

Contents

1 Introduction to the Study of Cell and Molecular Biology 1

1.1 The Discovery of Cells 2

1.2 Basic Properties of Cells 3

- Cells Are Highly Complex and Organized 3
- Cells Possess a Generic Program and the Means to Use It 5
- Cells Are Capable of Producing More of Themselves 5
- Cells Acquire and Utilize Energy 5
- Cells Carry Out a Variety of Chemical Reactions 6
- Cells Engage in Mechanical Activities 6
- Cells Are Able to Respond to Stimuli 6
- Cells Are Capable of Self-Regulation 6
- Cells Evolve 7

1.3 Two Fundamentally Different Classes of Cells 7

- Characteristics That Distinguish Prokaryotic and Eukaryotic Cells 8
- Types of Prokaryotic Cells 14
- Types of Eukaryotic Cells: Cell Specialization 15
- The Sizes of Cells and Their Components 17
- Synthetic Biology 17
- **THE HUMAN PERSPECTIVE:** The Prospect of Cell Replacement Therapy 20

1.4 Viruses 23

- Viroids 26
- **EXPERIMENTAL PATHWAYS:** The Origin of Eukaryotic Cells 26

2 The Chemical Basis of Life 32

2.1 Covalent Bonds 33

- Polar and Nonpolar Molecules 34
- Ionization 34

2.2 Noncovalent Bonds 34

- **THE HUMAN PERSPECTIVE:** Free Radicals as a Cause of Aging 35
- Ionic Bonds: Attractions between Charged Atoms 35
- Hydrogen Bonds 36
- Hydrophobic Interactions and van der Waals Forces 36
- The Life-Supporting Properties of Water 37

2.3 Acids, Bases, and Buffers 39

2.4 The Nature of Biological Molecules 40

- Functional Groups 41
- A Classification of Biological Molecules by Function 41

2.5 Four Types of Biological Molecules 42

- Carbohydrates 43

Lipids 47

Proteins 50

- **THE HUMAN PERSPECTIVE:** Protein Misfolding Can Have Deadly Consequences 66

Nucleic Acids 77

2.6 The Formation of Complex Macromolecular Structures 79

- The Assembly of Tobacco Mosaic Virus Particles and Ribosomal Subunits 79
- **EXPERIMENTAL PATHWAYS:** Chaperones: Helping Proteins Reach Their Proper Folded State 80

3 Bioenergetics, Enzymes, and Metabolism 86

3.1 Bioenergetics 87

- The Laws of Thermodynamics and the Concept of Entropy 87
- Free Energy 89

3.2 Enzymes as Biological Catalysts 94

- The Properties of Enzymes 95
- Overcoming the Activation Energy Barrier 96
- The Active Site 97
- Mechanisms of Enzyme Catalysis 99
- Enzyme Kinetics 102
- **THE HUMAN PERSPECTIVE:** The Growing Problem of Antibiotic Resistance 106

3.3 Metabolism 108

- An Overview of Metabolism 108
- Oxidation and Reduction: A Matter of Electrons 109
- The Capture and Utilization of Energy 110
- Metabolic Regulation 115

4 The Structure and Function of the Plasma Membrane 120

4.1 An Overview of Membrane Functions 121

4.2 A Brief History of Studies on Plasma Membrane Structure 123

4.3 The Chemical Composition of Membranes 125

- Membrane Lipids 125
- The Asymmetry of Membrane Lipids 128
- Membrane Carbohydrates 129

4.4 The Structure and Functions of Membrane Proteins 130

- Integral Membrane Proteins 130
- Studying the Structure and Properties of Integral Membrane Proteins 132

