Nucleic Acids, Proteins, and Enzymes
Key Concepts

3.1 Nucleic Acids Are Informational Macromolecules

3.2 Proteins Are Polymers with Important Structural and Metabolic Roles

3.3 Some Proteins Act as Enzymes to Speed up Biochemical Reactions

3.4 Regulation of Metabolism Occurs by Regulation of Enzymes
How does an understanding of proteins and enzymes help to explain how aspirin works?
Nucleic acids are polymers that store, transmit, and express hereditary (genetic) information.

DNA = deoxyribonucleic acid

RNA = ribonucleic acid

The monomers are nucleotides.
Concept 3.1 Nucleic Acids Are Informational Macromolecules

Nucleotide: pentose sugar + N-containing base + phosphate group

Nucleosides: pentose sugar + N-containing base
Concept 3.1 Nucleic Acids Are Informational Macromolecules

Bases:
- **Pyrimidines**—single rings
- **Purines**—double rings

Sugars:
- DNA contains **deoxyribose**
- RNA contains **ribose**
Figure 3.1 Nucleotides Have Three Components

Base + Ribose or deoxyribose = Nucleoside + Phosphate = Nucleotide

Pyrimidines

- Cytosine (C)
- Thymine (T)
- Uracil (U)

Purines

- Adenine (A)
- Guanine (G)
Nucleotides bond in condensation reactions to form **phosphodiester bonds**.

The linkage is between the #5 carbon of one ribose and the #3 carbon of the next ribose.

Nucleic acids grow in the 5’ to 3’ direction.
Figure 3.2 Linking Nucleotides Together

Rest of polymer

Pyrimidine base

Condensation reaction

Phosphodiester bond + H₂O

PRINCIPLES OF LIFE 2e, Figure 3.2
Oligonucleotides have up to 20 monomers.

- Example: small RNA molecules important for DNA replication and gene expression.

DNA and RNA are polynucleotides, the longest polymers in the living world.
<table>
<thead>
<tr>
<th>Nucleic acid</th>
<th>Sugar</th>
<th>Bases</th>
<th>Strands</th>
</tr>
</thead>
<tbody>
<tr>
<td>RNA</td>
<td>Ribose</td>
<td>Adenine, Cytosine, Guanine, Uracil</td>
<td>Single</td>
</tr>
<tr>
<td>DNA</td>
<td>Deoxyribose</td>
<td>Adenine, Cytosine, Guanine, Thymine</td>
<td>Double</td>
</tr>
</tbody>
</table>
Complementary base pairing:

Thymine (T) pairs with Adenine (A) via hydrogen bonds.

Cytosine (C) pairs with Guanine (G) via hydrogen bonds.

Polar bonds also connect the bases.
Base pairs are linked by hydrogen bonds, favored by the arrangement of polar bonds in the bases.

There are so many hydrogen bonds in DNA and RNA that they form a fairly strong attraction, but not as strong as covalent bonds.

Thus, base pairs can be separated with only a small amount of energy.
In RNA, the base pairs are A–U and C–G.

RNA is usually single-stranded, but may be folded into 3-D structures by hydrogen bonding.

Folding occurs by complementary base pairing, so structure is determined by the order of bases.
Figure 3.3 RNA

(A) RNA (single-stranded)

Phosphate
Ribose

3' end
5' end

(B) RNA structure with 3' to 5' and 5' to 3' directions indicated.
DNA is usually double stranded.  

Two polynucleotide strands form a “ladder” that twists into a double helix.  

Sugar-phosphate groups form the sides of the ladder, the hydrogen-bonded bases form the rungs.
Figure 3.4 DNA

(A) DNA (double-stranded)

- Deoxyribose
- Pyrimidine base
- Purine base
- Phosphate
- Hydrogen bond

5' end

(B) 3' end

[Image showing the structure of DNA with the 5' and 3' ends labeled]
The two strands are antiparallel (running in opposite directions), and the double helix is right-handed.
Concept 3.1 Nucleic Acids Are Informational Macromolecules

DNA’s information is encoded in the sequence of bases. DNA has two functions:

- Replication
- Information is copied to RNA and used to specify amino acid sequences in proteins.
DNA replication and transcription depend on base pairing:

5′-TCAGCA-3′

3′-AGTCGT-5′

transcribes to RNA with the sequence 5′-UCAGCA-3′.
DNA replication: the entire molecule must be replicated completely so that each new cell receives a complete set of DNA.

