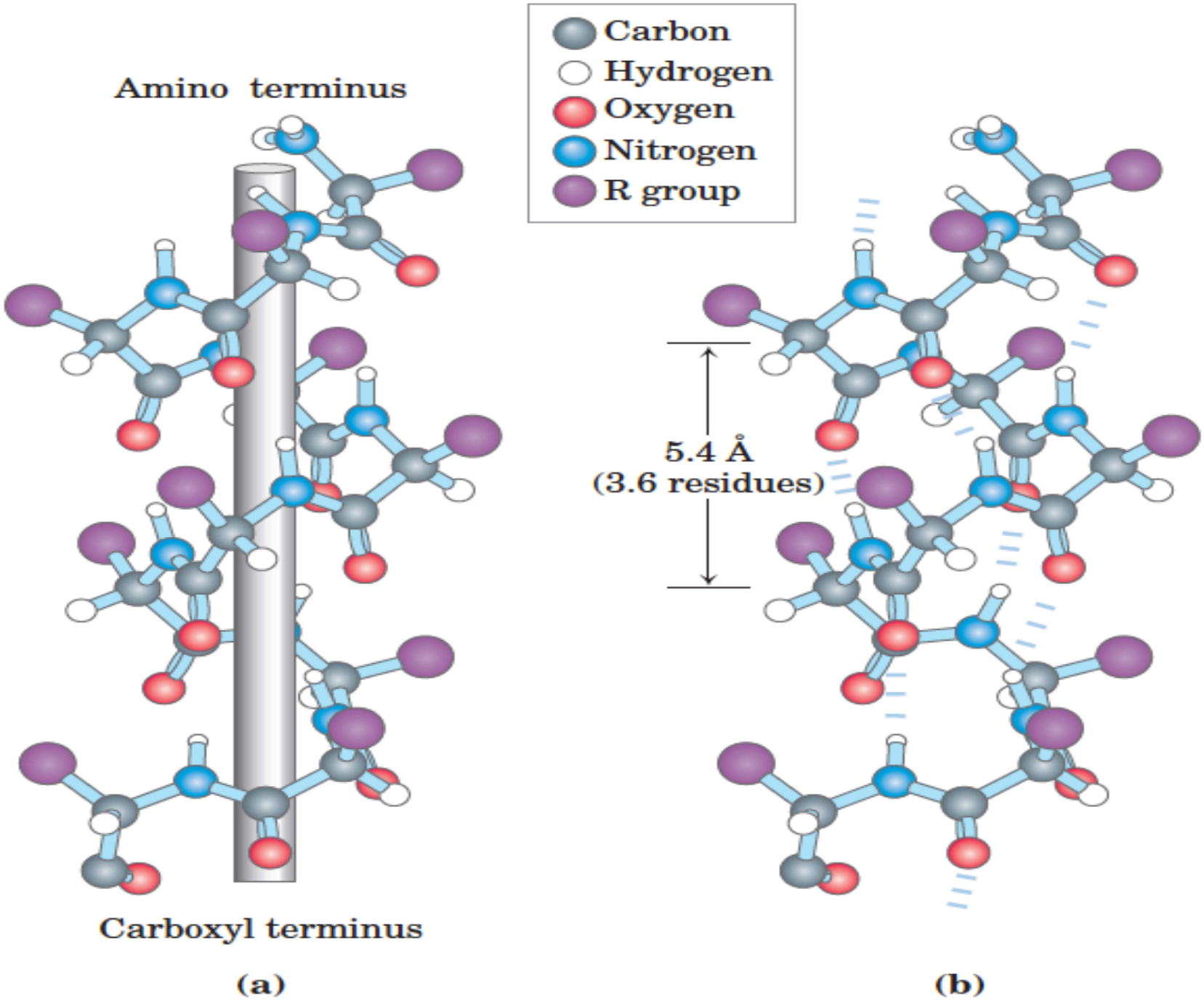
The known three-dimensional structures of proteins are archived in the Protein Data Bank, or PDB (www.rcsb.org/pdb).