Peripheral Membrane Proteins	137
Lipid-Anchored Membrane Proteins	137
4.5 Membrane Lipids and Membrane Fluidity	138
The Importance of Membrane Fluidity	139
Maintaining Membrane Fluidity	139
Lipid Rafts	139
4.6 The Dynamic Nature of the Plasma Membrane	140
The Diffusion of Membrane Proteins after Cell Fusion	141
Restrictions on Protein and Lipid Mobility	142
The Red Blood Cell: An Example of Plasma Membrane Structure	145
4.7 The Movement of Substances Across Cell Membranes	147
The Energetics of Solute Movement	147
Diffusion of Substances through Membranes	149
Facilitated Diffusion	156
Active Transport	157
● THE HUMAN PERSPECTIVE: Defects in Ion Channels and Transporters as a Cause of Inherited Disease	162
4.8 Membrane Potentials and Nerve Impulses	164
The Resting Potential	164
The Action Potential	165
Propagation of Action Potentials as an Impulse	167
Neurotransmission: Jumping the Synaptic Cleft	168
● EXPERIMENTAL PATHWAYS: The Acetylcholine Receptor	171
5 Aerobic Respiration and the Mitochondrion	178
5.1 Mitochondrial Structure and Function	179
Mitochondrial Membranes	180
The Mitochondrial Matrix	182
5.2 Oxidative Metabolism in the Mitochondrion	183
The Tricarboxylic Acid (TCA) Cycle	185
The Importance of Reduced Coenzymes in the Formation of ATP	186
● THE HUMAN PERSPECTIVE: The Role of Anaerobic and Aerobic Metabolism in Exercise	188
5.3 The Role of Mitochondria in the Formation of ATP	189
Oxidation-Reduction Potentials	189
Electron Transport	190
Types of Electron Carriers	191
5.4 Translocation of Protons and the Establishment of a Proton-Motive Force	198
5.5 The Machinery for ATP Formation	199
The Structure of ATP Synthase	200
The Basis of ATP Formation According to the Binding Change Mechanism	201

Other Roles for the Proton-Motive Force in Addition to ATP Synthesis 205

5.6 Peroxisomes 206

● **THE HUMAN PERSPECTIVE:** Diseases that Result from Abnormal Mitochondrial or Peroxisomal Function 207

6 Photosynthesis and the Chloroplast 211

6.1 Chloroplast Structure and Function 213

6.2 An Overview of Photosynthetic Metabolism 214

6.3 The Absorption of Light 216

Photosynthetic Pigments 216

6.4 Photosynthetic Units and Reaction Centers 218

Oxygen Formation: Coordinating the Action of Two Different Photosynthetic Systems 218

Killing Weeds by Inhibiting Electron Transport 225

6.5 Photophosphorylation 225

Noncyclic Versus Cyclic Photophosphorylation 226

6.6 Carbon Dioxide Fixation and the Synthesis of Carbohydrate 226

Carbohydrate Synthesis in C₃ Plants 226

Carbohydrate Synthesis in C₄ Plants 231

Carbohydrate Synthesis in CAM Plants 232

7 Interactions Between Cells and Their Environment 235

7.1 The Extracellular Space 236

The Extracellular Matrix 236

7.2 Interactions of Cells with Extracellular Materials 244

Integrins 244

Focal Adhesions and Hemidesmosomes: Anchoring Cells to Their Substratum 247

7.3 Interactions of Cells with Other Cells 250

Selectins 251

The Immunoglobulin Superfamily 252

Cadherins 253

● **THE HUMAN PERSPECTIVE:** The Role of Cell Adhesion in Inflammation and Metastasis 255

Adherens Junctions and Desmosomes: Anchoring Cells to Other Cells 257

The Role of Cell-Adhesion Receptors in Transmembrane Signaling 259

7.4 Tight Junctions: Sealing The Extracellular Space 260

7.5 Gap Junctions and Plasmodesmata: Mediating Intercellular Communication 262

Plasmodesmata 265

7.6 Cell Walls 266

Structure, Function, and Membrane Trafficking 270

8.1 An Overview of the Endomembrane System 271

8.2 A Few Approaches to the Study of Endomembranes 273

Insights Gained from Autoradiography 273

Insights Gained from the Use of the Green Fluorescent Protein 273

Insights Gained from the Biochemical Analysis of Subcellular Fractions 275

Insights Gained from the Use of Cell-Free Systems 276

Insights Gained from the Study of Mutant Phenotypes 277

8.3 The Endoplasmic Reticulum 279

The Smooth Endoplasmic Reticulum 280

Functions of the Rough Endoplasmic Reticulum 280

From the ER to the Golgi Complex: The First Step in Vesicular Transport 289

8.4 The Golgi Complex 290

Glycosylation in the Golgi Complex 292

The Movement of Materials through the Golgi Complex 292

8.5 Types of Vesicle Transport and Their Functions 295

COPII-Coated Vesicles: Transporting Cargo from the ER to the Golgi Complex 296

COPI-Coated Vesicles: Transporting Escaped Proteins Back to the ER 298

Beyond the Golgi Complex: Sorting Proteins at the TGN 298
Targeting Vesicles to a Particular Compartment 300