**Genome**—complete set of DNA in a living organism

**Genes**—DNA sequences that encode specific proteins and are transcribed into RNA
Gene expression: transcription and translation of a specific gene.

Not all genes are transcribed in all cells of an organism.
Figure 3.5 DNA Replication and Transcription

(A) Replication

DNA

DNA

DNA

(B) Transcription

DNA

RNA for protein 1

RNA for protein 2
DNA base sequences reveal evolutionary relationships.

Closely related living species should have more similar base sequences than species that are more distantly related.

Scientists are now able to determine and compare entire genomes of organisms to study evolutionary relationships.
Major functions of proteins:

- *Enzymes*—catalytic molecules
- *Defensive proteins* (e.g., antibodies)
- *Hormonal and regulatory proteins*—control physiological processes
- *Receptor proteins*—receive and respond to molecular signals
- *Storage proteins*—store amino acids
Concept 3.2 Proteins Are Polymers with Important Structural and Metabolic Roles

- **Structural proteins**—physical stability and movement
- **Transport proteins**—carry substances (e.g., hemoglobin)
- **Genetic regulatory proteins**—regulate when, how, and to what extent a gene is expressed
Protein monomers are **amino acids**.

Amino and carboxyl functional groups allow them to act as both acid and base.
The **R group** (side chain) differs in each amino acid.

Only 20 amino acids occur extensively in the proteins of all organisms.

They are grouped according to properties conferred by the R groups.
TABLE 3.2 The Twenty Amino Acids in Proteins (Part 1)

A. Amino acids with electrically charged hydrophilic side chains

Positive

Arginine (Arg; R)

Histidine (His; H)

Lysine (Lys; K)

Negative

Aspartic acid (Asp; D)

Glutamic acid (Glu; E)
<table>
<thead>
<tr>
<th>B. Amino acids with polar but uncharged side chains (hydrophilic)</th>
<th>C. Special cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serine (Ser; S)</td>
<td>Cysteine (Cys; C)</td>
</tr>
<tr>
<td><img src="image1" alt="Serine structure" /></td>
<td><img src="image2" alt="Cysteine structure" /></td>
</tr>
<tr>
<td>Threonine (Thr; T)</td>
<td>Glycine (Gly; G)</td>
</tr>
<tr>
<td><img src="image3" alt="Threonine structure" /></td>
<td><img src="image4" alt="Glycine structure" /></td>
</tr>
<tr>
<td>Asparagine (Asn; N)</td>
<td>Proline (Pro; P)</td>
</tr>
<tr>
<td><img src="image5" alt="Asparagine structure" /></td>
<td><img src="image6" alt="Proline structure" /></td>
</tr>
<tr>
<td>Glutamine (Gln; Q)</td>
<td></td>
</tr>
<tr>
<td><img src="image7" alt="Glutamine structure" /></td>
<td></td>
</tr>
<tr>
<td>Tyrosine (Tyr; Y)</td>
<td></td>
</tr>
<tr>
<td><img src="image8" alt="Tyrosine structure" /></td>
<td></td>
</tr>
</tbody>
</table>

**TABLE 3.2** The Twenty Amino Acids in Proteins *(Part 2)*

*PRINCIPLES OF LIFE 2e, Table 3.2 (Part 2)*
### TABLE 3.2 The Twenty Amino Acids in Proteins (Part 3)