The helix is a common protein secondary structure

- The simplest arrangement the polypeptide chain could assume with its rigid peptide bonds (but other single bonds free to rotate) is a helical structure, which Pauling and Corey called the α-helix as in the coming figure.

- (a) Formation of a right-handed helix.

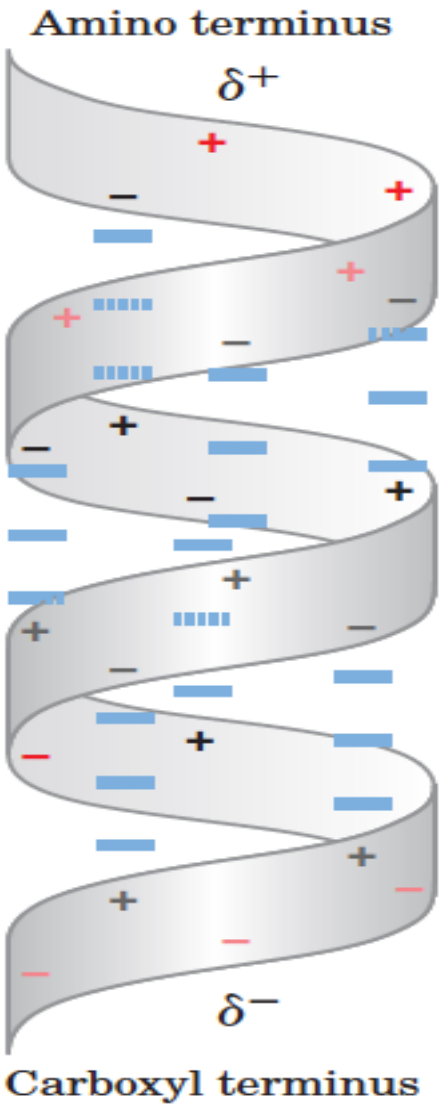
- (b) Ball-and-stick model of a right-handed α helix, showing the intrachain hydrogen bonds.