8.6 Lysosomes 303

Autophagy 304

● **THE HUMAN PERSPECTIVE:** Disorders Resulting from Defects in Lysosomal Function 306

8.7 Plant Cell Vacuoles 307

8.8 The Endocytic Pathway: Moving Membrane and Materials into the Cell Interior 308

Endocytosis 308

Phagocytosis 315

8.9 Posttranslational Uptake of Proteins by Peroxisomes, Mitochondria, and Chloroplasts 316

Uptake of Proteins into Peroxisomes 316

Uptake of Proteins into Mitochondria 316

Uptake of Proteins into Chloroplasts 318

● **EXPERIMENTAL PATHWAYS:** Receptor-Mediated Endocytosis 319

9 The Cytoskeleton and Cell Motility 324

9.1 Overview of the Major Functions of the Cytoskeleton 325

9.2 The Study of the Cytoskeleton 326

The Use of In Vitro and In Vivo Single-Molecule Assays 327

The Use of Fluorescence Imaging Techniques to Monitor the Dynamics of the Cytoskeleton 329

9.3 Microtubules 330

Structure and Composition 330

Microtubule-Associated Proteins 331

Microtubules as Structural Supports and Organizers 332

Microtubules as Agents of Intracellular Motility 333

Motor Proteins that Traverse the Microtubular Cytoskeleton 334

Microtubule-Organizing Centers (MTOCs) 339

The Dynamic Properties of Microtubules 341

Cilia and Flagella: Structure and Function 345

● **THE HUMAN PERSPECTIVE:** The Role of Cilia in Development and Disease 349

9.4 Intermediate Filaments 354

Intermediate Filament Assembly and Disassembly 354

Types and Functions of Intermediate Filaments 356

9.5 Microfilaments 356

Microfilament Assembly and Disassembly 358

Myosin: The Molecular Motor of Actin Filaments 360

9.6 Muscle Contractility 364

The Sliding Filament Model of Muscle Contraction 366

9.7 Nonmuscle Motility 371

Actin-Binding Proteins 372

Examples of Nonmuscle Motility and Contractility 374

10 The Nature of the Gene and the Genome 386

10.1 The Concept of a Gene as a Unit of Inheritance 387

10.2 Chromosomes: The Physical Carriers of the Genes 388

The Discovery of Chromosomes 388

Chromosomes as the Carriers of Genetic Information 389

Genetic Analysis in *Drosophila* 390

Crossing Over and Recombination 390

Mutagenesis and Giant Chromosomes 392

10.3 The Chemical Nature of the Gene 393

The Structure of DNA 393

The Watson-Crick Proposal 394

DNA Supercoiling 397

10.4 The Structure of the Genome 398

The Complexity of the Genome 399

● **THE HUMAN PERSPECTIVE:** Diseases that Result from Expansion of Trinucleotide Repeats 404