#### D. Amino acids with nonpolar hydrophobic side chains

<table>
<thead>
<tr>
<th>Alanine (Ala; A)</th>
<th>Isoleucine (Ile; I)</th>
<th>Leucine (Leu; L)</th>
<th>Methionine (Met; M)</th>
<th>Phenylalanine (Phe; F)</th>
<th>Tryptophan (Trp; W)</th>
<th>Valine (Val; V)</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Alanine" /></td>
<td><img src="image" alt="Isoleucine" /></td>
<td><img src="image" alt="Leucine" /></td>
<td><img src="image" alt="Methionine" /></td>
<td><img src="image" alt="Phenylalanine" /></td>
<td><img src="image" alt="Tryptophan" /></td>
<td><img src="image" alt="Valine" /></td>
</tr>
</tbody>
</table>

*PRINCIPLES OF LIFE 2e, Table 3.2 (Part 3)*

The glycine side chain is a single hydrogen atom—small enough to fit into tight corners in the interior of a protein molecule.

Proline has a ring structure that limits its hydrogen-bonding ability and its ability to rotate. It often functions to stabilize bends or loops in proteins.
Cysteine side chains can form covalent bonds called **disulfide bridges**.
Oligopeptides or peptides—short polymers of 20 or fewer amino acids (some hormones and signaling molecules)

Polypeptides are larger polymers. Functional proteins are made up of one or more polypeptides.

Proteins range in size from insulin, with 51 amino acids, to huge molecules such as the muscle protein titin, with 34,350 amino acids.
Amino acids are linked in condensation reactions to form **peptide bonds**.

Polymerization takes place in the amino to carboxyl direction.
Figure 3.6 Formation of a Peptide Bond

The formation of a peptide bond occurs when an amino group reacts with a carboxyl group in the presence of water (H₂O). This reaction results in the formation of a covalent bond between the amino group of one amino acid and the carboxyl group of another amino acid, releasing a water molecule (H₂O) in the process.

The amino group is denoted as (+H₃N) and the carboxyl group as (COO⁻). The process involves the transfer of negative charge from the carboxyl group to the amino group, forming a stable peptide bond between the two amino acids:

- **N terminus (⁺H₃N)**
- **C terminus (COO⁻)**
- **Peptide bond formation**
Primary structure of a protein is the sequence of amino acids.
Figure 3.7  The Four Levels of Protein Structure (Part 1)

(A) Primary structure

Peptide bond

Amino acid monomers
Secondary structure—regular, repeated spatial patterns in different regions, resulting from hydrogen bonding

- $\alpha$ (alpha) helix—right-handed coil
- $\beta$ (beta) pleated sheet—two or more sequences are extended and aligned
Figure 3.7  The Four Levels of Protein Structure (Part 2)

(B) Secondary structure

α Helix

Hydrogen bond

(C) β Pleated sheet

Hydrogen bond

PRINCIPLES OF LIFE 2e, Figure 3.7 (Part 2)
Tertiary structure—polypeptide chain is bent and folded; results in the definitive 3-D shape
Figure 3.7 The Four Levels of Protein Structure (Part 3)

(D) Tertiary structure

- β Pleated sheet
- Hydrogen bond
- α Helix
- Disulfide bridge

(E) Quaternary structure

- Subunit 1
- Subunit 2
- Subunit 3
- Subunit 4
Interactions between R groups determine tertiary structure:

- *Disulfide bridges* hold folded polypeptides together
- *Hydrogen bonds* stabilize folds
- *Hydrophobic* side chains can aggregate in the protein interior
- *van der Waals interactions* between hydrophobic side chains
Figure 3.8  Noncovalent Interactions between Proteins and Other Molecules
Concept 3.2 Proteins Are Polymers with Important Structural and Metabolic Roles

- Ionic interactions form salt bridges. They can also be deep within a protein, away from water.
Figure 3.9 The Structure of a Protein
Secondary and tertiary protein structure derive from primary structure.

**Denaturing**—heat or chemicals disrupt weaker interactions in a protein, destroying secondary and tertiary structure.

The protein can return to normal when cooled or the chemicals are removed—all the information needed to specify the unique shape is contained in the primary structure.
INVESTIGATION

HYPOTHESIS

Under controlled conditions that simulate the normal cellular environment a denatured protein can refold into a functional three-dimensional structure.