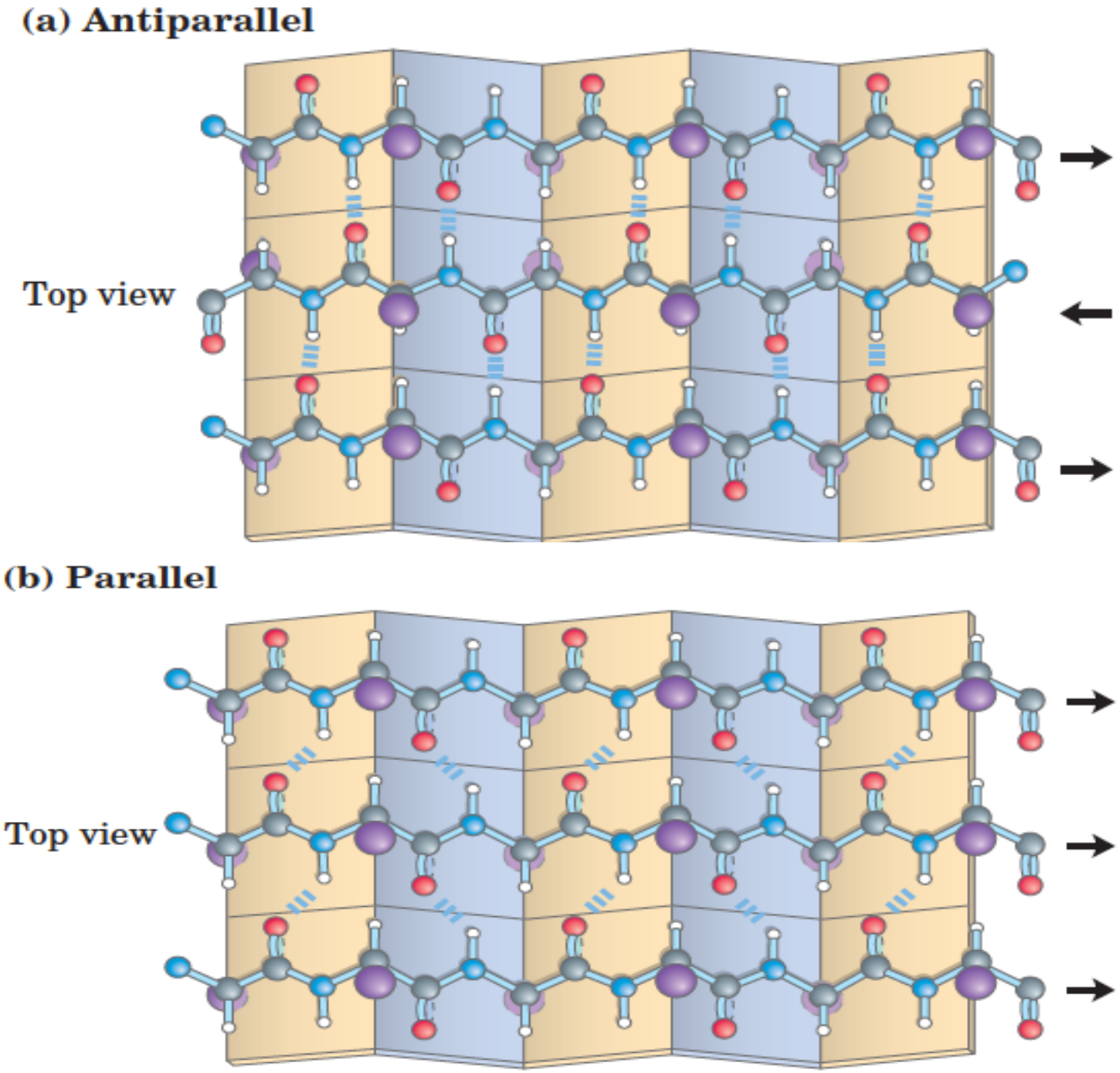

Carbon
Hydrogen
Oxygen
Nitrogen
R group
Amino terminus
Carboxyl terminus
5.4 Å
(3.6 residues)
(a)
(b)

- α-Helix can form in polypeptides consisting of either L- or D-amino acids. However, all residues must be of one stereoisomeric series; a D-amino acid will disrupt a regular structure consisting of L-amino acids, and vice versa.

Helix dipole

Non-hydrogen-bonded amino and carbonyl constituents in the peptide bonds near each end of the α-helical region are shown in red.




(a) Antiparallel
Top view
(b) Parallel
Top view

- In considering these higher levels of structure, it is useful to classify proteins into two major groups:

1) Fibrous proteins, having polypeptide chains arranged in long strands or sheets, and

2) globular proteins, having polypeptide chains folded into a spherical or globular shape.

Protein denaturation and folding

- All proteins begin their existence on a ribosome as a linear sequence of amino acid residues.