10.5 The Stability of the Genome 406

Whole-Genome Duplication (Polyploidization) 406

Duplication and Modification of DNA Sequences 407

"Jumping Genes" and the Dynamic Nature of the Genome 408

10.6 Sequencing Genomes: The Footprints of Biological Evolution 411

Comparative Genomics: "If It's Conserved, It Must Be Important" 413

The Genetic Basis of "Being Human" 414

Genetic Variation Within the Human Species Population 416

● **THE HUMAN PERSPECTIVE:** Application of Genomic Analyses to Medicine 417

● **EXPERIMENTAL PATHWAYS:** The Chemical Nature of the Gene 420

11 Gene Expression: From Transcription to Translation 426

11.1 The Relationship between Genes, Proteins, and RNAs 427

An Overview of the Flow of Information through the Cell 428

11.2 An Overview of Transcription in Both Prokaryotic and Eukaryotic Cells 429

Transcription in Bacteria 432

Transcription and RNA Processing in Eukaryotic Cells 433

11.3 Synthesis and Processing of Eukaryotic Ribosomal and Transfer RNAs 435

Synthesizing the rRNA Precursor 436

Processing the rRNA Precursor 437

Synthesis and Processing of the 5S rRNA 440

Transfer RNAs 440

11.4 Synthesis and Processing of Eukaryotic Messenger RNAs 441

The Machinery for mRNA Transcription 441

Split Genes: An Unexpected Finding 444

The Processing of Eukaryotic Messenger RNAs 448

Evolutionary Implications of Split Genes and RNA Splicing 454

Creating New Ribozymes in the Laboratory 454

11.5 Small Regulatory RNAs and RNA Silencing Pathways 455

● **THE HUMAN PERSPECTIVE:** Clinical Applications of RNA Interference 458

MicroRNAs: Small RNAs that Regulate Gene Expression 459

piRNAs: A Class of Small RNAs that Function in Germ Cells 460

Other Noncoding RNAs 461

11.6 Encoding Genetic Information 461

The Properties of the Genetic Code 461

11.7 Decoding the Codons: The Role of Transfer RNAs 464

The Structure of tRNAs 465

11.8 Translating Genetic Information 468

Initiation 468

Elongation 471

Termination 474

mRNA Surveillance and Quality Control 474

Polyribosomes 475

● **EXPERIMENTAL PATHWAYS:** The Role of RNA as a Catalyst 477

12 Control of Gene Expression 483

12.1 Control of Gene Expression in Bacteria 484

Organization of Bacterial Genomes 484

The Bacterial Operon 484

Riboswitches 487

12.2 Control of Gene Expression in Eukaryotes:

Structure and Function of the Cell Nucleus 488

The Nuclear Envelope 488

Chromosomes and Chromatin 493

● **THE HUMAN PERSPECTIVE:** Chromosomal Aberrations and Human Disorders 504

Epigenetics: There's More to Inheritance than DNA 509

The Nucleus as an Organized Organelle 510

12.3 An Overview of Gene Regulation in Eukaryotes 512

12.4 Transcriptional Control 514

The Role of Transcription Factors in Regulating Gene Expression 517

The Structure of Transcription Factors 519

DNA Sites Involved in Regulating Transcription 522

Transcriptional Activation: The Role of Enhancers, Promoters, and Coactivators 525

Transcriptional Repression 530

12.5 RNA Processing Control 533

12.6 Translational Control 536

Initiation of Translation 536

Cytoplasmic Localization of mRNAs 537

The Control of mRNA Stability 538

The Role of MicroRNAs in Translational Control 539

12.7 Posttranslational Control: Determining Protein Stability 541

13 DNA Replication and Repair 545

13.1 DNA Replication 546

Semiconservative Replication 546

Replication in Bacterial Cells 549

The Structure and Functions of DNA Polymerases 554

Replication in Eukaryotic Cells 558

13.2 DNA Repair 564

Nucleotide Excision Repair 565

Base Excision Repair 566

Mismatch Repair 567

Double-Strand Breakage Repair 567

13.3 Between Replication and Repair 568

- **THE HUMAN PERSPECTIVE:** The Consequences of DNA Repair Deficiencies 569

14 Cellular Reproduction 572

14.1 The Cell Cycle 573

- Cell Cycles in Vivo 574
- Control of the Cell Cycle 574

14.2 M Phase: Mitosis and Cytokinesis 581

- Prophase 583
- Prometaphase 588
- Metaphase 590
- Anaphase 592
- Telophase 597
- Motor Proteins Required for Mitotic Movements 597
- Cytokinesis 597

14.3 Meiosis 602

- The Stages of Meiosis 603
- **THE HUMAN PERSPECTIVE:** Meiotic Nondisjunction and Its Consequences 608
- Genetic Recombination During Meiosis 610
- **EXPERIMENTAL PATHWAYS:** The Discovery and Characterization of MPF 611

15 Cell Signaling and Signal Transduction: Communication Between Cells 617

15.1 The Basic Elements of Cell Signaling Systems 618

15.2 A Survey of Extracellular Messengers and Their Receptors 621

15.3 G Protein-Coupled Receptors and Their Second Messengers 621

- Signal Transduction by G Protein-Coupled Receptors 622
- **THE HUMAN PERSPECTIVE:** Disorders Associated with G Protein-Coupled Receptors 625
- Second Messengers 627
- The Specificity of G Protein-Coupled Responses 630
- Regulation of Blood Glucose Levels 631
- The Role of GPCRs in Sensory Perception 634

15.4 Protein-Tyrosine Phosphorylation as a Mechanism for Signal Transduction 636

- The Ras-MAP Kinase Pathway 640
- Signaling by the Insulin Receptor 644
- **THE HUMAN PERSPECTIVE:** Signaling Pathways and Human Longevity 647
- Signaling Pathways in Plants 648