METHOD

Chemically denature functional ribonuclease, so that only its primary structure (i.e., an unfolded polypeptide chain) remains.

RESULTS

When the disruptive agents are removed, three-dimensional structure is restored and the protein once again is functional.
CONCLUSION

In normal cellular conditions, the primary structure of a protein specifies how it folds into a functional, three-dimensional structure.

ANALYZE THE DATA

Initially, disulfide bridges (S—S) in ribonuclease were eliminated because the sulfur atoms in cysteine were reduced (—SH). At time 0, reoxidation began and at various times, the amount of disulfide bond re-formation (blue circles) and the function of ribonuclease (enzyme activity; red circles) were measured by chemical methods. Here are the data:

A. At what time did disulfide bridges begin to form?
B. At what time did enzyme activity begin to appear?
C. Explain the difference between your answers for the times of (A) and (B).
Quaternary structure—two or more polypeptide chains (subunits) bind together by hydrophobic and ionic interactions and hydrogen bonds.
Figure 3.7  The Four Levels of Protein Structure (Part 3)
Factors that can disrupt the interactions that determine protein structure (denaturing):

- Temperature
- Change in concentration of H⁺
- High concentrations of polar substances
- Nonpolar substances
Proteins interact with other molecules.

R groups on the surface may form weak interactions (e.g., hydrogen bonds) with groups on the surface of another molecule.

This can change the tertiary structure and thus the shape of the protein.

Protein structure can also be modified by covalent bonding of a chemical group to the side chain of one or more of its amino acids.
Figure 3.11 Protein Structure Can Change

(A) Protein

Unbound molecule

Bound molecule

(B) Unmodified amino acid

Modified amino acid
Living systems depend on reactions that occur spontaneously, but at very slow rates.

**Catalysts** are substances that speed up the reactions without being permanently altered.

No catalyst makes a reaction occur that cannot otherwise occur.

Most biological catalysts are proteins (**enzymes**); a few are RNA molecules (**ribozymes**).
An exergonic reaction releases free energy \((G)\), the amount of energy in a system that is available to do work.

Without a catalyst, the reaction will be very slow because there is an energy barrier between reactants and products.

An input of energy initiates the reaction (activation energy or \(E_a\)), which puts reactants into a transition state.
Figure 3.12 Activation Energy Initiates Reactions

(A) 
- Energy barrier
- Transition state intermediate (unstable)
- Reactants (stable)
- Products

Free energy (G)

Time course of reaction

(B) 
- Stable state
- Less stable state (transition state)

Free energy (G)

Time course of reaction
Activation energy can come from heat—the molecules have more kinetic energy.

This would not work in living systems because all reactions would be accelerated, including destructive ones.
Enzymes lower the activation energy by enabling reactants to come together and react more easily.

Example: A molecule of sucrose in solution may hydrolyze in about 15 days; with sucrase present, the same reaction occurs in 1 second!
Figure 3.13 Enzymes Lower the Energy Barrier

Enzymes lower the energy barrier for a chemical reaction. The graph shows the change in free energy (G) over time for both catalyzed and uncatalyzed reactions. The energy barrier (E_a) is lower in the catalyzed reaction, allowing the reaction to proceed more efficiently. The difference in free energy between reactants and products (ΔG) is also shown for both types of reactions.
Enzymes are highly specific—each one catalyzes only one chemical reaction.

Reactants are substrates: they bind to specific sites on the enzyme—the active sites.

Specificity results from the exact 3-D shape and chemical properties of the active site.
Figure 3.14 Enzyme Action

Enzyme (sucrase)

Active site

Sucrose

Glucose

Fructose

H₂O

Water

PRINCIPLES OF LIFE 2e, Figure 3.14
The enzyme–substrate complex (ES) is held together by hydrogen bonding, electrical attraction, or temporary covalent bonding.

\[ E + S \rightarrow ES \rightarrow E + P \]

The enzyme is not changed at the end of the reaction.
Enzymes use one or more mechanisms to catalyze a reaction:

- *Inducing strain*—bonds in the substrate are stretched, putting it in an unstable transition state.
• *Substrate orientation*—substrates are brought together so that bonds can form.