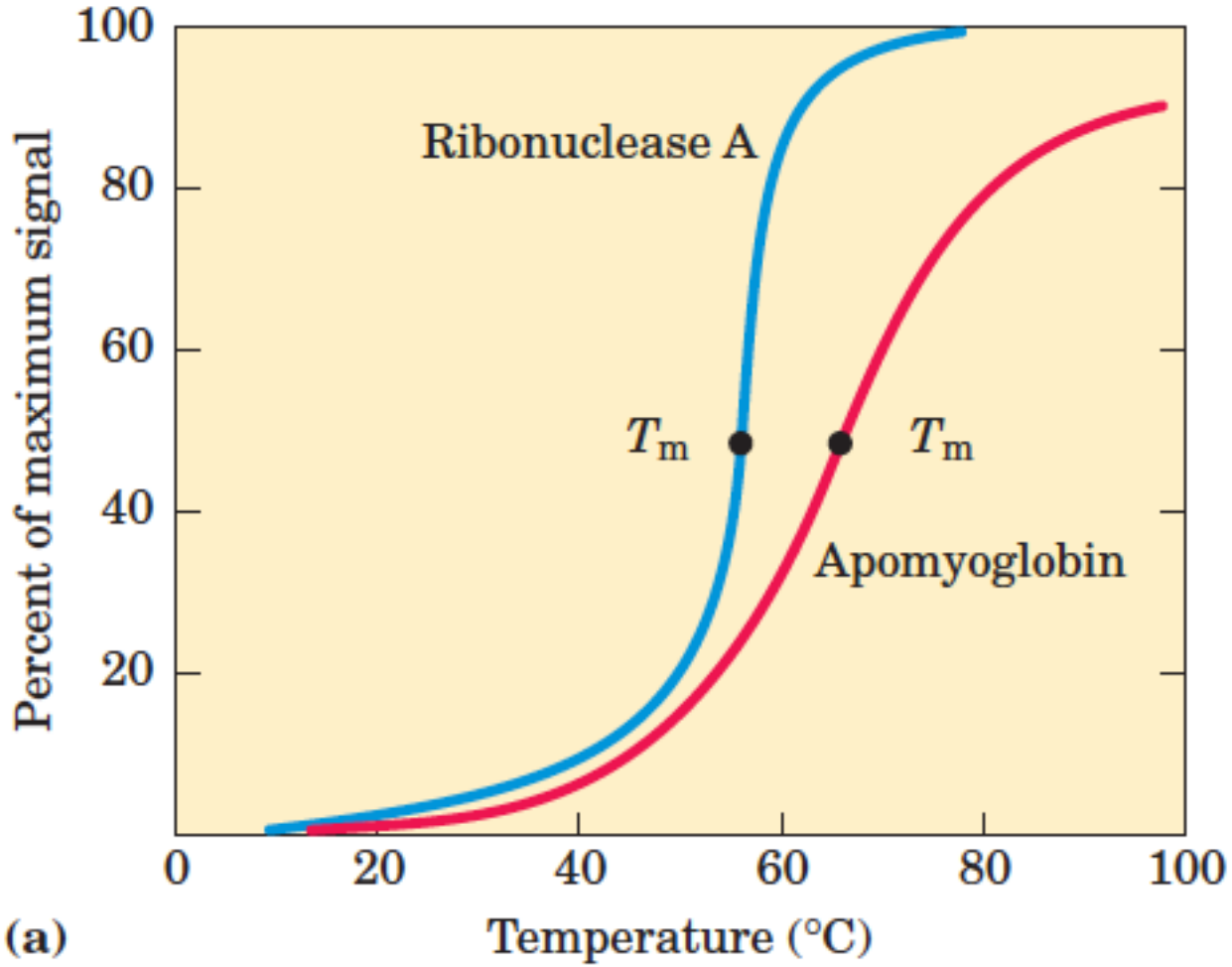
- This polypeptide must fold during and following synthesis to take up its native conformation.

How can proteins be denatured?

- Most proteins can be denatured by heat, which affects the weak interactions in a protein (primarily hydrogen bonds) in a complex manner.

- If the temperature is increased slowly, a protein's conformation generally remains intact until an abrupt loss of structure (and
- function) occurs over a narrow temperature range.