15.5 The Role of Calcium as an Intracellular Messenger 648

- Regulating Calcium Concentrations in Plant Cells 652

15.6 Convergence, Divergence, and Cross-Talk Among Different Signaling Pathways 653

- Examples of Convergence, Divergence, and Cross-Talk Among Signaling Pathways 654

15.7 The Role of NO as an Inter cellular Messenger 655

15.8 Apoptosis (Programmed Cell Death) 656

- The Extrinsic Pathway of Apoptosis 658
- The Intrinsic Pathway of Apoptosis 659

16 Cancer 664

16.1 Basic Properties of a Cancer Cell 665

16.2 The Causes of Cancer 667

16.3 The Genetics of Cancer 669

- Tumor-Suppressor Genes and Oncogenes: Brakes and Accelerators 671
- The Cancer Genome 683
- Gene-Expression Analysis 685

16.4 New Strategies for Combating Cancer 687

- Immunotherapy 688
- Inhibiting the Activity of Cancer-Promoting Proteins 689
- Inhibiting the Formation of New Blood Vessels (Angiogenesis) 692
- **EXPERIMENTAL PATHWAYS:** The Discovery of Oncogenes 694

17 The Immune Response 699

17.1 An Overview of the Immune Response 700

- Innate Immune Responses 700
- Adaptive Immune Responses 703

17.2 The Clonal Selection Theory as It Applies to B Cells 704

- Vaccination 706

17.3 T Lymphocytes: Activation and Mechanism of Action 707

17.4 Selected Topics on the Cellular and Molecular Basis of Immunity 710

- The Modular Structure of Antibodies 710
- DNA Rearrangements that Produce Genes Encoding B- and T-Cell Antigen Receptors 713
- Membrane-Bound Antigen Receptor Complexes 716
- The Major Histocompatibility Complex 716
- Distinguishing Self from Nonself 721
- Lymphocytes Are Activated by Cell-Surface Signals 722
- Signal Transduction Pathways in Lymphocyte Activation 723
- **THE HUMAN PERSPECTIVE:** Autoimmune Diseases 724
- **EXPERIMENTAL PATHWAYS:** The Role of the Major Histocompatibility Complex in Antigen Presentation 727

18 Techniques in Cell and Molecular Biology 732

18.1 The Light Microscope 733

- Resolution 733
- Visibility 734
- Preparation of Specimens for Bright-Field Light Microscopy 735
- Phase-Contrast Microscopy 735
- Fluorescence Microscopy (and Related Fluorescence-Based Techniques) 736
- Video Microscopy and Image Processing 738
- Laser Scanning Confocal Microscopy 739
- Super-Resolution Fluorescence Microscopy 740

18.2 Transmission Electron Microscopy 740

- Specimen Preparation for Electron Microscopy 742

18.3 Scanning Electron and Atomic Force Microscopy 746

- Atomic Force Microscopy 748

18.4 The Use of Radioisotopes 748

18.5 Cell Culture 749

18.6 The Fractionation of a Cell's Contents by Differential Centrifugation 752

18.7 Isolation, Purification, and Fractionation of Proteins 752

- Selective Precipitation 752
- Liquid Column Chromatography 753
- Polyacrylamide Gel Electrophoresis 756
- Protein Measurement and Analysis 757

18.8 Determining the Structure of Proteins and Multisubunit Complexes 758

18.9 Fractionation of Nucleic Acids 760

- Separation of DNAs by Gel Electrophoresis 760
- Separation of Nucleic Acids by Ultracentrifugation 760

18.10 Nucleic Acid Hybridization 762

18.11 Chemical Synthesis of DNA 764

18.12 Recombinant DNA Technology 764

- Restriction Endonucleases 764
- Formation of Recombinant DNAs 766
- DNA Cloning 766

18.13 Enzymatic Amplification of DNA by PCR 769

- Applications of PCR 770

18.14 DNA Sequencing 771

18.15 DNA Libraries 773

- Genomic Libraries 773
- cDNA Libraries 774

18.16 DNA Transfer into Eukaryotic Cells and Mammalian Embryos 775

18.17 Determining Eukaryotic Gene Function by Gene Elimination or Silencing 778

- In Vitro Mutagenesis 778
- Knockout Mice 778
- RNA Interference 780

18.18 The Use of Antibodies 780

Glossary G-1

Additional Readings A-1

Index I-1