• *Adding chemical groups*—R groups may be directly involved in the reaction.
Enzyme 3-D structures are so specific that they bind only one or a few related substrates.

Many enzymes change shape when the substrate binds.

The binding is like a baseball in a catcher’s mitt. The enzyme changes shape to make the binding tight—“induced fit.”
Figure 3.15  Some Enzymes Change Shape When Substrate Binds to Them
Some enzymes require ions or other molecules (cofactors) in order to function:

- *Metal ions*

- *Coenzymes* add or remove chemical groups from the substrate. They can participate in reactions with many different enzymes.

- *Prosthetic groups* (nonamino acid groups) permanently bound to their enzymes.
### TABLE 3.3 Some Examples of Enzyme Cofactors

<table>
<thead>
<tr>
<th>Type of cofactor</th>
<th>Role in catalyzed reactions</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>METAL IONS</strong></td>
<td></td>
</tr>
<tr>
<td>Iron (Fe$^{2+}$ or Fe$^{3+}$)</td>
<td>Oxidation/reduction</td>
</tr>
<tr>
<td>Copper (Cu$^{+}$ or Cu$^{2+}$)</td>
<td>Oxidation/reduction</td>
</tr>
<tr>
<td>Zinc (Zn$^{2+}$)</td>
<td>Helps bind NAD</td>
</tr>
<tr>
<td><strong>COENZYMES</strong></td>
<td></td>
</tr>
<tr>
<td>Biotin</td>
<td>Carries $-$COO$^-$</td>
</tr>
<tr>
<td>Coenzyme A</td>
<td>Carries $-$CO$-$CH$_3$</td>
</tr>
<tr>
<td>NAD</td>
<td>Carries electrons</td>
</tr>
<tr>
<td>FAD</td>
<td>Carries electrons</td>
</tr>
<tr>
<td>ATP</td>
<td>Provides/extracts energy</td>
</tr>
<tr>
<td><strong>PROSTHETIC GROUPS</strong></td>
<td></td>
</tr>
<tr>
<td>Heme</td>
<td>Binds ions, O$_2$, and electrons; contains iron cofactor</td>
</tr>
<tr>
<td>Flavin</td>
<td>Binds electrons</td>
</tr>
<tr>
<td>Retinal</td>
<td>Converts light energy</td>
</tr>
</tbody>
</table>
Rates of catalyzed reactions:
  • There is usually less enzyme than substrate present, so reaction rate levels off when all enzyme molecules are bound to substrate molecules.
    • The enzyme is said to be saturated.
Figure 3.16 Catalyzed Reactions Reach a Maximum Rate

Maximum rate

Reaction with enzyme

Reaction without enzyme

Concentration of substrate

Reaction rate
• Maximum reaction rate is used to calculate enzyme efficiency—molecules of substrate converted to product per unit time (turnover).
  • Turnover ranges from 1 to 40 million molecules per second!
Enzyme-catalyzed reactions operate in **metabolic pathways**.

- The product of one reaction is a substrate for the next reaction.
- Each step is catalyzed by a specific enzyme.

Cell have hundreds of enzymes that participate in interconnecting metabolic pathways, forming a metabolic system.
Figure 3.17 A Biochemical System
Systems biology is a new field that describes the components of metabolic pathways mathematically.

Computer algorithms are used to make predictions about what would happen if a component were altered.
Cells can regulate metabolism by controlling the amount of an enzyme.

Cells often have the ability to turn synthesis of enzymes off or on.

Activity of enzymes can also be regulated, which is often faster.
Chemical inhibitors can bind to enzymes and slow reaction rates.

Natural inhibitors regulate metabolism.