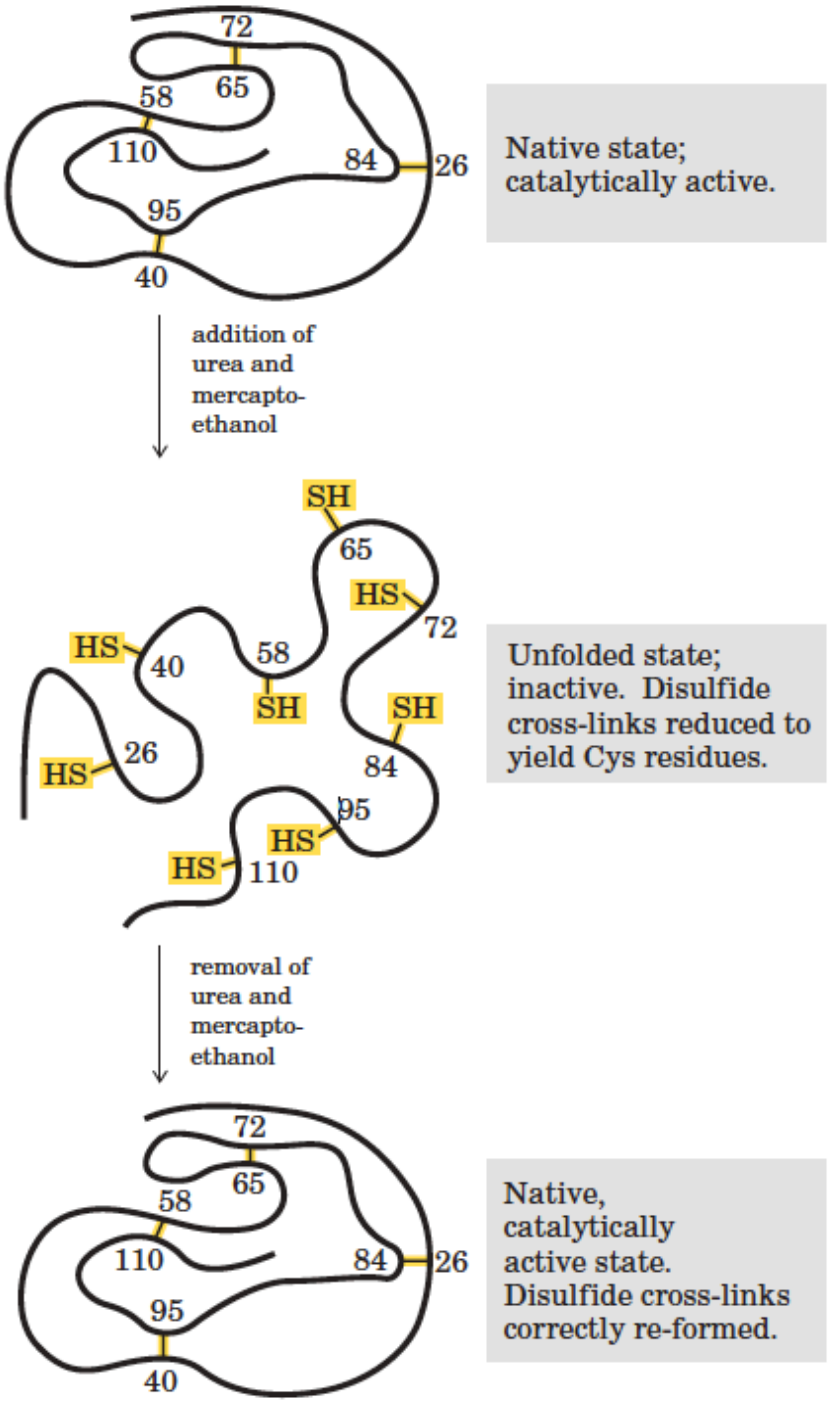
Thermal protein denaturation



The midpoint of the temperature range over which denaturation occurs is called the melting temperature, or Tm.

Renaturation of unfolded, denatured ribonuclease.

Certain globular proteins denatured by heat, extremes of pH, or denaturing reagents will regain their native structure and their biological activity if returned to conditions in which the native conformation is stable. This process is called renaturation.



Urea is used to denature ribonuclease, and mercaptoethanol (HOCH2CH2SH) to reduce and thus cleave the disulfide bonds to yield
eight Cys residues.

Renaturation involves reestablishment of the correct disulfide cross-links.