Artificial inhibitors are used to treat diseases, kill pests, and study enzyme function.
Irreversible inhibition:

- Inhibitor covalently binds to a side chain in the active site.
- The enzyme is permanently inactivated.
- Some insecticides act in this way.
Figure 3.18  Irreversible Inhibition

Acetylcholinesterase

Active site

DIPF
Reversible inhibition:

- A **competitive inhibitor** binds at the active site but no reaction occurs.
  - It competes with the natural substrate.
Reversible inhibition:

- A **noncompetitive inhibitor** binds at a site distinct from the active site, causing change in enzyme shape and function.
  - It prevents substrate binding or slows the reaction rate.
Figure 3.19 Reversible Inhibition

(A) Competitive inhibition

Competitive inhibitor → Active site → Substrate

(B) Noncompetitive inhibition

Substrate → Active site → Noncompetitive inhibitor
Allosteric regulation—non-substrate molecule binds a site other than the active site (the allosteric site)

The enzyme changes shape, which alters the chemical attraction (affinity) of the active site for the substrate.

Allosteric regulation can activate or inactivate enzymes.
Concept 3.4 Regulation of Metabolism Occurs by Regulation of Enzymes

Allosteric sites can be modified by:

- *Noncovalent binding* (reversible)
- *Covalent binding* of a molecule or chemical group, such as phosphorylation (reversible)
Figure 3.20  Allosteric Regulation of Enzyme Activity

(A) Inactive enzyme

Activator
Active site open
Substrate
Active enzyme
Products

(B) Inactive enzyme

Protein kinase
Active site open
Substrate
Active enzyme
Products
Phosphorylation by protein kinases is an important regulatory mechanism.

Phosphorylation can change a hydrophobic region to hydrophilic. The enzyme twists and exposes the active site.

Protein phosphatases reverse the process by removing phosphate groups.
Metabolic pathways:

The first reaction is the commitment step—the other reactions then happen in sequence.

**Feedback inhibition** (end-product inhibition)—the final product acts as an inhibitor of the first enzyme, which shuts down the pathway.
Figure 3.21 Feedback Inhibition of Metabolic Pathways

Threonine (starting material) → \( \alpha \)-Ketobutyrate (intermediate product) → Isoleucine (end product)
pH affects protein structure and enzyme activity:

- Acidic side chains generate $\text{H}^+$ and become anions.
- Basic side chains attract $\text{H}^+$ and become cations.
Example:

\[
\text{glutamic acid—COOH} \leftrightarrow \text{glutamic acid—COO}^- + H^+
\]

The law of mass action: the higher the H\(^+\) concentration, the more the reaction is driven to the left, to the less hydrophilic form.

This can affect enzyme shape and function.
Protein tertiary structure (and thus function) is very sensitive to the concentration of \( \text{H}^+ \) (pH) in the environment.

All enzymes have an optimal pH for activity.
Temperature affects protein structure and enzyme activity:

- Warming increases rates of chemical reactions, but if temperature is too high, noncovalent bonds can break, inactivating enzymes.

All enzymes have an optimal temperature for activity.
Figure 3.22 Enzyme Activity Is Affected by the Environment

(A) Reaction rate vs. pH for Chymotrypsin, Pepsin, and Arginase.

(B) Reaction rate vs. Temperature with an optimal temperature for maximum rate.
Isozymes catalyze the same reaction but have different composition and physical properties.

Isozymes may have different optimal temperatures or pH, allowing an organism to adapt to changes in its environment.
Aspirin binds to and inhibits the enzyme cyclooxygenase.

Cyclooxygenase catalyzes the commitment step for metabolic pathways that produce:

- Prostaglandins—involved in inflammation and pain
- Thromboxanes—stimulate blood clotting and constriction of blood vessels
Figure 3.23  Aspirin: An Enzyme Inhibitor

Arachidonic acid

2 O₂ → Cyclooxygenase → Aspirin

Prostaglandin H₂
Aspirin binds at the active site of cyclooxygenase and transfers an acetyl group to a serine residue.

Serine becomes more hydrophobic, which changes the shape of the active site and makes it inaccessible to the substrate.
Figure 3.24  Inhibition by Covalent Modification